Beta-casein proteins and infant growth and development

Milk formula is usually based on cows' milk with the addition of essential nutrients and vitamins. A major protein component of cows' milk is β-casein of which there are two primary variants, A1 and A2. Studies have linked a digestive product of A1, but not A2, to an increased risk of type 1 diabetes in some infants, adverse immune responses, digestive disorders and respiratory dysfunction. The A2 protein is more comparable to human β-casein protein. Formula based on the A2 protein, excluding A1 protein, may more closely mimic breast milk and may help to maintain optimal growth and development in the infant.

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Key points

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1. The A2 variant of β-casein in cows' milk is structurally more comparable to the β-casein protein in human breast milk.
2. Digestion of A1 yields BCM-7, which is associated with adverse effects including increased risk of type 1 diabetes, intolerance reactions and digestive disorders.
3. Levels of BCM-7 derived from A1-containing formulae in infants correlate with delays in psychomotor development and respiratory dysfunction.
4. Consumption of an infant formula containing only A2 β-casein may help to maintain a range of functions in growing and developing infants.

Breast milk is the preferred source of nutrition for infants. The World Health Organization (WHO) recommends that infants should be exclusively breastfed for the first six months of life and that breastfeeding should be continued for up to two years or beyond, together with appropriate solid foods[1]. However, not all infants can be breastfed and some may not have access to donor breast milk. In such situations, the family will need to use infant formula instead. Most infant formulas are produced from cows' milk as it is a relatively cheap source of protein and nutrients and is abundantly available. However, the protein composition of breast milk differs substantially from that of cows' milk. For example, breast milk is whey dominant, with approximate casein to whey ratio of 40:60, ranging from 10:90 in early lactation to 50:50 in late lactation. In contrast, cows' milk and infant formula have casein to whey ratios as high as 80:20[2].

β-casein is a major protein expressed in human and cows' milk and is present in many food products derived from milk. Like other proteins, β-casein is an important source of amino acids and facilitates mineral transport, but can be broken down into smaller bioactive peptides. In cows' milk, two primary variants of β-casein, termed A1 and A2, and several rare sub-variants have been identified. A1 and A2 β-casein differ in their protein structure by a substitution of the amino acid at position 67 (**FIGURE 1**). A1 β-casein contains a histidine residue at position 67, which allows cleavage of the preceding seven amino acid residues, generating the peptide β-casomorphin-7 (BCM-7). A2 β-casein contains a proline residue at position 67, which prevents cleavage of this peptide[3]. The protein structure of β-casein in breast milk is similar to that of A2 β-casein in cows' milk (**FIGURE 2**) and hence human β-casein is not susceptible to this mode of cleavage.

BCM-7 has a demonstrated potential to cross the gastrointestinal wall, enter the systemic circulation and influence systemic and cellular activities via opioid receptors. Moreover, BCM-7 and other derivatives of β-casein are potent exogenous agonists – exorphins – for opioid receptors, with the greatest affinity for μ receptors[4]. Consequently, BCM-7 has the potential to influence the activities of a variety of organs/systems, notably the digestive system and immune cells. It may also be involved in various disorders in infants, including type 1 diabetes[5] and respiratory dysfunction[6], and may influence central nervous system activity[7].

A1-derived BCM-7 and the digestive system

It is reported that chronic constipation and the development of anal fistulas in infants are significantly associated with the volume of cows' milk consumed and a shorter duration of breastfeeding[8]. This phenomenon may be related to the

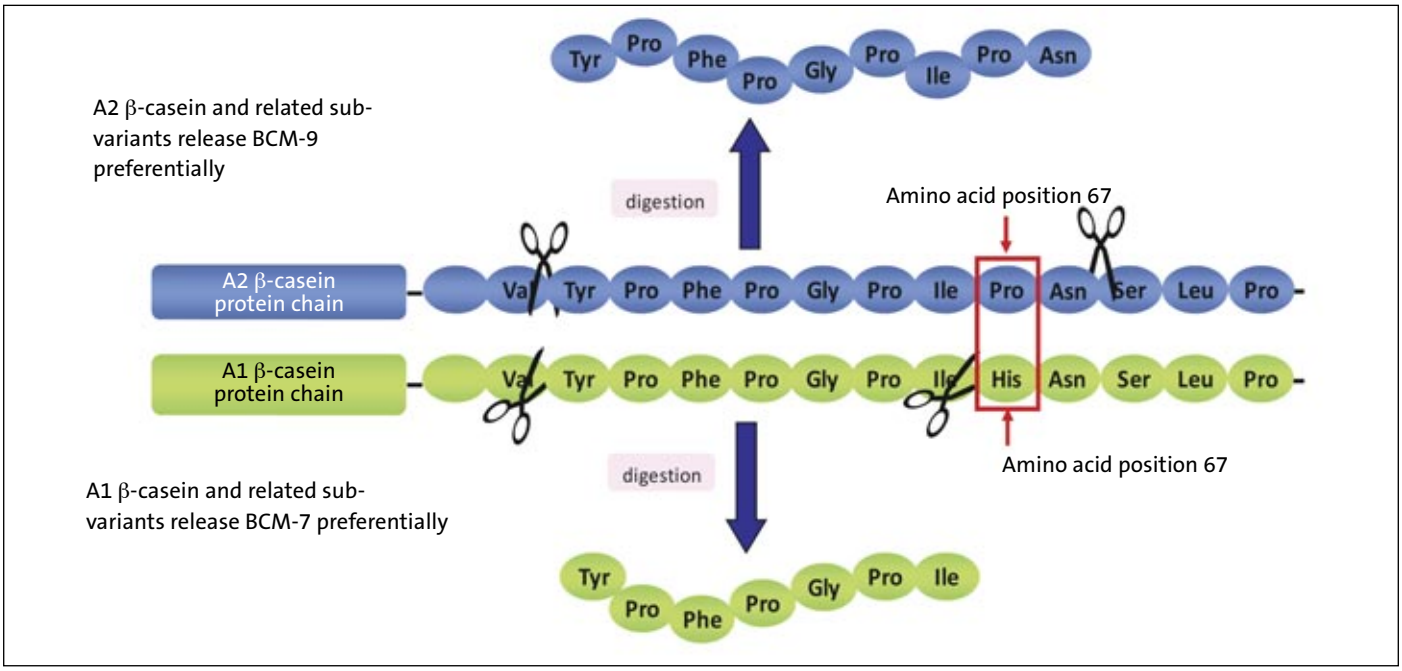

FIGURE 1 Cleavage of A1 β-casein at position 67 yields the peptide β-casomorphin-7 (BCM-7).

morphine-like effects of BCM-7[9,10].

The digestive tract of infants is very immature, particularly in terms of enzyme expression profiles and commensal bacteria[11], and undergoes continual development from birth to weaning[12]. Because proteins are principally digested in the intestinal tract in infants, rather than in the stomach as in adults, the likelihood of incomplete digestion of β-casein to amino acids is much greater in infants. Furthermore, the neonatal gut is designed to absorb relatively large macromolecules, particularly lactoglobulin (the main whey protein) from breast milk. A consequence of these essential features of the infant gut may include increased generation and uptake of BCM-7, which may adversely affect the functions of the digestive tract by slowing gastrointestinal transit, altering mucus secretion and facilitating the development of anal fistulas. The protein fragments may also have important roles in adverse immunological and allergic reactions[13].

A1-derived BCM-7 and immune function

While the immunomodulatory effects of morphine are generally well established, the potential immunomodulatory effects of β-casein and its cleaved peptides were first identified in the 1980s[14,15]. Since then, it has become apparent that exorphins, including BCM-7, have immunomodulatory properties. For example, BCM-7 was reported to trigger histamine release from peripheral leukocytes[16] and to stimulate secretion by peritoneal mast cells[17]. Studies have shown that BCM-7 alters lymphocyte proliferation *in vitro* through a pathway mediated by opiate receptors[18,19]. The first of these studies showed suppressive effects of BCM-7 on lymphocyte proliferation at all concentrations tested[18], while the second study showed suppressive effects of low BCM-7 doses and stimulatory effects at higher doses[19].

Clinically, BCM-7 may induce allergic reactions by stimulating excessive histamine release, which may lead to localised 'pseudoallergic' skin reactions or airway inflammation[16,20]. Impaired immune function may also increase susceptibility to infection and other potentially severe diseases, as has been reported for morphine[21]. Additional studies are needed to establish the specific immunomodulatory effects of BCM-7 and related peptides and to determine their clinical implications. Intervention studies are also warranted to assess whether the potential for these adverse events may be avoided by excluding A1 β-casein from the diet.

Tyr^{60}-Pro^{61}-Phe^{62}-Pro^{63}-Gly^{64}-Pro^{65}-Ile^{66}-His^{67}	Bovine β-casein A1
Tyr^{60}-Pro^{61}-Phe^{62}-Pro^{63}-Gly^{64}-Pro^{65}-Ile^{66}-Pro^{67}	Bovine β-casein A2
Tyr^{51}-Pro^{52}-Phe^{53}-Val^{54}-Glu^{55}-Pro^{56}-Ile^{57}-Pro^{58}	Human β-casein

FIGURE 2 Sequence comparisons of A1 and A2 β-casein in cows' milk and the corresponding sequence in human β-casein.

A1 β-casein and type 1 diabetes

Type 1 diabetes is characterised by autoimmune-mediated destruction of pancreatic cells. Its incidence is progressively increasing in many countries[22,23]. One explanation is that environmental factors play a major role in its pathogenesis[24].

A link between cows' milk and type 1 diabetes in animals was first reported in 1984[25], while a link to type 1 diabetes in humans was first reported in 1990[26]. A subsequent study proposed that early exposure to cows' milk may increase the risk of type 1 diabetes by approximately 1.5 times[27]. Since then, several published studies have supported this association[28-30], although other studies have found no association between antibodies to cows' milk and the risk of type 1 diabetes[31-33].

The identification of A1 and A2 β-casein and the increased understanding of their differing effects on immune function prompted the hypothesis that the discrepancies in epidemiological findings may be, at least partly, attributable to the main type of β-casein consumed in each country. In 1999, in an analysis of children aged 0-14 years across 10 countries/regions, it was reported that, while total cows' milk protein consumption (including from dairy foods) was not significantly

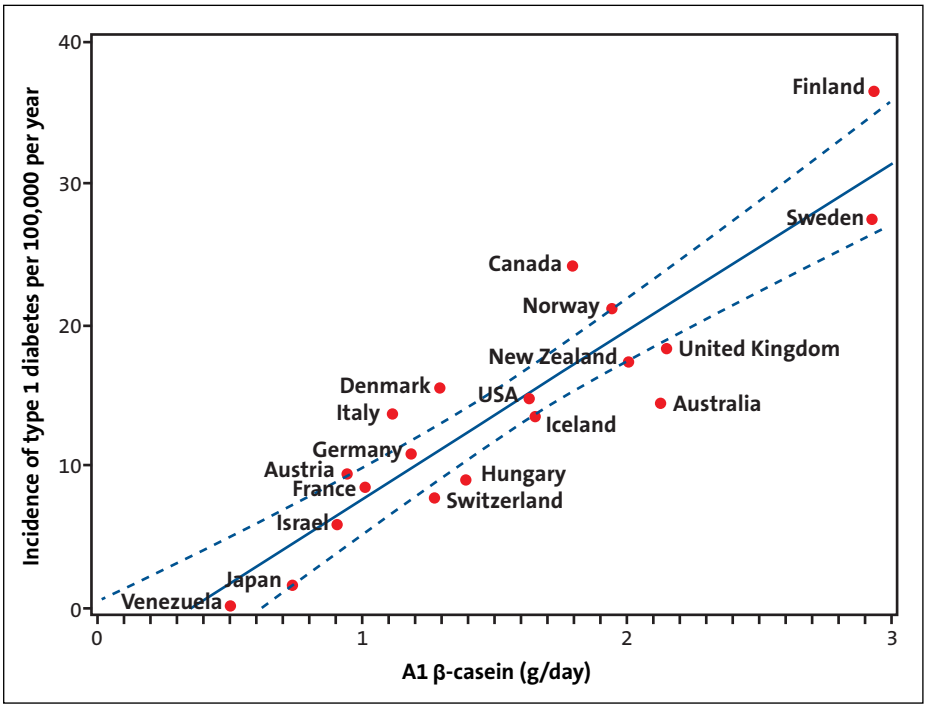

FIGURE 3 Correlation between A1 β-casein supply per capita in 1990 and incidence of type 1 diabetes (1990-1994) in children aged 0-14 years in 19 countries (r=0.92; 95% confidence interval 0.72-0.97; p<0.0001). The dotted lines show the 95% confidence limits of the regression line. Data from Laugesen and Elliott (2003)[35].

correlated with the incidence of type 1 diabetes (r=0.402), there was a correlation with the consumption of A1 β-casein (r=0.726)[34]. These findings were confirmed by two other independent studies involving a larger number of countries/regions (**FIGURE 3**)[35,36]. A study published in 2006 provided further support for the diabetes-producing effects of A1 β-casein[37].

To better understand the relationship between A1 β-casein and risk of diabetes, Birgisdottir et al compared the risk of type 1 diabetes among children and adolescents in Iceland and Scandinavia[37]. The consumption of A1 β-casein was calculated from milk and milk products consumption data and cows' milk protein concentration. A significant difference was found between the calculated intakes of A1 β-casein in two-year old children in Iceland, compared to Scandinavia. Consumption of A1 β-casein at this age was lower in Iceland than in Scandinavia and correlated with the incidence of type 1 diabetes in 0-14 year-old children (r=0.9, p=0.037). There was no difference in consumption of A1 β-casein in 11-14 year olds and no association with the incidence of type 1 diabetes in this age group. While not demonstrating cause and effect, these observational data suggest that avoiding the consumption of A1 β-casein during infancy and early childhood may reduce the risk of developing type 1 diabetes in adolescence.

A1-derived BCM-7 and respiratory function

Peptides derived from casein, including BCM-7, have been implicated in the aetiology of sudden infant death syndrome[6]. For example, Wasilewska et al noted that infants with apparent life-threatening events had higher serum levels of BCM-7 after apnoea compared with healthy infants of the same age[38]. Similar findings were reported for other BCMs and β-endorphins[39-41]. Hedner and Hedner noted that BCMs can readily cross the blood-brain barrier in newborn rabbits and cause dose-related depressions of respiratory frequency and tidal volume[42]. They found that BCM-7 was equipotent to morphine and its effects were reversed or prevented by naloxone, a μ-receptor antagonist.

Role for milk formula based on A2 protein

There are some data to suggest that consumption of dairy products containing predominantly A1 β-casein may be associated with adverse clinical outcomes in some susceptible infants and young children, including digestive disorders, immune disorders, type 1 diabetes and respiratory dysfunction. By contrast, infants who are mainly given breast milk, which contains β-casein that is more comparable in terms of structure and digestion patterns to A2 than to A1 β-casein in cows' milk, are at a lower risk of developing these disorders. While further research is warranted, for infants requiring milk formula because of limited availability of breast milk the data published to date suggest that milk formula (and dairy products in older infants) excluding A1 β-casein may help to reduce the risk of a range of adverse effects or reactions.

Conflict of interest and acknowledgement

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