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## **The gut microbiota and diabetic cardiomyopathy in humans**

Marie Bastin, Fabrizio Andreelli

Diabetology–Metabolism Department, Pitié-Salpêtrière Hospital, Assistance Publique  
Hôpitaux de Paris, Sorbonne University, INSERM, NutriOmics Research Team, Paris, France

### **Correspondence to:**

Fabrizio Andreelli

Pitié-Salpêtrière Hospital, Diabetology–Metabolism Department, E3M Building, 47–83  
Boulevard de l’Hôpital, 75651 Paris cédex 13

**Email:** [fabrizio.andreelli@aphp.fr](mailto:fabrizio.andreelli@aphp.fr)

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## **Abstract**

Type 2 diabetes (DT2) increases the risk of cardiovascular events and cardiac insufficiency. This insufficiency is mostly post-ischaemic in nature, but other aetiologies are possible in this high-risk population. In patients with DT2, diabetic cardiomyopathy is a recognized cause of cardiac insufficiency secondary to chronic hyperglycaemia and myocardial lipotoxicity, which promotes cardiomyocyte hypertrophy (and, frequently, apoptosis of these cells), interstitial fibrosis and a decrease in myocardial contractile performance. Several studies have shown that diabetic cardiomyopathy is associated with modifications to the intestinal microbiota, and changes in the synthesis of bacterial metabolites and their diffusion into the host, some of which appear to have direct deleterious effects on cardiac contractility. These findings open up new perspectives for pathophysiological studies by establishing the presence of a ‘microbiota–myocardium’ axis and raising the possibility of innovative new treatments. Correction of intestinal dysbiosis in patients with cardiac insufficiency could, therefore, constitute an innovative therapeutic approach to cases of this disease with a poor prognosis.

**Keywords:** Butyrate; Cardiac insufficiency; Cardiovascular event; Diabetes; Intestinal microbiota; Myocardium; Obesity; TMA/TMAO

## **Introduction**

Cardiometabolic disorders are the most frequently found non-transmissible diseases [1], and many studies have suggested that changes to the microbiota may be contributing to their pathophysiology. The bacterial families comprising the microbiota have now been classified into five phyla: Actinobacteria; Bacteroidetes; Firmicutes; Proteobacteria; and Verrucomicrobia. These phyla are present in roughly similar proportions in healthy individuals, whereas quantitative changes in these proportions have been observed in cardiometabolic diseases. The pioneering work of J.I. Gordon and his team [2] demonstrated an association between obesity and an imbalance between Firmicutes and Bacteroidetes favouring Firmicutes. This finding was subsequently confirmed by other studies in mouse models and in humans [3].

Enrichment of the microbiota with Firmicutes bacteria was initially found to correlate with a greater ability to extract calories from food, thereby promoting excessive weight gain, in rodents. However, it was soon revealed that the five phyla of bacteria present in the microbiota had specific metabolic capacities, resulting in the generation of bacterial metabolites that were then released into the host, albeit often in perfect symbiosis with the host (for example, the synthesis of vitamins and short-chain fatty acids). On the other hand, in cases of quantitative and/or qualitative imbalances among the phyla (intestinal dysbiosis), certain metabolites could have deleterious effects on host physiology. Thus, changes in the microbiota could be contributing to the risk of cardiovascular events and/or to changes in myocardial contractile function through the effects of certain circulating metabolites. These findings demonstrated the presence of a ‘microbiota–myocardium’ axis, certain aspects of which are here explored, taking cardiac insufficiency in the type 2 diabetes (DT2) patient population as an example.

## **Cardiac insufficiency and type 2 diabetes**

DT2 is a disorder known to be associated with high cardiovascular risk. Indeed, epidemiological studies have shown that both the incidence and severity of ischaemic cardiovascular events are greater in DT2 patients than in the general population [4], essentially due to the cumulative effects of a large number of risk factors, including arterial hypertension, atherogenic dyslipidaemia with small, dense low-density lipoprotein (LDL) and low levels of high-density lipoprotein (HDL), insulin resistance and platelet hyperaggregation [5]. Cardiac insufficiency is a serious consequence of myocardial ischaemic disease, and its prognosis is poor. One cause of non-ischaemic cardiac insufficiency was recently

demonstrated in a population of DT2 patients—namely, diabetic cardiomyopathy of metabolic origin [6]. This condition arises in certain DT2 patients who have never had a cardiovascular event, but who have nonetheless developed myocardial dysfunction to the stage of clinical cardiac insufficiency.

While the pathophysiology of non-ischaemic diabetic cardiomyopathy is complex and still imperfectly understood, it is known to involve an exaggerated use of lipids by the heart, the phosphatidylinositol 3-kinase (PI3K)/Akt pathway, reactive oxygen species (ROS) accumulation, certain lipid metabolites (such as ceramides), micro-ribonucleic acids (microRNAs) and epigenetics. Ectopic accumulation of lipids in the heart triggers myocardial hypertrophy, followed by cardiomyocyte necrosis and myocardial fibrosis in a context of changes to the myocardial microcirculation [7], together with the production of excessive amounts of oxidative stress molecules. These processes eventually result in a profound change in the ultrastructure of myocardium, leading to left ventricular dysfunction followed by clinical cardiac insufficiency [8]. Pathogenesis of the myocardial metabolic dysfunction is correlated with excessive abdominal adiposity, dyslipidaemia, presence of hepatic steatosis and its inflammatory activity score, and the amount of ectopic lipids present in skeletal muscle.

It is known that a lipotoxic setting alters adipokine secretion and functionality. Recent evidence has demonstrated that changes in adipokine secretion and signalling are associated with inotropic defects in diabetic cardiomyopathy [9]. Adiponectin, the most abundant peptide secreted by adipocytes, promotes lipid oxidation and glucose uptake, and has protective effects on acute vascular endothelial dysfunction as well as antioxidant properties [10]. Low adiponectin plasma levels, as observed in diabetes, are correlated with the oxidative damage seen in diabetic cardiomyopathy [11,12]. In addition, adiponectin exhibits potent antihypertrophic activity in cardiomyocytes, reduces cardiac cell death by inhibiting inflammation [13,14] and protects the myocardium against the endothelial dysfunction in microvessels of the heart that can lead to diabetic cardiomyopathy [12]. Interestingly, Shaver et al. [15] found that levels of adiponectin are more diminished in diabetes patients with diabetic cardiomyopathy than in diabetes patients without cardiomyopathy or in patients with diastolic dysfunction alone, suggesting that adiponectin may be a biomarker of diabetic cardiomyopathy. Also, there is an inverse relationship between adiponectin and leptin, with circulating levels of leptin being higher in patients with diabetes and diabetic cardiomyopathy

than in patients with diastolic dysfunction alone, suggesting that the adiponectin/leptin ratio may play a role in the pathophysiology of diabetic cardiomyopathy [15].

Emerging evidence has also suggested that resistin may play a role in the development of cardiovascular dysfunction [9]. Plasma resistin levels are elevated both in patients with coronary heart disease [16] and in those with congestive heart failure not only in cross-sectional studies [17], but also in prospective cohort studies [18]. However, even though these studies suggest an important role of resistin in ischaemic heart disease, there is no established evidence that resistin participates in the pathophysiology of diabetic cardiomyopathy. Altered secretion of adipokines is frequently observed in patients with excess fat mass, which is also a recognized contributor to an imbalanced gut microbiota. In this context, certain changes to the microbiota (intestinal dysbiosis) have recently been described, shedding new light on the pathophysiology of cardiac metabolic dysfunction.

### **Intestinal permeability**

The intestinal microbiota interacts in a complex manner with different types of cells in the digestive epithelium, with mucus acting as an intermediary. These interactions between bacteria and the digestive epithelium are bidirectional, and involve bacterial metabolites, antimicrobial peptides of digestive origin, the digestive tract immune system, enteroendocrine intestinal cells and the intestinal nervous system. Symbiosis between various phyla of the microbiota and the host requires that the digestive barrier be intact (and, therefore, of limited permeability to various diffusible bacterial compounds). Intestinal dysbiosis can profoundly modify the nature of the bacterial metabolites synthesized, thereby favouring the emergence of diseases [19], and bacterial metabolites can be transferred to the host bloodstream if the integrity of the intestinal barrier is lost [20]. This integrity may be altered by hypoperfusion of intestinal blood, infections, toxins, drugs, lipid-rich diets and/or changes to the intestinal immune system [21]. Obesity, a key factor of cardiometabolic risk, combines disequilibrium of the intestinal microbiota with changes in the homeostasis of the intestinal immune system. Monteiro-Sepulveda et al. [22] reported an infiltration of T lymphocytes into the jejunal lamina propria of obese subjects that was not observed in healthy subjects. They also found that the activated T lymphocytes secreted cytokines locally, which altered insulin signalling of the jejunal enterocytes and increased intestinal permeability in a pathological manner [23]. The degree of pathological infiltration of the jejunum with these immunocompetent cells

correlated with body mass index (BMI), presence of hepatic steatosis and lipid parameters. This suggests that, in obese patients, systemic changes in metabolic homeostasis may begin in the intestines with an association of dysbiosis, inflammation and increase in intestinal permeability.

### **The microbiota and cardiac insufficiency**

In cardiac insufficiency, a decrease in left ventricular ejection fraction (LVEF) and oxygenation alters intestinal functionality, favouring the development of anaerobic bacteria [24]. Patients with cardiac insufficiency also develop intestinal oedema secondary to a decrease in venous return (thereby reducing the capacity to absorb substrates and favouring bacterial proliferation) [25] and an increase in intestinal permeability [26]. Several studies have demonstrated disequilibrium of the microbiota in patients with cardiac insufficiency especially in post-ischaemic heart failure and, more recently, in diabetic cardiomyopathy (Table I). Intestinal dysbiosis can be characterized by various parameters, such as bacterial diversity and imbalances between bacterial families. As an example, Luedde et al. [27] showed that, in 20 patients with heart failure and a reduced ejection fraction due to ischaemic or dilated cardiomyopathy, the diversity of their microbiota was less than that of the controls, particularly in those who were obese or had DT2. Reduced diversity of gut microbiota is a hallmark of obese populations with cardiometabolic comorbidities and is correlated with insulin resistance, dyslipidaemia and a more pronounced inflammatory phenotype [28]. For this reason, it is difficult to estimate the respective contributions of obesity and heart failure to loss of the rich diversity of the gut microbiome observed in both patient populations.

Beyond diversity, quantitative differences in the faeces of patients with cardiac insufficiency have been reported, with an abnormal abundance of species from the phylum Proteobacteria [gram-negative bacteria that produce lipopolysaccharides (LPS), including *Campylobacter*, *Shigella* and *Salmonella* genera] [29] and genus *Lactobacillus* (gram-positive bacteria from the phylum Firmicutes) [30], as demonstrated in 60 patients with chronic heart failure of various aetiologies. In addition, Kummel et al. [31] performed a microbiota study of considerable importance as it included both a large number of patients with stable cardiac insufficiency and controls, and also adjusted their results for a large number of confounding factors capable of introducing a bias. The study confirmed that patients with stable cardiac insufficiency (due to mixed aetiologies) had less bacterial diversity after adjusting for BMI, age, gender and diabetes.

Cardiac insufficiency has been associated with a smaller proportion of bacteria from the family Lachnospiraceae, which includes several species capable of synthesizing butyrate. This bacterial decrease was inversely correlated with concentrations of circulating CD25, a marker of T-cell activation, thereby suggesting a link between inflammation, the decrease in Lachnospiraceae species and loss of butyrate synthesis capacity. This correlation was particularly significant in patients who died during follow-up or were on a waiting list for heart transplantation, providing the first evidence that microbiota parameters may be predictive of severe cardiac insufficiency. On the other hand, a decrease in bacteria from the phylum Bacteroidetes and genus *Faecalibacterium* (including *F. prausnitzii*, a species with anti-inflammatory properties known to protect against intestinal permeability) was observed in 22 patients with heart failure (including six with ischaemic heart failure and seven with diabetic cardiomyopathy) in a study by Kamo et al. [30]. In patients with heart failure, Luedde and colleagues [27] reported significant decreases in Coriobacteriaceae, Erysipelotrichaceae and Ruminococcaceae bacteria on the family level, and in *Blautia*, *Collinsella*, uncl. Erysipelotrichaceae and uncl. Ruminococcaceae on the genus level. Yet, in all of these studies, no differences were observed according to diabetes status of the studied populations, suggesting that heart failure may be more dependent on microbiota dysbiosis than on metabolic status.

It was therefore considered important to determine whether these quantitative changes in the bacterial phyla and genera of the microbiota might have physiological consequences, such as changes in the availability of bacterial metabolites. To this end, Katsimichas et al. [32] compared microbiota data from 28 patients with non-ischaemic heart failure and reduced ejection fraction (HFrEF) with those of control subjects matched for age, and then classified the genes of their bacterial species according to metabolic function. This study revealed that the two study groups differed in terms of bacterial enzymatic pathways. The 10 most abundant bacterial metabolic pathways enriched in the HFrEF patients involved glutathione metabolism, inositol phosphate metabolism, lipoic acid metabolism, metabolism of xenobiotics by cytochrome P450, naphthalene degradation, phosphate and phosphinate metabolism, and taurine and hypotaurine metabolism. Conversely, pantothenate and coenzyme A (CoA) biosynthesis, and phenylalanine, tyrosine and tryptophan biosynthesis pathways were depleted in HFrEF patients compared with the controls.

Beyond the metagenomics studies to determine the functional metabolic profiles of bacterial communities, several different methods can be combined to identify these differentially expressed bacterial metabolic pathways. Once the phyla and genera of the bacteria presented



differentially in the two groups have been determined, analysis of their genes can predict the possibilities for metabolite synthesis. The most likely metabolites can then be quantified in the blood or faeces using metabolomic techniques. This strategy makes it possible to assess the bacterial family of interest and to make genetic predictions as to which metabolites are likely to be synthesized, followed by screening for their presence in the blood and/or faeces. When Cui et al. [33] applied this approach to 53 patients with cardiac insufficiency (including 28 patients with DT2) and 41 control subjects, they found that the genes of the microbiota involved in the synthesis of tryptophan, trimethylamine (TMA) and LPS (with TMA and LPS being pro-inflammatory) were more strongly expressed in patients with cardiac insufficiency, regardless of its aetiology (ischaemic or HFrEF). In contrast, the genes encoding for butyrate–acetoacetate CoA transferase (a key enzyme in butyrate synthesis with beneficial metabolic properties), and for proteins involved in the synthesis and transport of amino acids or for iron transport, were less strongly expressed in patients with cardiac insufficiency [33].

Going from looking at genes to looking at the bacteria instead, the study then found that such patients were deficient in species capable of synthesizing beneficial short-chain fatty acids such as butyrate (including *F. prausnitzii*), and had an overabundance of bacteria capable of synthesizing TMA, which has deleterious cardiovascular effects following its transformation to trimethylamine oxide (TMAO) in the liver (see below). Consistent with the genetic analyses, quantification of metabolites in the blood showed that intestinal dysbiosis was associated with increased levels of 49 metabolites (including TMA, TMAO and sphingosine-1-phosphate, a cell-signalling sphingolipid with cardioprotective effects) in subjects with cardiac insufficiency.

While it remains difficult to explain these differences in metabolite levels and their physiological consequences in the context of cardiac insufficiency, studies of this type nevertheless open up new perspectives for a greater understanding of the disease. Interestingly, even though patients with heart failure and controls differed at the genus level, there were some genera differentially enriched between ischaemic cardiomyopathy and HFrEF patients, suggesting that each aetiology of heart failure may come with its own distinct gut microbiota dysbiosis. Nevertheless, the causality of these relationships has yet to be tested in experimental models and in specific studies taking into account the aetiology of heart failure.

### **Bacterial metabolites and cardiac insufficiency**

It is clear that cardiac insufficiency is associated with multiple changes in the microbiota and secretion of bacterial metabolites (Fig. 1). If the hypothesis of a ‘microbiota–myocardium’ axis is applied to non-ischaemic diabetic cardiac insufficiency, can it then be argued that changes in the secretion of certain bacterial metabolites can cause myocardial contractile dysfunction? The first bacterial metabolite reported to play a role in cardiometabolic risk was TMA. The microbiota has enzymatic equipment enabling it to produce TMA from phosphatidylcholine, choline, carnitine and food [34]; the TMA thus synthesized is then oxidized in the liver to generate TMAO, which spreads throughout the host body *via* the bloodstream. TMAO has been shown to be atherogenic and to influence not only cardiovascular risk, but also the risk of developing cardiac insufficiency [34]. In addition, high levels of TMAO in the bloodstream are predictive of long-term mortality in patients with cardiac insufficiency independently of traditional cardiovascular risk factors [35] and levels of B-type natriuretic peptide (BNP) [36]. But how does TMAO contribute to the pathophysiology of cardiac insufficiency? While the exact mechanisms remain unclear, it is possible that TMAO favours the production of inflammatory cytokines such as tumour necrosis factor (TNF)- $\alpha$ , interleukin (IL)-1 $\beta$  and IL-6 [37,38], thereby potentially aggravating myocardial fibrosis and microvascular dysfunction independently of its proatherogenic effects [39,40] and worsening the patient’s prognosis [41,42]. In a veritable vicious circle, TNF- $\alpha$ , IL-1 $\beta$  and IL-6 all increase intestinal permeability, thereby further increasing the diffusion of metabolites such as TMA or of bacterial fragments such as LPS (see below) that, in turn, also have deleterious effects on myocardium [43].

In cardiac insufficiency, the observed modifications in the phyla present in the microbiota favour not only the production of TMAO, but also of endotoxin (LPS). Like TMAO, LPS aggravates cardiac insufficiency [44]. These bacterial fragments can reach the host systemic circulation by interacting with receptors on the surface of the intestinal mucosa [42]. Once in the bloodstream, LPS interacts with Toll-like receptor 4 (TLR4) [44] in a multitude of tissues and cell types, including macrophages, dendritic cells, cardiomyocytes and cardiac fibroblasts [45]. Circulating LPS concentrations increase in cases of acute decompensation of cardiac insufficiency (as related to intestinal oedema) and decrease once this decompensation is controlled [46–48]. Interestingly, LPS levels are higher in portal than in peripheral blood, suggesting a digestive origin of the LPS detected [48]. Finally, LPS is itself a mediator that can alter intestinal permeability in a vicious circle that further increases systemic diffusion of LPS from the digestive system [49].

End-stage renal disease (a possible complication of diabetes) increases the risk of a cardiovascular event and cardiac insufficiency in a population already at risk due to the presence of diabetes and its associated comorbid conditions. However, little is known of the microbiota changes in patients with renal insufficiency, although the deleterious effects of certain metabolites of bacterial origin have been reported in this context. Indoxyl sulphate and p-cresyl sulphate are uraemic toxins derived from the intestinal microbiota *via* fermentation of tryptophan and tyrosine. Their circulating concentrations are correlated with cardiovascular and all-cause mortality and with myocardial contractile dysfunction [50]. In rodents, indoxyl sulphate has hypertrophic and profibrotic properties in the myocardium and kidney [51]. In addition, excess indoxyl sulphate favours the production of certain osteoblastic proteins involved in the development of vascular calcification, a process strongly correlated with a poor cardiovascular prognosis [50].

It is now evident that cardiac insufficiency is associated with a decrease in the bacterial families responsible for short-chain fatty-acid production. These fatty acids play an important role in supplying the energy needs of colon cells, although preclinical studies in a hypertensive mouse model have also indicated that they can help to prevent arterial hypertension and the development of myocardial fibrosis and cardiac insufficiency [52]. In addition, a small proportion of short-chain fatty acids diffuse into the bloodstream of the host, where they activate certain cellular targets, such as adenosine monophosphate (AMP)-activated protein kinase in the liver and skeletal muscle (and, potentially, the heart), thereby favouring lipid oxidation and improving sugar homeostasis in rodents [53]. Thus, a decrease in the numbers of bacteria producing butyrate may be associated with both the presence of DT2 and cardiac insufficiency [54]. Similarly, a loss of microbiota diversity appears to be an alteration common to both dysglycaemia and cardiac insufficiency [28]. Thus, there is an overlap between the qualitative and quantitative changes observed in the microbiota in these two diseases, suggesting a common pathophysiology for cardiometabolic diseases.

Bile acids provide a final example to illustrate the link between the metabolism of bacterial flora and risk of cardiac insufficiency. These acids play a key role in the stabilization of lipids to enhance their intestinal absorption, but are also involved in the regulation of metabolism and inflammation *via* several receptors, including the farnesoid X receptor (FXR) [55]. Colon bacteria can metabolize primary bile acids (such as cholic acid and chenodeoxycholic acid) to generate secondary bile acids (principally lithocholic and ursodeoxycholic acids), which are then broken down by the enterohepatic cycle. However, changes in the microbiota observed in cases of cardiac insufficiency lead to changes in the ratio of these two types of bile acids [56].

Activators of FXR decrease the inflammatory response by repressing the nuclear factor kappa-light-chain enhancer of activated B cells (NF- $\kappa$ B) nuclear receptor. This receptor is essential for the development of myocardial hypertrophy independently of all other risk factors. It has therefore been suggested that modulators of FXR might have beneficial effects on cardiac insufficiency [57]. Furthermore, ursodeoxycholic acid (UDCA) can form micelles that bind to LPS, thereby limiting its diffusion into the host systemic circulation. Through this mechanism, UDCA could potentially improve cardiac insufficiency. von Haehling et al. [58] tested this hypothesis in a randomized double-blind, placebo-controlled, clinical trial including 17 patients with LVEF levels < 45%. The study demonstrated that UDCA improved peripheral haemodynamics in 16 of the 17 patients with no effects on their physical capacity to exercise. Additional studies are now required to confirm the therapeutic benefits of these molecules for cardiac insufficiency.

### **Therapeutic perspectives**

Modification of the intestinal microbiota to prevent metabolic cardiac insufficiency and improve its prognosis in DT2 patients is a therapeutic strategy that is currently in development. Dietary changes can modify the microbiota and the metabolites it produces in a favourable manner, with positive effects on the risk or progression of cardiac insufficiency. In a US study of the determinants of weight gain during the prospective follow-up of three cohorts, involving a total of 120,877 patients, Mozaffarian et al. [59] showed that weight increases were correlated principally with the consumption of processed foods, sodas and red meat, whereas weight loss was favoured by a diet based on vegetables, fibre and yoghurt. Dietary recommendations along these lines were integrated into the Dietary Approaches to Stop Hypertension (DASH), which have been shown to decrease circulating TMAO levels as well as the incidence of cardiac insufficiency in observational studies [60]. The Mediterranean diet, which traditionally contains low levels of TMAO precursors, has also been found to reduce the number of repeat hospitalizations for cardiac insufficiency in the Mediterranean Diet in Acute Heart Failure (MEDIT-AHF) clinical trial [61].

Probiotics (live bacteria that can be ingested and are capable of modifying the proportions of bacterial families in the microbiota) such as *Saccharomyces boulardii* [62] have been shown to have beneficial effects in cases of cardiac insufficiency (LVEF improvement in seven patients with cardiac insufficiency in a randomized double-blind clinical trial), but now require confirmation in larger studies like the GutHeart study, for which recruitment is currently underway, in which 150 patients with LVEF levels < 40% will be randomized to

receive either an antibiotic, a probiotic (*S. boulardii*) or a placebo [63]. The principal outcome measure of this study will be the change in LVEF in the three groups after 3 months. Faecal transplantation, a technique already in use for cases of *Clostridium difficile* infection, could also become an innovative treatment for cardiometabolic diseases in the future [64].

Improving the management of cardiac insufficiency with pharmacological treatments that modify the microbiota is another possible innovative strategy. For example, 3,3-dimethyl-1-butanol is a structural analogue of choline that inhibits the bacterial TMA lyase and, thus, decreases circulating concentrations of TMAO in rodents [65]. Intakes of fibre and short-chain fatty acids (acetate) reduce cardiac hypertrophy in hypertensive mice [66], while oral administration of an activated charcoal adsorbent (AST-120) decreases both circulating concentrations of indoxyl sulphate and cardiac hypertrophy in rats with renal insufficiency [67]. The mechanism involved here is the AST-120 blockade of the first steps of conversion of dietary precursors of indoxyl sulphate resulting in a reduction in circulating concentrations of the molecule. As already mentioned, indoxyl sulphate has profibrotic effects not only on nephrons, but also on cardiomyocytes, suggesting that approaches to decrease its circulating concentrations could also be used as a treatment against cardiac fibrosis [68].

The reduced incidence of cardiovascular events independently of circulating lipid levels in a clinical trial of the monoclonal anti-IL-1 $\beta$  antibody canakinumab [in the Canakinumab Anti-inflammatory Thrombosis Outcome Study (CANTOS)] has paved the way for the use of treatments that control inflammation as innovative cardiometabolic therapies [69]. Based on the same concept, the Diastolic Heart Failure Anakinra Response Trial 2 (DHART2) involved the blockade (by anakinra) of IL-1 $\alpha$  and IL-1 $\beta$  receptors in obese patients with cardiac insufficiency but with preserved ejection fraction, with the principal aim of increasing VO<sub>2max</sub> [70,71]. Although this trial had negative results—perhaps because the effect of the molecule was too weak or because obesity in itself decreased ventilatory capacity regardless of myocardial function—it nevertheless showed that the treatment decreased circulating levels of C-reactive protein and N-terminal-pro-BNP, thus confirming an effect of anakinra on these inflammatory parameters. Other studies should now be performed to determine whether this innovative strategy for treating cardiac insufficiency could yield good results.

## **Conclusion**

There is growing evidence to suggest that changes in the intestinal microbiota can contribute to the pathophysiology of metabolic cardiac insufficiency and/or its progression in patients with diabetes, and that disruption of the symbiotic interactions between the microbiota and the

host triggers changes in the synthesis and diffusion (into the host) of a large number of bacterial metabolites, many of which have yet to be identified. Nevertheless, it has been shown that some of these metabolites can alter myocardial contractility independently of cardiovascular risk factors and/or arterial hypertension, thereby demonstrating the concept of the ‘microbiota–myocardium’ axis. However, this highly innovative concept still requires confirmation in randomized double-blind clinical trials to determine whether changes to the microbiota can indeed improve cardiac insufficiency.

Table I

Principal changes to the intestinal microbiota observed in cases of diabetic cardiomyopathy

[Table]

BG-: beta-glucan-negative; LPS+: lipopolysaccharide-positive

### **Figure legend**

Fig. 1. Principal changes to metabolites of bacterial origin in cases of diabetic cardiomyopathy. TMA/TMAO: trimethylamine/trimethylamine oxide; TNF: tumour necrosis factor; IL: interleukin; LPS: lipopolysaccharide; TRL4: Toll-like receptor 4; AMPK: 5' adenosine monophosphate-activated protein kinase.

## References

- 1- Lim SS, Vos T, Flaxman AD, Danaei G, Shibuya K. A comparative risk assessment of burden of disease and injury attributable to 67 risk factors clusters in 21 regions, 1990-2010: a systematic analysis for the Global Burden of Disease Study 2010. *Lancet* 2012; 380: 2224-60.
- 2- Ley RE, Turnbaugh PJ, Klein S, Gordon JI. Microbial ecology: human gut microbes associated with obesity. *Nature* 2006; 444:1022-3.
- 3- Bouter KE, van Raalte DH, Groen AK, Nieuwdorp M. Role of the gut microbiome in the pathogenesis of obesity and obesity-related metabolic dysfunction. *Gastroenterology* 2017; 152:1671-8
- 4- Shah AD, Langenberg C, Rapsomaniki E, Denaxas S, Pujades-Rodriguez M, Gale CP, et al. Type 2 diabetes and incidence of cardiovascular diseases: a cohort study in 1.9 million people. *Lancet Diabetes Endocrinol* 2015; 3:105–13.
- 5- Rydén L, Grant PJ, Anker SD, Berne C, Cosentino F, Danchin N, et al. ESC guidelines on diabetes, pre-diabetes, and cardiovascular diseases developed in collaboration with the EASD: The Task Force on Diabetes, Pre-Diabetes, and Cardiovascular Diseases of the European Society of Cardiology (ESC) and Developed in Collaboration. *Eur Heart J* 2013; 34:3035–87.
- 6- Boonman-de Winter LJM, Rutten FH, Cramer MJM, Landman MJ, Liem AH, Rutten GE, et al. High prevalence of previously unknown heart failure and left ventricular dysfunction in patients with type 2 diabetes. *Diabetologia* 2012; 55:2154–62.
- 7- Joubert M, Manrique A, Cariou B, Prieur X. Diabetes-related cardiomyopathy: The sweet story of glucose overload from epidemiology to cellular pathways. *Diabetes Metab* 2019; 45:238-47.
- 8- Lourenço AP, Leite-Moreira AF, Balligand JL, Bauersachs J, Dawson D, de Boer RA, et al. An integrative translational approach to study heart failure with preserved ejection fraction: a position paper from the Working Group on Myocardial Function of the European Society of Cardiology. *Eur J Heart Fail* 2018; 20:216–27.
- 9- Lebeche D. Diabetic cardiomyopathy: is resistin a culprit? *Cardiovasc Diagn Ther* 2015; 5:387-93.
- 10- Kita S, Maeda N, Shimomura I. Interorgan communication by exosomes, adipose tissue, and adiponectin in metabolic syndrome. *J Clin Invest* 2019; 129:4041-9.
- 11- Li J, Su S, Zong X. Analysis of the association between adiponectin, adiponectin receptor 1 and diabetic cardiomyopathy. *Exp Ther Med* 2014; 7:1023-7.
- 12- Knapp M, Tu X, Wu R. Vascular endothelial dysfunction, a major mediator in diabetic cardiomyopathy. *Acta Pharmacol Sin* 2019; 40:1-8.
- 13- Battiprolu PK, Lopez-Crisosto C, Wang ZV, Nemchenko A, Lavandero S, Hill JA. Diabetic cardiomyopathy and metabolic remodeling of the heart. *Life Sci* 2013; 92:609-15.
- 14- Shibata R, Ouchi N, Ito M, Kihara S, Shiojima I, Pimentel DR, et al. Adiponectin-mediated modulation of hypertrophic signals in the heart. *Nat Med* 2004; 10:1384-9.



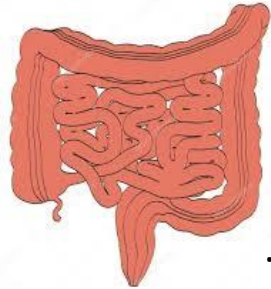
- 15- Shaver A, Nichols A, Thompson E, Mallick A, Payne K, Jones C, et al. Role of Serum Biomarkers in Early Detection of Diabetic Cardiomyopathy in the West Virginian Population. *Int J Med Sci* 2016; 13:161-8.
- 16- Pischon T, Bamberger CM, Kratzsch J, Zyriax BC, Algenstaedt P, Boeing H, et al. Association of plasma resistin levels with coronary heart disease in women. *Obes Res* 2005; 13:1764-71.
- 17- Takeishi Y, Niizeki T, Arimoto T, Nozaki N, Hirono O, Nitobe J, et al. Serum resistin is associated with high risk in patients with congestive heart failure--a novel link between metabolic signals and heart failure. *Circ J* 2007; 71:460-4.
- 18- Frankel DS, Vasani RS, D'Agostino RB Sr, Benjamin EJ, Levy D, Wang TJ, et al. Resistin, adiponectin, and risk of heart failure the Framingham offspring study. *J Am Coll Cardiol* 2009; 53:754-62.
- 19- Schroeder BO, Bäckhed F. Signals from the gut microbiota to distant organs in physiology and disease. *Nat Med* 2016; 22:1079-89.
- 20- Zhang K, Hornef MW, Dupont A. The intestinal epithelium as guardian of gut barrier integrity. *Cell Microbiol* 2015; 17:1561-9.
- 21- Bischoff SC, Barbara G, Buurman W, Ockhuizen T, Schulzke JD, Serino M, et al. Intestinal permeability--a new target for disease prevention and therapy. *BMC Gastroenterol* 2014; 14:189.
- 22- Monteiro-Sepulveda M, Touch S, Mendes-Sá C, André S, Poitou C, Allatif O, et al. Jejunal T cell inflammation in human obesity correlates with decreased enterocyte insulin signaling. *Cell Metab* 2015; 22:113-24.
- 23- Genser L, Aguanno D, Soula HA, Dong L, Trystram L, Assmann K, et al. Increased jejunal permeability in human obesity is revealed by a lipid challenge and is linked to inflammation and type 2 diabetes. *J Pathol* 2018; 246:217-30.
- 24- Sandek A, Swidsinski A, Schroedl W, Watson A, Valentova M, Herrmann R, et al. Intestinal blood flow in patients with chronic heart failure: a link with bacterial growth, gastrointestinal symptoms, and cachexia. *J Am Coll Cardiol* 2014; 64:1092-102.
- 25- Schiattarella GG, Sannino A, Toscano E, Giugliano G, Gargiulo G, Franzone A, et al. Gut microbe-generated metabolite trimethylamine-N-oxide as cardiovascular risk biomarker: a systematic review and dose-response meta-analysis. *Eur Heart J* 2017; 38:2948-56.
- 26- Sandek A, Bauditz J, Swidsinski A, Buhner S, Weber-Eibel J, von Haehling S, et al. Altered intestinal function in patients with chronic heart failure. *J Am Coll Cardiol* 2007; 50:1561-9.
- 27- Luedde M, Winkler T, Heinsen FA, Rühlemann MC, Spehlmann ME, Bajrovic A, et al. Heart failure is associated with depletion of core intestinal microbiota. *ESC Heart Fail* 2017; 4:282-90.
- 28- Le Chatelier E, Nielsen T, Qin J, Prifti E, Hildebrand F, Falony G, et al. Richness of human gut microbiome correlates with metabolic markers. *Nature* 2013; 500:541-6.
- 29- Pasini E, Aquilani R, Testa C, Baiardi P, Angioletti S, Boschi F, et al. Pathogenic gut flora in patients with chronic heart failure. *JACC Heart Fail* 2016; 4:220-7.

- 30- Kamo T, Akazawa H, Suda W, Saga-Kamo A, Shimizu Y, Yagi H, et al. Dysbiosis and compositional alterations with aging in the gut microbiota of patients with heart failure. *PLoS One* 2017; 12:e0174099.
- 31- Kummén M, Mayerhofer CCK, Vestad B, Broch K, Awoyemi A, Storm-Larsen C, et al. Gut microbiota signature in heart failure defined from profiling of 2 independent cohorts. *J Am Coll Cardiol* 2018; 71:1184-6.
- 32- Katsimichas T, Ohtani T, Motooka D, Tsukamoto Y, Kioka H, Nakamoto K, et al. Non-ischemic heart failure with reduced ejection fraction is associated with altered intestinal microbiota. *Circ J* 2018; 82:1640-50.
- 33- Cui X, Ye L, Li J, Jin L, Wang W, Li S, et al. Metagenomic and metabolomic analyses unveil dysbiosis of gut microbiota in chronic heart failure patients. *Sci Rep* 2018; 8:635.
- 34- Tang WHW, Li DY, Hazen SL. Dietary metabolism, the gut microbiome, and heart failure. *Nat Rev Cardiol* 2019; 16:137-54.
- 35- Tang WH, Wang Z, Fan Y, Levison B, Hazen JE, Donahue LM, et al. Prognostic value of elevated levels of intestinal microbe-generated metabolite trimethylamine-N-oxide in patients with heart failure: refining the gut hypothesis. *J Am Coll Cardiol* 2014; 64:1908-14.
- 36- Suzuki T, Heaney LM, Bhandari SS, Jones DJ, Ng LL. Trimethylamine N-oxide and prognosis in acute heart failure. *Heart* 2016; 102:841-8.
- 37- Mann DL. Innate immunity and the failing heart: the cytokine hypothesis revisited. *Circ Res* 2015; 116:1254–68.
- 38- Deswal A, Petersen NJ, Feldman AM, Young JB, White BG, Mann DL. Cytokines and cytokine receptors in advanced heart failure: an analysis of the cytokine database from the Vesnarinone trial (VEST). *Circulation* 2001; 103: 2055–9.
- 39- Bozkurt B, Kribbs SB, Clubb JF Jr, Michael LH, Didenko VV, Hornsby PJ et al. Pathophysiologically relevant concentrations of tumor necrosis factor- $\alpha$  promote progressive left ventricular dysfunction and remodeling in rats. *Circulation* 1998; 97: 1382–91.
- 40- Kubota T, McTiernan CF, Frye CS, Slawson SE, Lemster BH, Koretsky AP et al. Dilated cardiomyopathy in transgenic mice with cardiac-specific overexpression of tumor necrosis factor- $\alpha$ . *Circ Res* 1997; 81: 627–35.
- 41- Nagatomo Y, Tang WH. Intersections between microbiome and heart failure: revisiting the gut hypothesis. *J Card Fail* 2015; 21:973-80.
- 42- Brown JM, Hazen SL. The gut microbial endocrine organ: bacterially derived signals driving cardiometabolic diseases. *Annu Rev Med* 2015; 66:343-59.
- 43- Katsimichas T, Antonopoulos AS, Katsimichas A, Ohtani T, Sakata Y, Tousoulis D. The intestinal microbiota and cardiovascular disease. *Cardiovasc Res* 2019 Jun 4. pii: cvz135.
- 44- Lu YC, Yeh WC, Ohashi PS. LPS/TLR4 signal transduction pathway. *Cytokine* 2008; 42: 145–51.
- 45- Frangogiannis NG. The inflammatory response in myocardial injury, repair, and remodelling. *Nat Rev Cardiol* 2014; 11: 255–65.
- 46- Niebauer J, Volk HD, Kemp M, Dominguez M, Schumann RR, Rauchhaus M, Poole-Wilson PA et al. Endotoxin and immune activation in chronic heart failure: a prospective cohort study. *Lancet* 1999; 353: 1838–42.

- 47- Sandek A, Bjarnason I, Volk HD, Crane R, Meddings JB, Niebauer J, et al. Studies on bacterial endotoxin and intestinal absorption function in patients with chronic heart failure. *Int J Cardiol* 2012; 157:80-5.
- 48- Peschel T, Schönauer M, Thiele H, Anker SD, Schuler G, Niebauer J. Invasive assessment of bacterial endotoxin and inflammatory cytokines in patients with acute heart failure. *Eur J Heart Fail* 2003; 5: 609–14.
- 49- Hietbrink F, Besselink MG, Renooij W, de Smet MB, Draisma A, van der Hoeven H, et al. Systemic inflammation increases intestinal permeability during experimental human endotoxemia. *Shock* 2009; 32: 374–8.
- 50- Lekawanvijit S. Role of gut-derived protein-bound uremic toxins in cardiorenal syndrome and potential treatment modalities. *Circ J* 2015; 79:2088-97.
- 51- Yang K, Wang C, Nie L, Zhao X, Gu J, Guan X, et al. Klotho protects against indoxyl sulphate-induced myocardial hypertrophy. *J Am Soc Nephrol* 2015; 26:2434-46.
- 52- Marques FZ, Nelson E, Chu PY, Horlock D, Fiedler A, Ziemann M et al. High-fiber diet and acetate supplementation change the gut microbiota and prevent the development of hypertension and heart failure in hypertensive mice. *Circulation* 2017; 135: 964–77.
- 53- den Besten G, Lange K, Havinga R, van Dijk TH, Gerding A, van Eunen K, et al. Gut-derived short-chain fatty acids are vividly assimilated into host carbohydrates and lipids. *Am J Physiol Gastrointest Liver Physiol* 2013; 305:G900-10.
- 54- Qin J, Li Y, Cai Z, Li S, Zhu J, Zhang F, et al. A metagenome-wide association study of gut microbiota in type 2 diabetes. *Nature* 2012; 490:55-60.
- 55- Wang H, Chen J, Hollister K, Sowers LC, Forman BM. Endogenous bile acids are ligands for the nuclear receptor FXR/BAR. *Mol Cell* 1999; 3: 543–53.
- 56- Mayerhofer CCK, Ueland T, Broch K, Vincent RP, Cross GF, Dahl CP et al. Increased secondary/primary bile acid ratio in chronic heart failure. *J Card Fail* 2017; 23: 666–71.
- 57- Gordon JW, Shaw JA, Kirshenbaum LA. Multiple facets of NF-kappaB in the heart: to be or not to NF-kappaB. *Circ Res* 2011; 108: 1122–32.
- 58- von Haehling S, Schefold JC, Jankowska EA, Springer J, Vazir A, Kalpra PR et al. Ursodeoxycholic acid in patients with chronic heart failure: a double-blind, randomized, placebo-controlled, crossover trial. *J Am Coll Cardiol* 2012; 59: 585–92.
- 59- Mozaffarian D, Hao T, Rimm EB, Willett WC, Hu FB. Changes in diet and lifestyle and long-term weight gain in women and men. *N Engl J Med* 2011; 364:2392-404.
- 60- Tang WHW, Li DY, Hazen SL. Dietary metabolism, the gut microbiome, and heart failure. *Nat Rev Cardiol* 2019; 16:137-54.
- 61- Miro O, Estruch R, Martin-Sanchez FJ, Gil V, Jacob J, Herrero-Puente P, et al. Adherence to Mediterranean diet and all-cause mortality after an episode of acute heart failure: results of the MEDIT-AHF study. *JACC Heart Fail* 2018; 6: 52–62.
- 62- Costanza AC, Moscovitch SD, Faria Neto HC, Mesquita ET. Probiotic therapy with *Saccharomyces boulardii* for heart failure patients: a randomized, double-blind, placebo-controlled pilot trial. *Int J Cardiol* 2015; 179:348-50.
- 63- Mayerhofer CCK, Awoyemi AO, Moscovitch SD, Lappegård KT, Hov JR, Aukrust P, et al. Design of the GutHeart-targeting gut microbiota to treat heart failure-trial: a Phase II, randomized clinical trial. *ESC Heart Fail* 2018; 5:977-84.

- 64- Marotz CA, Zarrinpar A. Treating obesity and metabolic syndrome with fecal microbiota transplantation. *Yale J Biol Med* 2016; 89:383-8.
- 65- Wang Z, Roberts AB, Buffa JA, Levison BS, Zhu W, Org E, et al. Non-lethal inhibition of gut microbial trimethylamine production for the treatment of atherosclerosis. *Cell* 2015; 163:1585-95.
- 66- Marques FZ, Nelson E, Chu PY, Horlock D, Fiedler A, Ziemann M, et al. High-fiber diet and acetate supplementation change the gut microbiota and prevent the development of hypertension and heart failure in hypertensive mice. *Circulation* 2017; 135:964-77.
- 67- Lekawanvijit S, Kompa AR, Manabe M, Wang BH, Langham RG, Nishijima F, et al. Chronic kidney disease-induced cardiac fibrosis is ameliorated by reducing circulating levels of a non-dialysable uremic toxin, indoxyl sulfate. *PLoS One* 2012; 7:e41281.
- 68- Lekawanvijit S, Adrahtas A, Kelly DJ, Kompa AR, Wang BH, Krum H. Does indoxyl sulfate, a uraemic toxin, have direct effects on cardiac fibroblasts and myocytes? *Eur Heart J* 2010; 31:1771-9.
- 69- Ridker PM, Everett BM, Thuren T, MacFadyen JG, Chang WH, Ballantyne C, et al. Antiinflammatory therapy with canakinumab for atherosclerotic disease. *N Engl J Med* 2017; 377: 1119-31.
- 70- Van Tassell BW, Buckley LF, Carbone S, Trankle CR, Canada JM, Dixon DL, et al. Interleukin-1 blockade in heart failure with preserved ejection fraction: rationale and design of the Diastolic Heart Failure Anakinra Response Trial 2 (D-HART2). *Clin Cardiol* 2017; 40:626-32.
- 71- Van Tassell BW, Trankle CR, Canada JM, Carbone S, Buckley L, Kadariya D, et al. IL-1 blockade in patients with heart failure with preserved ejection fraction. *Circ Heart Fail* 2018; 11:e005036.

↑ Intestinal permeability



↓ Bacterial diversity

Metabolites of bacterial origin

↑ TMA/TMAO

TNF alpha, IL-1 β and IL-6

↑ LPS

TLR4

↑ Butyrate

↓ AMPK activity

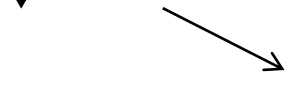
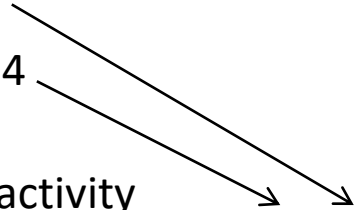
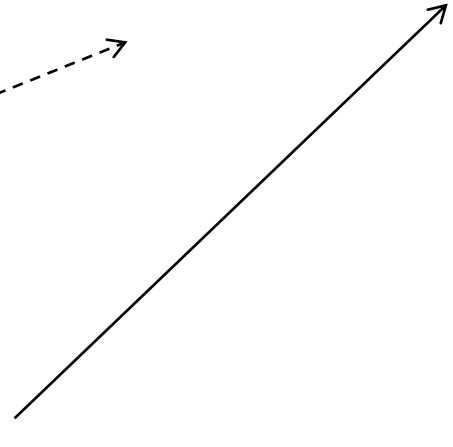
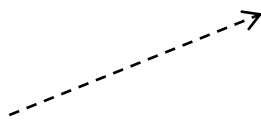
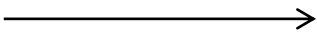
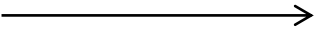
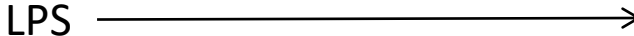
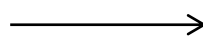
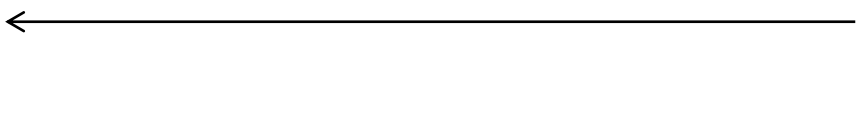
Kidney failure  
*p*-cresyl sulfate  
Indoxyl sulfate

Cardiomyocyte hypertrophy  
Interstitial fibrosis

↑ Sphingosine-1-phosphate  
(cardioprotection)

↓ Ricinoleic acid  
(anti-inflammatory)

↑ Secondary/primary bile acid ratio



Phylum	Genus	Cardiac insufficiency
Proteobacteria	<i>Campylobacter, Shigella, and Salmonella (BG-, LPS+)</i>	Increased
Firmicutes	<i>Lactobacillus</i> and Lachnospiraceae (butyrate producers)	Decreased
Bacteroidetes	<i>Faecalibacterium</i> (including <i>Faecalibacterium prausnitzii</i> , anti-inflammatory and maintains the integrity of the intestinal barrier)	Decreased