Milk, yoghurts and dental caries

Abstract

Milk is an important part of the human diet; after weaning cow's milk (bovine milk) predominates and this chapter considers the effect of bovine milk on dental caries. Yoghurt is a milk product and is also considered. Several published reviews have concluded that milk is of very low cariogenicity and may have some caries protective potential. For example, WHO reviewed the strength of the evidence in 2003 and concluded that a 'decreased risk' of dental caries from milk was 'possible'. The evidence comes from several types of study: epidemiological studies (interventional and observational), animal experiments, plaque pH studies, and *in vivo* and *in vitro* enamel and dentine slab experiments. More recent observational epidemiological studies have adjusted for potential confounders and have reported that milk consumption is associated with lower caries experience or incidence. Other types of study generally support this conclusion. Reasons for these favourable caries-related properties include the lower acidogenicity of lactose compared with other dietary sugars and the protective effects of calcium, phosphate, proteins and fats. There is less research concerning yoghurts but it is likely that the cariogenic potential of plain yoghurt is similar to milk. The addition of sucrose to milk increases caries risk. Milk is an important part of the human diet: for infants, it is the only source of essential nutrients. Its importance declines after weaning, so that the average consumption for all adolescents 11-18 years, in the UK from 2008 – 2012, was 141g of milk per day; males consuming 172g and females 109g [1]. Worldwide, using data from the Food and Agriculture Organisation, dairy production and supply (total and *per capita*) have increased since 1980 [2]. Consumption in China has tripled since 1982. In most countries, cow's (or 'bovine') milk is the most common milk consumed, and there is little information on dental properties of milk other than bovine. Human milk will not be considered in this review, but this has been considered elsewhere [3].

Some of the earliest investigations regarding milk and dental caries were carried out by Sprawson [4-8], who concluded that milk improved oral health. Mellanby and Coumoulos [9] attributed an improvement in children's teeth in London between 1929 and 1944 to improved diet, notably the introduction of cheap milk in 1934. In 1958, the Research Committee of the Canadian Dental Association [10] reviewed the evidence that milk consumption was associated with a reduction in caries incidence. Since then, much research, both clinical and non-clinical, has been published and, almost uniformly, milk is not seen as a cause of dental caries. The Department of Health, COMA [11] report on dietary sugars and human disease concluded that: "Although lactose alone is moderately cariogenic, milk also contains factors which protect against dental caries, so that milk without added sugars may be considered to be virtually non-cariogenic" (report section 6.7). In a review of diet, nutrition and chronic diseases, the World Health Organisation [12] classified evidence linking diet to several diseases including dental caries: the strength of the evidence for a 'decreased risk' of dental caries from milk was classified as 'possible'. Other reviews of the relation between milk consumption and oral health have been published in the USA [13-16], UK [17] and Sweden [18] and these comment on the favourable role of milk in the control of oral disease. Milk is considered a suitable vehicle for substances beneficial to oral health, principally fluoride [19] probiotics [20-22] and possibly vitamin D [15].

Review of evidence - milk and dental caries.

Eighty per cent of carbohydrate in milk is lactose. Various other components of milk have been considered to be protective against dental caries, namely the minerals, casein and other proteins and lipids. Cow's milk contains about 4.8g lactose per 100g milk. This amount could be sufficient to classify milk as cariogenic, but there is much evidence that lactose is the least cariogenic of the common dietary sugars [3]. In addition, the high concentrations of calcium and phosphorus in milk will help to prevent dissolution of enamel and other components, principally casein, are likely to be protective as well. Thus, it is possible that milk could be caries-promoting (due to the lactose content), caries-preventing, or somewhere between these two. This review considers these aspects.

Milk and dental caries – epidemiological studies.

There appears to have been just one intervention study and about 23 observational studies into the relationship between milk consumption and dental caries. While the majority of studies identify bovine milk as such, in a few studies 'dairy products' are listed: these studies have been included where it is clear that the majority of the 'dairy products' was bovine milk. As elsewhere in this review, formula milks are not included or considered.

The one intervention study was conducted a long time ago – in 1929 – and involved 319 4-16-yearold children, living with their parents in New York, USA [23]. The three intervention groups received supplements of milk, fresh fruit and vitamin D, and caries development over 13 months was compared with that of a control group. To quote the author: "With the cooperation of the parents and the supplementation of the diets with milk and fruit, we have proved that an adjustment of the diets and the arrest of caries are possible even under the poorest conditions." "Our good diets which contained a minimum of two or three pieces of fruit daily gave similar results to the diets which contained 1 quart [~ 1.14 litres] of milk without the additional fruit."

Two large-scale observational studies were conducted many years ago – one in the UK by Read and Knowles [24] and the other in New Zealand by Hewat [25]. The UK study involved 2,894 6-13-year-old children, while the New Zealand study involved over 2,000 children aged 7-16 years. In both studies, no relationship was found between milk consumption and caries experience, although a positive relation was recorded between consumption of sweets (sugar confectionery) and caries experience in the UK study. Read and Knowles [24] stated that "The value of milk in preventing caries does not appear to be supported." Lacto-vegetarian children in Australia were reported by Gillman and Lennon [26] to have lower caries experience than other children although, in their Finnish study, Linkosalo and Markkanen [27] reported no difference in caries experience.

In observational studies of diet and disease, the potential for confounding factors to influence relationships between diet and disease is substantial and should be taken into account in study design and data analysis. There have been four observational studies where the analyses of relationships between milk consumption and dental caries experience or increment were not controlled for possible confounding factors. Of these studies, Rugg-Gunn *et al.* [28] recorded a positive, but not statistically significant, relation between consumption of cow's milk and dental caries increment: the difference between the daily milk consumption in the high caries children (269ml) and the caries-free children (242ml) was small. Mattos-Graner *et al.* [29] reported that, in young Brazilian children, milk in bottles was not associated with dental caries unless sugar, or sugar and cereal, was added to the milk when the relationship became positive. Zita *et al.* [30] reported a weak negative correlation (-0.08) between milk intake and caries experience, while Potgieter *et al.* [31] reported "a marked and consistent drop in DMF [caries] rate with increase in the number of cups of milk consumed", in 864 10-16-year-old children in Connecticut, USA.

Greater computing power has allowed routine use of multivariate statistical analyses where the relationships between diet, namely milk consumption, and caries experience can be controlled for effects of possible confounding variables. Sixteen such studies have been published. In 15 of the 16 studies, milk consumption was associated with lower caries experience, although the results were not always clear-cut [32 – 46]. In the sixteenth study [47] "milk had a neutral association with caries" in their study of 642 pre-school children in Iowa, USA. In the study of Serra Majem *et al.* [32], 893 5-14-year-old Spanish children were examined: while skimmed milk had a "protective effect", the effect of whole milk was neutral. Petridou *et al.* in 1996 [33] reported that milk and dairy products were negatively associated with dental caries in 380 Greek adolescents aged 12-17 years. Petti *et al.* [34] reported an inverse relation between milk and caries that was strongest in children

with the highest frequency of sucrose consumption in 890 6-11-year-old Italian children who, on average, drank about 209ml of milk per day. Levy et al. [35] examined the relationship between caries experience of children in Iowa, USA, at age 5 years with diet during the previous years: higher milk consumption at 24-36 months was related to reduced risk of caries at 5 years. In a follow-up study of the same children (n=377) with mean age of 5 years, Chankanka et al [46] reported that greater milk consumption at meals was protective. Sohn et al [36] analysed data from the US NHANES III study (1988-1994) which involved nearly 6,000 children aged 2-10 years: "children with a high milk consumption pattern had a tendency toward lowest caries experience". The study of Kolker et al. [37] of 436 3-6-year-old children in Detroit, USA, revealed that increase in milk consumption was associated with lower caries experience. Levine et al. [38] related diet of 317 children in the UK at ages 7-11 years and at 11-14 years with caries experience at 11-14 years: moderate consumption of 'dairy products' was associated with less caries, although higher as well as lower frequency of consumption tended to be associated with greater caries experience. Llena and Forner [39] examined 369 6-10-year-old Spanish children: "in general, more frequent consumption of sugary foods was associated with a rise in the caries indices, while the children who consumed non-sugared milk and dairy products more often suffered lower caries rates". Lim et al. [40] analysed data from the Detroit Dental Health Project: 369 children aged 3 years or over were examined in 2002-3 and 2004-5. The authors concluded: "Children who consumed more soft drinks, relative to milk and 100% fruit juice, as they grew older were at a greater risk of developing dental caries." Johansson et al. [41] studied 1,206 1-5-year-old children in Boston, USA: caries prevalence was considerably lower (6%) in children who consumed milk with snacks compared with children who consumed a sugared drink with snacks (29%). In a study of 442 Australian Aboriginal young adults, Jamieson et al [42] reported that more frequent milk consumption was associated with lower caries experience. In 16 Inuit communities in northern Canada, Pacey et al. [43] found that higher frequency of milk consumption was protective against dental caries in children. In a study of 935 young Saudi Arabian women, Musaiger et al. [44], milk consumption was observed to protect against dental caries. Lempert et al. [45] found that milk consumption was associated with lower caries experience and 3-year caries incidence in young Danish children. A number of studies have reported that consumption of milk in children to be inversely related to consumption of added sugar drinks [48 - 50].

One study by Papas *et al* [51] has examined the relationship between consumption of 'dairy products' and root caries experience. This study involved 141 adults aged 47-83 years living in Boston, USA: while there was a strong relation between consumption of 'dairy products' and root caries, only cheese consumption was significantly associated with lower occurrence of root caries.

In summary, while there is an absence of controlled clinical trials, on balance the results of the above epidemiological studies strongly suggest that milk consumption is associated with lower experience of dental caries. This is especially so for the more recent studies which have examined their data using multivariate analysis. In 15 out of these 16 studies, higher milk (or dairy) consumption was associated with lower caries experience. The one study to examine root caries also recorded lower caries experience with higher consumption of dairy products, although the major effect in this study would appear to be from consumption of cheese.

Milk and dental caries – animal experiments.

Evidence from animal experiments not only indicates that cow's milk is non-cariogenic, but also strongly suggests an anti-cariogenic effect. The extensive studies of Schweigert et al. [52], Shaw [53] Dreizen et al. [54] and Stephan [55] labelled milk as non-cariogenic. Early indications that milk was not only non-cariogenic but anti-cariogenic [56, 57] were followed up by Reynolds and Johnson [58]. They found that supplementation of a cariogenic diet with cow's milk reduced substantially dental caries incidence and, importantly, that this was not due to reduced consumption of the cariogenic diet. In a review, Bowen and Pearson [59] came to the same conclusion. The study of Shaw et al. [57] also showed that the caries protective effect of milk was a post-eruptive effect (an intra-oral effect on erupted teeth) and not a pre-eruptive effect (an effect during tooth development). The caries-preventive effect of milk, in comparison with water, has also been reported by König [60], Bánóczy et al. [61] Stösser et al. [62] and Ghasempour et al [63]. A severe test of the cariogenic or cariostatic properties of milk was developed by Bowen et al. [64], using desalivated rats which are therefore much more caries-susceptible. In these experiments, the rats given milk or lactosereduced milk remained essentially caries-free, while those given sucrose or lactose in water developed caries. Using the same model a few years later, Bowen et al. [65] found a similar result regarding the very low caries potential of milk. The authors concluded "that [cow's] milk does not promote caries, even in the highly caries-conducive environment engendered", and "that milk or lactose-reduced milk can be used safely by hyposalivatory patients as a saliva substitute". In a study in Brazil, Peres et al. [66], also using desalivated rats, "concluded that cow's milk was not cariogenic". Bowen and Lawrence [67] compared the cariogenicity of cow's milk, human milk, Cola and honey in desalivated rats, once again concluding "that cow milk is essentially noncariogenic."

Caution is always needed when extrapolating the findings of animal experiments to the human situation, and this may be important when dietary phosphate is thought to play a caries-preventive role [3, 68] as is the case with milk. Although some caution is advisable, the results of the above numerous animal experiments give considerable weight to the evidence that bovine milk is non-cariogenic and may be anti-cariogenic.

Milk and dental caries – plaque pH experiments.

Several studies have shown that the fall in plaque pH after drinking milk is negligible [69–75]. In the studies of Rugg-Gunn *et al.* [73], 14 volunteers rinsed their mouths with cow's milk, human milk, lactose solution, or sucrose solution. Sucrose solution caused substantial falls in plaque pH, while the milks depressed plaque pH only slightly. An exception is the report by Birkhed [75] which showed that dental plaque microflora may adapt to lactose in milk leading to a greater ability to ferment this lactose following frequent milk consumption. In their experiment, 10 subjects rinsed six times a day for four weeks with low-fat milk: the fall in plaque pH with milk consumption was substantially greater at the end of the four weeks compared with measurements before the four weeks of rinsing. The authors speculate that adaption may have been less if the milk had been full-fat due to the protective effect of the higher casein content in full-fat milk. Naval *et al.* [76] demonstrated that milk consumed after a sugary food reduced the fall in plaque pH.

While plaque pH studies are a useful guide to the fermentability of sugars in foods, such experiments do not consider the presence and effect of substances in foods which may protect against dental

caries, such as, for example, calcium, phosphate and casein in milk.

Milk and dental caries - in vitro and in vivo studies.

Artificial caries-like lesions can be induced in small slabs of enamel by exposing their surface to acid buffers or by covering the slab with a biofilm which is then challenged with a variety of substrates. This method has been used both in the laboratory (in vitro), and in the mouth (in vivo) where they are held by a purpose-built intra-oral appliance, to examine the ability of foods to demineralise or remineralise tooth enamel. The early experiments of Weiss and Bibby [77] examined the in vitro effect of bovine milk (raw, pasteurised, whole or skimmed) and found that all the milks "reduced the solubility of enamel." Subsequently, Bibby et al. [78], using an artificial mouth test system (Orofax), found that the inclusion of milk solids reduced the cariogenicity of sugar-containing foods, while similar results were obtained by Thomson et al. [79] using an enamel slab intra-oral device. The same team reported that flavoured milk, containing 5% sucrose, caused considerably less demineralisation compared with an apple-based fruit juice [80] and bovine milk caused little demineralisation, much less than human milk [81]. Jensen et al. [82], using a similar system which held slabs of enamel or dentine in the mouth of volunteers, showed that milk (with three levels of fat content) was not cariogenic for enamel or dentine; Gedalia et al. [83] reported that milk remineralised enamel previously softened by Cola, and Walker et al. [84,85] reported that milk remineralised previously demineralised enamel slabs in vivo.

In addition to the plaque pH studies mentioned in the previous section, Jenkins and Ferguson [69] conducted *in vitro* comparisons of 4 % lactose solutions and cow's milk. They concluded that, within the limits of their experiments, their results "gave no grounds for suggesting that milk has a local effect on the teeth which would favour caries", and suggested that the negligible fall in plaque pH was partly due to milk's high buffering power, and the low level of dissolution of test enamel was due to the protective action of milk's high levels of calcium and phosphate. Rugg-Gunn *et al.* [73] also reported that both cow's milk and human milk protected enamel from dissolution in *in vitro* experiments, compared with sucrose or lactose, but that human milk was less protective than cow's milk, as would be expected from their different calcium and phosphorus content.

Eight further *in vitro* studies have investigated the caries inducing or caries preventive effect of milk. McDougall [86] showed that (a) demineralisation of enamel in an acid buffer was reduced by intermittent exposure to milk, and (b) that milk aided the remineralisation of demineralised enamel. Mor and Rodda [87] reported that milk remineralised artificially-induced lesions in enamel slabs. Arnold *et al.* [88] showed that milk inhibited enamel demineralisation compared with saline or a remineralising solution, and Ivancakova *et al.* [89] and Arnold *et al.* [90] reported that milk reduced the rate of root caries progression. Ongtenco *et al.* [91] reported that milk reduced demineralization of enamel slabs compared with deionised water.

Not all *in vitro* studies have suggested that milk resists demineralisation or encourages remineralisation. Prabhakar *et al.* [92] incubated enamel slabs with human or bovine milk *in vitro* and concluded that plain bovine milk was relatively cariogenic in the absence of saliva. Likewise, Muñoz-Sandoval *et al.* [93] and Giacaman and Muñoz-Sandoval [94] report that bovine milk caused demineralisation of enamel and dentine slabs and concluded that "Whole milk... may be less

cariogenic than sucrose but not anticariogenic." and "Bovine milk should not be considered cariessafe, mainly for root dentin." They also reported that skimmed and semi-skimmed milks are associated with more demineralisation that whole milk. These differences may be explained by differences in experimental methods. While the studies discussed in the previous paragraph used cell-free demineralisation, the three studies discussed in this paragraph used active biofilms, for example, of Streptococcus mutans, which might result in a greater challenge.

Although there is not total agreement in conclusions, both *in vitro* and *in vivo* enamel slab experiments suggest that bovine milk has little ability to cause demineralisation and, indeed, is capable of remineralising previously demineralised enamel.

The actions of the constituents of milk.

The principal constituents of bovine milk, other than carbohydrate (4.8%), are fat (up to 3.8%), protein (3%), calcium (118mg/100g) and phosphorus (96mg/100g), with some small variation. It has been mentioned previously that lactose is the least cariogenic dietary sugar. When Thomson *et al.* [81] increased the concentration of lactose in bovine milk from 5% to 7% (the level in human milk) no increase in the already low cariogenic potential was observed.

Oral clearance is influenced by the ingredients of foods as well as by salivary flow, action of the tongue, cheeks and lips, and by other factors. One ingredient which accelerates oral clearance is fat [95 - 98]. This is probably due to a physical action of holding all the particles together.

Proteins are adsorbed well onto enamel surfaces. Weiss and Bibby [77] investigated the ability of milk to reduce demineralisation in an *in vitro* enamel slab experiment. They found that milk components adhered to the enamel surface and that these could not be removed by washing with water. The protective effect was, however, removed by a protein solvent, and restored by application of casein alone. Muhler [99] showed that the protective effect, which he surmised was organic, was removed by heat-treating the milk powder prior to feeding to the experimental animals: this would de-nature proteins. Pearce and Bibby [100] tested 11 proteins and found that casein and globulins were adsorbed in greatest amounts, and albumins the least.

Casein is a phosphoprotein and represents about 87% of all proteins present in milk: it is considered to be one of the main ingredients responsible for the caries-protective action of milk. Decreases in the development of dental caries in rats have followed increases in the casein content of their diet [101-104]. The reasons for the caries preventive effect of caseins have been studied [105-108]. Casein appears to prevent adherence of salivary components and bacteria to enamel and pellicle, and to reduce the activity of glucosyltransferase, thus reducing glucan formation and plaque adherence. In a series of rat caries experiments, Guggenheim *et al.* [109] demonstrated the marked caries preventive effect of 'milk micellar casein' when incorporated into a cariogenic diet. Large reductions in the proportion of *S. sobrinus* were observed leading the authors to conclude that the micellar casein interfered with the adhesion of some plaque bacteria. Research into the favourable properties of casein, particularly its ability to concentrate calcium and phosphate in plaque, has led to the development of a compound known as CPP-ACP [110-112].

Harper et al. [68] questioned whether casein was the most caries-protective constituent in milk, following experiments in rats which tested the caries-reducing potential of three mineral-rich milk concentrates with various levels of whey protein, calcium, and phosphorus, but negligible levels of casein. The results suggested that considerable protection could be afforded by calcium and phosphate compounds in the absence of casein. In another animal experiment, Beighton et al. [113] concluded that the increased cariogenic potential of freeze-dried milk compared with whole milk could be due to lower calcium and phosphate concentrations in their freeze-dried milk. While the caries protective effect of phosphates in the diets of rats needs to be interpreted with care [3, 68] the results agree with other in vitro studies which demonstrate the favourable role of the high concentrations of calcium and phosphorus in milk [69, 73]. Using the in vivo enamel slab model, Thomson et al. [81] reported that when the calcium and phosphorus concentrations in human milk (~ 22mg/100g and 10mg/100g, respectively) were raised to the concentrations found in bovine milk (~114mg/100g and 96mg/100g, respectively) demineralisation of enamel was reduced by 70%. In agreement with these findings, Bowen and Lawrence [67] stated: "It seems, therefore, that the difference in the cariogenicity of the milk resides for the most part in the mineral content. Clearly, other factors such as casein content cannot be discounted." Support for the protective role of the mineral content of milk came from the studies of Grenby et al. [114] which showed, in in vitro experiments, that the "removal of lactose, fat, casein and other proteins had little influence on the protective effect of the milk fractions. Besides calcium and phosphorus, milk contains other more powerful protective factors against demineralisation, which they identified as proteose-peptone fractions 3 and 5." [114]. The in vitro studies of Shetty et al [115] suggested that both whey protein and casein were protective: casein more so in human milk and whey protein more so in bovine milk. The protective role of other components of milk has received some attention. Oho and workers reported that lactoferrin reduced binding of Streptococcus mutans to other cells [116] and to salivacoated hydroxyapatite beads [117], while Aimutis [111] discusses possible roles of lysozyme and lactoperoxidase, present in milk, in reducing cariogenic oral microflora.

The non-cariogenic and protective properties of bovine milk would appear to be due to several factors. First, the lactose content is low and of limited cariogenic potential; second, the high calcium and phosphate content resists demineralisation and aids remineralisation of enamel and dentine; third, casein reduces demineralisation of tooth tissue and, finally, other components of milk may reduce the ability of plaque microflora to adhere to enamel and produce acids.

Yoghurts.

Fermentation of milk leads to production of lactic acid and the resulting fall in pH inhibits growth of many pathogenic organisms. One of the most important of these fermented foods is yoghurt which has been traditionally fermented with *Lactobacillus bulgaricus*. The lactose content reduces substantially during fermentation although some galactose remains: other constituents are unchanged [118]. A high proportion of commercial yoghurts contain added sugars and flavouring, complicating interpretation of research into cariogenicity.

Two observational epidemiological studies have been identified. Marshall *et al.* [47] compared the diets of children in Iowa, USA, between the ages to 1 and 5 years with caries experience at about 5-6 years. While milk consumption had a neutral relation with caries, "high intakes of non-milk dairy

foods were associated with an increased extent of caries." However, they continue: "Yoghurt and dairy desserts are made with added sugar (e.g., sucrose and/or high-fructose corn syrup) which is a substrate for oral bacteria." The second study was by Tanaka *et al.* [119] who compared the diets of 2058 3-year-old Japanese children with their caries experience. They reported: "compared with yogurt consumption in the lowest tertile, its intake at the highest level was significantly associated with a lower prevalence of caries, showing a clear dose-response relationship." There is no record of these products being sweetened. An *in vivo* enamel slab study by Jensen *et al.* [82] found that consumption of sweetened (strawberry) yogurt increased demineralisation of enamel and dentine compared with milk. However, the sugars content of this (sweetened) product was not given. In an *in vitro* enamel slab experiment, Ferrazzano *et al.* [120] reported that yogurt with enhanced levels of naturally-occurring casein phosphopeptides inhibited demineralisation and promoted remineralisation of dental enamel.

Information on the dental effects of yoghurt is limited. As long as sugars are not added, there is no reason for suspecting that the effects of yoghurt would differ from that of milk.

Addition of sucrose and cocoa to milk.

In some countries, it is common to flavour milk, especially milk for children, with sugar and other flavourings. In most countries, the sugar added is sucrose and the most common other flavouring is cocoa. It is reasonable to assume that adding sucrose to milk will increase cariogenicity, but at what concentration will the added sucrose overcome the caries-protective properties of plain milk, is an interesting question. The question is complicated by the knowledge that cocoa itself has caries-protective properties [3, 121].

The effect of adding sucrose to cow's milk has been investigated in a variety of studies. In an uncontrolled, observational epidemiological survey, Mattos-Graner *et al.* [29] recorded that children who had drunk milk with added sugar had higher caries experience than children who had drunk milk with added sugar. Dunning and Hodge [122] reported results of a two-year clinical trial in American children and young adults. Caries increment was slightly higher (of borderline statistical significance) in children drinking milk with 6% sugar added, compared with children drinking plain milk. In a case-control study of rampant (extensive) caries in young Chinese children, Ye *et al.* [123] reported that, after multivariate analyses, bottle feeding with sugar-containing bovine milk was associated with rampant caries.

Shaw *et al.* [57] conducted animal experiments on milk-based products including 'chocolate drink' and 'chocolate milk'. There was a "general trend" for chocolate drink and chocolate milk to be less protective than plain milk. The authors did not give the sugars content of these products, but comment, "one would tend to suspect the reason for these differences to be the increased carbohydrate content of the preparations containing chocolate rather than chocolate itself." Also, using the rat caries model, Bowen and Pearson [59] studied the effect of adding 10% sucrose or 10% fructose to milk. There was little difference in cariogenicity between these two sugars and both were more cariogenic than milk alone but less cariogenic than 10% sucrose in water. A further aspect of this experiment showed that 4% lactose in water was of very low cariogenicity. In a second series of experiments, these authors found that caries development was similar in groups of animals

receiving 2%, 5% or 10% sucrose in milk. This caries development was greater than that recorded for rats receiving milk alone and less than that recorded for rats receiving 10% sucrose in water. From these results, they concluded "that lactose has little capacity to promote caries"; "It is clear that the addition of as little as 2% sucrose to milk enhances the caries activity of milk even though the milk-sucrose solutions are significantly less cariogenic than the water-sucrose solutions" and "the practice of adding any sugar to milk should be discouraged." More recently, Al-Jobair and Khounganian [124] reported rat caries studies which showed that flavoured milks (chocolate, strawberry or banana) containing 5-6% added sucrose, were more cariogenic than plain milk but less cariogenic than 10% sucrose in water.

A small and statistically non-significant increase in enamel softening was recorded by Thomson *et al.* [79] when 5% sucrose was added to cow's milk, in an intra-oral enamel slab experiment. However, softening was much greater when the enamel was exposed to 5% sucrose in water. Two plaque pH studies have indicated that the addition of 5% and 10% sucrose to cow's milk increased acidogenicity, but this increase was less than when sucrose was added to water [73,125]. In an *in vitro* enamel slab experiment, Bibby *et al.* [78] reported that "commercial chocolate milk was much more destructive" than whole milk. The chocolate milk contained 6% added sugars (75% corn syrup, 25% sucrose). In another, more recent, *in vitro* study, Prabhakar *et al.* [92] compared the cariogenic potential of various milks. They commented that: "It is a common household practice to add sugar to bovine milk to make it more palatable for infants [in India]". They concluded that "it was clearly evident that supplementation with an external carbohydrate source (sucrose) enhanced the cariogenic potential of milk." This is supported by the *in vitro* enamel slab and dentine slab experiments of Giacaman and Munoz-Sandoval [94] who reported that the addition of 10% sucrose increased the cariogenic potential markedly.

Two studies have compared the effect on caries development of cocoa with any other flavouring. Using the rat caries model, Al-Jobair and Khounganian [124] suggested that chocolate-flavoured milk was less cariogenic than strawberry or banana-flavoured milks. In the intra-oral enamel slab experiment of Thomson *et al.* [79] mentioned above, there was a hint that milk flavoured with cocoa caused less enamel softening than milk flavoured with strawberry. In all these studies, milks contained about 5% sucrose.

Thus, it is probable that adding sugar to milk increases risk of caries development: this evidence comes from several types of study – human, animal and laboratory. The concentration of added sugar at which caries development might begin is uncertain but may be as low as 2%. There is too little information on the effect of cocoa flavouring to draw conclusions.

Conclusion

Bovine milk can be considered non-cariogenic; its anti-cariogenic role is possible. Evidence from several types of study contributes to these conclusions. While early epidemiological evidence is equivocal, more recent observational studies which have used multivariate analyses have strongly favoured milk being associated with lower caries experience. Information from animal experiments shows clearly milk's non-cariogenicity as well as the caries-protective role of milk – some caution in interpreting this latter finding in animal experiments is needed. *In vivo* and *in vitro* demineralisation

and remineralisation (enamel slab) experiments also tend to indicate the low cariogenic potential of milk and its caries-protective role. These actions would appear to be due to (a) lactose being the least cariogenic of dietary sugars, (b) the protective role of casein and possibly fats, and (c) the protective role of calcium and phosphorus. There would appear to be no reason why yoghurt should be any different from milk with regard to dental effects. The favourable dental profile of milk and yoghurt is likely to be compromised by the addition of sucrose.

Compiled by Professor Andrew Rugg-Gunn and Margaret Woodward June 2017

References

- Public Health England: National Diet and Nutrition Survey. Results from Years 1, 2, 3 and 4 (combined) of the Rolling Programme (2008/2009 – 2011/2012). London, Public Health England, 2014.
- 2. Wang Y, Li S: Worldwide trends in dairy production and consumption and calcium intake: is promoting consumption of dairy products a sustainable solution for inadequate calcium intake? Food Nutr Bull 2008;29:172-185.
- 3. Rugg-Gunn AJ: Nutrition and Dental Health. Oxford, Oxford University Press, 1993.
- 4. Sprawson E: Freedom from and immunity to dental caries. Br Dent J 1932;52:174-177.
- 5. Sprawson E: Concerning raw milk and immunity to dental caries. Br Dent J 1932;52:642-646.
- 6. Sprawson E: Preliminary investigation of the influence of raw milk on teeth and lymphoid tissue. Proc Roy Soc Med 1932;25:11.
- 7. Sprawson E: Diet and dental caries. Br Dent J 1934;56:125-131.
- 8. Sprawson E: Foods and feeding as they affect teeth and their environment. Br Dent J 1947;83:227-235.
- 9. Mellanby M, Coumoulos H: The improved dentition of 5-year-old London school-children. Br Med J 1944;1:837-840.
- 10. Canadian Dental Association: Statement of the relation of milk consumption to dental caries. The Research Committee. J Can Dent Assoc 1958;24:558.
- 11. Department of Health: Dietary sugars and human disease. Report on health and social subjects 37. London, HMSO, 1989.
- 12. World Health Organisation: Diet, nutrition and the prevention of chronic diseases.

Technical Report Series 916. Geneva, WHO, Geneva, 2003.

- 13. Tinanoff N: Dietary determinants of dental caries and dietary recommendations for preschool children. J Publ Health Dent 2000;60:197-206.
- 14. Merritt J, Qi F, Shi W: Milk helps build strong teeth and promotes oral health. Calif Dent Assoc J 2006;34:361-366.
- 15. Dietrich T, Sheshah A, Nunn M: Dairy products and oral health. In: Wilson M (ed): Food constituents and oral health. Cambridge, Woodhead Publishing, 2009, pp 152-162.
- 16. Dror DK, Allen LH: Dairy product intake in children and adolescents in developed countries: trends, nutritional contribution, and a review of association with health outcomes. Nutr Rev 2014;72:68-81.
- Moynihan P: Foods and factors that protect against dental caries. Nutr Bull 2000;25:281-286.
- 18. Johansson I: Milk and dairy products: possible effects on dental health. Scand J Nutr 2002; 46:119-122.
- 19. Bánóczy J, Petersen PE, Rugg-Gunn AJ: Milk fluoridation for the prevention of dental caries, ed 2. Geneva, World Health Organization, 2009.
- 20. Meurman J H: Probiotics and oral health. In: Wilson M (ed): Food constituents and oral health. Cambridge, Woodhead Publishing, 2009, pp 421-432.
- 21. Stecksén-Blicks C, Sjöström I, Twetman S: Effect of long-term consumption of milk supplemented with probiotic lactobacilli and fluoride on dental caries and general health in pre-school children: a cluster-randomized study. Caries Res 2009;43:374-381.
- 22. Petersson L, Hakestam U, Baigi A, Magnusson K, Twetman S: Root caries development after intake of fluoridated milk and probiotics. Acta Odontol Scand 2011;69:321-327.
- 23. Brodsky RH: Factors in the etiology and arrest of dental caries. J Am Dent Assoc 1933;20: 1440-1458.
- 24. Read TT, Knowles EM: A study of the diet and habits of school children in relation to freedom from or susceptibility to dental caries. Br Dent J 1938;64:185-197.
- 25. Hewat RET: Field studies on dental caries in New Zealand; interim report. NZ Dent J 1948; 44:163-191.
- Gillman J, Lennon D: The biology of children of Hopewood House, Bowral, NSW, II. Observations extending over five years (1952-1956 inclusive). 4. Diet survey. Aust Dent J 1958;3:378-382.

- 27. Linkosalo E, Markkanen H: Dental erosions in relation to lactovegetarian diet. Scand J Dent Res 1985;36:56-60.
- 28. Rugg-Gunn AJ, Hackett AF, Appleton DR, Jenkins GN, Eastoe JE: Relationship between dietary habits and caries increment assessed over two years in 405 English adolescent schoolchildren. Archs Oral Biol 1984;29:983-992.
- 29. Mattos-Graner, RO, Zelante F, Line RCSR, Mayer MPA: Association between caries prevalence and clinical, microbiological and dietary variables in 1.0 to 2.5-year-old Brazilian children. Caries Res 1998;32:319-23.
- 30. Zita AC, McDonald RE, Andrews AL: Dietary habits and the dental caries experience in 200 children. J Dent Res 1959;38:860-865.
- 31. Potgieter M, Morse EH, Erlenbach FM, Dall R: The food habits and dental status of some Connecticut children. J Dent Res 1956;35:638-644.
- 32. Serra Majem L, Closas RG, Ramón JM, Manau C, Cuenca E, Krasse B: Dietary habits and dental caries in a population of Spanish schoolchildren with low levels of caries experience. Caries Res 1993;27:488-494.
- 33. Petridou E, Athanassouli T, Panagopoulos H, Revinthi K: Sociodemographic and dietary factors in relation to dental health among Greek adolescents. Community Dent Oral Epidemiol 1996;24:307-311.
- 34. Petti S, Simonetti R, Simonetti D'Arca A: The effect of milk and sucrose consumption on caries in 6-11-year-old Italian schoolchildren. Eur J Epidemiol 1997;13:659-664.
- 35. Levy SM, Warren JJ, Broffitt B, Hillis SL, Kanellis MJ: Fluoride, beverages and dental caries in the primary dentition. Caries Res 2003;37:157-165.
- 36. Sohn W, Burt BA, Sowers MR: Carbonated soft drinks and dental caries in the primary dentition. J Dent Res 2006;85:262-266.
- 37. Kolker JL, Yuan Y, Burt BA, Sandretto AM, Sohn W, Lang SW, Ismail AI: Dental caries and dietary patterns in low-income African American children. Pediatr Dent 2007;29:457-464.
- 38. Levine RS, Nugent ZJ, Rudolf MCJ, Sahota P: Dietary patterns, toothbrushing habits and caries experience of schoolchildren in West Yorkshire, England. Community Dent Health 2007;24:82-87.
- 39. Llena C, Forner L: Dietary habits in a child population in relation to caries experience. Caries Res 2008;42:387-393.

- 40. Lim S, Sohn W, Burt BA, Sandretto AM, Kolker JL, Marshall TA, Ismail AI: Cariogenicity of soft drinks, milk and fruit juice in low-income African-American children. J Am Dent Assoc 2008;139:959-967.
- 41. Johansson I, Holgerson PL, Kressin NR, Nunn ME, Tanner AC: Snacking habits and caries in young children. Caries Res 2010;44:421-430.
- 42. Jamieson LM, Roberts-Thomson KF, Sayers SM: Dental caries risk indicators among Australian Aboriginal young adults. Community Dent Oral Epidemiol 2010;38:213-221.
- 43. Pacey A, Nancarrow T, Egeland GM: Prevalence and risk factors for parental-reported oral health of Inuit preschoolers: Nunavut Inuit Child Health Survey, 2007-2008. Rural Remote Health; 2010;10:1368.
- 44. Musaiger AO, Al-Mannai M, Abduljawad E: Association of oral hygiene habits and food intake with the risk of dental caries among undergraduate university women in Saudi Arabia. Int J Adolesc Med Health 2014; 26:585-589.
- 45. Lempert SM, Christensen LB, Froberg K, Raymond K, Heitmann BL: Association between dairy intake and caries among children and adolescents. Results from the Danish EYHS follow-up study. Caries Res 2015;49:251-258.
- 46. Chankanka O, Levy SM, Marshall TA, Cavanaugh JE, Warren JJ, Broffitt B, Kolker JL: The associations between dietary intakes from 36 to 60 months of age and primary dentition non-cavitated caries and cavitated caries. J Public Health Dent 2015;75:265-273.
- Marshall TA, Levy SM, Broffitt B, Warren JJ, Eichenberger-Gilmore JM, Burns TL, Stumbo PJ. Dental caries and beverage consumption in young children. Pediatrics 2003;112:184-191.
- 48. Harnack L, Stang J, Story M: Soft drink consumption among US children and adolescents: nutritional consequences. J Am Diet Assoc 1999;99:436-441.
- 49. Skinner JD, Carruth BR, Moran J, Houck K, Coletta F: Fruit juice intake is not related to children's growth. Paediatrics 1999;103:58-64.
- 50. Marshall TA, Eichenberger-Gilmore JM, Broffitt B, Stumbo PJ, Levy SM: Diet quality in young children is influenced by beverage consumption. J Am Coll Nutr 2005;24:65-75.
- 51. Papas AS, Joshi A, Palmer CA, Giunta JL, Dwyer JT: Relationship of diet to root caries. Am J Clin Nutr 1995;61(suppl):423S-429S.
- 52. Schweigert BS, Shaw JH, Zeppelin M, Elvehjem CA: Dental caries in the cotton rat, VI. The effect of the amount of protein, fat and carbohydrate in the diet on the incidence and extent of carious lesions. J Nutr 1946;31:439-447.

- 53. Shaw JH: Effect of dietary composition on tooth decay in the albino rat. J. Nutr 1950;41: 13-24.
- 54. Dreizen S, Dreizen JG, Stone RE: The effect of cow's milk on dental caries in the rat. J Dent Res 1961;40:1025-1028.
- 55. Stephan RM: Effects of different types of human foods on dental health in experimental animals. J Dent Res 1966;45:1551-1561.
- 56. Sperling G, Lovelace F, Barnes LL, Smith CAH, Saxton JA Jr, McCay CM: Effect of long time feeding of whole milk diets to white rats. J Nutr 1955;55:399-414.
- 57. Shaw JH, Ensfield BJ, Wollman DH: Studies on the relation of dairy products to dental caries in caries-susceptible rats. J Nutr 1959;67:253-273.
- 58. Reynolds EC, Johnson H: Effect of milk on caries incidence and bacterial composition of dental plaque in the rat. Arch Oral Biol 1981;26:445-451.
- 59. Bowen WH, Pearson SK: Effect of milk on cariogenesis. Caries Res 1993;27:461-466.
- 60. König KG: Pre- and post-eruptive inhibition of experimental rat caries by fluoride administered in water, milk and food. Helv Odont Acta 1960;4:66-71.
- 61. Bánóczy J, Ritlop B, Solymosi G, Gombik A, Adatia A: Anticariogenic effect of fluoridated milk and water in rats. Acta Physiol Hungarica 1990;76:341-346.
- 62. Stösser L, Kneist S, Grosser W: The effects of non-fluoridated and fluoridated milk on experimental caries in rats. Adv Dent Res 1995;9:122-124.
- 63. Ghasempour M, Rajabnia R, Ashrafpour M, Ehsani A, Moghadamnia AA, Gharekhani S, Shahandashti EF, Bagheri M: Effect of milk and yogurt on streptococcus sobrinus counts and caries score in rats. Dent Res J (Isfahan) 2015;12:569-73.
- 64. Bowen WH, Pearson SK, VanWuyckhuyse BC, Tabak LA: Influence of milk, lactosereduced milk, and lactose on caries in desalivated rats. Caries Res 1991;25:283-286.
- 65. Bowen WH, Pearson SK, Rosalen PL, Miguel JC, Shih AY: Assessing the cariogenic potential of some infant formulas, milk and sugar solutions. J Amer Dent Ass 1997;128:865-871.
- Peres RC, Coppi LC, Franco EM, Volpato MC, Groppo FC, Rosalen PL: Cariogenicity of different types of milk: an experimental study using animal model. Braz Dent J 2002; 13:27-32.

- 67. Bowen WH, Lawrence RA: Comparison of the cariogenicity of Cola, honey, cow milk, human milk, and sucrose. Pediatrics 2005;116:921-926.
- 68. Harper DS, Osborn JC, Clayton R, Hefferen JJ: Modification of food cariogenicity in rats by mineral-rich concentrates in milk. J Dent Res 1987;66:42-45.
- 69. Jenkins GN, Ferguson DB: Milk and dental caries. Br Dent J 1966;120:472-477.
- 70. Frostell G: Effects of milk, fruit juices and sweetened beverages on the pH of dental plaques. Acta Odont Scand 1970;28:609-622.
- 71. Edgar WM, Bibby BG, Mundorff S, Rowley J: Acid production in plaques after eating snacks: modifying factors in foods. J Am Dent Ass 1975;90:418-425.
- 72. Mor BM, McDougall WA, Effects of milk on pH of plaque and salivary sediment and the oral clearance of milk. Caries Res 1977;11:223-230.
- 73. Rugg-Gunn AJ, Roberts GJ, Wright WG: The effect of human milk on plaque *in situ* and enamel dissolution *in vitro* compared with bovine milk, lactose and sucrose. Caries Res 1985;19:327-334.
- 74. Saeed S, Al-Tinawi M: Evaluation of acidity and total sugar content of children's popular beverages and their effect on plaque pH. J Ind Soc Pedodont Prev Dent 2010;28:189-192.
- 75. Birkhed D, Imfeld T, Edwardsson S: pH changes in human dental plaque from lactose and milk before and after adaption. Caries Res 1993;27:43-50.
- 76. Naval S, Koerber A, Salzmann L, Punwani I, Johnson BR, Wu CD: The effects of beverages on plaque acidogenicity after a sugary challenge. J Am Dent Assoc 2013;144:815-822.
- 77. Weiss ME, Bibby BG: Effects of milk on enamel solubility. Arch Oral Biol 1966;11:49-57.
- 78. Bibby BG, Huang CT, Zero D, Mundorff SA, Little MF: Protective effect of milk against *in vitro* caries. J Dent Res 1980;59:1565-1570.
- 79. Thomson ME, Dever JG, Pearce EIF: Intra-oral testing of flavoured sweetened milk. NZ Dent J 1984;80:44-46.
- Dever JG, Thomson ME, Hampton M: Fruit juice and flavoured milk: effects on enamel in an intra-oral model. NZ Dent J 1987;83:7-10.
- 81. Thomson ME, Thomson CW, Chandler NP: *In vitro* and intra-oral investigations into the cariogenetic potential of human milk. Caries Res 1996;30:434-438.

- Jensen ME, Donly K, Wefel JS: Assessment of the effect of selected snack foods on the remineralisation/demineralisation of enamel and dentine. J Contemp Dent Pract 2000;1: 1-12.
- 83. Gedalia I, Dakuar A, Shapira L, Lewinstein I, Goultschin J, Rahamin E: Enamel softening with Coca-Cola and rehardening with milk or saliva. Am J Dent 1991;4:120-122.
- 84. Walker G, Cai F, Shen P, Reynolds C, Ward B, Fone C, Honda S, Koganei M, Oda M, Reynolds E: Increased remineralization of tooth enamel by milk containing added casein phosphopeptide-amorphous calcium phosphate. J Dairy Res 2006;73:74-78.
- 85. Walker GD, Cai F, Shen P, Bailey DL, Yuan Y, Cochrane NJ, Reynolds C, Reynolds EC: Consumption of milk with added casein phosphopeptide-amorphous calcium phosphate remineralizes enamel subsurface lesions *in situ*. Aust Dent J 2009;54:245-249.
- 86. McDougall WA: Effect of milk on enamel demineralisation and remineralisation *in vitro*. Caries Res 1977;11:166-172.
- 87. Mor BM, Rodda JC: *In vitro* remineralisation of artificial caries-like lesions with milk. NZ Dent J 1983;79:10-15.
- Arnold WH, Cerman M, Neuhaus K, Gaengler P: Volumetric assessment and quantitative element analysis of the effect of fluoridated milk on enamel demineralisation. Arch Oral Biol 2003;48:467-473.
- 89. Ivancakova R, Hogan MM, Harless JD, Wefel JS: Effect of fluoridated milk on progression of root surface lesions *in vitro* under pH cycling conditions. Caries Res 2003;37:166-171.
- 90. Arnold WH, Heidt BA, Kuntz S, Naumova EA: Effects of fluoridated milk on root dentin remineralization. PLoS One 2014;9:e104327.
- 91. Ongtenco KL, Anthonappa RP, Itthagarun A, King NM, Lalloo R, Nair RG: Remineralization of initial enamel carious lesions using fluoridated milk *in vitro*. Acta Odontol Scand 2014; 72:737-744.
- 92. Prabhakar AR, Kurthukoti AJ, Gupta P: Cariogenicity and acidogenicity of human milk, plain and sweetened bovine milk: an *in vitro* study. J Clin Pediatr Dent 2010;34:239-248.
- 93. Muñoz-Sandoval C, Muñoz-Cifuentes MJ, Giacaman RA, Ccahuana-Vasquez RA, Cury JA. Effect of bovine milk on Streptococcus mutans biofilm cariogenic properties and enamel and dentin demineralization. Pediatr Dent 2012;34:e197-201.
- 94. Giacaman RA, Muñoz-Sandoval C: Cariogenicity of different commercially available bovine milk types in a biofilm caries model. Pediatr Dent 2014;36:1e-6e.

- 95. Bibby BG, Goldberg HJV, Chen E: Evaluation of caries-producing potentialities of various foods. J Am Dent Ass 1951;42:491-509.
- 96. Swenander Lanke L: Influence on salivary sugar of certain properties of foodstuffs and individual oral conditions. Acta Odontol Scand 1957;15(Suppl 23):1-156.
- 97. Frostell G: Dental plaque pH in relation to intake of carbohydrate products. Acta Odont. Scand 1969;27:3-29.
- 98. Brudevold F, Kashket S, Kent RL: The effect of sucrose and fat in cookies. J Dent Res 1990; 69:1278-1282.
- 99. Muhler JC: The effect of powdered milk on dental caries in the rat. J Nutr 1957;61:281-287.
- 100. Pearce EIF, Bibby BG: Protein adsorption on bovine enamel. Archs Oral Biol 1966;11:329-336.
- 101. Bavetta LA, McClure FJ: Protein factors and experimental rat caries. J Nutr 1957;63:107-117.
- 102. Holloway PJ, Shaw JH, Sweeney EA: Effects of various sucrose:casein ratios in purified diets on the teeth and supporting structures of rats. Arch Oral Biol 1961;3:185-200.
- 103. Reynolds EC, Black CL: Reduction of chocolate's cariogenicity by supplementation with sodium caseinate. Caries Res 1987;21:445-451.
- 104. Reynolds EC, Black CL: Confectionery composition and rat caries. Caries Res 1987;21:538-545.
- 105. Vacca-Smith AM, Van Wuyckhuyse BC, Tabak LA, Bowen WH: The effect of milk and casein proteins on the adherence of streptococcus mutans to saliva-coated hydroxyapatite. Arch Oral Biol 1994;39:1063-1069.
- 106. Vacca-Smith AM, Bowen WH: The effect of milk and kappa-casein on streptococcal glucosyltransferase. Caries Res 1995;29:498-506.
- 107. Vacca-Smith AM, Bowen WH: The effects of milk and kappa-casein on salivary pellicle formed on hydroxyapatite discs *in situ*. Caries Res 2000;34:88-93.
- 108. Danielsson Niemi L, Hernell O, Johansson I: Human milk compounds inhibiting adhesion of mutans streptococci to host ligand-coated hydroxyapatite. Caries Res 2009;43:171-178.
- 109. Guggenheim B, Schmid R, Aeschlimann JM, Berrocal R: Powdered milk micellar casein prevents oral colonization by Streptococcus sobrinus and dental caries in rats: a basis for

caries-protective effect of dairy products. Caries Res 1999;33:446-454.

- 110. Reynolds EC: Anticariogenic complexes of amorphous calcium phosphate stabilized by casein phosphopeptides: a review. Spec Care Dent 1998;18:8-16.
- 111. Aimutis WR: Bioactive properties of milk proteins with particular focus on anticariogenesis. J Nutr 2004;134:989S-995S.
- 112. Cochrane NJ, Reynolds EC: Casein phosphopeptides in oral health. In: Wilson M (ed): Food constituents and oral health. Cambridge, Woodhead Publishing, 2009, pp 185-224.
- 113. Beighton D, McIntosh HA, McDougall WA: Bacteriological studies of the effects of cow's milk on dental plaque and dental caries in rats. J Appl Microbiol 1979;47:255-262.
- 114. Grenby TH, Andrews AT, Mistry M, Williams RJH: Dental caries-protective agents in milk and milk products: investigations *in vitro*. J Dent 2001;29:83-92.
- 115. Shetty V, Hegde AM, Nandan S, Shetty S: Caries protective agents in human milk and bovine milk: an *in vitro* study. J Clin Pediatr Dent 2011;35:389-392.
- 116. Mitoma M, Oho T, Shimazaki Y, Koga T: Inhibitory effect of bovine milk lactoferrin on the interaction between a streptococcal surface protein antigen and salivary agglutinin. J Biol Chem 2001;276:18060-18065.
- 117. Oho T, Mitoma M, Koga T: Functional domain of bovine milk lactoferrin which inhibits the adherence of Streptococcus mutans cells to a salivary film. Infec Immun 2002;70:5279-5282.
- 118. Southgate DAT: Milk and milk products, fats and oils. In: Garrow JS, James WPT, Ralph A (eds): Human nutrition and dietetics. Edinburgh, Churchill Livingstone, 2000, pp 375-383.
- 119. Tanaka K, Miyake Y, Sasaki S: Intake of dairy products and the prevalence of dental caries in young children. J Dent 2010;38:579-583.
- 120. Ferrazzano GF, Cantile T, Quarto M, Ingenito A, Chianese L, Addeo F: Protective effect of yogurt extract on dental enamel demineralization *in vitro*. Aust Dent J 2008;53:314-319.
- 121. Gustafsson BE, Quensel CE, Lanke LS, Lundquist C, Grahnen H, Bonow BE, Krasse B: The Vipeholm dental caries study. The effect of different levels of carbohydrate intake on caries activity in 436 individuals observed for five years. Acta Odont Scand 1954;11:232-364.
- 122. Dunning JM, Hodge AT: Influence of cocoa in milk on dental caries incidence. J Dent Res 1971;50:854-859.

- 123. Ye W, Feng XP, Liu YL: Epidemiological study of the risk factors of rampant caries in Shanghai children. Chin J Dent Res 1999;2:58-62.
- 124. Al-Jobair A, Khounganian R, 2015 Evaluating the cariogenic potential of flavored milk: an experimental study using rat model. J Contemp Dent Pract 2015;16:42-47.
- 125. Moynihan PJ, Gould ME, Huntley N, Thorman S: Effects of glucose polymers in water, milk and a milk substitute on plaque pH *in vitro*. Int J Paediatr Dent 1996;6:19-24.