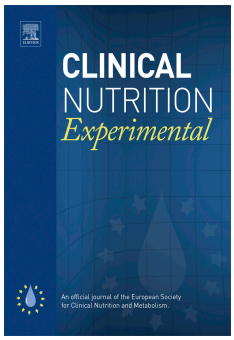
Losing weight for a better health: role for the gut microbiota

Maria Carlota Dao, Amandine Everard, Karine Clément, Patrice D. Cani

Accepted Manuscript



Losing weight for a better health: role for the gut microbiota

Maria Carlota Dao, Amandine Everard, Professor Karine Clément, Prof Patrice D. Cani

PII: S2352-9393(15)00020-2

DOI: 10.1016/j.yclnex.2015.12.001

Reference: YCLNEX 9

To appear in: *Clinical Nutrition Experimental*

Received Date: 10 November 2015

Accepted Date: 12 December 2015

Please cite this article as: Dao MC, Everard A, Clément K, Cani PD, Losing weight for a better health: role for the gut microbiota, *Clinical Nutrition Experimental* (2016), doi: 10.1016/j.yclnex.2015.12.001.

Losing weight for a better health: role for the gut microbiota

Maria Carlota Dao[1,2,3], Amandine Everard[4], Karine Clément[1,2,3], Patrice D. Cani[4]

[1]Institute of Cardiometabolism and Nutrition, ICAN, Assistance Publique Hôpitaux de Paris, Pitié-Salpêtrière hospital, Paris, France

[2]INSERM, UMR S U1166, Nutriomics Team, Paris, France

[3]Sorbonne Universités, UPMC University Paris 06, UMR_S 1166 I, Nutriomics Team, Paris, France

[4]Université catholique de Louvain, Louvain Drug Research Institute, WELBIO (Walloon Excellence in Life sciences and BIOtechnology), Metabolism and Nutrition research group, Av. E. Mounier, 73 Box B1.73.11, B-1200 Brussels, Belgium.

All authors equally contributed to this work

Correspondence should be addressed to Professor Karine Clément, Institute of Cardiometabolism and Nutrition (ICAN), bâtiment E3M, 83 boulevard de l'Hôpital, Bureau 616, 75013 Paris, France; E-mail: karine.clement@aphp.fr

And Prof Patrice D. Cani, Université catholique de Louvain, LDRI, Metabolism and Nutrition research group, Av. E. Mounier, 73 box B1.73.11, B-1200 Brussels, Belgium. E-mail: patrice.cani@uclouvain.be

Abstract

In recent years, there have been several reviews on gut microbiota, obesity and cardiometabolism summarizing interventions that may impact the gut microbiota and have beneficial effects on the host (some examples include [1–3]). In this review we discuss how the gut microbiota changes with weight loss (WL) interventions in relation to clinical and dietary parameters. We also evaluate available evidence on the heterogeneity of response to these interventions. Two important questions were generated in this regard: 1) *Can response to an intervention be predicted?* 2) *Could pre-intervention modifications to the gut microbiota optimize WL and metabolic improvement?* Finally, we have delineated some recommendations for future research, such as the importance of assessment of diet and other environmental exposures in WL intervention studies, and the need to shift to more integrative approaches of data analysis.

WEIGHT LOSS INTERVENTIONS, HEALTH OUTCOMES AND THE ROLE FOR GUT MICROBIOTA

Effect of calorie restriction on gut microbiota – can we predict host responses based on pre-intervention health status and microbiota composition?

Several studies in animal models and humans have addressed the impact of WL through calorie restriction (CR) on microbiota composition and its association with clinical outcomes. Some of these studies have analyzed whether certain phenotypes before WL may impact or predict the effect of the intervention on health outcomes.

Rodent models

Studies in rodent models have shed light on the role that gut microbiota may be playing in obesity. It has been demonstrated in rodents that an obese phenotype can be transmitted via the microbiota. Gut microbiota, depending on its composition and function, may be involved in several mechanisms leading to fat mass gain and eventually obesity. Among the mechanism the role of energy harvest from food (shown to be more efficient in certain bacterial groups) has been proposed. Germ free mice are resistant to diet-induced obesity,[6,7] but gain weight upon transfer of gut microbiota from conventionally raised mice or *ob/ob* mice, potentially through increased capacity for energy harvest.[8] Gut microbiota may also impact host metabolism in the development of rodent obesity through the induction of hepatic lipogenesis, and suppression of *Fiaf* in the gut epithelia, leading to upregulation of LPL activity and increased fat storage.[6] There is also a direct interaction between the gut microbiota, the gut-associated immune system, and adipose tissue through metabolic endotoxemia.[9–11] Therefore, other effects such as the regulation of lipogenesis and gluconeogenesis, gut hormone secretion and induction of inflammatory response have also been demonstrated in rodents.[5] In addition, rodent models have been used to investigate the relationship between genetics and gut microbiota,[12] and these studies have shown that different genetic backgrounds can lead to very diverse host-environment interactions.

Gut microbiota changes due to CR can be significant and depend on the type of intervention. For example, duration of CR can impact both gut microbiota composition and health outcomes. Zhang et al. showed in mice that lifelong CR led to large and

consistent changes in gut microbiota composition.[13] In this study, there was lower midlife serum LPS binding protein (LBP, a surrogate of metabolic endotoxemia) in mice fed a low fat and calorie diet, as opposed to other dietary compositions. Phyla that inversely correlated with LBP were positively correlated with lifespan, emphasizing on the important of low-grade inflammation in this context.

In humans

Divergence in human gut microbiota composition is associated to multiple factors. Microbiota enterotypes have been defined in different populations around the world. Differentiation into these enterotypes cannot be explained by individual factors such as age or degree of corpulence, geographical location, or by dietary modifications of short duration.[14] Instead, long-term dietary habits and certain clinical characteristics seem to be stronger determinants for these compositional differences.[15]

Obese and non-obese subjects have a different gut microbial profile.[16–20] Ley et al. showed that obese subjects have lower *Bacteroidetes* to *Firmicutes* ratio than lean subjects.[8] However, these findings have not been consistent in the literature.[21] Another study showed greater abundance in the *Firmicutes* group *Eubacterium rectale / Clostridium coccoides* in obese women with metabolic syndrome versus obese women with no metabolic complications and non-obese women.[19] There was a correlation between this bacterial group and certain clinical outcomes such as visceral adiposity. These findings suggest a different energy harvesting potential, consistent with the capacity of *Firmicutes* species to degrade non-digestible polysaccharides, although this remain to be proven.

An important aspect of gut microbial composition in relation to host health is microbial richness, referring to diversity in the gut ecosystem. Microbial richness is overall higher in lean vs. obese subjects, and this correlates with a healthier metabolic profile.[16,22] However even in subjects with different corpulence (lean vs. obese), metagenomic sequencing has revealed that different patterns of low or high diversity exist. When considering abundance of individual species, higher abundance of certain species such as *Faecalibacterium prausnitzii* (*F. prausnitzii)*[16,23,24] and *Akkermansia muciniphila (A. muciniphila)*[25,26] have been repeatedly associated with a healthier status.

In CR studies there have been some consistent shifts in microbial composition. Interestingly, it appears that certain characteristics in the gut, together with diet, associate with individual response to CR and lifestyle interventions. Such baseline differences and varied outcomes have been identified in the MICRO-Obes study, where a population of 49 overweight and obese individuals has been thoroughly studied in terms of gut microbiota composition, clinical parameters, and dietary intake. It was first shown that these individuals could be clustered by their response profile to 6 weeks of CR followed by a 6 week weight stabilization period. There were baseline differences in clinical parameters and microbiota among the three WL response clusters. Namely, *Lactobacillus/Leuconostoc/Pediococcus* group, was most abundant at baseline in the cluster of worst responders to CR and WS. However, the response to the intervention could be better predicted by baseline insulin sensitivity and inflammatory parameters illustrating the fact that we need deeper insight into the predictive potential of gut microbiota in dietary intervention.[27]

More recently, it was shown in both the MICRO-Obes and MetaHIT studies that individuals can be stratified by their microbial richness, and those with higher richness (about 60-80%) tend to have a healthier metabolic status [22] and dietary intake.[28] MICRO-Obes subjects that had higher baseline microbial richness tended to respond better to the dietary intervention in terms of blood lipids, insulin sensitivity and low-grade inflammation.

Finally, as it will be described in more detail in the following section, higher baseline *A. muciniphila* was associated with a healthier metabolic profile in the same study.[26] Individuals with a higher baseline abundance of this species had better outcomes from the intervention, namely a greater reduction in waist circumference, blood lipids, and increase in insulin sensitivity. Individuals with higher *A. muciniphila* in the context of higher microbial richness were also the most metabolically healthy throughout the intervention, illustrating the importance to take into account the overall gut microbial ecosystem, rather than focusing solely on one species.

The functional capacity of the gut microbiota in CR can be studied through modelisation of metagenomic information and through direct measure of metabolites in fluids (metabolomics). In a randomized cross-over study comparing a 4-week high protein/low carbohydrate diet to a high protein/medium carbohydrate regime in obese men, a reduction in abundance of *Roseburia* spp. and *E. rectale*, as well as fecal butyrate, correlated with lower carbohydrate intake.[29] Total fecal short chain fatty acids (SCFA), acetate, propionate, isovalerate and valerate increased with higher carbohydrate intake. On the other hand, the high protein/low carbohydrate diet was characterized by a potentially deleterious fecal metabolite profile, high in branched chain

fatty acids, phenylacetic acid and *N*-nitroso compounds.[30] Similarly, another study in obese adults found lower fecal SCFA production in an 8-week low carbohydrate/high fat regime. This was accompanied by an exacerbation of bowel habits and a decrease in *Bifidobacterium*.[31]

CR interventions in obese adolescents have also demonstrated changes in microbial composition.[32,33] Interestingly, baseline microbial composition differences were found between good (>4 kg WL) and bad (<2 kg WL) responders to CR, and changes in certain bacterial groups were associated with WL or improvement in clinical outcomes (**Table 1**).

Given the intricate relationship between the gut microbiota and host, a key question is *whether modification of gut microbiota before interventions through diet and/or prebiotic treatment (defined later in this review) has the potential to optimize WL and metabolic improvement*. Studying baseline differences between responders and non-responders is key to answer this question (**Figure 1**).

In conclusion, baseline profiles in microbiota and metabolic status, together dietary macronutrient intake, may play a role in outcomes from CR interventions. More detail is needed on the role of micronutrients. An interaction between diet and microbiota has been identified in the development of obesity in human-to-mouse microbial transplantation studies.[34,35] This evidence shows the importance of analyzing diet in CR interventions. For the most part, intervention periods have lasted most commonly 1 to 3 months, with a few exceptions going up to 6 months. Longer follow up periods should be included in future studies.

While these studies have adequately phenotyped the changes in gut microbiota composition with dietary interventions, it is difficult to go beyond strong correlations and

elucidate mechanisms from these results. As shall be discussed in the last section, data integration approaches allow the simultaneous analysis of environment, gut microbiota and host, which may lead to the identification of mechanistic links and therapeutic targets.

Effects of prebiotic and probiotic on host metabolism: putative links with gut microbes

Numerous studies have demonstrated that manipulating the gut microbiota with dietary intervention (i.e., prebiotics and probiotics) may affect host metabolism (i.e., glucose, lipid and energy metabolism) (**Figure 2**). In this section, we briefly discuss examples showing the impact of such intervention in preclinical models as well as recent evidence suggesting that dietary interventions using pre and probiotics may also be linked with gut microbes in humans.

Twenty years ago, Gibson and Roberfroid have developed the prebiotic concept, recently revised as "*A non digestible compound that, through its metabolization by microorganisms in the gut, modulates composition and/or activity of the gut microbiota, thus conferring a beneficial physiological effect on the host*".[36] Over the last decades, this concept has led to the investigation of key questions such as how changes in the gut microbiota induced by prebiotics but also specific bacteria contribute to regulate energy intake, fat mass development and glucose/lipid metabolism? We will first discuss data obtained in rodents and in the second part the effectiveness of such interventions on human health.

Animal models

More than a decade ago, Cani et al. described that the three different prebiotics (i.e. inulin-type fructans, which varied according to their degree of polymerization (i.e., number of fructose moieties), differentially affected gut peptides secretion. They found that the administration of prebiotic compounds profoundly changes the gut microbiota composition and metabolic function contributing to the upregulation of two gut peptides involved in reduced food intake, namely Glucagon-like peptide-1 (GLP-1) and PYY, and a decreased plasma levels of the orexigenic peptide, ghrelin.[37,38] By using culture and non-culture dependent tools it has been shown that the three prebiotics used were able to change the gut microbiota in favor of *Bifidobacterium* spp. The abundance of *Bifidobacterium* spp. was inversely associated with body weight, fat mass as well as metabolic endotoxemia and inflammation.[39] More recently, thanks to metagenomics tools, novel results have clearly shown that the modulation of the gut microbiota was more complex than a simple change in *Bifidobacterium* spp., indeed, dozens of taxa were changed upon prebiotic treatment in obese and diabetic rodents.[40] Among the taxa increased by the treatment, *Akkermansia muciniphila* was increased by about 100 fold.[40] Interestingly, the abundance of this bacteria was positively associated with a lower fat mass, an improved glucose tolerance and gut barrier function as well as with the number of intestinal L cells secreting GLP-1 and PYY.[40] Since this discovery, several studies have shown that the administration of *Akkermansia muciniphila* in obese and diabetic rodents reduces fat mass gain, insulin resistance, metabolic endotoxemia and low grade inflammation,[12,41,42] thereby showing that this bacteria may play a crucial role. Although the overall mechanisms are not fully elucidated, this bacterium reinforced the gut barrier function and contribute to regulate energy homeostasis.[41]

Thus, taken together, a variety of rodent model studies indicate that prebiotics may elicit beneficial impacts in metabolic disorders associated with obesity and diabetes. Moreover, several studies indicate that some of these effects may be obtained with specific bacteria often misinterpreted as probiotic. Notably, the term probiotic is often misused (see the International Scientific Association for Probiotics and Prebiotics published a consensus statement clarifying the scope of and appropriate use for the term 'probiotic' (for a review, see [43]).

Besides this important opinion, various strains of *Lactobacillus* and *Bifidobacterium* have demonstrated beneficial effects, most of the time by maintaining glucose homeostasis and decreasing inflammation and hepatic steatosis. Importantly, some of these strains also affect body weight and fat mass development, whereas others do not (for comprehensive reviews on this topic).[44,45]

In summary, abundant literature have reported the impact of specific *Lactobacillus* or *Bifidobacterium* strains on obesity and associated disorders in rodents, however strains are not equally potent in terms of body weight and fat mass loss or improvement of glucose/lipid metabolism and inflammatory markers.

The following examples illustrate the concept that strains are not equipotent. *Lactobacillus gasseri* BNR17 reduces body weight and fat mass in overweight rats,[46] whereas in diet-induced obese mice, *Lactobacillus plantarum* 14 reduces the mean adipocyte size and *Lactobacillus paracasei* F19 induces a reduction of total fat mass and plasma triglycerides.[47] Conversely, *Lactobacillus acidophilus* NCDC supplementation did not affect body fat mass and/or hepatic steatosis and muscle fat in obese mice.[48] *Lactobacillus casei* Shirota reduces insulin resistance and metabolic endotoxemia, without affecting fat mass and body weight in diet-induced obese mice.[49] Finally,

Lactobacillus plantarum WCFS1 did not change body weight, fat mass or inflammation in diet-induced obese mice.[41] These examples clearly illustrate that although they are all *Lactobacillus*, specific strains are efficient on metabolic parameters whereas other not.

Similar to the *Lactobacillus* spp. examples, specific strains of *Bifidobacterium* have been shown to metabolic disorders in obese and diabetic models.[44] For example, a recent study has shown that *Bifidobacterium pseudocatenulatum* CECT 7765 reduces body weight gain, fat mass, plasma glucose and inflammation in in diet-induced obese mice.[50] In a similar model, *Bifidobacterium longum* supplementation has been found to reduce body weight gain, fat mass, insulin resistance, systolic blood pressure, and metabolic endotoxemia.[51] Another study demonstrated that supplementation with *Bifidobacterium animalis* subsp *lactis* 420 reduced inflammation and improved insulin in obese and diabetic mice.[52] Again, these selected examples also illustrate that *Bifidobacterium* strains may affect metabolism, not always by inducing a body weight loss but most likely by improving intestinal barrier.

In humans

A limited number of studies have evaluated whether effects observed in rodents can similarly be achieved in humans. Among these studies, the impact of fermentable carbohydrates (including prebiotics) feeding on enteroendocrine hormones such as GLP-1, PYY and ghrelin, the reduced plasma glucose and inflammatory tone has been generally replicated in both healthy or obese humans,[53–55] however, the impact on fat

mass and body weight remain limited.[56] Interestingly, in these studies the gut microbiota composition was not studies, except in Dewulf et al. 2013, who shows that specific bacteria are positively and negatively correlated with fat mass, metabolic endotoxemia and glucose/lipid markers.[56]

A study using synbiotic approaches that is a supplementation with prebiotics and probiotic (inulin-type fructans and *Bifidobacterium longum*) has shown in 66 overweight patients with non-alcoholic steatohepatitis a reduced steatosis, metabolic endotoxemia, insulin resistance, and inflammation.[57] Excluding these studies using prebiotic supplementation, only few studies have reported a beneficial impact of probiotics on obesity and type 2 diabetes in humans, with again a certain strain specificity (for review[58]). More recently, similar to the results obtained in rodents, it has been shown that important variations of *Akkermansia muciniphila* quantity may be observed in the intestine of obese/overweight subjects. Although, no one knows with precision the level of *Akkermansia muciniphila* required to detect beneficial/healthy versus pathological situation, as discussed earlier in this review, Dao et al. have recently demonstrated in human that below a given fecal amount of *Akkermansia muciniphila* obese/overweight subjects were less disposed to respond to the beneficial effect of a caloric restriction diet in terms of improved cardiometabolic risk factors (i.e., plasma cholesterol, inflammation, insulin resistance and glycemia).[26]

Bariatric surgery induces substantial shifts in gut microbiota composition

Gut microbiota changes have been thoroughly assessed in bariatric interventions both in animal models and humans. In general, bariatric surgery leads to a dramatic

improvement of pre-surgical obesity co-morbidities, with some differences observed between the types of bariatric interventions. The gastric band, for example, leads to a more attenuated WL than sleeve, although they are both considered restrictive procedures. Roux-en-Y gastric bypass (RYGB) leads to the most important changes in health outcomes, potentially due to a change in the gut architecture and gut hormonal secretion, together with extensive WL (**Table 2**). This particular intervention causes greater improvements in type 2 diabetes and other obesity co-morbidities.[59] The effect of bariatric surgery on health has been extensively reviewed in previous publications. [60–63]

Rodent models

Studies in mice that have compared different bariatric surgery procedures with non-operated or SHAM operated mice have allowed the definition of surgery-specific changes in gut microbiota. Liou et al. compared mice that had undergone RYGB, non-operated controls weight matched to the RYGB group, and Sham operated mice fed a HFD *ad libitum*.[64] Gut microbial composition from Sham and weight-matched groups was different from that in the RYGB group. Of interest, among other phylogenetic changes, there was an increase in abundance of *Verrucomicrobia* (genus *Akkermansia*) and *Gammaproteobacteria* (genus *Escherichia*) with RYGB, which correlated with improved metabolic outcomes. Gut microbiota transfers (i.e. transfer of postsurgery caecal content) to germ-free mice led to weight improvement. This study showed that microbial changes in RYGB are due to gastrointestinal reconfiguration and not just to WL, changes in diet or intestinal transection. The RYGB group had the highest fecal energy output.

Vertical sleeve gastrectomy (VSG) is becoming popular practice in bariatric interventions. It was previously believed to be a purely restrictive procedure, but there is now evidence suggesting that several aspects of digestion, bile acid metabolism and gastrointestinal hormonal secretory profile are modified. To this point, it was recently published that circulating bile acids are altered in mice undergoing VSG, which was correlated with shifts in gut microbial composition.[65] Furthermore, knockout of the bile acid receptor FXR reduced WL and clinical improvement.

A recent study by Tremaroli et al. compared phenotypes in mice receiving fecal transfer from morbidly obese women, or women that had undergone either RYGB or vertical banded gastroplasty.[66] One unique feature of this study was that microbiota composition was studied long term, with fecal samples obtained 9 years after surgery, when the women were weight-stable. Changes in microbiota were not only maintained over time, they were also surgery-specific but independent of BMI. Even though the phenotype was transmitted from the two surgical groups to the mice, there were some functional and compositional differences in microbiota, such as higher *Proteobacteria* in the RYGB group, and lower abundance of *E. rectale* and *Roseburia intestinalis* in the sleeve group compared to the obese group. The fecal and circulating metabolite profiles were different between groups. This study provides compelling evidence of the role of microbiota in long term weight maintenance of bariatric patients.

In humans

The potential role of microbiota in human health improvement stemming from bariatric surgery has been recently summarized.[67,68] As in mouse studies, the composition of gut microbiota in humans is extensively changed with bariatric surgery

(**Table 2**). For example, Furet et al. showed important changes in microbiota measured with 16S qPCR, after bypass. This included an increase in *F. prausnitzii*, which was inversely associated with inflammation regardless of diet.[17] Later, Kong et al. published more detailed gut microbiota information on this group obtained with 16S pyrosequencing.[69] This analysis showed that microbial richness increased after RYGB, and that approximately half of the correlations seen between diet and gut microbiota could be explained by dietary intake.

Damms-Machado et al., compared the effect of a very low calorie diet (VLCD) to VSG over 6 months, with 3 patients per group. They saw a reduction in *Firmicutes* to *Bacteroidetes* ratio, less butyrate fermentation, and more NEFA and bile acid secretion in the VSG group.[70] The authors argue that the decrease in proportion of *Firmicutes* would account for the decrease capacity to ferment SCFA, leading to less calorie extraction from diet and therefore greater benefit from the intervention. It is difficult to link this to clinical outcomes because the VSG group was heavier at baseline than the VLCD group.

Other bariatric interventions have included a small number of subjects.[71–73] Their design has been either cross-sectional, or with short-term follow-up (**Table 2**). Some changes in gut microbiota have been consistent, such as a decrease in *Firmicutes* after surgery, increase *Proteobacteria* and a tendency towards an increase in *Verrucomicrobia* (*Akkermansia*).

Most importantly, very few bariatric intervention studies assessing microbiota have included dietary information and food intake behavior or other kinds of environmental exposures. Our group has recently reported that dietary quality in bariatric patients is poor, particularly protein intake.[74] In addition to change in food intake after

bariatric surgery, these subjects also receive protein supplementary that could impact on gut microbiota. Therefore, it will also be important to focus on dietary quality of bariatric patients before and after surgery to optimize response and increase the likelihood of a shift to a healthier gut microbiota.

Interpretation of microbial changes with human bariatric interventions need to be made with caution and with a thorough knowledge of the clinical background of the patients, as morbidly obese populations are usually taking multiple medications. The effect of polypharmacy, including metformin and other diabetes treatments, on the gut microbiota and its relation to health is only now being elucidated.[75,76]

INTEGRATION OF KNOWLEDGE AND POTENTIAL FOR FUTURE

Throughout this review we have discussed the interactions between three main elements: the host, the gut microbiota, and the environment. The advancement of available technologies for the assessment of gut microbiota is key in the work presented here. The field is shifting from targeted measurement of specific bacterial groups to a gut microbiota ecology approach. This is complementary to the thorough analysis of particular species of interest. With these advances in technology, microbiota will be more thoroughly characterized and quantified. This will include RNAseq and more detailed functional annotations. Other relevant measures include the gut environment, architecture and ecosystem, in conjunction with functional characteristics of the gut microbiota as a metabolic organ through the use of metabolomics.

From a clinical point of view, extensive phenotyping of populations is mandatory to identify subgroups that may be responding differently to an intervention. Indeed, even if a population seems uniform in terms of BMI, there is non-negligible heterogeneity in

body composition, which in turn would be associated with different profiles of metabolic health, as explained by Ahima and Lazar.[77] Clinical parameters, pathologies and other traits of the host must be studied in detail to identify subgroups that may respond differently to interventions.

Regarding the environment, there is a wide array of exposures influencing host and gut microbiota that are very difficult to measure. Diet is the factor with the greatest potential to influence the gut microbiota and, although it is often assessed, it is very difficult to measure it reliably. Dietary intake and habits should be routinely taken into consideration in the kinds of interventions we have covered in this review. At the same time, there are many other environmental factors that could be influencing microbiota, including drug intake, pollution and physical activity.

The gut microbiota is at the interphase between environment and host. It is important to study profiles from these three elements in parallel using data integration and systems biology approaches.[78,79] This would allow a more profound understanding of the factors that may be influencing, or may be influenced, by gut microbiota,[80] as well as differentiation of individual subpopulations that may undergo different responses after a WL intervention (**Figure 1**).

Ecosystem modelisation: a first step toward truly personalized nutrition?

An example of a potential approach for personalized improvement in metabolic status can be seen in the recently published work by Shoaie et al.[81] Given the complexity of the intestinal bacterial ecosystem characterized by microbe-microbe interactions, and interactions between microbes, the environment and the biology of the

host, informatics and mathematics experts have used novel approaches to model these interactions. These modelisation approaches aim at better understanding at the individual level the interactions between the microbiota ecosystem and dietary intake, and to infer the potential impact on metabolic health (**Figure 3**). As such, knowledge of the individual composition in gut microbiota lead to the identification of metabolites produced in excess or otherwise deficient and to propose appropriate individualized diet to correct a potential imbalance. Although this approach may seem a bit theoretical, a first step has been taken with the modeling of amino acid exchanges between different bacterial groups. Dietary protein and amino acids are, in fact, important substrates for colonic fermentation, where they serve as a nitrogen source for the microbiota. A model called CASINO (Community And Systems-level INteractive Optimization) was applied to analyze these exchanges in people with enriched or depleted microbiota of the MICRO-Obes study. CASINO was actually able to predict differences in production of SCFA and amino acids (such as phenylalanine and branched chain amino acids) between subjects. Fecal and blood metabolomics analysis allowed validation of the relevance of this theoretical model. Actually subjects with lower microbial richness had a greater elevation of amino acids such as phenylalanine and branched chain amino acids (valine, leucine, isoleucine). Blood elevation of some of these amino acids has been linked to insulin resistance and also identified as risk factor for type 2 diabetes (e.g. phenylalanine). The dietary intervention led to a significant decrease of these metabolites together with increased gut microbiota richness. CASINO also modeled which specific bacterial groups contributed significantly to the production of these “deleterious” metabolites. Finally, by comparing subjects with low or high gut microbiota richness during the

intervention, the model proposed what specific dietary changes (i.e. food categories) individuals with low richness potentially should consume to improve their metabolism.

CONCLUSIONS

Several studies described a positive impact of CR, bariatric surgery and dietary interventions such as prebiotic and probiotic supplementation on diet-induced metabolic disorders in rodents and in humans. Additional studies are warranted to suggest the use of one or another strain as therapeutic tool in the current clinical practice. It is worth noting that evidence suggests that body weight loss is not a prerequisite to observe beneficial impact upon health. This implies that changes in gut microbiota may contribute to the improvement of metabolic disorders via complex mechanisms that can be indirectly related to energy homeostasis.

ACKNOWLEDGEMENTS

PDC is a research associate at FRS-FNRS (Fonds de la Recherche Scientifique), Belgium. AE is postdoctoral researcher at FRS-FNRS, Belgium. PDC is the recipient of grants from FNRS (convention J.0084.15, convention 3.4579.11), PDR (Projet de Recherche, convention: T.0138.14) and ARC (Action de Recherche Concertée - Communauté française de Belgique convention: 12/17-047). This work was supported by the Fonds de la Recherche Scientifique - FNRS for the FRFS-WELBIO under grant: WELBIO-CR-2012S-02R. This work was supported in part by the Funds InBev-Baillet Latour (Grant for Medical Research 2015). PDC is a recipient of an ERC Starting Grant

2013 (European Research Council, Starting grant 336452-ENIGMO). KC and MCD have received funding from the European Union's Seventh Framework Programme for research, technological development and demonstration under grant agreement HEALTH-F4-2012-305312, from Clinical Research programs (PHRC, Microbiota), and National Agency of Research (ANR-MicroObse and Investissement d'Avenir IHU).

REFERENCES

1 Clarke SF, Murphy EF, Nilaweera K, *et al.* The gut microbiota and its relationship to diet and obesity: new insights. *Gut Microbes* 2012;**3**:186–202. doi:10.4161/gmic.20168

2 Hansen TH, Gøbel RJ, Hansen T, *et al.* The gut microbiome in cardio-metabolic health. *Genome Med* 2015;**7**. doi:10.1186/s13073-015-0157-z

3 Tremaroli V, Bäckhed F. Functional interactions between the gut microbiota and host metabolism. *Nature* 2012;**489**:242–9. doi:10.1038/nature11552

4 Alang N, Kelly CR. Weight gain after fecal microbiota transplantation. *Open Forum Infect Dis* 2015;**2**:ofv004. doi:10.1093/ofid/ofv004

5 Delzenne NM, Cani PD, Everard A, *et al.* Gut microorganisms as promising targets for the management of type 2 diabetes. *Diabetologia* 2015;**58**:2206–17. doi:10.1007/s00125-015-3712-7

6 Bäckhed F, Ding H, Wang T, *et al.* The gut microbiota as an environmental factor that regulates fat storage. *Proc Natl Acad Sci U S A* 2004;**101**:15718–23. doi:10.1073/pnas.0407076101

7 Bäckhed F, Manchester JK, Semenkovich CF, *et al.* Mechanisms underlying the resistance to diet-induced obesity in germ-free mice. *Proc Natl Acad Sci U S A* 2007;**104**:979–84. doi:10.1073/pnas.0605374104

8 Turnbaugh PJ, Ley RE, Mahowald MA, *et al.* An obesity-associated gut microbiome with increased capacity for energy harvest. *Nature* 2006;**444**:1027–31. doi:10.1038/nature05414

9 Cani PD, Amar J, Iglesias MA, *et al.* Metabolic endotoxemia initiates obesity and insulin resistance. *Diabetes* 2007;**56**:1761–72. doi:10.2337/db06-1491

10 Vila IK, Badin P-M, Marques M-A, *et al.* Immune cell Toll-like receptor 4 mediates the development of obesity- and endotoxemia-associated adipose tissue fibrosis. *Cell Rep* 2014;**7**:1116–29. doi:10.1016/j.celrep.2014.03.062

11 Everard A, Geurts L, Caesar R, *et al.* Intestinal epithelial MyD88 is a sensor switching host metabolism towards obesity according to nutritional status. *Nat Commun* 2014;**5**:5648. doi:10.1038/ncomms6648

12 Org E, Parks BW, Joo JWJ, *et al.* Genetic and environmental control of host-gut microbiota interactions. *Genome Res* 2015;**25**:1558–69. doi:10.1101/gr.194118.115

13 Zhang C, Li S, Yang L, *et al.* Structural modulation of gut microbiota in life-long calorie-restricted mice. *Nat Commun* 2013;**4**. doi:10.1038/ncomms3163

14 Arumugam M, Raes J, Pelletier E, *et al.* Enterotypes of the human gut microbiome. *Nature* 2011;**473**:174–80. doi:10.1038/nature09944

15 Wu GD, Chen J, Hoffmann C, *et al.* Linking long-term dietary patterns with gut microbial enterotypes. *Science* 2011;**334**:105–8. doi:10.1126/science.1208344

16 Le Chatelier E, Nielsen T, Qin J, *et al.* Richness of human gut microbiome correlates with metabolic markers. *Nature* 2013;**500**:541–6. doi:10.1038/nature12506

17 Furet J-P, Kong L-C, Tap J, *et al.* Differential adaptation of human gut microbiota to bariatric surgery-induced weight loss: links with metabolic and low-grade inflammation markers. *Diabetes* 2010;**59**:3049–57. doi:10.2337/db10-0253

18 Ley RE, Bäckhed F, Turnbaugh P, *et al.* Obesity alters gut microbial ecology. *Proc Natl Acad Sci U S A* 2005;**102**:11070–5. doi:10.1073/pnas.0504978102

19 Munukka E, Wiklund P, Pekkala S, *et al.* Women with and without metabolic disorder differ in their gut microbiota composition. *Obes Silver Spring Md* 2012;**20**:1082–7. doi:10.1038/oby.2012.8

20 Turnbaugh PJ, Hamady M, Yatsunenko T, *et al.* A core gut microbiome in obese and lean twins. *Nature* 2009;**457**:480–4. doi:10.1038/nature07540

21 Schwiertz A, Taras D, Schäfer K, *et al.* Microbiota and SCFA in lean and overweight healthy subjects. *Obes Silver Spring Md* 2010;**18**:190–5. doi:10.1038/oby.2009.167

22 Cotillard A, Kennedy SP, Kong LC, *et al.* Dietary intervention impact on gut microbial gene richness. *Nature* 2013;**500**:585–8. doi:10.1038/nature12480

23 Sokol H, Pigneur B, Watterlot L, *et al.* Faecalibacterium prausnitzii is an anti-inflammatory commensal bacterium identified by gut microbiota analysis of Crohn disease patients. *Proc Natl Acad Sci U S A* 2008;**105**:16731–6. doi:10.1073/pnas.0804812105

24 Munukka E, Pekkala S, Wiklund P, *et al.* Gut-adipose tissue axis in hepatic fat accumulation in humans. *J Hepatol* 2014;**61**:132–8. doi:10.1016/j.jhep.2014.02.020

25 Zhang X, Shen D, Fang Z, *et al.* Human gut microbiota changes reveal the progression of glucose intolerance. *PloS One* 2013;**8**:e71108. doi:10.1371/journal.pone.0071108

26 Dao MC, Everard A, Aron-Wisnewsky J, *et al.* Akkermansia muciniphila and improved metabolic health during a dietary intervention in obesity: relationship with gut microbiome richness and ecology. *Gut* Published Online First: 22 June 2015. doi:10.1136/gutjnl-2014-308778

27 Kong LC, Wuillemin P-H, Bastard J-P, *et al.* Insulin resistance and inflammation predict kinetic body weight changes in response to dietary weight loss and maintenance in overweight and obese subjects by using a Bayesian network approach. *Am J Clin Nutr* 2013;**98**:1385–94. doi:10.3945/ajcn.113.058099

28 Kong LC, Holmes BA, Cotillard A, *et al.* Dietary patterns differently associate with inflammation and gut microbiota in overweight and obese subjects. *PloS One* 2014;**9**:e109434. doi:10.1371/journal.pone.0109434

29 Duncan SH, Belenguer A, Holtrop G, *et al.* Reduced Dietary Intake of Carbohydrates by Obese Subjects Results in Decreased Concentrations of Butyrate and Butyrate-Producing Bacteria in Feces. *Appl Environ Microbiol* 2007;**73**:1073–8. doi:10.1128/AEM.02340-06

30 Russell WR, Gratz SW, Duncan SH, *et al.* High-protein, reduced-carbohydrate weight-loss diets promote metabolite profiles likely to be detrimental to colonic health. *Am J Clin Nutr* 2011;**93**:1062–72. doi:10.3945/ajcn.110.002188

31 Brinkworth GD, Noakes M, Clifton PM, *et al.* Comparative effects of very low-carbohydrate, high-fat and high-carbohydrate, low-fat weight-loss diets on bowel habit and faecal short-chain fatty acids and bacterial populations. *Br J Nutr* 2009;**101**:1493–502. doi:10.1017/S0007114508094658

32 Nadal I, Santacruz A, Marcos A, *et al.* Shifts in clostridia, bacteroides and immunoglobulin-coating fecal bacteria associated with weight loss in obese adolescents. *Int J Obes 2005* 2009;**33**:758–67. doi:10.1038/ijo.2008.260

33 Santacruz A, Marcos A, Wärnberg J, *et al.* Interplay between weight loss and gut microbiota composition in overweight adolescents. *Obes Silver Spring Md* 2009;**17**:1906–15. doi:10.1038/oby.2009.112

34 Fei N, Zhao L. An opportunistic pathogen isolated from the gut of an obese human causes obesity in germfree mice. *ISME J* 2013;**7**:880–4. doi:10.1038/ismej.2012.153

35 Ridaura VK, Faith JJ, Rey FE, *et al.* Gut microbiota from twins discordant for obesity modulate metabolism in mice. *Science* 2013;**341**:1241214. doi:10.1126/science.1241214

36 Bindels LB, Delzenne NM, Cani PD, *et al.* Towards a more comprehensive concept for prebiotics. *Nat Rev Gastroenterol Hepatol* 2015;**12**:303–10. doi:10.1038/nrgastro.2015.47

37 Cani PD, Dewever C, Delzenne NM. Inulin-type fructans modulate gastrointestinal peptides involved in appetite regulation (glucagon-like peptide-1 and ghrelin) in rats. *Br J Nutr* 2004;**92**:521–6.

38 Delzenne NM, Cani PD, Daubioul C, *et al.* Impact of inulin and oligofructose on gastrointestinal peptides. *Br J Nutr* 2005;**93 Suppl 1**:S157–61.

39 Cani PD, Neyrinck AM, Fava F, *et al.* Selective increases of bifidobacteria in gut microflora improve high-fat-diet-induced diabetes in mice through a mechanism associated with endotoxaemia. *Diabetologia* 2007;**50**:2374–83. doi:10.1007/s00125-007-0791-0

40 Everard A, Lazarevic V, Gaïa N, *et al.* Microbiome of prebiotic-treated mice reveals novel targets involved in host response during obesity. *ISME J* 2014;**8**:2116–30. doi:10.1038/ismej.2014.45

41 Everard A, Belzer C, Geurts L, *et al.* Cross-talk between Akkermansia muciniphila and intestinal epithelium controls diet-induced obesity. *Proc Natl Acad Sci U S A* 2013;**110**:9066–71. doi:10.1073/pnas.1219451110

42 Anhê FF, Roy D, Pilon G, *et al.* A polyphenol-rich cranberry extract protects from diet-induced obesity, insulin resistance and intestinal inflammation in association with increased Akkermansia spp. population in the gut microbiota of mice. *Gut* 2015;**64**:872–83. doi:10.1136/gutjnl-2014-307142

43 Hill C, Guarner F, Reid G, *et al.* Expert consensus document. The International Scientific Association for Probiotics and Prebiotics consensus statement on the scope and appropriate use of the term probiotic. *Nat Rev Gastroenterol Hepatol* 2014;**11**:506–14. doi:10.1038/nrgastro.2014.66

44 Cani PD, Van Hul M. Novel opportunities for next-generation probiotics targeting metabolic syndrome. *Curr Opin Biotechnol* 2015;**32**:21–7. doi:10.1016/j.copbio.2014.10.006

45 Park S, Bae J-H. Probiotics for weight loss: a systematic review and meta-analysis. *Nutr Res* 2015;**35**:566–75. doi:10.1016/j.nutres.2015.05.008

46 Yun SI, Park HO, Kang JH. Effect of Lactobacillus gasseri BNR17 on blood glucose levels and body weight in a mouse model of type 2 diabetes. *J Appl Microbiol* 2009;**107**:1681–6. doi:10.1111/j.1365-2672.2009.04350.x

47 Takemura N, Okubo T, Sonoyama K. Lactobacillus plantarum strain No. 14 reduces adipocyte size in mice fed high-fat diet. *Exp Biol Med Maywood NJ* 2010;**235**:849–56. doi:10.1258/ebm.2010.009377

48 Arora T, Anastasovska J, Gibson G, *et al.* Effect of Lactobacillus acidophilus NCDC 13 supplementation on the progression of obesity in diet-induced obese mice. *Br J Nutr* 2012;**108**:1382–9. doi:10.1017/S0007114511006957

49 Naito E, Yoshida Y, Makino K, *et al.* Beneficial effect of oral administration of Lactobacillus casei strain Shirota on insulin resistance in diet-induced obesity mice. *J Appl Microbiol* 2011;**110**:650–7. doi:10.1111/j.1365-2672.2010.04922.x

50 Moya-Pérez A, Neef A, Sanz Y. Bifidobacterium pseudocatenulatum CECT 7765 Reduces Obesity-Associated Inflammation by Restoring the Lymphocyte-Macrophage Balance and Gut Microbiota Structure in High-Fat Diet-Fed Mice. *PloS One* 2015;**10**:e0126976. doi:10.1371/journal.pone.0126976

51 Chen JJ, Wang R, Li X, *et al.* Bifidobacterium longum supplementation improved high-fat-fed-induced metabolic syndrome and promoted intestinal Reg I gene expression. *Exp Biol Med Maywood NJ* 2011;**236**:823–31. doi:10.1258/ebm.2011.010399

52 Amar J, Chabo C, Waget A, *et al.* Intestinal mucosal adherence and translocation of commensal bacteria at the early onset of type 2 diabetes: molecular mechanisms and probiotic treatment. *EMBO Mol Med* 2011;**3**:559–72. doi:10.1002/emmm.201100159

53 Cani PD, Lecourt E, Dewulf EM, *et al.* Gut microbiota fermentation of prebiotics increases satietogenic and incretin gut peptide production with consequences for appetite sensation and glucose response after a meal. *Am J Clin Nutr* 2009;**90**:1236–43. doi:10.3945/ajcn.2009.28095

54 Pedersen C, Lefevre S, Peters V, *et al.* Gut hormone release and appetite regulation in healthy non-obese participants following oligofructose intake. A dose-escalation study. *Appetite* 2013;**66**:44–53. doi:10.1016/j.appet.2013.02.017

55 Bonsu NKA, Johnson CS, McLeod KM. Can dietary fructans lower serum glucose? *J Diabetes* 2011;**3**:58–66. doi:10.1111/j.1753-0407.2010.00099.x

56 Dewulf EM, Cani PD, Claus SP, *et al.* Insight into the prebiotic concept: lessons from an exploratory, double blind intervention study with inulin-type fructans in obese women. *Gut* 2013;**62**:1112–21. doi:10.1136/gutjnl-2012-303304

57 Malaguarnera M, Vacante M, Antic T, *et al.* Bifidobacterium longum with fructo-oligosaccharides in patients with non alcoholic steatohepatitis. *Dig Dis Sci* 2012;**57**:545–53. doi:10.1007/s10620-011-1887-4

58 Druart C, Alligier M, Salazar N, *et al.* Modulation of the gut microbiota by nutrients with prebiotic and probiotic properties. *Adv Nutr Bethesda Md* 2014;**5**:624S – 633S. doi:10.3945/an.114.005835

59 Still CD, Wood GC, Chu X, *et al.* Clinical factors associated with weight loss outcomes after Roux-en-Y gastric bypass surgery. *Obes Silver Spring Md* 2014;**22**:888–94. doi:10.1002/oby.20529

60 Aron-Wisnewsky J, Julia Z, Poitou C, *et al.* Effect of bariatric surgery-induced weight loss on SR-BI-, ABCG1-, and ABCA1-mediated cellular cholesterol efflux in obese women. *J Clin Endocrinol Metab* 2011;**96**:1151–9. doi:10.1210/jc.2010-2378

61 Dixon JB, Zimmet P, Alberti KG, *et al.* Bariatric surgery: an IDF statement for obese Type 2 diabetes. *Diabet Med J Br Diabet Assoc* 2011;**28**:628–42. doi:10.1111/j.1464-5491.2011.03306.x

62 Miras AD, le Roux CW. Metabolic surgery: shifting the focus from glycaemia and weight to end-organ health. *Lancet Diabetes Endocrinol* 2014;**2**:141–51. doi:10.1016/S2213-8587(13)70158-X

63 Sjöström L, Peltonen M, Jacobson P, *et al.* Bariatric surgery and long-term cardiovascular events. *JAMA* 2012;**307**:56–65. doi:10.1001/jama.2011.1914

64 Liou AP, Paziuk M, Luevano J-M, *et al.* Conserved shifts in the gut microbiota due to gastric bypass reduce host weight and adiposity. *Sci Transl Med* 2013;**5**:178ra41. doi:10.1126/scitranslmed.3005687

65 Ryan KK, Tremaroli V, Clemmensen C, *et al.* FXR is a molecular target for the effects of vertical sleeve gastrectomy. *Nature* 2014;**509**:183–8. doi:10.1038/nature13135

66 Tremaroli V, Karlsson F, Werling M, *et al.* Roux-en-Y Gastric Bypass and Vertical Banded Gastroplasty Induce Long-Term Changes on the Human Gut Microbiome Contributing to Fat Mass Regulation. *Cell Metab* 2015;**22**:228–38. doi:10.1016/j.cmet.2015.07.009

67 Aron-Wisnewsky J, Dore J, Clement K. The importance of the gut microbiota after bariatric surgery. *Nat Rev Gastroenterol Hepatol* 2012;**9**:590+.

68 Madsbad S, Dirksen C, Holst JJ. Mechanisms of changes in glucose metabolism and bodyweight after bariatric surgery. *Lancet Diabetes Endocrinol* 2014;**2**:152–64. doi:10.1016/S2213-8587(13)70218-3

69 Kong L-C, Tap J, Aron-Wisnewsky J, *et al.* Gut microbiota after gastric bypass in human obesity: increased richness and associations of bacterial genera with adipose tissue genes. *Am J Clin Nutr* 2013;**98**:16–24. doi:10.3945/ajcn.113.058743

70 Damms-Machado A, Mitra S, Schollenberger AE, *et al.* Effects of Surgical and Dietary Weight Loss Therapy for Obesity on Gut Microbiota Composition and Nutrient Absorption. *BioMed Res Int* 2015;**2015**:e806248. doi:10.1155/2015/806248

71 Graessler J, Qin Y, Zhong H, *et al.* Metagenomic sequencing of the human gut microbiome before and after bariatric surgery in obese patients with type 2 diabetes: correlation with inflammatory and metabolic parameters. *Pharmacogenomics J* 2013;**13**:514–22. doi:10.1038/tpj.2012.43

72 Ward EK, Schuster DP, Stowers KH, *et al.* The Effect of PPI Use on Human Gut Microbiota and Weight Loss in Patients Undergoing Laparoscopic Roux-en-Y Gastric Bypass. *Obes Surg* Published Online First: 22 May 2014. doi:10.1007/s11695-014-1275-1

73 Zhang H, DiBaise JK, Zuccolo A, *et al.* Human gut microbiota in obesity and after gastric bypass. *Proc Natl Acad Sci U S A* 2009;**106**:2365–70. doi:10.1073/pnas.0812600106

74 Verger EO, Aron-Wisnewsky J, Dao MC, *et al.* Micronutrient and Protein Deficiencies After Gastric Bypass and Sleeve Gastrectomy: a 1-year Follow-up. *Obes Surg* Published Online First: 24 July 2015. doi:10.1007/s11695-015-1803-7

75 Shin N-R, Lee J-C, Lee H-Y, *et al.* An increase in the Akkermansia spp. population induced by metformin treatment improves glucose homeostasis in diet-induced obese mice. *Gut* 2014;**63**:727–35. doi:10.1136/gutjnl-2012-303839

76 Tilg H, Moschen AR. Microbiota and diabetes: an evolving relationship. *Gut* 2014;**63**:1513–21. doi:10.1136/gutjnl-2014-306928

77 Ahima RS, Lazar MA. Physiology. The health risk of obesity--better metrics imperative. *Science* 2013;**341**:856–8. doi:10.1126/science.1241244

78 Beebe K, Sampey B, Watkins SM, *et al.* Understanding the apothecaries within: the necessity of a systematic approach for defining the chemical output of the human microbiome. *Clin Transl Sci* 2014;**7**:74–81. doi:10.1111/cts.12131

79 Kussmann M, Morine MJ, Hager J, *et al.* Perspective: a systems approach to diabetes research. *Front Genet* 2013;**4**:205. doi:10.3389/fgene.2013.00205

80 Heinken A, Thiele I. Systems biology of host-microbe metabolomics. *Wiley Interdiscip Rev Syst Biol Med* 2015;**7**:195–219. doi:10.1002/wsbm.1301

81 Shoaie S, Ghaffari P, Kovatcheva-Datchary P, *et al.* Quantifying Diet-Induced Metabolic Changes of the Human Gut Microbiome. *Cell Metab* 2015;**22**:320–31. doi:10.1016/j.cmet.2015.07.001

TABLES

Table 1. Overview of CR studies reporting changes in microbiome composition and/or function, along with clinical outcomes and/or dietary intake.

							MICROBIOTA VS.:		
First author	**Study design**	**Population**	**Method**	**Diet reported?**	**Changes in gut microbiome**	**Clinical or dietary outcome associated with gut microbial changes**	**CLINICAL**	**METABOLITES**	**DIET**
Ley et al. (2006)	CR intervention: two diets (low carb or low fat) for 1 yr.	12 obese adults	16S rRNA sequencing	No	↑ *Bacteroidetes:Firmicutes*	Increase in *Bacteroidetes* abundance correlated with %WL.	only weight loss reported		
Duncan et al. (2007)	Randomized cross-over study: two 4-wk diets high in protein with low or medium carb, with a 3-day high carb maintenance diet before each regime.	19 obese men, no co-morbidities	FISH targeting 16S rRNA of 10 dominant bacterial groups and total bacteria	Yes	↑ total bacteria in maintenance diet. ↓ carb intake correlated with ↓ *Roseburia* spp/*E. rectale* group and *Bifidobacteria*. Abundance of other groups did not change.	●When compared to medium or low carb diets, SCFA content was higher in the maintenance diet. ●Butyrate production was positively correlated with carb intake.			
Santacruz et al. (2009)	CR and exercise intervention for 10 wks.	36 overweight /obese adolescents. There were low (WL<2kg) and high (WL>4kg0 responders	16S rRNA qPCR of 11 bacterial groups and total bacteria	Yes	Different *E. coli, B. longum and B. adolescentis* between low and high responders before and after the intervention. Greater change in bacterial group abundance for high responders.	●*Bacteroides* and *Lactobacillus* groups were positively correlated, and *E. coli* inversely correlated, with WL. ●Complex carb intake was negatively correlated with *B. fragilis.* ●There was no difference in dietary intake between groups.			
Nadal et al. (2009)	CR and exercise intervention for 10 wks.	39 overweight /obese adolescents. There were low (WL<2.5kg) and high (WL>4kg0	FISH targeting 16S rRNA of 11 dominant bacterial groups and total bacteria	Yes	In high responders: ↓ *C.histolyticum, E. rectale/C. coccoides, and C. lituseburense;* ↑ *Bacteroides/Prevotella.* No changes were seen for low	●Change in *C. histolyticum* and *E. rectale/C. coccoides* were positively correlated with WL. ●*E. rectale/C. coccoides* correlated with BMI z-score reduction. ●Changes in fasting glucose correlated positively with *E. rectale/C.coccoides* and negatively with Gram-negative bacteria. ●Changes in LDL cholesterol were inversely			only total calories reported

		responders			responders.	correlated with *C. lituseburense.*			
Brinkworth et al. (2009)	CR intervention: low-carb/high-fat vs. high-carb/low-fat diet for 8 wks.	91 overweight /obese adults	Culture system used for detection of *Bifidobacteria*, *Lactobacillus*, *E. coli*, total anaerobes and aerobes	Yes	↓ *Bifidobacteria* in low carb/high fat diet group at 8 wks. ↑ Fecal anaerobes in high carb/low fat diet group at 8 wks.	●Low carb/high fat group had more WL than high carb/low fat group. ●Fecal output and concentration of fecal butyrate, acetate, propionate, and total SCFA were lower in the low carb/high fat diet group.	only weight loss reported		
Wu et al. (2011)	Cross-sectional study (COMBO study). Subgroup was part of a 10-day controlled feeding study (CAFE study) (randomized to high-fat/low-fiber or low-fat/high-fiber).	Normal weight to obese subjects with no chronic co-morbidities. COMBO: N=98, 2-50y; CAFE: N=10, 18-40y	16S rRNA sequencing, with a subset of shotgun metagenomics. Rectal biopsy for CAFE.	Yes	Two main enterotypes identified: *Bacteroides* and *Prevotella*. In CAFE, microbiome composition shifted after 24h, but intra-subject variation < inter-individual variation, and enterotype classification remained constant.	●Nutrient groups had opposing correlation patterns with enterotypes: fats vs. plant-derived nutrients, proteins/amino acids vs. carbs, and fats vs. carbs. ●A more Western-style diet profile in *Bacteroides* enterotype. Carb-rich diet in *Prevotella* enterotype. ●Taxa that correlated with fat-derived nutrients and with calories were also correlated with BMI. ●Associations between nutrients and enterotypes were seen for the FFQ data (habitual diet) and not for 24h recalls. ●Demographic data recorded (ethnicity, income, etc) but no associations seen with BMI.	only BMI		
Walker et al. (2011)	CR intervention: 1-wk maintenance diet followed by 3-wk diet high in resistant starch or non-starch polysaccharides using a cross-over design. Finally, 3-wk high-protein CR diet was consumed.	14 overweight men	16S rRNA sequencing, 16S qPCR, and denaturing gradient get electrophoresis	Yes	Microbiota composition changed rapidly with diet, but inter-individual differences > within-subject changes. ↑ *E. rectale, C. aerofaciens,* and *R. Bromii* with resistant starch diet.	●Starch digestibility was greatest for non-starch polysaccharides. ●No emphasis on clinical data analysis.			
Russell et al. (2011)	Cross-over study: 1-wk maintenance diet followed by 4-wk high	17 obese men, no co-morbidities	FISH for detection of dominant bacterial groups and	Yes	↓ *Roseburia/E.rectale* group and *Bacteroides* spp. in high protein/low carb diet.	●Total SCFA were lower in high protein/low card diet. ●Both diets increased fecal branched-chain fatty acids, phenylacetic acid and N-nitroso compounds, and decreased butyrate and			

	protein/moderate carb or high protein/low carb diet.		total bacteria			fiber-derived antioxidants.			
Kong et al. (2013)	CR intervention: 6-wk CR followed by 6wk WS. Diet was high in protein and fiber, with low glycemic index. Subjects were clustered into 3 groups according to their WL response.	49 overweight /obese adults	16S rRNA qPCR of 7 dominant bacterial groups	Yes	↑ baseline *Lactobacillus/Leuconostoc/Pediococcus* group in non-responders.	●*Lactobacillus/Leuconostoc/Pediococcus* group abundance correlated to weight regain during the WS period, and with consumption of starch in the diet. ●Weight regain profiles could not be fully explained by differences in clinical parameters or dietary intake.			
Cotillard et al. (2013)	CR intervention: 6-wk CR followed by 6wk WS. Diet was high in protein and fiber, with low glycemic index.	49 overweight /obese adults	Shotgun metagenomic sequencing	Yes	↑ microbial richness in subjects with low baseline gene richness. 26 out of 39 gene clusters varied significantly with time; ↓ *E. rectale* and *Bifidobacterium* spp. ↓ several gene clusters during WS.	●Higher baseline microbial richness correlated with improved metabolic status after intervention. ●Change in abundance of the 26 gene clusters were correlated with changes in diet, clinical outcomes, or both.			
Dao et al. (2015)	CR intervention: 6-wk CR followed by 6wk WS. Diet was high in protein and fiber, with low glycemic index.	49 overweight /obese adults	Shotgun metagenomic sequencing and 16S rRNA qPCR of *A. muciniphila*	Yes	↓ *A. muciniphila*in subjects with highest baseline abundance, but it remained 100 times more abundant than in subjects with low baseline abundance. There was a core to 26 MGS associated with *A. muciniphila* abundance at least one point during the intervention.	●Higher baseline *A. muciniphila* abundance was associated with a metabolically healthier status and with better outcomes from the dietary intervention. ●The most metabolically healthy subgroup was characterized by higher *A. muciniphila* abundance and microbial richness. ●Baseline correlation between *A. muciniphila* and serum acetate, which decreased after the intervention. ●No correlation between *A. muciniphila* and diet, including diet quality index.			

Carb, carbohydrate; CR, calorie restriction; sAT, subcutaneous adipose tissue; SCFA, short chain fatty acids; wk(s), week(s); WL, weight loss; WS, weight stabilization; yr, year

Table 2. Effect of bariatric intervention on gut microbiota composition in humans.

							MICROBIOTA VS.:		
First author	**Study design**	**Population**	**Method**	**Diet reported?**	**Changes in gut microbiome**	**Clinical or dietary outcome associated with gut microbial changes**	**CLINICAL**	**METABOLITES**	**DIET**
Damms-Machado et al. (2015)	Comparison of LSG vs. VLCD, 6-mo follow up.	10 morbidly obese women, but microbiome data available for 6 (3 VLCD, 3 LSG).	Shotgun metagenomic sequencing	No	↓ *Bacteroidetes* in VLCD group; ↓ *Firmicutes* in LSG group. In LSG, ↓ *Firmicutes:Bacteroidetes, E. rectale,* and *F. prausnitzii* and ↑ *F. prausnitzii.* ↑ Several *Firmicutes* species in VLCD.	●The VLCD had an improvement in blood lipids while the LSG did not. ●Metabolic capacity for butyrate fermentation was increased for the VLCD group after the intervention, but there was no difference in SCFA between groups. ●Fecal excretion of NEFA and bile acids was increased after LSG.			
Zhang et al. (2009)	Cross-sectional comparison of lean, obese and RYGB patients.	3 lean, 3 morbidly obese, 3 unrelated BS patients 8-15 mo after surgery.	16S rRNA sequencing	No	↓ *Firmicutes,* ↑ *Gammaproteobacteria* in RYGB. ↑ *Prevotellaceae* (H2 producers) and *Archaea* (H2 consumers) in obese. *Verrucomicrobia* variable in normal weight group, undetectable in obese group and highest in RYGB.	None reported.			
Furet et al. (2010)	Bariatric intervention (RYGB) with 3 and 6-mo follow-up, and comparison to lean controls.	13 lean and 30 morbidly obese adults (7 with T2D)	16S rRNA qPCR of total bacteria and 7 select bacterial groups	Yes	↓ *F. prausnitzii* in obese diabetic patients. ↓ *Bacteroides/Prevotella* group in both obese groups. After RYGB: ↑ *Bacteroides/Prevotella and E. coli;* ↓ *Bifidobacterium* and *Lactobacillus/Leuconostoc/Pediococcus* groups. *F. prausnitzii* ↑ at 3 mo and remained stable at 6 mo.	● *F. prausnitzii* inversely related to inflammation independently of diet. There was an inverse correlation between Leptin decrease and E. coli increase after surgery. ●Some of the associations were observed between gut microbiota, corpulence and energy intake.			only total calories reported
Kong et al. (2013)	Bariatric intervention (RYGB) with 3 and 6-mo follow-up.	30 morbidly obese adults (7 with T2D)	16S rRNA sequencing	Yes	↑ microbial richness 3 mos post-surgery and then stabilized. Microbiome composition shifted throughout intervention. ↑ *Bacteroides, Escherichia, and Alistipes* increased;↓	●There were more correlations between microbial genera and sAT gene expression 3 mos after RYGB. ●Changes in the abundance of 14 discriminant genera were associated with changes in clinical parameters and sAT			only total calories reported

					Lactobacillus, Dorea, Blautia,and Bifidobacterium.	gene expression, but most of these genera were also associated with calorie intake.			
Graessler et al. (2013)	Bariatric intervention (RYGB) with 3-mo follow-up.	6 morbidly obese adults (5 with T2D). Lean controls for microbiome analysis only.	Shotgun metagenomic sequencing	No	Microbial composition shifted after BS, including changes in 22 microbial species. ↑ obese vs. lean differences after surgery. ↓ *Firmicutes* and *Bacteroidetes;* ↑ *Proteobacteria* and *Verrucomicrobia.* Some species level changes: ↓ *F. prausnitzii* and ↑ *A. mucniphila*.	●From PCA analysis, species from component 1 (characterized by *Enterobacter cancerogenus*) were correlated to BMI and CRP. Most correlations observed between CRP and bacterial species were BMI-dependent. ●There were 10 species associated with blood lipids and 2 with HbA1c and *F. prausnitzii* correlated with fasting glucose.			
Ward et al. (2014)	Bariatric intervention (RYGB) measuring effect of PPI use on gut microbiota before and 6 mo after RYGB.	8 morbidly obese adults	16S rRNA sequencing	No	↑ *Firmicutes* ↓ *Bacteroides* pre-surgery in PPI users. ↑ *Akkermansia* abundance pre-surgery in PPI users and increased in both groups.	PPI users tended to have less excess weight loss than non-users.	only weight loss reported		

HbA1c, hemoglobin A1c; LSG, laparoscopic sleeve gastrectomy; PCA, principal component analysis; PPI, proton pump inhibitor; RYGB, Roux-en-Y gastric bypass; T2D, type 2 diabetes; VLCD, very low calorie diet; yr, year

FIGURE LEGENDS

Figure 1. Comparing responses to weight loss interventions through extensive phenotyping and data integration. There are phenotypic and behavioral traits that differentiate responders vs. non-responders to weight loss interventions. These differences can be compared 1) at baseline, between responders (status Y) and non-responders (status X) for prediction (yellow profile vs. orange profile), and 2) before vs. after the intervention (yellow profile vs. blue profile) to study mechanisms that may be involved in a good response to the intervention. Environment may refer to diet, exercise, behavior, and other environmental exposures. Omics may refer to genomics, epigenomics, transcriptomics, proteomics and metabolomics in different tissues.

Figure 2. Dietary intervention such as prebiotic supplementation as well as gastric surgery impact gut microbiota and host metabolism and thereby represent interesting approaches for the treatment of obesity and metabolic disorders. Obesity is associated with alterations in metabolism and energy homeostasis. Gastric bypass surgery is associated with changes in gut microbiota composition and metabolic functions and represents one of the more effective approaches to treat obesity and metabolic disorders. Dietary interventions targeting the gut microbiota, such as prebiotics, induce changes in gut microbiota composition that are associated with modification of the secretion of gut enteroendocrine hormones as well as with a reduction in metabolic inflammation, and glucose, lipid and energy homeostasis dysfunctions.

Figure 3. Modelisation of the gut ecosystem as a first step for personalized nutrition. Individuals with low and high gut microbial richness differ in certain clinical parameters, dietary intake and metabolite profile. The CASINO toolbox predicts, at the individual level, differences in metabolite production by gut bacteria and proposes changes in dietary intake for individuals with low gene richness to improve their gut microbiome metabolism. BCAAs, branched chain amino acids.