

Review

The Role of Dairy Foods in Weight Management

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Dietary calcium appears to play a pivotal role in the regulation of energy metabolism and obesity risk. High calcium diets attenuate body fat accumulation and weight gain during periods of over-consumption of an energy-dense diet and to increase fat breakdown and preserve metabolism during caloric restriction, thereby markedly accelerating weight and fat loss. This effect is mediated primarily by circulating calcitriol, which regulates adipocyte intracellular Ca^{2+} . Studies of human adipocyte metabolism demonstrate a key role for intracellular Ca^{2+} in regulating lipid metabolism and triglyceride storage, with increased intracellular Ca^{2+} resulting in stimulation of lipogenic gene expression and lipogenesis and suppression of lipolysis, resulting in adipocyte lipid filling and increased adiposity. Moreover, the increased calcitriol produced in response to low calcium diets stimulates adipocyte Ca^{2+} influx and, consequently, promotes adiposity, while higher calcium diets inhibit lipogenesis, promote lipolysis, lipid oxidation and thermogenesis and inhibit diet-induced obesity in mice. Notably, dairy sources of calcium exert markedly greater effects in attenuating weight and fat gain and accelerating fat loss. This augmented effect of dairy products versus supplemental calcium has been localized, in part, to the whey fraction of dairy and is likely due to additional bioactive compounds, such as angiotensin converting enzyme (ACE) inhibitors in dairy, as well as the rich concentration of branched chain amino acids, which act synergistically with calcium to attenuate adiposity; however, these compounds do not fully account for the observed effects, as whey has significantly greater bioactivity than found in these compounds. These concepts are confirmed by epidemiological data as well as recent clinical trials which demonstrate that diets which include at least three daily servings of dairy products result in significant reductions in body fat mass in obese humans in the absence of caloric restriction and markedly accelerates the weight and body fat loss secondary to caloric restriction compared to low dairy diets. These data indicate an important role for dairy products in both the ability to maintain a healthy weight and the management of overweight and obesity.

Key teaching points:

- Dietary calcium modulates circulating calcitriol (1,25-dihydroxyvitamin D) levels that in turn regulate intracellular calcium which affects fat metabolism in human adipocytes.
- Reducing calcitriol levels by increasing dietary calcium results in reduction of body fat in the absence of caloric restriction, substantially increases body weight and fat loss during caloric restriction and reduces weight and fat regain following successful weight loss.
- Dairy sources of calcium are markedly (50–100%) more effective than supplemental calcium in reducing body weight and body fat during caloric restriction. A portion of this additional anti-obesity bioactivity is attributable to the ACE-inhibitory activity of dairy and to the rich concentration of branched chain amino acids.
- This anti-obesity effect of dietary calcium/dairy is supported by cellular mechanistic studies, animal studies human epidemiological studies and clinical trials.
- Incorporating dairy into weight management regimens is associated with significant preservation of lean body mass during caloric restriction.

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INTRODUCTION

A substantial body of data has emerged over the last five years to indicate that dietary calcium and dairy foods modulate adipocyte lipid metabolism and energy partitioning between adipose tissue and lean body mass, resulting in a significant “anti-obesity” effect. This effect is supported by a clear mechanistic framework, prospective and retrospective epidemiological reports and observational studies, secondary analyses of past clinical trials originally conducted with other primary endpoints (i.e., skeletal, cardiovascular) and prospective clinical trials. Further, these findings are evident in populations of multiple ages and ethnicities, suggestive of a generally robust effect, as discussed in this review.

MECHANISMS

A compelling mechanism for the anti-obesity effect of dietary calcium was provided by our studies of the mechanism of action of the *agouti* gene in regulating murine and human adipocyte metabolism [1–21]. These studies demonstrated a key role for intracellular Ca^{2+} in the regulation of adipocyte metabolism, resulting in modulation of adipocyte triglyceride stores; intracellular Ca^{2+} is regulated by calcitrophic hormones, and this provides the primary mechanistic basis for the anti-obesity effect of dietary calcium.

This regulation of adipocyte lipid metabolism by intracellular Ca^{2+} provides the key framework for dietary calcium modulation of adiposity. We have found both parathyroid hormone [4] and $1,25\text{-(OH)}_2\text{-D}$ [22,23] stimulate rapid increases in human adipocyte intracellular Ca^{2+} ; accordingly, suppression of these hormones by increasing dietary calcium facilitates re-partitioning of dietary energy from lipid storage to lipid oxidation and thermogenesis. Although both parathyroid hormone and $1,25\text{-(OH)}_2\text{-D}$ both modulate adipocyte intracellular Ca^{2+} , a growing body of evidence indicates that $1,25\text{-(OH)}_2\text{-D}$ plays a pivotal role in modulation of lipid metabolism, although an additional possible role for parathyroid hormone has not been excluded. Human adipocytes possess membrane (non-genomic) vitamin D receptors which transduce a rapid intracellular Ca^{2+} response to $1,25\text{-(OH)}_2\text{-D}_3$ [23,24]; consequently, $1,25\text{-(OH)}_2\text{-D}_3$ treatment of human adipocytes results in coordinated activation of fatty acid synthase expression and activity and suppression of lipolysis, leading to an expansion of adipocyte lipid storage [22,24,25]. However, it should be noted that while these data provide a plausible mechanism of action based on *in vitro* studies in human adipocytes, the direct effect of calcitrophic hormones on human adipocyte metabolism has not yet been assessed utilizing *in vivo* techniques, such as microdialysis. Nonetheless, a potential role of $1\alpha,25\text{-(OH)}_2\text{-D}_3$ in human obesity is suggested by other data. Polymorphisms in the nuclear vitamin D receptor (nVDR) gene are associated with susceptibility to obesity in humans [26,27], and several

lines of evidence demonstrate an alteration of the vitamin D-endocrine system in obese humans, with an increase in circulating $1\alpha,25\text{-(OH)}_2\text{-D}_3$ level [28,29]. These observations, coupled with the direct effects of $1\alpha,25\text{-(OH)}_2\text{-D}_3$ on adipocyte lipid metabolism, strongly implicate the increase in $1\alpha,25\text{-(OH)}_2\text{-D}_3$ found on low calcium diets as a contributory factor to excess adiposity.

In addition to regulating adipocyte metabolism via a non-genomic membrane receptor (the membrane-associated rapid response to steroid, or MARRS protein) [23,30,31], $1\alpha,25\text{-(OH)}_2\text{-D}_3$ also acts via the “classical” nuclear vitamin D receptor in adipocytes to inhibit the expression of uncoupling protein2 (UCP2) [32]; further, suppression of $1,25\text{-(OH)}_2\text{-D}_3$ levels by feeding high calcium diets to mice results in increased adipose tissue UCP2 expression and attenuation of the decline in thermogenesis which otherwise occurs with energy restriction [25], suggesting that high calcium diets may also affect energy partitioning by suppressing $1,25\text{-(OH)}_2\text{-D}_3$ -mediated inhibition of adipocyte UCP2 expression. However, the role of UCP2 in thermogenesis is not clear, and the observed thermogenic effect may be mediated by other, as of yet unidentified mechanisms. Moreover, thermogenic effects of dietary calcium and/or dairy products have not yet been demonstrated in humans. Nonetheless, in addition to inducing a mitochondrial proton leak, UCP2 serves to mediate mitochondrial fatty acid transport and oxidation, suggesting that $1,25\text{-(OH)}_2\text{-D}_3$ suppression of UCP2 expression may still contribute to decreased fat oxidation and increased lipid accumulation on low calcium diets [32].

Recent data demonstrate that $1,25\text{-(OH)}_2\text{-D}_3$ may also modulate adiposity by inhibiting adipocyte apoptosis [33]. This effect is mediated, in part, via inhibition of UCP2 expression and a consequent increase in mitochondrial potential, a key regulator of apoptosis, and in part via $1,25\text{-(OH)}_2\text{-D}_3$ regulation of cytosolic Ca^{2+} and of Ca^{2+} flux between endoplasmic reticulum and mitochondria [33 and unpublished data]. Consequently, adipocyte apoptosis is significantly impaired in association with increased $1,25\text{-(OH)}_2\text{-D}_3$ levels in mice fed low calcium diets, while there is a marked increase in adipocyte apoptosis in mice fed high calcium and/or high dairy diets [33]. An integrated summary of these mechanisms is shown in Fig. 1.

Increasing dietary calcium may also result in increased fecal fatty acid excretion and, accordingly, it is possible that the resultant increase in fecal energy loss could contribute to the anti-obesity effects of dietary calcium. In support of this concept, Papakonstantinou et al [34] demonstrated that a high calcium diet produced a substantial increase in fecal fat and energy excretion, and attributed the observed reduction in adiposity to fecal energy loss, although a marked decrease in circulating $1,25\text{-(OH)}_2\text{-D}_3$ was found as well. More recently, Jacobsen et al [35] reported that a short-term increase in calcium intake from 500 to 1800 mg/day increased fecal fat excretion ~2.5-fold, from 5.9 to 14.2 g/day. However, while such an increase in fecal fat loss will clearly contribute to a

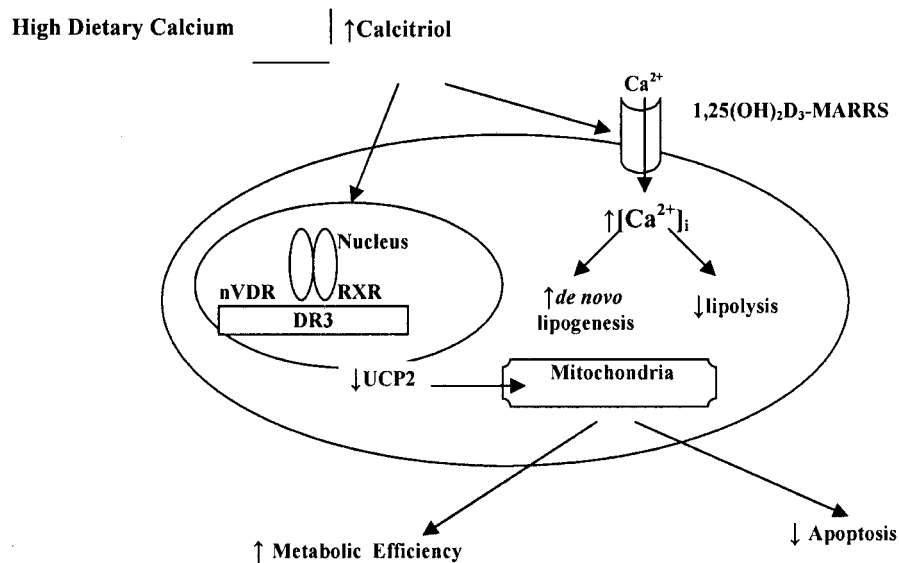


Fig. 1. An integrated summary of mechanisms.

reduction in energy balance, it required a larger level of calcium (1800 mg vs. 1200 mg used in clinical trials of calcium and obesity) to produce a quantitatively small effect (8.3 g additional fecal fat, representing a 75 kcal/day loss) which is insufficient to explain the magnitude of the effects observed in clinical trials (discussed later in this review). Previous studies also demonstrated that large increases in dietary calcium (2–4 g/day) result in statistically significant, but modest, increases in fecal fat loss [36–38]. For example, a supplement of 2 g calcium increased fecal fat excretion from 6.8% to 7.4% of total fat intake [37]. In contrast, in order to achieve a clinically meaningful (albeit modest) contribution to weight loss, the pancreatic lipase inhibitor orlistat must produce approximately a 30% inhibition of total dietary fat absorption, versus the approximately 1–2% found with dietary calcium. Thus, while calcium-inhibition of fat absorption may contribute to an anti-obesity effect, this effect is too small to explain the observed effects. Instead, the primary effect appears to be inhibition of calcitrophic hormone effects on adipocyte energy storage and utilization.

Other Dairy Components

Although dietary calcium appears to inhibit adiposity via the aforementioned 1,25-(OH)₂-D₃ mechanisms, data from clinical trials, rodent studies and population studies all indicate a substantially (~two-fold) greater effect of dairy versus supplemental sources of calcium in attenuating adiposity. Accordingly, it is important to identify the additional component(s) of dairy that may be responsible for this augmentation. Our preliminary studies in mice isolate a portion of this additional dairy-derived bioactivity to the whey fraction [39]. Likely candidates for this additional bioactivity include the branched

chain amino acid content of dairy protein and specific bioactive whey-derived peptides.

Dairy contains a number of bioactive compounds, which may act either independently or synergistically with calcium to affect lipogenesis, lipolysis, lipid oxidation and/or energy partitioning. Among these, the significant angiotensin converting enzyme (ACE) inhibitory activity contained in whey protein may be relevant to adipocyte lipid metabolism. Angiotensin II upregulates adipocyte fatty acid synthase expression [reviewed in 40], and ACE inhibition mildly attenuates obesity in both mice and in hypertensive patients. Consequently, since adipose tissue has an autocrine renin-angiotensin system, it is possible that a whey-derived ACE inhibitor may contribute to the anti-obesity effects of dairy.

In support of these concepts, a whey-derived ACE inhibitor significantly augmented the effects of dietary calcium on weight and fat loss in energy-restricted mice [39]. However, the combination of the calcium and ACE inhibitor was markedly less potent than either milk or whey in reducing body fat; moreover, milk and whey both substantially preserved skeletal muscle mass during energy restriction while calcium and the calcium/ACE inhibitor combination were without effect. Consequently, although calcium plays a significant role in weight management, and this effect is enhanced by whey-derived ACE-inhibition, a significant portion of the dairy effect remains unexplained. While it is likely that the protective effects of dairy on muscle mass may be attributable to the branched chain amino acid content of whey protein (discussed below), this is unlikely to explain the additional effects of whey on adiposity. An evaluation of whey-derived mineral mix versus calcium carbonate indicates that the other minerals contained in whey do not contribute to the anti-obesity effects of whey [39, and unpublished data]. Present studies in progress are directed

towards identification of the additional components which contribute to the additional anti-obesity bioactivity of dairy.

Although it may be tempting to speculate that the protein content of dairy may play a role in mediating the anti-obesity effect, studies demonstrating an anti-obesity effect of dairy products in both rodents and humans have maintained constant levels of protein intake. Accordingly, the protein content of dairy and whey *per se* cannot be responsible for the additional bioactivity. However, the amino acid composition of dairy protein may play a role. Dairy proteins have a high protein quality score and contain a high proportion (~26%) of branched chain amino acids (BCAA) [41,42]. In addition to supporting protein synthesis, the BCAA (leucine, isoleucine and valine) play specific metabolic roles as energy substrates and in the regulation of muscle protein synthesis, and their potential to participate in these additional metabolic processes are limited by their availability, with first priority provided to new protein synthesis [recently reviewed by Layman, 41]. Accordingly, only diets which provide leucine at levels which exceed requirements for protein synthesis can fully support the intracellular leucine levels required to support additional signaling pathways [41]. Consequently, the abundance of leucine in both casein and whey is of particular interest, as it plays a distinct role in protein metabolism and a pivotal role in translation initiation of protein synthesis [43]. Accordingly, the high concentration of BCAA, and leucine in particular, in dairy products may be an important factor in the re-partitioning of dietary energy from adipose tissue to skeletal muscle [44–46]. This suggests an interaction between the high levels of calcium in dairy in combination with the BCAA content of dairy protein, possibly in concert with other dairy-derived bioactive compounds may work in synergy to minimize adiposity and maximize lean mass.

Modulation of Central Adiposity

Both rodent and human studies demonstrate a shift in the distribution of body fat loss on high versus low calcium diets during energy restriction. In rodents, high calcium and high dairy diets produce a preferential loss of visceral adipose tissue [22,25], while clinical trials demonstrate a preferential loss of fat from the trunk region (i.e. an increase in trunk fat loss as a percentage of total fat loss) [47–50]. Recent studies describing the role of autocrine production of cortisol by adipose tissue provide a plausible and likely mechanism for this effect as well.

Human adipose tissue expresses significant 11 β -hydroxysteroid dehydrogenase-1 (11 β -HSD-1), which can generate active cortisol from cortisone, and visceral adipose tissue exhibits greater 11 β -HSD-1 expression than does subcutaneous adipose tissue [51,52]. Further, selective overexpression of 11- β -HSD-1 in white adipose tissue of mice results in central obesity [53,54], while homozygous 11 β -HSD-1 knockout mice exhibit protection from features of the metabolic syndrome [55]. We have recently found 1,25-(OH)₂-D₃ to exert

both short-term and long-term regulation of 11 β -HSD-1 in human adipocytes, resulting in ~2-fold increases in 11 β -HSD-1 expression and up to 6-fold increases in net cortisol production [56]. Thus, the increase in 1,25-(OH)₂-D₃ found on low calcium diets is likely to cause selective expansion of visceral adipose tissue, while the observed selective loss of central adiposity on high calcium and high dairy diets appears to be attributable to a reduction in cortisol production by visceral adipocytes [56].

ANIMAL STUDIES

We have confirmed the anti-obesity effect of dietary calcium and dairy products in a series of studies conducted in transgenic mice which express the *agouti* gene in adipose tissue under the control of the aP2 promoter, similar to the human pattern of expression of *agouti* and other obesity-associated genes [22,25,57–60]. These mice are not obese when fed standard chow diets but are susceptible to adult-onset diet-induced obesity. They respond to low calcium diets with accelerated weight gain and fat accretion, while high calcium diets markedly inhibit lipogenesis, accelerate lipolysis, increase thermogenesis and suppress fat accretion and weight gain in animals maintained at identical caloric intakes [22]. Further, low calcium diets impede body fat loss while high calcium diets markedly accelerate weight and fat loss in transgenic mice subjected to identical levels of caloric restriction [25,57–60]. However, there is one report indicating lack of effect of increasing calcium intake on body weight and body fat in rats and mice [61]. The reason for this difference is not apparent, but may be related to the use of older animals with more fully established obesity, as well as the lack of an energy restriction protocol. However, studies in other animal models (Zucker lean and obese rats, Wistar rats and Spontaneously Hypertensive rats) confirm the observation that increased calcium intake lowers body weight and fat content [34,62,63].

Dietary calcium and dairy also alter the partitioning of dietary energy during re-feeding following weight loss in aP2-*agouti* transgenic mouse model [64]. Although post-obese mice fed a low calcium diet rapidly regained all of the weight and fat that had been lost, re-feeding high calcium diets prevented the suppression of adipose tissue lipolysis and fat oxidation that otherwise accompanies post-dieting repletion and markedly upregulated indices of skeletal muscle fat oxidation [64]. Consequently, although animals re-fed low calcium diets rapidly regained all of the weight and fat that had been lost, animals fed high calcium diets exhibited a 50–85% reduction in weight and fat gain; moreover, dairy exerted markedly greater effects than supplemental calcium on fat oxidation and fat gain [64]. These data are supported by both clinical trials and observational data, as described in the next sections.

CLINICAL STUDIES

The original concept of calcium and dairy modulation of body composition and weight management emerged from data from a hypertension clinical trial, with subsequent corroboration via secondary analysis of other clinical trials originally conducted with skeletal outcomes and finally prospective clinical trials to evaluate the effects of calcium and dairy on adiposity. In the hypertension study, dietary calcium was increased from ~400 to ~1,000 mg/day in obese African Americans without altering dietary energy or macronutrient content. Although body weight did not change, there was a 4.9 kg reduction in body fat [22], which led to the subsequent mechanistic investigations already described. Heaney and colleagues subsequently re-analyzed a series of calcium intervention studies originally designed with primary skeletal endpoints that support a calcium-body weight linkage [65–67]. In an analysis of nine studies, including three controlled trials and six observational studies, a significant negative association between calcium intake and body weight was noted for all age groups studied (third, fifth and eight decades of life). The odds ratio for being overweight was 2.25 for young women below the median calcium intake compared to those above median calcium intake [65], and the controlled trials supported this relationship [65–67]. Overall, increased calcium intake was consistently associated with reduced indices of adiposity (body weight, body fat and/or weight gain); the aggregate effect was each 300 mg increase in daily calcium intake was associated with a 3 kg lower weight in adults and a 1 kg decrease in body fat in children.

Randomized Clinical Trials

Several clinical trials have been conducted to evaluate the effects of dietary calcium and/or dairy on adiposity; to date, all available randomized clinical trial data available are from adults. In the first trial [47], 32 obese adults were maintained on balanced caloric-deficit diets (500 kcal/day deficit) and randomized to control (0–1 serving/day and 400 to 500 mg Ca/day supplemented with placebo), high calcium (control diet supplemented with 800 mg Ca/day), or high dairy (3–4 servings of milk, yogurt and/or cheese/day, total Ca intake of 1200–1300 mg/day). Control subjects lost 5.4% of their body weight over a 24-week study, and this loss was increased to 8.6% on the high calcium diet and to 10.9% on the high dairy diet ($p < 0.01$). Fat loss (via DEXA) followed a similar trend, with the high calcium and high dairy diets augmenting the fat loss found on the low calcium diet by 38 and 64%, respectively ($p < 0.01$). This was accompanied by a marked change in the distribution of body fat loss [47], as fat loss from the trunk region represented 19% of the total fat lost on the low calcium diet, and this was increased to 50% of the fat lost on the high calcium diet and 66% on the high dairy diet; this effect has now

been explained via calcium/ $1,25\text{-(OH)}_2\text{-D}$ modulation of adipose tissue cortisol production [56], as discussed in a preceding section. These findings demonstrate that increasing dietary calcium from suboptimal to adequate levels can enhance the efficacy of an energy-restricted diet in weight and fat loss, while a markedly greater enhancement is found when dairy foods are used compared to calcium supplements [47].

The effects of dairy in augmenting weight and fat loss secondary to caloric restriction have been confirmed in additional clinical trials. A recent follow-up clinical trial of 34 obese subjects consuming a diet supplemented with three servings of yogurt (total calcium intake of ~1,100 mg/day) compared to a placebo control group (calcium intake of 400–500 mg/day) on a balanced calorie-deficit (–500 kcal/day) for 12 weeks supports these findings [48]. Both groups lost weight, but the yogurt group lost 61% more fat (4.43 vs. 2.75 kg) and 81% more trunk fat (3.16 vs. 1.74 kg) than the control group ($p < 0.001$). Similar to the first clinical trial, the fraction of fat lost from the trunk was markedly higher on the yogurt diet vs. control (60.0 vs. 26.4%). Moreover, there was a significant 31% reduction in the loss of lean tissue mass during energy restriction in the yogurt group compared to the control group. No adverse effects were observed on any serum lipid fraction in either of these trials, and there was an improvement in insulin sensitivity, glucose tolerance and blood pressure in the dairy groups in both trials [47,48]. These findings have been extended in a multi-center trial of 105 overweight and obese adults conducted at The University of Tennessee, Purdue University, USDA, ARS, Western Human Nutrition Research Center at the University of California-Davis, and The Ohio State University [50]. The design was similar to the first clinical trial, with subjects randomized to low calcium, high calcium and high dairy groups on balanced deficit (–500 kcal/day) diets for 12-weeks. Although the calcium supplement exerted little effect, the high dairy diet resulted in significant, marked (~2-fold) increases in fat loss and trunk fat loss, similar to that seen in the first trial [48]. However, in contrast to the first clinical trial [47], the calcium supplement was without significant effect.

These findings have also been replicated in a six-month clinical trial in obese African Americans [49], with essentially similar results. Inclusion of three daily servings of dairy into a balanced deficit diet with no alterations in dietary macronutrients results in ~two-fold increase in weight, fat and trunk fat loss versus those maintained on a low dairy diet. These findings were extended to a six-month study of obese African-American adults in the absence of energy deficit [49]. Isocaloric substitution of three daily servings of dairy products into the diets of obese African-American adults maintained on eucaloric diets for six months results in a 5.4% reduction in total body fat and a 4.6% decrease in trunk fat ($p < 0.01$ for both) in the absence of any change in body weight while the control group maintained on a low calcium/low dairy diet with identical macronutrient composition exhibited no significant changes in total body fat or trunk fat [49]. Bowen et al [68] recently reported

that dairy failed to enhance weight loss during 12 weeks of energy restriction in subjects on high protein diets. However, that work utilized a much higher level of protein intake than that used in the aforementioned trials (34% of energy versus 18%), making a direct comparison difficult, as higher protein intakes have been shown in some studies to be associated with greater weight loss. Indeed, the weight loss found by Bowen et al was approximately twice as high 9.7 vs. 4.99 kg) as that found in the control group in the preceding 12-week study (48). At this higher rate of weight loss (0.8 kg/week), a maximal rate of fat mobilization may already be approached, making additional increments due to dairy (or other factors) unlikely. Moreover, the baseline calcium intakes in the Bowen study were considerably higher (899 and 787 mg/day for men and women, respectively, assigned to the dairy protein diet, and 935 and 737 mg/day for those assigned to the mixed protein diet) than in the aforementioned clinical trials [47,48], in which baseline calcium intakes were <600 mg/day. This was considered critical in order to ensure that the effects of correcting suboptimal intakes were studied, rather than the effects of supplementing near-adequate intakes.

Finally, preliminary data demonstrate that a eucaloric high dairy diet markedly attenuates regain of body weight following successful weight loss compared to a low dairy diet (3.03 vs. 1.02 kg weight regain on low vs. high dairy diet, $p < 0.05$) [69]. Similarly, the high dairy diet attenuated regain of body fat (1.959 vs. 0.773 kg on low vs. high dairy diet, $p < 0.01$), and trunk fat (1.546 vs. 0.218 kg on low vs. high dairy diet, $p < 0.01$), indicating that dairy-rich diets attenuate short-term (12-week) weight, fat and trunk fat regain following weight loss. However, longer term assessments are needed to fully evaluate this phenomenon, and are presently in process.

To date, two short-term clinical trials have been conducted to evaluate the mechanisms of the anti-obesity effects of dairy. Both were randomized crossover design studies conducted to evaluate the effects of one-week on each diet and utilized whole-room calorimeters. In the first, level of calcium intake was without effect on 24-hour energy expenditure or fat oxidation, but significantly increased fecal fat and energy excretion [35], as previously discussed. The second study was based upon an observational study in which calcium intake was positively correlated with whole-body fat oxidation in a whole-room calorimeter, with measured calcium intake explaining ~10% of the variance in 24-hour fat oxidation [70]. In the follow-up study, consumption of a high dairy (3–4 servings/day) significantly increased 24-hour fat oxidation by 30 g/day [71]; however, this effect was only significant under conditions of energy deficit (–600 kcal/day) produced by a combination of caloric restriction and physical activity. The high dairy diet also resulted in a decreased respiratory quotient during periods of heightened metabolic activity [71]. Thus, the discrepancy between these findings and those of the previous study may be accounted for by the positive energy balance experienced by subjects in the first study [35], while the increased fat oxidation

was only significant in the second study during negative energy balance [71].

OBSERVATIONAL AND EPIDEMIOLOGICAL STUDIES

Although there have been a limited number of clinical trials to date, these clinical data are supported by multiple lines of evidence, including observational data noting an inverse relationship between dietary calcium and/or dairy and body weight and/or body fat in children and adolescents [72–76], younger and older women [77–79], African-American women [78], as well as by epidemiological data from NHANES I [79], NHANES III [22], NHANES 1999–2000 [79], the Continuing Study of Food Intake of Individuals [80], the HERITAGE study [81], the Quebec Family Study [82], the CARDIA study [83] and the Tehran Lipid and Glucose study [84].

In a retrospective analysis of a two-year prospective study of 54 normal-weight Caucasian women participating in an exercise intervention, the dietary calcium:energy ratio and the dairy calcium:energy ratio were significant negative predictors of changes in both body weight and body fat [77]. There was a notable interaction between dietary calcium and energy intake in predicting changes in body fat, as calcium, but not energy, intake predicted changes in body weight and body fat for women below the median energy intake (1,876 kcal/day), while energy intake alone predicted changes in weight and fat in women at higher levels of energy intake. Further, the reported effects of calcium appeared to be specific to dairy sources, as dairy calcium predicted changes in body weight and fat, while non-dairy calcium did not [77]. An inverse relationship between energy-adjusted dietary calcium intake and body mass index was also reported in lactose tolerant, but not lactose intolerant, African-American women [78]. Although the reason for the lack of effect in the lactose intolerant group cannot be definitively inferred from this cross-sectional study, the lactose-intolerant group exhibited a uniformly low calcium intake, presumably due to aversion to dairy products, and the lack of women with adequate calcium intakes in this group therefore precluded a clear relationship emerging as it did for the lactose tolerant women.

While most studies reporting the relationship between dietary calcium and/or dairy and indices of adiposity are in adults, there have been a few studies in children and adolescents [72–76,85,86]. Although one study recently reported no relationship between dietary calcium or dairy consumption in a longitudinal assessment of adolescent females [85], the authors noted that dairy consumption was significantly higher for their study cohort compared to that reported by CSFII for a nationally representative survey of the same age group (428 vs. 269 g/day of milk and milk products). Moreover, overall reported median dairy intake was 2.9 servings of dairy and 827 mg of dairy-derived calcium per day. Accordingly, it is possible that

this cohort represented a relatively high dairy consuming population and therefore was sufficiently above a yet-to-be determined threshold of dairy intake to observe an effect on indices of adiposity. In contrast, several other studies of children and adolescents suggest a protective effect of dairy [73–76,86].

A significant inverse relationship between dietary calcium and body fat was reported in a five-year longitudinal study of preschool children studied from two months of age ($R^2 = 0.51$) [72]. The group subsequently extended these longitudinal findings to eight years of age [73]. Overall, in predictive equations that explain 26–34% of the variability in body fat, variations in dietary calcium explained 7–9% of the variability in adiposity [73]. Notably, these longitudinal data strongly suggest that dairy and calcium intake within the first year of life are significant inverse determinants of body fat levels at age 8 [72,73]. Consistent with these findings, longitudinal data from the Framingham Children's Study indicate that higher intakes of calcium early in life (ages 3–5) were associated with decreased gain of body fat over time (early adolescence), with dairy servings being more strongly correlated to reduced body fat than dietary calcium *per se*.

The associations between dairy intake and incidence of the major components of the insulin resistance syndrome (IRS), including obesity, was evaluated in a 10-year population based prospective study of 3,157 black and white adults [83]. Overweight individuals who consumed the most dairy products had a 72% lower incidence of IRS compared to those with the lowest dairy intakes. Moreover, the cumulative incidence of obesity in those who started the study in the overweight category was significantly reduced from 64.8% in those consuming the least amount of dairy foods to 45.1% in the highest dairy food consuming group. Notably, the inverse relationship between dietary calcium and either IRS or obesity incidence in the CARDIA study was explained solely by dairy intake and was not altered by adjustment for dietary calcium, indicating the presence of an additional effect of dairy beyond the mechanisms already cited for dietary calcium in modulating adiposity and obesity risk; this is consistent with both the experimental animal and clinical trial data which also suggest that other dairy components, in addition to calcium, contribute to an anti-obesity effect.

SUMMARY AND CONCLUSIONS

An anti-obesity effect of dietary calcium and dairy foods is now evident from animal studies, observational and population studies and clinical trials. It is important, however, to interpret these findings within the context of overall energy balance. For example, Berkey et al [87] recently reported that adolescents who consume excess calories from milk exhibit higher gains in body mass index than those who do not; however, when adjusted for energy intake, this effect was not evident. Consistent with this, the reported effects of calcium and dairy on body

weight and body composition demonstrate accelerated weight and fat loss on energy restricted diets and improvements in body composition with isocaloric substitution of dairy for other components of the diet. Accordingly, these data should not be interpreted to suggest that increasing dairy intake exerts an anti-obesity effect independent of energy balance.

It is also important to interpret these findings to place these findings within the context of optimal calcium and dairy intake. It appears that the effects of calcium on healthy weight management result from correcting suboptimal intakes and thereby preventing the endocrine response ($PTH - 1\alpha,25-(OH)_2D_3$ axis) which favors adipocyte energy storage. Accordingly, once adequate dietary calcium levels are achieved, minimal responses would be anticipated from further increases in calcium intake, and the available data support this concept. Similarly, the available data indicate that substantial improvements in adiposity are unlikely to result from increasing dairy intake beyond an optimal range (approximately three daily servings).

While there is a strong theoretical framework in place to explain the effects of dietary calcium on energy metabolism, the precise mechanisms whereby dairy products exert substantially greater effects than equivalent amounts of calcium are not yet clear. However, the additional dairy effect appears to be mediated, in part, by several bioactive compounds, including angiotensin converting enzyme inhibitors, the high concentration of branched chain amino acids in dairy protein and other components which have not yet been identified. These data provide the framework for the development of strategies to utilize dairy products and dairy ingredients for the prevention of overweight and obesity and, in conjunction with controlling energy balance, for effective weight management.

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