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REVIEWS: CURRENT TOPICS

The relation of saturated fatty acids with low-grade inflammation and cardiovascular disease

Begoña Ruiz-Núñez*, D.A. Janneke Dijck-Brouwer, Frits A.J. Muskiet

Department of Laboratory Medicine, University of Groningen, University Medical Center Groningen, Groningen, The Netherlands

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Abstract

The mantra that dietary (saturated) fat must be minimized to reduce cardiovascular disease (CVD) risk has dominated nutritional guidelines for decades. Parallel to decreasing intakes of fat and saturated fatty acids (SFA), there have been increases in carbohydrate and sugar intakes, overweight, obesity and type 2 diabetes mellitus. The "lipid hypothesis" coined the concept that fat, especially SFA, raises blood low-density lipoprotein-cholesterol and thereby CVD risk. In view of current controversies regarding their adequate intakes and effects, this review aims to summarize research regarding this heterogenic group of fatty acids and the mechanisms relating them to (chronic) systemic low-grade inflammation, insulin resistance, metabolic syndrome and notably CVD. The intimate relationship between inflammation and metabolism, including glucose, fat and cholesterol metabolism, revealed that the dyslipidemia in Western societies, notably increased triglycerides, "small dense" low-density lipoprotein and "dysfunctional" high-density lipoprotein, is influenced by many unfavorable lifestyle factors. Dietary SFA is only one of these, not necessarily the most important, in healthy, insulin-sensitive people. The environment provides us not only with many other proinflammatory stimuli than SFA but also with many antiinflammatory counterparts. Resolution of the conflict between our self-designed environment and ancient genome may rather rely on returning to the proinflammatory/antiinflammatory balance of the Paleolithic era in consonance with the 21st century culture. Accordingly, dietary guidelines might reconsider recommendations for SFA replacement and investigate diet in a broader context, together with nondietary lifestyle factors. This should be a clear priority, opposed to the reductionist approach of studying the effects of single nutrients, such as SFA.

Keywords: Saturated fat; Coronary artery disease; Cholesterol; Immune system; Metabolism

1. Introduction

The global burdens of the metabolic syndrome, and its consequences, notably type 2 diabetes mellitus (DM2) and cardiovascular disease (CVD) are alarmingly rising, producing enormous losses of life quality in both developed and developing nations [1]. Most of these burdens are preventable, since they are largely due to suboptimal lifestyle, including excessive caloric intake, unbalanced diet, physical inactivity, insufficient sleep, chronic stress, unhealthy environment (*e.g.* smoking) and abnormal microbial flora [2–4]. Hominins have faced major changes in both dietary and physical activity patterns and body composition since our Paleolithic ancestors emerged on Earth some 2.5 million years ago. Nowadays, there are striking differences in dietary habits and rates of chronic diseases worldwide [5], and therefore, the identification and targeting of dietary factors with the greatest potential for reducing chronic diseases, notably DM2 and CVD, are of major public health importance [6].

Fat, carbohydrates (CHO) and proteins are the primary energy-containing macronutrients consumed on a routine basis by humans. In this context, the quality, rather than the quantity, of dietary CHO and fat has become a relevant issue in the nutritional origins of cardiometabolic conditions [6]. Among the macronutrients, fat contains the highest amount of energy per gram. A "consumed

Abbreviations: AHA, American Heart Association; CHO, carbohydrates; CLA, conjugated linoleic acid; CRP, C-reactive protein; CVD, cardiovascular disease; DHA, docosahexaenoic acid; DM2, type 2 diabetes mellitus; DNL, *de novo* lipogenesis; EPA, eicosapentaenoic acid; FA, fatty acids; GI, glycemic index; GL, glycemic load; GPR, G-protein-coupled receptor; HDL, high-density lipoprotein; HDL-C, high-density lipoprotein-cholesterol; HPA, hypothalamus-pituitary-adrenal; HPG, hypothalamus-pituitary-gonadal; LA, linoleic acid; LDL, low-density lipoprotein; LDL-C, low-density lipoprotein-cholesterol; LPS, lipopolysaccharide; LBP, LBP, lipopolysaccharide-binding protein; MCFA, medium-chain fatty acids; MESA, Multi-Ethnic Study of Atherosclerosis; MUFA, monounsaturated fatty acids; NAFLD, nonalcoholic fatty liver disease; NASH, nonalcoholic steatohepatitis; NFκB, nuclear factor kappa B; PUFA, polyunsaturated fatty acids; RCT, randomized controlled trial; SCFA, short-chain fatty acids; SFA, saturated fatty acids; TLR, toll-like receptor; TC, total cholesterol; TG, triglyceride; VLDL, very low-density lipoprotein.

* Corresponding author. Department of Laboratory Medicine, University Medical Center Groningen Rijksuniversiteit Groningen, Building 33, 3rd Floor, Room Y3.181, Internal Zip Code EA61, Hanzeplein, 1, P.O. Box 30.001, 9700 RB Groningen, The Netherlands. Tel.: +31-50-361-0363; fax: +31-50-361-2290.

E-mail address: bego@becafisiosalud.com (B. Ruiz-Núñez).

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calorie" is, however, not the same as an "available calorie", since isocaloric diets with different macronutrient compositions have different effects on resting and total energy expenditure [7]. Total and resting energy expenditures decrease in the sequence: very low CHO diet (CHO:fat:protein = 10:60:30 energy%)>low glycemic diet (40:40:20 energy%)>low fat diet (60:20:20 energy%) [8], implying that, on an isocaloric basis, the diet with the highest protein and fat contents gives rise to the lowest weight gain.

The interest in the relation between dietary fat and CVD arose from animal studies indicating that dietary cholesterol caused arterial lesions, largely mediated through an elevation of blood cholesterol levels [9]. Since then, the relation between dietary fat and CVD risk has been intensively investigated, using different approaches, including controlled feeding studies, randomized controlled trials (RCTs) and large cohort studies [10]. One of the larger studies examining CVD risk factors, the World Health Organization Multinational Monitoring of Trends and Determinants in Cardiovascular Disease, already concluded, at the end of last century, that there was no clear relationship between total cholesterol (TC) and CVD [11]. Even more striking is the observation that two thirds of people admitted for acute coronary events suffer from the metabolic syndrome, but 75% of these exhibit completely normal TC and low-density lipoprotein-cholesterol (LDL-C) concentrations [12].

The "lipid hypothesis" of CVD originated from the investigations of Ancel Keys in the 1950s [13] and became exacerbated after his Seven Countries Study in the 1970s [14]. Keys claimed that there was a correlation between high dietary fat content, particularly saturated fatty acids (SFA), and both elevated serum TC and LDL-C, and thereby of CVD risk (for a historical review, see Ref. [10]). Since then, dietary fat, and especially the consumption of SFA, has been consistently demonized [15]. Yet, the reduction in LDL-C from reducing SFA intake seems to be specific for large and buoyant low-density lipoprotein (LDL) particles, while the small and dense LDL particles are in fact the ones implicated in CVD [16,17]. Accordingly, the levels of small dense LDL have been shown to increase in response to low-fat/high-CHO diets [18,19].

Most of the studies on SFA solely focused on their tendency to alter lipoprotein metabolism and thereby influence the concentrations of lipoproteins carrying cholesterol in blood. Therefore, the question of what constitutes the healthiest overall mixture of the different classes of dietary fats still remains unanswered. Both agricultural and food industries are guided by recommendations to the public to decrease SFA intakes to "as low as possible", but in dietary guidelines, no lower limit of SFA intake has yet been identified [20].

In view of the controversy regarding adequate intakes and the complex effects of fatty acids (FA), notably SFA, this review aims to summarize research findings and observations regarding this particular group of FA and the mechanisms that relate them to (chronic) systemic low-grade inflammation, insulin resistance, the metabolic syndrome and eventually the development of CVD, among many other typically Western diseases associated with the metabolic syndrome.

2. SFA: Function and occurrence

The study of lipids, and FA as their major structural elements, remains one of the most enigmatic and complex research fields in biology and nutrition. As one of the energy-producing macronutrients in our diet, fat provides essential FA, and dissolves and assists in the absorption of fat-soluble vitamins and other essential nutrients. Dietary fat induces metabolic effects that are a complex consequence of the FA composition of food, its timing and intraindividual and interindividual variations. FA are key elements of all tissues. They are required for several basic functions in animals, playing pivotal roles as energy sources [21], elements of membrane phospholipids [22] and the main fuel (50%) for the production of prolonged low-intensity shivering, under cold circumstances [23]. The type of dietary fat affects vital functions of the cell and its ability to resist dysfunction, *e.g.* by influencing the functions of membrane-embedded receptors, enzymes and transport systems, by determining basic membrane characteristics and by producing highly active lipid mediators [24,25].

SFA, as part of FA, are used as, among others, energy source, building blocks for structural elements, for protein modification and for regulation of gene transcription [21]. Compositional analyses have shown remarkable specificities for particular SFA in cellular compartments [26], though the metabolic aspects and health effects of the individual SFA are hard to examine [27]. Adipose tissue and liver own the capacity to *de novo* synthesize and store SFA, particularly palmitate (16:0), from polar precursors, notably glucose [28,29]. In addition to 16:0, the mammary gland owns the means to produce other specific SFA, such as myristic (14:0) and lauric (12:0) acids, providing a source of easily available energy, emulgating capacity and microbial protection to ensure growth, development and survival of the mammalian offspring [30].

SFA usually account for 30–40 g% of total FA in human tissues, distributed among 16:0 (15–25 g%), stearic acid (10–20 g%), 14:0 (0.5–1 g%) and 12:0 (less than 0.5 g%) [31]. Palmitic and stearic acids are universally found in natural fats, while 12:0 is especially abundant in coconut (39–54 g%) and palm kernel (44–51 g%) oils (Table 1). Major sources of 14:0 are butter fat and both coconut and palm kernel oils [27]. However, the principal dietary sources of SFA in most Western countries are full-fat dairy products, red meat and grain-based dishes (*e.g.* pizza, pasta and desserts) [32–34], the latter also being rich in *trans* FA, low in polyunsaturated fatty acids (PUFA) of the ω3 series and with a high glycemic load (GL), a composition remotely away from the diet consumed by our Paleolithic ancestors.

Without yet entering the debate on the deleterious or beneficial effects of the different types of SFA, it has become clear that they cannot be considered as a single group in terms of structure, metabolism or cellular function [35,36]. Our body is capable of synthesizing SFA from CHO, *via de novo* lipogenesis (DNL) [28], which are basically the same FA as present in dietary fats of animal origin, mainly 16:0, but also 18:0, 14:0 and 12:0 to a lesser extent [37]. Myristic and palmitic acids are directly involved in two classes of posttranslational protein modifications, namely N-terminal myristoylation and side-chain palmitoylation [38]. The reversible attachment of 16:0 to the sulfur atom of cysteine facilitates protein–membrane interactions and the intracellular movement of proteins, and it is involved in a variety of signal transduction pathways. Myristoylation includes key components in intracellular signaling pathways, oncogenes, structural viral proteins and common constitutive eukaryotic proteins [31]. The so-called medium-chain SFA [medium-chain fatty acids (MCFA); 6–12 carbons] are mainly oxidized in the liver, as *e.g.* demonstrated by the low-fat deposition in adipose tissue from rats overfed with a medium-chain triglyceride (TG) diet compared with rats overfed with isocaloric long-chain TGs [39].

Complex CHO, such as dietary fiber, are metabolized by the colon microbiota and then fermented to short-chain fatty acids (SCFA; less than 6 carbons), mainly acetate, propionate and butyrate [40]. The latter is used as a fuel metabolite by the colonic epithelial cells and simultaneously prevents autophagy in these colonocytes [41]. Acetate and propionate are transported to the liver and peripheral organs, where they become substrates for gluconeogenesis and lipogenesis [40]. In addition to serving as energy sources, SCFA, notably butyrate, also affect colonic gene expression *via* histone deacetylase inhibition [42] and *via* metabolic regulation through signaling *via* G-protein-coupled receptors, such as GPR43. For example, SCFA have been shown to suppress inflammation through GPR43 signaling in immune cells [43] and to modulate glucagon-like peptide 1 secretion, thereby improving insulin secretion [44]. Moreover, butyrate induces

Table 1
Fat and FA content (saturated, monounsaturated and polyunsaturated) in different foods in g%

	Oils					Dairy products				Meat and fish					
Fat and FA	Coconut oil[a]	Palm oil[a]	Fish oil (cod liver)[a]	Olive oil[a]	Sunflower oil (high LA, >65%)[a]	Butter[a]	Cheese, parmesan[a]	Milk, cow[a]	Milk, sheep[a]	Beef, tenderloin, raw[a]	Beef, sirloin, steak, raw[b]	Chicken (white meat)[b]	Game meat, antelope, raw[a]	Game meat, moose, raw[a]	Fatty fish (salmon, Australian)[c]
Fat content	100.00	100.00	100.00	100.00	100.00	81.11	25.83	3.25	7.00	21.83	11.13	1.60	2.03	0.74	11.10
SFA	86.50	49.30	22.61	13.80	10.30	61.92	63.53	57.23	65.76	41.04	40.35	23.13	36.45	29.73	28.03
12:0	44.60	0.10	NA	0.00	0.00	2.79	3.38	2.37	3.41	0.18	0.22	0.00	0.00	NA	NA
14:0	16.80	1.00	3.57	0.00	0.00	10.01	11.27	9.14	9.43	3.07	3.16	0.63	0.99	0.00	0.80
16:0	8.20	43.50	10.63	11.30	5.90	26.17	26.97	25.51	23.17	23.96	24.29	16.25	16.75	12.16	19.02
18:0	2.80	4.30	2.80	2.00	4.50	12.06	8.91	11.23	12.84	12.69	12.36	6.25	18.23	17.57	6.10
MUFA	5.80	37.00	46.71	73.00	19.50	28.13	29.09	24.98	24.63	42.42	42.81	30.00	23.65	20.27	25.80
18:1ω9	5.80	36.60	NA	71.30	19.50	25.03	25.77	24.98	22.26	37.29	37.80	25.00	23.65	18.92	16.70
PUFA	1.80	9.30	22.54	10.50	65.70	3.69	2.20	6.00	0.31	3.99	3.80	25.00	21.67	32.43	40.00
LA	1.80	9.10	0.94	9.80	65.70	2.25	1.05	3.69	2.59	2.57	2.44	15.00	12.32	18.92	2.90
AA	0.00	0.00	0.94	0.00	0.00	0.00	0.00	0.00	0.00	0.04	0.02	NA	0.12	0.07	5.10
ALA	0.00	0.20	0.94	0.80	0.00	1.45	1.15	2.31	1.81	1.15	1.19	0.63	3.45	4.05	0.80
EPA	0.00	0.00	6.90	0.00	0.00	0.00	0.00	0.00	NA	NA	0.00	0.00	NA	NA	6.60
DHA	0.00	0.00	10.97	0.00	0.00	0.00	0.00	0.00	NA	NA	0.00	1.25	NA	NA	24.60

Abbreviations: AA, arachidonic acid; ALA, alpha-linolenic acid; DHA, docosahexaenoic acid; EPA, eicosapentaenoic acid; LA, linoleic acid; MUFA, monounsaturated fatty acids; NA, not available; PUFA, polyunsaturated fatty acids; SFA, saturated fatty acids.
[a] Data from [312].
[b] Data from [318].
[c] Data from [314].

apoptosis in a variety of tumor cells [45] while other SFA may also influence apoptosis *via* the ceramide pathway through the induction of ceramide *de novo* synthesis at several steps, including serine condensation with palmitoyl-CoA [46].

3. Human milk SFA

The FA in milk deserve special attention, since milk is the only food that is produced by the animals' own biochemical machinery and therefore provides insight into the animal's physiological needs and evolutionary past. Human milk may consequently harbor information on the importance of many nutrients, including SFA. Nevertheless, the optimal human milk composition has been subject of study for decades. There is *e.g.* no gold standard for the human milk FA composition. This composition is dependent on the short- and long-term maternal diet [47], while there is a lack of consensus regarding the optimal maternal diet [48]. The large worldwide variability of the human milk FA composition is testimony of the wide variety of foods tolerated by human beings, with dietary FA and CHO as major determinants [48]. There is, however, some degree of unity, and this is notably the case for SFA.

SFA represent around 40–60 g% of human milk FA [49], while 14:0 and shorter-chain SFA (less than 12 carbons) usually represent each about 10 g% [31] and 16:0 is about 22 g% [48] (Table 2). Despite the large biological variation in the composition of human milk FA, palmitate exhibits the lowest worldwide biological variation [48]. Myristic acid, 12:0 and shorter-chain SFA are produced in the mammary gland under the influence of a unique uncoupling protein that functions in the local *de novo* FA synthesis (DNL) from glucose. These MCFA are largely incorporated into milk TGs, following a CHO-rich meal [50]. It is, however, unlikely that, in the past, abundant dietary CHO served as a major substrate for MCFA production in the mammary gland, since the routine consumption of CHO-rich diets was not part of our culture until the start of the Agricultural revolution, some 10,000 years ago [51]. The inhabitants of the island of Chole and of Dar-es-Salaam, both in Tanzania, present low-CHO intakes from grains and corn and high consumptions of coconut. Their milks exhibit the highest caprylic acid (8:0) and 12:0 contents [51] (Table 2), confirming MCFA incorporation into human milk lipids following coconut consumption [51,52].

Milk MCFA may confer many favorable properties to the newborn. They serve as easily absorbable energy sources and exhibit broad-spectrum antiviral and antimicrobial properties. Milk MCFA content increases with advancing lactation [30,53], and the colostrum of mothers delivering preterm presents a higher MCFA content than those delivering at term [54]. MCFA in TGs entering the neonatal gastrointestinal tract are easily released by tongue lipase. Subsequently, the pancreatic- and milk-stimulated biliary lipases can process more effectively the already partially digested TGs [55]. Because of their polarity, the released MCFA are already largely absorbed in the stomach of the neonate, and thereby acting as a rapidly available energy source for the breastfed child. Lauric acid exhibits antimicrobial properties, against, among others, *Helicobacter pylori* [56], and operates synergistically with 1-monomyristyl as a bacteriostatic agent in the form of 1-monolauryl [57]. Finally, it is noteworthy that, in the developing brain, the essential FA alpha-linolenic acid is mostly converted into SFA and cholesterol [58], which are the two main culprits in the "lipid hypothesis" of CVD [14].

Summarizing the past two paragraphs, it is clear that SFA are not essential nutrients by definition. They have, nevertheless, important functions that deserve acknowledgement. Stigmatizing SFA and removing them from our diet may not be the "magic bullet" in the fight against the burden of typically Western diseases of affluence. The indisputably high human milk SFA content is testimony of their beneficial effects, at least in breastfed infants.

4. The "lipid hypothesis" of Keys and its consequences

The "lipid hypothesis" supports the concept that fat, especially SFA, as well as dietary cholesterol, raises blood cholesterol and thereby contributes to CVD risk [13]. However, the preliminary data from 22 different countries did not support the hypothesis that fat intake was unambiguously related with CVD (Fig. 1A) [59]. Data from 15 countries were excluded (Fig. 1B) in the published version of the Seven Countries Study [13]. Nevertheless, the findings gave rise to the first set of national dietary recommendations [59]. These recommendations advised us to reduce total fat to 30 energy% and SFA to 10 energy%. They occurred despite the negative outcomes of RCTs published between 1965 and 1978, *i.e.* prior to the first issue of these recommendations in 1977 and 1983 in the USA and UK, respectively. A

Table 2
FA content (saturated, monounsaturated and polyunsaturated) of milk in several populations and FA composition of various human tissues in g%

FA	Breast milk, Chole, TZ[a]	Breast milk, Maasai, TZ[a]	Breast milk, Jerusalem[b]	Breast milk, NL[b]	Subcutaneous adipose tissue, newborns, Curaçao, The Netherlands Antilles[c]	Subcutaneous adipose tissue, non pregnant women, Curaçao, The Netherlands Antilles[c]	RBC, newborns, Maasai, TZ[d]	RBC, adults, Maasai Wasso, TZ[e]	RBC, adults, NL[e]
SFA	75.30	56.14	54.25	58.78	53.20	32.37	52.40	55.51	45.44
12:0	20.17	7.82	9.67	8.20	0.30	0.58	NA	NA	NA
14:0	21.19	9.22	7.98	7.89	3.51	2.76	0.47	0.70	0.32
16:0	24.90	27.90	18.97	23.21	42.26	23.19	27.40	25.75	20.26
18:0	3.64	6.04	4.93	7.18	5.44	5.19	16.80	18.52	16.20
MUFA	17.85	33.89	33.18	33.04	41.60	48.75	16.60	18.61	18.73
18:1ω7	1.59	1.68	1.74	3.13	2.14	4.05	2.10	NA	NA
18:1ω9	12.79	27.66	28.14	26.49	28.22	39.32	9.35	12.83	11.90
PUFA	6.86	10.84	19.90	15.53	5.20	18.88	34.20	35.74	35.19
LA	4.23	8.79	16.57	12.84	3.56	16.90	3.01	9.98	8.76
AA	0.50	0.37	0.48	0.37	0.80	0.35	16.60	13.97	14.11
ALA	0.28	0.63	0.97	1.02	0.01	0.63	0.09	0.35	0.15
EPA	0.13	0.07	0.04	0.05	0.00	0.01	0.11	0.66	0.51
DHA	0.73	0.20	0.16	0.19	0.32	0.11	4.09	2.59	4.21

Abbreviations: AA, arachidonic acid; ALA, alpha-linolenic acid; DHA, docosahexaenoic acid; EPA, eicosapentaenoic acid; LA, linoleic acid; MUFA, monounsaturated fatty acids; NA, not available; NL, The Netherlands; PUFA; polyunsaturated fatty acids; RBC, red blood cells; SFA, saturated fatty acids; TZ, Tanzania.
[a] Data from [51].
[b] Data from [48].
[c] Data from [313].
[d] Data from [315].
[e] Data from [150].

recent metaanalysis of these trials, investigating mortality reduction by either lower fat intake or SFA replacement by vegetable oils, revealed that serum cholesterol decreased, but not total nor CVD mortality [60].

At the end of last century, Hayes [61] highlighted the possible flaws of the equations presented by Keys and the other Seven Countries coworkers and pointed at the pivotal role of environmental and genetic variables affecting individual responses. For example, Keys *et al.* assumed a standard response to dietary fat, but later studies showed that hormones and circulating lipids, among others, influence the abundance of LDL receptors in the liver [62] and thereby plasma LDL clearance [63]. Moreover, the presence of dietary ω3-PUFA was ignored, and all the (positive) effects caused by PUFA were granted to linoleic acid (LA) [61]. In addition, Keys did not show an association between dietary fat and mortality from all causes [59], and therefore, their data indicating that an increased fat intake increases the risk of death should not be considered as legitimate [64].

Consequently, by virtue of lack of data, the actual American Dietary Guidelines Advisory Committee Report does not provide sufficient evidence to conclude that decreases of dietary SFA, but also of *e.g.* salt and animal protein, would lead to positive health outcomes [65]. Nevertheless, from the 1970s to 2012, CHO intake in the USA increased from 44.0 to 48.7 energy%, protein intake decreased from 16.5 to 15.7 energy% and fat decreased from 36.6 to 33.7 energy% [66], with a concomitant reduction of SFA intake from 13.5 to about 11 energy% in men and that from 13 to 11 energy% in women [67] (Fig. 2). There has been a concomitant increase in the consumption of high-fructose corn syrup together with a decrease in the consumption of refined sugar, to the extent that, nowadays, the consumptions of these two forms of "fast" CHO are similar [68]. In addition, in the USA and The Netherlands, as in most Western countries, the consumption of LA is currently higher than ever along the evolution of the *Homo sapiens sapiens* and probably all of its extinct forefathers up to the common ancestor with the chimpanzee, some 6 million years ago [69–71].

The mantra that (saturated) fat must be removed from the diet to reduce CVD risk has dominated both dietary advices and guidelines for decades [72]. However, parallel to the decrease in fat and SFA intake and the increase in CHO intake (particularly monosaccharides and disaccharides) [73], there has also been an increase in the prevalence of overweight, obesity and DM2 [74–77]. In particular, since the "diet-heart hypothesis" of Keys [13], the prevalence of obesity has increased from 11.9% in 1971 to 33.4% in 2012 (men) and from 16.6 to 36.5% (women) [74,75] (Fig. 3).

5. Current intakes and recommendations: Far away from a Paleolithic diet

Dietary fat is, after CHO, the main energy source in Western countries. For instance, the dietary intakes of fat in the USA [78] and The Netherlands [33] comprise about 33 and 34 energy%, respectively. The current macronutrient composition and their quality (*e.g.* animal/vegetable protein, FA composition, complex/simple CHO) contrast with the dietary composition of our ancestors during the Paleolithic period [68], from which we do not differ much genetically and certainly not with respect to our main metabolic pathways. Northeast Africa is the region currently thought most pertinent to the establishment of the contemporary human genome. There, our earliest behaviorally modern ancestors of some 150,000 years ago might have obtained, on average, about 35 energy% from fats, 35 energy% from CHO and 30 energy% from protein [79]. Nevertheless, fat intake by Paleolithic hunter-gatherers may have varied drastically with latitude, *e.g.* >60% fat in Arctic regions and <25% fat in certain tropical locations [80]. Although dietary patterns varied with, among others, latitude, season, weather and culture, all ancestral diets shared some common key features. Food sources were limited to unprocessed plants and to foraged and hunted land and marine animals that only consumed natural foods from the local environments [68]. Traditional hunter-gatherers, unlike typically Western society members, consume all edible components of the animals they kill, including muscle meat, brain, organs, bone marrow and storage depots [81]. With the advent of both the Agricultural Revolution and animal husbandry, between 5,000 and 10,000 years ago, and more recently, the Industrial Revolution, we have dramatically altered the nutrient balance by consuming the foods typical for Western societies [82]. Game and wild plant foods contain relatively more protein, more roughage and more micronutrients per unit of weight than the foods typically available in the current Western supermarkets [81]. In addition, while its composition varies with season, the fat of wild animals tends to have more monounsaturated fatty acids (MUFA) and PUFA and less SFA than their farm-raised counterparts [79] (Table 1).

Low-fat/high-CHO diets are within the current range of recommendations of the Institute of Medicine [83] to consume 10–

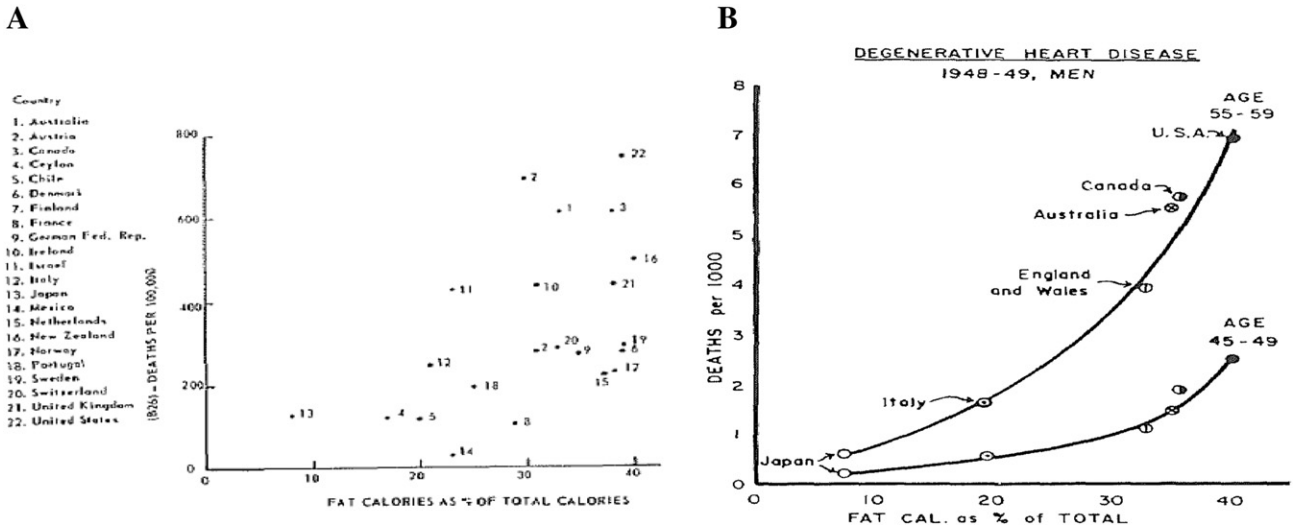

Fig. 1. Data from the "Seven Countries Study" showing mortality from arteriosclerotic and degenerative heart disease as a function of fat calories. Panel A. Mortality from arteriosclerotic and degenerative heart disease as a function of dietary fat calories in 55- to 59-year-old males. Data on mortality are in deaths per 100,000 inhabitants; fat calories represent the percentage of total calories. Data from [59] with permission from The Medical Society of the State of New York. Panel B. Mortality from degenerative heart disease as a function of dietary fat calories. Data on mortality are in deaths per 1,000 inhabitants; fat calories represent the percentage of total calories. Degenerative heart diseases are categories 93 and 94 in the Revision of 1938, Categories 420 and 422 in the Revision of 1948, International List. National vital statistics are from official sources. Fat calories and percentage of total calories were calculated from National Food Balance Data of 1949 supplied by the Nutrition Division, Food and Agriculture Organization of the United Nations. Data from [13] with permission from Journal of Mount Sinai Hospital.

35 energy% protein, 45–65 energy% CHO and 20–35 energy% fat. Such diets are, however, being increasingly criticized. It was already observed in the early 1960s that very low fat diets (where ≤15% of total calories are derived from fat) result in hypertriglyceridemia [84]. This effect was later attributed to increased rates of hepatic DNL [29,85] and the subsequent production of hepatic TG-rich particles, causing higher concentrations of very-low density lipoprotein (VLDL)-cholesterol and lower high-density lipoprotein-cholesterol (HDL-C), without yielding concomitant decreases in LDL-C [86]. Metaanalyses of RCTs did not find any potential health benefit of a low-fat diet in the general population [87,88]. An updated review of RCTs suggested a small (14%) but potentially important reduction in CVD risk with the modification of the *type* of dietary fat, but not with the reduction of total fat [89]. As a matter of fact, short-term consumption of a low-fat diet only shows beneficial effects on plasma lipid concentrations when accompanied by weight loss [90]. A recent systematic review and metaanalysis demonstrated the beneficial effects of a high-fat *versus* a low-fat diet on blood pressure and also TG, HDL-C and fasting glucose levels in both prediabetic subjects and patients with DM2 [91,92].

Nowadays, partially acculturated Greenland Eskimos obtain 4–8 energy% from SFA, but in the USA, SFA, MUFA and PUFA intakes represent about 11 [67], 12 and 7 energy% [93], respectively, and 12.5, 12.7 and 6.3 energy% in The Netherlands [33]. The estimated SFA intake by Paleolithic humans amounted to 5–7 energy%, while *trans* fats contributed in negligible amounts [79]. More recent estimates from various models studied by Kuipers *et al.* [70] show median Paleolithic SFA intakes ranging from 11.4 to 12.0 energy%, while the total range from all scenarios amounted to 6.8–19.0 energy%. The World Health Organization [94] and the 2010 US Dietary Guidelines [20] recommend consuming less than 10 energy% from SFA, and the American Heart Association (AHA)

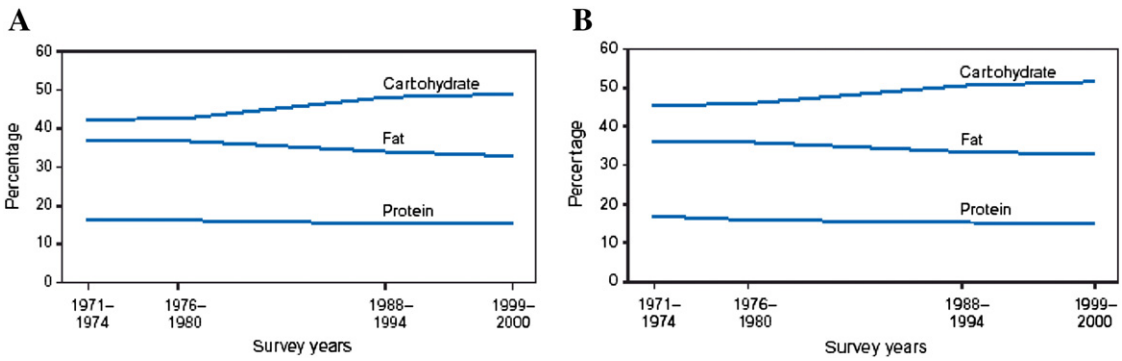

Fig. 2. Energy percentages from macronutrient intake among men (panel A) and women (panel B) aged 20–74 years from 1971 to 2000 in the USA. From 1971 to 2000, mean energy intake in kilocalories increased, while energy% from CHO increased and energy% from total fat decreased. Data from the Health and Nutrition Examination Surveys (NHANES), United States, 1971–2000. For men (panel A), the energy% from CHO increased between 1971–1974 and 1999–2000 from 42.4% to 49.0% ($P<0.01$); for women (panel B), from 45.4% to 51.6% ($P<0.01$). The energy% from total fat decreased from 36.9% to 32.8% ($P<0.01$) for men (panel A) and from 36.1% to 32.8% ($P<0.01$) for women (panel B). The energy% from SFA decreased from 13.5% to 10.9% ($P<0.01$) for men and from 13.0% to 11.0% ($P<0.01$) for women (data not shown). A slight decrease was observed in the energy% from protein, from 16.5% to 15.5% ($P<0.01$) for men (panel A) and from 16.9% to 15.1% ($P<0.01$) for women (panel B). Sample sizes ranged from 1,730 men and 2,003 women in NHANES 1999–2000 to 6,630 men and 7,537 women in NHANES III. Adapted from the Centers of Disease and Control (CDC) [67] with permission from the CDC.

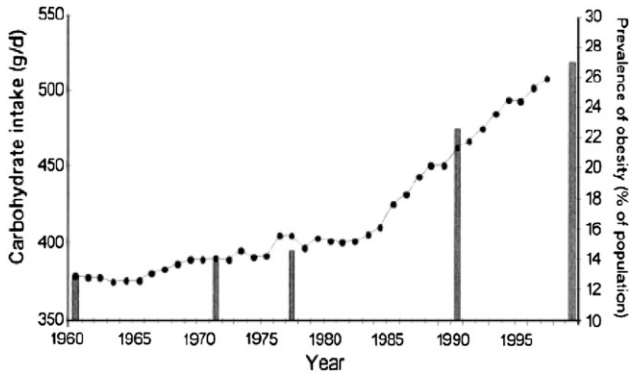

Fig. 3. CHO intake and prevalence of obesity in the USA from 1960 to 1997. Mean CHO intake (dotted line) is in grams per day (g/day) and obesity (body mass index>30 kg/m^2, vertical bars) is in percent (%) of total population. The intake of corn syrup sweeteners (as a representative for refined CHO) in 1997, which were almost nonexistent at the beginning of the century, comprised 20% of the total daily CHO intake and 10% of the daily total energy intake, representing an increase of 2100% in this period. The authors found a strong association between an increased consumption of refined CHO in the form of corn syrup, a decreased consumption of dietary fiber and an increasing trend in the prevalence of type 2 diabetes in the United States during the 20th century. Data from [206] with permission from The American Society for Nutrition.

advices on less than 7 energy% [95], while aiming at 5–6 energy% for patients with high LDL-C [96]. The AHA also recommends ω6-PUFA intakes (*i.e.* LA) of 5–10 energy% [97], while the median LA intakes from our ancient diet varied from 2.3 to 3.6 energy% [70].

In The Netherlands, the average fish consumption hardly amounts to three times per month, while the recommendation is twice per week, including one portion from fatty fish. The median intakes of the sum of eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) by adults amounts to 75–107 and 77–110 mg per day by women and men, respectively [33], while it is recommended to consume 450 mg/day [73] and the median intake from an East African Paleolithic diet ranged from 2.26 to 17.0 g/day, dependent on scenario [70]. Thus, in contrast to the present intakes, dietary SAFA intake was accompanied in the past by high intakes of EPA and DHA, emphasizing the importance of a balance between SAFA and PUFA and between omega-3 and omega-6 FA, as opposed to the evaluation and recommendation of single nutrients. In addition, the median intake of vegetables and fruits by Dutch adults barely reaches 200 g per day (recommendation 400 g/day; *i.e.* 200 g from fruits and 200 g from vegetables) [33]. Not surprisingly, the major contributors to diet-associated mortality in both the USA and The Netherlands were, by far, insufficient intakes of fish, vegetables and fruits, with less important roles for the intakes of saturated and *trans* FA [98].

6. (Saturated) fat and CVD risk

Diminishing SFA consumption is recommended to reduce CVD, by virtue of its influence on lipoprotein-cholesterol levels. This chapter explores this relation and discusses the evidence linking SFA with CVD risk. It also explores the SFA–CVD risk relation in the context of nutrient replacement and embarks into the relation between CVD and the consumption of dairy and meat, the latter being the principal sources of SFA in the typical Western diet.

6.1. Lipoproteins, FA, cholesterol and CVD risk

It is almost universally accepted that high serum TC and especially LDL-C are risk factors for CVD, while a high level of HDL-C is, on the other hand, protective. Serum TC/HDL-C ratio is the consensus CVD risk factor employed for CVD risk assessment of the individual subject. In men, one unit increase in the TC/HDL-C ratio is considered to confer a 53% higher CVD risk [99]. A 1% reduction in TC and LDL-C corresponds with a 2% and 1.7% CVD risk reduction, respectively [100], while an increase in HDL-C of 0.025 mmol/L (1 mg/dl) corresponds with a 2–3% decrease in CVD risk [101]. The emphasis related to SFA is usually on the seemingly unfavorable relation with LDL-C (see below). However, the causal relationship between LDL-C and CVD [102] is still subject of debate [103,104]. There is no doubt that statins reduce TC, LDL-C and CVD risk in both primary and secondary prevention studies [105]. However, the reduction of CVD risk is accompanied by a decrease of both LDL-C and C-reactive protein [105,106]. The pleiotropic effect of statins [107] and, in particular, their antiinflammatory properties [108], may represent a solid example of the intimate relationship between (low-grade) inflammation and metabolism [3,109].

The metabolic syndrome (later coined the "insulin resistance syndrome" [110]) is characterized by excessive body weight, impaired glucose homeostasis, hypertension and atherogenic dyslipidemia (the "deadly quartet"). This combination is a major risk factor for CVD and other "typically Western" diseases [111]. The coinciding atherogenic dyslipidemia is composed of elevated TGs, small dense LDL particles and reduced HDL-C (the "deadly lipid triad") [112]. Small dense LDL particles are susceptible to oxidation and thereby to structural modification [113]. They easily pass into the subendothelial space and may thereby promote atherosclerotic plaque formation [114] through foam cell generation, local inflammation and endothelial dysfunction [115]. Both oxidized and small dense LDL have been related to increased CVD risk [113,116] and they occur in conjunction with the high TG and low HDL-C features of the metabolic syndrome. Under these conditions, also high-density lipoprotein (HDL) particles change into what has unfortunately been coined "dysfunctional HDL" because of its proinflammatory character, in contrast to "normal HDL" ([115]; see below). Hence, it may be of more diagnostic value to employ plasma TG/HDL-C concentration ratio to define an atherogenic profile and thereby identify individuals with low insulin sensitivity and subsequent increased CVD risk [110,117].

6.1.1. Trans FA

It is universally accepted that *trans* FA increase CVD risk, most probably through their proinflammatory properties [118,119]. *Trans* FA comprise industrially produced, artificial, *trans* FA (found, among others, in many fast foods, bakery products and margarines) and naturally occurring *trans* FA (ruminant, *e.g.* present in dairy and meat) [120]. For instance, conjugated linoleic acid (CLA) is a *trans* FA naturally present in small amounts in ruminant fat [120]. Both observational studies on the relation of *trans* FA with risk [121,122] and metabolic studies on the relation of *trans* FA with lipoproteins [119,123,124] indicated that industrial *trans* FA have detrimental effects on cardiovascular health, increase LDL-C and lower HDL-C blood levels, the latter being distinct from the effect of SFA.

A metaanalysis of observational studies showed that the replacement of 2 energy% CHO, SFA and *cis*-MUFA or *cis*-PUFA by 2 energy% industrially produced *trans* FA corresponds with 20–30% higher risk of myocardial infarction and CVD mortality [125]. Not all *trans* FA seem equal: a recent metaanalysis of prospective studies suggested that the intake of industrial *trans* FA may be positively related to CVD, while the intake from ruminant *trans* FA was not [126]. A more recent review showed that, on a weight basis, CLA, ruminant and industrial *trans* FA, exhibited the same effect on blood lipoproteins [120]. However, the potential impact of the naturally occurring (ruminant) *trans* FA is not convincing because of the large scatter (*trans* FA intake ranged from 2.8 to 10.0 g/day) and a lack of sufficient observations at high intakes [120]. Moreover, the share of natural *trans* FA in our diet is usually <0.5% [69]. Therefore, the aforementioned risks might not be applicable to the naturally occurring *trans* FA [127].

6.1.2. MUFA and PUFA

The substitution of 1 energy% CHO by 1 energy% PUFA and, to a lesser extent, MUFA is accompanied by a reduction in LDL-C (mean: −0.019 and −0.009 mmol/L, respectively) and TC/HDL-C ratio (−0.032 and −0.026 mol/mol) and an increase in HDL-C (0.006 and 0.008 mmol/L) [128]. Thus, based on the TC/HDL-C ratio, MUFA and PUFA appear as the healthier options, compared with CHO. On the other hand, the relation between serum lipoprotein-cholesterol concentrations and CVD risk is more complicated than until recently assumed. This has become clearly illustrated by the failures of cholesteryl ester transfer protein inhibitors to reduce CVD risk despite their ability to greatly increase HDL-C [129,130], and the aforementioned pleiotropic effects of statins, which include a reduction of low-grade inflammation next to their cholesterol-lowering action [105,131]. The intimate relationship between inflammation and metabolism [132] (further reviewed), including cholesterol metabolism [115], indicates that the changes in "cholesterol" concentrations in Western societies are influenced by many lifestyle factors that may collectively cause low-grade inflammation and thereby CVD [2,3]. The conflicting evidence on the association between SFA and blood lipids suggests that SFA may not be the main culprit in raising blood lipid levels and, thereby, may not be a major risk factor in the development of CVD [133].

The initial GISSI-Prevenzione [134] and JELIS [135] studies supported the use of fish oil supplements in the secondary prevention of coronary heart disease, but a recent metaanalysis [136] and systematic review [137] yielded little or no effects, probably because fish oil may not contribute to a preventive effect on top of the highly effective current treatment with drugs [138]. EPA and DHA cause no change in TC and slightly increase both HDL-C and LDL-C [139,140]. They hold important antiarrhythmic, antithrombotic, antiatherosclerotic and antiinflammatory effects, while they also reduce serum TG and blood pressure and improve endothelial function [141]. Animal studies demonstrated that the combination of ω3-PUFA with SFA increases ω3-PUFA concentration in plasma and liver lipids [142] and that dietary SFA raise blood lipids (cholesterol and TG) only when the diet is deficient in ω3-PUFA [143]. Studies in humans showed that combining SFA and ω3-PUFA caused a synergistic beneficial effect of both types of FA, increasing red blood cell membrane fluidity; decreasing TC, LDL-C and TG; and increasing HDL-C [144,145]. On the other hand, a high ω6-PUFA diet was shown to increase hepatic cholesterol in mice when compared with a high-SFA diet [142]. The amount of ω3-PUFA required to cause any beneficial effect was maximized for mice consuming background diets with a low ω6-PUFA/SFA ratio [143]. EPA and DHA, synthesized from linseed oil or directly obtained from fish oil, are more efficiently incorporated into rat microsomal phospholipids at a SFA (hydrogenated beef tallow)-rich diet than at an LA (safflower oil)-rich diet, causing a reduction of AA and a higher EPA/AA ratio. A higher EPA/AA ratio relates to less inflammation, more vasodilatation, reduced coagulation and reduced growth. The LA-rich diet also increased the esterified cholesterol content and the unesterified cholesterol/phospholipid ratio in hepatic microsomes [142]. The same group also showed that, following ALA supplementation of rats, the extent of ALA, EPA and 22:5ω3 incorporation into liver and plasma lipids decreased with the background diet in the order SFA+ALA>MUFA+ALA>ω6-PUFA+ALA [143]. In addition, investigations with humans revealed that moderate intakes of 14:0 (from butter) and ALA (from rapeseed oil) were synergystically associated with more favorable plasma lipids, higher ω3-PUFA in plasma lipids and higher erythrocyte membrane fluidity [144,145]. Jointly, these studies demonstrate the superiority of a combination of dietary SFA and ω3-PUFA to cause higher ω3-status and thereby its associated favorable health effects, when compared with a combination of dietary LA and ω3-PUFA [143]. This effect appears in line with the so-called "Israeli paradox": the inhabitants of Israel present dietary habits low in calories, fat and SFA and high in ω6-PUFA, while their rates of Western illnesses are about the same as in the USA [146]. Altogether, these data confirm the recommendation of a diet high in ω3-PUFA with a concomitant reduction in the consumption of ω6-PUFA [133], which is basically what we have eaten in the past [147].

6.1.3. SFA

The harmful effect of dietary fat, notably SFA, and its relation with CVD, is increasingly questioned [69,72,148–151]. Recently, a revised metaanalysis of RCTs showed that the substitution of SFA (and possibly *trans* FA) by ω6-PUFA without simultaneously increasing ω3-PUFA presents no indication of benefit and it is even likely to increase CVD risk [152] (Fig. 4).

To venture into this discussion, it is important to review the historical relation between SFA intake and TC (Fig. 5). This relation is subject to a strong interindividual variation: SFA consumptions of 15% and 4% may correspond with TC values of 3 and 6 mmol/L, respectively, illustrating that SFA intake only explains a small part of TC variation. Other factors influencing the effects of SFA intake on LDL-C are the amount of dietary cholesterol, the apolipoprotein E4 allele, obesity, insulin resistance, hypertriglyceridemia and female gender [153]. Moreover, the average TC increase due to SFA intake is low: the replacement of 1 energy% CHO by 1 energy% SFA corresponds with an LDL-C increase of 0.033–0.045 mmol/L, but also with a frequently ignored concomitant increase of HDL-C with 0.011–0.013 mmol/L [83]. These changes take place in the context of an also frequently ignored lower TG (mean: −0.021 mmol/L) and without any significant effect on the TC/HDL-C ratio and ApoB [128,154]. For instance, in The Netherlands, a reduction of SFA intake from the current 13 energy% to the target intake of 10 energy% would result in an irrelevant CVD risk reduction at the individual level [69], when estimated from the SCORE algorithm for CVD risk assessment. Apart from the TC/HDL-C ratio, this algorithm also takes other risk factors into account, notably gender, age, smoking and systolic blood pressure [155].

It has been established that adipose tissue SFA content does not reliably reflect SFA intake and that adipose tissue SFA is at the same time inversely related to CVD [156,157]. Others have found that SFA status does not correlate well with dietary SFA [158]. We [150] recently showed, in an observational study of healthy African and Dutch subjects, that 14:0 *status*, 16:0 *status* and SFA *status* correlate positively with the TC/HDL-C ratio. SFA *intakes* in RCTs are, on the other hand, unrelated to this frequently used ratio for the calculation of individual CVD risk. The discrepancy between SFA *intake* and SFA *status* might derive from the often-overlooked endogenous SFA synthesis (DNL; notably 16:0) from CHO and other precursors, which, in contrast to the widespread believe, may contribute to a sizeable extent to SFA status [29,159]. Estimations of CVD risk with the

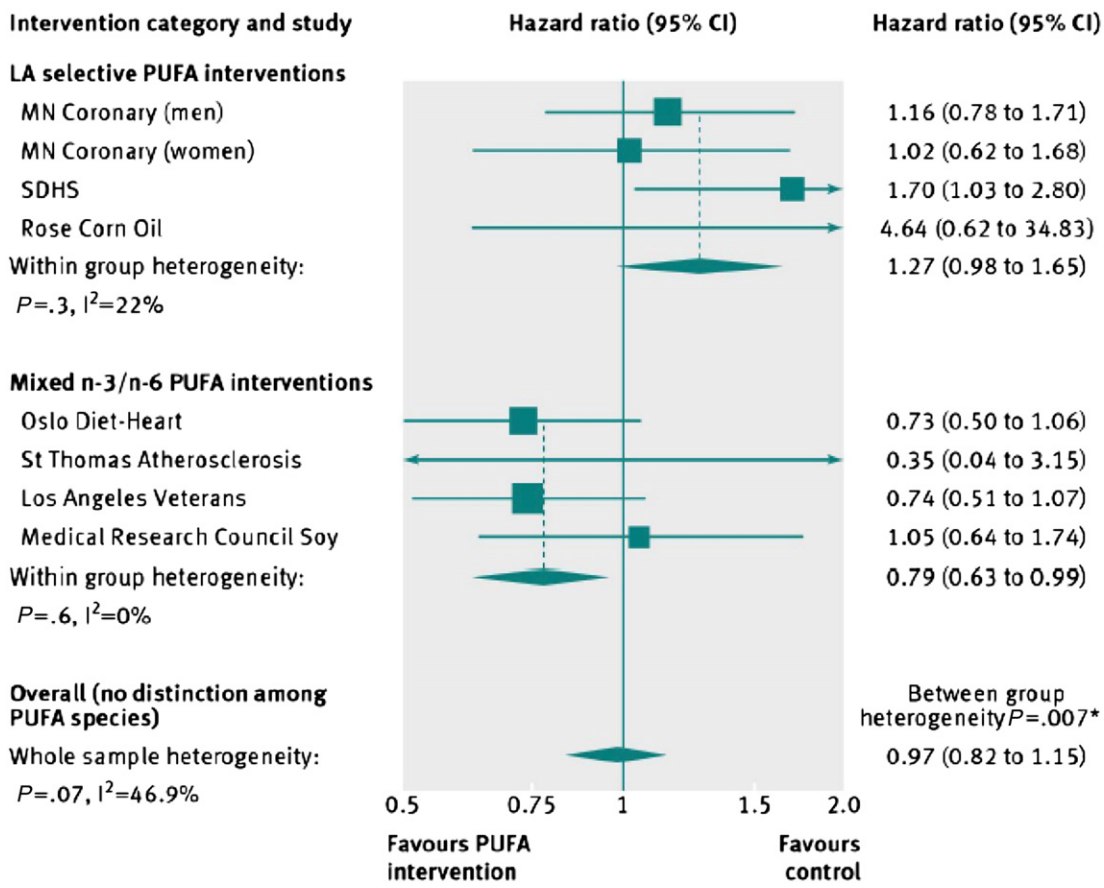

Fig. 4. Metaanalysis of effects of LA-selective interventions and mixed ω3/ω6 PUFA interventions on risk of cardiovascular death. LA selective interventions selectively increased intake of ω6 LA without a concurrent increase in ω3 PUFA. Mixed PUFA interventions increased intake of ω3 PUFA and ω6 LA. PUFA interventions replaced high-SFA control diets in each trial. *Significant heterogeneity between groups. Adapted from Ramsden *et al.* [214] with permission from Cambridge University Press.

TC/HDL-C ratio as proxy suggest that the Dutch, who are representative for a population with typical Western lifestyle, would have lower CVD risk than the African populations [150]. This is highly counterintuitive, considering that more than 75% of the current adult Dutch population does not adhere to the recommendations for adults of 200 g fruits and 200 g vegetables per day, nor to the consumption of *at least* two servings of fish per week (one time fatty fish), while also their intake of CHO with high glycemic index (GI) is high [33]. These observations raise the question of whether the TC/HDL-C ratio is a suitable marker for CVD risk estimation, and underline the usage of the aforementioned TG/HDL-C ratio as a more sensitive marker for the insulin resistance (metabolic) syndrome [110,117].

6.2. Observational and prospective studies on saturated fat: How big is the risk?

Recent observations in a prospective cohort study of older adults, the Cardiovascular Health Study, demonstrated divergent associations of circulating 16:0 *versus* 18:0 and 20:0 with atrial fibrillation. Higher levels of circulating 16:0 were associated with higher risk, while higher 18:0 and 20:0 were associated with lower risk, following adjustment for other atrial fibrillation risk factors [160]. Accordingly, 18:0 does not show any deleterious effect on CVD risk [161] and the evidence for its putative effect on thrombotic susceptibility appears inconsistent [162,163]. An update of the Nurses' Health Study showed that SFA intake alone was not a predictor of CVD and that only a higher PUFA intake was associated with decreased CVD risk, whereas a higher *trans* FA intake was associated with increased CVD risk, independent of other dietary and CVD risk factors [164]. Furthermore, a recent study in 2412 patients with established CVD was unable to find an association between dietary SFA intake and the incidences of both coronary events and mortality [165].

One of the strongest arguments against the presumed unfavorable effects of dietary SFA are the recent metaanalyses showing that, despite the undeniable (weak) relationship with cholesterol, dietary SFA are not associated with hard endpoints [153,166,167], even after adjustment for serum TC [168]. Mente *et al.* [166] studied the outcome of prospective studies and RCTs, which analyzed the relationship between various nutritional factors and CVD. There were strong and moderate degrees of evidence found for the beneficial effects of, among others, fruit, nuts, Mediterranean diet and MUFA, fish and fish oil FA, while strong negative effects were found for *trans* FA, foods with a high GI and GL and Western food. At the same time, there was insufficient evidence for SFA, PUFA, total fat, alpha-linolenic acid, meat, eggs and milk. Siri Tarino *et al.* [153] performed a metaanalysis of 21 prospective studies following 347,747 people during 5–23 years. Within this time frame, there was no relationship found between SFA intake and the development of CVD. More recently, The Health ABC Study, involving subjects with ages between 70 and 79 years, did not find any significant association between SFA consumption and CVD risk [169]. Finally, a recent study in rats has shown that virgin (highly saturated) coconut oil reduces CVD risk by exerting beneficial effects on lipid parameters through reducing DNL and enhancing the rate of FA catabolism [170].

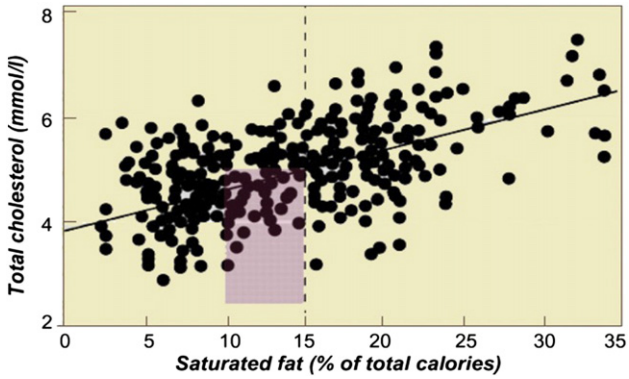

Fig. 5. Relationship between energy% intake of dietary saturated fat and serum TC. The slope amounts to 0.067 mmol/L serum cholesterol per energy% SFA (2.6 mg/dl/%). The shaded portion shows the expected benefit if saturated fat intake is lowered from 15 energy% to the currently recommended 10 energy%. This dietary change corresponds with an average serum cholesterol reduction of 0.34 mmol/L (13 mg/dl). The sizeable scatter indicates that SFA intake is an only modest determinant of TC in the population. Adapted from Volek *et al.* [172] with permission from Elsevier.

Forsythe *et al.* [171] and Volek *et al.* [172] clearly illustrated that SFA *intake* does not necessarily predict SFA *status* and CVD risk (further reviewed). Their valuable data suggest that it is not only the dietary SFA content but especially the entire dietary composition, and within this context the CHO energy%, that determines whether SFA intake is associated with detrimental outcomes or not [150]. Populations with low CVD risk show a wide range of fat intake, varying from 15% in China to 40% in some Mediterranean populations [173], but all of them present minimal intakes of *trans* FA, high intakes of ω3-PUFA and low intakes of ω6-PUFA. Most of these low-CVD-risk populations consume low amounts of SFA, but there are a few notable exceptions with high-SFA intakes. These include the Pacific Islander populations of Tokelau [174] and Kitava [175] and also the Maasai [176] and inhabitants of Chole [150] in Africa. In this context, it is noteworthy that the CVD mortality risk associated with eating too little fruit, vegetables and fish has been estimated to be 10 times greater than that due to SFA [98].

6.3. The controversy with milk and meat as saturated fat sources

Despite the sizeable contribution of both dairy products and meat to the SFA content of our diet, there is no consistent evidence that their consumption is associated with higher CVD risk [177–179]. A recent metaanalysis comparing the effects on cardiometabolic risk markers of increasing daily intake of low-fat dairy *versus* whole-fat dairy (mean: 3.6 servings/day for 26 weeks) showed neither improvement nor difference between both groups [180]. In agreement with these findings, a dose–response metaanalysis of prospective studies [181] indicated that milk intake is not associated with total mortality and may even be inversely associated with overall CVD risk. This finding was confirmed in the recent Multi-Ethnic Study of Atherosclerosis (MESA) [179]. More recently, in two large prospective US cohorts (the Health Professionals Follow-Up Study and the Nurses' Health Study), circulating biomarkers of dairy fat were not associated with stroke [182], while another metaanalysis provided further evidence supporting a beneficial effect of dairy consumption on CVD [183].

Results from short-term intervention studies indicate that a diet higher in SFA from whole milk and butter increases both LDL-C and HDL-C when substituted for CHO or unsaturated FA and might therefore not affect or even lower the TC/HDL-C ratio [184]. A study with healthy 15-year-old Swedish boys and girls found inverse associations between the dietary SFA content, mainly derived from milk, and serum concentrations of cholesterol and ApoB. These data suggest that milk fat contains, or is associated with, some component, or some other characteristics of food intake or lifestyle, that counteracts the expected negative effect of SFA intake on serum lipids [185]. Suggestions include CLA [186] and *trans* palmitoleic acid [187]. The latter has recently been associated with higher HDL-C levels and lower TC/HDL-C ratio, adiposity, TGs, C-reactive protein (CRP), fasting insulin, blood pressure and onset of DM2 [187]. However, despite all the currently available evidence, whole fat dairy is still not recommended in most food guidelines [98,188,189] because of the concern that SFA in dairy food may have an adverse effect on serum lipids, increasing CVD risk.

Evidence regarding meat shows conflicting results. While the recent MESA study revealed an association between high meat intake and increased CVD risk [179], a metaanalysis of 2010 displayed no association between red meat consumption and CVD and blamed its processing for substantially increasing CVD and DM2 risks [178]. The Global Burden of Disease Study [190] identified high blood pressure, tobacco smoke and alcohol use as the three leading risk factors for disease. A diet high in red meat ranked lowest in a list of 43 risk factors contributing to the global burden of disease. Therefore, recommendations to reduce the consumption of unprocessed red meats may be unnecessarily restrictive [191]. The traditionally living Maasai, with intakes of both meat and milk that highly surpass the median Western (saturated) fat intake, present the highest 16:0 content ever measured by our group, both in red blood cells [150] and breast milk [51]. They have an average TC of 4.9 mmol/L [150] and almost no evidence of CVD [176,192].

6.4. Effects of replacing saturated fat by other nutrients

Several questions regarding the relationship of SFA intake with CVD risk remain unanswered, especially whether the health effects of reducing SFA consumption are different depending on the replacement nutrient [67].

6.4.1. Saturated fat replacement by CHO

Replacing SFA with CHO with high GI values is associated with higher risk of myocardial infarction, while its replacement with CHO with low GI is associated with lower risk [193]. A recent systematic review and metaanalysis demonstrated that higher *ad libitum* CHO intakes are associated with increased TG levels [194], while exchanging either CHO or protein for fat improves the lipid-related CVD risk profile in overweight men and women [195]. Hence, the reduction of SFA intake without any consideration on the replacement nutrient may have substantial unfavorable effects on disease risk, especially

because the most common replacement nutrient in populations has been CHO [154]. Prioritizing SFA replacement with CHO, notably refined CHO and added sugars, has been associated with atherogenic dyslipidemia and increased CVD risk [153]. In fact, foods containing high amounts of CHO have been implicated in the etiology of obesity and DM2 [196,197] and are associated with nonalcoholic fatty liver disease (NAFLD) and nonalcoholic steatohepatitis (NASH) [29,198] (further reviewed). SFA [199] and CHO [200] intakes are strongly associated with NAFLD in obese children, as well as a low consumption of dietary fiber and ω3-PUFA [199], which underlines the importance of the multiple interacting factors playing a role in the ultimate (metabolic and inflammatory) status.

6.4.2. Low-fat/high-CHO diets

Over the last years, an increasing number of study outcomes have challenged the low-fat dietary approach [92]. A diet lower in CHO and concomitantly higher in fat and protein seems a much better option for weight loss and for the secondary prevention of chronic, typically Western, diseases [201,202]. Liu *et al.* [203] suggested that the high-CHO/low-fat diet currently recommended in the USA may increase the risks of insulin resistance and glucose intolerance and that a high dietary GL from refined CHO increases CVD risk, independent of other CVD risk factors. As an example, among subjects at high CVD risk, a Mediterranean diet (around 40 energy% fat, 40 energy% CHO and 16 energy% protein) fortified with either extravirgin olive oil or nuts achieved a 30% improvement over a low-fat diet in terms of cardiovascular events [204]. These data are further strengthened by an RCT comparing a low-fat diet (with <10 energy% SFA) *versus* a low-CHO diet (12 energy% from CHO) [171,172], showing that the low-CHO diet caused greater improvements on numerous endpoints such as body fatness, lipids, glucose tolerance, inflammation and thrombogenic markers (more details in paragraph "SFA and inflammation"). Severely obese subjects (mean body mass index: 43 kg/m^2) with a high prevalence of DM2 or the metabolic syndrome lost about three times more weight during 6 months on an *ad libitum* CHO-restricted diet (<30 g CHO/day, no calorie restriction) than on a calorie-restricted (calorie deficit, 500 kcal/day) and fat-restricted (fat intake<30 energy%) diet. Subjects following the CHO-restricted diet exhibited a relative improvement in insulin sensitivity and TG levels, even after adjustment for weight loss [205]. Increasing refined CHO intake, concomitant with decreasing intakes of fiber, paralleled the upward trend in the prevalence of both obesity and DM2 observed during the 20th [206] and 21st [75] centuries (Fig. 2). Meanwhile, there is no doubt that added sugars and sugar-containing soft drinks are important determinants of blood pressure, serum lipids and body weight within the free-living population consuming an *ad libitum* diet [207,208]. From different mechanistic points of view, high-CHO diets have been reported to reduce body fat oxidation [209], increase blood TGs [210,211] and reduce satiety [212] compared to low-fat diets, prompting the question of their suitability for patients suffering from the metabolic syndrome, CVD and DM2 [92].

6.4.3. Saturated fat replacement by MUFA and PUFA

Evidence suggests that SFA replacement by MUFA lowers CVD risk [154], although no RCT has directly tested this relationship [213]. On the other hand, metaanalyses of RCTs have shown that partial replacement of dietary SFA for LA, the dominating dietary PUFA, is unlikely to provide the intended benefits and may actually increase the risks of both CVD and death (Fig. 4) [152,214]. An excessive ω6-PUFA intake from refined vegetable oils has been identified as a contributor to cancer and CVD [215]. In line with these data, a recent systematic review, metaanalysis and metaregression of RCTs showed no significant benefit of SFA replacement by PUFA in the secondary prevention of CVD [216]. Another metaanalysis of 11 cohort studies and 8 RCTs where dietary SFA was replaced by PUFA [217], concluded that the benefits on CVD risk (in terms of TC/HDL-C reduction) could not distinguish between the potential benefits of increasing PUFA and reducing SFA intake. It was also noted that "the relatively modest magnitude of plausible benefit (about 10% lower CVD risk for 5 energy% replacement) indicates a need for a substantial policy focus on other dietary risk factors for CVD, in particular, high consumption of salt and low consumption of seafood, whole grains, fruits and vegetables [217]". These outcomes contrast with the current recommendations of the AHA to limit SFA intake to <7 energy% [188] and augment LA intake to *at least* 5–10 energy% [97]. As argued by Ramsden *et al.* [152], this advice may not provide "the most convincing" and "decisive" evidence base, with "immediate implications" for "population and individual level recommendations" to substitute ω6-PUFA-rich vegetable oils for SFA. Finally, the arterial plaque is primarily composed of unsaturated fats, particularly PUFA (and not SFA) [215], while SFA have been suggested as the preferred fuel for the heart [218].

Taken together, recent systematic reviews and metaanalyses do not support the current guidelines encouraging a high PUFA (*i.e.* LA) and low-SFA consumption [216,219], and hence, dietary guidelines should strongly reconsider their recommendations for replacing SFA with ω6-PUFA [64].

7. The connection between saturated fat and inflammation

Since the discovery that obesity is associated with accumulation of macrophages in adipose tissue [220], the mechanisms by which the latter becomes inflamed, resulting in insulin resistance, have become an important research question. Several studies demonstrated that SFA might cause adipose tissue inflammation by processes involving, among others, toll-like receptor (TLR) TLR-4, a sensor that binds bacterial lipopolysaccharide (LPS) [221] and thereby plays a key role in the innate immune response [132]. This chapter embarks into the relation of SFA with inflammation.

7.1. DNL and insulin resistance

It has been known for at least 60 years that either fasting or dietary CHO removal results in the virtual absence of DNL [222]. The normal response to fasting, *i.e.* the lowering of glucose and insulin, accelerated breakdown of stored body fat and increased release of free FA (adipose tissue) and ketones (liver), is mimicked by a very low CHO diet [223,224], also named ketogenic diet. Even small decreases in insulin are associated with large increases in fat breakdown and fat oxidation [225]. Conversely, due to limited storage capacity, a high-CHO intake promotes the conversion of CHO to SFA (DNL), a process that becomes stimulated by preexisting insulin resistance [29] and by the rapidity by which both glucose (high GL) and fructose enter the body [226] (Fig. 6). The flux towards acetyl-CoA in the liver determines whether its origin from fat, CHO, protein or alcohol will be used only for energy generation or concomitant conversion to fat, explaining the occurrence of both alcoholic fatty liver disease and NAFLD when overloaded.

Adipose tissue acts as a buffer capable of accumulating and depositing excess lipids. Impairment of this buffering mechanism, such as in insulin resistance, results into a radical remodeling of lipid flow that promotes accumulation of lipids in the liver, skeletal muscles and pancreatic beta cells [227]. Insulin resistance and its associated metabolic adjustments are important determinants of CVD [111]. Diets rich in fat, mainly SFA and *trans* FA, as well as CHO-rich diets, favor insulin resistance, independent of adiposity [228,229], while dietary fiber intake is inversely associated with the risk of developing insulin resistance in adults [228].

NAFLD and its sequel, NASH, are the hepatic manifestations of the metabolic syndrome (also named the insulin resistance syndrome [110]). These conditions are among the most profound stimulators of DNL from CHO [159], while insulin resistance also stimulates fat mobilization from the adipose tissue in the form of free FA. The prevalence of NAFLD is estimated at 25–30% in the adult Western

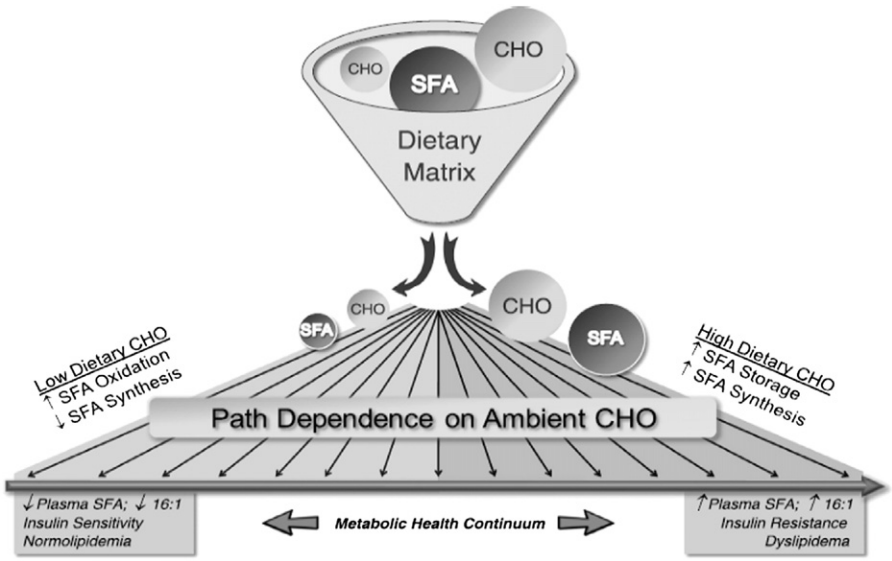

Fig. 6. Importance of dietary CHO in the synthesis and oxidation of saturated fat. Right side of the figure: high-CHO diets promote the initial utilization of CHO for energy generation, causing sparing of the concomitantly consumed dietary SFA and promoting DNL (*i.e.* synthesis of SFA and MUFA). This occurs notably with hypercaloric CHO-rich diets and CHO with high GI and in subjects with the metabolic syndrome (eventually causing NAFLD [29]). Concomitant DNL and sparing of dietary SFA contribute to increasing SFA status, which under such conditions becomes increasingly disconnected from dietary SFA intake and may become deleterious when uncompensated by other factors. Left side of the figure: low dietary CHO causes oxidation of dietary SFA, low DNL and low-SFA status, contributing to less SFA-induced inflammation. Adopted from [316] with permission from Wolters Kluwer Health.

population [230]. In patients with NAFLD, more than 25% of the FA in the liver and VLDL are *de novo* synthesized compared to less than 15% deriving from the diet [159]. In the Korean general population, the energy% from CHO is positively related to serum aminotransferase activity and metabolic syndrome prevalence, while the dietary fat percentage is inversely related [231,232]. Other prominent sources of DNL are fructose and alcohol, which bear great resemblance with CHO in their tendency to be converted into fat in the liver [226]. However, also in this context, it is the dose that makes the poison. Therefore, the total caloric intake from (other) sugars and other macronutrients [233–235], the rapidity of entry and the metabolic context of reduced insulin sensitivity should also be taken into consideration. Not surprisingly, subjects with NAFLD have been estimated to consume five times more CHO from soft drinks than their healthy counterparts [236].

The contribution of CHO to DNL (notably 16:0) and to a sizeable extent to SFA status [237] was illustrated by Forsythe *et al.* [171], Volek *et al.* [172] and, more recently, Volk *et al.* [238]. Forsythe and Volek [171,172] studied subjects with the metabolic syndrome and compared the effects of a high-fat/high-SFA (58.9 energy% fat, 36.4 g SFA/day)/very-low-CHO (12.4 energy%)/hypocaloric diet (1500 kcal) *versus* an equally hypocaloric diet with low fat (23.8 energy%), low SFA (11.7 g/day) and high CHO (55.8 energy%). They found that the high-fat/high–SFA (36.4 *versus* 11.7 g/day)/very-low-CHO-hypocaloric diet did not only cause a more pronounced decrease in SFA status (in cholesterol esters and TG) but also improvements in markers of the metabolic syndrome (greater weight loss, decreased adiposity, improved glycemic control and insulin sensitivity and more favorable TG, HDL-C and TC/HDL-C responses), markers of "atherogenic dyslipidemia" and CVD risk (*e.g.* postprandial lipidemia, apolipoproteins, LDL particle distribution and postabsorptive and postprandial vascular function), inflammatory markers (cytokines, chemokines and adhesion molecules) and markers of oxidative stress (urinary isoprostanes). In the recent study of Volk *et al.* [238], subjects with the metabolic syndrome were fed six different 3-week diets (300 kcal/day energy deficit) that progressively increased CHO intake (from 47 to 346 g/day) with concomitant decreases in total fat and SFA (from 84 to 32 g/day). At baseline, subjects consumed diet with 333 g CHO/day with 46 g/day SFA. CHO intake was adjusted every three weeks and total fat decreased proportionately to keep total energy constant. SFA intake was 40 g% of total fat for all phases. Despite a marked increase in dietary SFA intake from baseline to the first diet (46 to 84 g/day) and then a progressive decrease to 32 g/day in the last diet, the proportions of total SFA in plasma TG, CE and phospholipids of 16:0 in plasma TG and CE were not affected and were not associated with dietary CHO or SFA. Conversely, plasma palmitoleic acid, a biomarker associated with increased risk of, among others, insulin resistance, metabolic syndrome and DM2 [239], increased in all-three lipid fractions as CHO intake increased. These data suggest that it is not only the dietary SFA content but the entire dietary composition, and especially the CHO content, that determines whether SFA intake is associated with detrimental outcomes or not. Current data indicate that when the diet is low in SFA and high in CHO, dietary SFA are spared and additionally *de novo* synthesized from the abundant dietary CHO, but when the diet is low in CHO, dietary SFA are used for energy generation.

Taken together, it has become clear that high-CHO diets promote the initial utilization of CHO for energy generation, causing sparing of the concomitantly consumed dietary SFA and promoting DNL (*i.e.* synthesis of SFA, notably 16:0, and MUFA, notably 18:1ω9). This situation especially occurs with hypercaloric CHO-rich diets, high-CHO intake (notably CHO with high GI) and high-fructose and alcohol consumption and in subjects with the metabolic syndrome [29]. Accumulating fat in the liver eventually causes NAFLD. Concomitant DNL and sparing of dietary SFA increase SFA status, which under such conditions becomes disconnected from dietary SFA intake. The outcome may be expected to become deleterious when uncompensated by antiinflammatory factors and/or during circumstances of massive fat (SFA) mobilization.

7.2. SFA and inflammation

High-fat diets, notably those rich in SFA [240,241], have been shown to promote LPS (from Gram-negative bacteria) uptake in the gut [242] (Fig. 7). The subsequent postprandial (low-grade) endotoxemia may cause low-grade systemic inflammation [243], insulin resistance and obesity [244]. TLRs play a key role in recognizing

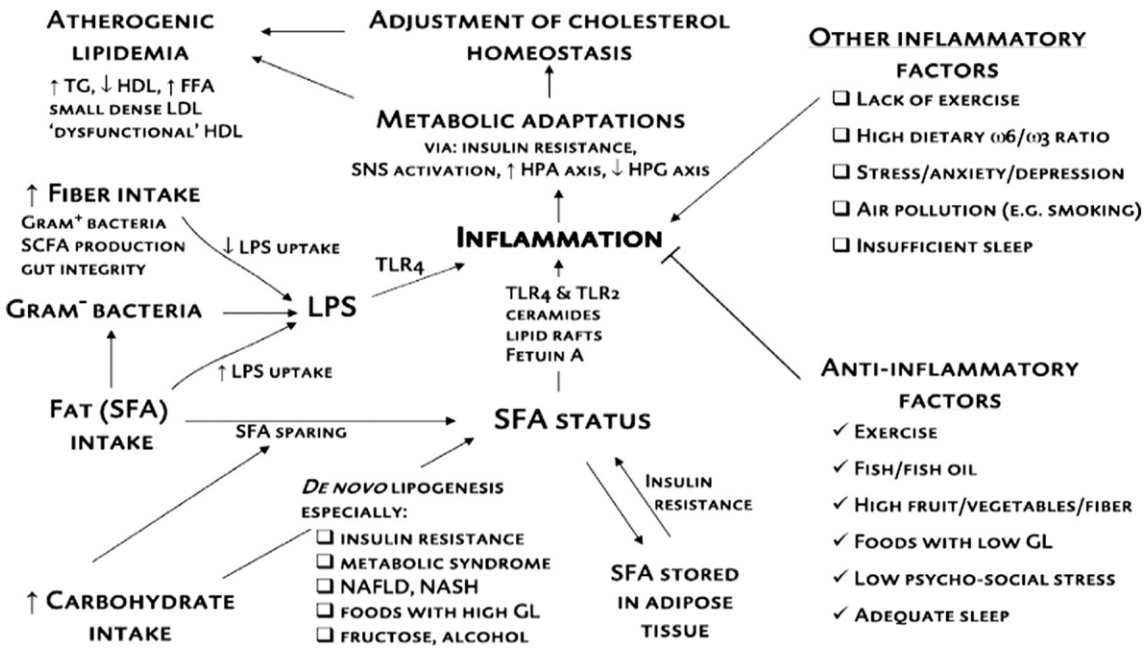

Fig. 7. Factors determining SFA status and its role in chronic systemic low-grade inflammation and atherogenic dyslipidemia. Chronic systemic low-grade inflammation is central in this pathophysiological cascade. Starting in the upper left of the figure. High-fiber intake may increase Gram-positive Firmicutes, resulting in a higher production of SCFA, which together support gut integrity and may decrease LPS uptake [293,294]. High-fat diets, especially those rich in SFA, have been shown to increase LPS uptake in the gut [241,242]. LPS may cause inflammation by its binding to TLR-4 [317]. A high-CHO intake induces DNL and the production of SFA, while it also promotes sparing of dietary SFA. DNL is also stimulated by insulin resistance, the metabolic syndrome, NAFLD/NASH and foods with high GL, fructose and alcohol [226]. A high-SFA status may cause inflammation *via* the activation of TLR-4 and TLR-2, ceramide production and by the formation of lipid rafts. Fetuin A, a liver-derived circulating glycoprotein, functions as an adaptor protein directly linking SFA to TLR-4 activation and promoting lipid-induced insulin resistance [269,270]. However, fetuin A acts as a pleiotropic molecule [273], which can also promote wound healing [277]. Excessive storage of SFA in adipose tissue may cause high free SFA in insulin-resistant subjects and upon fasting and thereby contribute to inflammation. Inflammation induces adaptations in metabolic (*e.g.* insulin resistance), hormonal (*e.g.* reduced insulin sensitivity, up-regulation of the HPA axis, down-regulation of the HPG axis) and nervous pathways [*e.g.* sympathetic nervous system (SNS) activation] that are jointly meant for the reallocation of energy-rich nutrients that spare glucose for the brain and immune system, and it forces other organs to use lipids for energy generation [3]. Among these changes, we find alterations in lipoprotein metabolism (high TGs, high free fatty acids, low HDL) and in cholesterol homeostasis (low HDL, small dense LDL, "dysfunctional" HDL), which are jointly known as the "atherogenic dyslipidemia" of the metabolic syndrome. All of these inflammation-induced adaptations aim at the short-term redistribution of energy, modulation of the inflammatory reaction and the repair of the damage produced by the infectious agent and immune system. However, in the long run, they cause the typically Western diseases of the metabolic syndrome. Whether SFA plays a relevant contributing role in the development of chronic systemic low-grade inflammation (the central factor in this pathophysiological cascade) is dependent on many other proinflammatory factors and their balance with antiinflammatory counterparts. Among the inflammatory factors are lack of exercise, high dietary ω6/ω3 ratio, chronic stress, anxiety and depression, air pollution (smoking included) and insufficient sleep. Antiinflammatory factors are *e.g.* physical exercise, fish and fish oil, high fruits, vegetables and fiber, foods with low GL, low psychosocial stress and adequate sleep [2]. Adapted from [150] with permission from Elsevier.

pathogen-associated molecular patterns and the subsequent activation of innate immune responses for host defense [221]. They are also crucial in shaping the adaptive immune response from its initiation to the development of immunological memory [245], in the sensing of metabolic disturbances and in linking immune responses to metabolic adjustments [109]. Among the different TLR subtypes, TLR-4 and TLR-2 on macrophages become activated in response to bacterial infection, tissue damage and, according to some authors (further explained), SFA [246,247]. LPS is among the natural ligands of the TLR-4 complex, while TLR-2 can recognize lipoproteins/lipopeptides of Gram-positive bacteria and mycoplasma [248]. LPS acts therefore as a signal molecule to detect the presence of the infectious source, with TLR-4 and TLR-2 constituting the sensors. The "lipid A" moiety (also named endotoxin) of LPS contains 12:0 and 14:0 and their hydroxyl derivatives. These are essential for the interaction with TLR-4 [249] and abundant in coconut oil (Table 1). It is now well documented that TLRs form homooligomers or heterooligomers [250] and that the homodimerization of TLR-4 is the initial step necessary for its activation [251]. TLR-4 is located on macrophages and adipocytes, which derive from a common precursor [252]. When activated, TLRs facilitate translocation of nuclear factor kappa B (NFκB) to the nucleus [253], inducing inhibition of insulin signaling, hepatic insulin resistance, activation of hepatic ceramide synthesis and other adaptations [254].

SFA can also become incorporated into specific "lipid raft" domains of the plasma membrane, where they enhance TLR-4 dimerization, tyrosine-protein kinase CSK recruitment [251,255] and the activation of downstream signaling pathways (*i.e.* JNK/AP-1) that may eventually inhibit insulin action [256]. Ceramides are produced *via* sphingomyelinase cleavage of membrane sphingolipids, from free FA by *de novo* synthesis or by recycling of more complicated glycosylated ceramides [257]. They modulate a variety of cellular responses including cell death, autophagy, inflammation and insulin signaling [258] and can lead to insulin resistance *via* different mechanisms. First, they can induce dephosphorylation of AKT2, an important signaling molecule in the insulin signaling pathway that is required for glucose transport, and thereby lower insulin signaling [259]. Second, ceramides can prevent insulin action through the activation of PKCζ, an atypical isoform of protein kinase C that binds AKT2 and thereby suppresses its activation [260] and involvement in the insulin signaling cascade [227]. SFA are involved in *de novo* ceramide biosynthesis, where serine and palmitoyl-CoA are condensed to form 3-ketosphinganine *via* a synergistic signaling of TLR-4 and LPS through a nontranscriptional mechanism [261]. Erridge [262] recently showed that apparently unspoiled food stuff contain large quantities of TLR-2 and TLR-4 stimulants (*i.e.* a sufficient SFA-containing microbial load) and can thereby trigger inflammatory signals. In line with these findings, experimental administration of the ligands of TLR-2 and TLR-4,

namely bacterial lipopeptides and LPS, to animal models of atherosclerosis and insulin resistance resulted in marked amplification of both conditions [240,263–265].

Chronic systemic low-grade tissue inflammation is emerging as a major cause of obesity-induced insulin resistance [266] with a strikingly strong accumulation of activated macrophages [267] in the adipose tissue of obese subjects [268] and a strong expression of proinflammatory genes. Although TLRs appear to be a basic element of SFA-induced insulin resistance [246,247], recent work indicates that SFA are not direct ligands for TLR-4 [269,270] and that TLR-2, TLR-4 nor ceramide signaling is directly required for SFA-induced hepatic insulin resistance [271,272]. The liver secretory glycoprotein fetuin A owns the ability to interact with a variety of receptors, including the insulin receptor and a variety of TLRs [273]. Fetuin A binds to and activates TLR-4 [269] and can also be secreted by adipose tissue, polarizing adipose tissue M2 macrophages towards the proinflammatory M1 subtype [274]. SFA, like 16:0, have been demonstrated to upregulate fetuin A mRNA expression in the liver by stimulating the binding of NFκB to the fetuin A promoter [273], inducing both inflammatory signaling and impairing insulin sensitivity [256]. Fetuin A has been presented as a biomarker of chronic inflammatory diseases, such as DM2 and atherosclerosis [270,275]. However, recent findings have recognized fetuin A to be a pleiotropic molecule [273], as witnessed by its ability to also act as a negative acute phase reactant in the setting of sepsis and endotoxemia [276] but also to promote wound healing [277]. These apparently contradictory effects are conceivable from the logics that an inflammatory stimulus causes not only damage but also protection from damage and stimulation of repair.

The involvement of SFA in low-grade inflammation may vary depending on the type of SFA (*i.e.* chain length) [154], other dietary components and lifestyle factors other than diet [3]. Several sources of SFA might ultimately cause inflammation *via* the abovementioned pathways. SFA intake is clearly one of these, but the aforementioned DNL from a CHO-rich diet (notably with high GL), presence of the metabolic syndrome, insulin resistance, NAFLD and high intakes of fructose and alcohol are other sources. Insulin resistance might increase the inflammatory potential of SFA by its release from adipose tissue as free FA, pointing at the important role of (long-term) dietary habits determining adipose tissue FA composition. The intimate connection between SFA and LPS, although mechanistically yet poorly understood, reappraises the important role of the gut flora.

7.3. The role of the gut

Studies in septic patients [278] and healthy volunteers injected with low-dose LPS [279] support the concept that the systemic inflammatory response is associated with increased levels of TG and decreases in TC, HDL-C and LDL-C, together with alterations in the composition of the lipoprotein particles, such as the appearance of both small dense LDL and the aforementioned "dysfunctional HDL" [115]. "Dysfunctional HDL" is proinflammatory, prooxidant and proatherogenic and does not support reverse cholesterol transport [280]. This newly formed HDL carries, among others, serum amyloid A instead of ApoA1, contains less esterified cholesterol, is rich in sphingolipids and rather binds to macrophages than to hepatocytes, exhibiting immunological activity. In contrast to its "dysfunctionality", these properties are highly functional during acute inflammatory circumstances, aiding in the inflammatory reaction, while, due to the active inhibition of reverse cholesterol transport, the loss of "normal" function causes, in an direct manner, the accumulation of cholesterol at the places where it is needed for repair [3,115]. However, in the long run, all of the above inflammation-induced allostatic adaptations in glucose and lipid homeostasis turn into the well-known impaired glucose homeostasis and "atherogenic dyslipidemia" components of the metabolic syndrome, which were in reality meant for the short-term energy redistribution, immune defense and repair.

As part of the host-defense mechanism against infection, the purposes of changing lipoprotein composition are the redistribution of the glucose and lipid energy sources among organs [3,281], the prevention of an exaggerated inflammatory response by the binding of LPS [282] and also the immediate promotion of repair. LPS binds to lipid particles, which may be part of a humoral detoxification mechanism. In healthy subjects, the LPS-binding protein (LBP) circulates in association with LDL, VLDL and chylomicrons, the latter being lipoproteins carrying lipids from the intestine to other tissues and the particles with the highest LPS-inactivating capacity [283]. The association of LBP with LDL and VLDL appears to result in part from the high affinity of LBP for ApoB. Consequently, LBP plays an important role in the reduction of LPS activity by catalyzing LPS transfer from micelles to circulating lipoproteins [284]. In addition, the LBP–LPS complexes may be part of a local gut defense mechanism to fight against translocated bacterial endotoxin [21]. The enhanced concentration of circulating LBP during an acute phase response may be crucial, since it diminishes the transfer of LPS to monocytes, reduces cytokine secretion and thereby modulates the immune response [285].

An important connection between potentially pathogenic bacteria, LPS and SFA might be that the bacterial cell membrane contains important amounts of SFA and MUFA, but not PUFA [249,286,287]. Circulating cell wall components from Gram-negative bacteria (*e.g.* "lipid A" moiety) profoundly activate TLR-2 and TLR-4, even in the central nervous system [288,289]. In the context above, it is conceivable that SFA intake increases both LDL-C and HDL-C concentrations [102,154,290]. We might be dealing with an evolutionary conserved immune response (further explained) to SFA and MUFA, either from bacteria or food [291], that is actually not meant to harm but on the contrary provides a survival advantage by means of protection/prevention from bacterial overgrowth in the gut, infection and bacterial LPS toxicity [292].

A high-fiber diet, as observed in rural African populations, decreases the ratio between the Gram-positive Firmicutes and the Gram-negative Bacteroidetes. This causes increased production of SCFA (less than 6 carbons) [293], which in turn improves gut integrity [294] and reduces paracellular uptake of LPS. Surprisingly, a recently identified Gram-negative microorganism constituting 3–5% of the gut bacterial community in healthy subjects, *Akkermansia muciniphila* [295], has been considered beneficial in the prevention of obesity and DM2. Gut colonization by *A. muciniphila* is related to decreased metabolic endotoxemia arising from a high-fat diet and its activity at the mucosal surface seems to help keeping the mucosal layer in shape [295].

As Paracelsus (1493–1541) already stated, "It is the dose (and circumstances) that makes the poison". Our food is composed of complex biological systems, such as meat, fish, vegetables and fruits, in which the nutrients, SFA included, obey to the balance that comes along with living material [3]. It is this balance on which hominins have evolved that may at best support our health, and it is therefore important to gain insight into the interaction of the various lifestyle factors.

7.4. Lifestyle and compensatory factors

Dietary SFA is only one of the many lifestyle factors playing a role in chronic systemic low-grade inflammation and the subsequent metabolic adaptations, including those causing changes in the concentrations of circulating lipids and lipoprotein-cholesterol. The environment provides us not only with many other proinflammatory stimuli than SFA but also with many compensating antiinflammatory stimuli [150] (Fig. 7). Among the inflammatory factors, we can find not

only other dietary components (*e.g.* a high dietary ω6/ω3 ratio) but also lack of exercise, chronic stress, anxiety and depression, air pollution (*e.g.* smoking, fine dust) and insufficient sleep. Examples of counteracting antiinflammatory stimuli are fish/fish oil, vegetables, fruits and fiber, low GL foods, exercise, low psychosocial stress and adequate sleep [2,3].

Some evidence for the concept of "compensatory factors" might come from observations of populations with traditional lifestyles. The traditionally living Maasai exhibit high intakes of SFA from milk and meat and very low ω3-PUFA intakes from fish, if any. They might be well protected from CVD by other factors (*e.g.* physical fitness and a temporary total abstinence from refined CHO during warrior hood; *i.e.* up to age 30 years), resulting in capacious coronary vessels [176]. In Tanzania, it is well known that the members of the Maasai tribe become at risk of developing the features of the metabolic syndrome when they move from rural to urban areas and adopt an urban lifestyle [296]. The inhabitants of the island of Kitava are at first glance suitable candidates for high CVD risk, as they eat high quantities of CHO (69 energy%) and SFA (17 energy%) and exhibit clear metabolic signs of DNL. Nevertheless, they do not suffer from the metabolic syndrome or CVD "despite" these seemingly unfavorable habits [297], perhaps due to their high intake of fish, high level of physical activity [297] or other compensatory antiinflammatory factors [2,3]. The CVD prevalence in France has been named "paradoxical", as there is high-SFA intake together with low mortality from CVD [298], an effect that has been attributed to diet quality and diversity [299] and also to the beneficial effects of moderate wine intake [300]. Nondiabetic urbanized Australian Aborigines subjected to a short-term (two weeks) and longer-term (three months) temporary reversion to their traditional diet (*i.e.* low CHO/high protein), and lifestyle in general, show an improvement in glucose tolerance and reduction of hyperinsulinemia and plasma TG concentrations [301,302]. In addition, the major metabolic abnormalities of DM2, *i.e.* fasting glucose, postprandial glucose clearance, fasting plasma insulin, insulin response to glucose and plasma TGs, were either greatly improved or completely normalized in another group of Australian Aborigines after a 7-week reversal of the urbanization process by living as hunter-gatherers in their traditional environment [303]. At least three factors known to improve insulin sensitivity (*i.e.* weight loss, low-fat diet and increased physical activity) were operating in this intervention study and might have jointly contributed to the favorable metabolic changes observed. Moreover, different responses to modern *versus* historically consumed foods (*i.e.* foods at which humans are clearly adapted) have been demonstrated [304,305]. As an example, a "newly" introduced form of beef (wagyu) induced a significantly greater postprandial inflammatory response than a traditional kind of meat (kangaroo) [305]. Whilst kangaroo is native to Australia, wagyu beef is at the opposite end of the spectrum in terms of human adaptation. Nevertheless, wagyu presents high proportions of MUFA and ω3 FA relative to other beef, and therefore, the difference in the inflammatory reaction may have been even greater between kangaroo and other "newer" and more processed meats. Finally, a recent systematic review of RCTs comparing the effects of a Paleolithic nutritional pattern (CHO:fat:protein = 30:40:30 energy%) with control diets based on the current dietary guidelines (45–60:25–30:10–20 energy%) showed greater short-term improvements in waist circumference, TGs, blood pressure, HDL-C and fasting blood sugar in the Paleolithic diet group than in the control diet group in participants with one or more components of the metabolic syndrome [202].

8. The role of fat in evolution: Lipids, metabolism and immunity

Our current Western lifestyle has been proposed as the cause of many interacting false inflammatory triggers, leading to a state of chronic systemic low-grade inflammation, insulin resistance, the metabolic syndrome and eventually the typically Western diseases [3]. The host response to infection/inflammation comprises profound changes in lipid metabolism and cholesterol fluxes, including increased hepatic DNL, lipolysis and cholesterol synthesis and cholesterol redistribution [306]. Inflammation induces several metabolic adaptations *via* hormonal (*e.g.* reduced insulin sensitivity) and nervous pathways [sympathetic nervous system and hypothalamus-pituitary-adrenal (HPA) and hypothalamus-pituitary-gonadal (HPG) axes], which jointly lead to the reallocation of energy-rich nutrients, notably glucose and fat, but also protein [281,307] (Fig. 7). In this scenario of allostasis, lipids have been identified as part of the innate immune response, while they also play an active role in the defense against invading pathogens [308].

The abovementioned mechanisms indicate a functional link between lipid metabolism and innate immunity that employs both pathogen- and nutrient-sensing pathways. The link may trace back to an evolutionary need for survival, resulting in the simultaneous development of organ systems and signaling pathways that mediate both processes [132]. Cells involved in metabolic and immune responses show evidence of coordination and coevolution. For instance, macrophages and adipocytes share a common lineage from mesodermal stem cells, they both carry the TLR-4 [252], both secrete cytokines and both become activated by pathogen-associated components, such as LPS [309]. As mentioned before, these evolutionary conserved mechanisms, now responsible of systemic low-grade inflammation and Western diseases, originally yielded an enormous survival advantage by protecting us from gut bacterial overgrowth and infection [291].

In view of the above, it seems logical to ponder that survival pressure would have favored both energy efficiency and storage ("thrifty genotype"), to prepare for times of food deprivation and to organize a potent immune response to defend the host against infectious agents [109]. When distressed, this system may aim at sparing glucose for the immune system and the brain *via* the induction of insulin resistance in selected insulin-dependent tissues and their switching to the fat burning mode while at the same time modulating the immune response and restoring the inflicted damage [3]. However, in our current society, these metabolic adaptations, meant for survival, threat to defeat us now that we chronically respond to the products of the cultural and industrial globalization in the 21st century [310]. By constituting an allostatic load, they contribute, together with other lifestyle factors, to the pathogenesis of the metabolic syndrome and its sequels, such as atherosclerosis [115], CVD and other typically Western diseases [3]. The underlying lifestyle causes of the metabolic syndrome, and notably their persistence, have never been encountered during human evolution until (very) recently.

9. Conclusions

We content that it is the interaction between many lifestyle factors that determines whether SFA, and as a matter of fact any nutrient, contributes to systemic low-grade inflammation, changes in lipoprotein metabolism and ultimately CVD risk (Fig. 7). The dysbalance between proinflammatory and antiinflammatory stimuli in our Western society does not originate from a single cause and, consequently, may also not become solved by a single "magic bullet". Resolution of the conflict between our self-made environment and our ancient genome may rather rely on returning to the lifestyle of the Paleolithic era according to the culture of the 21st century [3,68]. Accordingly, dietary guidelines might reconsider recommendations for replacing SFA, since "food, not nutrients, is the fundamental unit in nutrition" [311]. Researching the entire diet and, even better, diet in a broader context together with nondietary lifestyle factors, is a clear research priority, as opposed to the reductionist approach of studying

the effects of single nutrients such as SFA, CHO or PUFA, even if they have been conducted in an RCT design.

Conflict of interest

FAJM has received grants for studying human milk nutrients, including the human milk FA composition. There are no other potential conflicts to declare.

Authorship

BRN and FAJ formulated the research questions. BRN, DAJ and FAJ designed the study. BRN and FAJ wrote the article.

References

[1] Mathers CL, Loncar D. Projections of global mortality and burden of disease from 2002 to 2030. PLoS Med 2006;3:e442.
[2] Egger G, Dixon J. Non-nutrient causes of low-grade, systemic inflammation: support for a "canary in the mineshaft" view of obesity in chronic disease. Obes Rev 2011;12:339–45.
[3] Ruiz-Núñez B, Pruimboom L, Dijck-Brouwer DAJ, Muskiet FAJ. Lifestyle and nutritional imbalances associated with western diseases: causes and consequences of chronic systemic low grade inflammation in an evolutionary context. J Nutr Biochem 2013;24:1183–201.
[4] Danaei G, Ding EL, Mozaffarian D, Taylor B, Rehm J, Murray CJL, et al. The preventable causes of death in the United States: comparative risk assessment of dietary, lifestyle, and metabolic risk factors. PLoS Med 2009:e1000058.
[5] Popkin B. Global nutrition dynamics: the world is shifting rapidly toward a diet linked with noncommunicable diseases. Am J Clin Nutr 2006;84:289–98.
[6] Mozaffarian D, Appel L, Van Horn L. Components of a cardioprotective diet: new insights. Circulation 2011;123:2870–91.
[7] Bray GA, Redman L, de Jonge L, Covington J, Rood J, Brock C, et al. Effect of protein overfeeding on energy expenditure measured in a metabolic chamber. Am J Clin Nutr 2015 [ajcn. 091769].
[8] Ebbeling C, Swain J, Feldman H, Wong W, Hachey D, Garcia Lago E, et al. Effects of dietary composition on energy expenditure during weight-loss maintenance. JAMA (Chicago, Ill) 2012;307:2627–34.
[9] Anitschkow NN. A history of experimentation on arterial atherosclerosis in animals. Cowdry's arteriosclerosis: a survey of the problem. Springfield, IL: Charles C Thomas; 1967 21–44.
[10] Willett WC. Dietary fats and coronary heart disease. J Intern Med 2012;272: 13–24.
[11] Ecological analysis of the association between mortality and major risk factors of cardiovascular disease The World Health Organization MONICA ProjectInt J Epidemiol 1994;23:505–16.
[12] Sachdeva A, Cannon C, Deedwania P, Labresh K, Smith S, Dai D, et al. Lipid levels in patients hospitalized with coronary artery disease: an analysis of 136,905 hospitalizations in get with the guidelines. Am Heart J 2009;157:111–7 [e2].
[13] Keys A. Atherosclerosis: a problem in newer public health. J Mt Sinai Hosp N Y 1953;20:118–39.
[14] Keys A. Seven countries: a multivariate analysis of death and coronary heart disease. London, England: Harvard University Press, Cambridge, Massachusetts; 1980.
[15] Grundy SM, Bilheimer D, Blackburn H, Brown WV, Kwiterovich PO, Mattson F, et al. Rationale of the diet-heart statement of the American Heart Association. Report of nutrition committee. Circulation 1982;65:839A–54A.
[16] Krauss R. Lipoprotein subfractions and cardiovascular disease risk. Curr Opin Lipidol 2010;21:305–11.
[17] St-Pierre A, Cantin B, Dagenais G, Mauriège P, Bernard P, Després J, et al. Low-density lipoprotein subfractions and the long-term risk of ischemic heart disease in men: 13-year follow-up data from the Québec cardiovascular study. Arterioscler Thromb Vasc Biol 2005;25:553–9.
[18] Faghihnia N, Tsimikas S, Miller ER, Witztum JL, Krauss RM. Changes in lipoprotein(a), oxidized phospholipids, and LDL subclasses with a low-fat high-carbohydrate diet. J Lipid Res 2010;51:3324–30.
[19] Berneis K, Krauss R. Metabolic origins and clinical significance of LDL heterogeneity. J Lipid Res 2002;43:1363–79.
[20] U.S. Department of Agriculture and U.S., Department of Health and Human Services. Dietary guidelines for Americans, 2010. 7th ed. Washington, DC: U.S. Government Printing Office; 2010.
[21] German JB, Dillard CJ. Saturated fats: what dietary intake? Am J Clin Nutr 2004; 80:550–9.
[22] Simons K, Vaz WL. Model systems, lipid rafts, and cell membranes. Annu Rev Biophys Biomol Struct 2004;33:269–95.
[23] Haman F, Peronnet F, Kenny GP, Massicotte D, Lavoie C, Scott C, et al. Effect of cold exposure on fuel utilization in humans: plasma glucose, muscle glycogen, and lipids. J Appl Physiol (1985) 2002;93:77–84.
[24] Schwenk RW, Holloway GP, Luiken JJFP, Bonen A, Glatz JFC. Fatty acid transport across the cell membrane: regulation by fatty acid transporters. Prostaglandins Leukot Essent Fatty Acids 2010;82:149–54.
[25] Brookheart RT, Michel CI, Schaffer JE. As a matter of fat. Cell Metab 2009;10: 9–12.
[26] Watkins SM, Reifsnyder PR, Pan H, German JB, Leiter EH. Lipid metabolome-wide effects of the PPARγ agonist rosiglitazone. J Lipid Res 2002;43: 1809–17.
[27] Zock PL, de Vries JH, Katan MB. Impact of myristic acid *versus* palmitic acid on serum lipid and lipoprotein levels in healthy women and men. Arterioscler Thromb Vasc Biol 1994;14:567–75.
[28] Hellerstein MK. *De novo* lipogenesis in humans: metabolic and regulatory aspects. Eur J Clin Nutr 1999;53(Suppl. 1):S53–65.
[29] Schwarz J, Linfoot P, Dare D, Aghajanian K. Hepatic *de novo* lipogenesis in normoinsulinemic and hyperinsulinemic subjects consuming high-fat, low-carbohydrate and low-fat, high-carbohydrate isoenergetic diets. Am J Clin Nutr 2003;77:43–50.
[30] Gibson RA, Kneebone GM. Fatty acid composition of human colostrum and mature breast milk. Am J Clin Nutr 1981;34:252–7.
[31] Legrand P, Rioux V. The complex and important cellular and metabolic functions of saturated fatty acids. Lipids 2010;45:941–6.
[32] U.S. Department of Agriculture and U.S., Department of Health and Human Services. Report of the DGAC on the dietary guidelines for Americans, 2010; 2010.
[33] van Rossum C, Fransen HP, Verkaik-Kloosterman J, Buurma-Rethans E, Ocké MC. Dutch National Food Consumption Survey 2007–2010. Diet of children and adults aged 7 to 69 years; 2011 [RIVM Report 350050006].
[34] Varela-Moreiras G, Ruiz E, Valero T, Ávila JM, Pozo Sd. The Spanish diet: an update. Nutr Hosp 2013;28.
[35] Hughes TA, Heimberg M, Wang X, Wilcox H, Hughes SM, Tolley EA, et al. Comparative lipoprotein metabolism of myristate, palmitate, and stearate in normolipidemic men. Metab Clin Exp 1996;45:1108–18.
[36] Rioux V, Lemarchal P, Legrand P. Myristic acid, unlike palmitic acid, is rapidly metabolized in cultured rat hepatocytes. J Nutr Biochem 2000;11:198–207.
[37] Lambert JE, Ryan EA, Thomson AB, Clandinin MT. *De novo* lipogenesis and cholesterol synthesis in humans with long-standing type 1 diabetes are comparable to non-diabetic individuals. PLoS One 2013;8:e82530.
[38] Towler D, Gordon J, Adams S, Glaser L. The biology and enzymology of eukaryotic protein acylation. Annu Rev Biochem 1988;57:69–97.
[39] Geliebter A, Torbay N, Bracco EF, Hashim SA, Van Itallie TB. Overfeeding with medium-chain triglyceride diet results in diminished deposition of fat. Am J Clin Nutr 1983;37:1–4.
[40] Tremaroli V, Bäckhed F. Functional interactions between the gut microbiota and host metabolism. Nature 2012;489:242–9.
[41] Donohoe DR, Garge N, Zhang X, Sun W, O'Connell TM, Bunger MK, et al. The microbiome and butyrate regulate energy metabolism and autophagy in the mammalian colon. Cell Metab 2011;13:517–26.
[42] Davie JR. Inhibition of histone deacetylase activity by butyrate. J Nutr 2003;133: 2485S–93S.
[43] Sina C, Gavrilova O, Forster M, Till A, Derer S, Hildebrand F, et al. G protein-coupled receptor 43 is essential for neutrophil recruitment during intestinal inflammation. J Immunol 2009;183:7514–22.
[44] Tolhurst G, Heffron H, Lam YS, Parker HE, Habib AM, Diakogiannaki E, et al. Short-chain fatty acids stimulate glucagon-like peptide-1 secretion *via* the G-protein-coupled receptor FFAR2. Diabetes 2012;61:364–71.
[45] Belakavadi M, Prabhakar B, Salimath BP. Purification and characterization of butyrate-induced protein phosphatase involved in apoptosis of Ehrlich ascites tumor cells. Biochim Biophys Acta Gen Subj 2007;1770:39–47.
[46] Stiban J, Caputo L, Colombini M. Ceramide synthesis in the endoplasmic reticulum can permeabilize mitochondria to proapoptotic proteins. J Lipid Res 2008;49:625–34.
[47] Read WW, Lutz PG, Tashjian A. Human milk lipids. II. The influence of dietary carbohydrates and fat on the fatty acids of mature milk. A study in four ethnic groups. Am J Clin Nutr 1965;17:180–3.
[48] Smit EN, Martini IA, Mulder H, Boersma E, Muskiet F. Estimated biological variation of the mature human milk fatty acid composition. Prostaglandins Leukot Essent Fatty Acids 2002;66:549–55.
[49] Koletzko B, Thiel I, Abiodun PO. The fatty acid composition of human milk in Europe and Africa. J Pediatr 1992;120:S62–70.
[50] Insull Jr W, Hirsch J, James T, Ahrens Jr EH. The fatty acids of human milk. II. Alterations produced by manipulation of caloric balance and exchange of dietary fats. J Clin Invest 1959;38:443–50.
[51] Kuipers RS, Smit EN, van der Meulen J, Dijck-Brouwer DAJ, Boersma ER, Muskiet FAJ. Milk in the island of Chole [Tanzania] is high in lauric, myristic, arachidonic and docosahexaenoic acids, and low in linoleic acid reconstructed diet of infants born to our ancestors living in tropical coastal regions. Prostaglandins Leukot Essent Fatty Acids 2007;76:221–33.
[52] Francois CA, Connor SL, Wander RC, Connor WE. Acute effects of dietary fatty acids on the fatty acids of human milk. Am J Clin Nutr 1998;67:301–8.
[53] Lammi-Keefe CJ, Jensen RG. Lipids in human milk: a review. 2. Composition and fat-soluble vitamins. J Pediatr Gastroenterol Nutr 1984;3:172–98.
[54] Kuipers RS, Luxwolda MF, Offringa PJ, Boersma ER, Dijck-Brouwer DAJ, Muskiet FA. Gestational age dependent changes of the fetal brain, liver and adipose tissue fatty acid compositions in a population with high fish intakes. Prostaglandins Leukot Essent Fatty Acids 2012;86:189–99.

[55] Hamosh M, Iverson S, Mehta N, Spear M, Bitman J. In: Ghisolfi J, Putet G, editors. Lipids in human milk and their digestion by the newborn infant. Essential fatty acids and infant nutrition. Paris: John Libbey Eurotext; 1992. p. 119–37.
[56] Sun CQ, O'Connor CJ, Roberton AM. Antibacterial actions of fatty acids and monoglycerides against *Helicobacter pylori*. FEMS Immunol Med Microbiol 2003; 36:9–17.
[57] Batovska DI, Todorova T, Tsvetkova V, Najdenski HM. Antibacterial study of the medium chain fatty acids and their 1-monoglycerides: individual effects and synergistic relationships. Pol J Microbiol 2009;58:43–7.
[58] Cunnane SC, Ryan MA, Lin YH, Lim S, Salem N. Suckling rats actively recycle carbon from α-linolenate into newly synthesized lipids even during extreme dietary deficiency of n-3 polyunsaturates. Pediatr Res 2006;59:107–10.
[59] Yerushalmy J, Hilleboe HE. Fat in the diet and mortality from heart disease; a methodologic note. N Y State J Med 1957;57:2343–54.
[60] Harcombe Z, Baker JS, Cooper SM, Davies B, Sculthorpe N, DiNicolantonio JJ, et al. Evidence from randomised controlled trials did not support the introduction of dietary fat guidelines in 1977 and 1983: a systematic review and meta-analysis. Open Heart 2015;2:e000196.
[61] Hayes KC. Saturated fats and blood lipids: new slant on an old story. Can J Cardiol 1995;11(Suppl. G):39G–46G.
[62] Mustad VA, Ellsworth JL, Cooper AD, Kris-Etherton PM, Etherton TD. Dietary linoleic acid increases and palmitic acid decreases hepatic LDL receptor protein and mRNA abundance in young pigs. J Lipid Res 1996;37:2310–23.
[63] Shepherd J, Packard CJ, Grundy SM, Yeshurun D, Gotto Jr AM, Taunton OD. Effects of saturated and polyunsaturated fat diets on the chemical composition and metabolism of low density lipoproteins in man. J Lipid Res 1980;21:91–9.
[64] DiNicolantonio JJ. The cardiometabolic consequences of replacing saturated fats with carbohydrates or Ω-6 polyunsaturated fats: do the dietary guidelines have it wrong? Open Heart 2014;1:e000032.
[65] Hite A, Feinman R, Guzman G, Satin M, Schoenfeld P, Wood R. In the face of contradictory evidence: report of the dietary guidelines for Americans committee. Nutrition 2010;26:915–24.
[66] Centers for Disease Control and Prevention. National health and nutrition examination survey. Available at: http://www.cdc.gov/nchs/nhanes.htm. [(cited 21 December 2010). Available from: http://www.cdc.gov/nchs/nhanes.htm].
[67] Trends in intake of energy and macronutrients — United States, 1971–2000Morb Mortal Wkly Rep 2004;53:80–2.
[68] Cordain L, Eaton SB, Sebastian A, Mann N, Lindeberg S, Watkins BA, et al. Origins and evolution of the western diet: health implications for the 21st century. Am J Clin Nutr 2005;81:341–54.
[69] Laboratoriumgeneeskunde UMC, Amsterdam CV. Het faillissement van de verzadigd vethypothese van cardiovasculaire ziektes. Ned Tijdschr Klin Chem Labgeneesk 2012;37:192–211.
[70] Kuipers RS, Luxwolda MF, Dijck-Brouwer DA, Eaton SB, Crawford MA, Cordain L, et al. Estimated macronutrient and fatty acid intakes from an east African Paleolithic diet. Br J Nutr 2010;104:1666–87.
[71] Blasbalg TL, Hibbeln JR, Ramsden CE, Majchrzak SF, Rawlings RR. Changes in consumption of omega-3 and omega-6 fatty acids in the United States during the 20th century. Am J Clin Nutr 2011;93:950–62.
[72] Malhotra A. Saturated fat is not the major issue. Br Med J 2013;347:f6340.
[73] Gezondheidsraad 2006. Richtlijnen goede voeding 2006 — achtergronddocument. Gezonheidsraad; 2006.
[74] Ogden C, Carroll M, Kit B, Flegal K. Prevalence of obesity in the United States, 2009–2010. NCHS data brief, no 82. Hyattsville, MD: National Center for Health Statistics; 2012.
[75] Ogden CL, Carroll MD, Kit BK, Flegal KM. Prevalence of childhood and adult obesity in the United States, 2011–2012. JAMA 2014;311:806–14.
[76] RIVM. Nationaal kompas. Trends in de diabetes mellitus in Nederland; 2010.
[77] RIVM Nationaal Kompas Volksgezondheid. Trends overgewicht en obesitas in Nederland; 2010.
[78] Ernst ND, Sempos CT, Briefel RR, Clark MB. Consistency between US dietary fat intake and serum total cholesterol concentrations: the National Health and Nutrition Examination Surveys. Am J Clin Nutr 1997;66:965S–72S.
[79] Eaton SB. The ancestral human diet: what was it and should it be a paradigm for contemporary nutrition? Proc Nutr Soc 2006;65:1–6.
[80] Cordain L, Miller JB, Eaton SB, Mann N, Holt SH, Speth JD. Plant–animal subsistence ratios and macronutrient energy estimations in worldwide hunter-gatherer diets. Am J Clin Nutr 2000;71:682–92.
[81] Eaton SB, Eaton SB, Konner MJ. Paleolithic nutrition revisited: a twelve-year retrospective on its nature and implications. Eur J Clin Nutr 1997;51:207–16.
[82] Ghebremeskel K, Crawford MA. Nutrition and health in relation to food production and processing. Nutr Health 1994;9:237–53.
[83] Institute of Medicine (US). Panel on macronutrients, institute of medicine (US). standing committee on the scientific evaluation of dietary reference intakes. Dietary reference intakes for energy, carbohydrate, fiber, fat, fatty acids, cholesterol, protein, and amino acids; 2005.
[84] Ahrens EH, Hirsch J, Oette K, Farquhar JW, Stein Y. Carbohydrate-induced and fat-induced lipemia. Trans Assoc Am Physicians 1961;74:134–46.
[85] Chong MF, Hodson L, Bickerton A, Roberts R, Neville M, Karpe F, et al. Parallel activation of *de novo* lipogenesis and stearoyl-CoA desaturase activity after 3 d of high-carbohydrate feeding. Am J Clin Nutr 2008;87:817–23.
[86] Lichtenstein AH, Van Horn L. Very low fat diets. Circulation 1998;98:935–9.
[87] Nordmann A, Briel M, Keller U, Yancy W, Brehm B, Bucher H. Effects of low-carbohydrate vs low-fat diets on weight loss and cardiovascular risk factors: a meta-analysis of randomized controlled trials. Arch Intern Med 2006;166: 285–93.
[88] Hu F, Willett W. Optimal diets for prevention of coronary heart disease. JAMA 2002;288:2569–78.
[89] Hooper L, Summerbell C, Thompson R, Sills D, Roberts F, Moore H, et al. Reduced or modified dietary fat for preventing cardiovascular disease. Cochrane Database Syst Rev 2011;5:CD002137.
[90] Lichtenstein AH, Ausman LM, Carrasco W, Jenner JL, Ordovas JM, Schaefer EJ. Short-term consumption of a low-fat diet beneficially affects plasma lipid concentrations only when accompanied by weight loss. Hypercholesterolemia, low-fat diet, and plasma lipids. Arterioscler Thromb 1994;14:1751–60.
[91] Schwingshackl L, Hoffmann G. Comparison of the long-term effects of high-fat *v.* low-fat diet consumption on cardiometabolic risk factors in subjects with abnormal glucose metabolism: a systematic review and meta-analysis. Br J Nutr 2014;111:2047–58.
[92] Feinman RD, Pogozelski WK, Astrup A, Bernstein RK, Fine EJ, Westman EC, et al. Dietary carbohydrate restriction as the first approach in diabetes management: critical review and evidence base. Nutrition 2015;31:1–13.
[93] Xu J, Eilat-Adar S, Loria C, Goldbourt U, Howard BV, Fabsitz RR, et al. Dietary fat intake and risk of coronary heart disease: the strong heart study. Am J Clin Nutr 2006;84:894–902.
[94] Diet, nutrition and the prevention of chronic diseasesWorld Health Organ Tech Rep 2003;916 [i-viii, 1].
[95] Kris-Etherton PM, Harris WS, Appel LJ, AHA Nutrition Committee, American Heart Association. *Omega*-3 fatty acids and cardiovascular disease: new recommendations from the American Heart Association. Arterioscler Thromb Vasc Biol 2003;23:151–2.
[96] Eckel RH, Jakicic JM, Ard JD, Miller NH, Hubbard VS, Nonas CA, et al. 2013 AHA/ACC guideline on lifestyle management to reduce cardiovascular Risk. A report of the American College of Cardiology/American Heart Association task force on practice guidelines. J Am Coll Cardiol 2014;63:2960–84.
[97] Harris WS, Mozaffarian D, Rimm E, Kris-Etherton P, Rudel LL, Appel LJ, et al. *Omega*-6 fatty acids and risk for cardiovascular disease: a science advisory from the American Heart Association nutrition subcommittee of the council on nutrition, physical activity, and metabolism; council on cardiovascular nursing; and council on epidemiology and prevention. Circulation 2009;119: 902–7.
[98] National Institute for Public Health and the Environment. The Netherlands (RIVM). *Our food, our health. Healthy diet and safe food in The Netherlands*; 2006 270555009.
[99] Stampfer MJ, Sacks FM, Salvini S, Willett WC, Hennekens CH. A prospective study of cholesterol, apolipoproteins, and the risk of myocardial infarction. N Engl J Med 1991;325:373–81.
[100] Gotto Jr AM, Grundy SM. Lowering LDL cholesterol: questions from recent meta-analyses and subset analyses of clinical trial DataIssues from the interdisciplinary council on reducing the risk for coronary heart disease, ninth council meeting. Circulation 1999;99:E1–7.
[101] Gordon DJ, Probstfield JL, Garrison RJ, Neaton JD, Castelli WP, Knoke JD, et al. High-density lipoprotein cholesterol and cardiovascular disease. Four prospective American studies. Circulation 1989;79:8–15.
[102] Clarke R, Frost C, Collins R, Appleby P, Peto R. Dietary lipids and blood cholesterol: quantitative meta-analysis of metabolic ward studies. BMJ 1997; 314:112–7.
[103] Ravnskov U, Allen C, Atrens D, Enig MG, Groves B, Kauffman JM, et al. Studies of dietary fat and heart disease. Science 2002;295:1464–6.
[104] Weinberg S. The diet-heart hypothesis: a critique. J Am Coll Cardiol 2004;43: 731–3.
[105] O'Keefe JH, Carter MD, Lavie CJ. Primary and secondary prevention of cardiovascular diseases: a practical evidence-based approach. 2009;84: 741–57.
[106] Ridker PM. C-reactive protein, inflammation, and cardiovascular disease: clinical update. Tex Heart Inst J 2005;32:384–6.
[107] Vaughan CJ, Murphy MB, Buckley BM. Statins do more than just lower cholesterol. Lancet 1996;348:1079–82.
[108] Antonopoulos A, Margaritis M, Lee R, Channon K, Antoniades C. Statins as anti-inflammatory agents in atherogenesis: molecular mechanisms and lessons from the recent clinical trials. Curr Pharm Des 2012;18:1519–30.
[109] Hotamisligil GS, Erbay E. Nutrient sensing and inflammation in metabolic diseases. Nat Rev Immunol 2008;8:923–34.
[110] Reaven GM. The insulin resistance syndrome: definition and dietary approaches to treatment. Annu Rev Nutr 2005;25:391–406.
[111] Reaven G. Insulin resistance and coronary heart disease in nondiabetic individuals. Arterioscler Thromb Vasc Biol 2012;32:1754–9.
[112] Adiels M, Olofsson SO, Taskinen MR, Boren J. Overproduction of very low-density lipoproteins is the hallmark of the dyslipidemia in the metabolic syndrome. Arterioscler Thromb Vasc Biol 2008;28:1225–36.
[113] Verhoye E, Langlois MR. Circulating oxidized low-density lipoprotein: a biomarker of atherosclerosis and cardiovascular risk? Clin Chem Lab Med 2009;47:128–37.
[114] Steinberg D, Parthasarathy S, Carew TE, Khoo JC, Witztum JL. Beyond cholesterol. N Engl J Med 1989;320:915–24.
[115] Khovidhunkit W, Kim M, Memon RA, Shigenaga JK, Moser AH, Feingold KR, et al. Thematic review series: the pathogenesis of atherosclerosis. Effects of infection and inflammation on lipid and lipoprotein metabolism mechanisms and consequences to the host. J Lipid Res 2004;45:1169–96.

[116] Henriksen T, Mahoney EM, Steinberg D. Enhanced macrophage degradation of biologically modified low density lipoprotein. Arterioscler Thromb Vasc Biol 1983;3:149–59.
[117] McLaughlin T, Abbasi F, Cheal K, Chu J, Lamendola C, Reaven G. Use of metabolic markers to identify overweight individuals who are insulin resistant. Ann Intern Med 2003;139:802–9.
[118] Wallace S, Mozaffarian D. Trans-fatty acids and nonlipid risk factors. Curr Atheroscler Rep 2009;11:423–33.
[119] Brouwer I, Wanders A, Katan M. Effect of animal and industrial trans fatty acids on HDL and LDL cholesterol levels in humans — a quantitative review. PLoS One 2010;5:e9434.
[120] Brouwer I, Wanders A, Katan M. Trans fatty acids and cardiovascular health: research completed&quest. Eur J Clin Nutr 2013;67:541–7.
[121] Willett WC, Ascherio A. Trans fatty acids: are the effects only marginal? Am J Public Health 1994;84:722–4.
[122] Mozaffarian D, Katan MB, Ascherio A, Stampfer MJ, Willett WC. Trans fatty acids and cardiovascular disease. N Engl J Med 2006;354:1601–13.
[123] Katan MB, Zock PL, Mensink RP. Trans fatty acids and their effects on lipoproteins in humans. Annu Rev Nutr 1995;15:473–93.
[124] Ascherio A, Katan MB, Zock PL, Stampfer MJ, Willett WC. Trans fatty acids and coronary heart disease. N Engl J Med 1999;340:1994–8.
[125] Mozaffarian D, Aro A, Willett WC. Health effects of trans-fatty acids: experimental and observational evidence. Eur J Clin Nutr 2009;63(Suppl. 2): 5–21.
[126] Bendsen NT, Christensen R, Bartels EM, Astrup A. Consumption of industrial and ruminant trans fatty acids and risk of coronary heart disease: a systematic review and meta-analysis of cohort studies. Eur J Clin Nutr 2011; 65:773–83.
[127] Jakobsen MU, Overvad K, Dyerberg J, Heitmann BL. Intake of ruminant trans fatty acids and risk of coronary heart disease. Int J Epidemiol 2008;37:173–82.
[128] Mensink RP, Zock PL, Kester AD, Katan MB. Effects of dietary fatty acids and carbohydrates on the ratio of serum total to HDL cholesterol and on serum lipids and apolipoproteins: a meta-analysis of 60 controlled trials. Am J Clin Nutr 2003; 77:1146–55.
[129] Schwartz G, Olsson A, Abt M, Ballantyne C, Barter P, Brumm J, et al. Effects of dalcetrapib in patients with a recent acute coronary syndrome. N Engl J Med 2012;367:2089–99.
[130] Miller NE. CETP inhibitors and cardiovascular disease: time to think again. F1000Research; 2014 3.
[131] Ridker P. Statin therapy for low-LDL, high-hsCRP patients: from JUPITER to CORONA. Clin Chem 2010;56:505–7.
[132] Hotamisligil GS. Inflammation and metabolic disorders. Nature 2006;444:860–7.
[133] Dias C, Garg R, Wood L, Garg M. Saturated fat consumption may not be the main cause of increased blood lipid levels. Med Hypotheses 2013.
[134] Marchioli R, Barzi F, Bomba E, Chieffo C, Di Gregorio D, Di Mascio R, et al. GISSI-prevenzione investigators. Early protection against sudden death by n-3 polyunsaturated fatty acids after myocardial infarction: time-course analysis of the results of the gruppo italiano per lo studio Della sopravvivenza nell'infarto miocardico (GISSI)-prevenzione. Circulation 2002;105:1897–903.
[135] Yokoyama M, Origasa H. Effects of eicosapentaenoic acid on cardiovascular events in Japanese patients with hypercholesterolemia: rationale, design, and baseline characteristics of the Japan EPA Lipid Intervention Study (JELIS). Am Heart J 2003;146:613–20.
[136] Mariani J, Doval HC, Nul D, Varini S, Grancelli H, Ferrante D, et al. N-3 polyunsaturated fatty acids to prevent atrial fibrillation: updated systematic review and meta-analysis of randomized controlled trials. J Am Heart Assoc 2013;2:e005033.
[137] Andreasen J, Schmidt E. Therapeutic potential of marine n-3 fatty acids in CABG patients. Curr Opin Pharmacol 2012;12:142–6.
[138] Kromhout D, Giltay EJ, Geleijnse JM. n–3 fatty acids and cardiovascular events after myocardial infarction. N Engl J Med 2010;363:2015–26.
[139] Balk EM, Lichtenstein AH, Chung M, Kupelnick B, Chew P, Lau J. Effects of omega-3 fatty acids on serum markers of cardiovascular disease risk: a systematic review. Atherosclerosis 2006;189:19–30.
[140] Eslick GD, Howe PR, Smith C, Priest R, Bensoussan A. Benefits of fish oil supplementation in hyperlipidemia: a systematic review and meta-analysis. Int J Cardiol 2009;136:4–16.
[141] Mozaffarian D. Fish and n-3 fatty acids for the prevention of fatal coronary heart disease and sudden cardiac death. Am J Clin Nutr 2008;87:1991S–6S.
[142] Garg ML, Thomson AB, Clandinin MT. Interactions of saturated, n-6 and n-3 polyunsaturated fatty acids to modulate arachidonic acid metabolism. J Lipid Res 1990;31:271–7.
[143] MacDonald-Wicks LG, Garg ML. Incorporation of n-3 fatty acids into plasma and liver lipids of rats: importance of background dietary fat. Lipids 2004;39:545–51.
[144] Dabadie H, Motta C, Peuchant E, LeRuyet P, Mendy F. Variations in daily intakes of myristic and α-linolenic acids in sn-2 position modify lipid profile and red blood cell membrane fluidity. Br J Nutr 2006;96:283–9.
[145] Dabadie H, Peuchant E, Bernard M, LeRuyet P, Mendy F. Moderate intake of myristic acid in sn-2 position has beneficial lipidic effects and enhances DHA of cholesteryl esters in an interventional study. J Nutr Biochem 2005;16: 375–82.
[146] Dubnov G, Berry EM. Omega-6/omega-3 fatty acid ratio: the Israeli paradox; 2004.
[147] Simopoulos AP. Importance of the omega-6/omega-3 balance in health and disease: evolutionary aspects of diet. World Rev Nutr Diet 2011;102:10–21.
[148] Hoenselaar R. Saturated fat and cardiovascular disease: the discrepancy between the scientific literature and dietary advice. Nutrition 2012;28:118–23.
[149] Ravnskov U. A hypothesis out-of-date. The diet-heart idea. J Clin Epidemiol 2002;55:1057–63.
[150] Ruiz-Núñez B, Kuipers R, Luxwolda M, De Graaf D, Breeuwsma B, Dijck-Brouwer D, et al. Saturated fatty acid (SFA)-status and SFA-intake exhibit different relations with serum total cholesterol and lipoprotein-cholesterol: a mechanistic explanation centered around lifestyle-induced low grade inflammation. [
] J Nutr Biochem 2014;25:304–12.
[151] Kuipers RS, de Graaf DJ, Luxwolda MF, Muskiet MH, Dijck-Brouwer DA, Muskiet FA. Saturated fat, carbohydrates and cardiovascular disease. Neth J Med 2011;69: 372–8.
[152] Ramsden CE, Hibbeln JR, Majchrzak SF, Davis JM. n-6 fatty acid-specific and mixed polyunsaturate dietary interventions have different effects on CHD risk: a meta-analysis of randomised controlled trials. Br J Nutr 2010;104:1586–600.
[153] Siri Tarino P, Sun Q, Hu F, Krauss R. Meta-analysis of prospective cohort studies evaluating the association of saturated fat with cardiovascular disease. Am J Clin Nutr 2010;91:535–46.
[154] Micha R, Mozaffarian D. Saturated fat and cardiometabolic risk factors, coronary heart disease, stroke, and diabetes: a fresh look at the evidence. Lipids 2010;45: 893–905.
[155] CBO 2006. Cardiovasculair risicomanagement 2006; 2010.
[156] Pedersen JI, Ringstad J, Almendingen K, Haugen TS, Stensvold I, Thelle DS. Adipose tissue fatty acids and risk of myocardial infarction — a case–control study. Eur J Clin Nutr 2000;54:618–25.
[157] Clifton P, Keogh J, Noakes M. Trans fatty acids in adipose tissue and the food supply are associated with myocardial infarction. J Nutr 2004;134:874–9.
[158] Fuhrman B, Barba M, Krogh V, Micheli A, Pala V, Lauria R, et al. Erythrocyte membrane phospholipid composition as a biomarker of dietary fat. Ann Nutr Metab 2006;50:95–102.
[159] Donnelly KL, Smith CI, Schwarzenberg SJ, Jessurun J, Boldt MD, Parks EJ. Sources of fatty acids stored in liver and secreted *via* lipoproteins in patients with nonalcoholic fatty liver disease. J Clin Invest 2005;115:1343–51.
[160] Fretts AM, Mozaffarian D, Siscovick DS, Djousse L, Heckbert SR, King IB, et al. Abstract P001: plasma phospholipid saturated fatty acids and incident atrial fibrillation: the cardiovascular health study. Circulation 2014;129:AP001.
[161] Hunter JE, Zhang J, Kris-Etherton PM. Cardiovascular disease risk of dietary stearic acid compared with trans, other saturated, and unsaturated fatty acids: a systematic review. Am J Clin Nutr 2010;91:46–63.
[162] Tholstrup T, Miller GJ, Bysted A, Sandstrom B. Effect of individual dietary fatty acids on postprandial activation of blood coagulation factor VII and fibrinolysis in healthy young men. Am J Clin Nutr 2003;77:1125–32.
[163] Thijssen MA, Hornstra G, Mensink RP. Stearic, oleic, and linoleic acids have comparable effects on markers of thrombotic tendency in healthy human subjects. J Nutr 2005;135:2805–11.
[164] Oh K, Hu F, Manson J, Stampfer M, Willett W. Dietary fat intake and risk of coronary heart disease in women: 20 years of follow-up of the Nurses' Health Study. Am J Epidemiol 2005;161:672–9.
[165] Puaschitz NG, Strand E, Norekvål TM, Dierkes J, Dahl L, Svingen GFT, et al. Dietary intake of saturated fat is not associated with risk of coronary events or mortality in patients with established coronary artery disease. J Nutr 2015 [jn. 114.203505].
[166] Mente A, de Koning L, Shannon H, Anand S. A systematic review of the evidence supporting a causal link between dietary factors and coronary heart disease. Arch Intern Med 2009;169:659–69.
[167] Skeaff CMM, Jody. Dietary fat and coronary heart disease: summary of evidence from prospective cohort and randomised controlled trials. Ann Nutr Metab 2009;55:173–201.
[168] Scarborough P, Rayner M, van Dis I, Norum K. Meta-analysis of effect of saturated fat intake on cardiovascular disease: overadjustment obscures true associations. Am J Clin Nutr 2010;92:458–9.
[169] Houston DK, Ding J, Lee JS, Garcia M, Kanaya AM, Tylavsky FA, et al. Dietary fat and cholesterol and risk of cardiovascular disease in older adults: the Health ABC Study. Cardiovasc Dis 2011;21:430–7.
[170] Arunima S, Rajamohan T. Influence of virgin coconut oil-enriched diet on the transcriptional regulation of fatty acid synthesis and oxidation in rats — a comparative study. Br J Nutr 2014;111:1782–90.
[171] Forsythe CE, Phinney SD, Fernandez ML, Quann EE, Wood RJ, Bibus DM, et al. Comparison of low fat and low carbohydrate diets on circulating fatty acid composition and markers of inflammation. Lipids 2008;43:65–77.
[172] Volek JS, Fernandez ML, Feinman RD, Phinney SD. Dietary carbohydrate restriction induces a unique metabolic state positively affecting atherogenic dyslipidemia, fatty acid partitioning, and metabolic syndrome. Prog Lipid Res 2008;47:307–18.
[173] Ramsden C, Faurot K, Carrera Bastos P, Cordain L, De Lorgeril M, Sperling L. Dietary fat quality and coronary heart disease prevention: a unified theory based on evolutionary, historical, global, and modern perspectives. Curr Treat Options Cardiovasc Med 2009;11:289–301.
[174] Stanhope JM, Sampson VM, Prior IA. The Tokelau island migrant study: serum lipid concentration in two environments. J Chronic Dis 1981;34:45–55.
[175] Lindeberg S, Nilsson Ehle P, Terént A, Vessby B, Scherstén B. Cardiovascular risk factors in a Melanesian population apparently free from stroke and ischaemic heart disease: the Kitava study. J Intern Med 1994;236:331–40.
[176] Mann GV, Spoerry A, Gray M, Jarashow D. Atherosclerosis in the Masai. Am J Epidemiol 1972;95:26–37.

[177] German JB, Gibson R, Krauss R, Nestel P, Lamarche B, van Staveren W, et al. A reappraisal of the impact of dairy foods and milk fat on cardiovascular disease risk. Eur J Nutr 2009;48:191–203.
[178] Micha R, Wallace S, Mozaffarian D. Red and processed meat consumption and risk of incident coronary heart disease, stroke, and diabetes mellitus: a systematic review and meta-analysis. Circulation 2010;121:2271–83.
[179] de Oliveira Otto MC, Mozaffarian D, Kromhout D, Bertoni A, Sibley C, Jacobs D, et al. Dietary intake of saturated fat by food source and incident cardiovascular disease: the Multi-Ethnic Study of Atherosclerosis. Am J Clin Nutr 2012;96: 397–404.
[180] Benatar J, Sidhu K, Stewart RAH. Effects of high and low fat dairy food on cardio-metabolic risk factors: a meta-analysis of randomized studies. PLoS One 2013;8: e76480.
[181] Soedamah-Muthu S, Ding E, Al Delaimy W, Hu F, Engberink M, Willett W, et al. Milk and dairy consumption and incidence of cardiovascular diseases and all-cause mortality: dose–response meta-analysis of prospective cohort studies. Am J Clin Nutr 2011;93:158–71.
[182] Yakoob MY, Shi P, Hu FB, Campos H, Rexrode KM, Orav EJ, et al. Circulating biomarkers of dairy fat and risk of incident stroke in U.S. men and women in 2 large prospective cohorts. Am J Clin Nutr 2014;100:1437–47.
[183] Qin Li-Qiang LQ. Dairy consumption and risk of cardiovascular disease: an updated meta-analysis of prospective cohort studies. Asia Pac J Clin Nutr 2015; 24(3):90–100.
[184] Huth P, Park K. Influence of dairy product and milk fat consumption on cardiovascular disease risk: a review of the evidence. Adv Nutr 2012;3:266–85.
[185] Samuelson G, Bratteby LE, Mohsen R, Vessby B. Dietary fat intake in healthy adolescents: inverse relationships between the estimated intake of saturated fatty acids and serum cholesterol. Br J Nutr 2001;85:333–41.
[186] Stachowska E, Baskiewicz M, Marchlewicz M, Czupryńska K, Kaczmarczyk M, Wiszniewska B, et al. Conjugated linoleic acids regulate triacylglycerol and cholesterol concentrations in macrophages/foam cells by the modulation of CD36 expression. Acta Biochim Pol 2010;57:379–84.
[187] Mozaffarian D, de Oliveira Otto MC, Lemaitre RN, Fretts AM, Hotamisligil G, Tsai MY, et al. Trans-palmitoleic acid, other dairy fat biomarkers, and incident diabetes: the Multi-Ethnic Study of Atherosclerosis (MESA). Am J Clin Nutr 2013; 97:854–61.
[188] Lichtenstein A, Appel L, Brands M, Carnethon M, Daniels S, Franch H, et al. Diet and lifestyle recommendations revision 2006: a scientific statement from the American Heart Association nutrition committee. Circulation 2006;114:82–96.
[189] National Health and Medical Research Council. Eat for health. Australian dietary guidelines. Providing the scientific evidence for healthier Australian diets. Canberra: National Health and Medical Research Council; 2013.
[190] Lim SS, Vos T, Flaxman AD, Danaei G, Shibuya K, Adair-Rohani H, et al. A comparative risk assessment of burden of disease and injury attributable to 67 risk factors and risk factor clusters in 21 regions, 1990–2010: a systematic analysis for the global burden of disease study 2010. Lancet 2013;380:2224–60.
[191] Binnie MA, Barlow K, Johnson V, Harrison C. Red meats: time for a paradigm shift in dietary advice. Meat Sci 2014;98:445–51.
[192] Mann GV, Shaffer RD, Rich A. Physical fitness and immunity to heart-disease in Masai. Lancet 1965;2:1308–10.
[193] Jakobsen M, Dethlefsen C, Joensen A, Stegger J, Tjønneland A, Schmidt E, et al. Intake of carbohydrates compared with intake of saturated fatty acids and risk of myocardial infarction: importance of the glycemic index. Am J Clin Nutr 2010; 91:1764–8.
[194] Schwingshackl L, Hoffmann G. Long-term effects of low-fat diets either low or high in protein on cardiovascular and metabolic risk factors: a systematic review and meta-analysis. Nutr J 2013;12:48.
[195] Kratz M, Weigle D, Breen P, Meeuws K, Burden V, Callahan H, et al. Exchanging carbohydrate or protein for fat improves lipid-related cardiovascular risk profile in overweight men and women when consumed *ad libitum*. J Invest Med 2010; 58:711–9.
[196] Salmerón J, Ascherio A, Rimm EB, Colditz GA, Spiegelman D, Jenkins DJ, et al. Dietary fiber, glycemic load, and risk of NIDDM in men. Diabetes Care 1997;20: 545–50.
[197] Salmerón J, Manson JE, Stampfer MJ, Colditz GA, Wing AL, Willett WC. Dietary fiber, glycemic load, and risk of non-insulin-dependent diabetes mellitus in women. JAMA 1997;277:472–7.
[198] Toshimitsu K, Matsuura B, Ohkubo I, Niiya T, Furukawa S, Hiasa Y, et al. Dietary habits and nutrient intake in non-alcoholic steatohepatitis. Nutrition 2007;23:46–52.
[199] Papandreou D, Karabouta Z, Pantoleon A, Rousso I. Investigation of anthropo-metric, biochemical and dietary parameters of obese children with and without non-alcoholic fatty liver disease. Appetite 2012;59:939–44.
[200] Papandreou D, Rousso I, Malindretos P, Makedou A, Moudiou T, Pidonia I, et al. Are saturated fatty acids and insulin resistance associated with fatty liver in obese children? Clin Nutr 2008;27:233–40.
[201] Gardner CD, Kiazand A, Alhassan S, Kim S, Stafford RS, Balise RR, et al. Comparison of the Atkins, zone, ornish, and LEARN diets for change in weight and related risk factors among overweight premenopausal women: the a TO Z weight loss study: a randomized trial. JAMA 2007;297:969–77.
[202] Manheimer EW, van Zuuren EJ, Fedorowicz Z, Pijl H. Paleolithic nutrition for metabolic syndrome: systematic review and meta-analysis. Am J Clin Nutr 2015; 102:922–32.
[203] Liu S, Willett WC, Stampfer MJ, Hu FB, Franz M, Sampson L, et al. A prospective study of dietary glycemic load, carbohydrate intake, and risk of coronary heart disease in US women. Am J Clin Nutr 2000;71:1455–61.
[204] Estruch R, Ros E, Salas-Salvadó J, Covas M, Corella D, Arós F, et al. Primary prevention of cardiovascular disease with a Mediterranean diet. N Engl J Med 2013;368:1279–90.
[205] Samaha FF, Iqbal N, Seshadri P, Chicano KL, Daily DA, McGrory J, et al. A low-carbohydrate as compared with a low-fat diet in severe obesity. N Engl J Med 2003;348:2074–81.
[206] Gross LS, Li L, Ford ES, Liu S. Increased consumption of refined carbohydrates and the epidemic of type 2 diabetes in the United States: an ecologic assessment. Am J Clin Nutr 2004;79:774–9.
[207] Malik VS, Pan A, Willett WC, Hu FB. Sugar-sweetened beverages and weight gain in children and adults: a systematic review and meta-analysis. Am J Clin Nutr 2013;98:1084–102.
[208] Te Morenga LA, Howatson AJ, Jones RM, Mann J. Dietary sugars and cardiometabolic risk: systematic review and meta-analyses of randomized controlled trials of the effects on blood pressure and lipids. Am J Clin Nutr 2014; 100:65–79.
[209] Wolfe RR. Metabolic interactions between glucose and fatty acids in humans. Am J Clin Nutr 1998;67:519S–26S.
[210] Wolfe, BM Giovannetti, PM. Short-term effects of substituting protein for carbohydrate in the diets of moderately hypercholesterolemic human subjects. Metab Clin Exp. 1991;40:338–343.
[211] Brinkworth GD, Noakes M, Parker B, Foster P, Clifton PM. Long-term effects of advice to consume a high-protein, low-fat diet, rather than a conventional weight-loss diet, in obese adults with type 2 diabetes: one-year follow-up of a randomised trial. Diabetologia 2004;47:1677–86.
[212] Ludwig DS, Majzoub JA, Al Zahrani A, Dallal GE, Blanco I, Roberts SB. High glycemic index foods, overeating, and obesity. Pediatrics 1999;103:E26.
[213] Flock MR, Fleming JA, Kris-Etherton PM. Macronutrient replacement options for saturated fat: effects on cardiovascular health. Curr Opin Lipidol 2014;25:67–74.
[214] Ramsden C, Zamora D, Leelarthaepin B, Majchrzak Hong S, Faurot K, Suchindran C, et al. Use of dietary linoleic acid for secondary prevention of coronary heart disease and death: evaluation of recovered data from the Sydney Diet Heart Study and updated meta-analysis. Br Med J 2013;346:e8707.
[215] Felton CV, Crook D, Davies MJ, Oliver MF. Dietary polyunsaturated fatty acids and composition of human aortic plaques. Lancet (Lond, Engl) 1994;344:1195–6.
[216] Schwingshackl L, Hoffmann G. Dietary fatty acids in the secondary prevention of coronary heart disease: a systematic review, meta-analysis and meta-regression. BMJ Open 2014;4 [e004487-2013-004487].
[217] Mozaffarian D, Micha R, Wallace S. Effects on coronary heart disease of increasing polyunsaturated fat in place of saturated fat: a systematic review and meta-analysis of randomized controlled trials. PLoS Med 2010;7:e1000252.
[218] Lawson LD, Kummerow FA. Beta-oxidation of the coenzyme a esters of elaidic, oleic, and stearic acids and their full-cycle intermediates by rat heart mitochondria. Biochim Biophys Acta 1979;573:245–54.
[219] Chowdhury R, Warnakula S, Kunutsor S, Crowe F, Ward HA, Johnson L, et al. Association of dietary, circulating, and supplement fatty acids with coronary RiskA systematic review and meta-analysis. Ann Intern Med 2014;160:398–406.
[220] Xu H, Barnes GT, Yang Q, Tan G, Yang D, Chou CJ, et al. Chronic inflammation in fat plays a crucial role in the development of obesity-related insulin resistance. J Clin Invest 2003;112:1821–30.
[221] O'Neill LAJ. How toll-like receptors signal: what we know and what we don't know. Curr Opin Immunol 2006;18:3–9.
[222] Masoro EJ, Chaikoff IL, Chernick SS, Felts JM. Previous nutritional state and glucose conversion to fatty acids in liver slices. J Biol Chem 1950;185:845–56.
[223] Klein S, Wolfe RR. Carbohydrate restriction regulates the adaptive response to fasting. Am J Physiol 1992;262:E631–6.
[224] Phinney SD, Bistrian BR, Wolfe RR, Blackburn GL. The human metabolic response to chronic ketosis without caloric restriction: physical and biochemical adaptation. Metab Clin Exp 1983;32:757–68.
[225] Jensen MD, Caruso M, Heiling V, Miles JM. Insulin regulation of lipolysis in nondiabetic and IDDM subjects. Diabetes 1989;38:1595–601.
[226] Lustig R. Fructose: metabolic, hedonic, and societal parallels with ethanol. J Am Diet Assoc 2010;110:1307–21.
[227] Penkov DN, Egorov AD, Mozgovaya MN, Tkachuk VA. Insulin resistance and adipogenesis: role of transcription and secreted factors. Biochem Mosc 2013;78: 8–18.
[228] McAuley K, Mann J. Thematic review series: patient-oriented research. Nutritional determinants of insulin resistance. J Lipid Res 2006;47:1668–76.
[229] Glass C, Olefsky J. Inflammation and lipid signaling in the etiology of insulin resistance. Cell Metab 2012;15:635–45.
[230] Leclercq I, Horsmans Y. Nonalcoholic fatty liver disease: the potential role of nutritional management. Curr Opin Clin Nutr Metab Care 2008;11:766–73.
[231] Kwon OW, Jun DW, Lee SM, Lee KN, Lee HL, Lee OY, et al. Carbohydrate but not fat is associated with elevated aminotransferases. Aliment Pharmacol Ther 2012.
[232] Kim H, Nam C, Jee S, Han K, Oh D, Suh I. Normal serum aminotransferase concentration and risk of mortality from liver diseases: prospective cohort study. BMJ. Br Med J 2004;328:983.
[233] Chiu S, Sievenpiper J, de Souza R, Cozma A, Mirrahimi A, Carleton A, et al. Effect of fructose on markers of non-alcoholic fatty liver disease (NAFLD): a systematic review and meta-analysis of controlled feeding trials. Eur J Clin Nutr 2014;68:416–23.
[234] Moore JB, Gunn PJ, Fielding BA. The role of dietary sugars and *de novo* lipogenesis in Non-alcoholic fatty liver disease. Nutrients 2014;6:5679–703.
[235] Ochoa M, Lallès J, Malbert C, Val-Laillet D. Dietary sugars: their detection by the gut–brain axis and their peripheral and central effects in health and diseases. Eur J Nutr 2014;54:1–24.

[236] Nseir W, Nassar F, Assy N. Soft drinks consumption and nonalcoholic fatty liver disease. World J Gastroenterol 2010;16:2579–88.
[237] Lemaitre R, King I, Sotoodehnia N, Knopp R, Mozaffarian D, McKnight B, et al. Endogenous red blood cell membrane fatty acids and sudden cardiac arrest. Metab Clin Exp 2010;59:1029–34.
[238] Volk BM, Kunces LJ, Freidenreich DJ, Kupchak BR, Saenz C, Artistizabal JC, et al. Effects of step-wise increases in dietary carbohydrate on circulating saturated fatty acids and palmitoleic acid in adults with metabolic syndrome. PLoS One 2014;9:e113605.
[239] Mozaffarian D, Cao H, King IB, Lemaitre RN, Song X, Siscovick DS, et al. Trans-palmitoleic acid, metabolic risk factors, and new-onset diabetes in US AdultsA cohort study. Ann Intern Med 2010;153:790–9.
[240] Deopurkar R, Ghanim H, Friedman J, Abuaysheh S, Sia C, Mohanty P, et al. Differential effects of cream, glucose, and orange juice on inflammation, endotoxin, and the expression of toll-like receptor-4 and suppressor of cytokine signaling-3. Diabetes Care 2010;33:991–7.
[241] Mani V, Hollis J, Gabler N. Dietary oil composition differentially modulates intestinal endotoxin transport and postprandial endotoxemia. Nutr Metab 2013; 10:6.
[242] Moreira APB, Texeira TFS, Ferreira A. Influence of a high-fat diet on gut microbiota, intestinal permeability and metabolic endotoxaemia. Br J Nutr 2012; 108:801–9.
[243] Erridge C, Attina T, Spickett CM, Webb DJ. A high-fat meal induces low-grade endotoxemia: evidence of a novel mechanism of postprandial inflammation. Am J Clin Nutr 2007;86:1286–92.
[244] Cani PD, Delzenne NM. The role of the gut microbiota in energy metabolism and metabolic disease. Curr Pharm Des 2009;15:1546–58.
[245] Wolowczuk I, Verwaerde C, Viltart O, Delanoye A, Delacre M, Pot B, et al. Feeding our immune system: impact on metabolism. Clin Dev Immunol 2008;2008.
[246] Nguyen MT, Audrey, Favelyukis S, Nguyen A, Reichart D, Scott P, et al. A subpopulation of macrophages infiltrates hypertrophic adipose tissue and is activated by free fatty acids *via* toll-like receptors 2 and 4 and JNK-dependent pathways. J Biol Chem 2007;282:35279–92.
[247] Huang S, Rutkowsky J, Snodgrass R, Ono Moore K, Schneider D, Newman J, et al. Saturated fatty acids activate TLR-mediated pro-inflammatory signaling pathways. J Lipid Res 2012.
[248] Uematsu SA. Toll-like receptors and innate immunity. J Mol Med 2006;84: 712–25.
[249] Raetz CR, Reynolds CM, Trent MS, Bishop RE. Lipid a modification systems in Gram-negative bacteria. Annu Rev Biochem 2007;76:295–329.
[250] Gay N, Gangloff M, Weber ANR. Toll-like receptors as molecular switches. Nat Rev Immunol 2006;6:693–8.
[251] Wong SW, Kwon M, Choi AMK, Kim H, Nakahira K, Hwang DH. Fatty acids modulate toll-like receptor 4 activation through regulation of receptor dimerization and recruitment into lipid rafts in a reactive oxygen species-dependent manner. J Biol Chem 2009;284:27384–92.
[252] Margioris AN. Fatty acids and postprandial inflammation. Curr Opin Clin Nutr Metab Care 2009;12:129–37.
[253] Kawai T, Akira S. TLR signaling. Cell Death Differ 2006;13:816–25.
[254] Chavez J, Summers S. A ceramide-centric view of insulin resistance. Cell Metab 2012;15:585–94.
[255] Holzer R, Park E, Li N, Tran H, Chen M, Choi C, et al. Saturated fatty acids induce c-Src clustering within membrane subdomains, leading to JNK activation. Cell 2011;147:173–84.
[256] Johnson AMF, Olefsky J. The origins and drivers of insulin resistance. Cell 2013; 152:673–84.
[257] Gault C, Obeid L, Hannun Y. An overview of sphingolipid metabolism: from synthesis to breakdown. Adv Exp Med Biol 2010;688:1–23.
[258] Grösch S, Schiffmann S, Geisslinger G. Chain length-specific properties of ceramides. Prog Lipid Res 2012;51:50–62.
[259] Teruel T, Hernandez R, Lorenzo M. Ceramide mediates insulin resistance by tumor necrosis factor-alpha in brown adipocytes by maintaining Akt in an inactive dephosphorylated state. Diabetes 2001;50:2563–71.
[260] Fox T, Houck K, O'Neill S, Nagarajan M, Stover T, Pomianowski P, et al. Ceramide recruits and activates protein kinase C zeta (PKC zeta) within structured membrane microdomains. J Biol Chem 2007;282:12450–7.
[261] Schilling J, Machkovech H, He L, Sidhu R, Fujiwara H, Weber K, et al. Palmitate and LPS trigger synergistic ceramide production in primary macrophages. J Biol Chem 2012.
[262] Erridge C. The capacity of foodstuffs to induce innate immune activation of human monocytes *in vitro* is dependent on food content of stimulants of toll-like receptors 2 and 4. Br J Nutr 2011;105:15–23.
[263] Cani P, Amar J, Iglesias M, Poggi M, Knauf C, Bastelica D, et al. Metabolic endotoxemia initiates obesity and insulin resistance. Diabetes 2007;56: 1761–72.
[264] Mullick AE, Tobias PS, Curtiss LK. Modulation of atherosclerosis in mice by toll-like receptor 2. J Clin Invest 2005;115:3149–56.
[265] Lehr HA, Sagban TA, Ihling C, Zahringer U, Hungerer KD, Blumrich M, et al. Immunopathogenesis of atherosclerosis: endotoxin accelerates atherosclerosis in rabbits on hypercholesterolemic diet. Circulation 2001;104:914–20.
[266] Gregor M, Hotamisligil G. Inflammatory mechanisms in obesity. Annu Rev Immunol 2011;29:415–45.
[267] Lumeng CN, Deyoung SM, Saltiel AR. Macrophages block insulin action in adipocytes by altering expression of signaling and glucose transport proteins. Am J Physiol Endocrinol Metab 2007;292:E166–74.
[268] Weisberg SP, McCann D, Desai M, Rosenbaum M, Leibel RL, Ferrante Jr AW. Obesity is associated with macrophage accumulation in adipose tissue. J Clin Invest 2003;112:1796–808.
[269] Heinrichsdorff J, Olefsky J. Fetuin-A: the missing link in lipid-induced inflammation. Nat Med 2012;18:1182–3.
[270] Pal D. Fetuin-A acts as an endogenous ligand of TLR4 to promote lipid-induced insulin resistance. Nat Med 2012;18:1279–85.
[271] Erridge CS, Nilesh. Saturated fatty acids do not directly stimulate toll-like receptor signaling. Arterioscler Thromb Vasc Biol 2009;29:1944–9.
[272] Galbo T, Perry R, Jurczak M, Camporez J, Alves T, Kahn M, et al. Saturated and unsaturated fat induce hepatic insulin resistance independently of TLR-4 signaling and ceramide synthesis *in vivo*. Proc Natl Acad Sci U S A 2013.
[273] Mukhopadhyay S, Mondal SA, Kumar M, Dutta D. Pro-inflammatory and anti-inflammatory attributes of fetuin-A: a novel hepatokine: modulating cardiovascular and glycemic outcomes in metabolic syndrome. Endocr Pract 2014;1-18.
[274] Chatterjee P, Seal S, Mukherjee S, Kundu R, Ray S, Mukhopadhyay S, et al. Adipocyte fetuin-A contributes to macrophage migration into adipose tissue and their polarization. J Biol Chem 2013.
[275] Stefan N, Häring H. The role of hepatokines in metabolism. Nat Rev Endocrinol 2013;9:144–52.
[276] Li W, Zhu S, Li J, Huang Y, Rongrong Z, Fan X, et al. A hepatic protein, fetuin-A, occupies a protective role in lethal systemic inflammation. PLoS One 2011;6: e16945.
[277] Wang H, Sama AE. Anti-inflammatory role of fetuin-A in injury and infection. Curr Mol Med 2012;12:625–33.
[278] Levels JHM, Lemaire LCJM, van den Ende AE, van Deventer SJH, van Lanschot JJB. Lipid composition and lipopolysaccharide binding capacity of lipoproteins in plasma and lymph of patients with systemic inflammatory response syndrome and multiple organ failure. Crit Care Med 2003;31:1647–53.
[279] Hudgins LC, Parker TS, Levine DM, Gordon BR, Saal SD, Jiang X, et al. A single intravenous dose of endotoxin rapidly alters serum lipoproteins and lipid transfer proteins in normal volunteers. J Lipid Res 2003;44:1489–98.
[280] Otocka Kmiecik A, Mikhailidis D, Nicholls S, Davidson M, Rysz J, Banach M. Dysfunctional HDL: a novel important diagnostic and therapeutic target in cardiovascular disease? Prog Lipid Res 2012;51:314–24.
[281] Straub RH, Cutolo M, Buttgereit F, Pongratz G. Energy regulation and neuroendocrine-immune control in chronic inflammatory diseases. J Intern Med 2010;267:543–60.
[282] Liao W, Florén C. Hyperlipidemic response to endotoxin — a part of the host-defence mechanism. Scand J Infect Dis 1993;25:675–82.
[283] Vreugdenhil ACE, Rousseau CH, Hartung T, Greve JWM, van 't Veer C, Buurman WA. Lipopolysaccharide (LPS)-binding protein mediates LPS detoxification by chylomicrons. J Immunol 2003;170:1399–405.
[284] Wurfel MM, Hailman E, Wright SD. Soluble CD14 acts as a shuttle in the neutralization of lipopolysaccharide (LPS) by LPS-binding protein and reconstituted high density lipoprotein. J Exp Med 1995;181:1743–54.
[285] Zweigner J, Gramm HJ, Singer OC, Wegscheider K, Schumann RR. High concentrations of lipopolysaccharide-binding protein in serum of patients with severe sepsis or septic shock inhibit the lipopolysaccharide response in human monocytes. Blood 2001;98:3800–8.
[286] Alikunhi NM, Narayanasamy R, Kandasamy K. Fatty acids in an estuarine mangrove ecosystem. Rev Biol Trop 2010;58:577–87.
[287] Braunthal SD, Holt SC, Tanner AC, Socransky SS. Cellular fatty acid composition of *Actinobacillus actinomycetemcomitans* and *Haemophilus aphrophilus*. J Clin Microbiol 1980;11:625–30.
[288] Laflamme N, Soucy G, Rivest S. Circulating cell wall components derived from Gram-negative, not Gram-positive, bacteria cause a profound induction of the gene-encoding toll-like receptor 2 in the CNS. J Neurochem 2001;79:648–57.
[289] Laflamme N, Rivest S. Toll-like receptor 4: the missing link of the cerebral innate immune response triggered by circulating Gram-negative bacterial cell wall components. FASEB J 2001;15:155–63.
[290] Mensink RP, Zock PL, Katan MB, Hornstra G. Effect of dietary cis and trans fatty acids on serum lipoprotein[a] levels in humans. J Lipid Res 1992;33:1493–501.
[291] Viarengo G, Sciara MI, Salazar MO, Kieffer PM, Furlan RL, Garcia Vescovi E. Unsaturated long chain free fatty acids are input signals of the *Salmonella enterica* PhoP/PhoQ regulatory system. J Biol Chem 2013;288:22346–58.
[292] Pajkrt D, Doran JE, Koster F, Lerch PG, Arnet B, van der Poll T, et al. Antiinflammatory effects of reconstituted high-density lipoprotein during human endotoxemia. J Exp Med 1996;184:1601–8.
[293] De Filippo C, Cavalieri D, Di Paola M, Ramazzotti M, Poullet JB, Massart S, et al. Impact of diet in shaping gut microbiota revealed by a comparative study in children from Europe and rural Africa. Proc Natl Acad Sci U S A 2010;107: 14691–6.
[294] Macia L, Thorburn A, Binge L, Marino E, Rogers K, Maslowski K, et al. Microbial influences on epithelial integrity and immune function as a basis for inflammatory diseases. Immunol Rev 2012;245:164–76.
[295] Everard A, Belzer C, Geurts L, Ouwerkerk J, Druart C, Bindels LG, 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.
[296] Carlin L, Aspray T, Edwards R, Hayes L, Kitange H, Unwin N. Civilization and its discontents: non-communicable disease, metabolic syndrome and rural–urban migration in Tanzania. Urban anthropology and studies of cultural systems and world economic development; 2001 51–70.
[297] Lindeberg S. Paleolithic diet and evolution medicine: the key to diseases of the western world. Lakartidningen 2005;102:1976–8.

[298] Renaud S, de Lorgeril M. Wine, alcohol, platelets, and the French paradox for coronary heart disease. Lancet 1992;339:1523–6.
[299] Drewnowski A, Henderson SA, Shore AB, Fischler C, Preziosi P, Hercberg S. Diet quality and dietary diversity in France: implications for the French paradox. J Am Diet Assoc 1996;96:663–9.
[300] Lippi G, Franchini M, Favaloro EJ, Targher G. Moderate red wine consumption and cardiovascular disease risk: beyond the "French paradox", 31; 2010 059–70.
[301] O'Dea K, Spargo RM, Akerman K. The effect of transition from traditional to urban life-style on the insulin secretory response in Australian aborigines. Diabetes Care 1980;3:31–7.
[302] O'Dea K, Sinclair AJ. Increased proportion of arachidonic acid in plasma lipids after 2 weeks on a diet of tropical seafood. Am J Clin Nutr 1982;36:868–72.
[303] O'Dea K. Marked improvement in carbohydrate and lipid metabolism in diabetic Australian aborigines after temporary reversion to traditional lifestyle. Diabetes 1984;33:596–603.
[304] Cordain L, Eaton S, Brand Miller J, Mann N, Hill K. Original communications-the paradoxical nature of hunter-gatherer diets: meat-based, yet non-atherogenic. Eur J Clin Nutr 2002;56:S42.
[305] Arya F, Egger S, Colquhoun D, Sullivan D, Pal S, Egger G. Differences in postprandial inflammatory responses to a "modern" v. traditional meat meal: a preliminary study. Br J Nutr 2010;104:724–8.
[306] Grunfeld C, Feingold KR. Regulation of lipid metabolism bycytokines during host defense. Nutrition 1996;12:S24–6.
[307] Straub RH. Evolutionary medicine and chronic inflammatory state — known and new concepts in pathophysiology. J Mol Med 2012;90:523–34.
[308] Thormar H, Hilmarsson H. The role of microbicidal lipids in host defense against pathogens and their potential as therapeutic agents. Chem Phys Lipids 2007; 150:1–11.
[309] Chung S, Lapoint K, Martinez K, Kennedy A, Boysen-Sandberg M, McIntosh M. Preadipocytes mediate lipopolysaccharide-induced inflammation and insulin resistance in primary cultures of newly differentiated human adipocytes. Endocrinology 2006;147:5340–51.
[310] Wellen K, Hotamisligil G. Inflammation, stress, and diabetes. J Clin Invest 2005; 115:1111–9.
[311] Jacobs DR, Tapsell LC. Food, not nutrients, is the fundamental unit in nutrition. Nutr Rev 2007;65:439–50.
[312] U.S. Department of Agriculture, Agricultural Research Service. USDA national nutrient database for standard reference, release 26. Nutrient data laboratory home page; 2014.
[313] Offringa P. Perinatal studies in the Caribbean. Aspects of nutrition and metabolism. University of Groningen (thesis) 1990:125–52.
[314] Gibson RA. Australian fish—An excellent source of both arachidonic acid and ω-3 polyunsaturated fatty acids. Lipids 1983;18(11):743–52.
[315] Kuipers RS, Luxwolda MF, Sango WS, Kwesigabo G, Dijck-Brouwer DA, Muskiet FA. Postdelivery changes in maternal and infant erythrocyte fatty acids in 3 populations differing in fresh water fish intakes. Prostaglandins Leukot Essent Fatty Acids 2011;85:387–97.
[316] Volek JS, Phinney SD. A new look at carbohydrate-restricted diets: separating fact from fiction. Nutr Today 2013;48:E1–7.
[317] Lee JY, Zhao L, Youn HS, Weatherill AR, Tapping R, Feng L, et al. Saturated fatty acid activates but polyunsaturated fatty acid inhibits toll-like receptor 2 dimerized with toll-like receptor 6 or 1. J Biol Chem 2004;279:16971–9.
[318] European Food Safety Authority (EFSA). Scientific Opinion on Dietary Reference Values for fats, including saturated fatty acids, polyunsaturated fatty acids, monounsaturated fatty acids, trans fatty acids, and cholesterol. EFSA J 2010;8: 1461.