Using the gut microbiome to predict changes in muscle mass and function: Preliminary findings from a diet and exercise intervention study

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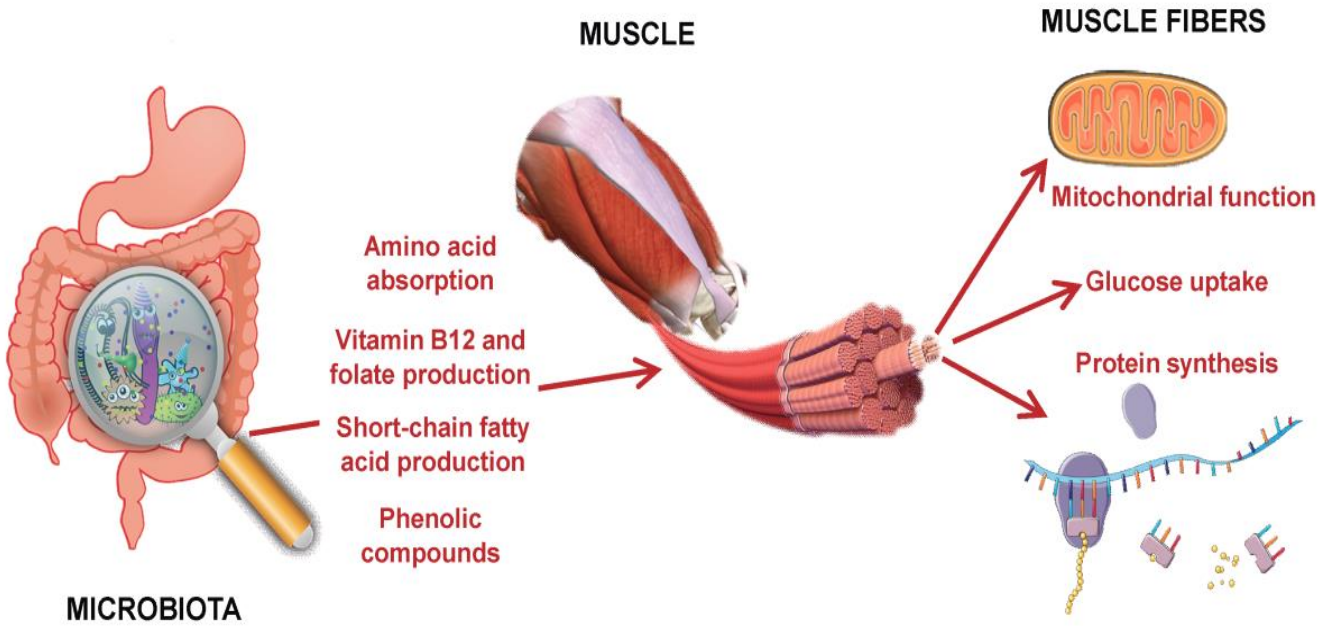


Ticinesi A, et al., Gut Microbiota, Muscle Mass and Function in Aging: A Focus on Physical Frailty and Sarcopenia. Nutrients. 2019 Jul 17;11(7).

Introduction

- The human gastrointestinal (GI) tract contains an abundant and diverse microbial community that gathers more than 100 trillion microorganisms.

- The collection of bacteria, archaea and eukarya colonising the GI tract is termed the ‘gut microbiota’ and forms an intricate and mutually beneficial relationship with host.
  - Contains over 100 times the amount of genomic content (microbiome) as the human genome.

- Taxonomically, bacteria are classified according to phyla, classes, orders, families, genera, and species.
  - Only a few phyla are represented, accounting for more than 160 species.
  - Two phyla Firmicutes and Bacteroidetes representing 90% of gut microbiota.

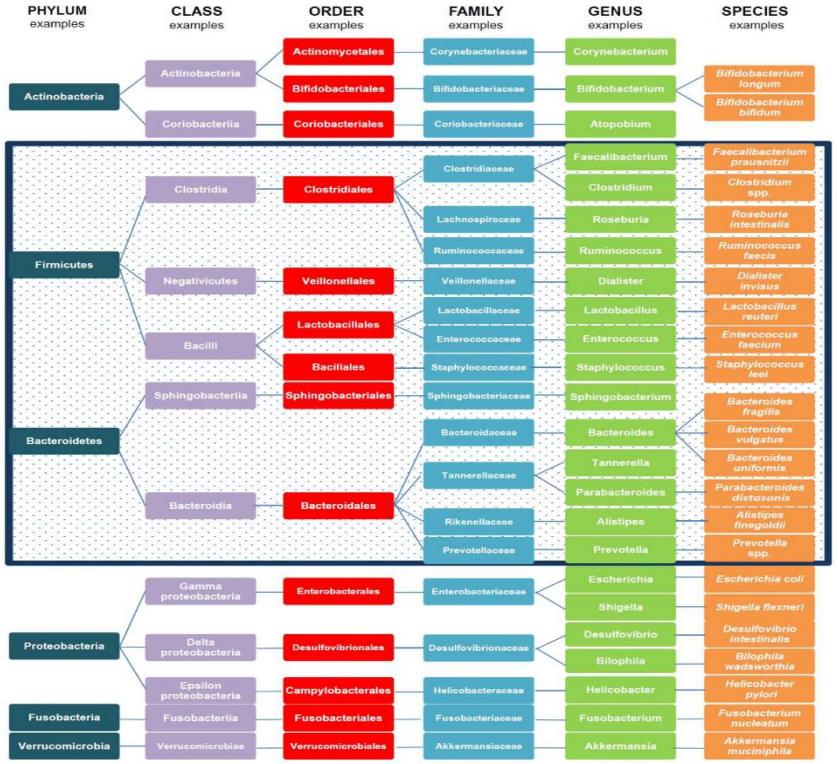


Figure 1. Examples of taxonomic gut microbiota composition. In the box are cited examples of bacteria belonging to Phyla Firmicutes and Bacteroidetes, representing 90% of gut microbiota.

Rinninella, E., Raoul, P., Cintoni, M., Franceschi, F., Miggiano, G., Gasbarrini, A., & Mele, M. C. (2019). What is the Healthy Gut Microbiota Composition? A Changing Ecosystem across Age, Environment, Diet, and Diseases. Microorganisms, 7(1), 14. doi:10.3390/microorganisms7010014

Introduction

- The microbiome (and microbiota) has received increasing attention over the last 15 years.

- Recent investigations have moved beyond classical infectious diseases and focused on its role in nutrient extraction, synthesis, absorption, immune function and metabolism.

- The "gut-brain axis" has also recently received a lot of attention
  - The gut not only receives regulatory signals from the central nervous system (CNS), but it is also able to send signals to the brain, and the brain receives them.
    - Recent evidence suggests that certain bacteria taxa are strongly related with cognitive and behavioural functions.

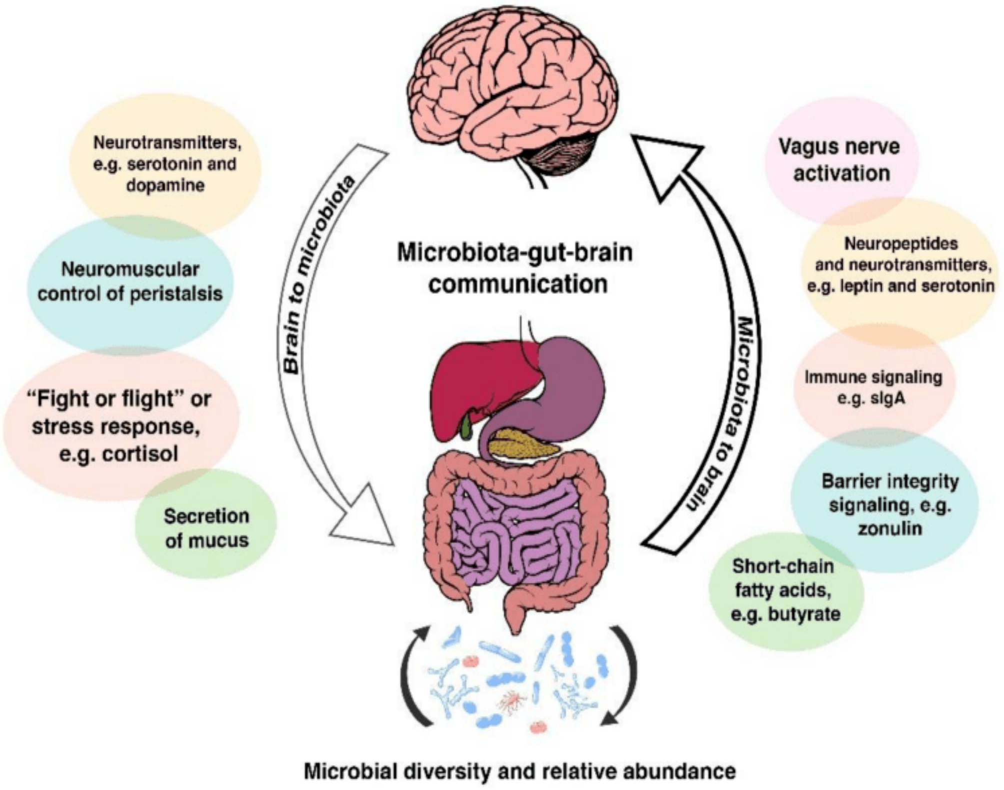


Figure 3. Main function of bacteria in the gut. Laukens D, et al., Heterogeneity of the gut microbiome in mice: guidelines for optimizing experimental design. FEMS Microbiol Rev. 2016 Jan;40(1):117-32.

Figure 2. Microbiota-gut-brain interaction. Taken from: https://www.scientistlive.com/the-microbiota-gut-brain-axis-from-...-biomarkers-for-clinical-treatment/

Introduction

- Growing recognition regarding the role of diet on modulating the composition and metabolic activity of the human gut microbiota, which in turn influences host health.
- Very few studies have examined how exercise alone impacts the gut microbiota.
- While changes in microbiota composition and functionality have been reported, further research in humans is needed.
  - Moreover, whether a potential **"gut-muscle axis"** reported in animal models plays a role is unknown.

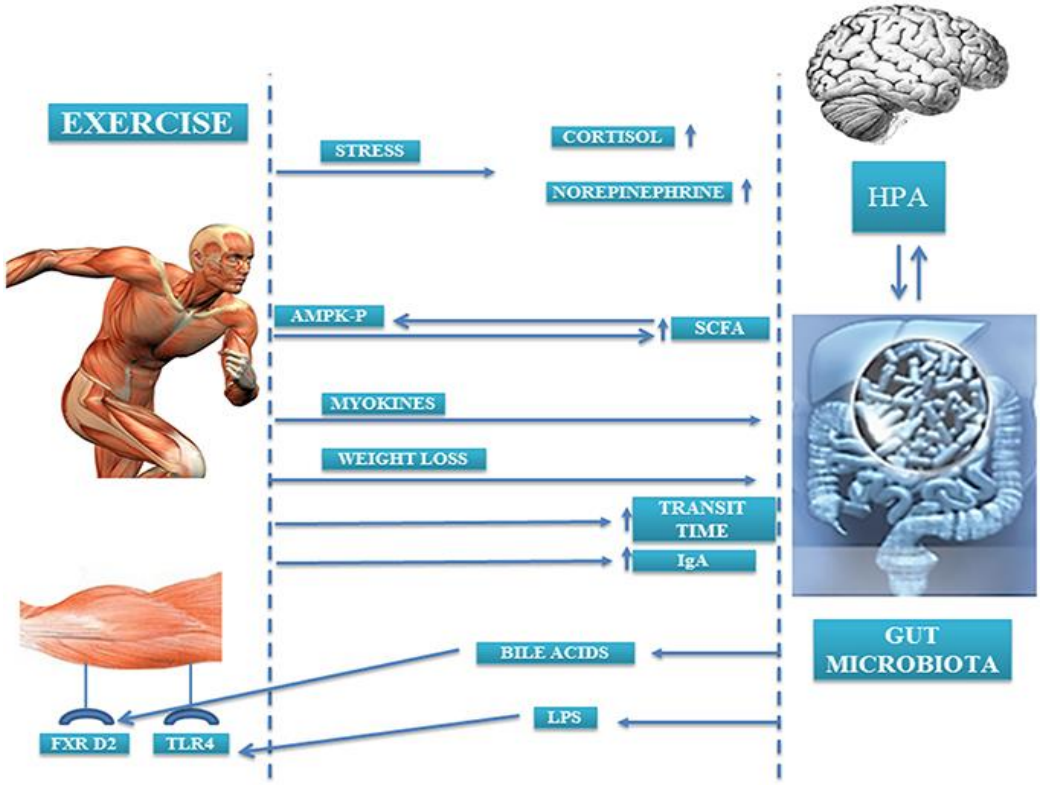


Figure 4. Gut microbiota-exercise interaction mechanisms. Cerdá B, et al., Modification: Another Piece in the Puzzle of the Benefits of Physical Exercise in Health? Front Physiol. 2016 Feb 18;7:51.

Aims and Hypothesis

Aims

- To investigate the effect of two calorie restricted diets (intermittent fasting and continuous calorie restriction) combined with exercise on lean mass retention and strength adaptations.
- To determine whether changes in lean mass retention and strength adaptations are associated with altered gut microbiota composition.

Hypothesis

- Greater muscle accretion and gains in muscle strength will be associated with altered microbiota composition, and/or higher production of short chain fatty acids and lower inflammation

Research Design: Overview

- Forty-four male and female participants recruited.
- Randomised into either “IF + RT*” or “CER + RT*” for 12 weeks.
- Both groups aimed to be isocaloric (average energy restriction of 20% EER), and isonitrogenous consuming an average protein intake of 1.5 grams per kilogram of bodyweight per day.

Screened and eligible participants

Randomised

IF - 5:2

(Fasting day = 30% EER*)

3 x RT sessions/week
(2 x gym + 1 home)

Program followed for 12 weeks

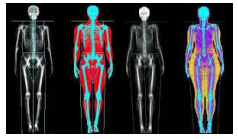
CER - 80%

3x RT sessions/week
(2 x gym + 1 home)

** IF= Intermittent fasting, CER= Continuous Energy Restriction*
RT= Resistance Training, EER=Estimated Energy Requirements

Baseline Assessment

Dexa Scan



pQCT Scan (thigh)

Ultrasound (thigh)

3RM strength testing

Faecal and blood sampling

12 week Intervention Period

Week 1 — Week 6 — Week 12

3- day food diary

3-day food diary

3- day food diary

Every Week

Bioelectrical impedance (BIA)

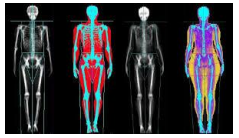
Everyday

Mood, hunger and fullness questionnaire

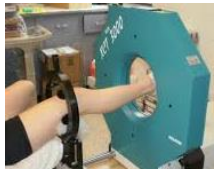
*Faecal samples were sequenced on an Illumina MiSeq platform

Final Assessment

Dexa Scan

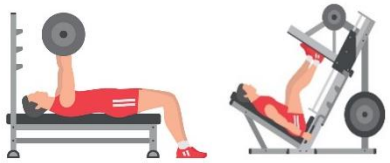


pQCT Scan (thigh)



Ultrasound (thigh)

3RM strength testing



Faecal and blood sampling

Statistical Analysis

- Data is presented as means ± standard deviation.
- Body composition and strength data was analysed using an intention-to-treat linear mixed model, assuming AR(1) dependence across time.
  - In addition, a change analysis was conducted for participants who completed both the baseline and 12 week assessments.
- Bioinformatics analysis involved de-multiplexing, quality control, operational taxonomic unit (OUT) clustering, and taxonomic classification.
  - Spearman's correlation coefficient was to used to determine the association between baseline bacteria species richness, relative abundance of phylum bacteria with lean body mass (LBM) and upper and lower body strength pre- and post-intervention.

Preliminary Results

Intervention Group	Body Weight (kg)			Body Mass Index (BMI)			Body Fat Percentage (%)			Lean Body Mass (kg)		
	Baseline	Post-Intervention	Δkg	Baseline	Post-Intervention	ΔBMI	Baseline	Post-Intervention	Δ%	Baseline	Post-Intervention	Δkg
Intermittent Fasting (n=17)	80.1±13.8	76.4±12.5	-3.7*	27.0±2.7	25.8±2.6	-1.2*	35.8±8.6	29.3±7.6	-6.5*	54.3±12.7	56.2±12.2	1.8*
Continuous Energy Restriction (n=17)	79.6±13.5	75.5±11.8	-4.0*	27.1±2.9	25.8±2.7	-1.3*	36.7±7.9	29.7±8.7	-6.9*	53.3±13.1	54.8±12.1	1.6*

Table 1. The effect of 12 weeks of IF and CER combined with exercise on body weight and body composition. Data is presented as means ± SD. * represents significant time effect $p < 0.01$.

Preliminary Results

Intervention Group	3 Repetition Max (RM) Bench Press (kg)			3 Repetition Max (RM) Leg Press (kg)		
	Baseline	Post-Intervention	Δkg	Baseline	Post-Intervention	Δkg
Intermittent Fasting (n=17)	43.3±18.5	47.7±17.45	4.4*	112.1±54.1	145.8±57.2	33.8*
Continuous Energy Restriction (n=17)	39.5±19.5	45.1±18.5	5.6*	108.1±65.8	137.6±59.6	29.6*

Table 2. The effect of 12 weeks of IF and CER combined with exercise on 3RM upper and lower body strength. Data is presented as means ± SD. * represents significant time effect $p < 0.01$.

Preliminary Results

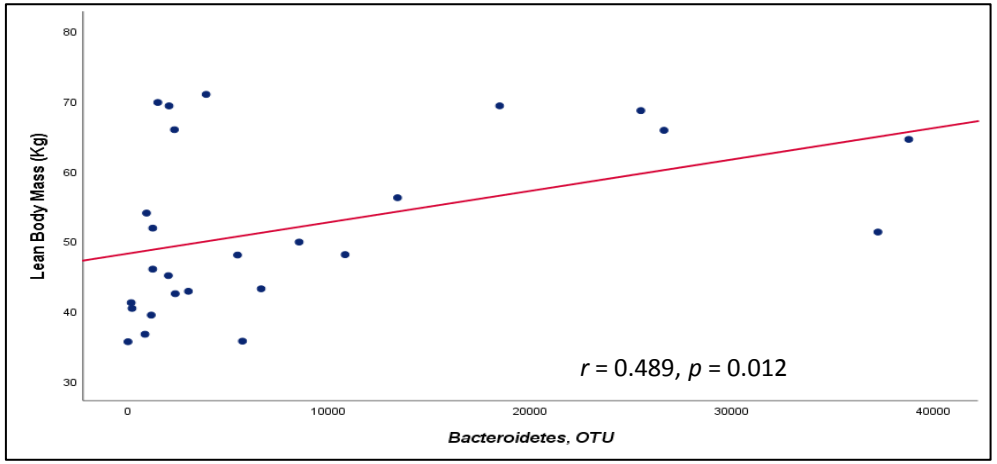


Figure 5. The association between baseline LBM and Bacteriodetes.

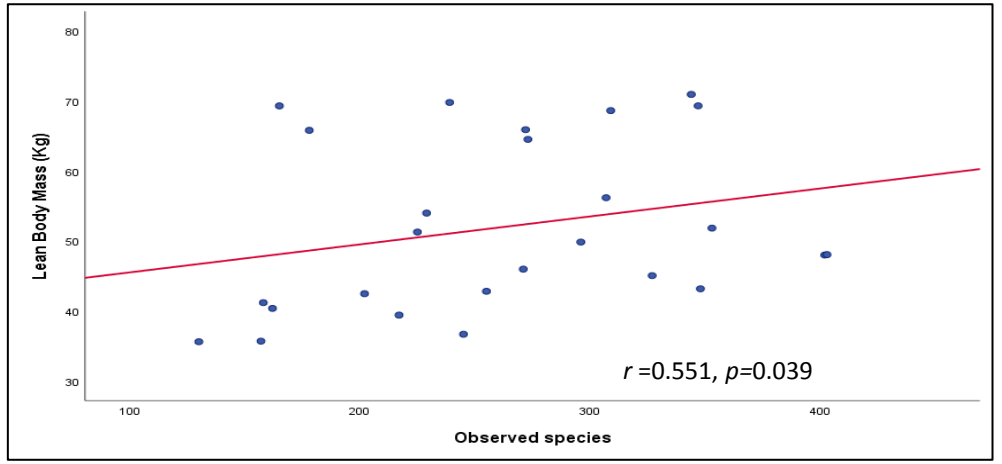


Figure 6. The association between baseline LBM and species richness.

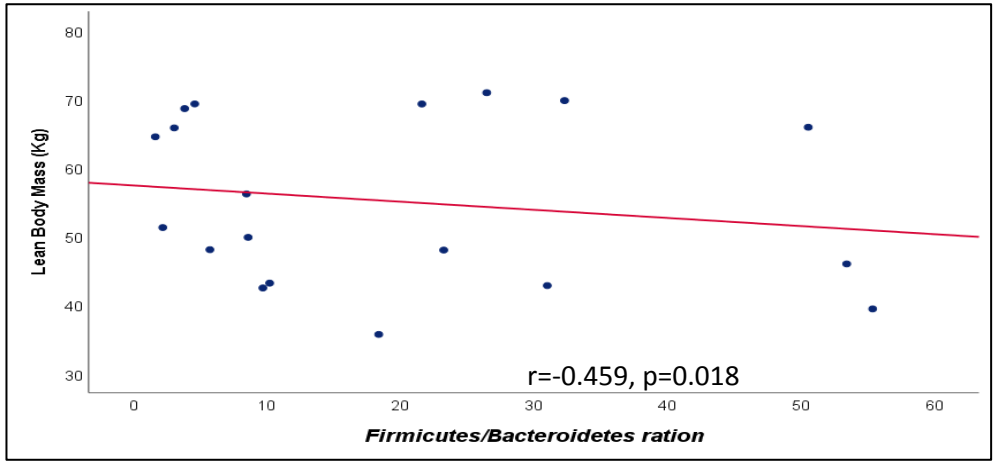


Figure 7. The association between baseline LBM and Firmicutes/Bacteriodietes ratio.

r: Spearman's correlation coefficient

Preliminary Results

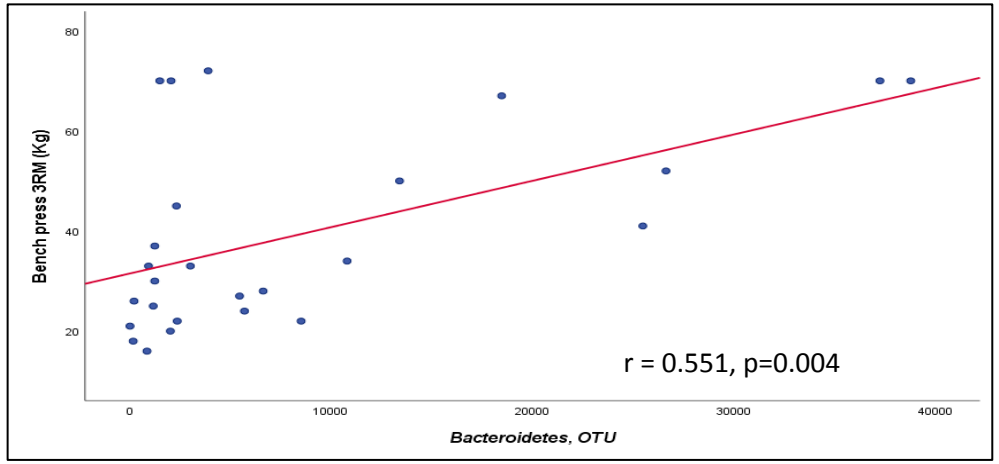


Figure 8. The association between baseline 3RM Bench Press strength and Bacteriodietes.

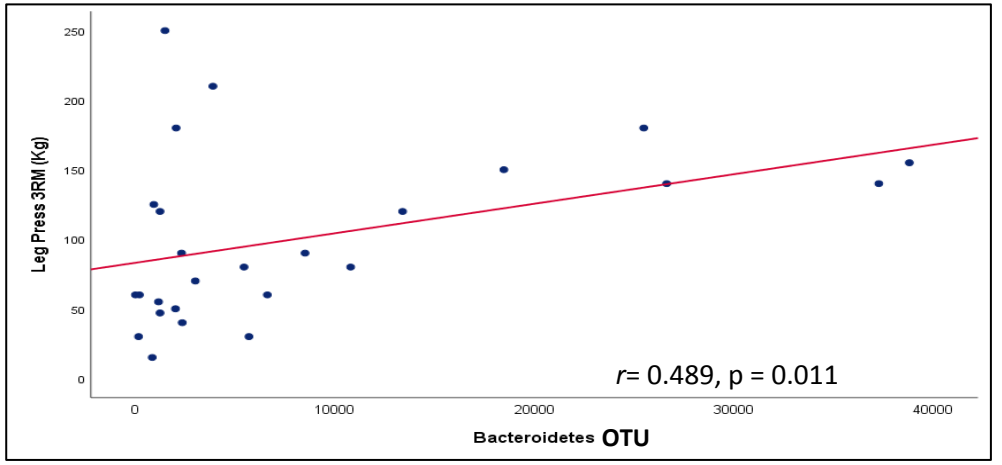


Figure 9. The association between baseline 3RM Leg Press strength and Bacteriodietes.

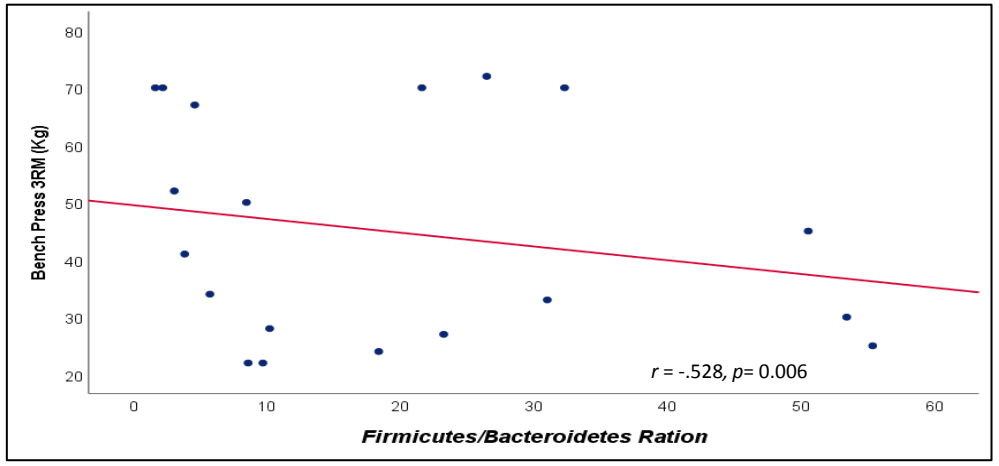


Figure 10. The association between baseline 3RM Bench Press strength and Firmicutes/Bacteriodietes ratio.

r: Spearman's correlation coefficient

Preliminary Results

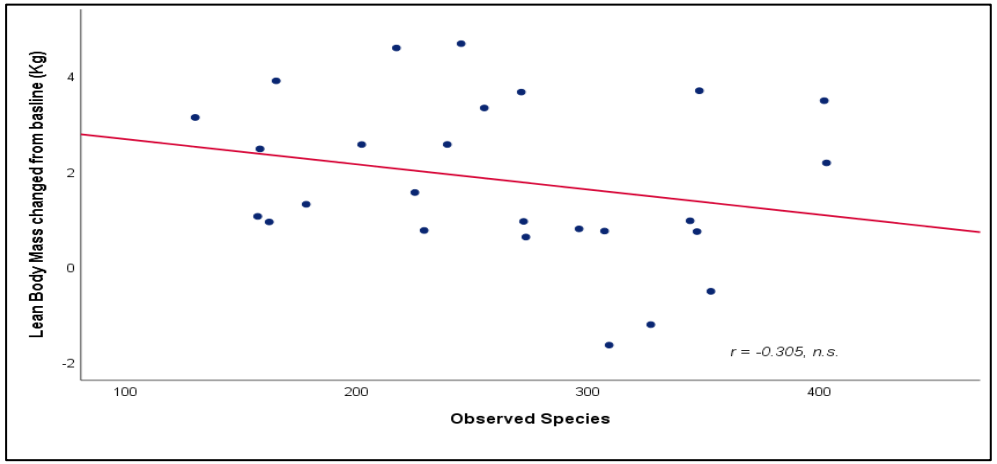


Figure 11. The association between change in LBM and species richness.

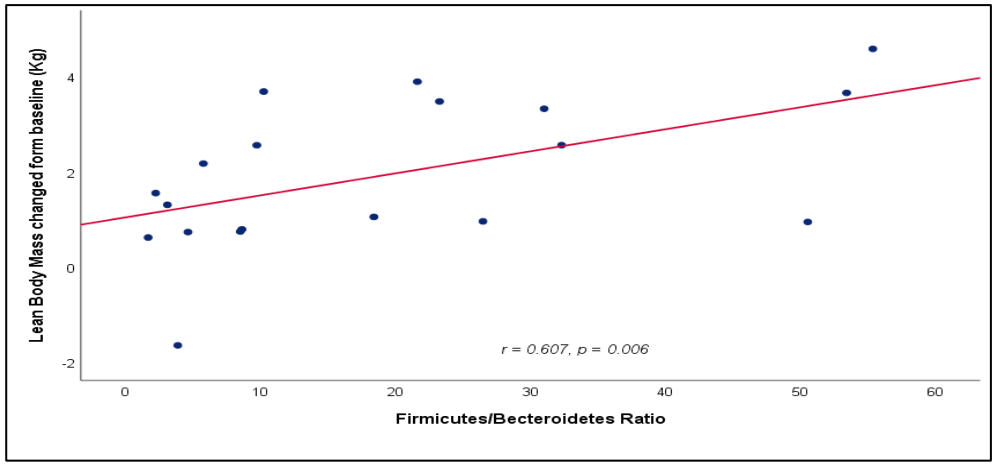


Figure 12. The association between change in LBM and Firmicutes/Bacteriodietes ratio.

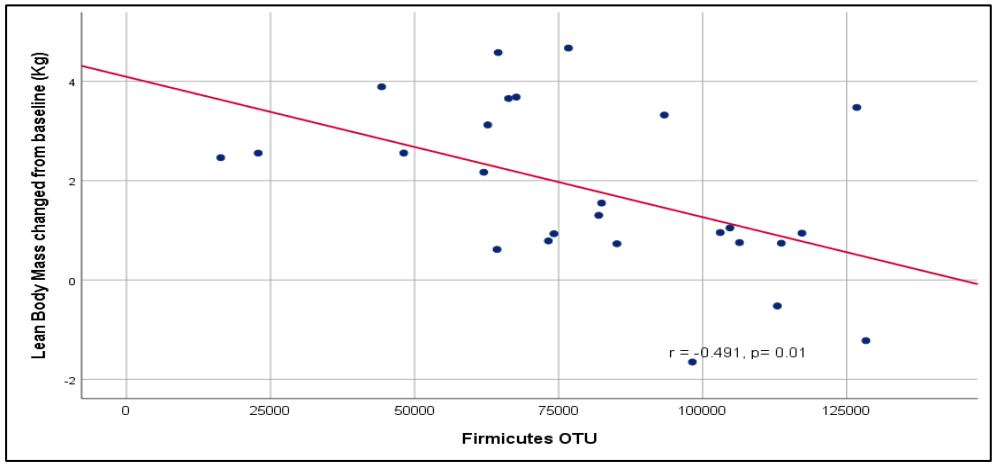


Figure 13. The association between change in LBM and Firmicutes

r: Spearman's correlation coefficient

Conclusions

- Across both diet groups, males and females experienced a significant reduction in bodyweight while, on average, increasing LBM.
  - Neither diet was more effective for bodyweight reduction, however there was a trend towards greater LBM accrual in IF compared to CER males.
  - Higher protein diet maybe important for muscle mass retention and/or growth during times of energy restriction

- Exploratory correlation analysis of bacterial phyla and lean body mass and upper and lower body strength revealed:
- At Baseline -
  - Species diversity was associated with LBM
    - Positive correlation between species diversity and LBM
  - Bacteroidetes abundance and Firmicutes/Bacteroidetes ratio was associated with LBM, upper and lower body strength
    - Positive correlation between bacteroidetes abundance and LBM, upper and lower body strength
    - Negative correlation between Firmicutes/Bacteroidetes ratio and LBM and upper body strength

Conclusions

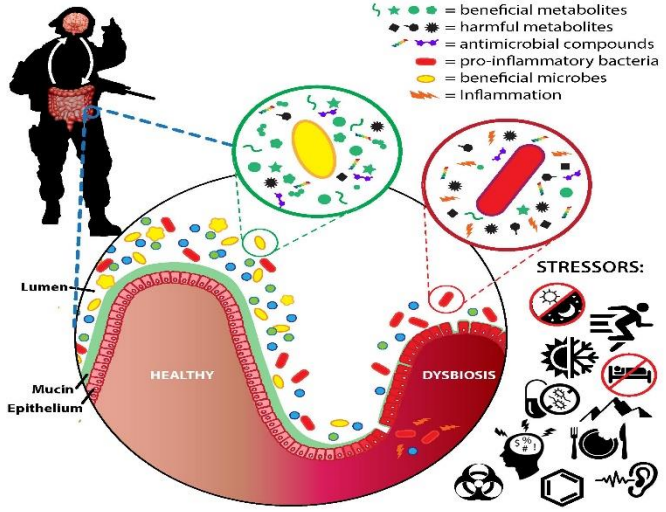
- Changes in LBM following a 12 week lifestyle intervention was associated with Firmicutes abundance and Firmicutes/Bacteroidetes ratio
  - Negative correlation between Firmicutes/Bacteroidetes ratio, Firmicutes abundance and LBM

- Individuals with lower baseline scores in relative abundance of specific microbial phyla and lower Firmicutes/Bacteroidetes ratio could be more responsive to the effects diet and/or exercise intervention
  - Resulting in greater changes in abundance of microbial phyla (and potentially diversity)
  - Enhancement of body composition and muscle strength
  - To be confirmed following analysis of post faecal samples

- Future Work
  - Analysis of Post-training data
    - Confirm association results
    - Identify possible mediators
      - Analysis faecal and plasma metabolomics
      - Markers of intestinal integrity and
      - Inflammation.

Practical Applications for Defence

- Army personal may under-consume energy for periods of their training, with deficits in CHO intake and PRO (compared to recommended optimal PRO intakes to enhance training adaptations)
  - Could lead to loss of muscle mass and impact function and performance
- Our preliminary findings showed that gains in muscle mass and strength can still occur despite calorie deficit
  - As long as protein consumption is high
- Microbiota data suggest that microbial diversity and specific microbial phyla – Firmicutes and Bacteroidetes maybe associated with baseline LBM and strength, but also adaptations in response to diet and exercise



Karl, J. P., et al., (2018). Effects of Psychological, Environmental and Physical Stressors on the Gut Microbiota.
Frontiers in microbiology, *9*, 2013. doi:10.3389/fmicb.2018.02013

Acknowledgements

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