The endotoxin-lipoprotein hypothesis – an update

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Abstract

Hypercholesterolaemia is a well-defined risk factor for both morbidity and mortality in coronary artery disease. However, in chronic heart failure high levels of plasma cholesterol have been associated with better survival. The reason for this inverse epidemiology is potentially explained by the ability of plasma lipoproteins to bind and detoxify bacterial lipopolysaccharide (LPS, endotoxin), a cell-wall component of Gram-negative bacteria. This hypothesis has been termed the "endotoxin-lipoprotein hypothesis", first published in 2000. LPS is one of the strongest inducers of pro-inflammatory cytokine release in heart failure. This article summarises the known pathophysiology of increased endotoxin concentrations entering the blood stream via the hypoperfused oedematous bowel wall. Furthermore, it discusses technical problems with the measurement of LPS, and reviews its signalling cascade and the biological effects of LPS as well as the potentially protective ability of serum lipoproteins in chronic heart failure.

Key words: heart failure, immune activation, lipids, cholesterol, endotoxin, LPS, pathophysiology.

Introduction

Hypercholesterolaemia is a well-defined risk factor for both morbidity and mortality in coronary artery disease (CAD). However, it is less clear what happens once CAD progresses to chronic heart failure (CHF). In fact, the role of total serum cholesterol and other serum lipids in this chronic condition is not entirely understood. Patients with advanced CHF often have low serum cholesterol concentrations, which has consistently and paradoxically been associated with a poor prognosis in these patients. Higher concentrations of total serum cholesterol in CHF predicted an improved survival in a relatively small study of 114 patients irrespective of disease aetiology, age, left ventricular ejection fraction and exercise capacity [1]. Horwich et al. [2] evaluated follow-up data of a larger patient cohort consisting of 1,134 patients and found that those in the lowest quintile of total serum cholesterol had a twofold increase in the relative risk of death during five years of follow-up (hazard ratio 2.5, 95% confidence interval 1.8-3.3). Higher concentrations of total cholesterol (144±8 mg/dl vs. 90±7 mg/dl) have been shown to be associated with a lower mortality rate in patients with severe systolic CHF (NYHA IV, cardiac index <2.0 l/min/m^2) supported by a left ventricular assist device [3].

This inverse relationship has been termed the "cholesterol paradox" [4]. It has been observed not only in patients with symptomatic CHF, but also in patients on haemodialysis and those with chronic obstructive pulmonary disease, AIDS, or advanced age [5]. A potential and plausible interpretation of this unexpected association is not easy but is possible as follows: First it may reflect the benefit of a greater metabolic reserve [1, 2] in a patient with an increased resting expenditure [6] and catabolic drive as in CHF [7], as this disease is a metabolically demanding chronic condition. Second, it may reflect relevant interactions between serum lipoproteins with bioactive endotoxin (also named lipopolysaccharide – LPS) in the human serum [8].

Bacterial translocation and the endotoxin-lipoprotein hypothesis

LPS is regarded as one of the strongest inducers of pro-inflammatory cytokine release. LPS, a cell-wall component of Gram-negative bacteria, can enter the circulation through the gut wall if barrier function is impaired as in various diseases, such as burn injury, sepsis, liver cirrhosis and ischaemic reperfusion injury [7-10]. The origin of inflammation in patients with CHF with elevated concentrations of pro-inflammatory cytokines is still a matter of debate [9-11]. Several hypotheses have been put forward [12] including local production by the myocardium itself [13] or by invaded pro-inflammatory cells [14], local secretion as a response to hypoxia [15], sympathetic or neurohormonal activation [16], central suppression of the parasympathetic nervous system [17] and LPS-triggered cytokine release [18]. The latter hypothesis is supported by the growing evidence of increasing amounts of LPS entering the circulation through an oedematous, hypoperfused bowel wall, thus leading to a severely disturbed intestinal microcirculation in CHF [18-22]. A multitude of conditions may contribute to making the bowel wall less resistant to such a translocation. Intramucosal acidosis, which occurs in about 50% of patients with circulatory failure [23-25] points to an inadequate oxygen supply and intestinal ischaemia [26]. An increase in gastric intramucosal carbon dioxide pressure occurs in decompensated CHF patients even at low levels of exercise [27] . Diminished gut circulation due to redistribution of splanchnic blood flow away from the gut under enhanced systemic sympathetic tone, and disturbed microcirculation are thought to contribute to local oedema of the bowel wall and to malabsorption and barrier dysfunction of the mucosa [22]. This mechanism seems plausible to explain the finding that bioactive LPS concentrations are higher in blood samples from the hepatic veins than in those of the left ventricle in patients with acute decompensated CHF [21]. Furthermore, a selective decontamination study of the gut in patients with CHF resulted in a decrease in some inflammatory markers such as monocyte CD14 expression. This underscores the potential importance of gut bacteria as one source of systemic inflammation in CHF [28]. Bowel wall oedema may contribute to fat malabsorption, which has been shown to occur in patients with cardiac cachexia [29]. Furthermore, loss of proteins has been described due to CHF [30]. We have recently reported altered intestinal morphology and function with increased bowel wall thickness, increased intestinal permeability of both the small and the large intestine, and decreased carrier-mediated D-xylose absorption, all together indicating bowel ischaemia and dysfunction of transport proteins. Moreover, we found higher concentrations of adherent bacteria within the sigmoid mucus and higher serum levels of immunoglobulin (Ig) A-anti-LPS in patients with stable CHF compared to healthy control subjects [22]. In the absence of systemic inflammatory responses these higher IgA-anti-LPS (*E. coli*-J5-endotoxin) levels in the patients may reflect higher LPS bioactivity and mucosal interaction. We speculate that this interaction presumably takes place in the intestinal mucosa with the gut being the largest source of LPS structures. Theoretically, further mucosal regions, such as lung tissue, could also be affected. There is considerable variation in the response to inhaled LPS in the literature. Under laboratory conditions it potentially causes a mild dose-dependant airflow obstruction, dry cough, and a mild increase in bronchial reactivity, and is able to cause systemic complaints, a rise of body temperature, and laboratory signs of systemic inflammation [31, 32]. In a small dose-response relationship study in 9 healthy volunteers the most sensitive markers of LPS-induced inflammation were the blood polymorphonuclear neutrophil (PMN) count with their level of activation, the blood CRP concentration, and the sputum's PMN count. The no-response threshold to acute inhalation of LPS was less than 0.5 µg [33]. However, the possibility of *E. coli* being present in the lung is of course conceivable, but the intestinal presence of *E. coli* is higher. Therefore we think the gut has greater potential to contribute to the higher IgA-anti-*E. coli* J5-endotoxin level in the patients.

Alterations of the intestinal mucosa may contribute to both chronic inflammation and malnutrition, with slightly lower cholesterol seen in patients with CHF predicting poor survival. The particularly devastating prognosis of patients with cardiac cachexia [34], a catabolic condition [35] where circulating cytokine levels are known to be the highest [7, 36, 37], underlines this view. Importantly, cachectic CHF patients do not have lower serum cholesterol levels than those who are metabolically stable.

In 2000 the endotoxin-lipoprotein hypothesis [8] was published, postulating that lipoproteins in the plasma may form micelles around LPS – a cell wall component from Gram-negative bacteria. Elevated plasma concentrations of LPS are present in CHF patients during oedematous decompensations, but also in progressive severe CHF [19, 38]. We have shown that very small amounts of LPS are able to induce TNF secretion in an ex vivo model (whole blood culture) of patients with CHF [19]. In the circulation of CHF patients, LPS may activate monocytes and macrophages to release pro-inflammatory mediators such as tumour necrosis factor alpha (TNF), thus leading to a pro-inflammatory response.

According to our hypothesis, serum lipoproteins serve as important regulators of LPS bioactivity by binding this substance [39]. Circulating lipoproteins detoxify LPS by preventing its biological function and are potentially beneficial by serving as a buffer for LPS at times of increased exposure, thereby limiting systemic inflammatory activation. The ability of lipoprotein fractions to bind LPS may explain the inverse relationship between lower lipoprotein levels and higher concentrations of soluble TNF-receptor-1/2 and TNF [40]. Importantly, inflammatory markers, and particularly soluble TNF-receptor-1, are a powerful predictors of impaired survival in patients with CHF [41].

Low Lipoproteins and Pro-Inflammation: Causality or Epiphenomenon?

Concept A

Endogenously and/or exogenously induced reductions of circulating lipoproteins can cause a loss of the serum's capacity to bind and detoxify LPS. Increased LPS bioactivity in turn leads to the production of pro-inflammatory cytokines that may, through a negative feedback mechanism, further lower serum lipoproteins. The systemic inflammatory response due to circulating LPS and pro-inflammatory cytokines, in concert with other maladaptive mechanisms, may result in worsening heart failure and, ultimately, increased heart failure-related mortality (Figure 1).

Concept B

Bacterial translocation may occur, but does not substantially contribute to elevated cytokine concentrations in patients with CHF. Inflammatory cytokines, presumably derived from other than mononuclear cells, exert catabolic effects at different sites of the human body, leading to an imbalance of anabolic/catabolic drive. As a result, lipoprotein levels fall like other surrogate markers, pointing to increased mortality in end-stage heart failure. Lipoproteins are not causally linked to the progression of heart failure, and are an end-stage epiphenomenon (Figure 2).

Measurement of LPS

LPS of Gram-negative bacteria can be measured by the Limulus amoebocyte lysate (LAL) assay, in which LAL clots in the presence of LPS. While the gel-clot LAL assay is biased due to individual and subjective evaluation of a given extent of clotting, the chromogenic LAL assay is less observer- but more temperature- and time-dependent. Therefore, turbidimetric assessment is often used. The LAL method has advantages over the possible rabbit-based pyrogen testing because it is more sensitive, requires a lower amount of sample specimen, and the assays can easily be repeated [42]. This test has a very high sensitivity with a lower limit of detection

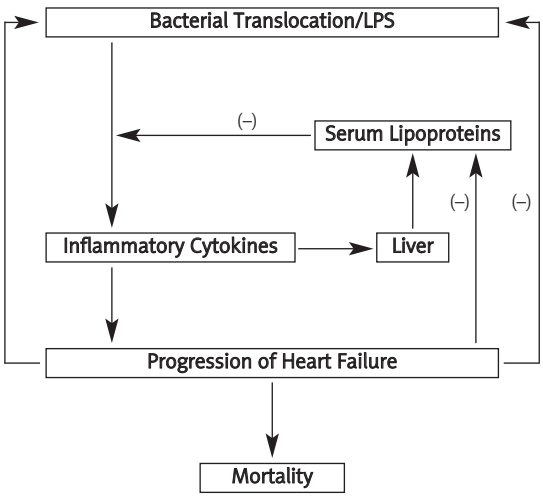

Figure 1. Concept A. Low serum lipids causally trigger pro-inflammation

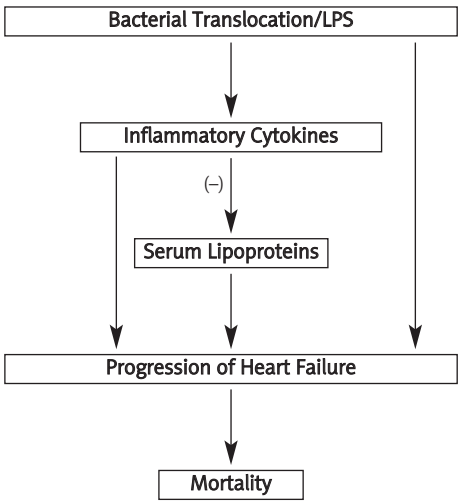

Figure 2. Concept B. Low serum lipids as an epiphenomenon triggered by pro-inflammation

of 1-5 pg/ml of blood LPS in humans, particularly in healthy individuals where this limit is often lower [43]. Furthermore, a variety of substances, such as bile acid, proteins, lipoproteins, heparin, and antibiotics like aminoglycosides and penicillin are able to build complexes with LAL, thereby interfering with the testing. Therefore, test validity depends on the material tested, with sufficient specificity in urine and cerebrospinal fluid. Due to the high sensitivity of this test, contaminations during or after taking of the sample may create false positive results. Moreover, there is a variety of endotoxins reacting each in their individual pattern, with differing standard curves for different LAL preparations, which makes interpretation of the test results difficult. Another important fact is that only free LPS is measured, whereas tests fail to detect membrane-bound endotoxin. Thus, interpretation of an individual endotoxin concentration is influenced by individual receptor configurations. For all these reasons measurement of LPS bioactivity by assessing immunoglobulin (Ig)-anti-LPS poses a more accurate diagnostic avenue.

All considered, so far precise testing for LPS in humans is not possible and needs to be approached with caution. However, available tests are usable for research purposes but one needs to keep in mind that none of the tests are certified for human samples. Further input from the industry and more scientific work are required to overcome these limitations, leading to a readily available testing system for humans. In the meantime, measuring IgA-anti-LPS may be an alternative.

Effects of inflammation in CHF

There are direct and indirect as well as time-dependent effects of inflammatory cytokines on the myocardium, the endothelium, and cells of the innate immune system [12]. All affected organ systems finally lead to disturbances of the immunological balance [12, 17], separately or jointly supporting the progression of CHF. Inflammatory effects on the myocardium include negative inotropic effects [44], cardiomyocyte hypertrophy [45], and apoptosis [46]. The activation of the immune system contributes to endothelial dysfunction. Interleukins and TNF are inhibitors of food intake [47]. This may also contribute to anorexia and body wasting in CHF [34, 35].

CHF is a state of neurohumoral imbalance and an enhanced sympathetic tone [48, 49]. In that condition unfavourable additional LPS-stimulated catecholamine release by granulocytes and phagocytes as shown by Flierl et al. [50] should be prevented. This is of particular importance because these cells in turn are able to promote immuno-stimulation and sympathetic overdrive even more. This points to an increasingly recognized cross-talk between the autonomic nervous system and the immune system during inflammation in CHF [50]. These mechanisms may aggravate restricted bowel perfusion and contribute to extended LPS translocation followed by even more enhanced inflammation.

LPS signalling

It is current thinking that LPS signals are mediated via two receptors on the cell membrane. Initially free LPS binds to LPS-binding protein (LBP). This is followed by a transfer of LPS to membrane-bound or soluble CD14 [51]. For complete LPS recognition the trans-membrane Toll-like receptor 4 (TLR4) [46, 52, 53] and an MD2 molecule [52] are necessary. The final TLR4-MD2 complex in response to LPS interaction forms complexes with heat-shock protein (Hsp) 90 and Hsp70, the levels of the latter being higher in patients with CHF than in healthy control subjects [54].

The vascular endothelium is one key target of LPS and the first host tissue barrier to encounter circulating LPS that is shed from replicating or dying Gram-negative bacteria [53]. LPS is regarded as one of the most powerful triggering factors of cytokine release but beyond this has further direct and indirect effects on the cardiovascular system. LPS stimulation has been demonstrated to result in expression of adhesion molecules by endothelial cells, which leads to increased binding of thrombocytes to endothelial cells und within one hour after injection [42] to an elevated number of platelet-monocyte aggregates. After intravenous administration of LPS to healthy volunteers Kälsch et al. [55] observed activation of platelets and monocytes with upregulation of pro-atherogenic CD40L on platelets. LPS triggered tissue factor (TF) is a lipoprotein that promotes the extrinsic pathway of the coagulation system, thereby critically contributing to microthrombus formation [56, 57]. Plasma TF levels have been shown [58] to predict poor prognosis in CHF, possibly via developing thrombotic complications [59]. Thus, one may speculate that blocking LPS could potentially stabilize the coagulation system. Interestingly, Mizuochi et al. [59] recently reported that carvedilol, a non-selective β-adrenoceptor antagonist with α_1-adrenoceptor blocking action, inhibited the production of TNF and TF in LPS-stimulated monocytes in vitro via pleiotropic effects, independent of its adrenoceptor inhibitory activities in monocytes. Thus, additional pleiotropic inhibitory effects on the LPS cascade could potentially contribute to the observed reduction in mortality by 35% in CHF patients receiving carvedilol compared to placebo in the Carvedilol Prospective Randomized Cumulative Survival Study (COPERNICUS) [60]. Several attempts have been made previously in order to block the

LPS cascade and to prevent pro-inflammatory stimulation. Initial studies on TNF antagonists as one of those approaches showed disappointing results in CHF, possibly due to problems not directly linked to the drug used [61]. Interestingly, direct blocking of the CD14-recognition receptor for LPS by IC14, a recombinant anti-CD14 monoclonal antibody [62], has been demonstrated to inhibit LPS-induced cytokine release in healthy subjects [63]. In 16 healthy male volunteers its administration followed by infusion of LPS (4.0 ng/kg) led to significant reductions in the release of TNF, IL-6, and IL-10. Furthermore, IC14 has been shown to successfully suppress ex vivo endotoxin stimulated TNF in patients with CHF [64], thereby being well tolerable and safe in administration.

Among the substances that have the property of forming micelles around LPS, thereby inactivating it, are conjugated bile salts, which have recently been reported [65] to reduce permeability of endotoxin through intestinal epithelial cells in vitro, finally suppressing inflammatory cytokine production dose-dependently. To date, there are no prospective studies investigating bile salts beside cholesterol as a predictive factor for survival in patients with CHF.

Specific protective role of cholesterol in the setting of increased endotoxin exposure

Serum lipoproteins can protect against lethal endotoxaemia and severe Gram-negative infections, as shown in animal models [66-68]. Reconstituted HDL appears to act as a potent inhibitor of TNF production by LPS-stimulated whole human blood [66], and similar LPS-buffering effects have been shown for other lipoprotein moieties [69]. In-vitro studies in humans using highly purified plasma lipoproteins demonstrated that they can inhibit endotoxin activity in a time- and dose-dependent fashion [70]. Lipoproteins in humans have been shown to inhibit the effects of LPS by binding and neutralizing it [69], resulting in reduced CD14 expression on monocytes [71] and reduced cytokine and pro-coagulatory responses [72, 73].

We found an inverse relationship between whole blood LPS stimulated TNF release and serum cholesterol which was strongest at 0.6 ng/ml of LPS in CHF patients ($r=-0.53$, $p=0.002$) [73]. Further evidence for the anti-inflammatory action of cholesterol comes from a study by Englund et al. [74] that demonstrated cholesterol to be able to inhibit inflammatory cytokine production at both translational and transcriptional levels. Incubation of macrophages in vitro with cholesterol was shown to decrease LPS-induced TNF release and also mRNA expression, demonstrating an effect similar to LPS desensitization [19, 75].

The proposed mechanism of action is to expand the phospholipid surface on plasma lipoproteins and thereby facilitate LPS neutralization. In this regard, administration of other lipid formations such as fish oil, whose intake is being discussed in order to improve survival in CHF patients, is gaining interest. Intravenous administration of n-3 fatty acids has been observed to result in reduced mononuclear cytokine generation provoked by ex vivo endotoxin challenge [76]. This finding is consistent with previous reports demonstrating that dietary n-3 fatty acids may suppress TNF and IL-1 release from mononuclear cells [77, 78].

Lipoproteins may act as a shield against inflammation, especially when the liver becomes less capable of clearing portal venous blood of endotoxins. Liver blood congestion is a well-known feature of the heart failure syndrome which presents particularly with an increase in pressure of the central veins and hypoxaemia of the periportal acini. This may result in rising LPS concentrations. The latter is seen in CHF patients with oedematous decompensation – where increased plasma levels of LPS are present [9]. However, absolute LPS blood load is difficult to assess, especially when the liver's clearing function is sustained and micelle forming substances like cholesterol act as buffers. This is the reason why even patients with severe ulcerative colitis can display a normal amount of free LPS in systemic blood [79]. Therefore, rather than LPS, IgA-anti-LPS, which has been shown to be higher in patients with CHF compared to controls [22] provides a useful alternative for measuring endotoxin bioactivity and interaction in the individual patient.

Interestingly, in patients with liver cirrhosis, portal hypertension, consecutive fluid congestion, and microcirculatory problems, similar LPS buffering abilities of lipoproteins such as of HDL have been observed [80]. In sepsis with highly upregulated systemic inflammation, higher lipoprotein levels are known to be related to a better outcome [81-84] and to fewer infectious complications [85]. Although it has long been believed that LPS is cleared by resident hepatic macrophages (Kupffer cells) and then delivered to hepatocytes, more recently, hepatocytes have been reported to internalize LPS independently of Kupffer cells [86], and the rate at which hepatocytes internalize LPS increases markedly not only during sepsis [87] but also with the infusion of lipoproteins [80, 88]. Chylomicron-bound LPS attenuates the hepatocellular response to pro-inflammatory cytokines, as shown in a rodent model [89]. This induction of cytokine tolerance in hepatocytes follows the internalization of chylomicron-LPS complexes, a process regulated by the LDL receptor, and may indicate a negative regulatory mechanism for the hepatic response to sepsis, serving to effectively downregulate the acute phase response [89].

Statins in CHF

Theoretically, cholesterol lowering statin therapy may potentially act at least in part in a pro-inflammatory manner due to the reduction of the plasma buffering capacity via the reduction of lipoproteins. Statins have been reported to exert beneficial effects in CAD not only related to plasma cholesterol, but also due to pleiotropic actions such as the reduction of plaque thrombogenicity, inhibition of cellular proliferation and migration, and improvement of endothelial function [53]. Statins are thought to inhibit the production of cytokines such as IL-1β in the endothelium [90] and have been shown to reduce inflammation [91], free-radical production in the vascular wall, and to increase the expression and activity of endothelial nitric oxide synthase [92] and the expression of endothelial progenitor cells that are involved in vascular repair. By upregulation of LDL receptors on hepatocytes [53], as observed in animal sepsis models, statins may contribute to the internalization of lipoprotein-LPS complexes, which at least for chylomicrons has been shown to induce cytokine tolerance in hepatocytes. This may contribute to a negative regulatory mechanism for the hepatic response to sepsis, serving to effectively downregulate the acute phase response [89].

Two potential disadvantages of statins in the particular setting of CHF are the ubiquinone- and lipid-lowering actions. Statins decrease the concentration of ubiquinone (coenzyme Q10), dietary supplementation of which has been shown to improve symptoms in patients with CHF [93]. Ubiquinone is known to exert antioxidative and membrane-stabilizing properties and is an essential intermediate in mitochondrial phosphorylation, necessary for the production of ATP. Its degree of depletion has been shown to be correlated with the severity of CHF [94]. However, intervention strategies to supply coenzyme Q10 did not result in survival benefits in CHF [95].

The role of lipid lowering therapy in patients with CHF is being discussed controversially. It could be potentially harmful in CHF due to decreased endotoxin defence, thereby counterbalancing the various beneficial pleiotropic effects of statins in CHF patients. Post-hoc analyses of secondary prevention trials found statin use to be associated with reduced occurrence of de novo heart failure [96], but most patients with CHF did not enter these studies because LVEF <40% was an exclusion criterion.

Small prospective studies have demonstrated promising results. Liao et al. [97] found in a study of 63 patients after a short intervention period of only 14 weeks of treatment with simvastatin an improved left ventricular ejection fraction in the verum group, accompanied by lower plasma concentrations of TNF, IL-6 and brain natriuretic peptides. Similar improvement in myocardial function was observed with atorvastatin [98]. Retrospective analysis of the OPTIMAAL (Optimal Therapy In Myocardial infarction with the Angiotensin II Antagonist Losartan) trial showed that statin treatment, prescribed in addition to a beta-blocker and an inhibitor of the renin-angiotensin system, was associated with better survival and less development of heart failure after acute myocardial infarction [99].

However, until only recently we did not have strong evidence to support a role for statins in the routine management of CHF patients as underlined by level C of evidence in the current guidelines [100]. Recently published data from the CORONA (Controlled Rosuvastatin Multinational Trial in Heart Failure) study [101] document for the first time in a prospective, randomized, placebo-controlled, multicentre large-scale statin intervention trial evidence from 5,011 elderly patients with CHF of ischaemic origin. The primary endpoint was a composite of cardiovascular death, nonfatal myocardial infarction or nonfatal stroke. The secondary endpoints were death from any cause, any coronary event, death from cardiovascular causes, and the number of hospitalizations for cardiovascular causes, unstable angina, or worsening heart failure. Although 10 mg of rosuvastatin, the statin used in CORONA, did reduce hospitalization for worsening CHF ($p=0.01$) and for all cardiovascular reasons ($p<0.001$) in this landmark trial, there was no benefit regarding the primary composite endpoint of cardiovascular death, nonfatal myocardial infarction or nonfatal stroke in CHF patients during the observational period of 38 months. At the end of the study, the composite primary end-point had occurred in 692 patients in the rosuvastatin and in 732 patients in the placebo group (HR 0.92, 95% CI 0.83-1.02, $p=0.12$). For the time being, one can only speculate that a lower dose of rosuvastatin (decreasing cholesterol less powerfully) might have exerted survival benefits in these patients. The counterbalancing effects of anti-inflammatory activity versus pro-inflammatory, lipid-dependent mechanisms make it difficult to study the pure effects of changes in lipids over time against anti-inflammatory effects over time. Importantly, the lipid lowering drug ezetimibe without statin-like pleiotropic effects did not show improvements in endothelial function [102]. In regard to CORONA we remain with the question of whether the concept of statin therapy in CHF does not work due to different modes of death. Did the protocol not meet the requirements of a CHF population? Alternatively, we may ask whether it is a drug or class effect, i.e. could we expect different results using other statins? Or is it a dose-dependent effect, leading to an imbalance between pleiotropic and lipid-lowering effects?

So far, the scientific community is not able to give a clear-cut indication for the use of statins in patients with CHF. With respect to the results of CORONA we only know that a powerful statin like rosuvastatin given to patients with ischaemic, systolic CHF in NYHA class II-IV does not result in improved outcome as regards to the above-mentioned endpoints. Further ongoing studies such as GISSI (n-3 polyunsaturated fatty acids and rosuvastatin in symptomatic congestive heart failure) and JUPITER (Justification for the Use of statins in Primary prevention: an Intervention Trial Evaluating Rosuvastatin) will hopefully shed more light on this important clinical question.

Conclusion

CHF has long been recognized as a chronic condition with systemic inflammation. Paradoxically in CHF, higher levels of lipoproteins have consistently been related to better survival – a phenomenon called the "cholesterol paradox". According to the endotoxin-lipoprotein hypothesis this bizarre interaction could be explained by the ability of serum lipoproteins to detoxify LPS through micelle formation mediated by lipoproteins. Alternatively, lipoproteins could just reflect a better metabolic reserve as the syndrome of CHF is a catabolic state. However, we do not have data to evidence the discussion in either way, nor do we have data to reject our hypothesis. Since immune activation and metabolic demands have been under research for years without finding the key to this question, we still have a clear task to do further basic research and clinical studies. This may provide additional evidence for the intriguing role of lipids buffering endotoxin. Furthermore and in addition to CHF, we should focus on the pathophysiology of LPS in other chronic conditions such as liver cirrhosis, COPD, rheumatoid arthritis, and sepsis. There is a big field for clinical research; the time has come to address this issue.

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