EVALUATION OF THE INFLUENCE OF RECOMBINANT BOVINE SOMATOTROPIN (RBST) ON HEALTH PERFORMANCE, SOME BIOCHEMICAL PARAMETERS AND BODY WEIGHT IN FATTENING CALVES

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SUMMARY

The objective for this study was to assess the effect of recombinant bovine somatotropin (rbST) in health performance, some biochemical parameters and body weight. Two groups of growing fatting calves; Five calves in each; were used in this experiment which carried out in the farm of Faculty of Veterinary Medicine, Beni-Suef, Cairo University. The first group acted as control and the second was treated with Somatech® (recombinant bovine somatotropin) 500 mg for each. Routine traditional clinical examination before, during, and 4 weeks after finishing the experiment was carried out. Biweekly blood serum samples were collected from each animal of both groups for estimating total protein, albumin, Globulin and A/G ratio, some enzymes activities (ALT, AST, ALP, and LDH), levels of T_3 and T_4, Ca, P, urea, and creatinine. Biweekly live bodyweights were also recorded.

The clinical examination revealed that the health performance of rbST-treated calves was not adversely affected. Local swellings at site of injections were observed, the swellings were disappeared 8 days after each injection. The serum total protein, albumin and A/G ratio followed the same trends of decrease in rbST-treated calves comparing with those of the control calves. Conversely, globulin showed increase trend in rbST-treated calves. T_3 and T_4 followed the same trends of decrease in rbST-treated group comparing with that of the control group.

The changes in the serum enzyme activities (ALT, AST, ALP, and LDH), Ca, P, urea and creatinine were insignificant. The results of bodyweight gain in rbST-treated calves and the control calves were insignificant. This study emphasizes that at young age when the natural growth potential is high, an effect of rbST cannot be expected.

INTRODUCTION

Somatotropin was discovered over 50 years ago. Initial investigations showed that when growing rats were injected with crude pituitary extract, growth rate was increased. The first study conducted on dairy cows using recombinantly derived bovine somatotropin (rbST) was by *Bauman et al.,(1982)*. Since that time the quantity and scope of the researches with bST have increased exponentially.

Somatotropin hormone is a protein in nature produced by anterior pituitary gland, with 190 or 191 amino acids long and can have either one or two of different amino acids (leucine and valine) at position number 126 in protein sequence , *Wood et al., (1989). Chillard, (1988)* mentioned that exogenous bST must be administered daily in order to continue an augmented milk response because of rapidly clearance of bST from the blood stream and not stored in the body. Hence , the biotechnical synthesis of somatotropin (rbST) for farm animal has greatly affected live- stock production *Holzer et al., (1999)*. Studies with this neotechnology carried out on steers ,*Rumsey et al.(1996*) have invariably demonstrated a shift in partitioning of nutrients toward a considerable increase of protein and decrease of fat deposits.

A portion of the biological actions of somatotropin may be mediated by insulin like growth factor-1 (IGF-1), Administration of bST to steers may increase O_2 consumption by the liver *(Early et al., 1990)*, which in turns conceivably increase total heat production. This could be the way by which bST induce its action as growth- promoter *Stelwagen et al.,(1992). Scacchi et al., (2003)* reported that the growth hormone (GH) plays a key role not only in the promotion of linear growth but also in regulation of intermediary metabolism , body composition, and energy expenditure. They added the growth hormone appears to direct fuel metabolism towards the preferential oxidation of lipids instead of glucose and proteins and to convey the energy derived from metabolic processes towards the synthesis of protein. Furthermore, the nutritional status has to be regarded as a major determinant in the regulation of the somatotropin-somatomedin axis in animals and humans.

Because of the dearth of information on the effect of rbST on the metabolism of young growing calves, besides the

widespread using of rbST among fattening and dairy farms in Egypt, this study aimed to evaluate the influence of the effect of injection of rbST on health status, some blood serum biochemical parameters and body weight of growing fattening calves.

MATERIALS AND METHODS

Animals:

The current study was conducted on 10 mixed breeds calves of 6 – 8 months age, and weighing range from 211 to 226 Kg belonging to the farm of Faculty of Veterinary Medicine, Beni-Suef, Cairo University. The animals under the experiment were proven to be apparently healthy and free from internal and external parasites and apt for fatting. All animals were housed in a hygienic barn.

Nutrition:

The task of nutrition in this study was formulate a balanced ration with , crude protein , calcium , phosphorous, and vitamins for fattening calves. The *Table of National Research Council NRC (2001)*, were used to calculate the dry matter intake requirement for the fattening calves, each separately , according to different live bodyweight. Simplifying the feeding system, the ingredients were restricted to commercial mixture (14 % crude protein), rice straw, and commercial minerals and vitamins supplement. The amount of concentrates was adjusted weekly according to body weight of each calf.

Experiment:

The animals were divided into two equal groups , the control one and other treated with commercial rbST *(Somatech®)* . *Somatech®* is a sterile , sustained release injectable formulation of the recombinant-DNA drove bovine somatotropin (rbST) analogue in a single dose syringe (each contain 500 mg zinc methionyl rbST), produced by *Monsanto company, St. Louis, Missouri, USA*. The calves of the treated group were injected biweekly *S/C* with *Somatech®* at the area just behind the shoulder lasting for7 successive injections.

The clinical and laboratory examinations were performed daily for all calves during experiment and lasted for four weeks after the experiment. The weight of each animal in both groups were recorded biweekly.

Samples

The blood samples were collected at 7 a.m. from jugular vein in a clean, dry, sterile centrifuge tubes biweekly from all investigated calves on 9th days post-injection. The serum samples were separated in clean Eppendrof tubes and kept at –20 °C till the time of biochemical analysis.

Methods:

-The traditional clinical and laboratory examinations were performed daily for all calves during experiment and lasted for four weeks after experiment according to *Radostits et al., (2000).*

-The serum samples were used for determination of ; amino transferases (ALT& AST) activities by (*Reitman and Frankel , 1957)*, ALP activity by *(Kind and king , 1954),* , LDH activity by *Kachmar and Moss (1976)*, total protein by *Peters (1968)*, albumin by *Drupt (1974)*, total globulins and A/G ratio were calculated mathematically. T_3 and T_4 by *Chopra et al., (1972)* and *Tietz (1976) respectively,* by using test kits supplied by *Tinty Biotech (USA).* The serum calcium by *Robertson (1968)*, inorganic phosphorous by *Fiske and Subbarow (1925),* urea by *Patton and Crouch (1977),* and the kinetic estimation of creatinine by *Houot (1985).*

The data of this study were statistically analyzed by PC-State program according to *Mohan et al, (1985).*

RESULTS

Daily traditional preliminary clinical examination and inspection conducted throughout the experimental period revealed that there was no differences between the control group and rbST treated group in physical condition , mental status, posture and gait, rectal body temperature (38.5 - 39.2 °C) and (38.5 - 39.5 °C), respectively. respiration rate (30 - 39/min.) and (30 - 40/min.), respectively, and pulse rate (75 -105/min.) and (70 - 110/min), respectively. Detailed clinical examination of body system revealed no differences between the two groups. It is noticeable that all animals received rbST injections developed mild local inflammatory swellings at the site of injections. The swellings were in the form of subcutaneous inflammatory reactions tender and firm in palpation. The swellings disappeared within 8 days after injection and had no effect on rectal body temperature. The swellings were regressed and became non detectable four weeks after the last injection.

The results of on serum concentrations of total protein , albumin, globulins (g/dl) and A/G ratio in the control and rbST-treated calves are shown in Table (1).

The effect of rbST on serum enzyme activities of Alanine aminotransferase (ALT), Aspartate aminotransferase (AST) and alkaline phosphatase (ALP) and lactate dehydrogenase (LDH) in control and treated calves are shown in Table (2).

The results of serum levels of T_3, T_4 and concentrations of Ca and P in the control and rbST-treated calves are tabulated in Table (3).

The results of serum concentrations of urea and creatinine in the control and rbST-treated calves are shown in listed (4).

The results of the body weight (Kg) in the control and rbST-treated calves are shown in Table (5) and illustrated in Figure (1).

DISCUSSION

Based on clinical examination, the health performance of rbST treated calves was not adversely affected. These findings were in agreement with that mentioned by *Eisemann et al., (1986)* and *Neathery et al.,(1991)*. The local swellings observed in rbST-treated calves at the site of injections were similar to those described by Whitaker et al.,(1988) and *Debackere (1990)* in dairy cattle. Local swellings in the site of injections in both dairy cattle and fattening calves suggest that this reaction may be due to local irritation caused by adjuvant substances in *Somatech®*.

Effect of rbST on the blood serum total protein was the net of its effect on both albumin and globulins (Table 1). Generally the trend of total protein of rbST treated calves was less than that of control calves. Irrespective of significant decreases in the 2^{nd} , 4^{th} and 5^{th} periods of rbST treated calves , the changes were within normal physiologic limits mentioned by *Benjamin (1984).*

The results of albumin in the present study followed virtually the trend of total protein, where albumin concentrations of rbST treated calves were less than that of the control calves at any period of experiment. The significant decreases were noticeable at 1^{th} , 3^{rd} , 5^{th} and 7^{th} periods comparing with control calves. The changes in albumin in this study were within the normal physiologic

range reported by *Benjamin (1984).* Conversely , the results of globulins showing increases of mean values of rbST-treated calves comparing with those of the control calves. Apart of significant increases at 3 rd, 5 th and 6 th periods , the changes were within the physiologic limit mentioned by *Benjamin (1984).* A/G ratio followed virtually the trend of total protein and albumin. The mean values of A/G ratio in rbST treated calves were less than that of the control calves. Insignificant decreases of A/G ratio in rbST treated calves were observed in the 2nd and 4th periods post treated only , while the decreases of A/G ratio of rbST in the rest of periods were significantly decrease. A/G ratio ranges in both rbST treated calves and control calves were within the normal physiologic ranges, *Benjamin (1984).*The results of blood serum protein in this study were supported by the results of body weight (table , 5 and fig. 1), in which the average daily gain was decreased in rbST treated calves , comparing with those of the control calves.

These results were not consistent with many studies as that reported by *Fabry et al.,(1991)* ad *Rumsey et al.,(1996).* They have shown a considerable shift in the partitioning of nutrients toward an increase in protein and decrease in fat deposit. *Kitagawa et al., (2001)* accounted the decrease in serum protein level and body gain – in spite of normal appetite – to higher serum somatotropin and lower serum insulin like growth factor-1. These suggested defects in somatotropin- IGF-1 axis such as in the somatotropin receptor.

Concerning the effect of rbST on ALT, AST , ALP and LDH activity (Table 2), the mean values of rbST-treated calves changed insignificantly, when compared with those of the control calves. With exception of mean values of AST at 3 rd and 6 th periods, where there were significantly increases in rbST treated calves , comparing with that of the control calves. The changes in ALT, AST, ALP, and LDH activities in rbST treated calves and the control calves were within the normal physiologic ranges mentioned by *Benjamin (1984). Vicini et al., (1990)* and *Graf et al., (1991)* obtained similar results in dairy cattle. Similar results of ALP activity in calves were reported by *Neathery et al.,(1991).*

It is important to mentioned that conversion of thyroxine (T_4) into metabolically active hormone triiodothyronine (T_3) is catalyzed by 5-monodeiodinase, mainly in extrathyroidal tissues *(Kaneko et al., 1997).*

The results of T_3 in rbST treated calves (Table 3) showed decreases in different periods of experiment comparing with that of control calves. The decreases of T3 in rbST treated calves were significant at 3 rd and 6th periods of experiment comparing with control calves. The results of T_4 were similar to the results of T_3 but the decrease of T_4 in rbST-treated calves was significant at the 1st and 2nd periods comparing with control calves. The results were in accordance with that reported *by Holzer et al. (2000)*. The results of T_3 and T_4 could be explained on basis of increase of serum somatotropin in rbST treated calves which exert an inhibitory effect on hypothalamus by stimulating the release of somatostatin which in turn has inhibitory effect on thyroid stimulating hormone TSH *(Kaneko et al., 1997).*

The results of calcium and phosphorus in rbST-treated calves (Table 3) were insignificantly changed comparing with those of control calves. These results are in agreement with that reported by *Neathery et al., (1991).*

The results of urea and creatinine of rbST treated calves (Table 4) showed insignificant changes from that of the control calves . These results are in agreement with that obtained by *Guillermo et al., (1990)* and *Neathery et al., (1991).*

The results of body weight were unexpected changes in this study (Table 5 & Figure 1), the average body gain of rbST-treated calves were 1, 9.25, 32.25 and 7.75 kg/ 14 days comparing with 11 , 12.75 ,39.25 and 2.25 Kg/ 14 days in control calves. These results were in agreement with *Eismann et al., (1989), Neathery et al., (1991)* and *Holzer et al., (2000)*, who conducted the studies on calves approximately of the same age. Meanwhile, these results were disagree with that reported by *Moseley et al., (1992)* and *Preston et al., (1995),* who conducted their studies on steer (over 11 months).

It is concluded that the age of the calves has a crucial factor, in context of application of other factors as nutrition and health of animals, in using somatotropin. This study emphasize that at young age, when the natural growth potential is high, an effect of rbST treatment cannot be expected

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Table (1): Serum concentrations of total protein , albumin, globulins and A/G ratio of the control and rbST-treated calves

Periods weeks	Total proteins (g/dl)		Albumins (g/dl)		Globulins (g/dl)		A/ G ratio	
	Control	BST-treated	Control	BST-treated	Control	BST-treated	Control	BST-treated
1st 2wks	6.68 ± 0.23	6.67 ±0.17	3.58 ± 0.20	3.25 * ±0.13	3.15 ±0.24	3.50 ±0.17	1.11 ±0.10	0.91 * ±0.01
2nd 4 wks	7.37 ±0.20	6.75 * ±0.004	3.58 ±0.27	3.48 ±0.26	3.16 ±0.11	3.46 ±0.33	1.12 ±0.003	1.02 ±0.13
3rd 6wks	6.94 ±0.26	6.86 ±0.01	3.44 ±0.13	3.10* ±0.13	3.22 ±0.30	3.82 * ±0.01	1.08 ±0.12	0.82 * ±0.01
4th 8 wks	7.10 ±0.01	6.81 * ±0.01	3.40 ±0.11	3.26 ±0.19	3.35 ±0.23	3.59 ±0.17	1.02 ±0.004	0.91 ±0.01
5th 10 wks	7.15 ±0.01	6.96 * ±0.12	4.14 ±0.12	3.37* ±0.16	3.12 ±0.19	3.59 * ±12.34	1.33 ±0.001	0.94 * ±0.01
6th 12 wks	7.32 ±0.01	7.03 ±0.26	4.14 ±0.14	3.83 ± 0.24	3.14 ±0.01	3.42 * ±0.19	1.31 ±0.01	1.12 * ±0.10
7th 14wks	7.17 0.15 ±	7.02 ±0.27	4.15 ±0.14	3.83 * ±0.17	3.29 ±0.22	3.64 ±0.22	1.29 ±0.11	0.93 * ±0.01

* Significant difference from control at P < 5% .

Table (2): Serum enzymes activities of ALT, AST, ALP and LDH in the control and rbST-treated calves

Period/week	(ALT) u/l		(AST) u/l		ALP u/l		(LDH) u/l	
	Control	BST-treated	Control	BST-treated	Control	BST-treated	Control	BST-treated
1st (2wks)	38.2 ±1.19	37.29 ±2.82	161.07 ±15.04	175.34 ±0.98	132.25 ±21.52	126.87 ±25.85	357.62 ±8.29	347.73 ± 15.58
2nd (4wks)	37.24 ±1.15	36.01 ±2.00	160.10 ±7.92	168.52 ±10.50	116.82 ±9.73	137.98 ± 12.33	347.73 ±12.48	346.08 ±11.42
3rd (6wks)	39.04 ±1.92	38.78 ±3.69	157.49 ±11.00	174.90* ±1.56	122.33 ±7.95	126.80 ±10.08	351.04 ±13.61	342.78 ±5.38
4th (8wks)	39.78 ±2.05	38.78 ±3.30	165.20 ±6.75	172.24 ±4.82	128.20 ±8.49	138.18 ±8.20	346.08 ±12.62	329.60 ±16.15
5th 10wks	40.12 ±1.36	37.69 ±3.31	164.88 ±13.47	174.55 ±1.56	135.30 ±11.23	146.05 ±12.34	352.68 ±3.81	347.73 ±8.30
6th 12wks	40.27 ±1.70	39.55 ±3.28	159.03 ±13.84	174.1 * ±2.50	137.46 ±12.64	148.56 ±9.17	352.68 ±3.81	347.73 ±8.30
7th 14wks	40.49 ±1.95	39.39 ±3.79	162.80 ±10.17	173.95 ±3.74	134.18 ±12.46	146.12 ±8.35	352.68 ±3.81	344.4 ±11.26

* Significant difference from control at P < 5% .

Table (3): Serum levels of T_3 and T_4 and concentration of calcium and phosphorus of the control and rbST-treated calves

Periods/ weeks	T_3 ng/dl		T_4 ug/dl		Calcium mg/dl		-Phosphorous mg/dl	
	Control	BST-treated	Control	BST-treated	Control	BST-treated	Control	BST-treated
1st 2Wks	3.17 ±0.56	2.76 ±0.41	8.51 ±0.51	7.33* ±0.32	10.21 ±0.34	10.40 ±0.37	5..49 ±0.18	5.43 ±0.024
2nd 4wks	3.37 ±0.75	2.70 ±0.29	8.80 ±0.21	7.22* ±0.71	10.46 ±0.12	10.35 ±0.30	5.60 ±0.39	5.57 ±0.46
3rd 6wks	2.90 ±0.35	2.03* ±0.26	8.20 ±0.65	7.56 ±0.76	9.85 ±0.53	10.42 ±0.49	5.47 ±0.12	5.46 ±0.25
4th 8wks	3.01 ±0.24	2.70 ±0.36	8.07 ±0.68	7.72 ±0.48	9.85 ±0.16	10.18 ±0.50	5.44 ±0.17	5.51 ±0.23
5th 10wks	3.35 ±0.79	2.90 ± 0.32	8.47 ±1.22	7.54 ±1.06	10.00 ±0.54	10.13 ±0.01	5.59 ±0.19	5.62 ±0.38
6th 12wks	3.23 ±0.27	2.57* ±0.26	8.39 ±1.09	7.68 ± 0.88	10.09 ±0.40	10.34 ±0.41	5.54 ±0.20	5.42 ±0.28
7th 14Wks	3.09 ±0.77	3.06 ±0.70	8.37 ±0.99	7.51 ±0.68	10.09 ±0.40	10.34 ±0.41	5.39 ±0.01	5.40 ±0.35

* Significant difference from control at $P < 5\%$.

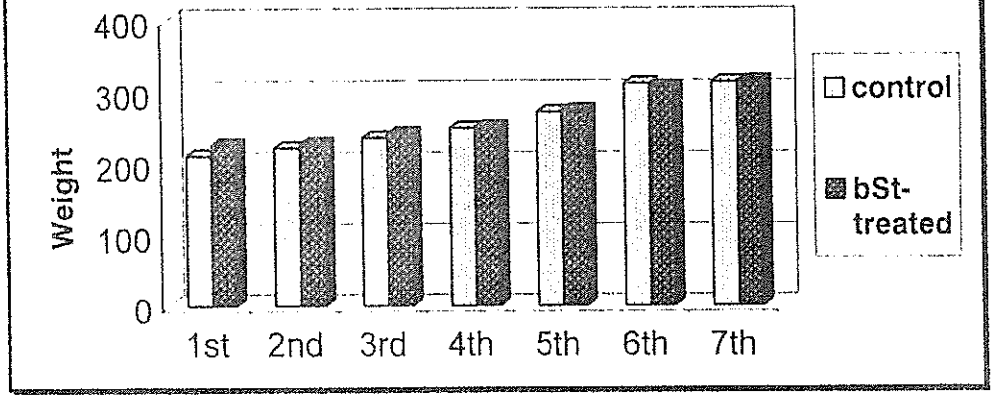
Table (4): Serum concentrations of urea and creatinine of the control and rbST-treated calves

Periods/weeks		1st (2 W)	2nd (4W)	3rd (6W)	4th (8 W)	5th (10 W)	6th (12 W)	7th (14 W)
Urea Mg/dl	(control)	29.99 ± 2.69	29.98 ± 2.94	30.35 ± 2.38	30.28 ± 1.60	30.49 ± 1.20	30.11 ± 2.50	30.118 ± 3.02
	(bST-treated	33.28 ± 1.89	33.56 ± 0.40	33.33 ± 0.80	33.54 ± 0.54	33.47 ± 1.63	32.50 ± 1.65	34.17 ± 2.18
Creatinine Mg/dl	(control)	1.43 ± 0.22	1.30 ± 0.01	1.34 ± 0.11	1.35 ± 0.03	1.33 ± 0.01	1.30 ± 0.01	1.34 ± 0.12
	BST-treated	1.42 ± 0.23	1.40 ± 0.14	1.37 ± 0.14	1.35 ± 0.13	1.39 ± 0.12	1.40 ± 0.14	1.50 ± 0.01

Table (5): The body weight (Kg) of the control and bST-treated calves

Periods/ weeks		1 st (2 W)	2nd (4W)	3 rd (6W)	4th (8 W)	5th (10W)	6th (12W)	7th (14W)
Body weight (Kg)	Control	211.00 ± 37.57	222.00 ± 30.10	236.00 ± 37.86	248.75 ± 41.23	271.50 ± 40.04	310.75 ± 33.55	313.00 ± 39.06
	BST-treated	226.25 ± 21.21	227.25 ± 28.92	241.50 ± 24.53	250.75 ± 21.75	273.75 ± 29.14	306.00 ± 27.90	313.75 ± 23.46

Fig. (1): The body weight (kg) in the control and rbST-treated calves



الملخص العربي

تقييم تأثير هرمون النمو البقرى المخلق على المؤشرات الصحية وبعض القياسات البيوكيميائية ووزن الجسم فى عجول التسمين

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استهدف هذا البحث دراسة تقييم تأثيرات هرمون النمو البقرى المحضر وراثيا والمستخدم تجاريا)سوماتيك) على صحة عجول التسمين , وبعض القياسات الكيميائية الحيوية وكذا تأثيره على معدل النمو ولتحقيق هذا الهدف تم إجراء التجربة على عدد 10 عجول تسمين تتراوح أعمارهم من 6 – 8 أشهر وتتراوح أوزانهم بين 211- 226 كجم. وقسمت إلى مجموعتين متساوية الأولى ضابطة والثانية معالجة بالحقن الموضعي تحت الجلد بعقار السوماتيك بمعدل حقنة كل 14 يوم ودام ذلك 7 حفنات متتالية.

وأظهرت النتائج الآتي:- ليس هناك فروقا واضحة بين المجموعة الضابطة والمعالجة في الحالة الصحية العامة ولكن وجد التهاب موضعي مكان الحقن والذي يختفي تلقائيا في خلال 8 أيام من الحقن . البروتين الكلى والألبومين ونسبة الألبومين للجلوبيولين كان لهم نفس المسلك حيث وضح قلتهم بالنسبة للمجموعة الضابطة ولكن في حدود المستوى الطبيعي بينما الجلوبيولين زاد نسبيا عن المجموعة الضابطة ولكن في حدود المستوى الطبيعي, لم يكن هناك تغيرات ملحوظة في مستوى نشاط الإنزيمات الآتية (ALT, AST, ALP LDH) وكذا لم يكن هناك تغيرات في تركيز الكالسيوم والفسفور والبولينا والكريلتنين ,بينما كانت هناك انخفاضا عاما والذي كان معنويا في بعض فترات التجربة لهرمونات الغدة الدرقية , T3 T4وكانت نتائج معدل النمو غير متوقعة حيث أنها بوجه عام أقل من المجموعة الضابطة . ونخلص من هذا البحث أنه لايمكن استعمال عقار هرمون النمو البقرى المحضر وراثيا في السن الصغير الذي يكون فيه معدل النمو الطبيعي عاليا.

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