Accepted manuscript

Digging deep for nutrients and metabolites derived from high dietary protein intake and their potential functions in metabolic health

Sarah Gilsenan[1,2], Dara Leong[1,2], Paul D. Cotter[1,2,4], Lorraine Brennan[2,3], Kanishka N. Nilaweera[1,2*]

[1]Food Biosciences Department, Teagasc Food Research Centre, Moorepark, Fermoy, County Cork 6 P61 C996, Ireland.

[2]VistaMilk Research Centre, Teagasc, Moorepark, Fermoy, County Cork P61 C996, Ireland.

[3]UCD School of Agriculture and Food Science, UCD Institute of Food and Health, Belfield, UCD, Dublin 4, Ireland

[4]APC Microbiome Ireland, University College Cork, Cork T12 YT20, Ireland 11

***Corresponding Author:** Kanishka N. Nilaweera, E mail: kanishka.nilaweera@teagasc.ie., Tel.: 003532542674

Keywords: proteins, surgery, microbiota, metabolites, metabolic health

Shortened title: Meabolites of dietary proteins

This peer-reviewed article has been accepted for publication but not yet copyedited or typeset, and so may be subject to change during the production process. The article is considered published and may be cited using its DOI.

10.1017/S0954422424000374

Nutrition Research Reviews is published by Cambridge University Press on behalf of The Nutrition Society

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

Abstract

Intake of high quantities of dietary proteins sourced from dairy, meat or plants can affect body weight and metabolic health in humans. To improve our understanding of how this may be achieved, we reviewed the data related to the availability of nutrients and metabolites in the faeces, circulation and urine. All protein sources (≥20% by energy) increased faecal levels of branched chain fatty acids and ammonia, and decreased the levels of butyrate. There were metabolites responding to dairy and meat proteins (branch chain amino acids) as well as dairy and plant proteins (p-cresol), which were increased in faecal matter. Specific to dairy protein intake, the faecal levels of acetate, indole and phenol were increased, whereas plant protein intake specifically increased the levels of kynurenine and tyramine. Meat protein intake increased the faecal levels of methionine, cysteine and alanine, and decreased the levels of propionate and acetate. The metabolite profile in the faecal matter following dairy protein intake mirrored availability in circulation or urine. These findings provide an understanding of the contrasting gut versus systemic effects of different dietary proteins, which we know to show different physiological effects. In this regard, we provide directions to determining the mechanisms for the effects of different dietary proteins.

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

Introduction

All living organisms require a constant supply of nutrients that can be metabolised in tissues, acting as fuels for growth and development, as well as regulators of nutrient (energy) homeostasis. This process is controlled, in part, by the small intestine by allowing digestion to take place, breaking complex nutrients into forms that can easily be absorbed into the circulation and/or by producing signalling molecules that communicate the availability of nutrients in the gastrointestinal tract to other tissues (Ma and Lee, 2020, Martinez-Guryn et al., 2018, Saps and Miranda, 2017). In contrast, the colon receives much less nutrient load compared to the small intestine because of absorption through the latter tissue and yet, a diverse range of metabolites are produced in the colon from metabolism of dietary nutrients by the gut microbiota inhabiting this tissue, resulting in a range of metabolic health outcomes (Figure 1) (Rajilić-Stojanović and de Vos, 2014, Wan et al., 2019, Nychyk et al., 2021, Clarke et al., 2012, Turnbaugh et al., 2009, Covasa et al., 2019). In this article, we focused attention on the nutrient and metabolite profiles created by high dietary protein (HDP) intake, which differ in source, to gaining knowledge on how the different dietary proteins influence body weight and metabolic health.

Effects on physiology and metabolic health

A renewed focus to understand the relationship between diet and metabolic health has arisen in part due to the increased prevalence of obesity and associated comorbidities over the last 100 years, mostly due to high calorie intake, particularly an increased intake of dietary fat which affects metabolic health (Wang et al., 2020, Le et al., 2020, Morselli et al., 2014, Lee et al., 2021, Hu et al., 2018, Santesso et al., 2012). A particular interest in protein intake has emerged with many weight loss or weight maintenance recommendations promoting increased protein intake, generally above 20% of total energy intake within the 10-30% acceptable macronutrient range for proteins (Wolfe et al., 2017). Data show that HPD intake reduced body weight gain or cause weight loss up to 10%, with a reduction in fat mass and an increased lean mass in over-weight and obese humans of both sexes (Table 1)(Sacks et al., 2009, Pesta and Samuel, 2014, Moon and Koh, 2020, Santesso et al., 2012). The effects extended to include reduction in plasma insulin levels, triglycerides, high density lipoproteins and blood pressure (Table 1). Notably, whilst these effects have been shown relative to baseline measurements or in comparison to carbohydrate intake (Table 1), there is evidence

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

that the quality of the protein also impacts on metabolic health (Table 1). For instance, whey protein (WP) intake reduced waist circumference, circulating ghrelin and insulin-like growth factor (IGF)1 levels in obese humans in comparison to soy intake (Table 1). Relative to collagen, WP caused a reduction in visceral fat, with similar effects shown for milk proteins, containing both WP and casein and in comparison to controls fed milk proteins and soy (Table 1). These effects reported during *ad libitum* intake, have been extended to include calorie restriction, with WP showing a greater improvement of metabolic health than other protein sources (Table 1), albeit there are few exceptions (Table 1)(Piccolo et al., 2015, Kjolbaek et al., 2017). It is also important to highlight that there are also data showing unhealthy outcomes of HPDs. For instance, red meat intake has been associated with increased risk of colorectal cancers and kidney disease (Lew et al., 2017, Aykan, 2015). The different effects of proteins on metabolic health can be related to the quantity and composition of the amino acids, how the proteins are digested and absorbed through the gut as well as the impact on the gut microbiota and their functional capacity to produce metabolites, which ultimately impacts on host health (Gorissen et al., 2018, Bilsborough and Mann, 2006, Agus et al., 2021, Cunningham et al., 2021). For this review, we focused attention on dairy, meat or plant protein intake and their impact on the abundance of metabolites produced in the gut (and hence detected in faeces) as well as that emerge in circulation/urine, to better understand how different proteins affect host metabolic health. Our focus was on data related to human studies albeit in few cases, we have mentioned rodent studies to draw conclusions. The search include effects of HPD, where the protein content was equal or greater than 20% of total energy intake.

Effect on the gut microbiota

The gastrointestinal tract is inhabited by the microbiota, with the colon containing a much higher density of microbiota (10^{10} to 10^{11} cells per millilitre of contents), as well as a much more diverse microbial composition compared to other parts of the intestine (Eckburg et al., 2005, Vuik et al., 2019). This complexity in microbial communities is further supported by the colonic structure and functions. Notably, the colon is made up of a number of colonic epithelial cells (Figure. 1), with many of these cells, particularly goblet cells, capable of producing enzymes contributing to the metabolism of nutrients in the intestinal mucosa before they reach the circulation (Husted et al., 2017). The colonic microbiome also acts as a

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

vital part of the digestive process by breaking down complex carbohydrates, proteins and fats, which are not broken down enzymatically in the preceding parts of the digestive system (Tremaroli and Bäckhed, 2012). The transit rate in the colon is much slower compared to the small intestine, allowing for increased microbial action on the food material (Roager et al., 2016). Indeed, microbial metabolism of digested dietary proteins results in the production of a range of nutrients and metabolites, which have diverse physiological functions (See below).

The microbiota composition plays an important role in metabolic health (Ley et al., 2006, Ley, 2010, Cunningham et al., 2021). Notably, the alpha and beta microbial diversity, measured by the richness of diversity and evenness, and relative differences in the overall diversity of taxa, respectively, highlight the similarities and differences in the microbiota across the different interventions, and associated metabolic states. A rich and diverse gut microbiota composition generally reflects a microbiota that is more resilient and capable of functioning better, with a loss in species diversity a common finding in several disease states (Manor et al., 2020). The importance of the gut microbiota in mediating protein effects was highlighted by recent work showing that WP reduced body weight gain in high fat fed mice and that this effect can be transferred via faecal matter onto mice fed casein (Nychyk et al., 2021, Boscaini et al., 2021, Boscaini, 2020). In contrast to animal studies (Wu et al., 2022), only few studies show an impact of dietary proteins on the gut microbiota in humans in the over-weight and obese categories (Table 2). Of note, subjects ingesting varied quantities of dietary fats, whilst co-ingesting proteins at 25% energy from various sources (red and white meat and plants), show no effect on the alpha or beta diversities (Lang et al., 2018). However, in the latter study, when the main effect of dietary fat on the gut microbiota was removed, an effect of dietary proteins can be seen on these micro-organisms, which were largely due to any source of protein rather than the quality of the protein consumed (Table 2). Similar data have been generated to show an impact of protein quantity on the composition of the gut microbiota (e.g., with or without fish intake or high and low gluten intake; Table 2). Where the impact of the source of proteins was investigated (pork versus chicken intake), the only changes in the gut microbiota was seen relative to baseline intake for each protein type (Dhakal et al., 2022)(Table 2). Imposing a calorie restriction for 8 weeks also did not affect the gut microbiota regardless of the hydrolysed state of the WP proteins (Beaumont et al., 2017b, Sun et al., 2022)(Table 2). In contrast to the above studies, which used 16S rRNA sequencing to uncover microbial changes, the study by Bel Lassen et al (2021) used Metagenomics sequencing to explore the impact of an extended calorie restriction (12 weeks)

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

on subjects consuming milk proteins supplemented with amino acids. The latter intervention was found to increase the microbial potential to produce amino acids compared to pea and casein intake (Table 2). This suggests that the interaction between the quality of the protein and the gut microbiota may be more subtle (at a functional level), requiring a greater depth of sequencing to uncover, but with the potential to influence the luminal pool of amino acids and their derivatives that are accessible by the host.

Effects on nutrients and metabolites

Most digested proteins are absorbed in the small intestine as amino acids, but some undigested proteins, especially following HPD intake, reach the colon where they are further broken down by proteolytic bacteria for the synthesis of other amino acids, and/or into amino acid derivatives that have been associated with numerous health outcomes including regulating digestion and absorption (Table 3). Of note, lysine, arginine, glycine and the branched chain amino acids (BCAAs), namely leucine, iso-leucine and valine are the most preferred amino acid (AA) substrates of colonic microbiota (Dai et al., 2011).

BCAAs and their derivatives

The BCAAs are building blocks of lean tissue, and are capable of modulating gene expression and signalling pathways, including regulating dietary nutrient absorption, partake in lipolysis, lipogenesis, glucose metabolism, and intestinal barrier function (Nishimura et al., 2010, Doi et al., 2003, Zhang et al., 2017). However, BCAAs have also been shown to have negative effects on metabolism with increased consumption of BCAAs been correlated with a more unhealthy metabolic state (Orozco-Ruiz et al., 2022), although these effects may be mediated somewhat by changing the levels of individual BCAAs (Yu et al., 2021). Increased BCAA intake has been shown to result in increased insulin resistance (Bishop et al., 2022, De Bandt et al., 2023) (Vanweert et al., 2022). Negative effects of BCAA intake may also include an increased risk of cancer (Rossi et al., 2022). Conversely, a reduction in BCAA intake has been shown to have positive effects on metabolic health (Cummings et al., 2018).

Intake of HPDs increased faecal levels of BCAAs, specifically following dietary casein and red and white meat intake (Table 4). In contrast, circulating levels of BCAA increased regardless of the type of protein consumed in both fasted and non-fasted states after prolong intake (3-4 weeks), as well as after acutely challenges, where the post-prandial

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

plasma increase was higher after milk protein consumption compared to plant protein intake (within 5h), WP intake compared to casein intake (3h) and following red meat intake compared to baseline measurements (within 4h) (Table 5). It is interesting that HPDs and BCAA should have both positive and negative outcomes on metabolic health. Whilst this suggests a potential functional relationship in the way dietary proteins affect metabolic health, it is important to highlight the role of the gut microbiota as a modulator of the effects. This is because these micro-organisms can convert BCAA into short chain fatty acids (SCFAs) and branched chain fatty acids (BCFAs) (Table 3), which have diverse metabolic health effects (discussed below). Indeed, and in agreement with the BCAA availability in the faeces and circulation, HPD intake also increases BCFA in faeces with some reaching the urine (Table 4 and 5). In contrast, the availability of SCFA in faeces and urine was either unaffected or decreased, with the exception of acetate, which was increased in faeces and urine following casein intake (Table 4 and 5 and further detailed below). The data suggest a potential microbial preference for conversion of amino acids into BCFA over SCFA in a background of HPD intake, which generally accompanies a low carbohydrate intake (Beaumont et al., 2017a, Sattari Najafabadi et al., 2019, Andriamihaja et al., 2010).

Aromatic amino acids (AAAs) and derived metabolites

Tryptophan: Evidence is emerging that dietary supply of tryptophan impacts on host metabolic health by directly or indirectly, the latter following microbial fermentation into numerous metabolites (Sridharan et al., 2014, Roth et al., 2021, Su et al., 2022). Indole, a tryptophan metabolite, acts as a signalling molecule capable of modulating the secretion of the satiety hormone, glucagon-like peptide (GLP)-1 from colonic enterendocrine L cells (Chimerel et al., 2014). Indole improves the intestinal epithelial barrier, upregulating genes responsible for tight junction organization, actin cytoskeleton, mucin production, and adherens junction, suggesting the strengthening and maintenance of the epithelial barrier, which directly affects intestinal permeability (Bansal et al., 2010). The tryptophan breakdown also produces indole-3-propionic acid (IPA), indole acetic acid (IAA) and kynurenine, which are also associated with several positive health outcomes (Table 3). Of note, like indole, IPA improves epithelial barrier function, and reduces inflammation and body weight (Hu et al., 2022). This molecule also improves insulin sensitivity, as does IAA (Hu et al., 2022). These effects are in part because of the indole moiety, which acts as ligands for aryl hydrocarbon receptor (Vyhlidalova et al., 2020), whereby receptor activation can suppress inflammatory

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

response and affect energy metabolism (Girer et al., 2020). Serotonin can be synthesized by the gut microbiota from tryptamine, a metabolite of tryptophan, and the latter amino acid and its derivative regulate the serotonin levels in the colon and blood (Wikoff et al., 2009, Roth et al., 2021). Tryptamine is also capable of inducing the release of serotonin from enteroendocrine cells, as well as potentiate the inhibitory response of cells to serotonin (Gao et al., 2018, Takaki et al., 1985). While there are many health benefits of breakdown of tryptophan, either in host cells and by microbial activity, the co-production of ammonia is a concern because of the damage caused to the mucosal layer in the colon, which impairs the absorptive capacity of the tissue (Yao et al., 2016).

In relation to protein source, intake of high quantities of proteins increased the faecal levels of indole-derivatives (milk proteins) and ammonia (all protein sources; Table 4). This raises the possibility that these dietary proteins increase the microbial activity related to metabolism of tryptophan in the gut. In support of this suggestion, intake of milk proteins supplemented with amino acids was found to increase the gut microbial potential to produce amino acids (Table 2). Beyond the gut, indole-derivatives have been found to increase in urine following dairy protein (casein) intake (Table 5), whilst other tryptophan metabolites, namely kynurenine and quinolinic acid show no consistency in terms of availability in faeces and circulation/urine based on the source of the protein consumed (Table 4 and 5). The presence of indole in faeces and circulation/urine following chronic intake of dairy proteins (>1 week; Table 4 and 5), is striking, and this contrasts with intake of non-dairy proteins, which only seem to increase indole levels in urine (by soy or meat/plant protein intake; Table 5). The difference may be related to the differential impact of dairy and plant proteins on the functional potential of the gut microbiota (mostly affected by dairy proteins; Table 2) combined with the host tissue- accessibility and metabolism of tryptophan that we know to be higher in quantity in milk proteins compared to plant proteins (Gorissen et al., 2018).

Tyrosine: Microbial metabolism of tyrosine can lead to the production of phenols, p-cresol-derivatives and tyramine (Table 3)(Oliphant and Allen-Vercoe, 2019). Tyramine is a neurotransmitter facilitating norepinephrine release, which is known to affect respiration and glucose levels in blood (Table 3). Both phenol and p-cresol are known to decrease integrity of the gut epithelium (Oliphant and Allen-Vercoe, 2019). Similar to tryptophan, the faecal availability of this AAA was not influenced by protein source, but the related metabolites,

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

phenol, p-cresol derivatives and tyramine were increased in faecal matter by dairy (phenol and p-cresol) and plant (p-cresol and tyramine) intake (Table 4). Data are limiting on the availability of tyrosine derived metabolites in circulation, except for the increased urinary levels of p-cresol detected following dairy (casein) protein intake (Table 5). The data suggest that the quality of protein associated with HPD, which can deliver high quantities of tryptophan and tyrosine, can provide beneficial effects (by producing indoles) as well potential harmful effects (by producing ammonia, phenol and p-cresols).

Non-essential amino acids: Dietary AAs are absorbed through the gut or act as substrates for microbial production of AAs, for their own utilisation and/or for supply to the host (Table 3). For instance, glutamine supplementation is found to impact the overall AA composition and content in the GI, including raising the concentration of Asp, Glu and Ala in the blood (Ma and Ma, 2019, Walker and van der Donk, 2016, Corpeleijn et al., 2010). Similarly, in the host, serine can be used to produce glycine or this process can be reversed (Wang et al., 2013). Glycine has many biological effects including being used for protein synthesis, bile acid metabolism, and hence contributing to digestion and absorption of dietary lipids and vitamins, as well as reducing body weight and fat and associated improvement in insulin sensitivity (Wang et al., 2013). Given the wide range of routes of amino acid synthesis (host tissue-metabolism and the gut microbiota), it is no surprise that intake of dietary proteins should cause an increase in the levels of tyrosine and glycine in circulation following chronic and acute challenges (all protein source; Table 5). Interestingly, whilst intake of red and white meat did not cause any changes in circulatory levels of these amino acids, it should be noted that the related data were generated from non-fasted state following 4 weeks of intervention (Connolly-Schoonen et al., 2023)(Table 5), contrasting with other studies showing a post-prandial increase in the AAs (4-5h) following an acute dietary protein challenge (Table 5), presumably reflecting a greater absorption in the small intestine.

Short chain fatty acids: There is a large body of evidence relating to the beneficial health impacts of SCFAs, namely acetate, butyrate and propionate, in particular in regulating energy metabolism, specifically in reducing hepatic glucose production and adiposity and stimulating the release of satiety related hormones such as peptide YY (Chambers et al., 2015, Hong et al., 2005, Frost et al., 2014, Donohoe et al., 2011). The SCFAs also partake in the maintenance of the gut, including improving the integrity of intestinal epithelial cells, promoting expression of tight junction associated proteins, cell proliferation and increasing

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

mucin production (Louis et al., 2007, Wang et al., 2019). These effects are depended upon the type of SCFA produced and how and where they act. Of note, SCFA are absorbed into the colonocytes or those that escape metabolism in cells, are transported into the liver via the portal system. It should be mentioned that only a minor fraction of SCFAs produced in the colon reach the circulatory system. Despite this, some contrasting responses of SCFAs need to be highlighted. Of note, acetate can be utilised for cholesterol synthesis, while propionate decreases the activity of the related pathway in the liver (Portune et al., 2016). The higher levels of SCFA also decrease the production of hydrogen sulphide, which is well-established to be detrimental to colonic health (Table 2), including as a contributing factor to ulcerative colitis (Teigen et al., 2019, Khalil et al., 2014) and as a potential trigger of colorectal cancer (Wolf et al., 2022). The effects of SCFA are mediated by G protein coupled receptors, namely GPR41, 43 and 109a, which are expressed in different tissues within the body (Layden et al., 2013). It should also be noted that some SCFAs have negative effects on health. Notably, propionate has been shown to increase liver lipogenesis (Gao et al., 2024). Additionally, acetate, propionate and butyrate have been shown to reduce gut dysbiosis-driven lung inflammation, as well as cause a pro-inflammatory response in human primary lung fibroblasts (Gao et al., 2024).

The SCFAs synthesised in the colon are mainly produced by the microbial fermentation of indigestible carbohydrates such as fibre, with increasing fibre intake increasing SCFA-producing bacteria and colonic SCFAs (Cani, 2014, Duncan et al., 2007, den Besten et al., 2015, Lu et al., 2016). However, AAs can also function as synthetic precursors of SCFAs in the colon (Portune et al., 2016), with the type and quantity of SCFA produced depending on the AA substrate available (Table 1), as well as microbiota present (Louis and Flint, 2017, Topping and Clifton, 2001, Blachier et al., 2007). Likely, as a result of the availability of AAs in the colon, HPD with low carbohydrates, have been shown to influence the production of SCFAs (Beaumont et al., 2017a, Sattari Najafabadi et al., 2019, Andriamihaja et al., 2010). Of note, proteins from dairy (casein), meat (red and white meat) and plant (soy) all decreased butyrate-producing microbiota, and further decreased butyrate levels in faeces (Table 4). In contrast, dairy (casein) proteins increased acetate levels in the faecal matter (Table 4) and also in urine (Table 5). Available evidence suggest that the source of protein influences the type of SCFAs produced in the gut, with some (acetate) reaching the urine, presumably via the circulation.

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

Branched Chain Fatty Acids (BCFAs): Further microbial fermentation of BCAAs results in the formation of branched SCFAs (BSCFAs) or BCFAs, including isovalerate and isobutyrate. The latter can also be produced from bacterial fermentation of some amino acids such as glycine (which can produce acetate), threonine (which can produce butyrate) or alanine (which can produce propionate) (Portune et al., 2016). The levels of BCFAs in the colon highlight proteolytic fermentation, as BCFAs are elevated when saccharolytic fermentation is minimal and protein fermentation is significantly enhanced in the colon (Diether and Willing, 2019, Yao et al., 2016). Similarly to SCFAs, BCFAs are shown to have positive impacts on metabolic health (Table 3), being associated with weight loss and maintenance (Taormina et al., 2020) as well as showing an inverse correlation with lipotoxicity and improved insulin sensitivity (Heimann et al., 2016, Taormina et al., 2020). Likely due to the availability of BCAAs in the colon (Table 4), HPDs increased the levels of BCFAs in the faecal matter and circulation including urine (Table 4 and 5). This effect was seen for proteins sourced from dairy, meat and plants, with the exception of gluten (Table 5). The largely similar effects of different proteins on the availability of BCFA both in faecal matter and in circulation suggest an important role for these metabolites in mediating the metabolic health effects of HPDs.

Exploring the potential mechanisms

All protein sources increased BCFAs in faecal content, probably from the increased gut availability of BCAA (Figure 2A), suggesting a greater bacterial conversion of BCAA to BCFA with intake of different proteins, albeit we cannot exclude the contribution of other amino acids for this process. Dairy protein intake specifically increased faecal levels of indole and acetate (Figure 2A). Alongside these health promoting metabolites, several other metabolites emerge in faecal matter with known unhealthy outcomes. These were phenol (dairy), p-cresol derivatives (dairy and plant), ammonia (all protein sources) and tyramine (plant). In circulation, and regardless of the source of proteins, amino acids including BCAA increased (Figure 2B). Additionally, for dairy proteins, the impact on the faecal availability of acetate, indole, p-cresol, BCFA and butyrate mirrored availability in the circulatory system or urine (Figure 2B), suggesting both gastro-intestinal and systemic effects of these metabolites. This contrasts with metabolites produced following plant and meat protein intake, which show fewer common responses in faecal matter and circulation (and urine)(Figure 2). The contrasting levels of metabolites in the gut and circulation/urine following dairy, meat or

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

plant protein intake could be related to the differences in the amino acid composition and the three-dimensional structure of the proteins accessible for enzymatic digestion and how the resulting digested peptides and amino acids are utilised by the dietary protein-sensitive gut microbiota to produce metabolites, which ultimately reach the gut and/or enter the circulatory system (Gorissen et al., 2018, Bilsborough and Mann, 2006, Agus et al., 2021, Cunningham et al., 2021).

In exploring the mechanisms for the physiological outcomes of HPD intake, the post-prandial increase in circulatory levels of AAs including BCAA is notable because their increase have been associated with increased satiety in human subjects, in particular following WP consumption compared to casein intake. The effect can be related to increased circulatory levels of satiety related hormones, namely cholecystokinin, (GLP)-1 and glucose-dependent insulinotropic polypeptide (Hall et al., 2003b). Whilst these data have emerged from acute challenges, the long term intake of HPD do not appear to cause changes in energy intake in humans, suggesting other mechanisms at play (Nilaweera and Cotter, 2023). In this regard, the increased availability of indole in the gut lumen by dairy protein intake is interesting because this metabolite and its derivatives are known to increase the release of GLP-1 from enteroendrocrine cells (Hu et al., 2022), and the activity of GLP-1 has been linked to roles beyond the reduction in food intake to include effects on adiposity (Nogueiras et al., 2009). In addition, and separate to effects on GLP-1, indoles and its derived have direct impact on adiposity, reducing adipogenesis and increasing thermogenesis (Hu et al., 2022), and their detection in urine following dairy (and plant) protein, presumably by the cross over from circulation, supports circulatory effects. Given that BCFAs have been associated with a reduction in body weight (Taormina et al., 2020), it is not surprising that these should also increase in circulation following HPD intake (Figure 2). The data suggest that indole and BCFA generated by HPD intake may contribute to the reduction in body weight either through effects on intake, energy expenditure and/or via direct effects on lipid metabolism. These metabolites could account, at least in part, for the greater impact of dairy proteins on metabolic health compared to other sources of proteins. In contrast to body weight and adiposity, the effect of HPD on insulin sensitivity is inconstantly reported (Santesso et al., 2012, Gannon and Nuttall, 2004, Clifton et al., 2008, Drummen et al., 2018). This may be due in part to increased circulatory levels of BCAA, and increased faecal and circulatory levels of p-cresol and its derivatives, which are known to reduce insulin sensitivity (Bishop et al., 2022, De Bandt et al., 2023), counterbalanced by the increased levels of acetate detected

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

mainly following dairy protein intake. Whilst HPD intake is known to reduce or cause no change in butyrate and propionate levels, the increased acetate level in faecal matter is significant because of important roles of these SCFA in energy balance regulation and insulin sensitivity (Morrison and Preston, 2016, Pham et al., 2024, Hernandez et al., 2019). Overall, it is clear that proteins from different sources produce common and distinct metabolites and their unique mechanisms of actions in the gut and/or via circulation presumably underlie the differences in physiological and metabolic outcomes of HPDs.

Future directions

There is limited data on the effect of high protein intake on the composition and functional potential of the gut microbiota. Further studies are also needed to ascertain how plant proteins other than soy and gluten influence the nutrients and metabolite profiles in the gut, given the increased focus on these proteins as a sustainable production source for human consumption (Langyan et al., 2021). Extending these lines of investigation, work is also needed to clarify the role of sex, since this parameter influences the protein quantity consumed, the composition and the functional potential of gut microbiota and physiological and metabolic parameters (Santos-Marcos et al., 2019, Bredella, 2017, Bennett et al., 2018). While the focus of our review was on human data, the cross species investigations can provide a greater understanding of the role played by nutrients and metabolites identified here as potential mediators of metabolic health effects of HPDs. These studies could involve transfer of faecal matter from humans to other species such as rodents within each sex and/or supplementation or depletion of the nutrients or metabolites in the diet to ascertaining their biological significance.

Conclusions

The amino acids derived from dietary proteins play important roles in physiological processors and in turn in metabolic health and, in some instances, in the pathophysiology of metabolic disorders. This functional relationship extends to include metabolites formed in the gut by the activity of the microbiota. A comparison of HPDs, which included the limited number of studies on plant proteins (soy and gluten), revealed similarities and differences in the metabolite profiles in faeces and circulation/urine, highlighting the contrasting gut versus circulatory effects of protein source within HPDs. This understanding will help to elucidate the complex mechanisms of action of HPDs and in turn, improve the efficacy of the interventions.

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

References

AGUS, A., CLEMENT, K. & SOKOL, H. 2021. Gut microbiota-derived metabolites as central regulators in metabolic disorders. *Gut,* 70**,** 1174-1182.

ALAMSHAH, A., SPRECKLEY, E., NORTON, M., KINSEY-JONES, J. S., AMIN, A., RAMGULAM, A., CAO, Y., JOHNSON, R., SALEH, K., AKALESTOU, E., MALIK, Z., GONZALEZ-ABUIN, N., JOMARD, A., AMARSI, R., MOOLLA, A., SARGENT, P. R., GRAY, G. W., BLOOM, S. R. & MURPHY, K. G. 2017. l-phenylalanine modulates gut hormone release and glucose tolerance, and suppresses food intake through the calcium-sensing receptor in rodents. *International Journal of Obesity,* 41**,** 1693-1701.

ANDRIAMIHAJA, M., DAVILA, A.-M., EKLOU-LAWSON, M., PETIT, N., DELPAL, S., ALLEK, F., BLAIS, A., DELTEIL, C., TOMÉ, D. & BLACHIER, F. 2010. Colon luminal content and epithelial cell morphology are markedly modified in rats fed with a high-protein diet. *American Journal of Physiology-Gastrointestinal and Liver Physiology,* 299**,** G1030-G1037.

ARCIERO, P. J., BAUR, D., CONNELLY, S. & ORMSBEE, M. J. 2014. Timed-daily ingestion of whey protein and exercise training reduces visceral adipose tissue mass and improves insulin resistance: the PRISE study. *J Appl Physiol (1985),* 117**,** 1-10.

AYKAN, N. F. 2015. Red Meat and Colorectal Cancer. *Oncol Rev,* 9**,** 288.

BAER, D. J., STOTE, K. S., PAUL, D. R., HARRIS, G. K., RUMPLER, W. V. & CLEVIDENCE, B. A. 2011. Whey protein but not soy protein supplementation alters body weight and composition in free-living overweight and obese adults. *J Nutr,* 141**,** 1489-94.

BANSAL, T., ALANIZ, R. C., WOOD, T. K. & JAYARAMAN, A. 2010. The bacterial signal indole increases epithelial-cell tight-junction resistance and attenuates indicators of inflammation. *Proc Natl Acad Sci U S A,* 107**,** 228-33.

BEAUMONT, M., PORTUNE, K. J., STEUER, N., LAN, A., CERRUDO, V., AUDEBERT, M., DUMONT, F., MANCANO, G., KHODOROVA, N., ANDRIAMIHAJA, M., AIRINEI, G., TOMÉ, D., BENAMOUZIG, R., DAVILA, A.-M., CLAUS, S. P., SANZ, Y. & BLACHIER, F. 2017a. Quantity and source of dietary protein influence metabolite production by gut microbiota and rectal mucosa gene expression: a randomized, parallel, double-blind trial in overweight humans. *The American Journal of Clinical Nutrition,* 106**,** 1005-1019.

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

BEAUMONT, M., PORTUNE, K. J., STEUER, N., LAN, A., CERRUDO, V., AUDEBERT, M., DUMONT, F., MANCANO, G., KHODOROVA, N., ANDRIAMIHAJA, M., AIRINEI, G., TOME, D., BENAMOUZIG, R., DAVILA, A. M., CLAUS, S. P., SANZ, Y. & BLACHIER, F. 2017b. Quantity and source of dietary protein influence metabolite production by gut microbiota and rectal mucosa gene expression: a randomized, parallel, double-blind trial in overweight humans. *Am J Clin Nutr,* 106**,** 1005-1019.

BEL LASSEN, P., ATTAYE, I., ADRIOUCH, S., NICOLAOU, M., ARON-WISNEWSKY, J., NIELSEN, T., CHAKAROUN, R., LE CHATELIER, E., FORSLUND, S., BELDA, E., BORK, P., BACKHED, F., STUMVOLL, M., PEDERSEN, O., HERREMA, H., GROEN, A. K., PINTO-SIETSMA, S. J., ZWINDERMAN, A. H., NIEUWDORP, M., CLEMENT, K. & ON BEHALF OF METACARDIS, C. 2021. Protein Intake, Metabolic Status and the Gut Microbiota in Different Ethnicities: Results from Two Independent Cohorts. *Nutrients,* 13.

BENNETT, E., PETERS, S. A. E. & WOODWARD, M. 2018. Sex differences in macronutrient intake and adherence to dietary recommendations: findings from the UK Biobank. *BMJ Open,* 8**,** e020017.

BILSBOROUGH, S. & MANN, N. 2006. A review of issues of dietary protein intake in humans. *Int J Sport Nutr Exerc Metab,* 16**,** 129-52.

BINGHAM, S. A., PIGNATELLI, B., POLLOCK, J. R., ELLUL, A., MALAVEILLE, C., GROSS, G., RUNSWICK, S., CUMMINGS, J. H. & O'NEILL, I. K. 1996. Does increased endogenous formation of N-nitroso compounds in the human colon explain the association between red meat and colon cancer? *Carcinogenesis,* 17**,** 515-23.

BISCHOFF, S. C., MAILER, R., PABST, O., WEIER, G., SEDLIK, W., LI, Z., CHEN, J. J., MURPHY, D. L. & GERSHON, M. D. 2009. Role of serotonin in intestinal inflammation: knockout of serotonin reuptake transporter exacerbates 2,4,6-trinitrobenzene sulfonic acid colitis in mice. *American Journal of Physiology-Gastrointestinal and Liver Physiology,* 296**,** G685-G695.

BISHOP, C. A., MACHATE, T., HENNING, T., HENKEL, J., PÜSCHEL, G., WEBER, D., GRUNE, T., KLAUS, S. & WEITKUNAT, K. 2022. Detrimental effects of branched-chain amino acids in glucose tolerance can be attributed to valine induced glucotoxicity in skeletal muscle. *Nutrition & Diabetes,* 12**,** 20.

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

BLACHIER, F., MARIOTTI, F., HUNEAU, J. F. & TOME, D. 2007. Effects of amino acid-derived luminal metabolites on the colonic epithelium and physiopathological consequences. *Amino Acids,* 33**,** 547-62.

BOSCAINI, S. 2020. *Novel insights into the mechanisms underlying the anti-obesity effects of whey protein.* Doctoral University College Cork.

BOSCAINI, S., CABRERA-RUBIO, R., GOLUBEVA, A., NYCHYK, O., FULLING, C., SPEAKMAN, J. R., COTTER, P. D., CRYAN, J. F. & NILAWEERA, K. N. 2021. Depletion of the gut microbiota differentially affects the impact of whey protein on high-fat diet-induced obesity and intestinal permeability. *Physiol Rep,* 9**,** e14867.

BRATLIE, M., HAGEN, I. V., HELLAND, A., ERCHINGER, F., MIDTTUN, O., UELAND, P. M., ROSENLUND, G., SVEIER, H., MELLGREN, G., HAUSKEN, T. & GUDBRANDSEN, O. A. 2021. Effects of high intake of cod or salmon on gut microbiota profile, faecal output and serum concentrations of lipids and bile acids in overweight adults: a randomised clinical trial. *Eur J Nutr,* 60**,** 2231-2248.

BREDELLA, M. A. 2017. Sex Differences in Body Composition. *Adv Exp Med Biol,* 1043**,** 9-27.

CANI, P. D. 2014. Metabolism in 2013: The gut microbiota manages host metabolism. *Nat Rev Endocrinol,* 10**,** 74-6.

CHAMBERS, E. S., VIARDOT, A., PSICHAS, A., MORRISON, D. J., MURPHY, K. G., ZAC-VARGHESE, S. E. K., MACDOUGALL, K., PRESTON, T., TEDFORD, C., FINLAYSON, G. S., BLUNDELL, J. E., BELL, J. D., THOMAS, E. L., MT-ISA, S., ASHBY, D., GIBSON, G. R., KOLIDA, S., DHILLO, W. S., BLOOM, S. R., MORLEY, W., CLEGG, S. & FROST, G. 2015. Effects of targeted delivery of propionate to the human colon on appetite regulation, body weight maintenance and adiposity in overweight adults. *Gut,* 64**,** 1744.

CHANDEL, N. S. 2021. Amino Acid Metabolism. *Cold Spring Harb Perspect Biol,* 13.

CHENG, Y., JIN, U. H., ALLRED, C. D., JAYARAMAN, A., CHAPKIN, R. S. & SAFE, S. 2015. Aryl Hydrocarbon Receptor Activity of Tryptophan Metabolites in Young Adult Mouse Colonocytes. *Drug Metab Dispos,* 43**,** 1536-43.

CHIMEREL, C., EMERY, E., SUMMERS, D. K., KEYSER, U., GRIBBLE, F. M. & REIMANN, F. 2014. Bacterial metabolite indole modulates incretin secretion from intestinal enteroendocrine L cells. *Cell Rep,* 9**,** 1202-8.

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

CHIU, S., WILLIAMS, P. T., DAWSON, T., BERGMAN, R. N., STEFANOVSKI, D., WATKINS, S. M. & KRAUSS, R. M. 2014. Diets high in protein or saturated fat do not affect insulin sensitivity or plasma concentrations of lipids and lipoproteins in overweight and obese adults. *J Nutr,* 144**,** 1753-9.

CLARKE, S. F., MURPHY, E. F., NILAWEERA, K., ROSS, P. R., SHANAHAN, F., O'TOOLE, P. W. & COTTER, P. D. 2012. The gut microbiota and its relationship to diet and obesity: new insights. *Gut Microbes,* 3**,** 186-202.

CLIFTON, P. M., KEOGH, J. B. & NOAKES, M. 2008. Long-term effects of a high-protein weight-loss diet. *The American journal of clinical nutrition,* 87**,** 23-9.

COKER, R. H., MILLER, S., SCHUTZLER, S., DEUTZ, N. & WOLFE, R. R. 2012. Whey protein and essential amino acids promote the reduction of adipose tissue and increased muscle protein synthesis during caloric restriction-induced weight loss in elderly, obese individuals. *Nutr J,* 11**,** 105.

CONNOLLY-SCHOONEN, J., DANOWSKI, L., BISTRICER, M., CAMPO CATALAN, L., AILAWADI, S., SICINSKI, E. M., SCHOONEN, M., INGRAM, B. & MONTROSE, D. C. 2023. A Pilot Controlled Feeding Trial Modifying Protein Intake in Healthy Subjects to Assess Adherence and the Metabolome. *Nutr Cancer,* 75**,** 1499-1510.

CORPELEIJN, W. E., RIEDIJK, M. A., ZHOU, Y., SCHIERBEEK, H., HUANG, Y., CHEN, C. & VAN GOUDOEVER, J. B. 2010. Almost all enteral aspartate is taken up in first-pass metabolism in enterally fed preterm infants. *Clinical Nutrition,* 29**,** 341-346.

COVASA, M., STEPHENS, R. W., TODEREAN, R. & COBUZ, C. 2019. Intestinal Sensing by Gut Microbiota: Targeting Gut Peptides. *Front Endocrinol (Lausanne),* 10**,** 82.

CRUZAT, V., MACEDO ROGERO, M., NOEL KEANE, K., CURI, R. & NEWSHOLME, P. 2018. Glutamine: Metabolism and Immune Function, Supplementation and Clinical Translation. *Nutrients,* 10.

CUMMINGS, N. E., WILLIAMS, E. M., KASZA, I., KONON, E. N., SCHAID, M. D., SCHMIDT, B. A., POUDEL, C., SHERMAN, D. S., YU, D., ARRIOLA APELO, S. I., COTTRELL, S. E., GEIGER, G., BARNES, M. E., WISINSKI, J. A., FENSKE, R. J., MATKOWSKYJ, K. A., KIMPLE, M. E., ALEXANDER, C. M., MERRINS, M. J. & LAMMING, D. W. 2018. Restoration of metabolic health by decreased consumption of branched-chain amino acids. *J Physiol,* 596**,** 623-645.

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

CUNNINGHAM, A. L., STEPHENS, J. W. & HARRIS, D. A. 2021. A review on gut microbiota: a central factor in the pathophysiology of obesity. *Lipids Health Dis,* 20**,** 65.

CUOMO, P., CAPPARELLI, R., IANNELLI, A. & IANNELLI, D. 2022. Role of Branched-Chain Amino Acid Metabolism in Type 2 Diabetes, Obesity, Cardiovascular Disease and Non-Alcoholic Fatty Liver Disease. *Int J Mol Sci,* 23.

DAHL, W. J., HUNG, W. L., FORD, A. L., SUH, J. H., AUGER, J., NAGULESAPILLAI, V. & WANG, Y. 2020. In older women, a high-protein diet including animal-sourced foods did not impact serum levels and urinary excretion of trimethylamine-N-oxide. *Nutr Res,* 78**,** 72-81.

DAI, Z. L., WU, G. & ZHU, W. Y. 2011. Amino acid metabolism in intestinal bacteria: links between gut ecology and host health. *Front Biosci (Landmark Ed),* 16**,** 1768-86.

DAVIE, J. R. 2003. Inhibition of histone deacetylase activity by butyrate. *J Nutr,* 133**,** 2485S-2493S.

DE BANDT, J.-P., COUMOUL, X. & BAROUKI, R. 2023. Branched-Chain Amino Acids and Insulin Resistance, from Protein Supply to Diet-Induced Obesity. *Nutrients,* 15**,** 68.

DEHHAGHI, M., KAZEMI SHARIAT PANAHI, H. & GUILLEMIN, G. J. 2019. Microorganisms, Tryptophan Metabolism, and Kynurenine Pathway: A Complex Interconnected Loop Influencing Human Health Status. *Int J Tryptophan Res,* 12**,** 1178646919852996.

DEN BESTEN, G., BLEEKER, A., GERDING, A., VAN EUNEN, K., HAVINGA, R., VAN DIJK, T. H., OOSTERVEER, M. H., JONKER, J. W., GROEN, A. K., REIJNGOUD, D. J. & BAKKER, B. M. 2015. Short-Chain Fatty Acids Protect Against High-Fat Diet-Induced Obesity via a PPARgamma-Dependent Switch From Lipogenesis to Fat Oxidation. *Diabetes,* 64**,** 2398-408.

DHAKAL, S., MOAZZAMI, Z., PERRY, C. & DEY, M. 2022. Effects of Lean Pork on Microbiota and Microbial-Metabolite Trimethylamine-N-Oxide: A Randomized Controlled Non-Inferiority Feeding Trial Based on the Dietary Guidelines for Americans. *Mol Nutr Food Res,* 66**,** e2101136.

DIETHER, N. E. & WILLING, B. P. 2019. Microbial Fermentation of Dietary Protein: An Important Factor in Diet(-)Microbe(-)Host Interaction. *Microorganisms,* 7.

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

DOI, M., YAMAOKA, I., FUKUNAGA, T. & NAKAYAMA, M. 2003. Isoleucine, a potent plasma glucose-lowering amino acid, stimulates glucose uptake in C2C12 myotubes. *Biochem Biophys Res Commun,* 312**,** 1111-7.

DONOHOE, D. R., GARGE, N., ZHANG, X., SUN, W., O'CONNELL, T. M., BUNGER, M. K. & BULTMAN, S. J. 2011. The microbiome and butyrate regulate energy metabolism and autophagy in the mammalian colon. *Cell Metab,* 13**,** 517-26.

DRUMMEN, M., TISCHMANN, L., GATTA-CHERIFI, B., ADAM, T. & WESTERTERP-PLANTENGA, M. 2018. Dietary Protein and Energy Balance in Relation to Obesity and Co-morbidities. *Front Endocrinol (Lausanne),* 9**,** 443.

DUNCAN, S. H., BELENGUER, A., HOLTROP, G., JOHNSTONE, A. M., FLINT, H. J. & LOBLEY, G. E. 2007. Reduced dietary intake of carbohydrates by obese subjects results in decreased concentrations of butyrate and butyrate-producing bacteria in feces. *Appl Environ Microbiol,* 73**,** 1073-8.

ECKBURG, P. B., BIK, E. M., BERNSTEIN, C. N., PURDOM, E., DETHLEFSEN, L., SARGENT, M., GILL, S. R., NELSON, K. E. & RELMAN, D. A. 2005. Diversity of the human intestinal microbial flora. *Science,* 308**,** 1635-8.

FROST, G., SLEETH, M. L., SAHURI-ARISOYLU, M., LIZARBE, B., CERDAN, S., BRODY, L., ANASTASOVSKA, J., GHOURAB, S., HANKIR, M., ZHANG, S., CARLING, D., SWANN, J. R., GIBSON, G., VIARDOT, A., MORRISON, D., LOUISE THOMAS, E. & BELL, J. D. 2014. The short-chain fatty acid acetate reduces appetite via a central homeostatic mechanism. *Nature Communications,* 5**,** 3611.

GANNON, M. C. & NUTTALL, F. Q. 2004. Effect of a high-protein, low-carbohydrate diet on blood glucose control in people with type 2 diabetes. *Diabetes,* 53**,** 2375-82.

GAO, J., XU, K., LIU, H., LIU, G., BAI, M., PENG, C., LI, T. & YIN, Y. 2018. Impact of the Gut Microbiota on Intestinal Immunity Mediated by Tryptophan Metabolism. *Frontiers in Cellular and Infection Microbiology,* 8.

GAO, Y., YAO, Q., MENG, L., WANG, J. & ZHENG, N. 2024. Double-side role of short chain fatty acids on host health via the gut-organ axes. *Anim Nutr,* 18**,** 322-339.

GEYPENS, B., CLAUS, D., EVENEPOEL, P., HIELE, M., MAES, B., PEETERS, M., RUTGEERTS, P. & GHOOS, Y. 1997. Influence of dietary protein supplements on the formation of bacterial metabolites in the colon. *Gut,* 41**,** 70-6.

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

GIGLIO, B. M., SCHINCAGLIA, R. M., DA SILVA, A. S., FAZANI, I. C. S., MONTEIRO, P. A., MOTA, J. F., CUNHA, J. P., PICHARD, C. & PIMENTEL, G. D. 2019. Whey Protein Supplementation Compared to Collagen Increases Blood Nesfatin Concentrations and Decreases Android Fat in Overweight Women: A Randomized Double-Blind Study. *Nutrients,* 11.

GIRER, N. G., TOMLINSON, C. R. & ELFERINK, C. J. 2020. The Aryl Hydrocarbon Receptor in Energy Balance: The Road from Dioxin-Induced Wasting Syndrome to Combating Obesity with Ahr Ligands. *Int J Mol Sci,* 22.

GORISSEN, S. H. M., CROMBAG, J. J. R., SENDEN, J. M. G., WATERVAL, W. A. H., BIERAU, J., VERDIJK, L. B. & VAN LOON, L. J. C. 2018. Protein content and amino acid composition of commercially available plant-based protein isolates. *Amino Acids,* 50**,** 1685-1695.

GRATZ, S. W., HAZIM, S., RICHARDSON, A. J., SCOBBIE, L., JOHNSTONE, A. M., FYFE, C., HOLTROP, G., LOBLEY, G. E. & RUSSELL, W. R. 2019. Dietary carbohydrate rather than protein intake drives colonic microbial fermentation during weight loss. *Eur J Nutr,* 58**,** 1147-1158.

HALL, W. L., MILLWARD, D. J., LONG, S. J. & MORGAN, L. M. 2003a. Casein and whey exert different effects on plasma amino acid profiles, gastrointestinal hormone secretion and appetite. *The British journal of nutrition,* 89**,** 239-48.

HALL, W. L., MILLWARD, D. J., LONG, S. J. & MORGAN, L. M. 2003b. Casein and whey exert different effects on plasma amino acid profiles, gastrointestinal hormone secretion and appetite. *Br J Nutr.,* 89**,** 239-48.

HANSEN, L. B. S., ROAGER, H. M., SONDERTOFT, N. B., GOBEL, R. J., KRISTENSEN, M., VALLES-COLOMER, M., VIEIRA-SILVA, S., IBRUGGER, S., LIND, M. V., MAERKEDAHL, R. B., BAHL, M. I., MADSEN, M. L., HAVELUND, J., FALONY, G., TETENS, I., NIELSEN, T., ALLIN, K. H., FRANDSEN, H. L., HARTMANN, B., HOLST, J. J., SPARHOLT, M. H., HOLCK, J., BLENNOW, A., MOLL, J. M., MEYER, A. S., HOPPE, C., POULSEN, J. H., CARVALHO, V., SAGNELLI, D., DALGAARD, M. D., CHRISTENSEN, A. F., LYDOLPH, M. C., ROSS, A. B., VILLAS-BOAS, S., BRIX, S., SICHERITZ-PONTEN, T., BUSCHARD, K., LINNEBERG, A., RUMESSEN, J. J., EKSTROM, C. T., RITZ, C., KRISTIANSEN, K., NIELSEN, H. B., VESTERGAARD, H., FAERGEMAN, N. J., RAES, J., FROKIAER, H., HANSEN, T., LAURITZEN, L.,

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

GUPTA, R., LICHT, T. R. & PEDERSEN, O. 2018. A low-gluten diet induces changes in the intestinal microbiome of healthy Danish adults. *Nat Commun,* 9**,** 4630.

HECTOR, A. J., MARCOTTE, G. R., CHURCHWARD-VENNE, T. A., MURPHY, C. H., BREEN, L., VON ALLMEN, M., BAKER, S. K. & PHILLIPS, S. M. 2015. Whey protein supplementation preserves postprandial myofibrillar protein synthesis during short-term energy restriction in overweight and obese adults. *J Nutr,* 145**,** 246-52.

HEIMANN, E., NYMAN, M., PALBRINK, A. K., LINDKVIST-PETERSSON, K. & DEGERMAN, E. 2016. Branched short-chain fatty acids modulate glucose and lipid metabolism in primary adipocytes. *Adipocyte,* 5**,** 359-368.

HERNANDEZ, M. A. G., CANFORA, E. E., JOCKEN, J. W. E. & BLAAK, E. E. 2019. The Short-Chain Fatty Acid Acetate in Body Weight Control and Insulin Sensitivity. *Nutrients,* 11.

HONG, Y.-H., NISHIMURA, Y., HISHIKAWA, D., TSUZUKI, H., MIYAHARA, H., GOTOH, C., CHOI, K.-C., FENG, D. D., CHEN, C., LEE, H.-G., KATOH, K., ROH, S.-G. & SASAKI, S. 2005. Acetate and Propionate Short Chain Fatty Acids Stimulate Adipogenesis via GPCR43. *Endocrinology,* 146**,** 5092-5099.

HU, S., WANG, L., YANG, D., LI, L., TOGO, J., WU, Y., LIU, Q., LI, B., LI, M., WANG, G., ZHANG, X., NIU, C., LI, J., XU, Y., COUPER, E., WHITTINGTON-DAVIES, A., MAZIDI, M., LUO, L., WANG, S., DOUGLAS, A. & SPEAKMAN, J. R. 2018. Dietary Fat, but Not Protein or Carbohydrate, Regulates Energy Intake and Causes Adiposity in Mice. *Cell Metab,* 28**,** 415-431 e4.

HU, W., YAN, G., DING, Q., CAI, J., ZHANG, Z., ZHAO, Z., LEI, H. & ZHU, Y. Z. 2022. Update of Indoles: Promising molecules for ameliorating metabolic diseases. *Biomed Pharmacother,* 150**,** 112957.

HUSTED, A. S., TRAUELSEN, M., RUDENKO, O., HJORTH, S. A. & SCHWARTZ, T. W. 2017. GPCR-Mediated Signaling of Metabolites. *Cell Metab,* 25**,** 777-796.

JENNIS, M., CAVANAUGH, C. R., LEO, G. C., MABUS, J. R., LENHARD, J. & HORNBY, P. J. 2018. Microbiota-derived tryptophan indoles increase after gastric bypass surgery and reduce intestinal permeability in vitro and in vivo. *Neurogastroenterol Motil,* 30.

KHALIL, N. A., WALTON, G. E., GIBSON, G. R., TUOHY, K. M. & ANDREWS, S. C. 2014. In vitro batch cultures of gut microbiota from healthy and ulcerative colitis

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

(UC) subjects suggest that sulphate-reducing bacteria levels are raised in UC and by a protein-rich diet. *Int J Food Sci Nutr,* 65**,** 79-88.

KJOLBAEK, L., SORENSEN, L. B., SONDERTOFT, N. B., RASMUSSEN, C. K., LORENZEN, J. K., SERENA, A., ASTRUP, A. & LARSEN, L. H. 2017. Protein supplements after weight loss do not improve weight maintenance compared with recommended dietary protein intake despite beneficial effects on appetite sensation and energy expenditure: a randomized, controlled, double-blinded trial. *Am J Clin Nutr,* 106**,** 684-697.

LANG, J. M., PAN, C., CANTOR, R. M., TANG, W. H. W., GARCIA-GARCIA, J. C., KURTZ, I., HAZEN, S. L., BERGERON, N., KRAUSS, R. M. & LUSIS, A. J. 2018. Impact of Individual Traits, Saturated Fat, and Protein Source on the Gut Microbiome. *mBio,* 9.

LANGYAN, S., YADAVA, P., KHAN, F. N., DAR, Z. A., SINGH, R. & KUMAR, A. 2021. Sustaining Protein Nutrition Through Plant-Based Foods. *Front Nutr,* 8**,** 772573.

LAYDEN, B. T., ANGUEIRA, A. R., BRODSKY, M., DURAI, V. & LOWE, W. L., JR. 2013. Short chain fatty acids and their receptors: new metabolic targets. *Transl Res,* 161**,** 131-40.

LE, T. H., DISEGNA, M. & LLOYD, T. 2020. National Food Consumption Patterns: Converging Trends and the Implications for Health. *EuroChoices,* n/a.

LEE, J. H., DUSTER, M., ROBERTS, T. & DEVINSKY, O. 2021. United States Dietary Trends Since 1800: Lack of Association Between Saturated Fatty Acid Consumption and Non-communicable Diseases. *Front Nutr,* 8**,** 748847.

LEW, Q. J., JAFAR, T. H., KOH, H. W., JIN, A., CHOW, K. Y., YUAN, J. M. & KOH, W. P. 2017. Red Meat Intake and Risk of ESRD. *J Am Soc Nephrol,* 28**,** 304-312.

LEY, R. E. 2010. Obesity and the human microbiome. *Curr Opin Gastroenterol,* 26**,** 5-11.

LEY, R. E., PETERSON, D. A. & GORDON, J. I. 2006. Ecological and evolutionary forces shaping microbial diversity in the human intestine. *Cell,* 124**,** 837-48.

LOUIS, P. & FLINT, H. J. 2017. Formation of propionate and butyrate by the human colonic microbiota. *Environ Microbiol,* 19**,** 29-41.

LOUIS, P., SCOTT, K. P., DUNCAN, S. H. & FLINT, H. J. 2007. Understanding the effects of diet on bacterial metabolism in the large intestine. *J Appl Microbiol,* 102**,** 1197-208.

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

LU, Y., FAN, C., LI, P., LU, Y., CHANG, X. & QI, K. 2016. Short Chain Fatty Acids Prevent High-fat-diet-induced Obesity in Mice by Regulating G Protein-coupled Receptors and Gut Microbiota. *Scientific Reports,* 6**,** 37589.

MA, N. & MA, X. 2019. Dietary Amino Acids and the Gut-Microbiome-Immune Axis: Physiological Metabolism and Therapeutic Prospects. *Compr Rev Food Sci Food Saf,* 18**,** 221-242.

MA, Z. F. & LEE, Y. Y. 2020. Chapter 7 - Small intestine anatomy and physiology. *In:* RAO, S. S. C., LEE, Y. Y. & GHOSHAL, U. C. (eds.) *Clinical and Basic Neurogastroenterology and Motility.* Academic Press.

MANOR, O., DAI, C. L., KORNILOV, S. A., SMITH, B., PRICE, N. D., LOVEJOY, J. C., GIBBONS, S. M. & MAGIS, A. T. 2020. Health and disease markers correlate with gut microbiome composition across thousands of people. *Nat Commun,* 11**,** 5206.

MARTINEZ-GURYN, K., HUBERT, N., FRAZIER, K., URLASS, S., MUSCH, M. W., OJEDA, P., PIERRE, J. F., MIYOSHI, J., SONTAG, T. J., CHAM, C. M., REARDON, C. A., LEONE, V. & CHANG, E. B. 2018. Small Intestine Microbiota Regulate Host Digestive and Absorptive Adaptive Responses to Dietary Lipids. *Cell Host & Microbe,* 23**,** 458-469.e5.

MOLLER, G., ANDERSEN, J. R., JALO, E., RITZ, C., BRAND-MILLER, J., LARSEN, T. M., SILVESTRE, M. P., FOGELHOLM, M., POPPITT, S. D., RABEN, A. & DRAGSTED, L. O. 2020. The association of dietary animal and plant protein with putative risk markers of colorectal cancer in overweight pre-diabetic individuals during a weight-reducing programme: a PREVIEW sub-study. *Eur J Nutr,* 59**,** 1517-1527.

MOON, J. & KOH, G. 2020. Clinical Evidence and Mechanisms of High-Protein Diet-Induced Weight Loss. *J Obes Metab Syndr,* 29**,** 166-173.

MORRISON, D. J. & PRESTON, T. 2016. Formation of short chain fatty acids by the gut microbiota and their impact on human metabolism. *Gut Microbes,* 7**,** 189-200.

MORSELLI, E., CRIOLLO, A., RODRIGUEZ-NAVAS, C. & CLEGG, D. J. 2014. Chronic High Fat Diet Consumption Impairs Metabolic Health of Male Mice. *Inflamm Cell Signal,* 1**,** e561.

MUTCH, D. M., FUHRMANN, J. C., REIN, D., WIEMER, J. C., BOUILLOT, J. L., POITOU, C. & CLEMENT, K. 2009. Metabolite profiling identifies candidate

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

markers reflecting the clinical adaptations associated with Roux-en-Y gastric bypass surgery. *PLoS One,* 4**,** e7905.

NILAWEERA, K. N. & COTTER, P. D. 2023. Can dietary proteins selectively reduce either the visceral or subcutaneous adipose tissues? *Obes Rev,* 24**,** e13613.

NISHIMURA, J., MASAKI, T., ARAKAWA, M., SEIKE, M. & YOSHIMATSU, H. 2010. Isoleucine prevents the accumulation of tissue triglycerides and upregulates the expression of PPARalpha and uncoupling protein in diet-induced obese mice. *J Nutr,* 140**,** 496-500.

NOGUEIRAS, R., PEREZ-TILVE, D., VEYRAT-DUREBEX, C., MORGAN, D. A., VARELA, L., HAYNES, W. G., PATTERSON, J. T., DISSE, E., PFLUGER, P. T., LOPEZ, M., WOODS, S. C., DIMARCHI, R., DIEGUEZ, C., RAHMOUNI, K., ROHNER-JEANRENAUD, F. & TSCHOP, M. H. 2009. Direct control of peripheral lipid deposition by CNS GLP-1 receptor signaling is mediated by the sympathetic nervous system and blunted in diet-induced obesity. *The Journal of neuroscience* 29**,** 5916-25.

NYCHYK, O., BARTON, W., RUDOLF, A. M., BOSCAINI, S., WALSH, A., BASTIAANSSEN, T. F. S., GIBLIN, L., CORMICAN, P., CHEN, L., PIOTROWICZ, Y., DEROUS, D., FANNING, A., YIN, X., GRANT, J., MELGAR, S., BRENNAN, L., MITCHELL, S. E., CRYAN, J. F., WANG, J., COTTER, P. D., SPEAKMAN, J. R. & NILAWEERA, K. N. 2021. Protein quality and quantity influence the effect of dietary fat on weight gain and tissue partitioning via host-microbiota changes. *Cell Rep,* 35**,** 109093.

OLIPHANT, K. & ALLEN-VERCOE, E. 2019. Macronutrient metabolism by the human gut microbiome: major fermentation by-products and their impact on host health. *Microbiome,* 7**,** 91.

OROZCO-RUIZ, X., ANESI, A., MATTIVI, F. & BRETELER, M. M. B. 2022. Branched-Chain and Aromatic Amino Acids Related to Visceral Adipose Tissue Impact Metabolic Health Risk Markers. *J Clin Endocrinol Metab,* 107**,** e2896-e2905.

PAL, S. & ELLIS, V. 2010. The chronic effects of whey proteins on blood pressure, vascular function, and inflammatory markers in overweight individuals. *Obesity,* 18**,** 1354-9.

PARADA VENEGAS, D., DE LA FUENTE, M. K., LANDSKRON, G., GONZALEZ, M. J., QUERA, R., DIJKSTRA, G., HARMSEN, H. J. M., FABER, K. N. & HERMOSO, M. A. 2019. Short Chain Fatty Acids (SCFAs)-Mediated Gut Epithelial

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

and Immune Regulation and Its Relevance for Inflammatory Bowel Diseases. *Front Immunol,* 10**,** 277.

PESTA, D. H. & SAMUEL, V. T. 2014. A high-protein diet for reducing body fat: mechanisms and possible caveats. *Nutr Metab (Lond),* 11**,** 53.

PHAM, N. H. T., JOGLEKAR, M. V., WONG, W. K. M., NASSIF, N. T., SIMPSON, A. M. & HARDIKAR, A. A. 2024. Short-chain fatty acids and insulin sensitivity: a systematic review and meta-analysis. *Nutr Rev,* 82**,** 193-209.

PICCOLO, B. D., COMERFORD, K. B., KARAKAS, S. E., KNOTTS, T. A., FIEHN, O. & ADAMS, S. H. 2015. Whey protein supplementation does not alter plasma branched-chained amino acid profiles but results in unique metabolomics patterns in obese women enrolled in an 8-week weight loss trial. *J Nutr,* 145**,** 691-700.

PINCKAERS, P. J. M., KOUW, I. W. K., GORISSEN, S. H. M., HOUBEN, L. H. P., SENDEN, J. M., WODZIG, W., DE GROOT, L., VERDIJK, L. B., SNIJDERS, T. & VAN LOON, L. J. C. 2023. The Muscle Protein Synthetic Response to the Ingestion of a Plant-Derived Protein Blend Does Not Differ from an Equivalent Amount of Milk Protein in Healthy Young Males. *J Nutr,* 152**,** 2734-2743.

PORTUNE, K. J., ANNE-MARIE, D., DANIEL, T., FRANÇOIS, B., MARTIN, B. & YOLANDA, S. 2016. Gut microbiota role in dietary protein metabolism and health-related outcomes: The two sides of the coin. *Trends in food science & technology.,* 57**,** 213-232.

PRODHAN, U. K., PUNDIR, S., CHIANG, V. S., MILAN, A. M., BARNETT, M. P. G., SMITH, G. C., MARKWORTH, J. F., KNOWLES, S. O. & CAMERON-SMITH, D. 2020. Comparable Postprandial Amino Acid and Gastrointestinal Hormone Responses to Beef Steak Cooked Using Different Methods: A Randomised Crossover Trial. *Nutrients,* 12.

RAJILIĆ-STOJANOVIĆ, M. & DE VOS, W. M. 2014. The first 1000 cultured species of the human gastrointestinal microbiota. *FEMS microbiology reviews,* 38**,** 996-1047.

ROAGER, H. M., HANSEN, L. B., BAHL, M. I., FRANDSEN, H. L., CARVALHO, V., GOBEL, R. J., DALGAARD, M. D., PLICHTA, D. R., SPARHOLT, M. H., VESTERGAARD, H., HANSEN, T., SICHERITZ-PONTEN, T., NIELSEN, H. B., PEDERSEN, O., LAURITZEN, L., KRISTENSEN, M., GUPTA, R. & LICHT, T. R. 2016. Colonic transit time is related to bacterial metabolism and mucosal turnover in the gut. *Nat Microbiol,* 1**,** 16093.

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

ROSSI, M., TURATI, F., STRIKOUDI, P., FERRARONI, M., PARPINEL, M., SERRAINO, D., NEGRI, E. & LA VECCHIA, C. 2022. Dietary intake of branched-chain amino acids and pancreatic cancer risk in a case-control study from Italy. *Br J Nutr*, 1-19.

ROTH, W., ZADEH, K., VEKARIYA, R., GE, Y. & MOHAMADZADEH, M. 2021. Tryptophan Metabolism and Gut-Brain Homeostasis. *Int J Mol Sci*, 22.

RUSSELL, W. R., GRATZ, S. W., DUNCAN, S. H., HOLTROP, G., INCE, J., SCOBBIE, L., DUNCAN, G., JOHNSTONE, A. M., LOBLEY, G. E., WALLACE, R. J., DUTHIE, G. G. & FLINT, H. J. 2011. High-protein, reduced-carbohydrate weight-loss diets promote metabolite profiles likely to be detrimental to colonic health. *The American Journal of Clinical Nutrition*, 93**,** 1062-1072.

SACKS, F. M., BRAY, G. A., CAREY, V. J., SMITH, S. R., RYAN, D. H., ANTON, S. D., MCMANUS, K., CHAMPAGNE, C. M., BISHOP, L. M., LARANJO, N., LEBOFF, M. S., ROOD, J. C., DE JONGE, L., GREENWAY, F. L., LORIA, C. M., OBARZANEK, E. & WILLIAMSON, D. A. 2009. Comparison of weight-loss diets with different compositions of fat, protein, and carbohydrates. *N Engl J Med*, 360**,** 859-73.

SALVI, P. S. & COWLES, R. A. 2021. Butyrate and the Intestinal Epithelium: Modulation of Proliferation and Inflammation in Homeostasis and Disease. *Cells*, 10.

SANTESSO, N., AKL, E. A., BIANCHI, M., MENTE, A., MUSTAFA, R., HEELS-ANSDELL, D. & SCHUNEMANN, H. J. 2012. Effects of higher- versus lower-protein diets on health outcomes: a systematic review and meta-analysis. *Eur J Clin Nutr*, 66**,** 780-8.

SANTOS-MARCOS, J. A., HARO, C., VEGA-ROJAS, A., ALCALA-DIAZ, J. F., MOLINA-ABRIL, H., LEON-ACUNA, A., LOPEZ-MORENO, J., LANDA, B. B., TENA-SEMPERE, M., PEREZ-MARTINEZ, P., LOPEZ-MIRANDA, J., PEREZ-JIMENEZ, F. & CAMARGO, A. 2019. Sex Differences in the Gut Microbiota as Potential Determinants of Gender Predisposition to Disease. *Mol Nutr Food Res*, 63**,** e1800870.

SAPS, M. & MIRANDA, A. 2017. Gastrointestinal Pharmacology. *Handb Exp Pharmacol*, 239**,** 147-176.

SATTARI NAJAFABADI, Z., SKAU NIELSEN, T. & SKOU HEDEMANN, M. 2019. Dietary protein source and butyrylated high-amylose maize starch included in a high-

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

protein diet determines the urinary metabolome of rats. *Int J Food Sci Nutr,* 70**,** 255-266.

SITES, C. K., COOPER, B. C., TOTH, M. J., GASTALDELLI, A., ARABSHAHI, A. & BARNES, S. 2007. Effect of a daily supplement of soy protein on body composition and insulin secretion in postmenopausal women. *Fertil Steril,* 88**,** 1609-17.

SRIDHARAN, G. V., CHOI, K., KLEMASHEVICH, C., WU, C., PRABAKARAN, D., PAN, L. B., STEINMEYER, S., MUELLER, C., YOUSOFSHAHI, M., ALANIZ, R. C., LEE, K. & JAYARAMAN, A. 2014. Prediction and quantification of bioactive microbiota metabolites in the mouse gut. *Nat Commun,* 5**,** 5492.

SU, X., GAO, Y. & YANG, R. 2022. Gut Microbiota-Derived Tryptophan Metabolites Maintain Gut and Systemic Homeostasis. *Cells,* 11.

SUN, Y., LING, C., LIU, L., ZHANG, J., WANG, J., TONG, X., HIDAYAT, K., CHEN, M., CHEN, X., ZHOU, H., XU, J., QIN, L., ZHU, W. & YANG, J. 2022. Effects of Whey Protein or Its Hydrolysate Supplements Combined with an Energy-Restricted Diet on Weight Loss: A Randomized Controlled Trial in Older Women. *Nutrients,* 14.

TAKAHIRA, M., NODA, K., FUKUSHIMA, M., ZHANG, B., MITSUTAKE, R., UEHARA, Y., OGAWA, M., KAKUMA, T. & SAKU, K. 2011. Randomized, double-blind, controlled, comparative trial of formula food containing soy protein vs. milk protein in visceral fat obesity. -FLAVO study. *Circ J,* 75**,** 2235-43.

TAKAKI, M., MAWE, G. M., BARASCH, J. M., GERSHON, M. D. & GERSHON, M. D. 1985. Physiological responses of guinea-pig myenteric neurons secondary to the release of endogenous serotonin by tryptamine. *Neuroscience,* 16**,** 223-40.

TAORMINA, V. M., UNGER, A. L., SCHIKSNIS, M. R., TORRES-GONZALEZ, M. & KRAFT, J. 2020. Branched-Chain Fatty Acids-An Underexplored Class of Dairy-Derived Fatty Acids. *Nutrients,* 12.

TEIGEN, L. M., GENG, Z., SADOWSKY, M. J., VAUGHN, B. P., HAMILTON, M. J. & KHORUTS, A. 2019. Dietary Factors in Sulfur Metabolism and Pathogenesis of Ulcerative Colitis. *Nutrients,* 11.

TOMÉ, D. 2021. Amino acid metabolism and signalling pathways: potential targets in the control of infection and immunity. *Nutrition & Diabetes,* 11**,** 20.

TOPPING, D. L. & CLIFTON, P. M. 2001. Short-chain fatty acids and human colonic function: roles of resistant starch and nonstarch polysaccharides. *Physiol Rev,* 81**,** 1031-64.

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

TREMAROLI, V. & BÄCKHED, F. 2012. Functional interactions between the gut microbiota and host metabolism. *Nature,* 489**,** 242-249.

TRINIDAD, T. P., WOLEVER, T. M. & THOMPSON, L. U. 1996. Effect of acetate and propionate on calcium absorption from the rectum and distal colon of humans. *Am J Clin Nutr,* 63**,** 574-8.

TURNBAUGH, P. J., RIDAURA, V. K., FAITH, J. J., REY, F. E., KNIGHT, R. & GORDON, J. I. 2009. The effect of diet on the human gut microbiome: a metagenomic analysis in humanized gnotobiotic mice. *Sci Transl Med,* 1**,** 6ra14.

VANWEERT, F., SCHRAUWEN, P. & PHIELIX, E. 2022. Role of branched-chain amino acid metabolism in the pathogenesis of obesity and type 2 diabetes-related metabolic disturbances BCAA metabolism in type 2 diabetes. *Nutrition & Diabetes,* 12**,** 35.

VERMEULEN, M. A., DE JONG, J., VAESSEN, M. J., VAN LEEUWEN, P. A. & HOUDIJK, A. P. 2011. Glutamate reduces experimental intestinal hyperpermeability and facilitates glutamine support of gut integrity. *World J Gastroenterol,* 17**,** 1569-73.

VUIK, F., DICKSVED, J., LAM, S. Y., FUHLER, G. M., VAN DER LAAN, L., VAN DE WINKEL, A., KONSTANTINOV, S. R., SPAANDER, M., PEPPELENBOSCH, M. P., ENGSTRAND, L. & KUIPERS, E. J. 2019. Composition of the mucosa-associated microbiota along the entire gastrointestinal tract of human individuals. *United European Gastroenterol J,* 7**,** 897-907.

VYHLIDALOVA, B., KRASULOVA, K., PECINKOVA, P., MARCALIKOVA, A., VRZAL, R., ZEMANKOVA, L., VANCO, J., TRAVNICEK, Z., VONDRACEK, J., KARASOVA, M., MANI, S. & DVORAK, Z. 2020. Gut Microbial Catabolites of Tryptophan Are Ligands and Agonists of the Aryl Hydrocarbon Receptor: A Detailed Characterization. *Int J Mol Sci,* 21.

WALKER, M. C. & VAN DER DONK, W. A. 2016. The many roles of glutamate in metabolism. *J Ind Microbiol Biotechnol,* 43**,** 419-30.

WAN, Y., WANG, F., YUAN, J., LI, J., JIANG, D., ZHANG, J., LI, H., WANG, R., TANG, J., HUANG, T., ZHENG, J., SINCLAIR, A. J., MANN, J. & LI, D. 2019. Effects of dietary fat on gut microbiota and faecal metabolites, and their relationship with cardiometabolic risk factors: a 6-month randomised controlled-feeding trial. *Gut,* 68**,** 1417.

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

WANG, G., HUANG, S., WANG, Y., CAI, S., YU, H., LIU, H., ZENG, X., ZHANG, G. & QIAO, S. 2019. Bridging intestinal immunity and gut microbiota by metabolites. *Cellular and Molecular Life Sciences,* 76**,** 3917-3937.

WANG, L., WANG, H., ZHANG, B., POPKIN, B. M. & DU, S. 2020. Elevated Fat Intake Increases Body Weight and the Risk of Overweight and Obesity among Chinese Adults: 1991-2015 Trends. *Nutrients,* 12.

WANG, W., WU, Z., DAI, Z., YANG, Y., WANG, J. & WU, G. 2013. Glycine metabolism in animals and humans: implications for nutrition and health. *Amino Acids,* 45**,** 463-77.

WIKOFF, W. R., ANFORA, A. T., LIU, J., SCHULTZ, P. G., LESLEY, S. A., PETERS, E. C. & SIUZDAK, G. 2009. Metabolomics analysis reveals large effects of gut microflora on mammalian blood metabolites. *Proc Natl Acad Sci U S A,* 106**,** 3698-703.

WOLF, P. G., COWLEY, E. S., BREISTER, A., MATATOV, S., LUCIO, L., POLAK, P., RIDLON, J. M., GASKINS, H. R. & ANANTHARAMAN, K. 2022. Diversity and distribution of sulfur metabolic genes in the human gut microbiome and their association with colorectal cancer. *Microbiome,* 10**,** 64.

WOLFE, R. R., CIFELLI, A. M., KOSTAS, G. & KIM, I. Y. 2017. Optimizing Protein Intake in Adults: Interpretation and Application of the Recommended Dietary Allowance Compared with the Acceptable Macronutrient Distribution Range. *Adv Nutr,* 8**,** 266-275.

WU, S., BHAT, Z. F., GOUNDER, R. S., MOHAMED AHMED, I. A., AL-JUHAIMI, F. Y., DING, Y. & BEKHIT, A. E. A. 2022. Effect of Dietary Protein and Processing on Gut Microbiota-A Systematic Review. *Nutrients,* 14.

YAO, C. K., MUIR, J. G. & GIBSON, P. R. 2016. Review article: insights into colonic protein fermentation, its modulation and potential health implications. *Aliment Pharmacol Ther,* 43**,** 181-96.

YE, Z., WANG, S., ZHANG, C. & ZHAO, Y. 2020. Coordinated Modulation of Energy Metabolism and Inflammation by Branched-Chain Amino Acids and Fatty Acids. *Frontiers in Endocrinology,* 11.

YOSHIDA, H., ISHII, M. & AKAGAWA, M. 2019. Propionate suppresses hepatic gluconeogenesis via GPR43/AMPK signaling pathway. *Arch Biochem Biophys,* 672**,** 108057.

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

YU, D., RICHARDSON, N. E., GREEN, C. L., SPICER, A. B., MURPHY, M. E., FLORES, V., JANG, C., KASZA, I., NIKODEMOVA, M., WAKAI, M. H., TOMASIEWICZ, J. L., YANG, S. E., MILLER, B. R., PAK, H. H., BRINKMAN, J. A., ROJAS, J. M., QUINN, W. J., 3RD, CHENG, E. P., KONON, E. N., HAIDER, L. R., FINKE, M., SONSALLA, M., ALEXANDER, C. M., RABINOWITZ, J. D., BAUR, J. A., MALECKI, K. C. & LAMMING, D. W. 2021. The adverse metabolic effects of branched-chain amino acids are mediated by isoleucine and valine. *Cell Metab,* 33**,** 905-922.e6.

ZHANG, S., ZENG, X., REN, M., MAO, X. & QIAO, S. 2017. Novel metabolic and physiological functions of branched chain amino acids: a review. *J Anim Sci Biotechnol,* 8**,** 10.

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

Table 1: Impact of protein quantity and quality on body weight and metabolic health in humans

	Test protein	Control	Dur:	BMI	Sex	Diet	Health outcomes	Ref:
Impact of protein quantity	WP	CHO	23W	~31.1	M/F	Ad lib:	↓ Body mass, fat mass, weight circumference and plasma insulin	(Baer et al., 2011)
	Soy	CHO	23W	~31.1	M/F	Ad lib:	↓ Plasma insulin	(Baer et al., 2011)
	Soy	CHO	3W	25-30	M/F	Ad lib:	↓Systolic blood pressure	(Beaumont et al., 2017b)
	WP	Baseline	16W	~28	M/F	Ad lib:	↓Body weight, body fat, plasma triglycerides and HDL	(Arciero et al., 2014)
	Casein	Baseline	12W	~30.5	F	Ad lib:	↑ Total abdominal body fat and subcutaneous fat	(Sites et al., 2007)
	MP	Baseline	20W	~29	M/F	Ad lib:	↓body weight, visceral fat and subcutaneous fat and systolic blood pressure	(Takahira et al., 2011)
	Soy	Baseline	20W	~29	M/F	Ad lib:	↓Diastolic blood pressure	(Takahira et al., 2011)
	Red/white meat	High v Low proteins	64W	~32.8	F	CR+ WMD	↑ weight loss	(Clifton et al., 2008)
Impact of protein quality	WP	Soy	23W	~31.1	M/F	Ad lib:	↓ Waist circumference, circulatory ghrelin and IGF1	(Baer et al., 2011)
	MP	Soy + MP	20W	~29	M/F	Ad lib:	↓ visceral fat	(Takahira et al., 2011)
	WP	Collagen	8W	30.9 -31.1	F	Ad lib:	↓ visceral fat	(Giglio et al., 2019)
	WP	Casein	12W	~31.3	M/F	Ad	↓Augmentation	(Pal and

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

						lib:	index (arterial stiffness)	Ellis, 2010)
	WP	Soy	2W	28-50	M/F	CR	Decline in muscle protein synthesis reduced	(Hector et al., 2015)
	MP + amino acids	Pea + casein	12W	~33	M/F	CR	↓ Visceral fat and ↓ subcutaneous fat	(Bel Lassen et al., 2021)
	Gelatin	Whey	8W	~36	F	CR	↓ waist circumference	(Piccolo et al., 2015)
	WP + EAA	Casein	8W	~31.3	M/F	CR	↓ body fat	(Coker et al., 2012)
	WPH	WP	8W	24-35	F	CR	↓ HOM-IR	(Sun et al., 2022)
	Whey	Soy	32W	27.6 -40.4	M/F	CR + WMD	CR induced metabolic improvement were not influenced by protein source	(Kjolbaek et al., 2017)

The direction of change is shown by arrows, as (↑) increase, (↓) decrease or (↔) no change. Abbreviations: Ad lib; ad libitum, CHO; carbohydrate, CR; calorie restriction, Dur; duration, EAA; essential amino acids, F; female, HDL; high density lipoproteins, IGF; insulin-like growth factor, M; male, MP; milk proteins, WMD; weight maintenance diet; WP; whey proteins, WPH; whey protein hydrolysate, W; weeks.

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

Table 2: Impact of protein quantity and quality on the composition and functional potential of the gut microbiota in humans

Test Protein	Comparison	Dur:	BMI	Sex	Intervention:	Impact on gut microbiota	Ref
Red meat, white meat, and plant proteins	High v Low fat	18W	18-26	M/F	Ad lib:	-Dietary fat main driver of changes in the gut microbiota. - Within each level of fat, dietary proteins, regardless of the source, affected *Akkermansia, Bacteroides, Sutterella, Cantenibacterium, Faecalibacterium, Megasphaera, Oscillospira* and *Methanobrevibacter*	(Lang et al., 2018)
Salmon	No fish	8W	~31	M/F	Ad lib:	-**[↓]** *Bacteroidetes* Phylum, *Clostridiales* order of *Firmicutes* -**[↑]** *Selenomonadales* order of *Firmicutes.*	(Bratlie et al., 2021)
Low Gluten	High Gluten	22W	~28	**?**	Ad lib:	-No changes in diversity. -**[↓]** *B. longum, B. angulatum, B. pseudocatenulatum, B. adolescentis, D. longicatena, B. wexlerae, Lachnospiraceae 1, A. hadrus, E. hallii.* -**[↑]** Clostridiales, Lachnospiraceae 2 -**[↓]** Functional potential for carbohydrate degradation	(Hansen et al., 2018)
Casein	Soy	3W	25-30	M/F	Ad lib:	-No changes in diversity.	(Beaumont et al., 2017b)
Pork	Chicken	3W	~30	M/F	Ad lib:	-No changes in diversity. -**[↑]** *Ruminiclostridium 5* pre- v post pork group and **[↓]** pre- v post chicken group	(Dhakal et al., 2022)
MP+ amino acid	Pea+ casein	12W	~32	M/F	CR	**[↑]** microbial function potential to synthesis amino acids	(Bel Lassen et al., 2021)
WPH	WP	8W	24-35	F	CR	No effect on the gut microbiota	(Sun et al., 2022)

The direction of change is shown by arrows, as (↑) increase, (↓) decrease or (↔) no change.

Abbreviations: Ad lib; ad libitum, CR; calorie restriction, Dur; duration, F; female, M; male, MP; milk proteins, WP; whey proteins, WPH; whey protein hydrolysate, W; weeks**.**

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

Table 3: Metabolic effects of dietary protein or microbial-derived amino acids and their metabolites.

Dietary protein derived nutrient	**Metabolites**	**Host metabolic effects of metabolites**	References
AA	Microbial-derived AA	Protein synthesis; Immunity; Regulation of metabolic pathways; Energy homeostasis.	(Cruzat et al., 2018, Ye et al., 2020, Cuomo et al., 2022, Chandel, 2021, Agus et al., 2021, Tomé, 2021, Alamshah et al., 2017, Vermeulen et al., 2011, Oliphant and Allen-Vercoe, 2019, Portune et al., 2016)
Aromatic AA-Tryptophan	Tryptamine Indole Kynurenine/Quinolinic Serotonin	Neurotransmitter (Immunity and intestinal motility). Improves intestinal barrier and satiety. Immunity. Neurotransmitter (Mood, appetite and immunity).	(Oliphant and Allen-Vercoe, 2019, Dehhaghi et al., 2019, Bischoff et al., 2009, Bansal et al., 2010, Chimerel et al., 2014, Jennis et al., 2018, Portune et al., 2016, Su et al., 2022, Roth et

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

			al., 2021, Hu et al., 2022, Cheng et al., 2015)
Aromatic AA-Tyrosine	Tyramine Phenol p-cresol-derivatives	Neurotransmitter linked to hypertension. Reduces integrity of gut epithelium. Reduces integrity of gut epithelium.	(Oliphant and Allen-Vercoe, 2019)
Aromatic AA-Phenylalanine	Phenylethylamine	Neurotransmitter. Releases catecholamine/serotonin.	(Oliphant and Allen-Vercoe, 2019)
Glutamate, Arginine	GABA	Neurotransmitter (stress modulation).	(Oliphant and Allen-Vercoe, 2019, Portune et al., 2016)
Basic AA-Arginine	Agmatine Putrescine Spermidine/spermine	Anti-inflammatory and inhibits intestinal cell proliferation. Promotes intestinal cell proliferation. Reduces oxidative stress.	(Oliphant and Allen-Vercoe, 2019)
Basin AA-Histidine	Histamine	Neurotransmitter (wakefulness and learning).	(Oliphant and Allen-Vercoe, 2019, Portune et al., 2016)
Sulphur-AA Cysteine Methionine	H_2S Methanethiol	Impedes cellular respiration and cause apoptosis.	(Portune et al., 2016, Yao et al., 2016)
Basic AA-Lysine, Histidine Aromatic AA-	Ammonia	Reduces intestinal activity.	(Portune et al., 2016, Yao et al., 2016)

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

Tyrosine, tryptophan			
BCAA- Leucine /Isoleucine	BCFA- Isovalerate/Isobutyrate	Lipid and glucose metabolism	(Heimann et al., 2016, Taormina et al., 2020)
Glutamine, Glutamate, Alanine, Histidine, Serine, Threonine Cysteine, Methionine, Lysine	Butyrate (SCFA)	Reduces intestinal permeability and affects cell proliferation; energy source for gut cells; mucin production: immunity: epigenetic effects.	(Parada Venegas et al., 2019, Salvi and Cowles, 2021, Davie, 2003)
Glutamine/Glutamate, Alanine, Glycine, Histidine, Serine, Threonine, Cysteine, Proline, Lysine	Acetate (SCFA)	Colonic calcium absorption; appetite suppression; liver lipogenesis and cholesterol metabolism	(Trinidad et al., 1996, Frost et al., 2014)
Aspartate, Alanine, Threonine Methionine	Propionate (SCFA)	Production of satiety hormones and appetite suppression; intestinal permeability; impedes liver gluconeogenesis cholesterol metabolism.(Portune et al., 2016)	(Chambers et al., 2015, Yoshida et al., 2019)

Abbreviations: AA; Amino acids, BCAA; Branched chain amino acids, BCFA; Branched chain fatty acid, GABA; gamma-aminobutyric acid; SCFA; short chain fatty acid.

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

Table 4: Impact of dietary proteins on the metabolite profiles in the faeces in humans.

Nutrient or metabolite		High protein diets			Reference
		Dairy	Meat	Plant	
BCAA		↑CAS (3W)	↑RED/WHT (4W)	?	(Beaumont et al., 2017a, Connolly-Schoonen et al., 2023)
EAA- Methionine		?	↑ RED/WHT (4W)	?	(Connolly-Schoonen et al., 2023)
Non-EAA-Cysteine, Alanine		?	↑ RED/WHT (4W)	?	(Connolly-Schoonen et al., 2023)
Non-EAA-Tyrosine		?	↔ RED/WHT (4W)	↔GLTN (22W)	(Connolly-Schoonen et al., 2023, Hansen et al., 2018)
BCFA		↑ MP (1W) ↑CAS (3W)	↑ RED/WHT (4W)	↑ SOY (3W) ↔ GLTN (22W)	(Beaumont et al., 2017a, Geypens et al., 1997, Russell et al., 2011, Gratz et al., 2019, Hansen et al., 2018)
SCFA-Butyrate		↓ CAS (3W)	↓ RED/WHT (4W)	↓ SOY (3W) ↔GLTN (22W)	(Beaumont et al., 2017a, Moller et al., 2020, Russell et al., 2011, Gratz et al., 2019, Hansen et al., 2018)
		↔ DAIRY/MEAT (1.5W)			
		↔ DAIRY/ANIMAL/PLANT (48W)			
SCFA-Propionate		↔ CAS (3W)	↓RED/WHT (4W)	↔ SOY (3W) ↔GLTN (22W)	(Beaumont et al., 2017a, Moller et al., 2020, Russell et al., 2011, Gratz et al., 2019, Hansen et al., 2018)
		↔ DAIRY/MEAT (1.5W)			
		↔ DAIRY/ANIMAL/PLANT (48W)			
SCFA-Acetate		↑ CAS (3W)	↓ RED/WHT (4W)	↔ SOY (3W) ↔GLTN (22W)	(Beaumont et al., 2017a, Moller et al., 2020, Russell et al., 2011, Gratz et al., 2019, Hansen et al., 2018)
		↔ DAIRY/MEAT (1.5W)			
		↔ DAIRY/ANIMAL/PLANT (48W)			
Tryptophan Derivatives	Indole-Derivatives	↑ MP (1W)	↔ RED/WHT (4W)	↔GLTN (22W)	(Geypens et al., 1997, Russell et al., 2011, Gratz et al., 2019, Hansen et al.,

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

					2018)
	Kynurenine	?	?	↑ GLTN (22W)	(Hansen et al., 2018)
	Quinolinic	?	?	↔GLTN (22W)	(Hansen et al., 2018)
Tyrosine Derivatives	Phenol	↑ MP (1W)		?	(Geypens et al., 1997, Moller et al., 2020)
		↔ DAIRY/MEAT (1.5W)			
		↔ DAIRY/ANIMAL/PLANT (48W)			
	p-cresol-derivatives	↑ MP (1W)	↔ RED/WHT (4W)	↑ SOY(3W)	(Beaumont et al., 2017a, Gratz et al., 2019, Geypens et al., 1997)
	Tyramine	↔ CAS (3W)	?	↑ SOY(3W) ↔GLTN (22W)	(Beaumont et al., 2017a, Hansen et al., 2018)
Tryptophan, Lysine, Histadine, Tyrosin	Ammonia	↑ MP (1W) ↔ CAS (3W)	↑RED (3W) ↔ RED/WHT (4W)	↔ SOY(3W)	(Beaumont et al., 2017a, Geypens et al., 1997, Russell et al., 2011, Moller et al., 2020, Bingham et al., 1996)
		↑ DAIRY/MEAT (48W)			
		↑ DAIRY/ANIMAL/PLANT (48W)			
Cysteine, methionine -derived	Methanethiol	↑ MP (1W)	?	?	(Geypens et al., 1997)

The direction of change is shown by arrows, as (↑) increase, (↓) decrease or (↔) no change of metabolites. The length of the dietary challenge is shown in subscript in weeks (W). Abbreviations, BCAA; Branch chain amino acids, BCFA; Branched chain fatty acids, CAS; casein, EAA; essential amino acids, MP; milk proteins, RED; red meat, SCFA; short chain fatty acids, GLTN; gluten, SCFA; short chain fatty acids; WHT, white meat.

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

Table 5: Impact of dietary proteins on the metabolite profiles in circulation or urine in humans.

Nutrient or metabolite	High protein diets			References
	Dairy	Meat	Plant	
EAA-BCAA	↑CAS (3W:PLASMA: FASTED) ↑ WP (4W:PLASMA; FASTED) ↑ WP (3H:PLASMA; POSTPRANDIAL) ↑MP (5H;PLASMA: POSTPRANDIAL)	↑RED (4H:PLASMA; POSTPRANDIAL) ↑RED/WHT (4W:SERUM: NOT FASTED)	↑SOY (3W:PLASMA:FASTED) ↑ MIX PLANT (5H;PLASMA: POSTPRANDIAL)	(Connolly-Schoonen et al., 2023, Beaumont et al., 2017a, Prodhan et al., 2020, Chiu et al., 2014, Pinckaers et al., 2023, Hall et al., 2003a)
EAA_Threonine	↑ MP (5H;PLASMA: POSTPRANDIAL)	↑RED (4H:PLASMA; POSTPRANDIAL) ↔ RED/WHT (4W:SERUM:NOT FASTED)	↑ MIX PLANT (5H;PLASMA: POSTPRANDIAL)	(Connolly-Schoonen et al., 2023, Pinckaers et al., 2023, Prodhan et al., 2020)
EAA_Tryptophan, Methionine, Lysine	↑ MP (5H;PLASMA: POSTPRANDIAL)	↑ RED (4H:PLASMA; POSTPRANDIAL) ↔ RED/WHT (4W:SERUM:NOT	↑ MIX PLANT (5H;PLASMA: POSTPRANDIAL)	(Connolly-Schoonen et al., 2023, Prodhan et al., 2020, Pinckaers et al., 2023)

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

		FASTED)		
Non-EAA_Tyrosine	↑ MP (5H;PLASMA: POSTPRANDIAL) ↑ CAS (3W:PLASMA: FASTED)	↑ RED (4H:PLASMA; POSTPRANDIAL) ↔ RED/WHT (4W:SERUM:NOT FASTED)	↑ SOY (3W:PLASMA:FASTED) ↑ MIX PLANT (5H;PLASMA: POSTPRANDIAL)	(Connolly-Schoonen et al., 2023, Beaumont et al., 2017a, Prodhan et al., 2020, Pinckaers et al., 2023)
Non-EAA_Glycine	↑ MP (5H;PLASMA: POSTPRANDIAL)	↑RED (4H:PLASMA; POSTPRANDIAL) ↔ RED/WHT (4W:SERUM:NOT FASTED)	↑ MIX PLANT (5H;PLASMA: POSTPRANDIAL)	(Connolly-Schoonen et al., 2023, Prodhan et al., 2020, Pinckaers et al., 2023)
BCFA	↑ CAS (3W:URINE: FASTED)	↑ RED/WHT (4W:SERUM:NOT FASTED)	↑ SOY (3W:URINE:FASTED) ↔ GLTN (22W:URINE;FASTED)	(Connolly-Schoonen et al., 2023, Hansen et al., 2018, Beaumont et al., 2017b)
SCFA-Butyrate	↓ CAS (3W:URINE: FASTED)	?	↓ SOY (3W:URINE:FASTED) ↔ GLTN (22W:URINE;FASTED)	(Hansen et al., 2018, Beaumont et al., 2017a)
SCFA-Propionate	↔ CAS (3W:URINE: FASTED)	?	↔ SOY (3W:URINE:FASTED) ↔ GLTN (22W:URINE;FASTED)	(Beaumont et al., 2017a, Hansen et al., 2018)
SCFA-Acetate	↑CAS (3W:URINE:	?	↔ SOY (3W:URINE:FASTED)	(Beaumont et al., 2017a, Hansen et al., 2018)

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

		FASTED)		↔ GLTN (22W:URINE;FASTED)	
Tryptophan Derivatives	Indole-derivatives	↑ CAS (3W:URINE: FASTED)	↔ RED/WHT (4W:SERUM:NOT FASTED)	↑ SOY (3W:URINE:FASTED) ↔ GLTN (22W:URINE;FASTED)	(Connolly-Schoonen et al., 2023, Dahl et al., 2020, Hansen et al., 2018, Beaumont et al., 2017a, Mutch et al., 2009)
			↑ MEAT/PLANT (2W:URINE/SERUM;FASTED)		
	Kynurenine	?	↔ RED/WHT (4W:SERUM:NOT FASTED)	↓ GLTN (22W:URINE;FASTED)	(Connolly-Schoonen et al., 2023, Hansen et al., 2018)
	Quinolinic	?	?	↔ GLTN (22W:URINE;FASTED)	(Hansen et al., 2018)
Tyrosine Derivatives	p-cresol-derivatives	↑CAS (3W:URINE: FASTED)	↔RED/WHT (4W:SERUM:NOT FASTED)	↔SOY (3W:URINE:FASTED)	(Connolly-Schoonen et al., 2023, Beaumont et al., 2017a, Dahl et al., 2020)
			↔MEAT/PLANT (2W:URINE/SERUM;FASTED)		
Tryptophan, Tyrosine, Histidine	Ammonia	↔CAS (3W:URINE: FASTED)	?	↔SOY (3W:URINE:FASTED)	(Beaumont et al., 2017a)

The direction of change is shown by arrows, as (↑) increase, (↓) decrease or (↔) no change of metabolites. The length of the dietary challenge is shown in subscript in weeks (W) or hours (H) along with the medium in which the metabolite was detected and whether the subjects were fasted or non-fasted. Abbreviations, BCAA; Branch chain amino acids, BCFA; Branched chain fatty acids, CAS; casein, EAA; essential amino acids, MP; milk proteins, RED; red meat, SCFA; short chain fatty acids, GLTN; gluten, SCFA; short chain fatty acids; WHT, white meat.

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

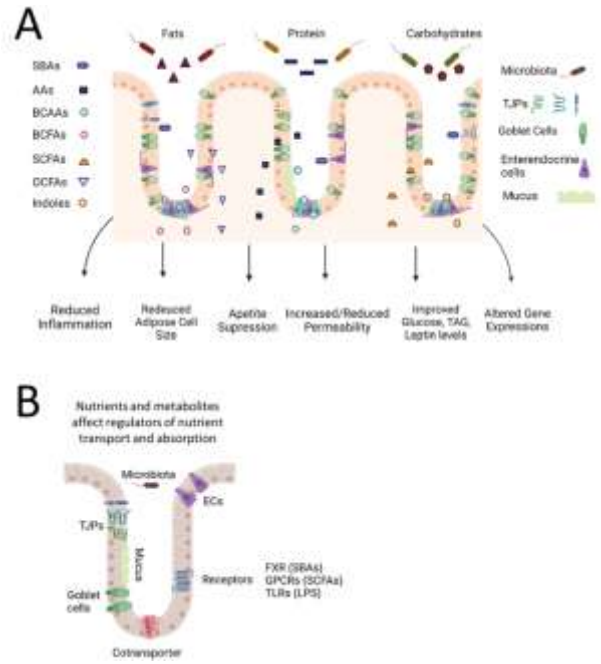

Figure 1: The impact of nutrients on the colonic epithelium. (A) Digested macronutrients either pass through the epithelium or they are metabolised by the gut microbiota, resulting in different metabolites been produced, with diverse roles. (B) shows a colon intestinal crypt, and associated cells and receptors that respond to nutrients and metabolites involved in many signalling mechanisms. Abbreviations: AA, amino acids; BCAA, branched chain amino acids; BCFA, branched chain fatty acids; EC, enteroendocrine cells; FXR, Farnesoid X receptor; SBAs, GPCRs, G-protein-coupled receptors; OCFA; odd chain fatty acids; Secondary bile acids; SCFA, short chain fatty acids; TAG; triacylglycerol; TLRs, Toll-like receptors; LPS, Lipopolysaccharides; TJB, tight junction associated proteins.

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press

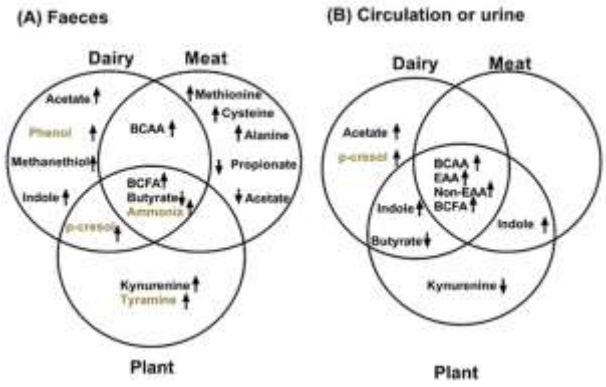

Figure 2: Impact of protein quality on the metabolites in (A) faeces and (B) circulation or urine. The direction of change is shown by arrows, as (↑) increased or (↓) decreased. Metabolites highlighted in red colour are known to cause unhealthy outcomes in humans. Abbreviations, BCAA; Branch chain amino acids, BCFA; Branched chain fatty acids, EAA; essential amino acids.

https://doi.org/10.1017/S0954422424000374 Published online by Cambridge University Press