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Effects of Pituitary-derived Bovine Growth Hormone on Production Parameters and Biokinetics of Key Metabolites in Lactating Dairy Cows at Peak and Mid-lactation

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Abstract

Changes in production parameters and metabolite biokinetics induced by treatment with pituitary-derived bovine growth hormone (bGH) were monitored at peak (*c.* 40 days) and mid-lactation (*c.* 130 days) in dairy cows.

During treatment with bGH milk production increased by 6 and 14% at peak and mid-lactation respectively. At peak lactation the content of milk fat tended to increase, whereas milk protein tended to decrease and milk lactose decreased significantly. Yield of milk fat increased, but there was no change in the yield of milk protein and lactose. The content of milk fat tended to increase at mid-lactation. Milk protein decreased and there was no change in milk lactose. Yields of milk fat and lactose but not protein increased. Growth hormone exerted metabolic effects which differed with stage of lactation. At peak lactation plasma glucose concentration and its irreversible loss increased, plasma urea and acetate were unchanged and their irreversible losses tended to increase. No change was measured for plasma non-esterified fatty acids (NEFA) and the irreversible loss of NEFA decreased. At mid-lactation plasma concentrations of glucose and NEFA were increased, plasma urea decreased and acetate and 3-hydroxybutyrate tended to increase. Irreversible losses of NEFA increased, urea tended to decrease and acetate and glucose remained essentially constant.

The results show that exogenous pituitary bGH exerts metabolic effects which result in the supply of increased nutrients to support milk synthesis. The metabolic effects differ with the stage of lactation, reflecting differences in physiological and/or nutritional state.

Introduction

The galactopoietic effect of growth hormone (GH) is now well recognized (Cowie *et al.* 1980). Although it has been suggested that GH exerts homeorhetic control resulting in the supply of additional nutrients for milk synthesis (Bauman and Currie 1980), there are few data to support this contention. It is known that pituitary GH is able to influence, either directly or indirectly, aspects of protein, carbohydrate and fat metabolism (Hart 1973; Hart *et al.* 1984). Thus it might be expected that exogenous GH would exert metabolic effects resulting in the increased supply of nutrients for milk synthesis. To date there is only one brief report (Peel *et al.* 1982) of measurements of the biokinetics of key metabolites for milk synthesis.

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In this the lipolytic effects of GH were prominent and thought to be important for enhanced milk production.

The present study was designed to quantify the effects of exogenous pituitary GH on plasma concentrations and whole-body irreversible losses of key metabolites for milk synthesis. Biokinetics of glucose, non-esterified fatty acids (NEFA), acetate and urea (as an indirect measure of amino acids) were measured in lactating cows at peak and mid-lactation during treatment with either saline or bovine GH (bGH).

Materials and Methods

Experimental Cows

Five British Friesian cows (see Table 1) were selected from the herd at the National Institute for Research in Dairying and accustomed to the barn, in which they were to be housed, for several weeks before parturition. All cows calved uneventfully and after calving were placed in standings which were isolated from other animals.

Table 1. Details of cows and feed offered throughout the experiment

Parameter	Cow number					Mean
	2874	3211	3233	3276	3289	
Days from calving to start of						
First experimental period	42	25	37	43	31	35·6
Second experimental period	134	117	–	135	123	127·3
Liveweight (kg) at						
Calving	594	724	514	512	472	563
First experimental period	542	680	465	502	483	516
Second experimental period	512	661	–	516	497	547
Feed allocation (kg/day; air dry)						
Hay	6·5	6·8	5·4	5·4	6·2	6·1
Pellets	15·2	15·9	12·7	12·7	14·6	14·2

From the first week of lactation, and for the duration of the experiment, the cows were fed a ration of 70 : 30 concentrate pellets: long pasture hay (see Table 2) in sufficient quantity to meet calculated metabolizable energy (ME) requirements for a peak milk yield of 30 kg/day. It was assumed that the cows would lose liveweight at the rate of 0·4 kg/day during the first 10 weeks of lactation then regain liveweight. Throughout the experiment each cow was offered the quantity of diet calculated to meet its requirement for early lactation. Feed intakes were recorded daily.

Table 2. Composition of ration ingredients fed at peak and mid-lactation
Values expressed as g/kg dry matter

Ration ingredient	Hay		Pellets	
	Peak lactation	Mid-lactation	Peak lactation	Mid-lactation
Crude protein ($N \times 6{\cdot}25$)	83·1	83·8	204·4	178·8
M.A.D.F.	367·9	360·5	54·6	65·0
Fat	82·9	93·8	65·9	93·1
Ash	80·8	70·3	68·3	66·7
Metabolizable energy (MJ/kg)	8·85	8·95	13·24	13·19

For 2 weeks before and during experimental periods, the concentrate portion of the diet was fed using continuous feeders. The hay portion was offered in equal amounts four times daily. The cows remained in their individual standings except for short periods once weekly when liveweights were recorded.

The cows were milked at 0800 and 1600 hours daily. Milk yields were recorded daily and during experimental periods representative subsamples of pooled (morning and afternoon) milk were retained for weekly protein, fat and lactose analyses. Formalin (100 ml/1; 4 drops/10 ml milk) was added as preservative.

One cow (No. 3233) became lame during the fourth month of lactation, reduced feed intake and subsequently developed ketosis. She was removed from the second part of the experiment. Another cow (No. 3211) displayed intermittent subclinical mastitis in one-quarter throughout the experiment (assessed by counting somatic cells in milk from individual quarters of cows). Otherwise the cows remained in good health.

Each cow was fitted with polyvinyl chloride catheters (1·0 mm int. diam. by 2·0 mm outer diam.: Dural Plastics, Dural, N.S.W., Australia) in both external jugular veins 3 days before each experimental period. Catheters were flushed with small quantities of sterile heparinized saline (9 g/l NaCl; 2×10^5 units/l heparin) and remained patent for the 2 weeks of each experimental period.

Analytical Methods

Milk constituents

Milk constituents were measured with a Milko-Scan 203 (A/S N. Foss Electric, Denmark) appropriately calibrated for analysis of cow milk.

Plasma metabolites

Plasma glucose and urea were assayed using a ChemLab Autoanalyser (ChemLab Instruments Ltd, Hornchurch, Essex, U.K.) as described by the manufacturer. Plasma non-esterified fatty acids (NEFA) were measured using the method of Dole (1956) as modified by Kelley (1965). Acetate was quantified by gas–liquid chromatography as detailed by Pethick *et al.* (1981). Plasma 3-hydroxybutyrate was measured by the auto-analytical method of Zivan and Snarr (1973).

Metabolite biokinetics

The isotopically labelled metabolites, [U-^{14}C]glucose, [U-^{14}C]acetate, [U-^{14}C]urea and [9,10-^{3}H] palmitic acid were obtained from Amersham International plc (Amersham, U.K.). Measurements of irreversible losses of glucose, acetate and palmitate were made by continuous intravenous infusion of the appropriate metabolite via one jugular catheter and subsequent collection of blood samples from the contralateral catheter. Palmitic acid was bound to plasma proteins (using fresh plasma from individual animals) as described by Winkler *et al.* (1964). Infusion rates for each labelled metabolite were recorded. Irreversible loss of urea was measured after pulse infusion of [U-^{14}C]urea into one catheter and blood collection, over 12 h, for construction of the isotope dilution curve.

Doses and periods of infusion of respective isotopes were as follows: [U-^{14}C]glucose, $3{\cdot}7 \times 10^7$ Bq, 240 min; [U-^{14}C]sodium acetate, $3{\cdot}7 \times 10^7$ Bq, 90 min; [9,10(n)-^{3}H]palmitic acid, $18{\cdot}5 \times 10^7$ Bq, 90 min; [U-^{14}C]urea, $1{\cdot}85 \times 10^7$ Bq, pulse infusion.

Specific radioactivities of respective metabolites were measured according to the procedures outlined by Jones (1965) for glucose; Pethick *et al.* (1981) for acetate; Winkler *et al.* (1964) for NEFA. Thus glucose was isolated as glucose penta-acetate, acetate was isolated as the volatile acid and total NEFA were separated from other labelled metabolites. In connection with the latter, it was assumed that palmitic acid was representative of NEFA. For determination of the specific radioactivity of urea, plasma was not submitted to prior treatment before addition of scintillant. In each case the scintillant used was Instagel (Packard Instruments).

Plasma hormones

Concentrations of insulin and GH were measured using the radioimmunoassay described by Tindal *et al.* (1978). Values were expressed in terms of the standards bovine insulin (21 i.u./mg) and bovine GH (NIH-GH-B2, 1·5 U/mg) respectively. The sensitivity of the assays were respectively 0·4 μU/ml and 0·2 ng/ml for insulin and GH. To avoid differences stemming from interassay variation, samples which were to be compared were assayed in the same radioimmunoassay.

Assays were considered valid if intra- and interassay coefficients of variation were less than 15%.

Growth Hormone

The preparation of bGH used in the experiment was extracted from bovine pituitary glands using the procedure described by Ellis (1961). This preparation (ICH/bGH/9) had a biological activity of 1·690 U/mg as assessed in the bioassay based on growth of hypophysectomized rats.

The daily dose of the GH was 0·06 mg/kg liveweight (equivalent to 0·1 U/kg liveweight). Prior to injection, the lyophilized hormone was dissolved in 10 ml 0·9% (w/v) saline adjusted to pH 10 with 1 M NaOH. Injections were administered subcutaneously each day at 0730 hours.

Experimental Procedures

Responses to exogenous GH were recorded during experimental periods at peak lactation (*c.* day 40) and at mid-lactation (*c.* day 130). At each stage of lactation, measurements were made over 6 days when cows received daily subcutaneous injections of either saline (pH 10·0) or bGH. The following schedule was used within each 6-day period:

Day 1: collection of blood for measurement of insulin and GH.
Day 2: measurement of palmitate and acetate irreversible losses, 90 min infusion commencing 1130 hours.
Day 3: measurement of glucose irreversible loss, 240 min infusion commencing 0930 hours.
Day 4: measurement of urea irreversible loss, pulse infusion 0900 hours, collection of blood samples over the next 12 h.

Statistical Analyses

Comparisons between mean values for parameters measured during control and bGH treatment periods were made using the paired *t*-test. For milk and feed intake data, mean values for 6-day treatment periods were used for each animal.

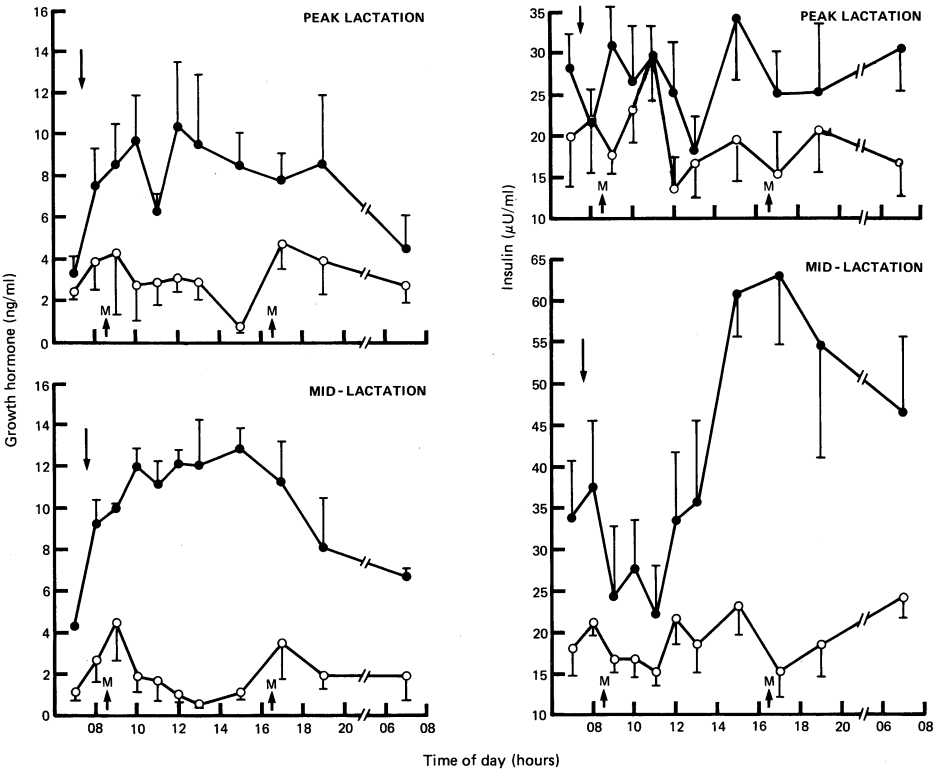

Fig. 1. Mean values for concentrations of growth hormone and insulin in plasma collected throughout the first day of each experimental period at peak and mid-lactation. Plotted points are mean values for five and four cows at peak and mid-lactation respectively, and standard errors are shown as vertical bars. Times of milking are marked (M). ○ Control. ● GH treatment. ↓ Time of injection.

Results

Plasma Hormone Concentrations

Plasma concentrations of GH and insulin measured throughout the first day of the saline and bGH treatment periods at peak and mid-lactation are shown in Fig. 1.

It is apparent that, in general, basal concentrations of GH were lower at mid- than at peak lactation. At peak lactation plasma concentrations were approximately 3·0 ng/ml throughout most of the period of measurement whereas at mid-lactation values varied between 1 and 2 ng/ml. At both stages of lactation the highest basal concentrations were measured in plasma collected after morning and afternoon milkings (i.e. 0900 and 1700 hours).

Injections of bGH significantly increased ($P < 0{\cdot}05$) plasma levels of the hormone within 1 h at both stages of lactation. Concentrations rose from basal values to between 10 and 12 ng/ml representing increases of around 3–4 and 6-fold at peak and mid-lactation respectively. Thereafter plasma levels were significantly raised above basal values for at least 12 h and remained high 24 h after injection.

Plasma insulin concentrations were reasonably constant at 15–20 μU/ml during control (saline injection) periods at both stages of lactation. There was a tendency for concentrations of insulin to increase after GH injection at peak lactation and this was particularly marked at mid-lactation ($P < 0{\cdot}05$).

Responses to bGH at Peak Lactation

Production responses

Changes in liveweight, feed intake, yield of milk and milk constituents and the content of major milk constituents at peak lactation are shown in Table 3. During treatment with bGH liveweight decreased significantly and there was a significant reduction in the content of milk lactose. Milk yield (actual and 4% fat corrected) increased significantly as did yield of milk fat. Although there was a tendency for milk fat content to increase and milk protein content to decrease changes were not significant. Feed intake remained constant.

Metabolite biokinetics

Plasma concentrations and whole-body irreversible losses of metabolites are shown in Table 4. Concentrations and irreversible loss of glucose were significantly increased by bGH treatment. Although, for other metabolites, the differences between values for the saline and GH treatment periods were not significant, trends were obviously apparent. Plasma urea concentrations were decreased during bGH treatment in four out of five cows and in all cows the irreversible loss of urea increased. There was no consistent change for plasma concentrations of NEFA but in four out of five cows the irreversible loss of NEFA decreased during treatment with the hormone. Similarly, no consistent changes were measured for plasma acetate concentrations but there was a tendency for its irreversible loss to be increased by bGH. Indeed in three out of five cows there was a marked increase in acetate irreversible loss, no appreciable change in one cow and an appreciable decrease for the other cow. For plasma 3-hydroxybutyrate, concentrations tended to decrease during GH treatment but differences were not significant.

Table 3. Production parameters, feed intake and liveweights of cows during treatment with saline or GH at peak lactation

Liveweights were measured on the last day of respective periods. Other values for individual cows are mean values for the 6 days of each treatment period. For saline *v.* GH mean values: $*P < 0{\cdot}1$; $**P < 0{\cdot}05$; $***P < 0{\cdot}01$; n.s., not significant

Parameter	Treatment	Cow number 2874	3211	3233	3276	3289	Mean
Liveweight (kg)	Saline	542	680	465	502	482	534
	GH	524	660	450	496	483	523**
Feed intake	Saline	171	105	158	170	180	157
(MJ ME/day)	GH	184	107	160	170	186	161$^{n.s.}$
Milk yield	Saline	33·9	23·1	24·9	22·3	29·9	26·8
(kg/day)	GH	35·5	23·6	27·4	24·7	30·8	28·4**
4% fat-corrected	Saline	32·0	27·3	28·0	26·0	27·4	28·1
milk yield (kg/day)	GH	35·7	29·4	28·7	29·2	29·3	30·5**
Milk fat	Saline	36·2	52·0	48·7	51·0	34·4	44·5
(g/kg)	GH	40·3	56·9	43·2	52·1	36·7	45·8$^{n.s.}$
Milk fat	Saline	1229	1204	1206	1139	1031	1162
(g/day)	GH	1434	1330	1183	1285	1128	1272**
Milk protein	Saline	30·3	30·7	26·5	27·4	32·1	29·4
(g/kg)	GH	28·3	31·0	26·7	26·4	31·2	28·7$^{n.s.}$
Milk protein	Saline	1028	706	658	609	964	793
(g/day)	GH	1003	744	729	651	963	818$^{n.s.}$
Milk lactose	Saline	50·5	45·7	49·2	50·6	50·9	49·4
(g/kg)	GH	48·6	42·5	48·4	47·4	49·0	47·2***
Milk lactose	Saline	1711	1056	1225	1128	1518	1328
(g/day)	GH	1722	1004	1324	1169	1507	1345$^{n.s.}$

Table 4. Concentrations of plasma metabolites and irreversible losses (IL) of metabolites during treatment of cows with saline and GH at peak lactation

For saline *v.* GH mean values: $*P < 0{\cdot}1$; $**P < 0{\cdot}05$; n.s., not significant

Parameter	Treatment	Cow number 2874	3211	3233	3276	3289	Mean
Plasma glucose	Saline	4·29	4·25	4·34	3·92	4·46	4·25
(mM)	GH	4·77	4·51	4·45	3·93	4·66	4·46*
Glucose IL	Saline	10·27	8·12	9·28	6·37	8·33	8·47
(mmol/min)	GH	12·61	9·31	11·36	9·39	11·70	10·87**
Plasma urea	Saline	4·65	3·72	5·63	6·98	5·75	5·35
(mM)	GH	6·51	3·42	5·47	6·24	4·29	5·19$^{n.s.}$
Urea IL	Saline	5·69	3·61	5·16	6·71	6·50	5·53
(mmol/min)	GH	10·73	3·94	5·85	7·73	7·60	7·17$^{n.s.}$
Plasma NEFA	Saline	407	362	402	426	214	362
(μM)	GH	370	412	340	398	231	350$^{n.s.}$
NEFA IL	Saline	9·07	9·88	8·14	7·94	2·59	7·52
(mmol/min)	GH	8·28	9·19	7·30	6·02	5·20	7·20$^{n.s.}$
Plasma acetate	Saline	1·30	0·89	1·20	2·60	0·90	1·38
(mM)	GH	1·32	1·12	1·19	1·78	0·88	1·26$^{n.s.}$
Acetate IL	Saline	54·5	62·9	31·4	110·3	100·3	71·9
(mmol/min)	GH	74·2	87·8	48·1	105·6	72·1	77·6$^{n.s.}$
Plasma 3-hydroxy-	Saline	1·04	1·76	1·78	6·64	0·81	2·41
butyrate (mM)	GH	0·82	2·19	1·10	3·69	0·76	1·72$^{n.s.}$

Responses to bGH at Mid-lactation

Production responses

Changes in liveweight, feed intake, yields of milk and milk constituents and the content of the major constituents of milk at mid-lactation are shown in Table 5. Significant increases were measured for milk yield (actual and 4% fat corrected), milk fat yield and milk lactose yield, in response to bGH treatment. Milk protein content decreased significantly and milk lactose content was unaffected. There was a trend for milk fat content to increase (in three out of four cows the content of fat increased whereas in one cow it decreased) but the difference between the mean values for saline and bGH periods was not significant. Similarly, in three out of four cows milk protein yield increased but the difference between mean values was not significant. Feed intake and liveweight were unchanged.

Table 5. Production parameters, feed intakes and liveweights of cows during treatment with saline or GH at mid-lactation

Liveweights were measured on the last day of respective periods. Other values for individual cows are mean values for the 6 days of each treatment period. For saline *v.* GH mean values: $*P < 0{\cdot}1$; $**P < 0{\cdot}05$; n.s., not significant

Parameter	Treatment	Cow number 2874	3211	3276	3289	Mean
Liveweight	Saline	514	670	526	506	554
(kg)	GH	518	658	520	516	553$^{n.s.}$
Feed intakes	Saline	198	207	170	195	193
(MJ/day)	GH	192	196	170	195	188$^{n.s.}$
Milk yield	Saline	22·0	17·7	17·7	19·7	19·3
(kg/day)	GH	24·7	21·5	18·7	22·9	22·0**
4% fat-corrected	Saline	21·4	15·1	19·4	20·3	19·1
milk yield (kg/day)	GH	23·0	19·0	22·6	26·5	22·8**
Milk fat	Saline	38·3	30·1	46·7	48·6	40·9
(g/kg)	GH	35·4	32·2	54·5	50·3	43·1$^{n.s.}$
Milk fat	Saline	865	544	824	828	765
(g/day)	GH	878	687	1012	1153	933*
Milk protein	Saline	37·8	36·6	34·4	37·1	36·5
(g/kg)	GH	37·5	36·0	31·9	34·2	34·9*
Milk protein	Saline	829	648	607	731	704
(g/day)	GH	925	770	597	781	768$^{n.s.}$
Milk lactose	Saline	44·5	42·9	45·8	46·7	45·0
(g/kg)	GH	44·2	45·0	46·5	46·1	45·5$^{n.s.}$
Milk lactose	Saline	976	760	808	920	866
(g/day)	GH	1091	964	867	1054	994**

Metabolite biokinetics

Concentrations and whole body irreversible losses of metabolites are shown in Table 6. During bGH treatment significant increases were measured for plasma glucose and NEFA concentrations and the irreversible loss of NEFA. On the other hand bGH treatment resulted in a significant reduction in plasma urea concentration and there was a marked tendency for the irreversible loss of urea to decrease. Even though the differences between mean values was not significant, there was a marked decrease in irreversible loss of urea in three out of four cows. No significant

change was measured for irreversible loss of glucose. Similarly, no significant changes were recorded for plasma acetate, acetate irreversible loss or plasma 3-hydroxybutyrate. In spite of this, in each case there was a trend for values to increase during the treatment period.

Table 6. Concentrations of plasma metabolites and irreversible losses (IL) of metabolites during treatment of cows with saline or GH at mid-lactation

For saline *v.* GH mean values: $*P < 0{\cdot}1$; $**P < 0{\cdot}05$; $***P < 0{\cdot}01$; n.s., not significant

Parameter	Treatment	Cow number 2874	3211	3276	3289	Mean
Plasma glucose	Saline	4·26	4·37	4·10	3·77	4·13
(mM)	GH	4·88	4·59	4·12	4·23	4·46*
Glucose IL	Saline	9·42	–	5·32	8·95	7·90
(mmol/min)	GH	9·93	–	5·10	9·27	$8{\cdot}10^{n.s.}$
Plasma urea	Saline	5·60	5·24	7·16	6·21	6·05
(mM)	GH	3·94	2·83	4·98	3·63	3·85***
Urea IL	Saline	6·69	7·30	5·79	6·99	6·69
(mmol/min)	GH	5·01	6·65	5·82	5·01	$5{\cdot}62^{n.s.}$
Plasma NEFA	Saline	176	184	182	151	173
(μM)	GH	448	348	279	302	344**
NEFA IL	Saline	5·55	5·99	4·93	4·12	5·14
(mmol/min)	GH	7·00	7·46	5·03	5·24	6·18**
Plasma acetate	Saline	1·08	0·94	1·00	1·25	1·07
(mM)	GH	0·77	0·98	1·55	1·56	$1{\cdot}22^{n.s.}$
Acetate IL	Saline	55·8	80·2	79·8	84·6	75·1
(mmol/min)	GH	65·2	70·0	97·8	99·2	$83{\cdot}1^{n.s.}$
Plasma 3-hydroxy-	Saline	0·95	1·36	1·87	1·47	1·41
butyrate (mM)	GH	0·82	1·36	2·87	2·06	$1{\cdot}78^{n.s.}$

Discussion

It is clear that the subcutaneous injections of bGH increased the plasma concentration of the hormone by 3–4-fold at peak lactation and 6-fold at mid-lactation. The fact that plasma concentrations of bGH during control periods were higher at peak than at mid-lactation is consistent with previous observations (Hart *et al.* 1978). Moreover, the increase in plasma GH recorded shortly after milking is consistent with reports that milking or suckling elicits GH release (Hart and Flux 1973).

Plasma concentrations of insulin tended to be increased after exogenous bGH at peak lactation and were significantly higher after the exogenous hormone at mid-lactation than during the corresponding control periods. This observation is consistent with previous reports (Bines *et al.* 1980). At both stages of lactation plasma glucose concentrations increased, reflecting a direct or indirect effect on glucose production and/or utilization in the body.

In conformity with several recent reports on the effects of bGH on milk production (see Johnsson and Hart 1986 for review) there were significant increases in milk production induced by bGH at both peak and mid-lactation. Clearly, exogenous GH was galactopoietic at peak lactation but in relative and absolute terms the increase in milk yield was less at peak than at mid-lactation (6 *v.* 14%; 1·6 *v.* 2·7 kg/day). Similar responses were reported by Richard *et al.* (1985).

The changes in milk composition and the yield of milk constituents induced by treatment with bGH varied according to stage of lactation. At peak lactation, there was a tendency for milk fat content to increase and milk protein content to decrease. Somewhat surprisingly, milk lactose content decreased consistently and significantly. Following bGH treatment at mid-lactation milk fat content tended to increase (not significant but in three out of four cows), milk protein content decreased significantly and there was no change in milk lactose. These changes were accompanied by significant increases in the yields of milk fat and lactose.

The above changes in milk composition probably reflected the nutritional states of the cows at the different stages of lactation and as discussed below, different metabolic effects of bGH at different stages of lactation.

It is apparent that feed intake (expressed as megajoules of metabolizable energy) was lower at peak than mid-lactation and cows were in negative energy balance at peak lactation and positive energy balance by mid-lactation. Average intakes during control periods were 157 and 193 MJ/day at peak and mid-lactation respectively (see Tables 3 and 5). This difference in feed intake during early and later lactation has been well documented (see Bines 1979), and occurred in the present study under conditions where the cows were offered the same quantity of the diet throughout the experiment. Treatment with bGH evoked essentially no change in feed intake. In this respect the results of this study are consistent with results of similar short-term studies (see Johnsson and Hart 1986) but differ from results of studies in which bGH has been administered over long periods to grazing (Peel *et al.* 1985) and concentrate-fed lactating cows (Eppard and Bauman 1984; Bauman *et al.* 1985). Under the latter conditions, food intake was markedly increased after several weeks of treatment.

The cows in the present study were fed to allow for a reduction in liveweight during early lactation. By the start of treatment at peak lactation liveweight loss was negligible in all cows but it is apparent that bGH treatment resulted in a substantial decrease in liveweight in four of the five cows. Although there was no significant effect of the hormone on the liveweight of cows at mid-lactation it is pertinent to point out that the cows were gaining weight at the time.

The metabolic effects of bGH differed with stage of lactation and presumably physiological state. At both stages of lactation plasma glucose concentrations increased but it was only at peak lactation that there was a significant increase in glucose irreversible loss. The latter presumably reflects increased utilization of glucose for milk synthesis and use as an oxidative fuel at this time. At mid-lactation it appears that glucose oxidation was spared as the glucose requirement for milk synthesis would have increased.

In connection with the latter, the well-documented lipolytic effects of GH (see Hart 1983) were not observed at peak lactation. At this stage of lactation no evidence for an increase in plasma concentrations of NEFA were obtained and irreversible loss of NEFA decreased in four out of five cows. Irreversible loss of NEFA only increased in the cow (No. 3289) which did not lose liveweight during GH treatment. The lipolytic effects of bGH were most evident at mid-lactation. Treatment with the hormone resulted in marked increases in both plasma concentration and the irreversible loss of NEFA, which is compatible with the tendency for bGH to reduce liveweight gain at this time.

Changes in acetate biokinetics are of interest even though effects of bGH

treatment were not significant. At peak lactation there was essentially no effect of bGH on plasma concentrations of acetate but there was a marked tendency for the irreversible loss of acetate to increase. In the three cows which lost the largest amounts of liveweight (Nos 2874, 3211 and 3233) acetate irreversible loss increased substantially. Essentially no change was measured for the cow in which the loss of liveweight was small (No. 3276) and there was a decrease in irreversible loss for the cow which maintained liveweight (No. 3289). Following treatment with bGH at mid-lactation, changes in acetate concentrations and irreversible losses were variable and there were no apparent correlations between irreversible losses of acetate and NEFA.

The virtual inverse relationship between irreversible losses of NEFA and acetate observed at peak lactation may reflect increased oxidation/utilization of acetate at peak lactation, in the absence of available NEFA. It would be of interest to measure the effects of bGH on tissue utilization/oxidation of these substrates.

Data for changes in plasma concentrations of 3-hydroxybutyrate are of interest with regard to the oxidation of NEFA. It would be expected that, if there was increased oxidation of NEFA, plasma concentrations of 3-hydroxybutyrate would increase (Kronfeld 1965). Although differences for plasma concentrations of 3-hydroxybutyrate were not significant, there was a tendency for concentrations to decrease after bGH at peak lactation and to increase at mid-lactation. Thus, although equivocal, the data are consistent with decreased oxidation of NEFA at peak lactation and increased oxidation at mid-lactation.

The changes measured for plasma urea and irreversible loss of urea in response to bGH are also of interest. Urea concentration and irreversible loss might be expected to reflect protein turnover. It is well documented that GH promotes synthesis of muscle protein (see Lindsay 1983) so the observations that, in early lactation, bGH had no effect on urea concentrations and that there was a consistent tendency for irreversible loss of urea to increase was unexpected.

A possible explanation for the increased irreversible loss of urea at peak lactation might be that amino acids were being utilized as gluconeogenic substrates. In this connection, it is known that several amino acids may be used as substrates for glucose synthesis and it seems plausible to suggest that in a situation where requirements for glucose increase (in this case for lactose synthesis at least) and glucose oxidation cannot be spared by provision of NEFA as oxidative fuel, that amino acids are used as gluconeogenic substrates.

The decreased concentrations and trend for decreased irreversible loss of urea observed at mid-lactation is consistent with bGH promoting muscle protein synthesis. Furthermore the significant decrease in milk protein content at mid-lactation would be consistent with a decreased availability of amino acids for milk protein synthesis associated with increased synthesis of muscle protein.

In summary, the results of the present study have reaffirmed that treatment of lactating dairy cows with exogenous bGH results in increased milk production. It is clear that the hormone exerts metabolic effects resulting in the provision of substrates to the mammary gland for milk biosynthesis. The metabolic effects of pituitary growth hormone are known to be heterogeneous (see Hart 1983) and it appears that different metabolic effects are exerted depending on physiological state. Overall, the results of the present study are consistent with GH exerting homeorhetic control, as suggested by Bauman and Currie (1980), resulting in the provision to

the mammary gland at the expense of other body tissues of nutrients for milk synthesis.

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References

Bauman, D. E., and Currie, W. B. (1980). Partitioning of nutrients during pregnancy and lactation: A review of mechanisms involving homeostasis and homeorhesis. *J. Dairy Sci.* **63**, 1514–29.

Bauman, D. E., Eppard, P. J., de Geeter, M. J., and Lanza, G. M. (1985). Responses of high-producing dairy cows to long-term treatment with pituitary somatotropin and recombinant somatotropin. *J. Dairy Sci.* **68**, 1352–62.

Bines, J. A. (1979). Voluntary feed intake. In 'Feeding Strategy for the High Yielding Dairy Cow'. (Eds W. H. Broster and H. Swan.) Ch. 3. pp. 23–48. (Granada Publishing Unit: St Albans.)

Bines, J. A., Hart, I. C., and Morant, S. V. (1980). Endocrine control of energy metabolism in the cow: the effect on milk yield and levels of some blood constituents of injecting growth hormone and growth hormone fragments. *Br. J. Nutr.* **43**, 179–88.

Cowie, A. T., Forsyth, I. A., and Hart, I. C. (1980). Hormonal control of lactation. In 'Monographs of Endocrinology'. Vol. 15. Ch. 4. (Springer-Verlag: Berlin, Heidelberg, New York.)

Dole, V. P. (1956). A relation between non-esterified fatty acids in plasma and the metabolism of glucose. *J. Clin. Invest.* **35**, 150–4.

Ellis, S. (1961). Studies on the serial extraction of pituitary proteins. *Endocrinology* **69**, 554–70.

Eppard, P. J., and Bauman, D. E. (1984). The effect of long-term administration of growth hormone on performance of lactating dairy cows. Proc. Cornell Nutr. Conf. Fd Manuf. pp. 5–12.

Hart, I. C. (1973). Effect of 2-bromo-α-ergocryptine on milk yield and the level of prolactin and growth hormone in the blood of the goat at milking. *J. Endocrinol.* **57**, 179–80.

Hart, I. C. (1983). Endocrine control of nutrient partition in lactating ruminants. *Proc. Nutr. Soc.* **42**, 181–94.

Hart, I. C., Bines, J. A., Morant, S. V., and Ridley, J. L. (1978). Endocrine control of energy metabolism in the cow: comparison of the levels of hormones (prolactin, growth hormone, insulin and thyroxine) and metabolites in the plasma of high- and low-yielding cattle at various stages of lactation. *J. Endocrinol.* **77**, 333–45.

Hart, I. C., Blake, L. A., Chadwick, P. M. E., Payne, G. E., and Simmonds, A. D. (1984). The heterogeneity of bovine growth hormone. Extraction from the pituitary of components with different biological and immunological properties. *Biochem. J.* **218**, 573–81.

Hart, I. C., and Flux, D. S. (1973). The release of growth hormone in response to milking in the goat during early and late lactation. *J. Endocrinol.* **57**, 177–8.

Johnsson, I. D., and Hart, I. C. (1986). Manipulation of milk yield with growth hormone. In 'Recent Advances in Animal Nutrition, 1986'. (Eds W. Haresign and D. J. A. Cole.) pp. 105–23. (Butterworths: London.)

Jones, G. B. (1965). Determination of the specific activity of labelled blood glucose by liquid scintillation using glucose pentaacetate. *Analyt. Biochem.* **12**, 249–58.

Kelley, F. (1965). Improved method for microtitration of fatty acids. *Analyt. Chem.* **37**, 1078–9.

Kronfeld, D. S. (1965). Growth hormone-induced ketosis in the cow. *J. Dairy Sci.* **48**, 342–6.

Lindsay, D. B. (1983). Growth and fattening. In 'Nutritional Physiology of Farm Animals'. (Eds J. A. F. Rook and P. C. Thomas.) Ch. 7. pp. 261–313. (Longman: London and New York.)

Peel, C. J., Sandles, L. D., Quelch, K. J., and Herrington, A. C. (1985). The effects of long-term administration of bovine growth hormone on the lactational performance of identical-twin dairy cows. *Anim. Prod.* **41**, 135–42.

Peel, C. J., Steinhour, W. D., Bauman, D. E., Tyrell, H. F., Brown, A. C. G., Reynolds, P. J., and Haaland, G. L. (1982). Administration of bovine growth hormone to high yielding Holstein cows. II. Influence on irreversible loss and oxidation rate of free fatty acids and glucose. *J. Dairy Sci.* **65** (Suppl. 1), 120–1.

Pethick, D. W., Lindsay, D. B., Barker, P. J., and Northrop, A. J. (1981). Acetate supply and utilization by the tissues of sheep *in vivo*. *Br. J. Nutr.* **46**, 97–110.

Richard, A. L., McCutcheon, S. N., and Bauman, D. B. (1985). Responses of dairy cows to exogenous bovine growth hormone administered during early lactation. *J. Dairy Sci.* **68**, 2385–9.

Tindal, J. S., Knaggs, G. S., Hart, I. C., and Blake, L. A. (1978). Release of growth hormone in lactating and non-lactating goats in relation to behaviour, stages of sleep, electroencephalograms, environmental stimuli, and levels of prolactin, insulin, glucose and free fatty acids in the circulation. *J. Endocrinol.* **76**, 333–46.

Winkler, B., Steele, R., Altszulter, N., and de Bodo, R. C. (1964). Effect of growth hormone on free fatty acid metabolism. *Am. J. Physiol.* **206**, 174–8.

Zivan, J. A., and Snarr, J. F. (1973). An automated colorimetric method for the measurement of 3-hydroxybutyrate concentrations. *Analyt. Biochem.* **52**, 456–61.

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