BRIEF COMMUNICATION

Lactation in transgenic mice expressing the ovine β-lactoglobulin gene

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Keywords Transgenic, milk composition, β-lactoglobulin, milk protein.

Milk protein composition can be altered by production of transgenic animals which express the inserted gene in mammary tissue and secrete the gene product in milk. This technique has been used in attempts to induce synthesis and secretion of exogenous milk proteins (Simons *et al.*, 1987), or to produce milk containing biologically-important non-milk proteins e.g. blood clotting factor IX (Clark *et al.*, 1989). This technology offers the potential for radical manipulation of milk composition in dairy animals. However, in most cases the effect of gene insertion on lactational performance was not investigated fully.

In this study we investigated the effect of expression and secretion of ovine-lactoglobulin (B-LG) on mouse mammary development and secretory activity. Transgenic C57BL/CBA mice from a line designated 45T.5 were used. These animals contained the entire ovine B-LG structural gene, and preliminary experiments indicated a high level of B-LG secretion (14-21% of total milk protein; Simons *et al.*, 1987). Animals with litters of 8-11 pups were studied in their first lactation, and compared with non-transgenic siblings fed and housed under the same conditions.

Mammary DNA content and enzymic indices of cell differentiation were measured as descibed by Shipman *et al.* (1987). Tissue weight, DNA concentration and total DNA content were similar in transgenic animals and non-transgenic controls on day 12 of lactation, indicating that B-LG expression had no effect on mammary growth. The total activities per cell of acetyl-CoA carboxylase, fatty acid synthetase, glucose-6-phosphate dehydrogenase and galactosyltransferase, which change in concert with the degree of secretory cell differentiation, were also not significantly different in transgenic and non-transgenic glands on day 12.

Pup weight gain up to day 12 was similar in the two groups of animals, and this was confirmed by direct measurement of mouse milk yield by $3H_2O$ dilution (Knight *et al.*, 1986) between days 9 and 13 of lactation. Milk composition was unaffected by insertion of the foreign gene. Surprisingly, in view of the high level of B-LG expression (Harris *et al.*, 1991), the milk protein concentration of transgenics was the same as that of controls (100.1±4.7mg/ml and 97.5±5.6 mg/ml respectively). This suggested that B-LG was synthesised at the expense of other milk proteins.

Ion exchange chromatography of transgenic milk proteins using an FPLC Mono-Q column (Pharmacia) and a gradient of 0 to 1.0M- NaCl in 10mM-imidazole pH 7.0 confirmed previous estimates of B-LG in milk. The fraction containing the foreign protein constituted 34±9.7% of total protein. Comparison of the relative concentrations of proteins in the other major peaks suggested further that B-LG secretion was not at the expense of one specific protein, but of endogenous milk proteins in general.

These results suggest that expression of a foreign protein in mammary tissue and its secretion in milk is not in itself sufficient to increase milk protein concentration. The mechanism by which B-LG suppressed the synthesis of endogenous milk proteins requires further investigation. It may be that the overall rate of milk protein synthesis may be determined by factors other than the relative abundance of their messenger RNAs, perhaps at the level of protein translation.

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