TITLE: Exercise and the microbiota

AUTHORS: Orla O'Sullivan, Owen Cronin, Siobhan F Clarke, Eileen F Murphy, Micheal G Molloy, Fergus Shanahan & Paul D Cotter

NOTICE: This is an electronic version of an article published in Gut Microbes, Volume 6, Issue 2, 2015 , pages 131-136. *Gut Microbes* is available online at:

www.tandfonline.com/10.1080/19490976.2015.1011875

Exercise and the microbiota

O'Sullivan O, [2] Cronin O, [3] Clarke SF, [1,2] Murphy EF, [4] Molloy MG, [3] Shanahan F [1,3] and Cotter PD [1, 2]

[1] Alimentary Pharmabiotic Centre,[2] Teagasc Food Research Centre, Moorepark, Fermoy, Cork, Ireland, [3] Department of Medicine, University College Cork, National University of Ireland, [4] Alimentary Health Ltd, Cork.

Keywords: 5-10 Key words

Exercise, diet, gut microbiota , diversity, host-microbe interactions

Original citation: Clarke SF, Murphy EF, O'Sullivan O, Lucey AJ, Humphreys M, Hogan A, Hayes P, O'Reilly M, Jeffery IB, Wood-Martin R, Kerins DM, Quigley E, Ross RP, O'Toole PW, Molloy MG, Falvey E, Shanahan F, Cotter PD. Exercise and associated dietary extremes impact on gut microbial diversity. Gut 2014; 63:1913-20; PMID: 25021423; http://dx.doi.org/10.1136/gutjnl-2013-306541).

Correspondence to:

Fergus Shanahan, MD, DSc

Department of Medicine,

Alimentary Pharmabiotic Centre

University College Cork, National University of Ireland

Cork, Ireland

Telephone +353-(0)21-4901226

FAX +353-(0)-21-4345300

Email: F.shanahan@ucc.ie

Disclosure of Potential Conflicts of Interest: No potential conflicts of interest are disclosed.

Abstract

Sedentary lifestyle is linked with poor health, most commonly obesity and associated disorders, the corollary being that exercise offers a preventive strategy. However, the scope of exercise biology extends well beyond energy expenditure and has emerged as a great 'polypill', which is safe, reliable and cost-effective not only in disease prevention but also treatment. Biological mechanisms by which exercise influences homeostasis are becoming clearer and involve multi-organ systemic adaptations. Most of the elements of a modern lifestyle influence the indigenous microbiota but few studies have explored the effect of increased physical activity. While dietary responses to exercise obscure the influence of exercise alone on gut microbiota, professional athletes operating at the extremes of performance provide informative data. We assessed the relationship between extreme levels of exercise, associated dietary habits and gut microbiota composition, and discuss potential mechanisms by which exercise may exert a direct or indirect influence on gut microbiota.

Introduction

Changes in human behaviour have influenced not only the external environment but also the inner microbial environment. This has been particularly evident as societies undergo socio-economic development. The microbiota may be considered as the proximate environmental factor conferring risk or resistance to a range of chronic inflammatory and metabolic disorders that are common in socio-economically developed societies. Underpinning host-environmental interactions is a signalling network among the microbiota, host metabolism and host immunity, with lifestyle factors such as diet influencing each component of this triad. Most of the elements of a modern society have a modifying effect on the indigenous microbiota; but one that has received comparatively little attention is exercise.

Exercise is well known for its metabolic and immunologic effects in the host but an impact on the microbiota has been uncertain.

Why is this important? Other than diet, few environmental variables in the modern lifestyle of a host exert such a level of control across a range of physiological events. We have shown that frailty and poor exercise capacity in the elderly is linked with low fecal microbial diversity that correlates with poor dietary diversity. [1] The possibility that exercise or combined dietary change might enhance microbial diversity has immediate implications not limited to the elderly. The growing list of reported health benefits from exercise now includes: limitation of age-related cognitive impairment, [2] prevention of colon cancer [3] and the treatment of diabetes, [4] irritable bowel syndrome [5] and depression. [6] The microbiota and diet-microbe-host immune and metabolic interactions have been implicated in the etiopathogenesis of all of these conditions.

The influence of physical activity on intestinal microbiota has largely been understudied. The majority of work to date pertains to murine models examining the combined effect of exercise and dietary interventions such as low energy or high fat diets. [7] However, the anti-inflammatory effect of regular exercise and modulation of infection risk has been the subject of much research. [8] Of late, exercise's ability to enable cross talk between skeletal muscle and multiple other organs (e.g. brain, bone, liver, adipose tissue, gut) has become increasingly recognized. [9, 10]

Training-related skeletal muscle plasticity, derives from a complex set of physiological and molecular processes, and is subject to the exercise mode engaged. In particular, peroxisome proliferator-activator receptor-G-coactivator-1α (PGC-1α), abundantly expressed in skeletal muscle tissue, is thought to play a key role in muscular adaptability in response to resistance or weights training. [11] PGC-1α, a regulator of mitochondrial biogenesis within

skeletal tissue, targets a diverse range of transcription factors in distant tissues, with multiple downstream effects. [12]

Here, we summarise recent work with professional athletes, operating at the extremes of performance, as a human model to explore diet-exercise-microbe inter-relationships and discuss potential mechanisms by which exercise might influence the microbiota.

A study of the extreme

To begin to explore the relationship among exercise, associated dietary adjustments, and the microbiota in humans, we reasoned that professional athletes operating at the extremes of fitness and performance would be an optimal study group in which to detect biologically significant deviations from controls. We studied the Irish International rugby football team during a pre-World Cup training camp in which dietary intake and physical activity were monitored.[13, 14] A comprehensive range of observational and laboratory assessments was performed which included: pattern and content of dietary intake, metagenomic fecal microbiota compositional analysis, body composition (by DEXA scanning), pro- and anti-inflammatory cytokines, and creatine kinase. It is noteworthy that many professional rugby athletes might be classified as overweight or obese, due to their large muscle mass, if assessed by body mass index (BMI) alone. Body size of rugby players is, in part, related to their specialist position within the team formation; forwards have high BMI and back players tend to have a lower or normal BMI. In anticipation of the possibility that BMI might directly or indirectly have a confounding influence on the interpretation of results, we recruited two control groups of healthy non- professional athletes with varying levels of physical activity, i.e. a low BMI group (BMI < 25) and a high BMI group (BMI > 28). As expected, the elite professional athletes had a higher lean body mass, lower body fat percentage and waist:hip ratios compared to controls. The rugby professionals grazed

throughout the day whereas controls ate at conventional meal times. The professional athletes also consumed significantly higher quantities of protein, fat, carbohydrates, sugar, cholesterol and saturated fat per day than both control groups and consumed significantly higher quantities of starch, fibre, mono-unsaturated fat and poly-unsaturated fat than the high BMI control group (details of diet were extrapolated from food frequency questionnaires). Remarkably, despite extremes of exercise as evidenced by extraordinarily high levels of creatine kinase in serum (serving as a proxy for degree of exercise) compared to the control groups, the professional athletes had lower levels of inflammatory cytokines than the controls. In addition, they had increased fecal microbial diversity. From a taxonomic perspective, this increased diversity was reflected by the presence of representatives of 22 phyla of bacteria in the athlete microbiota, in contrast to 11 and 9 phyla in the low and high BMI controls, respectively. Notably, among the taxa that were enriched in the athlete and low BMI populations relative to their high BMI counterparts was the lean phenotype-associated, *Akkermansia muciniphila*. [15, 16] The increased gut microbial diversity in athletes correlated with both protein intake and creatine kinase levels (Table 1).

Limitations and prospects

While we were careful in our interpretation of the data not to transmute correlation into causation it may be tempting to view increased microbial diversity as a benefit of exercise. However, this remains to be demonstrated by a prospective rather than cross-sectional study of volunteers undergoing a structured exercise programme which attempts to control for diet. Such a study is underway and will focus on healthy controls rather than the elite professional athlete. Furthermore, in addition to compositional analysis, whole genome sequencing for functional genomic analysis along with metabolomic profiling will be included to ascertain which, if any, functional pathways are altered during exercise training. Diet-exercise may

represent an inseparable interaction, the contributions of each being impossible to assess in isolation. It is noteworthy that exercise as a weight reduction strategy is often poorly efficacious unless combined with dietary restriction, [17] as an increase in physical activity by exercise-naïve individuals may result in unanticipated dietary change, driven by physiological responses to exercise and a risk of compensatory over-eating post-exercise. [18] Appetite response to exercise is complex and appears to be highly variable between individuals. [19] It is dependent on multiple factors including mental attitude towards exercise, [20] the intensity of the activity involved [21] and prior reliance on substrate availability for energy expenditure. [22] Moderately intense physical activity is followed by a transient surge in serum ghrelin levels in the post-exercise period. This rise is not apparent however following high intensity exercise. [21] The close relationship between diet and exercise obscures the study of exercise and microbiota in humans. Detailed analysis of baseline and subsequent dietary changes are essential.

Exercise biology and the microbiota – potential mechanism of interaction

Although a direct causal relationship between exercise and gut microbial composition or function has not been established and is inextricably linked with dietary adjustments, several potential mechanisms by which physical activity and fitness might modify the microbiota can be entertained. A schematic representation of some of the points of interaction is shown in Figure 1.

Abrupt exercise involves the production of multiple metabolites and inflammatory mediators, [23] but habitual exercise and fitness leads to suppression of basal pro-inflammatory cytokines (as found in our study) [13] indicating a regulatory loop between exercise biology and host immunity. Regular physical activity is a recognized treatment for multiple inflammatory-centred conditions. The anti-inflammatory effect of regular exercise results in improved

immunological profile in type 2 diabetes mellitus, [24] coronary artery disease, [25] peripheral arterial disease [26] and obesity. [27] In particular, combined aerobic and resistance training in type 2 diabetics leads to a decrease in pro-inflammatory cytokines IL-1β, IL-6, TNF-α, IFN-γ as well as an increase in anti-inflammatory modulators IL-4 and IL-10. [24] Amongst elderly subjects, aerobic and resistance training individually attenuate inflammatory tone but may take up to 6-months to take effect. [28]

Whether intestinal immunological responses to exercise mirror those of the systemic immune compartment is unclear but seems likely. Strenuous aerobic activity in healthy mice leads to an immediate increase in TNF-α expression in intestinal lymphocytes, [29] but appears to be suppressed below pre-exercise levels at 24-hours post-exercise. [30] Anti-inflammatory IL-10 expression in intestinal lymphocytes is also increased immediately post-exercise. [30, 31] Furthermore, regular low to moderate intensity exercise reduces colonic oxidative insult in a rat model of colitis. [32]

Of course, prolonged excessive exercise has a deleterious influence on intestinal function. Intense exercise redistributes blood from the splanchnic circulation to actively respiring tissues. [33] Prolonged intestinal hypo-perfusion impairs mucosal homeostasis and causes enterocyte injury. [34] Intestinal ischemia may result, particularly in the setting of dehydration, manifesting as abdominal cramps, diarrhoea or occasionally bloody diarrhoea. [35] This is particularly true in endurance sports. [36] As a result, increased intestinal permeability ensues, thought to be driven by the phosphorylation of several tight junction proteins. [37] These events render the gut mucosa susceptible to endotoxin translocation. [38] By contrast, regular moderate exercise mitigates the effects of stress-induced intestinal barrier dysfunction. Moderate exercise has been associated with a lesser degree of intestinal permeability, preservation of mucous thickness and lower rates of bacterial translocation along with up-regulation of anti-microbial protein production and gene expression in small

intestinal tissue (α-defensin, β-defensin, Reg IIIb and Reg IIIc). [39] In a mouse model of colitis, forced treadmill exercise training aggravated inflammatory indices and outcomes, while voluntary wheel training displayed a protective effect. [40] This suggests a biphasic and dose-response effect of exercise on gut health that has yet to be fully explored in humans.

The brain-gut axis is an established bi-directional avenue of signalling between two organs both of which are influenced by exercise. Several lines of evidence have extended this to include the microbiota. [41] Information exchange along the brain-gut-microbe axis is hard wired by the vagus nerve, neuroendocrine mediators including the hypothalamic-pituitary-adrenal axis and a variety of neurotransmitter and local hormones, some of which are also produced by the microbiota. [41, 42] This regulates several homeostatic processes including appetite, satiety and digestion. The vagus also regulates gastro-intestinal inflammatory tone, direct visualization of which has been shown with a pro-inflammatory response to vagotomy in mice. [43] The presence of a vagally driven anti-inflammatory pathway is now established [44] and cholinergic innervation to intestinal sub-mucosa may exert influence on mucosal innate immunity. [45] Evidence suggests that this parasympathetic neuro-immune reflex is dependent on vagal afferent neurons for the local release of intestinal inflammatory mediators in response to pathogenic gut bacteria. [46] Therefore, elevated vagal tone and parasympathetic influence, as seen in the resting state of athletes, may foster a preferential anti-inflammatory milieu at the intestinal-luminal interface, with an attendant conditioning influence on microbial composition.

Finally, exercise prescription is increasingly recognised as a safe and effective treatment for disorders ranging from depression [6] and irritable bowel syndrome [5] to inflammatory bowel disease [47] adding further support for the potential influence of exercise on the brain-gut-microbiome axis. By contrast, withdrawal of exercise from individuals accustomed to regular activity leads to deterioration in mood in as little as two weeks post-

exercise. [48] Furthermore, plasma kynurenine metabolite levels, which are strongly correlated with depression, [49] are subject to influence by exercise-related PGC-1α1 expression. Increased expression of muscle-specific PGC-1α1 in transgenic mice is associated with greater resilience to chronic stress and stress-induced CNS inflammation. [50] Thus, there are several points at which exercise biology and accompanying dietary adjustments may interact directly or indirectly with host-microbe interactions in the gut (Figure 1).

Conclusion

Whether the gut microbiota of sedentary subjects can be "trained" to resemble that of an elite athlete through exercise with increased protein intake or a combination of both is an intriguing prospect. It may be impossible to truly separate the physical act of exercise from the accompanying dietary adjustments that volunteers make, often unwittingly, during an exercise programme. In addition, it is presently unclear if there is a dose-response between exercise levels and beneficial alterations in microbial composition or function. Regardless, determination of optimal diet-exercise combination programmes which might give the elite athlete 'an edge' over competitors is worthy of careful pursuit including in sub-elite non-professional volunteers and may ultimately inform our advice regarding such activity in patients with a variety of disorders which could benefit from a personalised training programme.

Acknowledgements

The authors are supported in part by research grants from Science Foundation Ireland including a centre grant (Alimentary Pharmabiotic Centre; Grant Numbers SFI/12/RC/2273 and 12/RC/2273). Dr. Orla O'Sullivan is funded by a Starting Investigator Research Grant from Science Foundation Ireland (Grant number 13/SIRG/2160). Dr. Paul Cotter is funded by a Principal Investigator Award from Science Foundation Ireland P.D.C are supported by a SFI PI award (Grant number 11/PI/1137).

References:

1. Claesson MJ, Jeffery IB, Conde S, Power SE, O'Connor EM, Cusack S, Harris HM, Coakley M, Lakshminarayanan B, O'Sullivan O, Fitzgerald GF, Deane J, O'Connor M, Harnedy N, O'Connor K, O'Mahony D, van Sinderen D, Wallace M, Brennan L, Stanton C, Marchesi JR, Fitzgerald AP, Shanahan F, Hill C, Ross RP, O'Toole PW. Gut microbiota composition correlates with diet and health in the elderly. Nature 2012; 488:178-84.

2. Bherer L, Erickson KI, Liu-Ambrose T. A review of the effects of physical activity and exercise on cognitive and brain functions in older adults. Journal of aging research 2013; 2013:Article ID 657508.

3. Robsahm TE, Aagnes B, Hjartaker A, Langseth H, Bray FI, Larsen IK. Body mass index, physical activity, and colorectal cancer by anatomical subsites: a systematic review and meta-analysis of cohort studies. European journal of cancer prevention : the official journal of the European Cancer Prevention Organisation (ECP) 2013; 22:492-505.

4. Schwingshackl L, Missbach B, Dias S, Konig J, Hoffmann G. Impact of different training modalities on glycaemic control and blood lipids in patients with type 2 diabetes: a systematic review and network meta-analysis. Diabetologia 2014; 57:1789-97.

5. Johannesson E, Simren M, Strid H, Bajor A, Sadik R. Physical activity improves symptoms in irritable bowel syndrome: a randomized controlled trial. The American journal of gastroenterology 2011; 106:915-22.

6. Harkin A. Muscling In on Depression. The New England journal of medicine 2014; 371:2333-4.

7. Evans CC, LePard KJ, Kwak JW, Stancukas MC, Laskowski S, Dougherty J, Moulton L, Glawe A, Wang Y, Leone V, Antonopoulos DA, Smith D, Chang EB, Ciancio MJ. Exercise prevents weight gain and alters the gut microbiota in a mouse model of high fat diet-induced obesity. PLoS One 2014; 9:e92193.

8. Pedersen BK, Akerstrom TC, Nielsen AR, Fischer CP. Role of myokines in exercise and metabolism. J Appl Physiol (1985) 2007; 103:1093-8.

9. Handschin C, Spiegelman BM. The role of exercise and PGC1alpha in inflammation and chronic disease. Nature 2008; 454:463-9.

10. Hawley JA, Hargreaves M, Joyner MJ, Zierath JR. Integrative Biology of Exercise. Cell 2014; 159:738-49.

11. Ruas JL, White JP, Rao RR, Kleiner S, Brannan KT, Harrison BC, Greene NP, Wu J, Estall JL, Irving BA, Lanza IR, Rasbach KA, Okutsu M, Nair KS, Yan Z, Leinwand LA, Spiegelman BM. A PGC-1alpha isoform induced by resistance training regulates skeletal muscle hypertrophy. Cell 2012; 151:1319-31.

12. Lin J, Handschin C, Spiegelman BM. Metabolic control through the PGC-1 family of transcription coactivators. Cell metabolism 2005; 1:361-70.

13. Clarke SF, Murphy EF, O'Sullivan O, Lucey AJ, Humphreys M, Hogan A, Hayes P, O'Reilly M, Jeffery IB, Wood-Martin R, Kerins DM, Quigley E, Ross RP, O'Toole PW, Molloy MG, Falvey E, Shanahan F, Cotter PD. Exercise and associated dietary extremes impact on gut microbial diversity. Gut 2014; 63:1913-20.

14. Hold GL. The gut microbiota, dietary extremes and exercise. Gut 2014; 63:1838-9.

15. Walters WA, Xu Z, Knight R. Meta-analyses of human gut microbes associated with obesity and IBD. FEBS letters 2014; 588:4223-33.

16. Everard A, Belzer C, Geurts L, Ouwerkerk JP, Druart C, Bindels LB, Guiot Y, Derrien M, Muccioli GG, Delzenne NM, de Vos WM, Cani PD. Cross-talk between Akkermansia muciniphila

and intestinal epithelium controls diet-induced obesity. Proceedings of the National Academy of Sciences of the United States of America 2013; 110:9066-71.

17. Johns DJ, Hartmann-Boyce J, Jebb SA, Aveyard P. Diet or Exercise Interventions vs Combined Behavioral Weight Management Programs: A Systematic Review and Meta-Analysis of Direct Comparisons. Journal of the Academy of Nutrition and Dietetics 2014; 114:1557-68.

18. Thomas DM, Bouchard C, Church T, Slentz C, Kraus WE, Redman LM, Martin CK, Silva AM, Vossen M, Westerterp K, Heymsfield SB. Why do individuals not lose more weight from an exercise intervention at a defined dose? An energy balance analysis. Obesity reviews : an official journal of the International Association for the Study of Obesity 2012; 13:835-47.

19. Hopkins M, Blundell JE, King NA. Individual variability in compensatory eating following acute exercise in overweight and obese women. Br J Sports Med 2014; 48:1472-6.

20. Werle CO, Wansink B, Payne CR. Just thinking about exercise makes me serve more food. Physical activity and calorie compensation. Appetite 2011; 56:332-5.

21. Bilski J, Manko G, Brzozowski T, Pokorski J, Nitecki J, Nitecka E, Wilk-Franczuk M, Ziolkowski A, Jaszczur-Nowicki J, Kruczkowski D, Pawlik WW. Effects of exercise of different intensity on gut peptides, energy intake and appetite in young males. Annals of agricultural and environmental medicine : AAEM 2013; 20:787-93.

22. Pannacciulli N, Salbe AD, Ortega E, Venti CA, Bogardus C, Krakoff J. The 24-h carbohydrate oxidation rate in a human respiratory chamber predicts ad libitum food intake. The American journal of clinical nutrition 2007; 86:625-32.

23. Pedersen BK. Special feature for the Olympics: effects of exercise on the immune system: exercise and cytokines. Immunology and cell biology 2000; 78:532-5.

24. Balducci S, Zanuso S, Nicolucci A, Fernando F, Cavallo S, Cardelli P, Fallucca S, Alessi E, Letizia C, Jimenez A, Fallucca F, Pugliese G. Anti-inflammatory effect of exercise training in subjects with type 2 diabetes and the metabolic syndrome is dependent on exercise modalities and independent of weight loss. Nutrition, metabolism, and cardiovascular diseases : NMCD 2010; 20:608-17.

25. Lara Fernandes J, Serrano CV, Jr., Toledo F, Hunziker MF, Zamperini A, Teo FH, Oliveira RT, Blotta MH, Rondon MU, Negrao CE. Acute and chronic effects of exercise on inflammatory markers and B-type natriuretic peptide in patients with coronary artery disease. Clinical research in cardiology : official journal of the German Cardiac Society 2011; 100:77-84.

26. Tisi PV, Hulse M, Chulakadabba A, Gosling P, Shearman CP. Exercise training for intermittent claudication: does it adversely affect biochemical markers of the exercise-induced inflammatory response? European journal of vascular and endovascular surgery : the official journal of the European Society for Vascular Surgery 1997; 14:344-50.

27. Ho SS, Dhaliwal SS, Hills AP, Pal S. Effects of chronic exercise training on inflammatory markers in Australian overweight and obese individuals in a randomized controlled trial. Inflammation 2013; 36:625-32.

28. Martins RA, Neves AP, Coelho-Silva MJ, Verissimo MT, Teixeira AM. The effect of aerobic versus strength-based training on high-sensitivity C-reactive protein in older adults. European journal of applied physiology 2010; 110:161-9.

29. Pervaiz N, Hoffman-Goetz L. Immune cell inflammatory cytokine responses differ between central and systemic compartments in response to acute exercise in mice. Exercise immunology review 2012; 18:142-57.

30. Hoffman-Goetz L, Spagnuolo PA, Guan J. Repeated exercise in mice alters expression of IL-10 and TNF-alpha in intestinal lymphocytes. Brain, behavior, and immunity 2008; 22:195-9.

31. Hoffman-Goetz L, Pervaiz N, Packer N, Guan J. Freewheel training decreases pro- and increases anti-inflammatory cytokine expression in mouse intestinal lymphocytes. Brain, behavior, and immunity 2010; 24:1105-15.

32. Kasimay O, Guzel E, Gemici A, Abdyli A, Sulovari A, Ercan F, Yegen BC. Colitis-induced oxidative damage of the colon and skeletal muscle is ameliorated by regular exercise in rats: the anxiolytic role of exercise. Experimental physiology 2006; 91:897-906.

33. Qamar MI, Read AE. Effects of exercise on mesenteric blood flow in man. Gut 1987; 28:583-7.

34. van Wijck K, Lenaerts K, van Loon LJ, Peters WH, Buurman WA, Dejong CH. Exercise-induced splanchnic hypoperfusion results in gut dysfunction in healthy men. PLoS One 2011; 6:e22366.

35. de Oliveira EP, Burini RC. The impact of physical exercise on the gastrointestinal tract. Curr Opin Clin Nutr Metab Care 2009; 12:533-8.

36. Oktedalen O, Lunde OC, Opstad PK, Aabakken L, Kvernebo K. Changes in the gastrointestinal mucosa after long-distance running. Scandinavian journal of gastroenterology 1992; 27:270-4.

37. Zuhl M, Schneider S, Lanphere K, Conn C, Dokladny K, Moseley P. Exercise regulation of intestinal tight junction proteins. Br J Sports Med 2014; 48:980-6.

38. Brock-Utne JG, Gaffin SL, Wells MT, Gathiram P, Sohar E, James MF, Morrell DF, Norman RJ. Endotoxaemia in exhausted runners after a long-distance race. South African medical journal = Suid-Afrikaanse tydskrif vir geneeskunde 1988; 73:533-6.

39. Luo B, Xiang D, Nieman DC, Chen P. The effects of moderate exercise on chronic stress-induced intestinal barrier dysfunction and antimicrobial defense. Brain, behavior, and immunity 2014; 39:99-106.

40. Cook MD, Martin SA, Williams C, Whitlock K, Wallig MA, Pence BD, Woods JA. Forced treadmill exercise training exacerbates inflammation and causes mortality while voluntary wheel training is protective in a mouse model of colitis. Brain, behavior, and immunity 2013; 33:46-56.

41. Grenham S, Clarke G, Cryan JF, Dinan TG. Brain-gut-microbe communication in health and disease. Frontiers in physiology 2011; 2:94 eCollection 2011.

42. Stilling RM, Bordenstein SR, Dinan TG, Cryan JF. Friends with social benefits: host-microbe interactions as a driver of brain evolution and development? Front Cell Infect Microbiol 2014; 4:147.

43. O'Mahony C, van der Kleij H, Bienenstock J, Shanahan F, O'Mahony L. Loss of vagal anti-inflammatory effect: in vivo visualization and adoptive transfer. American journal of physiology Regulatory, integrative and comparative physiology 2009; 297:R1118-26.

44. Tracey KJ. The inflammatory reflex. Nature 2002; 420:853-9.

45. de Jonge WJ. The Gut's Little Brain in Control of Intestinal Immunity. ISRN gastroenterology 2013; 2013:630159.

46. Riley TP, Neal-McKinney JM, Buelow DR, Konkel ME, Simasko SM. Capsaicin-sensitive vagal afferent neurons contribute to the detection of pathogenic bacterial colonization in the gut. Journal of neuroimmunology 2013; 257:36-45.

47. Khalili H, Ananthakrishnan AN, Konijeti GG, Liao X, Higuchi LM, Fuchs CS, Spiegelman D, Richter JM, Korzenik JR, Chan AT. Physical activity and risk of inflammatory bowel disease: prospective study from the Nurses' Health Study cohorts. Bmj 2013; 347:f6633.

48. Kop WJ, Weinstein AA, Deuster PA, Whittaker KS, Tracy RP. Inflammatory markers and negative mood symptoms following exercise withdrawal. Brain, behavior, and immunity 2008; 22:1190-6.

49. Claes S, Myint AM, Domschke K, Del-Favero J, Entrich K, Engelborghs S, De Deyn P, Mueller N, Baune B, Rothermundt M. The kynurenine pathway in major depression: haplotype analysis of three related functional candidate genes. Psychiatry research 2011; 188:355-60.

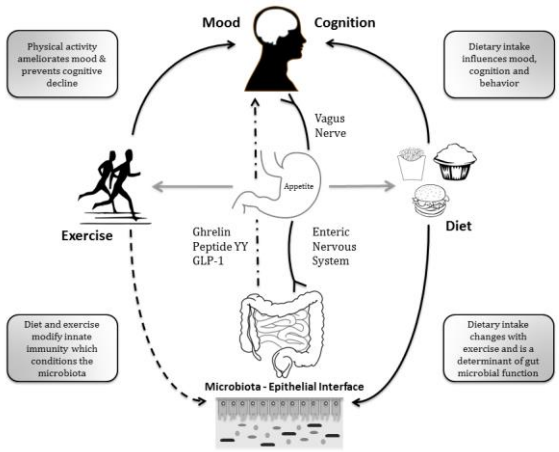
50. Agudelo LZ, Femenia T, Orhan F, Porsmyr-Palmertz M, Goiny M, Martinez-Redondo V, Correia JC, Izadi M, Bhat M, Schuppe-Koistinen I, Pettersson AT, Ferreira DM, Krook A, Barres R, Zierath JR, Erhardt S, Lindskog M, Ruas JL. Skeletal muscle PGC-1alpha1 modulates kynurenine metabolism and mediates resilience to stress-induced depression. Cell 2014; 159:33-45.

Table 1. Correlations between exercise intensity (creatine kinase), diet (protein intake) and diversity (estimated over 5 metrics; chao1, Simpson, Shannon, phylogenetic diversity and observed species). Table displays Pearson correlation coefficients followed by p-value in all incidences. P >0.05 was taken to be significant.

Figure 1.

Schematic overview of potential sites of interaction between the biological adaptations to exercise and the microbiota. This is intended to be representative not comprehensive. Exercise is linked with a diversity of biological responses including a modifying influence on the brain-gut-microbe axis, diet-microbe-host metabolic interactions, neuro-endocrine and neuro-immune interactions. For example, exercise is long known to increase vagal tone - the hard wiring of the gut - which is anti-inflammatory and immune-modulatory. The latter might represent an indirect means by which exercise conditions gut microbiota composition.

	Chao1	Simpson	Shannon	Phylogenetic diversity	Observed species
Creatine kinase (IU/L)	0.166/0.128	0.235/0.030	0.296/0.006	0.298/0.006	0.204/0.601
Protein intake (g/day)	0.258/0.017	0.236/0.030	0.241/0.007	0.427/<0.001	0.289/0.007


Mood
Cognition
Physical activity ameliorates mood & prevents cognitive decline
Dietary intake influences mood, cognition and behavior
Vagus Nerve
Appetite
Exercise
Diet
Ghrelin
Peptide YY
GLP-1
Enteric Nervous System
Diet and exercise modify innate immunity which conditions the microbiota
Dietary intake changes with exercise and is a determinant of gut microbial function
Microbiota - Epithelial Interface