Article:
Thorning, TK, Bertram, HC, Bonjour, J-P et al. (15 more authors) (2017) Whole dairy matrix or single nutrients in assessment of health effects: current evidence and knowledge gaps. The American Journal of Clinical Nutrition, 105 (5). pp. 1033-1045. ISSN 0002-9165

https://doi.org/10.3945/ajcn.116.151548

The American Journal of Clinical Nutrition
Official publication of the American Society for Nutrition

The American Journal of Clinical Nutrition
AJCN/2016/151548
Version 2

Whole dairy matrix or single nutrients in assessment of health effects: current evidence and knowledge gaps

Corresponding Author: Arne Astrup
Additional Authors: Tanja Kongerslev Thorning , Hanne Christine Christine Bertram , Jean-Philippe Bonjour, Lisette de Groot , Didier DUPONT , Emma L Feeney , Richard Ipsen , Jean-Michel Lecerf, Alan R Mackie , Michelle C McKinley, Marie-Caroline Michalski, Didier Rémond, Ulf Risérus , Sabita Soedamah-Muthu , Tine Tholstrup , Connie M Weaver, Ian D Givens

TITLE PAGE

1. Title: Whole dairy matrix or single nutrients in assessment of health effects: current evidence and knowledge gaps

2. Authors: Tanja Kongerslev Thorning[1], Hanne Christine Bertram[2], Jean-Philippe Bonjour[3], Lisette de Groot[4], Didier Dupont[5], Emma Feeney[6], Richard Ipsen[7], Jean Michel Lecerf[8], Alan Mackie[9], Michelle C. McKinley[10], Marie-Caroline Michalski[11,12], Didier Rémond[13], Ulf Risérus[14], Sabita S. Soedamah-Muthu[15], Tine Tholstrup[1], Connie Weaver[16], Arne Astrup[1], Ian Givens[17]

3. Affiliations:

[1]University of Copenhagen, Department of Nutrition, Exercise and Sports, Copenhagen, Denmark [2]Aarhus University, Department of Food Science, Aarslev, Denmark

[3]Department of Internal Medicine, University Hospitals and Faculty of Medicine, Geneva, Switzerland

[4]Wageningen University, Human Nutrition, Wageningen, The Netherlands

[5]INRA, Science and Technology of Milk and Eggs, Rennes, France

[6]Food for Health Ireland, Science Centre South, University College Dublin, Republic of Ireland

[7]University of Copenhagen, Department of Food Science, Copenhagen, Denmark

[8]Institut Pasteur de Lille, Lille Cedex, France

[9]University of Leeds, School of Food Science & Nutrition, Leeds, United Kingdom

[10]School of Medicine, Dentistry & Biomedical Science, Queen's University Belfast, Belfast, United Kingdom

[11]Univ-Lyon, CarMeN laboratory, INRA UMR1397, Université Claude Bernard Lyon 1, INSERM U1060, INSA-Lyon, IMBL, F-69621 Villeurbanne, France

[12]Centre de Recherche en Nutrition Humaine Rhône-Alpes (CRNH-RA), Centre Européen pour la Nutrition et la Santé (CENS), F-69600, Oullins, France

[13]INRA, Université d'Auvergne, UMR 1019, UNH, Clermont-Ferrand, France

[14]Uppsala University, Department of Public Health and Caring Sciences, Uppsala, Sweden

[15]Wageningen University & Research, Division of Human Nutrition, Wageningen, The Netherlands [16]Purdue University, Nutrition Science, West Lafayette, USA

[17]Institute for Food, Nutrition and Health, University of Reading, Reading, United Kingdom

4. Names for PubMed indexing: Thorning, Bertram, Bonjour, de Groot, Michalski, Rémond, Risérus, Soedamah-Muthu, Tholstrup, Weaver, Astrup, Givens

5. None

6. Disclaimers: none

7. Corresponding author: Arne Astrup, MD, DMSc, Head of Department, Professor

Department of Nutrition, Exercise and Sports, Faculty of Science, University of Copenhagen Noerre Alle 5, DK-2200 Copenhagen N, Denmark

E-mail: ast@nexs.ku.dk

Telephone: +45 35 33 24 76

8. Sources of Support: The University of Copenhagen received an unrestricted grant from GEIE European Milk Forum, France, to arrange an expert workshop in Copenhagen and this report is based on the presentations and discussions from that workshop. The sponsors had no influence on the meeting program, the selection of speakers, and were not present at the meeting. Moreover, they have not been involved in the production of this manuscript.

9. Short running head: Importance of the dairy matrix

10. Abbreviations: Bone mineral density, BMD; cardiovascular disease, CVD; coronary heart disease, CHD; high-density lipoprotein, HDL; glucose-dependent insulinotropic polypeptide, GIP; low-density lipoprotein, LDL; Milk Fat Globule Membrane, MFGM; short chain fatty acids, SCFA; saturated fatty acids, SFAs; triglycerides, TG; type 2 diabetes, T2D; ultra-high temperature, UHT

11: Not relevant

ABSTRACT

Foods consist of a large number of different nutrients contained in a complex structure. The nature of the food structure and the nutrients therein (i.e. the food matrix) will determine nutrient digestion and absorption, thereby altering the overall nutritional properties of the food. Thus, the food matrix may exhibit a different relationship with health indicators than that observed with the single nutrients studied in isolation. The evidence for a dairy matrix effect was presented and discussed by an expert panel at a closed workshop, and the following consensus was reached: 1) Current evidence does not support a positive association between intake of dairy products and risk of CVD (i.e. stroke and coronary heart disease) and type 2 diabetes. By contrast, fermented dairy products such as cheese and yogurt generally show inverse associations. 2) Intervention studies indicate that the metabolic effects of whole dairy may be different to that of single dairy constituents when considering effects on body weight, cardiometabolic disease risk, and bone health. 3) Different dairy products seem to be distinctly linked to health effects and disease risk markers. 4) Different dairy structures and common processing methods may enhance interactions between nutrients in the dairy matrix, which may modify the metabolic effects of dairy consumption. 5) In conclusion, the nutritional value of dairy products should not be considered equivalent to their nutrient contents, but rather be considered based on the biofunctionality of the nutrients within the dairy food structures. 6) Further research on health effects of whole dairy foods are warranted alongside the more traditional approach of studying the health effects of single nutrients. Future diet assessment and recommendations should carefully consider evidence on whole food effects alongside the evidence for individual nutrients. Current knowledge gaps and recommendations for priorities in future research on dairy were identified and presented.

Keywords: Whole dairy, dairy nutrients, dairy structure, fermented dairy, dairy protein, MFGM, calcium, phosphorous, blood lipids, bioavailability

1. INTRODUCTION

Nutritional evaluation of the relationship between diet and health has traditionally focused on individual food constituents such as proteins, fats, carbohydrates and micronutrients separately. This reductionist approach (1), linking one nutrient to one health effect, may partly explain some of the discrepancies between a food's predicted health effect based on its nutrient content and its actual health effect when consumed as a whole food. The diet does not consist of single nutrients, but of whole foods, either alone, or alongside many other foods as part of a meal. The foods have complex structures both physically and nutritionally which affect digestion and absorption and may generate interactions within the food matrix altering the bioactive properties of the nutrients in ways that are not predictable from the nutrition label information.

Recent studies have shown that the food matrix can modify the nutritional properties of a food (2,3). Plant-based foods contain cellular structures that require breakdown before the encapsulated nutrients and bioactive compounds can be released and absorbed. This may be achieved by processing (e.g. industrial and cooking) and by oral mastication, but bioavailability of nutrients may be limited in some foods (3,4). An example is almonds, for which the food matrix attenuates postprandial lipaemia after consumption (3). A slower release rate of nutrients from naturally encapsulated systems was shown to increase satiety after consumption (5). This paradigm may also apply to other foods, such as dairy products (6,7). Dairy products are major sources of high-quality protein and calcium, and their nutritional value is well recognized (8). However, dairy products are also a major contributor to saturated fat in the diet, and thus have been targeted as one of the main dietary causes of CVD, as saturated fat increases low density lipoprotein (LDL)-cholesterol (9). The majority of current dietary guidelines include dairy products as part of a healthy diet, but recommend the low-fat or fat-free versions, to reduce the intake of saturated fat (U.S. population goal is to reduce saturated fat intake below 10 % of total energy intake)(10). However, it is

questioned if the current dietary recommendations on dairy consumption takes full account of whole food effects or rely on extrapolations of health effects of the single nutrients contained in dairy. A difficulty with many epidemiological studies, which have examined dairy intake in relation to health, is the broad categorization of dairy foods. Some studies refer to dairy as one homogenous food group, while others attempt to divide dairy foods into low-fat and high-fat dairy. However, these terms lack a universal standard definition, and can lead to products being differentially categorized by different researchers. Further, these terms do not take into account the different dairy food matrices, i.e. the sum of dairy nutrients within the specific dairy food structure. Due to differences in dairy matrices between various types of dairy products, these foods may have distinct effects on health. Hence, focusing on the dairy matrix, instead of the dairy nutrients, allows investigation of the health effects of dairy products based on how they are actually consumed by the population.

2. AIM AND METHODS

The current paper is based on the presentations, discussions and conclusions from a workshop on the dairy matrix. The closed consensus workshop of invited scientists was arranged by the co-chairs Arne Astrup and Ian Givens and held at Bernstorff Slot, Gentofte, Denmark, 28-29 September 2016. The workshop program and speakers were selected by the co-chairs. A total of 19 experts were initially identified to represent different scientific areas within the dairy matrix. These 19 scientists were invited to the workshop, five declined to participate but two of these suggested replacements for their participation. Hence, 18 scientists consented to participate in the workshop, and out of these 10 were invited to present research on the dairy matrix within their scientific areas of expertise. Each presentation was allocated a discussant scientist, who was asked to challenge the content of the presentation in order to achieving a balanced view of the evidence. The overall aim of the workshop was to exploit the synergy between various scientific areas in order to reach of a more

comprehensive understanding of the dairy matrix, as well as to identify gaps in the existing knowledge and currently missing evidence. Prior to the workshop, speakers were asked to write an abstract based on 3-5 key papers within their area of research contributing with knowledge on the dairy matrix. A total of 42 background papers were chosen from the literature, and the data and viewpoints from these, including additional literature, were presented at the workshop. Each presentation was challenged by an extensive discussion held in plenum. The workshop was rounded off with group sessions, which aimed to identify gaps in the existing knowledge (**Table 1**) and, subsequently, these were presented and discussed in a plenary session. The outcome of the workshop is presented in this scoping review, which should act as a primer for future research. All workshop participants were from higher educational institutions or research institutes; hence no industrial representatives, or sponsors, participated in the workshop or contributed to the scientific paper.

3. EVIDENCE OF A DAIRY MATRIX EFFECT – SUMMARY OF CONSENSUS AND CONCLUSIONS FROM THE WORKSHOP

The major dairy components shown to impact the human health are fat, protein (whey and casein), minerals (calcium, magnesium and phosphate), sodium, and the components of the milk fat globule membrane (MFGM); i.e. the biological membrane surrounding the lipid droplets in milk. Dairy products are heterogeneous in terms of the content of these components but also their physical structure (**Table 2**). Although cheese has a high fat content, it is more similar in composition to yogurt and milk than to butter, due to the protein, mineral, and MFGM content. High-fat dairy products, with exception of butter, are specifically rich in MFGM (11). Yogurt and cheese are both fermented dairy products containing bacteria with a potential ability to produce bioactive peptides and short chain fatty acids (SCFAs). Compared to yogurt and milk, cheese, however, has a higher content of calcium, protein (mostly casein with only small amounts of whey protein), fat, and

sodium. Moreover, the structure of cheese is more solid, whereas yogurt has a gel structure and milk has a liquid structure. For cheese there are numerous methods of production and ripening, and these influence the extent of protein and fat degradation. In general, for fermented dairy products the bacterial culture used, as well as the type and amount of ingredients such as stabilizers, texturizers, and flavors is also highly variable. Due to differences in composition and structures of different dairy foods, it is plausible that they have different effects on health, and even more so compared to intake of a single nutrient, i.e. a dietary supplement. This concept was explored at the workshop and the evidence is summarized in the scoping review below, which is structured according to the categories of experimental designs and area of health or disease.

3.1. Evidence from meta-analyses on observational studies on dairy products and disease risk

The majority of observational studies on dairy products and disease risk have either focused on dairy components such as calcium, fat and protein or on dairy products either as a homogenous food group or according to high or low fat contents. However, disregarding the differences in composition and food matrix between various types of dairy products may have blurred the analyses, and may have led to misinterpretation of the true associations. The number of observational studies focusing on dairy products as whole foods is currently increasing, and some of these have in recent years been pooled in a number of meta-analyses with different end-points. The results from the meta-analyses on dairy product intake and risk of CVD, hypertension, and type 2 diabetes (T2D) are presented below.

3.1.1. Intake of dairy products and risk of CVD

In a meta-analysis of prospective cohort studies by Qin et al. (2015), total dairy consumption was inversely associated with overall risk of CVD (RR=0.88, 95% CI: 0.81- 0.96) and stroke (RR=0.87, 95% CI: 0.77- 0.99) (12). For the specific dairy products, only cheese intake was inversely associated with risk of stroke (RR=0.91, 95% CI: 0.84-0.98) and risk of coronary heart disease

(CHD) (RR=0.84, 95% CI: 0.71-1.00). This meta-analysis was, however, limited by broad categories of dairy and outcome variables, and also no dose–response analyses were conducted. Nevertheless, the findings were supported by a recent meta-analysis in which total dairy intake was associated with lower risk of stroke (RR=0.91, 95 % CI: 0.83-0.99), and cheese intake was associated with a lower risk of CHD (RR=0.82, 95 % CI: 0.72-0.93) and stroke (RR=0.87, 95 % CI: 0.77-0.99) (13). The dose-response analyses did, however, not support an inverse dose–response relationship between the dairy variables and CHD or stroke, after adjusting for within-study covariance. In the latest meta-analysis of dairy intake and stroke, which included 18 cohort studies with 8 to 26 years of follow-up and a total of 762,414 individuals and 29,943 stroke events, total dairy intake was not associated with stroke risk (14). For the specific dairy products, a daily 200 g increment in milk intake was associated with a 7% lower risk of stroke (RR=0.93; 95% CI: 0.88- 0.98; P=0.004), with considerable heterogeneity (I^2=86). RR were 0.82 (95% CI: 0.75–0.90) in East Asian and 0.98 (95% CI: 0.95–1.01) in Western countries (median intakes 38 and 266 g/d, respectively) with less but still considerable heterogeneity within the continents. Based on a limited number of studies, high-fat milk, but not low-fat milk, was directly associated with stroke risk. By contrast, cheese intake was marginally inversely associated with stroke risk (RR=0.97, 95% CI: 0.94–1.01 per 40 g/d), which is consistent with previous findings. No association was found for yogurt, but total intake of fermented dairy products (combining 2 of the products: cheese, yogurt, or sour milk) was borderline significantly associated with a 9% (RR=0.91, 95% CI: 0.82- 1.01) lower risk of stroke per 200 g/d, with no indications of a non-linear association. A beneficial effect of cheese intake was additionally supported by another meta-analysis of prospective cohort studies, in which cheese intake was inversely associated with total CVD (RR= 0.90, 95 % CI: 0.82-0.99), CHD (RR=0.86, 95% CI: 0.77-0.96) and stroke (RR=0.90, 95 % CI: 0.84-0.97) (15). The inverse association between cheese intake and stroke is of particular interest due to the high sodium content

of cheese. Furthermore, a recent meta-analysis suggested that butter consumption was not significantly associated with CVD, CHD, or stroke, despite the high content of SFAs (16).

3.1.1.1 Intake of dairy products and risk of hypertension

The latest meta-analysis assessing the relation between dairy intake and hypertension included 9 prospective cohort studies, with 57,256 individuals and 15,367 incident hypertension cases (17). Linear inverse associations were found between intake of total dairy, low-fat dairy, and milk and incident hypertension. Low-fat dairy intake was associated with a 4% lower risk of hypertension (RR=0.96, 95% CI: 0.93- 0.99) per 200 g/d, whereas high-fat dairy intake was not associated with hypertension (RR per 200 g/d= 0.99; 95% CI: 0.95- 1.03). A significant inverse linear association was found between milk intake and incident hypertension, with a pooled RR for total milk intake per 200 ml of 0.96 (95% CI: 0.93-0.99). No associations were found between total intake of fermented dairy, cheese, or yogurt and risk of hypertension. Thus, specifically low-fat dairy products and milk in particular seems to be linked to a lower risk of hypertension.

3.1.2. Intake of dairy products and risk of type 2 diabetes

The most recent meta-analysis on dairy intake and T2D included 22 cohort studies, with 579,832 individuals and 43,118 T2D cases (18). Total dairy intake was inversely associated with T2D risk (RR=0.97 per 200 g/d increment; 95% CI: 0.95-1.00; P=0.04), with a suggestive but similarly linear inverse association for low-fat dairy intake (RR=0.96 per 200 g/d; 95% CI: 0.92-1.00; P=0.072). The inverse association was strongest for populations >60 y (RR= 0.84 per 200 g/d; 95% CI: 0.77- 0.93). When analyzed according to the type of dairy products, a non-linear strong inverse association was found between yogurt intake and T2D (at 80 g/d, RR=0.86 vs. 0 g/d; 95% CI: 0.83- 0.90; P<0.001). At higher yogurt intakes, a levelling off in risk association was evident. In addition, low-fat fermented dairy was not associated with T2D risk, but when high-fat fermented dairy was included, a significant 12% lower risk was found with an intake of 40 g/d, above which there were

no further reduction in the RR of T2D. Cheese, cream, total milk, low-fat milk, high-fat milk, and total high-fat dairy intake were not found to be associated with T2D risk. Furthermore, butter was recently suggested to be inversely associated with incidence of T2D (RR=0.96, 95% CI: 0.93-0.99; P=0.021) (16). In summary, the dairy products yogurt and butter, as well as the group of high-fat fermented dairy products, seem to be inversely associated with T2D.

3.1.3. Considerations for future observational studies on dairy and cardiometabolic risk As observational studies have just recently started to examine associations for different types of dairy products, the analyses of individual dairy products in the presented meta-analyses were based on a low number of studies. Specifically, data on yogurt and cheese intake from observational studies are limited, which is likely due to a previous focus on milk intake. The latter however still warrant further investigation considering recent findings from two Swedish cohorts reporting that high total milk intake was associated with higher mortality (19). Based on the current literature it can be recommended that future epidemiological studies provide more extensive details about the types of dairy products, including fat content, as well as details within the specific dairy group (e.g. cheese types, yogurt types). In addition, as amounts and types of dairy products consumed vary across countries and continents, heterogeneity was present in several of the presented meta-analyses. Future studies should address the differences in intake across continents e.g. by use of stratification in the statistical analyses.

The advantages of observational studies are that they generally represent large populations, examine long-term associations with disease events, and assess real-life exposure prior to the occurrence of the outcome. An important disadvantage is, however, that confounding cannot be eliminated in observational studies and therefore evidence for causality is generally weak. The associations between dairy intake and disease risk could potentially be influenced by residual confounding. For instance, overall diet quality may differ between dairy and non-dairy consumers, and also different

types of dairy may cluster within different dietary patterns. Only a few epidemiologic studies have investigated how dairy intake affects the diet quality. One study showed that low-fat dairy product intake clusters within a healthy type dietary pattern , whereas the opposite seems to apply for intake of high-fat dairy products (20). Furthermore, another study showed that yoghurt consumption was associated with an improved diet quality, including higher consumption of fruits and vegetables, whole grains, and dairy products (21). Nevertheless, the impact of different types of dairy products on diet quality may very likely be population specific.

Observational studies support distinct relations between intake of specific types of dairy foods and risk of CVD, hypertension, and T2D, but does not provide evidence of causality, hence, randomized controlled trials have to be considered. In the following sections studies are presented comparing whole dairy products to dairy constituents or to other dairy products, with or without matching the dairy constituents.

3.2. Intervention studies comparing dairy products to dairy constituents (supplements) One way of investigating whether there is a specific effect of the dairy matrix on health is to examine intervention studies comparing whole dairy food with an isolated nutrient of interest from that food, to explore if there is a difference in outcome with the whole food versus the isolated nutrient. The results from intervention studies fitting this criterion are described below.

3.2.1. Effect of whole dairy foods compared to dairy constituents on body weight and body composition

Two studies have investigated if a whole dairy food has a different effect on body weight and body composition relative to calcium or milk protein. The first study in overweight/obese premenopausal women compared the effect of cows' milk (3 servings/d), soy milk fortified with calcium (3 servings/d), a calcium carbonate supplement (800mg/d), and a control diet (no addition)

on body weight, waist and hip circumference (22). All groups consumed an energy deficit diet (500 kcal/d) for a period of 8 weeks. The greatest reduction in body weight, BMI and body fat was in the cows' milk group. Weight loss in the cows' milk group was significantly greater than the weight loss in the control or soy milk group (weight loss 5.8%, 4.3% and 3.8%, respectively) but not relative to the calcium supplement (4.8% weight loss). These results suggest a dairy matrix effect for calcium and protein contained in milk. Another study compared 12-weeks intake of skimmed milk, casein or whey protein, versus water (0.5 L/d) on body composition and leptin levels in overweight adolescents (23). Compared to water, skimmed milk and the milk proteins increased lean mass, fat mass and leptin concentration; thus indicating a dairy protein effect rather than a dairy matrix effect.

3.2.2. Effect of whole dairy foods compared to dairy constituents on markers of cardiovascular disease risk

Effects of whole dairy compared to dairy constituents on cardiovascular outcomes have also been investigated in a number of studies. Previously, it was suggested that calcium supplements were associated with an increased risk of cardiovascular disease events and mortality (24) and, hence, it was assumed that calcium-rich foods, such as dairy, would exhibit the same association. However, an acute increase in calcium intakes from dairy products (milk and low-fat yogurt) was shown to attenuate postprandial lipaemia, whereas supplementary calcium carbonate did not exert a similar effect (25). Another acute study investigated the effects of a calcium supplement (500 mg) compared to a meal with a calcium supplement or a dairy product meal, or a calcium fortified juice, on serum calcium and phosphate up to 6 hours after consumption in a randomized cross-over manner (26). The elevation in serum ionized and total calcium concentration was delayed when the supplement was consumed with a meal, but was most delayed after the dairy product meal, thus indicating a dairy matrix effect. The authors speculated that the rapid increase in serum calcium

with the calcium supplement may partly explain the contrasting cardiovascular effects of calcium supplements and calcium in dairy foods. The different responses to calcium consumed as a supplement or as a dairy product, may be due to differences in the chemical form of calcium (inorganic calcium carbonate or citrate in supplements vs. organic calcium phosphate (hydroxyapatite) in dairy products), or to interactions between calcium and other components in the dairy matrix.

The effects of whole dairy foods on blood pressure may also differ from that of a calcium supplement. One study of older date investigated the effect of 6 weeks intake of milk (1180 mg calcium, 1650 mg potassium, and 110 mg magnesium) compared to a mineral-reduced milk (95 mg calcium, 580 mg potassium, and 10 mg magnesium) in an otherwise low-calcium diet (<500 mg/d) on blood pressure (27). The mean systolic blood pressure change associated with intake of milk (-4.1%) was significantly greater than the change observed with the mineral-reduced milk (-1.3%). This indicates a small hypotensive effect of milk intake, which was at least partly explained by its mineral content. More recently, a randomized double-blind crossover study investigated the blood pressure lowering effect of Grana Padano cheese (30g/d) in comparison to a placebo (consisting of flavored bread mixed with fats and salts in concentrations equal to those of Grana Padano cheese), in a 2-months cross-over study in mildly hypertensive subjects (28). Intake of the Grana Padano cheese resulted in a significant decrease in systolic and diastolic blood pressure compared to placebo. Therefore, SFAs and salt may have less effect on blood pressure when present in the cheese matrix, or other components in the cheese matrix (e.g. bioactive peptides) may have counteracted their effect.

3.2.3. Effect of whole dairy foods compared to dairy constituents on bone health

The importance of the dairy matrix on bone health has been studied in two trials comparing whole dairy products to dairy nutrients. Fermented dairy products may have a distinct positive effect on

mineral and bone balance, as in addition to the supply of calcium, phosphorus, and protein they also contain probiotic bacteria. The latter may alter the gut microbiota, and enhance the capacity to translocate calcium across the intestinal epithelium (29). A two-year randomized controlled trial examined the effects of calcium or calcium + vitamin D supplements, or a dairy product (cheese) on bone mass accrual and body composition in 10-12 year old girls with low habitual calcium intake (30). The cheese supplementation resulted in a higher percentage change in cortical thickness of the tibia compared to placebo, or calcium supplementation, or calcium + vitamin D supplementation. A per-protocol analysis found that cheese supplementation resulted in a higher whole-body bone mineral density (BMD) in those with >50% compliance. Thus, cheese had a superior effect on bone mass accrual than calcium or calcium + vitamin D supplementation. Another randomized controlled trial investigated the effect of dairy products or a calcium supplement, both providing 1200mg calcium/d, on markers of bone metabolism and BMD relative to a control group (31). After 12 months, the dairy group had greater improvements in pelvis, spine and total BMD than the calcium supplement group and the control group. Both trials support a dairy matrix effect on bone health.

3.2.4. Partial conclusion on dairy products compared to dairy constituents (supplements)

Studies to date examining the effect of whole dairy foods compared to single dairy constituents focusing on body weight, CVD risk or bone health provide indications that whole dairy products have a more beneficial effect on health compared to the single dairy constituents. This supports the concept that dairy products should not be considered as equal to a few nutrients, but as a function of the total nutrient content within the natural dairy structure. This proposition is worthy of investigation in future trials.

3.3. Intervention studies comparing effects of different dairy products on blood lipids, controlling for dietary factors

Different types of dairy products may affect risk markers of disease differently, and this could potentially be explained by additive effects of the active components contained in the dairy products. Comparison of one type of dairy product along with intake of specific dairy components to resemble the composition of another dairy product was investigated in two studies. Tholstrup et al. (2004) compared cheese, milk and butter in whole diets all balanced for the amount of fat (from cheese, milk and butter) and for the content of protein (80% casein and 20% whey protein) and lactose, but not calcium (32). No significant differences in blood lipids were found between cheese and milk. However, despite the addition of lactose and milk protein to the butter diet, butter caused significantly higher LDL-cholesterol and borderline higher total cholesterol concentrations compared to cheese whereas milk was intermediate and not significantly different to cheese and butter. Likewise, another study compared cheese to butter + casein and found a lower total cholesterol and a borderline lower LDL-cholesterol concentration after the cheese diet compared to the butter + casein diet (33). These studies suggest that protein and lactose are not the determinants of the difference between cheese and butter on blood lipids.

3.4. Intervention studies comparing effects of different dairy products on blood lipids, without controlling for dietary factors

Different types of dairy products as whole foods have also been compared in a number of intervention studies, illustrating the impact of these foods in the actual form they are consumed by the population.

Several studies with free-living or highly controlled full-diet designs, have shown that intake of cheese results in a lower LDL-cholesterol compared to intake of butter (2,34,35). These findings were substantiated by a recent meta-analysis of randomized controlled trials (36) (**Figure 1**). The trials suggest that fat consumed in isolation (butter) has a different effect to fat delivered in the cheese matrix, and supports a dairy matrix effect. One of the intervention studies also proposed that

cheese did not increase LDL-cholesterol concentration compared to a habitual diet with a lower saturated fat content (2). One of the intervention studies in addition to butter and cheese also included a milk comparison (34). In this study, cheese and milk were consumed in amounts providing similar amounts of fat, protein, and calcium. Compared to the butter-control diet, in which protein and fat, but not calcium, were balanced to that of the cheese and milk diets, both cheese and milk attenuated the increase in LDL-cholesterol compared to butter. This supports an impact of dairy calcium on LDL-cholesterol response, whereas fermentation does not seem to be involved because cheese is fermented but milk is not. It may be important that the fat and calcium are embedded in the same food matrix, and that a similar effect cannot be achieved by simply adding calcium to a diet with butter. A study in pigs showed that regular-fat cheese has a HDL-cholesterol increasing effect compared butter, whereas this effect was not found with a reduced-fat cheese + butter matching the milk-fat content of the regular-fat cheese (37). A tendency to an increased HDL-cholesterol by regular-fat cheese compared to reduced-fat cheese, without adjustments for fat or energy intake, was also shown in a larger study in humans (38). However, there was no difference in LDL-cholesterol concentration after intake of the regular- and reduced-fat cheese, despite a higher saturated fat intake with the regular-fat cheese, suggesting a moderation of the saturated fat within the regular-fat cheese matrix.

The effect of cheese and full-fat yogurt supplementation on blood lipids was also compared in a large parallel multi-center study (39). Despite slightly higher energy and calcium intakes with the yogurt compared to the cheese, the two dairy products did not affect the blood lipid profile differently.

Furthermore, a study compared the effect of 2-weeks intake of buttermilk (rich in MFGM), or of a larger intake of skimmed milk (providing similar amount of fat as the buttermilk), with intake of butter (low in MFGM) on blood lipid response (40). Although only few details were given, authors

reported that a smaller intake of buttermilk was equally efficient as a larger intake of skimmed milk in lowering total cholesterol, while butter intake increased total cholesterol. A more recent study investigated 12-weeks intake of traditionally produced buttermilk (rich in MFGM) compared to skimmed milk (low in MFGM) in diets supplemented with or without lutein-enriched egg yolk. Egg yolk consumption significantly increased total cholesterol and LDL-cholesterol concentrations. Buttermilk consumption did not prevent the total and LDL-cholesterol raising effect of egg yolk consumption compared to skimmed milk consumption, although there was a tendency to a lower total-cholesterol (41). In the two studies presented, calcium, found in both buttermilk and skimmed milk, may have attenuated the blood lipid response after consumption, whereas this would not apply to butter consumption. Hence, the dairy matrix may determine the impact size of the MFGM content. Nevertheless, the studies suggest a potential role of the MFGM in regulation of blood cholesterol balance.

3.5. How dairy matrix components interfere with the assumed effect of saturated fat on lipid metabolism

Calcium, phosphorus, MFGM, and starter cultures (in the fermented dairy types), are all dairy constituents suggested to contribute to modify blood lipid response to SFA intake. The blood lipid response is presumably attenuated by decreasing intestinal fat absorption and bile acid recycling or by modulation of gut microbiota or alteration of gene expression.

3.5.1. Lowering of fat digestibility by dairy constituents

A reduced intestinal absorption of fat from dairy products rich in calcium, phosphorus, and MFGM (such as cheese) has been demonstrated by an increased fat excretion in feces (34,42–44) and a reduced plasma chylomicron TAG concentration in the postprandial state (25). Differences within the dairy matrix may, however, influence this; a study in pigs showed a higher fecal energy excretion on a diet with regular-fat cheese compared to a diet with butter, whereas a diet with

reduced-fat cheese + butter caused a non-significant intermediate response (37). Furthermore, a study in humans examining equally high intakes of reduced-fat and regular-fat cheese, found no difference in body weight between groups after 3 months intake, despite a higher energy intake with the regular-fat cheese (38). These studies are suggestive of an enhanced fecal fat excretion with regular-fat cheese, thus supporting a dairy matrix effect.

3.5.1.1. Lowering of fat digestibility by calcium and phosphate

Two mechanisms for calcium, underlying a reduced intestinal fat absorption after dairy intake, have been suggested. The first mechanism is precipitation of calcium and free fatty acids as largely insoluble calcium-fatty acid soaps. The second mechanism is precipitation of calcium and phosphate in insoluble amorphous calcium phosphate, which adsorb bile acids and possibly also fatty acids to the surface and hence increases fecal bile acid and fat excretion (45–48). In vitro and in vivo, saponification was suggested to increase with increasing fatty acid saturation and chain length (45,49). However, a study in humans showed significantly higher fecal excretion of all fatty acid groups (SFA, MUFA, and PUFA) on a high-calcium diet (with low-fat dairy) compared to a low-calcium diet (50). Also, when fecal excretions of fatty acids were expressed as percentages of intake, the difference between diets was greater for MUFA than for SFA. The fecal excretion of fatty acids probably depends on the location of the fatty acids on the glycerol backbone, as fatty acids in the sn-1 and sn-3 position are released by lipase (51) and are, therefore, more susceptible to binding by calcium than fatty acids in the sn-2 position. The increase in fecal fat excretion by dairy calcium has been shown in several (34,43,50,52–54), but not all studies (2,55). A study by Weaver et al. (2011) found that the change in fecal calcium excretion (by dairy products or a supplement) predicted the change in fecal fat excretion as the fraction of the intake. However, this did not affect energy balance (55), which was believed to be influenced by excretion of calcium-fatty acid soaps mostly in infants (56).

The methods used for extracting fat vary between studies and may result in more or less absolute fat extraction from the feces samples. Also, it is important that fecal fat excretion is expressed as absolute amount (g/d) instead of concentration in feces, because feces volume varies within and between subjects. In addition, the studies which have chemically characterized the chemical form of fecal calcium are few and of older date. Hence, there is a need for new in vivo studies with chemical characterization of the calcium-fatty acid soaps and amorphous calcium phosphate compounds, preferably focusing on the ratios of which fat, calcium, and phosphate appear in whole dairy foods.

3.5.1.2. Lowering of cholesterol absorption by the MFGM

A study in mice has proposed a mechanism for the MFGM reducing intestinal cholesterol absorption after dairy intake based on inhibition of cholesterol micellar solubility (57). It was demonstrated in vitro that in the presence of buttermilk, the micellar solubility of cholesterol was reduced, probably due to the presence of sphingomyelin in buttermilk MFGM fragments (58). Because sphingomyelin is not completely hydrolyzed in the human small intestine, such sphingomyelin-cholesterol complexes suggests a potential cholesterol absorption limiting ability of the MFGM (59), however, such an effect was not confirmed in a study in humans measuring surrogate markers of intestinal fat absorption (60).

3.5.2. Link between fat digestibility and blood lipid response

There is evidence of a connection between an increased fecal fat excretion and an attenuated blood lipid response after intake of dairy products rich in calcium, phosphate and MFGM. An intervention study in humans showed a strong correlation between fecal fat excretion and changes in total and LDL-cholesterol after intake of diets with cheese or milk compared to a diet with butter (**Figure 2**) (34). Also, a cross-over study in humans, examining four full-diets with combinations of low-calcium or high-calcium and low-fat or high-fat, found that the response in LDL- and HDL-cholesterol depended on the saturated fat content of the diet, and that only the LDL-cholesterol

response was attenuated by a simultaneous high dietary calcium intake (61). As the phosphate content of the diets in this study were also high on the high-calcium diets and low on the low-calcium diets, phosphate may have contributed to the attenuation of LDL-cholesterol. Calcium phosphate was previously shown to increase fecal bile acid excretion and reduce LDL-cholesterol (62). This effect was presumably due to a decreased enterohepatic bile acid circulation and a consequently increased hepatic de novo bile acids synthesis from its precursor, cholesterol. This may have led to an upregulation of the hepatic LDL-receptor expression and an increased clearance of LDL-cholesterol from the circulation. Hence, amorphous calcium phosphate may influence blood lipids through fecal bile acid excretion.

3.5.3. Regulation of blood lipid response by the MFGM and milk-phospholipids

It is still unclear whether or not the MFGM (intact around the native milk fat globules or as free released fragments) affects the fecal fat excretion in humans. Some studies suggest that the MFGM could reduce or prevent an increase in fasting total cholesterol, LDL-cholesterol and triglycerides (TG) caused by SFA intake (60,63,64). In one study, a buttermilk-powder formulation (rich in MFGM fragments) significantly reduced fasting total cholesterol and TG compared with a placebo powder formulation, whereas the reduction in LDL-cholesterol was only borderline significant, and strongest in subjects with a high baseline LDL-cholesterol concentration (60). The changes in concentrations of LDL-cholesterol and TGs were not correlated, suggesting different metabolic pathways of regulation. In addition, the buttermilk formula was found to increase plasma lathosterol concentrations, a surrogate marker of endogenous cholesterol synthesis. In another randomized iso-energetic study, 8-weeks intake of whipping cream (rich in intact MFGM) did not raise plasma cholesterol in overweight adults compared to intake of butter oil (emulsified fat depleted of MFGM) which markedly raised LDL-cholesterol (63). Moreover, whipping cream intake differentially regulated nineteen genes and most of the changes in gene expression were correlated with the

changes in blood lipids. The effects of formulated drinks enriched with milk-phospholipids as MFGM concentrates from buttermilk, compared with other phospholipid sources on blood lipids have also been investigated in two studies. A parallel study tested a drink formulated from buttermilk concentrate, hereby enriched in sphingolipids (25% of MFGM polar lipids in cows' milk), compared to a drink formulated with skimmed milk enriched with egg-phospholipid and butter oil (64). No significant difference in blood lipids was found between groups. Another study, included two trials investigating 8 weeks intake of milk-phospholipids (2g) compared to milk-fat and to soya-phospholipids on lipid metabolism and other risk factors for CVD (65). In the first trial, subjects consumed milk enriched with either 2 g milk-phospholipids from a buttermilk concentrate or 2 g milk-fat for 8 weeks. In the second trial, subjects consumed milk enriched with either 3 g milk-phospholipids from the same buttermilk concentrate or 2.8 g soya-phospholipids for 7 weeks. Milk-phospholipid did not affect plasma lipids, insulin sensitivity, or inflammatory markers in the two trials. In summary, based on these few studies, the effects of formulated drinks enriched with MFGM through buttermilk concentrate appears to be less pronounced than the effect of intact MFGM, provided by buttermilk, cream, or cheese, which supports a stronger effect of whole dairy i.e. the dairy matrix compared to its isolated or reformulated constituents. The mechanisms induced by the MFGM are unclear and deserves further investigation, but might be associated with a decrease in intestinal cholesterol absorption and/or the regulation of expression of genes.

3.5.4. Regulation of blood lipid response by dairy matrix fermentation

Intake of fermented dairy products has been shown to have a beneficial effect on blood lipid concentrations. This may be due to fermented dairy products favoring a gut microbiota with a specific production of SCFA (66). The SCFAs are rapidly absorbed by the colon and metabolized in the liver and an altered production ratio of SCFAs may influence the blood lipid balance, as serum acetate: propionate ratio has been associated with serum total and LDL-cholesterol concentrations

(67–69). A metabolomics investigation of a study comparing milk, cheese or butter intake showed that both cheese and milk intake increased levels of fecal SCFAs (70). Specifically, cheese intake increased fecal butyrate, propionate, and malonate concentrations and decreased fecal acetate and glycerol concentrations. Furthermore, there were significant correlations between fecal propionate and butyrate levels and LDL-cholesterol concentrations. This indicates that dairy consumption and especially cheese, increases fecal SCFA levels or alters production ratio of these, and this is likely a result of modification of the gut microbiota. Low- and high-fat fermented dairy may not have a similar impact on gut microbiota. A previous study in pigs pointed towards distinct effects of reduced-fat cheese and regular-fat cheese on gut microbiota in diets with equal milk-fat (37). Regular-fat cheese caused a lower Firmicutes to Bacteroidetes ratio, and compared to a butter-control the changes in the relative abundance of more bacterial genera were found after intake of the regular-fat cheese e.g. a higher abundance of Lactobacillus and Oscillibacter.

3.6. Studies comparing similar dairy products with different structures or textures on digestion and absorption kinetics, appetite sensations and muscle protein synthesis

In addition to the nutrient matrix, the physical structures and textures of dairy products could influence the health effects of different types of dairy foods. Only a few studies have compared different structures or textures of dairy products in relation to absorption kinetics and appetite. A study investigated the effects of a semi-solid yogurt (378 g), drinkable yogurt (378 g), dairy beverage (400 mL), and fruit drink (400 mL), matched for palatability and energy content, on subjective appetite sensations (71). The semi-solid and liquid yogurts resulted in reduced hunger and increased feelings of fullness compared with the dairy beverage and fruit drink. However, subsequent food intake did not differ after intake of the beverages. Another study, comparing two iso-energetic meals with a liquid structure or a semi-solid structure, found a longer gastric emptying time and a prolonged satiety response after the semi-solid meal (grated cheese and low-fat yogurt)

(6). These studies suggest a prolonged satiety response after intake of a semi-solid dairy matrix and yogurt compared to a liquid dairy matrix.

Acid and rennet gels of dairy products with identical composition have been shown to exhibit major differences in the protein digestion kinetics and amino acid bioavailability in pigs (72). A rennet gel produced with heat-treated milk showed prolonged residence time in the stomach compared to an acid gel or a stirred acid gel. Also, the rennet gel caused slower release of milk proteins in the duodenum. A delayed gastric emptying was also seen after intake of yogurt compared to intake of milk (73), and yogurt was shown to lower and prolong the jejunal release of protein (74). Furthermore, different dairy matrix structures were found to impact the number of identified peptides released, with rennet gels inducing two and three times less peptides than milk and acid gels, respectively (75). This is consistent with the delayed protein digestion kinetics /amino acid absorption observed with rennet gels.

The dairy proteins, i.e. casein and whey are found in different ratios and levels in dairy foods (Table 2) and these proteins were shown to cause a distinct postprandial release of peptides. Medium-size peptides (750–1050 kDa) were released during 6h after casein ingestion, whereas larger peptides (1050–1800 kDa) were released during the first 3h after whey protein ingestion (76). In the jejunum, twice as many peptides were detected and sequenced after casein ingestion compared with whey protein ingestion, and accordingly -casein was shown to be the most important precursor of peptides. An acute study in humans, in which the protein ratio in the yogurt matrix was manipulated towards a higher whey protein content, decreased subsequent ad libitum energy intake compared to a yogurt with lower whey and higher casein content (77). Also, a study in a murine model revealed a lower body weight gain for mice fed whey protein as compared with casein (78). Metabolomics studies in both animals and humans suggest this to be a result of whey protein affecting endogenous metabolism through the Krebs cycle (78,79). Therefore, the balance between casein and whey

proteins in dairy products may be important for their overall effect on body weight, through mechanisms involving appetite regulation and endogenous energy metabolism. A high protein digestion rate is necessary in the elderly to induce protein metabolism and prevent age-related sarcopenia, because of an age-related higher threshold for inducement of an anabolic stimulus by plasma amino acids (80). The anabolic threshold is lower among younger adults, and therefore the protein kinetics may be less important in this population group, with the exception of elite athletes who have higher substrate requirements. Even small differences in protein digestion rate, may be sufficient to affect postprandial protein metabolism (81). For dairy products, pasteurization (72˚C for 20 seconds) does not seem to affect protein digestion rate, whereas ultra-high temperature (UHT) treatment (140˚C for 5 seconds) increased digestion rate (82). The latter is probably explained by heat-induced interactions with the whey protein (mainly -lactoglobulin) with subsequent partial dissociation of the casein micelles.

Homogenization strongly modifies the structure of milk fat globules, and therefore the metabolic impact of consumption of homogenized dairy products has been questioned (83). The effect of homogenization on the lipid absorption kinetics has not yet been investigated in vivo, but was investigated in vitro using nutrient-matched formulae; one based on raw milk and one based on homogenized milk (84). An increased postprandial release of free fatty acids during in vitro gastric digestion was observed with the homogenized formula. This was explained with the homogenized formula having a smaller lipid particle size with increased relative surface area, making the lipids more accessible for lipases. Finally, differences in fat droplet interface composition (proteins vs MFGM) was also suggested to impact the digestion kinetics of differently processed milk (85).

In summary, the micro- and macro-structures of the dairy matrix may influence the metabolic response after consumption. Therefore, different dairy matrix structures should be taken into consideration, in addition to the content of bioactive components, when evaluating the nutritional

properties of whole dairy foods. As different dairy structures may provide a better fit for the dietary needs of individual population groups, this should be further investigated and included when re-evaluating current dietary guidelines. For instance, elite athletes and the elderly may be given the advantage of a high and efficient nutrient release rate whereas obese or diabetic persons may benefit from a lower nutrient release rate.

3.7. How the matrix of fermented dairy may affect insulin sensitivity

It has been suggested that different protein sources (i.e. dairy, meat, fish, egg, and plants) are differently associated with risk of T2D (86). Among dairy products, yogurt and cheese in particular are associated with lower risk of T2D (18,87). This suggests that in addition to the protein and mineral content, other bioactive factors in fermented dairy products may influence T2D risk. Ripened dairy products contain branched chain amino acids and milk-derived peptides. Aside from the antihypertensive effect of milk-derived peptides, these may also be involved in regulation of insulinaemia and in stimulation of the satiety response (88). The bioactive peptides or amino acids produced during cheese ripening were recently shown to improve insulin sensitivity and reduce circulating free fatty acids in pigs (52). In humans, postprandial increments in branched chain amino acids from whey (i.e. leucine, valine, and isoleucine) was found to correlate with postprandial insulin response, and insulin responses to correlate with glucose-dependent insulinotropic polypeptide (GIP) concentrations (89). Furthermore, a cross-over study showed that 6 months intake of 4 daily servings of low-fat milk and yogurt reduced fasting plasma insulin by 9% and reduced insulin resistance by 11% in overweight and obese adults compared to a 6-month low-dairy control period (90).

Finally, stimulation of a beneficial gut microbiota, after intake of probiotics, has been suggested to modulate gut function through regulation of the immune system (91). This has also been

Owing to a low number of studies in humans in this field, more research on fermented dairy products and glucose homeostasis are needed.

4. CONCLUSIONS

Evidence to date indicates the dairy matrix has specific beneficial effects on health, as the metabolic impact of whole dairy on body weight, cardiometabolic disease risk, and bone health differ to that of single dairy constituents. Also, different types of dairy product types seem to be distinctly linked to various health effects and disease risk markers. In addition, different processing methods and dairy structures can enhance interactions in the dairy matrix, thereby modifying the metabolic effects of dairy consumption. The nutritional value of dairy products should therefore be considered as the biofunctionality of the sum of nutrients within the dairy matrix structures. Hence, there is a need for further research on health effects of whole dairy foods alongside the more traditional approach of studying the health effects of single nutrients. Such research would help to support dietary guidelines considering the effect of whole foods on health, rather than just focusing on a few individual nutrients within a food.

5. ACKNOWLEDGEMENTS

Authors' contributions to the manuscript: AA and IG designed research; TKT wrote the paper; all authors supplied valuable knowledge and scientific consultation throughout the manuscript preparation, IG had primary responsibility for final content. All authors read and approved the final content of the manuscript.

Conflicts of interest: HCB: has received research funding from Arla foods and Danish Dairy Research Foundation. JPB: is clinical research consultant for Yoplait France-General Mills USA. Member of The Scientific Committee of Candia, France. DD: has received funding from Lactalis. EF: has received honoraria and speaking honorariums from the National Dairy Council in Ireland.

574 Her research position in niversity College Dublin is funded by Food For Health Ireland, which
575 receives a portion of its funding from partners within the Irish dairy industry. RI: has received
576 funding as principal investigator or participant from the Danish Research Foundation for a number of
577 projects over the last 25 years. He has also collaborated extensively with the Danish dairy and
578 ingredient industry on projects funded partially by public funds and partially from industry. JML: is
579 President of the Scientific Advisory Board of FICT (Fédération Industries Charcuteries Traiteur) and
580 of GROS (Groupe de Réflexion Obésité Surpoids) and is a member of the Scientific Advisory Board
581 of GEMO (Groupe d'Expert en Micro Nutrition Oculaire), of APRIFEL (Agence pour la Recherche
582 et l'Information en Fruits et Légumes, and of ENSA (European Natural Soy and Plant Based Foods
583 Manufacturers Association), and he is a member of OCHA (Observatoire CNIEL des habitudes
584 alimentaires). He acts as an expert for Lactalis, Yoplait and Danone; is a member of OCHA
585 (Observatoire CNIEL des habitudes alimentaires), and has received funding for conferences for EMF
586 (European Milk forum). AM: has received funding from Arla Foods UK, Unilever plc, Dairy
587 Australia, Teagasc (Agriculture and Food Development Authority Ireland) and UK BBSRC; he is on
588 the editorial board of Food Research International. MM: has received speaking honoraria from the
589 Dairy Council for Northern Ireland and the European Milk Forum. MCM: has received research
590 funding from INRA (National Institute for Agricultural Research), ANR (National Research
591 Agency), SFD (French-speaking Society for Diabetes), Institut Carnot LISA (Lipids for Industry,
592 Safety and Health), SFN (French Society for Nutrition), Institut Benjamin Delessert
593 (public/academic/not-for-profit organizations) and from CNIEL (French Dairy Interbranch Sector),
594 Sodiaal-Candia R&D, Nutricia Research; she acts as a consultant for a number of different food
595 companies and the dairy sector; she is a member of the Scientific Committee of ITERG (Fats and
596 Oils Institute). UR: has received funding from a governmental grant (TvärLivs, 2012 to 2014, “role
597 of MFGM in dairy foods”) from the Swedish Research Council Formas that was also co-funded by

several food companies including Skånemejerier AB, Semper AB, Arla Foods Amba, and Dairy Sweden. SSM: has received unrestricted grants from several sponsors i.e. from Dutch Dairy Association for a meta-analysis dairy and stroke; from Global Dairy Platform, Dairy Research Institute and Dairy Australia for a meta-analysis dairy products and all-cause mortality, and for a meta-analysis on the effects of cheese on lipids. She received an international prize, i.e. The Wiebe Visser International Dairy Nutrition Prize from the Dutch Dairy Association's (NZO) Utrecht Group. The sponsors had no role in design and conduct of the study, data collection and analysis, interpretation of the data, decision to publish, or preparation of any of the manuscripts. TT: has received unrestricted grants from the Danish Dairy Board, Denmark; and from the Dairy Research Institute, Rosemont, IL. CW: has received funding from Tate & Lyle, Alliance of Potato Research and Education; she is an Advisory board Member for Pharmavite and International Life Science Institute; she receives honorarium from Tate & Lyle; she is on the Editorial Board of Nutrition Research Reviews. AA: has received research funding from: Arla Foods, DK; Danish Dairy Research Foundation; Global Dairy Platform; Danish Agriculture & Food Council. Advisory boards: Dutch Beer Knowledge Institute, NL; IKEA, SV; Lucozade Ribena Suntory Ltd, UK; McCain Foods Limited, USA; McDonald's, USA; Weight Watchers, USA. Consultant for: Nestlé Research Center, Switzerland; Nongfu Spring Water, China. Expert witness: Lantbrukarnas Riksförbund Mjölk, Stockholm 2015. Speaker for: LRF Mjölk, Gothenberg 2016; Global Dairy Platform, Berlin 2015; Dairy Council UK, Glasgow & Cardiff 2015; European Milk Forum, Dairy Councils of Republic of Ireland and Northern, Dublin & Belfast, 2014; European Milk Forum, Paris, 2014. Editorial boards: Associate Editor of American Journal of Clinical Nutrition, board member Annals of Nutrition and of Metabolism & Annual Review of Nutrition. Recipient of expenses and/or modest honoraria (<$2,000) for lectures given at meetings supported by corporate sponsors. IG: British Nutrition Foundation Scientific Advisory Committee Member; UK Food

Standards Agency, member of Advisory Committee; University College Dublin Institute of Food and Health, Scientific Advisory Panel Member; Estonian Biocompetance Centre of Healthy Dairy Products. Consultant to Scientific Panel; International Chair on Cardiometabolic risk, Member, Panel on Dairy/Health/Lipids; International Expert Movement to improve Dietary Fat Quality, Member; University of Aberystwyth, Research Assessment Panel for IBERS; European Healthy Lifestyle Alliance: Member of Discussion Panel on Obesity; International Expert Movement to Improve Dietary Fat Quality, Member; Consultant to the Dairy Council on Fats in Dairy Products and Cardiometabolic Disease; Co-operating with Universities of Copenhagen and Wageningen on Global Dairy Platform funded meta-analysis on dairy products and cardiometabolic disease; Recent/current dairy and health research funding from, UK BBSRC (Biotechnology and Biological Sciences Research Council), UK MRC Medical Research Council), Dairy Council, Agriculture and Horticulture Development Board Dairy (AHDB Dairy), The Barham Foundation Trust, various companies. LDG, DR, and TKT: have no conflicts of interest to declare.

6. REFERENCES

1. Fardet A, Rock E. Toward a new philosophy of preventive nutrition: from a reductionist to a holistic paradigm to improve nutritional recommendations. Advances in nutrition (Bethesda, Md). American Society for Nutrition; 2014;5:430–46.

2. Hjerpsted J, Leedo E, Tholstrup T. Cheese intake in large amounts lowers LDL-cholesterol concentrations compared with butter intake of equal fat content. The American Journal of Clinical Nutrition. 2011;94:1479–84.

3. Grundy MM, Grassby T, Mandalari G, Waldron KW, Butterworth PJ, Berry SE, Ellis PR. Effect of mastication on lipid bioaccessibility of almonds in a randomized human study and its implications for digestion kinetics, metabolizable energy, and postprandial lipemia. The American Journal of Clinical Nutrition. 2015;101:25–33.

4. Tydeman EA, Parker ML, Faulks RM, Cross KL, Fillery-Travis A, Gidley MJ, Rich GT, Waldron KW. Effect of carrot (Daucus carota) microstructure on carotene bioaccessibility in the upper gastrointestinal tract. 2. In vivo digestions. Journal of Agricultural and Food Chemistry. 2010;58:9855–60.

5. Hull S, Re R, Chambers L, Echaniz A, Wickham MSJ. A mid-morning snack of almonds generates satiety and appropriate adjustment of subsequent food intake in healthy women. European journal of nutrition. 2015;54:803–10.

6. Mackie AR, Rafiee H, Malcolm P, Salt L, van Aken G. Specific food structures supress appetite through reduced gastric emptying rate. American journal of physiologyGastrointestinal and liver physiology. 2013;304:G1038-43.

7. Vors C, Pineau G, Gabert L, Drai J, Louche-Pélissier C, Defoort C, Lairon D, Désage M, Danthine S, Lambert-Porcheron S, et al. Modulating absorption and postprandial handling of dietary fatty acids by structuring fat in the meal: a randomized crossover clinical trial. The American journal of clinical nutrition. 2013;97:23–36.

8. Drewnowski A, Fulgoni 3rd V. Nutrient profiling of foods: creating a nutrient-rich food index. Nutrition reviews. 2008;66:23–39.

9. Feeney EL, Nugent AP, Mc Nulty B, Walton J, Flynn A, Gibney ER. An overview of the contribution of dairy and cheese intakes to nutrient intakes in the Irish diet: results from the National Adult Nutrition Survey. The British journal of nutrition. 2016;115:709–17.

10. Millen BE, Abrams S, Adams-Campbell L, Anderson CA, Brenna JT, Campbell WW, Clinton S, Hu F, Nelson M, Neuhouser ML, et al. The 2015 Dietary Guidelines Advisory Committee Scientific Report: Development and Major Conclusions. Advances in nutrition. 2016;7:438–44.

11. Dewettinck K, Rombaut R, Thienpont N, Le TT, Messens K, Van Camp J. Nutritional and technological aspects of milk fat globule membrane material. International Dairy Journal. 2008;18:436–57.

12. Qin LQ, Xu JY, Han SF, Zhang ZL, Zhao YY, Szeto IM. Dairy consumption and risk of cardiovascular disease: an updated meta-analysis of prospective cohort studies. Asia Pacific

Journal of Clinical Nutrition. 2015;24:90–100.

13. Alexander DD, Bylsma LC, Vargas AJ, Cohen SS, Doucette A, Mohamed M, Irvin SR, Miller PE, Watson H, Fryzek JP. Dairy consumption and CVD: a systematic review and meta-analysis. The British journal of nutrition. 2016;115:737–50.

14. de Goede J, Soedamah-Muthu SS, Pan A, Gijsbers L, Geleijnse JM. Dairy Consumption and Risk of Stroke: A Systematic Review and Updated Dose-Response Meta-Analysis of Prospective Cohort Studies. Journal of the American Heart Association. 2016;5:1–22.

15. Chen GC, Wang Y, Tong X, Szeto IM, Smit G, Li ZN, Qin LQ. Cheese consumption and risk of cardiovascular disease: a meta-analysis of prospective studies. European journal of nutrition. 2016;1–11.

16. Pimpin L, Wu JHY, Haskelberg H, Del Gobbo L, Mozaffarian D. Is Butter Back? A Systematic Review and Meta-Analysis of Butter Consumption and Risk of Cardiovascular Disease, Diabetes, and Total Mortality. PLOS ONE. 2016;11:e0158118.

17. Soedamah-Muthu SS, Verberne LD, Ding EL, Engberink MF, Geleijnse JM. Dairy consumption and incidence of hypertension: a dose-response meta-analysis of prospective cohort studies. Hypertension. 2012;60:1131–7.

18. Gijsbers L, Ding EL, Malik VS, de Goede J, Geleijnse JM, Soedamah-Muthu SS. Consumption of dairy foods and diabetes incidence: a dose-response meta-analysis of observational studies. The American Journal of Clinical Nutrition. 2016;103:1111–24.

19. Michaelsson K, Wolk A, Langenskiold S, Basu S, Warensjo Lemming E, Melhus H, Byberg L. Milk intake and risk of mortality and fractures in women and men: cohort studies. BMJ (Clinical research ed). 2014;349:g6015.

20. Mursu J, Steffen LM, Meyer KA, Duprez D, Jacobs Jr DR. Diet quality indexes and mortality in postmenopausal women: the Iowa Women's Health Study. The American Journal of Clinical Nutrition. Divisions of Epidemiology and Community Health, University of Minnesota, Minneapolis, MN 55454, USA.; 2013;98:444–53.

21. Panahi S, Tremblay A. The Potential Role of Yogurt in Weight Management and Prevention of Type 2 Diabetes. Journal of the American College of Nutrition. a Department of Kinesiology , Universite Laval , Quebec City , Quebec , CANADA.; a Department of Kinesiology , Universite Laval , Quebec City , Quebec , CANADA.; b Centre de Recherche de l`Institut de Cardiologie et de Pneumologie de Quebec , Quebec City ; 2016;1–15.

22. Faghih S, Abadi AR, Hedayati M, Kimiagar SM. Comparison of the effects of cows' milk, fortified soy milk, and calcium supplement on weight and fat loss in premenopausal overweight and obese women. Nutrition, metabolism, and cardiovascular diseases: NMCD. 2011;21:499–503.

23. Larnkjaer A, Arnberg K, Michaelsen KF, Jensen SM, Molgaard C. Effect of increased intake of skimmed milk, casein, whey or water on body composition and leptin in overweight adolescents: a randomized trial. Pediatric obesity. World Obesity; 2015;10:461–7.

24. Bolland MJ, Avenell A, Baron JA, Grey A, MacLennan GS, Gamble GD, Reid IR. Effect of

calcium supplements on risk of myocardial infarction and cardiovascular events: meta-analysis. BMJ (Clinical research ed). 2010;341:c3691.

25. Lorenzen JK, Nielsen S, Holst JJ, Tetens I, Rehfeld JF, Astrup A. Effect of dairy calcium or supplementary calcium intake on postprandial fat metabolism, appetite, and subsequent energy intake. The American Journal of Clinical Nutrition. 2007;85:678–87.

26. Bristow SM, Gamble GD, Stewart A, Kalluru R, Horne AM, Reid IR. Acute effects of calcium citrate with or without a meal, calcium-fortified juice and a dairy product meal on serum calcium and phosphate: a randomised cross-over trial. The British journal of nutrition. 2015;113:1585–94.

27. Van Beresteijn EC, van Schaik M, Schaafsma G. Milk: does it affect blood pressure? A controlled intervention study. Journal of internal medicine. 1990;228:477–82.

28. Crippa G, Zabzuni D, Bravi E, Rossi F. Os 04-04 Antihypertensive Effect of Milk-Derivative Tripeptides. Randomized, Double-Blind, Placebo-Controlled Study on the Effects of Grana Padano Cheese Dop in Hypertensive Patients. Journal of hypertension. 2016;34 Suppl 1:e55-6.

29. McCabe L, Britton RA, Parameswaran N. Prebiotic and Probiotic Regulation of Bone Health: Role of the Intestine and its Microbiome. Current Osteoporosis Reports. 2015;13:363–71.

30. Cheng S, Lyytikainen A, Kroger H, Lamberg-Allardt C, Alen M, Koistinen A, Wang QJ, Suuriniemi M, Suominen H, Mahonen A, et al. Effects of calcium, dairy product, and vitamin D supplementation on bone mass accrual and body composition in 10-12-y-old girls: a 2-y randomized trial. The American Journal of Clinical Nutrition. 2005;82:1115–8.

31. Manios Y, Moschonis G, Trovas G, Lyritis GP. Changes in biochemical indexes of bone metabolism and bone mineral density after a 12-mo dietary intervention program: the Postmenopausal Health Study. The American Journal of Clinical Nutrition. 2007;86:781–9.

32. Tholstrup T, Hoy CE, Andersen LN, Christensen RD, Sandstrom B. Does fat in milk, butter and cheese affect blood lipids and cholesterol differently? Journal of the American College of Nutrition. 2004;23:169–76.

33. Biong AS, Muller H, Seljeflot I, Veierod MB, Pedersen JI. A comparison of the effects of cheese and butter on serum lipids, haemostatic variables and homocysteine. The British journal of nutrition. 2004;92:791–7.

34. Soerensen KV, Thorning TK, Astrup A, Kristensen M, Lorenzen JK. Effect of dairy calcium from cheese and milk on fecal fat excretion, blood lipids, and appetite in young men. American Journal of Clinical Nutrition. 2014;99:984–91.

35. Nestel PJ, Chronopulos A, Cehun M. Dairy fat in cheese raises LDL cholesterol less than that in butter in mildly hypercholesterolaemic subjects. European journal of clinical nutrition. 2005;59:1059–63.

36. de Goede J, Geleijnse JM, Ding EL, Soedamah-Muthu SS. Effect of cheese consumption on blood lipids: a systematic review and meta-analysis of randomized controlled trials. Nutrition

reviews. 2015;73:259–75.

37. Thorning TK, Raben A, Bendsen NT, Jørgensen HH, Kiilerich P, Ardö Y, Lorenzen JK, Kristiansen K, Astrup A. Importance of the fat content within the cheese-matrix for blood lipid profile, faecal fat excretion, and gut microbiome in growing pigs. International Dairy Journal. 2016;61:67–75.

38. Raziani F, Tholstrup T, Kristensen MD, Svanegaard ML, Ritz C, Astrup A, Raben A. High intake of regular-fat cheese compared with reduced-fat cheese does not affect LDL cholesterol or risk markers of the metabolic syndrome: a randomized controlled trial. The American Journal of Clinical Nutrition. American Society for Nutrition; 2016;104:973–81.

39. Schlienger JL, Paillard F, Lecerf JM, Romon M, Bonhomme C, Schmitt B, Donazzolo Y, Defoort C, Mallmann C, Le Ruyet P, et al. Effect on blood lipids of two daily servings of Camembert cheese. An intervention trial in mildly hypercholesterolemic subjects. International journal of food sciences and nutrition. 2014;65:1013–8.

40. Howard A, Marks J. Effect of milk-products on serum-cholesterol. The Lancet; Originally published as Volume 2, Issue 8149. 1979;314:957.

41. Severins N, Mensink RP, Plat J. Effects of lutein-enriched egg yolk in buttermilk or skimmed milk on serum lipids & lipoproteins of mildly hypercholesterolemic subjects. Nutrition, metabolism, and cardiovascular diseases: NMCD. 2015;25:210–7.

42. Thorning TK, Raziani F, Bendsen NT, Astrup A, Tholstrup T, Raben A. Diets with high-fat cheese, high-fat meat, or carbohydrate on cardiovascular risk markers in overweight postmenopausal women: A randomized crossover trial. American Journal of Clinical Nutrition. 2015;102:573–81.

43. Christensen R, Lorenzen JK, Svith CR, Bartels EM, Melanson EL, Saris WH, Tremblay A, Astrup A. Effect of calcium from dairy and dietary supplements on faecal fat excretion: a meta-analysis of randomized controlled trials. Obesity reviews: an official journal of the International Association for the Study of Obesity. 2009;10:475–86.

44. Jacobsen R, Lorenzen JK, Toubro S, Krog-Mikkelsen I, Astrup A. Effect of short-term high dietary calcium intake on 24-h energy expenditure, fat oxidation, and fecal fat excretion. International journal of obesity. 2005;29:292–301.

45. Jenkins TC, Palmquist DL. Effect of added fat and calcium on in vitro formation of insoluble calcium fatty acid soaps and cell wall digestibility. J Anim Sci. 1982;55:957.

46. Van der Meer R, De Vries HT. Differential binding of glycine- and taurine-conjugated bile acids to insoluble calcium phosphate. The Biochemical journal. 1985;229:265–8.

47. Van der Meer R, Termont DS, De Vries HT. Differential effects of calcium ions and calcium phosphate on cytotoxicity of bile acids. The American Journal of Physiology. 1991;260:142–7.

48. Govers MJ, Termont DS, Van Aken GA, Van der Meer R. Characterization of the adsorption of conjugated and unconjugated bile acids to insoluble, amorphous calcium phosphate. Journal of lipid research. 1994;35:741–8.

49. Gacs G, Barltrop D. Significance of Ca-soap formation for calcium absorption in the rat. Gut. 1977;18:64–8.

50. Bendsen NT, Hother AL, Jensen SK, Lorenzen JK, Astrup A. Effect of dairy calcium on fecal fat excretion: a randomized crossover trial. International journal of obesity (2005). 2008;32:1816–24.

51. Mu H, Hoy CE. The digestion of dietary triacylglycerols. Progress in lipid research. 2004;43:105–33.

52. Thorning TK, Bendsen NT, Jensen SK, Ardö Y, Tholstrup T, Astrup A, Raben A. Cheddar cheese ripening affects plasma nonesterified fatty acid and serum insulin concentrations in growing pigs. Journal of Nutrition. 2015;145:1453–8.

53. Lorenzen JK, Jensen SK, Astrup A. Milk minerals modify the effect of fat intake on serum lipid profile: results from an animal and a human short-term study. The British journal of nutrition. 2014;111:1412–20.

54. Buchowski MS, Aslam M, Dossett C, Dorminy C, Choi L, Acra S. Effect of dairy and nondairy calcium on fecal fat excretion in lactose digester and maldigester obese adults. International journal of obesity. 2010;34:127–35.

55. Weaver CM, Campbell WW, Teegarden D, Craig BA, Martin BR, Singh R, Braun MM, Apolzan JW, Hannon TS, Schoeller DA, et al. Calcium, dairy products, and energy balance in overweight adolescents: a controlled trial. The American journal of clinical nutrition. 2011;94:1163–70.

56. Michalski MC, Genot C, Gayet C, Lopez C, Fine F, Joffre F, Vendeuvre JL, Bouvier J, Chardigny JM, Raynal-Ljutovac K, et al. Multiscale structures of lipids in foods as parameters affecting fatty acid bioavailability and lipid metabolism. Progress in lipid research. 2013;52:354–73.

57. Chung R, Kamili A, Tandy S, Weir J, Gaire R, Wong G, Meikle P, Cohn J, Rye K. Dietary Sphingomyelin Lowers Hepatic Lipid Levels and Inhibits Intestinal Cholesterol Absorption in High- Fat- Fed Mice. PLoS One. 2013;8:e55949.

58. Conway V, Gauthier S, Pouliot Y. Effect of cream pasteurization, microfiltration and enzymatic proteolysis on in vitro cholesterol-lowering activity of buttermilk solids. Dairy Science & Technology. 2010;90:449–60.

59. Eckhardt E, Wang D, Donovan J, Carey MC. Dietary sphingomyelin suppresses intestinal cholesterol absorption by decreasing thermodynamic activity of cholesterol monomers. Gastroenterology. 2002;122:948–56.

60. Conway V, Couture P, Richard C, Gauthier SF, Pouliot Y, Lamarche B. Impact of buttermilk consumption on plasma lipids and surrogate markers of cholesterol homeostasis in men and women. Nutrition, metabolism, and cardiovascular diseases: NMCD. Elsevier B.V; 2013;23:1255–62.

61. Lorenzen JK, Astrup A. Dairy calcium intake modifies responsiveness of fat metabolism and blood lipids to a high-fat diet. The British journal of nutrition. 2011;105:1–10.

62. Ditscheid B, Keller S, Jahreis G. Cholesterol metabolism is affected by calcium phosphate supplementation in humans. The Journal of nutrition. 2005;135:1678–82.

63. Rosqvist F, Smedman A, Lindmark-Mansson H, Paulsson M, Petrus P, Straniero S, Rudling M, Dahlman I, Riserus U. Potential role of milk fat globule membrane in modulating plasma lipoproteins, gene expression, and cholesterol metabolism in humans: a randomized study. The American Journal of Clinical Nutrition. 2015;102:20–30.

64. Ohlsson L, Burling H, Nilsson A. Long term effects on human plasma lipoproteins of a formulation enriched in butter milk polar lipid. Lipids in health and disease. 2009;8:44.

65. Weiland A, Bub A, Barth SW, Schrezenmeir J, Pfeuffer M. Effects of dietary milk- and soyaphospholipids on lipid-parameters and other risk indicators for cardiovascular diseases in overweight or obese men - two double-blind, randomised, controlled, clinical trials. Journal of nutritional science. 2016;5:e21.

66. St-Onge MP, Farnworth ER, Jones PJ. Consumption of fermented and nonfermented dairy products: effects on cholesterol concentrations and metabolism. The American Journal of Clinical Nutrition. 2000;71:674–81.

67. Wolever TM, Spadafora P, Eshuis H. Interaction between colonic acetate and propionate in humans. The American Journal of Clinical Nutrition. 1991;53:681–7.

68. Wolever TM, Spadafora PJ, Cunnane SC, Pencharz PB. Propionate inhibits incorporation of colonic [1,2-13C]acetate into plasma lipids in humans. The American Journal of Clinical Nutrition. 1995;61:1241–7.

69. Wolever TM, Fernandes J, Rao A V. Serum acetate:propionate ratio is related to serum cholesterol in men but not women. The Journal of nutrition. 1996;126:2790–7.

70. Zheng H, Yde CC, Clausen MR, Kristensen M, Lorenzen J, Astrup A, Bertram HC. Metabolomics investigation to shed light on cheese as a possible piece in the French paradox puzzle. Journal of Agricultural and Food Chemistry. 2015;63:2830–9.

71. Tsuchiya A, Almiron-Roig E, Lluch A, Guyonnet D, Drewnowski A. Higher satiety ratings following yogurt consumption relative to fruit drink or dairy fruit drink. Journal of the American Dietetic Association. 2006;106:550–7.

72. Barbé F, Ménard O, Le Gouar Y, Buffière C, Famelart M-H, Laroche B, Feunteun S Le, Rémond D, Dupont D. Acid and rennet gels exhibit strong differences in the kinetics of milk protein digestion and amino acid bioavailability. Food chemistry. 2014;143:1–8.

73. Gaudichon C, Laurent C, Mahé S, Marks L, Tomé D, Krempf M. Rate of [15N]leucine incorporation and determination of nitrogenous fractions from gastro-jejunal secretion in fasting humans. Reproduction, nutrition, development. 1994;34:349–59.

74. Gaudichon C, Mahé S, Roos N, Benamouzig R, Luengo C, Huneau JF, Sick H, Bouley C, Rautureau J, Tome D. Exogenous and endogenous nitrogen flow rates and level of protein hydrolysis in the human jejunum after [15N]milk and [15N]yoghurt ingestion. The British journal of nutrition. 1995;74:251–60.

75. Barbé F, Le Feunteun S, Rémond D, Ménard O, Jardin J, Henry G, Laroche B, Dupont D.

Tracking the in vivo release of bioactive peptides in the gut during digestion: Mass spectrometry peptidomic characterization of effluents collected in the gut of dairy matrix fed mini-pigs. Food Research International. 2014;63:147–56.

76. Boutrou R, Gaudichon C, Dupont D, Jardin J, Airinei G, Marsset-Baglieri A, Benamouzig R, Tome D, Leonil J. Sequential release of milk protein-derived bioactive peptides in the jejunum in healthy humans. The American Journal of Clinical Nutrition. 2013;97:1314–23.

77. Doyon CY, Tremblay A, Rioux LE, Rheaume C, Cianflone K, Poursharifi P, Turgeon SL. Acute effects of protein composition and fibre enrichment of yogurt consumed as snacks on appetite sensations and subsequent ad libitum energy intake in healthy men. Applied physiology, nutrition, and metabolism. 2015;40:980–9.

78. Lillefosse HH, Clausen MR, Yde CC, Ditlev DB, Zhang X, Du Z-Y, Bertram HC, Madsen L, Kristiansen K, Liaset B. Urinary Loss of Tricarboxylic Acid Cycle Intermediates As Revealed by Metabolomics Studies: An Underlying Mechanism to Reduce Lipid Accretion by Whey Protein Ingestion? Journal of Proteome Research. American Chemical Society; 2014;13:2560–70.

79. Zheng H, Yde CC, Dalsgaard TK, Arnberg K, Mølgaard C, Michaelsen KF, Larnkjær A, Bertram HC. Nuclear magnetic resonance-based metabolomics reveals that dairy protein fractions affect urinary urea excretion differently in overweight adolescents. European Food Research and Technology. Springer Berlin Heidelberg; 2015;240:489–97.

80. Dardevet D, Rémond D, Peyron M-A, Papet I, Savary-Auzeloux I, Mosoni L. Muscle wasting and resistance of muscle anabolism: the anabolic threshold concept for adapted nutritional strategies during sarcopenia. TheScientificWorldJournal. 2012;2012:269531.

81. Rémond D, Machebeuf M, Yven C, Buffière C, Mioche L, Mosoni L, Patureau Mirand P. Postprandial whole-body protein metabolism after a meat meal is influenced by chewing efficiency in elderly subjects. The American journal of clinical nutrition. 2007;85:1286–92.

82. Lacroix M, Bon C, Bos C, Léonil J, Benamouzig R, Luengo C, Fauquant J, Tomé D, Gaudichon C. Ultra high temperature treatment, but not pasteurization, affects the postprandial kinetics of milk proteins in humans. The Journal of nutrition. 2008;138:2342–7.

83. Michalski M-C, Januel C. Does homogenization affect the human health properties of cow's milk? Trends in Food Science & Technology. 2006;17:423–37.

84. Bourlieu C, Ménard O, De La Chevasnerie A, Sams L, Rousseau F, Madec M-N, Robert B, Deglaire A, Pezennec S, Bouhallab S, et al. The structure of infant formulas impacts their lipolysis, proteolysis and disintegration during in vitro gastric digestion. Food chemistry. 2015;182:224–35.

85. Bourlieu C, Michalski M-C. Structure-function relationship of the milk fat globule. Current opinion in clinical nutrition and metabolic care. 2015;18:118–27.

86. Comerford K, Pasin G. Emerging Evidence for the Importance of Dietary Protein Source on Glucoregulatory Markers and Type 2 Diabetes: Different Effects of Dairy, Meat, Fish, Egg, and Plant Protein Foods. Nutrients. 2016;8:446.

87. Aune D, Norat T, Romundstad P, Vatten LJ. Dairy products and the risk of type 2 diabetes: a systematic review and dose-response meta-analysis of cohort studies. The American Journal of Clinical Nutrition. 2013;98:1066–83.

88. Ricci-Cabello I, Herrera MO, Artacho R. Possible role of milk-derived bioactive peptides in the treatment and prevention of metabolic syndrome. Nutrition reviews. 2012;70:241–55.

89. Nilsson M, Stenberg M, Frid AH, Holst JJ, Bjorck IM. Glycemia and insulinemia in healthy subjects after lactose-equivalent meals of milk and other food proteins: the role of plasma amino acids and incretins. The American Journal of Clinical Nutrition. 2004;80:1246–53.

90. Rideout TC, Marinangeli CP, Martin H, Browne RW, Rempel CB. Consumption of low-fat dairy foods for 6 months improves insulin resistance without adversely affecting lipids or bodyweight in healthy adults: a randomized free-living cross-over study. Nutrition journal. 2013;12:56.

91. Maynard CL, Elson CO, Hatton RD, Weaver CT. Reciprocal interactions of the intestinal microbiota and immune system. Nature. Nature Research; 2012;489:231–41.

92. Conway V, Gauthier S, Pouilot Y. Buttermilk: Much more than a source of milk phospholipids. Animal Frontiers. 2014;4:44–51.

93. The National Food Institute- Technical University of Denmark. Frida Food Data [Internet]. version 1. 2015 [cited 2017 Feb 8]. Available from: http://frida.fooddata.dk/

94. Michalski M-C. Specific molecular and colloidal structures of milk fat affecting lipolysis, absorption and postprandial lipemia. European Journal of Lipid Science and Technology. 2009;111:413–31.

7. TABLES

Table 1. Identified gaps in the existing knowledge and future research questions

Epidemiology
- How should high-fat and low-fat dairy foods be defined? Does it make sense to pool low-fat and high-fat dairy products when considering the differences in active components and structures between various types of dairy products?
- Epidemiological studies should recognize that milk, yogurt, cheese, butter and cream are each unique (in other ways apart from their fat content) and hence should be studied accordingly.
- Heterogeneity in observational studies needs to be thoroughly addressed in the analysis of future studies e.g. by stratification. Intake amounts, types and fortification practices vary between nations, genders and age groups, and genetic variation may also exist. - Analysis based on observational data from which certain foods are substituted with others is a new but promising approach. More of such studies are warranted in order to determine the optimal diet composition for disease prevention. However, analyses of substitution of dairy fat with other fats should be supplemented by analyses replacing different types of dairy foods with other relevant foods (e.g. replacement of cheese for plant oil is not a realistic dietary choice from a food perspective).
- To what extent is the intake of dairy products a marker of overall healthier or unhealthier diets and lifestyles?
- What are the facilitators and barriers to dairy product consumption in different age groups?
Bone health
- Protein requirements in children are extrapolated from nitrogen balance studies in adults. Should bone outcome measures be used to set protein requirements in children?
- Is the amino acid composition of protein important to bone health? Is there an optimal amino acid composition for bone remodeling?
- Does fermentation of dairy products have an impact on bone, and if so, what are the mechanisms?
Protein functionality
- More data are needed on protein ileal digestibility of dairy products as well as other protein sources.
- Is there an optimal protein source for the elderly and does it differ from that of the younger population?
- How can the anabolic protein threshold consistently be achieved in elderly? Is a certain intake of protein in each meal required for an optimal muscle protein synthesis? Should protein recommendations for elderly be increased due to the higher anabolic threshold?
- Proteins are essential for growth, but some amino acids can also act as signals and stimulate production of other compounds, like IGF-1. It is still unknown whether or not the endocrine functions of protein are affected by the dairy matrix.
- It is uncertain whether or not identified bioactive peptides from dairy can be absorbed into the blood intact, and whether or not these have a sufficiently long half-life to exert metabolic effects.
- Quantitative data on bioactive peptide release during digestion are lacking.
- Do dairy protein types or dairy processing cause different effects on the gut microbiota composition and activity?
Fat functionality
-Does it matter whether the membrane of the MFGM is disrupted, or is the presence of phospholipids the determinant for the metabolic effect of the MFGM? Could a potential difference be explained by the intrinsic proteins/enzymes of the MFGM?
- Could MFGM be involved in the metabolic programming of infants?
- What is the metabolic importance of the milk fat globule structure (homogenized vs. non-homogenized milk-fat)?
- RCT's in humans confirming the suggested specific effect of fermented dairy on insulin sensitivity remains to be conducted. What is the impact on health of a liquid and solid state of dairy fat e.g. melted vs solid cheese?
- What is the precise role of calcium in the reduced fat digestion? Fecal calcium-fatty acid soaps needs to be chemically confirmed.
- What is the role of dairy as bile acid sequestrants; is phosphate responsible for the increased bile acid excretion, and is there a dose response effect?
- Studies with free-living designs often find lower effect sizes than studies with a highly controlled design. It has to be clarified if the effects of the various mechanisms have a magnitude to impose an effect in whole diet setting?
- What is the role of fat in different dairy-matrices on regulation of non-lipid cardiometabolic risk markers e.g. inflammation and oxidative stress.
- Can metabolomics and lipidomics approaches be used to explain unknown metabolic pathways of action by various dairy matrices?
Digestion kinetics
- The structure a food adopts in the stomach is essential to understand its digestion behavior. Not sufficiently research has been conducted on the gastric residence time and postprandial responses to different dairy matrices.
- The postprandial kinetics after cheese intake may likely depend on the coagulation method used during cheese processing. It should be investigated whether cheese produced solely by acidification (e.g. cream cheese) or rennet (most cheeses) affects digestion kinetics and appetite differently. Also, it should be investigated if the postprandial kinetics after intake of soft and hard cheese or after intake of cheeses produced with different ripening methods differs.
- More research is needed on effects of dairy processing e.g. UHT and cooking, on digestion kinetics.

Table 2. Bioactive components and supramolecular structures in different dairy products[1]

	Calcium (mg/100 g)	Phosphorus (mg/100 g)	MFGM[2] (mg/100 g)	Protein[3] (g/100 g, type)	Fermented	Fat structure[4]	Protein network
Cheese[5] (25% fat)	659	510	150	23.2, casein	yes	MFG/ aggregates/ free fat	Solid/ viscoelastic
Milk (skimmed, 0,5% fat)	124	97	15	3.5, whey/casein	no	Tiny native MFG/ potential MFGM fragments	Liquid
Milk (whole, 3.5% fat)	116	93	35	3.4, whey/casein	no	Native MFG or homogenized milk fat droplets/ potential MFGM fragments	Liquid
Yoghurt (1.5% fat)	136	99	15	4.1, whey/casein	yes	Tiny native MFG/ potential MFGM fragments	Gel/ viscoelastic
Cream (38% fat)	67	57	200	2, -	no	Native MFG or homogenized milk fat droplets/ potential MFGM fragments	Liquid
Butter	15	24	-	<1 -	no/yes[6]	Continuous fat phase (water-in-oil emulsion)/ MFGM residue traces	-

Abbreviations: MFG, Milk fat globules; MFGM, Milk fat globule membrane.[1]approximate amounts, [2]based on Dewettinck et al. (2008)(11) and Conway et al. (2014)(92), [3]according to food composition tables from The Technical University of Denmark (93), [4]based on Michalski et al. (2009)(94) and Michalski et al. (2013)(56), [5]semi-hard Danbo type, [6]Depends on the production method used. With Indirect Biological Acidification (IBA) starter culture is added to the butter after churning.

8. FIGURE LEGENDS

Figure 1: Reprinted from de Goede et al. (2015) (36). Forest plot from meta-analysis of intervention studies comparing effects of cheese vs. butter consumption on plasma LDL-cholesterol levels. Data shown include the author names, year of publication, relative risks (RRs), 95% confidence intervals (95% CIs), and weight to the overall meta-analysis. Study-specific RRs and 95% CIs are represented as shaded squares. The area of the squares is within the overall meta-analysis. The diamond represents the pooled RR and the 95% CI. I^2 indicates the percentage of heterogeneity due to between-study variation. Abbreviations: LDL-C, LDL cholesterol; CI, Confidence Intervals; RR, Relative Risk.

Figure 2: Reprinted from Soerensen et al. (2014) (34). Correlations (95% CIs) between changes in LDL-C and fecal fat excretion during the butter-control (black), milk (open), and cheese (gray) periods ($R^2 = 0.163$, $P = 0.002$) ($n = 15$). Abbreviations: LDL-C, LDL-cholesterol.

Figure 2

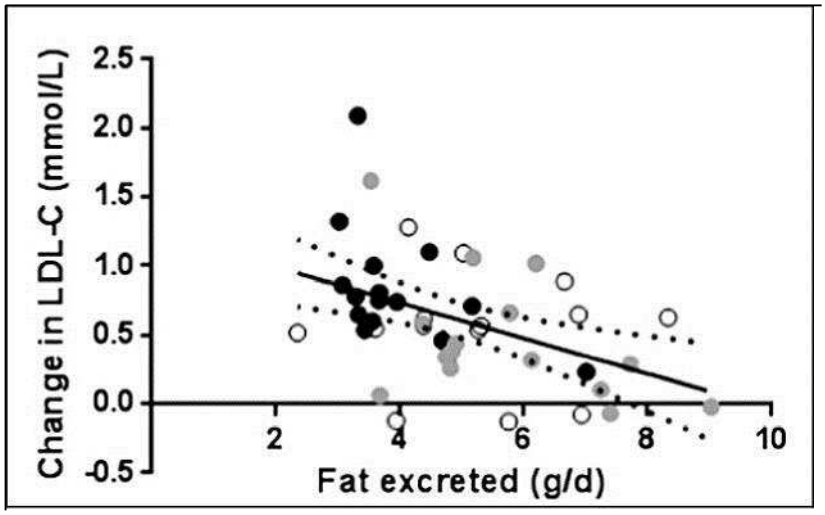
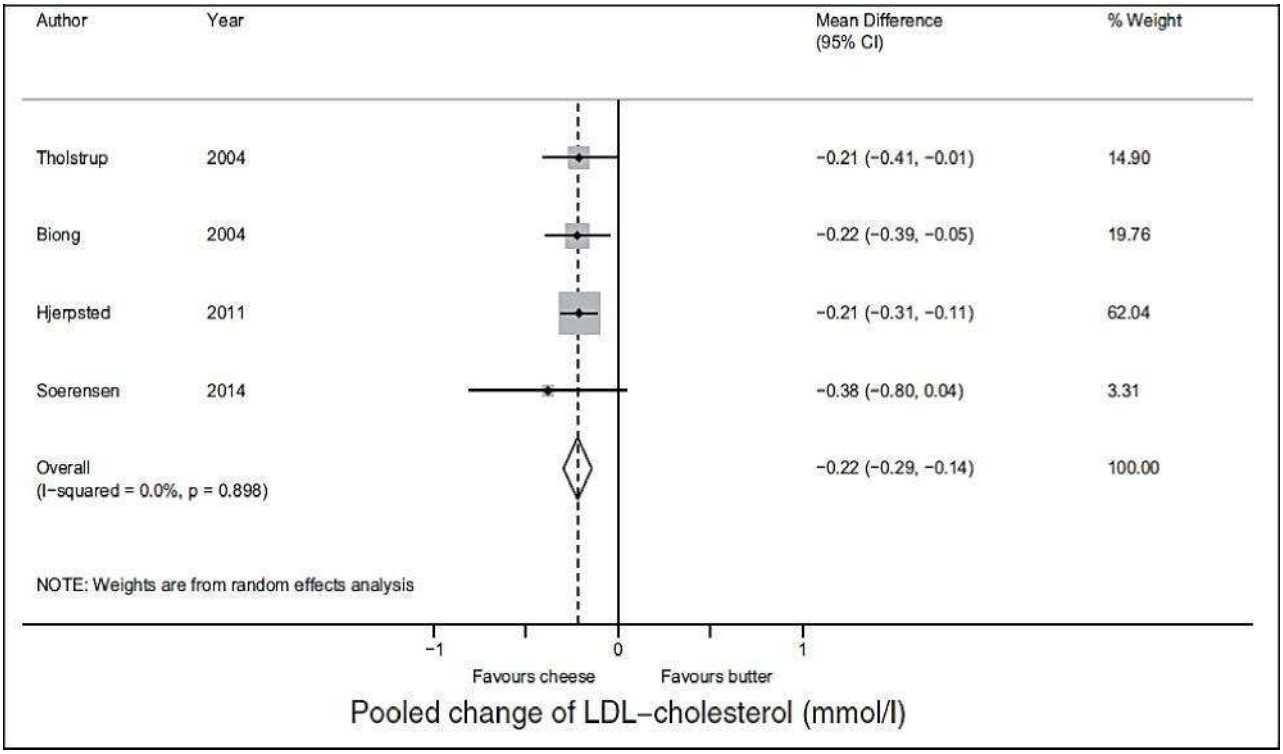

Change in LDL-C (mmol/L)
2.5
2.0
1.5
1.0
0.5
0.0
-0.5
2
4
6
8
10
Fat excreted (g/d)

Figure 1


Author
Year
Mean Difference (95% CI)
% Weight
Tholstrup 2004 −0.21 (−0.41, −0.01) 14.90
Biong 2004 −0.22 (−0.39, −0.05) 19.76
Hjerpsted 2011 −0.21 (−0.31, −0.11) 62.04
Soerensen 2014 −0.38 (−0.80, 0.04) 3.31
Overall (I−squared = 0.0%, p = 0.898) −0.22 (−0.29, −0.14) 100.00
NOTE: Weights are from random effects analysis
−1
0
1
Favours cheese
Favours butter
Pooled change of LDL−cholesterol (mmol/l)