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EIGHTH ANNUAL CONFERENCE ON SHOCK

Baltimore, Maryland
Sunday, June 9—Wednesday, June 12, 1985

Program

Sunday, June 9

1:00 to 5:00 p.m.	Registration	Assembly Area
1:00 to 5:00 p.m.	Council Meeting	Parlor F
7:30 p.m.	Opening Program	Ballroom
8:00 p.m.	Keynote address: "Application of Animal Shock Models to the Human" **Lerner B. Hinshaw, PhD** President-Elect University of Oklahoma	Ballroom
9:00 to 10:00 p.m.	Reception	Ballroom

Monday, June 10

7:30 a.m. to 6:00 p.m.	Registration	Assembly Area
8:30 a.m. to 8:45 a.m.	Introduction to Scientific Session	Hunt Room
8:45 to 10:45 a.m.	Symposium I: "Eicosanoids In and Out of Shock" Presiding: **James A. Cook, PhD** Medical University of South Carolina, Charleston, and **J. Raymond Fletcher, MD, PhD** Vanderbilt University 1) "Biological Importance of the Eicosanoids" **Bryan Smith, PhD** Temple University 2) "Role of Eicosanoids in Disease States Other Than Shock" **Perry V. Halushka, MD, PhD** Medical University of South Carolina, Charleston 3) "Importance of Cyclooxygenase Products in Shock-Like States" **James A. Cook, PhD** Medical University of South Carolina, Charleston 4) "Role of Leukotrienes in Endotoxin Action" **Dietrich Keppler, MD** University of Freiburg, Germany 5) "Significance of Eicosanoids in Medicine and Clinical Implications for Shock Therapy" **J. Raymond Fletcher, MD, PhD** St. Thomas Hospital, Nashville, Tennessee	
11:00 a.m. to 12:00 p.m.	Poster Session I, papers 1-36	Maryland 3

Program

12:00 to 12:45 p.m.	Poster Discussion	
	1) "Hypovolemia," papers 1–12 Chair: **Jureta W. Horton, PhD** University of Texas, Dallas	Maryland 1 & 2
	2) "Metabolism I," papers 13–24 Chair: **Leena M. Mela-Riker, MD** Oregon Health Sciences University	Maryland 4
	3) "Pulmonary," papers 25–36 Chair: **David N. Herndon, MD** University of Texas, Galveston	Garden Room
12:45 to 1:30 p.m.	Lunch	
1:45 to 3:15 p.m.	"Young Investigators Awards," papers 37–40 Presiding: **David G. Reynolds, PhD** University of Iowa 1) "Interactions Between Hepatocytes and Kupffer Cells During Sepsis" **Michael A. West, MD** University of Minnesota 2) "The Role of C5 in the Adult Respiratory Distress Syndrome" **Laurel Matthews Olson, MD** The University of Chicago 3) "Assessment of Soleus Muscle Amino Acid Transport Alterations in Endotoxic Rats" **Michael D. Karlstad, MS** Loyola University, Chicago 4) "Oxidant Injury and Cellular Morphology" **Daniel B. Hinshaw, MD** Scripps Clinic and Research Foundation	Hunt Room
3:15 to 3:30 p.m.	Coffee Break	Assembly Area
3:30 to 5:30 p.m.	Paper Session I, papers 41–48 Presiding: **W. Curtis Wise, PhD** Medical University of South Carolina, Charleston	Hunt Room
5:30 to 6:30 p.m.	Business Meeting	Hunt Room
7:00 to 8:00 p.m.	Reception	Ballroom Foyer
8:00 to 9:00 p.m.	Dinner	Ballroom
9:00 to 9:30 p.m.	Speaker: **Mr. Bob Willis** Director of Tourism for Baltimore "The New Baltimore"	

Tuesday, June 11

8:00 a.m. to 12:00 noon	Registration	Assembly Area
8:30 to 10:30 a.m.	Symposium II: "The Lung in Shock" Presiding: **Daniel L. Traber, PhD** University of Texas, Galveston, and **Ulf Haglund, MD, PhD** University Lund, Malmö, Sweden	Hunt Room

Program

	1) "ARDS - General Overview" **Roger G. Spragg, MD** University of California, San Diego 2) "Effect of Burn Injury to the Lung" **George C. Kramer, PhD** University of California, Davis 3) "Effect of Hemorrhagic Shock on the Lung" **Günther Schlag, MD** FAF Anesthesiologie, Vienna, Austria 4) "Sepsis and the Lung" **Robert H. Demling, MD** Harvard University	
10:45 to 11:45 a.m.	Poster Session II, papers 49-96	Maryland 3
11:45 to 12:30 p.m.	Poster Discussion	
	1) "Metabolism II," papers 49-60 Chair: **Irshad H. Chaudry, PhD** Yale University	Maryland 4
	2) "Opiates," papers 61-72 Chair: **Thomas Vargish, MD** West Virginia University	Parlor A
	3) "Cells," papers 73-84 Chair: **Peter A. Ward, MD** University of Michigan	Maryland 1 & 2
	4) "Eicosanoids," papers 85-96 Chair: **John T. Flynn, PhD** Jefferson Medical College	Garden Room
Free Afternoon		

Wednesday, June 12

8:30 a.m. to 12:00 p.m.	Registration	Assembly Area
8:00 to 10:00 a.m.	Symposium III: "Vascular Smooth Muscle Control" Presiding: **Robert F. Bond, PhD** Oral Roberts University, and **Bart Chernow, MD** Bethesda Naval Hospital 1) "Physiology of Vascular Smooth Muscle" **Harvey V. Sparks, Jr., MD** Michigan State University 2) "Regional Vascular Control During Hypotension and Shock" **Robert F. Bond, PhD** 3) "Vascular Smooth Muscle Membrane Potentials During Hypotensive Stress" **Julian H. Lombard, PhD,** Medical College of Wisconsin 4) "Pharmacological Manipulation of the Peripheral Vasculature in Shock States" **Bart Chernow, MD** 5) "Clinical Management of the Vasculature in Shock States" **Bryan L. Roth, MD** Bethesda Naval Hospital	Valley Room

Program

10:15 to 10:30 a.m.	Coffee Break	Assembly Area
10:30 a.m. to 12:30 p.m.	Paper Session II, papers 97–104 Presiding: **Thom E. Lobe, MD** University of Texas, Galveston	Valley Room
12:30 to 1:45 p.m.	Lunch	
1:45 to 3:45 p.m.	Workshop I: "Critical Issues in Shock Research" Presiding: **David G. Reynolds, PhD** University of Iowa, and **Charles L. Rice, MD** University of Washington, Seattle 1) "Is the Hyperdynamic Circulatory State Important in Sepsis?" **Irshad H. Chaudry, PhD** Yale University 2) "Brain Injury Research in 1985: Where Are We?" **Herbert J. Proctor, MD** University of North Carolina 3) "What Is the Status of Pharmacological Treatment in Spinal Cord Injury?" **Wise Young, MD, PhD** New York University 4) "What Are the Lymphokines and What Are They Doing in Shock?" **Satya N. Chatterjee, MD** University of California, Davis	Valley Room
3:45 to 5:45 p.m.	Paper Session III, papers 105–112 Presiding: **Robert R. Wolfe, PhD** University of Texas, Galveston	Valley Room
5:45 to 6:30 p.m.	Poster Session III, papers 113–151	Maryland 3 & 4
6:30 to 7:15 p.m.	Poster Discussion 1) "Endotoxin/Cardiovascular," papers 113–124 Chair: **H. Richard Adams, DVM, PhD** University of Missouri, Columbia 2) "General Session I," papers 125–136 Chair: **Linda T. Archer, PhD** University of Oklahoma 3) "General Session II," papers 137–151 Chair: **Thomas E. Emerson, PhD** Cutter Group of Miles Laboratories	Maryland 3 & 4
7:30 p.m.	Dinner	Valley Room
8:30 p.m.	Speaker: The Honorable **Doug Walgren** U.S. Representative, 18th Congressional District Pittsburgh, Pennsylvania Chairman, Subcommittee Science Research and Technology Member, Health & Environmental Subcommittee "Animals in Research: Achieving a Balance"	

Abstracts

1

DEPLETION OF CENTRAL SEROTONIN STORES EXACERBATES THE DELETERIOUS EFFECTS OF HEMORRHAGE. J.A. Spath, Jr. and P.S. Blum*. Department of Physiology, Thomas Jefferson University, Philadelphia, PA 19107.

Central serotonergic neurons impinging upon sympathetic preganglionic neurons have been implicated in the regulation of blood pressure. Using anesthetized cats we recorded the mean arterial blood pressure and splanchnic nerve activity before, during, and after hemorrhage. Ten cats were untreated, while five cats were given parachlorophenylalanine (p-CPA, 300 mg·kg^{-1}, i.p., 40 to 48 hours prior to hemorrhage) to deplete serotonin. All cats were hemorrhaged to 40 ± 2 mm Hg for one hour using a pressure-activated, servo-controlled pump. Thereafter, pressure regulation was stopped. After two hours of oligemia, the shed blood was reinfused, and the cats monitored for an additional two hours. The table gives the mean arterial blood pressures (mm Hg) of the two groups of cats:

	Control	Oligemia (90 min)	Post-oligemia (120 min)
Untreated	123 ± 8	32 ± 1	127 ± 9
Serotonin-depleted	133 ± 7	33 ± 2	74 ± 14*

*means ± SEM, $p<0.01$ vs untreated.

In cats with normal levels of serotonin, blood pressure was inversely correlated with nerve activity with oligemia ($r = -0.80$). In serotonin depleted cats there was a significantly less inverse correlation of these variables ($r = -0.46$, $p<0.05$). These results suggest that serotonergic neurons inhibit sympathetic outflow and that this inhibition is withdrawn early in oligemia.

2

TIME OF ADMINISTRATION DETERMINES THE EFFECTS OF PRAZOSIN IN HEMORRHAGIC SHOCK. G. JOHNSON, R. F. BOND. Oral Roberts University School of Medicine, Tulsa, OK 74171.

The purpose of this study was to evaluate the efficacy of prazosin when administered to rats during the compensatory vasoconstriction stage of hemorrhagic hypotension. The results of a previous study had shown that pretreatment with prazosin resulted in a prolongation of the decompensatory stage of hemorrhagic hypotension. In each experiment, two Sprague-Dawley rats were anesthetized and subjected to a modified Wigger's hemorrhagic shock protocol. Both rats were bled simultaneously to an AP of 60 and then 30 mm Hg. When the 30 mm Hg level was reached, one rat received a bolus dose of prazosin (5 mg/kg) and the other an equivolume amount of drug vehicle. Both were maintained at 30 mm Hg until they had taken back 20% of the maximum blood shed from the calibrated buret system, at which time the remainder of the blood was reinfused and pressures were monitored for two hours. The results indicated that there were no significant differences in shed blood volumes, compensatory times or decompensatory times. Significant differences between groups were observed in seven of the eight post-reinfusion pressure measurements with the prazosin group having the lower pressures. These results stand in marked contrast to a previous study in which prazosin had beneficial effects in hemorrhage with no differences in the post-reinfusion period. The efficacy of prazosin as a therapeutic agent in hemorrhage appears to be dependent upon the condition of the organism at the time of administration. (Supported by grants from the Tulsa Chapter of the AHA and Oral Roberts University research funds).

3

CHRONIC ETHANOLISM(ETOH) IMPAIRS CARDIOVASCULAR RESPONSE AFTER RESUSCITATION FROM HEMORRHAGIC SHOCK(HS). J.W. Horton, S. Taylor*. University of Texas Health Science Center at Dallas, Southwestern Medical School, Dallas, TX 75235.

Previous studies show a significant correlation between ETOH abuse and trauma. However, the protective or detrimental effect of ETOH in the trauma patient with HS is unclear. In this study, 20 dogs were fed a diet mixed with 3 gm/kg ETOH for 3 (Group I, N=5) and 9 (Group II, N=5) months; 10 dogs served as controls Blood ETOH

*Indicates nonmember of the Society.

levels 2-3 hrs after food consumption was 175-330 mg/100 ml. On experimental day, dogs were bled to a mean arterial blood pressure(MAP) of 35mmHg for 2 hrs. Resuscitation included shed blood and lactated Ringer's, 50 ml/kg. Chronic ETOH increased MAP and left ventricular end-diastolic pressure(LVEDP), but decreased cardiac output(CO), stroke work(SW), and pancreatic blood flow(PBF). Shock impaired cardiac function and regional blood flow to a similar extent in all dogs. After resuscitation, CO, SW, coronary blood flow(CBF), and PBF were significantly lower in the ETOH compared to control dogs. Our data indicate chronic ETOH impairs cardiac response to resuscitation from HS, likely due to pancreatic hypoperfusion.

	CONTROL			SHOCK			RESUSCITATION		
GROUPS	I	II	III	I	II	III	I	II	III
MAP,mmHg	133±4	146±6	117±5	35±2	35±1	35±1	137±6	130±4	132±8
CO,ml/min/kg	74±7	74±4	110±7	29±4	34±3	33±3	53±5	56±7	111±8
SW,gm/kg	1.3±.2	1.5±.2	1.6±.1	.09±.01	.09±.01	.08±.01	.9±.1	.8±.1	1.1±.1
LVEDP,mmHg	5.6±.6	6.6±.4	3.2±.6	1.4±.2	1.8±.4	1.4±.3	6.8±.2	8.4±.5	3.6±.4
CBF,ml/min/g	.9±.1	.9±.1	1.1±.2	1.4±.1	1.1±.1	.8±.1	1.3±.1	1.6±.1	1.8±.1
PBF,ml/min/g	.3±.06	.3±.01	.8±.02	.1±.01	.1±.01	.2±.01	.2±.04	.2±.03	.5±.06

4

RELOCATION OF NON-ALBUMIN PROTEINS AFTER ALBUMIN RESUSCITATION. R.W. SMITH,* A.M. LEDGERWOOD, C.E. LUCAS.* Wayne State University, Detroit, MI 48201.

Prior work showed that albumin (5%A) resuscitation lowered serum (S) globulin (G) and content (CPC) of fibrinogen (F), prothrombin (II), antithrombin III (AT III), and factor VIII (V III) compared to crystalloid (C) resuscitation. The effects of a lesser colloid load (1.25%A) on S and lymphatic (L) levels of non-albumin proteins (NAP) were studied in 40 splenectomized dogs (18-26 kg) subjected to reservoir shock (MAP = 60 torr/90 min, then 40 torr/30 min) and treated with (a) 20 ml/kg C or 1.25%A, (b) shed blood (c) 30 ml/kg C or 1.25%A, and (d) 250 ml autogenous bank blood. Fifty ml/kg of C or 1.25%A was infused daily for three days. Preshock, post-therapy, and daily S levels of G and CPC were measured; L levels for thoracic duct (TD) and skin (Sk) were measured on day three. 1.25%A resuscitation caused a reduction in serum NAP which was significant for F on day three (292 mg vs 402 mg), V III on days one and two (111 and 84%Act vs 169 and 124%Act), and II on day two (99%Act vs 111%Act). TD lymph levels of G and CPC parallelled S changes. In contrast, Sk lymph NAP rose significantly with 1.25%A compared to C resuscitation (Table).

Sk L	α_1G (g/dl)	α_2G (g/dl)	β G (g/dl)	δ G (g/dl)	F (mg/dl)	II (%Act)	V III (%Act)	AT III (sec)
1.25%A	.023±.01	.11±.05*	.11±.05*	.26±.12*	<40	23±10*	41±27*	14±11*
C	.017±.01	.04±.03	.04±.02	.09±.05	<40	12±6	17±12	6±7

*Indicates $p<0.05$ by unpaired student "t" test on day three studies

These findings suggest that 1.25%A resuscitation causes an extravascular relocation of NAP due to an oncotic homeostatic factor rather than decreased NAP production.

5

PREDICTION OF SURVIVAL AFTER VOLUME CONTROLLED HEMORRHAGIC SHOCK (HS) IN AWAKE RATS. M.J. McGlew, P. Safar, P. Stremple. Resuscitation Research Center and Department of Anesthesiology, University of Pittsburgh, Pittsburgh, PA 15260.

A volume controlled model of acute HS in awake Sprague-Dawley rats was developed in order to determine the shed blood volumes (SBV)/100g body weight (BW) that, without reinfusion, would result in both high and low mortality models 24h after hemorrhage (H). Following overnight recovery from femoral artery cannulation, unanesthetized, unrestrained rats were bled over 20 minutes a precalculated, randomly assigned SBV. Mean arterial blood pressure (MAP) was measured during H. No resuscitation was given; access to food and water was allowed starting 2h after H. Results Mortality at 24h correlated closely with SBV. A SBV of 2.50 cc/100g resulted in death of 6/23 rats (LD26), 2.75 in 13/24 (LD57), 3.00 in 20/25 (LD80) and 3.50 in 5/5 (LD100). Regression analysis showed that mortality with respect to SBV/100g could be calculated according to the formula % mortality = 72 (vol % BW) - 145 with a high degree of confidence ($r=.96$, $p<.05$). Mean MAPs of survivors and nonsurvivors during H were not significantly different for each SBV. MAPs during H varied considerably, but were not good predictors of survival. Conclusions. This HS rat model is useful in monitoring physiologic responses to H without the complications of anesthesia or restraint. The regression line relating mortality to SBV makes prediction of survival following H convenient and reliable. The LD80 model, with longer-term monitoring and observation

of dying pattern, will be used to evaluate protective and resuscitative potential, and the LD26 model for evaluating deleterious effects of adjunctive insults (e.g., trauma and sepsis).

6

QUANTITATIVE MORPHOMETRY OF MYOCARDIAL LESIONS IN SHOCK. Bill B. Curtis, II*, Jureta W. Horton, L. Max Buja*. Univ. of Texas Health Science Center at Dallas, Dallas, TX and Texas Tech Health Science Center School of Medicine, Lubbock, TX.

Previous studies show that hypovolemic shock damages the heart, resulting in myocardial lesions. Whether these lesions are reversible after volume replacement remains conjectural. In this study, 12 dogs were bled to a mean arterial blood pressure of 30mmHg and hypotension maintained for 2 hrs; 7 dogs were sacrificed without resuscitation; in 5 dogs, shed blood was returned and lactated Ringer's solution infused,50 ml/kg. Myocardium from shocked(S) and shock-treated(ST) dogs were compared to control non-shocked(NS) dogs. For light microscopy, posterior papillary muscles(PPM) were formalized, embedded in paraffin, stained with hematoxylin-eosin. A point count sampling technique was used to determine the mean percent of PPM involved with lesions and morphometric analysis was determined by the square lattice test probe method based upon the counting of points separated into 25 grids of 40 points/grid. The point ratio was obtained by the number of points falling on lesions divided by total number of test points. Lesions were graded 0-4, based on changes in myofibrils. A grade of 0=normal myocytes, 1=focal stretching or hypercontraction; 2=focal disruption; 3=diffuse hypercontraction; 4=diffuse disruption (contraction bands). NS dogs did not develop lesions. The major finding was the apperance of lesions in both groups of S dogs. The mean percent of PPM involvement was similar in the S (0.5%) and ST dogs (0.34%). The percent of type 3 and 4 lesions was 14.3 and 8 times, respectively, greater in the S when compared to ST (p=0.01). Our data indicate that shock-induced myocardial damage is reversible with crystalloid volume replacement.

7

RESPONSE OF HYPERTROPHIC HEART MYOCARDIAL GLYCOGEN TO GIK AND HYPOVOLEMIC SHOCK. C. WITTNICH*, R.C.-J. CHIU. Montreal Gen. Hosp.& McGill University, Mtl. P.Q.H3G1A4.

Glucose-insulin-potassium (GIK) given during myocardial ischemia or anoxemia results in improved myocardial function and augments energy reserves of myocardial glycogen(MG). As many patients with heart disease also have myocardial hypertrophy, our purpose was to examine whether similar elevations in MG can occur in hypertrophic hearts with GIK administration and study the effect of hypovolemic shock on those MG levels. Mongrel dogs (n=5) with myocardial hypertrophy induced by prior femoral arteriovenous shunts underwent a sternotomy for serial myocardial biopsies of the left(LV) and right(RV) ventricles. Blood samples were drawn for serum free fatty acids (FFA), glucose and insulin measurements. Control samples were followed by GIK infusion (14.5ml/kg/hr) for 2 hr. After post-infusion samples, the dogs were subjected to 2 hr of hypovolemic shock (Mean Arterial Pressure=40mmhg) prior to final sampling.

	MG(LV) (g%)	MG(RV)	FFA(mEq/l)	Glucose(mg%)	Insulin(μU/mg)
Control	.45±.10	.43±.08	.20±.10	102.56±16.73	41.76±46.16
Post GIK	.57±.17	.60±.09	.05±.01	203.79±22.36	990.38±286.22
Post Shock	.76±.09	1.61±.73	.38±.43	137.20±58.67	81.19±28.81

Values are expressed as Mean±SD.

These results indicate that the hypertrophic hearts can indeed respond to GIK infusion by increasing MG in both the RV and LV, as does the normal heart. These hearts then submitted to hypovolemic shock show a further elevation of MG. The elevated insulin levels post GIK results in supression of FFA. Thus GIK administration may have a sparing effect on energy stores of the heart during hypovolemic shock which could have clinical implications in the treatment of patients with hypertrophic myocardium.

8

CARDIAC ENERGY METABOLISM IN FOUR PHASES OF HEMORRHAGIC SHOCK. F.J. Pearce, R.J. Connett and W.R. Drucker. University of Rochester School of Medicine and Dentistry, Rochester, NY 14642.

Controversy continues regarding the contribution of the heart to the loss of physiologic compensation that develops during persisting hypovolemia. The present study was designed to evaluate the possibility that some of the discrepant results might be explained by the different phases of shock studied. Twenty rats were bled according to a modified Wigger's shock protocol in which the mean arterial blood pressure was maintained at 40 mmHg. Tissue high energy phosphates (HEP), free creatine (Cr), glycogen (GLY), lactate (LAC), pyruvate (PYR) and glucose-6-phosphate (G-6-P) levels were measured in four phases of hemorrhagic shock which were defined according to pre-determined levels of the fraction of maximal blood loss. The four phases were I) early compensatory (50% of projected maximum blood volume removed), II) maximal compensatory (peak of blood volume removal), III) early decompensatory (20% of shed volume reinfused) and IV) late decompensatory (75% of shed volume reinfused). The results showed significant increases in tissue levels of LAC and PYR in all phases of shock. The changes in tissue LAC paralleled the plasma level until phase IV when the tissue LAC change far exceeded the plasma change. Glycogen and G-6-P levels were increased twofold in phases II and III. In phase IV, G-6-P levels were 5-fold greater than control and GLY levels fell back to control levels. There were no significant changes in tissue HEPs until phase IV when ATP and CrP were decreased by 20% and 40%, respectively. These results indicate that although significant metabolic alterations occur in the early phases of hemorrhagic shock, cardiac energy balance is not affected until late in the decompensatory stage.

9

THE Td MODEL FOR STUDYING HEMORRHAGE IN RATS: EFFECT OF SURGERY OR STARVATION ON TOLERANCE TO BLOOD LOSS. ROBERT E. BURR*, DANLEY F. BROWN AND SHARON R. VELEZ*. Combat Casualty Care, Letterman Army Institute of Research, San Francisco, CA 94129.

Previous studies from our laboratory have shown that the time of death (Td) during a fixed rate hemorrhage in rats is highly reproducible. Bleeding, via a chronic left carotid catheter at a rate of 0.1% of body weight per minute in control rats, causes death in 54.4 ± 3.3 (SD) minutes at a mean blood volume removed (BVR) of 5.2 ± 0.3 ml per 100 grams of body weight. The small coefficient of variation and susceptibility of the results to parametric analysis have led us to examine the utility of this Td model for studies of factors that mitigate hemorrhagic shock in rats. Using 10 animals per group, we have found a significant difference in Td and BVR between bleeding the rats 1 day or 3 days after left carotid catheter implantation surgery ($p < 0.0001$ for both Td and BVR). In a second experiment, three groups of rats (n=10 per group) were studied. Group 1 were non-fasted controls, Group 2 were fasted 24 hours and Group 3 were fasted 48 hours. All animals had free access to water. Td and BVR were not different between Groups 1 and 2; but Group 3 was significantly different from both Groups 1 and 2 ($p < 0.0001$ for both Td and BVR). These data indicate that animals subjected to either surgical or nutritional stress are less tolerant of hemorrhage that non-stressed controls. Furthermore, the Td hemorrhage model can detect these differences in a sensitive and efficient fashion.

10

PHYSIOLOGIC EFFECTS OF PNEUMATIC ANTI-SHOCK GARMENT(PASG) APPLICATION. C.D. ZIPPE*, G.J.SLOTMAN, K.W.BURCHARD, and D.S.GANN. R.I. Hospital, Dept Surgery, Providence,R.I.

PASG compression is thought to compensate for hypotension by augmenting venous return(VR) or cardiac output(CO)and/or increasing systemic vascular resistance(SVR). The purpose of this study was:1)to correlate PASG pressure (PASGP)with cardiovascular responses and 2)to determine the magnitude, if any, of the autotransfusion effect. Eleven dogs were chronically prepared with thoracic arterial and Swan-Ganz catheters. Five normovolemic dogs received progressive increases in PASGP every 10 min and cardiovascular measurements were done.

PASGP(mmHg)	0	21±1	49±4	75±8	96±8
MAP (mmHg)	123±2	138±4*	147±4*	149±5*	155±6*
CVP (mmHg)	3.2±1.0	3.9±1.5	4.1±1.5	4.1±1.5	4.1±1.4
CI (lmin/m2)	3.09±.08	3.21±.12	2.95±.10	2.68±.15*	2.57±.28*
SVR (dyne-sec/cm5)	3.89±.12	4.21±.25	4.84±.23*	5.49±.39*	6.03±.15*

(*P<0.05 from baseline in all tables, data are means ± std. error.)

PASGP above 49mm Hg resulted in elevated MAP, diminished CI, and elevated SVR. At low PASGP, MAP elevations appeared to be influenced by augmentation of VR and CI. From this we concluded that the maximal autotransfusion effect would occur at PASGP<49mmHg. Subsequently, six dogs were hemorrhaged 12% of blood volume(avg 120 cc)

on separate days with and without PASG, using PASGP of 30mm Hg. Results before (C) and 1 minute post hemorrhage(H) include:

	MAP		CVP		CI		SVRI	
12%	C	H	C	H	C	H	C	H
NO PASG	145±8	147±9	0.5±.5	0.1±.1	3.01±.21	2.31±.13*	4.52±.38	6.28±.24*
PASG	128±6	128±8	1.1±1.0	1.2±.6	2.73±.06	2.37±.12*	4.47±.13	5.51±.18*

Low PASGP has no significant affect on MAP, CVP, CI, or SVR. Thus, if low PASGP results in an autotransfusion effect it is less than a 12% hemorrhage.

11

VOLUME CONTROLLED HEMORRHAGIC SHOCK (HS) CEREBRAL VIABILITY MODEL IN ANESTHETIZED RATS. E.L. Cerchiari, P. Safar, R. Sclabassi*, A. Lanier. Resuscitation Research Center & Departments of Anesthesiology & Neurosurgery*, University of Pittsburgh, Pittsburgh, PA 15260.

A short-term volume-controlled model of acute HS and reperfusion was developed in lightly anesthetized rats. It leaves individual responses to hemorrhage unaltered and allows the study of cerebral function and viability. Method: 40 Sprague-Dawley rats underwent tracheotomy, ventilation, cannulation of vessels, and insertion of 4 skull screws for electrocorticogram (ECoG) recording. Anesthesia was with N2O/O2 50/50% - fentanyl 20 μg/kg plus 10 μg/kg per h infusion. We studied 4 groups with random assignment: (1) sham anesthesia and surgery without bleeding; (2) shed blood volume (SBV) 2.0ml/100g rat; (3) SBV 2.5 ml/100g; and (4) SBV 3.0ml/100g. All rats were bled over 20 min, left hypovolemic for 30 min, then had the SBV reinfused over 20 min, and were observed for another 120 min. They were sacrificed by intracardiac paraformaldehyde for brain fixation and histologic examination. Results: All rats in group (1) survived. All in group (2) survived after transient moderate hypotension and acidemia. In group (3) 7/9 survived, wile 2/9 were resistant to reinfusion and died within 1h post-reinfusion. In group (4), the 4/9 survivors and 5/9 which died at the end of hypovolemia, developed hypotension and acidemia ($p<0.05$). There was marked variability in the MAP response to the same SBV, and in the MAP at which ECoG silenced. Conclusion: This model with SBV 3.0ml/100g is suitable for HS cerebral viability studies (ECoG spectrum, ICBF, brain CPK-BB, and brain lactate, confirmed by histopathologic damage scores).

12

EVIDENCE FOR INDEPENDENT CONTROL OF ADRENAL MEDULLARY AND CORTICAL BLOOD FLOW. M.J. Breslow, R.C. Koehler and R.J. Traystman. The Johns Hopkins Medical Institutions, Baltimore, MD 21205.

Despite the fact that the adrenal gland consists of two embryologically and functionally distinct secretory organs, prior studies of adrenal blood flow (Q) have not attempted to separate cortical and medullary flow. Morphologic data indicate that the vascular supply to these two regions are independent. This suggested the possibility of dissimilar regulatory mechanisms for medullary and cortical blood flow. To test this hypothesis we evaluated the radioactive microsphere technique for measurement of adrenal cortical and medullary Q. Preliminary studies showed that 15 μ spheres are almost completely entrapped (> 99%) in the adrenals, that injection of 4×10^6 spheres results in $\geq$ 400 spheres/sample without affecting subsequent Q determinations, and that streaming and skimming effects are neglible in these beds. We then studied the blood flow response of the adrenal medulla and cortex to hemorrhagic hypotension. Pentobarbital anesthetized dogs (n = 6, 23-31 kg) were hemorrhaged into a pressurized bottle system and Q was measured before and after hemorrhage. Systemic blood pressure decreased from 117 ± 3 (SE) to 67 ± 1 mm Hg with hemorrhage. Q to whole adrenal gland was unchanged with hemorrhage (167 ± 15 to 178 ± 14 ml/min/100gm). However Q to cortex decreased 32% (182 ± 17 to 138 ± 12) and Q to medulla increased fivefold (149 ± 13 to 745 ± 87). We conclude that adrenal cortical and medullary Q are controlled by independent regulatory mechanisms. The profound adrenal medullary vasodilation which occurs with hemorrhage may contribute to the release of epinephrine into the circulation, and thus be an important element of the early response to hypotension.

13

DEXAMETHASONE ALTERS PLASMA GLUCOSE AND LACTATE DYSHOMEOSTASIS DURING ENDOTOXICOSIS: POSSIBLE ROLE FOR DECREASED INSULIN SECRETION. M. R. YELICH, J. P. FILKINS. Dept. of Physiology, Loyola Univ. of Chicago, Stritch Sch. of Med., Maywood, IL 60153.

Dexamethasone (D) antagonizes hyperinsulinemia in the endotoxic (E) rat (*Circ. Shock* 9(2): 162, 1982) and thereby may ameliorate glucose (G) and lactate (L) dyshomeostasis of endotoxicosis. G and L were thus measured *in vivo* - simultaneously and sequentially - in anesthetized, fasted, endotoxic rats (3.3 mg/kg S. enteritidis LPS iv), with and without concurrent treatment with D (0.33 mg/kg iv). Glucose-induced insulin secretion also was evaluated using the *in vitro*, perfused pancreases from rats injected with LPS (16.7 mg/kg iv) with and without D three hrs prior. Stable levels of G (90 mg/dl) and L (0.8 mM) were obtained in control (C) rats during the 5 hr sampling period. In E rats, G increased to a maximum of 155 mg/dl at 90 min and then decreased to a hypoglycemic level of 52 mg/dl at 210 min. In E+D rats, G increased to 170 mg/dl at 90 min and remained above 112 mg/dl thereafter. In E rats, L increased steadily from C levels to hyperlactacidemic levels of 7 mM at 180 min, and declined to 5 mM by 300 min. In E+D rats, maximum L levels of 3.3mM occurred by 120 min and declined to 2 mM by 300 min. Significant insulin hypersecretion occurred from E pancreases compared to C (E:1845 μU/20 min ± 394 (SE(n=3) vs C: 608 ± 195 (4); $p \leq 0.05$). Insulin secretion from E+D pancreases was only moderately elevated above C (E+D:946 μU/20 min ± 562(4)). The *in vivo* results indicate that D prevented profound hypoglycemia and hyperlactacidemia in the endotoxic rat. D also antagonized E-induced insulin hypersecretion, which suggests that decreased insulin secretion may play a role in the amelioration of G and L dyshomeostasis. (Supported by NIH Grant GM 29619.)

14

INSULIN RESISTANCE IN ENDOTOXEMIA ESTIMATED BY A MODIFIED EUGLYCEMIC CLAMPING TECHNIQUE. E.JANE VALAS AND ROBERT J.ALTEVEER. HAHNEMANN UNIVERSITY, PHYSIOLOGY AND BIOPHYSICS, PHILADELPHIA, PA 19102.

Euglycemic (insulin) clamp experiments employ constant insulin infusions to obtain steady levels of insulinemia ([I]) while a glucose infusion rate (GIR) is adjusted to maintain euglycemia. Data sets of various levels of [I] and their matched GIR yield an S-shaped insulin dose-response curve (IDRC). Using mongrel dogs, we have maintained euglycemic clamps for 4+ hours. Within this time we are able to obtain control data, introduce various experimental variables, and collect data on their effect. This approach permits both a comparison of experimental values with controls obtained in the same experiment, and assessment of the transients immediately following the introduction of the experimental variable. Results show that during control periods the IDRC rises to a plateau of about 21 mg/kg.min GIR for [I] values > 1000 μU/ml, while, according to published reports, in humans the plateau occurs at 15 mg/kg.min GIR. After a sub-lethal dose (.25 mg/kg) of endotoxin, the IDRC shows a shift to the right and down. For insulin doses producing [I] between 80 μU/ml and 300 μU/ml a 30% decrease in GIR occurs. The IDRC plateau ([I] > 1000 μU/ml) is approximately 20% below control values. These shifts appear to be endotoxin-dose-related and imply insulin resistance. Although wound sites are known to increase non-insulin dependent glucose uptake, we conclude that after low doses of endotoxin, insulin-sensitive tissue shows a decrease in glucose uptake during hyperinsulinemia.

15

B[illegible]L AND INSULIN-STIMULATED 3-O-METHYL/GLUCOSE TRANSPORT IN THE SOLEUS MUSCLE OF BACTEREMIC RATS. M. WESTFALL*, M. KARLSTAD* and M. SAYEED. Dept. of Physiology, Loyola University of Chicago, Stritch School of Medicine, Maywood, IL 60153.

Many investigators find altered effects of insulin on glucose metabolism in various models of shock. To determine if the membrane glucose carrier in skeletal muscle is altered, basal and insulin-stimulated 3-O-methyl glucose (3MG) efflux was measured in the soleus muscle of bacteremic rats. Fed and fasted male Holtzman rats (80-100 g) were injected ip with Escherichia coli (5×10^{11}/kg). This dose of bacteria produced 55% lethality (at 12 hrs post-injection) in fed (N=45) and fasted (N=20) rats. Plasma glucose decreased from 8.1±0.2 (mM) in fed controls to 6.0±0.5 ($p<0.01$) in fed bacteremic rats, and from 6.7±0.5 in fasted controls to 5.0±0.5 ($p<0.05$) in

fasted bacteremic rats. To measure 3MG efflux, isolated rat soleus muscle was loaded with ^{14}C-3MG in Krebs-Ringer-bicarbonate (KRB) and then washed in non-radioactive media with or without insulin (10mU/ml). Efflux was calculated as fractional loss in units of %/minute. The efflux values were:

FED:	*Basal*	*Insulin*	FASTED:	*Basal*	*Insulin*
Control	2.01±0.20(11)	3.47±0.20^{a}(11)		2.29±0.18(10)	3.33±0.20^{a}(10)
E. coli	1.93±0.10^{b}(9)	3.77±0.33^{ab}(9)		2.40±0.19^{b}(9)	3.33±0.27^{ab}(9)

Values are Mean±SEM;()=N;a=p<0.05 as compared to basal;b=N.S. compared to controls. These data indicate no effect of bacteremia on the soleus muscle glucose carrier under basal conditions or after supramaximal insulin stimulation. Potential changes in the sensitivity of the carrier to submaximal insulin stimulation remains to be determined. (Supported by NIH GM 32288.)

16

INCREASED OXIDATION OF FAT AND KETONES BY MITOCHONDRIA IN HYPERDYNAMIC SEPSIS.
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Chronically catheterized rats were made septic by inoculations of E. coli, B. fragilis and S. aureus into a preformed subcutaneous abscess. All septic animals had intermittent E. coli bacteremia and significantly increased white cell counts (from control of 18.8±6x10^{3} to 31.6±4x10^{3}). Cardiac output, measured by thermodilution, increased to 146-436% of control. After 9-14 days of sepsis the rats were killed, skeletal muscle and liver collected for tissue carnitine determination and isolation of muscle and liver mitochondria and characterization of their fuel utilization functions. Control animals were catheterized and had noninfected abscesses. The following results were obtained: * p<0.05

	tissue carnitine (nmol/g)		mitochondrial activity (nmol/min/mg)			
	free	total	ΔO_2 pyr	ΔO_2 palmitate	ΔO_2βOH But	ΔLC-carnit.
control muscle	1085±68	1552±253	42±14	21±5.6	9±1.7	2.3±.3
septic muscle	961±72*	1347±177	62±15	100±30*	101±17*	5.3±1.8*
control liver	335±53	587±99	20±4	14±3.6	11±3	1.4±.3
septic liver	238±72	443±129	29±4.6	18±3.6	14±5	2.8±.8*

These results indicate 1) that skeletal muscle but not liver β-oxidation is highly activated, 2) long chain carnitine breakdown rates are elevated in both liver and muscle mitochondria, 3) tissue carnitine levels although reduced are not significantly below normal. The results suggest the dependence of skeletal muscle on lipid fuels in hyperdynamic sepsis. Supported by NIH grant GM 33267.

17

TITLE: ELEVATED RATE OF LIPOLYSIS IN BURN SUBJECTS. AUTHORS: E.J. PETERS,*, D.N. HERNDON, R.D. GOODENOUGH,* R.R. WOLFE, Shriners Burns Institute & The University of Texas Medical Branch, Galveston, TX 77550.

Nutritional support of burn victims is a crucial part of their medical care. It is known that burn victims are limited in their ability to utilize glucose as an energy source, but little is known about the role of lipids as an energy substrate. To address this question, the basal rate of lipolysis in 6 severely burned patients and 6 normal male volunteers was measured after an 8-12 hour fast. The burn subjects ranged in age from 8-43 years old and initially had been burned an average of 60% of their total body surface. Basal lipolysis was measured 13-56 days post burn by infusing stable isotopes (1-^{13}C-palmitate and d-5-glycerol) at a constant rate and measuring the resultant plasma enrichment of the isotopes by gas-chromatography mass-spectrometry. The 6 burn victims' basal rate of glycerol release was significantly ($p< 0.002$) greater than the controls' basal rate (5.78±1.20 μmole·kg^{-1}·min^{-1} vs 2.61±0.14 μmole·kg^{-1}·min^{-1} ($n = 14$), respectively). Likewise, the basal rate of free fatty acid (FFA) release in the 6 burn subjects was significantly ($p< 0.005$) elevated above the rate measured in 6 controls (15.6±2.0 μmole·kg^{-1}·min^{-1} vs 6.7±0.7 μmole·kg^{-1}·min^{-1} respectively). If the endogenous FFA were completely oxidized, it would provide approximately 175% of the total energy expenditure (measured by indirect calorimetry) in the burn subjects. Thus, lipolysis is dramatically stimulated in burn patients and the total energy requirements could theoretically be met by FFA oxidation alone. Therefore, little beneficial effect would be expected from the infusion of endogenous lipids for nutritional support in burn patients.

18

CHANGES OF CITRULLINE SYNTHESIS ACTIVITY IN SUBLETHAL BACTEREMIC RAT LIVER HOMOGENATES. H. AOYAMA*, F. HIRAI*, T. SATO, I.K. BEREZESKY*, AND B.F. TRUMP*. Department of Pathology, University of Maryland School of Medicine, Baltimore, MD 21201.

Our recent data suggested that despite no lethality and minimum organic damage, sublethal bacteremia induces altered liver mitochondrial (MT) activity, enhanced respiratory control ratio and citrulline synthesis (CIT-S) activity. For MT preparation, a relatively large amount of liver tissue is necessary; therefore, we developed a simpler and less time-consuming method of assaying citrulline synthesis in liver MT using liver homogenates. Spraque-Dawley rats (265-275 g) were fasted overnight. A sublethal dose of live E. coli (Serotype: O-18; 6.0 X 10^6 organisms/g BW) was injected through the tail vein; saline-injected rats were used as controls. At 6,9 and 12 hrs after treatment, rats were sacrificed, liver tissue homogenized, and CIT-S from ornithine and ammonium bicarbonate measured. Mortality rate was 0% at 12 hrs. Results, as shown below, suggest that sublethal bacteremia induces enhanced CIT-S, indicating hyper-mitochondria, and that this easily performed assay can be used as an indicator of mitochondrial function. (Supported by NIH GM32084.)

	6 HRS (n=4)	9 HRS (n=4)	12 HRS (n=4)	
CONTROL	0.674±0.060	0.766±0.064	0.66±0.045	MICROMOLES/g TISSUE/MIN
SUBLETHAL	1.562±0.166*	1.132±0.148	1.198±0.056*	*P<0.01

19

EFFECT OF ENDOTOXIN ON HEPATIC NITROGEN METABOLISM. L. Kilpatrick-Smith*, M. Yoder*, R. Polin* & M. Erecinska*(Introduced by: Ralph T. Geer). Depts. of Pharm. & Ped. University of Pennsylvania Medical School, Phila., PA 19104.

The effect of a sublethal dose of E. coli endotoxin (LPS) on hepatic nitrogen metabolism was examined in starved LPS-treated rats (E- 0.2mg/kg) and compared with appropriate controls (C- 5% dextrose in water). Plasma and freeze-clamped liver samples were taken at 1, 4 & 12 h after treatment. There were no significant differences in metabolic parameters investigated at 1 h post-treatment. However, progressive changes were observed at 4 & 12 h. In E as compared to C, plasma levels of ammonia (NH_4^+) rose by 48%, urea by 98%; these were accompanied by significant increases in concentrations of aspartate (asp), glutamate (glu), glutamine (gln) and alanine (ala). Hepatic urea rose by 67%, glu by 92%, gln by 55% and ala by 74% in E as compared to C. Liver NH_4^+ was somewhat elevated in E. The rates of urea and amino acid production and NH_4^+ utilization were measured in suspensions of hepatocytes (4 mg dry wt/ml) isolated from E and C rats 12 h post-treatment. At 37°C, with 1 mM oleate and 6 mM NH_4Cl, the rates of urea and gln synthesis by E hepatocytes increased to 300% as compared to C. However, while the $[NH_4^+]$ in C hepatocytes decreased by 43% after 45 min incubation, it only declined by 19% in E hepatocytes. When no NH_4^+ was added, the rate of urea synthesis was still greater in E by 34%. These data suggest that a sublethal dose of LPS stimulates hepatic protein breakdown. The consequent rise in the levels of amino acids and (possibly) NH_4^+ leads to augmented urea production. The increased plasma levels of NH_4^+ and urea could, therefore, be due to stimulated proteolysis rather than to impaired ureagenesis by the liver. Supported by Naval RC#N00014-84-K-0514 & NIH PO1-NS-17752-01.

20

RESPONSE OF LIVER GLYCOLYTIC INTERMEDIATES TO ENDOTOXIN IN THE POSTMATURE RAT. Robert E. Kuttner, Takao Sugai*, Ryuichi Denno*, William Schumer, Dept.Surgery, Univ. Health Sci./Chicago Med. Sch., No. Chicago, IL 60064.

Progressive decline during aging of the body's vital defensive functions against severe sepsis, trauma, and shock has been established. To ascertain if the pattern of metabolic response differed from younger rats, we investigated hepatic glycolytic activity in endotoxemic rats, 12-14 months old, a past prime maturity age manifesting reduced stress resistance and healing capacity. The rats were fasted for 18 h, IV injected with 2.0 mg/100 g BWt E. coli endotoxin (ET), etherized to take liver biopsies by liquid nitrogen cooled tongs, and killed at 3.0 and 4.5 h. Control liver glucose-6-phosphate (G6P) concentration was 295±17 nmole ± 1 S.D./g wet

tissue, a value (G6P) 50-100% higher than present and past G6P assays for 3-4 month old fasted rats (P<0.001). Phosphoenolpyruvate (PEP), was 205±17 nmole ± 1 S.D./g wet tissue, similar to young adult values. ET reduced G6P to 109±18 nmole and elevated PEP to 376±25 nmole/g wet tissue at 4.5 h (both changes P<0.001). Liver lactate rose 2.5-fold from control values of 1178±155 nmole/g (P<0.01). Same degree for G6P depletion was already evident at 3 h. Metabolite changes for postmature rats were similar to those of young rats. However, the amount of ET necessary for metabolic dyshomeostasis was much less. With young rats, 3 mg/100 g ET was LD_{50} in 48 h, while 2 mg ET in 12-14 month old rats killed 3 of 7 animals in 6 h. Decreased glucose-6-phosphatase activity in old rat livers may be responsible for higher G6P, and for diminished gluconeogenic potential in endotoxemic postmature animals. thus contributing to enhanced susceptibility to shock.(Aided by VA Med. Res. Serv.)

21

EFFECT OF GLUCOSE INFUSION ON GLYCOGEN REPLETION AND GLUCONEOGENESIS IN ENDOTOXIN-TREATED RATS. C.H. LANG, G.J. BAGBY, A.Z. BUDAY* AND J.J. SPITZER. Department of Physiology, LSU Medical Center, New Orleans, LA 70112.

This study evaluated the contribution of gluconeogenesis to hepatic and muscle glycogen repletion during glucose infusion in E. coli endotoxin (100 µg/100 g, LD 10) and saline (control) injected rats. Blood pressure decreased by 30% at 1.5 hrs post-endotoxin (ET) and returned to control levels by 4 hrs. Thereafter, ET and control (C) rats were infused with [6-^{3}H]-glucose in saline (C-S and ET-S groups) or glucose (230 µmmoles glucose/min/kg; C-G and ET-G groups) for up to 4 additional hrs. Compared to the C-S group, ET-S rats were normotensive, hyperglycemic, hyperlactacidemic and had an elevated rate of appearance (Ra) of glucose during this period. Glucose infusion increased plasma glucose concentration to a plateau of 16 mM in the C-G rats. The increase in plasma glucose of the ET-G group was similar during the first 2 hrs of glucose infusion, but thereafter increased to 29 mM at 4 hr. Glucose infusion increased lactate concentration to a similar degree in both groups (+1.6 mM). The calculated rate of total gluconeogenesis was elevated more than 200% in the ET-G rats compared to the C-G animals. Glucose infusion suppressed gluconeogenesis to a lesser extent in ET rats (46 ± 5%) compared to controls (81 ± 3%). Skeletal muscle from both groups repleted glycogen at similar rates. In contrast, hepatic glycogen repletion was decreased in ET-G by 73 and 60% at 2 and 4 hrs post-glucose. Thus, gluconeogenesis derived glucose-6-P is diverted from hepatic glycogen storage to hepatic glucose output in ET-treated rats during glucose infusion. (Supported by NIH GM32371).

22

Role of Hepatic Free Amino Acids as Gluconeogenic Precursors in Inflamation and Sepsis. J.H.SIEGEL, T.C.VARY*, B.PLACKO*, T.NAKATANI*, T.SATO, H.AOYAMA*. MIEMSS, Dept. Physiology and Pathology, University of Maryland, Baltimore,MD. 21202.

Gluconeogenesis is regulated both by substrate availability and hormonal stimulation. The relationship between plasma glucose and hepatic gluconegenic precursors was determined in an intra-abdominal abscess model. Plasma glucose levels were relatively unchanged in rats five days following an intraperitoneal introduction of a fecal-agar pellet of known bacterial flora which generated an abscess [sterile (I) or B. Fragilis + E. Coli abscess pellet (1.5ml) (LA)]. Control fed rats were not inoculated. The liver was frozen in situ and the tissue level of the gluconeogenic precursors, lactate and amino acids, were determined. Lactate levels were elevated in both I and LA, and the rise was greater in LA, than I. The pattern of hepatic amino acids, showed that tissue levels of threonine, serine, glycine, and alanine were significantly elevated ($p<.05$) in I and LA compared to control. Proline levels were also elevated but not significantly. In contrast, no alterations in the level of hepatic branched chain or aromatic amino acids were observed. Thus, precursors for glucose formation, both lacate and gluconeogenic amino acids are elevated in inflamation and chronic septic abscess models, while plasma glucose is relatively stable. These results suggest that in this model, the hepatic level of gluconeogenic substrates is probably not rate-limiting for glucose production in inflamation or sepsis. The accumulation of gluconegenic precursors may be related to the lack of proper hormonal stimulus for gluconeogenesis, or due to accelerated release of these gluconeogenic amino acids from non-hepatic tissues with increased hepatic uptake.

23

DEPRESSED ADRENOCORTICOSTEROID SECRETION IN NEONATAL ENDOTOXIN SHOCK.
LINDA WITEK-JANUSEK. Depts. of Physiology and Maternal Child Nursing, Loyola University of Chicago, Maywood, IL 60153.

The neonatal rat is highly sensitive to the lethal effects of bacterial endotoxin. Since the secretion of adrenocorticosteroids is essential to resisting endotoxin, plasma corticosterone was measured in the 10 day old endotoxic rat pup. Pups were removed from their dams and randomly injected with either saline or 0.1 mg/kg S. enteritidis endotoxin. This dose is 90% lethal at 24 hr. Pups were placed in an incubator (39°C) and at 4 hr post injection trunk blood and adrenal glands were collected. Corticosterone was measured using a competitive protein binding assay. Endotoxic pups were significantly ($p<.01$) hypoglycemic (35±8 vs. 73±4 mg/dl). Their adrenal glands were hemorraghic but gland weights did not differ from that of control pups (27.8±2.6 vs. 24.5±1.9 mg/100 gm). Plasma corticosterone increased 66% in endotoxic pups compared to control pups (8.2±0.9 vs 2.8±0.3 µg/dl; $p<.01$); however, this increment is markedly less than the adult rat's plasma corticosterone response to endotoxin (63.3±9 vs. 14.3±6.4 µg/dl; $p<.01$; *Endocrinology 102*: 947, 1978). Since adrenocorticotrophic hormone (ACTH) injection (Cortrosyn IP; 0.1 mU/gm) to the 10 day old pup did not elevate plasma corticosterone above that of saline injected control pups (1.9±0.1 vs. 1.0±0.1 µg/dl; $p>.05$), the lack of an adult pattern of adrenocortical secretion appears to be due in part to a decreased adrenocortical sensitivity to ACTH. Thus, the susceptibility of the neonatal rat pup to endotoxin may be due to its depressed ability to secrete adrenocorticosteroids.
(Loyola U. BRSG USPH RR05368).

24

PERSISTING GLUCOCORTICOID HORMONE PROTECTION IN ENDOTOXIC RATS WITH INHIBITED HEPATIC GLUCONEOGENESIS.
Takao Sugai*, Robert E. Kuttner, William Schumer, Dept. Surgery, Univ. Health Sci./Chicago Med Sch., N. Chicago, IL 60064.

Prolonged survival of endotoxic and septic animals by glucocorticoid hormones may be attributable to either one or two possible mechanisms; 1) stabilization of membranes preventing the leaky capillary syndrome, or 2) averting terminal hypoglycemia by accelerating gluconeogenesis. These mechanisms were studied by blocking gluconeogenesis with L-tryptophan (TY) in endotoxin (ET) treated rats simultaneously injected with dexamethasone (DMS). TY, 50 mg/100 g rat weight, was injected IP. TY rapidly forms quinolinic acid blocking glucose synthesis in the liver. E. coli ET, 3 mg/100 g (LD_{50} in 48 h) plus DMS, 1 mg/100 g, both IV, were given to fasted adult male rats (180-220 g). At 6 h, the ET+TY group had one survivor (N=16) while with DMS, 11 of 29 rats survived (P=0.02). However, only 3 rats survived 24 h even with DMS. The intensified lethality with TY showed the vital gluconeogenic capacity in endotoxemic rats. The ability of DMS to delay death suggests a nonmetabolic mechanism. In parallel experiments, freeze-clamp liver biopsies at 5 h were assayed for glycolytic intermediates. Phosphoenolpyruvate and glucose 6-phosphate were 58±24 nmole and 69±19 nmole ±1 S.D./g wet liver in the ET+TY group (N=7). With DMS treatment, these concentrations were 79±31 and 78±17 nmole/g respectively, not significantly different (N=9). Control values in the ET alone group were 222±79 and 137±31 nmole/g (N=8). DMS's failure in the ET+TY rats to replenish key intermediates shows that the TY blockade of gluconeogenesis was not overcome and that DMS acted by a second nonmetabolic mechanism. (Aided by VA Medical Research Service)

25

THE USE OF VENOVENOUS EXTRACORPOREAL MEMBRANE OXYGENATION IN SHEEP RECEIVING SEVERE SMOKE INHALATION INJURY.
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Smoke inhalation injury now represents the most frequent cause of death in burn patients, accounting for 20-80% of overall mortality. We have studied the use of extracorporeal membrane oxygenation (ECMO) to support sheep which have received lethal pulmonary smoke damage. Sheep (n=16) received inhalation injury induced by insufflation with smoke derived from burning cotton. The smoke was delivered with a bee smoker. The treatment group, those placed on ECMO (n=6), were heparinized and placed on a venovenous perfusion circuit consisting of a roller pump, membrane

oxygenator, and heat exchanger at the time of injury. Gas flow (100% oxygen) to the oxygenator was 10 L/min. Flow rate in the circuit approximated 20-25% of cardiac output. They remained on partial venovenous bypass until the termination of the experiment 96 hours post injury. All animals in the treatment group survived. The control sheep (n=7) received inhalation injury alone and had a 100% mortality (p=0.0015 ECMO vs. cont.) Ventilatory management of treatment and control groups was identical. At present we are studying a third group (n=3) comprised of animals receiving inhalation injury with systemic heparinization. This group is not placed on ECMO. To date this group has a 2/3 early mortality. Data must be collected from additional animals before statistically significant conclusions pertaining to this group may be drawn. From these studies we believe that venovenous ECMO may be a valuable adjunctive treatment modality in severe inhalation injury.

26

ACUTE CARDIOPULMONARY EFFECTS OF SMOOTH OR ROUGH STRAIN LIPOPOLYSACCHARIDES IN THE CONSCIOUS SHEEP. Gary Jesmok,* Julian Borgia* (Introduced by: William Schumer). Travenol Laboratories, Morton Grove, IL 60053.

We have characterized the acute cardiopulmonary response following smooth (055:B5) or rough strain (J5 and Re595) lipopolysaccharide (LPS) challenge (1 ug/kg over 10 minutes) in conscious sheep. LPS induced a pronounced increase in mean pulmonary artery pressure within 10 minutes with a more rapid onset with the rough strains. Arterial PO_2 fell between 20-30 torr following LPS infusion and the onset of hypoxemia was more rapid with the rough strains. Cardiac output and stroke volume declined following LPS challenge while systemic vascular resistance increased. Heart rate exhibited a biphasic pattern, with an initial increase followed by a return to baseline and a subsequent elevation. The arterial-venous oxygen content difference and the calculated oxygen consumption (Fick principle) increased following LPS infusion. The arterial hematocrit increased (30-35%) following LPS challenge. Coincident with these cardiopulmonary events, the arterial neutrophil count declined precipitously, > 98% over 30 minutes, with a more rapid decline with the rough strains. Results would indicate that the lipid A portion of the LPS is the active moiety of LPS and that neutrophil sequestration and/or activation is intimately associated with the induction of the acute cardiopulmonary effects of LPS administration.

27

PLASMA EXCHANGE IN EARLY FLUID RESUSCITATION FAILURE OF THERMAL INJURIES. C.W. CLINE*, I.W. GOLDFARB, R.SCHNARRS*, H.SLATER*, J.C. GAISFORD*, H.E. JACOB*, J.B. HANRAHAN*. The Western Pennsylvania Hospital, Pittsburgh, PA 15224.

To evaluate the response to plasma exchange (PE) in thermal injuries who did not respond in the anticipated manner to conventional fluid resuscitation, a protocol was designed including individuals 15 to 70 years of age with total body surface burns of greater than 30 percent, with or without inhalation injury. Materials include the Haemonetics V-50 apheresis machine in double arm continuous fashion utilizing type specific fresh frozen plasma replacing 1.5 times the calculated plasma volume. Appropriate data was obtained prior to and after each PE via invasive and non-invasive means. Parameters studied included Cardiac Index (CI), Stroke Index (SI), cardiac chamber pressures, systemic vascular resistance (SVR), pulmonary vascular resistance (PVR), and pulmonary dynamics. Five patients thus far studied are male, three with associated ARDS. All benefitted from PE with increased CI (55% ± 37%, $p < 0.01$) and SI (56% ± 47%, $p < 0.01$) lessened ventilatory support requirements, reduced volume infusion with maintenance of adequate urinary output, and decrease in PVR and SVR. We conclude that PE may benefit those patients with thermal injury who fail to respond in the anticipated manner to conventional fluid resuscitation.

28

EFFECTS OF SMOKE INHALATION ON AIRWAY BLOOD FLOW AND EDEMA FORMATION. DN Herndon, DL Traber, HA Linares, GC Kramer, Shriners Burns Inst., Galveston, and Univ. CA, Davis.

Smoke inhalation (SI) causes a major airway lesion which can progress to inflammation and edema of lung parenchyma. We measured distribution of bronchial blood flow and pulmonary pathology after SI in 6 sheep. A baseline measurement of bronchial blood flow was made with a double microsphere technique (Fed Proc 43:921). The next day each sheep was anesthetized and insufflated with smoke until carbon monoxide saturation was 20%. Animals recovered from anesthesia in 30 min. Respiratory distress, viz. labored breathing, coughing and decreased arterial PO_2 was not apparent before 6 hrs. A second blood flow determination was made after PO_2 fell to 60 mm Hg at 8 to 30 hrs after SI. Animals were sacrificed and samples taken for histology and gamma counting.

	Blood Flow (ml/min per 100 g) ± SE			
	Upper Trachea	Lower Trachea	Stem Bronchi	Lung Parenchyma
Pre-Injury	11.2 ± 3.1	14.4 ± 6.3	7.7 ± 3.2	40.0 ± 6.3
Smoke Inhalation	94.5 ± 15.0	169.6 ± 52.0	104.5 ± 46.0	37.4 ± 7.8

Blood flow in large airways was 10-20x pre-injury levels, while cardiac output was not changed. The most prominent histopathological lesion was in the trachea and stem bronchi which exhibited marked congestion, edema and polymorphonuclear infiltration. Pulmonary parenchyma surrounding injured airways exhibited varying degrees of congestion, interstitial and alveolar edema and neutrophil infiltration. Marked hyperemia of the bronchial circulation after SI contributes to airway edema and formation of tracheobronchial exudate and may participate in the development of interstitial and alveolar edema.

29

PULMONARY DYSFUNCTION FROM THE ENDOTOXIN STIMULATED BURN WOUND: PREVENTION BY TOPICAL IBUPROFEN. A. KATZ*, P.V. RYAN*, C. LALONDE*, K. WEST*, R.H. DEMLING. Longwood Area Trauma Center, Harvard Medical School, Boston, MA 02115.

Respiratory distress in the absence of bacteremia or endotoxin is a common finding after burn injury. Endotoxin 2 µg/kg injected beneath a full thickness eschar of 1000 cm^2 in adult sheep (n=8), results in an increase in pulmonary artery pressure (Ppa) from 18±3 to 28±5 mm Hg and a decrease in arterial oxygen content from 90±4 to 73±5 torr. Venous blood thromboxane, measured as TxB_2, increased from 220±100 to 380±130 pg/ml while levels seen in lymph draining the burn wound increased from 450±150 to over 2000 pg/ml. Levels in lung lymph were significantly less than that in burn lymph, indicating the wound to be the source. Topical ibuprofen in a 5% concentration was applied to the burn beginning 24 hrs prior to the subeschar endotoxin (n=7). Mean ibuprofen levels measured in lymph after application and prior to endotoxin was 1.9±5 mcg/ml while levels in plasma were below the minimal detectable concentration of 0.2 mcg/ml. Mean Ppa after endotoxin only increased to 21±4 mm Hg and PaO_2 decreased to 85±5. We conclude that, 1) thromboxane released from the endotoxin stiumulated burn wound can produce pulmonary hypertension and decreased PaO_2, and 2) topical ibuprofen can penetrate the burn tissue and attenuate the endotoxin response.

30

INCREASED PERMEABILITY IN LUNGVESSELS IN SEPTIC SHEEP. L.SMITH, S. ANDREASSON, B. RISBERG. Dept. of Surgery I, University of Goteborg, Sweden.

Permeability in capillaries can be characterized by the osmotic reflection coefficient (σ). In the chronic lung lymph drainage preparation in sheep we studied effects from septicemia on pulmonary microvascular exchange. Following infusion of live E. Coli we determined σ. To measure σ we increased left atrial pressure (P_{la}) by an inflatable balloon. σ was measured during a baseline elevation of P_{la} and following E. Coli infusion with maintained inflated balloon. Permeability surface area product (PS) and σ for total protein were calculated by iterative curve-fitting using the non-linear flux equation. Data were compared to σ achieved at high lymph flows when the lymph to plasma ratio (L/P) for total protein was filtration independant.

In this experiment we used 7 chronically instrumented sheep. The lunglymph fistula and left atrial balloon catheter were positioned 4 days prior to experiment. Catheters for hemodynamic monitoring (pulmonary artery, central vein and carotid artery) were placed on day of experiment. Following baseline re-

adings P_{la} was successively increased. Two readings of constant L/P at different high lymph flows were considered indicative of a filtration independant state. Live E. Coli (10^9/Kg BW) was then infused during 20 min with the balloon still inflated. After 4 hours of sepsis a new filtration independant state was reached.

During baseline σ was found to be 0.73 but after sepsis it was significantly decreased to 0.61. There was a good agreement between calculated and observed σ PS was not affected by sepsis.

We conclude from these experiments that sepsis increased the pulmonary microvascular permeability as indicated by reduction in σ.

31

CARDIOPULMONARY RESPONSE TO SEQUENTIAL DOSAGES OF ENDOTOXIN. D.L. TRABER, M. BROWN*, D.N. HERNDON, L.D. TRABER*. The University of Texas Medical Branch and Shriners Burns Institute, Galveston, TX 77550.

We have previously demonstrated that the administration of a 0.75 ug dosage of endotoxin (LPS) will produce a fall in cardiac output (CO) 3 hr after its administration but at 6 hr the CO begins a sustained elevation which remains in effect for twenty-four hours. The present study was accomplished to determine the effects of a second dosage of LPS administered at the beginning of the hyperdynamic response. The studies were accomplished on six sheep with chronic lung lymph fistulas and cardiopulmonary catheters. Their control CO's were 5.1±0.5 l/min. Six hours following LPS the outputs had risen to 6.2±0.5 l/min. At this time a second dosage of LPS was administered. The CO continued its rise until 8 hrs. at which time it showed a 1 liter dip (5.6±0.5 l/min) to a value still above control levels. The increase in CO was again significantly elevated at 10 hrs. and was still 700 ml above the control level 24 hr after LPS. During the time of the elevated CO there was a marked elevation in protein-rich lung lymph. The pulmonary artery pressure although showing a large response (18±1 to 35±6) with the first dosage of LPS was only minimal changed with the second (24±3 to 28±3). In like fashion the second blood neutrophil count fall (2916±798 to 1588±330) was not as great as that seen initially (2607 ± 774 to 820±260). The hyperdynamic and pulmonary responses to LPS are either unchanged or increased after administration of a second dosage of endotoxin administered during the response.

32

LEUKOPENIA MAY EXACERBATE THE INCREASED EXTRA VASCULAR LUNG WATER (EVLW) FOLLOWING HIGH DOSE ENDOTOXIN. R. WINN, R. MAUNDER* AND J. HARLAN*:(Intro by: R. Maier). Univ of Wash, Seattle, WA 98195.

It has been reported that granulocyte depletion prevents increased vascular permeability after low dose endotoxin infusion (0.5 µg/kg). We wished to determine the role of PMNs in the development of pulmonary edema following high dose endotoxin infusion (5 µg/kg). Six goats were prepared with pulmonary and systemic arterial catheters for measurement of pressures and thermal dilution cardiac output. Three animals were given 1.5 mg/kg nitrogen mustard approximately 3 days prior to the experiments. All 3 had a systemic leukocyte counts of less than 300 cells per mm^3 when they were studied. After assuring a stable baseline, a 1 hr endotoxin infusion was started (5 g/kg total dose). Animals were killed at 5 to 6 hrs or when their clinical condition dictated. All leukopenic goats were killed by 1.5 hours, whereas, all of the controls survived. Lungs were removed to determine EVLW. Values for each goat are given below (normal 6.6ml/kg).

Control EVLW (ml/kg)	Leukopenic EVLW (ml/kg)
>10	11.4
11.6	19.3
10.6	19.8

Vascular pressures and cardiac output changes were similar between the two groups. Pulmonary artery pressure increased sharply from approx 20 cmH_2O to a peak of approx 50 cmH_2O. Pulmonary wedge pressure was similar increasing from 7 cmH_2O to approx 21 cmH_2O. Aortic pressure decreased from 90 torr to 60 torr in controls and to 30 torr in leukopenics. Cardiac output declined to approximately 30% of baseline in both groups. We conclude that leukopenia due to nitrogen mustard does not protect and may, in fact, exacerbate pulmonary edema formation following high dose endotoxin infusion.

33

CLEARANCE OF BLOOD BORNE PSEUDOMONAS BY THE LUNG DURING SEPSIS. D.J. DEHRING,* R. FADER,* L.D. TRABER,* J. UNBEHAGEN,* D.L. TRABER . The University of Texas Medical Branch and Shriners Burns Institute, Galveston, TX 77550

Quantitative-bacterial cultures of the pulmonary arterial blood have shown that live Pseudomonas aeruginosa organisms are removed from the blood in the pulmonary circulation at a high rate in pigs (75%) and to a lesser extent in dogs (10%) and baboons (5%). We did serial quantitative bacterial cultures and white blood counts (WBC's) on pulmonary artery (PA) and systemic arterial (A) blood in the ovine chronic lung lymph preparation. The animals (n=5) received a solution of 1.15±0.34 x 10^8 (mean±SEM) washed Ps.aeruginosa cells over a 1-2 hr period to produce both the pulmonary hypertensive and permeability phases of lung edema. No bacterial growth was present at baseline. PA bacterial concentrations averaged $2x10^3$ during organism infusion, and increased 10 fold 15 minutes after bacterial infusion stopped and then decreased over the next four hrs. Bacterial concentrations in A were 10-20 fold less than PA. Clearance of bacteria across the lung was 80-95%. A severe persistent neutropenia occurred after 1 hour of pseudomonas infusion with a clearance of 10-40% across the lungs. Ovine and porcine septic models develop respiratory failure and clear a large percentage of bacteria in their lungs. The bacteria may be embolized in the PA circulation or phagocytized by WBC's or other phagocytic cells in the circulation of the lung.

34

THE EFFECT OF HEMORRHAGIC SHOCK ON ENDOTOXIN-INDUCED LUNG INJURY. M.M. KRAUSZ, E. MORIEL, E. KORONADO, A. PEREL. Hadassah University Hospital, Jerusalem Israel,91120.

We have previously observed that hemorrhagic shock prevents increased pulmonary shunting and permeability in response to complement activation by zymosan. The response to E. coli endotoxin during hemorrhagic shock was studied in 13 unanesthetized sheep with chronic lung lymph fistulas. Seven were given E. coli endotoxin (1µg/kg) alone and six were given the same dose after 2 h of hemorrhagic hypotension with a mean arterial pressure of 50 mmHg. A characteristic two phase lung response was seen: an initial pulmonary hypertension phase with a rise in mean pulmonary artery pressure (Ppa) to 49.7 ($p<0.001$), a rise in pulmonary microvascular pressure (Pmv) to 23.7 ($p<0.005$), and a second permeability phase with an increase in pulmonary lymph flow (Ql) to 28.6 ml/h ($p<0.01$) after one h and 36.9 ml/h ($p<0.005$) after 2 h. Pulmonary protein clearence (L/P·Ql) was 18.5 ($p<0.01$) and 24.3 ml/h ($P<0.025$) respectively. A concomitant fall in circulating WBC to 2900/mm^3 ($p<0.001$) and PaO_2 to 68mmHg ($p<0.01$) was also observed. During hemorrhagic hypotension administration of E. coli endotoxin led to a comparable rise in Ppa to 45mmHg, but Pmv increased only to 19.2mmHg which was significantly lower ($p<0.05$) from the control group. Ql in the hemorrhage group was 15.3ml/h after 1 h and 17.4ml/h after 2 h and L/P·Ql was 9.2ml/h and 10.6ml/h respectively. These values were all significantly reduced compared to the control group. The fall in circulating WBC and PaO_2 was comparable in both groups. It is concluded that hemorrhagic shock ameliorates the pulmonary microvascular hypertensive effect and the permeability phase in response to endotoxin, but does not affect hypoxemia.

35

COMBINED EFFECTS OF FAT EMBOLISM AND ENDOTOXEMIA IN SHEEP M.L. NERLICH, J.M. ALBES, D.H. WISNER, J.A. STURM, H.-J. Oestern Dept. of Trauma Surgery, Hannover Medical School, Germany

The role of fat embolism syndrome on the development of posttraumatic ARDS is unclear.An increased sensitivity of the pulmonary microcirculation for bacterial endotoxins after severe trauma is discussed. Our goal was to elucidate the interactions between a fracture-like bone marrow fat intravasation and a standardized endotoxemia We performed chronic lung lymph fistula operations in 24 sheep. 8 sheep were given. sterile bone marrow-fat (30mg/ kgBW)intravenously. Two hours later 2µg/kgBW E. coli endotoxin was given and the reaction observed over 24 hours. This group was compared with 8 sheep receiving only the identical endotoxin dose. Fat infusion alone (8 sheep) caused a pulmonary hypertension without any change in pulmonary capillary permeability lasting for approximately 4 hours.

	PAP mmHG				QL ml/0.5h				L/P RATIO			
	fat+endo		endo		fat+endo		endo		fat+endo		endo	
	x	SEM	x	SEM	x	SEM	x	SEM	x	SEM	x	SEM
Base	15.9	0.2	15.5	0.2	4.5	0.5	2.6	o.2	0.68	0.04	0.69	0.04
1 hour	37.1	1.6	24.8	1.0*	18.3	1.6	12.4	1.3*	0.63	0.03	0.61	0.04
5 hours	27.5	2.5	21.4	0.9*	12.7	1.8	8.4	1.0*	0.81	0.12	0.66	0.03*

(*=p<0.05)

The pulmonary injur y due to endotoxin was significantly increased after previous fat injury. This may be due to either increased eicosanoid products after fat inj. or RES blocade due to fat and subsequently prolonged circulating endotoxin.

36

CROSS REACTIVE AND NON-SPECIFIC IMMUNOPROPHYLAXIS OF GRAM NEGATIVE ENDOTOXEMIA. M.J. GIROTTI,* B. MOON,* T.R. TODD, K. HONG* Department of Surgery, University of Toronto and Queen's University, Ontario, Canada

Our previous work suggested that cross-reactive immunization was protective against heterologous endotoxin challenge (1). As the mechanism remained unclear we undertook to examine the role of different immunization procedures. Five groups of awake Suffolk sheep were challenged with 2 μg/Kg. of intravenous serratia marcenscens endotoxin (SE). Group C (control), Group A (active immunization with E.coli J5 core lipo-polysaccharide), Group WC (active immunization with E.coli J5 whole cells), Group B (active SE). Measured parameters included pulmonary artery pressure (PAP), cardiac output (CO - by thermodilution), arterial blood gases, and white cell counts (WBC). The following results were obtained, each value representing percent change from control.

	GpC(n=5)	CpA(n=5)	Gp(n=5)	GpB(n=5)	GpS(n=5)	
PAP	↑ 170%	63%*	93%	68%*	15%*	
CO	↓ 45%	13%*	18%*	27%	4%*	$p < 0.05$
PaO2	↓ 24%	11%*	26%	14%	8%*	Unpaired T
WBC	↓ 59%	24%*	59%	54%	9%*	

Our previous work suggested that cross reactive immunization was protected against heterologous endotoxin challenge. The results herein suggest that cross reactivity is largely confined to LPS fractions and not whole cells. In addition it suggests a possible though inferior role for non-specific immunization (BCG).

1. Journal of Applied Physiology. 56(3):582-589, 1984.

37

INTERACTIONS BETWEEN HEPATOCYTES AND KUPFFER CELLS DURING SEPSIS. MA West,* GA Keller,* BJ Hyland,* FB Cerra, RL Simmons.* University of Minnesota, Department of Surgery, Minneapolis, MN 55455.

Alterations in liver function are seen during sepsis or endotoxemia. We hypothesize that circulating bacterial products activate Kupffer cells (KC) which mediate alterations in hepatocyte (HC) function. We have shown that endotoxin (LPS) or killed E. coli (KEC) added to in vitro HC alone have no effect on HC protein synthesis. Co-culture of HC with KC (95% pure) markedly enhances HC protein synthesis (p=0.001) but addition of LPS or KEC to HC:KC co-culture results in significantly decreased HC protein synthesis (p=0.01) without changes in morphology or HC viability. We also showed that dexamethasone (10^{-8}M) blocks this decrease after addition of LPS or KEC. Indomethacin had no effect on protein synthesis of HC alone or HC:KC co-culture. When LPS was added to co-culture the decreases seen previously were not blocked by addition of indomethacin (10^{-7}-10^{-5}M). Similar results were seen whether indomethacin was given pre- or post-LPS triggering. More recently, we have found that transfer of HC:KC or KC supernatant, after LPS trigering, results in decreased protein synthesis of HC cultured alone. Early supernate (0-6° after LPS) was more effective than later supernate (6-24° after LPS). These results suggest that: 1) KC mediate biphasic functional alterations in HC, 2) dexamethasone but not indomethacin will block the decrease in HC protein synthesis in co-culture after LPS, and 3) a soluble KC-derived mediator may be responsible for the decreased HC protein synthesis after LPS is added to co-culture.

38

THE ROLE OF C5 IN THE ADULT RESPIRATORY-DISTRESS SYNDROME

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One proposed mechanism for the pathogenesis of the Adult Respiratory Distress Syndrome (ARDS) is that C5a triggers granulocytes to produce and release toxic oxygen radicals which damage cellular membranes in pulmonary capillaries. The purpose of this study was to evaluate the role of C5 in ARDS. The test animals were coisogenic twin mice strains which differed only in being either C5-sufficient (+C5) or C5-deficient (-C5). Cecal ligation and puncture (CLP) was used to produce lethal septicemia. One hundred mice were randomly assigned to 4 groups: +C5 anesthetic control, +C5 CLP, -C5 control and -C5 CLP and survival was recorded every 6 hrs. Eighty additional animals were similarly randomized. At 24 hours, 10 randomly selected animals from each group were sacrificed for morphometric analysis of lung specimens. Intracapillary granulocrit (gct) and air-blood barrier thickness (ABBT) were then determined. The remaining 40 animals were anesthetized and arterial blood was obtained from the abdomenal aorta of each mouse for arterial pO2 measurement. In this lethal sepsis model, mean survival time is significantly increased in -C5 septic mice ($p < .001$). Morphometric results demonstrate an increase in intracapillary gct and ABBT 24 hours after CLP in +C5 mice ($p < .001$) Similarly, mean arterial pO2 is decreased in the +C5 septic animals ($p < .001$). The importance of C5 in ARDS is illustrated by the finding of normal parameters in the -C5 septic twins of these animals.

39

ASSESSMENT OF SOLEUS MUSCLE AMINO ACID TRANSPORT ALTERATIONS IN ENDOTOXIC RATS.

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We previously reported a decrease in amino acid (AA) transport by endotoxic rat soleus muscle, *in vitro*. This study evaluated the kinetics of AA transport *in vitro* and the magnitude of change *in vivo* in endotoxic rats. *In vitro* measurements were made in soleus muscle from fasted male Holtzman rats (80-100g) 5 hrs after intravenous saline (SAL) or S. enteritidis endotoxin (ET) (20mg/kg). Kinetic parameters V_{max} and K_m were estimated from initial rates of uptake of the AA analog α-aminoisobutyric acid (AIB) during incubations in O_2-Krebs with various [AIB] (0.1mM-20mM). For *in vivo* studies fasted rats were injected with AIB (1mg/kg) and ET (20mg/kg) or SAL and killed 1-5 hrs later. AIB uptake by muscle *in vivo* was corrected for uptake into extracell space (EC) (measured as inulin distribution in muscle, *in vivo*) and expressed as ratios of [AIB]cell/[AIB]extracell to assess active transport.

	[AIB]cell/[AIB]extracell (hrs post-injection)				EC Space	V_{max}	K_m
	1	2	4	5	(ml/g)	(nmol/g/min)	(mM)
SAL	2.1±.2(6)	2.5±.2 (7)	4.7±.3 (12)	4.9±.3 (5)	.22±.01 (9)	55 (62)	1.7(62)
ET	1.7±.1(5)	1.8±.3[a](4)	2.8±.2[a](12)	3.5±.3[a](4)	.17±.01[a](6)	18[b](67)	1.6(67)

Values=Mean±SE, ()=# rats, a=p<0.01 ET vs SAL, b=p<0.01 (analysis of covariance)

The decrease in active AA transport *in vivo* in endotoxic rats is comparable to that measured previously *in vitro*. The altered transport is apparently not due to a change in membrane carrier affinity (K_m) for AA but rather a decrease in the number of the carriers and/or a decrease in their mobility in the membrane (V_{max}). Alterations in AA transport may limit the availability of AA for muscle protein synthesis during endotoxic shock. (Supported by NIH Grant GM 32288.)

40

OXIDANT INJURY AND CELLULAR MORPHOLOGY.

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Oxidants, known to be generated in inflammatory reactions, produce severe tissue damage. We have examined the effect of oxidants on the biochemistry and morphology of target cells. Cellular ATP levels of target P388D_1 cells fell to 10 percent control values within 20 min of exposure to 5mM H_2O_2. Intracellular Ca^{++}

($[Ca^{++}]_i$) rose from resting levels of ~100nM to >1500nM after 60 min of injury with 5mM H_2O_2. Plasma membrane blebbing followed the rise in $[Ca^{++}]_i$ and preceded loss of viability as seen by Trypan Blue exclusion. Cellular filamentous (F) actin content, measured by flow cytometry of fluorescent phallacidin stained cells and SDS-PAGE of Triton X-100 irsoluble cytoskeletons, increased within 30 min exposure to H_2O_2. The F-actin was seen to form large highly ordered aggregates progressively over 2 hr of injury by transmission electron microscopy (EM). Considerable rounding of injured cells adherent to a plastic substrate as well as diminished surface area of contact with the substrate was seen with fluorescent and electron microscopy. Microtubules, readily identifiable by transmission EM at earlier time points of injury, disappeared by 2 hr, a finding consistent with the rounding observed and the high $[Ca^{++}]_i$ seen by that time. The decreased levels of ATP are attributed to decreased glycolysis, produced by a fall in the essential coenzyme, NAD, which was secondary to stimulation of ADP ribose polymerase. Normal NAD, ATP, $[Ca^{++}]_i$, and F-actin were maintained when the polymerase was inhibited. These oxidant mediated changes could play a significant role in affecting the integrity of surfaces comprised of adherent cells (e.g. endothelium) during acute inflammation

41

HYPOXIA CAUSES ELEVATED PLASMA LPS CONCENTRATION IN MONKEYS. S.L. GAFFIN, J.G. BROCK-UTNE, M. WELLS, A. ZANOTTI, J.W. DOWNING. Departments of Physiology, Paediatric Surgery and Anaesthetics, University of Natal Medical School, Durban, South Africa.

The breathing of an hypoxic gas mixture leads to a reflex reduction in peripheral blood flow, including splanchnic blood flow, in favor of blood flow to the brain and heart. It is also known that intestinal ischemia damages the gut wall, allowing endotoxin present within the intestines to escape into the blood circulation. We examined here whether the reduced blood flow by the hypoxic reflex can lead to sufficient damage of the intestinal wall to cause endotoxemia. Anesthetized vervet monkeys breathed air for one hour, then an hypoxic gas mixture (F_{IO_2} = 0,13) for one hour and, finally, 100% O_2. Within 15 minutes of hypoxia the plasma LPS concentration rose significantly from baseline 0,39 ng/ml, reached a peak of 1,60 ng/ml ($p < 0,001$) after 30-40 minutes and then declined during the hypoxic episode to eventually reach baseline levels. Control monkeys breathing air or 70% N_2O (F_{IO_2} = 0,3) for three hours, showed no such elevation in plasma endotoxin concentration. We conclude, therefore, that hypoxia leads to a temporary endotoxemia in primates. We further found that RES damage caused by whole body X-irradiation (200r) led to elevated endotoxin levels lasting for a longer period of time during a standard hypoxic insult. On the other hand, in keeping with our previous reports, Anti-endotoxin hyperimmune plasma (Anti-LPS) reduced both the magnitude and duration of this hypoxia induced endotoxemia. This hypoxia model provides a simple non-invasive model for endotoxemia and possibly for RES function.

42

TYPE OF ANESTHESIA INFLUENCES OUTCOME IN A PORCINE SEPTIC SHOCK MODEL. U. PFEIFFER*, M. PERKER*, H. WELLHÖFER*, H. REICHLE*, G. BLÜMEL*, G. ZIMMERMANN* (Introduced by: H.-J. Oestern). Institute of Experimental Surgery, D-8000 Munich 80, FRG.

Pentobarbital (P, 15 mg/kg/h) anesthesia was compared to anesthesia with ketamine (K, 10 mg/kg/h), ketamine+metomidate (KM, 2.5+3 mg/kg/h), and halothane (H, 0.4 Vol%) in Pseudomonas aeruginosa sepsis in pigs with an average body weight of 29.3 kg. The animals were paralyzed with alcuroniumchloride (0.3 mg/kg/h) and put on artificial ventilation (F_{IO_2} = 0.33)). 2.5 x [illegible] /kg/h live organisms were infused till death of the pigs. Pulmonary capillary wedge pressure was well maintained by infusion of 6 % dextran 70. The mean survival times for the different groups (n = 8, each) were 2.2 hrs (P), 3.6 hrs (KM), 6.2 hrs (H), and 9,8 hrs (K). 2 hrs after the start of bacteria infusion pigs of groups KM and P as compared to groups K and H had significantly lower total systemic resistance, whilest cardiac output and pulmonary extravascular thermal volume (ETV) were not different. The animals of groups P, KM, and H died of septic circulatory failure, whilest ketamine monoanesthesia resulted in death of pulmonary edema (ETV 2.2 times of control at 8 hrs). Endotoxic

circulatory failure seems to be mediated in part by activation of the central adrenergic pathway and the - at least - transitory stabilization by ketamine could result - similar to the action of yohimbine - from an antagonizing effect at the central α_2-receptors, which are, as far as known, also involved in the regulation of ß-endorphine release. This study demonstrates, that results from septic animal models strongly depend on the type of anesthesia employed.

43

PROSTACYCLIN AND THROMBOXANE IN SEPTIC SHOCK: SPECIES DIFFERENCES. J.V. QUINN*, S.A. YELLIN*, D. NGUYEN*, K.W. BURCHARD, G.J.SLOTMAN. Dept. of Surgery, Brown University, Rhode Island Hospital, Providence, RI 02903

Prostacyclin and thromboxane A_2 have been implicated in hypotensive sepsis. Correlations between the human prostanoid response to sepsis and experimental paradigms are poorly understood. The purpose of this study was to compare changes in plasma levels of prostaglandin 6-keto-$F_{1\alpha}$(PGI) and thromboxane B_2 (TxB) during septic shock in Sprague-Dawley rats, adult female pigs, and in man. Severe sepsis followed by septic shock (systolic BP <90 mmHg) was induced in rats by subcutaneous innoculation of 10^9 Aeromonas hydrophila and in pigs by graded intravenous infusion of 1.0 X 10^9/ml Aeromonas hydrophila. Plasma PGI and TxB, pg/ml, were measured by radioummunoassay of control, septic, and septic shock experimental blood samples, and of aliquots from non-septic (control), septic, and septic shock (systolic BP <90 mmHg) S.I.C.U. patients. Results:

	CONTROL		SEPTIC		SEPTIC SHOCK	
	TxB	PGI	TxB	PGI	TxB	PGI
Rat (n=33)	60±8**	83±10	269±55*	263±49*	182±35*	407±140*
Pig (n=8)	413±68	141±21	1690±371*	284±68	2210±426*	1988±843*
Human (n=94)	108±21**	208±40	243±38	213±32	187±37	708±333*

*p<0.05 vs. control ** p<0.05 vs. pig ANOVA

Control and septic TxB levels vary significantly between species. PGI is increased in septic shock in all 3 species. Murine and porcine septic shock models are clinically relevant for studies of PGI in sepsis but may not be valid for TxB.

44

EICOSANOID METABOLISM IN ENDOTOXIN (LPS)-TOLERANT (TOL) MACROPHAGES (MØ): DIFFERENTIALLY ALTERED LIPOXYGENASE (LO) AND CYCLOOXYGENASE (CO) PATHWAYS T.S. Rogers,* P.V.Halushka,* W.C.Wise, and J.A.Cook. Med. Univ. of S.C., Charleston, S.C. 29425.

Altered MØ arachidonic acid (AA) metabolism may play a role in LPS shock and the phenomenon of LPS TOL induced by repeated sublethal injections of LPS. Studies were initiated to characterize both LO and CO metabolism in LPS TOL and non-TOL MØ in response to LPS and the calcium ionophore, A23187. Basal levels of the LO metabolites, immunoreactive (i) leukotriene (LT) B_4 and C_4/D_4, and the CO metabolites $iTxB_2$, i6-keto-PGF_{1a}, and $iPGE_2$ were decreased in LPS TOL vs. non-TOL MØ ($p<0.05$). $iLTB_4$ and $iLTC_4/D_4$, unlike the CO metabolites, were not significantly stimulated by LPS (50 ug/ml) in either the TOL or non-TOL group. Stimulation of PG and $iTxB_2$ synthesis by LPS was less in TOL MØ compared to controls ($p<0.05$). A23187 (1 uM)stimulated synthesis of all eicosanoids in both groups. However, A23187-stimulated $iLTC_4/D_4$ and $iPGE_2$ production in TOL MØ was increased, whereas, i6-keto-PGF_{1a} levels were lower. $iLTB_4$ and $iTxB_2$ levels were not different (TOL vs.non-TOL).

A23187 Stimulated Eicosanoid Production [ng/ml]

	$iLTB_4$	$iLTC_4/D_4$	$iTxB_2$	i6-keto PGF_{1a}	$iPGE_2$
non-TOL	13.9±2.3	9.2±0.7	33.7±1.7	9.7±1.8	8.4±0.5
TOL	14.7±0.7	32.9±1.7*	47.3±4.9	4.4±0.6*	17.4±2.0*

values are mean + S.E.M. * = $p<0.05$ non-TOL vs.TOL. n = 10-22.

These data demonstrate that LPS stimulates MØ synthesis of CO but not LO metabolites, and that AA metabolism by the LO and CO enzymes in MØ is differentially altered by LPS TOL (Supported by NIH GM 27673 and HL 29566).

45

Diethylcarbamazine, a Leukotriene Inhibitor, Improves Survival Of Endotoxemia in the Rat. F. Rogers*, R. Dunn*, P. Nolan*, A. Phuangsab*, J. Barrett*, Cook County Hospital/University of Illinois, Chicago. Ill. (Introduced by J. Ferguson).

Leukotrienes have been recently implicated in sepsis. This study investigated the effect of the leukotriene inhibitor, diethylcarbamazine (DEC), on rat endotoxemia mortality. Male Spraque-Dawley rats were randomly assigned to four groups of 3i animals each. All substances were administered by intraperitoneal injection. Group I was given E. Coli endotoxin (15mg./kg.) (lipopolysaccharide B, E. Coli 026:B6; Difco Labs), Group II received DEC in five doses over ten hours for a total of 750mg/kg. Group III was given endotoxin (15mg/kg) followed by DEC in five separate injections over a ten hour period for a total dose of 750mg/kg. Group IV was pretreated with a single injection of DEC (150mg/kg) one hour prior to receiving endotoxin (15mg/kg) followed by additional doses of DEC over a ten hour period for a total DEC dose of 750mg/kg. Survival rates after 48 hours:

	Gp I (endo)	Gp II (DEC)	Gp III (endo-DEC)	Gp IV (DEC-endo)
Surviving/Total	10/31	31/31*	23/31*	24/31*
Mortality rate:	68%	0%	26%	23%

* $p < 0.01$ compared to Group I (chi square)

Treatment with DEC significantly reduced rat endotoxemia mortality; rates declined by 62% and 66% compared to non-DEC treated controls. DEG has been previously shown to inhibit leukotriene synthesis. The results of this experiment show that leukotriene inhibition may be useful in the treatment of sepsis.

46

INFLAMMATORY EICOSANOIDS DECREASE STRESS FIBERS IN CULTURED AORTIC ENDOTHELIAL CELLS. S.L. WELLES[1*], D. SHEPRO[1,2], and H.B. HECHTMAN[3]. Departments of Biology[1] and Surgery[2], Boston University, and Department of Surgery[3], Harvard Medical School, Boston, MA, 02215.

Endothelial cell (EC) stress fibers (SF) are hypothesized to maintain intimal and microvascular integrity in situ, and SF decreases may be correlated with decreased EC barrier function. Accordingly, polymorphonuclear leukocyte (PMN) supernatant, leukotriene (LT) B_4, LTD_4 and thromboxane (TX) B_2, all implicated in edema formation, were tested as stimuli of SF disassembly. As revealed by fluorescence microscopy, EC treated for 15 min with PMN supernatant, 0.1μM LTB_4, LTD_4 or TXB_2 decreased SF numbers to 7%, 7%, 39% and 35%, respectively, of controls (3.8 ± 0.5 fibers/cell). All decreases were significant ($p<0.01$). Pretreatment of EC with FPL-55712, 13-APA, imidazole and ketoconazole prevented LTD_4 and TXB_2-stimulated SF decreases. LTB_4-stimulated SF disassembly was not inhibited by FPL-55712 and 13-APA, but was blocked by imidazole and ketoconazole. Pretreatment of EC with nifedipine or lanthanum antagonized LT or TX-stimulated SF disassembly. These data indicate that: 1) eicosanoid-stimulated SF disassembly may be correlated with edema formation, 2) SF disassembly may be receptor-mediated and stimulate de novo TXB_2 synthesis by EC, and 3) SF disassembly may be dependent, in part, on calcium influx. (Supported in part by USPHS grants HLB 16714 and 33104; GM 24891.)

47

EFFECT OF FRUCTOSE 1-6 DIPHOSPHATE ON THE PULMONARY CIRCULATION AND DAMAGE ASSOCIATED WITH ALPHA-NAPHTHYLTHIOUREA. A.K. Markov, A.L. Causey,* L. Dorroh,* R. Didlake,* J.A. Fletcher.* Univ. of Mississippi Medical Center, Jackson, MS 39216.

In treating patients in shock with fructose 1-6 diphosphate (FDP), in those who concomitantly had Adult Respiratory Distress Syndrome (ARDS) we noted significant hemodynamic, radiographic and pulmonary function improvement. In an effort to elicit the mechanism of the protective action of FDP in ARDS we simulated the condition in 25 anesthetized dogs by injecting them intravenously with 5 mg/kg Alpha-naphthylthiourea (ANTU). The animals were randomly assigned into two groups. Thirty minutes after administration of ANTU, 12 dogs were treated with FDP, and those serving as controls received a saline solution in the same volume. At 4 hours the dogs were killed, the lungs exsanguinated, and wet and dry weights recorded. The pulmonary pressures in the dogs treated with FDP remained unchanged, while in

those serving as controls, they increased from 12.9 ± 2.4 to 21.8 ± 3.14 mm/Hg ($p < 0.001$). There were no differences between the groups in left ventricular diastolic and arterial pressure, and cardiac output. However, pulmonary resistance in the dogs serving as controls was significantly higher ($p < 0.001$). Lung to body weight (gm/kg) for the FDP treated group was 9.83 ± 0.684 and for the controls 16.7 ± 0.99 ($p < 0.001$). The wet to dry lung weight ratio for the treated dogs was 4.32 ± 0.17, and for those receiving saline 6.18 ± 0.396 ($p < 0.001$). These results indicate that FDP prevents pulmonary vascular damage associated with ANTU. The attenuation of pulmonary microvascular damage by FDP in this model is attributed to the prevention of free radical formation (superoxides and peroxides) by the neutrophiles.

48

A CONTROLLED CLINICAL TRIAL OF PURIFIED FIBRONECTIN (FN) IN PATIENTS WITH SEVERE ABDOMINAL INFECTIONS (SAI). P. LUNDSGAARD-HANSEN, E. RUBLI*, J.E. DORAN, E. PAPP*, J.-J. MORGENTHALER*. University Hospital, CH 3010 Berne, Switzerland.

We prospectively studied 72 patients admitted with SAI, sequentially assigning them to conventional intensive care alone (Cntrls) or to intensive care supplemented with a dose of 0.8g purified Fn on Days 1-5 post admission. The Fn used was shown to be functionally active in gelatin-binding and phagocytosis-stimulating assays. Patients were monitored for 11 days in the ICU, then followed until their death in hospital or discharge to home (= ultimate death/survival). Three post-ICU deaths in the Fn group and two in the Cntrl group were unrelated to the SAI and were excluded from statistical analyses. Number of organ failures and presence of postoperative SAI on admission correlated with mortality; however Fn and Cntrl groups did not differ in these respects. All patients had counter-SAI surgery within 24 hrs of their admission to the study. Ultimate mortality in the Fn and Cntrl groups was not significantly different (9/33 and 13/34 respectively). As a group, survivors had higher levels of Fn, Antithrombin III, C3, C4, C3b INH and transferrin than did the nonsurvivors on Days 1-11 ($0.001 < p < 0.05$). In contrast, Fn recipients and Cntrls differed only in their Fn levels on Days 3-11. Fn levels ($\bar{x}$ ± SEM) on Days 1 and 5 were:

	+Fn Surv.	+Fn Deaths	Cntrl Surv.	Cntrl Deaths
Day 1	176 ±15	152 ±24	178 ±13	128 ±26
Day 5	544 ±40	314 ±44	285 ±32	170 ±28

Our normal reference range ($\bar{x}$ ± 2SD) for Fn is 195-545 ug Fn/ml. Using all complete ICU data, Fn correlated with the above mentioned proteins ($0.77 \geq r \geq 0.56$, N=233, $p < 0.001$), indicating parallel variations of these major regulatory plasma proteins.

49

^{31}P NUCLEAR MAGNETIC RESONANCE (NMR) STUDY OF REPERFUSED KIDNEY POST-SHOCK. R. S. RHODES, J. E. JENTOFT, R. G. BARR.* CWRU School of Medicine, Cleveland, OH 44106

The on-line, non-destructive advantages of ^{31}P NMR were used to study the phosphorus metabolites of isolated, perfused kidneys following shock. Thirteen anesthetized rats were subject to a modified Wiggers' model of hemorrhagic shock lasting 120 to 270 minutes. The right kidney was then perfused with oxygenated Krebs-Henseleit solution for 180 minutes while in the NMR spectrometer. Spectra were obtained every 15 minutes. Five kidneys from non-shocked rats served as controls and maintained high levels of ATP, low ADP, and normal intracellular pH during perfusion. The levels of phosphomonoester (sugar phosphates, AMP, etc.), inorganic phosphate (Pi), phosphodiester (PDE), and NAD also remained constant. Kidneys from shock animals had higher Pi ($p<.01$) and lower ATP ($p<.01$) than controls. ATP/ADP ratios remained high, however. When stratified as to early or late shock, 5 of 6 early shock but only 2 of 7 late shock kidneys had normal Pi or ATP. The levels of PDE were significantly reduced ($p<.001$) in both early and late shock suggesting greater sensitivity to injury in the medulla. Intracellular pH, Mg++, and all other phosphorus metabolites were similar to controls. The inability to restore ATP content to normal with reperfusion post-shock does not appear to be secondary to inability to perfuse ischemic tissue as there was no evidence of persistent acidosis.

50

A RANDOMIZED CLINICAL TRIAL ON ATP-$MgCl_2$ FOR POSTISCHEMIC ACUTE RENAL FAILURE (ARF).
H. HIRASAWA, K. SOEDA*, Y. OHTAKE*, M. OHKAWA, M. ODAKA*, H. SATO*. Department of Emergency and Critical Care Medicine, Department of Surgery, Chiba University School of Medicine, Chiba, Japan 280.

Previous experimental studies from our laboratory have shown that ATP-$MgCl_2$ has a beneficial effect on the recovery of renal function following ischemic insult to the kidneys. The present study was undertaken to investigate the efficacy of $MgCl_2$ administration for the treatment of postischemic ARF patients. Thirty-two patients, who were admitted to our institution from March 1979 to March 1984 and whose main pathogenesis of ARF was considered to be ischemic insult, were randomly divided into two groups. Sixteen patients received ATP-$MgCl_2$ (40-50 μmoles/kg) intravenously on the day of admission or on the following day (ATP Group) and were treated according to our regimen (i.e. hemopurification with hemodialysis and/or hemoadsorption, intravenous hyperalimentation, enhancement of reticuloendothelial system). The other sixteen patients (Control Group) were treated with the same regimen as the ATP Group, but did not receive ATP-$MgCl_2$. There were no significant differences in sex and age distribution between the two groups. The clinical course and survival between the two groups were compared. Mean serum creatinine levels at admission were 8.8 ± 1.0 mg/dl in the ATP Group and 8.2 ± 0.7 mg/dl in the Control Group (NS). The mean duration of hemopurification treatment was 9.8 ± 1.8 days in the ATP Group and 11.9 ± 2.7 days in the Control Group. The survival rate was 100% in the ATP Group and 73.3% in the Control Group ($p < 0.05$). These data suggest that ATP-$MgCl_2$ administration is beneficial for the treatment of postischemic ARF patients.

51

NMR EVALUATION OF VERAPAMIL TO PREVENT REPERFUSION INJURY AFTER RENAL ISCHEMIA.
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Verapamil has been shown to moderate ischemic renal damage during the reperfusion period. We chose to examine renal high energy phosphate metabolism during ischemia and reperfusion in an in vivo rabbit model using ^{31}P Nuclear Magnetic Resonance (NMR) spectroscopy. Renal blood flow (RBF) was measured using radioisotope labeled microspheres. Our purpose was to determine whether NMR is a useful technique to measure metabolic changes acutely, whether verapamil produced significant improvement in metabolic parameters, and whether these metabolic changes were flow related. Rabbits (n=12) were divided into control(C) or verapmil(V) treated groups. All animals were anesthetized with Halothane (1-2%), arterial and venous catheters inserted, and an NMR surface coil placed on one kidney with the vascular supply prepared for occlusion. NMR spectra of the designated kidney and microsphere injection were performed during control, occlusion, and reperfusion periods of 40min. each. The V group received a bolus of 0.1mg/kg 5 min before reperfusion followed by a .005mg/kg/min infusion for 15 min. Renal blood flow was 3.01±1.41ml/min/mg tissue during control periods. The occluded kidney had no demonstrable blood flow. During reperfusion RBF returned to 100% of control in both groups. Verapamil did not significantly increase the rate of ATP recovery or the end recovery ATP levels.

52

INCREASED TOLERANCE TO GLOBAL HYPOXIA FOLLOWING ATP-$MgCl_2$ TREATMENT.
P.S. Greene*, I.H. Chaudry, M.G. Clemens, M.J. Hull*, and A.E. Baue. Dept. of Surgery, Yale University School of Medicine, New Haven CT 06510

Since the susceptibility of the brain to hypoxic injury is thought to be due in part to its small stores of high energy phosphates, we studied the effect of ATP-$MgCl_2$ treatment on animals exposed to a low O_2 environment. Mongolian gerbils (45-60g) were given i.p. drug treatments and 30 min later exposed to 3% O_2:97% N_2. Saline treated animals survived 7.4±0.8 min (mean±SE). ATP-$MgCl_2$ (300 μmole/kg) increased survival time to 16.3±1.6 min ($p < .05$), which was longer than the 12.2±1.5 min seen with ATP alone ($p < .05$). Neither adenosine-$MgCl_2$ (300 μmoles/kg) nor $MgCl_2$ alone prolonged survival significantly over saline controls. In separate experiments, ^{14}C-ATP or ^{14}C-adenosine were given i.p. and 30 min later gerbils were decapitated and their organs harvested. Distribution of radioisotopes in percent of injected dose per g of tissue (x100) were:

Label	Brain	Blood	Heart	Liver	Kidney	Muscle
^{14}C-ATP	7.1±1.4	143±16	268±59	624±71	359±32	47.1±4.3
^{14}C-Aden	9.7±1.0	114±27	178±6.5	588±128	654±109	58.9±10

Thus neither ATP nor Adenosine treatment resulted in significant incorporation of labelled purine into the brain. Similar results were obtained in gerbils exposed to hypoxia. Although ATP-$MgCl_2$ has been found to improve cellular function and nucleotide levels following adverse circulatory conditions in other organs, the present results indicate that, in the brain, ATP-$MgCl_2$ mediates a protective effect without crossing the blood-brain barrier. (NIH grant HL-19673).

53

ATP-$MgCl_2$ IS DETRIMENTAL TO ISOLATED HEPATOCYTES FROM SHOCK INJURED LIVER. D.J. BARILLO*, B.F. RUSH, Jr., M.J. DONOHOE*, G.S. DIKDAN*. UMDNJ- New Jersey Medical School, Newark, N.J. 07103

The isolated hepatocyte from the shock injured liver is used in our lab to test the efficacy of drugs in hemorrhagic shock. This affords an opportunity to observe changes in the cells independent of the vascular system. Sprague-Dawley rats were shocked to 30 mmHg until 40% of blood was returned. The liver was perfused 1 hr post shock. Sodium, K+ and cell viability (V) were determined on the freshly isolated cells (I) and on cells incubated for 1 hr at 37 C in 1) Swim's S77 (C), 2) Swim's S77 with methylprednisolone (MP) (143 ug/cc) and 3) Swim's S77 with ATP-$MgCl_2$ (ATP) (0.18 uM/cc). V determined by trypan blue exclusion.

Group	Na+ (meq/l)	K+ (meq/l)	V
I (n=13)	53.9	82.2	85.6
C (n=8)	24.7≠	85.3	73.1
ATP (n=6)	27.2≠	80.5	66.7≠
MP (n=5)	23.7≠	89.6	76.2

significance ≠ $p<.05$ relative to no incubation, 1 hr post shock (I)

ATP-$MgCl_2$ incubation causes significant reduction in viability of shock injured hepatocytes. MP has no significant effect on V or intracellular electrolytes.

54

THE SICK CELL SYNDROME: A CATECHOLAMINE-INDUCED RESPONSE TO STRESS? M.G. CLEMENS, I.H. CHAUDRY and A.E. BAUE. Yale University School of Medicine, New Haven CT 06510.

In spite of compensatory mechanisms, severe hemorrhage produces rapid alterations in cell functions. An early such change is depolarization of the hepatocyte membrane. Since this effect is more rapid than expected from hypoperfusion alone, we investigated whether increased catecholamine levels might be responsible. Rats were anesthetized with Nembutal and a femoral artery was cannulated for blood pressure measurement. Portal pressure monitoring and drug infusion were via a double-lumen catheter in the splenic vein. Hepatic membrane potentials were measured with glass microelectrodes and liver blood flow with H_2 polarography. When epinephrine was infused into the portal vein at 1-5 µg/min it increased portal pressure (2-10 mmHg), and hyperpolarized hepatocytes from -42.0±1.1 to -50.2± 1.6 mv ($p<.001$) (mean ±SE) but did not affect systemic blood pressure or liver blood flow. High epinephrine doses (>10 µg/min), however, increased both portal and systemic pressures, decreased liver blood flow by 50% and rapidly depolarized the hepatocytes from -47.2±2.5 mv to -37.4±1.9 mv ($p< 001$). Thus, high epinephrine concentrations which might be found following hemorrhage or trauma produce a rapid depolarization of hepatocytes and decreased hepatic perfusion. Since epinephrine stimulates glycogenolysis it may also be responsible for the early hyperglycemia observed following hemorrhage and trauma. Thus, while epinephrine helps compensate for decreased circulating volume, it may also mediate early membrane and metabolic alterations in stress and injury. (Supported by NIH grant HL-19673)

55

REOXYGENATION INJURY IN ISOLATED HEPATOCYTES : EFFECTS OF ATP-$MgCl_2$. H. HAYASHI*, I.H. CHAUDRY, M.G. CLEMENS, M.J. HULL*, A.E. BAUE. Yale University School of Medicine, New Haven, CT 06510

In order to determine whether ATP-$MgCl_2$ ameliorates reoxygenation injury independent of humoral or vascular effects, we studied the effects of ATP-$MgCl_2$ treatment following anoxia/reoxygenation in isolated rat hepatocytes. Hepatocytes were incubated in Krebs HCO_3 buffer with substrates and albumin. Intracellular electrolytes and ATP levels (μmol/g wet, mean±S.E.) of control (oxygenated) and after 90 min anoxia + 60 min reoxygenation with and without 0.1mM ATP-$MgCl_2$ were:

	Na	K	Mg	Ca	ATP	Pi
CONTROL	12.8±1.3	69.7±1.8	8.8±0.2	1.9±0.2	2.1±0.2	2.8±0.4
ANOXIA	43.3±1.9*	49.3±2.8*	8.5±0.3	2.0±0.2	0.6±0.1*	10.9±0.4*
REOXYGENATION	22.1±2.0*	50.5±3.5*	6.8±0.2*	2.3±0.3*	0.6±0.1*	2.8±0.4
+ ATP-$MgCl_2$	17.2±3.2**	58.3±5.6**	7.5±0.3**	2.9±0.8	0.8±0.1**	3.7±0.2**

* $p<0.05$ compared to CONTROL, ** $p<0.05$ compared to REOXYGENATION

The accumulation of Pi and the lack of changes in Mg or Ca following anoxia indicate that plasma membrane permeability to divalent ions remains relatively intact during anoxia but increased with reoxygenation. Addition of ATP-$MgCl_2$ after reoxygenation significantly improved Na, K, Mg and ATP levels and produced an accumulation of Ca and Pi in the cell. Additions of ATP, $MgCl_2$, or Pi alone did not have the same effects. These results indicate that reoxygenation injury occurs in hepatocytes independent of circulatory defects and that ATP-$MgCl_2$ directly improves post-anoxic cell metabolism. (Supported by NIH grant HL-19673)

56

ENHANCEMENT OF ENDOTOXIC SHOCK BY GALACTOSAMINE (GAIN)-INDUCED LIVER INJURY. A. AL TUWAIJRI, A. AL AKWA, C. GASSIM, I. MARAWI, A. SAAD, E. SHAIL. Department of Physiology, King Saud University, Riyadh, Saudi Arabia.

This study was carried out to further evaluate the role of bacterial endotoxic shock in Galactosamine (GaIN) induced hepatitis in experimental animals. Several studies have shown that endotoxin is an important factor in GaIN liver injury. Wistar rats were given single IP injection of GaIN, and on different time intervals were given E. coli endotoxin. Various doses of GaIN and endotoxin were tested. Mortality rate and blood chemistry were determined. Animals pretreated with a minimal amount of GaIN become extremely sensitive to very small doses of endotoxin. Administration of 10 ug/kg endotoxin at 6 and 24 hours after GaIN (30 mg/kg) injection caused 100% mortality compared to animals which receive GaIN only. Endotoxin administration 12 or 24 hours prior GaIN failed to induce mortality. In addition, liver enzymes and bilirubin are markedly ($P< 0.01$) increased in endotoxin-GaIN treated rats compared to GaIN treated group. These findings indicate that a minimal amount of endotoxin (few micrograms) markedly aggrevate GaIN induced hepatitis. The present result strongly suggest the importance of endotoxin in GaIN induced hepatitis.

57

TOTAL HEPATIC BLOOD FLOW (THBF) IN EXPERIMENTAL PERITONEAL SEPSIS: THF EFFECTS OF GENTAMICIN AND STEROID. T.C. FABIAN, P. DIXIT*, C.R. PATTERSON*. University of Tennessee, Memphis, TN 38163

In male Sprague-Dawley rats indocyanine green (ICG) clearance was used to determine THBF at 6 and 18 hrs after either sham operation (control, n=16) or cecal ligation & puncture (septic, n=48). Septic animals received either no therapy, gentamicin alone (GS), or gentamicin and methylprednisolone (GS/MP). The last is a combination we have previously reported to improve survival in this model.

ICG $t_{½}$ (mins)
($\bar{x} \pm$ SEM)

		SEPTIC ANIMALS		
TIME (hrs)	CONTROL	No Rx	GS	GS/MP
6	1.81 ± 0.10	2.24 ± 0.22	2.02 ± 0.28	2.04 ± 0.18
8	1.95 ± 0.07	4.10 ± 0.52	2.79 ± 0.22	2.88 ± 0.24

There were no significant differences among the 6 hr groups. At 18 hrs untreated animals differed significantly from control animals ($p<.01$) and from gentamicin treated animals ($p<.05$). Animals treated with gentamicin and steroid did not differ from those treated with gentamicin only. These results confirm that peritoneal sepsis produces hepatic hypoperfusion; this effect is significantly reversed by gentamicin. The improved survival reported with added steroid therapy does not result from increased THBF.

58

SEPSIS DECREASES THROMBOXANE (TX) PRODUCTION IN THE ISOLATED PERFUSED RAT LIVER
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These studies were conducted to determine the effect of surgical stress and sepsis on hepatic Tx production. METHODS: Adult Male Sprague-Dawley rats(250-300 g) were randomly divided into 3 groups (N=10): Control (C) were unmanipulated, while 48h prior to study, Septic (CL) underwent laparotomy and cecal ligation, and Sham (SH) underwent laparotomy only. Livers were perfused at 25 ml/min in a non-recirculating system with Kreb's bicarbonate buffer containing 5mM glucose, 4% (w/v) human albumin, and 15% (v/v) human erythrocytes. Posthepatic samples of perfusate were analyzed for TxB_2 by RIA. In a subset of each group (N=5), zymosan activated plasma (ZAP) was infused prehepatically (0.3ml/min) after 30 min of perfusion. RESULTS: TxB_2 production in pg/g/min presented below as the Mean±SE

Group	Time (min) after starting perfusion							
	1	5	15	20	30	31	45	60
C	666±180	1403±306	1547±397	1628±392	1656±364		714±321	503±209
C+ZAP						2173±181	2152±217	1797±290
SH	706±212	1704±363	2341±728	2464±633	2139±452		1140±448	956±331
SH+ZAP						2907±413	2684±486	2175±314
CL *	143±32	362±81	644±178	910±270	1149±347		589±315	556±276
CL+ZAP						1944±557	2228±625	2210±437

* $p < .015$ vs SH (2-way ANOVA) CONCLUSION: Sepsis significantly decreases basal hepatic Tx production, but does not affect Tx release in response to ZAP. This suggests that C5a-mediated Tx synthesis is unaffected by sepsis.

59

A STREPTOCOCCAL PREPARATION (OK-432) IMPROVES SURVIVAL AND RETICULOENDOTHELIAL SYSTEM (RES) FUNCTION IN CIRRHOTIC SEPTIC RATS.
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It has been shown that cirrhotics are susceptible to infection due to depressed RES function. The present study was undertaken to investigate whether OK-432, a penicillin-, heat-treated lyophilized powder of Su-strain Streptococcus pyrogenes A3, would improve survival and RES function following sepsis in cirrhotic rats. Cirrhosis was produced by the subcutaneous injection of CCl_4 twice a week for 10 weeks. Either OK-432 in a dose of 0.1 KE/rat (OK-432 group) or saline (saline group) was injected ip on day five after the last CCl_4 injection. Sepsis was produced by ischemic intestinal loop method two days after OK-432 or saline injection. Global RES phagocytic activity was measured using ^3H labeled lipid emulsion method prior to the sepsis procedure. In vitro Kupffer cell and plasma opsonic activities were also studied using liver slice bioassay method. The survival was measured over a period of 7 days. The results were as follows.

	survival (%)	global RES phagocytic index	Kupffer cell activity (%ID/100mg)	Plasma opsonic activity (%ID/100mg)
Sham group	33	0.0402 ± 0.0029	4.82 ± 0.68	5.82 ± 0.57
OK-432 group	80**	0.0544 ± 0.0041**	6.13 ± 1.00	8.03 ± 0.77*

*$p < 0.05$, **$p < 0.01$ compared to sham group

Thus the OK-432 significantly improved the survival of cirrhotic rats following sepsis, through the improvement of RES function caused by increased opsonic protein.

60

SERA MEASUREMENTS IN SWISS WEBSTER (SW) MICE REFLECTING EARLY HEPATIC DYSFUNCTION DURING SEPSIS. N.A. SACCO*, R.S. MCCUSKEY. Department of Anatomy, West Virginia University, Morgantown, WV 26506-6302.

Limited data is available on the responses of mice to sepsis induced by cecal ligation and puncture (Urbaschek et al., Circ. Shock 14:209, '84). To further characterize this model in SW mice, hepatocyte function was monitored by indocyanine green (ICG) dye extraction and serum albumin, calcium and LDH levels were measured at 0, 1.5, 3, 6 and 12 h at which time animals began to die. The latter measurements were chosen in order to compare with histochemical evaluations of hepatic tissue in progress. To determine the percentage of ICG extraction, an i.v. injection (0.5 mg/kg body wt) was given to each animal twenty minutes prior to collection of cardiac blood for spectrophotometric analysis at 805 nm. Both 3 and 6 h groups showed significant ($P<.01$) retentions of ICG in sera (5% and 19.5%) suggesting failure of dye extraction by hepatocytes. This hepatic dysfunction correlated with decreases in serum albumin, up to 25%, seen at these times. At 12 h albumin remained depressed but sera retention of ICG had returned to normal. However, dye was now being excreted into the urine suggesting that reduced albumin levels may result in loss of unbound ICG through urine. Serum Ca^{2+} was inversely related to albumin levels. LDH levels were significantly increased (85%) by 12 h. We concluded that sepsis in mice, as in other species, results in early hepatocyte damage affecting cellular metabolism, protein synthesis and membrane permeability. Since these changes follow closely drastic reductions in hepatic sinusoidal blood flow (McCuskey et al., Circ. Shock 13:88, '84), hepatocellular hypoxia may be a significant factor contributing to liver failure during sepsis. Supported in part by the Amer. Heart Assoc., WV Affiliate.

61

CHOLECYSTOKININ-8 ANTAGONIZES OPIOID ANALGESIA, BUT NOT ENDOTOXIC HYPOTENSION IN RATS. D. MALCOLM*, S. TEICH*, L. BLACK*, B. CHERNOW, & J. HOLADAY. Walter Reed Army Institute of Research, Washington, D.C. 20307 & Naval Medical Research Institute, Bethesda, MD 20814.

Recently, cholecystokinin octapeptide (CCK-8) has been shown to antagonize opioid-induced analgesia and catalepsy, suggesting that this peptide may function as a physiologic antagonist of endogenous opioid systems. Since the endogenous opioids have been shown to contribute to the acute hypotension associated with models of septic shock, we sought to determine whether CCK-8 treatment would prevent or reverse endotoxin-induced hypotension in conscious rats. Indwelling arterial catheters were connected to a physiograph for continuous monitoring of mean arterial pressure (MAP). An indwelling venous catheter was used for injection of saline or CCK-8 (1, 10 and 100 µg/kg); CCK-8 alone caused a transient (<5 min) increase in MAP (saline vs. CCK-8: 2±2 vs. 13±3, 28±5, 15±3 mm Hg, respectively). Endotoxin (*E. coli* lipopolysaccharide; 30 mg/kg, iv; LD 80) injection caused an acute hypotension (>20 mm Hg drop), followed by a partial recovery and a secondary fall in MAP 45 min post-injection. Following the acute hypotension, administration of the opioid antagonist naloxone (5 mg/kg, iv) significantly improved MAP (20 mm Hg), however CCK-8 (10, 100 µg/kg, iv) was without effect. Furthermore, 15 min pretreatment with CCK-8 at a dose (5 µg/kg, intraperitoneally, ip), which blocked morphine (7.5 mg/kg ip) hot plate analgesia by 60 %, failed to block endotoxic hypotension. Neither treatment significantly affected 24 hr survival. The observation that CCK-8 antagonized opioid analgesia (primarily a µ receptor phenomenon) but failed to block or reverse the acute hypotension of shock (thought to be mediated by δ opioid receptors) suggests that CCK-8 may act to antagonize µ-mediated but not δ-mediated opioid actions *in vivo*.

62

NALOXONE AND RESPIRATORY COMPENSATION IN CONSCIOUS ENDOTOXIC RATS. W.R. LAW*and J.L. FERGUSON. U. of Ill. at Chicago, H.S.C., Chicago, Ill. 60680.

We investigated the effect of naloxone on respiratory compensation for metabolic acidosis in conscious, endotoxic rats. Male, Sprague-Dawley rats (300-400 gm) were surgically prepared with right carotid and jugular vein cannulas under Equi-Thesin (2.2 ml/kg) anesthesia. Twenty-four hours later rats received either E.coli endotoxin (etx; Difco batch #0127:B8, control #706951) or saline. Arterial blood gases and pH (Corning 168) were measured prior to (t=0) and 10, 30, and 60 min. after etx. At 25 min. post-etx rats received either saline (ES group) or 2 mg/kg naloxone (EN group). Means were compared using the

Least Significant Difference test after 2-way ANOVA between time and treatment. Changes in blood gases in ES rats were similar to those we have previously published, wherein PO_2 rose and PCO_2 fell in a stepwise fashion 10 and 30 min. post-etx. Arterial pH declined but was not significantly lowered until 60 min. post-etx. The changes in blood gases in EN rats were not significantly different from those of ES rats at any time measured. However, in contrast to the pH changes in the ES group, no significant alteration in pH was observed in EN rats at any time. In fact, at 60 min. post-etx the pH in EN rats (7.41 ± .02) was significantly greater than that in ES rats (7.19 ± .08). Naloxone alone had no effect on blood gases or pH. These results support our earlier finding that the increased ventilatory drive observed in endotoxic rats is not directly related to changes in arterial pH. The mechanism behind this increased ventilatory drive remains unclear.

63

ENDOTOXIC SHOCK ELICITS GREATER ENDORPHIN SECRETION THAN HEMORRHAGIC SHOCK. A.J. HAMILTON*, D.B. CARR*, P.McL. BLACK.* Massachusetts General Hospital, Boston, MA 02114. Introduced by: John F. Burke.

Endorphins may contribute to the pathophysiology of both endotoxic and hemorrhagic shock. To determine if endorphin secretion was similar in both types of shock, twenty-five sheep were divided into three groups: a saline control group (n=10), an endotoxin(ETX)-treated group (n=9) and a hemorrhaged group (n=6). Each sheep had baseline determinations of mean arterial pressure (MAP) and plasma levels of beta-endorphin-like immunoreactivity (iB-EP). Experimental animals either received ETX 450 ng/kg intravenously or underwent withdrawal of blood volume sufficient to diminish MAP by approximately one third of baseline values. MAP and iB-EP levels were determined every fifteen minutes for five hours. Data were averaged within each group and compared between groups. ETX and Hemorrhage-treated groups both showed a significant fall in MAP; however, this was greater in the hemorrhage-treated group ($p<.05$, t test). ETX-treated animals displayed a mean peak iB-EP level of 1550% above baseline as compared to only 201% in the hemorrhage-treated group. This difference in mean peak iB-EP response between the two shock groups is also significant ($p<.01$, t test). Peak iB-EP levels coincided with trough MAP values in the ETX-treated group while the peak iB-EP lagged behind the onset of trough MAP in the hemorrhage group. These results demonstrate that iB-EP secretory patterns differ in endotoxic versus hemorrhagic shock and suggest that distinct mechanisms of opiopeptide secretion occur in the two shock states.

64

THE CENTRAL NERVOUS SYSTEM (CNS) IS INVOLVED IN THE CARDIOVASCULAR RESPONSES TO NALOXONE (NAL) IN CANINE ENDOTOXIC (ES) BUT NOT HEMORRHAGIC SHOCK (HS). N. J. GURLL, E. GANES, D. G. REYNOLDS. Univ. of Iowa Hospital, Iowa City, IA 52242.

We have previously shown that NAL improves mean arterial pressure (MAP, mmHg), cardiac output (CO, L/min), left ventricular contractility (LV dp/dt max, mmHg x 10^3/sec) and survival in canine ES and HS. To determine involvement of CNS mechanisms, we perfused either artificial CSF or NAL 0.1 mg/kg over 30 min through the (cannulated and vented) IIIrd ventricle of 20 adult mongrel dogs subjected to either HS (bled over 30 min to MAP of 45 mmHg which was maintained by a reservoir for 30 min before perfusion) or ES (endotoxin 1 mg/kg i.v. 15 min before perfusion). NAL in ES improved MAP by 25±4, CO by 0.6±0.1, and LV dp/dt max by 0.8±0.1 which are significantly ($P<.05$) greater than the responses to CSF (5±3, 0.2±1 and 0.2±0.1 respectively). In contrast, the cardiovascular responses to NAL and CSF in HS are slight and not different. Similarly, there was no difference in cardiovascular responses between CSF (N=5) and NAL 0.1 mg/kg (N=5) injected intrathecally into the cisterna magna of anesthetized dogs subjected to HS (MAP at 45 mmHg for 1 hr before injection). Since this dose of NAL given into the coronary circulation in HS is markedly effective, we conclude that NAL works primarily in the CNS in ES but at the heart in HS. (supported by DOD contract DAMD 17-81-C-1177)

65

LATE ADMINISTRATION OF METHYLPREDNISOLONE DOES NOT ENHANCE NALOXONE EFFECT IN HYPOVOLEMIC SHOCK. K. BEAMER*, T. DALY*, T. VARGISH. Dept. of Surgery, West Virginia University, Morgantown, WV 26506.

The late addition of methylprednisolone(MP) to our hypovolemic shock protocol was evaluated to determine whether any hemodynamic enhancement of the naloxone(NAL) effect might be present. Twenty-five dogs, prepared for monitoring, were bled into a reservoir to a mean arterial pressure(MAP) of 40-45 mmHg. MAP was maintained for 45 minutes at which time the reservoir was clamped (t=0). Treatment was initiated with 0.9%NaCl(S) (2.5ml) IV bolus plus IV infusion(2.5 ml/hr) or NAL(2 mg/kg) IV bolus plus IV infusion(2 mg/kg/hr). In two other groups of animals, MP(30mg/kg) was added to the treatment regimen as an IV bolus at t=30 minutes. At t=60 minutes, the infusions were stopped and the shed blood was returned. Hemodynamic measurements were made and plasma endorphin-like activity(PELA) was measured throughout the treatment period. Values are presented as means ± SEM:

		MAP mmHg			CO l/min			PELA p mol/l		
Treatment	N	t=0	t=30	t=60	t=0	t=30	t=60	t=0	t=30	t=60
S	11	40±3	53±7	52±8	.54±.1	.68±.1	.63±.1	193±36	260	199±57
NAL	10	44±1	87±7●+	78±9●	.56±.0	.88±.1●	.78±.1●	164±24	102±23	98±17
S+MP	4	45±3	61±3●	65±9●	.70±.1	.78±.1	.84±.1	105±10	179±26	186±37
NAL+MP	4	44±2	83±9●+	81±9●	.62±.0	.98±.1●	1.0±.1●+	181±48	115±11	128±23

● $p < .05$ when compared with t=0 + $p < .05$ when compared with S group at same time

The addition of MP to the S or NAL treated animals did little to improve their hemodynamic performance or lower PELA.

66

LACK OF EFFICACY OF NALOXONE IN CANINE GRADED HEMORRHAGE. S.C. DRONEN*, R. FOUTCH*, P.A. MANINGAS*. (Introduced by: L. William Traverso) Department of Clinical Investigation, Madigan Army Medical Center, Tacoma, WA 98431.

Animal studies of naloxone's efficacy in hemorrhagic shock have primarily evaluated cardiovascular parameters in reservoir models. This study used a fixed volume hemorrhage model to evaluate naloxone's ability to prevent deterioration of cardiovascular and biochemical parameters during graded hemorrhage. Fifteen mongrel dogs were bled 50% of their estimated blood volumes over one hour. This was followed by a one hour stabilization period, then reinfusion over 30 minutes, and finally, an additional one hour monitoring period. Eight dogs received 2 mg/kg intravenous naloxone 30 minutes prior to hemorrhage and 2mg/kg/hr for the duration of the study period. Seven control dogs received an equivalent volume of saline without naloxone. Pulmonary capillary wedge pressure, central venous pressure, cardiac output, heart rate, blood pressure, arterial and mixed venous blood gases, and serum lactates were measured at 19 intervals throughout the study period. Mean arterial pressure, cardiac index, systemic vascular resistance and oxygen delivery, consumption and extraction were calculated for each sampling period. Overall there were not significant differences between the narcan and control groups in the mean data values ($p<0.05$, two-tailed independent t test). Furthermore, the degree of hemodynamic and biochemical deterioration observed during hemorrhage was not significantly different. Mortality was 25% in the narcan group and 0% in the controls. We conclude that naloxone does not have any beneficial hemodynamic effects in this fixed volume hemorrhage model. Effects shown in prior studies may reflect the use of the reservoir model.

67

SEROTONIN ANTAGONIST AND TRH: BENEFICIAL COMBINATION THERAPY IN HEMORRHAGIC SHOCK. FEUERSTEIN G., ELAM R.* Dept of Neurology, Unif Svcs Univ of the Hlth Sci, Bethesda, MD 20814; *Dept of Pharmacology, Hadassah Medical Sch, Jerusalem, Israel.

Thyrotopin Releasing Hormone (TRH) or serotonin (S) antagonists were shown to enhance cardiovascular recovery after bleeding. Since S and TRH are co-localized in brain nuclei we postulated that S serves as an inhibitory regulator of TRH action. This hypothesis was tested in pentobarbitone anesthetized cats instrumented with arterial catheters and a ventriculo-cisternal perfusion for S and catecholamine and leu^5-enkephalin (LE) assay. Bleeding (25 ml/kg) caused shock, blood pressure (BP) <55mmHg over 1 hr. Methysergide (0.2 mg/kg or the TRH analogue MK-771 (1 mg/kg) improved BP recovery to some degree but M+MK771 had a pronounced effect on BP recover. The M+MK-771 group had also improved survival rate over

MK-771 alone. Potentiation of MK-771 effect on BP was also seen in non-bled cats. Shock caused increase in CSF 5 and LE but decrease in catecholamines.

	Blood Pressure (mmHg)				
Treatment	T-0	T-10	T-30	T-60	0 time = control
Saline	125±5	36±5	51±2	55±4	period; drugs
M	138±5	42±6	73±8+	80±8+	injected after
MK-771	124±9	55±7	90±7*$	94±5*$	bleeding
M+MK771	130±9	55±7	118±6$	111±4$	T-10.n=6-9.

These studies indicate that combination therapy of S antagonist and TRH analogue might be a useful therapy in hemorrhagic shock.

68

CHANGES IN PLASMA ENDORPHINS DURING THERAPY OF CANINE HEMORRHAGIC SHOCK WITH THYROTROPIN RELEASING HORMONE. H.V. DEDHIA, L. TEBA, M. ZAKARIA, K.C. BEAMER. West Virginia University Medical Center, Morgantown, WV 26506. (Introduced by: Thomas Vargish)

Endogenous opiates seem to have a role in the pathogenesis of shock. Ten dogs anesthetized with I.V. pentobarbital were bled to a MAP of 50 mmHg and maintained for half an hour. Then, the animals were divided into two groups: A) received TRH 2 mg/Kg IV, B) received placebo (D5W). Cardiac output and hemodynamics were recorded. Arterial blood was collected for Beta endorphin (END) measurements. Samples were obtained before shock was induced, after half an hour of being in shock and at 15, 45, 75, 120 min. after TRH or placebo was given. The blood samples were analyzed in duplicate by radioimmunoassay. B END levels were compared and analyzed by Student's t test. Mean sequential values, ± SD of END are shown in the table and expressed as pmol/L. A significant increase in plasma END levels was observed in both groups after shock ($p < .05$). In the TRH treated group, a progressive decline in END was seen reaching statistical significance at 2 hours ($p < .05$). These changes were not observed in the placebo group. Our conclusions are: 1) A significant increase in plasma B- endorphin is seen during canine hemorrhagic shock and may play an important role in its pathogenesis. 2) Treatment with TRH causes a significant drop in endorphins level during hemorrhagic shock.

	Pre shock	after ½ hr of shock	15'	45'	75'	120'
TRH group(n=5)	14±8	122±92	63±38	55±59	75±47	23±16
Placebo group(n=5)	13±19	52±17	54±30	73±44	35±21	50±69

69

TRIIODOTHYRONINE (T_3) IMPROVES SURVIVAL IN CANINE HEMORRHAGIC SHOCK. H. SHIGEMATSU*, R.A. SMITH*, C.H. SHATNEY. University Hospital and Memorial Medical Center, Jacksonville, FL 32209

It is well documented that the euthyroid sick ("low T_3") syndrome occurs in circulatory collapse. Other investigators have noted beneficial hemodynamic effects of T_3 infusion in septic shock. To evaluate the potential therapeutic value of T_3 in hemorrhagic shock, 26 anesthetized (30 mg/kg pentobarbital), heparinized (200 U/kg) mongrel dogs were bled rapidly into a reservoir to a mean arterial pressure of 40 mmHg. After 60 min of hypotension animals were given 15 µg/kg of T_3 (13 dogs) or an equal volume of saline (13 dogs), and the reservoir line was clamped for 30 min. The shed blood was then reinfused over 30 min. After one hour of monitoring the dogs were returned to the kennel and observed for 3 days. Hemodynamic data were obtained via systemic and pulmonary arterial catheters. T_3 administration caused significant increases during the clamped period in cardiac output, stroke volume, mean arterial pressure, right and left ventricular stroke work and systemic vascular resistance, with a decrease in pulmonary vascular resistance. After reinfusion of shed blood, hemodynamic measurements revealed no significant intergroup differences, except for an increased pulmonary artery wedge pressure in the T_3 dogs. In the control group, 6 of 13 dogs died. However, only one of 13 T_3 dogs died ($p < 0.05$). In our previous studies, both thyroidectomy and TRH (thyrotropin releasing hormone) treatment improved survival in canine hemorrhagic shock. Although T_3 therapy might exert effects via the pituitary-thyroid axis, the possibility also exists that T_3 improves survival by acting on cardiovascular receptors to improve hemodynamic function at a critical stage of shock.

70

INVESTIGATION OF THE HYPERTENSIVE EFFECT OF TRH IN RABBIT HEMORRHAGIC SHOCK. J.A. SAMPSON*, B.L. BASS*, J.W. HARMON, J. HOLADAY. Walter Reed Army Institute of Research, Washington, D.C. 20307.

TRH has recently been shown to increase blood pressure in both hemorrhagic and endotoxic shock. This could be the result of increasing cardiac output (CO) or systemic vascular resistance (SVR). To differentiate these two possibilities we investigated the action of this drug in a model using conscious male New Zealand rabbits. Using radio-labelled microspheres and pressure transducers, we studied the changes produced in CO, mean arterial pressure (MAP) and calculated SVR. Following controlled hemorrhage where 30% of the estimated total blood volume was removed over 15 minutes, MAP fell to 51 $\pm$ 8 mmHg. Injection of TRH (2 mg/kg) produced a 100% increase in MAP post-treatment (103 $\pm$ 6 mmHg). This rise was not accompanied by an increase in CO, (200 $\pm$ 36 ml/min) pretreatment versus post treatment (202 $\pm$ 20 ml/min). However we found a marked increase in SVR from pre-treatment (281 $\pm$ 27 mmHg.min.L^{-1}) to post treatment values (545 $\pm$ 63 mmHg.min.L^{-1}). With the finding of an increased vascular resistance, we were concerned about the uniformity of this "pressor" effect on the vascular beds of critical organs. TRH had a tendency to decrease the percentage of CO to the small bowel, large bowel and kidneys. The blood flow distribution to the brain and heart were not decreased but instead showed a relative increase. Thus in the conscious rabbit model, TRH produces an increase in MAP, predominantly by elevating SVR. TRH's effects of increasing resistance with the constellation of a decreased flow to the GI tract and kidneys with sparing of the CNS and heart suggest the effects are mediated through the sympathetic nervous system.

71

INFLUENCE OF ANESTHETICS ON THE HEMODYNAMIC AND METABOLIC EFFECTS OF THYROTROPIN RELEASING HORMONE IN RATS. C.F. SCHAEFER, M.R. LERNER*, M.L. WILSON*, D.J. BRACKETT, P. TOMPKINS*, J.W. HOLADAY, L. FAGRAEUS, M.F. WILSON. Depts. Anesthesiology and Medicine, Univ. of Oklahoma Health Sciences Ctr., VA Medical Center, Oklahoma City, OK 73190 and Walter Reed Army Inst.Res., Washington, D.C. 20012.

Thyrotropin releasing hormone (TRH) has received much recent attention as a potential therapeutic agent in shock. Little is known regarding the hemodynamic effects of TRH under anesthesia. As part of a series of experiments related to TRH and endotoxin shock, we studied the effects of 2 mg/kg i.v. bolus of TRH on central hemodynamics, respiratory rate (RR), blood gases, and metabolic parameters in spontaneously breathing rats either awake (A) or anesthetized with enflurane (E, 2% E in O_2) or ketamine (K, 60 mg/kg/hr, i.v. and 100% O_2). Instrumentation was performed under E (for E and A) or K anesthesia after endotracheal intubation; catheters were implanted to measure blood pressure (BP), cardiac index (CI) by thermodilution, heart rate (HR), blood gases, hematocrit, glucose, and lactate. After a 20 min control period TRH was given and readings were continued for 250 min. Marked respiratory depression occurred with E ($\downarrow$RR, $\downarrow$pH, $\uparrow PCO_2$) which was reversed by TRH within 5 min but gradually returned as RR decreased to control by 30 min. No respiratory depression was noted with K relative to A and only a small increase in RR occurred with TRH in either K or A. Possibly due to greater cardiovascular depression by E than by K, stimulatory effects of TRH were quite notable with E ($\uparrow$BP, $\uparrow$HR, $\uparrow$CI) but less with K and least awake. In conclusion, TRH-stimulation of respiration and hemodynamics was most evident under anesthesia, particularly when the anesthetic depressed breathing and circulation.

72

EVALUATION OF VASOPRESSIN AND CATECHOLAMINE RELEASE AND HEMODYNAMIC RESPONSES AFTER TRH IN THE CONSCIOUS INSTRUMENTED RAT. D.J. BRACKETT, P. TOMPKINS*, M.R. LERNER*, M.L. WILSON*, C.F. SCHAEFER, J.W. HOLADAY, L. FAGRAEUS, M.F. WILSON. VA Medical Center, Depts. Anesthesiology and Medicine, Univ. of Oklahoma Health Sciences Center, Oklahoma City, OK 73190 and WRAIR, Washington, DC 20012.

This study was designed to investigate the mechanisms of the responses elicited by thyrotropin releasing hormone (TRH) to aid in clarifying its effect during shock. Sprague-Dawley rats were instrumented for measurement of cardiac output (CO) by ther-

mal dilution, arterial pressure (AP), central venous pressure (CVP), and heart rate (HR). In conscious animals arterial blood samples were obtained at control 5, 30, 60, and 240 min after TRH (2.0 mg/kg, i.v.) for measurement of plasma vasopressin (VP) radioimmunoassay (RIA) and catecholamine (CA) radioenzymatic assay (REA) concentrations. TRH did not affect VP release during the 4 hr observation period. CA measurements revealed a significant increase in epinephrine (EPI) plasma concentrations at 5 min returning to baseline for the period from 30 to 240 min. There was no increase in norepinephrine. Concurrent with the EPI increase was a significant rise in AP, HR, and respiration rate (RR). The AP returned to control by 5 min, but HR and RR did not return to control until 20 min. For the remaining 4 hrs there was no change in these parameters. There was no apparent effect on CO or CVP by TRH. The rapid transient pressor response to TRH in this study was associated with release of EPI, without an increase in plasma VP concentrations. Whether this response pattern is important in the action of TRH during shock, a situation which already features highly elevated CA concentrations, or if TRH affects other vasoactive mediators remains to be investigated.

73

NEUTROPHIL MEMBRANE-POTENTIAL CHANGES INDUCED BY LATEX AND PREOPSONIZED ZYMOSAN DIFFER. G.E. BROWN*, M.E. LANSER. MD Inst. Emer. Med. Ser., Univ. MD Cancer Center, Baltimore, MD 21201.

Neutrophil bactericidal activity is decreased following trauma and during sepsis. Membrane depolarization is the initial step, following receptor binding, in a series of reactions resulting in bactericidal oxygen species. A flow cytometric analysis of the neutrophil membrane depolarization response to phagocytosable targets has been performed. Purified normal neutrophils (1 x 10^5 ml) preloaded with a fluorescent membrane-potential indicator dye (3,3'-dipentyloxacarbocyanine) were exposed either to 10 ul of washed latex beads (0.81 u) or 0.5 mg of preopsonized zymosan. The membrane-potential of individual cells was followed by flow cytometry. Latex particles induced a large irreversible loss of dye from all cells. This response is qualitatively similar to that seen after addition of 1.6 uM of the secretagogue 12-O-tetradecanoylphorbol 13-acetate (TPA). In contrast, preopsonized zymosan induced a transient loss of dye from only 30% of the cells. Thus, although both phagocytic targets induced respiratory activation and superoxide production, the earliest known event in that process, altered membrane potential, appeared to differ both qualitatively and quantitatively. The stimulus specific pattern of membrane depolarization may in part explain the effects of trauma septic serum on oxidative metabolism induced by a variety of particulate and soluble stimuli. Flow cytometry in conjunction with fluorescent membrane-potential sensitive dyes promises to be a useful tool in investigating normal and altered phagocytic function following trauma and sepsis.

74

ADDITION OF FIBRONECTIN TO SEPTIC TRAUMA SERUM DEPRESSES NEUTROPHIL CHEMILUMINESCENCE (CL). M. E. Lanser, R. Mora, MD. Inst. Emerg. Med Serv.,Balto. MD 21201

Septic trauma serum contains an abnormal protein that suppresses the CL response of normal neutrophils. This suppressive factor reversably binds to neutrophil membranes and prevents membrane-ligand interaction. We hypothesized that addition of fibronectin (FN) would amplify the serum suppressive activity of septic sera by increasing the membrane-binding of the suppressor. Such amplification might permit earlier detection of serum suppressive activity. Eight serum samples (125 µl) from 3 multiple-trauma patients, drawn between 2 and 17 days prior to the detection of clinical sepsis (presepsis) were incubated with normal neutrophils ($1x10^6$) and increasing amounts of FN. CL was initiated by the addition of preopsonized zymosan. The 15 min integral CL response (millivolts) was compared to that obtained using nonseptic sera:

FN added (µg/ml) =	0	3	6	12
Nonseptic (N=8)	44985±3813	45933±4600	44761±5518	39839±5310
Preseptic (N=8)	41231±5950	38727±5709	39120±6024	25212±7230*†

*$p<0.05$ compared to 0; †$p<0.05$ compared to nonseptic.

The addition of FN to nonseptic sera did not alter the CL response. In contrast, the addition of increasing amounts of FN to preseptic serum significantly

($p<0.05$) depressed the CL response. Thus, employing CL, the addition of FN to trauma serum may be used to detect the future development of sepsis. The exact mechanism by which FN causes such a marked depression of the CL response in preseptic sera remains to determined.

75

BLOOD LEVELS OF INFLAMMATORY MEDIATORS IN TRAUMA PATIENTS - RELATION TO POSTTRAUMATIC COMPLICATIONS. H. REDL*, E. PAUL*, J.K.S. NUYTINCK**, R.J.A. GORIS**, P.J.J.v. MUNSTER***, G. SCHLAG* * LBI exp. Traumatol., Vienna, ** Dept. Gen. Surg., *** Dept. Pediat., St Radboud Hosp., Nijmegen

It has been suggested that lysosomal enzymes, and toxic oxygen products, released from activated granulocytes play an important role in the pathophysiology of the Adult Respiratory Distress Syndrome (ARDS) and possibly of Multiple Organ Failure (MOF).

In a series of 71 trauma patients we measured the inflammatory parameters CH_{50}, C_{3d}, C_{3a}, C_{5a} and the granulocyte enzyme elastase - antiprotease complex, at admission, after 24 hours and subsequently at weekly intervals. We investigated their relation to the severity of injury, as measured by the Injury Severity Score (ISS), to the incidence of ARDS and to the incidence and severity of MOF (own scoring system).

At admission ($r=0.413$, $p<0.01$) and after 24 hours ($r=0.482$, $p<0.01$) ISS correlated best with elastase. At admission, elastase was the best predictor for incidence of ARDS ($p<0.001$). Elastase persisted to do so 24 hours after the injury ($p<0.005$). Only elastase correlated well with the severity of MOF ($r=0.0685$, $p<0.001$ at day 9; $r=0.772$, $p<0.005$ at day 16; $r=0.791$ and $p<0.05$ at day 23) suggesting that in trauma patients granulocytes are activated presumably because of complement activation, though no sign. correlations for C_{3a}, C_{5a}, and only some for C_{3d}, CH_{50} could be found. Moreover, plasma elastase may be of great value in monitoring trauma patients as to the development of posttraumatic complications such as ARDS and MOF.

76

LEUKOCYTE FUNCTION AND RESPIRATORY FAILURE AFTER SEVERE TRAUMA. G.REGEL,J.A.STURN,A.DWENGER F.SCHWEITZER,D.H.WISNER,H.J.OESTERN. Dept.Trauma Surgery,Hannover Medical School, Germany.

It is known that the polymorphonuclear leukocytes(PMN) play a major role in the development of ARDS after blunt trauma. It is a matter of some debate whether the increased pulmonary capillary permeability associated with ARDS is a consequence of suppression or overactivation of the PMN function. We compared whole blood chemiluminescence (CL) with the following clinical parameters: Oxygen quotient (paO_2/FIO_2), dynamic compliance(COMPL) and extravascular lung water(EVLW). A total of 17 severely injured patients were prospectively followed up to 14 days, to determine whether there are any differences in PMN function between survivors (s) and non-survivors (ns). The survivor group (n=8) showed a significant increase in the peak maximum(CLPM) and a significant decrease in peak-time(CLPT) as compared to controls. Patients who ultimately died of respiratory failure (ns) demonstrated significantly longer peak-time as compared to both controls and survivors. The changes in CL were seen beginning 2 days post trauma. Changes in the measured clinical parameters were apparent 4 days post trauma.

Survivors (n=8)	CL-PM	CL-PT	paO_2/FIO_2	COMPL	EVLW
days after trauma	($\times 10^6$ cpm)	(sec)		(ml/cm H_2O)	(ml/kg BW)
2	0.47±0.04	25.9±1.11	380.2±85.2	51.9±11.1	6.92±1.25
4	3.23±2.39	21.38±1.26	366.2±66.9	57.0±16.7	6.39±0.9
7	2.56±2.00	19.9±3.64	380.3±105.5	56.5±14.6	8.06±2.5
Non-survivors (n=9)					
2	1.21±1.02	25.65±0.64	297.0±85.3	46.1±8.0	7.83±2.63
4	1.52±1.17	25.98±2.05	257.9±85.3	40.1±11.8	11.48±5.72
7	0.98±0.77	25.4±3.35	143.6±77.1	24.2±10.3	19.62±7.9

Peak-maximum of CL is a sign of the phagocytic activity of the PMN. Length of peak-time is inversely related to the degree of opsonizing ability. Both parameters reflected disturbed function in the ns group. We concluded that severe trauma leads to a suppression of the PMN and a decrease in opsonizing ability before respiratory failure appears.

77

ENDOTOXIC AND HYPEROXIC INJURY IN CULTURED HUMAN ENDOTHELIAL CELLS. H. LEVENTHAL*, D. SILVERMAN*, L. MARZELLA*. (Introduced by Dr. R Adams Cowley) Department of Pathology and M.I.E.M.S.S., University of Maryland, Baltimore, MD 21201.

Endotoxin protects against hyperoxic lung injury in rats. This protection is associated with an elevation of antioxidant enzymes in the lung. However, the precise mechanism of the protective effect and the cell(s) responsible are not known. We have tested the effects of endotoxin and hyperoxia on cultured human umbilical vein endothelial cells. Cells were plated ($3x10^5$) on Corning 35 mm^2 dishes in medium 199 with Earle's salts, 20% heat inactivated human serum, and a bicarbonate buffer. Cells were gassed with 95% air-5% CO_2 (controls) or 95% O_2-5% CO_2 (hyperoxia). E. coli 025:B6 lipopolysaccharide (LPS) was added to a dose of 1, 5 µg/ml (low) or 100 µg/ml (high). The percentage of dead cells attached or in suspension is shown below. Number of experiments is shown in parentheses.

Time	Control	LPS-Low	LPS-High	Hyperoxia	LPS-Low and Hyperoxia	LPS-High and Hyperoxia
24 hr	1.4 (5)	5.1 (3)	3.2 (5)	5 (5)	3.4 (3)	6.4 (5)
48 hr	4.2 (1)	5.7 (1)	9.9 (1)	8.5 (1)	15.4 (1)	8.1 (1)
72 hr	1.6 (5)	3.8 (3)	11.8 (5)	9.6 (5)	5.4 (3)	8.2 (5)
7 days	2.2 (5)	2.9 (2)	18.1 (4)	11.9 (5)	2.8 (3)	12.7 (4)

Assessment of injury by phase contrast and by transmission electron microscopy showed similar results. We conclude that the presence of low doses of endotoxin tends to protect endothelial cells against hyperoxia. In the presence of high dose endotoxin and hyperoxia, cell killing is lower than in high dose endotoxin alone.

78

INFLUENCES OF PHAGOCYTIZING LEUKOCYTES ON CULTIVATED RAT HEPATOCYTES. F. HIRAI*, H. AOYAMA*, T. SATO, I.K. BEREZESKY*, AND B.F. TRUMP*. Department of Pathology, University of Maryland School of Medicine, Baltimore, MD 21201.

From the standpoint of cellular injury, much attention has recently been focused on the contribution of superoxides (SR) generated by phagocytizing leukocytes in severe infectious insults. The purpose of this study was to investigate what influences were noted in cultivated monolayer hepatocytes (H) exposed to activated leukocytes (L) by heat-killed E. coli (EC). Changes were evaluated using two parameters specific for the liver; namely, % OCT release as an index of cell integrity and % urea synthesis as an index of function. After the initial urea synthesis assay, each dish was divided into: 1) H+L+EC; 2) H+L; 3) H+EC; and 4) H with or without SOD. At 6, 12, and 24 hrs later, supernatants were collected for OCT assay and urea synthesis measurement was performed. 100% cell destruction was accomplished using saponin. These data suggest that 1) the effect of SR appears late; 2) SOD has little beneficial effect; and 3) functional enhancement is observed despite integral damage which is in accordance with that seen in in vivo sublethal bacteremic rat models. A,C,D = SOD (+); B,D,F,H = SOD (-). (NIH GM 32084)

	% Control Ratio in % OCT Release								% Control Ratio in % Urea Synthesis							
	1		2		3		4		1		2		3		4	
	A	B	C	D	E	F	G	H	A	B	C	D	E	F	G	H
6 Hr:	97	98	147	115	112	107	109	100	110	117	114	112	90	96	96	100
12 Hr:	70	68	69	72	91	90	95	100	139	138	140	135	135	121	100	100
24 Hr:	189	171	155	135	104	105	91	100	118	126	123	93	93	93	100	100

79

LIPID PEROXIDATION AND ACUTE LUNG INJURY FOLLOWING SYSTEMIC COMPLEMENT ACTIVATION. G.O. Till, J.R. Hatherill, P.A. Ward. Department of Pathology, University of Michigan Medical School, Ann Arbor, MI 48109.

Previously we have demonstrated that systemic complement activation in rats following skin burns or intravenous injection of cobra venom factor (CVF) causes acute lung microvascular injury which is related to neutrophil-derived oxygen radicals. Our current studies show that acute lung injury in CVF-treated rats is paralleled by the appearance of lipid peroxidation products (conjugated dienes, lipid hydroperoxides, fluorochromic substances) in both plasma and tissue of lungs but not liver, kidney or spleen. In thermally injured rats, conjugated dienes appear sequentially both in the burned skin (at 15 min) and in the lungs (at 20 min) as well as in the plasma (with peaks at 30 and 180 min postburn). The appearance of lipid peroxidation products in plasma of both thermally injured and CVF-treated rats is greatly diminished if the animals are depleted of their blood neutrophils or

pretreated with catalase, iron chelators or scavengers of hydroxyl radical. Furthermore, the interventions that prevent lipid peroxidation also protect from acute lung injury. Our data suggest a linkage between activation of the complement system, generation by blood neutrophils of hydroxyl radical, lung injury, and appearance in tissues and plasma of products of lipid peroxidation. (Supported in part by NIH grants GM28499, GM29507, HL28442 and HL31963).

80

ENDOTOXIN-INDUCED LUNG INJURY:ROLE OF OXIDANTS AND EFFICACY OF STEROID. T.MATSUDA*,H. MIYASHITA*,N.KOJIMA*,K.EGAMI*,K.ADACHI*,K.YAMASHITA*,M.ONDA*,O.KAWANAMI*,(Introduced by:H.Hirasawa). Deps. of Surgery and Pathology,Nippon Medical School,1-1-5,Sendagi, Bunkyo-ku,Tokyo,113,JAPAN.

Our previous study demonstrated that leukocyte-derived oxidants produced acute lung injury in the endotoxin-administrated guinea pigs. In this study we evaluated an efficacy of steroid of these animals by the following methods. Guinea pigs (200g,male) received 0.7mg/kg of E.coli endotoxin(ET) in the peritoneal cavaty (Group-I) and the animals of Group-II received 30mg/kg of methylprednisolone(MP). The animals of Group-III had the MP followed by intraperitoneal injection of ET. Animals were sacrificed 30,60,180 min of ET or saline injection. Peripheral leukocytes counts and bronchoalveolar lavage(BAL) cell differentials were made. In order to analyse the activities of superoxide dismutase(SOD) and malonaldehyde(MDA),Pyrogallol and TBA methods were applied respectively. Significant leukopenia occurred in each group. In Group-I leukocytes especially eosinophils were recovered by BAL and the total cell number of the BAL fluid increased at 180 min of ET injection,but remarkable increase was not found in other two groups. SOD activity in each group diminished below the control level. MDA remarkablly increased at 60 min of ET injection in Group-I. In contrast the amount remained at the control level in both Group-II and III. Accordingly histlogical examinations revealed marked edematous changes in the alveolar structures in Group-I, but not significant in Group-II and III. These results suggest that ET certainly provokes vascular damages in the lung and MP pretreatment minimized the injury preventing edema formation. MP might have potential roles for altering ET-induced leukocyte chemotaxis and diminishing oxidants-induced lung injury in a quantitative manner.

81

A NEW SYNTHETIC COMPLEMENT INHIBITOR AND SEPTIC ADULT RESPIRATORY DISTRESS SYNDROME (ARDS) IN RATS. S. ODA*, H. HIRASAWA, S. KOBAYASHI*, M. OHKAWA, M. ODAKA*, H. SATO*
Department of Surgery, Department of Emergency and Critical Care Medicine, Chiba University School of Medicine, Chiba, Japan

It has been claimed that pulmonary neutrophil sequestration caused by activated complement plays an important role in pathophysiology of septic ARDS. The present study was undertaken to investigate the effect of a new synthetic complement inhibitor, FUT-175 (2-(6-aminido)naphthyl-4-guanidinobenzoate 2HCl), on complement level, pulmonary neutrophil sequestration, extravascular lung water (EVLW) and thromboxane B2 level in septic rats. Sepsis was produced by cecal ligation and punctures (CLP) in male Wistar rats. FUT-175 was infused intravenously in 5% dextrose at a dose of 4 mg/kg in the volume of 1 ml immediately after CLP, 4, 8 and 12 hrs after CLP, respectively (FUT group). The control septic rats received 1 ml of 5% dextrose at the same time to FUT group (control septic group). Complement C3 level and pulmonary ^{51}Cr labeled neutrophil counts were measured at 6 and 16 hrs after CLP. Thromboxane B2 and EVLW were also measured at 6 hrs after CLP. Among control septic rats C3 level at 6 hrs after CLP was 39.1 $\pm$ 0.5% of normal rats and neutrophil count in lung was 3.2 $\pm$ 0.3%TO/ID (normal rats: 1.01 $\pm$ 0.26). In FUT group C3 level was 52.0 $\pm$ 5.9% of normal rats and neutrophil count in lung was 2.6 $\pm$ 0.2, respectively. Therefore the complement activation and pulmonary neutrophil sequestration produced by sepsis were beneficially affected with FUT-175 treatment. The increases in thromboxane B2 level and in EVLW among septic control rats were also prevented among FUT-175 group. These data suggest that FUT-175 could be beneficial for the treatment of septic ARDS.

82

INTERACTION OF PROSTAGLANDINS, ACTIVATED COMPLEMENT, AND GRANULOCYTES IN CLINICAL SEPSIS AND HYPOTENSION. G.J. SLOTMAN, K.W. BURCHARD, J.A. WILLIAMS*, A. D'AREZZO*, S.A. YELLIN*. Dept. of Surgery, Brown University, Prov., RI 02903

Circulating complement, thromboxane A_2, prostacyclin, and activated granulocytes have been implicated in the hemodynamic dysfunction of sepsis. The purpose of this study was to evaluate the interaction of plasma levels of complement components C3a and C5a, thromboxane B_2 (TxB), prostaglandin 6-keto-$F_{1\alpha}$ (PGI) and granulocyte aggregation (GA) in clinical sepsis. Forty-five patients in the surgical intensive care unit were followed for up to 10 days. Daily arterial blood samples were analyzed for plasma C3a, C5a, TxB, and PGI, pg/ml, by radioimmunoassay. GA, % maximum aggregation of zymosan activated plasma standard curves, was performed with patient plasma and normal donor purified human leukocytes. Patients were studied in 3 groups: non-septic (n=16), septic (n=16), and septic shock (systolic BP <90 mmHg, n=13). Results:

	C3a	C5a	PGI	TxB	GA
NON-SEPTIC	868±230*	18±3	336±107*	102±118	16.5*+
SEPTIC	728±106*	18±3	232±43*	189±49	66
SEPTIC SHOCK	1439±209	14±2	751±362	191±37	60

*p<0.05 vs. Septic Shock +p<0.05 vs. Septic and Septic Shock ANOVA

Plasma C3a and PGI are increased in clinical septic shock, but not in normotensive sepsis or in non-septic patients. TxB appears unrelated to sepsis or hypotension. GA is increased in sepsis but does not correspond with plasma C3a and C5a.

83

GRAM-NEGATIVE BACTERIA ACTIVATE GRANULOCYTES WITHOUT COMPLEMENT OR PROSTAGLANDINS. S.A. YELLIN*, F.L. GARRITY*, G.J. SLOTMAN. DEPARTMENT OF SURGERY, BROWN UNIVERSITY, Rhode Island Hospital, Providence, RI 02903.

Cardiorespiratory dysfunction in gram-negative septicemia may be mediated by circulating complement, activated leukocytes, prostaglandins, or by a direct effect of endotoxin. The purposes of this study were to determine if bacteria produce these substances and to evaluate the direct effects of bacteria on granulocyte aggregation (GA). 10^9 E. Coli and Aeromonas hydrophila were incubated in broth to a concentration of 10^9/ml. Broth was filtered and analyzed by radioimmunoassay for complement components C3a and C5a, thromboxane B_2 (TxB), and prostaglandin 6-keto-$F_{1\alpha}$ (PGI) and by the Limulus amoebocyte lysate test (LAL) for endotoxin. GA, % of maximum zymosan activated aggregation, was performed with broth and normal purified human leukocytes in HBSS. Organisms were incubated in broth (B), broth + 0.135 mg/ml arachidonic acid (BA) and broth + arachidonic acid + indomethicin (BAI). Broth alone was control (C). Results: C3a, C5a, TxB, and PGI were not detectable in C broth or in any E. Coli or Aeromonas filtrate. LAL was positive in all bacterial filtrates, but negative in C broth. No GA response was produced by C broth. GA from E. Coli (B=60%, BA=72%, BAI=56%) and Aeromonas hydrophila (B=56%, BA=40%, BAI=68%) were strongly positive. E. Coli and Aeromonas hydrophila do not release TxB or PGI in detectable quantities. Endotoxin-positive broth stimulates GA in the absence of C3a and C5a. GA is mediated directly by endotoxin or by some other bacterial product.

84

HUMORAL FACTORS POTENTIATING DEATH IN PRIMATE ENDOTOXIC SHOCK. E.C. POHLSON,* M. MAY,* F. SIRI,* J.J. McNAMARA. Cardiovascular Research Lab, Queen's Medical Center, Honolulu, HI 96813.

In endotoxic shock numerous enzyme systems and biologically active substances are released from the cell and thought to contribute to morbidity and mortality. It is unclear whether or not these substances, when activated, are capable of self-propagation with resultant further tissue injury or require endotoxin as a catalyst. To evaluate this question 20mg/kg E. coli endotoxin was dissolved in normal saline (1mg/cc) and infused into 7 Rhesus macaques (toxin donor, TD). An additional 7 R. macaques received an equivalent volume of normal saline (control donor, CD). Animals were monitored for 12 hrs. Lactated Ringer's was infused to maintain a MABP of 40mmHg when necessary. Donor animals were exsanguinated and plasma ex-

tracted. Limulus lysate assay was utilized to determine the approximate amount of endotoxin present in TD plasma and this volume of endotoxin (.125mg/kg) was added to CD plasma. Plasma was infused into 7 toxin receiver (TR) and 7 control receiver (CR). Receiver animals were monitored for 24 hrs and then returned to their cages for 3 days or until death. Lungs were evaluated histologically for evidence of acute injury. Five of 7 TR developed hemorrhagic alveolar edema and died within 3 days. The other 2 TR were sacrificed at 3 days and edema was not present. Two of 7 CR developed nonhemorrhagic alveolar edema and none expired prior to the end of the 3-day survival watch. Our data suggests a humoral factor present in the plasma of TD capable of promoting severe tissue injury and death of recipient animals independent of endotoxin.

85

ROLE OF MEDIATORS IN GUINEA PIG ANAPHYLACTIC SHOCK. H. Darius, D.J. Lefer, J.B. Smith and A.M. Lefer. Department of Physiology, Jefferson Medical College, Thomas Jefferson University, Philadelphia, PA 19107.

The role of leukotrienes (LT) and thromboxane (Tx) A_2 was investigated in isolated lung parenchymal strips from ovalbumin (OA) sensitized and control guinea pigs and in anesthetized animals in vivo. When ovalbumin (40 mg) was injected into sensitized guinea pigs, they developed anaphylactic shock and died within 10 min due to a decrease in MABP to 5 mm Hg, at a time when circulating platelet count dropped 68 ± 15%. Pretreatment of the animals with the Tx-receptor antagonist, SQ-29,548 (3 mg/kg), or the leukotriene-receptor antagonist, FPL-55,712 (3 mg/kg), did not change the fatal outcome or the thrombocytopenia. LTC_4 (2-10 ng/ml) induced contractions of lung parenchymal strips were accompanied by release of TxA_2, measured by radioimmunoassay of TxB_2. Contractions and TxB_2 concentrations in the bath were diminished 80 to 90% after preincubation with SQ-29,548, the Tx-synthetase inhibitor CGS-13,080, and cyclooxygenase inhibitors. This was specific for LTs, because contractions induced by histamine (100-500 ng/ml) did not increase TxB_2 concentrations in the bath and were unaltered by blocking agents. We observed no potentiation of the bronchoconstrictor effects of histamine or LTs in pulmonary strips from sensitized animals. However, addition of OA (20 µg/ml) to the lung parenchymal strips was followed by a marked contraction which was only slightly reduced by Tx, LT, histamine or serotonin blockers. Thus, immunologically induced contractions of isolated lung parenchymal strips are probably not dependent on the release of TxA_2 or LTs from lung tissue. The anaphylaxis of OA appears to be due to release of some other mediator or interaction between mediators.

86

ABDOMINAL SURGERY, BURN INJURY, AND BONE FRACTURE ENHANCE SYSTEMIC PEPTIDE LEUKOTRIENE CONCENTRATIONS IN THE RAT. C. DENZLINGER*, S. RAPP*, W. HAGMANN*, D. KEPPLER*. (Introduced by: A.M. Lefer). Biochemisches Institut, University of Freiburg i. Br., D-7800 Freiburg, West Germany.

Until now the hypothesis of leukotriene (LT) involvement in trauma is mainly based on pharmacological studies showing beneficial effects of inhibitors of LT biosynthesis or action. We provide direct evidence for enhanced systemic peptide LT production in the rat following severe tissue damage. LT concentrations were measured by the combined use of high-performance liquid chromatography and radioimmunoassay in bile and plasma, where N-acetyl-LTE_4 and LTE_4 were the predominant metabolites, respectively. Peptide LTs were rapidly eliminated from blood plasma into bile. Biliary N-acetyl-LTE_4 in controls amounted to 7.8 ± 6.1 nmol/l (mean ± SD); abdominal surgery, burn injury, and bone fracture resulted in LT concentrations of 62 ± 23, 86 ± 49, 121 ± 54 nmol/l bile, respectively. Blood plasma LTE_4 after abdominal surgery amounted to 1.7 ± 1.0 nmol/l. It is proposed that enhanced systemic peptide LT concentrations mediate some of the pathophysiological events accompanying severe tissue damage, such as enhanced vascular permeability, respiratory and circulatory dysfunction.

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87

ANTI-SHOCK ACTIONS OF LY-171883, A NEW LEUKOTRIENE D_4 RECEPTOR ANTAGONIST. Carl E. Hock and Allan M. Lefer. Department of Physiology, Jefferson Medical College, Thomas Jefferson University, Philadelphia, PA 19107.

The peptide leukotrienes are well suited for a role as mediators of circulatory shock and ischemia. We studied the effects of a specific LTD_4 receptor antagonist (LY-171883) on circulatory shock following trauma. Anesthetized rats were subjected to 575 revolutions in a Noble-Collip drum which resulted in a shock state characterized by a significant reduction in mean arterial blood pressure and heart rate ($p < 0.001$), a six-fold increase in plasma cathepsin D activity, a four-fold increase in plasma myocardial depressant factor (MDF) activity and a high mortality rate. LY-171883 was administered 10 minutes after the induction of trauma as a bolus (2 mg/kg) followed by an infusion (2 mg/kg/h) up to 4.8 hours. LY-171883 significantly attenuated the pressor effect of LTD_4 in anesthetized control rats. Administration of the antagonist had no significant effect on the release of the lysosomal enzyme cathepsin D (13.6 ± 1.5 vs 15.6 ± 2.3 U/ml) vehicle vs drug, respectively. However, LY-171883 significantly attenuated the accumulation of MDF in the plasma of traumatized rats. Furthermore, the antagonist signifcantly prolonged survival time in this severe shock model (1.7 ± 0.3 vs 2.7 ± 0.2 h, $p < 0.02$, vehicle vs drug, respectively). In addition, traumatized rats given a higher dose of the antagonist (4 mg/kg) survived 3.4 ± 0.6 h ($p < 0.01$) and maintained significantly lower circulating MDF activities than those given vehicle. Previous data from our laboratory, using lipoxygenase inhibitors, suggests a role for lipoxygenase metabolites in circulatory shock. The present results using a specific LTD_4 receptor antagonist indicate that peptide leukotrienes are important mediators of the pathogenesis of traumatic shock.

88

Diethylcarbamazine, a Leukotriene Inhibitor, Improves In-vitro Microcirulatory Flow During Endotoxemia. R. Dunn*, F. Rogers*, P. Nolan*, A. Phuangsab*, J. Barrett*, Cook County Hospital/U. of Ilinois, Chicago, Ill. (Introduced by Dr. J. Ferguson).

Sepsis is associated with alterations in microcirculatory flow as evidenced by increased A-V shunting and decreased peripheral vascular resistance. Leukotrienes, derivatives of arachidonic acid, have been implicated as mediators of the septic process. This study investigated the effect of diethylcarbamazine (DEC), a leukotriene inhibitor, on the flow of endotoxin-treated blood through a 5 micron pore polycarbonate filter according to the technique of Reid. The Reid method models microcirculatory flow in that the pores of the filter are approximately the size of capillaries. Flow of blood through the Reid apparatus is expressed as the Red Cell Flow Index (RCFI), where RCFI = flow (ml/sec)*hematocrit. The effects of E. Coli endotoxin (lipopolysaccharide B, E. Coli 026:B6; Difco Labs) and DEC on the RCFI of heparinized whole blood from 6 volunteers were determined. Results:

CONTROL	ENDOTOXIN	DEC	DEC THEN ENDOTOXIN	RCFI: MEAN ±S.E.
.83 ±.31	.05 ±.04*	1.38 ±.33*	.70 ±.21	*.p<.01

Endotoxin reduced the RCFI of whole blood, while DEC increased the RCFI. Pretreatment with DEC before the addition of endotoxin nearly abolished endotoxin's deleterious effect on flow. This study indicates inhibition of leukotrienes may be useful in preserving capillary flow during sepsis.

89

PROTECTIVE EFFECT OF LY171883, A SELECTIVE LEUKOTRIENE (LT) ANTAGONIST, ON ENDOTOXIC (LPS) SHOCK. J.A. Cook, W.C. Wise, and P.V. Halushka,* Medical University of South Carolina, Charleston, South Carolina 29425.

Previous studies have suggested that 5-lipoxygenase metabolism is enhanced in LPS shock but the potential pathogenic role of these LT mediators remains uncertain. In the present study, the effects of a selective LTD_4/E_4 antagonist, LY171883, 1-[2-hydroxy-3-propyl-4-[4-(1H-tetrazol-5-YL)-butoxy]-phenyl ethanone, (Lilly Pharm.), on LPS induced sequelae in the rat were assessed. LY171883 was given as a bolus (30 mg/kg) i.v. 10 minutes prior to Salmonella enteritidis LPS (40 mg/kg) in ketamine anesthetized (200 mg/kg) rats, followed by an infusion (10 mg/kg/hr) starting at 30

min post-LPS. Carotid artery blood pressures were determined at 10 minutes prior to and for 240 minutes post-LPS administration. Compared to shocked vehicle controls LY171883 attenuated the initial (0-30 min), but not the later, endotoxin induced hypotension (ANOVA, $P<0.01$). LY171883 effectively prevented (ANOVA, $P<0.001$) the neutropenia (0-180 min), but not the thrombocytopenia induced by LPS. Hemoconcentration resulting from LPS shock was reduced by LY171883 compared to the vehicle control group ($P<0.02$). In vitro studies confirmed no inhibitory effect of this antagonist on eicosanoid metabolism. Basal and calcium ionophore stimulated synthesis of immunoreactive (i)TxB_2, i6-keto-$PGF_{1\alpha}$ and iLTC_4/D_4 was not affected by LY171883 (10 and 50 μM). These data demonstrate that this LTD_4/E_4 antagonist has significant salutary actions in LPS shock, and suggest that leukotrienes D_4 and/or E_4 may contribute to some LPS induced sequelae. (Supported by NIH GM27673 and HL29566).

90

ENDOTOXIN-INDUCED EICOSANOID PRODUCTION. G.BOTTOMS, M.JOHNSON*, D.WARD*, J.FESSLER*, C.LAMAR*, J.TUREK*. Dept of Veterinary Physiology&Pharmacology, Dept of Veterinary Anatomy & Dept of Large Animal Clinics, Purdue University, W. Lafayette, IN 47907.

Eicosanoid concentrations were determined in the plasma of ponies given endotoxin every 6 hrs for 24 hrs and in the media of cultured endothelial and smooth muscle cells and freshly isolated blood cells incubated with endotoxin. Endotoxin injections resulted in cyclic increases of thromboxane (TxB_2) during the first 0.5 hr which decreased to near baseline by 4 hrs after endotoxin. Prostacyclin (6-keto-$PGF_{1\alpha}$) showed the same cyclic response, but was highest at 1.5 hrs and returned to baseline by 6 hrs after endotoxin. There was no significant change in plasma LTC_4 immunoreactive material over time during the 24 hour study. Endotoxin, as low as 5 ug/ml, stimulated cultured endothelial cells to produce prostacyclin and thromboxane, but concentrations up to 100 ug/ml had no significant effect on LTC_4 production. Endotoxin increased the production of prostacyclin by vascular smooth muscle cells, but had no effect on thromboxane or LTC_4. Endotoxin incubated with plasma containing WBCs' stimulated the production of thromboxane and immunoreactive like LTC_4, but had no effect on prostacyclin. Therefore, endotoxin is capable of perturbing cells directly and increasing the production of cyclooxygenase products. Some of these products may be detrimental and some may to be beneficial. The role of leukotrienes remains questionable since significant increases of LTC_4 were not detected in the plasma of animals injected with endotoxin. It is possible that changes in peripheral plasma LTC_4 are less than the sensitivity of our assay. (Supported by USDA and AQHA Grants)

91

EFFECTS OF METHYLPREDNISOLONE ON THROMBOXANE AND PROSTACYCLIN LEVELS IN A RAT ENDOTOXIN SHOCK MODEL. K.L. LINCOLN*, R. OCHOA, J.E. ZWIER-KOEZLER*, and R.R. GORMAN*. The Upjohn Company, Kalamazoo, MI 49001.

The purpose of this experiment was first to follow the progression of Thromboxane B2 and 6-keto-PGF1-alpha levels over time in a rat model of endotoxin shock, and secondly to determine the effect of Methylprednisolone sodium succinate on those levels. The treatment groups included: 1)saline + saline, 2)toxin + saline, and 3)toxin + 39.5 mg/kg Methylprednisolone sodium succinate. Endotoxin shock was induced in unanesthetized female Sprague-Dawley rats by infusing an LD_{85} (18 mg/kg) dose of E. coli endotoxin through a tail vein cannula over a period of 5 hours. Treatment was started 15 minutes after the initiation of the toxin. During the first 20 minutes of treatment, 2/3 of the therapy was administered. The remaining 1/3 of the therapy was administered over the next 120 minutes. Four rats from each treatment group were sacrificed at 10, 15, 30, 60, 120, 240, 300, 480, and 720 minutes after toxin administration initiation. Blood samples were withdrawn, quenched with indomethacin, and analyzed for Thromboxane B2 and 6-keto-PGF1-alpha levels using a radioimmunoassay. Both Thromboxane B2 and 6-keto-PGF1-alpha levels initially increased then decreased in all groups. Thromboxane B2 and 6-keto-PGF1-alpha levels remained low in saline + saline rats and gradually increased in toxin + saline rats. Administration of Methylprednisolone sodium succinate to toxin-treated rats prevented the rise of both arachidonates.

92

THE EFFECT OF A THROMBOXANE RECEPTOR BLOCKER IN ENDOTOXIN SHOCK. R. URBASCHEK*, H. PATSCHEKE[1*], K. STEGMEIER[2*], B. URBASCHEK. Dept. of Immunol. & Serol., Inst. of Hygiene & Med. Microbiology and [1]Inst. of Clin. Chem., Klinikum Mannheim of the University of Heidelberg, [2]Boehringer Mannheim GmbH, D-68 Mannheim, West Germany.

BM 13.177 and its more potent analogue BM 13.505 selectively antagonize thromboxane A_2 (TXA_2) and the cyclic endoperoxides PGG_2 and PGH_2 at the thromboxane receptor of platelets and arterial smooth muscle. Their protective effect on endotoxin-induced mortality was studied in NMRI mice. 1) BM 13.177 or BM 13.505 1.5 mg/mouse were injected i.v. 10 minutes before plus 6 hours after i.v. injection of 125 µg, 150 µg, 175 µg, or 200 µg of endotoxin in 10 mice each. Controls received saline. 2) Correspondingly, BM 13.177 or BM 13.505 were injected into mice sensitized to the effects of endotoxin by BCG infection ($1x10^8$ viable CFU) 14 days before injection of endotoxin 0.2 µg/mouse. 3) BM 13.505 was administered at different times after i.v. injection of 175 µg endotoxin/mouse. In experiment 1, a significant decrease in and a prolonged onset of lethality was observed after pretreatment with the TXA_2 receptor blockers. The drugs were also capable of reducing the lethality rate in BCG-sensitized animals in which death occurs within few hours following minute amounts of endotoxin. The third experiment revealed that the treatment with BM 13.505 is most effective when administered shortly before and after endotoxin. No effect was observed when the receptor blockers were given two hours or later after endotoxin. Although the pathophysiological events are multifactorial, the results described here clearly support the concept that TXA_2 is an important mediator of endotoxin-induced shock and death in mice.

93

EFFICACY OF NON-STEROIDAL ANTI-INFLAMMATORY DRUGS AND/OR STEROIDS ON ENDOTOXIN-INDUCED CHANGES IN CALVES. J.H.MARGOLIS*, G.D.BOTTOMS, J.F.FESSLER*. Department of Veterinary Physiology & Pharmacology and the Department of Large Animal Clinics, Purdue University, West Lafayette, Indiana 47907.

Numerous studies have shown non-steroidal anti-inflammatory drugs as well as steroids are beneficial in the treatment of endotoxic shock. Experiments were done to test the effficacy of these drugs in calves given endotoxin. Twelve male calves (34-57 kg) were divided into four equal groups: Group 1, untreated; Group 2, flunixin meglumine treated (FM; 1.1 mg/kg; a non steroidal anti-inflammatory drug); Group 3, dexamethasone treated (Dex.; 2 mg/kg); Group 4, FM plus Dex. treated. Each calf was given intravenous and intraperitoneal injections of Escherichia coli endotoxin at t=0, 2 and 4 hours. Mean arterial pressure (MAP), pulmonary artery pressure (PAP) and cardiac output (CO) were measured. Blood samples were collected for the determination of arterial blood gases, lactate, thromboxane (TxB_2), and prostacyclin (6-keto-$PGF_{1\alpha}$). MAP and PAP did not change significantly between groups. A comparison between Group 1 vs Group 4 revealed that, in Group 1, CO was lower ($p<0.05$) at 0.17 hours; heart rate was greater ($p<0.05$) at 2,3,5,6,18 and 24 hours; serum lactate was greater ($p<0.05$) at 2, 4 and 6 hours; HCO_3^- was lower ($p<0.05$) at 2,3,4,5 and 6 hours; TxB_2 was greater ($p<0.05$) at 2,3,4,5 and 6 hours and 6-keto-$PGF_{1\alpha}$ was greater ($p<0.05$) at 6 and 18 hours. Similar comparisons between Group 1 vs Group 2 or 3 revealed few differences. These results indicate that the combination treatment of FM and Dex. prevent many of the metabolic derangements observed during endotoxic shock in cattle. (Supported by AVMA Grant)

94

INDOMETHACIN AND DOPAMINE IN ENDOTOXIN SHOCK: IMPROVEMENT IN HEMODYNAMICS AND MORTALITY. A.J. Griffin*, M. Goto. Loyola University Stritch School of Medicine, Maywood, Illinois 60153.

Since indomethacin(IND) and dopamine(DOP) are used, often empirically, in endotoxin shock(ETX), this study was performed to quantitate mortality and effects of the treatment on the hemodynamics of the surviving animals of Salmonella endotoxin shock studied 24 hours after ETX. 116 Holtzman rats (180-260g) were divided into three groups: Group 1 (n=74) received 15mg/kg of ETX. Group 2 (n=26) received ETX + 1.5 mg/kg of IND, Group 3 (n=16) received ETX + IND + continuous DOP (5 mcg/kg/min) by implanted osmotic pressure pump. ETX, IND and DOP were all in-

jected into the right jugular vein. 24 hours after ETX injection, heart rate, mean arterial pressure (MAP) and cardiac output (CO) were measured in the surviving rats under pentobarbital anesthesia.

	Mortality(%)	MAP(mmHg)	CO(ml/min/kg)	
Group 1	90	67 ± 11	148 ± 27	
Group 2	50*	85 ± 8	143 ± 10	* p <0.5
Group 3	25*	100 ± 18*	209 ± 23*	(mean ± s.e.)

Indomethacin decreased the 24 hour mortality rate in endotoxin shock. Hemodynamics were not significantly different in the surviving rats from the IND treated and the untreated groups. The addition of dopamine to indomethacin further decreased the mortality rate. Remarkable improvement in hemodynamics were observed in only those rats receiving both dopamine and indomethacin.

95

PROSTANOID PRODUCTION BY CULTURED HUMAN ILIAC VEIN ENDOTHELIAL CELLS IN RESPONSE TO ZYMOSAN-ACTIVATED PLASMA. John T. Flynn, David Wojcik*, Kenneth P. Chepenik*, Elliot Levine* and Stuart K. Williams*. Thomas Jefferson University, Philadelphia, PA 19107.

Activated complement (AC) has been shown to stimulate arachidonic acid metabolism in several types of cells. The present study evaluates the ability of cultured human adult endothelial cells (HAEC) to respond to AC directly, and to subsequent ionophore challenge. Cells were grown to confluence in medium containing ECGF and heparin. The cells were then challenged for 1 hr with either vehicle, or human AC (Cordis) at 10, 1 and 0.1 units of activity per culture. The medium was collected, the cells refed, and incubated for 24 hrs. A23187 (10 μM) was then added to all cultures. Prostacyclin (PGI) concentrations (measured as 6 keto-$PGF_{1\alpha}$ by RIA) were 653±36 pg/culture in the control group, and 1382±81, 816±103, and 652±70 pg/culture in cells receiving 10, 1 and 0.1 U of AC respectively. While 1 and 0.1 U of AC did not statistically increase PGI production, cells receiving 10 U produced statistically more PGI ($p < 0.005$) than any other group tested. After challenge with A23187, 24 hrs. later, a similar situation was observed. The cells which previously had received 10 U of AC demonstrated a significantly increased production of PGI while all other groups were only moderately stimulated by the ionophore. Thromboxane B_2 (TXB) in these same groups of cells was measurable with a control group value of 380±35 pg/culture. As was seen with PGI, 10 U of AC produced a significant increase in TXB production to 1000±118 pg/culture ($p<0.001$) while lower doses of AC had no effect. When all groups of cells were subsequently challenged with the same dose of ionophore (10 μM), only the group which was previously treated with 10 U of AC demonstrated a significant increase in TXB production to 1240±118 pg/culture. These data demonstrate that AC can directly stimulate prostanoid production in cultured HAEC. In addition, it appears that AC may influence the long-term response of the arachidonic acid cascade to various stimuli. This work was supported in part by GM 28023.

96

EICOSANOIDS IN SPLANCHNIC ARTERIAL OCCLUSION (SAO) INDUCED SHOCK. W.C. Wise, M. Pinosky*, S.H. Ashton*, P.V. Halushka*, and J.A. Cook. Medical University of South Carolina, Charleston, South Carolina 29425.

SAO shock is a lethal form of shock, attributable to prolonged ischemia of the splanchnic region. To assess the potential role of eicosanoids in SAO shock, survival time and plasma levels of TxB_2 and 6-keto-PGF_{1a} were measured by RIA in normal, essential fatty acid deficient (EFAD), and Indomethacin (INDO) (10 mg/kg) pretreated (30 min) shocked and sham control rats. SAO shock was induced by occlusions of the coeliac and superior mesenteric arteries for 60 mins. Plasma was obtained from the vena cava 5 min after occlusion release for assay of $iTxB_2$ and 16-keto-PGF_{1a}.

	Plasma pg/ml		% Survival		
	$iTxB_2$	i6-keto-PGF_{1a}	1hr	2hr	3hr
SHAM	516 ± 87 (6)	195 ± 31 (6)	100	100	100
SAO	3020 ± 750 (7)*	1617 ± 272 (8)*	42	32	10
EFAD-SAO	502 ± 145 (7)	<150 (7)	82	47	18
INDO-SAO	<150 (4)	<150 (4)	43	14	0

*= P < 0.05 ANOVA to EFAD-SAO;

SAO shock caused a significant increase in $iTxB_2$ and i6-keto-PGF_{1a} but not in EFAD or INDO rats. EFAD but not INDO improved survival time from shock. Arterial blood pressure pre- and post-occlusion were not significantly different between any group. These observations raise the possibility that other arachidonic acid metabolites,

e.g. lipoxygenase products and/or other mediators, may play a major role in the pathophysiology of SAO shock. (Supported by NIH GM27673, HL29566 and MUSC Funding).

97

IN VITRO EFFECTS OF OPIATE PEPTIDES IN GUINEA PIG MYOCARDIUM. L.F. Hathorn*, J.L. Caffrey*, P. Kutsky, J.L. Parker+. Dept. Physiology, Texas College of Osteopathic Medicine, Ft. Worth, TX 76107, and Dalton Research Center+, University of Missouri, Columbia, MO 65211.

Inotropic and chronotropic effects of methionine-enkephalin (ME) were evaluated using spontaneously beating right atria and electrically driven left atria isolated from guinea pigs. In vitro addition of ME (10^{-8}-10^{-5}M) failed to alter basal heart rate, developed contractile tension (CT), or rates of tension development and relaxation (±dT/dt). Myocardial tissues were determined to possess a substantial capacity for enkephalin hydrolysis. However, attempts to inhibit hydrolysis by pretreatment with 3,4-dihydroxyphenylacetic acid (10^{-4}M) failed to facilitate responses to the added ME. Biologic activity of ME from the medium bathing the tissues was verified using a guinea pig ileal bioassay. CT concentration - response curves to norepinephrine (NE; 10^{-9}-10^{-4}M) were shifted to the right ($p<0.05$) after ME, but maximal responses were unaffected. Atrial tissues isolated from endotoxin shocked animals were inotropically depressed, but unlike control tissues, were completely insensitive to ME. Preliminary studies confirmed a lack of direct myocardial depression by ME, dynorphin and D-ala-D-leu-enkephalin in guinea pig ventricular papillary muscles. Thus, we conclude that opiate peptides do not produce direct inotropic or chronotropic depression of guinea pig myocardium. However, these studies also provide evidence supportive of opiate-adrenergic interactions in isolated cardiac tissues. (Supported by American Heart Association 82-855 and 83-1061, NHLBI T35-HL-07465, and AOA 82-11-078).

98

BETA-ENDORPHIN(BE) CAUSES CARDIOVASCULAR(CV) DEPRESSION IN DOGS. D.W. Tuggle*, J.W. Horton. University of Texas Health Science Center at Dallas, Southwestern Medical School, Dallas, Texas.

BE has been implicated in the CV depression in shock. While pharmacologic doses of BE cause hypotension, physiologic doses of BE have not been studied. In this study, 6 dogs (Group I) were given IV BE (peak concentrations previously determined in canine shock, 3200 pg/ml); 5 min prior to BE infusion, 4 dogs (Group II) were given Naloxone(NAL), 2 mg/kg bolus and continuous infusion, 2 mg/kg/hr. In Group I, BE caused stroke volume(SV, ml/kg), dP/dt (max, mmHg·sec), and coronary blood flow(CBF, ml/min/g) to fall, while heart rate(HR, bpm) and peripheral vascular resistance(PVR, dynes·cm·sec^{-5}) rose signficantly. NAL pretreatment maintained dP/dt, SV, and CBF with no change in HR or mean arterial pressure(MAP, mmHg). This study confirms that BE depresses contractility and CBF in normovolemic nonstressed dogs, suggesting that BE is in part responsible for CV depression in shock.

	CONTROL		15 MINUTES		60 MINUTES		120 MINUTES	
	I	II	I	II	I	II	I	II
MAP	131±10	129±12	134±10	131±12	119±9	131±12	129±13	136±13
CBF	2.50±.47	2.13±.16	1.73±.06	3.08±.09	1.68±.16	2.52±.40	.68±.11	2.18±.39
dP/dt	3167±140	3750±822	3500±287	4875±850	2980±525	4970±595	2875±412	4107±540
SV	0.88±.12	1.06±.11	.68±.08	1.04±.13	.58±.08	.99±.14	.57±.08	.99±.17
HR	117±23	87±11	138±13	80±9	127±13	88±10	118±15	90±12
PVR	5105±488	3506±285	7165±1759	4426±363	8469±1964	4488±1042	9472±1281	5640±160

99

EFFECT OF NALOXONE (Nal), ON ARACHIDONIC ACID (AA) METABOLISM IN ENDOTOXIC (LPS) SHOCK. Keith Coffee,* J.A. Cook, W.C. Wise, and P.V. Halushka.* Medical University of South Carolina, Charleston, S.C. 29425.

Both AA metabolites and the endogenous opiate system have been implicated in the sequelae of LPS shock. Cyclooxygenase inhibitors attenuate circulatory responses to enkephalins, suggesting that AA metabolites may modulate opiate receptor mediated events. Evidence for interaction of these potential pathogenic factors in shock however is lacking. The present studies assessed the action of Nal on LPS shock in the rat with regard to blood pressure, plasma TxB_2, thrombocytopenia, neutropenia, and hemoconcentration. Long Evans rats were pretreated with 5 mg/kg Nal iv bolus followed by an iv infusion (1 mg/kg/hr) 15 min prior to S. enteritidis LPS (30 mg/kg iv bolus) administration. Pretreatment with Nal significantly attenuated LPS induced hypotension (0-30 min, $P<0.05$, n=15) and neutropenia (0-180 min , $P<0.05$, n=6). Plasma TxB_2 levels were lower in Nal treated rats (1.1 ± 0.1 ng/ml, n=6) vs shocked controls (1.9 ± 0.3 ng/ml, $P<0.05$, n=6). Hemoconcentration and thrombocytopenia were not significantly altered. Additionally, the effect of Nal on in vitro AA metabolism by peritoneal macrophages was determined. Nal (10 ug/ml) significantly ($P<0.05$, n=4) inhibited LPS stimulated synthesis of TxB_2 and 6-keto-PGF_{1a} 66% and 60% respectively compared to controls. These studies suggest that a salutary action of this opiate receptor antagonist in LPS shock may be mediated, in part, by reduced AA metabolism. (Supported by NIH GM 27673 and HL 29566).

100

INTESTINAL ISCHEMIA ASSOCIATED WITH NALOXONE RESUSCITATION OF NEONATAL SEPTIC SHOCK. E.D. DOBKIN,* T.E LOBE, J. BHATIA,* K.T. OLDHAM,* H.A. LINARES,* D.L. TRABER.
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To evaluate the effectiveness of naloxone in peritonitis-induced septic shock, anesthetized pigs were monitored and peritonitis was induced by intraperitoneal injection of a fecal E.coli mixture. All pigs received identical colloid resuscitation and gentamicin, and acidosis was corrected. When cardiac output decreased by 20% of baseline, the pigs either received an initial IV bolus of naloxone (2 mg/kg) followed by a 2 mg/kg·hr infusion (Group I, n=9), or received no additional pharmacological intervention (Group II, n=7). Hemodynamic parameters assessed included mean arterial, pulmonary arterial, and central venous pressures; cardiac, stroke volume, and left ventricular stroke work indices; and systemic and pulmonary vascular resistance indices. There were no significant differences in any of the parameters assessed between Groups I and II, although peripheral vascular resistance in Group I was transiently elevated acutely after naloxone infusion began. Mean survival times in the two groups were also similar. Five of 9 Group I animals (56%), demonstrated gross and histologically proved intestinal ischemia, while none of the animals in Group II demonstrated any notable sequelae ($p<.02$). These data suggest that naloxone resuscitation results in an increase in vascular resistance without concomitant improvement in cardiac performance and that these changes are associated with significant intestinal ischemia in this model.

101

EFFECT OF THYROTROPIN RELEASING HORMONE IN CANINE HEMORRHAGIC SHOCK. L. TEBA, M. ZAKARIA, K.C. BEAMER, S. RYERSON. West Virginia University Medical Center, Morgantown, WV 26506. (Introduced by: Thomas Vargish)

Thyrotropin Releasing Hormone (TRH) appears to function as a "physiologic antagonist" of opiates. Hemodynamic response of canine hemorrhagic shock (CHS) to TRH is reported. Ten dogs (23-27 Kg) were anesthetized with pentobarbital. Femoral and pulmonary artery catheters were placed. Cardiac output (CO) and hemodynamics were obtained. All dogs were bled to a mean arterial pressure (MAP) of 50 mmHg and maintained at this level for half an hour. The above measurements were then repeated, and the animals were divided into two groups: A) received TRH 2 mg/Kg IV in 20 cc D_5W bolus, B) received placebo 20 ccD_5W bolus. CO and hemodynamics were recorded at 1, 8 and 15 min. and every 15 min. thereafter for up to 2 hours. Significance ($p < 0.05$) of this data was tested using a Student's t test. After TRH, MAP (table 1), and CO increased significantly. These changes were still significant 2 hours later. CO improved 36%, 56%, 64%, 64% at 1, 15, 60, 120 min. respectively. No significant changes were observed in CO in the placebo group. In both groups, the differences for HR and SVR were not significant. These results show that TRH improves MAP and

CO in CHS without affecting HR or SVR. These changes persisted for 2 hours.
Table - Mean Systemic Arterial Pressure (mmHg)± SD

	½ hour shock	1'	8'	15'	30'	45'	60'	75'	90'	105'	120'
Placebo n=5	49±4	58±7	61±14	68±10	64±14	67±15	65±17	65±7	60±14	68±13	62±13
TRH n=5	50±5	84±12	95±15	102±18	106±25	107±20	106±21	104±24	98±29	98±33	102±36

102

BENEFICIAL CARDIOVASCULAR EFFECT OF THYROTROPIN RELEASING HORMONE IN HEMORRHAGIC SHOCK IN THE PRESENCE OF ACIDEMIA. F. SCHIEBEL, L. TEBA, M. ZAKARIA, I. RAMOS. West Virginia University Medical Center, Morgantown, WV 26506 (Introduced by: Thomas Vargish)

The effect of thyrotropin releasing hormone in reversing prolonged hypotension in the presence of acidemia was studied. Five dogs (23-27 Kg) were anesthetized with pentobarbital. A femoral and pulmonary artery catheter were placed. Each dog was bled to a mean arterial pressure (MAP) of 50 mmHg. Without any further blood withdrawal to alter blood pressure, MAP was recorded every 15 min. Hypotension persisted (MAP 62±12 SD) for 2½ hours. Then baseline cardiac output and hemodynamics were obtained. Arterial blood was sampled for pH determination, and each dog received TRH 2 mg/Kg body weight. Afterwards, CO and hemodynamics were repeated at 1, 15 and 30 min. The significance ($p < .05$) of this data was tested using a Student's t test. At 2½ hours post hemorrhagic shock, arterial pH was 7.20 (7.15-7.24). Results are shown in the table.:

	Baseline	1'	15'	30' - Post TRH
MAP	55±14	81±9*	77±14*	77±13*
CO	1.2±.1	2±.5*	2.1±.3*	2.1±.4*
HR	185±11	199±11*	196±10	196±10
SVR	3623±648	3437±814	2843±237*	2808±449*

mean±SD, *$p < .05$

This data indicates that a significant increase in MAP and CO can be obtained with TRH 2 mg/Kg in prolonged hemorrhagic shock, even when arterial pH is below 7.25.

103

THYROTROPIN RELEASING HORMONE IN HEMORRHAGIC SHOCK: EFFECTS ON CARDIAC OUTPUT AND REGIONAL BLOOD FLOW. A.-L. SIREN,* E. POWELL, G. FEUERSTEIN. Department of Neurology, Uniformed Services University of the Health Sciences, Bethesda, MD 20814. Thyrotropin releasing hormone (TRH) reverses hypovolemic shock in the rat. This study aims to elucidate the effects of TRH on cardiac index (CI) and regional blood flow in hemorrhagic shock. Conscious rats, chronically instrumented for either CI (thermodilution, cardiomax) or blood flow (BF) measurements (directional pulsed Doppler technique) were used. BF to hindquarters (HQ), mesenteric (M) and renal (R) arteries and the mean arterial pressure (MAP) and heart rate (HR) were continuously monitored. Hemorrhagic shock, induced by bleeding, (5 or 8.5 ml/300g) caused decreases in MAP (-65 mmHg), CI (-50%), total peripheral resistance index (TPRI) (-20%) and in blood flows in HQ(-75%), M(-64%) and R(-67%). The TRH-analogue CG3703 (0.5 mg/kg i.a.), injected at the end of the bleeding, induced rises of MAP and HR (maximum changes of +67±6 mmHg and +123±30 bpm, respectively). Control group received equal volume of saline which did not alter the cardiovascular changes caused by the hemorrhage. CG3703 raised the TPRI (+101±2% vs +25±6% in the control ($p<0.001$) but had no effect on CI. The blood flows in control vs CG3703-treated groups 2 hr after the bleeding were: -32±6% vs -55±6% ($p<0.001$) in HQ, -9±8% vs -61±11% ($p<0.001$) in M and -2±9% vs -30±7% in R, and the vascular resistances +30±7% vs +149±36% in A ($p<0.001$), +4±8% vs +349±244% in M and -10±9% vs +88±112 in R. The 24 h survival was decreased by TRH-treatment. The results suggest that TRH induced reversal of hypotension is due to an increase in TPR and caused an impairment of the local blood flow in M and HQ vasculature, exacerbates the shock and leads to death.

104

HEMODYNAMIC AND METABOLIC RESPONSES, INTESTINAL PATHOLOGY, AND SURVIVAL TO ENDOTOXIN IN TRH PRE-TREATED, CONSCIOUS, INSTRUMENTED RATS. M.F. WILSON, D.J. BRACKETT, C.F. SCHAEFER, M.R. LERNER*, M.L. WILSON*, P. TOMPKINS*, J.W. HOLADAY, L. FAGRAEUS. VA Medical Center, Depts. Medicine and Anesthesiology, Univ. of Oklahoma Health Sciences Ctr., Oklahoma City, OK 73104 and Walter Reed Army Inst.Res.,Washington, D.C. 20012.

These studies were designed to evaluate possible benefits of thyrotropin releasing hormone (TRH) for circulatory and metabolic support to counter adverse effects of a lethal dose of endotoxin (E) given by intravenous bolus. Sprague-Dawley rats were instrumented under enflurane anesthesia for measurements of cardiac output (CO) by thermal dilution, arterial pressure (AP), central venous pressure, and heart rate (HR) by indwelling catheters. After recovery arterial blood samples were obtained at control and 1 and 4 hrs for blood gases, lactate (LACT), glucose, and hematocrit (HCT). Groups were TRH and saline, saline and E, TRH pre-treatment (Rx) and E. The TRH dose was 2.0 mg/kg; E dose was 20 mg/kg. With saline and E: all survived 4 hrs; all had a BP and CO fall with compensatory tachycardia. Over 4 hrs acidosis developed ($\downarrow$ pH) with respiratory compensation ($\downarrow PCO_2$), LACT and HCT increased, and initial hyper- and 4 hr hypoglycemia occurred. With TRH pre-Rx and E there was less initial BP and CO fall due to less pronounced stroke volume decrease and higher HR response resulting in the same peripheral resistance response. From 1 to 4 hrs the hemodynamic response was not different. With TRH and E compared to saline and E, LACT increased more, respiratory compensation was initially stronger (greater $\downarrow PCO_2$) and then less (less $\downarrow PCO_2$ and greater $\downarrow$ pH), 40% survived 4 hrs, and intestinal pathology was severe in both groups. In conclusion, a 2.0 mg/kg TRH pre-Rx did not benefit these conscious, instrumented rats challenged with a lethal E bolus.

105

EVALUATION OF THE METABOLIC ROLE OF GLUCAGON AND INSULIN IN BURN PATIENTS USING SOMATOSTATIN INFUSION AND STABLE ISOTOPIC TRACERS. R.R. WOLFE, F. JAHOOR,*, D.N. HERNDON, M.H. WOLFE,*, Shriners Burns Institute and The University of Texas Medical Branch, Galveston, TX 77550.

There is a persistent increase in the rate of gluconeogenesis from amino acids in burn patients that is a component of an accelerated rate of net protein breakdown. We have evaluated the role of glucagon (G) and insulin (I) in this process in 8 severely burned ($\bar{X}$ = 68%) BSA patients by suppressing the secretion of both hormones with somatostatin (S) infusion, and, in some cases, replacing insulin. Primed-constant infusions of 6,6-d_2-glucose and 3-^{13}C-alanine enabled the quantitation of glucose production and alanine flux, and plasma amino acid levels were measured as well. All studies were done after 8 h of fasting. Basal glucose production was elevated in burn patients (3.75±.54 vs 2.38±.08 mg/kg·min for control). Reduction of both I and G in patients by S infusion alone caused a modest fall in glucose production, but the most striking response was a significant increase in plasma glucose concentration due to a significant reduction in glucose clearance (from 3.83 ml/kg·min before S to 2.83 ml/kg·min after S, $p < .05$). Also, alanine flux increased significantly during S infusion, as did many of the plasma amino acid levels. When I was replaced at the basal level during S infusion, the basal glucose production rate fell further (to 2.38±.82 mg/kg·min), and glucose clearance was maintained at the basal level. Alanine flux also remained at the rate determined before the S and I infusion.

We conclude that in burned patients G stimulates glucose production and basal I both suppresses endogenous glucose production and stimulates glucose uptake.

106

TRIGLYCERIDE AND FREE FATTY ACID TURNOVER IN E. COLI ENDOTOXIN TREATED RATS. G.J. BAGBY, C.B. CORLL*, R.R. MARTINEZ*. LSU Medical Center, New Orleans, LA 70119.

Hypertriglyceridemia is a common metabolic disorder observed during bacterial sepsis and after endotoxin (ET) administration. Lipoprotein lipase (LPL), the enzyme responsible for clearing circulating triglycerides (TG) is decreased in tissues after ET or E. coli administration. However, the importance of this decrease to the development of hypertriglyceridemia has not been ascertained. This study was performed to determine if ET induced hypertriglyceridemia results from an increased rate of TG appearance or a decreased metabolic clearance rate (MCR) of TG.

Turnover of TG (VLDL-TG labeled with ^{3}H-palmitate) and FFA (^{14}C-palmitate) were simultaneously determined by the constant isotope infusion technique in conscious rats 18-20 hr after the administration of ET (0.15 mg/100g) or saline (control, C). ET and C rats had similar mean arterial blood pressures and heart rates during the infusion period. Plasma FFA concentration and turnover did not differ between ET and C animals but the MCR was significantly decreased in the ET treated rats ($p<0.05$). Plasma TG concentrations were increased about 3 fold in ET rats. Turnover of TG did not differ between ET and C rats. As a consequence, ET treatment reduced the MCR of TG to 22% of the C value (8.21 ± 1.56 ml x min^{-1} x kg^{-1}). The decrease in the MCR of TG and the absence of an elevated TG turnover indicate that the hypertriglyceridemic state after ET results from a reduced ability of tissues to take up circulating TG. ET induced suppression of tissue LPL activity is likely to be responsible for the elevation of plasma TG concentrations. (Supported by NIH grant GM 32654).

107

MONOKINE-INDUCED HYPERINSULINISM AND GLUCAN SENSITIZATION TO ENDOTOXIN. J. P. Filkins. Department of Physiology, Loyola Univ. Medical Center. Maywood, IL 60153.

Glucan, a β 1-3-linked glucopyranose derivative of the cell wall of the yeast Saccharomyces cerevisiae induces a marked proliferation of the macrophage elements of the reticuloendothelial system (RES) and also sensitizes to the metabolic effects of bacterial endotoxin. The present study evaluated the effects of S. enteritidis endotoxin (E) on plasma glucose (PG), immunoreactive insulin (IRI) and non-suppressible insulin-like activity (NSILA) in glucan vs saline-treated Holtzman rats.

GROUP	N	E(10μg/kg)	PG(mM)	IRI(μU/ml)	NSILA*
Saline Controls	6	-	5.8±.42	22±6	5.9±.48
	5	+	6.4±.31	28±7	6.4±.55
Glucan	10	-	5.2±.22	26±4	6.8±.55
	10	+	1.1±.09†	42±6†	14.2±1.2†

†=p<.05 for E+ vs E- * DPM $C^{14}O_2$ from glucose/g adipose tissue/hr x 10^4

Low dose endotoxicosis in glucan rats resulted in hypoglycemia and elevations in both IRI and NSILA. Since glucoregulatory monokines are related to endotoxic hyperinsulinism (*Fed. Proc. 44:* 28-32, 1985), the extreme sensitivity of glucan rat to endotoxin may be related to excessive monokine secretion, e.g., interleukin 1, uncompensated hyperinsulinism, and glucose dyshomeostasis. (Supported by GM 29619.)

108

GLUCOSE KINETICS AND OXIDATION IN NORMAL VOLUNTEERS, SEPTICAEMIC PATIENTS, AND PATIENTS WITH SEVERE PANCREATITIS. J H F SHAW, J JANUSZKIEWICZ*, R HORSBOROUGH*, Department of Surgery, Auckland Hospital, Auckland, New Zealand.

Five normal volunteers, seven severely septic patients, and five patients with severe pancreatitis were studied. Glucose turnover was measured using primed constant infusions of either 66d- or 6-^{3}H glucose, and glucose oxidation with either u-14c- or u-13c- glucose. Basal measurements were made during Period I (2 hrs), and then glucose was infused (4 mg/kg.min) during Period II (2 hrs).

		CONTROL	SEPTIC	PANCREATITIS
glucose production (Ra)	Period I	13.7 ± 0.4	22.0 ± 2.4*	15.2 ± 0.9*
(umole/kg.min)	Period II	21.7 ∓ 1.8	38.5 ∓ 4.2*	23.8 ∓ 1.0
glucose clearance (Cl)	Period I	2.7 ∓ 0.1	4.8 ∓ 0.4*	3.2 ∓ 0.3
(mL/kg.min)	Period II	2.9 ∓ 0.3	4.4 ∓ 0.5*	3.3 ∓ 0.5
glucose oxidation (Ox)	Period I	5.6 ∓ 0.5	9.0 ∓ 0.1*	5.7 ∓ 0.9
(umole/kg.min)	Period II	7.8 ∓ 0.8	12.6 ∓ 1.1*	7.6 ∓ 1.4
% glucose uptake	Period I	40.8 ∓ 4.0	42.3 ∓ 2.8	39.6 ∓ 8.5
oxidised	Period II	35.9 ∓ 3.6	34.4 ∓ 2.2	31.9 ∓ 5.7
% supression by g. infusion	Period II	98 ∓ 3%	34 ∓ 5%*	78 ∓ 9%*

We conclude that: (i) In septic patients glucose Ra, Cl, and Ox are all significantly higher than in volunteers, and glucose Ra is only partially supressed by

glucose infusion, (ii) Glucose turnover is significantly higher in patients with pancreatitis, and supression is less than in volunteers, (iii) The efficiency with which glucose is oxidised is similar in stressed patients and volunteers.

109

Potential Regulatory Mechanism for Depression of Ketogenesis in Sepsis. T.C. Vary*, J.H. Siegel, T. Nakatani*, T. Sato, H. Aoyama*. MIEMSS and Departments of Physiology and Pathology, University of Maryland, Baltimore, MD. 21201.

Several investigators have demonstrated a diminished relative rate of ketogenesis during inflamatory states or sepsis. Although the biochemical mechanism for this effect is unknown, decreased hepatic malonly-CoA levels have been implicated as an important regulatory stimulus for ketone body formation in diabetes and starvation, whereas increased malonyl-CoA levels inhibit ketogenesis. In the present study, the effect of an intra-abdominal abscess on the level of hepatic malonly-CoA was investigated in four groups of rats (control, sterile, small abscess, large abscess). Frozen liver samples were taken in situ five days following the intraperitoneal introduction of a rat-fecal agar pellet inoculated with a known bacterial flora which generated an abscess [sterile inflamatory; B. Fragilis 10^8/ml + E. Coli 10^2/ml (small=0.8ml or large 1.5ml) abscess pellet]. The level of malonyl-CoA in normal, fed rats was 5±0.6 nmol/gm wet wt (n=9). The malonyl-CoA level was not altered in animals with a sterile pellet. However, hepatic malonyl-CoA levels were significantly increased in small (10±1 nmol/gm wet wt)($p<.05$;n=9) or large (12±2 nmol/gm wet wt)($p<.01$;n=13) septic abscess rats compared to normal, fed animals. Hepatic ketone bodies (3-hydroxybutyrate and acetoacetate) did not increase in sepsis over control or inflamation. These results are compatible with the hypothesis that increased hepatic malonyl-CoA levels may be responsible in part for the reduced relative rate of ketogenesis in sepsis. Increased formation of malonyl-CoA may differentiate sepsis from inflammation.

110

BETA ENDORPHIN DOES NOT ALTER GLUCOSE METABOLISM IN SKELETAL MUSCLE:J.F. Amaral, C. Mendez, M.D. Caldwell*, and D.S. Gann*, Brown Univ/R.I. Hospital, Dept. Surgery, Providence, R.I.

We have previously reported that naloxone, a mu opiate antagonist, blunts the hyperglycemic response to hemorrhage and that it increases glucose uptake in skeletal muscle(SM). Other investigators have reported that the IV injection of beta-endorphin(BE), a mu and delta receptor opiate agonist, produces hypoglycemia, whereas injection of BE into the CSF produces hyperglycemia. The present studies were designed to examine the direct effect of BE in the presence and absence of insulin(INS) on glucose uptake and metabolism in rat SM using an isolated, recirculating, perfused hindlimb system(JBC 1978:253:6837). After a 20min pre-perfusion period, the isolated hindlimb SM was perfused with no hormones(CTL), with 10ng ins or with 4, 8, and 16ng BE per ml of perfusate, either alone or in combination(see Table), for an additional 80min. BE was undetectable, by RIA, in the perfusate at the onset of perfusion.

Group	Glucose Uptake um/min/100gBW	Pyruvate Release um/min/100gBW	Lactate Release um/min/100gBW	
CTL (n=11)	1.95±0.18	0.412±0.07	2.85±0.56	
4ng-BE (n=12)	1.74±0.24	0.270±0.05	2.61±0.33	
8ng-BE (n=9)	1.74±0.18	0.210±0.04	2.18±0.31	
16ng-BE (n=6)	1.74±0.36	0.304±0.13	1.72±0.36	
CTL (n=7)	1.22±0.15	0.456±0.11	1.93±0.26	
10ng INS (n=6)	2.32±0.32**	0.[illegible]8±0.10	2.47±0.42	
10ng INS 8ng-BE(n=6)	2.28±0.21**	0.3[illegible]6±0.13	2.14±0.44	**p<0.01
10ng INS 16ng-BE(n=6)	2.23±0.33**	0.[illegible]1±0.17	2.61±0.35	

As shown, BE did not alter SM glucose uptake or metabolism and the 2-fold increase in glucose uptake produced by INS was unaltered by the addition of BE. On a molar basis, the concentrations of BE were 2 and 4 times those of INS. This study supports in vivo studies that have found no difference in glucose clearance after the IV injection of BE. In addition, it suggests that the action of naloxone on glucose uptake in SM is not mediated through an antagonism of BE and that BE does not mediate insulin resistance in SM through a direct effect.(This work supported in part by a NIH grant(GM27946) and a scholarship from The American College of Surgeons.

111

IN VIVO MONITORING OF HIGH ENERGY PHOSPHATES IN SKELETAL MUSCLE DURING HEMORRHAGIC SHOCK AND RESUSCITATION. Chih-Hsiung Wu*, Brenda G Nichols*, James W Holcroft and George C Kramer, Depts. of Surgery and Human Physiology, University of California, Davis, USA, and Dept. of Surgery, Taipei Medical College, Taipei, ROC.

Phosphorous NMR spectroscopy allows non-invasive monitoring of intracellular energy substrates (J Trauma, 24:S154, 1984). In the present study we used an Oxford Topical Nuclear Magnetic Resonance System, TMR, to monitor intracellular levels of ATP, creatine-phosphate (Cr~P), inorganic phosphate (P_i) and pH in biceps femoris during shock and resuscitation. Inactin anesthetized rats (250-350 g) were bled to a mean arterial pressure (MAP) of 50 mm Hg for 45 minutes (group I) or 90 minutes (group II). Then both groups were resuscitated with lactated Ringers until MAP was 90 mm Hg. Resuscitation volumes were 3-4x shed blood volume. After 45 minutes of hemorrhage P_i was increased 300-500%; Cr~P was decreased to 35-50% of baseline; ATP was slightly decreased to 92-95% of baseline. Intracellular pH decreased from its baseline range of 7.0-7.1 to 6.8-6.9. After 90 minutes of hemorrhage P_i was increased over 500%; Cr~P averaged 15% of baseline; ATP was 74% and pH was 6.4. Intracellular ATP levels were well maintained until the Cr~P/P_i ratio fell to 0.2 or below. Resuscitation after 90 minutes of hemorrhage restored MAP, but failed to affect any intracellular variable. Resuscitation after 45 minutes caused a partial improvement of intracellular phosphate levels, but all values remained substantially below baseline. These data show that despite restoration of circulatory volume and pressure with large volumes of lactated Ringers a significant intracellular energy deficit remains in skeletal muscle after hemorrhagic shock.

112

EVIDENCE FOR MEDIATORS IN ALTERED GLUCOCORTICOID ACTION DURING ENDOTOXIC SHOCK. MOLLY R. HILL*, REX D. STITH*, AND RODERICK E. MCCALLUM. University of Oklahoma Health Sciences Center, Oklahoma City, OK 73190.

The purpose of this study was to determine if the effects of endotoxin (Etx) on altered glucocorticoid activity and plasma glucose levels (glc) are the result of mediators. Steroid binding in liver cytosol, glc, and corticosterone (cort) levels were assayed in C3HeB/FeJ (FeJ) LPS normoresponsive and C3H/HeJ (HeJ) LPS hyporesponsive mice to compare these animals for use as donors and recipients in passive transfer experiments. Mice received 400 µg (LD_{50}) of S. typhimurium Etx i.p. and were killed 6h later. In FeJ mice, Etx depressed the maximum number of glucocorticoid binding sites (Bmax) from 122±15 fmol/mg to 43±8 fmol/mg, but did not alter the affinity (Kd) of the receptor. Glc levels were decreased to 50% of control and cort levels increased 3-fold. No changes in these parameters were seen in HeJ mice given Etx. FeJ peritoneal exudate cells (PEC), elicited by thioglycollate, were transferred i.p. to HeJ mice (2.5×10^6 cells/mouse) and 30 min later, the HeJ recipients were injected with Etx (400 µg). Mice were killed 4-6h later. Steroid binding was depressed to 80% and glc levels to 82% of controls. Similar results were obtained when FeJ PEC were incubated in vitro with Etx and the supernatants transferred to HeJ mice. Decreased binding and glc levels were also seen in HeJ mice given mediator-rich BCG-Etx serum i.v. These results indicate that the perturbation seen in the molecular mode of glucocorticoid action during endotoxic shock is mediated. A likely source of mediator(s) is the mononuclear phagocyte.

113

SEPSIS INDUCED CORONARY VASODILATATION. R. SHAPIRO*, M.J. BRESLOW*, C. MILLER* and R.J. TRAYSTMAN*. (INTRODUCED BY: M. ALLO). Department of Anesthesiology and Critical Care Medicine, The Johns Hopkins Medical Institutions, Baltimore, MD 21205.

Myocardial oxygen consumption (MVO_2) and coronary blood flow (CBF) decrease during hemorrhagic shock. In contrast, CBF is reported to increase during septic shock (SS) at similar levels of mean blood pressure (MBP). We measured CBF (microspheres) and MVO_2 in a porcine model of SS to determine if increased CBF in SS is due to increased metabolic activity of the heart. Ketamine anesthetized pigs (n=6, 40-50 kgs.) were ventilated to maintain $PaCO_2$ at 40 mmHg., and left atrial and great cardiac vein catheters were inserted. MBP, heart rate (HR), central venous and pulmonary wedge pressures, cardiac output (CO), MVO_2 and CBF were measured before and 90 mins. after the infusion of live E. coli (≈10^{12} organisms). E. coli infusion decreased MBP (105 ± 3 to 58 ± 4 mmHg), SVR (1277 ± 79 to 733 ± 40 dynes-$sec^{-1}cm^{-5}$) and did not change CO or HR. CBF did not change (105 ± 4 to 124 ± 15 ml/min) despite a 60% decrease in coronary perfusion pressure (80 ± 4 to 32 ± 3 mmHg). Coronary

vascular resistance decreased 63% (603 ± 40 to 224 ± 40 dynes-sec^{-1}cm^{-5}), MVO_2 decreased 31% (12.0 ± 0.8 to 8.3 ± 0.5 cc/min/100 gm.) and coronary venous O_2 content increased 311% (1.8 ± 0.2 to 5.6 ± 0.8 vol%). These results suggest that coronary arterial vasodilatation occurs during SS which is independent of myocardial metabolic activity. Coronary vasodilatation may be due to either direct vascular effects of E. coli or to indirect effects of mediators released during SS.

114

CHANGES IN CENTRAL HEMODYNAMICS AND SYMPATHETIC NERVE ACTIVITY DURING SEPTIC SHOCK IN CONSCIOUS RATS: S. LUNDIN*, J. PALSSON* & S.-E. RICKSTEN* (Introduced by: Hengo Haljamäe). Dept. of Physiology, University of Göteborg, Sweden.

Animal models may be of great value to study the pathophysiology of human septic shock. In this study lethal septic shock was induced by continuous infusion of live E. coli (10^9/h i.v.) in conscious rats (survival 2-4 h). Cardiac output (CO) was measured by dye-dilution technique. Renal sympathetic nerve activity (RSNA) was recorded in a separate group of conscious rats. The early response to bacteria infusion was a slight decrease in mean arterial pressure (MAP) from 119±3 mm Hg (n=9) before infusion to 112±3 mm Hg at 1 h while heart rate (HR) increased markedly (375±9 to 436±6 b/m at 1 h, p<0.001). Renal sympathetic nerve activity increased in parallell to the increase in HR. Respiratory rate increased from 90±6 to 119±10/min, pCO_2 decreased from 34±1 mm Hg to 26±2 mm Hg (p<0.001) while pH increased from 7.47±0.01 to 7.51±0.01 at 1 h (p<0.05). Cardiac output (CO) 25.0±3.1 ml/min x 100 g before infusion initially increased to 29.6±3.1 ml/min x 100 g (n.s.) at 1 h. At 2 h, when MAP was 107±4 mm Hg, a significant decrease in CO was observed (19.2±1.5 ml/min x 100 g b.w. although HR and RSNA remained markedly elevated. In this septic shock model a general excitation of autonomic centres appears to be present as shown by the increased sympathetic nerve activity, heart rate and ventilatory drive. The increased ventilation does not appear to be secondary to a metabolic acidosis but due to activation of respiratory centres. Studies of conscious, in contrast to anesthetized, animals appear to be of great value since anesthesia might influence these responses to experimental septic shock.

115

PLASMA CATECHOLAMINES IN CONSCIOUS RATS DURING SEPTIC PERITONITIS. S. B. JONES*, M. F. KOVARIK* and F. D. ROMANO*. (Introduced by: Mohammed M. Sayeed) Department of Physiology, Loyola University Medical Center, Maywood, IL 60153.

The present study examines plasma norepinephrine (NE), epinephrine (E) and dopamine (DA) as well as blood pressure (MAP) and heart rate (HR) following cecal ligation and puncture (CLP). Fasted male rats were subjected to carotid cannulation and CLP under halothane anesthesia between 1500 and 1700 hr. Mortality was 80% at 48 hrs. At 16,20,24 and 41 hrs following surgery, blood samples were withdrawn for catecholamine analysis. MAP and HR were monitored at these times for 30 to 60 min. Data are as follows:

Time Post CLP	16 hrs	20 hrs	24 hrs	41 hrs
Plasma NE*	961±79(10)	1233±158(9)	1273±133(7)	1702±306(4)†
Plasma E*	315±57(10)	330±93 (9)	211±66 (7)	193±87 (4)
Plasma DA*	353±54(10)	344±80 (9)	355±108(7)	278±63 (4)
MAP (mmHg)	127±6 (9)	128±5 (9)	127±9 (4)	115±7 (3)
HR (per min)	461±25 (5)	473±25 (5)		

values are mean ± SEM, ( ) = N, *=picograms/ml, †=p<0.05 vs 16 hrs

Cardioselective beta adrenergic blockade (Atenolol, IV) at 20 hrs significantly reduced heart rate within minutes by 130 bpm. MAP fell immediately and at 4 hrs was reduced 26 mmHg. Elevated plasma NE values (vs control not shown) suggest overflow of nerve-stimulated release and are consistent with increased peripheral NE turnover during CLP (*Fed. Proc. 43*: 415, 1984). Decreases in HR with beta blockade suggest increased adrenergic stimulation of the myocardium.(Loyola U. BRSG USPH5SO7RR05368)

116

DIMINISHED VASCULAR CONTRACTILITY TO NOREPINEPHRINE IN EXPERIMENTAL SEPSIS. T.M. MCKENNA*, F.A. BRIGLIA*, B. CHERNOW, B.L. ROTH*. Naval Medical Research Institute, Bethesda, Maryland 20814-5055.

Catecholamine unresponsiveness is a characteristic finding in patients with sepsis. We therefore compared vascular contractility in vitro in response to norepinephrine (NE) between thoracic aortic rings isolated from rats made septic by cecal ligation and bowel puncture for 24 h (N=4), and sham-operated controls (N=4). Aortic rings were suspended between two hooks within a perfused organ bath, under a resting tension of 3.3 g. Patency of the vascular endothelium was tested by application of acetylcholine (Ach) during a NE-induced contraction. Cumulative dose-responses to NE ($5X10^{-10}$ to 10^{-5} M) were determined, and the effect of NCDC (a phospholipase C inhibitor) on NE-induced contraction was measured. Ach induced a prompt relaxation and NCDC a slower decrease in contraction in septic and sham tissue, although both responses were qualitatively less in the septic preparation. Septic aortas displayed a significantly reduced contractile response to NE ($p < 0.05$ from 10^{-7} M to 10^{-5} M) with a 45% reduction in force at the highest dose (septic: 372±87.1 mg tension/mg tissue vs sham: 684±24.6). No difference in the ED_{50} (40nM) for NE was apparent. In conclusion: 1) A significant impairment of vascular contractility in response to NE, with no change in ED_{50}, persists in a septic vascular preparation in vitro. 2) The vascular endothelial response to Ach is intact at 24 h. We suggest that impairment of phospholipase C, by toxic factors or by loss of alpha-1 adrenoreceptors, may contribute to the decreased contractility in this model.

117

VASCULAR FUNCTIONAL RESPONSES IN ENDOTOXIN SHOCK. P. Kutsky, J. L. Parker. Dept. of Physiology, Texas College of Osteopathic Medicine, Fort Worth, TX 76107.

Previously we reported that in vitro examination of aortic strips and rings from a guinea pig model of endotoxin-induced circulatory shock revealed an increased contractile response to 60 mM KCl, but no alteration in responses to norepinephrine(NE). Preliminary experiments evaluating responses to added Ca in a zero Ca, 60 mM KCl-substituted solution indicated that shock tissue was more sensitive to low Ca concentrations (36-72 µM). These data suggest an alteration in extracellular Ca handling by vascular smooth muscle in endotoxin shock. Therefore, we examined the Ca dose response curve of control and shock aortic rings in zero Ca, 5.4 mM KCl solution. Shock rings exhibit greater tension development than control rings at physiological Ca concentrations. Pretreatment with 50 µM D600 reduced contractile tension in control rings at low Ca levels (<12 µM) and lowered the shock ring Ca dose response curve to the same level as the control response curve in the presence of D600. Gentamicin, which depresses vascular smooth muscle contraction via a Ca related mechanism, had similar inhibitory and relaxing effects on control and shock aortic rings whether contracted by 60 mM KCl or 10^{-7} M NE. Although electron microscopy demonstrated endothelial lifting in shock aorta, the relaxing effect of 10^{-6} M acetylcholine on 10^{-7} M NE induced contraction was not significantly altered in shock rings. Thus, aortic tissue isolated from endotoxin shocked guinea pigs has an altered Ca response which can be antagonized by D600.

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118

SENSITIVITY OF RIGHT ATRIA TO β-ADRENERGIC STIMULATION IN EARLY SEPSIS. L.W. SMITH*, S.A. WINBERRY*, H.I. MILLER, K.H. MC DONOUGH*. L.S.U.M.C., New Orleans, LA 70112.

Early in sepsis, in vitro depression of myocardial function has been observed, yet there is an apparent increase in chronotropic sensitivity to β-adrenergic stimulation. To eliminate the contributions of changes in inotropic state and in α-adrenergic and muscarinic effects, we measured the chronotropic sensitivity of isolated right atria (RA) to isoproterenol (IPR) stimulation. RA were removed from rats 24 hours following the induction of sepsis by cecal ligation and puncture (CP). At that time, in comparison to sham operated (SH) rats, CP rats demonstrated an elevated heart rate (HR). The chronotropic response of the RA to IPR was studied by incubation of the RA in 10 ml organ baths, using Krebs-Ringer bicarbonate buffer con-

taining 10 mM glucose, 5 μM 17-β-estradiol, 300 μM ascorbate, 2.25 mM $CaCl_2$, 30 μM EDTA $(Na)_2$, at pH 7.4, 34°C., equilibrated with $95\%O_2$:$5\%CO_2$. Prior to IPR stimulation, RA were incubated for 30 minutes with 10 μM dibenamine to irreversibly inhibit α-adrenergic and muscarinic receptors and subsequently were primed with 1 nM IPR. Two cumulative dose response curves were obtained for each RA. There was a significant decrease in the IPR dose at which the half-maximal HR response was seen in CP vs. SH rats (CP= $1.2 \pm .5 \times 10^{-11}$M; SH=$1.2 \pm .5 \times 10^{-9}$M) but no significant difference in the maximal HR response (CP = 371 ± 34; SH = 362 ± 29 beats per minute (BPM)). Basal HR was significantly higher in CP (Cp = 260 ± 17; SH = 215 ± 10 BPM). These results are consistent with our earlier results utilizing Langendorff perfused hearts in which increased chronotropic responsiveness was demonstrated in early sepsis. This increase is probably not mediated via α-adrenergic or muscarinic mechanisms.

119

SEPTIC AND ENDOTOXIC RAT HEART RESPONSE TO CATECHOLAMINES. A.L. ZARITSKY*, S. SHARP*, N. IVES*, B. CHERNOW. Naval Medical Research Institute, Bethesda, MD 20814-5055.

Myocardial dysfunction is a complication of septic and endotoxic shock, but is incompletely characterized. This study evaluated the response of the isolated heart from normal (N), septic (S), or endotoxic (Endo) rats (n=4, each group) to catecholamines. Hearts were rapidly removed from CO2-anesthetized rats at 48 hours post cecal ligation, or 30 minutes post a 20 mg/kg IV dose of E. coli endotoxin, and were perfused in a Langendorf preparation; a balloon was inserted into the LV for pressure measurement. Epinephrine (E, 0.1 μg/min), dopamine (DA, 2 μg/min), and dobutamine (Dob, 1 μg/min) were infused in random order for 10 mins each following a 10 min control period. Changes in heart rate (HR), developed LV pressure (dP), and dP/dt were compared to preceding control; the group responses were compared to those of the N rat. Blood cultures were positive in the S group only. E produced significant ($p < 0.05$) increases in HR and dP in N rats; HR increased in Endo and S rats. Dob increased HR and dP/dt only in S rats who also had an increased chronotropic response to E compared to N. Catecholamine response in S and Endo rat hearts was similar; however catecholamines caused more chronotropy in S rats than N rats. DA was a weak inotrope in all groups, even when infused at 2-20 times the rate of the other drugs. Conclusions: 1) S rats are more likely to have catecholamine-induced tachycardia than N rats. 2) In our models of experimental sepsis, Dob and E are superior inotropes to DA.

120

MYOCARDIAL PERFORMANCE AND ADRENERGIC MODULATION OF CYCLIC AMP LEVELS FOLLOWING ENDOTOXIN ADMINISTRATION. R.E. SHEPHERD*, C.H. LANG, AND K.H. McDONOUGH*. Department of Physiology, L.S.U. Medical Center, New Orleans, LA 70112.

The in vivo injection of E. coli endotoxin (ET) depresses myocardial performance in situ and in isolated heart preparations. We asked whether myocardial dysfunction was ET dose dependent and whether the observed depression was related to an impaired beta-adrenergic responsiveness. Hearts were removed from rats injected with saline or ET (10, 100 or 1000 ug/100 g) and studied 4 hr post-ET as isolated working heart preparations or were perfused with collagenase for isolation of myocytes. Starling curves of working hearts from animals injected with 100 ug or 1000 ug ET were depressed compared to controls; 10 ug of ET produced no significant myocardial depression. Myocytes from rats given 1000 ug ET showed blunted cyclic AMP (cAMP) accumulation in response to beta-adrenergic receptor challenge (1nM-10 uM ISO); animals injected with 10 and 100 ug ET did not show the blunted cAMP. Accumulation of cAMP in the presence of 1 uM Forskolin (FOR) was also depressed in myocytes from the high-dose ET-treated animals. Challenge with both ISO+FOR produced comparable cAMP levels to control values. This suggests a dose-dependency in the myocardial depression induced by ET with an apparent dissociation between the dysfunction noted in whole organ function compared to isolated myocyte beta-adrenergic responsiveness. Blunted myocyte hormonal responsiveness following ET may be attributed to alterations in both receptor coupling to adenylate cyclase and in the adenylate cyclase itself.

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121

HYPOCALCEMIA ASSOCIATED ELECTROMECHANICAL DISSOCIATION FOLLOWING TRANSFUSION OF CITRATED PLASMA DURING RESUSCITATION FROM HYPOTHERMIC HYPOVOLEMIC SHOCK. R.C. DENNIS, M.D. PALTER*, N.S. YESTON, R.C. GRASBERGER*, T.K. McINTOSH, C.R. VALERI*, Naval Blood Research Laboratory and Section of Critical Care Medicine, Division of Surgery, Boston University Medical Center, Boston, MA 02118

Disagreement exists whether administration of citrate phosphate dextrose (CPD) preserved blood products during resuscitation reduces blood ionized calcium. Nine male baboons were externally cooled (32°C) and bled to a mean arterial pressure of 45 mmHg. During initial hypothermia (from 37° to 32°C) and 30 minutes of shock no reduction in ionized calcium occurred. Following a 30 minute hypovolemic shock period the animals underwent an exchange transfusion of 1-3 times their blood volume with frozen washed red blood cells, CPD preserved fresh frozen plasma, and normal saline over 37 ± 12 minutes. Mean blood pressure was maintained at 45 mmHg and temperature at 32°C. Electromechanical dissociation, consisting of normal sinus ECG with an arterial pulse at one-half the ECG rate, was observed in all animals and was associated with a fall in mean blood ionized calcium from 4.27 ± 0.29 mg/dl to 1.84 ± 0.73 mg/dl. This cardiac arrhythmia was immediately reversed after the administration of 10% calcium chloride the restoration of ionized calcium levels. We conclude that rapid infusion of CPD preserved blood products may produce electromechanical dissociation during hypothermic, hypovolemic shock which can be reversed with calcium chloride.

122

MYOCARDIAL ENERGY DEFICIT DURING ACUTE ENDOTOXIN SHOCK IN THE DOG. R.M. Raymond, D.M. Klein, P.M. Kober, D.A. Gibbons, Depts. of Surg. and Physiol., Loyola Univ. Med. Ctr. 60153 and the VA Hosp., Hines, IL 60141

Several reports have described the occurrence of myocardial failure during various shock protocols. Since there is lack of information regarding myocardial energy metabolism during shock, the present study was undertaken to define the temporal changes in myocardial high energy phosphate during shock. Mongrel dogs (18-25kg) were anesthetized with Nembutal, intubated and ventilated with room air. Serial transmural biopsies (3mm dia.) were harvested along the anterior free wall of the left ventricle, using an Alko pneumatic biopsy drill. Endotoxin shock was induced by injecting 1mg/kg i.v. E. coli endotoxin. The following table lists epicardial (epi) and endocardial (endo) ATP and CP concentrations during endotoxin shock. (*=p<0.05)

		Time (hrs)			
	Control	1	2	3	agonal
ATP (μM/g wet wt.)					
Epi	6.7	5.5	6.0	5.2	3.6*
Endo	5.7	5.5	5.2	3.1*	4.0*
CP (μM/g wet wt.)					
Epi	13.6	6.6*	4.9*	5.4*	0.2*
Endo	13.2	7.6*	6.3*	7.9*	0.2*

These data demonstrate that endotoxin shock induces a myocardial energy deficit, which occurs only as an agonal event. Reductions in ATP result from an inability by CP to maintain ATP within normal range and suggest reductions in ATP by 30-40% are consistent with decreases in myocardial performance and death. (Supported by the V.A.)

123

ALTERATIONS IN α_1 ADRENERGIC RECEPTOR BINDING IN A RAT MODEL OF CHRONIC SEPSIS. M.R. MCMILLAN*, B. CHERNOW, B.L. ROTH*. Naval Medical Research Institute, Bethesda, Maryland 20814-5055.

Radioligand binding assays were performed in the cecal ligation 2-puncture rat model of chronic sepsis (Wichterman, et al, J Surg Res 29:189, 1980) 48 hours after ligation and puncture. Using [^{3}H] prazosin, rat liver was identified as a rich source of α_1 adrenergic receptors. Binding data were analyzed using a computerized non-linear least-squares curve fitting technique (LIGAND) as previously described (Roth and Coscia, J. Neurochem. 42:1677, 1984). Antagonist/-antagonist competition binding data were best fit to a one site model of receptor-ligand interaction in control and experimental groups. Binding parameters (mean ±

S.D., n = 3) were determined to be: K_D: 0.34±0.06 nM (control) and 0.22±0.16 nM (experimental) ($p > 0.05$ vs control); Bmax: 172±13 fmole/mg (control) and 102±10 fmole/mg (experimental) ($p < 0.05$ vs control). Some heterogeneity was noted in the down-regulation of α_1-receptors in experimental animals which seemed related to the degree of sepsis.[1] The data demonstrate significant down regulation of the α_1 adrenergic receptor with no change in antagonist affinity in this model of chronic sepsis; this adrenergic receptor abnormality may underlie catecholamine unresponsiveness in clinical sepsis.

124

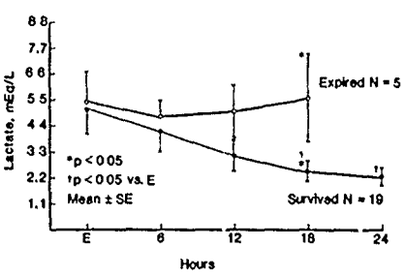
REGIONAL BLOOD FLOW DURING CHRONIC, NONLETHAL ENDOTOXEMIA. J.A. Spitzer, C.H. Lang and R.E. Fish, Dept. of Physiology, LSU Medical Ctr., New Orleans, LA 70112.

We have previously described metabolic alterations in hepatocytes and adipocytes isolated from rats continuously infused with nonlethal doses of E. coli endotoxin (ET). Alterations in cardiac output (CO) and regional blood flow during chronic endotoxemia were examined in order to evaluate the influence of in vivo hemodynamic changes on tissue metabolism. ET was infused iv from a subcutaneously implanted pump (Alzet) at a rate of 0.6mg/100 g/day. CO and regional blood flow were determined in chronically catheterized conscious rats at 6 and 30 hrs post-ET using radiolabeled microspheres (15 μm, ^{141}Ce, ^{85}Sr, ^{46}Sc). Time-matched control animals had a stable MABP (105 ± 2 mmHg) and CO (373 ± 20 ml/min/kg) over the 30 hrs of the experiment. ET rats were hemodynamically stable throughout the 30 hrs with CO remaining at control levels and MABP showing only a 13% decrease after 30 hr. Pancreatic flow (ml/min/g) was reduced by 40% after 6 hrs, with no changes in epididymal fat or hepatic flow seen at this time. After 30 hrs of ET, the blood flow to both the pancreas and lung was reduced (48 and 63%, respectively). Hepatic arterial flow was elevated 4-fold, while portal venous and total hepatic flow were not significantly different. Blood flow to other organs was stable over the course of the experiment. These results indicate that chronic ET infusion induces cellular metabolic changes in the absence of a fall in CO and tissue hypoperfusion. (Supported by GM 30312 and HL 07098 and the Am. Heart Ass., LA).

125

DELAYED LACTATE CLEARANCE IN PATIENTS WITH SHOCK. J.L. FALK,* E.C. RACKOW, J.A. LEAVY,* M.H. WEIL. UHS/The Chicago Medical School, North Chicago, IL 60064

We studied 24 patients with circulatory shock to assess the time course of arterial lactate clearance after fluid resuscitation. Patients had an initial arterial lactate (L) > 2mEq/l, pulmonary artery wedge pressure (PAWP) < 15mmHg, and systolic intra-arterial pressure < 90 mmHg or cardiac index (CI) < 2.2 L/min/M^2. The CI and L were measured prior to fluid resuscitation, at the end of fluid resuscitation (E) and 6, 12, 18, and 24 hrs later. Survivors (N = 19) after resuscitation were compared to fatalities (N = 5).



The CI increased from 2.4 ± 0.2 (SE) to a peak level of 3.5 ± 0.2 L/min/M^2 in survivors and from 1.9 ± 0.3 to 2.9 ± 0.6 L/min/M^2 in fatalities ($P<0.05$). The L clearance is shown in the graph (mean ± SE, *$P<0.05$ between groups; **$P<0.05$ compared to E). Lactate progressively cleared in the survivors such that, at 18 hours, the L decreased from 5.2 + 1.0 to 2.5 + 0.3 mEq/l ($P<0.05$). By comparison, the patients who expired did not clear lactate but rather L increased. These data provide evidence that reduction in lactate in patients surviving shock is substantially delayed and that prolonged lactate clearance may not be an unfavorable indicator of outcome.

126

ADRENALECTOMY POTENTIATES THE ENDOTOXIN-INDUCED CARDIODEPRESSANT EFFECT OF SERUM IN THE RAT. A. CARLI, M.C. AUCLAIR, C. VERNIMMEN. Institut de Pharmacologie, 15 rue de l'Ecole de Médecine F-75006. Previously we showed that endotoxin administered to intact rats (IR) at a sublethal dose (2 mg/kg i.v.) was nonhypotensive but able to induce the early and prolonged (at least 4 hrs) release of a lipid soluble cardiodepressant factor (CDF), decreasing contractility of cultured rat heart cells (CRHC) by about 35%. In another connection it is established that adrenalectomy depresses cardiac contractility through an unclear mechanism and worsens shock mortality. The present study was designed to determine if the endotoxin-induced seric CD effect was potentiated in adrenalectomized (6 days before use) Na-supplemented rats (AR). In AR 2 mg/kg endotoxin caused severe hypotension and 100% lethality within the first 90 minutes. To obtain in AR the same seric CD activity on CRHC that in IR with a strict parallel in intensity and time course it was necessary to reduce the endotoxin dose by about 200 times (0.01mg/kg). The latter proved sublethal in AR and without seric CD effect in IR. When compared to sham-operated control, serum from AR (no endotoxin) induced a slight decrease in the CRHC contractility by about 9%. Conclusions : i) adrenalectomy potentiates the endotoxin-induced CD effect of serum in the rat; ii) cardiodepression observed in non-endotoxic AR could be at least partly humorally mediated. (Supported by INSERM CRL 825005)

127

THROMBIN ACTIVATED PROTEIN C AGAINST ENDOTOXIN. F.B. Taylor, A. Chang,* L. Hinshaw. Thrombosis/Hematology Research Program, Oklahoma Medical Research Foundation, Okla. City, OK 73104; and Veteran's Administration Hospital, Research Wing, Okla. City, OK 73104.

The purpose of these studies was to determine if an infusion of thrombin or activated protein C would protect dogs/baboons from endotoxin/Escherichia coli shock. Infusion of dogs with thrombin (0.5 U/kg/min) for 90 min significantly increased percent survival following infusion of endotoxin (0.06 mg/kg/min) for 30 min. Nine of fourteen dogs infused with thrombin survived more than seven days, whereas thirteen of fourteen dogs infused with saline died within 36 hours. Those dogs which survived responded to endotoxin with enhanced anticoagulant and fibrinolytic activity as measured by the Xa one-stage and fibrin degradation product assays, respectively. Those dogs receiving saline instead of thrombin did not respond with these activities until the end of the study. These studies were repeated on eight baboons. One received thrombin (1.5 U/kg/min) followed by Escherichia coli (E. coli) 2-3 x 10^{10} organisms/kg over 2 hours; two received activated protein C (0.01 mg/kg) every 10 min over 2 hours together with E. coli, and three received E. coli alone. The animals receiving thrombin or activated protein C had anticoagulant and fibrinolytic responses to E. coli and survived. The four control animals had minimal responses and died within 36 hours. We concluded that thrombin or activated protein C may protect dogs and baboons from endotoxin/E. coli shock and that this was associated with early anticoagulant/fibrinolytic response to endotoxin/E. coli infusion.

128

IBUPROFEN PRETREATMENT IN ENDOTOXIN SHOCK. R.R.Beck* and F.L.Abel. University of South Carolina School of Medicine, Columbia, SC 29208.

Dogs were pretreated with 10 mg/kg IV Ibuprofen (IBUP), a cyclooxygenase inhibitor, 30 minutes prior to the IV injection of 1 mg/kg E. Coli endotoxin (ETOX). Flow probes on the hepatic artery and portal vein and catheters in the femoral artery and vein and portal and hepatic veins allowed us to quantitate hepatic changes due to ETOX and the effects of IBUP pretreatment on these changes. With ETOX treatment alone arterial pressure was significantly decreased from control mean of 135 mmHg to 95 mmHg by 15 minutes and continued to slowly decline. Hepatic blood flow decreased by 15 minutes from 34 to 25 ml/min/kg and continued to fall. IBUP pretreatment prevented the fall in blood pressure and hepatic blood

flow during the measurement period, 180 minutes. Arterial glucose concentration was 141 mg/dl before ETOX treatment, increased to 240 mg/dl at 15 minutes then slowly decreased over the 3 hour measurement period. With IBUP pretreatment arterial glucose did not increase at 15 minutes and fell slowly during the measurement period although none of these animals had plasma glucose concentration less than 50 mg/dl. Hepatic glucose production was significantly higher in IBUP pretreated animals by 30 minutes, 10.2 mg/min/kg with ETOX alone vs 20.6 mg/min/kg with IBUP pretreatment, and remained higher for the remainder of the experiment. Hepatic uptake of lactate and glycerol was significantly greater in pretreated animals. Pretreatment with this nonsteroidal anti-inflammatory agent appears to improve some aspects of hemodynamics and hepatic metabolism.

129

THE RELATIONSHIP OF PLASMA GASTROINTESTINAL GLUCAGON CONCENTRATION TO GASTROINTESTINAL MUCOSAL HEMORRHAGE DURING SEVERE SEPSIS. K. ISHIDA, G. WILLIAMS*, L. ARCHER, L. HINSHAW. VA Medical Center and University of Oklahoma Health Sciences Center, Departments of Surgery, Physiology, and Pathology, Oklahoma City, OK 73104.

Sepsis is a frequent cause of multiple organ failure including hemorrhagic lesions of the gastrointestinal tract. We have reported that concentrations of plasma gastrointestinal-derived glucagon are markedly elevated in dogs administered LD_{100} E. coli. We hypothesized that these increased concentrations might be related to the degree of gastrointestinal mucosal hemorrhage. Eighteen adult dogs were separated into groups: group I - LD_{100} E. coli alone, group II - LD_{100} E. coli + tobramycin (TOB), group III - LD_{100} E. coli + TOB + methylprednisolone sodium succinate (MPSS). Animals were infused intravenously for 1 hr with 1.1×10^{10} E. coli/kg body wt, monitored for 6 hr, and observed for a 7-day recovery period. Glucagon concentrations were determined by means of an RIA utilizing polyethyleneglycol. Postmortem examinations were performed on all dogs. Percent survival (>7 days): I=0%, II=17%, III=83%. Gastrointestinal glucagon concentrations were markedly elevated in groups I and II. The early rise of gastrointestinal glucagon (0-6 hr) was attenuated in MPSS/TOB-treated dogs (group III) and progressively decreased after 18 hr. These decreases were associated with increased survival. Mucosal hemorrhages were observed in 60% of the animals in group I, 50% in group II, and 17% in group III. Results suggest that during severe sepsis increases in plasma gastrointestinal glucagon concentration may be related to the degree of mucosal hemorrhage in the gastrointestinal tract.

130

ANTIBIOTICS, CORTICOSTEROIDS AND FLUID INFUSIONS ALONE AND IN COMBINATIONS FOR TREATMENT OF EXPERIMENTAL SEPTIC SHOCK. J. OTTOSSON*, I. DAWIDSON, R. SIMONSEN*. University of Texas Health Science Center, Dallas, TX 75235.

The multidimensional pathophysiology of septic shock suggests several therapeutic approaches. This study evaluates intravascular volume, lung water changes, O_2 cons., and survival in septic shock in rats, treated with antibiotics, fluid infusion, and corticosteroids (CS) alone and in combinations. Sepsis was induced by IP injection of E. Coli (5×10^8 /100g bwt) bacteria. Untreated rats had decreased PV from 4.5 to 3.5 ml/100g bwt at 5.5 hrs ($p < 0.001$) and O_2 cons. from 20.5 to 14.3 ml/100g bwt ($p < 0.001$). The antibiotic drug given together with CS (Dexamethasone 8 mg/kg bwt) prevented further drop in PV but did not prevent further drop in O2 cons. (9.8 ml/kg). The combination of all three therapeutic agents resulted in a PV increase to 7 and 25% ($p < 0.01$) above preshock level for RL (30 ml/100g bwt) and Albumin 3% (7.5 ml/100g bwt) respectively. O_2 cons. was 17.8 and 7.0 ml/kg bwt for Albumin and RL respectively. No septic untreated animals survived 24 hrs. The combination of antibiotics and CS yielded a 60% survival rate and 100% when Albumin 3% was added ($p < 0.05$). Extravascular lung water (EVLW) was 73% in control animals. Septic animals increased this value to 77% and 80% at 5 and 9.5 hrs respectively ($p < 0.05$). Treatment with CS in combination with 3% Albumin had no further increase in lung water (77%). RL in combination with CS resulted in a significantly larger lung water increase to 83% at 9.5 hrs ($p < 0.01$). It is concluded that the treatment combination of antibiotics, CS and 3% colloid solution infusion is the most effective treatment of experimental septic shock.

131

SINGLE- VS. MULTIPLE-DOSE TOBRAMYCIN THERAPY FOR LETHAL SEPSIS IN THE DOG. B. BELLER, L. ARCHER, A. CHANG*, C. MURRAY*, K. ISHIDA, V. O'MALLEY*, D. FLOURNOY*, R. PASSEY*, L. HINSHAW. Veterans Administration Medical Center, Okla. City, OK 73104.

Antimicrobial effectiveness and effect on survival of single- vs multiple-dose aminoglycoside antibiotic therapy (with and without steroid) for lethal sepsis was evaluated. Thirty adult mixed-breed dogs were anesthetized, divided into 5 equal groups and infused iv for 1 hr with E. coli. Group A was given no drug. Group B was given a 45 mg/kg 10-min iv injection (inj) of tobramycin (TOB) at 65 min. Group C was given TOB: 3 mg/kg 10-min iv inj at 65 min, then 8.25 mg/kg iv infusion for 285 min, and 11.25 mg/kg im inj at 6, 12, and 18 hr (total 45 mg/kg). Group D was given the same TOB regimen as B plus a 30 mg/kg iv inj and 30 mg/kg iv infusion of methylprednisolone sodium succinate (MPSS) from 15 to 360 min. Group E was given the same TOB regimen as C plus the same MPSS regimen as D. Treated dogs also received 11.25 mg/kg TOB daily for 4 days. Percent survival (>7 days): A=0%, B=0%, C=17%, D=83%, E=83%. By 4 hr, TOB-treated groups had fewer E. coli/ml blood than group A ($p<0.05$) and group B had fewer than C, D, or E ($p<0.05$). High "trough" serum TOB concentrations were associated with death and very low levels with recovery. Serum urea nitrogen and creatinine increased in all groups, but returned to normal by 7 days in survivors. Blood pressure decreased in all groups, but began to recover by 3 hr in D and E. Serum glucose levels of D and E did not decrease. Serum cortisol levels increased in all groups, but were lower than normal from 48-96 hr in groups D and E. In summary, single-dose TOB was an effective antimicrobial agent. However, recovery from E. coli sepsis was dependent on TOB plus MPSS and was not achieved with TOB single- or multiple-dose therapy alone.

132

ANTIBIOTICS BUT NOT NALOXONE OR METHYLPREDNISOLONE INCREASE SURVIVAL IN A RAT MODEL OF SURGICAL SEPSIS. S.J. Hollenbach*, L.R. DeGuzman*, R.F.Bellamy* (Sponsor: W.L. Traverso). Letterman Army Institute of Research, Presidio of San Francisco, Ca. 94129.

We used the peritonitis model of Witcherman et al (J. Surg. Res. 29: 189-201, 1980) (cecal ligation and puncture) to determine the effects of gentamicin (4.5mg/kg/day)- clindamycin (30mg/kg/day), naloxone (2mg/kg/hr), or methylprednisolone (2mg/kg/hr) on survival. Studies were carried out using 350 gram Sprauge-Dawley rats. Drugs were infused by Alzet minipumps (model 2001) for seven days. Rats were observed daily for three weeks or until they died. All nonsurvivors had purulent peritonitis and positive blood cultures.

Treatment Group	Number of Rats	%Surviving 5 days	%Surviving 10 days	$p<0.01$
Untreated (U)	63	46	46	* A vs U,N
Antibiotics (A)	43	86*	86**	** A vs U,N,S
Naloxone (N)	43	35	30	***S vs U
Methylprednisolone (S)	43	70	14***	

($p<0.01$ ANOVA and t-test with Bonferroni maneuver).

The median time of death in the untreated animals was 36 hours compared to 22 hours in animals given naloxone. Antibiotics were clearly life-saving, while naloxone had no perceptible beneficial effect on survival. Methylprednisolone had a curiously dichotomous effect, and early increase in survival followed by death of nearly all animals after the first week.

133

ARTERIO-VENOUS PLASMA THROMBOXANE IN EXPERIMENTAL AND CLINICAL RESPIRATORY DISTRESS SYNDROME (RDS)

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The lung has been implicated as a primary source of arachidonic acid metabolites in experimental and clinical RDS. This study was performed to determine if the lung is a source of thromboxane (Tx) production in dogs and patients with RDS. Lung injury was induced in dogs with 0.2 mg/kg oleic acid (OA) IV. All patients were septic with evidence of RDS. Simultaneous samples of blood were obtained from

pulmonary artery (V) and systemic arterial (A) catheters to measure immunoreactive (i)TxB_2. Results are expressed as iTxB_2 (pg/ml) ± SEM.

	Arterial		Venous
Control (n=7)	401 ± 63	Dogs	774 ± 211
0.2 mg/kg OA (n=7)	1783 ± 401		1640 ± 463
RDS (n=11)	484 ± 101	Patients	559 ± 131

Plasma iTxB_2 rose significantly ($P < 0.02$) following oleic acid in the dog. In patients plasma iTxB_2 also rose significantly ($P < 0.005$) compared to controls (91 ± 18 pg/ml n=6). However, there were no significant A-V differences in either the experimental or patient groups. The results show that plasma iTxB_2 is elevated by oleic acid and during sepsis with RDS. Since there was no A-V difference in iTxB_2, these results raise the possibility that there may also be extrapulmonary sources of Tx_2 in RDS.

134

COMPARISON OF LIPID A AND LIPID X ON VASCULAR ENDOTHELIAL CELL CULTURES. SUSAN L. GARTNER*. Naval Medical Research Institute, Bethesda, MD 20814-5055. (Introduced by: Bart Chernow)

Cultured bovine aortic endothelial cells are being used as a sensitive target system to study the mechanism of Lipid A-induced injury. Endothelial cell cultures were grown until confluent on 24-well culture dishes and exposed to various concentrations of either Lipid A (Calbiochem) or Lipid X (a gift from Christian R. H. Raetz). After 24 hours of exposure the cell release was determined by counting the cells in the supernatant.

Concentration (μg/ml)	Cell Release (% of Control ± SD) 0	0.1	1.0	10	100
Lipid A	100 ± 18	156 ± 19*	280 ± 130**	985 ± 390**	884 ± 64**
Lipid X	100 ± 23	85 ± 11	99 ± 25	78.5 ± 8.0	76.6 ± 11

* $p < 0.05$ ** $p < 0.01$

Data show that small amounts of Lipid A disaccharide were sufficient to cause endothelial cell detachment while Lipid X was not. Lipid X is a monosaccharide precursor of Lipid A which has been reported to share some of the same biological properties of Lipid A, such as B-lymphocyte mitogenecity, gelation of limulus lysate, and activation of protein kinase C. Lipid A and Lipid X, being chemically defined, may provide important probes for studying the mechanism of action of the parent lipopolysaccharide. Supported by Naval Medical Research and Development Command Work Unit No. MR041.2001.0436.

135

Effect of hemorrhage on regional microvascular reserve and flow in rat brain, G.J. Grover*, E. Francois-Dainville*, E. Buchweitz*, H.R. Weiss* Dept. of Physiol. & Biophys., Heart & Brain Circ. Lab., UMDNJ-Rutgers Medical School, Piscataway, N.J. 08854

The purpose of this study was to determine if mobilization of the microvascular reserve and redistribution of blood flow are used by the brain to compensate for hemorrhagic hypotension. Conscious Long-Evans rats served as controls or were bled to 40-45 mm Hg pressure and held at this pressure 30 min. Regional cerebral blood flow was then determined in half of these rats using ^{14}C-labelled iodoantipyrine as a marker. In the other half, fluorescein isothyocyanate-dextran was injected for viewing of the perfused microvessels. The brains were then analyzed for both perfused and total microvascular morphology. Mean cerebral flow was reduced from 58±28 (± S.D.) ml/100g/min in controls to 26±11 ml/100g/min with hemorrhage, with no regional differences. The overall percent of arterioles perfused was not different between hemorrhage (65±21%) and control animals (51±18%), though this was higher in several regions in hemorrhaged animals (hippocampus, hypothalamus, medulla). The overall percentage of capillaries perfused was higher in hemorrhaged rats (60±17%) compared to controls (45±11%). This mobilization of capillaries was especially pronounced in the pons region. Thus, with acute hemorrhagic hypotension in rat, the brain does not seem to redistribute blood flow regionally, but does appear to mobilize some of its unperfused microvascular reserve to compensate for a decreased flow, especially in the brainstem.

136

ALCOHOL INCREASES MORTALITY IN MURINE HEAD INJURY. C.D. FRANCO*, C.R. SPILLERT, K.R. SPILLERT*, E.J. LAZARO*, Department of Surgery, UMDNJ-New Jersey Medical School, Newark, N.J. 07103.

Head injury is a major factor in the mortality of traumatized patients accounting for about 50% of the resulting fatalities. Alcohol intoxication is frequently (25-50%) associated with head injuries. This study was undertaken to investigate the effects of alcohol on head trauma in a standardized animal model. Swiss Webster mice (25±2g) were given 0.2cc of either saline or 50% ethanol in saline IP. Thirty minutes later, under light ether anesthesia, severe concussion was produced by dropping a 39.5g lead weight from a height of 30cm. The trauma was centered on the midskull by channelling the weight through a vertical tube 1.2cm in diameter. Animals were observed daily for eight days. Survival rates (excluding those animals that did not survive the anesthesia) for each group at 4 and 8 days are shown below:

GROUP	SURVIVALS* 4 Days	8 Days	
Saline	12/12 (100%)	8/12 (67%)	*Significance p< .002
Alcohol	10/21 (48%)	1/21 (5%)	(Fisher's Exact Method)

Conclusion: This study clearly demonstrates that alcohol increases the lethality of standardized head trauma in mice. The mechanism by which alcohol modifies the effects of craniocerebral trauma remains to be elucidated.

137

COMPARISON OF ENDOTOXIN AND E. COLI INFUSION IN AWAKE YOUNG PIGS. E. Schrauwen, E. Cox, and A. Houvenaghel, Lab. of Vet. Physiol., State University Center, Antwerp, Belgium.

In conscious pigs (n=16; ±15 kg), the influence of E. coli endotoxin (2.5 mg/kg) and E. coli bacteria (5.10^8/kg) IV infusion (over 1 hour) on clinical and hemodynamic parameters, survival time, and necropsy changes was studied. Endotoxin induced nausea, vomiting, dyspnoea, cyanosis, excitement followed by depression, marked edema and hemorrhages in several organs, as well as significant changes in hemodynamic variables, e.g. a decrease in arterial pressure and cardiac output, an increase in heart rate and total peripheral and pulmonary vascular resistances. Survival at 30 hours amounted to 33%. E. coli bacteria provoked comparable clinical signs and necropsy findings, although often less pronounced. Small abcesses in kidneys and lungs were regularly observed. Hemodynamic variables changed similarly as in the endotoxin group. Survival at 30 and 96 hours amounted to 71 and 57% respectively. Endotoxin infusion therefore mimics E. coli bacteria infusion, probably as the result of an activation of similar pathophysiological mechanisms. Taking into account an endotoxin content of 10% of the bacterial mass, the E. coli bacteria infusion was the equivalent of ±15 µg/kg of endotoxin. Infused in this dose-range, however, endotoxin produces no mortality, comparable clinical signs, and less pronounced hemodynamic changes. (Supp. by I.W.O.N.L., Belgium).

138

THE EFFECT OF A SEROTONIN ANTAGONIST AND A THROMBOXANE SYNTHETASE INHIBITOR ON THE PULMONARY VASCULAR RESPONSE TO I.V. INFUSION OF LIVE E.COLI. E SVARTHOLM, J LJUNGBERG, D BERGQVIST & U HAGLUND. Dept of Surgery, Malmö General Hospital, Malmo, Sweden.

A release of serotonin and increased synthesis of thromboxane A_2 have been implicated as important factors in the pulmonary vascular changes following experimental bacteriemia. This was further explored in experiments on anesthetized pigs given live i.v. E.coli (10^9/ml; 1 ml/kg x min for 2 min followed by 1 ml/kg x h for 3 h). The animals were ventilated artificially. Cardiac output and pulmonary arterial pressure were monitored by means of a Swan-Ganz catheter. Different coagulation factors and variables reflecting fibrinolysis were followed. The following series of bacteriemic pigs was studied: untreated controls (n=6), pretreatment with kentaserin (a serotonin receptor blocker, Janssen; n=6), pretreatment with

UK 38 485 (a thromboxane A_2 synthetase inhibitor, Pfizer; n=7), or combined treatment with ketanserin and UK 38 485 (n=9).

Untreated pigs had a significant increase of pulmonary arterial pressure (from 16±1 to 41±3 mmHg) and signs of activated coagulation/fibrinolysis. Ketanserin had no effect on the pulmonary arterial pressure. However, coagulation factors were not equally activated. UK 38 485 attenuated but did not prevent the increase of pulmonary arterial pressure (14±2 to 28±4). The combination of ketanserin and UK 38 485 was not more effective than UK 38 485 alone.

It was concluded that thromboxane but not serotonin was an important factor for the pulmonary vascular response in pigs following i.v. bacteria.

139

PULMONARY HEMODYNAMIC AND VENTILATORY CORRELATES IN PORCINE SEPTIC SHOCK: THE EARLY ONSET OF PERMEABILITY CHANGES. G.J.-H. CHUANG☆ C.X. GAO☆ D.S. MULDER☆ R.C. J. CHIU. The Montreal General Hospital/McGill University, Montreal, H3G 1A4.

The purpose of this study is to correlate the temporal sequence of pulmonary hemodynamic, ventilatory and permeability changes in porcine septic shock, since the early responses of lung to sepsis remain controversial. Piglets(13-21kg) were lightly anesthetized, and breathed room air spontaneously. Gr.I (control, n=5) received i.v. normal saline (N/S) at 6-8ml/kg/hr. Gr.II (sepsis, n=7) received same volume of N/S with live E. coli (U9-41) at 4.5-10x10^8cfu/hr for 6 hours or until death. Hemodynamic and ventilatory (CO,PAP,PVR,BP physiologic shunts Qsp/QT; dead space VD/VT), blood gases Hct and wbc were monitored serially. Right lymphatic ducts were cannulated for lymph flow and lymph/serum protein ratio. Extravascular lung water (EVLW) was measured by gravimetric method at the end of experiments.

RESULTS:

	Base Line				1hr. post infusion(sepsis)				
	PAP (mmHg)	PaO2 (mmHg)	Qsp/QT (%)	VD/VT	PAP	PaO2	Qsp/QT	VD/VT	EVLW (ml/kg)
Gr.I	11±1.9	81±6.0	5.1±2.1	0.58±0.03	12.5±2.3	76±2.2	9.9±1.1	0.55±0.04	2.53±0.17
Gr.II*	8.6±1.6	79±4.7	10.2±2.2	0.55±0.04	34±1.7	42±2.2	48.8±4.2	0.70±0.01	5.51±0.67
P. Value	all> 0.05				<.001	<.001	<.001	<.01	<.01

Two pigs in Gr.II (*) died within 1hr. also with marked increase in EVLW(5.2 and 7.5 ml/Kg). Lung lymph flow increased in the 1st hr, reaching a peak at 2-3 hr. Striking leucopenia and hemoconcentration also occurred within 1hr. Thus this study elucidates the pathophysiologic progression of "Septic lung" in pigs and supports the concept that increase in pulmonary vascular permeability occurs early in septic shock.

140

ACUTE COR PULMONALE DUE TO PLATELET-ACTIVATING FACTOR OR TXA_2 ANALOG. F LAURINDO,* D EZRA,* J CZAJA,* G FEUERSTEIN, R GOLDSTEIN* USUHS, Bethesda, MD 20814.

Platelet-activating factor (PAF), a mediator released during anaphylaxis and endotoxemia, induces severe and even fatal shock state, while releasing thromboxane A_2 (TXA_2). However, the mechanism of such effect is unclear. We infused PAF (40-280 pmol/kg/min iv) or TXA_2 analog (U46619, 0.2-1μg/kg/min iv) for 1-7 min to open-chest, anesthetized pigs instrumented with aortic flowmeter, left ventricular (LV) and Swan-Ganz catheters and dimension gauges on both ventricles. With each PAF infusion (n=7) the primary hemodynamic change was a 5-120 fold rise in pulmonary vascular resistance (PVR; from 244±45(SE) to 11590±3279 dyneseccm^{-5}). Mean pulmonary arterial pressure rose from 19±1 to 45±2 mm Hg, while cardiac output (CO) fell from 2.3±0.1 to 0.3±0.1 L/min. Arterial pO_2 showed no consistent change. Despite increased systemic vascular resistance (SVR), PVR/SVR rose 10-fold. Systemic arterial pressure (SAP) decreased late at critically high PVR, from 94±3 to 34±5 mmHg. Right ventricular (RV) segment shortening fell as end-diastolic length increased. Right atrial pressure rose, with evidence of tricuspid regurgitation. LV diastolic pressures and dimensions fell significantly, indicating LV underfilling during PAF. Each of the PAF effects were reproduced by U46619 (n=6) or mechanical pulmonary artery constriction (n=5). TXA_2 synthesis blockade (indomethacin, 6 mg/kg) totally prevented SAP fall but prevented only 70% of PVR rise and 59% of CO decrease; PVR rise was significantly delayed. Thus, acute RV pump failure due to severe PVR rise is the primary mechanism of PAF-induced low-output state. This effect is mediated mainly but not exclusively by TXA_2.

141

THE EFFECTS OF A PLATELET ACTIVATING FACTOR (PAF) ANTAGONIST ON PAF AND ENDOTOXIN HYPOTENSION IN THE RAT. A. W. Mikulaschek*, P. D. Toth. Department of Medical Research, Methodist Hospital of Indiana, Inc., Indianapolis, IN 46202.

Research in recent years has implicated many vasoactive mediators in causing or maintaining the shock process. One of the newer candidates to be considered is PAF. The present group of experiments examined the effects of a reported specific PAF antagonist, CV 3988, on PAF and endotoxin hypotension. Sprague Dawley male rats (250-350 gram) were used in all experiments. Animals were lightly anesthetized with halothane and instrumented to measure mean arterial pressure (MAP) and placed in acrylic restrainers. Hypotension induces by PAF (3.0 ug/kg i.v.) was attenuated only by CV 3988 (10 mg/kg). The other compounds tested which were unsuccessful were TRH, benoxaprofen, fenoprofen, FPL 55712, naloxone, diphenhydramine, verapamil, and methylprednisolone. In another set of rats, hypotension was induced by endotoxin (E.coli - 0127:B8) (40 mg/kg). Animals were treated with saline or CV 3988 (10 mg/kg i.v.) 5 minutes prior to endotoxin administration. Below are listed representative MAP (mmHg) (mean ± SEM) (data were measured 60 minutes after endotoxin administration):

Saline (n = 5)	72 ± 7	
CV 3988 (n = 6)	99 ± 5 *	*$p \leq 0.05$

In another series of rats which were pithed and vagotomized, hypotension was induced by endotoxin (6 mg/kg i.v.). Animals were treated with saline or CV 3988 (10 mg/kg i.v.) 5 minutes prior to endotoxin administration. Below are listed representative MAP (mmHG) (mean ± SEM) (data were measured 30 minutes after endotoxin administration):

Saline (n = 7)	33 ± 5	
CV 3988 (n = 7)	51 ± 1 *	*$p \leq 0.05$

These data are consistent with the concept that CV 3988 seems to be a specific PAF antagonist. These data also demonstrate that it can attenuate endotoxin hypotension and is partially active peripherally. Its central acting effects remain to be examined. These data also suggest that PAF may participate in endotoxin hypotension. Further work is needed to explore these observations, to determine if CV 3988 antagonizes other mediators, and to better characterize its metabolic and hemodynamic effects in these and other shock models.

142

THE EFFECTS OF E.COLI ENDOTOXIN ON RAT SERUM ACE ACTIVITY IN VITRO AND IN VIVO. D.M. Deitz,* K.R. Swartz*, E. Murphy*, M. Wright*, R.S. Connell, M.W. Harrison. Oregon Health Sciences University, Portland, OR 97201.

Adult respiratory distress syndrome (ARDS) is the clinical expression of diffuse injury of both the pulmonary capillary endothelial cells and the alveolar epithelium. The etiology is multifactorial with sepsis being the most frequent precipitator. Previous investigation of the relationship between septicemia and the activity of angiotensin converting enzyme (ACE), an enzyme located in capillary endothelial cells including those in the lung, demonstrated reduced ACE activity in humans with septic ARDS. Using the synthetic ACE substrate BPAP and the assay described by Catravas and Gillis, we investigated the effect of E.coli endotoxin on serum ACE activity in the rat. In vitro, a statistically significant concentration dependent reduction in ACE activity was demonstrated ($p<.005$). In vivo an intravenous dose of endotoxin (20mg/kg) alone resulted in no significant change in serum ACE activity. However, the combination of intravenous endotoxin (20mg/kg) and mild hemorrhage (5-10% of blood volume) caused a statistically significant reduction in serum ACE activity by 15 minutes as compared to control rats with hemorrhage only (39% vs. 66%, $p<.005$). This reduction persisted at 30 and 60 minutes. However, by 180 minutes ACE activity was no longer statistically different from control values. We have demonstrated an acute reduction of serum ACE activity in the endotoxemic rat which appears to be dependent on the amount of circulating endotoxin and the presence of mild blood loss.

143

COMPARATIVE EFFECTS OF VARIOUS GAMMAGLOBULIN PREPARATIONS ON SURVIVAL IN A LETHAL MODEL OF ABDOMINAL SEPSIS. H. Shennib, R. Chiu, S. Karp*, G. Richards*, J. Prentis*, D. Mulder* The Montreal General Hospital & McGill University, Montreal, Que. H3G 1A4.

The purpose of this study was to examine if available gammaglobulin preparations can have a beneficial effect in the treatment of abdominal sepsis. A lethal rat model of abdominal sepsis was developed by ligating 5cm of proximal small bowel and an I.P. implantation of a gelatin capsule containing a mixture of sterile rat stools, live E.coli $6x10^5$, Ps. aeruginosa $9x10^4$, bact.fragilis $2x10^6$ and St. faecalis $3x10^6$. Four groups of Sprague Dawley rats (n=24 each) were used, each had either of the following injected I.V. at the time of induction of abdominal sepsis [Control normal saline (N); Gammaimmune, Cutter lab (G); Sandoglobulin, Sandoz (S);Atoxin, Univ. Natal R.S.A.(A)].

Two more groups (n=12 each) were given either Cefotaxime alone (C), or Cefatoxime and Atoxin (C+A). All bacteria used were found sensitive to Cefatoxime,

Times (hrs)	% Survival N	A	G	S	*P 0.01 compared to (N)(chi square) C	C+A
24	70.8	100*	75	91.7	100	100
36	54	73.9	50	70.8	83.3	100*
48	33.3	39.1	29.2	33.3	66.6	83.3*
60	29.1	26.1	20.8	20.8	50	58.3

In this inevitabely lethal model of abdominal sepsis, out of all the gammaglobulin preparations examined, Atoxin was found to provide protection in the early stages of sepsis. Atoxin when used in combination with selective antibiotic therapy allowed further prolongation of survival.

144

FACTORS INFLUENCING SURVIVAL IN SEPTIC RATS. G.T. ROBINSON, S. FORBES, T. HIRANO, T.S. VELKY, A.G. GREENBURG. San Diego VA Medical Center and Univ. of California San Diego, San Diego, CA 92161

Survival following a traumatic or septic insult is influenced by multiple factors. Alteration of host defenses by impaired reticuloendothelial system (RES) function increases mortality. The present study assesses the impact of perioperative insults on RES function and overall survival in an animal model of sepsis. Adult male Sprague-Dawley rats underwent exploratory laparotomy (EL), splenectomy (S), cecal ligation and perforation (CL), or cecal ligation, perforation and splenectomy (CLS). RES activity was evaluated by measuring fibronectin (FN) levels and by assessing response to intravenous gelatin-lecithin-triolein-glycerol (GLTG, 50 mg/Kg). The cause of death was evaluated at necropsy. Fibronectin levels significantly increased in all groups 24 hours postoperatively, with the highest levels in the S, CL, and CLS groups. Intravenous GLTG transiently depleted FN without increasing mortality 24 hours postoperatively, however, administration seven days postoperatively depressed FN levels in the groups S, CL, and CLS, with a 17-57%* mortality rate among previously septic animals. Sepsis alone results in excess mortality of 17-25%*, while mortality from blood drawing by cardiac puncture is 13%* among septic animals. Splenectomy is associated with an 8-9% excess mortality from sepsis and 3-25% excess mortality from cardiac tamponade. Overall mortality thus results from the combination of increased susceptibility to both sepsis and post-puncture cardiac tamponade. Prolonged alterations in RES function observed following sepsis predispose the animals to late complications. (*$p<.05$ by Students T-test or Fishers' Exact test.)

145

INCREASED SURVIVAL IN RATS PRE-TREATED WITH FIBRONECTIN DURING SALMONELLA TYPHIMURIUM BACTEREMIA. J.M. LURTON*, M.M. EVERETT*, T.E. EMERSON, JR., Physiology Research Department, Cutter Group of Miles Laboratories, Inc., Berkeley, CA 94710.

Preliminary data reported earlier by our group support the hypothesis that pre-treatment with fibronectin (FN) increases survival during Salmonella typhimurium (S. typ.) bacteremia. However, while the average values were numerically smaller in the FN vs saline groups (55% vs 71% mortality, respectively at days 9-16 of sepsis), the numbers of rats per group were too small to show statistical significance. In the present study, the numbers of animals per group were increased from 29 to 108 in the FN group and from 21 to 101 in the saline group. To achieve this, four separate sub-groups of FN and saline pre-treated rats were completed with the following numbers of rats in the FN and saline groups, respectively: a) 9 and 10; b) 20 and 11; c) 49 and 50; d) 30 and 30. Sub-groups "a" and "b" are from the earlier study. All rats were injected i.p. with purified human FN (50 mg/kg) or equivolume amounts of saline immediately prior to an i.p. injection of S. typ., 3 x 10^9 cells/rat, SL 1027. These groups were then combined and compared statistically by Chi Square analysis (*$p<0.05$; **$p<0.01$). Percent mortality against time for the combined groups is shown below.

Time(Days)	N	1	2	3	4	5	6	7	8	9	10	11	12	13	14
FN	108	31%	37%*	39%*	44%*	46%*	50%**	51%**	53%*	54%*	54%*	55%*	56%*	56%*	56%*
NaCl	101	44%	54%	54%	60%	64%	69%	70%	70%	71%	71%	71%	71%	71%	72%

This study demonstrates that FN pre-treatment increases survival time and permanent survival during S. typ. bacteremia in the rat.

146

EFFECTS OF TREATMENT WITH FIBRONECTIN (FN) AFTER ONE HOUR OF ENDOTOXIN SHOCK ON CARDIOVASCULAR, METABOLIC AND COAGULATION VARIABLES IN THE DOG. J.G. HAUPTMAN*, E.A. BEDNAR*, A.T. DAVIS*, T.E. EMERSON, JR., Michigan State University, East Lansing, MI 48824, Butterworth Hospital, Grand Rapids, MI 49503; and Cutter Group of Miles Laboratories, Inc., Berkeley, CA 94710.

Earlier work suggests that pre-treatment with (FN) is efficacious during endotoxin shock in the rat and dog. The present study was to determine the effects of treatment with FN 1 h after induction of endotoxin shock. Dogs were anesthetized with Nembutal and the following variables determined: 1) cardiac output, arterial pressure, total peripheral resistance, pulmonary artery and wedge pressures, and heart rate; 2) arterial BUN, glucose, lactate, albumin, globulin, ALT, alkaline phosphatase, cations, pH, HCO_3-, PCO_2, PO_2 and O_2 SAT; 3) platelets, fibrinogen, partial thromboplastin time, prothrombin time, thrombin time and FDP. Six dogs were injected i.v. with purified human FN (50 mg/kg) and 6 received equivolume i.v. injections of saline 1 h after i.v. injection of E. coli endotoxin (0.5 mg/kg). Determinations were made periodically for 6 h of shock. In another 5 dogs, plasma FN levels were followed for 5 h after i.v. E. coli endotoxin (0.5 mg/kg). There was no significant difference between the groups in any variable examined. In the other group, plasma FN levels fell by 54%, 56%, 51% and 54% at 1, 2, 3, 4 and 5 h of shock, respectively. Failure to demonstrate efficacy with FN post-shock treatment may be related to a number of factors, but the very rapid depletion of plasma FN within 1 h of shock suggest that the mechanism by which FN may act is already irreversibly damaged by the first h of endotoxin shock.

147

MORPHINE DEPRESSION OF MYOCARDIAL FUNCTION. T.R. RIGGS*, Y.YANO*, T. VARGISH. Dept. of Surgery, West Virginia University, Morgantown, WV 26506

A modified Langendorff rat heart preparation with an isolated working rat heart was used to establish the presence of opiate receptors in the myocardium and to demonstrate the effects of an opiate agonist on myocardial function in the absence of neuronal and humoral factors. Twenty-five Sprague-Dawley rats weighing 400-450 gm were anesthetized and had their hearts excised and attached to the Langendorff perfusion apparatus. After a 30 minute control period, the Krebs-Henseleit Buffer(KHB) perfusion solution was altered by the addition of 0.9%NaCl(S) or morphine sulfate(MS) which resulted in a final concentration of $2x10^{-4}$M or $3x10^{-4}$M MS in the KHB. Temperature, preload, and afterload were kept constant while heart rate(HR), cardiac output(CO), and aortic dP/dt max were measured. The data represents measurements taken at control and times(minutes) after the addition of S or MS to the KHB presented as means ± SEM:

		N	Control	t=15	t=30	t=45
H.R.	S	8	236±10	231±9	232±8	231±7
(beats/min)	$2x10^{-4}$M	9	265±8#	216±3●	213±4●#	216±7●
	$3x10^{-4}$M	8	236±8	193±6●#	189±5●#	185±6●#
C.O.	S	8	55±1	57±1	57±1	56±1
(ml/min)	$2x10^{-4}$M	9	57±2	55±2	54±2	54±2
	$3x10^{-4}$M	8	55±1	52±1●	51±1●#	50±1●#
dP/dt max	S	8	3159±101	2993±128	2931±122	2875±123
(mm Hg/sec)	$2x10^{-4}$M	9	3141±48	2972±63	2956±59	2917±54
	$3x10^{-4}$M	8	3152±73	2819±90●	2819±67●	2731±88●

●p<.05 when compared with control #p<.05 when compared with NS group at same time period.

The data suggests that opiate receptors are present in the myocardium and that morphine sulfate directly effects the myocardium causing a significant decrease in HR, CO, and aortic dP/dt max.

148

EFFECTS OF PRE-TREATMENT WITH IMMUNOGLOBULIN G ON HEPATIC CLEARANCE OF INDOCYANINE GREEN (ICG) DYE DURING SALMONELLA TYPHIMURIUM BACTEREMIA IN THE RAT. T.E. EMERSON, JR., J.M. LURTON*, M.S. COLLINS*. Physiology and Microbiology Research Departments, Cutter Group of Miles Laboratories, Inc., Berkeley, CA 94710.

We reported previously that a recently developed, native immunoglobulin G preparation for i.v. use (IGIV-pH 4.2; 500 mg/kg) is efficacious during severe Salmonella typhimurium (S. typ.) bacteremia; this product prevented hypotension and acidosis, ameliorated abnormalities of indices of organ damage, including elevations of alkaline phosphatase, SGOT, and GGTP, and dramatically decreased mortality. The present study compares hepatic clearance of ICG dye in non-infected Sprague-Dawley

rats (250-300 gms) with rats given i.p. injections of IGIV-pH 4.2 (500 mg/kg) or albumin (500 mg/kg) 2 h prior to i.p. infection with S. typ., SL 1027 at 3 x 10^9 cells/rat. At 15-19 h post-infection, rats were anesthetized with Nembutal. ICG (5 mg/kg) was injected i.v. and plasma clearance kinetics determined. ICG clearance is dependent upon hepatic blood flow as well as hepatocyte function at this dose but a dose which saturates the hepatocyte receptors was lethal in these sick rats. Mean $t_{\frac{1}{2}}$ values ± SEM for the non-infected (N=6), IGIV-pH 4.2 (N=11) and albumin (N=10) groups were 2.9 ± 0.09, 4.1 ± 0.24 and 5.4 ± 0.47 min, respectively. These values were all significantly different ($P<0.05$). Thus, pre-treatment with IGIV-pH 4.2 at a dose well tolerated clinically ameliorates the depressed hepatic clearance of ICG dye during S. typ. bacteremia in the rat. These data also support and extend earlier observations demonstrating that prophylaxis with IGIV-pH 4.2 is efficacious in impending bacteremic shock.

149

HEMODYNAMIC EFFECTS OF HYPERTONIC NaCl DURING SURGICAL TREATMENT OF AORTIC ANEURISMS J J C AULER JR* (Introduced by G C Kramer), M H C PEREIRA*, M ROCHA-E-SILVA*, A D J.TENE*, F PILEGGI*. Instituto do Coração, Hospital das Clínicas, Faculdade de Medicina, Universidade de São Paulo, 05403, São Paulo, Brasil.

Hypertonic NaCl (HS: 2400mOsm/l; 4ml/kg) effectively resuscitates dogs from severe blood loss. During thoracoabdominal aortic surgery, major volemic shifts follow aortic unclamping. This study compares hemodynamic effects of HS and isotonic NaCl (IS) on the severe hypotension which follows aortic unclamping. 10 consecutive patients with chronic aortic aneurisms were operated under the same anesthetic and technical conditions. 5 received HS, 5 IS, 4 ml/kg, 10 min after aortic unclamping. Cardiac index (CI - $l.min^{-1}.sq\ m^{-1}$), mean systemic (MAP) and pulmonary (PAP) arterial pressure (mmHg), pulmonary capillary pressure (PCP - mmHg), systemic (SVR) & pulmonary (PVR) vascular resistance ($dynes.sec.cm^{-5}$) were measured after anesthesia (A) aortic clamping (B), unclamping (C), after HS/IS (D), & at the end of the operation (E). Duration of clamping was similar for both groups (64±36min). The HSxIS "t" test showed no differences at A B C E. At D: CI: 4.9±0.5(HS)x2.5±1.0(IS). MAP: 65±6(HS)x 73±7(IS). PAP: 28±3(HS)x15±1(IS). PCP: 22±3(HS)x9±1(IS). SVR: 626±79(HS)x1418±212 (IS). PVR: 46±11(HS)x141±32(IS). Only MAP showed no difference HSxIS. Total crystal loid infused was 575±48(HS)x768±81(IS) ml/h; total blood was 2040±180(HS)x3880±270 (IS) ml. Thus HS effectively restores normal circulatory function after aortic clamping, with higher CI, lower SVR/PVR, in spite of lower requirement of crystalloid and blood transfusion. It is suggested that the known vascular effects of HS are mainly responsible for the observed differences between HS and IS treated patients.

150

HYPERTONIC RESUSCITATION FROM SEVERE BLOOD LOSS: ONLY NaCl INDUCES REFLEX FEMORAL VASOCONSTRICTION. M ROCHA-E-SILVA* (Introduced by: G.C. Kramer), G.A. NEGRAES*, L. LOPPNOW*, V PONTIERI*. Instituto do Coração, Hospital das Clínicas, Faculdade de Medicina, Universidade de São Paulo, 05403, São Paulo, Brasil.

Hypertonic NaCl (2400 mOsm/l) effectively resuscitates dogs from severe blood loss, but only with intact cervical vagi. Other equally hypertonic solutes are ineffective. This study examines the effects of 2400 mOsm/l NaCl or glucose (4 ml/kg) on femoral, renal and mesenteric flows of 45 anesthetized dogs bled to, and held at 40 mm Hg mean arterial pressure (MAP) over 45 min. Hypertonic NaCl was given to dogs with intact or locally anesthetized cervical vagi. Femoral flow was measured in intact and denervated hindlimbs. Removed blood (46±3 ml/kg) was discarded. Results as follows: A: Hemorrhage induced a 4.1-fold renal vasoconstriction, 6.2-fold mesenteric constriction, 5.8-fold intact hindlimb constriction, 7.2-fold denervated hindlimb constriction. B: Hypertonic NaCl (intact vagi) restored MAP to a stable 93±5 mm Hg, redilated renal, mesenteric and denervated hindlimb vessels to pre-hemorrhage levels; innervated hindlimbs were transiently dilated, but reverted to severe & sustained constriction. C: Hypertonic NaCl (blocked vagi) induced transient recovery of MAP and restored all measured flows to pre-hemorrhage levels. D: Hypertonic glucose was similar to "C" but MAP fell faster after initial recovery. Thus hypertonic NaCl, but not glucose, induces reflex femoral vasoconstriction, while NaCl and glucose pro

duce renal and mesenteric dilation. This reflexly controlled, selective femoral vaso constriction may be a critical aspect of the survival response to hypertonic NaCl.
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151

BENEFICIAL EFFECTS OF GLUCAGON IN ENDOTOXIC SHOCK IN RATS. S. TEICH,* D. MALCOLM,* G. ZALOGA,* J. HOLADAY & B. CHERNOW. Naval Medical Research Insititue, Bethesda MD 20814 & Walter Reed Army Institute of Research, Washington, D.C. 20307.

Glucagon has effects which are potentially beneficial in shock including positive cardiac inotropic and chronotropic actions. We sought to determine if glucagon treatment would alter cardiovascular parameters and survival in endotoxic shock. Arterial catheters were connected to a physiograph for continuous monitoring of mean arterial pressure (MAP) and heart rate (HR). Drugs were administered through a venous catheter. Venous blood was withdrawn (volume restored) for CBC, lactate and glucose determinations prior to and two hrs following endotoxin injection. Endotoxin (E. coli LPS; 30 mg/kg, iv; LD80) injection caused an acute hypotension (>20 mm Hg drop), followed by a partial recovery and a secondary fall in MAP 45 min post-injection. Glucagon or saline were administered when the criteria for acute hypotension were met. Glucagon at 2 but not 1 mg/kg elevated MAP at 45 and 60 minutes post injection (mean of 12 & 10 mm Hg, respectively) compared to saline controls. Glucagon caused a dose related increase in HR throughout the 105 minutes observed. At 2 hr post-endotoxin, there were no difference in white cells or lactate levels between saline and glucagon-treated animals. Glucose levels in control animals decreased (-25±17%) but were elevated in glucagon-treated (2 mg/kg) animals (42±14%). Glucagon (1 & 2 mg/kg) significantly increased 6 hour survival (50% controls vs. 88% glucagon; $p<.04$) but not 24 hr survival (2 of 10 controls vs. 5 of 17 glucagon-treated rats survived). Results indicate that glucagon improves hemodynamic and metabolic variables that may contribute to enhanced survival. From these findings, we suggest that glucagon may be beneficial in the treatment of endotoxic shock.

Author-Abstract Index

Abel, F.L., 128
Adachi, K, 80
Akwa, A.L., 56
Albes, J.M., 35
Alteveer, J., 14
Amaral, J.F., 110
Andreasson S., 30
Aoyama, H., 18, 22, 78, 109
Archer, L.T., 129, 131
Ashton, S.H., 91, 96
Auclair, M.C., 126

Badsha, N., 48
Bagby, G.J., 21, 106
Barr, R.G., 49
Barrett, J., 45, 88
Barrillo, D.J., 53
Bartos, D., 16
Bartos, F., 16
Bass, B.L., 70
Baue, A.E., 52, 54, 55
Beamer, K.C., 65, 68, 101
Beck, R.R., 128
Bednar, E.A., 146
Bellamy, R.F., 132
Beller, B., 131
Berezesky, I.K., 18, 78
Bergqvist, D., 138
Bhatia, J., 100
Black, L., 61
Black, P. McL., 63
Blümel, G., 42
Blum, P.S., 1
Bond, R.F., 2
Borgia, J., 26
Bottoms, G.D., 90, 93
Brackett, D.J., 71, 72, 104
Breslow, M.J., 12, 113
Briggs, R., 51
Briglia, F.A., 116
Brock-Utne, J.G., 41
Brown, G.E., 73
Brown, D.F., 9
Brown, M., 25, 31
Bryant, R.E., 16
Buchweitz, E., 135
Buday, A.C., 21
Buja, L.M., 6
Burchard, K.W., 10, 43, 82
Burr, R.E., 9

Caffrey, J.L., 97
Caldwell, M.D., 110
Carli, A., 126
Carr, D.B., 63
Causey, A.L., 47
Cerchiari, E.L., 11
Cerra, F.B., 37
Chang, A.C.K., 127, 131
Chaudry, I.H., 52, 54, 55
Chepenik, K.P., 95
Chernow, B., 61, 116, 119, 123, 151
Chiu, R.C.-J., 7, 139, 143
Chuang, G.J.-H., 139
Clemens, M.G., 52, 54, 55
Cline, C.W., 27
Cochrane, C.G., 40
Coffee, K., 99
Collins, M.S., 148
Connell, R.S., 16, 142
Connett, R.J., 8
Cook, J.A., 44, 89, 96, 99
Corll, C.B., 106
Cox, E., 137

Curtis, B.B., 6
Czaja, J., 140

Daly, T., 65
D'Arezzo, A., 82
Darius H., 85
Das Gupta, T.K., 38
Davis, A.T., 146
Dawidson, I., 130
Deahia, H.V., 68
Deguzman, L.R., 132
Dehring, D.J., 33
Deitz, D.M., 142
Demling, R.H., 29
Dennis, R.C., 121
Denno, R., 20
Denzlinger, C., 86
Didkan, G.S., 53
Didlake, R., 47
Dixit, P., 57
Dobkin, E.D., 100
Donohoe, M.J., 53
Doran, J.E., 48
Dorroh, L., 47
Downing, J.W., 41
Dronen, S.C., 66
Drucker, W.R., 8
Dunn, R., 45, 88
Dwenger, A., 76

Egami, K., 80
Elam, R., 67
Emerson, T.E., Jr., 145, 146, 148
Erecinska, M., 19
Erwin, L., 16
Everett, M.M., 145
Ezra, D., 140

Fabian, T.C., 57
Fader, R., 33
Fagraeus, L., 71, 72, 104
Falk, J.L., 125
Fehr, D., 51
Ferguson, J.L., 62
Fessler, J.F., 90, 93
Feuerstein, G., 67, 103, 140
Filkins, J.P., 13, 107
Fink, M.P., 58

Fish, R.E., 124
Fletcher, J.A., 47
Fluornoy, D., 131
Flynn, J.T., 95
Forbes, S., 144
Foutch, R., 66
Franco, C.D., 136
Francois-Dainville, E., 135

Gaffin, S.L., 41
Ganes, E., 64
Gann, D.S., 10, 110
Gao, C.X., 139
Gardiner, W.M., 58
Garrity, F.L., 83
Gartner, S.L., 134
Gasiford, J.C., 27
Gassim, C., 56
Gibbons, D.A., 122
Girotti, M.J., 36
Goldfarb, I.W., 27
Goldstein, R., 140
Goodenough, R.D., 17
Goris, R.J.A., 75
Gorman, R.R., 91
Goss, J.R., 16
Goto, M., 94
Grasberger, R.C., 121
Greenburg, A.G., 144
Greene, P.S., 52
Griffin, A.J., 94
Grogan, J.B., 56
Grover, G.J., 135
Gurll, N.J., 64

Haglund, U., 138
Hagmann, W., 86
Halushka, P.V., 44, 89, 96, 99, 133
Hamilton, A.J., 63
Hanrahan, J.B., 27
Harlan, J., 32
Harmon, J.W., 70
Harris, J., 51
Harrison, M.W., 16, 142
Hatherill, J.R., 79
Hathorn, L.F., 97
Hauptman, J.G., 146
Hayashi, H., 55

Hechtman, H.B., 13, 46
Herndon, D.N., 17, 25, 28, 31, 105
Hill, M.R., 112
Hinshaw, D.B., 40
Hinshaw, L., 127, 129, 131
Hirai, F., 18, 78
Hirano, T., 144
Hirasawa, H., 50, 59, 81
Hock, C.E., 87
Holaday, J., 61, 70, 71, 72, 104, 151
Holcroft, J.W., 111
Hollenbach, S.J., 132
Hong, K., 36
Hooper, J., 25
Horsborough, R., 108
Horton, J.W., 3, 6, 98
Houvenaghel, A., 137
Hull, M.J., 52, 55
Hyland, B.J., 37
Hyslop, P.A., 40

Ishida, K., 129, 131
Ito, Y., 59
Ives, N., 119

Jacob, H.E., 27
Jahoor, F., 105
Januszkiewicz, J., 108
Jatene, A.D., 149
Jentoft, J.E., 49
Jesmok, G.J., 26
Johnson, G., III, 2
Johnson, M., 90
Jones, S.B., 115
Jungberg, J.L., 138

Karlstad, M., 15, 39
Karp, S., 143
Katz, A., 29
Kawanami, O., 80
Keller, G.A., 37
Keppler, D., 86
Kilpatrick-Smith, L., 19
Klein, D.M., 122
Kobayashi, H., 59
Kobayashi, S., 59, 81
Kober, P.M., 122
Koehler, R.C., 12
Kojima, N., 80
Koronado, E., 34
Kovarik, F., 115
Kramer, G.C., 28, 111
Krausz, M.M., 34
Kutsky, P., 97, 117
Kuttner, R.E., 20, 24

Lalonde, C., 29
Lamar, C., 90
Lang, C.H., 21, 120, 124
Lanier, A., 11
Lanser, M.E., 73, 74
Laurindo, F., 140
Law, W.R., 62
Lazaro, E.J., 136
Leavy, J.A., 125
Ledgerwood, A.M., 4
Lefer, A.M., 85, 87
Lefer, D.J., 85
Lerner, M.R., 71, 72, 104
Leventhal, H., 77
Levine, E., 95
Linares, H.A., 28, 100
Lincoln, K.L., 91
Lobe, T.E., 100
Loppnow, L., 150
Lucas, C.E., 4
Lundin, S., 114
Lundsgaard-Hansen, P., 48
Lurton, J.M., 145, 148

Malcolm, D., 61, 151
Maningas, P.A., 66
Marawi, I., 56
Margolis, J.H., 93
Markov, A.K., 47
Martin, L., 51
Martinez, R.R., 106
Marzella, L., 77
Matsuda, T., 80
Maunder, R., 32
May, M., 84
McCallum, R.E., 112
McCuskey, R.S., 60
McDonough, K.H., 118, 120
McGee, M., 51
McGlew, M.J., 5

McIntosh, T.K., 121
McKenna, T.M., 116
McMillan, M.R., 123
McNamara, J.J., 84
Mela-Riker, L., 16
Mendez, C., 110
Mikulaschek, A.W., 141
Miller, C., 113
Miller, H.I., 118
Miyashita, M., 80
Moon, B., 36
Mora, R., 74
Morgenthaler, J.J., 48
Moriel, E., 34
Moss, G.S., 38
Mulder, D.S., 139, 143
Munster, P.J.J.v., 75
Murotani, N., 59
Murphy, L., 142
Murray, C.E., 131
Myashita, M., 80

Nakatani, T., 22, 109
Negraes, G.A., 150
Nerlich, M.L., 35
Nguyen, D., 43
Nichols, B.G., 111
Nolan, P., 45, 88
Nuytinck, J.K.S., 75

Ochoa, R., 91
Oda, S., 81
Odaka, M., 50, 59, 81
Oestern, H.J., 35, 76
Ohkawa, M., 50, 81
Ohtake, Y., 50
Olanoff, L.S., 133
Oldham, K.T., 25, 100
Olson, L.M., 38
O'Malley, V., 131
Onda, M., 80
Ottoson, J., 130

Palsson, J., 114
Palter, M.D., 121
Papp, E., 48
Parker, J.L., 97, 117
Passey, R., 131
Patscheke, H., 92
Patterson, C.R., 57
Paul, E., 75
Pearce, F.J., 8
Pelsson, J., 114
Pereira, M.H.C., 149
Perel, A., 34
Perker, M., 42
Peters, A., 51
Peters, E.J., 17
Pfieffer, U., 42
Phuangsab, A., 45, 88
Pileggi, F., 149
Pinosky, M., 96
Placko, B., 22
Pohlson, E.C., 84
Polin, R., 19
Pontieri, V., 150
Powell, 103
Prentis, J., 143

Quinn, J.V., 43

Rackow, E.C., 125
Ramos, I., 102
Rapp, S., 86
Raymond, R.M., 122
Redl, H., 75
Regal, G., 76
Reichle, H., 42
Reines, H.D., 133
Reynolds, D.G., 64
Rhodes, R.S., 49
Richards, G., 143
Ricksten, S.-E., 114
Riggs, T.R., 147
Risberg, B., 30
Robinson, G.T., 144
Rocha-E-Silva, M., 149, 150
Rogers, F., 45, 88
Rogers, T.S., 44
Romano, F.D., 115
Roth, B.L., 116, 123
Rubli, E., 48
Rush, B.F., Jr., 53
Ryan, P.V., 29
Ryerson, S., 101

Saad, A., 56
Sacco, N.A., 60
Safar, P., 5, 11
Sampson, J.A., 70
Sato, H., 50, 59, 81
Sato, T., 18, 22, 78, 109
Sayeed, M., 15, 39
Schaefer, C.F., 71, 72, 104
Schapiro, R., 113
Schiebel, F., 102
Schlag, G., 75
Schnarrs, R., 27
Schraifstatter, I.U., 40
Schrauwen, E., 137
Schumer, W., 20, 24
Schweitzer, F., 76
Sclabassi, R., 11
Shail, E., 56
Sharp, S., 119
Shatney, C.H., 69
Shaw, J.H.F., 108
Shennib, H., 143
Shepherd, R.E., 120
Shepro, D., 46
Shigematsu, H., 69
Siegel, J.H., 22, 109
Silverman, D., 77
Simmons, R.L., 37
Simonsen, R., 130
Siren, A.-L., 103
Siri, F., 84
Sklar, L.A., 40
Slater, H., 27
Slotman, G.J., 10, 43, 82, 83
Smith, J.B., 85
Smith, L., 30, 118
Smith, L.W., 118
Smith, R.A., 69
Smith, R.W., 4
Soeda, K., 50
Spath, J.A., Jr., 1
Spillert, C.R., 136
Spillert, K.R., 136
Spitzer, J.A., 124
Spitzer, J.J., 21
Spragg, R.G., 40
Stegmeier, K., 92
Stith, R.D., 112
Stremple, P., 5
Sturn, J.A., 35, 76
Sugai, T., 20, 24
Svartholm, E., 138
Swartz, K.R., 142

Tankersley, S., 133
Taylor, F.B., 127
Taylor, S., 3
Teba, L., 68, 101, 102
Teich, S., 61, 151
Till, G.O., 79
Todd, T.R., 36
Tompkins, P., 71, 72, 104
Toth, P.D., 141
Traber, D.L., 25, 28, 31, 33, 100
Traber, J., 33
Traber, L.D., 25, 31, 33
Traystman, R.J., 12, 113
Trump, B.F., 18, 78
Tuggle, D.W., 98
Turek, J., 90
Tuwaijri, A.L., 56

Unbehagen, J., 33
Urbaschek, B., 92
Urbaschek, R., 92

Valas, E.J., 14
Valeri, C.R., 121
Vargish, T., 65, 147
Vary, T.C., 22, 109
Velez, S.R., 9
Velky, T.S., 144
Vernimmen, C., 126

Ward, D., 90
Ward, P.A., 79
Weil, M.H., 125
Weiss, H.R., 135
Welles, S.L., 46
Wellhöfer, H., 42
Wells, M., 41
West, K., 29
West, M.A., 37
Westfall, M., 15
Widener, L., 16
Williams, G., 129