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UNITED STATES AIR FORCE

GRADUATE STUDENT RESEARCH PROGRAM

1988

PROGRAM TECHNICAL REPORT

UNIVERSAL ENERGY SYSTEMS, INC.

VOLUME 3 of 3

PROGRAM DIRECTOR, UES
RODNEY C. DARRAH

PROGRAM ADMINISTRATOR, UES
SUSAN K. ESPY

PROGRAM MANAGER, AFOSR
LT. COL. CLAUDE CAVENDER

SUBMITTED TO
AIR FORCE OFFICE OF SCIENTIFIC RESEARCH
BOLLING AIR FORCE BASE
WASHINGTON, DC

DECEMBER 1988

UNITED STATES AIR FORCE

GRADUATE STUDENT RESEARCH PROGRAM

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Program Director, UES
Rodney C. Darrah

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Lt. Col. Claude Cavender

Program Administrator, UES
Susan K. Espy

Submitted to

Air Force Office of Scientific Research

Bolling Air Force Base

Washington, DC

December 1988

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PREFACE

The United States Air Force Graduate Student Research Program (USAF-GSRP) is conducted under the United States Air Force Summer Faculty Research Program. The program provides funds for selected graduate students to work at an appropriate Air Force Facility with a supervising professor who holds a concurrent Summer Faculty Research Program appointment or with a supervising Air Force Engineer/Scientist. This is accomplished by the students being selected on a nationally advertised competitive basis for a ten-week assignment during the summer intersession period to perform research at Air Force laboratories/centers. Each assignment is in a subject area and at an Air Force facility mutually agreed upon by the students and the Air Force. In addition to compensation, travel and cost of living allowances are also paid. The USAF-GSRP is sponsored by the Air Force Office of Scientific Research, Air Force Systems Command, United States Air Force, and is conducted by Universal Energy Systems, Inc.

The specific objectives of the 1988 USAF-GSRP are:

(1) To provide a productive means for the graduate students to participate in research at the Air Force Laboratories/Centers;

(2) To stimulate continuing professional association among the Graduate Students and their professional peers in the Air Force;

(3) To further the research objectives of the United States Air Force;

(4) To enhance the research productivity and capabilities of the graduate students especially as these relate to Air Force technical interests.

During the summer of 1988, 107 graduate students participated. These researchers were assigned to 23 USAF laboratories/centers across the country. This three volume document is a compilation of the final reports written by the assigned students members about their summer research efforts.

LIST OF 1988 PARTICIPANTS

NAME/ADDRESS	DEGREE, SPECIALTY, LABORATORY ASSIGNED
Ben A. Abbott Electrical Engineering Dept. Vanderbilt University Nashville, TN 37240 (615) 332-2723	Degree: B.S., Computer Science, 1983 Specialty: Electrical Engineering Assigned: Arnold Engineering Development Center
Antoinne C. Able Meharry Medical College 1005 D.B. Todd Blvd. P O Box 882 Nashville, TN 37208 (615) 361-5303	Degree: M.S., Biology, 1982 Specialty: Biology Assigned: Wilford Hall Medical Center
Stanley D. Adams College of Medicine Meharry Medical College 1005 D.B. Todd Blvd. Nashville, TN 37208 (615) 327-6204	Degree: B.S., Cellular & Molecular Biology, 1987 Specialty: Physiology Assigned: Wilford Hall Medical Center
John D. Allison Dept. of Psychology Univ. of Texas at Austin Mezes Hall 330 Austin, TX 78712 (512) 471-5857	Degree: M.A., Psychology, 1987 Specialty: Comparative Neurobiology Assigned: Human Resources Laboratory: Manpower and Personnel Div.
James E. Angelo Dept. of Physics Univ. of Minnesota - Duluth Duluth, MN 55812 (218) 726-7124	Degree: B.S., Math/Physics, 1986 Specialty: Applied Physics Assigned: Materials Laboratory
John E. Bambery Dept. of Physics University of Pennsylvania Indiana, PA 15701 (412) 357-2611	Degree: B.S. Physics, 1987 Specialty: Computer Analysis Assigned: Avionics Laboratory

Daniel W. Barineau Dept. of Engineering Science Virginia Tech. 1300-B Terrace View Apts. Blacksburg, VA 24060 (703) 552-7867	Degree: Specialty: Assigned:	B.S., Chemical Eng., 1987 Engineering Mechanics Harry G. Armstrong Aerospace Medical Research Laboratory
John W.J. Barnaby Dept. of Electrical Engineering University of Alabama Box 6169, 317 Houser Hall Tuscaloosa, AL 35487-6169 (205) 348-6351	Degree: Specialty: Assigned:	B.S., Electrical Eng., 1987 Electrical Engineering School of Aerospace Medicine
Kathleen M. Bennett Dept. of Engineering Mgmt. University of Dayton 300 College Park Dayton, OH 45469 (513) 229-2699	Degree: Specialty: Assigned:	B.S., Mechanical Eng., 1984 Engineering Management Flight Dynamics Laboratory
Mark N. Beorkrem Dept. of Psychology Washington University One Brookings Drive Campus Box 1125 St. Louis, MO 63130 (314) 889-6536	Degree: Specialty: Assigned:	B.S., Psychology, 1987 Organizational Behavior Human Resources Laboratory: Operations Training Div.
Joel L. Berg Dept. of Eng. Science & Mech. Virginia Polytechnic Inst.&S.U. Blacksburg, VA 24061 (703) 961-6326	Degree: Specialty: Assigned:	M.S., Engr. Mechanics, 1984 Structural Vibrations Astronautics Laboratory
Darwin L. Boyd Dept. of Physics Kent State University Smith Laboratory of Physics Kent, OH 44242 (216) 672-2880	Degree: Specialty: Assigned:	B.S. Physics, 1982 Condensed Matter Physics Materials Laboratory
George C. Boynton Dept. of Physics University of Miami P O Box 248046 Coral Gables, FL 33124 (305) 284-2323	Degree: Specialty: Assigned:	M.S., Physics, 1983 Physics Armament Laboratory

Mark L. Brusseau
Dept. of Soil Science
University of Florida
2169 McCarty Hall
Gainesville, FL 32611-0151
(904) 392-1951

Degree: M.S., Geology, 1984
Specialty: Contaminant Hydrology
Assigned: Engineering & Services Center

Bruce W. Bullard
Dept. of Electrical Eng.
University of Colorado
1420 Austin Bluffs Pkwy.
P O Box 7150
Colorado Springs, CO 80933-7150
(719) 593-3351

Degree: B.S., Electrical Eng., 1988
Specialty: Electrical Engineering
Assigned: Frank. J. Seiler Research Lab.

Franklin A. Bynum
Dept. of Physics
Miami University
Culler Hall
Oxford, OH 45056
(513) 529-5657

Degree: M.S., Physics, 1988
Specialty: Physics
Assigned: Weapons Laboratory

Kevin L. Carmichael
Dept. of Physics
Wright State University
Dayton, OH 45435
(513) 873-2954

Degree: B.S., Physics, 1987
Specialty: Solid State Physics
Assigned: Avionics Laboratory

Lance H. Carter
Dept. of Aerospace Eng.
Virginia Polytechnic Inst.&S.U.
817 Claytor Square
Blacksburg, VA 24060
(703) 953-2289

Degree: B.S., Aerospace Eng., 1987
Specialty: Engineering Mechanics
Assigned: Astronautics Laboratory

David B. Chenault
Dept. of Physics
University of Alabama
Center for Applied Optics
Huntsville, AL 35899
(205) 895-6102

Degree: B.S. Physics, 1986
Specialty: Physics
Assigned: Armament Laboratory

Daniel B. Cook
Dept. of Electro-Optics
University of Dayton
300 College Park
Dayton, OH 45469
(513) 228-4111

Degree: B.S., Physics, 1987
Specialty: Image Processing
Assigned: Avionics Laboratory

Patricia P. Cooper
Dept. of Applied Psychology
Francis Marion College
Florence, SC 29501
(803) 661-1378

<u>Degree:</u> M.A., Information Sci., 1974
<u>Specialty:</u> Psychology
<u>Assigned:</u> Human Resources Laboratory: Operations Training Div.

Otis Cosby, Jr.
School of Medicine
Meharry Medical College
1005 D.B. Todd Blvd.
Nashville, TN 37208
(615) 327-6223

<u>Degree:</u> B.S., Natural Science, 1983
<u>Specialty:</u> Natural Science
<u>Assigned:</u> School of Aerospace Medicine

Richard E. Courtney
Dept. of Computing Info. & Sci.
Kansas State University
234 Nichols Hall
Manhattan, KS 66506
(913) 532-6350

<u>Degree:</u> M.S., Computer Science, 1986
<u>Specialty:</u> Computer Science
<u>Assigned:</u> Rome Air Development Center

Jerry W. Dillon
School of Dentistry
Meharry Medical College
1005 D.B. Todd Blvd.
Nashville, TN 37208
(615) 327-6207

<u>Degree:</u> M.S., Microbiology, 1988
<u>Specialty:</u> Microbiology
<u>Assigned:</u> School of Aerospace Medicine

Charles C. Drake
Dept. of Computer Science
Jackson State University
1400 Lynch StreetTraining Systems
Jackson, MS 39203
(601) 968-2105

<u>Degree:</u> B.S., Computer Science, 1987
<u>Specialty:</u> Computer Science
<u>Assigned:</u> Human Resources Laboratory:

Susan M. Dumbacher
Dept. of Aerospace Eng.
University of Cincinnati
Cincinnati, OH 45225
(513) 475-6185

<u>Degree:</u> B.S., Aerospace Eng., 1986
<u>Specialty:</u> Controls
<u>Assigned:</u> Flight Dynamics Laboratory

Michael K. Ellis
Dept. of Computer Science/Eng.
University of Arkansas
1900 N. Garland
Fayetteville, AR 72703
(501) 575-0722

<u>Degree:</u> B.S., Computer Sci., 1988
<u>Specialty:</u> Neural Network
<u>Assigned:</u> Harry G. Armstrong Aerospace Medical Research Laboratory

Bryan C. Foos
Dept. of Civil Engineering
Ohio State University
2070 Neil Avenue
Columbus, OH 43210
(614) 292-2771

Degree: B.S., Civil Engineering, 1988
Specialty: Geotechnical and Materials
Assigned: Flight Dynamics Laboratory

Ernest J. Freeman
Dept. of Biological Sciences
Kent State University
Kent, OH 44240
(216) 672-2363

Degree: B.S., Zoology, 1985
Specialty: Neurochemistry
Assigned: School of Aerospace Medicine

Peter Gaddis, Jr.
Dept. of Sociology
Jackson State University
1400 J.R. Lynch Street
Jackson, MS 39217
(601) 968-2350

Degree: M.A., Sociology, 1988
Specialty: Alcohol and Drug Studies
Assigned: Human Resources Laboratory: Manpower and Personnel Div.

Douglas P. Gagne
Dept. of Mechanical Eng.
University of New Hampshire
Durham, NH 03824
(603) 868-6160

Degree: B.S., Mechanical Eng., 1988
Specialty: Systems Modeling & Dynamics
Assigned: Materials Laboratory

William L. Geisler
College of Polymer Science
University of Akron
Akron, OH 44325
(216) 375-7500

Degree: B.S., Chemical Eng., 1988
Specialty: Polymeric Materials
Assigned: Astronautics Laboratory

Robert L. Goetz
Dept. of Mechanical Eng.
Ohio University
Athens, OH 45701
(614) 594-3499

Degree: B.S., Mechanical Eng., 1987
Specialty: Mechanical Design
Assigned: Materials Laboratory

David L. Graham
Dept. of Mechanical Eng.
Northwestern University
Tech. 2524
Evanston, IL 60208
(312) 491-3589

Degree: B.S., Mechanical Eng., 1988
Specialty: Mechanical Engineering
Assigned: Astronautics Laboratory

Gary E. Griesheim Dept. of Civil Engineering Southern Illinois University Carbondale, IL 62901 (618) 536-2368	Degree: Specialty: Assigned:	B.S., Eng. Mechanics, 1987 Composite Materials Design Astronautics Laboratory
Edward A. Grissom Dept. of Electrical Eng. Tennessee Tech. University 1217 Springdale Cookeville, TN 38501 (615) 526-1036	Degree: Specialty: Assigned:	B.S., Electrical Eng., 1983 Digital Signal Processing Avionics Laboratory
Virginia A. Gunther Dept. of Psychology State Univ. of New York at Binghamton Binghamton, NY 13901 (607) 777-4610	Degree: Specialty: Assigned:	M.A., Exp. Psychology, 1978 Cognitive Psychology Harry G. Armstrong Aerospace Medical Research Laboratory
Douglas R. Hansen Dept. of Civil Engineering Colorado State University Room B302 Ft. Collins, CO 80523 (303) 491-8353	Degree: Specialty: Assigned:	B.S., Wildlife Biology, 1981 Environmental Engineering Engineering & Service Center
Thomas K. Harkins Dept. of Mechanical Eng. Louisiana State University 1636 Applewood Road Baton Rouge, LA 70808 (504) 766-3671	Degree: Specialty: Assigned:	B.S., Mechanical Eng., 1986 Flight Dynamics Armament Laboratory
Gary A. Hellenga Dept. of Mathematical Sciences Montana State University 1116 South Hedges Bozeman, MT 59715 (406) 994-5360	Degree: Specialty: Assigned:	B.S., Mathematics, 1983 Applied Mathematics Rome Air Development Center
Andrew Hensley Dept. of Mechanical Eng. University of Detroit Detroit, MI 48603 (313) 927-1242	Degree: Specialty: Assigned:	B.S., Mechanical Eng., 1988 Process Modeling Materials Laboratory

Norman C. Holmes
Dept. of Mechanical Engineering
University of New Hampshire
Durham, NH 03824-3541
(617) 662-6386

Degree: B.S., Mechanical Eng., 1984
Specialty: Mechanical Engineering
Assigned: Flight Dynamics Laboratory

Stephen R. Jenei
Dept. of Biology
University of Dayton
300 College Park Drive
Dayton, OH 45469-0001
(513) 229-2135

Degree: B.S., Biology, 1986
Specialty: Endocrine Physiology
Assigned: Harry G. Armstrong Aerospace Medical Research Laboratory

Alan C. Jewell
Dept. of Civil Engineering
Colorado State University
Fort Collins, CO 80521
(303) 491-5048

Degree: B.S., Geophysical Eng., 1984
Specialty: Geophysical Engineering
Assigned: Weapons Laboratory

Jennifer A. Joyce
Dept. of Chemistry
Texas A&M University
College Station, TX 77843-3255
(409) 845-5345

Degree: B.S., Chemistry, 1988
Specialty: Inorganic Chemistry
Assigned: Frank. J. Seiler Research Lab.

Steven P. Kahn
Dept. of Eng. Science & Mech.
Virginia Tech. University
Blacksburg, VA 24060
(703) 953-1966

Degree: B.S., Eng. Science and Mechanics, 1988
Specialty: Engineering Mechanics
Assigned: Astronautics Laboratory

Elizabeth J. Kavran
Dept. of Biology
University of Dayton
300 College Park Road
Dayton, OH 45429
(513) 229-7660

Degree: B.A., Biology, 1987
Specialty: Biology
Assigned: Harry G. Armstrong Aerospace Medical Research Laboratory

Phyllis Y. Keys
Dept. of General Engineering
University of Illinois
909 S. Fifth Street
Champaign, IL 61820
(217) 332-5010

Degree: B.E., General Eng., 1987
Specialty: Human Factors
Assigned: Occupational and Environment Health Laboratory

Thomas E. Kimble
Center for Space Sciences
Physics Program
Univ. of Texas at Dallas
2601 North Floyd Road
P O Box 830688, M.S. FO-22
Richardson, TX 75083-0688
(214) 690-2884

Degree: B.A., Economics, 1981
Specialty: Space Sciences
Assigned: Air Force Geophysics Lab.

Charles L. King
Dept. of Industrial Eng.
University of Arkansas
4207 Bell Engineering Ctr.
Fayetteville, AR 72701
(501) 575-3156

Degree: B.S., Industrial Eng., 1988
Specialty: Management
Assigned: Human Resources Laboratory: Logistics & Human Factors Div.

Scharine Kirchoff
Geophysical Institute
Univ. of Alaska - Fairbanks
P O Box 83328
Fairbanks, AK 99708
(907) 479-5866

Degree: M.A., Geology, 1986
Specialty: Geophysics
Assigned: Air Force Geophysics Lab.

Christopher G. Kocher
Dept. of Civil Engineering
Southern Illinois University
Carbondale, IL 62901
(618) 536-2368

Degree: B.S., Eng. Mechanics, 1986
Specialty: Engineering Mechanics
Assigned: Astronautics Laboratory

Michael J. Koharchik
Dept. of Engineering Mechanics
The Pennsylvania State Univ.
State College, PA 16802
(814) 237-6527

Degree: B.S., Aerospace Eng., 1987
Specialty: Aerospace Structures
Assigned: Astronautics Laboratory

Keith A. Krapels
Dept. of Electrical Eng.
Memphis State University
Memphis, TN 38152
(901) 454-3312

Degree: M.S., Electrical Eng., 1986
Specialty: Electrical Engineering
Assigned: Arnold Engineering Development Center

Richard J. Kunze
Dept. of Psychology
University of Missouri-Columbia
210 McAlester Hall
UMC Campus
Columbia, MO 65211
(314) 882-4351

Degree: B.A., Psychology, 1986
Specialty: Experimental Psychology
Assigned: Harry G. Armstrong Aerospace Medical Research Laboratory

Thomas E. Lane
Dept. of Chemistry
Ball State University
Muncie, IN 47306
(317) 285-8078

Degree: B.S., Biology, 1988
Specialty: Immunology & Microbiology
Assigned: School of Aerospace Medicine

Bobby L. Larry
Dental School
Meharry Medical College
1005 D.B. Todd Blvd.
Nashville, TN 37208
(615) 322-9819

Degree: B.S., Biology, 1978
Specialty: Dentistry
Assigned: Wilford Hall Medical Center

Aleshia C. Lewis
Dept. of Biology
Meharry Medical College
1005 D.B. Todd Blvd.
Nashville, TN 37208
(615) 327-6111

Degree: B.S., Microbiology, 1986
Specialty: Microbiology
Assigned: Wilford Hall Medical Center

Yuhong Y. Li
Dept. of Computer Science
University of Nebraska
Lincoln, NE 68588
(402) 472-7211

Degree: M.S., Chemical Eng., 1977
Specialty: Chemical Engineering
Assigned: Avionics Laboratory

Yolanda A. Malone
Dept. of Pharmacology
Meharry Medical College
1005 D.B. Todd Blvd.
Nashville, TN 37208
(615) 327-6111

Degree: M.S., Medicine, 1988
Specialty: Medicine
Assigned: School of Aerospace Medicine

Randal L. Mandock
School of Geophysical Science
Georgia Inst. of Technology
Atlanta, GA 30332
(404) 894-3890

Degree: M.S., Atmospheric Sci., 1986
Specialty: Atmospheric Science
Assigned: Avionics Laboratory

David L. Mayfield
Dept. of Ind./Org. Psychology
University of Georgia
Athens, GA 30602
(404) 542-3053

Degree: B.S., Psychology, 1987
Specialty: Industrial/Organ. Psychology
Assigned: Human Resources Laboratory: Manpower and Personnel Div.

John E. McCord Dept. of Chemistry Murray State University Murray, KY 42071 (502) 762-4490	Degree: Specialty: Assigned:	B.S., Chemistry, 1987 Physical Chemistry Weapons Laboratory
David B. McKenzie Dept. of Civil Engineering Michigan Technological Univ. Houghton, MI 49931 (906) 482-4882	Degree: Specialty: Assigned:	B.S., Civil Eng., 1988 Environmental Engineering Engineering & Services Center
Salvatore P. Miceli Dept. of Aerospace Eng. Sciences Univ. of Colorado - Boulder Campus Box 429 Boulder, CO 80309-0429 (303) 492-6417	Degree: Specialty: Assigned:	B.S., Aerospace Eng., 1988 Unsteady Aerodynamics Frank. J. Seiler Research Lab.
Hisook L. Min Division of Basic Studies Jarvis Christian College Hawkins, TX 75765 (214) 769-2174	Degree: Specialty: Assigned:	M.S., Computer Science, 1987 Applied Physics Armament Laboratory
Deborah J. Mitchell Dept. of Chemistry Prairie View A&M University Drawer C Prairie View, TX 77446 (409) 857-3910	Degree: Specialty: Assigned:	B.S., Chemistry, 1986 Biochemistry Harry G. Armstrong Aerospace Medical Research Laboratory
William A. Moran Dept. of Chemistry Calif. State Univ. -Northridge 18111 Nordhoff Street Northridge, CA 91330 (818) 885-3381	Degree: Specialty: Assigned:	B.S., Chemistry, 1986 Chemistry Astronautics Laboratory
William D. Morse Dept. of Electrical Eng. Ohio State University 205 Dresser Laboratory 2015 Neil Avenue Columbus, OH 43210-1272 (614) 292-2572	Degree: Specialty: Assigned:	B.S., Electrical Eng., 1987 Electrical Engineering Flight Dynamics Laboratory

Lisa F. Weinstein
Dept. of Psychology
Univ. of Illinois
at Urbana-Champaign
Aviation Research Lab., Q5
#1 Airport Road
Savoy, IL 61874
(217) 244-8728

Degree: M.A., Experimental Psy., 1987
Specialty: Engineering Psychology
Assigned: Harry G. Armstrong Aerospace Medical Research Laboratory

Michael A. Zmuda
Dept. of Computer Science
Wright State University
Dayton, OH 45324
(513) 873-2491

Degree: B.S., Compt. Sci. & Math, 1987
Specialty: Pattern Recognition
Assigned: Avionics Laboratory

Robert G. Petroit
Dept. of Elect. & Compt. Eng.
Illinois Institute of Tech.
3300 So. Federal
Chicago, IL 60616
(312) 567-3400

Degree: B.S., Electrical Eng., 1987
Specialty: Communications Systems
Assigned: Rome Air Development Center

Peter E. Pidcoe
Dept. of Bioengineering
University of Illinois
Box 4348
Chicago, IL 60680
(312) 996-2331

Degree: B.S., Environ. Mgmt., 1988
Specialty: Signal Processing
Assigned: Human Resources Laboratory: Operations Training Div.

Steven J. Pierce
Dept. of Civil Engineering
Geotechnical Program
Colorado State University
Fort Collins, CO 80523
(303) 491-5048

Degree: B.S., Geology, 1985
Specialty: Geotechnical Engineering
Assigned: Engineering & Services Center

Julia Rennenkampff
Dept. of Mathematics
New York University
251 Mercer Street
New York, NY 10012
(212) 998-3140

Degree: M.S., Mathematics, 1987
Specialty: Waves in Random Media
Assigned: School of Aerospace Medicine

Robert J. Riley
Dept. of Mech. & Aero. Eng.
Cornell University
105 Upson Hall
Ithaca, NY 14853
(607) 255-3623

Degree: B.S., Mechanical Eng., 1987
Specialty: Combustion, Fluid Dynamics
Assigned: Aero Propulsion Laboratory

Mary C. Ritter
Dept. of Biology
Trinity University
715 Stadium Dr.
Box 937
San Antonio, TX 78284
(512) 737-4782

Degree: B.A., Biology, 1988
Specialty: Biological Oceanography
Assigned: School of Aerospace Medicine

Jacqueline Roberts
Dept. of Chemistry
Wright State University
229 Oelman Hall
Dayton, OH 45435
(513) 873-2855

Degree: B.S., Chemistry, 1986
Specialty: Toxicology
Assigned: Harry G. Armstrong Aerospace Medical Research Laboratory

James D. Roberts
Dept. of Engineering
Univ. of Texas-San Antonio
San Antonio, TX 78285
(512) 691-4490

Degree: B.S., Electrical Eng., 1988
Specialty: Order Statistic Filters
Assigned: School of Aerospace Medicine

Matthew S. Rubin
Dept. of Elect. & Compt. Eng.
Ohio University
West Green Drive
Stocker Center 329
Athens, OH 45701-2979
(614) 593-1568

Degree: B.S., Electrical Eng., 1987
Specialty: Microwave Network Theory
Assigned: Rome Air Development Center

John Y. Salinas
Dept. of Medicine
Meharry Medical College
1005 D.B. Todd Blvd.
Nashville, TN 37208
(615) 327-4537

Degree: M.S., Biochemistry, 1984
Specialty: Medicine
Assigned: Wilford Hall Medical Center

Eric O. Schmidt
Dept. of Geophysical Science
Georgia Inst. of Technology
Atlanta, GA 30332
(404) 894-3897

Degree: M.S., Physics, 1983
Specialty: Atmospheric Science
Assigned: Avionics Laboratory

Gregory A. Schoeppner
Dept. of Civil Engineering
Ohio State University
470 Hitchcock
2070 Neil Avenue
Columbus, OH 43210
(614) 292-7304

Degree: M.S., Civil Eng., 1987
Specialty: Civil Engineering
Assigned: Flight Dynamics Laboratory

Douglas J. Sego
Dept. of Organ./Behavior Mgmt.
Michigan State University
232 Eppley East
Lansing, MI 48824
(517) 353-5414

Degree: B.A., Business Info., 1987
Specialty: Organization/Behavior
Assigned: Human Resources Laboratory: Training Systems

Anne L. Siegman
Dept. of Mathematics
University of Miami
1107N 1239 Dickinson Dr.
Coral Gables, FL 33146
(305) 284-2925

Degree: B.A., Mathematics, 1986
Specialty: Mathematics
Assigned: Armament Laboratory

Jeff P. Simmons
Dept. of Engineering
Carnegie Mellon University
Pittsburgh, PA 15213
(412) 268-2684

Degree: M.S., Engineering, 1985
Specialty: Materials Science
Assigned: Materials Laboratory

Kimberly F. Smith
Dept. of Medicine
Meharry Medical College
P O Box 935
1005 D.B. Todd Blvd.
Nashville, TN 37208
(615) 327-6308

Degree: B.S., Microbiology, 1986
Specialty: Epidemiology
Assigned: Wilford Hall Medical Center

Brian K. Spielbusch
Dept. of Electrical Eng.
Univ. of Missouri-Columbia
Truman Campus
600 W. Mechanic
Independence, MO 64050
(816) 276-1250

Degree: B.S., Electrical Eng., 1985
Specialty: Electro Optics
Assigned: Weapons Laboratory

Daryl W. Sprehn
Dept. of Elect. Eng. Tech.
Oregon Inst. of Technology
3201 Campus Drive
Klamath Falls, OR 97601-8801
(503) 882-6890

Degree: B.S., Electrical Eng., 1988
Specialty: Electromagnetic Propagation
Assigned: Rome Air Development Center

Christopher Sullivan
Dept. of Psychology
Colorado State University
Fort Collins, CO 80523
(303) 491-7184

Degree: M.S., Psychology, 1986
Specialty: Experimental Psychology
Assigned: Harry G. Armstrong Aerospace Medical Research Laboratory

David A. Swick
Dept. of Mechanical Eng.
Ohio University
723 Carriage Hill
Athens, OH 45701
(614) 594-4818

Degree: B.S., Mechanical Eng., 1987
Specialty: Mechanical Design
Assigned: Materials Laboratory

Richard A. Swift
Dept. of Aero. & Mech. Eng.
University of Notre Dame
Notre Dame, IN 46556
(219) 239-5430

Degree: B.S., Aeronautical Eng., 1987
Specialty: Structural Mechanics
Assigned: Flight Dynamics Laboratory

Paul R. Tanner Dept. of Physiology Meharry Medical College Nashville, TN 37212 (615) 269-0873	Degree: Specialty: Assigned:	B.A., Psychology, 1986 Sensory Neurophysiology Wilford Hall Medical Center
David F. Thompson School of Mechanical Eng. Purdue University West Lafayette, IN 47906 (317) 494-4903	Degree: Specialty: Assigned:	M.S., Engineering, 1985 Computer Information Flight Dynamics Laboratory
Ronald C. Tomlinson Dept. of Chemistry Wright State University Dayton, OH 45435 (513) 873-2855	Degree: Specialty: Assigned:	B.S., Chemistry, 1987 Polymer Synthesis Materials Laboratory
Robert W. Tramel Dept. of Mathematics Univ. of Tennessee Space Inst. Tullahoma, TN 37388-8897 (615) 455-0631	Degree: Specialty: Assigned:	B.S., Physics, 1986 Computational Fluid Mechanics Arnold Engineering Development Center
Tien N. Tran Dept. of Elect. & Comp. Eng. University of Cincinnati Cincinnati, OH 45221 (513) 475-4247	Degree: Specialty: Assigned:	M.S., Electrical Eng., 1988 Image Coding Avionics Laboratory
John P. VanTassel Dept. of Computer Science Wright State University Dayton, OH 45435 (513) 873-2491	Degree: Specialty: Assigned:	B.A., Computer Studies, 1987 Formal Specifications Avionics Laboratory
Deborah L. Vezie Dept. of Chemical, Bio., and Materials Engineering Arizona State University COB B210 Tempe, AZ 85287 (602) 965-3313	Degree: Specialty: Assigned:	B.S., Biomedical Eng., 1987 Materials Science & Eng. Materials Laboratory
Oden L. Warren Dept. of Chemistry Iowa State University Ames, IA 50011 (515) 294-6342	Degree: Specialty: Assigned:	B.S., Chemistry, 1988 Physical Chemistry Materials Laboratory

Lisa F. Weinstein
Dept. of Psychology
Univ. of Illinois
at Urbana-Champaign
Aviation Research Lab., Q5
#1 Airport Road
Savoy, IL 61874
(217) 244-8728

<u>Degree:</u> M.A., Experimental Psy., 1987
<u>Specialty:</u> Engineering Psychology
<u>Assigned:</u> Harry G. Armstrong Aerospace Medical Research Laboratory

Michael A. Zmuda
Dept. of Computer Science
Wright State University
Dayton, OH 45324
(513) 873-2491

<u>Degree:</u> B.S., Compt. Sci. & Math, 1987
<u>Specialty:</u> Pattern Recognition
<u>Assigned:</u> Avionics Laboratory

PARTICIPANT LABORATORY ASSIGNMENT

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RESEARCH REPORTS

1988 GRADUATE STUDENT RESEARCH PROGRAM

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7	MULTIGRAPH Kernel for Transputer Based Systems	Ben Abbott
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Final Report

Improvements in the Control of Robotic Arm Simulations Using the Articulated Total Body (ATB) Model

Prepared by: Daniel William Barineau

Academic Rank: Masters Student

Department and University: Engineering Mechanics, Virginia Tech

Research Location: AAMRL/BBM, Wright-Patterson AFB, Dayton OH 45433-6573

Date: 1 SEPT 1988

Contract No: F49620-88-C-0053

Improvements in the Control of Robotic Arm Simulations Using the Articulated Total Body (ATB) Model

by

Daniel William Barineau

Abstract

Modifications were made to the control of torque generation in the Air Force's ATB model. Improved controller feedback was created by adding an integral control term to the existing control equation. Motion studies were performed using a Merlin robot simulation to determine control variables for single joint rotations. A joint rotation range from -180 degrees to +180 degrees was examined.

Values determined from the above study for the six joints in question were added to the input description as a tabular set of data, which the computer could access depending on the joint target angles set by the operator. Simultaneous multi-joint rotations were also studied using the same controlling values as were used in single joint rotations. It was found that these numbers produced accurate results for all joint rotations, as long as either the shoulder or elbow joints were held at their initial angular positions. The error produced when the target angles for both the shoulder and elbow were non-zero was less than two degrees of arc. This problem can be overcome by modifying the control equation with some function of the robot's inertial properties.

Acknowledgements

I would like to offer my most sincere thanks to the Air Force Office of Scientific Research and the Air Force Systems Command for giving me such an invaluable research opportunity this past summer. I must also thank Universal Energy Systems for their very competent management of the program. Many students, myself included, would lack the ability to perform necessary research were it not for the farsighted nature of this program.

My summer fellow experience was considerably enhanced by the highly knowledgeable people with whom I worked. To Captain Taylor and Lieutenants Urema and Detereman I give a hearty thanks for the computer oriented assistance I could not have done without. To Sherri, Annette, and Therese, my SRL office-mates, I would hope that my constant questions were not too annoying. I would also like to thank Terri McFarland and Ric Rasmussen for improving my overall enjoyment of the summer at Wright-Patterson.

The concern, guidance, and stimulating conversation of Dr. Ints Kaleps throughout my research period was greatly appreciated. Finally, to the person without whom I could not have accomplished half as much as I did, I would like to commend Louise Obergefell for never being too busy to take a few minutes of her time and listen to my questions.

I. INTRODUCTION:

The Articulated Total Body (ATB) Model is a computer simulation program used to analyze human body responses in dynamic environments. It was originally developed by Calspan Corporation in the 1970's for the National Highway Traffic Safety Administration (NHTSA) for simulating three dimensional dynamics in automobile crashes. That initial program, called CVS for Crash Vehicle Simulator, underwent extensive changes by the Air Force to arrive at the current ATB model (Ref. 1). Included in these modifications is the ability to generate torques at the joints of a described body model.

The Modelling and Analysis Branch of the Harry G. Armstrong Aerospace Medical Research Lab at Wright-Patterson AFB has recently been using the ATB model to simulate robotic motion and its control. The active generation of torque at the model's joints enables the simulation to interact within its environment by means of internal rather than external forces. The resulting kinematic and dynamic data can provide valuable information in perfecting robotic usage in the aerospace field and the development of active human body simulations.

My research interests in the biomedical field have included the analysis and modelling of human muscle and its control. During the 1987-88 school year I installed the ATB program on the AT&T 3B15 computer system in the Engineering Mechanics department of Virginia Tech. By doing this I became quite familiar with many of the 130+ subprograms and over 16,000 lines of Fortran code. The object of this work was a thesis oriented goal of simulating robotic motion in a coordinated fashion within specified error limits. The addition of similar actuator elements into a mannikin description would create a more realistic model of the human body that not only passively responded but could actively resist external environmental forces. The robot simulations are an initial step in this direction.

II. OBJECTIVES OF THE RESEARCH EFFORT:

In their current forms, crash and ejection simulations run on the ATB use mannikin descriptions that can only passively react to their environment. Therefore, an obvious improvement would be the ability to generate torques at the joints of the model. This would create an approximation of human muscle contraction across the joints in an attempt either to move or remain stationary during a simulation. Another application in which torque generation at joints would be necessary, is the use of the ATB program to analyze robotic motion. Aerospace operations including satellite retrieval, waste disposal, and the movement of heavy objects provide opportunities for the use of remotely controlled robots. With this in mind, the ATB could provide kinematic and dynamic data by simulating the desired task. An initial step towards attaining these goals is the generation and accurate control of joint torques in a robotic arm simulation.

In its current form, the ATB model is based on the rigid body dynamics of coupled systems with Lagrange type constraint equations. This allows a system to be described as a set of rigid segments, coupled at joints which allow the application of torques as functions of joint orientations and rate of change of orientations (Ref. 2). The current simulation is that of an American Cimflex MR6500 Merlin robot with six articulations. The physical description that is input into the program contains the values of mass, moment of inertia, and segment geometry.

The control of angular displacement created by the joint torques is contained in a subroutine that can be modified to represent whatever controlling equation the user desires. At the beginning of the summer the controlling equation being used was :

$$T = f_2(\Theta-\Theta_0) - f_3(\dot{\Theta}) \qquad (1)$$

Where: $\Theta_0 = f_1(t)$,
T is the joint torque applied by the actuator,
Θ is the joint angle,
Θ_0 is the joint target angle,
$\dot{\Theta}$ is the joint angular velocity,
t is time, and
f_i's are input functions.

In this case, $f_1(t)$ is a constant function equal to Θ_0, while $f_2(\Theta-\Theta_0)$ and $f_3(\dot{\Theta})$ are equal to $K_1*(\Theta-\Theta_0)$ and $K_2*\dot{\Theta}$ respectfully. This represents both proportional and derivative control of the joint torque feedback, but does not contain an integral control term. Oftentimes, the lack of this term will create a steady state angular error of the shoulder and elbow joints due to forces such as gravity.

My assignment as a participant in the 1988 Graduate Summer Research Program was to add an integral control term to the existing control equation. The modified equation appears as:

$$T = f_2(\Theta-\Theta_0) - f_3(\dot{\Theta}) - f_4\left(\int_0^t(\Theta-\Theta_0)dt\right) \qquad (2)$$

Where all terms are the same as before, and $f_4\left(\int_0^t(\Theta-\Theta_0)dt\right)$ is equal to $K_3*\int_0^t(\Theta-\Theta_0)dt$. In addition to this, I was to examine the values of the K_i terms for functional relationships over an angular rotation range from -180 to +180 degrees. This information would allow me to modify the input description to the ATB program so that in changing rotational angles in subsequent simulations, an operator would only have to specify the desired target angles. Finally, I was to attempt the simultaneous movement of all joints in the model, to determine if inertial properties needed to be taken into account in simulating coordinated motion.

III.

a. In adding integral control to the ATB program, modifications were made to subroutine USER, in which a user defined equation controls the joint feedback information. As previously mentioned, the modification appeared as constant term times the integral of the error function (Ref. 3). This was approximated by the summation of $(\Theta - \Theta_o)$ at the given time intervals multiplied by the differential time step dt. Operational verification was limited to the observation of joint output to see if the new equation could remove the steady state error, while at the same time creating a characteristic integral term instability.

b. The results of these program changes were the removal of the steady state angular error in both the shoulder and elbow joints for single rotations. A small amount of oscillatory instability, less than .010 degrees, was also introduced into the system as expected.

IV.

a. Functional dependencies for K_1, K_2, and K_3 were initially determined for single joint rotations of the shoulder and elbow. Therefore, the isolated joint's motion and control parameters were unaffected by inertial effects from other segments. The shoulder and elbow were examined first because of their relatively large moments of inertia and the resulting steady state displacements due to gravity. After these two, the waist, forearm, wrist and palm joints were examined for the same relationships. In all cases, rotational ranges from -180 to 180 degrees were examined, and for this initial work, contact between segments was not taken into consideration. For all joints, a similar initial robot orientation was used as the model for tuning of the control parameters. The control variables of K_1, K_2, and K_3 were varied until an angular displacement curve was achieved that had the following qualities:

(1) less than one-half percent overshoot,

(2) ultimate angular displacement errors of less than 0.03 degrees, and

(3) a rise time to the target angle between .25 and .35 seconds.

As initial approximations, values for the K_i's were obtained from quarter decay and critically damped control equations (Ref. 5). These were then modified to fulfill the conditions stated above.

b. Initial results produced by using the control parameter estimating equations were unsatisfactory in attaining the desired target angle. Initial positioning of the model, and physical dimensions needed to be taken into account before proper responses were obtained. K_1 terms for all joints other than the shoulder varied with relative angle displacements as functions of $\cos(\Theta_0)$. The K_1 term for the shoulder varied as a function of Θ_0^{-1}. K_2 values for all joints studied were also periodic as some function of $\cos(\Theta_0)$. Relative values for both K_1 and K_2 were proportional to the inertia of the segments being moved by the joint, i.e. K_2shoulder > K_2elbow > K_2forearm > ... etc. For K_3 values, the shoulder and elbow were once again functions of $\cos(\Theta_0)$ while the other four joints had constant values of zero due to negligible gravitational displacements. For graphical depictions of these relationships, see Figures 1-3.

V.

a. It was decided that the values for K_1, K_2, and K_3 over the range of relative angular displacements from -180 to 180 degrees, would be input to the ATB in tabular form. Due to the way in which the program retrieves data from the input deck, this required changes in the equations defined in subroutine USER. Verification of results would be accomplished by comparing results from previous simulations with those using the tabular data set.

b. The results obtained from using the tabular set of K_1 descriptions was much more flexible once it was operating correctly. As noted earlier, after modification, the operator need only specify the desired relative angular displacement and the computer looks up the necessary values for K_1, K_2, and K_3.

VI.

a. For producing simultaneous six joint motion, the previously input values for single joint motions were used. This was initially completed for the four joints, that had not experienced any significant displacement due to gravity, namely the waist, forearm, wrist, and palm. Subsequent trials included one of the two remaining joints, either the shoulder or the elbow. Final simulations included all six joints rotating simultaneously to six different relative angles.

b. The initial tests with four joints, and the subsequent ones including five joints were well coordinated in the same manner as the single joint rotations. In adding the sixth joint, either the shoulder or the elbow, the K values were no longer sufficient to maintain the previously specified accuracy (less than 0.03 degrees error). This inaccuracy was approximately one percent of the relative angular displacement for both the shoulder and elbow joints. These effects are most likely created by the configuration and inertial qualities of the upper two arm segments.

VII. RECOMMENDATIONS

a. The way in which the present system controls the simulated robotic arm should be sufficient for most modelling cases. The current set-up will allow the accurate control of the angular displacement profile for one to five joint motion. Kinematic and dynamic data for the allowed five joint simulations should prove useful in robotic motion modelling and analysis.

b. The first suggestion for follow-on research is to incorporate some form of adaptive control into the present controlling equation in order to remove the error in simultaneous shoulder-elbow rotations. Another area to be looked into is the validation of the simulation results against the actual motion studies of a similar robot. This should not be overly difficult in that there is already a robot of this kind in the department. The expansion of the number of modelled joints to include all those found in the human body is another possible research area. Finally, a very useful way to use this torque generation and control scheme is to incorporate it into mannikin models. This would allow the active movement of the simulated humans in either ejection or car crash simulations.

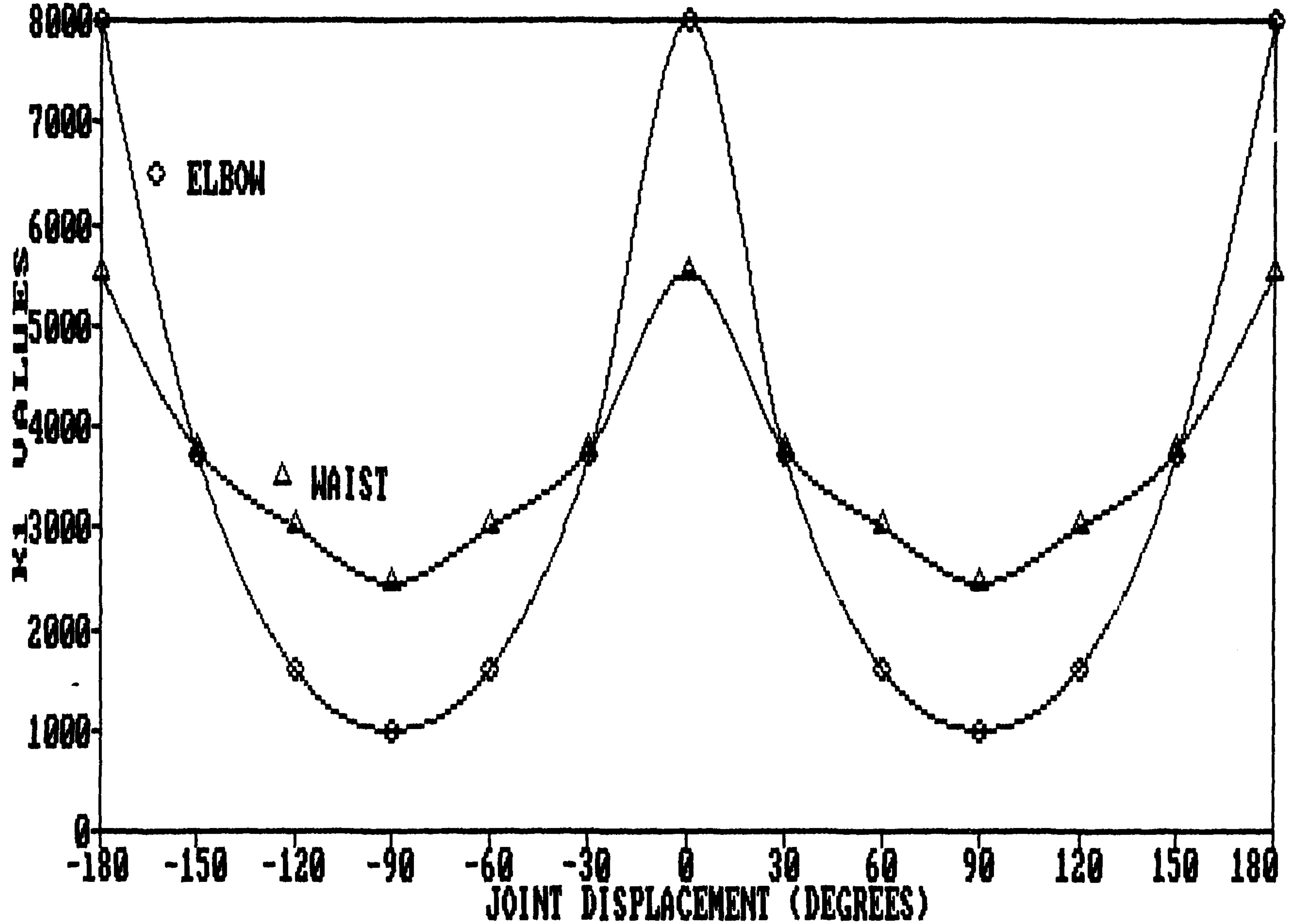

Figure 1. Periodic relationships between the controlling equation variable K_1 and the relative angular displacement θ_o.

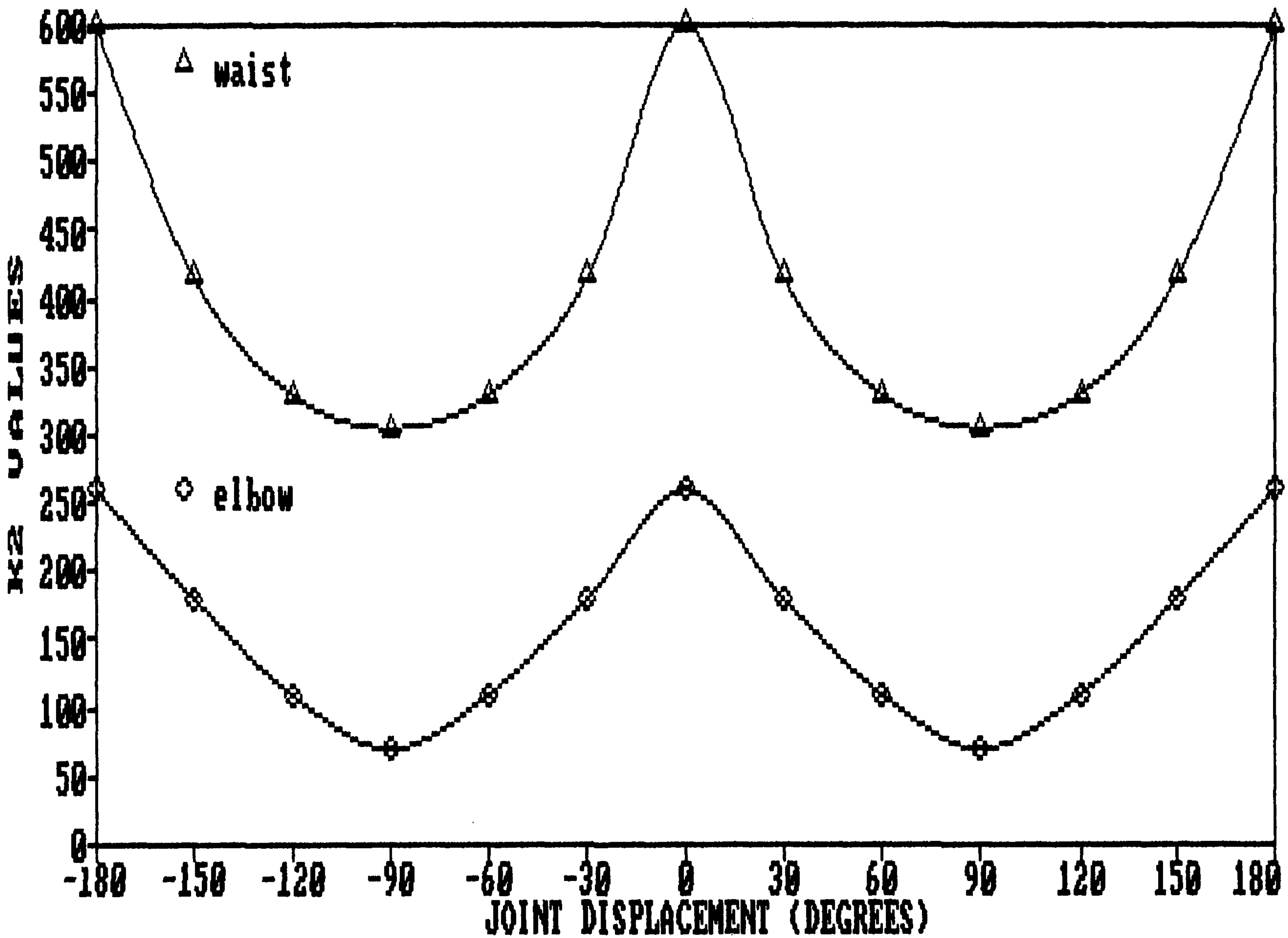

Figure 2. Periodic relationships between the controlling equation variable K_2 and the relative angular displacement θ_o.

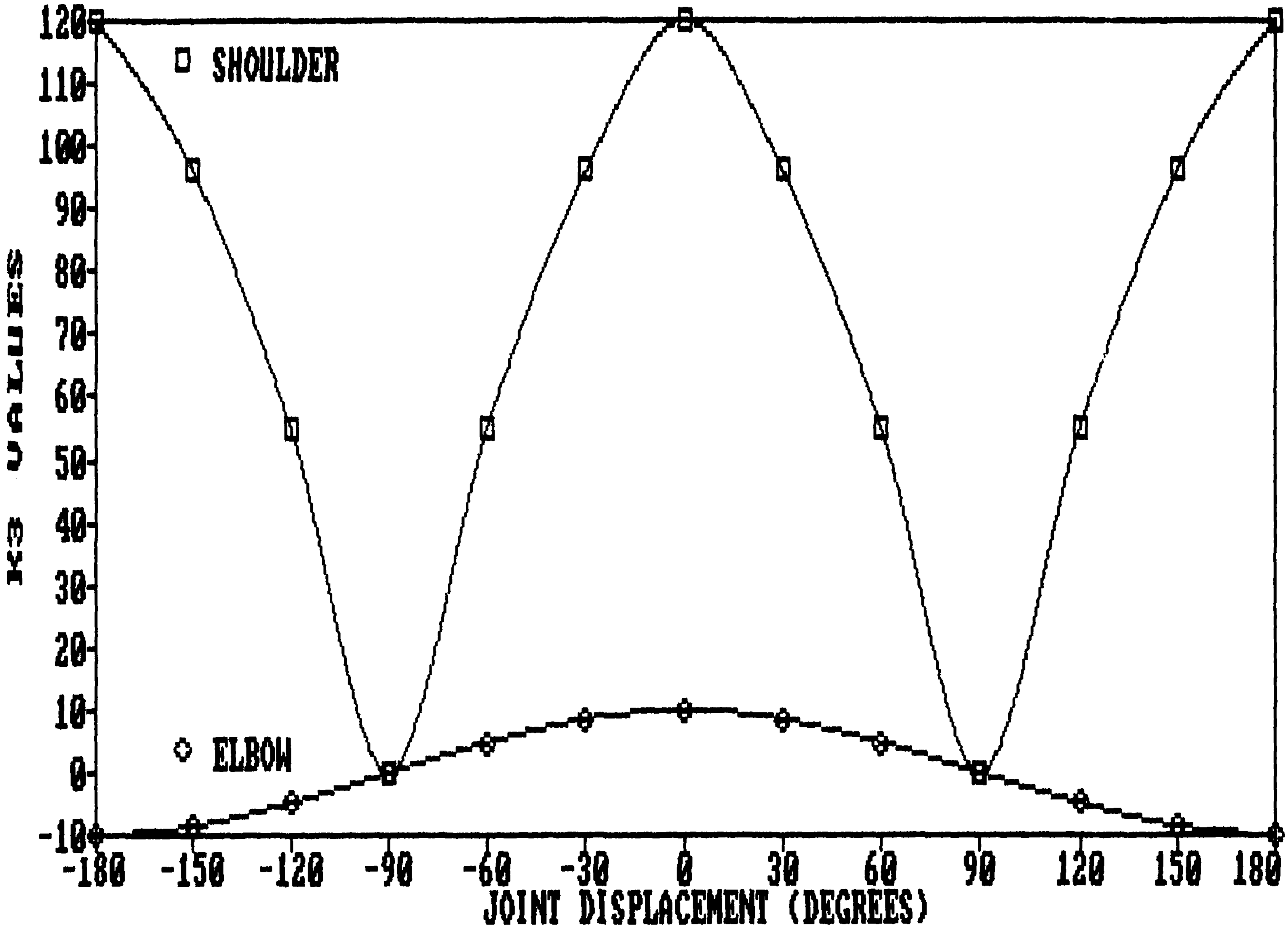

Figure 3. Periodic relationships between the controlling equation variable K_3 and the relative angular displacement θ_o.

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<u>FINAL REPORT</u>

Auditory Modeling

Prepared By:	C. David Covington, Ph.D. and Michael K. Ellis, Graduate Student
Academic Rank:	Assistant Professor
Department and University:	Department of Electrical Engineering University of Alabama
Research Location:	AAMRL/BBA Wright-Patterson AFB, OH 45433
USAF Researcher:	Timothy R. Anderson

Date:	15 August 1988
Contract No.	F49620-88-C-0053

SAME REPORT AS
DR. DAVID COVINGTON
HARRY G. ARMSTRONG AEROSPACE
MEDICAL RESEARCH LABORATORY # 110

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FINAL REPORT

Performance in a Visual Monitoring Task with Serial and Simultaneous Display Formats

Prepared By: David G. Payne, Ph.D. and Virginia Gunther

Academic Rank: Assistant Professor and Graduate Student

Department and University: State University of New York
University Center at Binghamton

Research Location: AAMRL/HEX
Wright-Patterson AFB, OH 45433

USAF Researcher: Kenneth R. Boff, Ph.D

Date: 19 August 1988

Contract No. F49620-88-C-0053

SAME REPORT AS
DR. DAVID PAYNE
HARRY G. ARMSTRONG AEROSPACE
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FINAL REPORT

Prepared by:	Stephen R. Jenei, B.Sc.
Academic Rank:	Graduate Student
Department and University	Department of Biology University of Dayton
Research Location:	AAMRL/THT Wright-Patterson AFB Dayton, OH
USAF Researcher:	David R. Mattie, Ph.D.
Date:	15 August 1988

EVALUATION OF THE TOXIC EFECTS OF A 90-DAY CONTINUOUS EXPOSURE OF RATS AND TO SHALE DERIVED JP-4 JET FUEL

by

Stephen R. Jenei

ABSTRACT

Many volitile hydrocarbons are widely used as additives in jet and automobile fuels. JP-4 is a complex mixture of aliphatic and aromatic hydrocarbons with various additives and is used as a fuel additive. This investigation looked at the effects of JP-4 on the kidneys and livers of Fischer F344, male rats. Sections of kidneys containing the glomeruli were processed and sectioned for transmission electron microscopy (TEM). Also, as part of the summer program, tissues collected from ALCAP implantable ceramic drug delivery capsules were obtained from animals implanted for three months and examined for inflammatory and immune response. A new procedure utilizing scanning electron microscopy for examination of spermatozoa was also developed for the Air Force.

ACKNOWLEDGENENTS

I would like to thank the Air Force Systems Command, the Air Force Office of Scientific Research and Universal Energy Systems, Inc. for the sponsorship and direction of this project. Although many should be thanked for technical assistance and support, special mention should be made to Dr. David R. Mattie who has directed my work under this summer project and is a member of my graduate committee, Dr. Hixon, Dr. Grabau, Sgt. Maslanka, Sgt. Chase, Jay Kennard, Doug Helton, Sgt. Russell and all the mebers of the toxic Hazards Branch.

I. INTRODUCTION

The Air Force has evaluated the compatibility of shale derived turbine fuels with aircraft engines such as the shale derived JP-4 jet fuel. Acute and subcrhonic toxicity evaluations were preformed to provide information dealing with the health and safety concerns. Previous studies of hydrocarbon fuels have shown morphological changes in the kidneys of male rats (MacEwen and Vernot, 1978; 1981).

II. OBJECTIVES OF THE RESEARCH EFFORT

The objectives of this investigation was to determine the toxic effects of a 90 day continuous exposure of rats to shale derived JP-4 vapours for comparison with a previous 90 day petroleum JP-4 study by MacEwen and Vernot (1980).

Because of mechanical difficulties with the transmission electron microscope (TEM) and to expand training in electron microscopy, part of the summer research time was spent on evaluating the tissue which encapsulates an implantable ceramic drug delivery device which the Air Force uses to deliver volitile materials. A second project was to develope a satisfactory procedure for evaluating spermatozoa and implants by scanning electron microscopy (SEM) for the Air Force.

MATERIALS AND METHODS

In this investigation, the shale derived JP-4 samples (test fuels) to be used was supplied by the USAF. The JP-4 shale fuel was a complex mixture of aliphatic and aromatic hydrocarbon compounds including various additives.

The rats were exposed to 500 mg/m^3 and 1000 mg/m^3 shale JP-4 vapour on a continuous basis for 90-days. Three Thomas Dome Isolation Chambers were used with the control group being eposed only to the natural air. Each chamber housed 95 male and 75 female Fischer 344 rats. After the 90 day continuous exposure, 15 rats of each sex from each group were sacrificed. The remainder of the animals were sacrificed at 2 weeks, 2 months, and 9 months post-exposure. The final sacrifice was then 24 months post-exposure. It is the 2 month, post-exposure group consisting of 10 male rats which were investigated under the Graduate Student Summer Support Program.

The kidneys and livers taken from the JP-4 exposed animals 2 months post-exposure were fixed in 1.5% Karnovski's fixative for 36 h and then minced into 1 mm^3 cubes for processing for transmission electron microscopy (TEM). The tissues were embedded in Epon synthetic resin and then sectioned (1 micron) using an ultramicrotome. Upon examination of the 1 micron or thick sections, areas of interest were marked and the tissue samples trimmed down for 600 angstrom or thin sectioning. The thin sections were then stained by uranyl acetate and lead citrate heavy metal stains and placed on a copper grid for placing into the transmission electron microscope.

Alumino-calcium-phosphorus oxide (ALCAP) ceramic capsules which had been implanted intraperitoneally in rats for 3 months were investigated. These capsules, which contained a protein mixture, had elicited a host reponse and were completely encapsulated with tissue. The tissue surrounding the capsules was examined for an inflammatory and/or immune response by the host.

Tissues collected from the ALCAP capsules were first fixed in 10% neutral buffered formalin for two weeks. The tissues were then sliced in 1 mm^3 sections and processed for paraffin embedding. After embedding in paraffin blocks, the tissues were cut in 5 micron slices on a rotary microtome and the sections placed on glass histology slides. The histology slides were stained by standard hematoxalin and eosin stains and coverslipped using a synthetic resin mounting agent. The slides prepared were examined for the presence of inflammatory cells and immune cells along with collagenous scar tissue and accompanying vascularization.

The ceramic implants and spermatozoa collected from 3 month post-implant rats were prepared and examined by SEM. The specimens were fixed in 10% neutral buffered formalin for two weeks and the prepared by freeze drying to preserve the natural state of the tissues. The samples were placed onto specimen stubs and immediately place into liquid N_2. The specimens were then transferred to an Emscope FD500 freeze drier and dried overnight. The specimens were then coated with gold using a sputter coater and examined using Amray SEM.

RESULTS TO DATE

The work involved in the study of the effects of the shale JP-4 on the kidneys of male rats is in progress at the time of the writing of this final report. The data obtained to date has shown that the proximal tubules of rats exposed to 500 mg/mm^3 of the shale JP-4 had more hyaline droplets than the proximal tubules of the control animals. Furthermore, the proximal tubules of animals exposed to 1000 mg/mm^3 of the shale JP-4 had even greater numbers of hyaline droplets present and dialated spaces where the renal tubular cells interdigitate. This study will continue in the Ultrastructural Laboratories of AAMRL/THT under the direction of Dr. David Mattie.

The study involved in the evaluation of fibrous tissue encapsulating the ALCAP ceramic capsule implants and spermatozoa has been completed. The study so far has shown that collagenous fibrous scar tissue with accompanying vascularization encapsulates the implant. Within the tissue, there was found a zone of hypercellularity within the tissue sections. Along the inside surface of the tissue was found numerous multinuclear-giant cells (MNGC), macrophages, undifferentiated and differentiated lymphocytes, and granulocytes. The presence of these cells indicate both an immune response and an inflammatory response has taken place against the implant and/or the protein within the ALCAP capsule.

Since there was an abundance of differentiated plasma cells present within the tissue, it could be assumed that antibodies are being produced against the proteins held within the capsule. However, without the aid of special immunostaining techniques, positive identification of the materials for which the antibodies are directed against. The presence of the MNGC's, which result from the fusion of macrophages attempting to digest indigestable material, are an indicator of a chronic immune and/or inflammatory response.

The studies involving the evaluation of spermatozoa by scanning electron microscopy showed that the spermatozoa could be preserved intact using the methods for freeze-drying developed during the summer. Spermatozoa were able to be viewed clearly for precense of malformations, decapitation, split tails, or other morphological changes in the structure of the spermatozoa.

IV. RECOMMENDATIONS

The results of the shale JP-4 study are not complete and will require completion of the investigation before the complete conclusions and recommendations can be made.

The ALCAP ceramic drug delivery device would probably not be a viable delivery system for proteins unless an immune tolerance to the protein could be developed by the host. The delivery device could, however, be used to elicit an immune response for the production of specific antibody.

The use of freeze-drying for examination of spermatozoa by SEM can be incorporated by the air force as an additional tool in the evaluation of the effects of chemicals on the male reproductive system. The incorporation of immunogold labelling techniques for detection of inhibin or similar glycoproteins using the SEM in conjunction with x-ray microanalysis and backscattering techniques could prove vital in determining the function and mechanism of action of the interaction of inhibin with spermatozoa.

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FINAL REPORT

CERAMIC COMPOSITE MATERIALS FOR ESTABLISHING INGROWTH OF BONE AND DEVELOPING BONE REMODELING MODELS

Prepared by:	Elizabeth J. Kavran
Academic Rank:	Graduate Student
Department and University:	Biology Department University of Dayton
Research Location:	AAMRL/BBD Wright Patterson Air Force Base Dayton, Ohio 45433
USAF Researcher:	Ed Eveland
Date:	30 September 1988
Contract No:	F49620-88-C-0053

CERAMIC COMPOSITE MATERIALS FOR ESTABLISHING

INGROWTH OF BONE AND DEVELOPING

BONE REMODELING MODELS

ABSTRACT

The primary objective of this study was to prepare a Tricalcium Phosphate (TCP) bone composite. X-ray diffraction analysis confirmed that the aqueous precipitation and sulfate calalysis procedure employed in this investigation yielded 100% pure TCP. This investigation also looked at the setting properties of different TCP and polyfunctional acid composites. Results obtained indicated that addition of malic acid to TCP yielded the best composite. This composite set and hardened within 15 minutes. The pH studies confirmed that a TCP composite with malic acid and calcium hydroxide on dissolution maintain an alkaline pH. The effect of implanting an ALCAP, malic acid and calcium hydroxide composite on the blood profile of rhesus monkeys was also observed in this investigation. The data obtained showed that the composite implants did not change the blood chemistry profile of the rhesus monkeys.

Acknowledgements

I wish to thank the Air Force Systems Command and the Air Force Office of Scientific Research for sponsorship of this research. I also wish to thank the department of Biodynamics and Bioengineering at W.P.A.F.B. for their laboratory use, as well as Ed Eveland Ph.D. who provided technical and histomorphometric advice. David Mattie Ph.D. and Sgt. Joseph Maslanka provided scanning electron micrograph technical advice and instruction. I wish also to thank Universal Energy Systems for their concern and help in all administrative and directional support of this program.

Acknowledgements are also due to the Department of Biology of the University of Dayton for providing the composites needed for this investigation. The help and guidance of Dr. P. K. Bajpai clearly added to every aspect of this research project.

I. INTRODUCTION:

Bone defects can result from removal of bone from donor sites, diseased bone, excision of tumors, pathologic resoprtion of bone, trauma, and non-unions of comminuted fractures. According to Sevitt (1981) the healing of experimental lesions parallels the healing of fractures. The implantation of a porous material can be used as a model to examine bone healing and bone growth.

Composites of calcium phosphate ceramics, such as tricalcium phosphate (TCP) and aluminum calcium phosphorus oxide (ALCAP) with biodegradable organics (such as polyfunctional carboxylic acids, amino acids and vitamins) and calcium hydroxide have been developed to repair these lesions (Sevitt, 1981). Bone resorption and formation occurs in pockets of normal bone, and according to Hori and Takahashi (1985) occurs by an "on-off" mechanism. Thus, these events can be quite difficult to follow in bone. Hence, it follows that bone substitutes can provide a matrix for investigating bone remodeling.

Ceramics have been made using various combinations of calcium aluminate and calcium mixed with phosphates (deGroot, 1980 & Hulbert et. al., 1974). Calcium phosphate ceramics attempt to reproduce the mineral phase of bone. Throughout implantation in many animals, TCP resorbs relatively rapidly and is biocompatible. Blocks of TCP have been implanted in tibia of rats (Bhaskar et. al., 1971) and in the femur, mandible, iliac crest, and inferior orbital rim in dogs (Camron et. al., 1977, Ferraro, 1979, Mors & Kaniski, 1975). In monkeys, chipped or powdered TCP has been used to repair defects in premolars and molars (Levin et. al., 1974), in apexification of teeth and in capping of pulp (Koening, 1974, Heller, 1975). TCP alone has not been reported to cause any adverse reactions in animal or human tissue (Metsger et. al., 1982).

This procedure has the ability to validate the on-gowing mechanisms involved in bone ingrowth and remodeling. It can also explain the data obtained from on-going and competed studies at BBD on the effects of prolonged vibration and mechanical stress on remodeling of bone. This procedure can also be incorporated into a comprehensive technique for analysis of bone, either as part of biodynamic studies or toxicity testing.

II. <u>OBJECTIVES OF THE RESEARCH EFFORT</u>:

The primary objective of this investigation was to prepare a Tricalcium phosphate ceramic (TCP) and polyfunctional acid (alpha-ketoglutaric acid or malic acid) composite for further study on bone ingrowth and remodeling with the use of a synthetic bone substitute implanted in bone. This fabrication process and characteristic study would then be used to further enhance a comprehensive analytical procedure to collect and process bone acquired in biodynamic and toxicity studies.

A procedure such as this has not been attempted to date. To date, normal bone samples have been used for histomorphometric analysis, X-ray analysis, and scanning electron microscopy. This combined approach would reduce sample size, as well as time and eliminate technical problems presently encountered at BBD and THT. It would also provide a more complete description of biomechanical responses which occur in hard tissues subjected to mechanical or toxicologic stresses. A second objective of this investigation was to obtain information on blood samples obtained during a bone remodeling and labelling process.

III MATERIALS AND METHODS

a. Preparation of TCP and ALCAP

The TCP used in this investigation was prepared by an aqueous precipitation and sulfate catalysis procedure (Bajpai et.al., 1981). The theoretical calcium/phosphorus ratio of 1.5 for TCP is involved in this reaction. Ammoniacal solutions of reagent grade calcium nitrate and ammonium hydrogen phosphate were reacted in glass vessels. The sludge was centrifuged and washed three times with one hour intervals. The final centrufigation was washed with a 3% ammonium sulfate solution. The percipitate was dried overnight at 72 C. The dried TCP percipitate was sintered in alumina crucibles and heated at a rate of approximately 9 C per minute to 1150 C and held for one hour. The dried powder was then sized to 33 um and less. X-ray diffraction analysis was used to identify the sintered percipitate derived and to assure the purity of the finished ground powder. Aluminum oxide, calcium oxide and phosphorous oxide powders were mixed in a ratio of 50:34:16 by means of a ball mill roller. The mixture was calcined at 1250 C for 12 hours in aluminate crucibles. The calcined material was ground and sieved to obtain particles of 33 um and less. These particles were then sintered for 36 hours at 1450 C. These were then set with 15% $Ca(OH)_2$ in a 2:1 ratio with malic acid in a syringe to yield a 1.3 cm 0.25 cm shape (see figure 1).

b. Preparation of composites

Composites for implantation were prepared by mixing 133.3 mg of either TCP or ALCAP and 30.0 mg calcium hydroxide with 66.7 mg of either ketoglutaric acid or malic acid. For studying the effects of vitamin-D, 50.0 mg of 1-25 dihydrocholecalciferol (DHCC) was added to both ketoglutaric and malic acid composites (see figure 2).

The six TCP composites were placed into 8 mm x 4 mm x 2 mm flexible rubber molds, and 60 ul deionized water was added to each composite. These composites were allowed to dry overnight. These forms were then placed into a 3.3 cm flat width Spectrapor standard dialysis-tube bags. The dialysis bags were tied with silk suture. These bags were placed into a 100 ml serum bottle containing 70 ml sterile saline 7.02 buffer. The serum bottles were placed into an oscillating water bath at 75 cycles per minute and maintained at 37 C. The pH of each bottle was measured at 1, 24 and 48 hours.

The setting property of each sample was measured in a similar manner. The TCP composites were placed into a deep welled glass slide, and 60 ul of deionized water was added to each composite. The hardness of each sample was measured every 5 minutes for one hour and then at 24 hours.

c. Blood analysis

The on-going investigation entitled "Use of a Porous Implant to Develop an Optimum Method to Examine Bone Growth/Repair for Biodynamic and Toxicologic Analysis" supplied this investigator the data on blood chemistry for statistical analysis. This investigation is similar in nature to the investigation proposed for TCP composites.

Four monkeys were implanted with two ALCAP ceramic implants. Surgeries were performed at AAMRL/VS. Blood was removed from the monkeys four weeks prior to labelling (physical). Then the animals were labelled involving an alternating series of fluorescein-tetracycline, xylenol orange and dicarbomethylaminomethyl fluorescein. Blood was then removed after labelling (baseline). Two weeks after labelling, the monkeys were implanted with the ceramics. The monkeys were then sacrificed one month later and blood was removed for sampling (sacrifice). Blood was analyzed for the following parameters (ppm); glucose, BUN, creatine, UA, sodium, potassium, chlorine, carbon dioxide, PO4, calcium, total preoteins, albumin, globulin, alkaline phosphates, SGOT, SGPT, LDH, bilirubin, white blood cells, red blood cells, hemoglobin, HCT, MCV, MCH, MCHC, neutrophil, lymphocytes, monocytes, eosinophols, basophils.

IV RESULTS AND DISCUSSION:

a. Preparation of TCP and ALCAP

The synthesis procedure described has been used by the University of Dayton. The theoretical yield calculated for this reaction is 86.3 grams. The actual yield, after sintering, was approximately 60 grams with major losses attributed to losses in the centrifugation and washing processes.

X-ray diffraction analysis performed on the first batch of material indicated that the sintered form was pure beta-calcium orthophosphate (see figure 3). Based on this result, seven additional precipitation runs were conducted.

b Composites

Table 1 represents the pH of the different composites over time. It has been suggested (Bindermann, 1987) that an alkaline environment promotes bone growth. Addition of vitamin D in the presence of malic acid or alpha ketoglutaric acid alone lowered the pH to an unacceptable level. TCP with vitamin D alone maintained an alkaline pH upto 48 hours. On suspension in neutralized saline buffer (pH 7.4) TCP alone also maintaianed an alkaline pH environment. The data obtained on the pH characteristics of the composites suggest that the appropriate composites for studying bone remodeling should include: TCP with malic acid and calcium hydroxide, TCP, TCP with the addition of DHCC, and TCP with the addition of malic acid, calcium hydroxide and DHCC.

Table 2 indicates the setting hardness characteristics of each composite. It is evident that malic acid or alpha ketoglutaric acid is required to set TCP. The addition of DHCC did not effect the rate of setting to hardness, but it did demonstrate the ability to set TCP to hardness.

However, after prolonged exposure to air, these composites became brittle and broke upon touch. Morris (1988) has suggested that lysine, proline, or cysteine set TCP rapidly and the composites remain strong.

c Pore size

The scanning electron microscope at the Toxic hazard Division of W.P.A.F.B. was used to take photographs of the fractured edges of the composites for determining the pore sizes of the composites. It was determined that there was no significant difference among pore sizes of the different set and hardened TCP composites. Figures 4 and 5 show scanning electron micrographs (SEM) of TCP and ALCAP respectively. A semiautomatic computerized videoplan was employed to determine area of each pore in the photograph. The TCP composites had 10 % of their surface area covered by pores, whereas, only 4 % of the surdace area of the ALCAP ceramic was covered by pores.

d Blood analysis

Because the protocol was approved at a late date and complications were encountered with the quarantine of the rats, the study on TCP and TCP components was postponed. However a similar study implanting ALCAP composites in rhesus monkeys was being conducted. Blood obtained from these monkeys was analyzed to observe the effects of ALCAP implants in these animals. Since both studies involve implantation and remodeling, the data obtained in the ALCAP study can be correlated with the rat study.

The differences among the animals did not have a significant effect on the blood chemistry parameters. However, differences in the amount of total bilirubin in the blood collected at the three time periods were significantly different. They were within the normal range for total bilirubin in the blood of rhesus monkeys. The level of total bilirubin in the plasma decreased significantly after labelling. Thus, it appears that the labelling decreases the degradation of erythrocytes.

V RECOMMENDATIONS:

It is recommended that TCP implantation in rats should be followed up, possibly under a mini grant. This would enable Air Force personnel to further enhance an interdiciplinary approach to the analysis of bone ingrowth and remodeling. This investigation parralels the ongoing study with BBD and TDH ALCAP implantaion. The techniques employed in both of these investigations can be used to investigate the effects of prolonged vibration and mechanical stress on bone in Air Force pilots.

The use of biomaterial implants, instead of autografts, for repairing traumatized tissues in civilians, as well as Air Force personnel, will become a routine in the near future.

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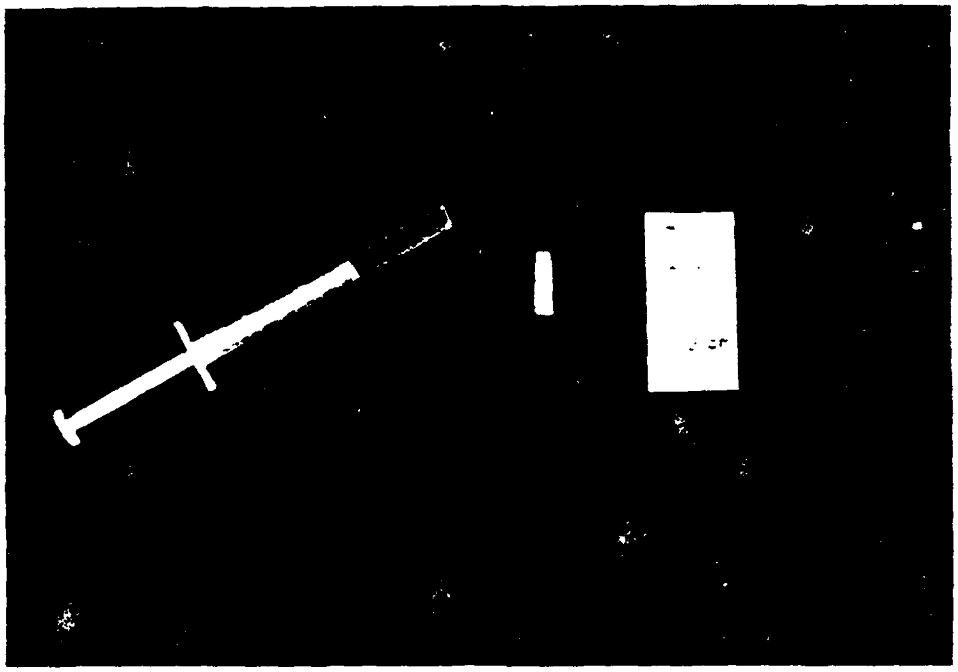

FIGURE 1: This photograph shows the procedure to form the cylindrical molds. The syringe on the left held the wet composite until it set to hardness.

FIGURE 2: The powder on the left is the TCP mixture. 133.3 mg of it was placed in the watch glass on the left and mixed with 66.7 mg of malic acid and 60 ul of distilled water. The watch glass on the left shows the hardened composite 24 hours after addition of water.

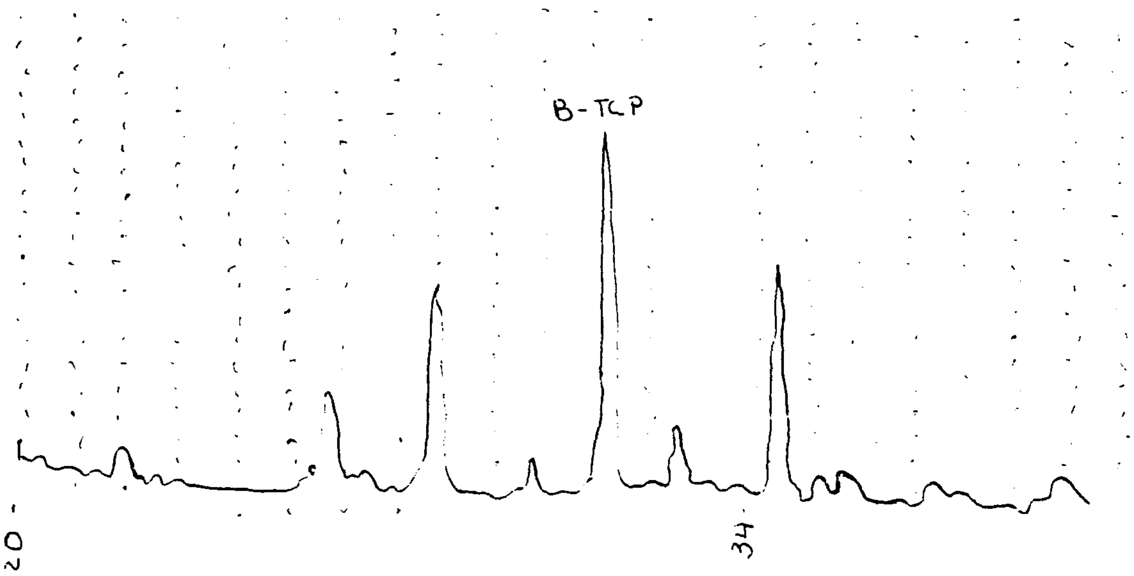

FIGURE 3: The X-ray diffraction pattern of TCP. Units are degree/DIV 1.

TABLE 1: The pH measurements for the TCP composites suspended for two days in 70 ml neutralized saline (37 C).

TREATMENT	1 Hr	24 Hr	48 Hr
saline	7.02	6.70	6.50
TCP	10.44	7.00	7.22
TCP + ketoglutaric acid	4.02	4.10	4.42
TCP + malic acid	4.24	4.22	4.22
TCP + ketoglutaric acid + CaOH2	11.64	6.65	6.60
TCP + malic acid + CaOH2	9.80	10.20	10.13
TCP + ketoglutaric acid + CaOH2 + DHCC	12.61	4.64	4.67
TCP + malic acid + CaOH2 + DHCC	11.19	6.06	6.14
TCP + DHCC	6.56	10.88	10.88

TABLE 2: Setting hardness of calcined TCP powder mixtures with 60 ul of deionized water added. A rating of 1 indicates the composite did not set. A rating of 5 indicates the composite attained consistency of gum, while a rating of 10 indicates the composite attained a consistency of plaster.

	TIME IN MINUTES								
TREATMENT	5	10	15	20	25	30	45	60	1440
TCP	1	1	1	1	1	1	1	1	1
TCP + DHCC	2	4	8	10	10	10	10	10	10
TCP + ketoglutaric acid + CaOH2	8	9	10	10	10	10	10	10	10
TCP + malic acid + CaOH2	1	3	10	10	10	10	10	10	10
TCP + ketoglutaric acid + CaOH2 + DHCC	2	3	8	10	10	10	10	10	10
TCP + malic acid + CaOH2 + DHCC	1	10	10	10	10	10	10	10	10

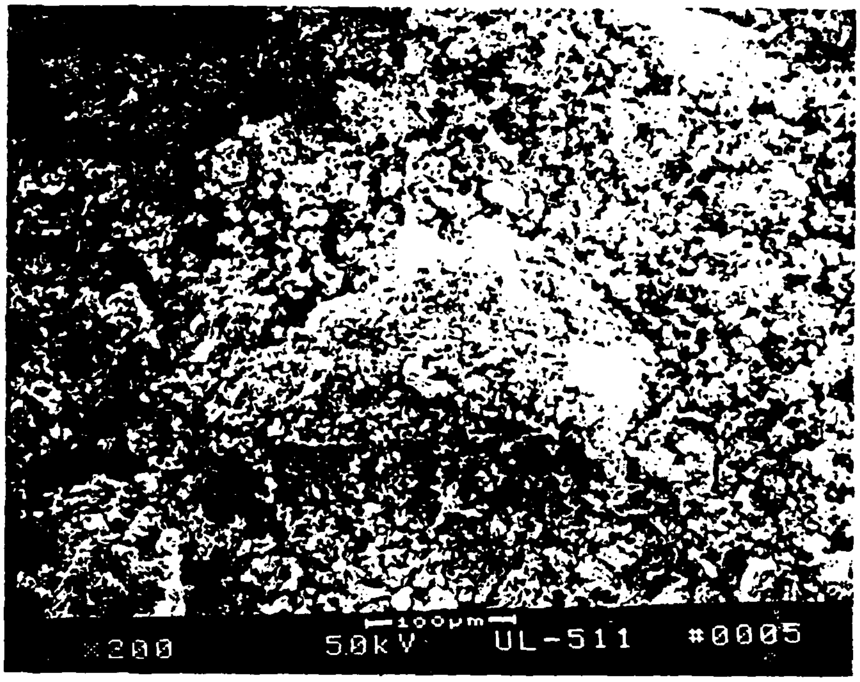

FIGURE 4: Scanning electron micrograph of tricalcium phosphate, alpha ketoglutaric acid, 15% calcium hydroxide, and vitamin D composite after setting to hardness.

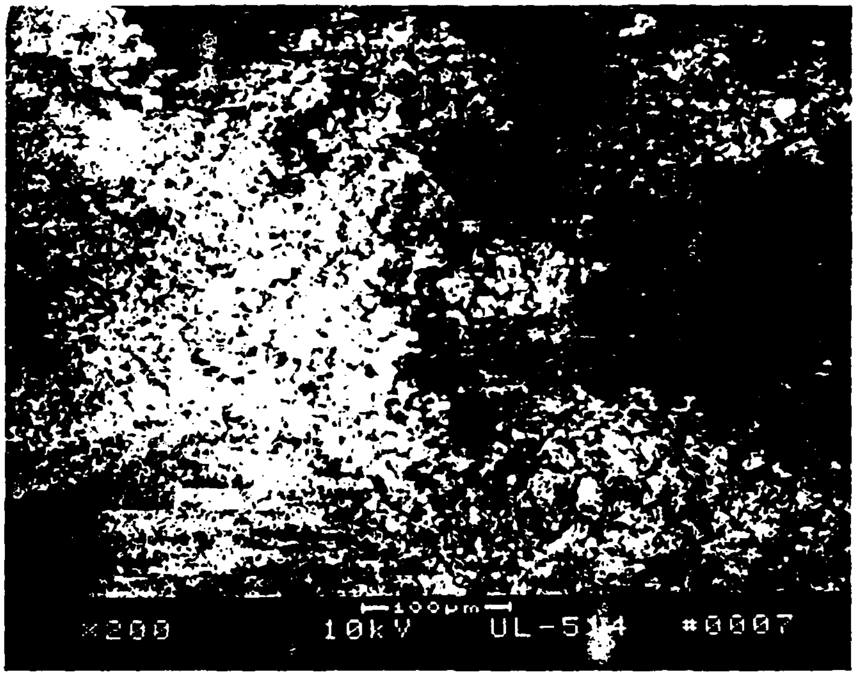

FIGURE 5: Scanning electron micrograph of ALCAP, malic acid, and 15% calcium hydroxide composite set to hardness.

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FINAL REPORT

VISUAL-SPATIAL LOCALIZATION WITH A HELMET-MOUNTED DISPLAY

Prepared by: Richard J. Kunze

Academic Rank: Graduate Student

Department and University: Department of Psychology, University of Missouri-Columbia

Research Location: Armstrong Aerospace Medical Research Laboratory - AAMRL/HEA
Visual Display Systems Branch
Wright-Patterson AFB
Dayton, OH 45433-6573

USAF Researcher: Dr. Michael Venturino

Date: 20 September 88

Contract No.: F49620-88-C-0053

VISUAL-SPATIAL LOCALIZATION WITH A HELMET-MOUNTED DISPLAY

by

Richard J. Kunze

ABSTRACT

An experiment to investigate design factors and performance factors involved in the development of helmet-mounted displays and the ability of humans to locate objects in virtual space yielded three major findings. (1) The number of targets displayed (3, 6, or 9) played a significant role in the subjects' ability to to remember target locations, with subjects decreasing in accuracy as the number of targets increased. (2) Properties of initial location of targets in virtual space (e.g. elevation) explained a significant proportion of target replacement error. (3) The size of the helmet-mounted display field-of-view affected subjects' ability to search for and find targets.

Acknowledgements

I would like to thank the Air Force Systems Command and the Air Force Office of Scientific Research for sponsorship of this research, and Universal Energy Systems for their help with the administrative aspects that arose throughout the program.

I especially wish to extend my thanks to Mike Venturino for following through with his promise of what this research program had in store me. To describe it as an extremely enjoyable and enriching experience does not begin to do it justice.

Special thanks to Bob Osgood and Max Wells for their input, encouragement, and friendship. Likewise to Ken Aldrich and Jim Shaw who endured what must have seemed an endless string of computer crashes, technical difficulties, developmental revisions, and source code changes.

I. INTRODUCTION

As computer and optical technologies have grown, numerous applications of these technologies have been proposed for use in present and future aircraft. The benefits of systems employing newly developed hardware and software are often viewed as being obvious aids to the pilot. They can detect what the human eye cannot, make computations and decisions faster than the human brain, and provide numerous types of information that have never before been available. However, increasing the amount and types of information on display systems can overwhelm a pilot or, in some cases, important information may be displayed in a less than optimal manner. As new ways are developed to take advantage of the ever growing technological world, serious considerations must be taken to assure that the advanced systems are providing advanced information that is useful and usable to the pilot without having deleterious effect on his performance.

One of the next steps for instrumentation displays in aircraft is to develop helmet mounted displays (HMD's) that afford pilots the types of information (e.g. heading, airspeed, attitude, computer sights, etc.) that heads-up displays (HUD's) do , but at all times, not merely when the pilot is looking straight ahead. Such HMD's will employ advanced computer and optical technologies to provide pilots with vital information while flying. That information would presumably include variations of HUD-type displays, but may also include new means of displaying information in more meaningful, more efficient ways (e.g. better symbology). HMD's also provide the means to display other types of information that are not currently possible on conventional displays (e.g. virtual 3 dimensional displays corresponding to the 3 dimensional world the pilot operates in).

The development of HMD's depends not only on the available technology, but on empirically determining the optimal means of applying the technologies. Use of advanced systems can afford one myriads of types and forms of information, but again, it must be empirically determined which types of information are most valuable and what form the information will take so its communicative value will be optimized. To make such empirical determinations, numerous experiments must be performed to investigate not only the systems themselves, but the human factors that influence and effect the use (or misuse) of the systems (i.e. the design of such systems must be consistent with the human operator's ability to gather, interpret and use the information that the systems provide, as well as their ability to take full advantage of the systems. The operator must also be able to easily understand and operate them with a minimal amount of cognitive effort).

The areas of emphasis in my graduate studies have, in general, been cognitive psychology and information processing. My research interests have been focused on the encoding of spatial information. These areas were heavily drawn upon in this summer's research effort. In addition to the above, the research also demanded integrating aspects of human sensation and perception.

II. OBJECTIVES OF THE RESEARCH EFFORT

The objective of this summer's research was to investigate human factors involved in the development and use of new visual display systems (particularly HMD's) for future aircraft The Visually Coupled Airborne Systems Simulator (VCASS) at the Armstrong Aerospace Medical Research Laboratory (AAMRL) has been used in the investigation of human factors for such display systems. VCASS was the primary tool employed in this summer's research. Upon my arrival at AAMRL, a number of on-going experiments using the VCASS system had been looking into various aspects of HMD's and human performance using said devices. My efforts were directed toward the evaluation of these experiments as well as to the design of subsequent experiments to further clarify previous results.

Initial variables to be manipulated include field-of-view (FOV) (i.e. the size of display window in the HMD that provides enhanced information), the cognitive load on the subject (i.e. the number of targets they were required to find and remember), and the physical location attributes of targets (i.e. the contribution of initial target elevation and azimuth to target replacement error). A number of dependent measures were taken on the subjects' performance. They included total search time, average search time per target, total replacement time, average replacement time per target, and average replacement error.

One study was completed during my ten week stay, and three others were planned and in the development process. This report will detail the procedure and results of the one completed study. The results of it and the subsequent studies should lead to a better understanding of the constraining factors that must be accounted for in the final design and development of workable visual display systems and HMD's of the future.

III. METHOD

Subjects

Subjects were paid volunteers obtained from an AAMRL subject pool that is maintained by Systems Research Laboratories under U.S.A.F. contract. All subjects were right handed, male subjects with 20/30 visual acuity or better, ranging in age from 18 to 30. A total of 12 subjects began the experiment, however, 2 were dropped following the first session. One was dropped when he failed to return for the data collection session that followed the practice session, and the other because he was unable to return for the data collection session within a reasonable time.

Stimuli and Apparatus

This experiment was performed using the Visually-Coupled

Airborne Systems Simulator (VCASS) at the Armstrong Aerospace Medical Research Laboratory. The display used consisted of two helmet-mounted CRT displays with a combined maximal field-of-view of 120 deg azimuth and 80 deg elevation with 40 deg binocular overlap. The position of the helmet was measured and updated at 20 Hz. Readings were taken on all 6 axes (3 rotational and 3 translational) with an electromagnetic helmet position tracker. The helmet position information was used to present space-stabilized images on the displays.

While in the helmet, the subjects viewed a computer-generated stroke-drawn world of 4 pi steradian at optical infinity. The entire scene was updated at 20 Hz. Subjects sat in a simulated generic fighter aircraft cockpit with a centrally located control stick.

Head-stabilized viewing windows (henceforth referred to as the fields-of-view or FOV's) were constantly maintained at the center of the helmet's combined display (i.e. information from the head tracker updated the position and moved the FOV with the head). The FOV for each trial measured 20x20, 30x30, 45x42.5, 60x50, or 90x60 (degrees azimuth x degrees elevation). At the center of the FOV was a constantly visible reticle cross (approximately 3.4 deg high and wide). The FOV's were used to locate targets. In order for a target to be seen it had to be inside the FOV. Terrain could be seen anywhere within the overall 120 deg by 80 deg display regardless of whether it was inside or outside the FOV. The terrain scene was composed of recognizable landmarks. The scene had a horizon at zero deg elevation and subtended 360 deg in azimuth.

Targets were small squares approximately 2.2 deg high and wide with numbers within their perimeter. They were presented at stationary positions in a gaming area that extended 120 deg left and right as well as 90 deg upward from straight ahead. The gaming area was sectioned off into 9 bands equidistant from straight ahead. The bands can be described as concentric circles of increasing size radiating outward on the inside of the virtual sphere with their centers located straight ahead. In a given trial, subjects were presented with 3, 6, or 9 targets located in the 9 bands.

Target positions were determined pseudo-randomly to ensure equal dispersion throughout the gaming area. In a three target condition targets were located in bands 1,4, and 9. In a six target condition they were in bands 1, 3, 4, 6, 7, and 9. All nine bands were sampled in the nine target condition. There was an additional stipulation that all targets had to be separated by at least 10 deg.

In order to somewhat equate the difficulty of target patterns over subjects, 15 target patterns were generated (1 for each target density/FOV combination). Each subject received the same patterns in the same conditions. The numbering of the targets was achieved by assigning consecutive numbers to the targets based on azimuth. Each target density/FOV combination was viewed twice by all subjects (once labelled in a right-to-left manner and a second time labelled left-to-right). The actual order of presentation of patterns was randomized for each subject.

Procedure

On the first day (practice session), subjects read instructions detailing the experiment. The experimenter then placed the subject in the cockpit and further described the task. The subject was informed about how the helmet would project a virtual space. A transparent half-sphere was used to show how far the around the virtual space extended and where targets would be in it in relation to the subject. The sphere was also used to demonstrate how the targets would be numbered.

The FOV's and reticle were described and subjects were informed that these images would move with them and remain directly in front of the helmet at all times. Subjects were advised to move freely in the cockpit and to use any strategy they found useful (short of using cues from outside the helmet) to find and remember the target locations. The subject would then be shown the switches they would be using on the control stick and how to use them.

The helmet was then placed on the subjects head, boresighted, and checked for optical clarity. Subjects were advised of the sensitivity of the optics and then instructed on guessing procedures. They were encouraged to set a high criterion for what they labelled as a guess.

Subjects received 20 training trials during the practice session. The first 15 trials were a step through procedure in which the subject received each target density (3, 6, then 9) for each FOV size (90,60,45,30, then 20). In the last 5 practice trials, the five different FOV's were presented in random order, each with 6 targets. At the beginning of the practice trials, the experimenter "walked the subject through" the first few trials until the subject reported feeling comfortable with the procedure.

During the experimental session on day two the subjects completed 33 trials. The first three were warm-up trials designed to re-acquaint the subject with the procedure. The warm-up trials were followed by two blocks of 15 trials. Each block contained all fifteen possible combinations of target densities and FOV's and were presented in random order. In addition, if a trial occurred in the first block that was labeled left-to-right, its corresponding trial in the second block was labeled right-to-left.

Design

A 2 X 2 repeated measures design was employed in this study. The manipulated variables were target density (3, 6, or 9 targets), and FOV size (20,30,45,60,90 degrees).

IV. RESULTS

Two-way repeated measures ANOVA's where performed on the data for each of five primary dependent measures, total search time, average search time per target, average replacement time per target, total replacement time, and average replacement error. Chi squared tests were run to determine whether the frequency of guessing was differentially effected by the major independent variables (FOV and target density). ANOVA's were also performed on the data from each target density condition to determine the effect of serial position. In addition, a regression analysis was performed on the data to determine the significance of target azimuth and elevation as predictor variables for replacement error.

Total Search Time

The total search time a subject used in each of the FOV by target density conditions was taken to be the average of the search times for the two trials in each condition. The results of the ANOVA indicated a large main effect for FOV $F(4,36)=26.94$, $p<.0001$. With smaller FOV's subjects presumably had more difficulty finding and rehearsing the target patterns. Not surprisingly, the results also indicated a profound main effect for target density $F(2,18)=100.13$, $p<.0001$. When the number of targets increased, the search times increased. A significant FOV by density interaction was also present $F(8,72)=7.97$, $p<.0001$.

Average Search Time per Target

By simple virtue of the fact that it takes longer of search for and rehearse the locations of 9 targets than it does for 3 or 6, it can well be argued that total search time is an obvious difference that should be expected between the three target densities. It was for this reason that we also looked at average search time per target, thereby adjusting, or accounting, for the different number of targets viewed. Therefore, the search times for each trial were divided by the number of targets searched for. In doing so, it allowed us to examine the effect of the variables on the amount of time subjects devoted (on the average) to learn each target. It was found that even after giving a subject a score based solely on a per target basis, that the main effects and the interaction were significant. For FOV $F(4,36)=20.29$, $p<.0001$, for target density $F(2,18)=4.79$, $p<.025$, and $F(8.72)=6.46$, $p<.0001$ for the interaction (see figures 1 and 2).

Total Replacement Time

Unlike total search time, replacement time was not effected by FOV $F(4,36)=0.78$, $p>.50$. However, as one would expect, it was effected by target density $F(2,18)=109.92$, $p<.0001$. It took longer for subjects to replace 9 targets, less for 6, and even less for 3. Here too, there was a significant interaction $F(8,72)=2.31$, $p<.05$.

Replacement Time per Target

As with the problem identified above with total search time, total replacement time is also problematic when viewed in the context of its absolute value. Here again, times were divided by the number of targets to obtain a measure that indicates how quickly (on average) each target was replaced. This measure may indicate the care and thought the subjects invested in each target replacement. On a per target basis, there were no significant main effects, and the interaction was just shy of significant $F(4,36)=1.90$, $F(2,18)=2.33$, and $F(8,72)=2.03$ for FOV, target density and the interaction, respectively (see figures 1 and 2).

Replacement Error

Mean replacement error was obtained by averaging error scores for each of the two trials in each condition. While there was no effect for FOV $F(4,36)=0.83$, $p>.5$, there was a large main effect for target density $F(2,18)=46.88$, $p<.0001$. With more targets, subjects made larger errors at replacement time. There was also a significant interaction between the variables $F(8,72)=2.79$, $p<.01$ (see figure 3).

Frequency of Guessing

A chi squared analysis was run on each target density conditions for each of the major independent variables (FOV and target density) as well as for serial position. In order to run a chi squared test based on the initial azimuths and elevations, target azimuth and elevation were broken into discrete categories spanning 30 deg of absolute displacement from straight ahead. There were no significant differences on any variable.

Target Variables

By labelling targets and requiring subjects to replace a specific target in a specific position, it was possible to do one-to-one match-ups between initial target locations and the subsequent replacement positions rather than using the least error method that was employed in previous experiments (Wells, Venturino, & Osgood, in press). In addition, the one-to-one correspondence afforded the opportunity of examining the effects of individual target location properties (specifically azimuth, elevation, and serial position) on individual replacement errors. Use of the least error method had not afforded this option in a clean manner as it made assumptions about the replacement positions and minimized error without regarding the subject's intended performance.

Serial Position

ANOVA's were performed for each density condition to examine the effect of serial position on replacement error. Although there was no effect for serial position in the three target condition, there was an effect in the six and nine target conditions. In the six target condition, $F(5,45)=3.21$, $p<.025$. Further analysis revealed that position 5 yielded significantly higher error scores than position 1. In the nine target condition, $F(8,72)=9.96$, $p<.0001$. Position 8 yielded more error than positions 1, 4, 5, 6, and 9, error in position 7 was greater than in positions 1, 4, 5, and 6, position 3 had more error than position 5, and position 2 was also associated with more error than position 5 (see figure 3).

As can be seen in figure 3, the graph of error versus position looks very little like one would expect when looking for serial position effects. Further investigation revealed that an unforeseen confound had occurred between the band in the gaming area where the target was drawn from and the number a target was labelled with. Particularly in the nine target condition, the number 5 target had always been in band five (i.e. it had always been located straight ahead with very little variation in positioning).

Azimuth and Elevation

The overall correlation, across all conditions, between initial target elevation and replacement error was .0915, p,.0001. Individual regression analyses performed on each target density condition produced R-squared values of .1576, .0460, and .0533 for the 3, 6, and 9 target conditions, respectively. All were significant at $p<.0001$. Azimuth only made a significant contribution to the regression equation in the 9 target condition.

The outcome of this study replicates findings of past studies (e.g. Wells, Venturino, & Osgood, in press). It was found the the size of the HMD field-of-view affected performance aspects of human subjects differentially. While the size of the FOV had a large significant effect on the subjects' ability to find and rehearse target locations, it did not affect their ability to replace a target once learned. Evidently, when the FOV is providing unique information that is only available when properly positioned, the smaller FOV's contribute to large decrements in performance. However, once the subject has obtained the information provided by the FOV, he can perform equally well whether that information was obtained with a large FOV or a small one.

On the other hand, the number of targets appears to affect the subjects' performance pretty much across the board. The greater the number of targets, the poorer the performance. In addition, this variable has significant interactions with FOV size making it more difficult (considering the afore mentioned trade-offs) to determine an optimal FOV size that will maximize the ability of the user to perform well under all conditions.

Additionally, it was found that human subjects tend to perform less well with variations in target elevation than with variation in azimuth. Specifically, targets with high elevations tended to have much more error associated with them than low elevation targets. Although this finding is currently in the process of being replicated, it would seem that this may be a possible area to focus on in the development of HMD's. If operating with elevations is indeed a problem for humans, perhaps displays can be developed to help overcome this "handicap."

V. RECOMMENDATIONS

The potential benefits of employing computer and optical technologies within the cockpit are emormous. HMD's have the ability to display 3 dimensional images thereby opening a whole new world of possible information displays. HMD's can have other systems slaved to their movement for better (more versatile) use (e.g. slaving radar, guns, missiles, etc. so they are operative in whatever direction the pilot is looking). With the enormous amount of information that pilots must handle in everyday (let alone combat) situations, it is vitally important to continue the development of systems that enhance pilots' abilities, not restrict them.

This experiment, and the subsequent ones to follow, merely scratch the surface investigating aspects of HMD's. In order to unlock the full potential of such display devices, much further research must be done. Additionally, the communication of the availablity and potential of these technologies should spawn even more new and innovative applications of these displays.

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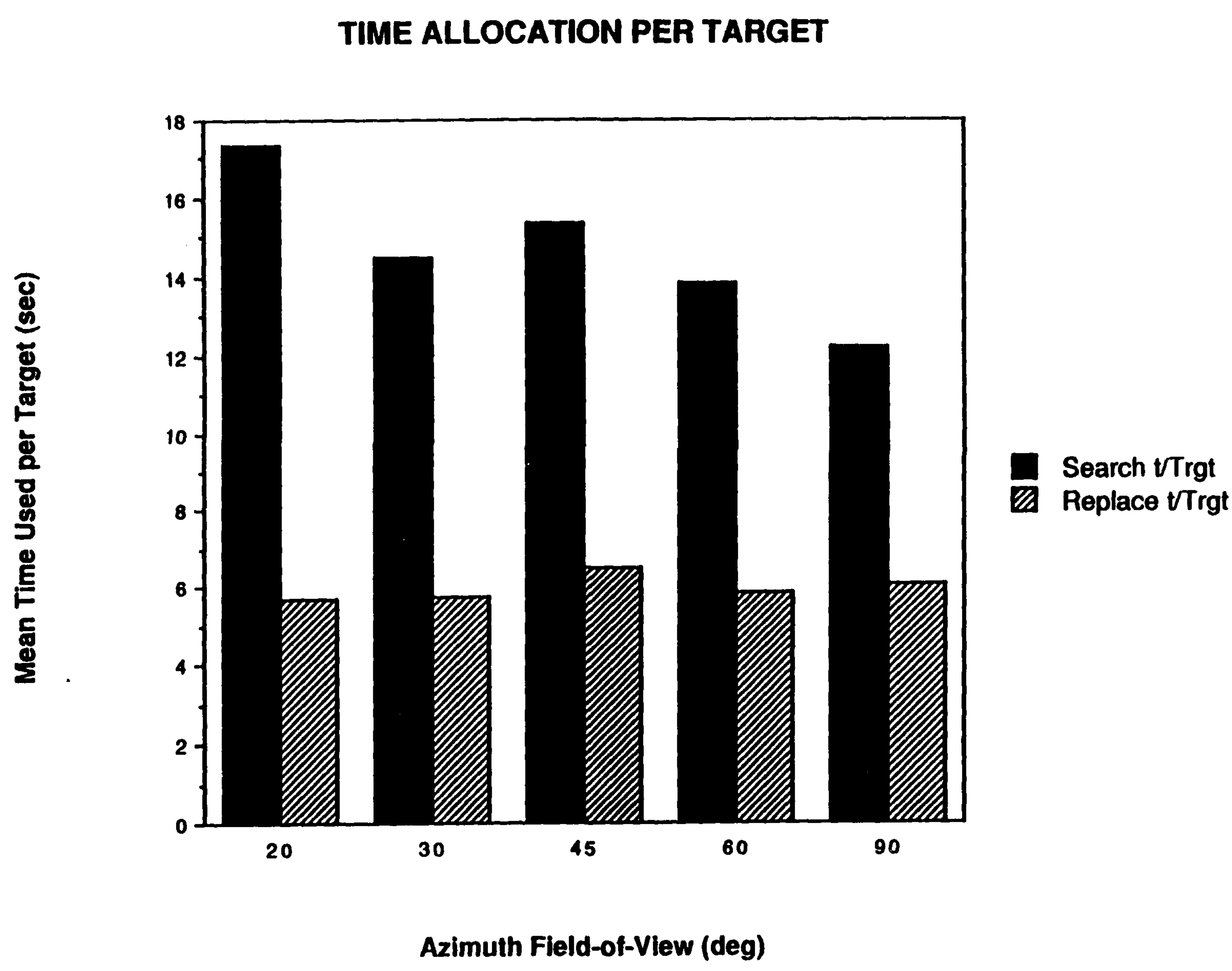

TIME ALLOCATION PER TARGET
Mean Time Used per Target (sec)
18
16
14
12
10
8
6
4
2
0
20
30
45
60
90
Azimuth Field-of-View (deg)
Search t/Trgt
Replace t/Trgt

FIGURE 1.

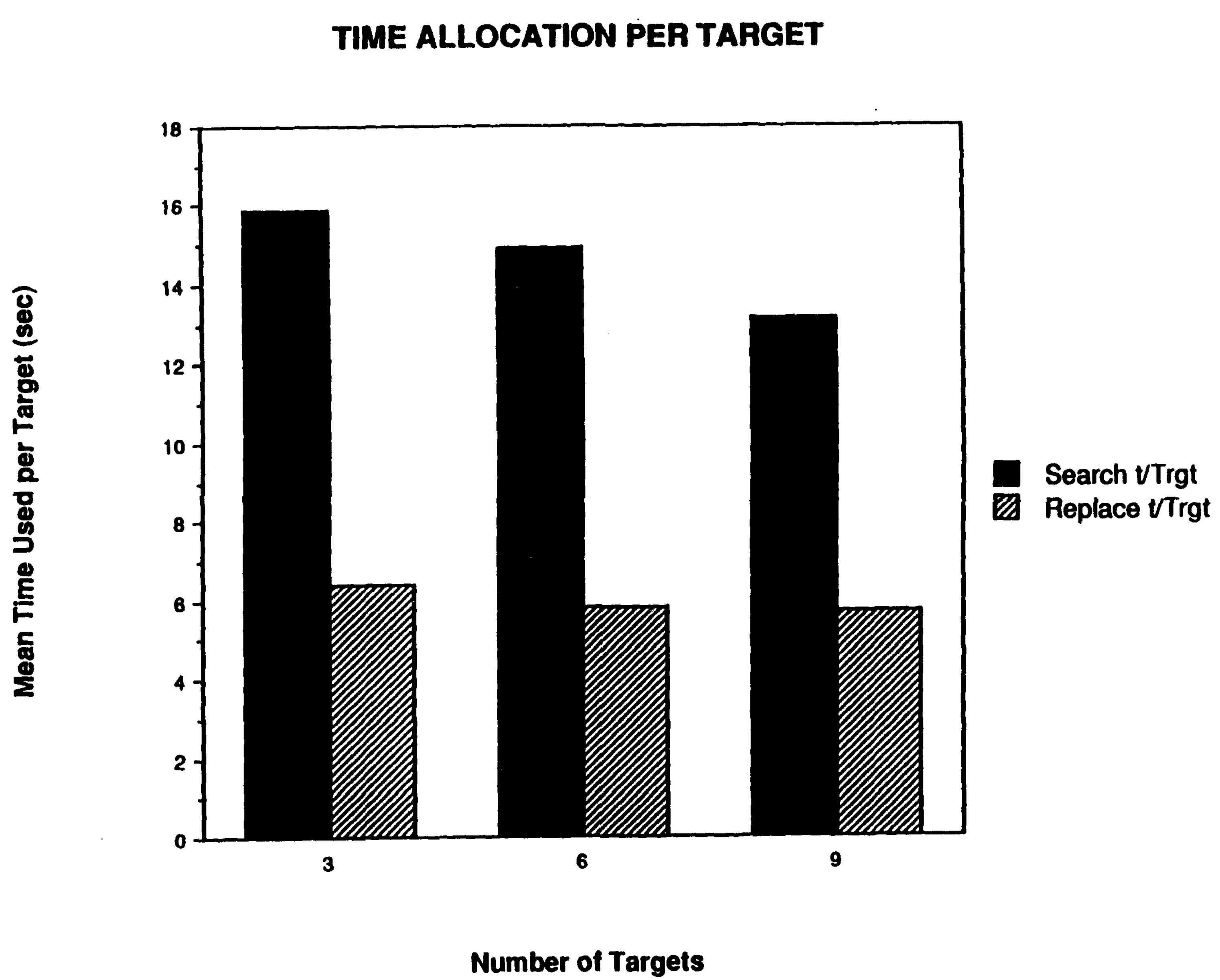
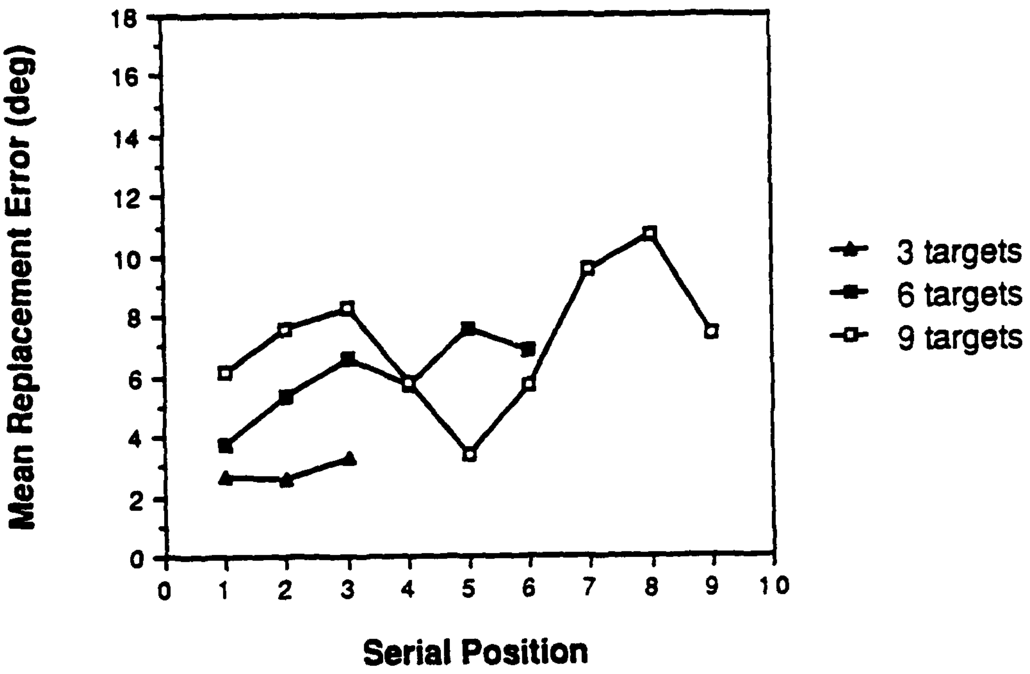

TIME ALLOCATION PER TARGET
Mean Time Used per Target (sec)
18
16
14
12
10
8
6
4
2
0
3
6
9
Number of Targets
Search t/Trgt
Replace t/Trgt

FIGURE 2.

REPLACEMENT ERROR (Averaged over FOV's)



DEVIATION OF REPLACEMENT ERROR

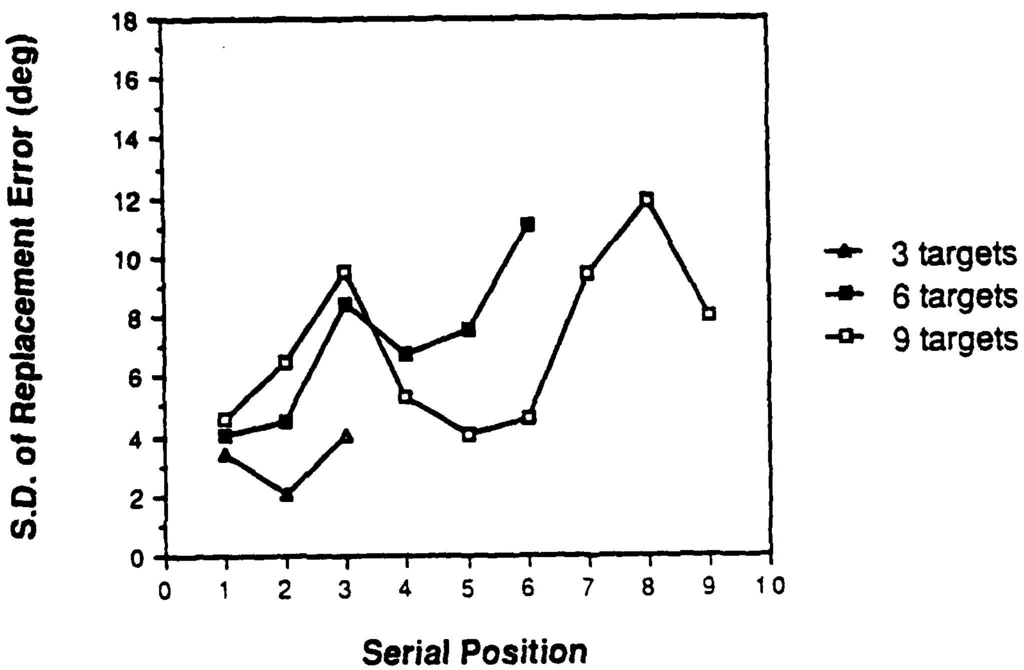


FIGURE 3.

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FINAL REPORT

EVALUATION OF AN EXTRACTION PROCEDURE FOR THE ANALYSIS OF SERUM STEROIDS

Prepared by:	Robert E. Masingale, Sr., Ph.D.
Academic Rank:	Professor
Department and	Chemistry Department
University:	Jarvis Christian College
	Deborah J. Mitchell
	Graduate Student
	Chemistry Department
	Prairie View A&M University
Research Location:	Naval Medical Research Institute
	Detachment (Toxicology)
	Wright-Patterson AFB
	Dayton, OH 45433
USN Researcher:	Linda A. Lininger, LT, MSC, USN

Date:	19 September 1988
Contract No:	F49620-88-C-0053

SAME REPORT AS
PROF. ROBERT MASINGALE
HARRY G. ARMSTRONG AEROSPACE MEDICAL RESEARCH LABORATORY # 112

1988 USAF-UES Summer Faculty Research Program

Graduate Student Research Program

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Universal Energy Systems, Inc.

FINAL REPORT

Prepared by: Jacqueline Roberts-Johnson

Academic Rank: Masters Degree

Department/University: Chemistry Department of Wright State University

Research Location: Harry G. Armstrong Aerospace Medical Research Laboratory
Bldg. 79, WPAFB
Dayton, Ohio 45433

USAF Researcher: Dave Mattie

Date: September 29, 1988

Contract No: F49620-85-C-0013

NEPHROTOXICITY OF 2,5-DMH IN FISCHER 344 RATS

by

Jacqueline Roberts

ABSTRACT

2,5-Dimethylhexane (2,5-DMH) was studied as a potential carcinogen in Fischer 344 rats. This was carried out using rats dosed with 2,5-DMH and collecting the urine after a 48 hour period. Analysis via gas chromatograph and gas chromatograph/mass spectrometer produced two unidentified peaks which were established as metabolites produced by these rats. These metabolites were identified by comparison of purchased and synthesized chemicals. With the knowledge of these identified metabolites it was then hoped to determine if structures of these metabolites induced the nephrotoxicity found in these animals.

Acknowledgements

I would like to thank the Air Force Systems Command and the Air Force Office of Scientific Research for sponsoring my research. I would also like to thank Universal Energy Systems for their concern for this work.

My advisor, Dr. Paul Serve', played a big role in guiding me along the way and Dr. Dave Mattie, my effort focal point, helped keep the enthusiasm of this project alive. Thanks also goes to Kyung Yu, GC/MS analyzer, and Gayle McDonald, animal doser.

I. INTRODUCTION:

Air Force personnel are constantly being exposed to jet-fuel fumes. These fuels are composed of various hydrocarbons. Several type hydrocarbons have been studied in depth to determine if they could produce harmful effects in man. Fischer 344 rats were chosen as the species to perform the research on because hydrocarbon-induced exposure produced nephrotoxic effects in these animals.

The compound of interest, 2,5-DMH, is a branched hexane hydrocarbon. It is an isomer of octane similar in structure to 2,3,4-TMP and 2,2,4-TMP which were found to produce nephrotoxicity in these rats.[1] The research period was spent trying to determine if 2,5-DMH produced the same effect.

My research interests have been in the area of cyclic hydrocarbons with side chains and their effects on Fischer 344 rats. Specifically, my thesis research was on Nephrotoxic Effects of Ethylcyclohexane in Fischer 344 Rats. I was also part of the research efforts that were performed on t-butylcyclohexane and isopropylcyclohexane at the Harry G. Armstrong Aerospace Medical Research Laboroatory. All this previous experience contributed to my assignment for the 2,5-DMH probe.

II. OBJECTIVES OF THE RESEARCH EFFORT:

The goal of this research was to see if there exists a specific structural requirement for inducing the nephropathy seen in these rats. The identification of urinary and kidney metabolites produced by this hydrocarbon (2,5-DMH) was attempted for this purpose.

Since at the present time one metabolite is yet unidentified, a brief synopsis of the efforts undertaken will be reviewed.

III.

Slices of each kidney (control and dosed) were sent to pathology for typical hyaline droplet formation seen in previous studies.

Eight rats were dosed with 2,5-DMH and four rats were dosed with water for controls. The urinary and kidney extracts were then injected into the Varian GC for analysis.

Next, chemicals had to be either purchased or synthesized for metabolite identification. The following chemicals were purchased:

A. WILEY ORGANICS, 1245 South Sixth St., Coshocton, OH 43812
2,5-Dimethyl-2-hexanol; 2,5-Dimethyl-3-hexanol; 2,5-Dimethyl-2,5-hexanediol; 2,5-Dimethyl-2-hexene; 2,5-Dimethyl-1-hexene; 2,5-Dimethyl-1,5-hexadiene; and 2,5-Dimethyl-1,4-hexadiene.

B. Aldrich Chemical Co., P. O. Box 2060, Milwaukee, WI 53201
osmium tetroxide; tetraethylammonium acetate tetrahydrate; 90% t-butylhydroperoxide; borane in tetrahydrofuran

These chemicals had to be synthesized in the laboratory:

A. 2,5-Dimethyl-2,3-hexanediol was prepared by the osmium tetroxide of 2,5-Dimethyl-2-hexene using the procedure of Sharpless.[2]

B. 2,5-Dimethyl-3,4-hexanediol was isolated by the sodium borohydride reduction of 2,5-Dimethyl-4-hydroxy-3-hexanone using the Brown procedure.[3]

C. 2,5-Dimethyl-1,6-hexanediol was synthesized by the dihydroboration of 2,5-Dimethyl-1,5-hexadiene using the procedure of Brown.

D. 2,5-Dimethyl-1-hexanol was prepared by the hydroboration of 2,5-Dimethyl-1-hexene using the procedure of Brown.[4]

E. 2,5-Dimethyl-1,2-hexanediol wassynthesized by the osmium tetroxide oxidation of 2,5-Dimethyl-1-hexene using the procedure of Sharpless.

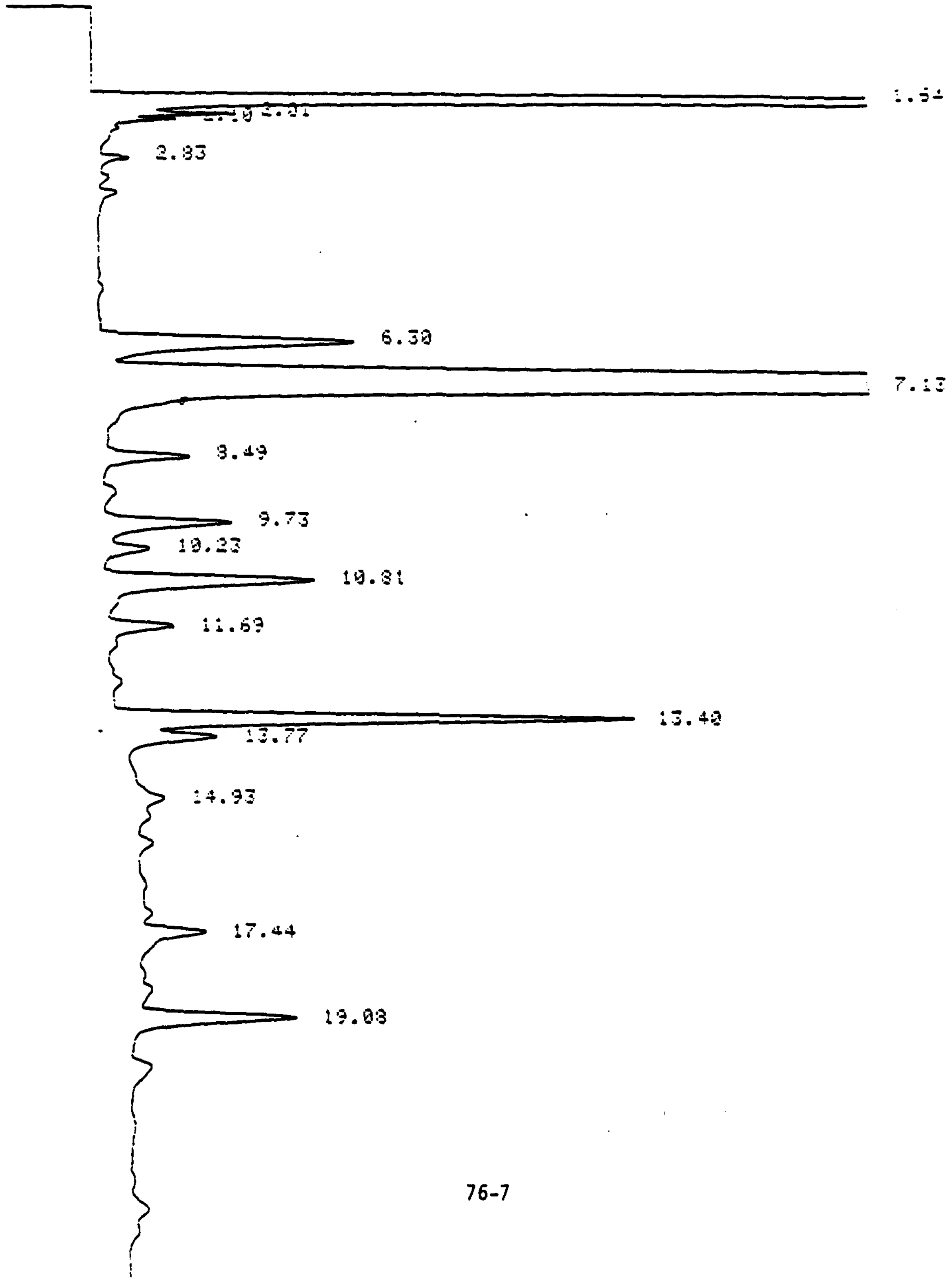
F. 2,5-Dimethyl-1-hexanoic acid was prepared by the chromic acid oxidation of 2,5-Dimethyl-1-hexane using the procedure of Bruson.[5]

All these possible metabolites were injected into the Varian 6500 GC as well as the Hewlett-Packard GC/MS for comparison with urinary metabolite data. A final test for presence of metabolite was to spike a pooled urine sample with each of the suspected metabolites and inject into the GC.

b. The following results were obtained. [1]Pathology reports indicated that there was moderate hyaline droplet formation in the dosed animals whereas there was only mild hyaline droplet formation found in the control animals. [2]Only two urinary and no kidney metabolites were found. Figure 1 represents a GC trace of a 48 hour pooled urine sample. All peaks are control peaks except those with retention times of 6.30, and 7.13, respectively.

FIGURE #1

POOLED URINE 48HRS 2,5-DIMETHYLHEXANE 1UL CW-20M 25 METER 100/1/5/180/20
LIST ATTN 2^
ATTN = 2^3



At the present time, it is believed that the peak at retention time of 6.30 corresponds to the carboxylic acid (2,5-Dimethyl-1-hexanoic acid) but absolute identification has not been made at the present date.

The peak at retention time of 7.13 was positively identified as 2,5-Dimethyl-2,5-hexanediol. GC/MS analysis solidifies this finding.

GC/MS analysis was performed on all the synthesized compounds in an effort to solve the identity of the peak at retention time of 6.30. Problems arose when fragmentation patterns of these chemicals bore no resemblance to the fragmentation patterns of the pooled urine peak unidentified at retention time 6.30. (See Figures 2-13).

IV. RECOMMENDATIONS

a. This research can be used along with all the other hydrocarbons studied to get a better idea of the cause of nephrotoxicity in Fischer 344 rats.

b. It is apparent that 2,5-DMH produces nephrotoxicity in these animals, but the question remains unanswered concerning the fact, 'Is there a structural requirement for inducing nephrotoxicity?'

c. With .rther research on hydrocarbons that are components of jet fuels, it is hoped that the U. S. Air Force will some day answer the question that it has sought after. My research has added to the list of hydrocarbons that do produce this effect.

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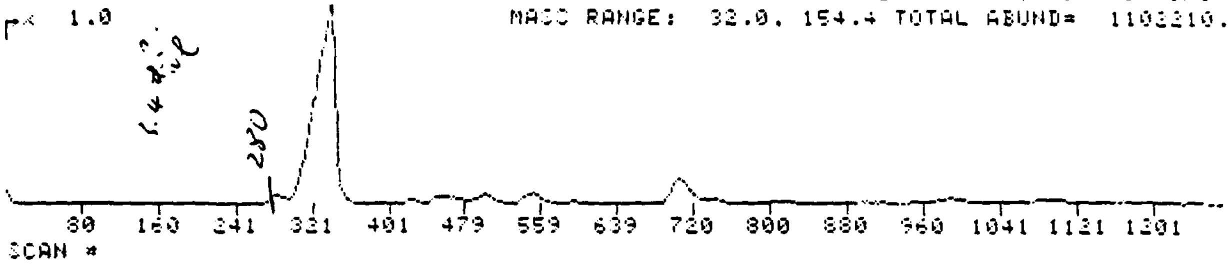
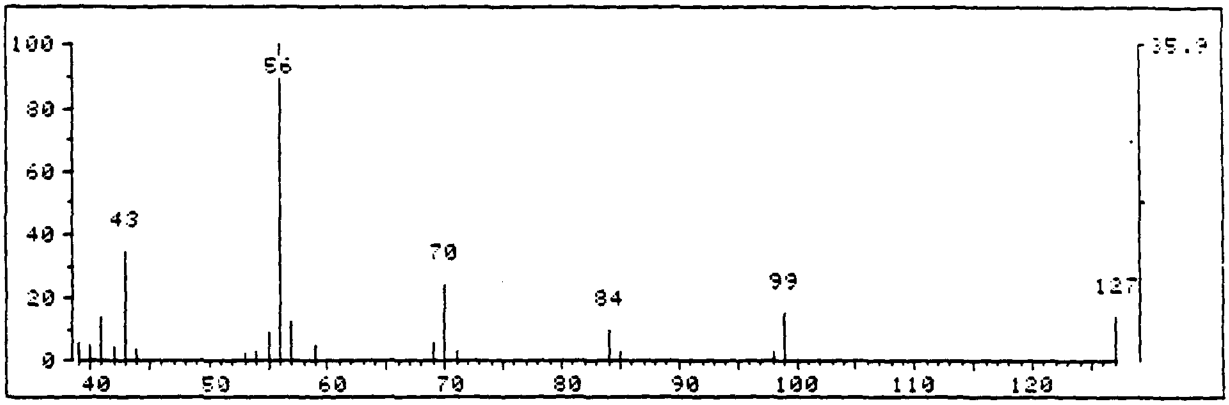
POOLED URINE OF RATS TO 3,5-DIMETHYLHEXANE 05JULY88 FRN 12229, CRN 39
CW-20M 25METER 100/1 5/180/20 5:1 1272 SCANS (1272 SCANS, 13.43 MIN)



280 RET. TIME: 7.23 TOT ABUND= 1201. BASE PK/ABUND: 56.3/ 431.

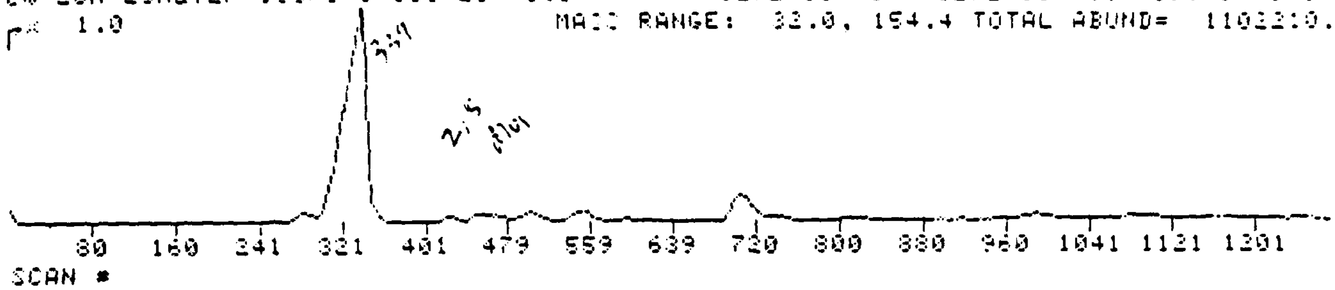


FRN 12229, SPECTRUM # 280 RET.TIME: 7.23, 20 PEAKS

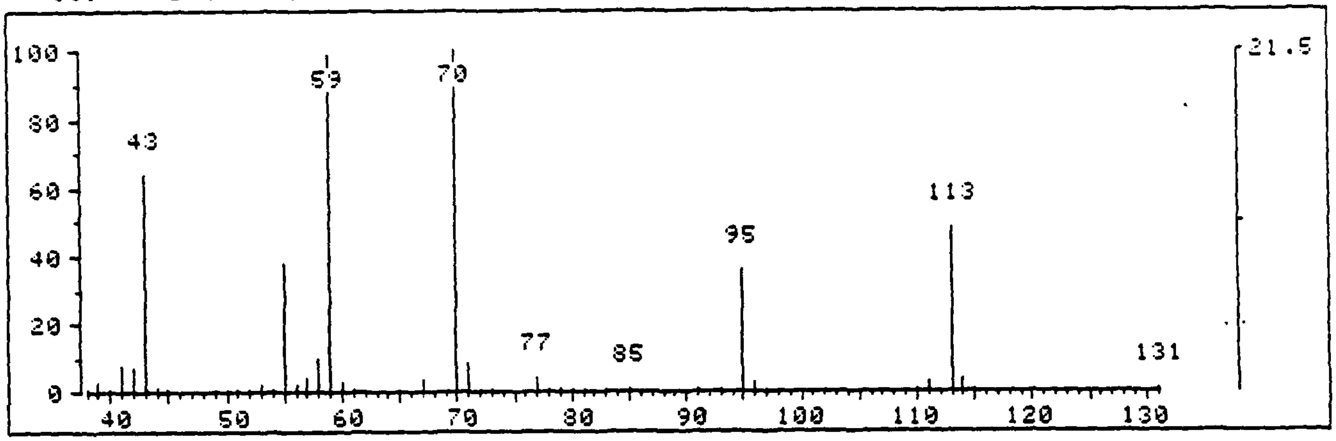
M/Z	REL ABUND	M/Z	REL ABUND	M/Z	REL ABUND	M/Z	REL ABUND
39	6.0	44	3.7	57	13.0	84	10.2
40	5.1	53	2.3	59	5.1	85	3.0
41	14.2	54	2.8	69	6.0	98	2.8
42	4.4	55	9.0	70	24.1	99	15.3
43	34.8	56	100.0	71	3.0	127	14.2

>PAUSE

POOLED URINE OF RATS TO 2,5-DIMETHYLHEXANE 05JULY88 FRN 12229, CRN 89
CW-20M 25METER 100 1 5 180 20 5:1 1272 SCANS (1272 SCANS, 19.43 MINS)
MASS RANGE: 33.0, 154.4 TOTAL ABUND= 1103210.



339 RET. TIME: 8.15 TOT ABUND= 23742. BASE PK/ABUND: 70.3/ 5100.

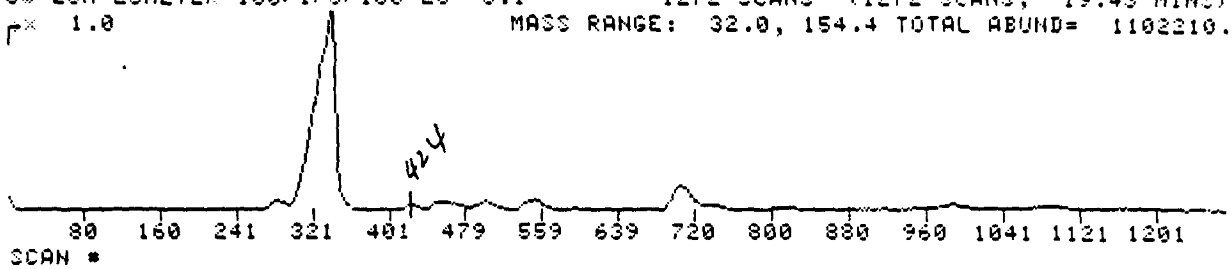


FRN 12229, SPECTRUM # 339 RET.TIME: 8.15, 45 PEAKS

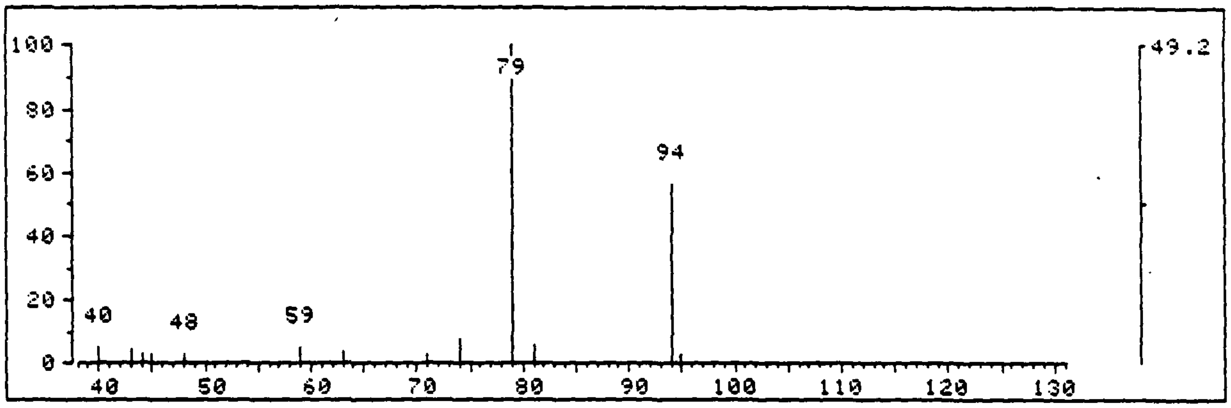
M/Z	REL ABUND	M/Z	REL ABUND	M/Z	REL ABUND	M/Z	REL ABUND
38	.3	53	2.2	71	8.6	95	35.7
39	2.8	54	.9	72	.8	96	2.8
40	.5	55	38.3	73	.6	97	1.1
41	7.7	56	2.2	77	4.3	110	.6
42	7.4	57	4.3	78	.2	111	3.0
43	64.0	58	9.6	79	.6	113	48.4
44	1.7	59	98.6	80	.3	114	3.6
45	1.3	60	3.2	81	.5	115	.5
49	.3	61	.8	83	.5	131	1.0
50	.4	65	.5	85	.7		
51	.4	67	3.6	91	.4		
52	.2	70	100.0	93	.9		

>PAUSE

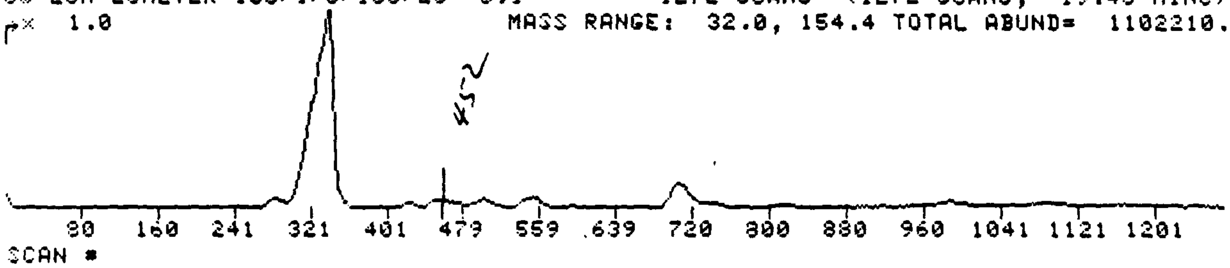
POOLED URINE OF RATS TO 2,5-DIMETHYLHEXANE 05JULY88 FRN 12229, CRN 89
CW-20M 25METER 100/1/5/180/20 5:1 1272 SCANS (1272 SCANS, 19.43 MINS)
MASS RANGE: 32.0, 154.4 TOTAL ABUND= 1102210.



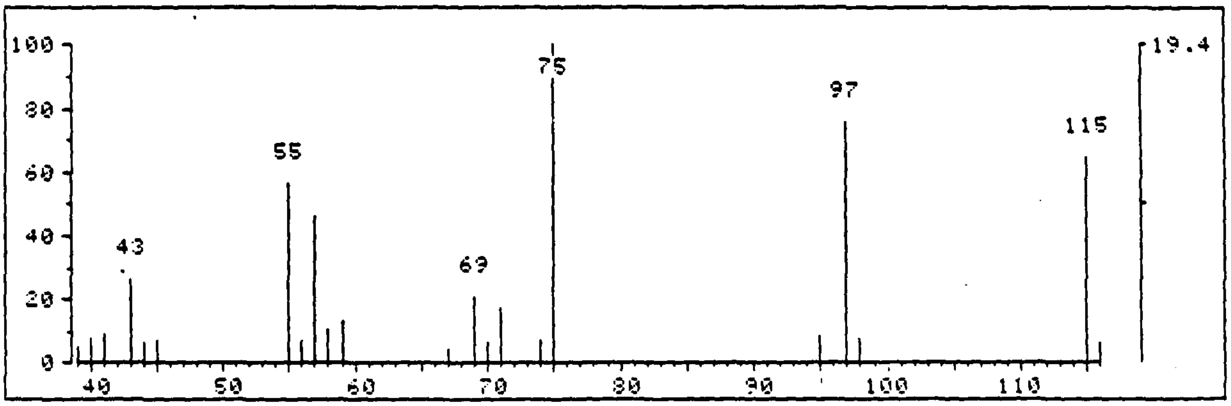
\# 424 RET. TIME: 9.43 TOT ABUND= 673. BASE PK/ABUND: 79.2/ 331.



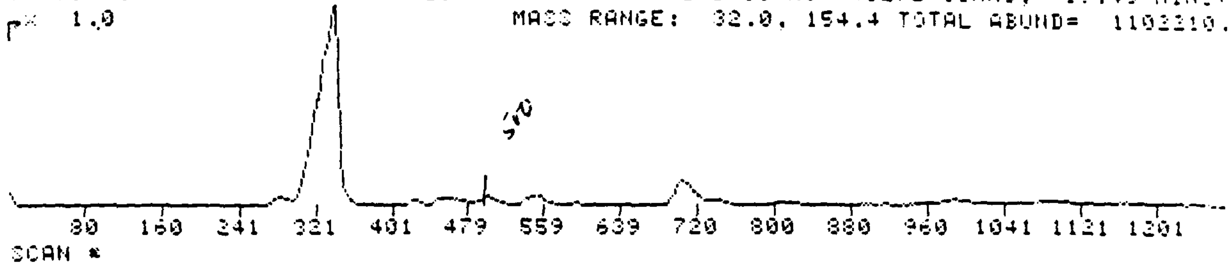
POOLED URINE OF RATS TO 2,5-DIMETHYLHEXANE 05JULY88 FRN 12229, CRN 89
CW-20M 25METER 100/1/5/180/20 5:1 1272 SCANS (1272 SCANS, 19.43 MINS)
MASS RANGE: 32.0, 154.4 TOTAL ABUND= 1102210.



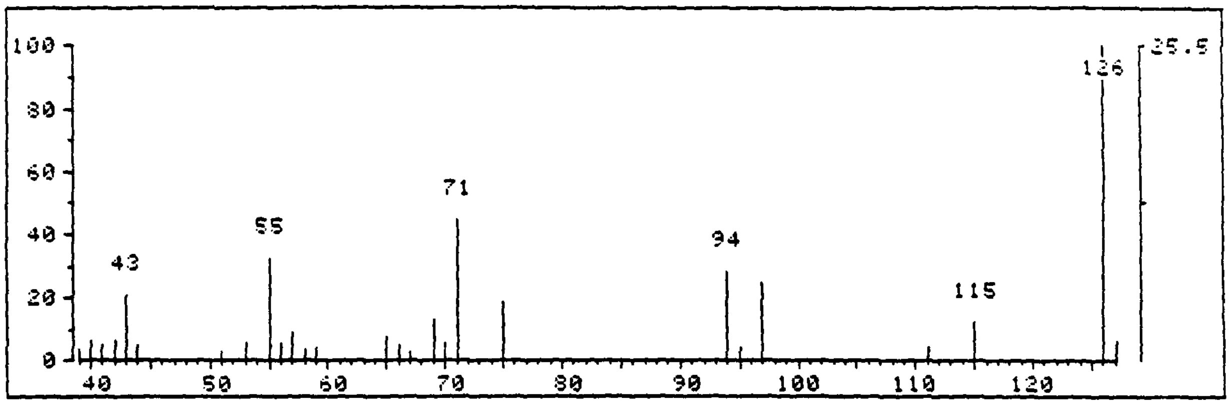
\# 452 RET. TIME: 9.87 TOT ABUND= 1084. BASE PK/ABUND: 75.3/ 210.



POOLED URINE OF RATS TO 2,5-DIMETHYLHEXANE 05JULY88 FRN 12229, CRN 38
CW-20M 25METER 100/1/5/180/20 5:1 1272 SCANS (1273 SCANS, 19.43 MIN)
MASS RANGE: 32.0, 154.4 TOTAL ABUND= 1103310.



500 RET. TIME: 10.60 TOT ABUND= 1224. BASE PK/ABUND: 126.3/ 312.

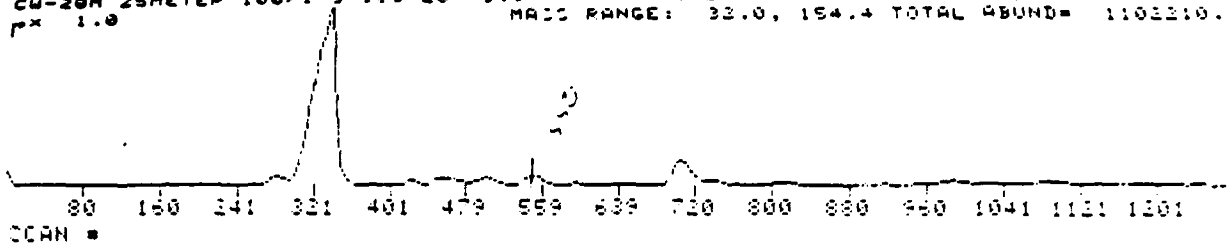


FRN 12229, SPECTRUM # 500 RET.TIME: 10.60, 27 PEAKS

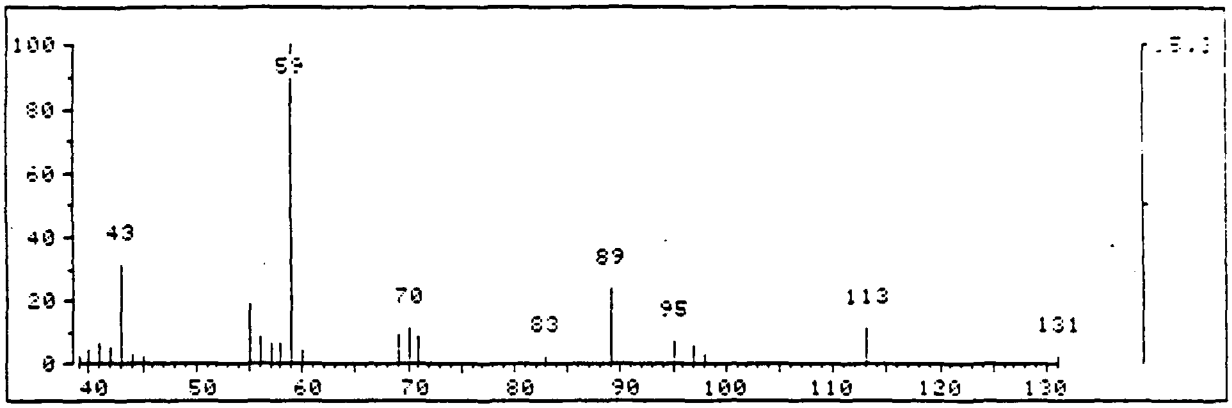
M/Z	REL ABUND	M/Z	REL ABUND	M/Z	REL ABUND	M/Z	REL ABUND
39	3.8	53	6.1	66	5.1	95	4.5
40	6.4	55	32.4	67	3.2	97	24.7
41	4.8	56	6.1	69	13.5	111	4.2
42	6.4	57	9.0	70	5.8	115	12.8
43	21.2	58	3.8	71	45.2	126	100.0
44	5.1	59	4.5	75	18.6	127	6.7
51	3.2	65	7.7	94	28.5		

>PAUSE

POOLED URINE OF RATS TO 2,5-DIMETHYLHEXANE 05JULY88 FRN 12229, CRN 39
CW-20M 25METER 100/1.5 180/20 5:1 1272 SCANS (1272 SCANS, 19.-- MIN)
R× 1.0 MASS RANGE: 33.0, 154.4 TOTAL ABUND= 1102210.



550 RET. TIME: 11.37 TOT ABUND= 1332. BASE PK ABUND: 59.4 470.

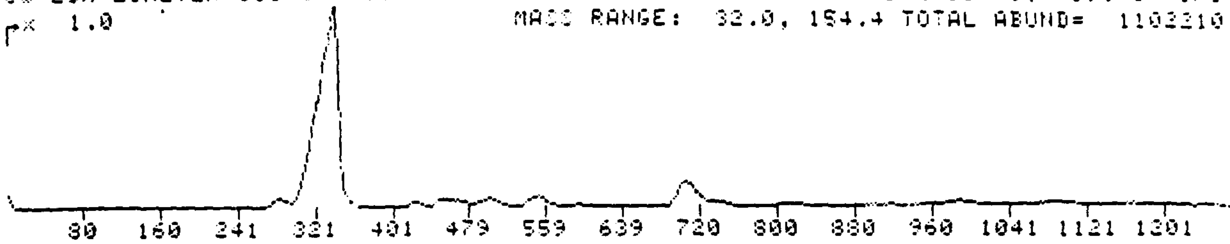


FRN 12229, SPECTRUM # 550 RET.TIME: 11.37, 23 PEAKS

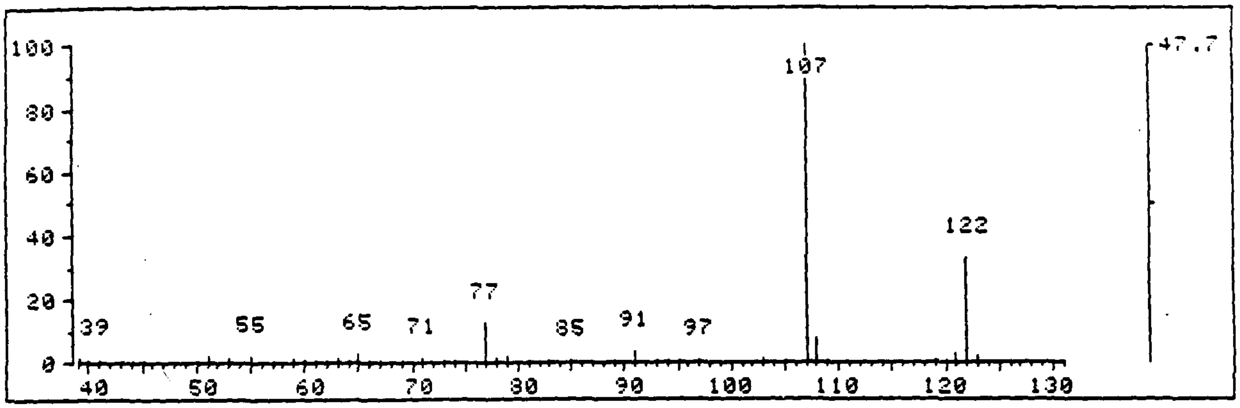
M/Z	REL ABUND	M/Z	REL ABUND	M/Z	REL ABUND	M/Z	REL ABUND
39	2.1	45	2.3	60	4.3	95	7.4
40	4.5	55	18.5	69	9.4	97	5.5
41	6.8	56	8.5	70	11.3	98	2.8
42	5.1	57	6.6	71	8.3	113	11.1
43	31.5	58	6.2	83	2.3	131	2.3
44	3.2	59	100.0	89	23.6		

>PAUSE

POOLED URINE OF RATS TO 2,5-DIMETHYLHEXANE 05JULY88 FRN 12339, CRN 39
CW-20M 25METER 100/1.5/180/20 5:1 1272 SCANS (1272 SCANS, 19.43 MIN)
MASS RANGE: 32.0, 154.4 TOTAL ABUND= 1102210.



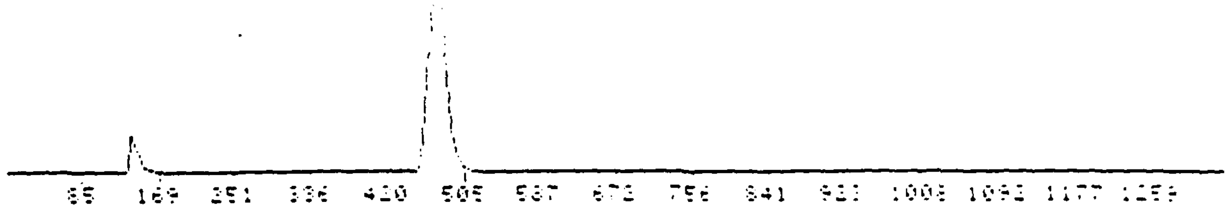
* 706 RET. TIME: 13.75 TOT ABUND= 3091. BASE PK/ABUND: 107.3/ 1475.



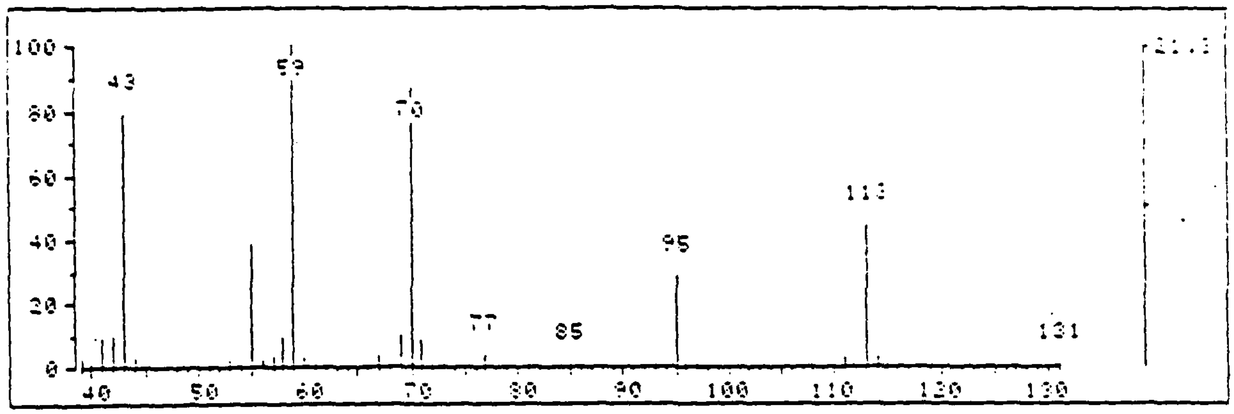
2,5-DIMETHYL-2,5-DIOL IN MEOH 23AUG88 FRN 12237, CRN

W-20M 25METER 100 1 5 180 10 1341 SCANS

1.0 MASS RANGE: 32.0, 131.0 TOTAL ABUND=



* 476 RET. TIME: 7.50 TOT ABUND= 9928. BASE PK ABUND: 59.0 2115.

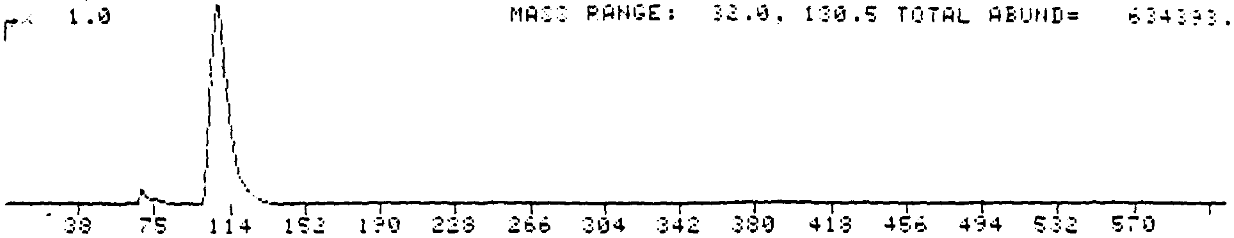


FRN 12237, SPECTRUM # 476 RET.TIME: 7.50, 39 PEAKS

M/Z	REL ABUND	M/Z	REL ABUND	M/Z	REL ABUND	M/Z	REL ABUND
39	3.1	54	1.0	69	10.0	95	28.1
40	1.7	55	38.8	70	87.1	96	2.1
41	9.4	56	2.3	71	8.5	97	1.0
42	9.7	57	4.0	72	.8	110	.5
43	78.9	58	9.3	73	.6	111	3.0
44	3.0	59	100.0	77	4.1	113	44.1
45	1.2	60	3.1	79	.8	114	3.4
50	.6	61	.5	81	.5	115	.5
51	.6	67	3.5	85	.6	131	.8
53	2.2	68	.6	93	.8		

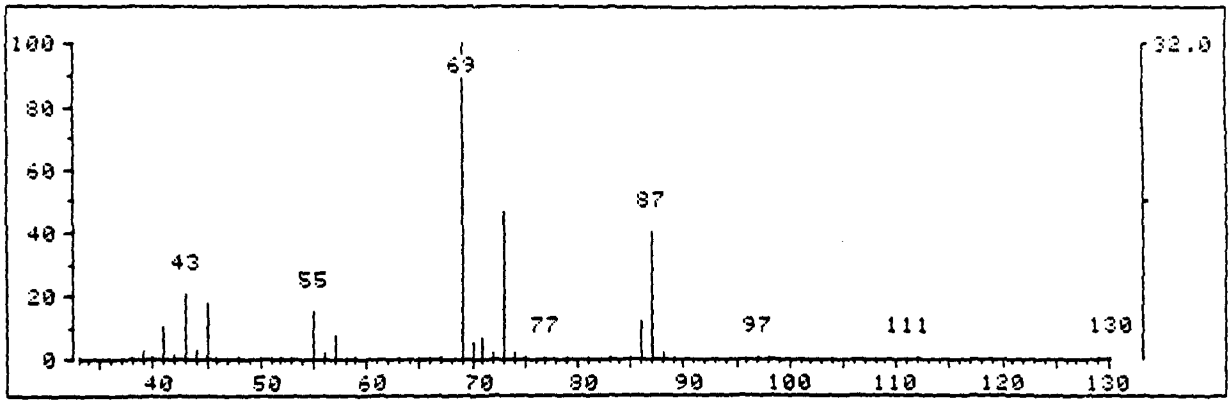
>PAUSE

2,5-DIMETHYL-3-HEXANOL IN MEOH 23AUG88 FRN 12233, CRN 39
CW-20, 25METER 100/1/5/180/30 616 SCANS (616 SCANS, 9.07 MIN)
× 1.0 MASS RANGE: 32.0, 130.5 TOTAL ABUND= 634393.



SCAN #

\# 108 RET. TIME: 3.10 TOT ABUND= 52204. BASE PK/ABUND: 69.3/ 16697.

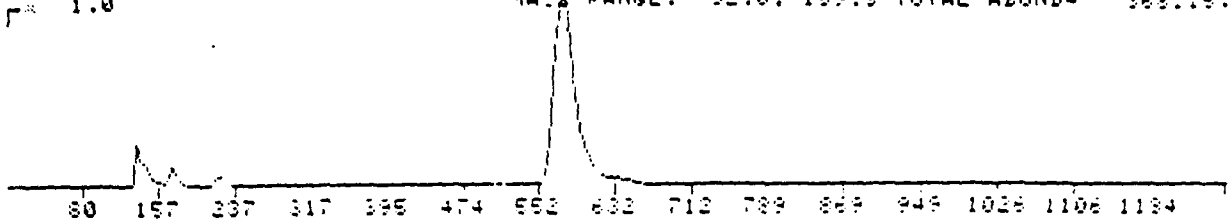


FRN 12233, SPECTRUM # 108 RET.TIME: 3.10, 47 PEAKS

M/Z	REL ABUND	M/Z	REL ABUND	M/Z	REL ABUND	M/Z	REL ABUND
33	.1	50	.1	67	.8	85	.5
38	.1	51	.3	69	100.0	86	13.0
39	2.8	52	.1	70	5.9	87	41.0
40	.6	53	1.1	71	7.3	88	2.2
41	10.4	55	15.4	72	2.4	89	.2
42	1.4	56	2.3	73	47.1	97	1.1
43	21.2	57	8.0	74	2.2	98	.1
44	3.1	58	1.0	75	.1	111	.2
45	18.1	59	1.1	77	.1	112	.1
46	.4	63	.1	79	.1	129	.1
47	.1	65	.2	81	.1	130	.1
48	.1	66	.1	83	.1		

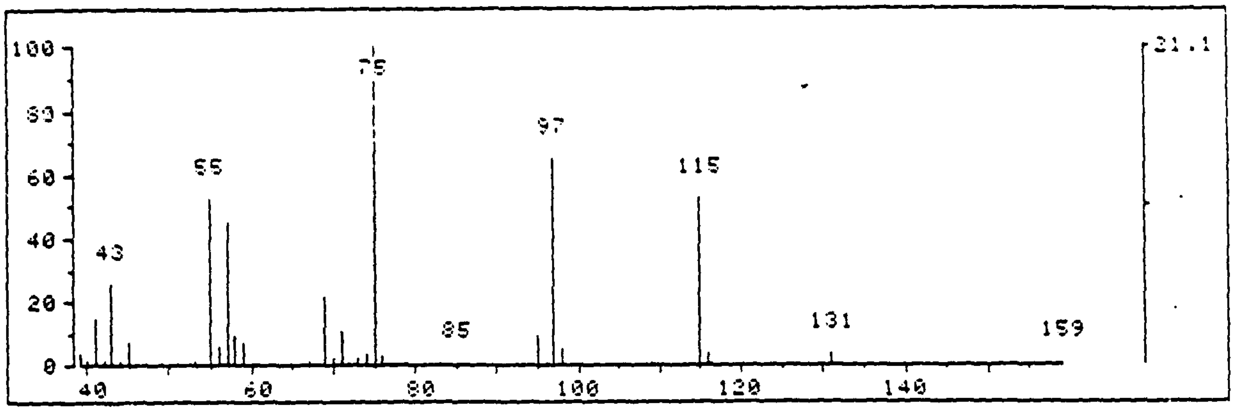
>PAUSE

2,5-DIMETHYL-1,2-HEXANEDIOL IN MEOH 23AUG88 FRN 12236, CRN 89
CW-20M 25METER 100 1 5 180 10 1266 SCAN (1266 SCAN), 18.67 MIN)
MAIN RANGE: 32.0, 159.5 TOTAL ABUND= 368115.



SCAN #

577 RET. TIME: 9.00 TOT ABUND= 9689. BASE PK ABUND: 75.3 2048.

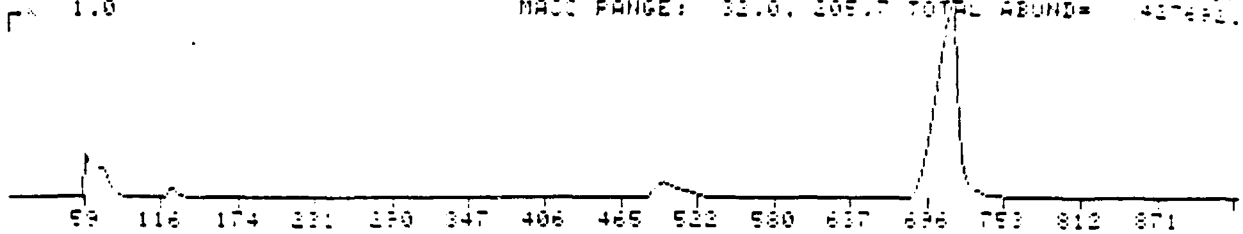


FRN 12236, SPECTRUM # 577 RET.TIME: 9.00, 41 PEAKS

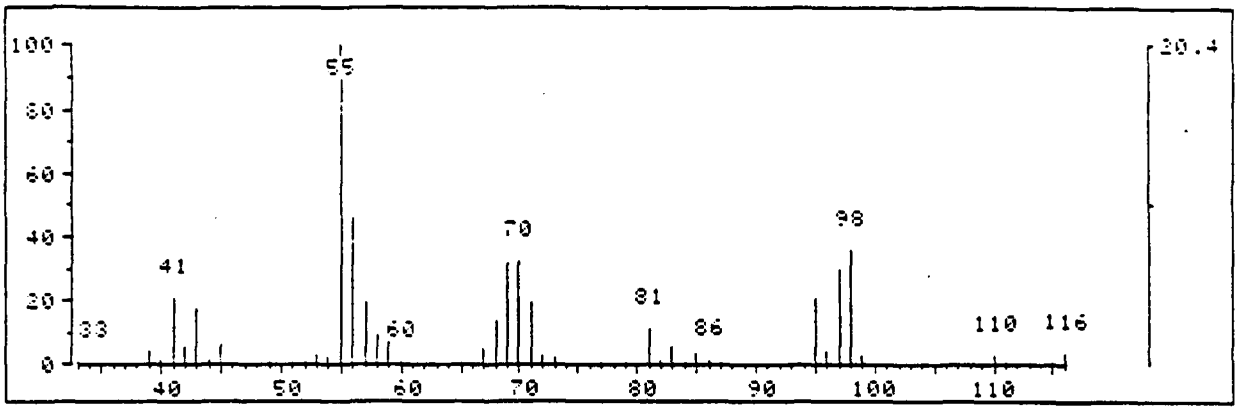
M/Z	REL ABUND	M/Z	REL ABUND	M/Z	REL ABUND	M/Z	REL ABUND
39	4.1	57	45.0	74	4.0	97	64.6
40	2.0	58	8.9	75	100.0	98	4.9
41	14.6	59	7.4	76	3.3	109	.6
42	1.8	60	.6	77	.8	113	.5
43	25.4	65	.5	79	.5	115	52.7
44	1.8	67	1.9	81	.8	116	4.0
45	7.0	69	21.8	83	.5	131	3.9
49	.7	70	2.5	85	1.0	159	.5
53	1.7	71	10.4	93	.6		
55	52.2	72	1.3	95	9.0		
56	5.8	73	2.6	96	.7		

>PAUSE

3,5-DIMETHYLHEXANE-1,6-DIOL IN MEOH 30AUG88 FRN 12239, CRN 59
CW-20M 25METER 100 1 5 180 20 931 SCANS 1 931 SCANS, 17.88 MINS
1.0 MASS RANGE: 33.0, 205.7 TOTAL ABUND= 427892.



713 RET. TIME: 15.17 TOT ABUND= 14309. BASE PK ABUND: 55.3 2913.

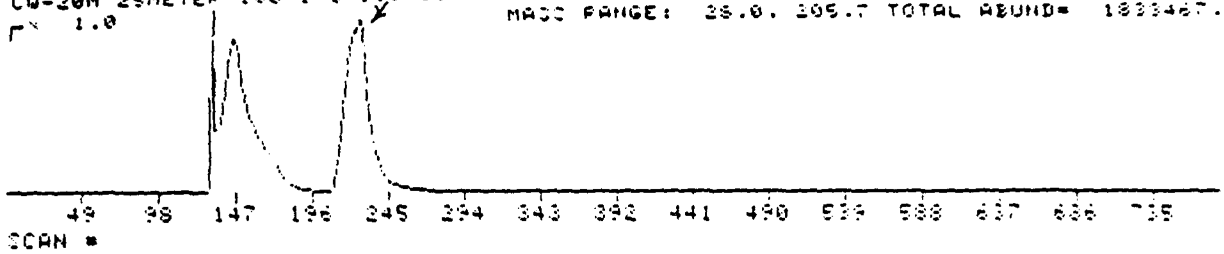


FRN 12239, SPECTRUM # 713 RET.TIME: 15.17, 48 PEAKS

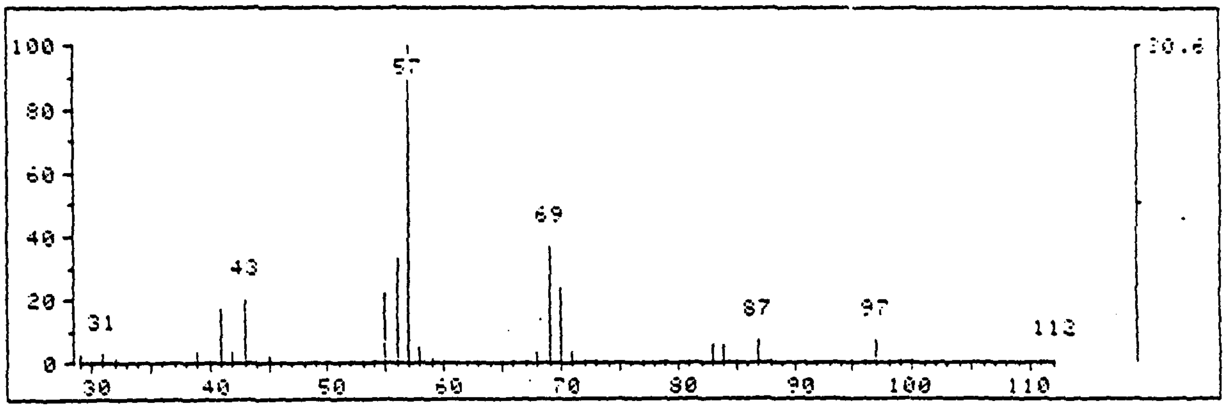
M/Z	REL ABUND	M/Z	REL ABUND	M/Z	REL ABUND	M/Z	REL ABUND
33	.3	54	2.6	71	19.3	93	.5
39	4.5	55	100.0	72	3.1	95	21.1
40	1.6	56	46.1	73	2.3	96	4.5
41	20.9	57	19.7	77	.7	97	29.6
42	6.0	58	9.5	79	1.3	98	36.1
43	17.5	59	8.9	81	11.0	99	3.1
44	1.4	60	.4	82	1.9	109	.9
45	6.7	65	.9	83	5.5	110	3.4
49	.5	67	5.2	84	1.1	111	.5
51	.5	68	14.2	85	3.9	113	.4
52	.4	69	31.5	86	1.6	115	1.2
53	2.9	70	32.6	87	.7	116	3.5

>PAUSE

2,5-DIMETHYL-1-HEXANOL IN MEOH 23AUG88 ??? SCANS (??? SCANS, 11.60 MIN) FRN 12235.
CW-20M 25METER 100 1 5 150 10 MASS RANGE: 25.0, 205.7 TOTAL ABUND= 1833467.



225 RET. TIME: 3.87 TOT ABUND= 40803. BASE PK ABUND: 57.3 12497.



FRN 12235, SPECTRUM # 225 RET.TIME: 3.87, 50 PEAKS

M/Z	REL ABUND	M/Z	REL ABUND	M/Z	REL ABUND	M/Z	REL ABUND
29	3.2	51	.4	67	1.8	85	.8
30	.2	52	.2	68	4.0	87	7.2
31	3.4	53	1.6	69	36.6	88	.4
32	1.5	54	.8	70	23.5	95	.2
38	.1	55	22.3	71	4.0	96	.2
39	3.7	56	33.4	72	.2	97	7.2
40	.9	57	100.0	73	.5	98	1.1
41	17.7	58	5.1	77	.2	99	.9
42	4.0	59	1.8	79	.3	100	.1
43	20.4	60	.1	81	.5	111	.3
44	.9	63	.1	82	.3	112	.6
45	2.4	65	.3	83	6.1		
50	.2	66	.1	84	5.5		

>PAUSE

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FINAL REPORT

Physiological Measures of Workload During Actual Flight

Prepared by:	Christopher Sullivan
Academic Rank:	Graduate Student, M.S.
Department and Universtiy:	Depatment of Psychology Colorado State University
Research Location:	Armstrong Aerospace Medical Research Laboratory Workload and Ergonomics
USAF Researcher:	Glenn F. Wilson, Ph.D.
Date:	September 19, 1988
Contract No:	F49620-88-C-0053

Physiological Measures of Workload

During Actual Flight

by

Christopher Sullivan

Abstract

Five physiological measures of workload--heart rate, respiration, eye-blink, electroencaphalogram and event-related brain potential--were recorded from ten experienced F4 attack aircraft crews. Each crew flew the same air-to-ground pratice maneuver in the wing position of either a two-ship or four-ship formation. Each mission was comprised of low level terrain following, high G maneuvers, bombing runs, a high altitude cruise to base, and a formation approach ending in single-ship landings. A preliminary analysis of the data indicates that ERPs can be a useful measure of workload in this exceedingly complex and noisy enviroment. Preliminary analysises on the remaining measures have not been completed.

Acknowledgements

I wish to thank the Air Force Office of Scientific Research for sponsoring me through GSRP. I would also like to thank Universal Energy Systems for their help with administrative details.

My experience at AAMRL/HEG was not only intellectually enriching but also personally rewarding and for this many bright and warm people were responsible. Iris Davis and Penny Fullenkamp were great friends and exceptional coworkers. I am also greatful to Celia, Carolyn, Rod, Gloria, Claude, Cathy, Merry, Barb, Ann, Al, Paul, and Alex. Finally I wish to thank especially Dr. Glenn Wilson for his help, guidance, and friendship.

I. INTRODUCTION:

The Workload and Ergonomics group of AAMRL/HEG is concerned primarily with the psychophysiological measurement of workload. Since 1979 this group has undertaken the task of developing a psychophysiological battery of workoad tests. The product of this effort is the Neuropsychological Workload Test Battery (NWTB). In addition, Glenn Wilson and his colleagues have conducted a number of studies demonstrating the usefulness of physiological measures of workload in highly complex real world settings such as the cockpit of highspeed aircraft. In their most recent effort, Glenn and his colleagues are recording five physiological measures of workload from ten experienced F4 attack aircraft crews while they are engaged in an air-to-ground practice maneuver.

My research interests have been in the area of learning and memory and more recently, in the area of the event-related brain potential as a measure of attention and workload. Also, I am interested in control-engineering models of manual control and workload. These interests contributed to my assignment to the F4 study.

II. OBJECTIVES OF THE RESEARCH EFFORT:

The importance of the explication of pilot/crew workload in the cockpit of highspeed aircraft have been underscored in recent years by a number of studies that have demonstrated differences in the workload of simulated and actual flight (see Wilson, 1987). Emerging as a group of promising measures of pilot/crew workload are physiological measures. Although some physiological measures--heart rate, eye-blink, respiration, electroencephalogram (EEG) and event-related brain potential (ERP)--have been shown to be valid and reliable measures of workload in the laboratory and flight simulator, an adequate data base for recording these measures when the pilot and/or crew are actually flying an aircraft does not exist. In fact the ERP has not yet been recorded during flight. Therefore the present effort was designed to increase the data base on physiological measures of workload recorded in flight. More specifically five measures--heart rate, respiration, eye-blink, EEG and ERP--were recorded from ten experienced F4 attack aircraft crews while they were flying an air-to-ground practice maneuver.

Given the scope of this project, my assignment as a participant in the 1988 Graduate Student Research Project (GSRP) was to oversee the recording and analysis of the ERP data. In addition I assisted in the training and instrumenting of the F4 crews. The body of this report will be concerned with the ERP as a measure of attention and workload.

III. GOALS ACCOMPLISHED

a. Background

In addition to heart-rate, eye-blink, respiration, and EEG, another promising measure of workload is the ERP. The ERP is a transient series of voltage fluctuations that can be recorded from the scalp in response to the presentation of discrete stimuli (Donchin, 1975). This time-locking of the ERP to the eliciting stimulus is what differentiates ERPs from the ongoing EEG (Kramer, Sirevaag & Braune, 1987). In short, the ERP represents the processing of a highly specific event.

In recent years an extensive series of investigations have demonstrated the utility of the P300 component of the ERP complex as a measure of perceptual-central workload (for a brief review see Donchin, Kramer & Wickens, 1986). In this type of experiment the subject is assigned two tasks, a primary task and a secondary task. The primary task is performed as well as possible whereas the secondary task (an auditory oddball task) is performed only to the extent that primary-task performance remains stable (Donchin, Kramer and Wickens, 1986). If the primary task is difficult, or in other words, if the primary task workload is high, it will require maximal processing resources for stable performance. Minimal resources, only, will remain available for performing the secondary task. In short changes in the demand or workload of the primary task will be reflected in the amplitude of the P300. One advantage of this technique is that the stimulus eliciting the ERP can be coupled with a wide variety of primary tasks (e.g. perceptual monitoring, manual tracking, mathematical processing, etc.). In addition since the response is covert (target stimuli are counted), the ERP measure, unlike other secondary task measures requiring overt responses, is non-intrusive (Wickens, 1979).

b. Instrumentation and Procedure

Each crew consisted of the pilot and the Weapons System Officer (WSO). The pilot was instrumented with a Del Mar Avionics ambulatory phsyiological recorder and an AIWA am/fm cassette stereo. The AIWA recorder presented a three minute segment of tones to the pilot during the low level terrain following and the high altitude cruise to base. In addition, it sent a trigger pulse 150 milliseconds prior to the tone, to the physiological recorder so that the pilot's processing of the tone could be extracted from the ongoing EEG. Both instruments were modified to meet the needs of the study. The WSO was instrumented also with a Del Mar Avionics ambulatory physiological recorder, an AIWA am/fm stereo cassette recorder and, in addition, an accelerometer. The AIWA stereo was modified to record cockpit communications and keep a precise time record during the mission. In this way segments of the mission (for example, low level terrain following or a particular type of bomb drop) could be correlated precisely with the physiological data.

Each crew was instrumented two hours prior to their pre-flight briefing. During the latter part of this period baseline data were collected. Each baseline task had either a low or a high level of workload. For the pilot there were five baseline tasks--an auditory oddball task, a tracking task with two levels of difficulty, and an auditory oddball task combined with each level of tracking. The WSO performed twice at each level the tracking task only. In addition to the baseline tasks, for each crew member data that might potentially contanimate the raw EEG (i.e. muscle movement due to moving the eyes, jaw, head, etc.) were recorded.

Following the baseline tasks there was a half-hour briefing in which the goals of the mission were discussed. Each crew flew the same air-to-ground mission in the wing position of either a two-ship or four-ship formation. Each mission was comprised of low level terrain following, high G maneuvers, bombing runs, a high altitude cruise to base, and a formation approach ending in single-ship landings.

During those segments of the mission in which ERP data were collected, the primary task was considered to be that complex of activities engaged to successfully accomplish that leg of the mission. Therefore component ERP amplitudes were considered a measure of workload for that segment of the mission.

c. Problems Encountered During the In-Flight Recording of the ERPs

In the area of pilot workload, ERPs have not yet been recorded in flight. In addition to the exorbitant cost of flight time, of central importance here, is the high level of noise in the cockpit. Exceedingly high electromagnetic activity as well as artifact due to muscle movement and cockpit communications both greatly reduce the signal-to-noise ratio of the biological signal of interest.

Fortunately, the Workload and Ergonomics group at AAMRL/HEG is equipped with excellent technical support. Based upon a previous study of in-flight pilot workload conducted by Glenn Wilson, it was decided that the signal-to-noise ratio could be improved by electrically shielding all scalp and heart electrodes, and by placing scalp electrodes in a bipolar arrangement.

An additional problem we faced in the F4 study was time-locking the ERP to the eliciting stimulus. This was a formidable problem given that a number of different measures of workload were being investigated, and given that each crew member could carry safely only two to three peices of equipment.

d. Results

At this time data has been collected on five crews. Although a complete analysis of this data is far from completion, a preliminary analysis of the data indicates that the noise reduction and time-locking techniques were quite successful. In addition, a preliminary analysis of the peak and latency of various ERP components indicates that ERPs can be a useful measure of workload in the cockpit. Particularly promising candidate measures of workload are the P300, and more surprisingly, the N100.

The bipolar arrangement of the scalp electrodes proposed an interesing problem to the analysis of the data. You will recall that such an arrangement of the electrodes was required to increase the signal-to-noise ratio. Since, however, the P300 is an ubiquitously occurring potential, systematic influences on this component are erased due to the arrangment. Therefore a monopolar electrode arrangement with the source at Cz was derived from the addition of two of the bipolar arrangements, Cz-Fz and Cz-Pz.

IV. RECOMMENDATIONS

a. Implications of Results

The use of the event-related brain potential as a measure of resource allocation and workload has several implications for the study of pilot state. Traditionally, the analysis of pilot/crew performance has been conducted by detailing observable aspects of tasks in the flight simulator and in the laboratory. Although these procedures provide an accurate description of the behavior exhibited, it has been shown that the workload during flight is greater than that in either the simulator or laboatory. Given the increasing complexity of aircraft cockpits, it is important that we be able to accurately assess workload as it pertains to successful mission accomplishment (Wilson, 1987). Since, ERPs as well as other physiological measures of workload, can be employed successfully in the cockpit, these measures should prove very helpful in assessing operator state. In addition, it is often useful to pinpoint the source of increases in workload. Most tasks in the cockpit are multi-dimensional requiring the pilot and crew to attend to a number of modalities of information while implementing one or more responses. Increases in workload might be a function of one or two of these components only. Since the P300 is sensitive to perceptual-central processing it should contribute to componential analysis of the workload of multidimensional tasks.

b. Future Research

In addition to the P300, the N100 was reliably recorded from each pilot. Previous research has demonstrated that the N100 indexes the processing distribution of resources during high load conditions. However, unlike the P300, the specific resource "pools" to which the N100 is sensitive are not known. Identifying those unknown resources should provide researchers with a measure that would be very useful in the microanalysis of pilot workload. A mini-grant proposal to evaluate the ne difference" ERP (which includes N100) as a secondary task measure of resource allocation and workload has been submitted to UES.

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FINAL REPORT

Ground-Texture Information for Aimpoint Estimation

Prepared by:	Lisa F. Weinstein
Academic Rank:	Doctoral candidate
Department and University:	Psychology Department University of Illinois
Research Location:	USAF-AAMRL/HEF Wright-Patterson AFB Dayton, Ohio 45433
USAF Researcher:	Richard Warren, Ph.D.
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Ground-Texture Information for Aimpoint Estimation

by

Lisa F. Weinstein

ABSTRACT

This experiment examined the effects of ground texture on aimpoint estimation in a landing-judgment task. Subjects viewed a simulated landing approach which stopped "in midair". Subjects were asked to estimate the point at which they would land if they continued on the current path. Results indicate that structured texture, and expanding texture elements improved performance. Furthermore, with dot patterns, performance was better when the aimpoint was represented by a specific point on the display.

Acknowledgments

I wish to thank the Air Force Systems Command and the Air Force Office of Scientific Research for sponsorship of this program. In addition I would like to thank the Armstrong Aeromedical Research Laboratory for allowing me to spend the summer at their facility. Also, I would like to acknowledge the concern and assistance in administrative matters shown to me by Universal Energy Systems.

My sincere thanks are extended to Dr. Rik Warren for providing me guidance, encouragement, and a truly enjoyable work experience. Also, I wish to thank Bob Todd for his time and effort programming the experimental displays. Finally, I would like to express deep thanks to Dr. Lawrence Wolpert and Kim Reardon for their endless assistance with data analysis and interpretation.

I. INTRODUCTION:

Determining what visual information a pilot uses when landing an aircraft has implications for the design of computer-generated visual scenes for flight simulators.

The PACLAF (Perception & Control in Low Altitude Flight) Laboratory of AAMRL (Armstrong Aerospace Medical Research Laboratory/HEF) is interested in examining the types of information a pilot might use for the perception and control of an aircraft during low-level flight. The research includes exploring the optical bases for self-motion and the importance of scene detail. In particular, experiments have examined the perception of self-motion and scene detail in a simulated approach to landing. PACLAF research attempts to identify visual cues for a pilot, make recommendations, and convey findings to the operational crewstation design and simulator communities.

My research interests are in the area of visual perception and the application of theoretical knowledge to the design of visual displays for flight simulators. I have conducted experiments examining the role of ground texture in the control of altitude during low-level flight. My interest is in isolating the relative contributions of various textures to altitude control. As a result of my interest in low-level flight and simulation, I was assigned to the PACLAF laboratory at AAMRL.

II. OBJECTIVES OF THE RESEARCH EFFORT:

With the rapid increase in computer-graphics technology, it is tempting to create as realistic a visual scene as possible in flight simulators. However, there is some evidence that maximum fidelity is not desirable (Warren & Riccio, 1985).

In addition, when attempting to land a flight simulator, pilots tend to land short of their estimated landing point and at higher velocities (Armstrong, 1968; Buckland, Monroe & Mehrer, 1980). Therefore, it is desirable to determine what visual information a pilot uses when flying and landing an aircraft in order to design effective flight simulators with optimal computer-generated visual scenes.

My primary objective as a participant in the 1988 Graduate Student Summer Research Program was to explore the various types of visual information a pilot might use when landing an aircraft. In particular, I was interested in examining the relative contributions of several types of visual information in the determination of the point at which a pilot would contact the ground (his aimpoint).

III. THE EXPERIMENT:

A. Introduction

When designing flight simulators, designers must determine what information to present in a visual display. The trend has been to provide the most information available given the current technology. This approach to design is not only expensive, but may not be effective. As mentioned earlier, Warren and Riccio (1985) suggest that maximum fidelity does not always mean maximum efficiency. Therefore, it is important to determine what information a pilot needs in order to successfully land an aircraft, and provide designers with that information.

The point to which a plane is heading is known as the aimpoint. In terms of the optic array, this point has been called the focus of expansion (Langewiesche, 1944; Gibson, 1947), and is defined as the point that optically remains stationary while all other points radially expand outward from the direction in which one is moving. But it is an empirical question as to which type of information is best for detecting the focus of expansion. Gibson (1966) suggested that the type of information presented (as long as enough points are present to generate a flow pattern) should not affect a pilot's ability to locate the focus of expansion.

It is a well established fact that pilot performance varies under different visual conditions. For example, when viewing night scenes in a simulator pilots trend to land short of their desired aimpoints and at higher velocities (Armstrong, 1968; Buckland, Monroe, & Mehrer, 1980). Buckland et al. (1980) found that additional ground texture improved performance on a simulated landing task. In addition, Carel (1961) found that subject's performance on a heading-judgment task was more accurate with a grid pattern than with random dots. However, no systematic analysis has been done to determine why additional ground texture is beneficial. Once we determine what aspects of texture are beneficial, designers will have a guide as to what types of visual information will be most effective in a simulator.

The purpose of this experiment was to evaluate the relative contributions of several types of ground texture with a simulated approach-to-landing task. I compared the contribution of structured textures to random textures. The benefit of texture elements which change in perspective (squares) over elements which do not change in perspective (dots) was also examined. Additional benefits from having the aimpoint explicit on the display were explored as well.

It was hypothesized that the addition of structure, as Carel found, and the presence of expanding objects would improve aimpoint estimation. Furthermore, having the aimpoint explicit on the display should also improve performance.

B. Method

Task. In all conditions, subjects viewed a simulated landing approach to the ground. The display froze "in midair" after 10 seconds. The subject's task was to estimate the point at which they would contact the ground if they continued on the same path.

Subjects. Twelve paid volunteers participated in the experiment. Subjects were recruited from the local area, and the experiment was run at Wright-Patterson AFB Armstrong Aeromedical Research Laboratory. All had normal or corrected to normal vision. None had previous piloting experience.

Apparatus. The experimental displays were presented on a Silicon Graphics 3020 IRIS computer system. A high-resolution (1024 by 768 pixels) color monitor updating at 20 frames/s presented the displays. Subjects were seated 63.5 cm from the monitor, resulting in a 31 x 24 degree field of view. The experiment was conducted in a darkened booth, and subjects viewed the displays directly. Subjects entered their responses by using a three-button mouse.

Design. Each subject participated in two 1-hour sessions. All subjects experienced the same experimental conditions, presented in different random orders.

There were four displays: a random dot pattern, a structured dot pattern, a checkerboard pattern, and a pattern of random squares. Both the checkerboard pattern and the random squares expanded when approached.

The simulated flights involved approaching the ground at four path angles (5, 9, 12, & 14 deg). In addition, on half of the trials the aimpoint was represented as an actual point on the display. For example, in the "point present" condition with the dot displays, a dot would have been located at the actual aimpoint while in the checkerboard condition, the aimpoint appeared as the center of a square.

All subjects received all possible combinations of these factors in random order. Each subject received four replications of each display over the two-day testing period (2 on Day 1 and 2 on Day 2).

Therefore, the design was a 4 (path angles) x 4 (displays) x 2 (point explicit/not explicit) x 4 (replications) within-subjects design. Displays were further decomposable as a 2 (dots vs checkerboard) x 2 (structured vs random).

Procedure. During each session, subjects received 10 trials of training to familiarize themselves with the task. Feedback was presented to the subjects only during these familiarization trials. After the subjects were familiar with the task, they performed 64 experimental trials each day.

After 10 sec the display froze 50 ft above the ground, and an arrow would appear in the center of the display. The arrow could be manipulated in depth on the display by pressing the mouse buttons. The task was to move the arrow on the display to the perceived aimpoint.

The top button moved the arrow up the screen, or away from the subject in depth. The bottom button moved the arrow down on the screen, or closer to the subject in depth. The top button moved the arrow up at a fast rate, while the bottom button moved the arrow down at a slower rate. This enabled the subjects to fine tune their responses.

Once the arrow was positioned at the estimated aimpoint, the subject pressed the center mouse button twice to continue to the next trial.

C. Results

Repeated-measures analyses of variance were performed on four dependent measures: absolute angle error, absolute distance error, signed angle error, and signed distance error. Absolute error measures are reported to show an estimate of global error, while signed errors indicate the direction of error (i.e., overshoot or undershoot of the aimpoint). Errors are presented in both distance (ft) and angle (deg) measures. All four ANOVAs yielded significant results (see Table 1).

Absolute angle error

Subjects tended to underestimate their aimpoint at the 5 and 9-deg path angles, and overestimate their aimpoint at the 12 and 14-deg angles. The pattern of results indicates a tendency to point toward the center of the visible ground. This pattern was apparent in all four error measures and will therefore only be reported here.

A significant interaction was found between day and type of display. Over time, performance improved with the dot displays, while there was no change in performance with the checkerboard displays (see Figure 1). In addition, initial performance was better with the checkerboard than with the dots. A similar pattern of results was obtained between day and type of texture. Over time, performance improved with the random textures, while there was almost no change in performance with the structured displays (see Figure 2). Furthermore, initial performance was superior with the structured textures.

The path angle by point-explicit interaction was also significant with judgments improving at steeper path angles with the point explicit. At the 5 and 9-deg angles, there was no change in performance when the point was explicit.

The point explicit by display type interaction revealed that performance was significantly better with the dot displays when the point was explicit, while having the point explicit did not improve performance on the checkerboard displays (see Figure 3).

There was also a three-way interaction among path angle, texture type and display type. However, an examination of the means revealed no logical pattern.

Absolute distance error

Subjects improved their performance from the first session (mean error= 100 ft) to the second session (mean error = 63 ft). In addition there was a significant day by display type interaction. The pattern was the same as for absolute angle error (see Figure 1).

Signed angle error

The main effect of texture type was significant. Performance was better with the structured (mean error = .42 deg) than with the random texture (mean error = .71 deg). There was also a main effect of display type. Performance with checkerboards (mean error = .15 deg) was better than with dots (mean error = .99 deg).

There was a significant interaction between the path angle and the point explicit. It appears that with the exception of the 9-deg path angle, having the point explicit improved performance.

The three-way interaction among point explicit, texture type and display type was also significant. Performance with the point explicit was better only for the random dot display.

There was also a three-way interaction among path angle, texture type and display type. As with absolute angle error, an examination of the means revealed no logical pattern.

Signed distance error

As in the signed angle error, the main effect of texture type was significant. Performance was better with the structured texture (mean error = -6 ft) than with the random texture (mean error = 14 ft). There was also a main effect of display type. Performance with checkerboards (mean error = -16 ft) was better than with dots (mean error = 24 ft).

The day by display type interaction was also significant. The pattern is similar to that shown in Figure 1.

Finally, there was a day by path angle interaction. Although performance improved at each path angle over time, the improvement at the 12 deg angle was approximately twice the change of that at the other angles.

D. Discussion

In general, subjects were surprisingly accurate with errors, i.e., on the order of 1 to 3 deg. These results indicate more accurate performance than has been found in similar studies (Llewellyn, 1971; Johnston, White, & Cumming, 1973).

Results support the hypothesis that structured texture and expanding objects improve initial performance on an aimpoint estimation task. This result supports Carel's (1961) finding that subjects performed a heading-judgment task more accurately with a structured grid pattern than with a random dot pattern.

However, these findings seem to contradict Gibson's (1966) assertion that the type of texture elements in a display should not affect a person's ability to locate the focus of expansion. I suggest that while the information in the optic array may be sufficient for the detection of the aimpoint, the addition of structure and expanding texture elements enhances the ability to detect the focus of expansion or aimpoint.

In addition, it appears that when viewing dot displays, performance is improved when the aimpoint is co-incident with a dot in the display. This improvement in performance may be due to the increased ease of seeing a dot which remains stationary as opposed to locating a stationary point on a non-textured background.

The significant effect of path angle appears to be due to central tendency in which subjects tend to point toward the center of the visible ground. Reardon (1988) noticed this trend in a similar study, but found that training helped to minimize this tendency. Future research on aimpoint estimation should employ training to minimize this effect.

The training effects found with dot patterns raise questions concerning the use of dot patterns in aimpoint studies. If there is a training effect, we must be careful to achieve stable performance before collecting measurements. Furthermore, the reason for differential learning effects between displays still needs to be pursued.

Table 1: Results from the four analyses of variance.
Row 1 = F values
Row 2 = Percent of variance accounted for

	Absolute Distance Error	Absolute Angle Error	Signed Distance Error	Signed Angle Error
Path Angle	13.54** 2.60	15.69** 4.80	38.12** 29.50	28.41** 13.20
Day	18.14** .93			
Type			6.19* 1.80	5.18* .93
Texture			14.31** .22	5.11* .24
Day by Path Angle				5.12** .76
Day by Texture		4.81* .13		
Day by Type	5.64* .27	6.83* .32		4.81* .29
Path Angle by Point Exp.		3.40* .36	3.19* .13	
Type by Point Exp.		8.63** .30		
Path Angle by Type by Texture		3.37* .41	3.47* .34	
Point Exp. by Type by Texture			7.76* 24	

* = p < .05
** = p < .01

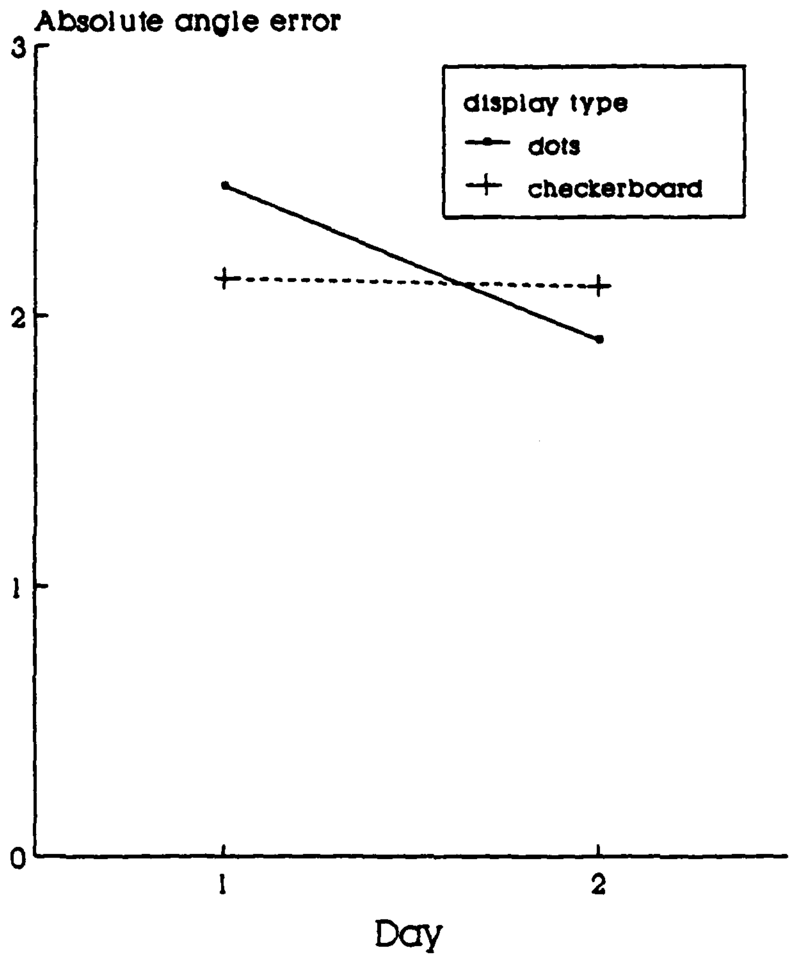

Figure 1: Day by type of display interaction

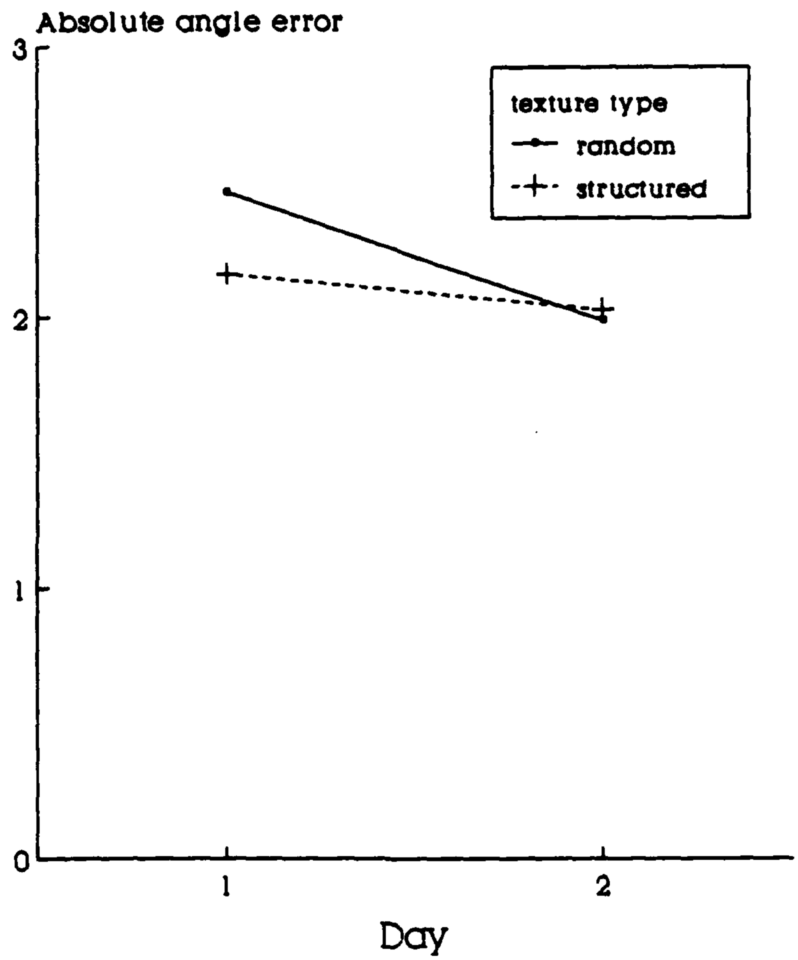

Figure 2: Day by type of texture interaction

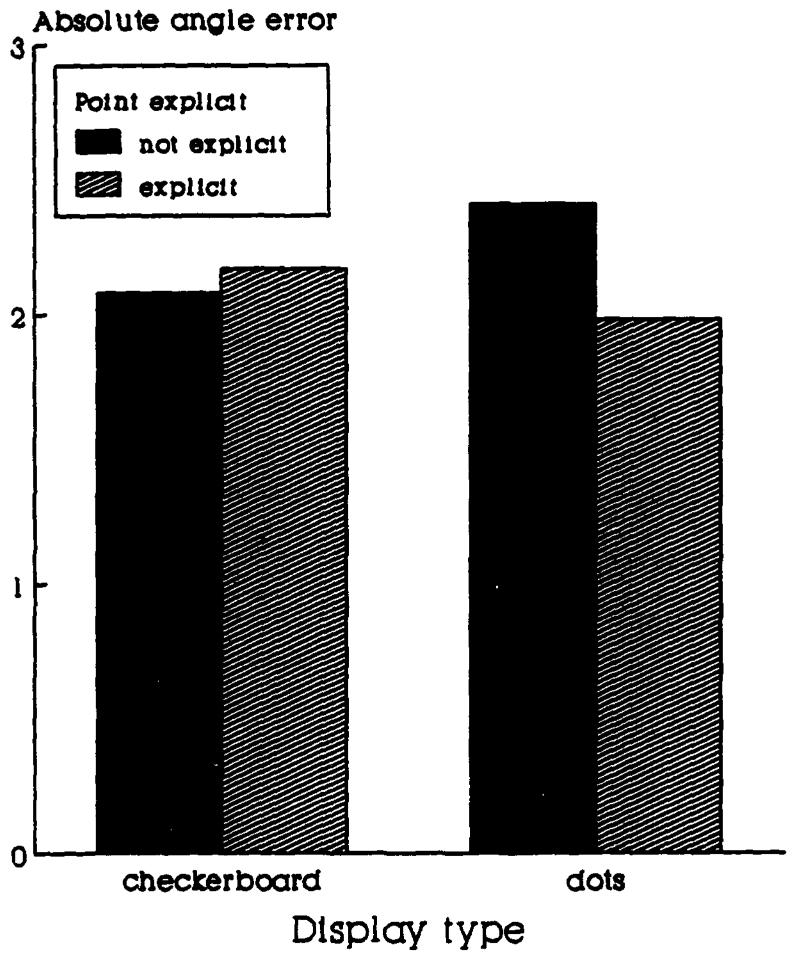

Figure 3: Display type by point explicit /not explicit interaction

IV. RECOMMENDATIONS:

From these results it is hard to recommend design guidelines for visual displays in simulators. An active flight task may require different visual information than a passive landing-judgment task. Therefore, I suggest that a similar study be conducted using active control.

In addition, since naive subjects were used in this study, a replication might be performed using experienced pilots. It is important to determine if the visual information used by novices is the same as that used by experts.

As mentioned earlier, it appears that subjects were biased by a central tendency effect in the current study. There is some evidence that training can minimize the effects of central tendency in a landing-judgment task. As a result, if a similar task is used in the future sufficient training should be included to minimize central tendency effects. The data should also be examined for other indications of training effects.

The current study helped to answer some of the preliminary questions concerning visual information in landing. More research is necessary to examine the many issues still unresolved. Once a coherent set of results is obtained from a program of research, a set of design guidelines can be provided to simulator designers.

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Prepared By: Robert K. Young, Ph.D. and John Allison

Academic Rank: Professor / Graduate Research Assistant

Department and University: Psychology, University of Texas at Austin

Research Location: Human Resources Laboratory/MOEL
Brooks AFB, TX 78235

USAF Researcher: Scott R. Chaiken

Date: 12 August 1988

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Prepared by:	L. Alan Witt, Ph.D.
Academic Rank:	Assistant Professor
Department and University:	Department of Psychology Western Illinois University
and:	Mark N. Beorkrem
Academic Rank:	Graduate Student
Department and University:	Department of Psychology Washington University (St. Louis)
Research Location:	Human Resources Laboratory/Operations Training Division, Williams Air Force Base, Arizona
USAF Researcher:	Elizabeth L. Martin, Ph.D.

Date:	26 August 1988
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Prepared by: Sunita S. Rana, Ph. D.

Charles Drake (Graduate Student)

Academic Rank: Assistant Professor

Department and University: Computer Science, Jackson State University

Research Location: Human Resources Laboratory, Brooks AFB, Texas 78235-5601

USAF Researcher: Dr Kurt Steuck

Date: 9 Aug 88

Contract No: F 49620-87-R-0004

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PROF. SUNITA RANA
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FINAL REPORT

Prepared by: Peter Gaddis, Jr.

Academic Rank: Graduate Student

Department: Sociology

University: Jackson State University

Research Location: Brooks Air Force Base
AFHRL San Antonio,
Texas 78235-5601

USAF Researcher: Dr. William E. Alley

Date: September 14, 1988

Contract No: F49620-88-C-0053

Underlying Distributions of The New PACE Variables

by

Peter Gaddis, Jr.

ABSTRACT

This study investigated the underlying distributions of the variables in the payoff algorithm in the new PACE system. The results from the goodness-of-fits test indicated that only one variable- "objective interest", seems to follow a normal distribution in all three batches of recruits included in the analysis. Some of the other variables were found to follow a normal distribution in one batch and a non-normal distribution in the remaining batches of recruits. To assess interaction between job properties and individual characteristics in the new PACE system, intracorrelations among payoffs of various jobs in the Adiministrative area of a week group were computed for a particular group of the Air Force trainees. The payoffs of only one out of eight jobs in the Administrative area of a week group were negatively correlated. In addition, data analysis involving application of general linear model was performed to assist Dr. Rana in analyzing interaction in the new PACE system.

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<u>Underlying Distributions of The New PACE Variables</u>

I. INTRODUCTION

To classify enlisted personnel, the Air Force of the United States of America employs two computerized systems: a) Processing and Classification of Enlistees (PACE), and b) Procurement and Management Information System (PROMIS). PACE is a post-enlistment system managed by the Headquarters, Air Training command while the PROMIS is a pre-enlistment system managed by the USAF Recruiting Service. Every year, these two systems classify approximately fifty-six thousands recruits into one of nearly three-hundred jobs. After the PROMIS classifies fifty percent of the recurits through the Guaranteed Training Enlistment Program (GTEP) and the remaining fifty percent into four aptitude areas- Mechanical (M), Administrative (A), General (G), and Electronics (E), the trainees are finally assigned to specific Air Force jobs by the PACE system.

My research interests have been in the area of the behavioral sciences which includes both psychology and sociology. As an undergraduate, my research efforts stemmed from a psychological perspective. During that time, I participated in the Minority Access To Research Careers Training Program which layed the foundation of my research background. It introduced me to the fundamentals of research by giving me the opportunity to collect and analyze data, present and publish papers.

During my tenure as a graduate student, I participated in the Minority Institute Research Training Program. It helped me to expand my research interests and skills. I believe that my participation in the Minority Research Training Programs contributed to my selection by the Air Force Office of Scientific Research. My GRSP assignment to work on the person-job match project provides me with another opportunity to develop my research skills.

II. OBJECTIVES OF THE RESEARCH EFFORT

All the classification systems of the Armed Forces including The PROMIS and the new PACE of the Air Force are based on the assumption that the job properties interact substantially with personal characteristics of the recruits. A major challenge facing the researchers at the Air Force Human Resources Laboratory (AFHRL), is: How to maximize total payoff for the entire batch of recruits by manipulating interaction between the above two factors? The literature search indicates that the above problem has not been studied by the researchers. To provide some insight in the general problem of maximizing total payoffs, the following objectives were formulated jointly with Dr. Rana:

1. This study was designed to investigate the underlying distributions of the variables of payoff algorithm in the new PACE with particular emphasis on the "final payoff". To determine if the new PACE is more effective than the current PACE, the two systems should be compared in terms of their variables. This comparison involves testing of hypotheses on the parameters of

the distributions of the PACE variables. Thus, to compare the two classification systems, knowledge of the underlying distributions of the PACE variables is required.

2. The second objective of the study was to compute intra-correlations among final payoffs of each possible person-job match. The examination of the intra correlations will provide information which can be used to assess potential for increasing the total payoff by manipulating the interaction between job properties and personal characteristics.

III. BACKGROUND

The review of literature shows that there are no studies devoted to the investigation of underlying distributions of the PACE variables or to the examination intracorrelations among final payoffs of different AFSs. However, there are many studies addressing the development of the processing and classification systems of the Air Force.

Since this study focused on the underlying distributions of the variables and intracorrelations among final payoffs of different AFSs in the PACE system, a brief explanation may be useful here. The current PACE system is a batch process which classifies recruits in the Air Force every week. First, PROMIS classifies 50% of the recruits into four aptitude areas (M, A. G, or E) according to their Armed Services Vocational Aptitude Battery (ASVAB) composite score. For details of ASVAB see Ree, Welsh, Wegner, and Earles (1985). The recruits in these AFS areas are assigned to specific jobs by the PACE system. PACE uses three input data files - a trainee file, a quota file and an AFS prerequisite file. These three files are finally reduced to a qualification file which determines for each trainee in the week group all the AFS for which the trainee qualifies. A "sort" subroutine sorts the available jobs by priority and the qualified people by preference and aptitude, and then makes assignment from top down. The current PACE does not optimally assign recruits to AFSs and is largely driven by short-term priorities.

A major recent advance in classification techniques is the development of Payoff Algorithm (Pina, Emerson, Leighton and Cummings, 1988). Implementing the payoff algorithm within the current PACE results in the new PACE system. The Payoff algorithm is a mathematical model which uses information about the individual and the Air Force Speciality to generate a payoff. A payoff value represents the worth to the Air Force of assigning a particular recruit to a specific job. The variables in the payoff algorithm are selected to represent 10 fundamental concepts. Some of these variables are based on the findings of earlier studies. For example, job difficulty measures were developed according to task analysis (Weeks 1984). Intellectual ability variable was based on the results of Wilbourn, Valentine and Ree (1984). For details of the variable representing probability of completing first term, see Emerson and Pina (1987) and Finstuen and Alley (1983). Full discussion of all the variables and functions of the payoff algorithm is given in Pina, Emerson, Leighton, and Cummings (1988).

IV. RESEARCH DESIGN

Method

1. Sample Organization: The sample used in this study consisted of three conviently chosen week groups of Air Force trainees. These weekly classifications of enlisted personnel contained data on a large number of variables including the functions and variables of the payoff algorithm.

2. Procedure: The classification data for Administrative area of weekly batch PPJM.51 was used to investigate the distributions of PACE variables and compute intracorrelations among the final payoffs of different AFSs. To study the underlying distribution of the PACE variables, goodness-of-fit tests were performed by using SPSS/PC+ subroutines. Intracorrelations among the final payoffs were also computed by applying the correlations subprogram of the SPSS/PC+ software.

V. RESULTS

The Kolmogrov-Smirnov test was applied to the classification data of the Administrative area of PPJM.50, PPJM.51 and PPJM.52. The results are arranged in table 4.1 (Appendix A). As shown in table 4.1, the majority of the PACE variables (aptitude vs. job difficulty 2, effectiveness, return on investment, job fill economy, efficiency, (100-effective wt.) X efficiency, person job match, final payoff, academic background, restricted interest, probability of completing term, casual time, and fill priority) follow non-normal distributions in all three groups of recruit . It is clear from this table that only "objective interest" seems to follow a normal distribution in all three batches of recruits. Ability and effictive wt. X effectiveness followed the normal distribution in the PPJM.50 and PPJM.52 batches but not in the PPJM.51 batch. Aptitude vs. job difficulty, aptitude/job difficulty, and interest followed normal distributions in PPJM.50 but not in the PPJM.51 and PPJM.52 batches. Trainiability follows the normal distribution in the PPJM.51 batch but not in batches PPJM.50 and PPJM.52. The intracorrelations among the "final payoffs" of assigning recruits to different AFSs were computed for the Administrative area of the week group PPJM.51. The correlation matrix is given in table 4.2 (Appendix B).

The results in table 4.2 show that the intra correlations range from - 0.9565 to .9988. This range indicates that there exists a high potential for increasing the "total payoff" by manipulating assignments of recruits to various AFSs. However, in this batch, final payoffs of only one AFS were found to be negatively correlated. This perhaps reflects the practical limitations within the system to increase total payoff. However, to draw any reliable inference, more research is needed.

VI. RECOMMENDATIONS

a. In this current study, the sample for goodness-of-fit tests consisted of only 3 conveniently selected batches. Use a random sample consisting of at least 10 batches. Also obtain estimates of the mean and standard deviation required in the Kolmogrov-Smirnov test (for normal distribution) by using data from many batches.

b Compute intra correlations among final payoffs of different AFSs for all aptitude areas of many week groups. Analyze these intracorrelations carefully before making any inference about the scope of increasing total payoff by manipulating interaction.

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APPENDIX A

TABLE 4.1

KOLMOGROV-SMIRNOV GOODNESS-OF-FIT TEST
(NORMAL DISTRIBUTION)

VARIABLES	PPJM50 2-TAIL PROB.	PPJM51 2-TAIL PROB.	PPJM52 2-TAIL PROB.
APTITUDE VS. JOB DIFICULTY	.295	.000	.033
APTITUDE VS JOB DIFFICULTY 2	.000	.000	.000
APTITUDE /JOB DIFFICULTY	.295	.000	.033
TRAINIABILITY	.008	.149	.000
ABILITY	.207	.001	.263
INTEREST	.116	.008	.038
EFFECTIVENESS	.054	.015	.070
RETURN ON INVESTMENT	.000	.000	.000
JOB FILL ECONOMY	.000	.000	.000
EFFICIENCY	.000	.000	.000
EFFECTIVE WT. X EFFECTIVENESS	.170	.000	.240
(100-EFFECTIVE WT.) X EFFIC.	.000	.000	.000
PERSON JOB MATCH	.004	.000	.072
FINAL PAYOFF	.004	.000	.072

* Ho : THE DATA FOLLOW NORMAL DISTRIBUTION

TABLE 41.CONTINUED.

KOLMOGROV-SMIRNOV GOODNESS-OF-FIT TEST
(NORMAL DISTRIBUTION)

VARIABLES	PPJM50 2-TAIL PROB.	PPJM51 2-TAIL PROB.	PPJM52 2-TAIL PROB.
INTELLECTUAL ABILITY	.160	.158	.001
ACADEMIC BACKGROUND	.000	.000	.000
OBJECTIVE INTEREST	.183	.177	.105
RESTRICTED INTEREST	.000	.000	.000
PROB. OF COMP. TERM	.000	.004	.009
CASUAL TIME	.000	.000	.000
FILL PRIORITY	.000	.000	.000

APPENDIX B

TABLE 4.2

INTRACORRELATIONS AMONG FINAL PAYOFFS OF DIFFERENT AFSs.

	JOB1	JOB2	JOB3	JOB4	JOB5	JOB6	JOB7	JOB8
JOB1	1.0000	.9988	.8794	.8785	.3698	.9560	-.8610	.0402
	P=.	P=.000	P=.000	P=.000	P=.005	P=.000	P=.000	P=.770
JOB2	.9988	1.0000	.8812	.8814	.3515	.9508	-.8540	.0350
	P=.000	P=.	P=.000	P=.000	P=.009	P=.000	P=.000	P=.800
JOB3	.8794	.8812	1.0000	.9986	.3426	.8551	-.9565	.0130
	P=.000	P=.000	P=.	P=.000	P=.010	P=.000	P=.000	P=.925
JOB4	.8785	.8814	.9986	1.0000	.3313	. 8531	1.0000	-.0381
	P=.000	P=.000	P=.000	P=.	P=.013	P=.000	P=.000	P=.907
JOB5	.3698	.3515	.3426	.3313	1.0000	.5896	-.3418	.1133
	P=.005	P=.009	P=.010	P=.013	P=.	P=.000	P=.011	P=.410
JOB6	.9560	.9508	.8551	.8531	.5896	1.0000	-.8309	.0656
	P=.000	P=.000	P=.000	P=.000	P=.000	P=.	P=.000	P=.634
JOB7	-.8610	-.8540	-.9565	-.9461	-.3418	-.8309	1.0000	-.0381
	P=.000	P=.000	P=.000	P=.000	P=.011	P=.000	P=.	P=.782
JOB8	.0402	.0350	.0130	.0162	.1133	.0656	-.0381	1.0000
	P=.770	P=.800	P=.925	P=.907	P=.410	P=.634	P=.782	P=.

* P= LEVEL OF SIGNIFICANCE

. LEVEL OF SIGNIFICANCE CAN NOT BE COMPUTED

1988 USAF-UES GRADUATE STUDENT SUMMER RESEARCH PROGRAM

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FINAL REPORT

Development of a General Reliability Simulation Model

Prepared by:	Charles L. King
Academic Rank:	B. S. Industrial Engineering
Department:	Industrial Engineering
University:	University of Arkansas
Research Location:	USAFHRL/LRA
	WPAFB
	Dayton, Ohio 45433
USAF Researcher:	Alan Herner
Date:	30 Sept 88
Contract No:	F49620-88-C-0053

Development of a General Reliability
Simulation Model
by
Charles L. King

ABSTRACT

Recent advances in the capabilities of discrete-event simulation permit the development of a general simulation model to assist design and test engineers in the assessment of reliability. The process-oriented, discrete-event simulation language SIMNET allows the programming flexibility necessary to model any system whose component relationships can be represented by a functional block diagram. Alternative designs are modeled simply, by changing initial data entries to the model. The model was applied to an actual case study: the F-16 voice communications system. F-16 engineers assisted in development of the system functional block diagram and provided actual field reliability data (mean flight hours between failure), for the line replaceable units (LRU's). Upon complete development, this model will interface with an expert system.

ACKNOWLEDGEMENTS

The Fellow would like to express his gratitude to the staff of the Air Force Human Resources Laboratory. Their complete cooperation and willingness to fill his every need during his period of research was most helpful in yielding this final report. Universal Energy Systems is also to be commended for hosting this program, and for all of their cooperation during the research period. The Fellow would also like to express his gratitude to his Summer Faculty Research Advisor, Dr. Tom Landers, whose daily support and encouragement was vital to the Fellow's research efforts.

I. INTRODUCTION

Methods of assessing the maintainability and repairability of a system need to be incorporated into the weapons system development process. The Air Force Human Resources Laboratory is very interested in providing the design/test engineer with a tool to analyze design reliability through the accurate assessment of available data. Simulation provides such a tool. When integrated with an expert system, simulation can be a very effective tool for performing modeling and trade-off studies among design alternatives.

The Fellow's background has included courses in operations research and simulation. Because of experience and personal interest, the Fellow had the opportunity to investigate simulation modeling of reliability and maintenance at the AFHRL/LRA under the 1988 Graduate Student Summer Research Program.

II. OBJECTIVES OF THE RESEARCH EFFORT

In recent years the Air Force has placed a greater emphasis on reliability early in the design process. The necessary theory and tools are not yet available to most design and test engineers, however. There is a need for the development of a tool whereby design and test engineers can make more effective use of available reliability data in the process of creating and evaluating design alternatives.

This Fellow's major professor is Dr. Tom Landers, Assistant Professor, Department of Industrial Engineering, University of Arkansas. He was selected for the Summer Faculty Research Program at the Air Force Human Resources Laboratory, Wright-Patterson AFB. His research goal was the assessment of an expert system approach to provide the engineer with the capability of accurate reliability data analysis. The scope of his research included conceptual design, requirements definition, technology assessment, and initial prototype development, for an expert system approach to failure data analysis (see figure 1).

CONCEPTUAL DESIGN
DATA FLOWS

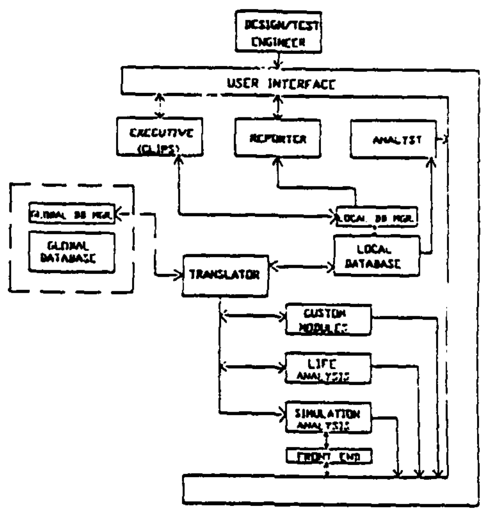

FIGURE 1

Due to recent developments in the capabilities of discrete-event simulation, a secondary goal of research was to investigate the potential for interfacing an external simulation module with the expert system. The simulation module should accept outputs from the expert system for purposes of modeling and trade-off studies among design alternatives. Simulation is the preferred approach, for reliability analysis, because it can account for complexities in system configuration (e.g., redundancies). Simulation also permits more realistic analysis than is possible by closed-form mathematical models. Time-between-failure distributions other than the exponential can be modeled. Also, patterns of growth or deterioration can be modeled. Discrete-event simulation is the tool of choice for manufacturing engineers (designing the production process) and logistics engineers (designing the support process). Therefore a discrete-event simulation tool should be present in the RAMCAD environment, to facilitate concurrent engineering.

This research has accomplished conceptual design and early development of the simulation module. The module can be interfaced with the expert system and provide subsystem or system modeling capability using the outputs of the expert system. The scope of the research included the following tasks:

a. literature review

b. selection of software

c. conceptual design and functional requirements definition for the development of a general reliability simulation model

d. demonstration of simulation model using actual Air Force field data

e. development of requirements for interfacing with the user and the expert system

f. formulation of future enhancements for model and expert system interface.

III. FINDINGS

a. Review of previous work

A review of the literature leads to the conclusion that previous simulation modeling has not provided the design engineer (working at a system or subsystem level) the necessary tools to evaluate design reliability of a system under study. The work done in this area has been directed toward either:

1.) Simulation of large scale models often containing several different systems including scope and detail beyond the immediate needs of an engineer designing at the system or subsystem level [1].

2.) Simulation of specific systems which have been modeled by system-specific applications using one of the various simulation languages [2,4]. It seems that this situation has resulted from two deficiencies: (1) limitations of the simulation language precluding construction of a general reliability model, or (2) the desire on the part of the programmer to narrowly focus the scope on a specific application. The majority of the discrete-event simulation languages available today do not possess the functional capabilities necessary to allow the analyst to develop a general reliability model for varied applications. Models developed using these languages are typically system-specific, requiring changes in coding to adapt to a new system for study. This forces an engineer to become skilled in the programming of some particular simulation software, or to rely heavily on the assistance of a simulation analyst. Neither of these options is desirable.

The principle cause of programming changes, in most simulation models, is the difference in the structure of functional block diagrams from one system to the next. The relationships among system components is modeled by the structure of the block diagram and each is usually unique to the system being considered. The diagram models the serial and parallel relationships among components and also accounts for instances of redundancy present in the system. These relationships must be represented in the simulation model as decision criteria for determining the operating status (failed/not failed) of a system.

b. SIMNET simulation approach

The network construction, and thus the programming, of most discrete-event simulation languages is dependent upon the structure of a system's functional block diagram and thus models are found to be system-specific [3,4]. The one exception to this shortcoming that the Fellow has identified is a simulation software called SIMNET. The Fellow was familiar with the software prior to this research project, having programmed with it while enrolled in a simulation course taught by the developer of SIMNET, Dr. Hamdy A. Taha of the Industrial Engineering Department at the University of Arkansas [3]. SIMNET has allowed the development of a general model requiring minimal input by the user [3]. Rather than re-programming the model, the user only inputs initial data to reflect a change in functional block diagram structure (component relationships), or data (such as component MTBF). This capability eliminates the need for the user to possess analyst-level knowledge of the software. Appendix A describes the key features and advantages of SIMNET, for reliability modeling. In the future, the integrated SIMNET model and user interface will be developed, further simplifying the user's task.

c. Simulation model

The modeling flexibility made possible by the programming features of SIMNET, allowed the construction of a general reliability model [3]. A compact network allows an analyst to easily trace the path to be followed by each transaction. The model flexibility also allows a representation of the true relationships depicted by the functional block diagram of a modeled system.

Upon analysis of the various run scenarios which could be incorporated in a general reliability model, the Fellow has chosen to represent two different model scenarios which seem to offer the most utility and represent varied potential applications. These scenarios are:

1. Mission scenario - Reliability for a

series of missions of some specified equal length of time t. At the completion of each mission any failed components are repaired. If a system failure should occur during a mission, that mission is terminated, repairs are effected on all failed components, and the next mission begun. In this scenario, the variables of primary importance are time between system failures, the mission success probability, and the percentage of times any particular component of interest was involved in a system failure.

2. Continuous operation scenario - Reliability and availability for some continuous specified time t, during which repair is effected upon component failures. Any component failure creating a system failure is given priority for repair. In this scenario, both mean time between failure, and mean time to repair data are input for each component. The variables of interest are mean time between system failures, mean time to repair, operational readiness (availability), utilization of repair facilities, and the percentage of time any particular component of interest was involved in a system failure.

These two scenarios represent a number of potential applications. The mission oriented scenario includes aircraft applications. This scenario models the same basic applications of the Mission Reliability Model software (MIREM) which is an analytic approach to mission reliability,but requires a great deal of space, and specialized training for the user [5,6]. The continuous operation scenario applies to production systems (maintenance test equipment, computer facilities, manufacturing work centers). One example is a parallel arrangement of computer control processor units (CPU's). When one unit fails, it begins to be serviced, while the remaining units continue to operate. The system is still operational until all CPU's are in the failed status.

The SIMNET general reliability model is built upon a number of basic assumptions, including:

1. The time between failures are independently and identically distributed.

2. The repair of components is always successful and assumed to be in as good as new condition.

3. Preventative maintenance is not considered in this model.

4. Simulation begins at time 0, with all components working, and the observation period begins at the end of the transient period.

5. The system being studied can be represented by a reliability block diagram.

6. Steady-state conditions for comparison of alternatives.

SIMNET possesses the ability to model numerous statistical distributions such as the exponential, poisson, beta, gamma, lognormal, normal, uniform, Weibull, and triangular. At this stage in the development of the simulation model the Weibull distribution is coded into the model because of its wide application in the reliability field, and because the exponential distribution is a special case of the Weibull distribution with a shape parameter of 1. In the current model, any distributions can be applied by altering the program statements in the appropriate locations. The ability to assign a distribution in response to a user prompt is planned as a part of the continued development of the model. This will allow for direct integration and complete utilization of the simulation module through the use of outputs from the expert system currently under development (see figure 1).

Another valuable feature of the general reliability model allows for the study and analysis of a critical component. The user may specify, at the outset of any run, a particular component of interest for study. During execution, a variable will accumulate the number of times this particular component is involved in a system failure, and calculate this as a percentage of total system failures. Sensitivity analyses, such as the incremental affect upon system reliability created by the increase in a component's MTBF, can thus be analyzed for the user by simply varying the MTBF in the initial input data. In this way, those components most responsible for system degradation can be easily pinpointed and desired improvements determined. This can serve to identify critical components which might not be inherently obvious to the user, but upon execution, show to be a liability to the system.

Model logic

At the outset of a run, the user will input information specific to the system to be modeled. These inputs include:

1. number of components in system.

2. number of repair resources available (for the continuous operation with repair scenario).

3. identification of a component within the system for individual analysis.

4. desired mission length (for mission scenario).

5. any transient period desired.

6. total run length.

7. incidence matrix containing the minimal cut sets of the system.

The incidence matrix is formed from the functional block diagram, representing component relationships (series/parallel) of a system. Each component in the system is assigned a unique number as identification (1...x). Then, the critical paths from the source to sink nodes are identified, and the components involved are assigned a 1 to identify a minimal cut set. Each of these critical paths are identified and accumulated in the incidence matrix (see figure A4). The incidence matrix is then input to the model in order to determine system status upon a component failure.

At the outset of a simulation run, a determination is made as to which model scenario will be in effect. The determination is made by scanning the data entered for analysis. If repair data has been entered, then the continuous operation with repair scenario is implemented; otherwise the mission scenario is assumed. At the same time, a transaction is created with an attribute to represent each component in the system being analyzed.

For the mission scenario, components are each assigned a time to next failure based upon the MTBF data input to the model. At these assigned times, the status of a component is changed from a 0 to a 1 to indicate failure. A check is then performed to see if the failure of this component has created a system failure. This is done by comparing the current status of the attribute string (representing all components) to the collection of cut sets contained in the incidence matrix input by the user. If the 0,1 configuration of the attribute string matches any entry in the matrix (or if more 1's are present) then this indicates that a system failure has occurred. At this point, the termination of a mission and a return to base is simulated by effecting repair of all failed components, and returning each component's attribute to a 0 operating status. If the failure of a component does not create a system failure, then such components remain failed until either: 1.) a system failure occurs, or 2.) the end of the mission. At the end of a mission length, repairs are effected to all failed components.

For the continuous operation with repair scenario, the model operates similarly except that upon the failure of a component, a repair process is begun, with repair times determined from the repair data entered by the user. If a component failure creates a system failure, then this component is given priority for repair, and the lives of remaining components are "frozen" at the time of system failure. This effects the "bad as old" status of the remaining components upon the repair of the component(s) critical to the operation of the system.

d. F-16 case study

To aid in the validation process and display the utility of the general reliability model, a case study was developed for the F-16C voice communication subsystem using actual Air Force data. Working jointly with F-16 engineers, actual field data was obtained from the F-16 MODAS. Also, with the aid of F-16 engineers, the functional block diagram was developed for the subsystem and the associated MTBFs for each subsystem component identified (see figure A3). For the case study, mission success probability was defined as secure voice radio communication (either UHF or VHF band). From the functional block diagram, the minimal cut sets were determined, and formed into an incidence matrix shown in figure A4. A cut set is a collection of components which, if failed, result in a system failure. It was decided to simulate a 2 hour mission length for this aircraft. An analysis of the TIME TO FAIL variable (representing the time between system failures) was performed using an arbitrary run length of 120,000 hours to determine an appropriate transient period for the simulation. Note that in actual operations, a specific aircraft would not accumulate this quantity of sorties or flying hours, and would therefore never reach steady state condition. However, steady state is a necessary assumption to make valid relative comparisons among alternatives. Through the use of the SIMNET $PLOT statement, which graphically displayed the value of the TIME TO FAIL variable at specified intervals of 1,000 hours, it was determined to take approximately 60,000 hours to reach steady-state conditions. (see figure A2) Therefore, the period of time was designated as the transient period for the run, and the observation period during which statistics were gathered, began at 60,000 hours. The observation period spanned 50,000 missions or 100,000 flight hours. It was also decided to single out component #4, the Processor Adapter Assembly KY-58 (work unit code 63C80) for individual analysis since it was involved in a serial relationship, was a necessary component for secure voice communication, and also possessed a relatively low MTBF (2047 hours). A histogram was produced as part of the output representing the time between failures for the communications subsystem. The histogram recorded all system failures falling between 100 flight hour increments from 0 to 1000 hours. The histogram indicates the distribution of the time-between-failures variable. The case study input data and program output are contained in Appendix A for reference.

The output from the simulation run (shown in Appendix A) summarizes variable calculations and the resulting histogram. The variable representing the mean flight time between mission critical system failures, TIME TO FAIL, was calculated to have a value of 662.1 hours. The number of updates recorded for this variable indicated that there were 151 system failures recorded during the 50,000 simulated missions. This yields a probability of success for the mission to be 99.7% (PROB SUCES variable). In other words, approximately 3 out of 1000 missions would experience failure due to loss of secure voice radio communications. The individual analysis of the Processor Adapter Assembly yielded 44 occurrences of this component being involved in a system failure (FAILS), and this represented 29.14% of the total mission failures. The Air Force has recently initiated a Twice R-Half M policy. This policy requires that a new item, developed as a replacement for an item in inventory, must exhibit double the MTBF and one-half of the maintenance required by the predecessor. The Fellow chose to investigate the impact of this policy on a potential design alternative: replacing the KY-58. When the MTBF for the KY-58 was doubled from 2047 to 4094 hours, the item's contribution to system failures was reduced from 29.14% to 21.85%. This brief analysis of the simulation output illustrates the value of such a tool for use by the design engineer during the early stages of system development.

The mean time between system failures of 662.1 hours took into account the serial and parallel relationships among the components of the subsystem. This result leads to an interesting and very valuable comparison. Often times during the design and testing stages, engineers assume strictly serial relationships exist between the components of a system. This assumption is made to avoid the often complex analysis presented by functional series/parallel relationships. If this assumption had been made in this case study, the resulting reliability for the subsystem would be 113.7 hours MTBF (98% mission success probability for the two-hour mission) This compares with the 662.1 hours MTBF (99.7% success probability) that is actually representative of the subsystem considering the redundancies for a mission. This demonstrates that the series-system approximation is pessimistic and misleading.

e. Integration requirements

In order to interface the simulation model with the expert system, the following exchange of data would occur:

1.) The expert system would analyze raw data, determine through statistical testing the most applicable probability model for the data, and calculate the associated MTBFs (or other parameters) for individual components.

2.) The expert system would write this output to an external ASCII file (local database) for use by the simulation model.

3.) The simulation model would read from the file and import the necessary data, then would prompt the user, through the interface, for other data inputs.

4.) The results obtained from the simulation run would be written to an external file (local database) for use by the expert system and/or the user.

Upon complete development, this simulation model can serve as a valuable tool in either a stand-alone analysis or as an external function to the expert system, depending upon the objectives and/or the skill of the user.

f. Future enhancements

A number of future enhancements to both the simulation model and the expert system interface would serve to increase the efficiency, utility, and outward appearance of the software. These suggested improvements include:

1.) Complete development of the expert system interface.

2.) Ability for the user to construct an on-screen schematic diagram of the functional block diagram representing the system being studied.

3.) Use of the on-screen diagram to identify cut-sets.

4.) Development of an algorithm to automatically translate the functional block diagram constructed by the user into an incidence matrix containing the minimal cut sets for direct entry into the simulation module. The work performed in this area at this time has been highly theoretical in nature [7,8].

5.) Construction of external file functions to read and write I/O from the simulation module.

6.) Generalization of the simulation model to accept any statistical distribution based upon user selection from a user interface.

7.) Development of additional graphical capabilities for the model such as plotting of the functional block diagram, construction of shaded histogram plots in color, printing of incidence matrix, and automatic development of a network diagram representing the model code.

8.) Suppression of SIMNET default outputs that are not of interest to the engineer and/or the development of an output file containing only pertinent output of interest, fully interpreted for the user.

9.) Development of the transient period analysis for easy use by the user through the interface.

IV. CONCLUSIONS

A review of the literature in the field of reliability simulation has indicated that there is no general discrete-event simulation model to assist the engineer in analyzing system reliability at the system or subsystem level in an integrated RAMCAD environment. The design engineer needs a general model to facilitate the analysis of reliability design alternatives. In the past, models have attempted to consider several different systems at once and simulate very complex systems, or analyst-level users have hard-coded their own system-specific models for applications. But due to a lack of flexibility on the part of the simulation languages, these models represented customized tools which would require re-programming for other applications.

SIMNET, a newly developed discrete-event simulation language, offers the flexibility necessary to construct such a general reliability model for application to virtually any system the typical design or test engineer might be analyzing [3]. This is accomplished by simply changing the initial data inputs utilized by the model. The ensuing development and validation of this model using actual F-16 field data, has served to illustrate the merit and value associated with the development of such a tool for use by engineers. At the current stage of development, the model is capable of modeling two scenarios: the mission scenario, and the continuous operation with repair scenario. This model will eliminate much of the judgement and erroneous assumptions that engineers have been forced to make in the past regarding the assessment of reliability.

V. RECOMMENDATIONS

The Fellow feels that the simulation model is consistent with the objectives of the RAMCAD program and would serve as a valuable tool at a RAMCAD workstation to assist the engineer in reliability analyses. The model would allow the engineer to assess the effects of design alternatives on system reliability from the analysis of data available on components comprising a system. This trade-off methodology is consistent with that of other existing RAMCAD modules. The simulation model also provides another tool for the promotion of concurrent engineering in the development and testing of a proposed design. Upon integration with the expert system, this software would serve as a valuable addition to a RAMCAD workstation by allowing an accurate and effective analysis of reliability data to be performed at the system or subsystem level.

Topics for future research include the continued development of both the user and expert system interface, along with the development of an efficient algorithm to translate a schematic representation of a system's functional block diagram into an incidence matrix containing the minimal cut sets of the system. Upon complete development of the user and expert system interface, the model should lead the user through the necessary inputs, thereby eliminating the need for the user to possess analyst-level knowledge of simulation software. An algorithm is also needed to identify the minimal cut sets, based upon a system's functional block diagram.

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FINAL REPORT

Development of a Candidate Task Taxonomy for Air Force Enlisted Specialties

Prepared by:	David L. Mayfield (with Charles E. Lance, Ph.D.)
Academic Rank:	Graduate Student
Department and University:	Department of Psychology University of Georgia
Research Location:	AFHRL/MOD Brooks AFB San Antonio, TX 78235-5601
USAF Researcher:	R. Bruce Gould Ph.D.

Date:	19 August 1988
Contract No:	F49620-88-C-0053

Development of a Candidate Taxonomy for

Air Force Enlisted Specialties

by

David L. Mayfield

ABSTRACT

Principal Component Analysis was conducted on the General Work Inventory and the Electronic Principals Inventory toward developing a taxonomy of tasks performed by USAF personnel. One of the primary purposes of the taxonomy is to permit the assessment of similarities between the job requirements across different Air Force specialties. Results from several analyses, along with input from AFHRL Scientists resulted in a 26-category candidate skill/task taxonomy. Definitions of the categories, comparisons between this candidate taxonomy and other taxonomies, and practical uses of this taxonomy are presented. The candidate taxonomy needs to be validated in terms of its ability to support accurate cross-AFS ease of movement predictions.

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I would also like to thank Lt.Col. Larry Short for supporting our summer work, Capt. Joe Filer for his ideas on estimating cross-AFS transferability of skills, and to Mr. Wayne Archer for his critical and insightful contributions at various stages of this research.

Finally, I am very grateful to Drs. Bruce Gould and Michael J. Kavanagh for the advanced planning of our summer work, for providing us with background literature prior to our summer work, and for their support and critical contributions during the summer research period. Drs. Gould and Kavanagh helped make this summer work possible.

INTRODUCTION:

Melton and Briggs (1960) saw a need for the development of a general task taxonomy that would allow the integration of human performance data into a useful, predictive tool. Since that time, a number of task, ability, intellect , temperament, personality, interest, environment, and motivation taxonomies have been proposed and developed (see for example, Dunnette, 1976; Farina, 1973; Fleishman & Quaintance, 1984; Peterson & Bownas, 1982; and Wheaton, 1973).

A task taxonomy can be defined as the scheme by which tasks and subtasks, identified through a task analysis, are classified into a logical framework (Companion & Corso, 1982); more specifically, "a taxonomy is a means of classifying objects or phenomena in such a way that useful relations among them are established" (Miller, 1967).

Fleishman & Quantance (1984) state the basic assumption behind the development of task taxonomies: "The world of human tasks is not impossibly diverse and that common task dimensions can be identified which will allow improved prediction of human performance on these tasks." They express the need for taxonomic development in the field of human performance as well as the usefulness and applied practical benefits of a human performance taxonomy. Behavior taxonomies may help to address many common concerns in basic and applied psychology and to integrate concepts and research in a number of seemingly diverse fields (Fleishman & Quaintance, 1984).

Peterson and Bownas (1982) separate taxonomies into two categories: task and human-characteristics taxonomies. The task characteristics approach defines the task as a set of conditions that elicits performance. The assumption is made that tasks can be described and differentiated in terms of intrinsic, objective properties which they may possess. Studies by Fitts (1962), Farina and Wheaton (1973), and Stolurow (1964) are representative of attempts to pursue the task characteristic approach. Human characteristic approaches produce taxonomies based on human cognitive abilities (see Dunnette, 1976); psychomotor and physical abilities (see Fleishman, 1964); personality (see French, 1973, and Gough, 1976); and vocational preference (Holland, 1976).

Acording to Companion and Corso (1982) an effective task taxonomy should: (a) simplify the description of tasks in the system, (b) employ terms that are compatible with the users, (c) be complete and internally consistent, (d) be compatible to the theory or system to which it will be applied, (e) provide some basis on which performance can be established or predicted, (f) have some practical utility, (g) be cost effective, (h) provide a framework around which all relevent empirical data can be integrated, and (i) account for the interaction of task properties and operator performance. In the present study, attention was given to these criteria in the development of a task taxonomy.

II. BACKGROUND:

Important human resource planning issues for the USAF concern estimating manpower, personnel, and training (MPT) requirements for new and modified weapon systems (WSs). The emphasis of this MPT issue is, given a specific WS design, to estimate alternative sets of personnel requirements for implementing, operating, and maintaining the WS. Each set of estimated personnel requirements would include projections for: (a) occupational structures, or configurations of jobs to support the WS, (b) manpower needs, or the numbers of people required to support the WS, (c) knowledge, skills, and abilities required for operation and maintainers of the WS, (d) training needs, and (e) costs of implementing the personnel requirements plan (Kavanagh & Gould, 1988). In order to generate these personnel forecasts the USAF needs a skill/task taxonomy for enlisted Air Force Specialties (AFSs).

While much research has been conducted on work taxonomies (see, for example, Dunnette, 1976; Fleishman & Quaintance, 1984; Peterson and Bownas, 1982; and Wheaton, 1973), the USAF has only recently begun work toward the development of a task taxonomy for enlisted AFSs (United States Air Force Occupational Measurement Center, 1984) (see Table 1, OMC Classification). The present work was intended to extend research efforts in the MPT Technology Branch of the Manpower and Personnel Division, Air Force Human Resources Laboratory (AFHRL/MOD) by developing a taxonomy of tasks which can be used in assessing similarities between the requirements across different Air Force specialties or in existing and emerging AFSs.

Statement of Problem

The USAF's Occupational Measurement Center (OMC) identifies tasks through reviewing job documentation, observing workers, interviewing and surveying workers and supervisors (Christal, 1974). The result is the creation of extremely large task inventories containing nearly every task performed in a particular specialty. Because each inventory is created within each Air Force Specialty (AFS), cross AFS comparisons are difficult. The USAF needs a general skill, knowledge, and ability taxonomy based on all AFSs in order to make cross-AFS comparisons. This study investigates one possible taxonomy.

III. PROCEDURE

Ideally, inductive development of a taxonomic system would include (a) a comprehensive definition of the domain of objects to be classified, (b) a thorough description of characteristics of objects within the domain on which classification decisions will be based, (c) devising a means for measuring these characteristics, (d) measurement of relevent characteristics of objects, (e) an initial empirical classification based on objects' similarities (e.g.,using pricipal component analysis), and (f) cross-validation of the classificatory system. In the present case this would require the development of a general work inventory for USAF jobs, empirical groupings of tasks, and cross-validation of the derived taxonomic system. However, present, practical constraints: (a) the absence of an general work inventory developed specifically for the Air Force, (b) budgetary restrictions, and (c) time constraints, required the use of existing data sets, none of which individually were entirely appropiate. Consequently, results from multiple analyses performed on a number of data sets were logically synthesized into a working Air Force task/skill taxonomy. Discussions with

USAFHRL scientists and subject matter experts also provided input to the present efforts to develop a taxonomy suited to the Air Force research needs.

The General Work Inventory

In the present study the General Work Inventory (GWI) was used as a substitute for a general Air Force work inventory. The (GWI) is a shorter and less technical version of the Occupational Analysis Inventory (OAI). The OAI was designed to be used in "describing, comparing, and grouping jobs and occupations for educational purposes" (Cunningham, Boese, Neeb, & Pass., 1983). The GWI's 268 job-rating items, or work elements, are based on selected factors and modified elements from the OAI, in addition to information from other sources. The GWI is organized into eight sections (A) Sensory Requirements, (B) Information Elements, (C) General Mental Requirements, (D) General Physical Requirements, (E) Physical Work Activities (F) Interpersonal Work Activities, (G) Work Conditions, and (H) Job Benefits/Opportunities. Conceptually, the major divisions of the GWI correspond to the components in the information processing paradigm presented by Cunningham et. al. (1983). The GWI was designed to achieve as much specificity in description as possible, and at the same time retaining applicability to the general population of jobs. In contrast to the Positions Analysis Questionaire (PAQ) (McCormick, 1976), which was designed to describe the general "worker oriented" characteristics of jobs, the GWI was designed to capture some of the more specific technology (or "job-oriented") content of jobs. However, both the GWI and the PAQ were designed to use in civilian rather than military contexts.

Sections A through F of the GWI are rated on a Part-of-the-Job scale which asked the rater to consider three factors in determining as element's part in the job: (1) how important or critical the element is to the job, (2) how often the element occurs, (3) how much time the worker spends with the element. Ballentine and Cunningham (1981) administered the GWI to 2,141 skilled (5-level) USAF personnel; this data set was used in the current study.

The first step in deriving a candidate task taxonomy was deciding what sections of the GWI to analyze. The intent was to create a taxonomy of tasks based on work behaviors. Sections A (sensory Requirements), G (Work Conditions), and H (Job Benefits) are not directly related to work behaviors. Therefore, only sections B through F of the GWI were used in the analysis. These sections contain 217 of the 268 items.

Principal Component Analysis of the GWI

Principal Component Analysis was conducted on sections B through F (hence forth refered to as the GWI) using MAX FACTOR (a factor analysis program which is designed to handle a large number of variables) and the first one hundred components were retained for a plot of the eigenvalues. Cattel's (1966) scree test was used to determine the appropriate number of components to retain. Results suggested twenty to thirty-five principle components should be retained in subsequent analyses.

Principle Component Analysis (PCA) of the GWI were conducted (again with MAX FACTOR) and 20, 28, 32, and 35 principle components (PCs) were retained for VARIMAX rotation. These analyses resulted in: (a) two "large" "Mechanical" and "Electrical/Electronic" first (PCs) which contained 19 to 28 items, (b) several interpreptable components, many of which were similar across solutions, and (c) a small number of uninterpretable PCs.

Next, separate PCAs were conducted on the items which loaded on (a) the two "large" "Mechanical" and "Electrical/Electronic" components and (b) the group of the "remaining items" (items in sections B-F minus the items loading on the "Electrical/Electronic" and "Mechanical" components). Using SPSSX, four-, five-, and six-component solutions were retained for VARIMAX rotation for the group of "Mechanical" and "Electrical/Electronic" items. 18-,26-, and 30-component solutions were retained for VARIMAX rotation for analysis of the "remaining items".

IV RESULTS:

In total, eleven different PCAs were performed on the GWI data: 100-, 20-, 28-, 32-, and 35-component solutions on sections B-F; 3-, 4-, and 5-component solutions on the items loading on the "large" "Electrical/Electronic" and "Mechanical" factors; and 18-, 26-, and 30-component solutions on the "remaining items". The analysis produced several interpretable components, many of which were similar across the different solutions (see Table 2 for the 26component "Remaining Items" solution and Table 3 for the 5-component "Electrical/Electronic" - "Mechanical" solution).

Synthesis of a set of meaningful task/skill categories, identified from examination of all the solutions (with particular attention given to the 'Electrical/Electronic' - 'Mechanical' and the 'remaining items' analyses), led to the generation of a draft candidate taxonomy of 33 categories (see Table 1, Draft Candidate Taxonomy).

Discussions with AFHRL/MOD Scientists

The Draft Candidate Taxonomy was reviewed by Drs. Bruce Gould and Michael Kavanagh, Mr. Wayne Archer, Capt. Joe Filer (USAF/MODJ), and the authors, and decisions to delete, combine, or expand certain categories were made. Categories were deleted because (a) the operational implications of thier task content were covered by other taxonomy categories (e.g. the activities in Transporting and Shipping are represented in Maintaining Inventories, Mechanical Equipment Operation, and Physical Labor), (b) had no operational value or relevance to the USAF (e.g. Apprenticing or Vending/Merchandising), or (c) were too 'job specific' (e.g. the category Policing refering to the general set of activities performed by the 811x0 AFS). Other categories were combined in order to balance the taxonomy such that it would be more congruent with proportions of Air Force Specialties (e.g. Scientific/Technical, Biological, Physical Sciences, Social Sciences/Services and Engineering were combined to form Science & Engineering).

The GWI, being a 'general' work inventory, did not contain the number of 'electrical/electronic' and 'mechanical' items that were needed to classify the large number of electrical/electronic or mechanical specialties in the USAF. In order to increase the specificity of description of the mechanical area, the Mechanical category was logically expanded into Mechanical Systems Operation and Mechanical Systems Maintenance.

Expansion of the Electrical/Electronic Category

Another measuring instrument, the Electronics Principles Inventory (EPI), was used in order to describe electrical/electronic categories in the proposed taxonomy. The EPI was developed at the USAF Occupational Measurement Center (OMC) for the purpose of course validation (USAFOMC, 1984). It contains 1257 items covering the full scope of electronics principles or fundamentals as defined by Air Training Command (ATC) fundamental courses and by instructors and supervisors of these courses. The EPI instructs job incumbents to indicate whether each principle is used on their present job (responses are in a binary yes/no format). The EPI has been used operationally by ATC to validate training courses for more than 5 years, which, along with extensive validation studies, attests to the validity of the instrument (Ruck, 1986).

The 1257 EPI items are divided into 39 classifications (categories). Measurements from each of the 2494 respondents were averaged within item classifications (categories) to yield a more manageable data set of 39 "percentage of classification (category)" scores. Principal components analysis was conducted using SPSS-X and the appropriate number of components, as determined by a scree test (Cattell, 1966) were retained for varimax rotation. A 5-component solution (shown in Table 4) was interpreted by Dr. Gould and a subject matter expert (electrical repair specialist) and was used to augment the candidate taxonomy. The five "Electrical/Electronic" categories are (1)

Complex Electronic Circuit Maintenance, (2) Digital Systems Maintenance, (3) Communication Systems Maintenance, (4) Basic Electrical/Electronic Repair, and (5) Electronic Peripherals Maintenance (see Table 4).

After these modifications were made, definitions were formulated for each of the new 26 categories based on the items from which they were designed (see Table 5).

V. CONCLUSIONS:

Although similar to the OMC Classification (see Table 1), the proposed candidate taxonomy (a) is based on empirical evidence (as opposed to logical analysis used in the development of the OMC Classification), (b) better defines the electrical/electronic and mechanical specialties, and (c) respecifies ambiguous OMC categories (e. g. Simple Physical Labor, General Tasks or Procedures, Simple Nontechnical Procedures and Special Talents) in more concrete terms (e.g. Manufacturing/Fabricating, Construction, Artistic, Food Preparation, and Animal Care). In general, the Candidate taxonomy specifies a more exhaustive and distinct set of task content categories than the OMC taxonomy.

The proposed taxonomy trades specificity for parsimony. Compared to the rather lengthy and complicated taxonomies such as the PAQ, containing 187 job elements (Mccormick, 1976) and the OAI with 617 work elements (Cunningham, et. al., 1983), the proposed taxonomy can be used for more general purposes (e. g. cross-AFS skill, knowledge, and ability comparisons). It also was designed with the guidance of USAF scientists which increases the liklihood that it will be useful for the Air Force's research needs.

The Candidate Taxonomy can be used in the USAFMPT research on (a) forecasting the task content and skill requirements for projected AFSs (b) anticipating training needs for new AFSs, (c) setting aptitude standards, and (d) determining optimal strategies for internal (cross-AFS) reassignments, and (e) designing alternative organizational structures for the operation and maintenance of emerging WSs,. By allocating tasks to the categories of the candidate taxonomy the USAF can make cross-specialty comparisons in order to establish ease of movement (EOM) indices for movements from one specialty to another or from existing to newly developed specialties.

Further Research Needs

The Taxonomy must now be validated in terms of (a) the distinctness of its categories, (b) the extent to which categories are descriptive of tasks performed by enlisted personnel, (c) its usefulness in distinguishing among AFSs, and (d) its ability to support accurate cross-AFS ease of movement predictions. A review by subject matter experts and the members of the OMC staff as well as empirical validation is needed. Lance (1988) has proposed such research.

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SCIENCE AND ENGINEERING Collecting/organizing/summarizing technical information/ information about people, events, places. Using systematic/ scientific methods to test theories/products/ equipment. Write reports of results/findings. Using chemical, physical, geological, topological, geographical, or meteorological information, such as aerial photos, barometric pressures, weather forecasts, maps, books/reports on mineral deposits, chemicals, radiation, or hydraulics. Using technical drawings and engineering plans such as blueprints, charts, wiring and circuit diagrams, and other designs/plans for machinery, equipment, structures, manufacturing processes, etc.

ARTISTIC - AUDIO/VISUAL Audio and Visual art design/production, e.g., photography, movies, recordings, drawing illustrations, designing layouts for publications, creating musical compositions decorating building interiors. Performing, which would include playing musical instruments, singing, dancing; and acting. Operating equipment such as turntables, stereos, videotape players, overhead or slide projectors, tripods, film developing equipment, etc.

FOOD PREPARATION Preparing/cooking food, using/producing menus, recipes, nutrition guides, food requests and estimates.

ANIMAL CARE Using books/reports/records/instructions to groom, exercise, train, treat, and tend animals.

FABRIC/ROPE WORK Work involving sewing, stitching, threading, weaving, combining, separating etc. of fabric, thread, rope, material, fiber, string, etc.

MANAGING OTHERS Managing/administering/supervising/evaluating others, e.g., determining goals and coordinating others' activities, assigning work to others and supervising their work activities, evaluating others' performance, making staffing decisions, conducting group meetings, settling conflicts and enforcing rules.

TRAINING Explaining ideas/procedures to others, demonstrating how a task is done, monitoring learner progress providing feed-back on mistakes, preparing lesson plans, course outlines, etc.

SURVEILLANCE Using codes/symbols (traffic control "lingo," flag, and hand signals), detecting, visualizing and recognizing objects that are difficult to see (e.g., ships and aircraft at a distance or criminal suspect in a crowd), tracking and pursuing moving targets/objects, using firearms or other handheld weapons, enforcing rules or laws.

COMMUNICATION SYSTEMS MAINTENANCE Maintaining and repairing communication systems such as antennas, microwave oscillators and amplifiers, signal generators, and transmitters and receivers. Making radio frequency measurements and calculations. Using osclloscopes.

BASIC ELECTRICAL/ELECTRONIC REPAIR Maintaining and repairing basic electro-mechanical equipment with a working knowledge of multimeters, direct and alternating current, and soldering and solderless connections.

ELECTRONIC PERIPHERALS MAINTENANCE Maintaining and repairing electronic peripheral devices such as storage type display tubes, microphones and speakers, and photosensitive devices.

PHYSICAL/MANUAL LABOR Nontechnical manual and physical tasks such as sweeping, lifting, carrying, cleaning, sawing, lubricating, bonding/sealing, drilling, cutting, hoisting, chipping, and planing. Using basic tools such as a hammer, paint scraper, shovel, or wheelbarrow.

MANUFACTURING/FABRICATING Making things from materials such as sheet metal, metal tubing, glass, brick, plastic, rubber, paper, and lumber. For example, Pressing, mixing, beating, forging, grinding, stitching, forming, melting, and chemically treating materials to manufacture things.

CONSTRUCTION Using construction information such as materials lists, building designs, etc. Building/maintaining structures made of brick, stone, lumber, asphalt, or concrete, such as walls, floors, cabinets, houses, bridges, towers, roads, run Laying/covering with roofing materials, floor coverings, wall paper.

MEDICAL - PATIENT CARE Verbally or physically interacting with patients, e.g., bandaging, giving injections, applying medicines, drawing blood, performing physical therapy. Reading/interpreting medical charts, thermometer readings, test results.

MEDICAL - TECHNICAL Performing technical procedures in a medical lab or operating room, e.g., operating X-rav machine, microscope, EKG machine, respirator, ultrasound machine.

ORAL AND WRITTEN COMMUNICATION Reading/speaking/writing, understanding words, and expressing ideas, including receiving/processing/initiating letters, books, reports, phone calls, orders, directions/instructions, lectures, contracts; attending/conducting meetings, presentations.

PLANNING/PROBLEM SOLVING Using available information to anticipate/figure out/solve problems, and plan the steps and procedures required to reach a solution to the problem (e.g., identifying a traffic problem and formulating a plan for re-routing traffic).

TABLE 5

Revised Task Taxonomy Category Definitions

CLERICAL Secretarial functions such as filing, preparing forms, answering telephones, taking dictation, typing reports, proof-reading copy, comparing lists/data. Operating office equipment/ machinery such as computers, typewriters, calculators, and computer terminals. Processing information related to military regulations, federal or state laws, leases, contracts, court records and decisions, and legal documents.

PERSONNEL Processing data/information about individuals, such as employment applications, performance reviews, disciplinary reports, media releases, production records, personnel forecasts, training records, counseling information, and social services.

MAINTAINING INVENTORIES Maintaining materials/merchandise/ supplies/equipment records. Ordering, receiving, maintaining, routing and accounting for inventory. Preparing, analyzing and maintaining records of financial dealings, property, assets.

COMPUTATIONAL Performing basic numerical operations such as adding, subtracting, multiplying, dividing. Computing statistics using formulas and equations. Locating statistics/data in graphs/ tables/charts. Using computing devices (calculators, adding machines) to compile statistics and solve math problems.

MECHANICAL SYSTEMS MAINTENANCE Performing mechanical repair and maintenance activities including maintaining, repairing, assembling, installing, troubleshooting, and adjusting/tuning mechanical systems. Lubricating, bonding and sealing, and using tools (such as a hammer, screwdriver, jack, clamp, or block and tackle) to repair mechanical systems.

MECHANICAL SYSTEMS OPERATION Operating mechanical equipment such as a meat slicer, sewing machine, printing press, bulldozer, road grader, forklift or tractor. Driving/operating/piloting vehicles. Using mechanical tools such as an electric drill, air wrench, chain saw, or jack hammer.

COMPLEX ELECTRONIC CIRCUIT MAINTENANCE Maintaining and repairing equipment containing complex electronic circuitry such as electron tube amplifier circuits, tubes, limiter and clamper circuits, wave shaping circuits, multivibrators, oscillators, coupling circuits power supply filters, resonant cavities, and magnetic amplifiers.

DIGITAL SYSTEMS MAINTENANCE Maintaining and repairing digital systems such as digital logic numbering systems, computers, television, laser, and infared systems. Maintaining and repairing equipment which contains digital circuits or processes digital to analog/analog to digital information.

Table

Comparison Between OMC and Candidate Task/Skills Taxonomies

OMC Classification	Revised Candidate Taxonomy	Draft Candidate Taxonomy
1. Clerical	1. Clerical	1. Clerical
	2. Personnel	2. Personnel
	3. Maintaining Inventories	3. Maintaining Inventories
		4. Legal/Contractual
2. Computational	5. Computational	5. Computational
3. Office Equipment		
4. Mechanical	6. Mechanical Systems Operation	6. Mechanical - Maintenance
5. Simple Mechanical Equip/ Systems Operation	7. Mechanical Systems Maintenance	
6. Complex Mechanical Equip/ Systems Operations		
7. Mechanical-Electrical		
8. Mechanical-Electronic		
9. Electrical	8. Complex Electronic Circuit Maintenance	7. Electrical/Electronic
10. Electronic		
11. Electrical-Mechanical	9. Digital Systems Maintenance	
12. Electrical-Electronic	10. Communication Systems Maintenance	
13. Electronic-Mechanical		
	11. Basic Electrical/ Electronic Repair	
	12. Electronic Peripherals Maintenance	
14. Simple Physical Labor	13. Physical/Manual Labor	8. Physical/Manual Labor
		9. Semi-Skilled Labor
		10. Transporting/Shipping
	14. Manufacturing/Fabricating	11. Manufacturing/Fabricating
	15. Construction	12. Building Construction*
		13. Construction: Other than Buildings
15. Medical-Patient Care	16. Medical - Patient Care	
16. Medical-Equip Oriented	17. Medical- Technical	14. Medical/Health
17. Medical-Procedures		
18. Simple Nontech Procedures		15. Apprenticing
19. Communication-Oral		
20. Communication-Written	18. Oral & Written Communication	16. Communication
21. General Tasks or Procs		
22. Reasoning/Planning/Organizing	19. Planning/Problem Solving	17. Problem Solving
23. Scientific Math Reasoning or Calculations	20. Science & Engineering	18. Scientific/Technical
		19. Biological
		20. Physical Sciences
		21. Engineering
		22. Social Sciences/Services
24. Special Talents	21. Artistic - Audio/Visual	23. Artistic - Visual
		24. Entertaining
		25. Vending/Merchandising
	22. Food Preparation	26. Food Preparation
	23. Animal Care	27. Animal Care
	24. Fabric/Rope Work	28. Fabric/Rope Work
		29. Operating Audio/Visual Equip
		30. Operating Rail Vehicles
		31. Flying Vehicles
25. Supervisory	25. Managing Others	32. Managing/Developing/ Influencing Others
26. Training	26. Training	
	27. Surveillance	33. Policing/Surveillance

Table 2

GWI Non-Mechanical/Electrical Items Pricipal Componects Analysis
26-Component Solution

Factor 1: Managing

C05	.4900	Social Judgment
F106	.5869	Managing/Administering
F107	.7440	Supervising
F108	.7718	Evaluating Others
F202	.5850	Leading Group Discussions/Meetings
F204	.5448	Settling Conflicts
F206	.4460	Influencing/Convincing
F207	.4780	Communicating
F211	.7328	Counseling
F212	.6387	Teaching
F216	.6097	Enforcing

Factor 2: Scientific/Technical

B20	.5592	Difficult Speaking
B22	.5788	Difficult Writing
B24	.4527	Clerical Operations
B25	.6368	Collecting/Organizing/Summarizing
B26	.6421	Studying/Evaluating
B27	.6521	Creating Information
B31	.5278	Professional Writing
B32	.4357	Researching
B33	.4710	Investigating
C82	.5718	Expressing Ideas
C83	.4611	Creating Ideas
B14	.4287	Tables/Graphs/Charts

Factor 3: Construction

B41	.5898	Using Structural/Construction Information
B42	.5302	Prod/Comm Structural/Construction Information
E151	.6268	Masoning/Trowling/Casting
E152	.5779	Earth Moving/Quarrying/Mining
E153	.6460	Laying/Covering
E162	.6182	Building Structures
E166	.6308	Maintaining/Repairing Structures
E184	.4390	Environmental/Terrain Features
E192	.6571	Exteriors of Buildings
E193	.5508	Interiors of Buildings
E194	.6645	Structures/Constructions other than Buildings

Factor 4: Artistic Activities - Visual Arts

B36	.6593	Visual Art Production
B57	.7796	Using Visual Art/Decorative Information
B58	.7634	Producing/Communicating Visual Arts/Decorative Info.
C04	.6274	Artistic Judgment/Creativity
E120	.4664	Recording/Developing Equipment
E158	.3696	Printing
E172	.5777	Decorating/Styling

Factor 5: Policing/Surveillance

B17	.6316	Visual and Hearable (sic) Codes/Symbols
C90	.5808	Dectecting Objects
C92	.6741	Position/Location Awareness
D100	.6508	Tracking
D106	.6083	Running
E124	.5217	Firearms and other Handheld Weapons
E128	.5379	Off-Road Vehicles

Factor 6: Medical/Health Related Activities

B47	.7106	Using Medical/Health Information
B48	.6806	Producing/Communicating Medical/Health Information
B75	.4601	Using Biological Information
E118	.7793	Medical/Health Equipment
E176	.8847	Medical/Health Treating/Caring
E163	.6053	Acting upon People
F213	.7079	Health Treating/Caring

Factor 7: Computational

B12	.6666	Numbers & Math Symbols
B13	.4295	Written Symbols
B14	.4538	Tables/Graphs/Charts
B29	.6780	Numerical Operations
C86	.6518	Mathematical Reasoning/Problem Solving
C87	.6005	Using Basic Arithmetic
E115	.5070	Computing Devices

Factor 8: Biological

B49	.6406	Using Plant-Life Information
B50	.6830	Producing/Communicating Plant-Life Information
B51	.6619	Using Information about Animals
B52	.6253	Producing/Communicating Information about Animals
E177	.5855	Handling/Caring for Animals
B75	.3066	Using Biological Information
B76	.4371	Producing/Communicating Biological Information
E171	.3870	Caring For Plant Life
E101	.4528	Acting upon Plant Life
E102	.7601	Acting upon Animals

Factor 9: Entertaining

B59	.6592	Using Performing Arts/Entertainment Information
B60	.7058	Prod/Communicating Perf. Arts/Entertainment Info.
E129	.6987	Musical Instruments
E180	.6526	Performing
F210	.6293	Diverting/Amusing

Factor 10: Personnel

B53	.6074	Using Information About Individuals
B54	.7276	Producing/Communicating Info. About Individuals
B55	.5578	Using Human Improvement Information
B56	.6373	Producing/Communicating Info. About Human Improvement
B63	.4340	Using Organizational Management/Administration Info
B64	.5119	Prod./Comm. Org Mgt/Administration Info.

Factor 11: Clerical

B30	.5530	Automated Data Processing
D104	.5701	Sitting
E113	.3940	Communication Equipment
E115	.5259	Computing Devices
E116	.7166	Keyboard Equipment
E117	.5509	Office Machinery/Equipment
E122	.4173	Hand-Held Writing/Drawing/Marking Devices
B24	.4241	Clerical Operations

Factor 12: Vending/Merchandising

B61	.6405	Using Sales/Merchandising Information

Table 2 (continued)

B62	.6891	Producing/Communicating Sales/Merchandising Info.
B67	.6614	Using Economic Information
B68	.6324	Producing/Communicating Economic Information
F214	.4891	Selling/Merchandising

Factor 13: Communication

B10	.6914	Written Words
B11	.7029	Spoken Words
B18	.5216	Reading
B19	.6149	Ordinary Speaking
B21	.5803	Routine Writing
B22	.3730	Difficult Writing

Factor 14: Physical Sciences/Engineering

B43	.4521	Using Info about Materials/Substances/Chemicals
B44	.5377	Prod/Comm Info about Materials/Substances/Chemicals
B70	.5583	Producing/Communicating Environmental Information
B71	.6585	Using Physical Science/Technology Information
B72	.6471	Prod/Comm Physical Science/Technology Info.
B74	.4327	Producing/Communicating Engineering Information
B69	.4940	Using Environmental Information

Factor 15: Manufacturing/Fabricating

E143	.7082	Material Forming
E147	.6748	Heat-Treating Objects/Materials/Substances
E163	.6002	Fabricating
E186	.5124	Processes/Finishes Materials/Substances
E195	.4640	Other Manufactured/Fabricated Products
E144	.4345	Crushing/Grinding/Mixing/Separating Materials/Substances
E145	.4386	Chemically Treating Materials/Substances
E146	.3781	Electrically Treating Materials/Substances

Factor 16: Fabric/Rope Working

E131	.5747	Boats/Ships
E149	.6614	Sewing/Stitching
E150	.6836	Fiber/Thread Working

Factor 17: Legal/Contractual

B65	.6230	Using Legal/Contractual Information
B66	.6025	Prod/Comm Legal/Contractual Information
F215	.5073	Litigating/Contracting

Factor 18: Social Science

B76	.4385	Producing/Communicating Biological Information
B78	.5437	Prod/Comm Social or Behavioral Science Info.
B79	.5041	Using Liberal Arts/Humanities Information
B80	.6138	Prod/Comm Liberal Arts/Humanities Info.
B77	.4339	Using Social or Behavioral Science Information

Factor 19: Problem Solving

C84	.5644	Detecting Problems
C85	.5295	Verbal Reasoning/Problem Solving
C81	.4637	Understanding Words
C89	.4491	Planning
C82	.4184	Expressing Ideas
C93	.3844	Memorizing/Remembering

Factor 20: Food Preparation

B45	.8192	Using Information About Food
B46	.7675	Producing/Communicating Information about Food
E148	.7433	Cooking/Preparing Food

Factor 21: Apprenticing

F200	.5054	Helping Superiors Perform Tasks
F203	.4673	Arguing/Debating
F199	.4368	Being Supervised
F201	.4232	Cooperating
F205	.4206	Bargaining

Factor 22: Policing

E1[illegible]	.4382	Grooming
E177	.4698	Handling/Caring For [illegible]
E1[illegible]	[illegible]	Handling [illegible]
[illegible]	[illegible]	Protecting [illegible]
E176	.3410	Protecting Property
E124	.3341	Firearms and Other Handheld Weapons

Factor 23: Engineering

B74	.4589	Producing/Communicating Engineering Information
B73	.4567	Using Engineering Information
B34	.4066	Bookkeeping/Accounting

Factor 24: Landscaping

E171	.6201	Caring for Plant Life
E181	.6110	Plant Life
E184	.4028	Environmental Terrain Features

Factor 25: ???????

E129	.4349	Rail Vehicles
E123	.3834	Sporing Equipment
E145	.3513	Chemically Treating Materials/Substances

Factor 26: Operating Audio-Visual Equipment

E120	.4084	Recording/Developing Equipment
E191	.4132	Other Equipment
E121	.4362	Audio/Visual Presentation Equipment

Table 3

GWI Mechanical - Electrical Items Principal Components Analysis: Five-Component Solution

Component 1: Electrical/Electronic

E168	.830	Maintaining/Repairing/Setting Up Electrical/Electronic Equipment
D30	.870	Using Electrical/Electronic Info.
E100	.783	Electrical/Electronic Equipment
E168	.767	Troubleshooting
E169	.710	Adjusting/Tuning
E112	.692	Electrical/Electronic Equipment
E114	.692	Measuring/Testing Devices
D40	.768	Producing/Communicating Electrical/ Electronic Info.
E133	.651	Precision Working
E140	.620	Fusing or Cutting By Heat
E160	.593	Installing
E161	.582	Assembling
D18	.576	Reading From Measurement/Testing Devices & Indicators
D16	.594	Drawings/Pictures/Diagrams
E108	.501	Small Handtools
E130	.466	Mechanical Connecting/Fastening/ Joining
E173	.455	Watching/Monitoring Machines or Equipment
C88	.489	Object Problem Solving/Invention
D96	.389	Working With Fingers
D37	.474	Using Mechanical Information

Component 2: Semi-Skilled Labor

E138	.772	Abrading
E136	.767	Sawing
E135	.747	Chipping/Planing/Milling
E137	.750	Drilling/Boring
E134	.567	Cutting By Blade
E109	.547	Large Handtools/Portable Hand-Held Equipment
E142	.537	Bonding/Sealing
E111	.401	Fixed-Location Machines/Equipment
E141	.491	Liquid Application/Coating
E110	.436	Portable Work Aids
E140	.413	Fusing or Cutting By Heat
E130	.406	Mechanical Connecting/Fastening/ Joining

Component 3: Mechanical - Maintenance

E164	.626	Maintaining/Repairing/Setting Up Machines
E155	.612	Lubricating
E180	.631	Machines/Mechanical Equipment
E154	.510	Cleaning

Component 4: Transportation/Shipping

E127	.753	Highway Vehicles
E190	.678	Transporting
D103	.613	Physical Endurance
D102	.500	Strength
D99	.584	Coordination and Balance
D107	.536	Back/Leg Bending Activities
E126	.551	Mechanized Work Equipment
E157	.488	Supporting/Hoisting
E110	.470	Portable Work Aids
E109	.415	Large Handtools/Portable Hand-Held Equipment

Component 5: Manual Labor

D98	.627	Steady Hands
D97	.625	Working With Hands
D101	.594	Quick Reactions
D96	.566	Working With Fingers
D99	.555	Coordination and Balance
C91	.481	Visualizing Objects
D102	.450	Strength
D103	.447	Physical Endurance
D107	.412	Back/Leg Bending Activities

Table 4

EPI Principal Components Analysis: Five-Component Solution

Factor 1: Complex Electronic Circuit Maintenance

ETUBECS	.7715	Electrontube Amplifier Circuits
TUBES	.7176	Tubes
LIMCCS	.6690	Limiter & Clamper Circuits
WAVES	.6640	Wave Shaping Circuits
MVIBS	.6636	Multivibrators
OSSILBS	.6707	Oscillators
COUPCS	.6662	Coupling Circuits
RCICS	.6616	Resistive Capacitive Inductive Circuits
POWSF	.6400	Power Supply Filters
FSFS	.6183	Frequency Sensitive Filters
RESONCS	.6178	Resonant Cavities
POWSVR	.5960	Power Supply Voltage Regulators
MAGAMPS	.5670	Magnetic Amplifiers

FACTOR 2: Digital Systems Maintenance

DLNSF	.7481	Digital Logic Numbering Systems
DIGCS	.7303	Digital Circuits
DIGAN	.6974	Digital To Analog & Vice Versa
COMPS	.6962	Computers
TV	.5372	TV & Laser & Infrared Systems

FACTOR 3: Communication Systems Maintenance

ANTEN	.7727	Antennas
CONNCTS	.7509	Connections
RFMSMTS	.7205	Radio Freq Measurements
RFCALCS	.6314	Radio Freq Calculations
SIGGEN	.5600	Signal Generators
OSCILL	.5420	Oscilloscopes
MOAS	.5350	Microwave Oscillators & Amps
TMITRS	.4767	Transmitters & Receivers

FACTOR 4: Basic Electrical/Electronic Repair

MULTIMET	.8024	Multimeters
ACDC	.7645	Direct-Alternating Current
ELEMECH	.7530	Electro-Mechanical Devices
SOLDER	.6787	Soldering & Solderless Connections

FACTOR 5: Electronic Peripherals Maintenance

STDTS	.7019	Storage Type Display Tubes
MICROPH	.5532	Microphones & Speakers
ELECEYE	.4070	Photosensitive Devices

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FINAL REPORT

FORM DISTORTIONS IN COMPUTER GENERATED MOVING OBJECTS: AN ASSESSMENT OF DISPLAY PARAMETERS.

Prepared by: Jerome I. Nadel

Academic Rank: Masters of Science, Doctoral Candidate

Department and University: Department of Psychology, Kansas State University

Research Location: AFHRL/OT
Williams Air Force Base
AZ, 85240-6457

USAF Researcher: Elizibeth Martin, Ph.D.

Date: 17 August 1988

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FORM DISTORTIONS IN COMPUTER GENERATED MOVING OBJECTS: AN ASSESSMENT OF DISPLAY PARAMETERS.

ABSTRACT

A study is reported that assessed the computer display parameters that yield systematic form distortion of computer generated small moving objects. Lindholm (1988) reported that an interlaced display with a 30 Hz update rate produced systematic form distortion as a function of object speed and direction of motion and line written first. The present study extended the Lindholm (1988) paradigm, testing predictions concerning the rate of movement of the object and presentation duration. We found that (a) the direction of distortion could be predicted by the line written first (top vs. bottom), (b) the magnitude of distortion could be predicted by the object speed, and (c) the reliability of the perceived distortion could be predicted by the presentation duration.

Acknowledgement

I wish to thank the Air Force Office of Scientific Research for sponsorship of this excellent research opportunity. As a returning summer fellow I am especially grateful because over a two summer period I have been given the opportunity to collaborate on two separate exciting and professionally fulfilling projects.

My most sincere appreciation and thanks go to Dr. Elizibeth Martin, my focal point and friend, for her concern with every aspect of research my fellowship. I would like to thank Dr. Julie Lindholm, my summer colleague, for allowing me to collaborate on a very exciting and compelling research project. Finally I would like to acknowledge the efforts of my six experimental observers who donated their time (and "eyes") for the good of science: Lt. Guy Booth, Lt. Vickie Rojas, Lt. Pam Lash, Mr. Pete Pidcoe, Mr. Mark Beorkrem, and Dr. Alan Witt.

I. INTRODUCTION:

The Human Resources Laboratory's Operations Training Division, located at Williams AFB, is particularly concerned with identifying simulator systems that generate perceptually veridical images. This necessitates an interdisciplinary effort combining disciplines of experimental psychology and applied engineering. As a doctoral student of Human Factors or Engineering Psychology I am primarily concerned with human performance and limitations when interacting in a machined environment.

Last summer, as a fellow on the 1987 GSRP, I collaborated on a human factors evaluation of the USAF's AVTS eye-tracking system. That project, in combination with my academic training contributed to my 1988 assignment to the Human Resources Laboratory. The research described in this final report deals with a basic visual phenomenon - apparent motion. More specifically, it attempts to identify the computer display parameters that lead to form distortions in apparent motion. The application of the results described in this report have considerable implications because they help define the image display parameters that yield veridical perception of computer generated moving objects.

II. OBJECTIVES OF THE RESEARCH EFFORT:

In flight simulators, each object is represented in the computer by a model. The resultant display image is composed of an array of picture elements (pixels) arranged in rows and columns. In a raster-refresh display, the pixels are activated by an electron beam that scans from left to right, and top to bottom. The image is commonly read out in the form of two half-frames. This half-frame representation, know as interlacing, writes all of the odd lines from top to bottom first and then returns to the top of the display to write the even lines (Freeman, 1986). In a 30 Hz interlaced refresh system each half-frame representation is written every 1/60 sec. Although displays can be either non-interlaced (with all horizontal lines displayed each vertical sweep) or interlaced (with half of the horizontal lines displayed on each sweep), most flight simulator displays are interlaced.

A second parameter affecting the spatiotemporal representation of a computer generated moving object is the image update rate. The image update rate is the rate at which display information, stored in computer memory, is modified. The most advanced flight simulators, such as the Advanced Visual Training System (AVTS) have image update rates of 60 Hz. However, lower cost simulators have image update rates of 30 Hz or 15 Hz.

Dr. Julie Mapes Lindholm, a University of Dayton Research Institute Research Psychologist, has conducted research attempting to identify the display parameters that lead to systematic form distortion for computer generated moving objects (Lindholm, 1988). In a study prior to my arrival at HRL, Dr. Lindholm assessed the perceptual effects of interlacing (interlacing vs. noninterlacing), image position update rate (30 Hz vs. 60 Hz) and image velocity (60, 120, or 240 pixels per second), when viewing a computer imaged moving object. For the interlaced display, the image could be drawn with either the top or bottom half presented first.

On each trial, one of three airplane-like shapes composed of three pixels on bottom and one on top was drawn; top on left, top in center, and top on right. However, in addition to the three generated object shapes, subjects sometimes saw the top pixel displaced to the left or right of the bottom row, or half-way between the left and center or right and center bottom pixels. The effects of the main variables in this study, image update rate and display update rate, varied with target speed, with the most reliable data occurring at the slowest speed. When the display was noninterlaced and the target moved only 60 pixels per second (one pixel per vertical scan), the perceived shape matched the computer shape on more than 95% of the trials, regardless of the image update rate. However, the results were much more complicated for the interlaced displays: The perceived form depended not only on the update rate, but also on the interlace order - top line vs. bottom line written first. For objects moving at 60 pixels per second in the 60 Hz update interlaced conditions, correct identification occured regardless of first line written - 70% for top first, 80% for bottom first. In contrast, when the image update rate was 30 Hz, the apparent relative position of the top pixel was systematically shifted. In the top-first condition, on 86% of the trials, subjects reported seeing the upper pixel displaced ahead of its physical location by one pixel. In the bottom-first condition, on 83% of the trials, subjects reported the top pixel as displaced behind from its physical location by one pixel.

The results from the Lindholm (1988) study suggested that the interlaced displays currently used in flight simulators may not support veridical perception of the forms of small moving objects. Therefore, it was necessary to replicate these findings and elaborate on the diplay parameters that lead to systematic form distortion of computer generated moving objects.

III.

a. **Experimental Approach:**

The Lindholm (1988) study found reliable form distortion with the 30 Hz update interlaced display, at the slowest object velocity. Further exploratory work suggested that consistent form distortion also occurred at the higher velocities if the observers were given sufficient time to view the stimulus. For example, in conditions where the airplane-like stimuli were presented continuously traversing a CRT, consistent form distortions occurred regardless of object velocity. Under these conditions the magnitude and direction of form distortion could be predicted based on two display parameters; object velocity and line written first (top vs. bottom). The amount of distortion (of the top relative to the bottom of the form) appeared to be half the distance between writes (measured in pixels) in the direction of motion if the top line is written first and opposite of the direction of motion if the bottom line is written first. Interestingly, at higher velocities this distortion could easily be negated by covering up the screen such that the horizontally moving stimulus passed through a small vertical slit. This raised questions about the effect of stimulus duration on the consistency and magnitude of form distortion.

The present experiment used an interlaced display with a 30 Hz update rate. As in the Lindholm (1988) study, top vs. bottom line written first and direction of movement (left-to-right vs. right-to-left) was compared. One stimulus, three pixels bottom with top on center, was used. The response alternatives (see figure 1) represented, in an analog fashion, the predicted magnitude of distortion for the three rates of movement used (60 pixels/s, 120 pixels/s, and 240 pixels/s).

This study also attempted to systematically manipulate subjects ability to process and perhaps visually track the moving stimulus. Three stimulus durations were included; 133 msec, 267 msec (replicating the Linholm 1988 study), and 533 msec. It was hypothesized that the 133 msec duration would be insufficient for tracking and would therefore produce minimal form distortion, while, in contrast, the 533 msec duration would yield sufficient tracking and/or processing time to produce very consistent form distortion.

Fisher and Brietmeyer (1987) have described disengagement as an attentional state that facilitates "express saccades"; a saccadic eye movement with a latency in the order of 100 to 120 msec. In their disengagement paradigm, a temporal gap of 200 msec between offset of fixiation and onset of peripheral target was identified as the optimal interstimulus interval (ISI) for producing express saccades. The present experimenters reasoned that express saccades should facilitate the initiation of visual tracking. Therefore, the Fisher et. al. 200 msec I.S.I. paradigm was adopted in the present study for a "gap" vs. "no gap" comparison. A center fixation vs. peripheral cue fixation condition was also added with the logic that the cue would facilitate tracking of the moving stimulus and therefore contribute to systematic form distortion.

b. Method:

Six subjects (2 female, 4 male) with normal or corrected to normal vision ran through 6 experimental sessions consisting of 144 stimulus presentations per session. Each session was divided into two blocks; 72 presentations with center fixation and 72 presentations of peripheral cue fixation. Fixation/cue order was counterbalanced. Within a given block the stimuli were presented in a random order.

Observers were run on-line with a PDP 11 microcomputer, a Hewlett-Packard 2648A Graphics Terminal with short persistence phosphor, and a VOTAN VTR 6050 Series II Voice Terminal. The Voice Terminal allowed for verbal responses from the observers which, once verified by the observer, were imputed directly into the computer data-set. The subjects viewed the stimuli through a velvet lined cowling from a distance of 70 cm, which for the three object movement rates gave respective angular velocities of 4.7 deg/s, 9.3 deg/s, and 18.5 deg/s.

c. **Results:**

The data were reduced by scoring each response with respect to the direction of motion. As stated previously, the "center plane" computer model was the physical stimulus presented on every trial. Therefore, a response of center was scored as a 0. The distribution of resonses ranged from -4 to +4, with response alternatives (reflecting pixel displacement) producing a distribution of -4, -2, -1, -.5, 0, .5, 1, 2, and 4.

Based on the results of the Lindholm (1988) study it was predicted that line written first (top vs. bottom) should direct the displacement of the top pixel; form distortion in the direction of motion if the top line is written first and opposite of the direction of motion if the bottom line is written first. Figure 2 graphically depicts this relationship for the 60 pixel/sec rate of motion. It also reveals the effect of exposure duration on the reliability of the form distortion; predicted form distortion (1 pixel displacement) increases from 43% at the 133 msec duration to 97% at the 533 msec duration.

Figure 3 further exemplifies the effect of exposure duration on the reliability of perceived form distortion. Note that rates of movement of 60 pixels/sec, 120 pixels/sec, and 240 pixels/sec should produce systematic top pixel displacements of 1,2, and 4 respectively; half the distance between updated writes on the interlaced display. These figures reflect an interaction between rate of movement and stimulus duration. At the 133 msec duration (3a) the predicted displacement is only marginally reflected for the slowest object speed (60 pixels/sec) - 1 pixel displacement. At the 267 msec duration (3b) (a replication of the Lindholm 1988 study) the predicted displacement is very reliable for the 60 pixels/sec speed and somewhat for the 120 pels/s speed.

However, for the 533 msec duration (3c) the predicted form distortions are strongly supported.

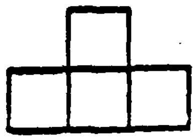
The temporal gap and fixation location variables, relevant to the role of attention/eye movements are presented in Tables 1 and 2. Tables 1 and 2 present comparisons of the effects of these variables for the three stimulus durations, collapsed over all other independent variables. Table 1 shows the predicted percent of distortion for these conditions at the three speeds. Although statistical analyses have not yet been conducted it appears that there are some marginal effects: mean percent no gap = 46.0, mean percent gap = 51.6; mean percent center = 46.9, mean percent offset = 50.9. Table 2 shows the mean percentage correct. It also shows marginal effect for the temporal gap and cue variables. However, these tables do exemplifiy the effect of stimulus duration on the reliability of perceived form distorion; as duration increases percent of form distortion increases, while percent of correct (veridical object perception) decreases.

IV. RECOMMENDATIONS:

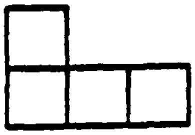
Formal statistical tests are planned for the results obtained in this study. This project was successful in replicating the effects of the Lindholm (1988) study as well as empirically verifying the predictions set forth concerning the magnitude and direction of form distortion on interlaced displays as a function of line written first, object direction, object speed, and presentation duration. Follow-up research is recommended, encorporating eye movement monitoring to more directly assess the mechanisms that yield the observed form distortions that occur with computer imaged moving objects. These basic research efforts, in combination with applied flight simulator research should identify the display parameters that yield systematic veridical perception of the forms of small moving objects

Figure 1

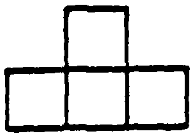
Stimulus Objects (Computer Models)



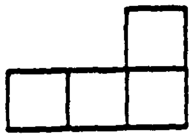
Response Alternatives



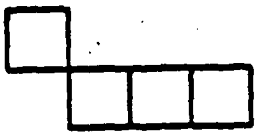
Left



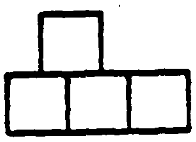
Center



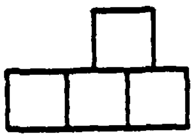
Right



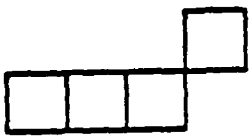
Off-on-the-left



Slightly Left



Slightly Right



Off-on-the-right

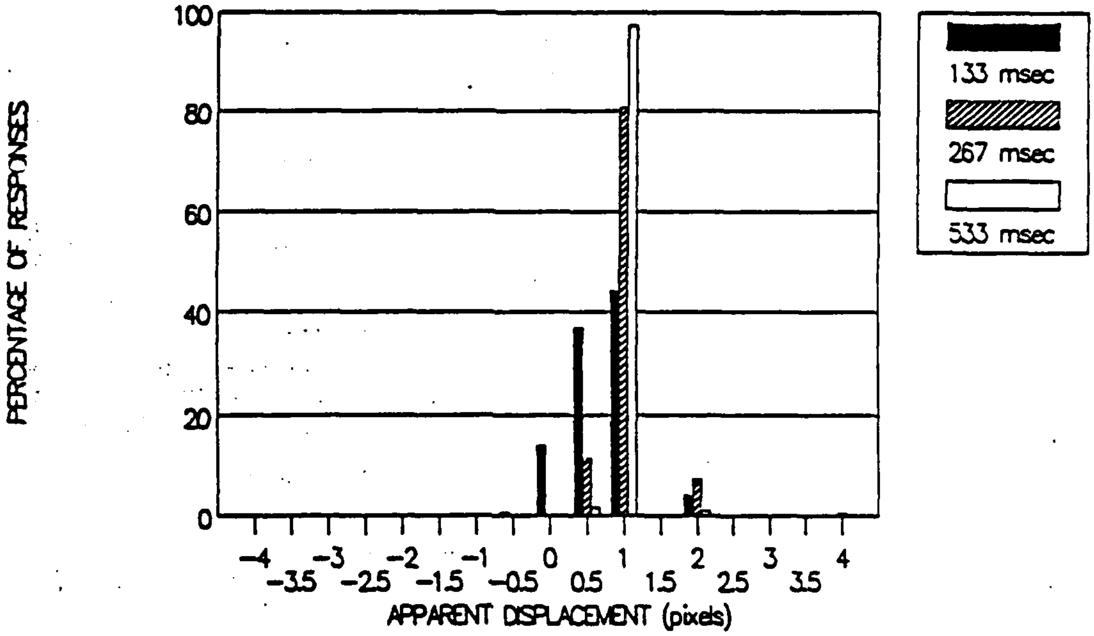
Way-off-on-the-left

Way-off-on-the-right

Figure 2

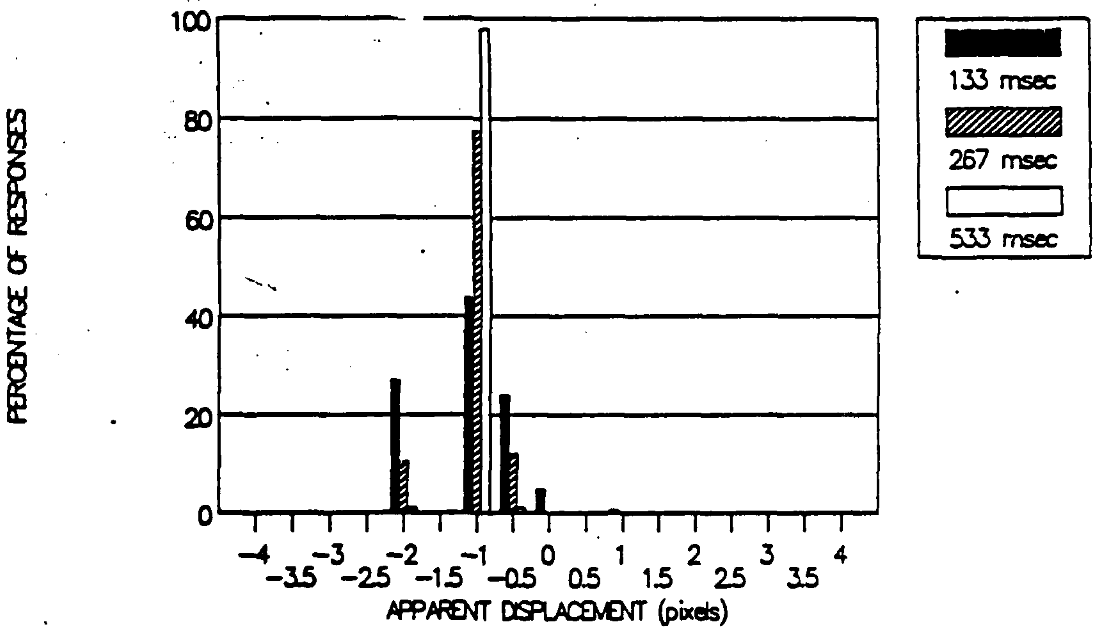
60 pixels/sec

Top First



60 pixels/sec

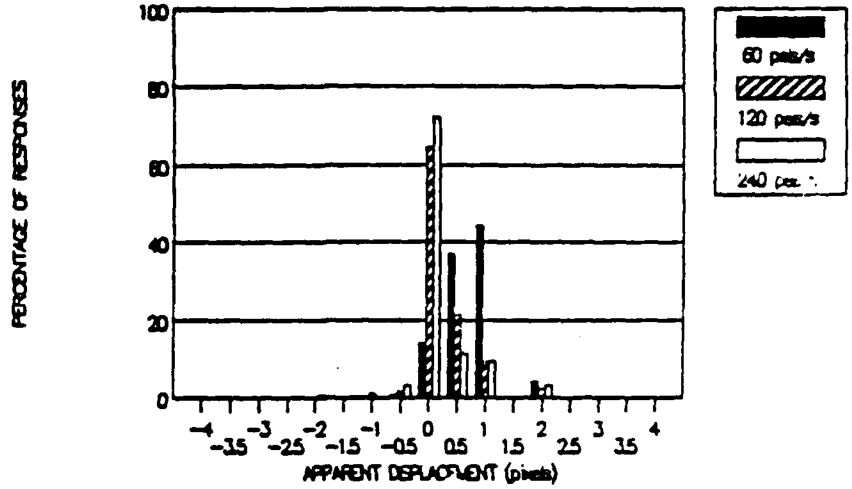
Bottom First



Duration - 133 msec

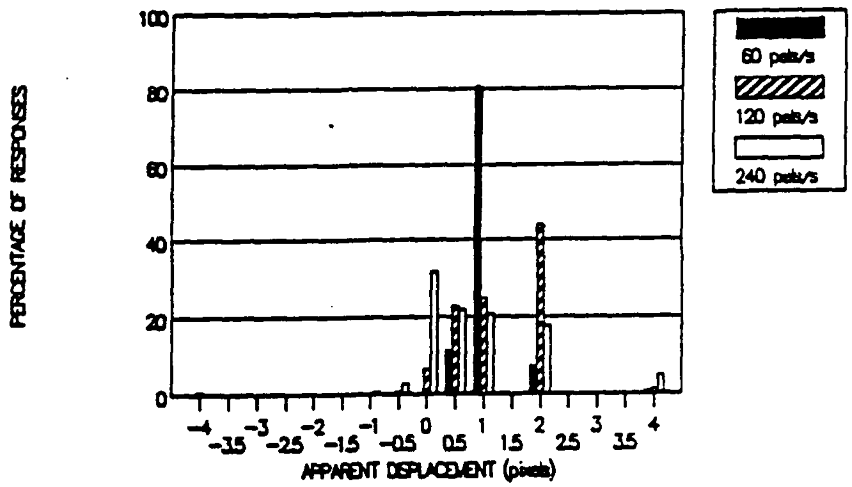
Top First

Figure 3a:



Duration - 267 msec

Figure 3b:



Duration - 533 msec

Figure 3c:

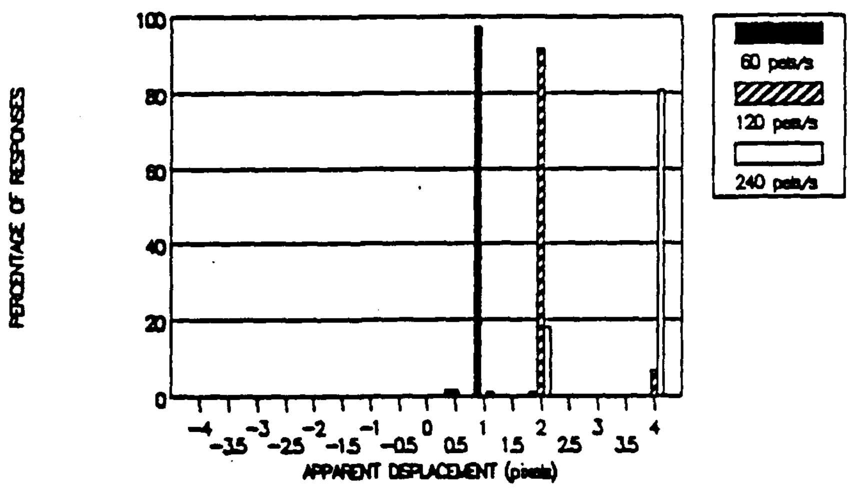


Table 1

Predicted Percent of Distortion

	No Gap Center	No Gap Offset	Gap Center	Gap Offset
133 msec	14.2	17.7	14.6	21.9
267 msec	33.3	38.9	45.8	45.8
533 msec	84.8	89.6	89.2	92.0

Table 2

Mean Percent Correct

	No Gap Center	No Gap Offset	Gap Center	Gap Offset
133 msec	43.1	43.4	43.8	38.5
267 msec	13.5	9.4	11.1	7.3
533 msec	0.0	0.0	0.0	0.0

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FINAL REPORT

OCULOMOTOR RESPONSE TO SINUSOIDAL STIMULI

Prepared by:	Peter E. Pidcoe
Academic Rank:	Doctoral Candidate
Department and	Department of Bioengineering
University:	University of Illinois at Chicago
Research Location:	AFHRL/OT Williams Air Force Base AZ, 85240-6457
USAF Researcher:	Elizabeth Martin, Ph.D.

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OCULAR MOTOR RESPONSE TO SINUSOIDAL STIMULI

Peter Pidcoe

ABSTRACT

A linear systems approach was taken to characterize the frequency response of the oculomotor system to a variety of horizontally tracked targets. The applied stimulus included single frequency sinusoids ranging in frequency from 0.1 Hz to 2.0 Hz and a random stimulus composed of six non-harmonically related sinusoids. The amplitude of all stimuli was ±2 degrees.

A discrete Fourier transform analysis (DFT) was performed on the sample population response at each frequency using the Chirp Z-transform algorithm (Rabiner, et al., 1968). The magnitude components from each response DFT were compared to actual components of the stimulus. Plotting the amplitude ratios of the single frequency stimuli provided a characterization of the frequency response of the oculomotor system. The amplitude ratios for the random stimulus were also computed and compared to the single frequency responses.

ACKNOWLEDGMENT

I wish to thank the Air Force Office of Scientific Research and the Human Resources Laboratory at Williams AFB for this research opportunity. A special thanks to Dr. Elizabeth Martin for her support and friendship.

I would also like to thank Dr. Paul Wetzel, my friend and summer advisor. The goals of this summer could not have been accomplished without his help and expertise. Finally I would like to acknowledge the efforts of my experimental observes whose time and patients were appreciated: George Kelly, Scott Smallwood, Jerome Nadel, Paul Wetzel, and Elizabeth Martin.

I. INTRODUCTION:

The Human Resources Laboratory's Operations Training Division at Williams AFB is actively involved in the research and development of advanced flight simulators for use in pilot training. One area of interest involves the measurement of combined head and eye position for control of a high resolution inset to be aligned with to the direction of gaze. This would allow the peripheral areas of a presented image to be displayed at a lower resolution and thus reduce the computational needs of the system. As a bioengineer with a background in system modeling and signal processing, I am interested in basic research to characterize the oculomotor system. This characterization may provide useful in the application of eye tracking devices to flight simulation systems.

II. OBJECTIVES OF THE RESEARCH EFFORT:

The objectives of this research effort are to characterize the frequency response for the smooth pursuit branch of the oculomotor system. The responses of both predictable sinusoidal and random stimuli will be compared. The frequency response of the oculomotor control system for steady-state sinusoidal inputs will be determined. And the differences between predictable sinusoidal and unpredictable random stimuli will be examined.

III. METHODS:

The data presented in this experiment represents the average of the responses of six subjects. During data collection, the subject's head was held in a fixed position using a bite bar attached to a table-mounted chin/head rest combination while eye position was measured. The stimulus was presented directly in front of the subject on an Hewlett Packard 1310B CRT Display at a distance of 1 meter. The display target was a point source subtending a visual angle of approximately 5 arc-minutes with an intensity of 1.8×10^{-1} foot-lamberts. To reduce the influence of target persistence, an overhead background light was used at an intensity of 0.005×10^{-1} foot-lamberts (measured at the screen). These light intensities were measured with a Photo Research Spectra Spotmeter.

The subjects (5 males and 1 female, no pilots) were dark adapted for approximately ten minutes prior to data collection. The stimulus consisted of ten single frequency sinusoids and one combination of six non-harmonically related sinusoids to create a random target movement. The frequencies of the single sinusoids were from 0.10 to 0.60 in 0.10 Hz intervals and 0.80, 1.00, 1.50, and 2.00 Hz. The amplitudes of these stimuli were ±2 degrees. The frequencies of the random stimulus were 0.12, 0.36, 0.65, 0.80, 1.00, and 1.43 Hz, each component having an amplitude of 0.5 degrees. The combination of these components provided a random stimulus with a ±2.0 degree displacement amplitude.

The order of stimulus presentation was randomized but consistent for each subject. All eleven stimuli were presented three times per subject and each stimulus was preceded by a calibration file containing fixed target positions at 0.0, ±1.5 and ±3.0 degrees. The maximum duration of each stimulus trial was limited to 30 seconds. Single frequency stimulus trials between 0.8 and 2.0 Hz were further reduced to 20 seconds. To prevent fatigue, subjects were given sufficient breaks between runs.

A differential infrared reflectance technique was used to monitor the position of the left eye. The eye position signal was sampled by a Data Translation DT2821 data acquisition board at a rate of 500 samples per second. The collected data was stored on a Compaq microcomputer for later analysis.

Data analysis was accomplished through the use of an interactive graphics display program and off-line analysis programs (Wetzel 1988). The data analysis procedure first involved an examination of each response and the removal of any blinks from the response which tend to produce false high frequency information in the analysis. After removal of the blinks, the responses of all subjects at each frequency were combined to create the sample population response. This consisted of 18 responses per stimulus trial. The sample population response file was then analysed.

The spectral analysis of the averaged data was accomplished using the Chirp Z-transform algorithm. This is a discrete Fourier transform method and has the advantage of not requiring the number of points in a data set to be equal to a power of two.

IV. RESULTS:

This section contains compiled results from the data collection and analysis methods described earlier. As an example of the sample population response to the stimuli, a graphical comparison of the stimulus and response to the 0.50 Hz sinusoidal stimulus (Figure 1) and random stimulus (Figure 2) are provided. These are typical results and illustrate the purity of the frequency response. The spectral analysis of these are displayed in Figures 3 and 4, again re-enforcing the purity of the frequency response of the oculomotor system. Figure 5 compares the response amplitudes for the single stimulus frequencies, illustrating the low pass filter characteristics of the system. Table 1 lists the response gains for all presented frequencies. The presented data is based on the average response for the sample population.

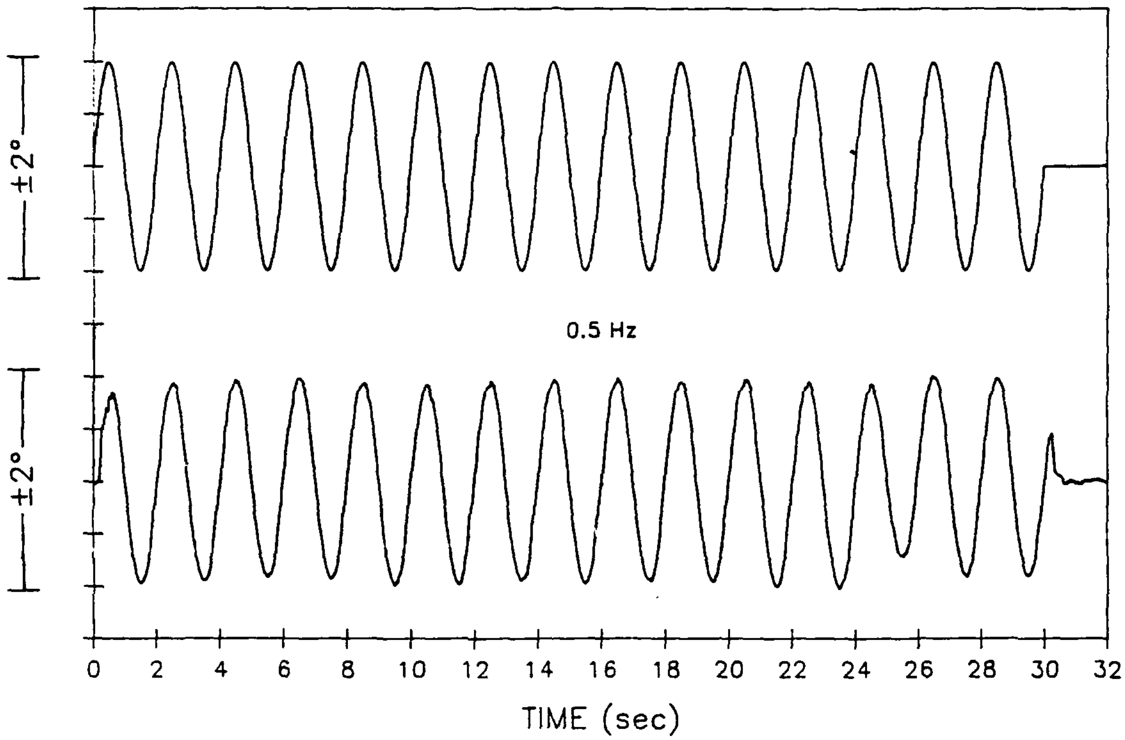

Figure 1. Sample population response to the 0.5 Hz stimulus. The stimulus is presented as the top curve. The response is the bottom curve.

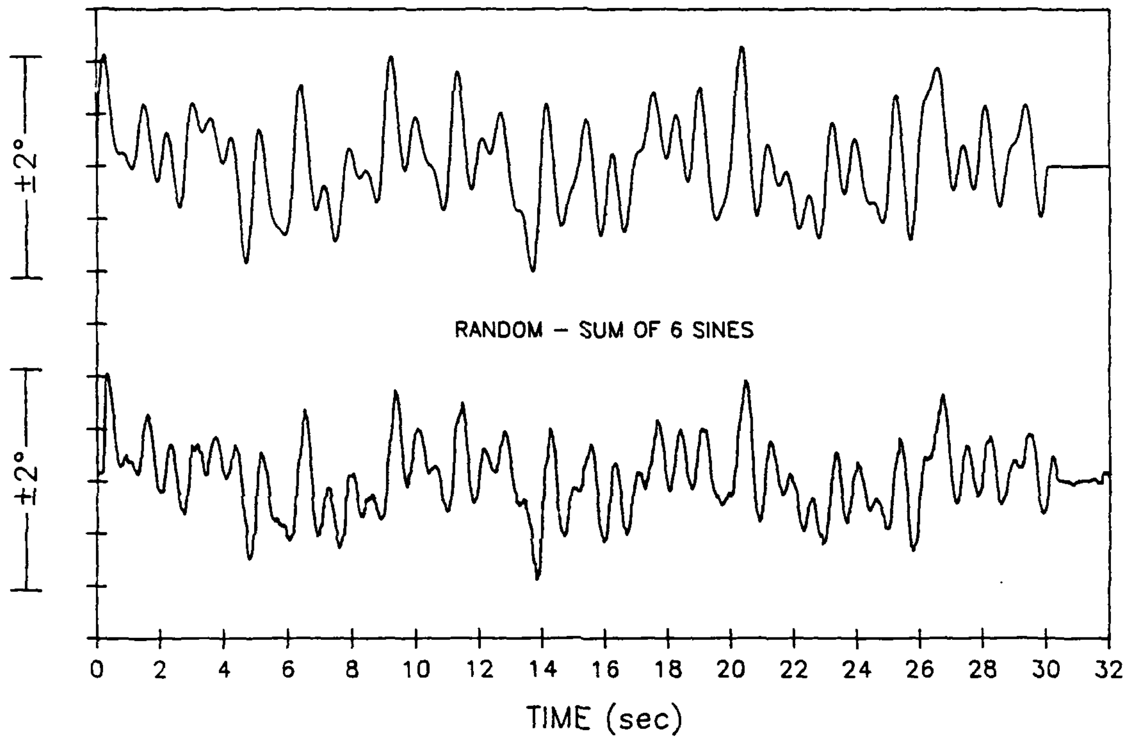

Figure 2. Sample population response to the random stimulus. The stimulus was a summation of six non-harmonically related sinusoids and is presented as the top curve. The response is the bottom curve.

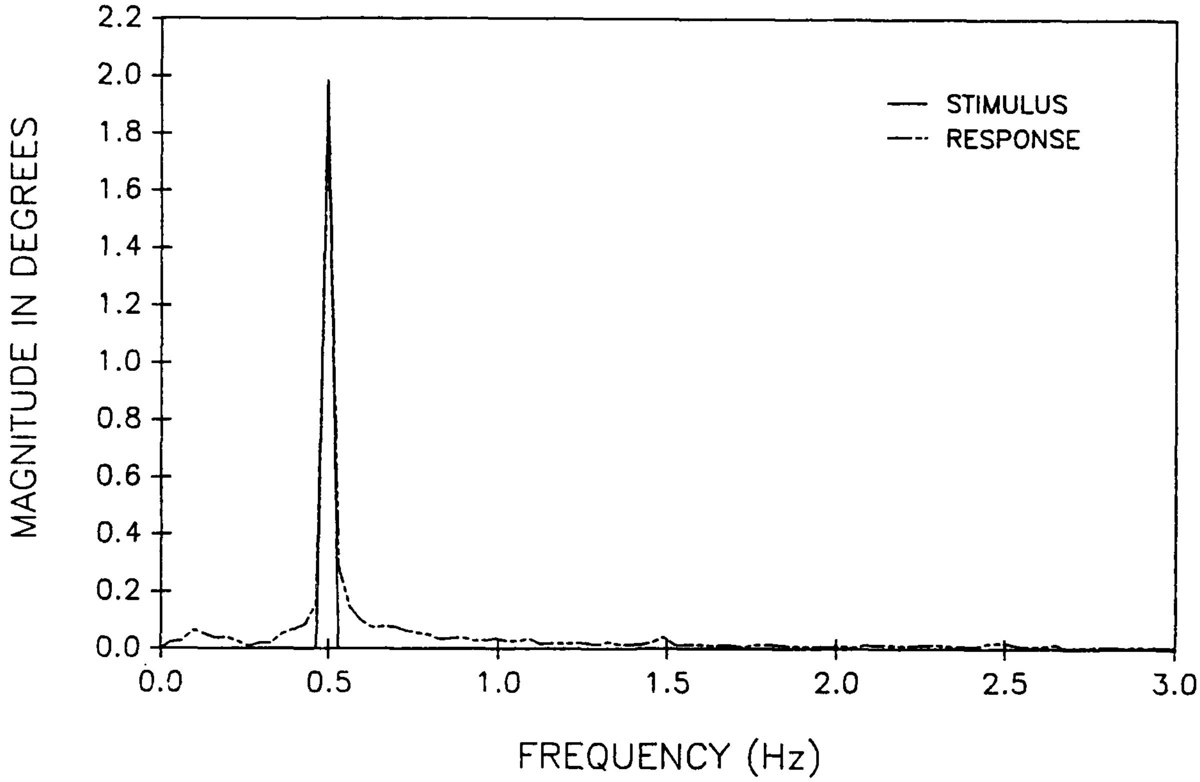

Figure 3. Comparison of the spectral components of the 0.5 Hz sinusoidal stimulus to those of the sample population response. The spectral components were generated using the Chirp Z-Transform algorithm.

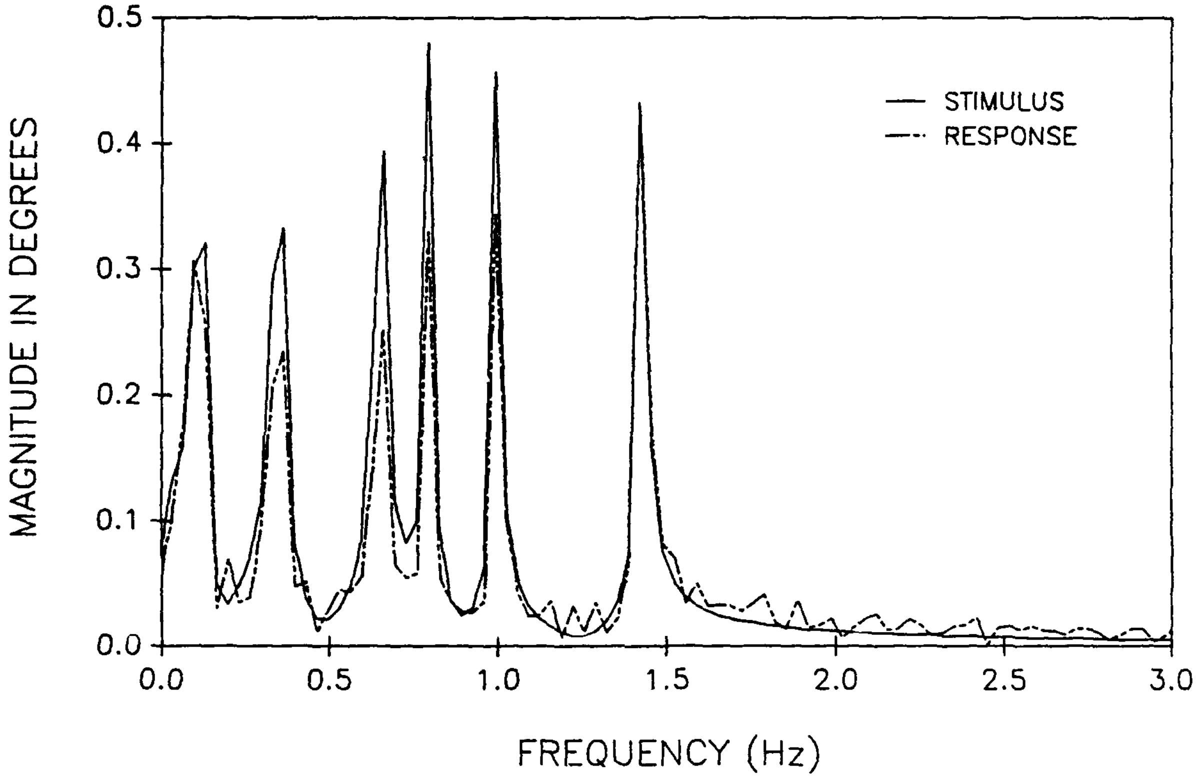

Figure 4. Comparison of the spectral components of the random stimulus to those of the sample population response. The spectral components were generated using the Chirp Z-Transform algorithm.

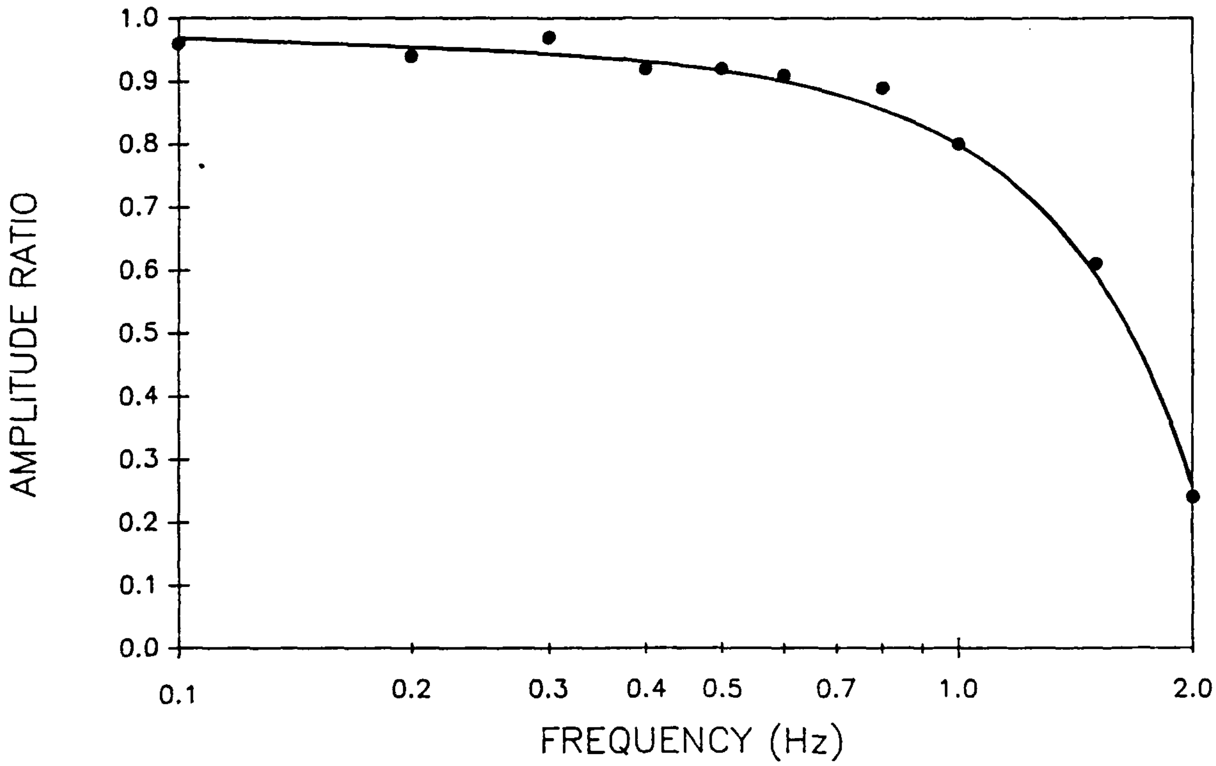

Figure 5. Comparison of the amplitude ratios of the sample population response for the single frequency sinusoidal stimulus. The cutoff frequency is approximately 1.2 Hz.

Table 1. Response gains for the various stimulation frequencies. The random stimulus contained components of each of the frequencies listed below.

SINGLE FREQ STIMULUS		RANDOM STIMULUS	
FREQ (Hz)	GAIN	FREQ (Hz)	GAIN
0.10	0.96	0.12	0.96
0.20	0.94	--	--
0.30	0.97	0.35	0.71
0.40	0.92	--	--
0.50	0.92	--	--
0.60	0.91	0.65	0.64
0.80	0.89	0.80	0.69
1.00	0.80	1.00	0.75
1.50	0.61	1.43	1.00
2.00	0.24	--	--

V. DISCUSSION:

Qualitatively comparing the sample population responses to single frequency stimuli shows the frequency response of the oculomotor system to be linear. Figure 1 illustrates the purity of the sample population response for the 0.5 Hz stimulus. This is typical for the entire range of frequencies tested. As the stimulus frequency was increased, the amplitude of the response decreased and is characteristic of a low-pass system. The response of the sample population to the random stimulus is provided in Figure 2. It is more difficult to examine the frequency response characteristics of this response due to its complex form.

A qualitative comparison of the stimulus-response data is not sufficient to distinguish measurable differences in frequency or amplitude. For this reason, the DFT using the Chirp Z-transform algorithm was introduced to allow the spectral components of each stimulus-response combination to be evaluated. A spectral analysis decomposes a data set into its individual frequency components. Each frequency component has an associated magnitude. These components can be quantitatively compared. The resolution of the analysis ranged from 0.03 to 0.05 Hz. This resolution is limited based on the sampling rate and the number of samples collected.

Ideally, the DFT of the stimulus would be a single point at the stimulation frequency whose magnitude is equal to the actual stimulus amplitude. Figure 3 is an example of the DFT for the 0.50 Hz stimulus and sample population response. Both the stimulus and the response data have a substantial frequency component only at 0.50 Hz. The component magnitude is very close to the actual 2.0 degree amplitude of the stimulus. The finite width of the component is due to the limited resolution of the analysis. The DFT of the other single frequency responses provided similar results. Evaluation of all the single sinusoid stimulus-response data provides the amplitude ratios used in later comparisons. The amplitude ratio, also refered to as the gain, is defined as the ratio of the output response to the input stimulus.

Spectral analysis of the random stimulus-response data is graphically depicted in Figure 4. Substantial components exist at each of the frequencies present in the stimulus, however the stimulus magnitudes are less than their actual 0.5 degree amplitudes. Again, this is due to the frequency resolution of the analysis and can be improved by increasing the number of collected samples. The analysis still provides a method of comparison between the stimulus and response since both undergo the same transformation.

The amplitudes of the frequency components of the sample population response to the random stimulus vary. It appears that in the random stimulus case, the oculomotor system tends to track the higher and lower frequencies at a higher gain than the intermediate ones. It is important to note that substantial components in the response data occur only at frequencies corresponding to the components of the stimulus file. This fact re-enforces the frequency linearity of the oculomotor system. Comparisons of gains for the single frequency stimulus and the random stimulus are provided in Table 1.

If the gains of the responses for the single stimulus data are plotted versus frequency, the general frequency characteristics of the system can be viewed. Figure 5 illustrates this comparison. The system was found to be band limited with a cutoff frequency of approximately 1.2 Hz. The characteristics are that of a low pass filter. These findings are consistent with Fender and Nye (1961) and Wetzel (1988). Fender and Nye (1961) also found the system to be non-linear by comparing the frequency responses of stimuli with different amplitudes.

VI. RECOMMENDATIONS:

This experiment examined the characteristic frequency response of the smooth pursuit branch of the oculomotor system for a single amplitude stimulus. It has been shown by Fender and Nye (1962) that using other stimulus amplitudes may result in similar frequency responses but with different gains, implying the system is non-linear with respect to amplitude. The sample population response to the random stimulus suggests that the oculomotor system appears to favors the tracking of the highest and lowest frequencies in a given stimulus. Modified experiments using random stimulus for various frequencies and amplitudes may result in further system characterization.

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Final Report

Prepared by:	Douglas J. Sego
Academic Rank:	Graduate Student
Department and University:	Management Department Michigan State University
Research Location:	AFHRL/IDE Brooks AFB San Antonio, Texas 78235
USAF Researcher:	Mark Teachout

Date:	22 August 1988
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AIR FORCE TRAINING EVALUATION SYSTEM: A CASE STUDY

by

Douglas J. Sego

ABSTRACT

The Instructional Systems Development (ISD) model used by the Air Force specifies a link between the training evaluation phase and the needs reassessment phase. This linkage is examined using the Aerospace Ground Equipment (AGE) career field Airman in Basic Residence (ABR) technical training course as a case study. Several sources of information were used in developing this case study. They are listed along with a description of the information they contain. How the evaluation system answers the question of content validity and over- or undertraining of the AGE ABR course is examined. The key issues are the matching information from two domains, such as the job and training content domains, and the level of specificity of that information. Limitations of the evaluation system are provided as well as future research directions.

I. INTRODUCTION:

The Instructional System Development (ISD) model is a five-step process followed by the Air Force in developing and modifying training courses. This system was developed to insure the job relevance of training and to monitor inadequacies in learning in a systematic manner (Montague & Wulfeck 1986). The ISD model is a closed loop system which specifies a connection between the evaluation phase and reassessing training needs to improve the quality of training. However, the model does not clearly specify how the link between training evaluation and training needs reassessment is made.

The Human Resource Labatory (HRL) at Brooks Air Force Base in San Antonio, Texas has been involved over the last seven years in a major research program aimed at developing job performance measures for enlisted personnel. The purpose of the Job Performance Measurement System (JPMS) project is to validate the Armed Services Vocational Aptitude Battery (ASVAB), the selection test used by all the Services, against actual job performance rather than training success. HRL personnel believe that the job performance measurement system developed for this project might also be useful during the evaluation phase of hte ISD process, and more specifically in reassessing training needs. HRL was interested in potential application of the JPMS technologies to training.

I have been working with Dr. Ford for the last two years as a research assistant. When Dr. Ford applied for the Summer Faculty Research Program he encouraged me to apply for a Graduate Student Research Program position to assist him in this research effort. My course work includes the area of training and compliments my familiarity with training systems in the military, as I spent eight years in the Navy.

II. OBJECTIVES OF THE RESEARCH EFFORT:

The objective of this case analysis is to specify how the linkage between training evaluation and training needs reassessment is currently made by addressing two questions with respect to training for Areospace Ground Equipment (AGE) specialists. First, how does the system operate to maintain job relevance in the AGE Airman Basic in Residence (ABR) technical training course? This is a content validity issue. Second, how is the issue of over- or undertraining examined in the current system? This is a training efficiency issue.

III. METHODOLOGY USED IN THIS CASE ANALYSIS:

A review of the training needs assessment, or front-end analysis literature, both military and cilivian, was conducted to determine what is currently known about the training evaluation to needs reassessment linkage. Information was also gathered on the Air Force training evaluation system and sources of data used to evaluate the AGE ABR training course.

Several site visits were made in support of this project. The first visit was to Bergstrom AFB, Texas to familiarize ourselves with an AGE shop work environment and to talk to supervisors and recent AGE ABR course graduates about AGE training, both training in the ABR course and on-the-job. The second was to Randolph AFB, Texas where personnel from the Occupational Measurement Center (OMC) were questioned about the occupational survey process and the development of the Occupational Survey Report (OSR) and the Training Requirements Analysis (TRA) report. The third visit was to Chanute AFB, Illinois Technical Training Center. Several groups were visited during our four days there, including personnel in the: Faculty Development Training Branch, Computer Based Instruction Branch, AGE Training Development Branch, Jet Engine Training Development Branch, Training Evaluation Division, and the on site OMC Detachment. A tour of the AGE training facilities was also conducted. Finally, a visit was made to the Security Police training course at Lackland AFB, Texas.

IV. CASE ANALYSIS INTRODUCTION:

The Aerospace Ground Equipment (AGE) career field maintains both powered and non-powered equipment used to support aircraft in the Air Force. The equipment varies from small jet engines used to start jet aircraft to tow bars used by ground personnel when moving aircraft. The Airman Basic in Residence (ABR) course for AGE lasts 18 weeks.

A conceptual model of the possible types of evaluative information is described by Ford (1988) and shown in Figure 1. The model contains four domains: job content domain, training content domain, job performance domain, training performance domain. Each domain contains information useful in evaluating a training course in terms of job revelancy and over- or undertraining.

The job and training content domains are conceptualized as consisting of two components: content and emphasis. The content component includes the tasks (or Knowledges, Skills & Abilities (KSAs)) that are performed on the job or taught in the training course. The emphasis component involves the importance of the tasks for job performance or the amount of emphasis placed on the tasks in training.

The job and training performance domains are conceptualized as consisting knowledge and performance components. The knowledge component focuses on how well an airman knows the correct way to perform job tasks or to what extent a trainee has learned the material covered in the training course. The performance component specifies how well an airman performs job tasks or a trainee performs the tasks taught in training.

Throughout this paper the term "task" is defined as, "a unit of work activity or operation which forms a significant part of a duty" (AFM 50-2). Tasks (T) are operationalized at different levels of specificity in different documents. Less specific than the task level is the "Cluster of Tasks" (CT) level. In this case, tasks are grouped, for example, by co-performance. More specific than the task level is the "Sub-task" (ST) level.

Here the task is broken down into requisite KSAs or the steps required to perform the task.

V. INFORMATION CONTAINED IN THE FOUR DOMAINS:

There are many sources of information about the four domains in the AGE career field. They are listed in Table 1 along with the type of information, level of specificity, and the relevant domain. The following is a detailed description of the information from the four domains identified in Table 1.

Job Content Domain

An OSR defines the job content of a career field and is based upon a comprehensive occupational survey of airmen within a target occupation. The survey is a job inventory of a career field and asks the respondents to indicate which task they presently perform. In the OSR this information is recorded in one of three time-in-service catagories: first enlistment (1-48 months), second enlistment (49-96 months), career personnel (97+ months). Tasks are analyzed to reflect the percentage of airmen performing each task in each catagory. These data provide content information about the job content domain by time-in-service.

The last OSR for AGE surveyed 2,629 airmen in 1983. About 75 senior AGE NonCommissioned Officers from different Major Commands (MAJCOM) complete two additional surveys. The purpose of these latter surveys was to gather "task difficulty" and "training emphasis" ratings information. Task difficulty is the relative difficulty to learn a task and is rated on a nine point scale from 1-extremely low to 9-extremely high. Relative difficulty represents the time needed, on the average, to learn to perform a task satisfactorily. Training emphasis is defined as the perceptions of the NCOs as to which tasks should be taught to first enlistment airmen in a structured training course. This scale ranges from 0-no training recommended to 9-extremely high training emphasis. These two pieces of data are part of the emphasis component of the job content domain. The OSR is at the task (T) level of specificity.

Another important document resulting from analysis of the occupational survey data is training extract. After the surveys have been returned and analyzed, personnel from OMC contact Subject Matter Experts (SME) to collect additional information. The SMEs go through the OSR tasks one by one and determine within which, if any, Specialty Training Standard (STS) statement the task belongs. In some cases, a number of OSR tasks are matched to one STS statement. For example, 50 OSR tasks fall within one AGE STS statement, 18A., "operational fundamentals of gas turbine compressors". Some STS statements are knowledge not task statements and so do not match with OSR tasks. The procedure is also used to match the OSR tasks to the Plan of Instruction (POI) objectives created by the AGE Training Development Branch (TDB). Information on what equipment is used in the career field is also included in the training extract. This information is broken down by time-in-service and/or by MAJCOMs.

The Specialty Training Standard (STS) lists the most common tasks, knowledges, and technical references necessary for AGE personnel to perform duties at the 3-, 5-, and 7-skill levels of proficiency (STS 423X5 Aug 1987). The level of specificity of the STS is at the CT level for tasks statements and general knowledge level for knowledge statements. Tasks are clustered mostly by co-performance. The 1983 OSR included 615 task items that defined the AGE career field. The AGE STS dated July 1982 consisted of 124 statements of which 80 were task statements.

The STS indicates which statements are ABR course requirements and the level of proficiency the graduate is expected to obtain as a result of the training. The STS is a contract between the MAJCOMs and the AGE school. It defines what needs to be trained in the ABR course and sets the criteria for knowledge and performance of the AGE ABR graduates. The statements are general in order to be as useful to as many users as possible (e.g., AGE STS statement #11. Reciprocating Engines, b. Diesel Engines, (2) inspect.).

Utilization and Training Workshops (U&TW) are held to make changes to the current STS. In December of 1983 a U&TW was held to discuss the OSR and training extract for AGE. The workshop included representatives from all the MAJCOMs, AGE technical school, Air Training Command, and OMC. The U&TW looked at the results of the OSR and training extract to determine what changes to the current STS were necessary.

Another reason for calling a U&TW is if there have been major changes in the career field that may not be reflected in the current STS or OSR. U&TWs are also called by MAJCOMs to request additional training in a course or to suggest removal of content perceived as no longer appropriate.

Training Content Domain

Training Evaluation Reports (TER) are produced annually or bi-annually by the Training Evaluation Division of each Technical Training Center. A TER has been done for the AGE ABR course every year for at least the last three years. The TER examines how well the ABR course is preparing graduates to perform their assigned tasks according to the proficiency level specified by the STS. Questionnaires are developed to collect this information with questionnaire items taken directly from the applicable STS statements.

For 1988, AGE ABR TER questionnaires were sent to 314 supervisors of recent graduates asking them how satisfied they were with the graduate's performance and if the tasks taught in the course were being utilized. If less than 80% of the graduates perform the required tasks satisfactorily then training is considered inadequate. If less than 30% of the supervisors indicate the graduates are performing a task taught in training, that task is considered for removal from the ABR course. The 1988 TER found no under-utilization of training. The TER Branch also interviewed 14 graduates and 13 supervisors in 1988 to verify the questionnaire findings. Information was also reviewed from external sources including: training quality reports, OSR, and utilization and training workshop minutes. Internal evaluation sources reviewed included: attrition rates, wing staff surveillance visits, standardization/evaluation reports, management effectiveness inspections, and student critiques.

Supervisors in the field also have a method of providing technical schools with feedback about training effectiveness in the form of Training Quality Reports (TQR). TQRs are submitted by a supervisor and report a graduate's lack of proficiency in one or more tasks or lack of knowledge that the STS states a graduate of the AGE ABR course should have. The school is required to respond to all TQRs. Since January 1985 only 7 TQRs have been submitted about the AGE ABR course.

The Plan of Instruction (POI) contains the requirements of the AGE ABR course in terms of training objectives. The POI is broken down by block and unit of instruction. Each unit has one or more objectives and includes the time spent training on the objective, the STS statement reference, STS proficiency level, and the form of criteria testing, either written or progress check. The POI is based on the STS and therefore is at the CT level of task specificity.

Training Performance Domain

In the AGE ABR course trainees are given knowledge tests at the end of each of the 11 blocks. The questions are written by the course instructors and are dirctly related to the course objectives. A major criteria for determining a good test question is when less than 50% of the trainees get the question wrong. The average grade on any given test is between 85% and 90%. Test questions are written at the ST level of specificity. In other words, questions are written from steps in Technical Orders (TO).

Every objective after Block I Unit I requires a performance progress check in the AGE ABR course. The progress check involves two or more trainees working together to perform a task on AGE equipment located at the technical school. The trainees use the relevant TO to complete the task and are observed at all times by an instructor. If no difficulties are encountered during the performance progress check the trainees are checked off as having completed the task. If the trainees do not pass, the instructor explains what was done wrong and trainees are alowed to perform the task again.

Job Performance Domain

A similar kind of performance check is conducted by the Quality Assurance (QA) branch personnel in field settings. On a regular basis, QA monitors airmen performing different tasks to check on their proficiency in that task. These are tasks they are presently certified on. QA personnel insure that tasks are completed in accordance with the procedures laid out in the TO and that there are no major discrepancies. If a major discrepancy is noted then recommendations are made which usually include decertification and/or additional training. This information is used by the MAJCOMs to monitor wing readiness, but is not presently used to provide performance feedback information to training programs. This information would be useful in determining the effective of a training program as explained later in this paper.

As part of the Air Force's Weighted Airmen Promotion System (WAPS), Speciality Knowledge Tests (SKT) are developed for each career field. Senior NCOs write test questions sampling the STS subject matter areas judged as appropriate for promotion to higher grades. These tests are given to airmen eligible for promotion. The tests items could be useful in identifying tasks that may require more training or tasks that less time could be spent on during training. However, at the present time this information is not used to evaluate training programs.

VI. LINKAGE OF EVALUATION INFORMATION WITH NEEDS REASSESSMENT:

When evaluating a training course, for issues of job relevancy or over- or undertraining, information from the training content domain must be linked with information from the job content or job performance domains. One way of making this link is a technique called the Matching Technique (Ford and Wroten 1984). It is important that the information being matched is at the same level of specificity (e.g. if task level of specificity describes the job content domain then the training content domain must also be at the task level). The type of information that is gathered depends on the purpose of the evaluation.

One of the purposes of an evaluation system is to insure that the training content is job relevant or content valid. This is one of the purposes of the AGE evaluation system and an example of how this is presently accomplished follows.

In 1983 a new OSR and training extract was completed for the AGE career field. The task information is used to make training content decisions. Air Training Command (ATC) has set a criterion standard to use when making decisions about which tasks to include in an ABR training course. If the OSR reports that 30% of the airmen in their first enlistment perform a task then the task is considered for possible inclusion in the course. Also taken into consideration by the AGE Training Development Branch (TDB) is the task difficulty and training emphasis ratings.

The decision to include or exclude tasks from the AGE ABR course is made a U&TW. A U&TW for AGE was last called in 1983 to examine the new information in the OSR and compare it to the existing STS and POI. According to the minutes of that meeting most of the recommendation for changes to the AGE ABR course and STS were made by the workshop attendees and support for the changes came from the OSR.

After the U&TW is completed the Training Development Branch (TDB) examines the new STS and the training extract to identify the STS statements that apply to the AGE ABR course. OSR tasks that are listed under the applicable STS statement are reviewed in the following manner. If a task is performed by over 50% of the airmen during their first enlistment then that task is recommended for knowledge and hands on training. If the percentage performing is between 30 - 49% then the task is recommended for knowledge training only. Below 30% tasks are generally not taught in the ABR course. The task difficultly and training emphasis ratings are also considered. A task is not recommended for training, no matter how high the percent performing rating is, if the task difficulty rating is statistically one standard deviation below the mean. A task is recommended for training if the training emphasis rating is statistically one standard deviation above the mean. The final criteria is the AGE school's ability to teach the task given the constraints the school faces with reference to time, equipment, qualified instructors, and building space.

The review process greatly reduces the number of OSR tasks that are applicable to the AGE ABR course. The TDB personnel then take the STS and the OSR tasks recommended for inclusion in training and make revisions to the ABR course. This includes writing objectives for the ABR course. The objectives are written from the STS statements and not from the OSR tasks. The OSR tasks are not referenced by either the STS or the POI.

At this point there is a change in the level of specificity of the information. The OSR is at the task (T) level i.e., "adjust turbine engine fuel system components" and "remove or install turbine engine atomizers". The STS statement is at the less specific cluster of tasks (CT) level. The two tasks mentioned above are clustered, along with 48 others, under AGE STS statement number 18A, "operational fundamentals of gas turbine compressors".

To cover all the material taught under a single STS statement more than one objective is used. An example of an objective for AGE STS statement 18A is, "Without reference, identify basic facts pertaining to compressor section components. A minimum of 9 of 12 components must be correctly matched to their functions".

This is the procedure used when an OSR has been completed for the AGE career field. At the present time there are no plans to create another OSR for AGE. Since the time between OSRs is often several years, another evaluation process is used to insure the continued job content revelance of the AGE ABR training course. This evaluation is conducted by Chanute's Training

Evaluation Division and results in a TER.

A TER was recently completed (2/88) for the AGE ABR course. After reviewing the findings of the questionnaires, interviews, and internal and external feedback sources, the training evaluation personnel made recommendations to the TDB at the AGE school. In this TER one of the action items, in responce to comments made by four supervisors, was to, 'make students more aware of non-powered aviation ground equipment in the field'. It is important to note that the AGE school is required by ATC to respond to the recommendations made in a TER.

In this case they included an orientation lesson in block I of the course and noted that there is no corresponding statement in the AGE STS so no further action was taken. This is an example of job content domain information being added to the ABR course by SME request and not by a more systematic means. It is not clear if this new course content was matched to applicable OSR information i.e., task difficulty and training emphasis ratings. The evaluation found that training was being utilized and the supervisors were satisfied with the graduates' performance. As a result no additional changes were recommended based on the TER questionnaire.

VII. LIMITATION OF THE AGE EVALUATION SYSTEM:

One of the objectives of this paper was to analyze how the AGE evaluation system evaluated training efficiency. This is done by answering the question, "is the training program over- or undertraining certain tasks"? The information required to answer this question is the importance of a task in the job content domain and the emphasis placed on that task in the training content domain (see Ford 1988). This analysis is not possible because the information needed is not available in a useful form at this time.

The training emphasis rating found in the OSR is the supervisor's perception of which OSR tasks should be taught in the AGE ABR course. The problem is that the training content domain has no comparable training emphasis information. Instructor ratings of the emphasis placed on tasks during training or the actual time spent in training on tasks are two ways of collecting emphasis information in the training content domain. However, this would not completely solve the problem. There is no document that lists what OSR tasks finally get incorporated in the ABR course.

Once the OSR information is matched with the appropriate STS statement and POI objective in the training extract the information is lost in the system. In the example given above the AGE STS statement 18A had 50 OSR tasks associated with it. Each OSR task was checked to determine if it should be included in the AGE ABR course. This process reduced the list of associated OSR tasks from 50 to an unknown quantity. Unknown because the OSR tasks that are finally included in the STS and in the training program are not recorded in any document. The STS statements make no references to the OSR tasks. This is not to say the tasks are not included in the training program, only that once included the OSR tasks are absorbed into the appropriate STS statement making references to those OSR tasks in the training content domain extremely difficult.

Similar problems are encountered when evaluating the content validity of the AGE ABR course. OSR tasks identify the job content domain, while POI objectives identify the training content domain. The OSR tasks and the POI objective, however, are not in forms that allow direct comparisons to be made. Content validity is consquently evaluated using STS statements. At a more general level of specificity the STS identifies both the job and training content domains. This is the level of specificity where AGE training evaluations are conducted. At this level the TERs do a good job of evaluating the content relevance of training. Evidence suggests that the job content domain as identified at the STS level is highly matched (content valid) with the training content domain as specified at the STS level. One could ask the question, if information were at the task level rather than the CT level would this provide more useful information about content validity of training? Thus training omission or training contamination could be identified.

As mentioned earlier the OSR tasks are not referenced in either the STS or the POI. One consquence of not referencing OSR tasks in the STS or the POI is that when a new OSR is created there is no simple, systematic way of incorporating the new information into the evaluation system. Judgements about where an OSR tasks belongs must be made by SMEs. Instructors knowledge of the course content is required to determine if an OSR task is currently being taught in the AGE ABR course. A situation that exacerbates the problem is the duty rotation of instructors. A set of instructors involved in transposing information from one source to another will be able to explain the linkages between documents. After this set of instructors leave, this information is lost to the evaluation system.

Another possible limitation to the current evaluation system is in not using performance information. Although performance information is available in both the job and training content domains, the information is neither used nor collected in a form that makes it useful to the training evaluation system. Performance information would be useful in determining over- or undertraining of tasks in the AGE ABR course. Per,ormance information could also be used as an indication of readiness or to identify the strengths as well as the weaknesses in a training program.

VIII. FUTURE RESEARCH:

The Job Performance Measurement System (JPM) developed by HRL could provide valuable information about over- or undertraining in a training program. The JPMS is of known reliability and validity and provides information at the ST and T levels of specificity. To be useful, however, the training content domain would require information at the same level of specificity. A methodology for doing this would have to be developed. Once this information was available in the training content domain the next step would be to develop a matching methodology. Finally, once the information was matched, criteria for making decisions about changing course content based on this information would be needed.

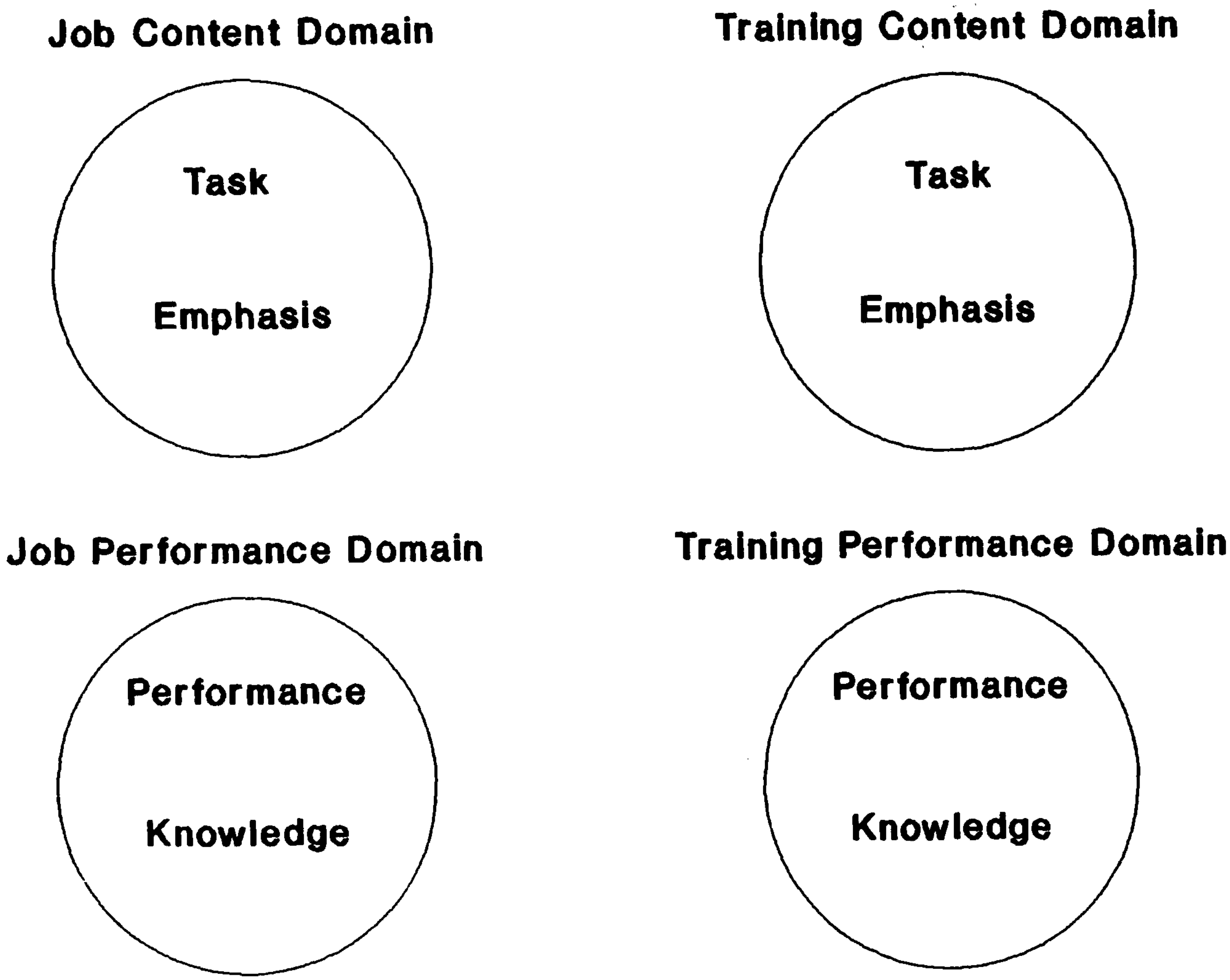

Figure 1. The sources and types of information available for evaluating a training program.

Table 1. The Available Sources of Evaluation Information.

Domain	Source of Information	Type of Information	* Level of Specificity
Job Content	OSR	Task	T
		Emphasis	T
	STS	Task	CT
		Knowledge	CT
	U&TW	Task	CT & T
Training Content	POI	Task	CT
	TER	Task	CT
		Performance	CT
	TQR	Task	CT
		Performance	CT
Job Performance	QA Performance Check	Performance	T & ST
	SKT	Knowledge	ST
Training Performance	Block Tests	Knowledge	ST
	Performance Progress Checks	Performance	T

* T => Task, CT => Cluster of Tasks, ST => Sub-task

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Training Extract	AFPT 90-423-459	May 1983
Training Evaluation Report	TER C 85-5	February 1986
Training Evaluation Report	TER C 86-28	February 1987
Training Evaluation Report	TER C 87-22	February 1988
AGE Utilization and Training Workshop	Minutes	December 1983
Study Guide/Workbook	C3AIR75135	June 1987
Plan of Instruction	C3ABR42335	August 1987
Training Plan	C3ABR42335	June 1987
Istructional System Development	AFM 50-2	July 1986

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1988 USAF-UES SUMMER FACULTY RESEARCH PROGRAM/

GRADUATE STUDENT RESEARCH PROGRAM

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Conducted by the

Universal Energy Systems, Inc.

<u>FINAL REPORT</u>

EVALUATION OF T-9 NOISE SUPPRESSOR SYSTEM AT McCONNELL AFB, KANSAS

Prepared by:	Phyllis Y. Keys, B.E.
Academic Rank:	Graduate Student I
Department and	General Engineering
University:	University of Illinois, UIUC
Research Location:	USAFOEHL/ECH
	Brooks AFB
	San Antonio, TX 78235
USAF Researchers:	Major John Seibert
	MSgt Abel de la Rosa

Date:	August 12, 1988
Contract No:	F49620-88-C-0053

Evaluation of T-9 Noise Suppressor System at McConnell AFB, Kansas

Phyllis Y. Keys

ABSTRACT

This report presents the topics of research studied during the Graduate Student Research Program which include the following: the effects of noise on humans, effective methods of measuring the noise and its effect, a description of a T-9 Noise Suppressor System, and a description of an experiment measuring nitrous oxide leakage in dental masks. Also included is sample output of a noise measurement from the T-9 Noise Suppressor System (T-9NSS) at McConnell AFB. Recommendations which should improve the quality of work at the laboratory in which I worked, as well as recommendations for the Research Program, are made at the end of this report.

ACKNOWLEDGMENTS

I want to thank the Occupational and Environmental Health Laboratory and the Air Force Office of Scientific Research for sponsorship of this research. I also want to thank Universal Energy Systems for giving me the opportunity to gain this experience.

The experience was very rewarding. I gained a wealth of knowledge. Having had no previous experience in noise evaluation, I was at first hesitant about the project. But, Major John Seibert and the acoustical technicians sparked an interest which will undoubtedly lead to further studies in this area. The help of Capt. Terry Fairman, Major Ellis, Lt Winston Shaffer, Capt. Carpenter, and MSgt. de la Rosa was greatly appreciated. The support of the General Engineering department at the University of Illinois also made this effort possible.

I. INTRODUCTION

High levels of noise and lengthy exposure to noise have been areas of concern for the United States Air Force since as early as 1956. Due to the increasing awareness of the potential for hearing loss on bases which have aircraft testing, noise suppressor systems are being built to reduce the noise levels to which workers and the surrounding communities are exposed.

The Noise Section of the USAF Occupational and Environmental Health Laboratory at Brooks Air Force Base is primarily a consulting branch for the entire United States Air Force. Consultations on recommended noise levels and methods of measuring the existing noise levels are given on a daily basis. Measurements and reports are their main objectives. Therefore, any methods which will improve techniques of data manipulation are highly desirable. Required skills include the ability to use state-of-the-art equipment and knowledge of extraneous factors which may affect noise measurements.

My research interests include evaluation of any hazard, danger, or potentially harmful work situation. The increasing number of cases involving hearing loss compensation makes noise an area of concern for all industry. My studies of the legal aspects of engineering design led me to the Occupational and Environmental Laboratory.

II. OBJECTIVES OF THE RESEARCH EFFORT:

Noise is defined as unwanted or unpleasant sound. Lengthy exposure or exposure to extremely loud levels of noise can cause irreparable damage to the human ear. When the Worker's Compensation Act was first passed, hearing loss and hearing damage were not even considered to be an injury for which workers could be compensated. Later, the amount of hearing loss that could be attributed to occupation was still questionable. And although a worker who had suffered hearing loss was awarded compensation in 1948 by the New York Court of Appeals, it wasn't until eight years later that Air Force Regulation 160-3 developed a program which would protect military personnel against noise hazards. This Air Force Regulation, however, preceded the Department of Labor's 1969 noise regulation to protect government workers.

Because of the increasing awareness of the possibility of noise-induced hearing loss occurring among industrial workers, measures are now being taken within the Air Force to decrease the both the length of time and the levels of noise to which workers are exposed. These measures include ear protection devices such as muffs and engineering control devices, to include the A/F32T-9 Jet Engine Noise Suppressor System.

Before the design of the A/F32T-9 Noise Suppressor System, a building referred to as a Hush House was used to test engines. The Hush House was used predominantly to test engines that were still installed in the plane. However, it is obvious that the size of any building is limited by economics, and engines which were used in larger aircraft had to be removed for testing. In order to be more cost effective, the Air Force asked for the design of a smaller building to be used mainly for large aircraft engine testing. Authorization for the building of twenty-six A/F32T-9 Large Turbo Fan Engine Enclosed Noise Suppressor Systems was given. The A/F32T-9 Jet Engine Noise Suppressor System is a building designed to protect the surrounding area from noise. Commonly called the T-9 Noise Suppressor System(T-9NSS), this suppressor system encloses a single engine and uses an air-cooled exhaust system to quiet noise levels.

The first of the twenty-six buildings was constructed at McConnell Air Force Base, Kansas in January, 1986. Although evaluations reported in May of 1987 showed that the noise levels at 100m from the building exceeded the established criterion of 77dB(A), the data was not considered highly reliable because of meteorological conditions and because of the reflection of acoustic waves off buildings located near the T-9NSS. After the second T-9NSS was built at Sky Harbor International Airport in Phoenix, Arizona, an evaluation was made of the noise levels there. It was found that even with stable meteorological conditions, the noise criterion was exceeded at nineteen of the twenty-four sampling positions.

The two noise surveys mentioned above were done primarily to determine whether building specifications had been met. Because of the high noise levels measured, a desire to evaluate the noise levels to which workers and the surrounding community were exposed led to a second evaluation of the McConnell T-9NSS.

The data was collected for eight engines at three power settings and eight different positions for a total of 192 data files. The data was stored on the diskettes of a Hewlett-Packard desk-top computer. USAFOEHL, Noise Section, has only one HP model computer. A Basic program transfers the data from the HP to the main VAX computer system for use at the Digital VT220 and VT240 terminals available to the other consultants in the division.

My task was to research VAX capabilities and the various computer packages available to the Occupational and Environmental Health Laboratory to determine the quickest and most efficient method of processing noise data similar to that gathered at McConnell AFB. This task gave me the opportunity to use some of my own skills while being taught the methods of a noise evaluator and becoming familiar with the equipment used in noise evaluations. I was also introduced to research methods used by several divisions of OEHL at Brooks AFB, and a second task of aiding in the data gathering for an evaluation of the effect of sterilization on the efficiency of a dental mask to control nitrous oxide leakage was assigned.

This report will consist of a discussion of the importance of noise evaluation and its impact on occupational safety, an explanation of how the original task was completed, a description of the second task, and the required section on recommendations which will include an evaluation of the relevance of my tasks at Brooks to my career objectives.

III. EFFECTS OF NOISE

The concern about noise exposure is very real. Noise can affect a human both physiologically and socially. Some problems arising from lengthy exposure and/or exposure to high levels of noise include tinnitus (ringing in the ears), paracusis (hearing inappropriate pitches), speech misconception, and loss of hair cells in the inner ear. The loss of these hair cells will cause permanent loss of hearing. Another problem that exists is a tendency to adjust to the noise level of your surroundings. This is called a temporary threshold shift. A temporary loss of hearing occurs. But, after time away from the source of the noise, the hearing level readjusts. Studies have shown that after repeated daily temporary shifts, a permanent threshold shift occurs and hearing is not regained.

The concern for human welfare is not the only factor in evaluating noise. The legal aspects of not evaluating noise play a major role. After a worker was compensated for hearing loss which may have been caused by his occupation, the Air Force began to consider what effects their operations might have on the hearing of military personnel. Soon after this, groups to assist industry in controlling noise exposures were created. Among these agencies are: the National Research Council Committee on Hearing, Bioacoustics, and Biomechanics (CHABA); the Occupational Safety and Health Administration (OSHA); and the American National Standards Institute (ANSI). Most states include occupational noise-induced hearing loss as compensable. Compensation can be as much as $33,000 for hearing loss in both ears.

Air Force Regulation 161-35, Hazardous Noise Exposure, requires ear-protection devices for all personnel exposed to noise levels exceeding 84 dB(A). OSHA regulations governing the civilian sector require hearing protection for levels exceeding 90 dB(A). Noise regulations are becoming more stringent as the number of cases of hearing loss increase.

Although engineering controls have been used to limit noise exposure in most industrial settings, the noise levels are often excessive even with the controls. Ear-protection devices have been developed to provide greater attenuation to workers who must be in the area of the noise source.

The anechoic chamber is a room used in the testing of protective devices. The walls, floor, and ceiling of this room are covered with absorptive material to eliminate reflections. Workers are placed in the chamber and asked to respond when a sound is heard. The anechoic chamber helps to cut out background noise during these tests.

To measure the noise exposure of an individual, a noise dosimeter is attached to the body of the person. Readings are taken for a specified period of time, and the average noise exposure is calculated. To measure the noise level of a particular room or area, sound level meters can be used for a quick analysis of whether the room has hazardous noise areas. For the measurements taken at McConnell, microphones were used to record the noise, and the recordings were analyzed in the lab using a spectrum analyzer.

ECH, the division of the Occupational Environmental Health Laboratory which houses the noise section does measurements of the noise levels. The Audiology Department does hearing tests on individuals to determine the effect of noise exposure and the quality of ear-protective devices. The Hearing Conservation Data Registry has been set up by the Audiology department to evaluate the effect of the hearing conservation program within the Air Force. Measurements of hearing before duty in the Air Force, during duty, and at the end of duty are taken. These measurements aid in determining if protective devices should be used and provide valuable information in the event of a compensation claim being filed.

IV. RESEARCH EFFORT (T-9NSS)

The research effort on the T-9 Noise Suppressor System began with becoming familiar with the procedures of the Air Force team which gathered data at McConnell AFB. Microphones were set up at various locations inside the T-9NSS and outside. Recordings of the noise levels were taken for a J-57 engine at three power level settings: idle, intermediate, and military. Pictures of the setup were taken by the evaluation crew.

Located at the front and the rear of the T-9NSS are acoustical attenuators which dampen the effect of the noise from the engine. An exhaust augmentor located at the rear on the exterior acts as a muffler for noise leaving the building. The engine is placed inside the T-9NSS by workers, but it is controlled by personnel stationed in a trailer that is not a part of the building. The observation windows on each side of the building provide visual access to the operation of the engine. For personnel who do enter the building during operations, earmuffs and plugs are required.

My tasks in this project included finding a method of speeding up the processing of data from the noise evaluations. Using the VAX system, a spreadsheet (DECalc), and a graphics package (Tellagraph), Major Seibert and I created a command file to process the noise data. We began with twenty-four data files from McConnell. Using the command files, we were able to compile the tables of data and files to print graphs for all twenty-four data files were compiled in less than one hour. Sample output from one file is shown in Figures 1 and 2.

A second task in the area of noise was the testing of the reverberation chamber. A reverberation chamber is a room with no parallel surfaces in which all surfaces are made as hard and reflective to noise as possible. Its purpose is to create a diffuse field in which the noise is uniformly distributed within the room. Measurements at any point in the room should be the same. The reverberation chamber at Brooks is used predominantly to test and calibrate equipment. Before using it as a standard calibrator, it must be certified. The certification process includes running tests to ensure that there are not large variations in measurements at different points in the room.

A microphone was moved to different locations in the room with a sound source operating. Measurements using a noise dosimeter were taken at each location. My task included calibrating the instruments used in the testing, setting up the equipment, and taking measurements.

V. RESEARCH EFFORT (NITROUS OXIDE/DENTAL MASK)

Evaluation was made of three types of masks used in administering nitrous oxide to dental patients. The Miran I Variable Filter Infrared Analyzer was used to measure nitrous oxide gas leakage. After preliminary testing, two of the masks were eliminated from the experiment. The objective of the experiment was to determine whether sterilization of the masks had any effect on leakage. A chamber constructed by Capt. Carpenter was used to hold the head of a mannequin and an air pump. Two masks of the same type were sterilized five times before each day's measurements. After one of the masks was placed onto the nose of the mannequin, three liters of nitrous oxide and three liters of nitrogen were then pumped into the mask. The pump was placed into the chamber to move the air from the chamber to the Miran Analyzer. A vacuum was used to remove excess nitrous oxide from the chamber. Actual measurements in a dental office at Wilford Hall on Lackland AFB will be taken at a later date. My tasks included recording data and observing the process.

VI. RECOMMENDATIONS (AND RELEVANCE OF EFFORT)

Recommendations for noise evaluations include: (1) A standardized format for all evaluations of similar nature should be enforced, and (2) A training course or one manual for each terminal should be provided for all VT users.

I feel that the noise evaluations are a necessary part of occupational safety enforcement. But, the technicians and engineers who perform these evaluations should receive more training in reporting techniques, technical writing, and computer aids.

Although I do not feel that my research this summer has been highly technical in nature, I do feel that I have benefited from the Graduate Student Research Program. The consultants with whom I worked were engineers with years of experience and knowledge. Discussions of their thesis projects and other projects completed while at Brooks were highly educational and interesting.

In the original application for GSRP I indicated that the program would aid in my search for a thesis topic. Although I have not confirmed a topic, that goal is closer.

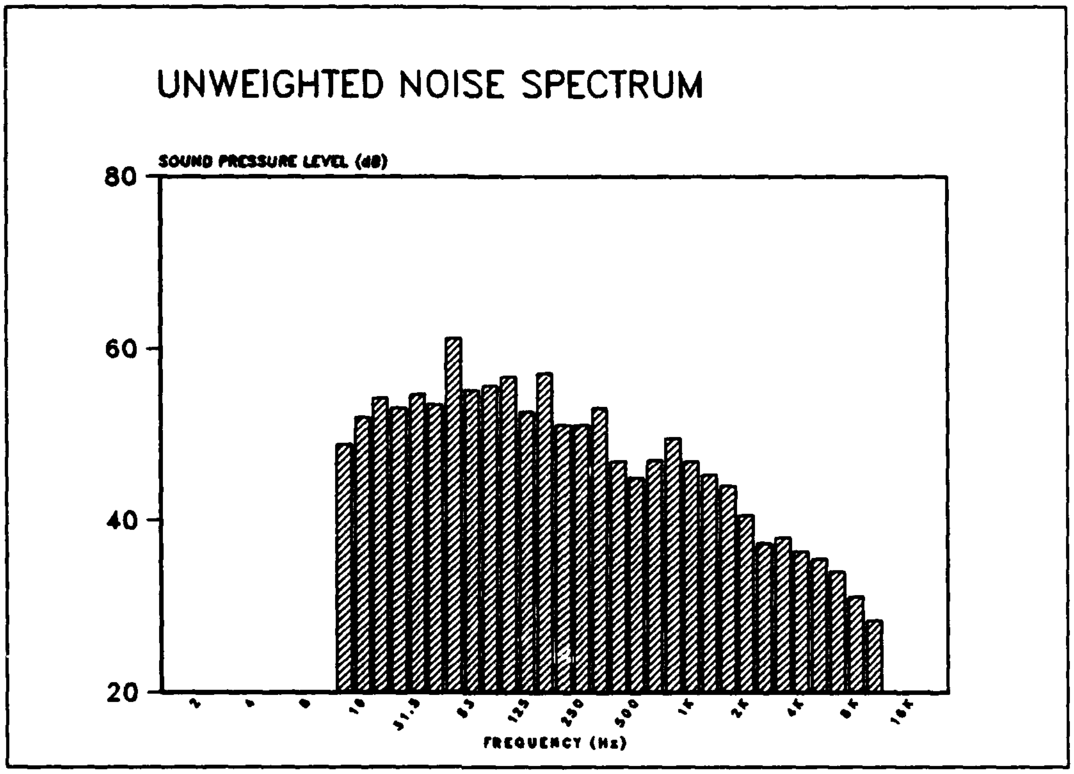
TABLE 1: MEASURED NOISE SPECTRUM LEVELS.
LOCATION: A/F32T-9NSS. McCONNELL AFB KS.
POSITION: PMEL
ENGINE: J-57; POWER: BACKGROUND NOISE.

FREQ (Hz)	SOUND PRESSURE LEVEL (dB)	A-WT SOUND LEVEL [dB(A)]	C-WT SOUND LEVEL [dB(C)]	OCTAVE BAND SPL (dB)	A-WT OCTAVE BAND SL [dB(A)]	C-WT OCTAVE BAND SL [dB(C)]
12.5	48.8	0.0	37.6			
16	51.9	0.0	43.4	56.9	6.4	49.6
20	54.2	3.7	48.0			
25	53.0	8.3	48.6			
31.5	54.6	15.2	51.6	58.5	20.6	55.5
40	53.4	18.8	51.4			
50	61.2	31.0	59.9			
63	55.1	28.9	54.3	63.0	36.1	62.0
80	55.6	33.1	55.1			
100	56.7	37.6	56.4			
125	52.6	36.5	52.4	60.7	45.3	60.5
160	57.1	43.7	57.0			
200	51.1	40.2	51.1			
250	51.0	42.4	51.0	56.6	48.5	56.6
315	53.0	46.4	53.0			
400	46.8	42.0	46.8			
500	45.0	41.8	45.0	51.1	48.0	51.1
630	47.0	45.1	47.0			
800	49.5	48.7	49.5			
1000	46.8	46.8	46.8	52.3	52.1	52.3
1250	45.3	45.9	45.3			
1600	44.0	45.0	43.9			
2000	40.6	41.8	40.4	46.2	47.3	46.1
2500	37.3	38.6	37.0			
3150	38.0	39.2	37.5			
4000	36.4	37.4	35.6	41.5	42.5	40.8
5000	35.5	36.0	34.2			
6300	34.1	34.0	32.1			
8000	31.1	30.0	28.1	36.6	35.9	34.0
10000	28.3	25.8	23.9			

OASPL = 67.1 dB OASLA = 56.1 dB
OASLC = 66.0 dB C-A = 9.9 dB

FIGURE 1: TABULAR OUTPUT FOR NOISE DATA
(ONE LOCATION ONLY)

TABLE 1: T9NSS NEAR FIELD NOISE
J–57 ENGINE AT BACKGROUND AT PMEL



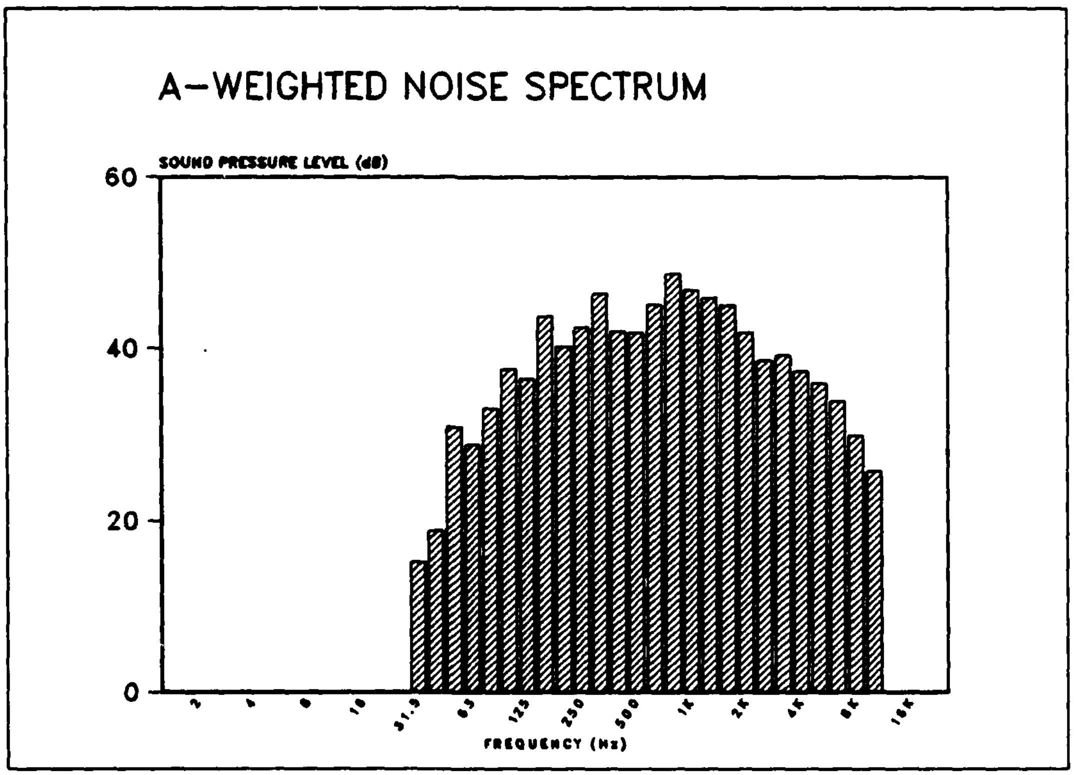


FIGURE 2: GRAPHICAL OUTPUT FOR NOISE DATA
(ONE LOCATION ONLY)

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MR. JOHN BARNABY

FINAL REPORT NUMBER 90

NO REPORT SUBMITTED

MR. OTIS COSBY

FINAL REPORT NUMBER 91

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FINAL REPORT

Standardization of DAIM Precipitate

Prepared By:	Jerry Dillon
Academic Rank:	Graduate Student
Department and University:	Denistry Meharry Medical College
USAF Researcher:	Dr. John Wright Brooks AFB, TX 78235-5301
Date:	30 September 1988
Contract No.	F49620-88-C-0053

Jerry Dillon

STANDARDIZATION OF DAIM PRECIPITATE

Abstract

DALM (Diazoluminolmelanin) a luminol derivitive has recently been discovered. This presentation marks the initial quantitating for this oxidize free radical scavenger in the presence of H_2O_2 and HRP (Horseradish Peroxidase). Quantitation results are compared to the mother compound luminol as a means to guage efficiency of use.

Acknowledgments

I would like to take this opportunity to thank the Air Force Systems Command, Air Force Office of Scientific Research, Dr Johnathan L. Kiel for this learning opportunity to grow educationally and develop into as sound a professional as they have all been this summer of 1988. I would like to give special thanks to Meharry School of Dentistry, to my family, and a special friend Mercedes Maynor for being such an inspiration.

Standardization of DAIM Percipitant

DAIM (Diazoluminomelanin) a derivative of Luminol (5-Amino -2, 3 dehydrophtholazine -1, 4-dione) as a light emitter has been the article of interest for the development of a summer project for U.E.S.

DAIM an in-house development has been found to possess qualities ideal for quantitating intracellular activation of certain cells present in the R.E.S. (Reticular Endotheliol System). There exists a large body of data which has clearly documented the existence of a "respiratory burst" of metabolic activity associated with phagocytosis [1]. The basic fundamental physiological significence of this phenomenon of the respiratory burst, with its consequent generation of reactive species of oxygen metabolites, is to provide the phagocytes with an efficient microbicidal mechanism [1].

Luminol had previously inherited the duty of quantitating the "respiratory burst" in phogocytes. The burst is characterized by a sharp increase in O_2 uptake occuring subsequent to phagocytosis [2]. This presentation is amongst the first articles cited about the new DAIM (Diazoluminomelanis) precipitant. My particular area of observation was directed toward the standardization of the DAIM precipitant by weight with the varying concentration of HRP (Horseradish Peroxidase), with a standard amount of H_2O_2. Secondly, to see what effect the DALM precipitant had on Macrophage growth rate on cell-line RAW 264.7.

Luminescence resulted from activation appear to be more pronounced for the DAIM precipitant than luminol while using less DAIM, measured by weight. Structural analysis of DALM precipitate is still in the developmental stages and are being passed through pattons customs.

Material & Methods

Diazoluminomelanin and Horseradish Peroxidase Solutions

Diazoluminomelanin was prepared previously by USAF proprietory. DALM 1mg was dissolved in 1ml NaOH. This stock solution was diluted to 1:100, 1:200, 1:400, 1:800, 1:1000, 1:10,000, and higher dilutions as needed in: ph 6.5, 0.1 M phosphate buffer, ph 7.4, 0.1M phosphate buffer or 0.1M Tris - buffer ph 9.5. A stock solution of horseradish peroxidase (HRP Type VI, Signa chemicals) was prepared in pH 7.4 phosphate buffer 0.01M. The HRP was diluted to 1:1000, 1:10,000, 1:100,000, 1:1 X 10^6, in the buffers containing DALM.

Luminescent Assay

One hundred microliters of 0.03% H_2o_2 in water was used to activate 900 ul - solutions of HRP/DALM solutiuons (at dilutions and in buffers presented above). Relative Light Unit were recorded n 16 sec intervals on Turner Designs 20e luminater before and after the addition of H_2O_2. Total integration time for all run was 10.67 minutes at 37°C. Ten backgrown recording of luminescence totaling 2.67 minutes were taken at 16 sec, 64 sec, 112 sec, 160 sec. Twenty sample reading totalling 5.33 minutes were recorded at: 16 sec, 80 sec, 160 sec, 240 sec, 320 sec. Ten post-reaction continual (background) recording totalling 2.67 minutes were taken at 16 sec, 64 sec, 112 sec, 160 sec.

Results and Discussions

The fact that 1 mole of luminol is sufficient to covert 1 mole of HRP-H_2O_2 complex to free enzymes presents a basic standard for which a study of this nature can use as a background to standardize derivitive of luminol. There have been some 19 oxygen radicals scavengers assessed which are based on the principle that all oxygen radicals produced a baseline chemiluminescence which may be luminol enhanced [2, 3, 4].

DAIM a luminol derivative has been found to luminesce and flouresce at very low concentration. Dilutions of DALM precipitant were made starting 1:100 dilution doubling and ending at 1:1000 dilution. Data is presented in tables.

The results of the various dilutions at Ph 6.5, 7.4 or 9.5 are presented in Tables 1 - 3. Luminescense reading were greatest at 1:100 DALM vs 1:1000 HRP for all the PH's in (6.5, 7.4, and 9.5). The PH 6.5 lumionescence results showed the largest differences between HRP catalyzed and non-catalyzed DALM luminescence. This fact will be of importance in future studies of DALM as a macrophage substrate. The chemiluminescent reaction of DALM at acid pH's is important in macrophage studies since the phago-lysosomes of macrophages attain a pH of about 5. This work suggests that activated macrophages will engulf DALM precipitate and free radicals produce by the cell will be quantitated by the measuring of light emission from the precipitate.

DALM was administered to an aggressive macrophage cell-line. Initial concentration of control and test groups were approximatley 35,000 cells per vial. Concentration counts taken by coulter counter were recorded at 24, 48, 72 hrs after initiation of exposure to DALM. Cells were counted and observed for growth differences. There were no significantly different counts from that of control using a 1:100 dilutiuon of DALM. In some cases DALM treated cells were in number higher than control in terms of growth rate. Cellular debris was seen in the test samples but it represented minor cellular lysis or shrinkage.

TABLE 1 Tris

PH 9.5

DALM Dilution

HRP DILUTION	1:100	1:200	1:400	1:800	1:1000	1:10,000
1:1000	4.402 ± 0.16	2.79 ± 0.08	1.8816	1.137	0.042	0.092
1:10,000	2.573 ± 0.079	1.674 ± 0.065	1.1143 ± 0.023	0.664 ± 0.111	0.1925 ± 0.015	0.0535 ± 0.0007
1:100,000	2.344 ± 0.035	1.6065 ± 0.033	1.202 ± 0.112	0.574 ± 0.002	0.178 ± 0.006	0.026 ± 0.02
1:1 X 10^6	2.3665 ± 0.08	1.528 ± 0.02	1.167 ± 0.023	0.643 ± 0.0004	0.173 ± 0.0006	0.04 ± 0.007

TABLE 2

PH 7.4

DALM Dilution

HRP DILUTION	1:100	1:200	1:400	1:800	1:1000
1:1000	8.316 ± 0.68	4.135 ± 0.10	3.125 ± 0.15	1.798 ± 0.021	1.575 ± 0.53
1:10,000	6.291 ± 0.23	4.658 ± 0.28	2.100 ± 0.19	1.214	0.885 ± 0.199
1:100,000	5.819 ± 0.24	4.6785 ± 0.66	N/R	N/R	N/R
1:1 X 10^6	4.6635 ± 0.06	N/R	N/R	N/R	N/R

N/R= No Run

TABLE 3

PH 6.5

DALM Dilution

HRP DILUTION	1:100	1:200	1:400	1:800	1:1000	1:10,000
1:1000	4.187 ± 0.45	2.322 ± 0.17	2.32 ± 0.21	1.5185 ± 0.70	1.2065 ± 0.58	0.616 ± 0.18
1:10,000	1.667 ± 0.23	1.391 ± 0.049	1.445 ± 0.65	1.3315 ± 0.26	0.558	0.429 ± 0.07
1:100,000	2.593 ± 0.20	1.247	0.880	0.593	0.340	0.091 ± 0.033
1:1 X 10^6	2.02 ± 0.158	1.096	1.187	N/R	N/R	N/R

N/R = No Run

Conclusion

DALM precipitate at this point has not been structurally defined. It appears to be clearly a derivitive of luminol but has some unique chemical differences. The conventional luminol system for chemiluminescence assays require an alkaline solution of luminol, an oxidant and a catalyst [5]. DALM seems to prefer an acidic environment but is activated by oxidizing free radicals.

This study was an effort to characterize the parameters for efficient use of DALM precipitate and to maximize its response as a free radical indicator. We feel that this concentration block study has been successful in answering some of the questions about the precipitate. I have particularly enjoyed this learning experience and hope to pursue a branch study that will answer some of the "dental questions" in dental graduate school.

Thanks for selecting me and I look forward to working with you in the future.

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FINAL REPORT

Prepared by: Ernest J. Freeman

Academic Rank: Graduate Student

Department and Biological Sciences

University: Kent State University

Research Location: USAFSAM/NGNS
Brooks AFB
San Antonio TX 78235

USAF Researcher: Dr. David M. Terrian

Date: 10 Sept. 88

Contract No: F49620-88-C-0053

MEMBRANE ALTERATIONS INVOLVED IN EVOKED RELEASE OF L-GLUTAMIC ACID FROM MOSSY FIBER SYNAPTOSOMES

by

Ernest J. Freeman

ABSTRACT

Mossy fiber synaptosomes were isolated from both hippocampal and cerebellar tissue obtained from 200-300 gram male Wistar rats. The synaptosomal preparation was applied to filters supported in incubated superfusion chambers. The preparation was kept covered continuously with appropriate buffer while fractions were collected at a rate of 0.5 ml/min. After fractions were collected they were frozen and lyophilized. The fractions were later reconstituted and assayed for the presence of L-glutamate using an enzyme coupled reaction in a florescence spectrometer. This allowed us to determine the stimulatory and inhibitory actions of various agents on the release of this acidic amino acid neurotransmitter from the mossy fiber synaptosomes.

ACKNOWLEDGEMENTS

I wish to thank the Air Force Systems Command and the Air Force Office of Scientific Research for sponsorship of this research effort. I also thank Universal Energy Systems for their generous support and fine directional and adminstrative assistance.

The Graduate Student Research Program proved to be an extremely rewarding experience in all respects. Dr. David Terrian provided me with an excellent atmosphere. He afforded me the opportunity to accomplish a major portion of research that is, and will be very important to me in the many months of research to come. The help of Peter Hernandez, Anna Marie Michals and Sudar Alagarsamy was greatly appreciated. The concern, interest and encouragement of Dr. Bob Gannon and Dr. Mike Rea proved to be invaluable. Again, I would like to thank Dr. Terrian for his direction and much needed expertise that made this program an outstanding success.

I. INTRODUCTION:

The transmission of neuronal impulses are dependent on the release of various neurotransmitter substances. In the mossy fibers,MF, one of these neurotransmitters is L-glutamate. The release of this neurotransmitter from the nerve terminal requires stimulus induced reversible alterations in the permeability of the membrane.

The Neuroscience Function of the USAF School of Aerospace Medicine at Brooks Air Force Base is particularly interested in the mechanisms involved in the release of glutamate from a mossy fiber synaptosomal preparation. To understand the mechanisms controlling the membrane alterations involved in neurotransmitter release would allow for manipulations that could either inhibit or promote the release of glutamate. In the hippocampus such manipulations could allow for enhanced long term potentiation and learning. While in the cerebellum enhanced motor activity control could be obtained.

My previous research has involved in investigations into the effects that K+ depolarization and arachidonic acid have on glutamate release from an identicle MF synaptosomal preparation that is utilized by Dr. Terrian's group. This experience and the collaboration between my advisor, Dr. Robert Dorman, at Kent State University and Dr. Terrian contributed to my assignment to the Neuroscience Function.

II. OBJECTIVES OF THE RESEARCH EFFORT:

Generally the aim of the research effort was to attempt to manipulate the environment of the nerve terminals to enhance or inhibit the release of glutamate by affecting the membrane alterations involved.

Specifically, I looked at the effects of K+ depolarization on glutamate release as well as arachidonic acid stimulated release of this acidic amino acid neurotransmitter. Exact protocols for optimizing the release profiles generated by addtion of these stimulating agents were also determined. In addition, since arachidonic acid can be broken down by enzymes contained in the MF synaptosomes various inhibitors of these pathways were used to more precisely determine the critical compound responsible for stimulating release of glutamate from the nerve terminals.

Research on this project is being continued at Kent State University where I am in process of determining the source of the arachidonic acid since it was found stimulate glutamate release from MF synaptosomes.

III.

a. Lipid metabolism of the MF synaptosomal membranes has been implicated as an important regulator of the MF synaptic processes. These experiments attempted to correlate endogenous lipid metabolism with synaptic transmission.

Since we proposed to look at exact pathways of lipid metabolism we employed a homogenous and simple synaptic model that was isolated with a high degree of purity. The preparation of the MF synaptosomes was carried out according to the methods described by Terrian et. al. (1985). The final pellets were suspended in a Krebs-bicarbonate medium (KBM; final concentrations: 122mM NaCL, 3.1mM KCL, 1.2 mM MgSO4, 0.4 mM KH2PO4, 5mM NaHCO3, 20 mM TES, 11 mM D-Glucose; pH 7.4; 295 mOs/kg). Calcium ions were omitted during preincubation to allow the synaptosomes to restore membrane polarity and cytosolic calcium concentrations. Samples were taken for protein determinations according to the Warburg- Christian method (Layne, 1957). This provided a rapid and convenient estimate that proved useful in minimizing the inter-experimental variability in the amount of synaptosomal protein layered onto the superfusion columns. The synaptosomes were preincubated at 30 C for 5 min prior to applying 2mg protein/chamber as determined by the Warburg-Christian method. The preincubated synaptosomes were superfused at a rate of 0.5 ml/min with a KBM containing 1.8 mM CaCl2. Superfusion conditions were previously described by Dorman et. al., 1986. Drugs to be tested were added to the superfusion medium 8 min prior to depolarization of the synaptosomes with either a 2 min pulse of 35 mM KCl in KBM, or a 12 min pulse of 200uM arachidonic acid in KBM as well. The 4 min superfusate fractions were later concentrated five to 20-fold by lyophilization and assayed for glutamic acid using a fluorometric assay (Graham and Aprison, 1966) with a Perkin- Elmer Model LS5B spectrofluorometer to measure the production of NADH by glutamic acid dehydrogenase. A linear relationship between light emitted (excitation 340 nm; emission 460 mn) and the amount of L-glutamate present in the quartz cuvette was obtained over a range of 0.25 to 2.5 nmoles. None of the test compounds used in the experiments interfered with this assay when present in concentrations found in the superfusion medium.

b. Using this experimental design the following was accomplished. It was shown that a 2 min pulse of 45 mM KCl stimulated the efflux of L-glutamate from both hippocampal and cerebellar mossy fiber synaptosomes (Fig. 1). The peak height of this response, in the hippocampus corresponded to 630 pmoles of L-glutamate where the cerebellar response was much lower, 437 pmoles, compared to the spontaneous efflux which was 71.0 and 182.0 pmoles respectively.

Exposure of the synaptosomes to 200uM arachidonic acid (20:4), also caused an increase in the efflux of L-glutamate in both the hippocampus and cerebellum (Fig. 2). The hippocampus again responded with greater release than the cerebellum as well as responding more quickly to the stimulus. The effects of 20:4 were seen to be reversible and the synaptosomes retained their ability to respond to an additional stimulus of 45 mM KCl (Fig. 2). Although, the cerebellar response to 45 mM KCl was drastically reduced when compared to the group that was not treated with 20:4. In contrast the hippocampus response to the subsequent K-depolarization was nearly identicle to the non-20:4 treated group

(Fig. 2).

Ouabain, a Na-KATPase inhibitor, was also used to determine its effect on the release of L-glutamate from the mossy fiber synaptosomes (Fig. 3). The initial experiments illustrated a somewhat dose-dependent response of the synaptosomes to ouabain, but more experiments must be done to obtain a more reliable profile for ouabain's effect. 0.3mM ouabain was used in subsequent experiments due to its more apparent stimulation of L-glutamate efflux.

In an effort to more clearly determine the site of action of 20:4 the mossy fiber synaptosomes were treated with 200uM 20:4 and 0.3 mM ouabain (Fig. 4). Figure 4, again needs more experimental data but a trend can be seen. The ability of both the compounds to cause an increase in the efflux of neurotransmitter, in both tissues, is greater when they are present together in the superfusion medium than when they are present alone.

Finally, to further clarify the critical compound in the stimulation of L-glutamate release nordihydroguaiaretic acid (NDGA), a lipoxygenase pathway inhibitor, was used. This compound inhibits the lipoxygenase enzyme's ability to break down 20:4. Figure 5 shows the dose dependent inhibition of L-glutamate release by NDGA. 35mM KCl was used since it caused a submaximal efflux, compared to 45mM KCl, of neurotransmitter and thus would allow for a more sensitive condition to illustrate NDGA's inhibitory effect. As is shown in figure 5, 1.0uM NDGA inhibits completely the ability of 35mM KCl to stimulate glutamate release whereas 0.1uM NDGA fails to inhibit release at all. The effect of NDGA was seen only in the hippocampus, NDGA was unable to inhibit the 35mM KCl stimulated efflux in the cerebellum (data not shown). In addition, NDGA was used to determine its effect on 200uM 20:4 stimulated efflux of L-glutamate in the hippocampus only since NDGA was not seen to work in cerebellum. As figure 6 illustrates NDGA shows the opposite effect with 20:4 stimulated efflux than it did with K-depolarization. The NDGA at 1.0uM concentration actually caused a potentiation of the 20:4 effect.

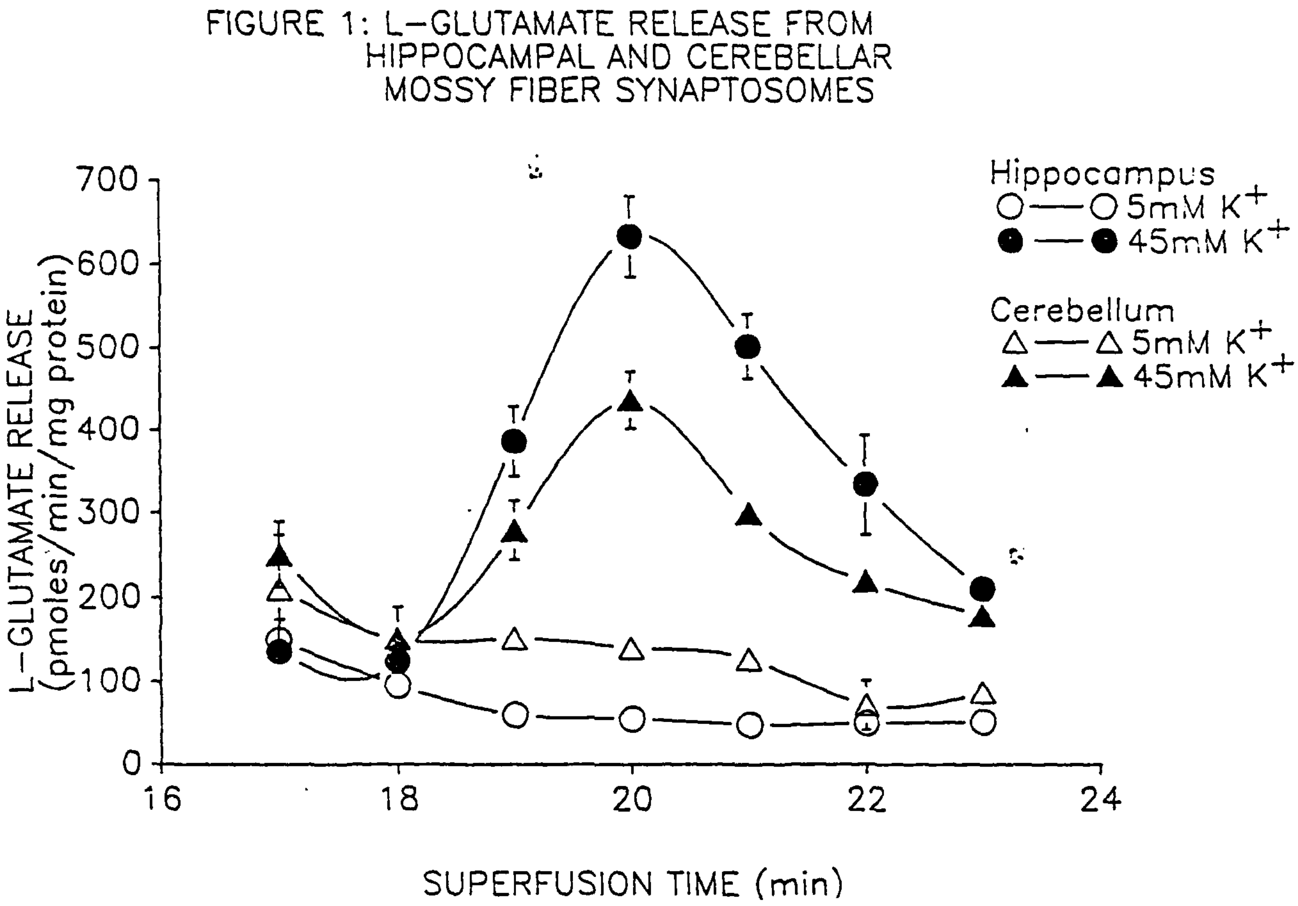

Figure 1: Effects of membrane depolarization on the efflux of L-glutamate from hippocampal and cerebellar mossy fiber synaptosomes. Synaptosomes were superfused with control buffer (KBM) for 16 min prior to the addition of control (5mM KCl) or depolarizing buffer (45mM KCl) for 2 min. Fractions were collected and L-glutamate quantitated. Results are expressed as pmoles/min/mg protein.

FIGURE 2a: ARACHIDONIC ACID STIMULATED
L-GLUTAMATE RELEASE FROM HIPPOCAMPUS

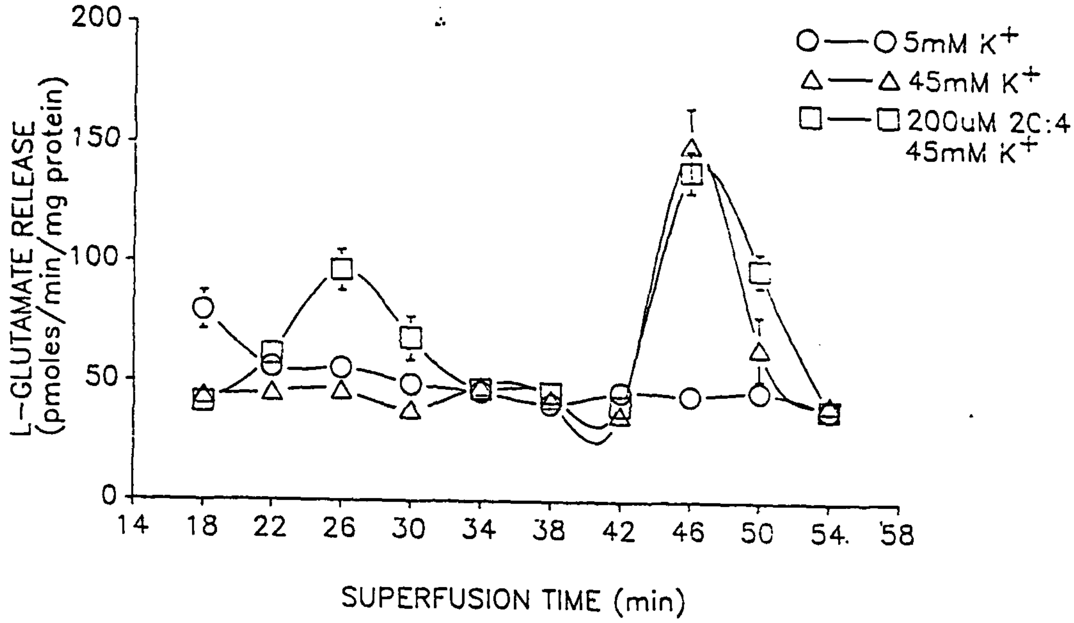

FIGURE 2b: ARACHIDONIC ACID STIMULATED
L-GLUTAMATE RELEASE FROM CEREBELLUM

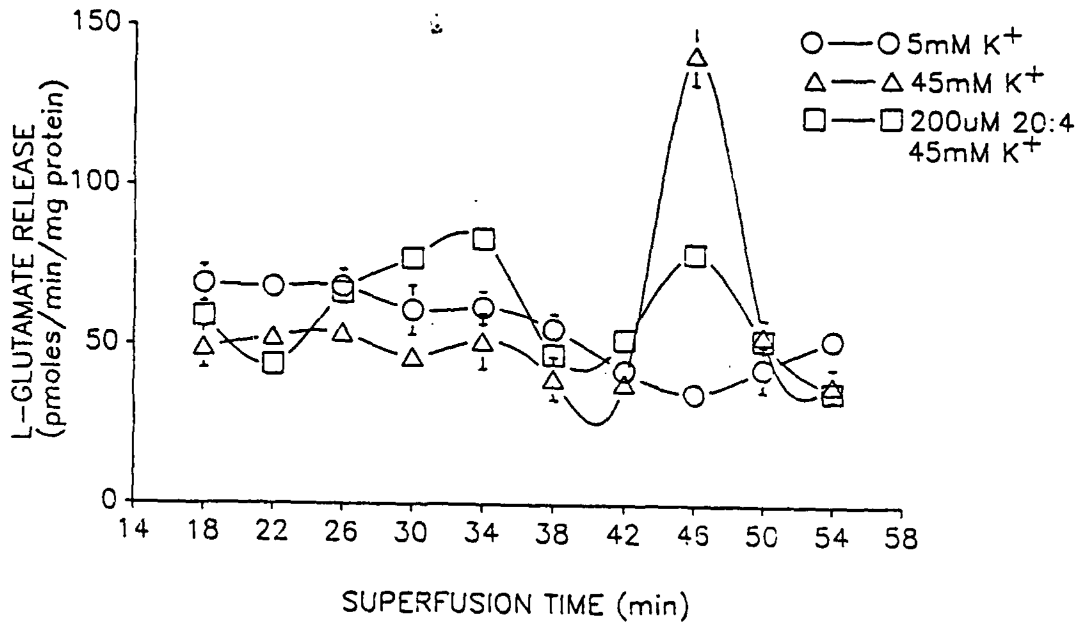

Figure 2a + 2b: Effects of exogenous arachidonic acid on the efflux of L-glutamate from mossy fiber synaptosomes. Synaptosomes were superfused with control buffer for 18 min. prior to the addition of 200uM arachidonate. Control buffer was reintroduced at 26 min. 45mM KCl buffer was added at 42 min. for 2min. Fractions were collected for an additional 12min. L-glutamate was quantitated as described and the results are expressed as pmoles/min/mg protein.

FIGURE 3: OUABAIN STIMULATED L-GLUTAMATE RELEASE IN HIPPOCAMPUS

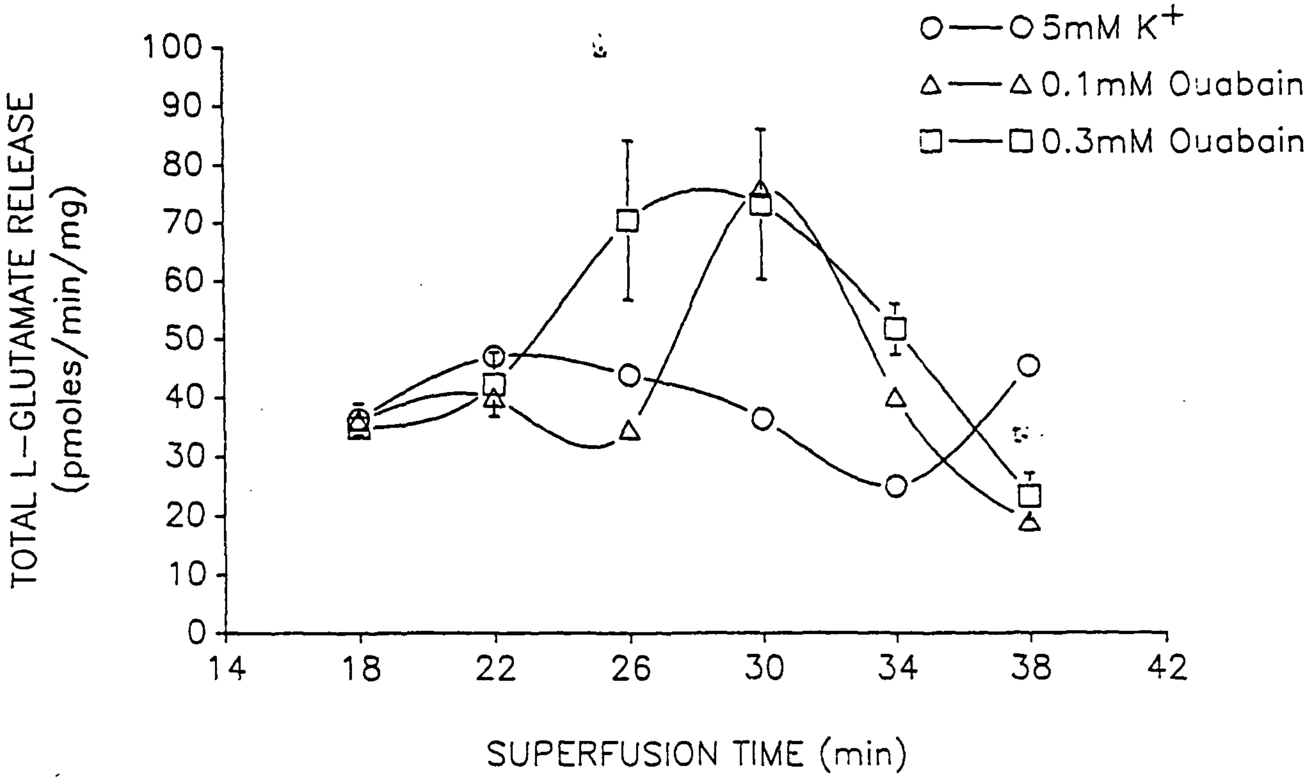

Figure 3: Effects of ouabain on the efflux of L-glutamate from mossy fiber synaptosomes. Synaptosomes were superfused with KBM (5mM KCL) for 18 min. prior to the addition of either 0.1mM or 0.3mM ouabain. Control buffer was reintroduced at 26 min. and 4 min. fractions were collected for another 12 min. L-glutamate was quantitated and reported as pmoles/min/mg protein.

FIGURE 4a: COMBINED EFFECTS OF 0.3mM OUABAIN AND 200μM ARACHIDONIC ACID IN HIPPOCAMPUS

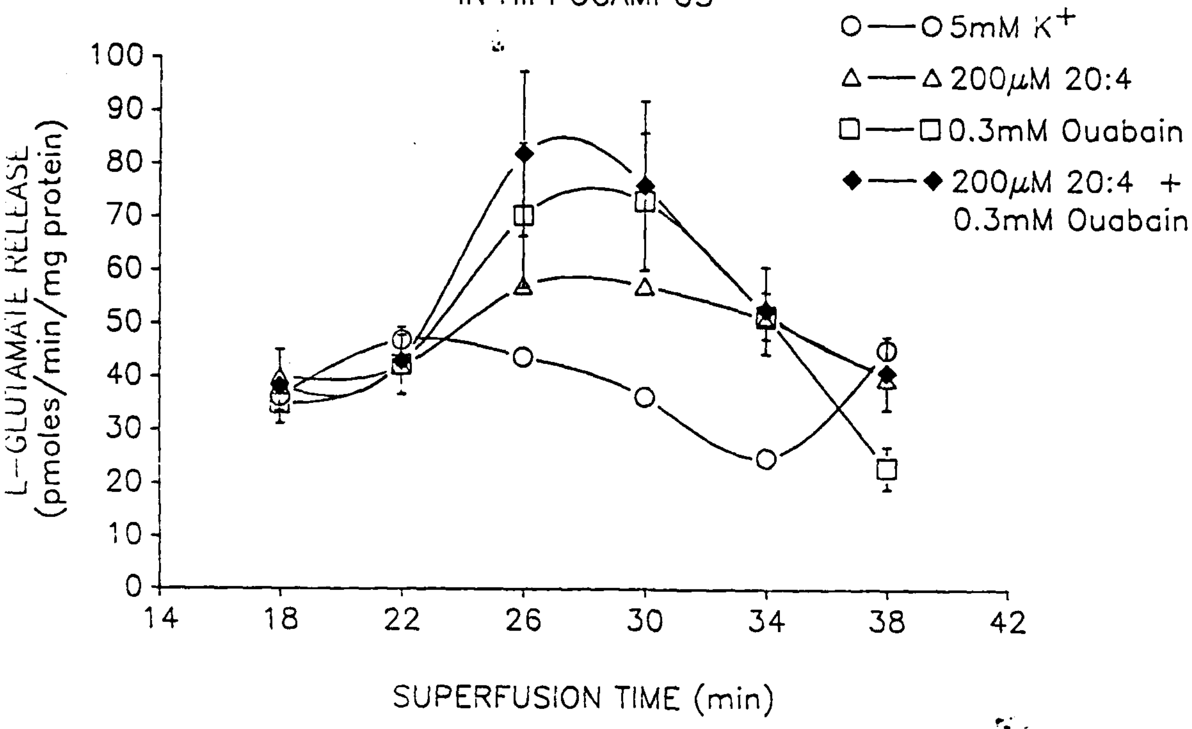

FIGURE 4b: COMBINED EFFECTS OF 0.3mM OUABAIN AND 200μM ARACHIDONIC ACID IN CEREBELLUM

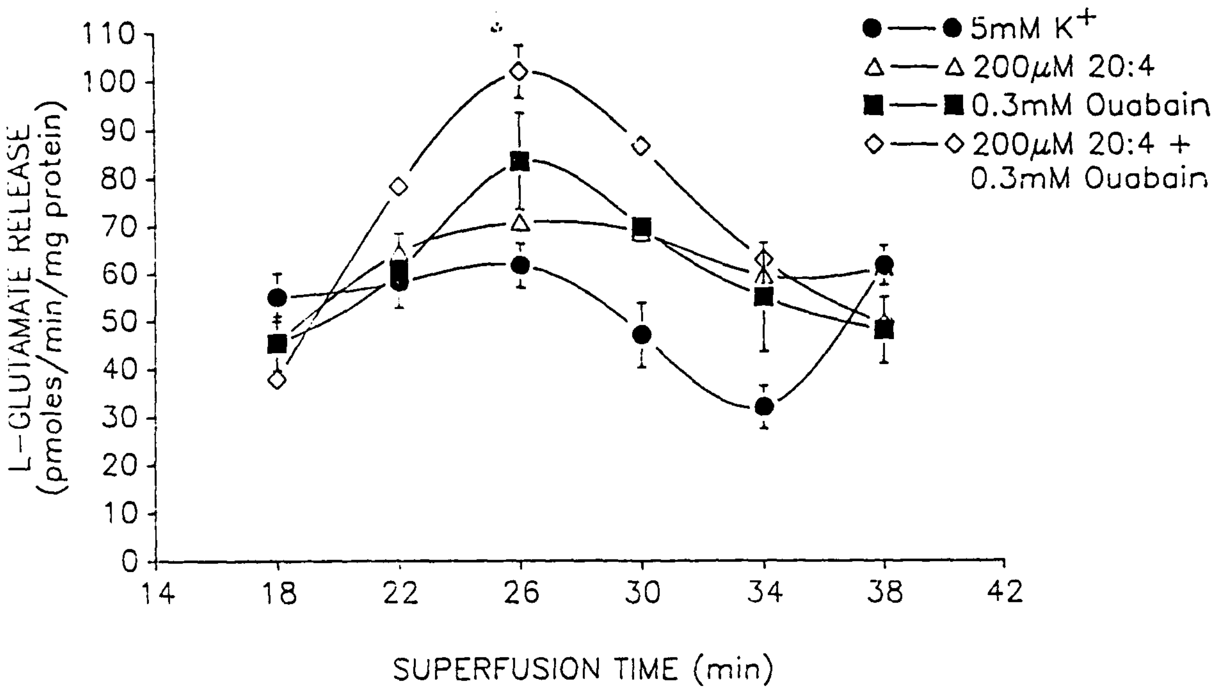

Figure 4a + 4b: The combined effects of 200uM arachidonate and 0.3mM ouabain on the efflux of L-glutamate from hippocampal and cerebellar mossy fiber synaptosomes. Preparations were superfused for 18 min. with control buffer prior to the addition of either 200uM 20:4, 0.3mM ouabain or a combination of the two compounds. Control buffer was reintroduced at 26 min. and 4min. fractions were collected for an additional 12min. L-glutamate was quantitated as described and the results are reported as pmoles/min/mg protein.

FIGURE 5: DOSE DEPENDENT INHIBITION OF L-GLUTAMATE RELEASE BY NORDIHYDROGUAIARETIC ACID

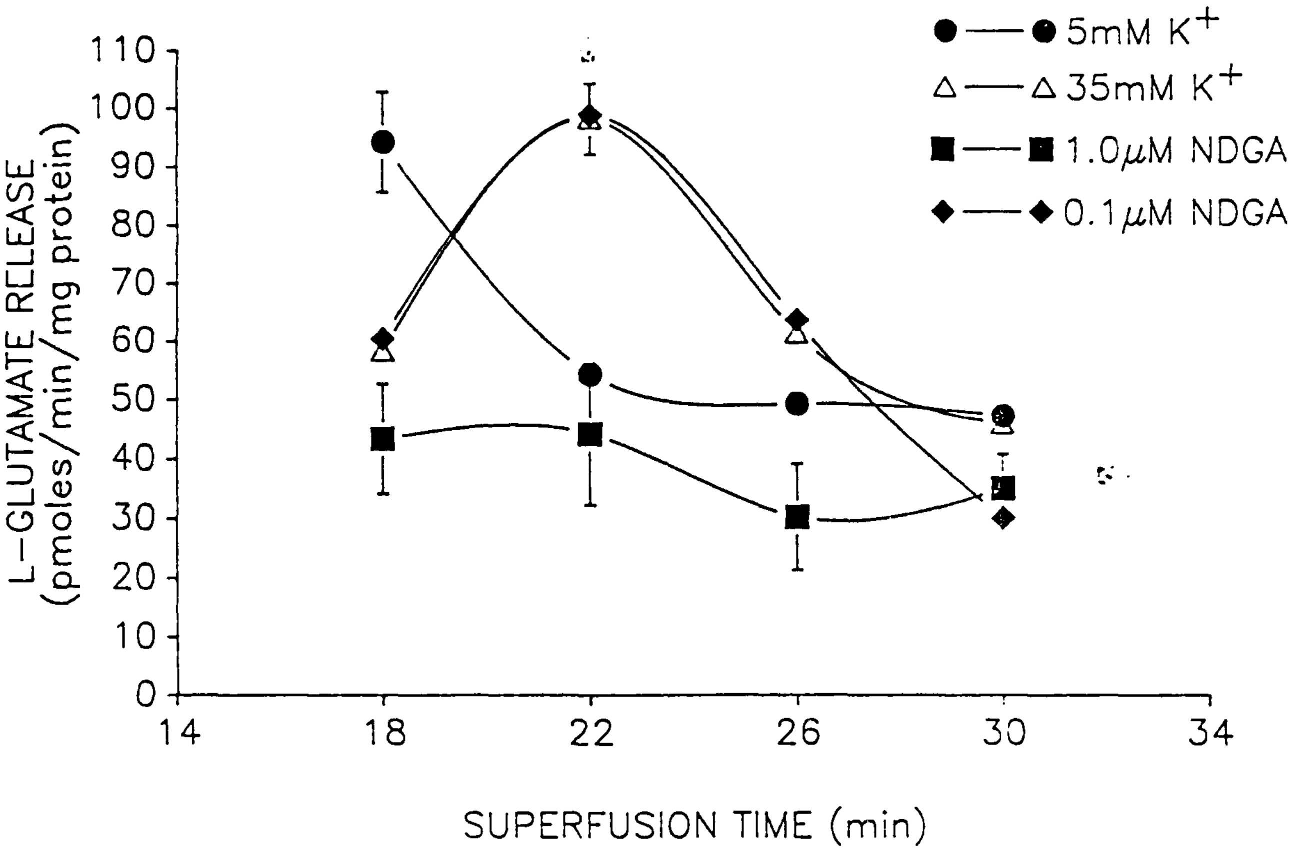

Figure 5: Dose dependent inhibition of L-glutamate efflux by nordihydroguaiaretic acid. Hippocampal mossy fiber synaptosomes were superfused for 10 min. prior to addition of the inhibitor. NDGA was present for 8 min. preceeding the 2 min. depolarization with 35mM KCl buffer. NDGA was also present in the 35mM KCl buffer. Fractions were collected for 10 min. after depolarization. L-glutamate was quantitated as described and results are expressed as pmoles/min/mg protein.

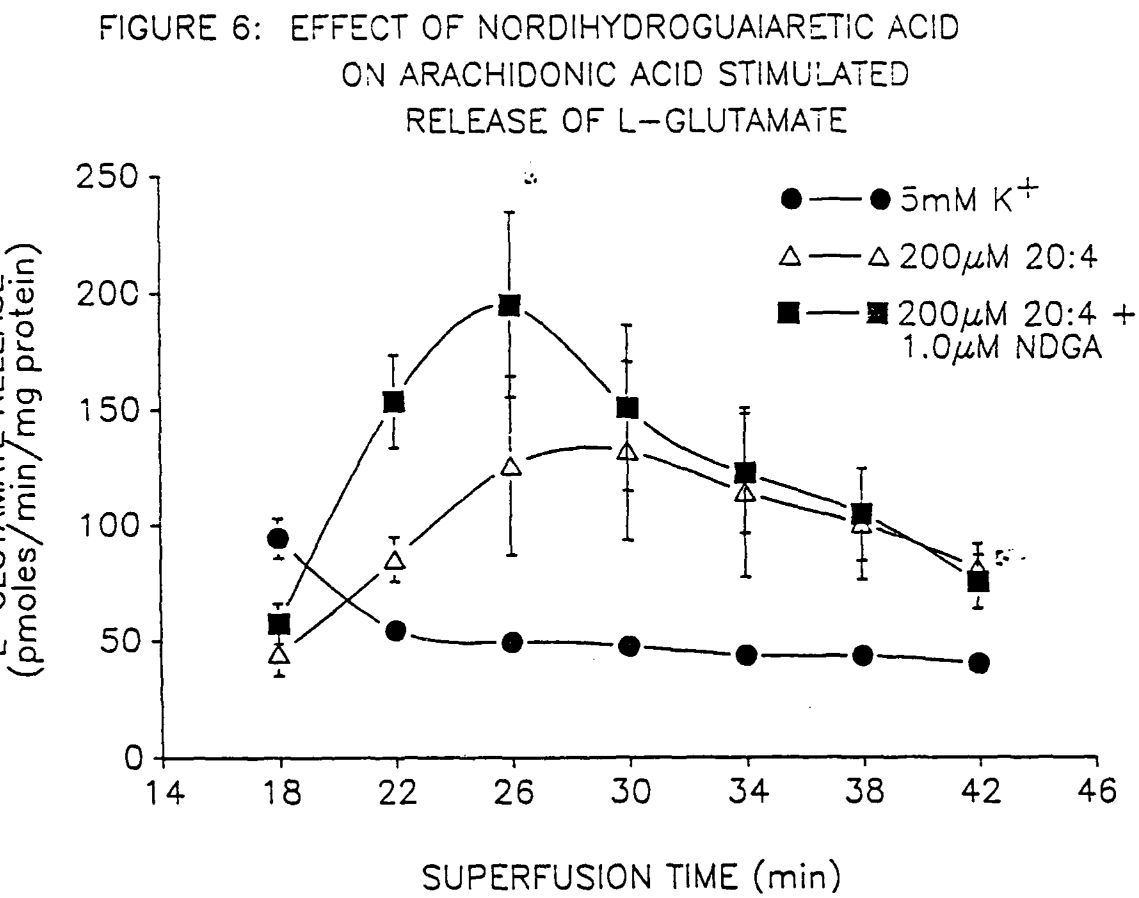

Figure 6: Effect of nordihydroguaiaretic acid on the arachidonic acid stimulated efflux of L-glutamate from hippocampal mossy fiber synaptosomes. Synaptosomes were superfused with control buffer for 10 min. prior to adding 1.0uM NDGA. 200uM arachidonic acid , in the presence of 1.0uM NDGA, was introduced at 18 min. Control buffer was returned at 30 min. and fractions were collected for another 12min. L-glutamate was quantitated as described and results are reported as pmoles/min/mg protein.

IV. RECOMMENDATIONS:

a. It has been shown here that lipid metabolites , specifically arachidonic acid , can evoke the release of endogenous L-glutamate from mossy fiber synaptosomes. Current investigations are based on the stimulated release of radiolabeled neurotransmitters that were preloaded into synaptosomal or brain slice preparations. The suitability of using these techniques to study the release mechanisms has been questioned, because it is not clear whether the radiolabeled neurotransmitters are gaining access to the physiologically relevant pool. The ability, illustrated here, to measure endogenous neurotransmitter release circumvents the above difficulties and allows for a more direct correlation between membrane lipid metabolism and nerve impulse transmission.

b. The ability to measure the evoked release of endogenous glutamate provides us with an opportunity to correlate lipid metabolism with pre- and postsynaptic neurotransmitter release mechanisms. Since we have observed stimulated efflux of glutamate with K-depolarization and in the presence of exogenous arachidonic acid, we are now in the position to manipulate lipid metabolism and relate the effects to the release of glutamate.

Since arachidonic acid is metabolized in the synaptosomes to either prostaglandins or eicosanoids, via the cyclooxygenase and lipoxygenase pathway respectively, it will be necessary to determine the exact compound responsible for the stimulation of glutamate efflux. This will be accomplished by utilizing various specific inhibitors of these two pathways. Initial experiments reported here using NDGA, a lipoxygenase pathway inhibitor, implicate arachidonic acid as the critical mediator stimulating efflux of neurotransmitter. Although, the effects of the cyclooxygenase inhibitors ibuprofen and indomethacin must now be investigated in order to rule out any stimulatory actions by metabolites of this pathway. In addition various metabolites of these two pathways, such as the prostaglandins F2-alpha, E2 and the eicosanoid 12-HETE, must be utilized in order to more directly observe their effects on release of glutamate from the mossy fiber synaptosomes.

Finally, looking in the other direction the source for the arachidonic acid must be determined. It has been shown that in response to K-depolarization synaptosomes have an increased phospholipid catabolism (Lazarewicz et. al., 1983), and free arachidonate accumulation (Lazarewicz et. al., 1983; Asakura and Matsuda, 1984). The increased phospholipid catabolism leads to an increase in diglycerides present, of which the arachidonate may be a constituent. Experiments will be done utilizing various diglycerides which contain arachidonic acid to assess their effects on glutamate efflux. Arachidonic acid is released from these diglycerides as a result of phopholipase activity. Thus, in order to determine if the diglycerides serve as the source for arachidonate, various phospholipase inhibitors will be used in an attempt to inhibit stimulated efflux of glutamate by inhibiting the release of arachidonate from the diglyceride.

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FINAL REPORT

Development of Improved Assays for Cholesterol and Major Lipoprotein Fractions

Prepared by: Eric R. Johnson, Ph.D. and Thomas Lane

Academic Rank: Professor and Graduate Student

Department and University: Department of Chemistry, Ball State University

Research Location: USAF SAM/NGIL, Brooks AFB, TX 78235

USAF Researcher: Harvey A. Schwertner

Date: 3 August 1988

Contract No: F49620-87-R-0004

SAME REPORT AS
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FINAL REPORT

GLAUCOMA IN US AIR FORCE AVIATORS-

USAF SCHOOL OF AEROSPACE MEDICINE STUDY GROUP

Prepared by:	Yolanda A. Malone
Academic Rank:	Medical Student, SMP III
Department and University:	School of Medicine Meharry Medical College
Research Location:	Ophthalmology Department USAF School of Aerospace Medicine Brooks Air Force Base, TX 78235-5000
USAF Researcher:	Captain Daniel R. Peters
DATE:	September 27, 1988
Contract No.:	F49620-88-C-0053

GLAUCOMA IN US AIR FORCE AVIATORS-
USAF SCHOOL OF AEROSPACE MEDICINE STUDY GROUP

BY

Yolanda A. Malone

ABSTRACT

A retrospective study was conducted on approximately 125 United Air Force Pilots diagnosed with glaucoma. Pilots were followed over a course of ten years. The developed protocol focused on glaucoma with regard to epidemiology, treatment, and aeromedical disposition. In addition the effects of medication and progression of various parameters such as: cup/disc ratio, visual field change and intraocular pressure ranges were examined. The data collected indicated a positive correlation between factors such as trauma, steroid use and heredity to glaucoma. There were few cases of treatment by laser trabeculoplasty, but in cases where this type of treatment was necessary the pilots were usually grounded. Pilots retaining flying status were controlled on medication. Patients with pigmentary dispersion glaucoma were excluded from this study.

ACKNOWLEDGEMENT

I would like to express my appreciation to the Air Force Systems Command and the Office of Scientific Research Department of Ophthalmology for sponsorship of this research effort. Many thanks to Universal Energy System for their administrative role. The summer was very enriching and rewarding due to the guidance and counsel of Captain Daniel Robert Peters. The concern of LTC Colonel Miller was inspiring and stimulating. The resourcefulness of Airman First Class Lisa Robinson and the knowledge she provided was very helpful. Many thanks to all at Brooks Air Force Base in the Department of Ophthalmology and Optometry who added to the different aspect of this research experience.

ratio ranging from .1 to .9 for the left and right eyes. from .1 to .9 for the left and right eye. Asymmetry of the cup/disc ratio between eyes of greater than .2, generalized enlargement of the cup/disc ratio in both eyes of greater than .6, or an increase in the vertical cup/disc ratio over the horizontal cup/disc ratio is highly suggestive of glaucomatous damage. However there may be a normal increase slowly and slightly with age. This is in contraindication to the more rapid cupping seen in glaucoma. In eyes with congenital disc anomalies, high myopia or cataract it may be difficult to determine the degree of cupping.

The optic disc may present with a progressive degeneration prior to the development of detectable visual field defects. In cases where glaucoma is poorly controlled cup/disc ratios vary with damage of neural rim tissue. Usually progressive, asymmetrical alterations are seen but concentric enlargement of the cup may occur. Selective loss of inferior or superior neural rim tissue leads to vertical cupping. This usually starts with focal notching when enough of the underlying lamina cribrosa tissue is bared a "lamina dot" sign is produced. Retinal vessels supported by the neural rim will bend away from the defect and become bayoneted. Other glaucomatous changes include splinter hemorrhages near the disc margin, baring of circumlinear vessels where neural rim tissue has

I. INTRODUCTION AND STATEMENT OF OBJECTIVES

Normally the optic nerve head is round or slightly ovoid, measuring about 1.5 mm in diameter. The physiologic cup (a pale depression) is usually found in the center of the optic disc. The outermost boundary of the cup is where the retinal nerve fibers change from a direction parallel with the plane of the retina and begin to course posteriorly. The central pallor results from the fact that the underlying lamina cribrosa is bare of overlying neural and glial elements. The tissue between the edge of the cup and the outer margin of the disc is termed the neural rim and is even around the cup. The capillary vascularity of the neural rim enhances the normal color which is due to the surface capillaries emanating from the retinal circulation and the deeper one from ciliary circulation. The retinal vessels usually enter the disc centrally and course nasally to then follow the edge of the cup.

Variation of the optic cup's size and shape is dependent on the individual and it is usually reported in terms of the horizontal cup/disc ratio. However, initial data collection for this research product included data of vertical cup/disc ratio. A genetic predisposition in relation to cup/disc ratio has been proposed. The majority of normal eyes have a cup/disc ratio of less than .4 in both the horizontal and vertical meridians. The data collected for pilots at Brooks AFB, TX had a sample cup/disc disappeared from a vessel and a progressive nasal shift of disc vessels.

Majority of patients reviewed seem to have ocular hypertension/pre-glaucoma. The elevated intra-ocular tension often progressed to glaucoma. Primary open angle glaucoma is the most common type of glaucoma seen in humans. There are no obvious ocular abnormalities but there does seem to be an increased incidence of this disease in hyperparathyroid and diabetic patients. In this condition both eyes are usually affected to the same degree. The disease is usually detected after the fourth decade of life. It has been detected in all existing races.

It has been suggested that disease is due to multifactorial heritable factors, trauma and steroid use. our study reviewed the correlation of each of these factors to the disease process. Most of our cases are discovered during a routine eye exam. The eyes are usually white with intraocular pressure ranging from 15-25mm Hg and sometimes as high as 40mm Hg. The cup/disc ratio is often grater than .3 with vertical elongation. In cases where intra-ocular pressure is normal but accompanied by cupping and visual field defects a diurnal curve may reveal an elevated intra-ocular pressure later during the day. Some patients with typical optic nerve alterations and visual field defects have low tension glaucoma. Therapeutic regimens utilized in the treatment of glaucoma are medications such as pilocarpine, timolol, timoptic, etc., laser trabeculoplasty or a variety of surgical filtering procedures which serve to decrease intra-ocular pressure.

MATERIALS AND METHODS

The patient pool for this study consisted of approximately 125 USAF Aviators diagnosed with glaucoma or ocular hypertension. The records of the patients were provided by the medical records department at Brooks Air Force Base. A protocol was developed where by pertinent information was extracted from the records. It was decided that the following factors would be compiled:

1. Race and sex of the patient.
2. Which eye(s) is affected.
3. History of: a. Trauma
   b. Steroid use
   c. Family Trait
4. Best visual acuity.
5. Visual field defects.
6. Age at the time of initial diagnosis.
7. Diurnal curve variations.
8. Cup/Disc ratio.
9. Treatment in terms of medication/surgery
10. Affect of glaucoma on flying status.

After compilation of the above factors the material was assimilated and coded for statistical analysis.

III. RESULTS

Both eyes were affected in the majority of the cases of ocular hypertension and as well as for glaucoma. The cup/disc ratio ranged from .1 to .9 for both the right and the left eye but usually it was the same or did not differ any more than .1 for any given patient. Visual acuity ranged from 20/12 to 20/400 between the different patients. In addition patients with extremely poor visual acuity for one eye did not necessarily have very poor visual acuity for the other eye. This is quite different from the cup/disc ratios in which both eyes had nearly the same numerical value. Applanation tonometry ranged from 12 to 32 with a majority of the pilots reported on being controlled by medication or having undergone surgery to decrease intra-ocular pressure. Most of the patients in this study received waivers as long as there were no visual field defects and the glaucoma was under control. However, in some of the cases where surgery was necessary and had to be repeated or the visual acuity was greatly diminished then flying waiver was denied and the pilot was grounded. Recall that this study seeks to follow pilots over a course of ten years in an effort to see the affects of glaucoma on pilot flying status therefore flying status may vary from one evaluation to the follow-up examination.

IV. DISCUSSION

The collected data indicates a linear progression of the disease process with respect to time. It seems that patients often presenting with ocular hypertension progress to full blown cases of glaucoma. In addition there is a positive correlation between the development of glaucoma and trauma to the affected eye as well as to steroid use. The drug of choice for the treatment of glaucoma is timolol. However, other agents such as pilocarpine, carbachol, and epinephrine were used alone or in combination by subjects in this study. Of the 125 cases reported only three indicated the use of laser trabeculoplasty as a component of treatment. Two of the three cases which utilized this procedure had to have the procedure again after a varying period of time. Patients which had the surgery were grounded and there was no documentation of them being returned to flying status.

V. CONCLUSION

The statistical analysis of this project is not available at this time. However, from the data collected it can be concluded that hereditary predisposition, steroidal use and trauma all serve as risk factors to the development of glaucoma. However it is possible for pilots to maintain flying status as long as their intra-ocular pressure is controlled by medication and there is no visual field deficits nor decrease in visual acuity. In many of the cases in which the pilot was grounded the cup/disc ratio was as high as .9 indicating an increased possibility of optic nerve damage. Thus it can be seen that it is possible to maintain flying status with glaucoma but great care must be taken to assure that the condition is controlled and that the pilot has properly functioning vision.

VI. RECOMMENDATIONS

1. Examine the time lapse between laser trabeculoplasty procedures when more than one is required in a given patient.
2. Examine the duration and dosage of steroid use in relation to the development of glaucoma.
3. Examine the time lapse between trauma and the development of glaucoma.
4. Continuation of the study of the effects of glaucoma on United States Air Force Aviators in an effort to improve upon treatment and flying status recommendations.

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FINAL REPORT

THE SEPARATION OF HDL2 AND HDL3 USING THE TECHNIQUE OF ULTRACENTRIFUGATION

Prepared by:	Joe M. Ross, Ph.D. and Conrad Murray
Academic Rank:	Assistant Professor / Graduate Student
Department:	Chemistry Department
University:	Central State University
Research Location:	USAFSAM/NGIL Brooks AFB San Antonio TX 78235
USAF Researcher:	Harvey Schwertner, Ph.D.

Date:	26 Aug 88
Contract No:	F49620-87-R-0004

SAME REPORT AS
PROF. JOE ROSS
SCHOOL OF AEROSPACE MEDICINE # 146

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FINAL REPORT

Light Beam Interaction Induced by a Transition Metal Complexed To A Tridentate Ligand

Prepared by:	Christine M. Nelson
Academic Rank:	Graduate Student
Department and University:	Chemistry Department, Trinity University
Research Location:	USAFSAM/NGOV Brooks AFB San Antonio, TX 78235-5301
USAF Researcher:	John Taboada, PhD

Date:	August 30,1988
Contract No:	F49620-88-C-0053

Light Beam Interaction Induced by a Transition Metal Complexed To A Tridentate Ligand

by

Christine M. Nelson

ABSTRACT

Transition metals when complexed with aromatic nitrogen donor ligands, such as 2,2'-bipyridine and 1,10-phenanthroline, exhibit very rich visible and near UV spectra. The multiple peaks are caused by the various transitions available to the electrons in the molecule, such as d to d, and ligand to ligand transitions and charge transfers between the metal and the ligand. The Schiff base derived from the condensation of 8-quinolinamine and pyridine-2-carboxaldehyde when complexed to a transition metal displays similar characteristics to other aromatic nitrogen donor ligands. A series of these Schiff base complexes, using different transition metals as the center, was synthesized. The samples were placed in orthogonally intersecting laser beams to investigate whether they demonstrated properties that would induce interaction between the beams.

Acknowledgements

I wish to thank the Air Force Systems Command and the Air Force Office of Scientific Research for sponsorship of this research. Universal Energy Systems must be mentioned for their concern and help to me in all administrative and directional aspects of this program.

My greatest appreciation is given to Dr. John A. Burke, Jr. for extending to me this opportunity to further my research experience. He offered me his encouragement, confidence and patience to make this a very educational and enjoyable research summer. I must also thank Dr. John Taboada for his interest in and support of our project. Dr. Dale Clark and Mr.Leo Mosser also have my appreciation for their southern hospitality in loaning us their spare laboratory and helping us set up housekeeping. I would also like to thank Daryl Bird, Ed Herna, and Leo Jehl of OEHL for their help with halide and metal analysis that was invalubie to this project. John Barnaby, Will Robinson, Chris Ritter, and Rob Siller also added to making this an enjoyable summer with their sense of humor.

I. Introduction

Much inorganic photochemistry in recent years has dealt with the behavior of molecules when in the excited state, since a variety of transitions were possible and in the excited state a variety of reactions may occur. Most molecules that have been studied have a primary absorber that is intensely colored and is excited by visible or near-UV radiation. It has been found that transition metals complexed with aromatic nitrogen donors, especially 2,2'-bipyridine and 1,10-phenanthroline, may possess many of these intense absorbers, leading to very rich spectra (1-7). The various peaks are due to the many possible transitions available to the electron in these complexes, not only are the d to d, and ligand to ligand transitions possible but also some charge transfers either from the ligand to the metal or vice versa from the metal to the ligand. Much research has blossomed in this field with the epiphany of the laser since reasonable concentrations of the excited states are now accessible. This has lead to greater insight into the processes that occur when transition metals absorb light, such as luminescence (8), electron-transfer (9), photochemistry (10), excited state absorbtion (11, 12), and solvatochromism (3, 13).

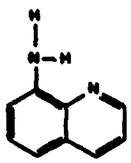
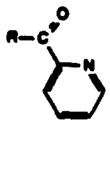
The Schiff base derived from the condensation of 8-quinolinamine and pyridine-2-carboxaldehyde has yielded complexes with cobalt (II) and iron (II) that are expected to demonstrate a similar metal nitrogen bonding scheme to those complexes containing 2,2'-bipyridine and 1,10-phenanthroline. (14) The Schiff base complexes, therefore, are expected to display similar spectral properties.

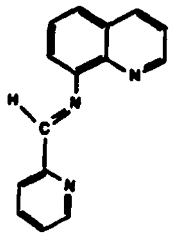
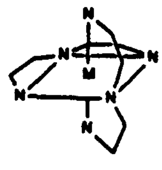
The excited state absorbance of these complexes is of great interest since processes therein may make it possible for two orthogonally oriented laser beams to interact. This property would be applicable to the development of a tunable optical switch using SEPI techniques. (15)

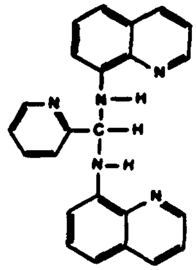
II. Objectives

The general goal of this summer internship was to synthesize a series of transition metal complexes (IV) with the tridentate ligand 8[(pyridine-2-methylene) amino]quinoline (SB) (III). This ligand is a Schiff base created by the condensation of 8-quinolinamine (I) and pyridine-2-carboxaldehyde (II).

The first step was to develop a more direct route by which the iron(II) complex could be synthesized. The hope was to be able to shorten the process outlined by Dwyer (1953) that required an intermediate step of synthesizing an aminal (V) before the ligand could be complexed to the metal. This process was then to be applied to other transition metals, such as ruthenium(II) and chromium(III), with the intent to synthesize a series of the Schiff base complexes. The complexes were then to be tested for their potential to create interaction between two othogonal laser beams of 633 nm and 532 nm light.



I. II. III. IV. V.

III.

a. Materials and chemicals were all either reagent grade or research grade. Ligand reactants were supplied by Sigma-Aldrich and were used without further purification. Solvents were also used as supplied by their source. Analytical services were provided by the Occupational and Environmental Health Laboratories (OEHL) at Brooks AFB. Infrared spectra were obtained using KBr disks with a Perkin-Elmer 1600 FTIR. Spectra of solutions in the near IR, visible and ultraviolet were measured with either a Cary 2400 or a Cary 2315 spectrophotometer using one centimeter quartz cuvettes. The lack of fluorescence was observed with a Perkin-Elmer MPF-44B Fluorescence Spectrophotometer using one centimeter fluorometric cells irradiated with a Xenon lamp.

Preparation of the aminal (pyridyl-N,N'-(di-8-quinolyl)-amino)-methane (PQAM). (14, 15) Pyridine-2-carboxaldehyde (1.68 g, 15.7 mmol) was added to a solution of 8-quinolinamine (4.5 g, 31.21 mmol) in ethanol (3.6 ml) and the mixture warmed to 40^{o}C for five minutes. After cooling the solution to room temperature, ether (150 ml) was added to the solution and a yellow precipitate formed which was isolated using suction filtration. The precipitate was recrystalized by dissolving it in ethanol and inducing precipitation with ether; 52% yield. The product was confirmed by comparing its melting point and infrared spectra to those listed in the literature. m.p. 124-125°C. (m.p. 123-125°C (14); m.p. 123.5°C (16)).

Preparation of the $[Fe(SB)_2]I_2$ starting with the aminal. A solution of ferrous sulfate (.054 g, .19 mmol) dissolved in water (1 ml), was added to a solution of PQAM (.129 g, .34 mmol) dissolved in hot methanol (25 ml). The mixture was heated to 70°C and turned a dark forest green. Water (40 ml) was then added to the mixture and the solution was filtered. A saturated solution of potassium iodide (40 ml) was added to the mixture and a dark precipitate formed. The product was filtered, recrystallized in water and washed with isopropyl alcohol followed by ether; 62% yield after recrystalization.

Preparation of $[Fe(SB)_2]I_2$ by a new direct reaction. Iron sulfate heptahydrate (.570 g, 2.05 mmol) was dissolved in water (15 ml) and added to a solution of 8-quinolinamine (.551 g, 3.82 mmol)dissolved in methanol (50 ml). This solution turned a deep golden color. Pyridine-2-carboxaldehyde (.41 g, .46 ml, 3.82 mmol) was added to this solution via a syringe. Upon addition of the pyridine-2-carboxaldehyde the solution immediately turned a deep forest greeen typical of the iron Schiff bass complex. Water (40 ml) was added and the solution was filtered. Then a saturated solution of potassium iodided dissolved in water (200 ml) was added to the solution and a dark precipitate formed. The precipitate was isolated using suction fitration and was recrystallized in hot water followed by washings with isopropyl alcohol and ether. the crystals were of a fine black crystalline powder. 72% yield after recrystalization. [Iron analysis [$Fe(SB)_2$]I_2 calculated Fe% - 7.217%; found Fe% - 7.994%]

The products synthesized via both routes exhibited identical characteristics, including infrared spectra, indicating the same product was formed. There are added benefits, however, to the direct reaction method. The reaction time is cut in half since the intermediate step of synthesizing the aminal is not necessary and the overall yield is dramatically higher. Not only is the yield with respect to the iron increased (62% yield - starting with the aminal; 72% yield - direct reaction), but also with respect to the ligand reactants. There was no loss of the 8-quinoline when it was spliced off of the aminal to form the ligand nor is it or the other ligand reactacts lost during the synthesis of the aminal.

A similar proceedure was used to isolate an optical isomer of the $Fe(SB)_2I_2$ complex. The direct reaction synthesis of the complex was run and the product was isolated using barium d-tartrate which was obtained in the following manner. Barium chloride [$BaCl_2 \cdot 2H_2O$ (.93 g, 3.8 mmol)] was dissolved in a minimal amount of water (3 ml). D-tataric acid (.57 g, 3.8 mmol) was dissolved in a minimal amount of water. The two solutions were heated to 90°C at which point they were combined and neutralized to a pH of 7 with ammonium hydroxide. A white precipitate formed which was added to the forest green solution containing the iron complex. The solution was then heated and stirred over low heat for 20 min and filtered. Excess solvent was evaporated and the solution was cooled in the refrigerator for 72 hours. Using suction filtration, a black precipitate was isolated and then washed with isopropyl alcohol and ether and then allowed to air dry. The solid was redissolved in a minimal amount of water and filtered. A concentrated solution of potassium iodide was added until a precipitate formed. The precipitate was isolated using suction filtration and was washed with isopropyl alcohol and ether. There was not enough time to do follow up work on the product, such as determining its optical rotation using a polarimeter.

Preparation of the ruthenium complex (RuSB1) and the chromium complex (CrSB1). $RuCl_3 \cdot xH_2O$ (.40 g, 1.91 mmol)) was dissolved in water (20 mL) and 8-quinolinamine (.55 g, 3.8 mmol) was dissolved in ethanol (20 mL). The mixtures were combined in a round bottom flask and a mixture of 1:1 ethanol:water (30 mL) was added. The pyridine-2-carboxaldehyde (.41 g, .46 ml, 3.81 mmol) was added via syringe. Oxalic acid was added to the solution to act as a reducing agent (.142 g, 1 mmol). At this point, the solution was intensely dark with a greenish tinge. The solution was allowed to reflux 96 hours and the solution took on a hue of maroon. The reaction solvent was rotary evaporated off until only a black solid remained. The black solid was redissolved in methanol and filtered. A concentrated solution of ammonium hexafluorophosphate (NH_4PF_6)dissolved in methanol was added until a black precipitate formed which was isolated using suction filtration. A thin layer chromatography of the product indicated that two products were present. The product was purified through a Al_2O_3 chromatography column (approximately 20 cm long) using 10:3:5 toluene:methanol: acetonitrile as the eluant.

The above reaction was run to synthesize the chromium complex, CrSB1 using $CrCl_3 \cdot 6H_2O$ (.51 g, 1.91 mmol) instead of $RuCl_3 \cdot xH_2O$ (.40 g, 1.91 mmol).

b. The 532 nm beam created by doubling the frequncy output from a YAG mode locked, Q-switched laser was diverted and aligned to intersect orthogonally with a 633 nm beam from a helium-neon laser. The 1064 beam from the YAG laser was blocked a short distance from the source for safety. A solution of the RuSB1 dissolved in methanol (164 mg/L) was placed at the point of intersection in a 0.2 cm fluorometric cell. The visible light intensities were monitored with a Thorn EMI, Inc. C-10 Photon Counter system. The photomultiplier tube was cooled electronically and monitored by an EMI Gencom Inc. FACT 50 MKIII device. During measurements, the tube temperatures were maintained between -23^{o}C and -26^{o}C. Uncertainty in the intensity measurements was less than 2%. This was estimated using standard deviations obtained from the average of repeated measurements. Each reading was corrected for dark counts obtained before, during and after the experiment. The beam intensities were adjusted with neutral density filters to read at least a ten-fold increase over the dark count readings.

Beam interaction was detected by monitoring the intensity of the 633 nm beam while switching the 532 nm beam on and off. Table 1 and Figure 1 summarize and analyze the spectral data indicating that the two beams exhibited some interaction. The magnitude of the interaction was dependant upon the power density of the 532 nm beam. Figure 1 indicates that the data are demontrating behavior typical to standard saturation. However, more data are necessary to confirm the interaction and the influence of RuSB1.

IV. RECOMMENDATIONS

The excited state spectra for the 8[(pyridine-2-methylene)amino]quinoline complexes should be investigated further for possible effects they may have on beam interaction. The study of them should be expanded to include a wider range of wavelengths. This may lead to the observance of a more dramatic effect or the development of switches that would be triggered by beams of different wavelengths.

Additional transition metal complexes of this and other similar Schiff base ligands need to be synthesized, and characterized. These complexes have the versatility in that they strongly absorb in the visible region and the position of the wavelength maximum can be adjusted using sutiable substituents on the aromatic ligands and the proper selection of the central transition metal.

The RuSB1 and CrSB1 synthesized this summer also need follow up research. Elemental analysis for carbon, hydrogen and nitrogen is needed to confirm their formulation and chemical nature. More research is needed to confirm the beam interaction observed using RuSB1 and the surface has only been scratched as to the possibility of CrSB1 creating an observable effect.

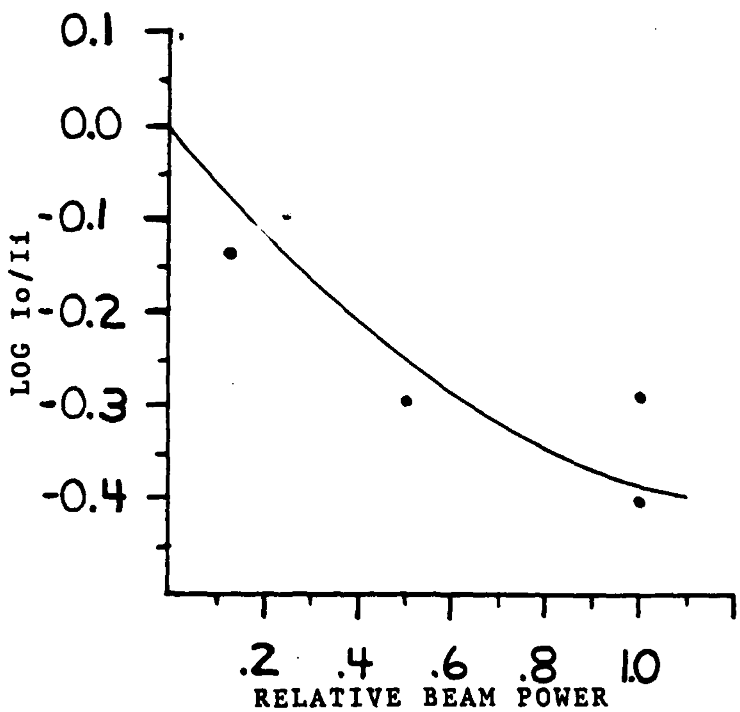
TABLE I. DATA FOR RuSB1 OPTICAL DENSITY

CHANGE AT 633 nm VS POWER OF 532 nm BEAM

N.D. FILTER OD	EXPERIMENTAL 10exp-OD	Io/Ii CORR.	LOG Io/Ii
0.0	1.000	.9358	-0.0288
0.0	1.000	.9122	-0.0399
0.3	0.501	.9343	-0.0295
0.6	0.251	.9786	-0.0094
0.9	0.126	.9722	-0.0122

Io and Ii refer to the intensities of the 633 nm beam with the 532 nm beam being switch off and on respectively.

FIGURE 1. PLOT OF DATA FROM TABLE I.



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FINAL REPORT

ELECTROMAGNETIC WAVE PROPAGATION IN A ONE-DIMENSIONAL RANDOM MEDIUM

Prepared By: Julia Rennenkampff
Academic Rank: Master of Science
Department and University: Courant Institute of Mathematical Sciences, New York University
Research Location: USAFSAM/RZM
Brooks AFB
San Antonio, Texas 78235
USAF Researcher: Dr. Richard A. Albanese, M.D.
Date: August 19, 1988
Contract No: F49620-88-C-0053

ELECTROMAGNETIC WAVE PROPAGATION IN A ONE-DIMENSIONAL RANDOM MEDIUM

by

Julia Rennenkampff

ABSTRACT

Given a slab of a temporally dispersive medium whose dielectric constant and conductivity vary deterministically on a large scale and randomly on a small scale compared to the width of an incident electromagnetic pulse, we characterize the approximate autocorrelation function for the reflected signal at the surface of the medium.

ACKNOWLEDGEMENTS

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I would also like to thank everyone in the Radiation Sciences Division at Brooks Air Force Base for welcoming me and for making my summer there as enjoyable as it was. In particular, I am grateful to Dr Albanese for all his friendship and support, and for all his help with this problem. I would like to thank Mr Medina and Mr Penn for their interest and support, Ms Wood for this excellent typing job, Lt Nielsen for his help in getting me settled in San Antonio and for being a great officemate, and Mr Mitchell and Dr Burton for their concern and encouragement.

I. INTRODUCTION

Dr Albanese and his group are interested in the propagation of electromagnetic pulses in human tissue. They would like to understand the direct problem, determining the electromagnetic field from the known structure of the medium, as well as the inverse problem, namely that of inferring the properties of the medium from the reflected or transmitted wave. Being able to solve the first problem would make it possible to set realistic safety standards for exposure to electromagnetic radiation, whereas a solution of the second may make it possible to use electromagnetic pulses as an imaging tool.

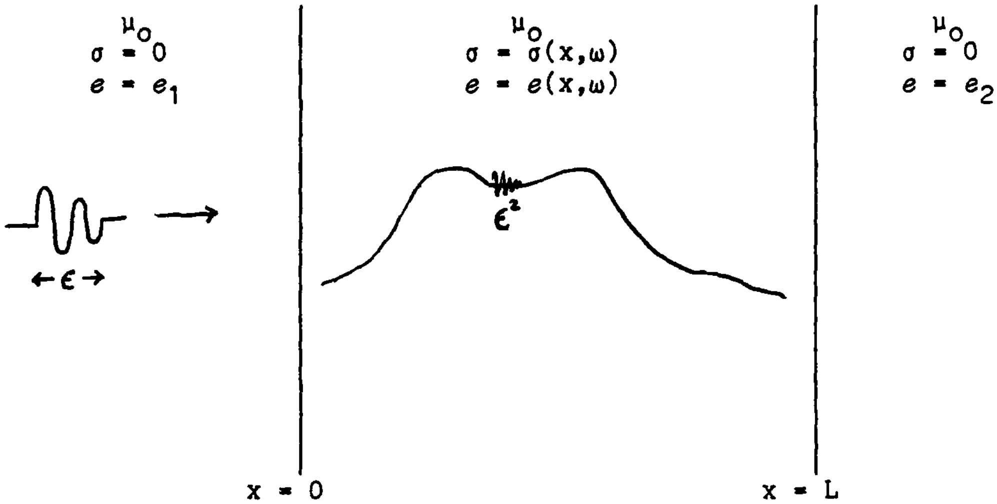
I have been working on the specific problem of characterizing the electromagnetic field resulting from a pulse which is incident on a randomly layered slab of a dispersive medium when the scale on which the medium varies randomly is small compared to the width of the pulse, which in turn is small compared to the scale on which the medium varies deterministically.

Human tissue is a dispersive medium, and may be regarded as random in several ways, so our model may be applicable.

II. OBJECTIVES

We consider a slab of a temporally dispersive medium located between the planes $x = 0$ and $x = L$, with the dielectric constant e and the conductivity σ depending on x, x/ε^2, and the frequency ω, with the dependence on the second variable being random. Let the half-spaces $x < 0$ and $x > L$ be occupied by homogeneous, nondispersive, nonconducting media, and let the magnetic permeability be constant throughout.

Figure 1



We would like to characterize the response of this medium to an electromagnetic pulse which is incident from the left ($x<0$), and whose width is of order ε, Figure 1.

Since the medium properties vary randomly, the electromagnetic field due to the pulse will be a stochastic process in t and x. We hope to characterize the approximate statistics of this process at $x = 0$ by asymptotic analysis of the relevant stochastic equations in the small parameter ε.

III. FORMULATION AND SCALING

Define $\hat{E}(x,\omega) = \int_{-\infty}^{\infty} E(x,t)e^{i\omega t}dt.$

From Maxwell's equations, we have that

$$\hat{E}_{xx}(x,\omega) = -\mu_0\omega^2\left(e(x,\omega) + \frac{i}{\omega}\sigma(x,\omega)\right)\hat{E}(x,\omega)$$

$$= -\mu_0\omega^2\alpha(x,\omega)\hat{E}(x,\omega).$$

To make the scaling assumptions above precise, we let

$E(x,t) = E(\frac{x'}{\varepsilon^2},\frac{t'}{\varepsilon^2}) = E'(x',t')$, and consequently

$\hat{E}(x,\omega) = \hat{E}(\frac{x'}{\varepsilon^2},\varepsilon\omega') = \hat{E}'(x',\omega')$.

Also, we assume that for $0 < x' < L$, $\alpha(x,\omega) = \bar{\alpha}(x',\varepsilon\omega')\bullet$ $(1 + \eta(x',x'/\varepsilon^2,\varepsilon\omega'))$, η with mean zero in the second variable.

We are interested, first, in the response to a δ-function incident pulse

$$E'(x',t') = (\frac{\mu_0}{e_1})^{1/4}\delta(t' - \frac{x'}{c_1}),$$

$$t' < 0,\ x' < 0,\ c_1^2 = \frac{1}{e_1\mu_0}.$$

The response to this pulse is described by the system (dropping primes)

$$\hat{E}_{xx}(x,\omega) = -\mu(\omega^2/\varepsilon^2)\alpha(x,x/\varepsilon^2,\varepsilon\omega)\hat{E}(x,\omega) \qquad 0 < x < L$$

$$\hat{E}(x,\omega) = (\mu_0/e_1)^{1/4}(e^{i\omega x/\varepsilon c_1} + \hat{R}(\omega)e^{-i\omega x/\varepsilon c_1}) \qquad x < 0 \qquad 3.1$$

$$\hat{E}(x,\omega) = (\mu_0/e_2)^{1/4}(\hat{T}(\omega)e^{i\omega x/\varepsilon c_2}) \qquad x > L$$

Denote the solution of 3.1 by $\hat{E}^\varepsilon(x,\omega)$.

The response to an incident pulse of the form $(1/\sqrt{\varepsilon})f([t-(x/c_1)]/\varepsilon)$ is then given by

$$E(x,t) = \int_0^t f(s/\varepsilon)E^\varepsilon(x,t-s)ds$$

$$= (1/\sqrt{\varepsilon})(1/\sqrt{2\pi})\int_{-\infty}^{\infty}\hat{f}(\omega)\hat{E}^\varepsilon(x,\omega)e^{-i\omega t/\varepsilon}d\omega,$$

where $\hat{f}(\omega) = \int_{-\infty}^{\infty} f(t)e^{i\omega t}dt.$

We are interested in the statistics of $E(x,t)$ at $x = 0$ and at times $t^{\varepsilon} = \tau + \varepsilon\sigma$ for τ fixed.

Let $\xi(x,\omega) = \int_0^x \sqrt{\mu_0 \bar{\alpha}(x,\omega)}\, dx.$

Let $\xi^{\varepsilon} = \xi(x,\varepsilon\omega)$, $\alpha^{\varepsilon} = \alpha(x,x/\varepsilon^2,\varepsilon\omega)$, $\bar{\alpha}^{\varepsilon} = \bar{\alpha}^{\varepsilon}(x,\omega)$.

$$\text{Let } \hat{E}(x,\omega) = (\mu_0/\bar{\alpha}^{\varepsilon})^{1/4}[\hat{A}(x,\omega)e^{i\omega\xi^{\varepsilon}/\varepsilon} + \hat{B}^{\varepsilon}(x,\omega)e^{-i\omega\xi^{\varepsilon}/\varepsilon}]$$

$$\text{and } \hat{E}_x(x,\omega) = i(\omega/\varepsilon)(\mu_0^3\bar{\alpha}^{\varepsilon})^{1/4}[\hat{A}(x,\omega)e^{i\omega\xi^{\varepsilon}/\varepsilon} - \hat{B}(x,\omega)e^{-i\omega\xi^{\varepsilon}/\varepsilon}]. \qquad 3.2$$

From 3.2 and 3.1, it follows that

$$\frac{\partial}{\partial x}\begin{pmatrix}\hat{A}(x,\omega)\\ \hat{B}(x,\omega)\end{pmatrix} = \frac{1}{2}\frac{i\omega}{\varepsilon}\eta^{\varepsilon}\sqrt{\mu_0\bar{\alpha}^{\varepsilon}}\begin{pmatrix}1 & e^{-2i\omega\xi^{\varepsilon}/\varepsilon}\\ -e^{2i\omega\xi^{\varepsilon}/\varepsilon} & -1\end{pmatrix}\begin{pmatrix}\hat{A}\\ \hat{B}\end{pmatrix}$$

$$+ (1/4)(\bar{\alpha}^{\varepsilon\prime}/\bar{\alpha}^{\varepsilon})\begin{pmatrix}0 & e^{-2i\omega\xi^{\varepsilon}/\varepsilon}\\ e^{2i\omega\xi^{\varepsilon}/\varepsilon} & 0\end{pmatrix}\begin{pmatrix}\hat{A}\\ \hat{B}\end{pmatrix}. \qquad 3.3$$

Let $\hat{E}^{\varepsilon}(x,\omega) = (\mu_0/\bar{\alpha}^{\varepsilon})^{1/4}[\hat{a}^{\varepsilon}(x,\omega)e^{i\omega\xi^{\varepsilon}/\varepsilon} + \hat{b}^{\varepsilon}(x,\omega)e^{-i\omega\xi^{\varepsilon}/\varepsilon}]$. Then $\hat{a}^{\varepsilon}$, $\hat{b}^{\varepsilon}$ satisfy 3.3 with boundary conditions

$$\hat{a}^{\varepsilon}(0,\omega) = \frac{1}{2}\left[(c_1+1/c_1)+(c_1-1/c_1)\hat{R}^{\varepsilon}(\omega)\right]$$

$$\hat{a}^{\varepsilon}(L,\omega) = \frac{1}{2}\left[(c_2+1/c_2)\hat{T}^{\varepsilon}(\omega)\right]e^{i\omega L/\varepsilon c_2 - i\omega\xi(L,\varepsilon\omega)/\varepsilon}$$

$$\hat{b}^{\varepsilon}(0,\omega) = \frac{1}{2}\left[(c_1-1/c_1)+(c_1+1/c_1)\hat{R}^{\varepsilon}(\omega)\right]$$

$$\hat{b}^{\varepsilon}(L,\omega) = \frac{1}{2}\left[(c_2 - 1/c_2)\hat{T}^{\varepsilon}(\omega)\right]e^{i\omega L/\varepsilon + i\omega\xi(L,\varepsilon\omega)/\varepsilon}$$

$$c_1 = \left(\overline{\alpha}^{\varepsilon}(0,\varepsilon\omega)/e_1\right)^{1/4}, \quad c_2 = \left(\overline{\alpha}^{\varepsilon}(L,\varepsilon\omega)/e_2\right)^{1/4}.$$

In the time domain, define,

$$A^{\varepsilon}_{f,\tau,x}(\sigma) = \int_0^{\tau+\varepsilon\sigma} f(s/\varepsilon)A^{\varepsilon}(x,\tau + \varepsilon\sigma - s)ds$$

$$= \frac{1}{\varepsilon}\frac{1}{4\pi^2}\int_{-\infty}^{\infty}\hat{f}(\omega)\hat{A}^{\varepsilon}(x,\omega)e^{-i\omega(\tau+\varepsilon\sigma)/\varepsilon}d\omega$$

$$B^{\varepsilon}_{f,\tau,x}(\sigma) = \int_0^{\tau+\varepsilon\sigma} f(s/\varepsilon)B^{\varepsilon}(x,\tau + \varepsilon\sigma - s)ds$$

$$= \frac{1}{\varepsilon}\frac{1}{4\pi^2}\int_{-\infty}^{\infty}\hat{f}(\omega)\hat{B}^{\varepsilon}(x,\omega)e^{-i\omega(\tau+\varepsilon\sigma)/\varepsilon}d\omega,$$

where $\hat{A}^{\varepsilon}(x,\omega) = \hat{a}^{\varepsilon}(x,\omega)e^{i\omega\xi^{\varepsilon}/\varepsilon}$

$$\hat{B}^{\varepsilon}(x,\omega) = \hat{b}^{\varepsilon}(x,\omega)e^{-i\omega\xi^{\varepsilon}/\varepsilon}$$

The correlation functions for the right- and left-going waves at any fixed τ and x are given by

$$E\left\{\begin{matrix} A^{\varepsilon}_{f,\tau,x}(\sigma)A^{\varepsilon}{}_{f,\tau,x}(0) \\ B^{\varepsilon}_{f,\tau,x}(\sigma)B^{\varepsilon}{}_{f,\tau,x}(0) \end{matrix}\right\}$$

$$= \frac{1}{\varepsilon}\frac{1}{4\pi^2}\int_{-\infty}^{\infty}\hat{f}(\omega_1)\overline{\hat{f}}(\omega_2)E\left\{\begin{matrix} \hat{a}^{\varepsilon}(x\ ,\omega_1)\overline{\hat{a}}^{\varepsilon}(x\ ,\omega_2)e^{i\omega(\xi_1^{\varepsilon}-\xi_2^{\varepsilon})/\varepsilon} \\ \hat{b}^{\varepsilon}(x\ ,\omega_1)\overline{\hat{b}}^{\varepsilon}(x\ ,\omega_2)e^{-i\omega(\xi_1^{\varepsilon}-\xi_2^{\varepsilon})/\varepsilon} \end{matrix}\right\}$$

$$\cdot\; e^{-i\omega_1(\tau/\varepsilon) - i\omega_1\sigma + i\omega_2(\tau/\varepsilon)} d\omega_1 d\omega_2,$$

where $\xi_i^\varepsilon = \xi(x,\varepsilon\omega_i)$, $i = 1,2$.

Let $\omega = (1/2)(\omega_1+\omega_2)$, $h = (1/\varepsilon)(\omega_2-\omega_1)$, so $\omega_1 = \omega-\varepsilon h/2$, $\omega_2 = \omega+\varepsilon h/2$.

The above equation then becomes

$$\frac{1}{4\pi^2}\int_{-\infty}^{\infty}\int_{-\infty}^{\infty}\hat{f}(\omega-(\varepsilon/2)h)\overline{\hat{f}}(\omega+(\varepsilon/2)h)$$

$$\cdot\; E\begin{Bmatrix}\hat{a}^\varepsilon(x,\omega-(\varepsilon/2)h)\overline{\hat{a}}^\varepsilon(x,\omega+(\varepsilon/2)h)e^{i\omega(\xi_1^\varepsilon-\xi_2^\varepsilon)/\varepsilon}\\ \hat{b}^\varepsilon(x,\omega-(\varepsilon/2)h)\overline{\hat{b}}^\varepsilon(x,\omega+(\varepsilon/2)h)e^{i\omega(\xi_1^\varepsilon-\xi_2^\varepsilon)/\varepsilon}\end{Bmatrix}$$

$$\cdot\; e^{ih\tau-i\omega\sigma+i(\varepsilon/2)h\sigma}d\omega dh$$

$$= \frac{1}{4\pi^2}\int_{-\infty}^{\infty}\int_{-\infty}^{\infty}\hat{f}(\omega-(\varepsilon/2)h)\overline{\hat{f}}(\omega+(\varepsilon/2)h)$$

$$\cdot\; E\begin{Bmatrix}\hat{a}_1^\varepsilon\;\overline{\hat{a}_2^\varepsilon}\;e^{i\omega(\xi_1^\varepsilon-\xi_2^\varepsilon)/\varepsilon}\\ \hat{b}_1^\varepsilon\;\overline{\hat{b}_2^\varepsilon}\;e^{-i\omega(\xi_1^\varepsilon-\xi_2^\varepsilon)/\varepsilon}\end{Bmatrix}\cdot\; e^{i\omega\tau-i\omega\sigma+i(\varepsilon/2)h\sigma}d\omega dh.$$

Let $u^\varepsilon(x,\omega,h) = E\begin{Bmatrix}\hat{a}_1^\varepsilon & \overline{\hat{a}}_2^\varepsilon\\ \hat{b}_1^\varepsilon & \overline{\hat{b}}_2^\varepsilon\end{Bmatrix}$.

Claim: $u^\varepsilon(x,\omega,h) \to u(x,\omega,h)$ as $\varepsilon\to 0$, and

$$E\begin{Bmatrix}A_{f,\tau,x}^\varepsilon(\sigma)A_{f,\tau,x}^\varepsilon(0)\\ B_{f,\tau,x}^\varepsilon(\sigma)B_{f,\tau,x}^\varepsilon(0)\end{Bmatrix}\to$$

$$\frac{1}{4\pi^2}\int_{-\infty}^{\infty}\int_{-\infty}^{\infty}|\hat{f}(\omega)|^2 u(x,\omega,h)e^{ih\tau-i\omega\sigma}d\omega dh. \qquad 3.4$$

We will describe this correlation function at $x = 0$.

Let $\hat{v}^{\varepsilon}(x) = \begin{pmatrix}\hat{A}_1^{\varepsilon}\\ \hat{B}_1^{\varepsilon}\\ \hat{A}_2^{\varepsilon}\\ \hat{B}_2^{\varepsilon}\end{pmatrix}$

where $\begin{pmatrix}\hat{A}_j^{\varepsilon}\\ \hat{B}_j^{\varepsilon}\end{pmatrix}$ are solutions of 3.3 with $\omega = \omega_j$, $j = 1,2$.

(I am omitting the dependence on ω,h for now.)

We will approximate

$$E_{v,\eta}\{f(\hat{v}^{\varepsilon}(x),\eta^{\varepsilon}(x))\} = E\{f(\hat{v}^{\varepsilon}(x),\eta^{\varepsilon}(x))|\hat{v}^{\varepsilon}(x_0) = v,\eta^{\varepsilon}(x_0) = \eta\},$$

for any smooth function f and any fixed x_0.

Since there exists a smooth function $f_u(Y^{\varepsilon}(x))$, where $Y^{\varepsilon}(x)$ is the fundamental solution of the differential equation satisfied by $\hat{v}^{\varepsilon}(x)$, such that $u^{\varepsilon}(o,\omega,h) = E\{f_u(Y^{\varepsilon}(0))|Y^{\varepsilon}(L) = I\}$, this will yield $u(0,\omega,h)$. $\hat{v}(x)$ satisfies

$$\frac{\partial}{\partial x}\hat{v}^{\varepsilon}(x) = \left(\begin{array}{l} \left[\frac{i}{2}\frac{\omega_1}{\varepsilon}\eta(x,x/\varepsilon^2,\varepsilon\omega_1)\sqrt{\mu_0\bar{\alpha}(x,\varepsilon\omega_1)}\begin{pmatrix} 1 & e^{-2i\omega_1^{\varepsilon}\xi_1/\varepsilon} \\ -e^{2i\omega_1\xi_1^{\varepsilon}/\varepsilon} & -1 \end{pmatrix}\right. \\ \left[\frac{i}{2}\frac{\omega_2}{\varepsilon}\eta(x,x/\varepsilon^2,\varepsilon\omega_2)\sqrt{\mu_0\bar{\alpha}(x,\varepsilon\omega_2)}\begin{pmatrix} 1 & e^{-2i\omega_2^{\varepsilon}\xi_2/\varepsilon} \\ -e^{2i\omega_2\xi_2^{\varepsilon}/\varepsilon} & -1 \end{pmatrix}\right. \end{array}\right. \tag{4.1}$$

$$+\ \frac{1}{4}\frac{\bar{\alpha}'(x,\varepsilon\omega_1)}{\bar{\alpha}(x,\varepsilon\omega_1)}\left.\begin{pmatrix} 0 & e^{-2i\omega_1\xi_1^{\varepsilon}/\varepsilon} \\ e^{2i\omega_1\xi_1^{\varepsilon}/\varepsilon} & 0 \end{pmatrix}\right]\begin{pmatrix}\hat{A}_1^{\varepsilon} \\ \hat{B}_1^{\varepsilon}\end{pmatrix}$$

$$\left.+\ \frac{1}{4}\frac{\bar{\alpha}'(x,\varepsilon\omega_2)}{\bar{\alpha}(x,\varepsilon\omega_2)}\left.\begin{pmatrix} 0 & e^{-2i\omega_2\xi_2^{\varepsilon}/\varepsilon} \\ e^{2i\omega_2\xi_2^{\varepsilon}/\varepsilon} & 0 \end{pmatrix}\right]\begin{pmatrix}\hat{A}_2^{\varepsilon} \\ \hat{B}_2^{\varepsilon}\end{pmatrix}\right)$$

Let $(i/2)\eta(x,x/\varepsilon^2,\varepsilon\omega\mp\varepsilon^2h/2)$

$$= \eta_0(x,x/\varepsilon^2,0) + \eta_1(x,x/\varepsilon^2,0)(\varepsilon\omega\mp\varepsilon^2h/2) + O(\varepsilon^2)$$

$$\frac{1}{4}\frac{\bar{\alpha}'(x,\varepsilon\omega\mp\varepsilon^2h/2)}{\bar{a}(x,\varepsilon\omega\mp\varepsilon^2h/2)} = \nu_0(x,0) + \nu_1(x,0)(\varepsilon\omega\mp\varepsilon^2h/2) + O(\varepsilon^2).$$

$$\sqrt{\mu_0\bar{\alpha}(x,\varepsilon\omega\mp\varepsilon^2h/2)} = \gamma_0(x,o) + \gamma_1(x,o)(\varepsilon\omega\mp\varepsilon^2h/2) + O(\varepsilon^2)$$

Let $p_j^{\varepsilon} = \begin{pmatrix} 1 & e^{2i\omega_j\xi_j^{\varepsilon}/\varepsilon} \\ -e^{2i\omega_j\xi_j^{\varepsilon}/\varepsilon} & -1 \end{pmatrix}$, $\eta_j = \begin{pmatrix} 0 & e^{2i\omega_j\xi_j^{\varepsilon}/\varepsilon} \\ e^{2i\omega_j\xi_j^{\varepsilon}/\varepsilon} & 0 \end{pmatrix}$, $j = 1,2.$

Then

$$\frac{\partial}{\partial x}\hat{v}^{\varepsilon}(x) = \begin{array}{l} \left[(1/\varepsilon)\left(\omega\eta_0\gamma_0 + \varepsilon(-h\eta_0\gamma_0/2 + \eta_0\omega^2\gamma_1 + \gamma_0\omega^2\eta_1) + O(\varepsilon^2)\right)p_1^{\varepsilon}\right. \\ \left[(1/\varepsilon)\left(\omega\eta_0\gamma_0 + \varepsilon(h\eta_0\gamma_0/2 + \eta_0\omega^2\gamma_1 + \gamma_0\omega^2\eta_1) + O(\varepsilon^2)\right)p_2^{\varepsilon}\right. \\ \left. + \left(\nu_0 + \varepsilon\nu_1\omega + O(\varepsilon^2)\right)\eta_1^{\varepsilon}\right]\begin{pmatrix}\hat{A}_1^{\varepsilon} \\ \hat{B}_1^{\varepsilon}\end{pmatrix} \end{array}$$

$$+ \left(\nu_0 + \varepsilon\nu_1\omega + O(\varepsilon^2)\right)\eta_2^\varepsilon\Big] \begin{pmatrix} \hat{A}_2^\varepsilon \\ B_2^\varepsilon \end{pmatrix}$$

$$= \frac{1}{\varepsilon} \begin{bmatrix} \omega\eta_0\gamma_0 p_1^\varepsilon & 0 \\ 0 & \omega\eta_0\gamma_0 p_2^\varepsilon \end{bmatrix} \hat{v}^\varepsilon$$

$$+ \begin{bmatrix} (-h\eta_0\gamma_0/2+\eta_0\omega^2\gamma_1)p_1^\varepsilon + \nu_0\eta_1^\varepsilon & 0 \\ 0 & (h\eta_0\gamma_0/2+\eta_0\omega^2\gamma_1)p_2^\varepsilon + \nu_0\eta_2^\varepsilon \end{bmatrix} \hat{v}^\varepsilon$$

$$+ \; O(\varepsilon)\hat{v}^\varepsilon$$

$$= \; \frac{1}{\varepsilon} G\left(x,\bar{\eta}(x,x/\varepsilon^2,0),\xi^\varepsilon/\varepsilon,\omega,h\right)\hat{v}^\varepsilon$$

$$+ \; H\left(x,\bar{\eta}(x,x/\varepsilon^2,0),\xi^\varepsilon/\varepsilon,\omega,h\right)\hat{v}^\varepsilon + O(\varepsilon)\hat{v}^\varepsilon.$$

Let $\eta^\varepsilon = \bar{\eta}(x,x/\varepsilon^2,0) = \left(\eta_0(x,x/\varepsilon^2,0),\eta_1(x,x/\varepsilon^2,0)\right)$.

We do the asymptotic analysis on this equation. We assume that $\bar{\eta}(x,z,0)$ is a stationary ergodic process with the transition function $P_x(Z,\eta,A)$, invariant measure $\bar{P}_x(A)$, and infinitesimal generator Q_x. Denote the expectation with respect to $\bar{P}_x$ by E'.

Let $\psi_x(\eta,A) = \int_0^\infty [P_x(z,\eta,A) - \bar{P}_x(A)]dz$.

For any smooth function $f(v,\eta)$, let $Y^\varepsilon(x,v,\eta) = E_{v,\eta}\{f(\hat{v}^\varepsilon(x),\eta^\varepsilon(x))\}$.

$$= y_0 + \varepsilon y_1 + \varepsilon^2 y_2 + \ldots$$

$$\frac{\partial y^\varepsilon}{\partial x} = \frac{1}{\varepsilon^2} Q_x y^\varepsilon + \frac{1}{\varepsilon} G^\varepsilon{}_v \cdot \nabla_v y^\varepsilon + ..^\varepsilon{}_v \cdot \nabla_v y^\varepsilon + O(\varepsilon)v \cdot \nabla_v y^\varepsilon,$$

$$G^\varepsilon = G(x,\eta,\chi^\varepsilon,\omega,h), H^\varepsilon = H(x,\eta,\chi^\varepsilon,\omega,h), \chi^\varepsilon = \xi^\varepsilon/\varepsilon.$$

Equating coefficients of equal powers of ε yields

$$O(1/\varepsilon^2):\quad 0 = Q_x y_0 \tag{4.2}$$

$$O(1/\varepsilon):\quad 0 = Q_x y_1 + G^{\varepsilon} v \cdot \nabla_v y_0 \tag{4.3}$$

$$O(1):\quad \frac{\partial y_0}{\partial x} = Q_x y_2 + G^{\varepsilon} v \cdot \nabla_v y_1 + H^{\varepsilon} v \cdot \nabla_v y_0. \tag{4.4}$$

4.2 implies that $y_0 = y_0(x,v)$.

The solvability condition for 4.3 is

$$E'\{G^{\varepsilon} v \cdot \nabla_v y_0\} = 0$$

Since y_0 depends only on x and v and $E'\{G^{\varepsilon}\} = 0$, this is satisfied.

Thus, $y_1(x,v,\eta) = \int \psi_x(\eta,dz) G(x,z,\chi^{\varepsilon},\omega,h) v \cdot \nabla_v y_0$.

The solvability condition for 4.4 gives

$$\frac{\partial y_0}{\partial x} = E'\{G^{\varepsilon} v \cdot \nabla_v y_1 + H^{\varepsilon} v \cdot \nabla_v y_0\}$$

$$= E'\left\{G^{\varepsilon} v \cdot \nabla_v \int \psi_x(\eta,dz) G(x,z,\chi^{\varepsilon},\omega,h) v \cdot \nabla_v y_0\right\} + H^{\varepsilon} v \cdot \nabla_v y_0$$

$$= \int_0^{\infty} d\phi E'\left\{G(x,\tilde{\eta}(x,0,0),\chi^{\varepsilon},\omega,h) v \cdot \nabla_v [G(x,\tilde{\eta}(x,\phi,0),\chi^{\varepsilon},\omega,h) v \cdot \nabla_v y_0]\right\}$$

$$+ H^{\varepsilon} v \cdot \nabla_v y_0.$$

Define $\langle \cdot \rangle_{\chi} = \lim_{\chi_0 \to \infty} \frac{1}{\chi_0} \int_0^{\chi_0} \cdot \, d\chi$.

Let $y(x,v,\eta)$ satisfy

$$\frac{\partial y}{\partial x} = \left\langle \int_0^\infty d\phi E'\{G(x,\tilde{\eta}(x,0,0),\chi^\varepsilon,\omega,h)v\cdot\nabla_v[G(x,\tilde{\eta}(x,\phi,0),\chi^\varepsilon,\omega,h)v\cdot\nabla_v y]\} + H^\varepsilon v \cdot \nabla_v y \right\rangle_{\chi^\varepsilon}$$

$$= \int_0^\infty d\phi E'\{\langle G(x,\tilde{\eta}(x,0,0),\chi^\varepsilon,\omega,h)v\cdot\nabla_v[G(x,\tilde{\eta}(x,\phi,0),\chi^\varepsilon,\omega,h)v\cdot\nabla_v y]\rangle_{\chi^\varepsilon}\}$$

$$= L_{x,\omega,h}y.$$

Claim: $y^\varepsilon(x,v,\eta) \to y(x,v,\eta)$ as $\varepsilon\to0$.

Let $Y^\varepsilon(x)$ be the fundamental solution of 4.1 with $Y^\varepsilon(L) = I$, the identity matrix. Then $u^\varepsilon(0,\omega,h) = E_I\{f_u(Y^\varepsilon(0))\}$, where the subscript I indicates the value of Y^ε at $x = L$.

From above, we have, then, that

$$u(0,\omega,h) = \lim_{\varepsilon\to0} u^\varepsilon(0,\omega,h) = y(0,I), \qquad 4.5$$

where $y(x,Y)$ is the solution o^

$$\frac{\partial y}{\partial x} = L_{x,\omega,h}y \quad , \quad 0 < x < L \quad , \quad \text{with } y(L,Y) = f_u(Y).$$

V. RECOMMENDATIONS

I have claimed that the correlation functions of $A^\varepsilon_{f,\tau,0}(\sigma)$ and $B^\varepsilon_{f,\tau,0}(\sigma)$ can be approximated, as $\varepsilon\to0$, by 3.4 with $x=0$ and $u(0,\omega,h)$ given by 4.5. This claim needs to be substantiated. I think that this can be accomplished, chiefly, by applying the theorem in the appendix of [1].

We have characterized the correlation functions, but in order to interpret this result, 4.5 will have to be calculated more explicitly. We are planning to do this, as well as to characterize the other statistics of the medium's response, using similar methods.

We are hoping that once these calculations have been carried out, we will be able to see more clearly the relationship between the deterministic structure of the medium and the statistics of the response at $x=0$.

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FINAL REPORT

Synchronization of the Chlamydomonas reinhardtii cell cycle through light-dark cycling for subsequent testing with infra-red laser light in experiments concerning human night vision

Prepared by:	Mary Christine Ritter
Academic rank:	Graduate Student
Department and University:	Biology Trinity University
Research location:	Lasar spectroscopy Clinical Applications Laboratory Clinical Services Division Scool of Aerospace Medicine Brooks Air Force Base San Antonio, TX 78235
USAF Researcher:	Dr. John Taboada

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Synchronization of the **Chlamydomonas reinhardtii** cell cycle through light-dark cycling for subsequent testing with infra red laser light in experiments concerning human night vision

by

Christine Ritter

ABSTRACT

Synchronization of Chlamydomonas reinhardtii strain 125N through light-dark cycling was attempted through a variety of means: 1) aerated liquid cultures, 2) agar plate cultures with a 5 ml liquid overlay, and 3) non-aerated liquid cultures. These were cycled through 12 hour light-12 hour dark intervals with 2) and 3) being cycled through 18 hour light -6 hour dark intervals as well. A set of cultures were grown under constant light as a control. The light cycled cultures did not double during the dark period or show strong cell size distribution peaks that are expected of synchronization.

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I. INTRODUCTION

Chlamydomonas reinhardtii is a single celled, phototactic algae of 10-15 μm in length. It is roughly spherical in shape, contains a single chloroplast cupped around a single nucleus and has a pair of anterior flagella used for locomotion. It has evolved a specialized means of regulating its exposure to light. This consists of a directional light antenna and the pair of flagella. The light antenna consists of an eye spot with associated structures. This eyespot utilizes the light detecting pigment rhodopsin. The rhodopsin molecule is also utilized in the human eye. It functions in detecting light and is thus an important factor in night vision.

This project was focussed on the synchronization of the Chlamydomonas reinhardtii cell cycle. The information obtained will be utilized in subsequent experiments at the Brooks Air Force Base Vision Biophysics Function in their research concerning the enhancement of night vision of Air Force pilots. The experiments planned will test the affect of infra-red laser light on the phototactic activity of the synchronized cells.

It is important that the cells be in synchrony since the infra-red light may have effects at specific points of the cell life cycle and not at others. The synchronization of the algal cells leads to a majority of the cells existing in more or less the same morphological and physiological state and thus an amplification of cellular events as they would occur in single cells (Surzycki, 1971).

In order to synchronize C. reinhardtii a basic understanding of its vegatative cell cycle is necessary. When grown in liquid media , the cells vegatative cell cycle is necessary. When grown in liquid media , the cells retract their flagella before they divide (Cavalier-Smith, 1974). Division is by multiple fission and may produce 2, 4, 8, 16, or even 32 daughter cells. This takes place in a time span of approximately three hours (Craigie and Cavalier-Smith, 1982). During division the protoplast divides into 2^n daughter cells which remain inside the mother cell wall for several hours until they are liberated by the enzymatic dissolution of this wall (Demets et. al., 1985; Craigie and Cavalier-Smith, 1982).

Craigie and Cavalier-Smith (1982) found that cell volume was a major determinant in the timing of C. reinhardtii cell division and that the larger cells typically yielded a greater number of daughter cells than the smaller so as to maintain a fairly uniform daughter cell size. They found that the larger fraction entered division first and that the timing of subsequent divisions was determined by the first. They also noted that light cycling directly affected the division times and rates and that light could directly initiate cell liberation.

Spudich and Sager (1980) performed experiments on the synchronization of C. reinhardtii. In their paper they note: "By growing cells in alternating periods of light and darkness, we have found that the synchronization of phototropically grown Chlamydomonas populations are regulated at two specific points in the cell cycle: the primary arrest (A) point located in early G , and the transition (T) point, located in mid G . At the A point, cell cycle progression becomes light dependent. At the T point, completion of the cell cycle becomes independent of light. Cells transferred from light to dark at cell cycle positions between the two regulatory points enter a reversible resting state in which they remain viable and metabolically active but do not progress through their cycle" (p.136).

II. OBJECTIVES

This research was originally geared towards a study of infra-red laser light affects on the phototaxis of a synchronized population of Chlamydomonas reinhardtii. As the project progressed and the difficulty of synchronization became apparent, the effort became concentrated on the synchronization of the algae through light and dark cycling and the infra-red phototaxis studies were left for a future project.

Synchronization of the C. reinhardtii cell cycle was attempted through several ways. HS media was used for all cultures though some cultures were grown in aerated liquid media, others in non-aerated liquid media and still others on agar plates with a 5 ml liquid overlay. All were subjected to light-dark cycling of 12 hour intervals. The non aerated cultures and the plates were also subjected to light-dark cycling of 18 hours light and 6 hours dark. Constant light cultures were grown as controls.

III. MATERIALS AND METHODS

A. Aerated Liquid Cultures

Several Chlamydomonas reinhardtii, strain 125N, cultures were grown on HS media so that they would be dependent on light for their metabolic processes and aerated for an ample CO2 supply. The parameters for these varried widely in an attempt to determine and achieve synchrony (SEE TABLE 1).

Each culture was aerated with air that had been filtered through a one foot long glass tube, stuffed firmly with cotton and bubbled through water contained in a side arm flask to be humidified. However, the overall set up of each experiment varied as widely as their parameters (SEE FIGURE 1).

Experiment 1 was sampled once a day for analysis on the model ZBI Coulter Counter whereas experiments 2,3, and 5 (the light cycled cultures) were sampled during the 3rd, 4th, 5th and 6th hours of the dark cycle . Experiment 4 was sampled simultaneously with 5.

The algal cells were counted electronically on the model ZBI Coulter Counter in Dr. Rex Moyer's laboratory at Trinity University (SEE TABLE 2 FOR PARAMETERS). A small sample was obtained asceptically from the culture, 100 µl of which was added to 10 ml of isoton and mixed by gentle inversion before being counted. Three counts were obtained and averaged. This average was then used to calculate the approximate number of cells per milliliter of the sample. The peak channels and the number of cells each represented were noted for each sample. A graph of the cell size distribution was obtained at key intervals, usually once a day as in the case of experiment 1. Cell counts were plotted against time for experiments 3, 4, and 5.

B. Non-Agitated Cultures

Chlamydomonas reinhardtii , strain 125N, was grown in 10 ml of liquid HS media in a 125 ml flask for a maximum air to surface ratio, and on HS agar plates with a 5 ml HS liquid overlay. Three cultures of each were grown in constant light as a control, three were subjected to 18 hour light - 6 hour dark cycling, and three were subjected to 12 hour light- 12 hour dark cycling. The light cycled sets were innoculated two hours before the beginning of the first light period . Each culture was tested for contamination at the beginning of the second complete dark cycle. During the third complete dark cycle, samples were taken once every one and a half hours with the first sample being taken at the beggining of the dark period and the last at the beginning of the next light period. The samples were mixed before sampling for a uniform cell concentration. Each sample was then analyzed on the model ZBI Coulter Counter. Integrated counts of each sample were noted for each ten channel interval. The constant light cultures were inoculated and sampled over the same time frame as the 12 hour light - 12 hour dark cycled cultures. A cell size distribution graph was made for the first and last samples of each culture.

Cell counts were plotted against time, percentages of total cell count were calculated for each ten channel integration, and the maximum percentage of cells which may have divided into two were calculated based on the beginning and ending cell counts for each respective algae culture.

IV. **RESULTS**

A. Aerated Liquid Cultures

Experiment 1 showed that as cell size increased there was a broader cell size distribution for cultures grown in constant light. It showed good growth and division with the highest division rate being found on DAY 2 with an overall cell increase of ten-fold (SEE TABLE3). No other cultures increased at this high of a rate.

Experiment two became contaminated and the results are considered to be invalid for the purposes of this project.

Experiments 3, 4, and 5 all had broad cell size distributions with a variety of peaks. They also did not double during the dark period as expected of synchronized cells . However, there was an increase in cell count from one day to the next. When the cell counts were plotted against time for each culture, a series of alternating peaks and dips were found. This showed inconsistent cell counts which varied widely from one sample period to another.

B. Non-Agitated Cultures

Only two cultures had more than fifty percent of their cells dividing into two during the third complete dark cycle (SEE TABLE 4). The 18 hour light - 6 hour dark cycled flasks showed a maximum of five percent dividing cells. The plate cultures yielded a higher percentage of dividing cells than the flask cultures with those plates that were light cycled having a greater percentage of dividing cells than those grown under constant light.

The constant light flasks yielded a fairly uniform cell size distribution. However, the cell size distribution graphs of the constant light plate cultures showed a sharp incline with three strong peaks and milder peaks along the broad slope. This showed a wide distribution of cell sizes and cell size concentrations in a variety of peaks. The 18 hour light - 6 hour dark cycled flasks yielded graphs with several small peaks over a broad range showing a wide distribution of cell sizes. The plates showed a few strong peaks with several smaller ones over a narrower range, but no sharp peaks that would be expected of synchrony were found. Flask #1 was the only 12 hour light - 12 hour dark cycled culture to show strong cell size distribution peaks in the small cell size range.

The 12 hour light - 12 hour dark cycled plates and flasks, and the constant light plates had a greater concentration of cells between channells 10-39 at the end of the third dark period than at the beginning. However, the concentration of cells in each channel window of the contant light flasks and the 18 hour light - 6 hour dark cycled plates showed only slight variations. The 18 hour light - 6 hour dark cycled flasks had an overall decrease in cell size concentration

V. DISCUSSION

There have been several papers published on the synchronization of Chlamydonmonas reinhardtii (Demets et. al., 1980; Lien and Knutsen,1979; Rooney et. al., 1977; and Surzycki,1971), however none of them give an adequate definition of synchronization or describe a method by which to determine if the cells are in synchrony. It has been assumed for the purposes of this project that synchrony refers to the cell cycle and that it may be determined by recognizing the uniform liberation of the daughter cells. This liberation should be denoted by at least a doubling in the cell count over a short period of time and a sharp peak in the lower channels of the cell size distribution graph. Using these to criteria, it has been determined that synchrony was not achieved for any culture grown for this project. This could be due to several possibilities.

There are many factors that affect synchronization : temperature and its fluctuation, the length and brightness of the light period, the media used, and the amount of aeration or agitation of the culture. Many of the synchronization papers have seemingly contradictory results. This is thought to be due to the variety of methods and conditions used. Surzycki (1971) found that variation in the temperature of more than one degree centigrade resulted in poor synchrony of aerated cultures where as Demets et. al. (1985) found that the cultures would not divide synchronously every twenty four hours if the media was agitated. They postulated that agitation may prevent the cells from entering the immobile phase before division.

The affects of the above conditions may have direct bearing on the interpretation of this project. Experiments 2 and 3 were grown inside of a box which would reach 41°C during the light cycle then fall back to 23°C during the dark. The aerated cultures may not have been capable of entering the immobile phase necessary for division. The light inside the box may have been so intense as to inhibit cell cleavage. Coleman (1982) noted that cell cleaxage appeared to be inhibited under high light intensity. However, it is not understood why the non-agitated cultures were not able to achieve synchrony. They were maintained at a constant temperature and were subjected to direct but not not intense light. It may be that they required a greater intensity of light or that the sampling procedures in some way inhibited cell division.

The difficulty of synchronization may rest in the type of media used. High Salt (HS) was used to require the algae to be dependent on photosynthesis. However, this would mean that the cells would not be able to grow during the dark period. Craigie and Cavalier-Smith (1982) noted that this alga could be synchronized through light-dark cycling if grown in media containing acetate. This would allow for cell growth during the dark period which may be necessary for synchronous division.

In cultures subjected to 12 hour light - 12 hour dark cycling, cell division is confined to the dark period (Cavalier-Smith, 1974). Surzycki (1971) noted that cell division occured synchronously between the 3rd and 6th hours of the dark period. Neither of these expected results were found in any of the light cycled cultures of this project. None of the cultures doubled during the dark period and broad cell size distribution ranges were found. There was a cell count increase from the end of one dark period to the beginnig of the next for the aerated liquid cultures.

The results obtained may not reflect accurate cell counts due to cells sticking to the culture container. Demets et. al. (1980) explains the dips and peaks found when cell counts are plotted against time by this sticking action and points out that only these settling cells are involved with division. This series of alternating peaks and dips was found in experiments 3, 4, and 5, as well as in all of the non-agitated cultures.

This project has raised two questions: 1) What defines synchrony, vegatative nuclear division or daughter cell liberation? and 2) Is synchrony attainable for an entire population? Craigie and Cavalier-Smith (1982) mention that the maximum attainable synchrony is limited by 1) the spread in cell size of newly produced zoospores; and 2) the rate of, and inter cellular variation in, growth during the light period. This implies that synchrony is limited in some way. This same article states that the larger cells divide before the smaller cells and the time span between the two is directly related to the cell length.

VI. RECOMMENDATIONS

There are several recommendations that can be made concerning the synchronization of the *Chlamydomonas reinhardtii* cell cycle, several of which are based on previous literature. Periodic temperature reductions have been used to achieve synchrony (Rooney et. al., 1971). Temperature reductions concurrent with light cycling may also be utilized. Lein and Knutsen (1979) suggest a 12:4 light dark cycle at 35°C. Surzycki (1971) has a method for selecting a suitable strain for synchronization.

It is recommended that synchrony be attempted on HSA plates under 12 hour light - 12 hour dark cycling. The 12 hour cycled plates gave the best results in this project and the acetate contained in the media would allow for cell growth during the dark period.

Cragie and Cavalier- Smith (1982) mention that different size fractions divide at different times. It may be possible to isolate a given size fraction and use it to inoculate a culture that will give rise to a synchronous population.

It is also suggested that a device be prepared that could maintain a continuous and synchronous C. reinhardtii culture from which samples could be readily drawn asceptically when needed.

Table 1: Experimental Parameters of the aerated cultures

Experiment	#1	#2	#3*	#4 *	#5
amount of media	500 ml	500 ml	250 ml	250ml	250ml
Initial concentration**	196	72	44	53	49
light intensity	strong	strong	strong	medium	medium
temperature	ambient	23-41 C	23-41 C	ambient	ambient
light cycling ***	no	12 hour	12 hour	no	12 hour

*Experiments 2 and 3 were contained in a wooden box.
**This number, times 1000, represents the number of cells per milliliter.
***12 hour refers to 12 hour light - 12 hour dark cycling.

Table 2: Coulter Counter Parameters

Amp	1
Aperture	100
Current	1/2 (1ma)
Thresholds	
Upper	3
Lower	100
Matching switch	20k
Gain trim	.6-.7
Count control	external
Count range	100
Base channel threshold	1
Window width	100

Table 3: Experiment 1: Cell Counts and Division Factors

DAY	COUNT	DIVISION FACTOR
1	196,400	
2	520,400	2.6
3	5,480,000	10.5
4	9,093,333	1.6
5	8,693,333	.9
6	8,240,000	.9
7	12,453,200	1.5
8	13,946,666	1.1

Table 4: Percent of Cells Dividing into two Daughter Cells

light cycling	18/6	12/12	constant light
Flask 1	4%	26%	7%
2	<1%	21%	20%
3	5%	27%	38%
Plate 4	51%	69%	48%
5	18%	13%	3%
6	16%	19%	20%

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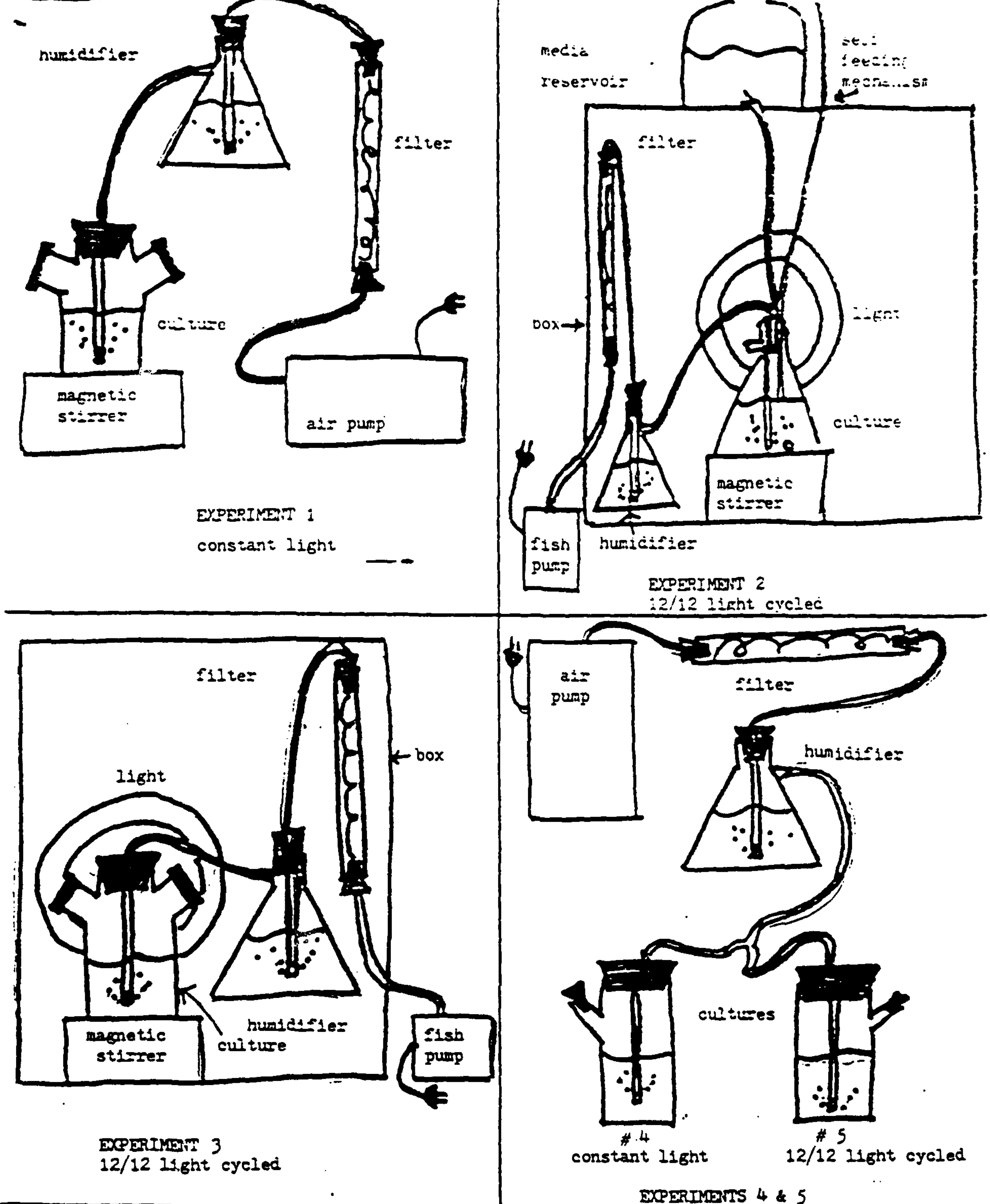
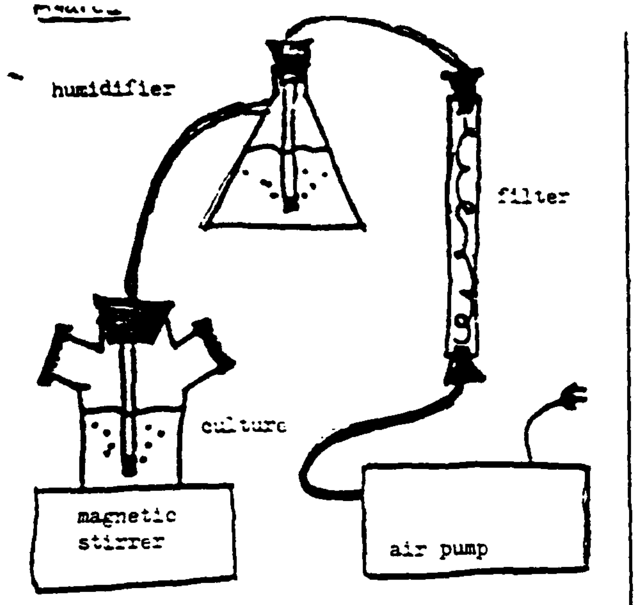
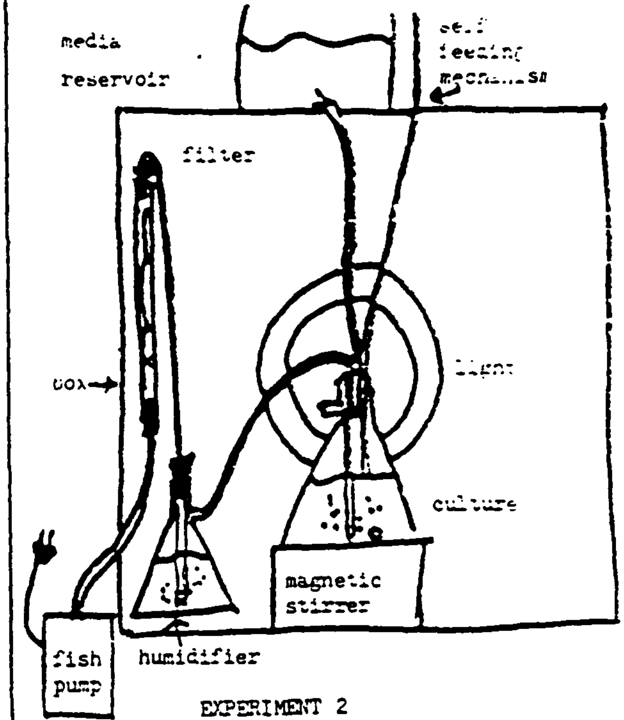
Figure 1



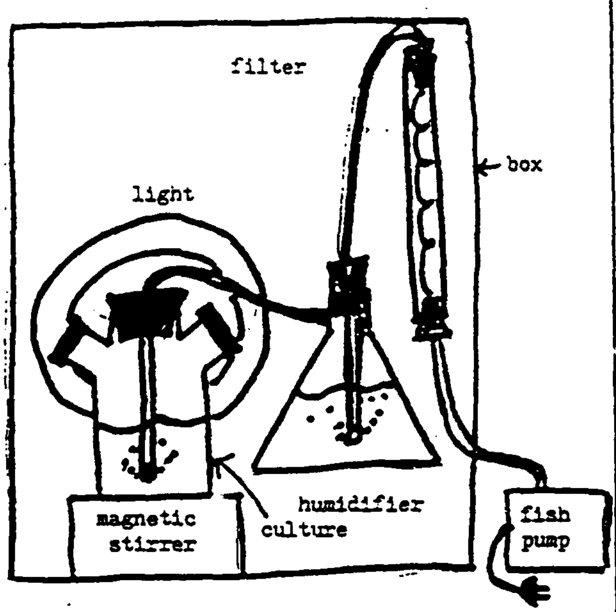
NOTE: Figures are not drawn to scale.



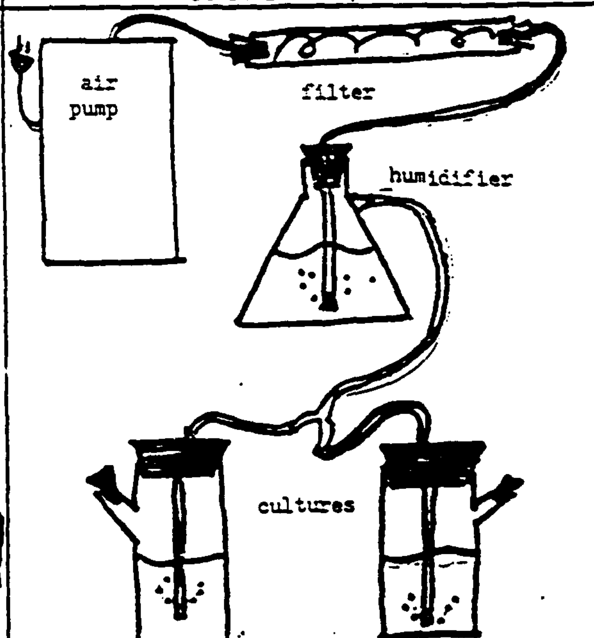
EXPERIMENT 1
constant light



EXPERIMENT 2
12/12 light cycled



EXPERIMENT 3
12/12 light cycled



4 constant light

5 12/12 light cycled

EXPERIMENTS 4 & 5

NOTE: Figures are not drawn to scale.

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FINAL REPORT

Prepared by:	Harold G. Longbotham, Ph.D. Jim Roberts, Graduate Student
Academic Rank:	Assistant Professor Electrical Engineering
Department and	Electrical Engineering
University:	University of Texas at San Antonio
Research Location:	USAFSAM/RZV Brooks AFB San Antonio, TX 78235
USAF Researcher:	Captain Norman Barsalou

Date:	9 Sept 88
Contract No:	F49620-87-R-0004

SAME REPORT AS
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1988 USAF-UES SUMMER FACULTY RESEARCH PROGRAM

GRADUATE STUDENT RESEARCH PROGRAM

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FINAL REPORT

Three Selected Areas of Research in Endocrinology and Metabolism:

I. Biochemical Changes in Saliva of Patients with Metabolic Disorders

II. Design and Validation of a Questionnaire Used to Assess The Nutrition Knowlege of Diabetic Patients

III. Evaluation of a "Kit" Used to Measure Glycosylated Hemoglobin

Prepared by: Antoinne C. Able

Academic Rank: Senior Medical Student

Department and University: School of Medicine, Meharry Medical College

Research Location: Wilford Hall USAF Medical
San Antonio, TX 78236

USAF Researcher: John O. Burgess, D.D.S
James M. Jacobson, M.D.

Date: 10 Sep 1988

Contract No: F49620-88-C-0053

Three Selected Areas of Research in Endocrinology And Metabolism

by

Antoinne C. Able

ABSTRACT

Several projects were undertaken at Wilford Hall Medical Center during my appointment there as a UES Summer Graduate Student Fellow. First, our laboratory addressed chemistry of saliva exist in patients with endocrine and metabolic dysfunction. Seven (7) patient were seen and saliva call will be used in future studies. Secondly, two questionnaires have been designed to be used to assess the nutritional knowledge of diabetic patients. The 40 item questionnaire was designed by Registered Dieticians and will be used to validate the 48 item questionnaire designed by the facilities endocrinologists. Thirdly, the special chemistry laboratory sought to determine the reliability of a glycosylated hemoglobin assay kit.

ACKNOWLEDGMENTS

Much appreciation is extended to the Universal Energy System, Inc. and to Dr. Jorge Sintes as they have made a way that could not be made by others.

Thanks are expressed to Dr John O. Borgess and Jim Jacobson for their time and assistance through out the duration of this project. To Drs. U. Fohlmeister and F. Wians I must express much gratitude for your exceptional support and guidance during my stay there. Your willingness to offer projects when work in other areas slowed down certainly made me feel welcome. No doubt that Col Cissik, Capt Bradley and the entire Clinical Investigation Directorate will be often thought of for their unyielding moral support.

Lastly, Ms. Margarita Lopez-Albano receives my most highest regard for her skilled artistic touch at the keyboard of life.

I. INTRODUCTION

The concerns of research in the Department of Endocrinology and Metabolism at Wilford Hall USAF Medical Center, Lackland Air Force Base extend from improving current methods of establishing diagnoses to developing methods that best enhance patient management. In so doing, many ramifications of a clinical situation may be called upon for contribution. Thus, to conduct clinical investigation in this area, it is important for one to disease processes, but also capable and knowldegeable of techniques and trends involved in the generation of laboratory data. Also, and most importantly, one should be skillful enough in patient interactions to collect the necessary samples and the appropriate historical data for the completion of the clinical record.

My experience in clinical research has revolved around a project that studied the effects of certain chemical derivatives on cholinergic transmission in isolated tissues from various animal models. My task as research assistant included the isolation and observation of standard muscle and nerve-muscle preparations from various species after exposure to different concentrations of the compound under investigation. In this capacity I was also responsible for performing spectrophotometric analysis, radioisotope assays and other biochemical techniques used in measuring cholinesterase activity. While completing the requirements for the degree of Masters of Science in Biology, I gained experience in the utilization electron microscopy, electrophoresis and statistical correlation. Thus, was provided adequate exposure to principles and techniques in laboratory procedure. As a senior medical student at Meharry Medical College, I have acquired ample knowledge of the basic medical sciences and have developed sufficient skills in patient interviewing and history taking to perform well as a Summer Graduate

Research Fellow.

II. OBJECTIVES OF THE RESEARCH EFFORT:

Several concerns were addressed as a result of my appointment to Lackland AFB. One project sought to test the hypothesis that severe and/or chronic biological stressors, such as hypo- or hyperthyroidism, adrenal insufficiency or steroid excess, and diabetes mellitus can create oral conditions that are conducive for dental caries formation. Since no studies exist which relate these conditions to the chemical changes of calcium binding capacity, salivary concentration of unbound calcium, total protein and amylase levels, this protocol has been designed to initiate a body of information that may provide some insight into the cariogenic effect of the conditions stated above. Subjects for this study were patients seen for metabolic disorders (hyper-, hypothyrodism, Diabetes mellitus, adrenal insufficiency, and Cushing's syndrome) and patients receiving steroid therapy for acute and chronic cases of asthma. From each available subject samples of whole saliva were collected, stored in 1-2ml aliquots at $-20^{0}C$, thawed, and assayed to determine levels of secreted corisol, amylase, calcium bioavailbility and the total protein content using the methods described below.

Another project involved the development of an objective test that could reliably measure the extent of a diabetic patient's knowledge and ability to apply his individually proposed American Diabetic Association Diet. While working closely with a certified nutritionist, several methods were reviewed and two instruments have been constructed to be used as tools to evaluate the patient's information base and skills in managing dietary intake. Here again, little infomation is currently available for review in this area although knowledge of dietary restrictions are essential in the efficacious management of individuals with diabetes mellitus.

The third project assigned to me as a Summer Fellow was one which involved the comparison of a commerically available kit used in measuring serum level of glycosylated hemoglobin to other kits with the same or similar purpose. Many corporations have developed and manufactured "kits" that contain all the necessary reagents to perform the desired assay. These kits are reported to deliver accurate and reproducible results at minimal expenditure, thereby making them ideal for massive screening efforts and for laboratories that process samples in large quantities. My responsibility was to determine if this kit provided reliable results in Wilford Hall USAF Medical Center Special Chemistry Clinical Laboratory and then determine how well these results correlated to other kits made available to the facility.

III.

There is a great need to quantify the changes in the levels of the different saliva constiuents that result from various biological stressors (is physical injury) and systemic disease processes (i.e., the fairly common metabolic disorders of hyper-, hypothyroidism, Diabetes mellitus, adrenal insufficiency and Cushing's syndrome). This information may may aid in the prediction and prevention of oral pathology.

Many have reported on the effects of psychosocial stressors (such as death of a spouse, divorce, retirement and a change in living conditions) on the activation of the well defined pathway of the stress reaction through rising circulating levels of cortiso. Morse and associates, (1983) report on the production of opaque appearing, viscous mucous like saliva secretions that are most evident during times of sympathetic nervous system activation. Also, this group tells of the inhibition of the immune response by corticosteroids. These hormones are released during stressful situations and then cariogenic bacteria multiply as a result of low saliva flow rates and immune inhibition. It is also reported that the production of large molecular weight glycoproteins allows for the adherence of placque forming bacteria that eventually initiate caries formation. Bennick (1982) has shown that certain proline rich proteins have roles in prevention of caries formation, but did not indicate how the levels of these proteins are altered in the presence of chronic systemic disease. While it appears to be an increasingly accepted fact in the medical community that certain physiologic changes in the human saliva create an environment for caries formation, few reports demonstrate the possibility of a correlation between caries formation and the stress of acute and chronic alterations in hormonal and metabolic disorders.

The work of Mandel (1980) suggests that the thirst and dry mouth of poorly controlled diabetes are factors which lend to oral disease. On the other hand, Tenovuo et. al. (1986) concluded that no differences in peridontal health or in the oral mucosa are likely in patients being treated for Diabetes mellitus in spite of their finding of increased total protein and levels of immunoglobulins, particularly Ig A (the major immunoglobulin secreted in the oral cavity). The latter group of authors adds that reports in this area are scanty and those published often provide conflicting data which makes the issue controversial. Obviously, a closer look at the relationship between diabetes and changes in human saliva is required.

More literature is available in regards to altered thyroid function and tends to provide slightly more insight to this novel area. Johnson and Kalu (1988) have shown that elevated thyroxine (T_4) serum concentrations in rats with hyperthyroidism enhance the synthesis of salivary proteins. T_4 also increases the number of beta (B) receptors, which are thought to be associated with protein synthesis. Mandel (1980) reported that humans patients treated with radioiodine for thyrotoxicosis exhibited a marked reduction in salivary flow rates and amylase concentrations, and caries formation was abundant. These studies do provide a basis for our hypothesis that saliva contents are altered in the presence of some biological stressors. Attempts at elucidating the biochemical changes in saliva in the presence of hyperthyroidism and hypothyroidism can be useful in the overall management of these patients.

As stated earlier, much has been reported on the changes in the level of salivary cortisol as a result of psychosocial stressors. Our emphasis was to determine the effect of the aforementioned metabolic derangements on salivary chemistry, as there is a paucity of relevant data. Laudat and colleagues (1987) performed studies assessing changes in salivary cortisol levels in patients with untreated Cushing's syndrome and patients with primary and secondary adrenal insufficiency. These authors were able to establish normal and pathological ranges for salivary cortisol measurements in the basal state and during dynamic testing of the pituitary-adrenal axis. And they further suggested that salivary cortisol measurements are an excellent index of free cortisol concentration. They mention the advantages of utilizing salivary samples to monitor the extent of the disease. These + include (1) not having the confounding factor of endogenous binding proteins, and (2) the ease of sampling allows multiple collections without the stress of venipuncture.

Even more closely related to the goals of this academic endeavor is the observation that sodium (Na) and potassium (K) concentrations are altered as a result of adrenocortical dysfunction. Mandel (1980) reported that the mean Na to K ratio in Addisonian patients was 5.0 and that was decreased to 1.8 with corticosteroid therapy. At the other end of the spectrum, it was shown that ratio in patients with Cushing's syndrome was 0.5 and the control value was 1.3. The author concluded that these reported values can be used both as a guide in making accurate diagnosis and as an early reference to prognosis and recovery. Bearing this in mind, the benefit of achieving our goal can therefore be expanded from just the prevention of caries formation and other pathological oral conditions, to a less stressful means of sample collection and the creation of standards by which both diagnostic and prognostic values may be compared.

Participants of this study included patients evaluated in the Endocrinology and Metabolism Service of the Internal Medicine Department of the Wilford Hall USAF Medical Center and from the population of patients in the Allergy Clinic receiving therapy for asthma. Each patient underwent appropriate diagnostic tests to confirm the disease and then was referred to my lab for saliva collection. The disease studied in this protocol included: hyperthyroidism, hypothyroidsim, adrenal insufficiency, steroid excess (as in therapy for treating asthmatics or Cushing's syndrome). Saliva collection from each patient was accomplished between 0800 and 1200 hours daily. Two samples were required for this protocol, one before and one following therapy. Saliva collection was done in a dental exam chair, thereby providing maximum comfort to the volunteer during collection of the specimen.

Prior to collection of pure parotid saliva, each subject were asked to allow approximately 1-2ml of saliva to accumulate in their mouths and then expectorate into vials that were kept on ice. This sample was used to determine baseline levels of cortisol. Pure parotid samples were then collected by use of a sterilized Lashley's saliva collection cup. The collection unit is composed of two concentric circles made of plastic capable of with standing ethylene oxide (gas) sterilization. The inner circle is placed over the opening of Stenson's duct on inner aspect of the cheek and secured in place via a vacuum which was created in the outer circle by a syringe connected through plastic tubing and three way stop-cock value. The collection receptacle was maintained in an insulating unit on ice during the collection process. Collection in the test tube was accomplished plastic tubing connected to the port of the inner circle of the Lashley unit. Saliva flow was stimulated with 2% citric acid until an initial amount of 1ml was collected. This was discarded and stimulation proceeded with citric acid applied with a cotton tipped swab to the lateral borders of the tongue every thirty seconds thereafter for ten minutes. Submandibular and the remaining parotid gland secretions were removed periodically using lights suctioning. Patients receiving propanolol were asked to avoid taking the medication 4-6 hours prior to providing saliva samples. Samples were then stored at -20^{0}C, thawed and centrifuged at 1200 x g to remove inteferring mucins and other particulate matter. The supernatant was then subjected to the various assays listed below. Total protein determinations were made using the Pierce Bradford Coumasie Blue assay (BCA), which utilizes the principles that when proteins react with copper II they produce copper I. The BCA protein assay reagent then reacts with copper I to form a intense purple color absorbed at 562mm.

Bovine serum albumin serves as the standard for quantifying protein. Cortisol concentration were to be determined using Diagnostic Products Corp's Coat-A-Count Cortisol Kits.

b. To date seven patients have contributed samples. These have been stored for future chemical and statistical analysis. Also, several for future chemical and statistical analysis. Also, several delays in shipping and delivery have prevented determinations of cortisol levels, calcium bioavailability and amylase activity.

IV.

a. It is our belief that emphasis on diet education and follow up in diabetic patient is lacking. Lockwood, et. al (1986) showed that patients and physicians agree that diet and diet-related issues constitute a very difficult problem.

Lorenz et. al (1985) recommended that all teaching should be followed by thorough and systematic assessment. In general current literature reflects the need for a validated instrument that is capable of measuring a patient's knowledge of his diabetic diet and its application there of. Clearly, determining whether or not the goals of education programs are being met can be beneficial both to the patient and to the institution rendering this service. Implementing individual assessments can enhance self management in diabetics by eliminating areas of incomplete understanding of the concepts in the diet regimen. By providing more personalized instruction patients' level of expectation improve and hence there will be improvement in their desire to help themselves.

The observed trends of patient responses in general areas of assessment can help the institution by revealing how well and what patients are learning. The areas of both effective and faulty instruction in this educational program can be established; then any necessary augmentation or modification in the curriculum can be made. Eventually the progression of the deletorious effects of diabetes may be slowed significantly.

Improvements in patient education through knowledge assessment also has social implication. Theorectically, increased patient knowledge of diet plans can destroy many barriers to participation in functions outside the home and increase confidence in themselves as managers of their well being. Improvements in self management though a better adherence to diet could reduce diabetes related hospitalization, saving money for the patient and to for cost conscious third party insurance agencies. Conclusively, objective monitoring of patient diet plans has many benefits to all involved.

The type and contents of the questions included in our questionnaire was determined through the expertise of endocrinologists and other physicians involved in diabetic patient management and associated with this project. The questionnaire was designed to be comprehensive yet easily interpreted. It consisted of 48 questions in three areas: Personal Dietary Requirements (18 quest), Concept of an exchange (15 questions), and estimating recipe content (15 questions). The questionnaire was to be administered by the diabetic teaching nurse both before and after instruction from the nutrition clinic. Validation of our test required that it be compared to a "gold standard". The nutrition clinic therefore developed a 40 item questionnaire that met the same criteria as that designed by the research group. The dieticians were responsible for the type and content of each question. They would also be responsible for the administration of their test prior to and following instruction in a manner which best suited them. Reliability for each test was provided for by pre-and post instruction administration. Statistical correlation of our assessment questionnaire with that of nutrition determine its validation.

The population of participants in the study are patients referred to the Wilford Hall Medical Center Nutrition clinic after a diagnosis of either diabetes mellitius or impared glucose tolerance had been made. The criteria for diabetes have been established by two methods and are as follows:

1. Fasting plasma glucose (glc) 140mg/d on at least two occassions.
2. Oral glucose tolerance test criteria (1.75mg glc/kg body weight to maximum of 75 gm glc).

a. Impaired glc. tolerance: plasma glc between 140 mgldl and 200 mgldl two hours following ingestions and >200 mgldl between time zero and two hours.

b. Diabetes mellitus: plasma glc > 200 mg/dl at two hours and one other point in the test.

Participants will be assigned a number using a table of random numbers as they begin therapy and instruction from the Diabetic Teaching Nurse. Each participants will be >18 years old and capable of completing the questionnaire. Other details will be provided, following completion of the project. 100-150 patients are expected to participate. Due to the lack of remaining time no data was obtained. However, the communication between the endocrinology service and the nutrition clinci has increased. It is assumed that the project will proceed as scheduling allows.

V.

a. Several kits have been made available to measure serum levels of glycosylated hemoglobin (GHB). Pacific Hemostasis division of Curtin Mathesion Scientific, Inc. (Ventura, CA) has created "Glycoscreen" a glycosylated hemoglobin (GHB) assay which will be evaluated for precision and compared to other kits. Measurements of GHB were introduced into clinical practice as an index of long term diabetic control (Davidson, 1986). GHB is formed non enzymatically through a two step process which ends in the addition of either sugar or sugar phosphates to reactive valine or lysine sites on the beta chain of hemoglobin molecule. Thus, the amount of GHB is a measure of the prevailing glucose concentration to which the red blood cells have been exposed. The pieces of GHB are determined by the sugar moiety attached to the hemoglobin molecule. HbA_{1c} results from the addition of glucose; HbA_{1a} results from the addition of a sugar phosphate.

Isolation of these species is facilitated by the resulting increase in Therefore, charge dependent separation techniques are employed in quantifying levels of GHB species. These include electrophoresis and ceation exchange chromatography. The latter has been the basis of many commerically available kits. The makers of glycoscreen report that this usual method is sensitive to temperature fluctuation, interference by abnormal hemoglobin (fetal hemoglobin and hemoglobin S seen in sickle cell anemia) and the presence of intermediate labile forms of GHB. They also add that based on their boronate affinity method these technical difficulties are eliminated.

The separation of GHB in the "Glycoscreen" assay is due to the specific binding characteristics of aminophenylboronic acid to the sugar or sugar phosphate adducts at lysine and valine residues of GHB. The affinity method is also free of interference by temperature, labile GHB and abnormal hemoglobins. Another advantage of Glycoscreen is that the assay utilizes a single tube for each patient, control and calibrator sample; therefore avoiding problems inherent in column chromatography. Mesurements of GHB were made following the manufactureers instructions. The process involved two absorbance reading on a dilute hemolysate and a second absorbance reading after a short incubation with an atiquat of affinity gel. The percentage of GHB is calculated using the following formula:

$$\frac{\text{Absorbance A-Absorbance B X 1.2) x 100 = \%GHB}}{\text{Absorbance A}}$$

Coefficients of variation (c.v) will serve as statistical model. Results will be compared be to results of the manufacturer and other reports.

b. Twenty assays were performed in duplicate using manufacturer's serum samples of normal and elevated levels of GHB. While they report a corrected GHB value of 7.3% (c.v = 4.5%) in the normal sample, we achieved only 4.04% (S= 1.64) with c.v.= 40.5%. Values obtained in the elevated sample were 11.51 (S=2.28) and c.v. of 19.8% when the manufacturer reported mean value of 14.1% and c.v. 3.1%. The discrepancies resulted incessation of the experiment until a company representative could be contacted.

VI. Recommendations

Due to the design of the protocol for determining the effects of biological stressors on saliva patients must provide another sample following therapy. The collection of this sample will be performed by colleagues at the University of Texas Health Sciences Center. Cooordination of the effort will be by personnel of Wilford Hall.

Remaining assays will be performed at this time.

The project involving the assesment of nutritional knowledge in diabetic patients has been preliminarily completed with the design of the questionnaire. Once duplication of the tests, manpower assignments, and scheduling of events have been completed, data can be collected and compiled for statistical analysis. Twenty five individuals should be assessed first. Depending on those results, more should be added to prove significance. It is my recommendation this project continue, as the results can aid tremendously in patient management once validation has been accomplished. Also since many believe that knowledge does not always result in adherence, I recommend that the issue of motivational and environmental factors be addressed by whatever means necessary in order to improve the self managment of diabetes. Also, consideration should be given toward making the questionnaire unbiased in terms of cultural factors that may also influence results. The evaluation of the "Glycoscreen" glycosylated hemoglobin assay has been halted and the company representative has been contacted. It is recommened that this suggestion be followed and that this project be continued as this information can be of significiant value to the manfacturer and especially to those institutions which to choose use this kit in their method of diagnosis and managment. Unless these discrepancies are resolved, future use of this kit may yield, erroneous results, thus resulting in the possibility of malpractice.

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FINAL REPORT

Utility of Sensitive Immunoradiometric Assay (IRMA) to Predict Results of Thyroliberin Stimulation Test.

Prepared by: Adams, Stanley D.

Academic Rank: Medical Student

Department and University: School of Medicine, Meharry Medical College

Research Location: Wilford Hall USAF Medical Center
SGHMME/SGHMMC
Lackland AFB,
San Antonio, TX 78236-5300

USAF Researchers: Jacobson, James M, M.D.
Grassman, Eric D, M.D.

Date: 12 August, 1988
Contract No: F 49620-88-C-0053

Utility of Sensitive Immunoradiometric Assay (IRMA) to Predict Results of Thyroliberin Stimulation Test.

Stanley D. Adams

ABSTRACT

The clinical utility of two "sensitive" immunoradiometric assay (IRMA) kits (Tandem-R TSH HS, Hybritech,Inc; Echoclonal TSH, Bio-Rad) was evaluated in relation to analytic and clinical performance for measurement of thyrotropin (TSH). Their performance was compared to that of a "regular" IRMA (ARIA-HT, Becton-Dickinson) in order to learn whether these sensitive assays might be clinically useful to predict results of the thyroliberin (TRF) stimulation test, thereby obviating the need to perform this expensive and time-consuming test.

Suitable patients were identified who had had a TRF test previously and 107 refrigerated serum samples were located, along with pertinent records associated with these samples. The samples were provided to the laboratory for high-sensitivity assay, and a preliminary data file was created. Descriptive statistics were done on these data, and groundwork was done to expedite the accomplishment of inferential statistics.

Additional research assistance was provided to two other projects: a research paper was revised from an intermediate draft to final drafts and submitted to a scientific journal and an on-going cardiological research project was given off-line computerized left-ventricular pressure/radioactive count data analysis.

ACKNOWLEDGEMENTS

I would like to thank the Air Force Systems Command and the Air Force Office of Scientific Research for making this research opportunity possible.

Dr James M. Jacobson was always willing to make time within his busy schedule to discuss ways to overcome encountered roadblocks. The Wilford Hall Endocrinology Service was a continual source of information, with Dr Bob Constant, Dr Paul Glowienka, Dr Steve Brietzke, and Gloria Ortiz, R.N. being especially supportive. Cliff Butzin, Ph.D. was generous with his time and a patient teacher of statistical methods.

Dr Eric D. Grassman and Dr Leo Spaccavento explained many technical cardiological points and were generally helpful. Finally, I would like to acknowledge the contributions of my wife, Junghee Park-Adams, not only for continual encouragement, but for hands-on assistance with my projects when the going got tough.

I. INTRODUCTION

The standard methods for assay of thyrotropin (thyroid stimulating hormone, TSH) were developed several years ago. More recently, with the advent of such developments as monoclonal antibodies, more sensitive tests have been developed. Collectively, these assays are known as "sensitive" or "highly-sensitive" assays, to distinguish them from the preexisting "regular" assays.

Improved analytical sensitivity is the hallmark of these sensitive assays, which augments the ability to distinguish primary hyperthyroidism from euthyroidism on the basis of TSH measurement alone(1,2). Analytic (3) and clinical(2) performance criteria have been recently introduced which allow categorizing TSH assays as "sensitive" or "regular".

The thyroliberin (thyrotropin-releasing factor,TRF) stimulation test has been the standard follow-up diagnostic tool to distinguish subclinical hyperthyroidism or hypothyroidism from euthyroidism for almost two decades (4,5). This test is time consuming, expensive, and can have adverse consequences to the patients (6). When the basal (unstimulated) concentration of TSH is below the "virtually ensured" assay detection limit, a TRF stimulation test is not required. Therefore, measurement of TSH with a sensitive assay can avoid the drawbacks of the TRF stimulation test by allowing a single-test strategy.

In healthy euthyroid individuals TSH concentrations are generally between 0.3 to 4.0 milli-international units per liter (mIU/L), whereas in untreated, overtly thyrotoxic patients with Graves' disease the serum TSH ranges from <0.005 to 0.190 (mean, 0.04) mIU/L (7). The recent emergence of "sensitive" assays has decreased the lower level of detectability (LLD) from approximately 0.4 mIU/L to values as low as 0.04 mIU/L (1,8,9).

It has been previously shown that certain high-sensitivity assay kits may obviate the need to perform TRF stimulation tests in selected patients (10,11) and may indicate thyroid status more reliably than simple free T3 or free T4 measurements (12). The Endocrinology Service at Wilford Hall USAF Medical Center at Lackland Air Force Base is particularly interested whether or not in a general unselected patient population a single "blood test" strategy can essentially give the same information as can the TRF (thyroliberin) stimulation test; a procedure which requires several hours of a patient's time, the services of a trained endocrinological nurse to perform, and the administration of an expensive and possibly dangerous diagnostic material. Therefore, an affirmative answer would mean both increased cost-effectiveness and greater patient safety.

Several factors prepared and guided me towards clinical research with the Endocrinology and Cardiovascular Services at Wilford Hall. Foremost was my basic interest in the areas of both endocrinological and cardiovascular physiology. Also, my undergraduate muscle biostructure laboratory experience, along with both undergraduate and medical school coursework in anatomy, physiology, and biochemistry was appropriate background for this research endeavor.

II. OBJECTIVES OF THE RESEARCH EFFORT

From several research projects that my USAF research colleague had either underway or "in the works", I initially chose two projects. Also, it was left as an open matter that if extra time became available, I would assist the Chief Cardiologist and his colleagues on an on-going research project.

a.) One of the early projects (Effect of Gemfibrozil on Reaction Time) was discontinued for the duration of my research time, so it was decided that I should undertake the final revisions of a paper (Hyperthyroidism in Mitral Valve Prolapse Syndrome, submitted to American Journal of the Medical Sciences) that existed in an intermediate draft. Free rein was given towards the accomplishment of this end.

In this manner, it was planned that by accomplishing this task, two goals would be met: I would be able to "learn the ropes", not only with regards to familiarizing myself with the in-house word processing system, but also the innumerable details required before a paper is allowed to be submitted to a major journal.

b.) The result of these efforts was that a body of research that had been previously accomplished, but had never been published, became realized as a tangible journal submission.

III. a.) The cardiologist who shared partial authorship of the paper which I was editing figured importantly during my research period for two reasons. First, it was imperative to have the paper reviewed by the cardiologist to assure technical accuracy and second, the cardiologists were interested in assistance with a project of their own.

b.) The cardiological comments with regards to technical accuracy were obtained and second, an on-going cardiological research project was given research assistance in the form of providing off-line computerized left-ventricular pressure/radioactive count data analysis (Left Ventricular Pressure-Volume Relationship in Patients with Coronary Artery Disease).

IV. a.) The USAF researcher and his colleagues had previously published research on the clinical utility of a recently developed "super-sensitive" assay of thyrotropin (TSH) as a possible replacement for the more expensive, time-consuming, and hazardous thyroliberin (TRF) stimulation test. The previous paper had used a highly selected population of patients in arriving at it's conclusions.

A logical follow-up report would concern similar research done on a general, unselected population; this population would be much like the population that might walk through an endocrinologist's door.

b.) With this in mind, the following has been accomplished. We retrospectively selected 107 serum samples from 102 patients who had undergone TRF stimulation tests. The 102 patients included 70 women and 32 men, aged 16 to 84 (mean=48.6± 16.6, median=48.25), who in addition to recieving the TRF stimulation test, also recieved standard thyroid function tests (T3RU, T3RIA, T4).

The TRF stimulation test had been performed by collection of a basal (unstimulated) blood sample, followed by the intravenous administration of 500 micrograms of TRF (Thyponine; Abbott Laboratories, North Chicago, IL) as a bolus over 15-30 seconds. Additional blood samples were obtained at 15 minute intervals for one hour after the administration of TRF.

Serum samples obtained for the initial TSH assays were accumulated over a twenty-six month period. Five patients contributed more than one serum sample. Serum from all patients was collected prior to performance of the TRF stimulation test. These initial serum samples were divided into two aliquots, one of which was put into refrigerated storage and the other which was assayed in the ARIA-HT instrument (Becton-Dickinson Immunodiagnostics, Salt Lake City, UT) for basal TSH and for total thyroxin (T4), and for triiodothyronine resin uptake (T3RU) by the MAAT3 method (Nuclear Diagnostic, Inc., Troy, MI).

These measurements were followed by calculation of the free thyroxin index (FTI)=[(T4)(T3RU)]/30. Serum samples collected after performing the TRF stimulation test were also divided into two aliquots, one of which was stored and one which was assayed for TSH rise in the ARIA-HT instrument.

All assays were performed by trained laboratory technicians and were carried out according to the manufacturer's instructions. In radioimmunoassays, bound ligand was measured by counting gamma radiation for one minute and analyzing results with an automatic data reduction procedure (Micromedic 4/600 with MACC; Micromedic, Inc., Horsham, PA) using a linear-logarithmic transformation. Stored samples were kept between -40 and -70 from 4 months to 26 months prior to high-sensitivity assay.

"Tandem-R TSH HS" (Hybritech Inc., San Diego, CA) and "EchoClonal hTSH" (Bio-Rad, Hercules, CA), both "sensitive" IRMA kits, were kindly provided as gifts by the manufacturers. We used one lot number for this second part of the study, performed in August, 1988.

For statistical analyses, we used SPSSx software (SPSS Inc., Chicago, IL 60611), to° calculate descriptive statistics. For the statistical analysis the procedures used will be correlation analysis between basal regular TSH and basal sensitive TSH, true positives, true negatives, false positives, false negatives, sensitivity, specificity, and to compare sensitivity and specificity by graphing the two as abscissa and ordinate in order to display a classic receiver-operator curve (ROC). The remainder of the statistical analysis will be performed by the primary investigator.

IV.a.) The relationship between left ventricular (LV) pressure and volume provides a highly accurate means of characterizing ventricular diastolic and systolic function with the "end-systolic" pressure volume relationship being a useful volume-independent measure of ventricular contractility(13).

Limiting the use of pressure-volume curves to the animal model only has formerly been the result of the inability to measure instantaneous left ventricular volumes. In animals, this measurement is possible using highly invasive methodology, involving the placement of myocardial markers (sonographic crystals); this technique is precluded in humans.

By injection of an appropriate radioisotope and use of the nuclear "stethoscope" (Bios, Inc.; Valhalla,NY), continuous measurement of beat to beat LV function can be made, which can provide a relatively accurate estimate of ventricular volumes. Since patients with coronary artery disease routinely have continuous LV pressures recorded, applying the nuclear stethoscope concurrently will allow simultaneous measurement of LV pressures and volumes. This data can be analyzed "off-line" after the coronary catheterization procedure, on specialized computer software to provide pressure-volume (p-v) curves, and from the point of maximum pressure to volume ratio on these curves, a line can be drawn (termed "Emax"); this is a measure of myocardial contractility.

b.) With regards to the above, the following has been accomplished. Pressure volume curves were computer generated (IBM AT; International Business Machines, Armonk, N.Y.) from data obtained at coronary catheterizations of five patients. A seperate p-v curve was generated for each of the following data records: 1.) baseline one; 2.)atrial pacing to coronary ischemia; 3.)baseline two; 4.)nitroglycerin infusion with pacing; 5.)baseline three. These data records allowed analysis of the resting state, the pacing induced ischemic state, and after nitroglycerin followed by pacing induced ischemia.

These curves were later plotted (HP Plotter; Hewett-Packard,Covallis, OR) by the primary investigator to be used as graphics to accompany the research article in progress.

V. RECOMMENDATIONS

a.) The use of high-sensitivity throtropin assays to predict results of the TRF stimulation test might provide a marked advance in the study of endocrine disease, provided that in the final analysis this research indicates an affirmative answer to this issue. If this is the case, then this result should be communicated throughout the USAF endocrinological community worldwide.

The TRF stimulation test will very probably remain the most sensitive method of confirming certain conditions, such as whether a patient with low serum thyroid hormone values is truly hypothyroid; also, this test is useful in distinguishing disorders of growth hormone or prolactin secretion. However, if the use of this test can be reduced, it would mean an advantage to the Air Force in greater cost-effectiveness and patient safety.

b.) The ability to measure pressure-volume loops would represent a milestone in the study of ventricular function. Since this is a highly experimental technique, more research must be done to correlate this with other measures of ventricular function, such as animal sonographic models. However, one can envision this promising work developing into methods of "real-time" on-line assessment of ventricular function.

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1988 USAF-UES SUMMER RESEARCH PROGRAM/

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FINAL_REPORT

DENTAL_MATERIALS

Prepared by:	Bobby L. Larry, B.S.
Academic rank:	Sophomore Dental Student
Department and	School of Dentistry
University:	Meharry Medical College
Research	Dunn Dental Clinic/WHMC
location:	Lackland AFB
	San Antonio, Texas 78236
USAF Researcher:	Col. John O. Burgess, D.C.
Date:	05 August 1988
Contract:	F49620-88-C-0053

DENTAL MATERIALS

by

Bobby L. Larry

ABSTRACT

The shear bond strength of commercial liners and bases to prepared dentin was investigated. Two liners which contain calcium hydroxide and are polymerized by visible light, (Prisma VLC Dycal) and Cavalite (Kerr), were compared with a capsulated "Bonded-base" cement (ESPE Kefac-Bond applicap) and two glass ionomer lining cements: Shofu lining cement and GC lining cement. These materials were also used to determine the amount of fluoride released when used as a liner or base.

The strength of different types of pin-retained amalgams was investigated. Stainless steel pins: Stabilok small diameter, MPS, TMS minum, DENLOK, TMS Link Series (Whaledent) and one SS goldplated TMS Minum (Wholedent) were compared with 3 titanium pins; Filpin, Stabilok 6mm and TMS Link Plus.

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I wish to thank the United States Air Force and the Benjamin Dunn Dental Clinic Research Division for sponsoring this research. Universal Energy Systems, Inc. must be mentioned for their concern and help to me in all administrative and directional aspects of this program.

My experience was rewarding and enriching because of the many helpful, cordial and dedicated people. Dr. John O. Burgess, Dr. Jorge L. Sintes and Dr. Jim Summitt, provided me with support, encouragement, understanding and a truly enjoyable working atmosphere. The help of Drs. Burgess, Sintes and Summitt was invaluable in overcoming many technical barriers. Their concerns and untiring efforts were greatly appreciated.

INTRODUCTION:

Prisma VLC Dycal (Caulk), Cavalite (Kerr), (ESPE) Ketac-bond applicap, Shofu lining cement and GC lining cement are used as filling material and cementing agents. These products have anticariogenic properties through the slow release of fluoride from the restored portions of the tooth to surrounding tooth structure. Teeth normally consist of hydroxyapatite crystals within its chemical composition. After a carious lesion is removed and prior to amalgam condensing, one of these products is utilized. The fluoride released from these materials replaces the hydroxide and forms fluorapatite crystals, thus creating a much stronger tooth.

Dental amalgam is often used to replace lost tooth structure. When large amounts of tooth structure is missing amalgam pins are inserted into the dentin to retain the amalgam restoration. Stainless steel and titanium pins are commonly used in this procedure. Perhaps, with the addition of these pins, the amalgam lasts longer and better resists tensile and compressive forces.

I am a sophomore dental student at Meharry Medical College, School of Dentistry. Currently, I rank in the upper 10% of the class of 1991. During the first year of my matriculation at Meharry Medical College, I learned about various materials, products and instruments, used in the field of dentistry. I also received valuable clinical and laboratory experience. The General Dentistry and Dental Research Department at the Benjamin Dunn Dental Clinic are quite diversified and provide additional practical training in dental health care delivery.

After receiving a Bachelor of Science Degree from Tougaloo College, I worked as a Laboratory Technician and a Pollution Control Technician. Here I learned how to operate meters, calibrators, acids, solutions, monitors and other laboratory equipment. The knowledge I gained during my past employment and my first year of dental school help me to interact smoothly with the on-going research in the above mentioned department.

II. OBJECTIVES:

a. To examine the resistance form of amalgam restorations retained with 10 different types of pins.

b. To determine the shear bond strength of liners and bases.

c. To determine the fluoride release of liners and bases.

III.

a. Extracted mandibular molars, both caries and restoration free, were selected then measured at the cemento-enamel junction. The measurements ranged from 9-12 mm. The specimens were stored in tap water except during preparation and testing. The occlusal surface of each tooth was reduced with a Torit Model Trimmer to provide a flat surface approximately 1 mm above the cemento-enamel junction. One hundred and ten mandibular teeth were prepared in this investigation. The roots of each tooth were notched for better retention and embedded in an aluminum mounting ring filled with orthodontic self-curing acrylic. Six pin holes were drilled approximately 0.5 mm inside the dentino-enamel junction of each tooth. Six 0.6 mm self-threading pins were inserted in each tooth following manufacturers instructions. After insertion, the pins were measured and cut 2 mm above the flattened tooth surface. Copalite (Plastodent cavity lining) was evenly applied on the flattened tooth surface as a sealant. A Tofflemeyer matrix band and Moyco Copper bands were secured in place and dental amalgam was condensed into these retainers using hand and pneumatic instruments. The amalgam height was measured distally, mesially, buccally and lingually. The amalgam crown was built to a height of 4 mm.

Presently, the first 40 specimens are being tested with an Instron Testing Machine (Instron Corp.). A test load and a constant crosshead speed of 0.05" (1.27 mm) per minute is applied until the amalgam fractures. The results will be recorded on an analysis chart.

b. My assignment as a participant in the 1988 Summer Graduate Research Program (SGRP) was to determine the bonding strength of five commercially available liners and bases. Prisma VLC Dycal (Caulk), Cavalite (Kerr), Ketac bond "bonded base" applicap (ESPE), Shofu lining cement and GC lining cement (Liv).

Seventy five extracted maxillary molars were reduced on their buccal surface to expose the dentin. A Torit model trimmer was used to develop a smooth flat surface. A small piece of teflon tape, with a 3mm hole punched in it, was attached to the flatten surface. A very small plastic tube (7 mm height, 3mm diameter) was placed over the hole, while the lining/base material was slowly inserted inside the tube's cavity. The teeth were separated in five sets of fifteen teeth per set, corresponding with the five different materials to be tested.

The specimens were then embedded in aluminum mounting rings filled with orthodontic self-curing acrylic resin. Mounting these specimens required patience and delicate handling, because the material very easily detached from the tooth. The specimens were stored in tap water except during testing. Initially, the retention of the liner/base material to the prepared dentin held quite firmly. However. water storage reduced the adhesiveness. In fact, several specimens dislodged from the teeth prior to testing. Approximately 5 teeth per group were tested by the Instron Testing machine (Instron Corp.). The results obtained were invalid due to the low number of specimens and the inconsistent range of recorded values. It was decided that the specimens be replaced. Specimens were re-prepared and an immediate shear bond strength performed.

c. The liner/base materials mentioned above are also used to determine the amount of fluoride being released. These materials were applied into the cavity of a small plastic tube (7mm height, 3 mm diameter) and allowed to harden. The hardened product was removed and placed in a plastic test tube. Here again, fifteen specimens per material were formulated. One millimeter of Type I distilled water was added to each test tube. The liner/base material will be stored in the distilled water for seven days. On the seventh day, the millivolts will be recorded for each material. After which 100 and 200 microliters of Tisab buffer solution will be added and read accordingly. Sgt. David Goddu, USAF Medical Technologist, constructed a curve to convert milliwatts to p.p.m. Therefore, the fluoride released will be measured in p.p.m.

IV. RECOMMENDATIONS:

A. No data was obtained from the pin retained amalgam study because the first fifteen specimens designed were not carefully controlled. Time did not permit us to finish this project before the ten week research period expired. Therefore, I recommend receiving results of this study and to participate in the development of the publication.

B. As indicated in III section b, data obtained from the shear bond strength of liners/bases was equivocal because of the high water solubility rate. Therefore, I recommend continuing this study at Meharry Medical College, School of Dentistry through a student's research award. Then to develop a publication for a dental journal and a presentation at I.A.D.R.

C. This project is for a one year period. After results are obtained, I would greatly appreciate helping in the development of a publication.

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FINAL REPORT

Prepared by: Aleshia C. Lewis

Academic Rank: Medical Student - Sophomore

Department: Microbiology

University: Meharry Medical College

Research Location: Wilford Hall USAF Medical Center
Lackland AFB
San Antonio, TX 78236-5300

USAF Researcher: Ronald E. Grimwood, Lt. Col, USAF,MC

Date: 5 September 88

Contact No: F49620-88-C-0053

In Vitro Culture of Human Keratinocytes with Subsequent Induction of Stratification

by

Aleshia C. Lewis

ABSTRACT

Human keratinocytes were cultured from cells obtained from newborn foreskin. The method used involves a two-phase technique that optimizes proliferation (Phase I) and differentiation (Phase II). Phase I consist of developing monolayers of highly proliferative basal cell-like keratinocytes in serum-free keratinocyte growth medium (KGM). Phase II involves the induction of differentiation and stratification of these confluent monolayers. The resulting epidermal sheets are removed from the medium and stained for specific keratinocyte cell markers (keratin, bullous pemphigoid, pemphigus vulgaris, and laminin) using immunofluorescent techniques.

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I would like to acknowledge the Air Force Systems Command and the Air Force Office of Scientific Research for sponsorship of this program. Special thanks are extended to Dr. Ronald E. Grimwood for his guidance and assistance in every aspect of completing my research assignment. Colonel John H. Cissik, Micheal Chastain, Major Patterson, Captain Bradley, Dr. Van Dellen, and all others from the Clinical Investigations Facility are appreciated for helping to creating an enjoyable working atmosphere.

I. INTRODUCTION

The decreased ability of severely traumatized tissue to respond to autographs or to undergo natural healing has lead to the search for alternative approaches of medical treatment for patients with chronic skin ulcers, mechanobullous disease, and extensive burns involving large percentages of the total body surface area. The grafting of in vivo cultured human keratinocytes has proved to be a successful method of treating severely traumatized tissue.

My interest in the application of cell culture research to clinical medicine led me to seek a position as a summer fellow in the area of laboratory sciences. My experience in developing primary cell cultures from human tissue was very limited. My previous work with fibroblast cultures and my work with immunochemical staining contributed to my assignment to the Clinical Investigation Facility (CIF) at Wilford Hall USAF Hospital.

The Principal Investigator in Dermatology at Wilford Hall is particularly concerned with the culture of human keratinocytes for subsequent use in the treatment of burn patients. However, before cultured human keratinocytes can be used for clinical treatment, it is necessary to develop and perfect those techniques necessary to generate monolayers of cells with the potential to form stratified sheets of epithelia. This paper describes the attempts made at developing and perfecting these techniques and the practical applications of in vivo cultured human keratinocytes.

II. OBJECTIVES OF THE RESEARCH EFFORT

As a summer fellow in the Graduate Student Research Program (GSRP), my assignment was to accomplish two main objectives. First, I was required to develop techniques for successful primary isolation of human keratinocytes from newborn foreskin. The cells derived from the foreskin tissue had to be planted on plastic tissue culture plates and allowed to grow to 80% confluence. The second major objective was to induce differentiation and stratification of the keratinocyte monolayers. It was therefore determined that a growth medium containing serum and an increased calcium concentration would accomplish this. Immunofluorescent staining was selected as the best method to determine whether or not the stratified sheets of cells were human keratinocytes.

III.

a. The defined medium for phase one was Keratinocyte Growth medium (KGM, Clonetics, San Diego, CA). This serum-free formulation contains hydrocortisone, bovine pituitary extract, insulin, epidermal growth factor, and antimicrobial agents. Phase two growth medium was Dulbecco's modified Eagle medium (Gibco) containing 10% fetal bovine serum and 1.8 mM calcium. As previously stated, calcium and serum facilitate stratification and differentiation.

The foreskin tissue obtained from the newborn nursery was placed in a holding medium containing Hank's balanced salt solution, Hepes buffer, and antimicrobial agents (Solution A). After being transported to the laboratory, the foreskins were rinsed in absolute ethanol for 5 seconds, followed by two 5 second rinses in sterile Solution A. Using sterile forceps and scissors, the foreskins were cut into small ($2mm^2$) pieces and placed into a 60 ml petri dish containing 7 mls of 25% trypsin (Sigma-type III or T-2271)-Solution A. The dish was covered and incubated for 2-3 hours at 37°C and 5% CO_2. Following the two hour incubation period, the epidermis was pulled off each $2mm^2$ piece of tissue using sterile forceps and placed into a petri dish containing 5 mls of Solution A. The solution was was pipeted up and down to break the epidermal sheets into single cells. The cell suspension was then pipet aspirated into a centrifuge tube containing 5 mls of KGM with 10% fetal calf serum. After being centrifuged for 5 minutes at 1,000 rpm the supernatant was poured off and 2 mls of KGM was added to the pellet. The pellet was then broken up by vigorous agitation and the resuspended cells were added to a $25cm^2$ tissue culture flask containing 7 mls of KGM. After 48-72 hours, the initial KGM was replaced by fresh media. After reaching 80% confluence, the cells were serially passed to a $75cm^2$ culture flask.

The passage of human keratinocytes involves the use of trypsin/EDTA (Gibco,IX) obtained from the clonetics corporation. The procedure involves replacing the 9 mls of KGM in the $25cm^2$ culture flask with 2 mls of trypsin/EDTA. The latter reagent is allowed to act for 30 to 60 seconds at room temperature. Following this short incubation period, the trypsin/EDTA is removed and the culture flask is sealed and allowed to remain at room temperature for 3-4 additional minutes. Once the cells have been released from the bottom of the flask, 2 mls of trypsin neutralizing solution (Clonetics) is added and the cell mixture is transfered to a sterile centrifuge tube. An additional 2 ml of trypsin neutralizing solution is added to the culture flask to recover any remaining cells. The cell suspension is then centrifuged at 1,000 rpm for 5 minutes, discarding the supernatant and resuspending the pellet in 2 mls of KGM. The cells are then counted using a hemocytometer. An average of 5.45×10^4 cells/ml are usually inoculated into new culture vessels ($75cm^2$). After reaching 80% confluence, the cultured keratinocytes are placed in Dulbecco's modified Eagle's medium (DMEM) for phase two of culture techniques.

The second phase of the culturing technique involves inducing differentiation of the keratinocytes so that they can form cohesive intact sheets. This was accomplished by replacing KGM with DMEM containing 10% serum and an elevated calcium concentration (1.8mM). The differentiating cultures were fed DMEM every two days, with cohesive stratified sheets forming within 13-16 days. Following the formation of stratified sheets, the tops of the plastic culture flasks were removed. The epidermal sheets were then lifted from the bottom of the flasks via enzymatic treatment with neutral protease (Dispase), 4u/ml, in solution A, for 5 minutes. The sheets were placed in 100 ml petri dishes, transported to the dermatology clinic, and frozen in O.T.C. for cryostat sectioning. The cryostat sections were immunostained for keratin, laminin, pemphigus vulgaris antigen, bullous pemphigoid antigen and vimentin as described below.

The immunofluorescent studies used to identify keratinocyte antigens were performed following both Phase I and Phase II. Briefly, proliferating keratinocytes obtained on first passage following Phase I were inoculated into bi-well slides using the techniques described above for cell passage. The cells were grown at 37°C in 5% CO_2 within the bi-well slide chambers. Once well formed colonies had been identified (2 days later), the media was removed, the cells were permeablized with acetone, and allowed to air dry. Monoclonal antibodies to human keratin (AEI-AE3), Hybritech, San Diego, CA), bullous pemphigoid, pemphigus vulgaris, and vimentin (Boehringer Mannhem, Indianapolis, IN) were used to test for the presence or absence of these keratinocyte cell markers by indirect immunofluorescence. Direct immunofluorescence was used to test for the presence of laminin. The positive and negative controls used for both immunofluorescent techniques were done on fresh foreskin tissue that had been previously cryopreserved in O.T.C.

b. The procedures described under Phase I of this project resulted in the foremation of isolated keratinocyte colonies that eventually joined together. After approximately 17 days, a single monolayer of keratinocytes developed, covering 80% of the culture flask bottom. Immunofluorescent staining of Phase I cells for the presence of the bullous pemphigoid antigen, the pemphigus vulgaris antigen, keratin, and laminin yielded positive results. Negative results were obtained following staining with the monoclonal antibody against vimentin.

The results derived from Phase II of this experiment demonstrated positive staining using immunofluorescence and mouse monoclonals against bullous pemphigoid, pemphigus vulgaris, keratin, and laminin. The multilayers were also stained with a monoclonal to vimentin, and once again, the results were negative. Two to three days following the addition of DMEM to keratinocyte cultures, the rough, pebbly appearance of the monolayer surface changed. The intercellular spaces between each keratinocyte decreased drastically, resulting in a smooth, continuous sheet of cells. Eight to ten days later, it was determined that the basal layer attached to the flask bottom was actively dividing. This determination was based on the presence of many surface keratinocytes floating in the growth media. Following their removal from the culture flask, the stratified sheets of epithelia were stained with hematoxylin and eosin. This staining revealed the presence of epidermal sheets two to four cell layers thick.

IV. RECOMMENDATIONS

Based on the described techniques, it has been shown that the growth of human keratinocyte monolayers and their subsequent stratification can be accomplished. The cultured cells were undoubtedly determined to be keratinocytes and not fibroblasts. This determination was based on the negative staining observed with the use of a monoclonal to vimentin (a fibroblast protein). Additional confirmation was seen with the positive immunofluorescent staining observed with the use of monoclonals to specific keratinocyte cell markers.

Now that the validity of cell culture technique has been determined, it is possible to investigate those areas in research that involve the application of cell culture to clinical treatment. Included among prospective projects and problems are: 1) The possible use of cell culture technology in the treatment of mechanobullous disease that fail to heal. 2) Comparison of tissue culture techniques with semipermeable and occlusive dressing techniques in the treatment of skin ulcerations and burns. 3) What is responsible for the loss of HLA-DR antigens in cultured keratinocytes and how can this loss be applied to the prevention of graft vs. host reactions in tissue transplants? 4) Can effective dermal equivalents be produced that will substitute for a missing dermis as found in full thickness 3rd degree burns? Although I did not attempt to resolve these problems during my 1988 GSRP, I was able to demonstrate the practicality of generating large numbers of undifferentiated Phase I cells and differentiated Phase II cells from human tissue.

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1988 USAF-UES SUMMER FACULTY RESEARCH PROGRAM

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FINAL REPORT

Autologous Bone Marrow Transplant for Poor Prognosis Lymphomas - A Pilot Dose Escalation Study of a BACE Regimen and Follow-Up

Prepared by : John Y. Salinas

Academic Rank : Medical Student

Department and University : Department of Medicine, Meharry Medical College

Research Location : Wilford Hall USAF Medical Center
Division of Hematology-Oncology
Lackland AFB , San Antonio Texas

USAF Researcher : Dr. James M. Thompson, Maj USAF

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Autologous Bone Marrow Transplant for Poor Prognosis Lymphomas- A Pilot Dose Escalation Study of a BACE Regimen and Follow-Up

by

John Y. Salinas

ABSTRACT

A pilot dose escalation study of autologous bone marrow transplant patients is currently in progress whereby the standard preparatory pre-transplant regimen of BACT established by the National Cancer Institute is being modified by substituting VP-16 Etoposide , for 6-thioguanine currently in use. VP-16 is a more active anti-lymphoma drug. The doses are sequentially being escalated in a patient population which is resistant to standard primary lines of treatment , with relapse of the aggressive lymphoma following a brief remission. The modified regimen , BACE , also involves escalation of BCNU and Ara-C. Initially all patients have been treated with 7 weeks of MACOP-B cytoreduction. Data has been extracted from medical files of already transfused patients. Methods of prospective data retrieval for specific parameters has been incorporated.

ACKNOWLEDGEMENTS

I wish to thank the Air Force Systems Command and the Air Force Office of Scientific Command for sponsorship of this research. Particularly, I would like to to thank the Oncology- Hematology Division at Wilford Hall USAF Medical Center for their support and guidance in this project , I hope I have contributed productively to their innovative and much needed work. My experience was a rewarding and enriching one because of the many skills, contacts, and learning situations which I was exposed to. Specific thanks to Dr. Jim Thompson , Dr. Mary Daly who enlightened my appreciation of medicine and its applications to save people in real life need. I would also like to thank Col. John Cissic who was very helpful during my interaction at Wilford Hall. And last but not least , I would like to thank UES for administartive support throughout this program

I. INTRODUCTION

Major progress in bone marrow transplantation treatments in the past decade have significantly affected prognosis of varied hematological disorders. Recently , bone marrow transplants have received increased attention in the treatment of malignant lymphomas, with particular emphasis to those cases resistant to standard lines of therapy. The major obstacle of such present treatment studies has been in the lack of sufficient patient populations in one center to allow clear statistical analysis of its efficacy and improvement.

The Hematology-Oncology section of the Wilford Hall USAF Medical Center is particularly concerned with the treatment of the many patients with diverse types of particular lymphomas, specifically those unresponsive to conventional primary lines of treatment. Wilford Hall USAF Medical Center has become a major center of referral for the treatment of such hematological disorders within the military and related community, providing an ample patient population for treatment studies and followup of this promising technique of bone marrow transplantation salvage therapy.

My research interests have been in the study and applica tion of hematologic and immunologic techniques in erythropoietic neoplasias which have subsequently led to my interest and assign ment at Wilford Hall USAF Medical Center.

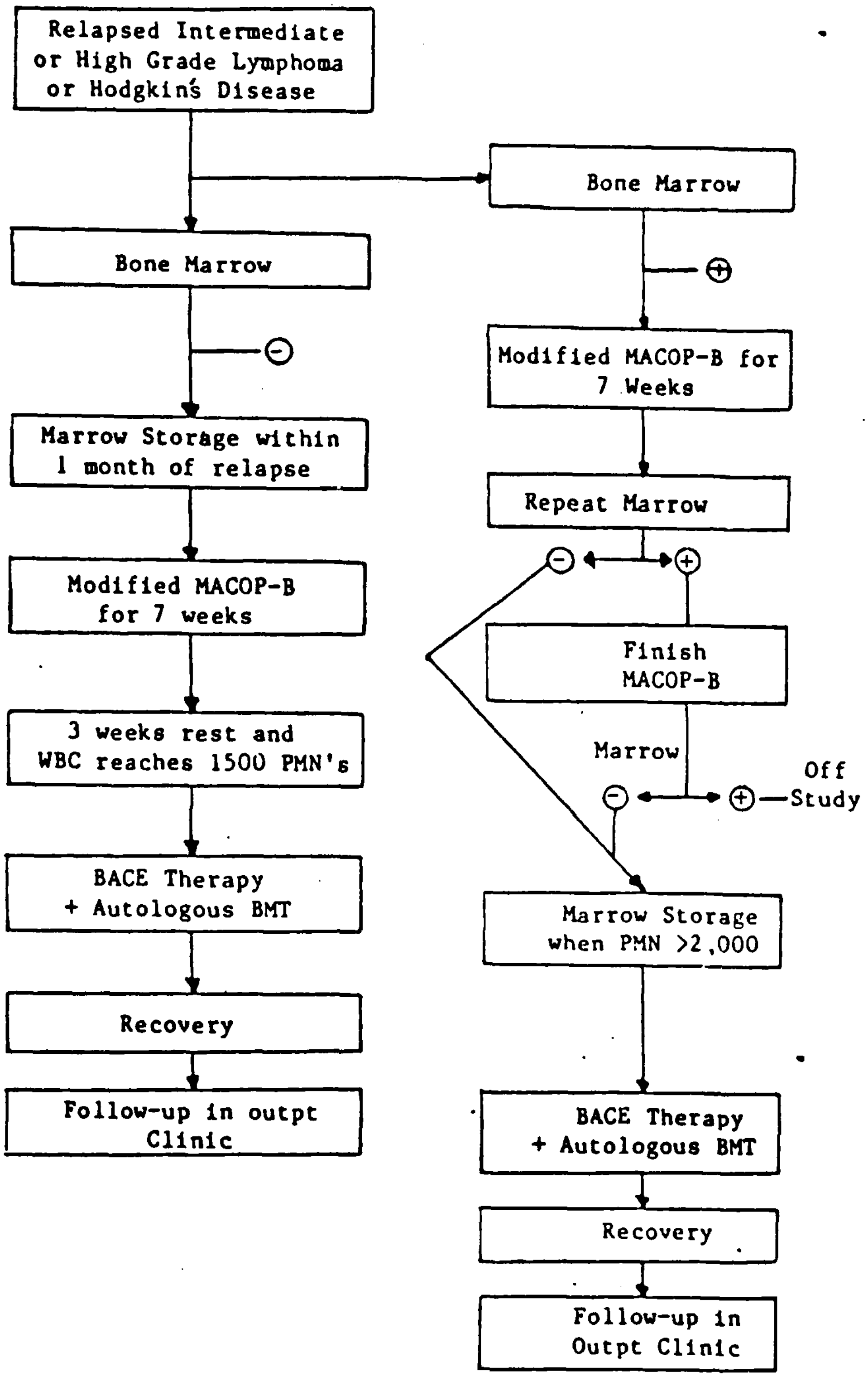
II. OBJECTIVES OF RESEARCH EFFORT

Autologous bone marrow transplantation is receiving much more attention and acceptance as a salvage modality in extreme situations of disease. Until better cytotoxic drugs are available for patients with refractory disease, the use of autologous bone marrow transplantation, which explores the sttep dose response curves of most chemotherapeutic agents remains the most promising approach to relapsing and aggres sive episodes of disease. With the promise of bone marrow transplantation, however, is associated the challenge of statistical study and follow-up of its patients, as well as improvement of therapeutic modalities and applications to appropriate candidates in order to improve its efficacy.

My assignment as a participant in the 1988 Summer Graduate Fellowship Program was to assist in a pilot dose escalation study of autologous bone marrow transplants for poor prognosis lymphomas by retrospective investigation of medical charts and follow-up of involved patients at Wilford Hall USAF Medical Center and involved facilities. It was determined that the accumulated data and results would pro vide a framework of parameters for prospective and ongoing studies of autologous bone marrow transplantation salvage therapy, as well as basis for future publication of work.

In particular, the medical charts of these bone marrow

TREATMENT FLOW SHEET



Atch 1

transplant patients were investigated for protocol evaluations, for isolation of treatment parameters for statistical analysis as well as for toxicities in dose escalations, patient response to dose escalations and for follow-up information for longterm patient outcome. The ultimate utility of this therapeutic approach will not be defined until the doses of active drugs in combination in this pilot study are maximally escalated in patients , with minimal residual disease present at the time of transplant and with satisfactory response to its effects.

In this pilot study as formulated, a modification of the BACT (BCNU , Ara C , Cytoxan , 6 Thioguanine) transplant preparative regimen of the National Cancer Institute was made by substituting VP-16 , a more active antilymphoma drug for 6 thioguanine, and then sequentially escalating the dosages of BCNU , VP-16 , and Ara-C as tolerated. Patients were grouped into the new dosage regimen, known as BACE , in combination schedules previously formulated. Initially all patients were treated with seven weeks of the MACOP-B regimen (Methotrexate, Adriamycin , Cytoxan , Vincristine , Prednisone , Bleomycin) planned to effect pretransplant tumor cytoreduction. There after BACE was administered. Dosage escalation of individual drugs of combination schedules occured in a carefully prescribed fashion to ultimately define the highest doses which would be safely tolerable. Toxicity criteria defined by the Fred Hutchinson Cancer Research Center in Seattle Washington was adopted to carefully monitor regimen related toxicity. The

CYTOREDUCTION MACOP-B	SCHEDULE ROUTE	DAY-69	DAY-62	DAY-55	DAY-48	DAY-41	DAY-34	DAY-27	DAY-20
ADRIAMYCIN 50mg/m^2	IV	X		X		X		X	
CYCLOPHOSPHAMIDE 350mg/m^2	IV	X		X		X		X	
METHOTREXATE 400mg/m^2	IV		X				X		
LEUKOVORIN 15mg	PO		Q6°X6 doses 24 hrs post MTX				Q6°X6 doses 24 hrs post MTX		
VINCRISTINE 1.4mg/m^2 (2.0mg max)	IV		X		X		X		
BLEOMYCIN 10units/m^2	IV				X				
PREDNISONE	PO	<------------75mg PO daily-------------->				<--30mg QD-->	<-----5mgQD------->		

NOTE: Bone Marrow harvested on/about D-71 if bone marrow negative for lymphoma pre-treatment.

Atch 2

TRANSPLANT PREPARATION:

RACE	SCHEDULE	DAY -6	DAY -5	DAY -4	DAY -3	DAY -2	DAY -1	DAY 0
DRUG	ROUTE IV	PATIENT # 1 - 3 350mg/m^2 IV D-6 4 - 6 350mg/m^2 IV D-6,-5 (All Patients $\geq$ 6 if toxicity acceptable)						T R
BCNU 350mg/m2		X	X					
ARA-C	IV	PATIENT # 16 - 19 100mg/m^2 IV Q 12h x 6 doses (days -3,-2,-1) 20 - 22 250mg/m^2 IV Q 12h x 6 doses 23 - 25 500mg/m^2 IV Q 12h x 6 doses (All patients $\geq$ 25, same as #23-25, if toxicity acceptable)			X	X	X	A N S
CYTOXAN 50mg/kg	IV	ALL PATIENTS X	ALL PATIENTS X	ALL PATIENTS X				P L
VP-16 (Etoposide) 250mg/m^2	IV	PATIENT # 1 - 6 125mg/m^2 IV Q 12h x 6 doses (Days -6,-5,-4) 7 - 9 166mg/m^2 IV Q 12h x 6 doses: 10 - 12 208mg/m^2 IV Q 12h x 6 doses: 13 - 15 250mg/m^2 IV Q 12h x 6 doses: (All patients $\geq$ 15, same as #13-15, if toxicity acceptable) X	X	X				A N T
SEPTRA DS	PO	<------DAY -9 TO DAY -2------>						

NOTE: Bone Marrow harvested on/about D-8 if bone marrow initially positive pre-treatment and subsequently negative on D-10. Bone Marrow reinfusion will be on Day 0 for all patients.

ultimate goal is to use the drug doses defined by the pilot study in a phase II trial in an attempt to significantly improve response rates and survival.

As previously documented, Hodgkins and Non Hodgkins lymphoma is curable by primary therapy in approximately 50% of the cases. The remaining cases relapse despite current therapy, the patients eventually succumbing to their disease. The brief remission does not adequately translate to cure. At this point in time, it is felt that bone marrow transplant is the only therapeutic approach that has demonstrated the potential for cure after relapse of an aggressive lymphoma. Nevertheless, the standard pretransplant preparative regimen of BACT currently used has offered only a 20-30% long term survival rate in this patient population, the majority succumb to their illness as well as suffer from well defined toxicities. The present pilot study undertaken by the staff at Wilford Hall a dose escalation study of a BACE regimen, relies on two oncologic principles. The first is the relationship of dose to response; the second is the principle of residual tumor burden. Specifically, the higher the dose of a drug that is administered, the better the response of the tumor treated, potentially the difference between cure and no cure, toxicity not withstanding. With the ability to store autologous bone marrow, capable of hematological reconstitution, drug dosages can be markedly increased taking advantage of this dose respnse relationship. As for residual tumor burden , a more likely curative benefit of the patients increased response to increased dose is evident in the patient with the lowest residual tumor burden at transplantation. The dose escalation study has been initiated with these objectives in mind to determine the utility of this therapeutic approach.

IV RESULTS and DISCUSSION

Preliminary facet of the investigation was to confirm the required MACOP-B regimen given for cytoreduction as per protocol for seven weeks, since most of the patients returned to their referring medical centers of treatment. Concurrence with protocol was examined. Notwithstanding the difficulty of follow-up medical retrieval from referring institutions, it was found that most patients received 90-98% of their treat ment , with some protocol violations evident. Also investigated was the toxicities observed , most commonly involving granulo cytopenia, mucositis, and acral hypoesthesias, all of which were transient. Severe toxicity was unusual in the first seven weeks of therapy, a finding concurrent with a similar MACOP B regimen by Klimo et al for twelve weeks. Bone marrow harvest was performed before or after MACOP B regimen depending upon the patients bone marrow disease status pre treatment. Following a three week rest period, BACE regimen was instituted 6 days prior to transplantation of autologous bone marrow. Many of the parameters extracted from the medical charts revolved around the date of transplant Intervals isolated were important for determination of

 C LAST MACOP-B TO TRANSPLANT

PRODUCT-LIMIT SURVIVAL ANALYSIS

TIME VARIABLE IS SURVIVAL

CASE LABEL	CASE NUMBER	TIME	STATUS	CUMULATIVE SURVIVAL	STANDARD ERROR	CUM DEAD	CUM LOST	REMAIN AT RISK
	3	9.00	DEAD	0.9375	0.0605	1	0	15
	14	12.00	DEAD	0.8750	0.0827	2	0	14
	16	16.00	DEAD	0.8125	0.0976	3	0	13
	13	31.00	DEAD	0.7500	0.1083	4	0	12
	8	33.00	DEAD			5	0	11
	11	33.00	DEAD	0.6250	0.1210	6	0	10
	6	34.00	DEAD	0.5625	0.1240	7	0	9
	10	37.00	DEAD	0.5000	0.1250	8	0	8
	4	40.00	DEAD	0.4375	0.1240	9	0	7
	15	42.00	DEAD	0.3750	0.1210	10	0	6
	2	44.00	DEAD	0.3125	0.1159	11	0	5
	9	45.00	DEAD	0.2500	0.1083	12	0	4
	1	46.00	DEAD	0.1875	0.0976	13	0	3
	5	49.00	DEAD	0.1250	0.0827	14	0	2
	12	55.00	DEAD	0.0625	0.0605	15	0	1
	7	119.00	DEAD	0.0000	0.0000	16	0	0

MEAN SURVIVAL TIME = 40.31 S.E. = 6.182

QUANTILE	ESTIMATE	ASYMPTOTIC STANDARD ERROR
75TH	31.00	9.81
MEDIAN (50TH)	37.00	6.00
25TH	45.00	1.73

BROOKMEYER-CROWLEY 95% CONFIDENCE INTERVAL FOR MEDIAN SURVIVAL TIME
(33.00 , 44.00)

NOTE: BROOKMEYER-CROWLEY CONFIDENCE INTERVAL ASSUMES NO TIES AMONG OBSERVED RESPONSE TIMES. AT LEAST ONE SUCH TIE OCCURRED.

(D)(1) TRANSPLANT TO ANC 500

PRODUCT-LIMIT SURVIVAL ANALYSIS

TIME VARIABLE IS SURVIVAL

CASE LABEL	CASE NUMBER	TIME	STATUS	CUMULATIVE SURVIVAL	STANDARD ERROR	CUM DEAD	CUM LOST	REMAIN AT RISK
	15	15.00	DEAD	0.9412	0.0571	1	0	16
	4	16.00	DEAD	0.8824	0.0781	2	0	15
	9	18.00	DEAD	0.8235	0.0925	3	0	14
	3	21.00	DEAD			4	0	13
	8	21.00	DEAD			5	0	12
	17	21.00	DEAD	0.6471	0.1159	6	0	11
	1	23.00	DEAD			7	0	10
	12	23.00	DEAD	0.5294	0.1211	8	0	9
	14	27.00	DEAD	0.4706	0.1211	9	0	8
	5	29.00	DEAD			10	0	7
	16	29.00	DEAD	0.3529	0.1159	11	0	6
	2	31.00	DEAD			12	0	5
	7	31.00	DEAD	0.2353	0.1029	13	0	4
	10	32.00	DEAD	0.1765	0.0925	14	0	3
	6	33.00	DEAD			15	0	2
	11	33.00	DEAD	0.0588	0.0571	16	0	1
	13	36.00	DEAD	0.0000	0.0000	17	0	0

MEAN SURVIVAL TIME = 25.82 S.E. = 1.582

QUANTILE	ESTIMATE	ASYMPTOTIC STANDARD ERROR
75TH	21.00	1.97
MEDIAN (50TH)	27.00	3.29
25TH	31.00	1.75

BROOKMEYER-CROWLEY 95% CONFIDENCE INTERVAL FOR MEDIAN SURVIVAL TIME
(21.00 , 29.00)

NOTE: BROOKMEYER-CROWLEY CONFIDENCE INTERVAL ASSUMES NO TIES AMONG OBSERVED RESPONSE TIMES. AT LEAST ONE SUCH TIE OCCURRED.

PAGE 2 BMDP1L 17-AUG-88 10:08:21 D ② TRANSPLANT TO ANC 1000

PRODUCT-LIMIT SURVIVAL ANALYSIS

TIME VARIABLE IS SURVIVAL

CASE LABEL	CASE NUMBER	TIME	STATUS	CUMULATIVE SURVIVAL	STANDARD ERROR	CUM DEAD	CUM LOST	REMAIN AT RISK
	6	18.00	DEAD	0.9444	0.0540	1	0	17
	17	19.00	DEAD	0.8889	0.0741	2	0	16
	13	22.00	DEAD	0.8333	0.0878	3	0	15
	2	23.00	DEAD	0.7778	0.0980	4	0	14
	11	24.00	DEAD	0.7222	0.1056	5	0	13
	14	27.00	DEAD	0.6667	0.1111	6	0	12
	15	29.00	DEAD	0.6111	0.1149	7	0	11
	5	32.00	DEAD			8	0	10
	18	32.00	DEAD	0.5000	0.1179	9	0	9
	4	34.00	DEAD	0.4444	0.1171	10	0	8
	3	35.00	DEAD			11	0	7
	12	35.00	DEAD	0.3333	0.1111	12	0	6
	16	36.00	DEAD	0.2778	0.1056	13	0	5
	7	37.00	DEAD	0.2222	0.0980	14	0	4
	10	39.00	DEAD	0.1667	0.0878	15	0	3
	9	40.00	DEAD	0.1111	0.0741	16	0	2
	8	42.00	DEAD	0.0556	0.0540	17	0	1
	1	51.00	DEAD	0.0000	0.0000	18	0	0

MEAN SURVIVAL TIME = 31.94 S.E. = 2.046

QUANTILE	ESTIMATE	ASYMPTOTIC STANDARD ERROR
75TH	24.00	3.17
MEDIAN (50TH)	32.00	3.54
25TH	37.00	2.35

BROOKMEYER-CROWLEY 95% CONFIDENCE INTERVAL FOR MEDIAN SURVIVAL TIME
(27.00 , 35.00)

NOTE: BROOKMEYER-CROWLEY CONFIDENCE INTERVAL ASSUMES NO TIES AMONG OBSERVED RESPONSE TIMES. AT LEAST ONE SUCH TIE OCCURRED.

(D) (3) TRANSPLANT TO PLT 20K

PRODUCT-LIMIT SURVIVAL ANALYSIS

TIME VARIABLE IS SURVIVAL

CASE LABEL	CASE NUMBER	TIME	STATUS	CUMULATIVE SURVIVAL	STANDARD ERROR	CUM DEAD	CUM LOST	REMAIN AT RISK
	2	0.00	DEAD	0.9412	0.0571	1	0	16
	6	14.00	DEAD			2	0	15
	16	14.00	DEAD	0.8235	0.0925	3	0	14
	8	17.00	DEAD	0.7647	0.1029	4	0	13
	1	18.00	DEAD			5	0	12
	11	18.00	DEAD			6	0	11
	14	18.00	DEAD	0.5882	0.1194	7	0	10
	13	26.00	DEAD	0.5294	0.1211	8	0	9
	5	29.00	DEAD	0.4706	0.1211	9	0	8
	7	33.00	DEAD	0.4118	0.1194	10	0	7
	10	34.00	DEAD	0.3529	0.1159	11	0	6
	15	36.00	DEAD			12	0	5
	17	36.00	DEAD	0.2353	0.1029	13	0	4
	4	37.00	DEAD	0.1765	0.0925	14	0	3
	3	41.00	DEAD	0.1176	0.0781	15	0	2
	9	43.00	DEAD			16	0	1
	12	43.00	DEAD	0.0000	0.0000	17	0	0

MEAN SURVIVAL TIME = 26.88 S.E. = 3.013

QUANTILE	ESTIMATE	ASYMPTOTIC STANDARD ERROR
75TH	18.00	2.03
MEDIAN (50TH)	29.00	10.29
25TH	36.00	1.75

BROOKMEYER-CROWLEY 95% CONFIDENCE INTERVAL FOR MEDIAN SURVIVAL TIME
(18.00 , 34.00)

NOTE: BROOKMEYER-CROWLEY CONFIDENCE INTERVAL ASSUMES NO TIES AMONG OBSERVED RESPONSE TIMES. AT LEAST ONE SUCH TIE OCCURRED.

(F) TRANSPLANT TO DISCHARGE

PRODUCT-LIMIT SURVIVAL ANALYSIS

TIME VARIABLE IS SURVIVAL

CASE LABEL	CASE NUMBER	TIME	STATUS	CUMULATIVE SURVIVAL	STANDARD ERROR	CUM DEAD	CUM LOST	REMAIN AT RISK
	13	19.00	DEAD	0.9444	0.0540	1	0	17
	11	23.00	DEAD			2	0	16
	18	23.00	DEAD	0.8333	0.0878	3	0	15
	12	24.00	DEAD	0.7778	0.0980	4	0	14
	10	29.00	DEAD			5	0	13
	16	29.00	DEAD	0.6667	0.1111	6	0	12
	5	37.00	DEAD			7	0	11
	7	37.00	DEAD	0.5556	0.1171	8	0	10
	15	38.00	DEAD	0.5000	0.1179	9	0	9
	17	40.00	DEAD	0.4444	0.1171	10	0	8
	4	42.00	DEAD			11	0	7
	6	42.00	DEAD			12	0	6
	8	42.00	DEAD	0.2778	0.1056	13	0	5
	2	43.00	DEAD	0.2222	0.0980	14	0	4
	1	48.00	DEAD	0.1667	0.0878	15	0	3
	3	54.00	DEAD	0.1111	0.0741	16	0	2
?	14	246.00	DEAD	0.0556	0.0540	17	0	1
	9	406.00	DEAD	0.0000	0.0000	18	0	0

MEAN SURVIVAL TIME = 67.89 S.E. = 23.165

QUANTILE	ESTIMATE	ASYMPTOTIC STANDARD ERROR
75TH	29.00	4.00
MEDIAN (50TH)	38.00	3.18
25TH	43.00	2.82

BROOKMEYER-CROWLEY 95% CONFIDENCE INTERVAL FOR MEDIAN SURVIVAL TIME
(29.00 , 40.00)

NOTE: BROOKMEYER-CROWLEY CONFIDENCE INTERVAL ASSUMES NO TIES AMONG OBSERVED RESPONSE TIMES. AT LEAST ONE SUCH TIE OCCURRED.

 J TRANSPLANT TO RELAPSE

PRODUCT-LIMIT SURVIVAL ANALYSIS

TIME VARIABLE IS SURVIVAL

CASE LABEL	CASE NUMBER	TIME	STATUS	CUMULATIVE SURVIVAL	STANDARD ERROR	CUM DEAD	CUM LOST	REMAIN AT RISK
	4	56.00	DEAD	0.7500	0.2165	1	0	3
	1	57.00	DEAD	0.5000	0.2500	2	0	2
	3	79.00	DEAD	0.2500	0.2165	3	0	1
	2	385.00	DEAD	0.0000	0.0000	4	0	0

MEAN SURVIVAL TIME = 144.25 S.E. = 80.425

QUANTILE	ESTIMATE	ASYMPTOTIC STANDARD ERROR
75TH	56.00	
MEDIAN (50TH)	57.00	11.50
25TH	79.00	142.03

LOWER ONE-SIDED BROOKMEYER-CROWLEY 95% CONFIDENCE LIMIT FOR MEDIAN SURVIVAL TIME = 56.00

of concurrence of drug administration with proposed protocol as well as for examination of intervals required for recovery from transplantation and disease, determined by Absolute Neutrophilic counts (ANC) greater than 500 , ANC $>$1000 , platelet counts $>$20,000 , and date of discharge or death. A summary of toxicity was also organized by organ system for patients involved in study.

It was observed as far as protocol was concerned, that the actual intervals from transplantation were often longer, probably arising from complications and noncompliance, and realistically not the ideal. The expected 70 day interval from initiation of MACOP B to transplant was for example a much longer mean of 90.69 $\pm$ 7.015. That for termination of MACOP B to transplantation was 40.31 $\pm$ 6.182 also longer than the ideal 27 days. The BACE regimen was for the most part close to ideal since it was supervised at Wilford Hall as opposed to MACOP-B regimens. Concurrence with protocol dosages, as stated previously was found to be approximately in the 95% range of compliance with protocol.

More importantly, however, was the observation of recovery from transplantation. Criterai were established as stated previously, such as ANC $>$500, ANC$>$1000, platelets $>$20K, and the intervals required to reach these criteria were tabulated for each patient. It was found that from transplant to ANC$>$500 took on the average 25.82 $\pm$ 1.5, a fairly consistent observation. To achieve an ANC$>$1000 the data revealed 31.94 $\pm$ 2.046. Platelets $>$20K were obtainable on the average at 26.88 $\pm$ 3.013, a result compatible with the ANC $>$500. The results from platelets were derived from a consistent 3 day plateau of platelet levels following many transfusions. With the exception of the two patients who suffered particular complications, the transplant-discharge interval values were found to display two predominant modes at 37 and 42 days. For those for whom transplant proved to be of lethal complications, an interesting mean interval of 107.71 $\pm$ 29.37 was determined. Probably the most interesting however, was the transplant-relapse interval mean of 144.25 $\pm$ 80.42 of significance to possibly determine roughly the criteria point for success / failure of an autologous bone marrow transplant.

V. RECOMMENDATIONS

Further studies with longer follow-up periods will assist in accurately determining the curative potential of this treatment modality. Longer follow-up periods are needed to prove that this technique can cure an acceptable proportion of patients before this strategy can be accepted for widespread clinical use. The present study serves as groundwork for the ongoing work at Wilford Hall. The substantial patient population and referrals available at Wilford Hall USAF Medical Center lend excellent advantages to available research data towards this therapeutic modality. It is hoped that the present work has identified needs and suggestions for data retrieval. Throughout the research period, various forms and retrieval . Throughout the research period , various forms and tables were designed for incorporation into departmental charts and individual patient files to facilitate the retrieval and proper recording of pertinent data towards this research effort. It is hoped that application for a mini grant will provide the necessary funding to continue this project with the completion of a publishable work. The data , resources, and potential for publication greatly favor support of such a project. Although further analysis of the data collected is currently in progress, the facilitation of data retrieval , and future recording of data , which this project has hopefully assisted, will permit a more detailed and accurate analysis of the value of autologous bone marrow transplantation in aggressive lymphomas with high dose chemo therapeutic regimens. It has been suggested that while the toxicity of autologous bone marrow transplants is significant, younger patients with multiple unfavorable prognostic variables, exposure to only one or two prior chemotherapy regimens, and a limited chance of cure by standard second line of therapy may benefit. This suggestion is an area which can be investi gated at Wilford Hall and compared in a future project with present and prospectively acquired data which this project has hopefully facilatated or served as an impetus for.

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1988 USAF-UES Summer Graduate Student Research Program

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Final Report

Abdominal Abscess Formation in the Mouse Model

Prepared by:	Kimberly F. Smith
Academic Rank:	Sophmore
Department and University:	School of Medicine Meharry Medical College
Research Location:	Clinical Investigations Directorate Wilford Hall Medical Center Lackland Air Force Base
USAF Researcher:	Capt. D. V. Bradley
Date:	Summer 1988
Contract No.:	F49620-88-C-0053

Kimberly Smith

ABDOMINAL ABSCESS FORMATION IN THE MOUSE MODEL

ABSTRACT

The purpose of this research project was to develop a reproducible model of intraabdominal abscess formation in mice. The sterilized cecal contents of twenty adult meat-fed Wistar rats were combined with a suspension of *Bacteroides distasonis* and *Streptococcus faecalis* and injected intaperitoneally into twenty juvenile C57/BL6 mice. The mice were allowed food and water on a continual basis for one week following injection and thereafter euthanized and examined for abdominal abscess formation.

ACKNOWLEDGEMENT

I would like to take this opportunity to acknowledge the following mentioned; The A.F. Systems Command, The A.F. Office of Scientific Research and The Lackland Air Force Clinical Investigations Directorate. I would also like to acknowledge the following individuals for their genuine concern and thoughtfulness in helping me acheive the desired research goals which I acheived during this summer project; Col. J. Cissik, Lt. Col. A. Vandellen, Major K. Lally, Major W. Patterson, Capt. D. Bradley, Capt. F. Rowan, Sgt. R. Taylor, Sgt. D. Goddu, Sgt. S. Boike and the animal care technicians. Without the guidance of the above mentioned, my summer research experience would not have been as fulfilling.

INTRODUCTION

The major research objective of this project was to develop a reproducible model of intraabdominal abscess formation in the mouse. Penetrating wounds of the abdomen are common in combat, resulting in fecal peritonitis with abdominal abscess which may lead to serious illness and/or death. Through various research efforts it was determined that the necessary constituents for abdominal abscess formation were cecal material and the anaerobic bacterial strain , Bacteriodes. Bacteriodes is a gram negative anaerobic bacillus, a group which comprises 99% of the normal fecal flora and is most commonly recovered from clinical anaerobic infections in conjunction with a facultative aerobe. B. fragilis, a member of this group, which is bile resistant, is the most resistant microorganism to antimicrobial agents than any other anaerobe typically found in clinical specimens and constitutes the dominant portion of normal colonic flora. It has also previously been shown that B. fragilis alone has the ability to cause abscesses without the aid of an accompanying facultative aerobic organism (e.g. Enterococcus) which is the opposite case of the organism of our choice, B. distasonis. In addition, B. distasonis lacks the capsule which is found surrounding B. fragilis. It has been suggested that this capsule may provide the neccesary constituents to produce abscesses which is afforded B. distasonis by the accompanying facultative organism. The cecal material is thought to provide the framework necessary for the development of abscesses but as of yet the virulent factors provided by the facultative organism have not been thoroughly characterized. The purpose of this study is to demonstrate the ability of _B. distasonis_ to also cause abscess formation when the two above mentioned constituents have been added.

The cecal contents were collected from twenty adult Wistar rats which were fed 25 grams of lean ground meat daily for two weeks before euthanization by exposure to carbon dioxide gas. The cecal contents were sterilized, mixed 50:50 with sterile peptone yeast glucose broth and frozen at -90 C for further use. Twenty C57/BL6 mice were allowed laboratory mice chow and water on a continual basis for one week to allow for acclimatization and then injected intraperitoneally with a sterile cecal contents/_B. distasonis_/_Strep. faecalis_ mixture. A control group was injected with a mixture of sterile cecal contents/saline solution. The mice were again allowed food and water ad libum for another week at the end of which they were euthanized via carbon dioxide gas. The mice were then autopsied and examined for the presence of abdominal abscess formation.

MATERIALS AND METHODS

Materials

Animals - Twenty adult male Wistar rats (Harlans Laboratories) weighing between 160-190 grams and twenty C57/BL6 male mice (Jackson Laboratories) weighing approximately 20 grams were incorporated into this project. The rats were fed 25 grams of lean ground meat daily and water. The mice were fed laboratory mouse chow and water ad libum.

Media - 5% sheep blood agar, CDC anaerobic blood agar and chocolate agar plates were used to culture the bacteria necessary for this experiment.

Inoculum- Streptococcus faecalis (ATCC 299T)

*Streptococcus faecalis (ATCC 29212)

Bacteroides distasonis (ATCC 8503) and sterilized rat cecal contents were combined and injected into mice as inoculum in two tenth milliliter quantities.

Miscellaneous- A sterile loop was used to transfer cultures to fresh media. An anaerobic hood, an anaerobic incubator 37 C and aerobic incubator 37 C were used to grow up the cultures. Swabs were used to transfer bacteria from the agar plates to 0.9% NaCl sterile solutions in order to make bacterial suspensions. TB syringes and 25 guage needles were used to inject the inoculum intraperitoneally into the mice.

MATERIALS AND METHODS

Methods

Twenty adult male Wistar rats were fed 25 grams of ground lean meat daily and water for two weeks. The rats were then euthanized by exposure to carbon dioxide gas, their cecums removed and the cecal contents pooled. The cecal contents were then sterilized (autoclave, 251 F for 1 hour), mixed 50:50 with sterile peptone yeast glucose broth, filtered through sterile gauze and deep froze at -90 C for later use.

Twenty C57/BL6 male mice were fed laboratory mouse chow and water ad libum for one week prior to injection to allow for acclimatization.

A suspension of _B. distasonis_ of 0.14 absorption reading (660 nm) and a suspension of _S. faecalis_ of 0.16 absorption reading (660 nm) was combined with the sterile cecal contents/peptone yeast glucose broth mixture in 25:25:50 portions and injected intraperitoneally in 0.2 ml quantities into seven groups of four mice using a TB syringe and a twenty five guage needle. Four control mice were injected with nonvirulent mixture of sterile cecal contents/peptone yeast glucose broth and saline in 50:50 portions by the same means as described for the virulent inoculum. The mice were again allowed laboratory mouse chow and water ad libum for one week and sacrified by exposure to carbon dioxide gas. All mice were then autopsied for the presence of abdominal abscess formation.

*_Strep. faecalis_ (ATCC 29212) was suspected of having lethal effects and was substituted with the ATCC 299T strain.

RESULTS

Estimation of Bacterial Counts of Inoculum

Dilutions of the microbial suspensions used to prepare the inoculum were used to determine the microbial count by use of spread plates (blood agar). The dilutions were prepared in sterile 0.9% (NaCl) saline solution, spread plated onto blood agar and incubated for 24 hours at 37 C. The number of colonies per specific dilutions was determined and the number of colony forming units per milliliter was determined. It was determined from the two strains of bacteria utilized in this experiment that a 0.14 absorption reading (660nm) suspension of B. distasonis (ATCC 8503) and a 0.16 absorption reading (660nm) suspension of S. faecalis (ATCC 299T) both produced a ten to the eighth power colony forming unit per milliliter count. Therefore it was determined that one fifth of this quantity was injected intraperitoneally into the mice as an 0.2 ml inoculum.

Bacterial *Inoculum*

The following tables describe the contents of the inoculums prepared and injected on the dates given;

Table 1 - First Inoculation, June 12, 1988

Group	Agent: sterile saline	cecal contents	B. dist (8503)	S. faec (29212)
1	-	+(.50ml)	+(.25ml)	+(.25ml)
2(control)	+(.50ml)	+(.50ml)	-	-

*Absorption readings(660nm); B. dist 0.14, S. faec 0.16

Table 2 - Second Inoculation, June 29, 1988

Group	cecal contents	B. frag (23745)	B. dist (8503)	S. faec (29212)	S. faec (299T)
3	+(.50ml)	-	+(.25ml)	+(.25ml)	-
4	+(.50ml)	-	+(.25ml)	-	+(.25ml)
5	+(.50ml)	+(.25ml)	-	-	+(.25ml)

*Absorption readings(660nm); B. dist 0.14, S. faec 0.16

Table 3 - Third Inoculation, July 18, 1988

Group	sterile saline	cecal contents	B. dist (8503)	S. faec (299T)
6	-	+(.50ml)	+(.25ml)	+(.25ml)
7	-	+(.50ml)	•+(.25ml)	+(.25ml)
8	+(.75ml)	-	•+(.25ml)	-

*Absorption readings (660nm); B. dist•0.17/0.14, S. faec 0.16

Summary

Two mortality cases resulted overnight from group 1 of the first inoculation (June 12,1988). No cause of death was revealed during autopsy of mortality cases. No abscesses observed after euthanization of the remaining mice nor in the mortality cases. Two mortality cases overnight from group 3, one mortality case two days after injection from group 4 and one mortality case nine days after injection from group 5 resulted from the second inoculation (June 29,1988). No cause of death was revealed during autopsy of any of these mice. No abscesses were observed in the remaining group 3 mice. Abscesses were observed in two out of four of the group 4 mice and in three out of four of the group 5 mice. In both groups, abscesses were found on the liver and intestines. There were no mortality cases from the third inoculation (July 18,1988). Upon euthanization, abscesses were observed in one out of four of the group 6 mice, two out of the four group 7 mice and in one of the group 8 mice.

RECOMMENDATIONS

The succesful development of a model for the study of intraabdominal abscess formation would surely prove profitable in the field of study of care for combat injuries, stab wounds, erupted appendices and the like. Although we were approaching the development of such a model, I feel that a few adjustments will be necessary to improve the results of our experimentations. First of all, as is stated in the introduction, B. distasonis has a considerably weaker ability to cause abscess formation than does B. fragilis, the microorganism which has already been proven to successfully provide an intraabdominal abscess model in mice. The cause for this difference in virulence has been thought to be the presence of a polysaccharide capsule around B. fragilis, where as B. distasonis lacks such a capsule. In the previous studies performed, in which B. fragilis was the microorganism of study, wheat bran was added to the inoculum to aid as an irritant to the contents of the peritoneal cavity thus allowing the microorganism to adhere and enter it's murtiplication-growth phase. Therefore I would suggest that the experiments be repeated with the addition of bran to the inoculum. Another note on the virulence of B. fragilis vs B. distasonis; since B. distasonis is considerably weaker at forming abscesses, it would be of interest to determine if whether or not an increase in the number of colony forming units per milliliter of B. distasonis injected per inoculation would not cause the results obtained in this experiment to more closely approximate those obtained from the B. fragilis studies in which formation of abscesses after injection was observed in nearly one hundred percent of the specimens.

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1988 USAF-UES SUMMER FACULTY RESEARCH PROGRAM/

GRADUATE STUDENT RESEARCH PROGRAM

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FINAL REPORT

I. Impact of Diabetes Mellitus on Overall Health and Functional Status: A Comparison of Two Instruments of Measurement

II. A Comparison of the Effects of Alprazolam, Amitriptyline, and Placebo on Reaction Time in Patients with Chronic Low Back Pain

Prepared by: Paul R. Tanner

Academic Rank: Medical Student

Department and University: School of Medicine, Meharry Medical College

Research Location: USAF WHMC/SGHMME, Lackland AFB, San Antonio, TX 78236

USAF Researcher: James M. Jacobson, M.D.

Date: 5 August 1988

Contract No.: F49620-88-C-0053

ABSTRACT

The Diabetic Effects Assessment and Recording Scale (DEARS) was shown to be reliable and valid in a study involving 206 diabetic patients. We took patient responses from 130 of the same patients and had the responses coded and entered into a data bank. After the data was checked for accuracy, data from the DEARS and the Locus of Control for Diabetes were to be statistically analyzed using analysis of variance to obtain interpretable results. We hypothesized that lower scores on the DEARS should correlate with internalized scores on the LOC. The importance of these results will be to show further validation for the DEARS as a measurement instrument to assess comprehensive health status in diabetes.

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My experiences at Wilford Hall USAF Medical Center have been very rewarding and have given me the opportunity to learn much about the qualities of joys of a researcher. Col James M. Jacobson has provided me with exceptional support and encouragement for my endeavors and proved to be a rich source of information and positive leadership. The help of Dr Stephen A. Brietzke and Dr Mark E. Romanoff as the principle heads of my two research projects was invaluable in helping to find solutions to the many roadblocks I faced. The friendliness and guidance of the entire Endocrinology staff, fellows, the secretary, Ms Hortencia Salazar, and A1C Mark Timmons helped to make an enjoyable working atmosphere for my research experiences. The patience and help of Miss Georgia Henderson with data processing was especially appreciated.

I. Introduction:

A) Diabetes Mellitus is a disease of the endocrine system which involves glucose metabolism by the body. There are many physical and psychosocial aspects of diabetes which affect patient functioning. Because of the number of complications involved with diabetes, many clinical instruments have been developed to measure specific aspects of diabetic functioning.

The Endocrinology Service at Wilford Hall USAF Medical Center at lackland AFB, San Antonio, TX was concerned with developing a comprehensive instrumentto measure overall health status in diabetes. They developed the Diabetes Effects Assessments and Recording Scale (DEARS) in a validation study involving 206 hospital inpatients and outpatients diagnosed as having diabetes mellitus. The DEARS is a 75 question survey grouped into 12 subscales to measure physiologic and psychosocial aspects of health status in diabetic patients. In order to prove the reliability and validity of DEARS, data from several other health status questionnaires including Locus of Control for Diabetes and the Sickness Impact Profile were collected from 130 of the patients who participated in the DEARS study.

My background in psychology and present status as a medical student helped to motivate my research interests involving the statistical analysis of the data involved in the diabetes study. My experiences as a senior research technician in a physiology lab exposed me to working with data collection and literary searches involved with experimental research. Finally, my exposure to and great interest in the endocrine system as a medical student contributed to my appointment as a summer research fellow with the Endocrinology Division.

B) Amitriptyline and Alprazolam are in a class of drugs known as antidepressants. Antidepressants have been used successfully in the treatment of chronic pain syndrome, but they exhibit sedative side effects in varying degrees of severity. These side effects can have an adverse affect on job performance and compliance with a medical regimen of treatment.

The Anesthesiology Service at Wilford Hall USAF Medical Center is concerned with testing the number and severity of side effects associated with Amitriptyline and Alprazolam through the use of a reaction time device developed by Repperger, Jacobson et al (1985). The testing device measures simple time (SRT), choice reaction time (CRT) and decision time (DT) involved with the processing of a one bit piece of information by the Central Nervous System.

Because Alprazolam is a new drug, the Anesthesiology Department wishes to test if Alprazolam can be shown not to impair job performance. Alprazolam costs considerable more than Amitriptyline, and the study is also designed to determine if Alprazolam will be effective enough to warrant the higher cost for its use by the Air Force.

Because I also have research interests in the area of sensory responses to external stimuli, I was assigned to work on this second project as part of my summer research appointment. My work experience with patient care in hospital settings, and my studies of patient management in Behavioral Sciences courses in college and medical school also contribute to my assignment to this reaction time study.

II. OBJECTIVES OF THE RESEARCH EFFORT

My assignment as a summer graduate research fellow was to participate in two different projects related to my current interests and background.

A) The first project concerned the correlation of the newly developed DEARS with the Locus of Control for Diabetes. Several initial goals were planned. the first goal was to have patient responses to four different questionnaires entered into computer data base, and to check the accuracy of the data after it had been compiled. The second goal was to complete an extensive literary review to gather information about the development, history, and uses for the DEARS and LOC in health status measurement. The final goal was to run a statistical analysis on the data for use in writing a paper for publication concerning correlations between the DEARS and the LOC in order to show further validation of the DEARS.

B) The second project concerned the use of the reaction time device to measure the different effects of Alprazolam, Amitriptyline, and Placebo on SRT, CRT, and DT in patients with low back pain. Twenty patients from the hospital's Pain Clinic were were to be tested over a period of nine weeks during which time they will receive two different doses of each study drug at two week intervals and placebo for one week. Responses for SRT and CRT are recorded using the reaction device, and patients will fill out a symptom score sheet at each testing session. At the conclusion of the study, the data will be analyzed using appropriate statistical measures.

III. The patient responses to the LOC, SIP, Diabetes Mellitus Treatment Scale, and the Diabetes Mellitus Optimism Scale were taken to a statistician at the Brady Green Clinic in San Antonio. The responses were entered into an IBM computer data base under a separate code for each patient set of responses over a period of two weeks. Data printouts for each questionnaire were then checked for accuracy against the original patient responses. Corrections were entered into the computer files and updated data printouts and floppy disks were produced. The data was then entered into a central file on the VAX computer system at Wilford Hall Medical Center.

For the literary search, 25 to 30 articles concerning the DEARS and its development were selected and obtained through the hospital medical library and the Inter-library loan program. I read the articles and grouped them into related topics for use in preparing the study paper. Fifty-five to 60 articles concerning the history, development, and use of the Locus of Control survey were also obtained from the library.

The original Locus of Control survey, (the I-E scale), developed by Rotter was a 29-item, forced-choice test based on Rotter's social learning theory. In 1976, Wallston et al developed the 11-item Health Locus of Control Scale (HLC) based on Rotter's original I-E scale. The HLC was designed to provide a more sensitive prediction of measured degree of internality to health related behaviors. The factor structure for the HLC was assessed for its use in experimental research by Boyle and Harrison.

In 1979, Wallston, Wallston, and DeVillis developed an 18-item, Multidimensional Health Locus of Control Scale (MHLC) based on their earlier HLC scale. The MHLC is a Likert-type scale divided into 3 subscales: Internal Health Locus of Control, Powerful Others Health Locus of Control, and Chance Health Locus of Control. The MHLC was modified by Bishop and Jacobson (1987) into the Locus of Control scale for Diabetes for correlation with the DEARS.

Because my ten-week assignment ended before data for the LOC and DEARS could be analyzed, the remainder of the work in completing the paper for publication will be continued by Dr Stephen A. Brietzke of the Endocrinology Service at WHMC.

B. Patients for the reaction time study were selected voluntarily from the hospital Pain Clinic. Patients were told that the study was a 9-week double-blinded study in which they would be given two doses of Alprazolam and Amitriptyline. A history of patients backpain problems and treatment were taken, and a consent form was signed before the study began.

The patients were then acquainted with the reaction time device before baseline measurements were taken. The reaction device is a sensitive microprocessor based device which measures time in milliseconds for the patient to respond to a light coming on. Patients start the device by depressing a "reset" switch on the test box with their left hand. After a random interval form 1.5 to 7 seconds, one of two red lights on the box turns on. The patient then uses a left-right toggle switch with their right hand to break an electrical connection and turn off the red light.

Patients are verbally encouraged to react as swiftly as possible to turn off the red light when they first notice the light coming on. Patients respond to two different situations. In one situation, either the left light or the right light only will turn on, and this measures the time to make a simple decision (SRT). In the other situation, either light is randomly set to come on, an the patient must choose which direction to move the turn-off switch. This measures the Choice Reaction Time for the patient's response. To obtain the Decision Time for reaction by the CNS, the Srt is subtracted from the CRT.

To date, eleven patients volunteered, and were tested for baseline measurements. Two patients failed to return for subsequent testing, and three more were dropped from the study due to adverse drug reactions. The remaining seven patients were tested at two week intervals for the first four weeks. An initial dose of either Alprazolam or Amitriptyline was given during the first two weeks. In the fifth week, all patients were given a placebo and were tested at the end of the week. For the sixth through the ninth weeks, patients will b given the other drug and again measured at two week intervals.

This study will be continued to its completion by the principal investigator, Dr Mark E. Romanoff of the Anesthesiology Department of Wilford Hall USAF Medical Center.

IV. RECOMMENDATIONS

A) The DEARS offers a valid and reliable measurement instrument for health status and functioning of diabetics. Hopefully, the results from correlational studies with the LOC and SIP will establish the DEARS as an acceptable test in the literature. Further studies utilizing the DEARS on different diabetic populations are encouraged to strengthen its validity and to modify any weak points in its construct.

B) The reaction time device is an instrument that seems to have endless possibilities for future research under many different situations. I recommend that the device should be utilized as a standard test for development of new drugs in their testing for effects in human studies.

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