The role of gut microbiota in human metabolism

Vrieze, A.

Citation for published version (APA):
Vrieze, A. (2013). The role of gut microbiota in human metabolism
CHAPTER 9

SUMMARY & FINAL CONSIDERATION
This thesis focussed on the role of gut microbiota in health and disease. The main focus of this thesis was to study the putative role of the gut microbiota in the development of human pathophysiology, including obesity and insulin resistance. Moreover, our most important clinical question was whether gut microbiota were causally involved in the occurrence of metabolic disease in humans by applying fecal microbiota transplantation. This procedure was originally developed for patients with recurrent *Clostridium difficile* infection, as described in chapter 3, but we also applied this procedure to subjects with metabolic syndrome.

In chapter 2 the relatively new concept of the gut microbiota and its possible relationship with obesity is described. Recent studies have shown that obese humans are characterized by a different gut microbiota composition and these data suggest that gut microbiota may play a role in the development of obesity and insulin resistance. Also animal studies revealed a specific relationship between the gut microbiota and human metabolism. Germ-free mice were shown to be protected from obesity and insulin resistance, whereas their colonization with microbiota derived from obese mice resulted in increased body fat content and insulin resistance. In line with the idea of causation, colonization of germ-free mice with an ‘obese’ microbiota caused a greater increase in body fat compared to colonization with a ‘lean’ microbiota.

Modulators that can influence the composition of the gut microbiota and subsequent metabolic traits are more elaborately reviewed in chapter 3. The influence of short-chain fatty acids, bile acids, prebiotics, probiotics, antibiotics and fecal transplant are discussed. We also describe how the specific changes in gut microbiota may affect metabolism and how these findings could be translated into novel therapeutic pathways for obesity and type 2 diabetes.

One possible way to modify the gut microbiota is fecal microbiota transplantation. In 1958, the surgeon Eiseman treated four patients with severe antibiotic-induced colitis with an enema that consisted of donor feces. Following this initial publication, infusion of feces from healthy donors has been reported as an effective treatment of recurrent *Clostridium difficile* infection in more than 300 patients. However, experience with
this procedure was limited by a lack of randomized trials supporting its efficacy and the non-appealing nature of the treatment. In chapter 4 the conventional antibiotic treatment was compared with the infusion of a donor feces solution in patients with recurrent *Clostridium difficile* infection. This study revealed that treatment with donor feces was superior to vancomycin. The mechanism underlying the efficacy of donor feces infusion is likely the restoration of the normal microbiota as a host-defense against *Clostridium difficile* and thus reveals an important role for intestinal microbiota in human pathophysiology and pathology.

As described in chapter 2 & 3, previous studies have shown that obesity and insulin resistance are associated with an altered composition of the gut microbiota. Taking a reductionist approach, as we used in chapter 4, restoration of a ‘lean’ gut microbiota may ameliorate obesity and insulin resistance in subjects with metabolic syndrome. In chapter 5 we studied the effects of infusion of ‘lean’ donor feces on gut microbiota composition and glucose metabolism. Eighteen male subjects with metabolic syndrome underwent bowel lavage and were randomized to allogenic (from male lean donors with body mass index<23 kg/m²) or autologous (reinfusion of own gut microbiota) microbial transfer. We found an improvement in peripheral insulin sensitivity 6 weeks after allogenic gut microbiota infusion, along with increased levels of (small) intestinal butyrate-producing intestinal microbiota. In conclusion, our data point toward a regulating role for butyrate produced by (small) intestinal microbiota leading to an improvement in insulin sensitivity. Moreover, we identified the specific (small) intestinal bacterial species responsible for this improvement and are currently testing the efficacy of oral administration of these microbiota as therapeutic agent to increase insulin sensitivity in subjects with insulin resistance.

In chapter 6 we further expand our investigation of manipulating the gut microbiota. In this chapter we examined the short term effects of oral broad spectrum antibiotic treatment on gut microbiota composition and subsequent bile acid- and glucose metabolism in humans. In this controlled trial, 20 male obese subjects with the metabolic syndrome were randomised to 7 days of amoxicillin or 7 days of vancomycin. Vancomycin reduced fecal microbial diversity with a decrease of
gram-positive bacteria and a compensatory increase in gram-negative bacteria. We also found an impaired bile acid dehydroxylation and subsequent decreased insulin sensitivity. This randomised study thus provides further evidence on the involvement of gut microbiota and bile acids in the glucose metabolism of subjects with the metabolic syndrome.

In chapter 7 we examined the role of gut microbiota composition in the inflammation of visceral adipose tissue. We hypothesized that altered fecal microbiota composition induces visceral adipose cell inflammation by enhanced bacterial translocation. Indeed, we were able to identify subtle differences in gut microbiota composition in subjects with high endotoxin plasma load, compared to low endotoxin plasma load. CD-68 staining of the fat biopsies further showed that gram negative endotoxemia was also associated with the degree of macrophage accumulation, i.e. inflammation. As we also showed that bacterial DNA was present in human visceral adipose tissue, we conclude that plasma LBP/endotoxin levels might be a marker of human visceral adipose tissue inflammation. Our finding of bacterial gut microbiota derived DNA being present in visceral adipose tissue lends further support to this hypothesis and is topic of ongoing research at our department.

Finally, we reviewed the role of fecal transplant as therapy for microbial disbalance and clinical disease in chapter 8. Fecal microbiota transplantation has been utilized sporadically for over 50 years, but the growing epidemic of Clostridium difficile infection has increased the use of fecal microbiota transplantation. In chapter 4 we already discussed the role of fecal transplant in patients with recurrent Clostridium difficile infection. Growing interest in fecal transplant has largely been driven by new research into the gut microbiota, which is now beginning to be appreciated as a microbial human organ with important roles in immunity and energy metabolism. This new paradigm raises the possibility that many diseases result, at least partially, from microbiota-related dysfunction and calls for investigation regarding the causal relationship between gut microbiota and IBD, IBS, neurodevelopmental disorders, autoimmune diseases and allergic diseases.
Chapter 9

**Final considerations**

Taken together, this thesis support the hypothesis that gut microbiota can be viewed as an ‘exteriorised organ’ that contributes to energy metabolism and the modulation of our immune system. Following Koch’s postulates, it has now been shown that gut microbiota are associated with metabolic disease and that these microbiota can be isolated from a diseased organism (e.g. visceral adipose tissue). The third postulate and ultimate proof of causality have to come from trials showing beneficial effects of treatment with identified bacterial strains aimed at improving insulin sensitivity in humans.