A bovine albumin peptide as a possible trigger of insulin-dependent diabetes mellitus


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IN BRIEF

Hypothesis and aim
Cow's milk has been implicated as a possible trigger of the autoimmune response that destroys pancreatic beta cells in genetically susceptible hosts, thus causing diabetes mellitus. Studies in animals have suggested that bovine serum albumin (BSA) is the milk protein responsible, and an albumin peptide containing 17 amino acids (ABBOS) may be the reactive epitope. Antibodies to this peptide react with p69, a beta-cell surface protein that may represent the target antigen for milk-induced beta-cell-specific immunity. The presence of anti-BSA and anti-ABBOS were investigated in insulin-dependent diabetes.

Experimental design and methods
Immunoaassays and Western blot analysis have been used to analyze anti-BSA antibodies in the serum of 142 children with insulin-dependent diabetes mellitus, 79 healthy children, and 300 adult blood donors. Anti-ABBOS antibodies were measured in 44 diabetic patients at the time of diagnosis, three to four months later, and one to two years later.

Results
All the diabetic patients had elevated serum concentrations of IgG anti-BSA antibodies (but not of antibodies to other milk proteins), the bulk of which were specific for ABBOS. The mean (+SE) concentration was 8.5±0.2 kilofluorescence units (kfU) per microliter, as compared with 1.3±0.1 kfU per microliter in the healthy children. IgA antibodies were elevated as well, but not IgM antibodies. The antibody concentrations declined after diagnosis, reaching normal levels in most patients within one to two years. The initial decline involved anti-ABBOS-specific antibodies almost exclusively. Much lower serum concentrations of anti-BSA antibodies were found in all 379 control subjects, but only 2.5 percent of them had small amounts of ABBOS-specific IgG.

Messages and perspectives
Patients with insulin-dependent diabetes mellitus have immunity to cow's milk albumin, with antibodies to an albumin peptide that are capable of reacting with a β-cell specific surface protein. Such antibodies could participate in the development of islet dysfunction.

Comment by Jill Norris

Approximately 1 in 300 children will develop insulin-dependent diabetes mellitus (IDDM), which is associated with increased morbidity and premature mortality. The ability to prevent such a serious disease lies in a clear definition of its causes. While genetic alterations have been linked to diabetes risk, it
appears that fewer than 5 percent of individuals possessing the currently identifiable "diabetes genes" will ever become overtly diabetic. There has been a long history of attempts to link the expression of diabetes to a variety of environmental events; and in the past 10 years, evidence has been mounting that the environmental factor may be present in the diet, potentially during infancy. The paper by Karjalainen et al. (1) has brought special attention to the possible relationship between early exposure to cow's milk protein and the development of IDDM. While this report provides an exciting conceptual framework and hypothesis, it is only one of a number of studies that have attempted to unravel the complexities of infant feeding practices and later development of IDDM.

In 1984, scientists in Scandinavia reported a nationwide decrease in breast-feeding and a corresponding increase in the incidence of IDDM between 1940 and 1980 (2). Subsequent studies have shown that (i) diabetic children were significantly less likely to have been breast-fed than nondiabetic children (3-6), (ii) there was no significant difference in the frequency of breast-feeding between diabetic and nondiabetic children (2, 6-13), or (iii) diabetic children were significantly more likely to have been breast-fed than nondiabetic children (14). Certain studies suggested that the longer children had been breast-fed, the lower was their risk for developing diabetes (2, 4, 5, 15-17), while other studies did not show increased protection with longer duration of breast-feeding (11-14). These inconsistent results suggest that breast-feeding initiation and duration may be inadequate measures of the diabetogenic exposure. For example, rather than simply regarding breast-milk as protective, the breast-feeding variable could be viewed as a surrogate for perinatal food diabetogens either in breast-milk substitutes or in the weaning foods. Indeed, several studies have reported that children with diabetes were more likely to have received any breast-milk substitutes prior to 3 or 4 months of age than children without diabetes (6, 15-17). This was also true when the analyses were limited to cow's milk-based breast-milk substitutes (6, 15-17). However, most associations were relatively weak and were not seen in all populations (6, 12, 13). A meta-analysis of selected studies suggests that children with diabetes are 60% more likely to have had an early exposure to cow's milk than nondiabetic children (18). A major limitation of most of the aforementioned studies is that the infant diet data were based on long-term maternal recall, which is subject to error (19). In fact, it is of concern that the studies using prospectively-collected records to assess infant diet (11, 12, 20) did not find the associations between IDDM and infant diet exposures found in studies that relied on recalled data. This suggests that there may be bias in the retrospective assessment of infant diet.

Although experimental data confirm that diet has a major diabetogenic effect in BB rats (21, 22) and NOD mice (22), the exact chemical identity of the food diabetogen(s) is not clear. While some studies have shown cow's milk-containing diets to be diabetogenic in the BB rat (22, 23), others have failed to replicate these findings (22, Eleanor Colle, personal communication). In addition, diets containing skim milk powder (24) or bovine serum albumin (BSA) (22) were not diabetogenic in the NOD mouse.

It has been known for several years that children with IDDM have an increased frequency of antibodies to a variety of cow's milk proteins and that this is particularly evident in those with early onset IDDM (25). Karjalainen et al.'s paper (1) greatly refines these observations. This report shows that 100% of newly diagnosed diabetic children in Finland had elevated levels of BSA antibodies, compared to less than 4 percent of nondiabetic children (1). The bulk of these antibodies were specific for a fragment of BSA, termed ABBOS, that shows two short regions of amino acid sequence similarity with a ß-cell surface protein (ICA69) of the human pancreas (26). There were no differences in titers of antibodies to casein and ß-lactoglobulin in these diabetic and nondiabetic children in the Finnish study (1). Based on these and other observations, the authors present in their paper an hypothesis of pathogenesis related to molecular mimicry with the two antigenic determinants (ABBOS and ICA69) playing a role in the induction of cow's milk induced ß-cell autoimmunity. Two points of their hypothesis that the authors emphasis yet provide little supporting data are that, 1) this process only occurs in hosts with diabetes-associated HLA Class II (DR/DQ) haplotypes capable of binding and presenting the antigen, and 2) early exposure to cow's milk protein initiates the process. A more indepth analysis of the genetic susceptibility of the study subjects and a connection between age at exposure to cow's milk protein and antibody level in susceptible individuals would have added support to this hypothesis.

Subsequent studies have found interesting but not entirely confirming results. In contrast to Karjalainen et al. (1), Atkinson et al. report that BSA antibodies were not detected significantly more often in patients with new-onset IDDM than in normal patients (27). In addition, many patients with other autoim-