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TITLE: Targeted Nutritional Approach to Improve Muscle Function and Physical Activity by Restoring Metabolic Deregulations During Recovery from Sepsis

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n/a

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n/a

7. PERFORMING ORGANIZATION NAME(S) AND ADDRESS(ES)

AND ADDRESS(ES)

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13. SUPPLEMENTARY NOTES

14. ABSTRACT

Purpose: The major goal to the project is to test the hypothesis that a unique formulation based on EAA will improve physical activity, and involuntary isometric skeletal muscle strength faster during the recovery of a sepsis event.
Scope: The proposed study in a catheterized pig model recovering from sepsis is innovative because, a) the targeted nutritional supplementation is a novel approach to attenuate tissue breakdown in sepsis and improve functional outcome and restore muscle mass, b) it provides insights into sepsis-induced severe tissue breakdown and physical outcome. The use of innovative, stable tracer methodology to measure metabolic fluxes within and across muscle, enables quantification of all metabolic endpoints. The results of the proposed study will have a positive impact by providing the basis to develop novel cost-effective nutritional approaches for patients recovering from sepsis to improve recovery and rehabilitation. It has a strong justification because of its rapid translation into clinical application.
Achievement of year 1:
Concerning the pig studies we obtained IACUC approval. We implemented logistics for monitoring physical activity and muscle strength measurement (buying testing equipment) and other surgery/animal care/pharmaceutical compounding/ analytical related preparations. Started with the animal studies and validated the approach and trained new personnel. No changes needed to be made concerning the approach.

15. SUBJECT TERMS

Sepsis, nutritional support, essential amino acids, muscle strength, pig, protein synthesis, protein breakdown, muscle breakdown.

16. SECURITY CLASSIFICATION OF:			17. LIMITATION OF ABSTRACT	18. NUMBER OF PAGES	19a. NAME OF RESPONSIBLE PERSON
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Standard Form 298 (Rev. 8-98)
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TABLE OF CONTENTS

INTRODUCTION

As the endotoxin-induced sepsis-like state is not comparable to real life human sepsis, we have developed a translational model in which sepsis is induced by continuous intravenous infusion with live bacteria (Pseudomonas aeruginosa) using a pig model. In this sepsis model, we observed complex multiorgan protein metabolic disturbances including muscle protein catabolism{Ten Have, 2015, S33}{Ten Have, 2017, 1-301}{Ten Have, 2019, G755-G762}, ultimately leading to bacterial translocation, increased cytokine release, and stimulated muscle breakdown. Our approach is to study the effects of targeted dietary EAA enriched nutritional supplementation versus a sham-control on muscle function and physical outcome in the recovery phase from sepsis (primary endpoint). Muscle function and physical outcome will be related to net protein synthesis on whole body and muscle level (secondary endpoint). In a randomized, sham controlled, blind (for nutritional intervention) 2 group design, we will use involuntary isometric skeletal muscle strength and physical activity measurements for functional outcome. Innovative, stable tracer technologies will be used to quantify metabolic endpoints like muscle net protein synthesis and muscle mass.

KEYWORDS

Sepsis, nutritional support, essential amino acids, muscle strength, pig, protein synthesis, protein breakdown, muscle breakdown.

ACCOMPLISHMENTS

What were the major goals of the project?

The major goal to the project is to test the hypothesis that a unique formulation based on EAA will improve physical activity, and involuntary isometric skeletal muscle strength faster during the recovery of a sepsis event.

What was accomplished under these goals?

Major Task 1: Pig studies:

We achieved “Subtask 1: Submit documents for AICUC approval” and obtain AICUC approval.
We achieved “Subtask 2: Implementation of logistics for monitoring physical activity and muscle strength measurement (buying testing equipment) and other surgery/animal care/pharmaceutical compounding/ analytical related preparations.”. Logistics are in place.
We started “Subtask 3: Performing animal studies. 62 animals, 4 animals/6 weeks”. Due to validation/personal training purposes and university COVID-19 logistic restrictions, we have done 2 animals/6 week. In total 8 animals.

Major Task 2: Analysing specimen - Activity (Video) monitoring

Nothing to report. Starts in year 2

Major Task 3: Data modeling - statistical data analysis

“Subtask 1: Data modeling - statistical data analysis”: nothing to report. Will start in the third year.
“Subtask 2: Writing scientific manuscripts”: manuscript preparations ongoing.

What opportunities for training and professional development has the project provided?

We (Deutz, ten Have) have setup the logistics of the procurement, housing, and care for the animals. Once the process was established, additional staff (Rice, Morse) were trained in the practical care and documentation needed to house the animals and ensure their well-being throughout the course of the study. We (Deutz, ten Have, Simbo) have also on-boarded and trained new staff (Rice, Morse) in the preparation, implementation, and analysis of samples/data generated during the metabolic study days. Rice and Morse have also developed professional skills from formal surgical prep, maintenance of chronic catheters, and animal care during critical illness and recovery. Rice and Morse have developed technical skills in experimental design, behavior analysis, continuous glucose monitoring, metabolic tracking, use of isotopes in experimental design and data analysis.

Assessing muscle function is one of the primary end points in this study. Dr. Peter Nghiem and his team (Alexis Rutledge) are considered one of the leading authorities on muscle testing in canine models for DMD and have published extensively in this field. Therefore, scientific knowledge and technique was extrapolated from their canine DMD studies to our current porcine project. In collaboration with the CTRAL team, Dr. Nghiem's team purchased and set up the muscle testing equipment from Aurora Scientific. Significant training and troubleshooting were completed to establish the muscle testing in pigs. Moreover, significant time and effort were given to assess anatomical difference in porcine cadavers to identify pelvic limb nerves and determine anatomical differences between canine and porcine. Finally, due to differences in muscle strength, a fatigue protocol (instead of an eccentric contraction decrement protocol, as performed in Dr. Nghiem's canine studies) was researched, instituted, and established for this current study.

The entire team meets weekly to assess progress and discuss the logistics of the implementation of the protocol and address any issues that may arise during the test days.

How were the results disseminated to communities of interest? Nothing to report.

What do you plan to do during the next reporting period to accomplish the goals? Nothing to report.

IMPACT

What was the impact on the development of the principal discipline(s) of the project? Nothing to report.

What was the impact on other disciplines? Nothing to report.

What was the impact on technology transfer? Nothing to report.

What was the impact on society beyond science and technology? Nothing to report.

CHANGES/PROBLEMS

Changes in approach and reasons for change. Nothing to report.

Actual or anticipated problems or delays and actions or plans to resolve them.

Due to validation/personal training purposes and university COVID-19 logistic restrictions, we have done less animals as indicated in the SOW. We expect to see improvement and some logistic adjustments in year 2 of this project to ensure the goals can be achieved.

Changes that had a significant impact on expenditures.

Due to validation/personal training purposes and university COVID-19 logistic restrictions, we have done less animals as indicated in the SOW.

Significant changes in use or care of human subjects. Not applicable.

Significant changes in use or care of vertebrate animals. Nothing to report.

Significant changes in use of biohazards, and/or select agents. Nothing to report.

PRODUCTS

Publications, conference papers, and presentations.

Journal publications. Nothing to report.

Books or other non-periodical, one-time publications. Nothing to report.

Other publications, conference papers, and presentations. Nothing to report.

Website(s) or other Internet site(s). Nothing to report.

Technologies or techniques.
Standardized and internationally recognized methods in rare disease, muscle function, and metabolism are used.

Inventions, patent applications, and/or licenses. Nothing to report.

Other Products. Nothing to report.

PARTICIPANTS & OTHER COLLABORATING ORGANIZATIONS

What individuals have worked on the project?

Name:	Nicolaas Deutz
Project Role:	PI
Research Identifier (e.g., ORCID ID):	0000-0001-5845-6447
Nearest person month worked:	1.23
Contribution to Project:	Responsible for all aspects of this study, including financial management, experimental design, stable tracer related methodological aspects.
Funding Support: Complete only if the funding support is provided from other than this award	NIH - 1R01HL132887-01A1; European Society for Clinical Nutrition and Metabolism, Internal

Name:	Gabriella Ten Have
Project Role:	Co-I
Research Identifier (e.g., ORCID ID):	0000-0003-2617-1193
Nearest person month worked:	9.73
Contribution to Project:	Responsible for design, implementing, coordinating and performing the animal experiments
Funding Support: Complete only if the funding support is provided from other than this award	NIH - R56HL141744, 5R01DK120296-02; Abbot Nutrition; American Society for Parenteral and Enteral Nutrition, European Society for Clinical Nutrition and Metabolism, Internal

Name:	Marielle Engelen
Project Role:	Co-I
Research Identifier (e.g., ORCID ID):	0000-0001-9884-2553
Nearest person month worked:	0.17

Contribution to Project:	Responsible for coordination of overall aim of the pig study activities, mentoring research staff
Funding Support: Complete only if the funding support is provided from other than this award	NIH - R56HL141744, 5R01AG064010-02; European Society for Clinical Nutrition and Metabolism, Internal

Name:	Peter Nghiem
Project Role:	Senior Personnel
Research Identifier (e.g., ORCID ID):	0000-0002-8796-8123
Nearest person month worked:	1.00
Contribution to Project:	Responsible for all measurements related to muscle function, endurance and strength.
Funding Support: Complete only if the funding support is provided from other than this award	NIH 5RO1NS094705-05, RO1EB028533, 5RO1AR049722-10; Edgewise Therapeutics; Internal

Name:	John Thaden
Project Role:	Senior Personnel
Research Identifier (e.g., ORCID ID):	0000-0003-3381-2198
Nearest person month worked:	3.26
Contribution to Project:	Responsible for supervising and performing laboratory preparations and GC-MS/MS and LC-MS/MS analysis
Funding Support: Complete only if the funding support is provided from other than this award	NIH - R56HL141744; Internal

Name:	Ryan Morse
Project Role:	Research Assistant
Research Identifier (e.g., ORCID ID):	n/a
Nearest person month worked:	10.94
Contribution to Project:	Responsible for day-to-day coordination and performing animal experiments and laboratory analysis
Funding Support: Complete only if the funding support is provided from other than this award	Internal

Name:	Sarah Rice

Project Role:	Post-doc Research Associate
Research Identifier (e.g., ORCID ID):	0000-0002-5481-4558
Nearest person month worked:	9.66
Contribution to Project:	Responsible for day-to-day coordination and performing animal experiments and laboratory analysis
Funding Support: Complete only if the funding support is provided from other than this award	European Society for Clinical Nutrition and Metabolism; Internal

Name:	Sunday Simbo
Project Role:	Research Associate
Research Identifier (e.g., ORCID ID):	n/a
Nearest person month worked:	2.67
Contribution to Project:	As an experienced researcher in the group, assisted wiht onboardin and others for day-to-day coordination and performing animal experiments and laboratory analysis
Funding Support: Complete only if the funding support is provided from other than this award	NIH - R56HL141744, 5R01DK120296-02; Internal

Name:	Alexis Rutledge
Project Role:	Research Staff
Research Identifier (e.g., ORCID ID):	n/a
Nearest person month worked:	3.40
Contribution to Project:	Assistant to Co-I, Dr. Nghiem, for all measurements related to muscle function, endurance, and strength
Funding Support: Complete only if the funding support is provided from other than this award	NIH 5RO1NS094705-05, RO1EB028533; Internal

Has there been a change in the active other support of the PD/PI(s) or senior/key personnel since the last reporting period?
Nothing to Report.

What other organizations were involved as partners?
Nothing to report.

SPECIAL REPORTING REQUIREMENTS

Quad Chart. Not applicable.