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Effect of Recombinant Bovine Somatotropin (Somidobove) in a Sustained Release Vehicle on Plasma Somatotropin Level and Lactational Performance of Dairy Cows

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Summary

The effects of administration of recombinantly derived bovine somatotropin (somidobove) in a sustained-release vehicle on the profiles of concentrations of bovine somatotropin (bST) in the blood plasma and on the milk yield of dairy cows of three herds were examined. Cows (36-87 days *post partum*) were treated subcutaneously with recombinant bST at 28-day intervals. In control animals, basal concentrations of bST averaged 1.4 ng.ml^{-1} in first-calf heifers and 1.5 ng.ml^{-1} in multiparous cows. In somidobove treated first-calf heifers, the concentration of bST was increased to 10.7, 14.5, and 27.0 ng.ml^{-1} at 24 h postinjection and in multiparous cows to 6.6, 11.0, and 11.7 ng.ml^{-1} on day 2 postinjection of 320, 640, and 960 mg of somidobove, respectively. On day 8 postinjection the average plasma bST levels of both parity groups are similar (on the average 3.4, 8.6, and 12.5 ng.ml^{-1} for three doses of somidobove respectively) and for the two highest doses being still significantly increased. During the 2nd week postinjection plasma bST concentration declined returning to control levels on day 15 postinjection. Somidobove-treated first-calf heifers produced 10.9, 16.7 and 17.9 % and multiparous animals 25.5, 24.2 and 32.5 % more milk than the controls when given 320, 640 and 960 mg somidobove, respectively. The cyclic pattern in milk yield within each 28-day injection interval was observed consistently in all herds. The milk yield increased to a maximum between day 4 to 8 postinjection and then slowly declined. Milk composition was not affected by somidobove treatment.

Key words

Somatotropin – Cow – Milk yield – Milk composition

Introduction

Farm animals represent one of man's most valuable renewable resources. A research problem facing agricultural scientists in developing countries is how to maintain an adequate supply of farm animal products in the face of rising human population without extending land areas for production of forages and without substantial increases of the number of farm animals. On the other hand, the supply of animal products in developed countries is adequate but the animal industry is faced with the problem of overproduction, i.e. the supply of milk is outstripping demand. The solution is to increase the efficiency of animal production enabling both to increase the volume of animal production in developing countries and to obtain the same amount of animal products (at a lower price) from fewer animals in developed countries. In the past, the efficiency of animal production was increased mainly by genetic selection over many generations and improving animal nutrition. At present the efficiency of animal production can be increased by use of new knowledge from the field of hormonal control of metabolism at the level of nutrient partitioning (Bauman *et al.* 1982). This holds particularly for the somatotropin stimulating effect on milk production in lactating cows, demonstrated over 50 years ago (Asimov and Krouze 1937). Although increases in milk yield were observed in cows given crude anterior pituitary derived somatotropin, the limitations in bST supply (only 5-15 mg bST can be purified from each pituitary) prevented its commercial application. However, recent advances in molecular biology and biotechnology have made it possible to produce bST by recombinant-DNA technology. Bauman *et al.* (1985) first demonstrated that daily injections of recombinant bST have a similar stimulatory effect on lactation as pituitary derived bST. Daily administration of bST is not practical, however, and therefore more sustained release vehicles have been developed recently. In the present study, the effects of recombinant bovine somatotropin in a

sustained release vehicle on plasma bST concentrations and lactational performance were studied.

Material and Methods

Recombinantly derived bST-somidobove in prolonged release vehicle (OPTIFLEX^{R}) was a gift from Eli Lilly/Elanco (Indianapolis, IN, U.S.A.). Recombinant somatotropin consists of the 190 amino acid form of pituitary bovine somatotropin with an additional nine amino acids at the aminoterminus. Somidobove is the nonproprietary name for recombinant bovine somatotropin established by US Adopted Names Program in accordance with The Federal Food, Drug and Cosmetic Art (Griffiths 1987). Pituitary-derived bST NIH-GH-B 1003A was a gift from the National Institutes of Health (Bethesda, MD, U.S.A.). Lactoperoxidase (B grade) was purchased from Calbiochem (Los Angeles, CA, U.S.A.). ^{125}I ($Na^{125}I$, carrier free) was bought from Amersham International plc, Amersham, England.

Trials have been conducted in 3 commercial herds (56 cows). Cows were Friesian and crossbred Bohemian spotted-Friesian or Bohemian spotted-Ayrshire. Cows were housed in tie stalls and offered corn silage, haylage, green alfalfa or clover hay and a concentrate mixture to meet nutrient requirements for maintenance and milk production (NRC, 1978). Commencing on day 36-87 *postpartum* cows received a subcutaneous injection of 0 (control), 320, 640, or 960 mg somidobove at 28-d intervals in an 84-d experiment after a 21-d pretreatment period. A 21-d pretreatment period was used to collect covariate data to generate least squares means for treatments. The cows were arranged in blocks of two or four for treatment assignment based on parity, calving date (cows differed by less than 20 d in duration of lactation), and milk production (cows differed less than 3 kg of average daily milk yield) during pretreatment. The milk yield was recorded daily, and weekly milk samples were analysed for fat by Gerber's method and protein by Kjeldahl (Kjeltec System I.: Tecator AB, Höganäs, Sweden). The milk yield and milk composition were subjected to analysis of covariance.

Blood for analysis was taken in the morning between 08.00 to 09.00 h by jugular vein puncture on heparin at intervals during treatment and plasma was stored at -20 °C until analysed. Concentration of bST in the plasma of cows was determined by radioimmunoassay (Dvořák *et al.* 1978) with pituitary-derived bST purified by ionex chromatography (potency: 1.4 IU.mg^{-1}) for iodination and reference standards. Iodination of bST was performed by the lactoperoxidase method (Thorell and Johansson 1971). Intraassay and interassay coefficients of variation averaged 5.9 and 10.2 %.

Results

The blood plasma immunoreactive bST concentrations were determined in first-calf heifers after the 3rd injection of somidobove when they were on day 92 to 137 of lactation and in multiparous cows after the 2nd injection of somidobove when they were on day 76 to 115 of lactation. Mean concentrations of bST in control and treated animals (day 1 or 2, 8, 15 and 22 after injection of somidobove) are shown in Fig. 1.

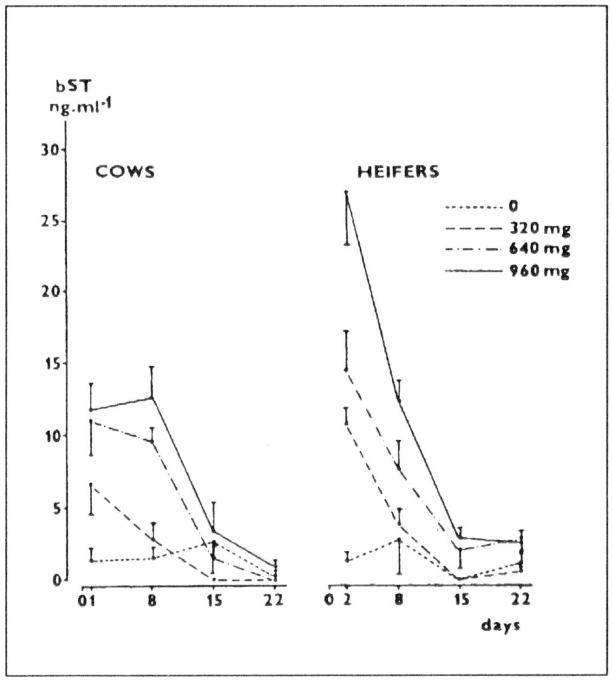

Fig. 1
Profiles of plasma somatotropin of multiparous cows and first-calf heifers after administration of recombinant bovine somatotropin (somidobove) in a sustained release vehicle. Crossbred Friesian-Bohemian spotted first-calf heifers and multiparous cows were injected (subcutaneously) with 0 (control), 320, 640 or 960 mg of somidobove at 28-d intervals. Blood for determination of somatotropin (bST) was taken by jugular vein puncture after second (in multiparous cows) or third (in first-calf heifers) injection of somidobove. The values are the mean concentrations of bST ± S.E.M. obtained from four animals.

In the control groups, mean basal concentrations of bST varied between 0.0 to 2.8 ng.ml^{-1} in first-calf heifers and between 0.3 to 2.7 ng.ml^{-1} in multiparous cows. Administration of 320, 640, or 960 mg of somidobove significantly ($P<0.01$) elevated the concentration of bST to 6.6, 11.0, and 11.7 ng.ml^{-1} respectively on day 2 postinjection in multiparous cows. Average bST level for the two highest doses was also significantly increased by postinjection day 8 and decreased thereafter rapidly reaching preinjection

concentration by day 15. In somidobove-treated first-calf heifers, the concentrations of bST in the plasma increased to 10.7, 14.5, and 27.0 ng.ml^{-1} at 24 h postinjection and then decreased by day 8 to a similar level as that found in multiparous cows. Average plasma bST levels of both parity groups were 3.4, 8.6, and 12.5 ng.ml^{-1} for three doses of somidobove respectively.

The milk yield from control first-calf heifers and multiparous cows decreased on the average by 9.8 and 7.0 % per 28 days, respectively. Milk yields of cows receiving different doses of somidobove decreased more slowly (in first-calf heifers) or increased (in multiparous cows) compared to the preliminary period. When the milk production response of bST treated cows was compared with milk production of control animals during the same experimental period, the bST treated first-calf heifers produced 2.3, 2.6 and 2.8 kg.d^{-1} or 10.9, 16.7 and 17.9 % and bST treated multiparous animals produced 4.0, 3.8 and 5.1 kg.d^{-1} or 25.5, 24.2 and 32.5 % more milk than the controls when given 320, 640 and 960 mg somidobove, respectively.

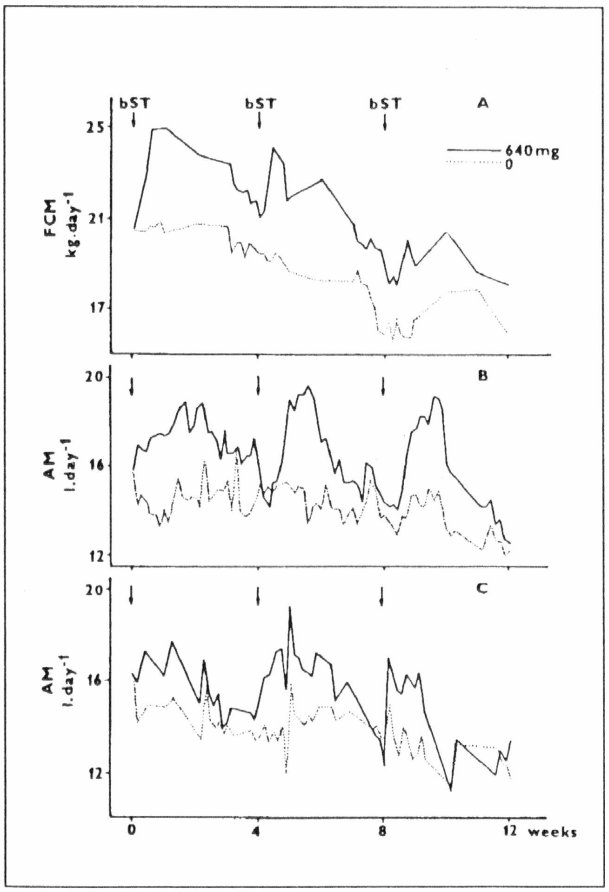

Fig. 2
Effect of somidobove in a sustained release vehicle on milk production in three herds of dairy cows. Cows received an injection of 640 mg of somidobove at 28-d intervals (arrows) in an 84-d experiment. FCM =4 % fat-corrected milk; AM = actual milk production; Herd A = Friesian and crossbred Friesian-Bohemian spotted dairy cows; herds B and C=crossbred Ayrshire-Bohemian spotted dairy cows.

Although cows received different amounts of somidobove, no dose-dependent response in multiparous cows was observed. Milk composition was not affected by somidobove treatment (Tab. 1). The daily pattern of milk yield during the three injection intervals in three herds (34 cows) is shown in Fig. 2. Daily 4 % fat-corrected milk yield (FCM) and actual milk yield of control cows of all herds gradually decreased during the experimental period. Cows treated with somidobove (640 mg at 28-d intervals) showed a cyclic pattern of daily milk yield within each injection period. Milk production increased to a maximum at 4 to 8 d postinjection and then slowly declined to 28 d. Treatment with somidobove increased the milk yield and FCM in all herds. However, in herd C with poor management conditions the milk yield response was lower (11 % increase) than that in herd B (an increase of 16 %) using the same breed of cows - crossbred Ayrshire x Bohemian spotted, at a similar level of milk production during pretreatment and stage of lactation.

Discussion

Milk production of cows subjected to long-term treatment involving daily injections of somatotropin increased and remained relatively constant for several weeks after the beginning of treatment (Bauman *et al.* 1985). However, in our experiments a cyclic pattern in milk yield within each 28-d injection interval was observed consistently in all three injection cycles. It seems to be related to a cyclic pattern of release of somidobove from the prolonged-release vehicle in the injection site. Milk production increased to a maximum at 4 to 8 d postinjection and then slowly declined as bST plasma concentrations returned to control levels.

The milk yield responses to different bST doses were not significantly dose-dependent in first-calf heifers but not consistently dose-dependent in multiparous cows after injection of 320, 640, or 960 mg.d^{-1} of somidobove. Bauman *et al.* (1985) using daily injections of bST in the range from 13.5 to 40.5 mg.d^{-1} for 180 d obtained a dose-dependent milk yield response but also no significant difference between the treated groups. Milk composition was unaffected by administration of bST. Similar findings were reported by Van den Bergh (1991) in cows as far as a positive energy balance is concerned. In cows on a negative energy balance the fat content of the milk will increase and protein content may decrease.

The mechanism by which bST exerts its galactopoietic activity is unclear. BST did not affect the synthetic activity of cultured mammary tissue (Škarda *et al.* 1982). Moreover, Mikuláš *et al.* (1981) and

Table 1

Effect of recombinant bovine somatotropin in a sustained release vehicle on milk, fat and protein yield and milk composition

	Preliminary period	Somidobove, mg.28 d^{-1} (1)			
		0	320	640	960
First-calf heifers					
Milk yield (2) kg.d^{-1}	20.3±0.8	15.6±0.6[a]	17.3±0.7[b]	18.2±1.1[b]	18.4±0.3[b]
% increase		0	10.9	16.7	17.9
Fat, %	3.74±0.12	3.89±0.11	3.85±0.12	3.68±0.09	3.76±0.11
Fat yield, kg.d^{-1}	0.79±0.04	0.62±0.03[a]	0.68±0.02[b]	0.70±0.04[b]	0.72±0.01[b]
Protein, %		3.29±0.12	3.02±0.13	2.99±0.14	2.96±0.05
Protein yield, kg.d^{-1}		0.53±0.02[a]	0.54±0.03[a]	0.57±0.03[b]	0.58±0.01[b]
Multiparous cows					
Milk yield, kg.d^{-1}	17.9±2.6	15.7±1.7[a]	19.7±1.9[b]	19.5±1.2[b]	20.8±2.8[b]
% increase		0	25.5	24.2	32.5
Fat, %	3.60±0.16	3.65±0.07	3.69±0.07	3.72±0.14	3.90±0.22
Fat yield, kg.d^{-1}	0.70±0.10	0.61±0.07[a]	0.76±0.08[b]	0.76±0.06[b]	0.81±0.09[b]
Protein, %		2.96±0.11	3.19±0.07	3.01±0.06	3.03±0.17
Protein yield, kg.d^{-1}		0.49±0.05[a]	0.65±0.05[b]	0.61±0.04[b]	0.63±0.07[b]

(1) Cows were injected (subcutaneously) with different doses of recombinant bovine somatotropin (somidobove) at 28-d intervals. All values are the least squares means from four animals, ± S.E.M. The results were obtained after 3rd injection of somidobove in first-calf heifers and after 2nd injection of somidobove in multiparous cows due to induction of superovulation and/or embryo transfer in some multiparous cows after 3rd injection of somidobove.

(2) 4 % fat corrected milk yield

[a,b] Means with different superscripts differ ($P<0.05$)

Gertler *et al.* (1984) using goat and bovine mammary membrane preparations came to the conclusion that bST receptors were not present in the mammary gland. The hypothesis that a portion of the specific tissue effects of exogenous bST is mediated by IGF-I has been proposed. Receptors for IGF-I have been detected in mammary tissue (Dehoff *et al.* 1988) and the level of IGF-I in the plasma was increased by administration of bST (Mielke *et al.* 1990, Škarda *et al.* 1990, Schams *et al.* 1991). According to Peel and Bauman (1987), treatment with somatotropin appears to coordinate metabolism of many tissues to preferentially partition nutrients to support higher milk production. The suppression of lipogenesis in adipose tissue of bST treated cows (Škarda and Mader 1991) supports the concept that bST partitions fatty acids from adipose tissue and circulation for use by the mammary tissue. The rapidity for the rise and fall of milk yield in response to bST injections and their cessation implies that galactopoiesis is achieved through increased productivity of existing secretory cells rather than an increase in cell numbers.

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