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STUDIES ON SELENIUM AND/OR VITAMINE ADMINISTRATION TO EGYPTIAN BUFFALO CALVES. I- EFFECT ON BLOOD SERUM SELENIUM LEVEL, DAILY GAIN AND SOME BLOOD CONSTITUENTS

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SUMMARY

A trial was conducted to determine the effect of selenium (Se) injection, vitamin E (orally) and Se+Vit E on blood serum selenium level, daily gain and some blood constituents (total protein, albumin, globulin, A/G ratio, total cholesterol, total lipids, GOT and GPT). Egyptian buffalo calves (15 males and 16 females) raised on Buffalo Research Station at Mehallet Mousa, Kafr El-Sheikh Governorate, were randomly assigned after weaning to four groups. The first group (8 calves) was injected with Se as sodium selenite intramuscularly in a dose of 0.125 mg/ kg body weight. The second group (8 calves) received vitamin E orally (200-400 mg/ head). The third group (8 calves) received Se+Vit E. Se or vitamin E divided into two doses and administered in two successive weeks and repeated every three months. The fourth group (7 calves) was kept as control. Treatment with Se+Vit E significantly (P<0.05) increased concentration of Se in blood serum of calves at intervals of 2, 14, 16 and 30 weeks after the onset of treatment. Daily body weight gain improved in treated groups by 42, 27 and 65 gm/ day for Se, Vit E and Se+Vit E groups, respectively. Blood biochemical constituents were not affected by different treatments, while interaction between treatment and intervals were significant (P<0.01) or (P<0.05) for blood constituents studied except total lipids.

Keywords: Buffalo, calves, selenium, vitamin

INTRODUCTION

Selenium is an essential nutrient for animals and diseases due to its inadequacy in livestock of a world wide distribution. Ruminants are subjected

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frequently to sever dietary deficiencies of this micro-element (Hidiroglou and Jenkins, 1975). Soil and therefore the pastures they carry, vary widely in their selenium content depending largely on their geological origin. A number of factors also reduce the availability of soil selenium to plants and in turn to animals. The animal essentiality for selenium is based on its incorporation into the enzyme glutathione peroxidase (GSH-Px). This enzyme catalyzes the breakdown of hydrogen peroxidas and lipid hydroperoxidases in body tissues and fluids. Depending upon this function, selenium plays a role in protection against hepatic damage due to oxidative stress (Stuart, 1987), in reproductive function in buffalo dams (Youssef et al., 1988), and buffalo bulls (Omaima-Mahmoud, 1994). In immunological reactivity (Hassanin et al., 1988); in muscular dystrophy and growth rate of calves (Gleed et al., 1983), in growth rate of suckling buffalo calves (El-Ayouty et al., 1996).

In the last few years, biological uses of selenium was considered as an economic tool in many breeding centers in different localities in ARE. Selenium functions are to a certain extent duplicated by vitamin E, but it is not capable for replacing the vitamin in all situations. Evidence suggests that requirement for vitamin E and Se are mostly independent (Stowe et al., 1988, and Van Saun et al., 1989).

No enough attention had been given to the Se and vitamin E status of the growing buffalo calves in Egypt. Some investigations on the buffalo calves at Mehallet Mousa Research Station declared that the growing calves are maintained slow growth rate (Metry, 1997), and encountered with some health problems, such as weakness. The favorable results obtained by Sesupplementation of buffalo dams (Awad et al., 1985), proved that these animals raised on Se-deficient area. Therefore, the major objective of this study was to monitor serum Se, daily weight gain and some serum metabolites of buffalo calves supplemented with Se and/or vitamin E (∞-tocopherol) at three months intervals during 30 weeks after weaning.

MATERIALS AND METHODS

The present study was conducted on 31 buffalo calves (15 males and 16 females) after weaning raised on Buffalo Research Station at Mehallet Mousa, belonging to Animal Production Research Institute, Ministry of Agriculture. The calves were divided randomly into four groups. The first group (8 calves) was injected intramuscularly with 0.125 mg Se/kg body wt. as sodium selenite. The second group (8 calves) was supplemented orally with 200-400 mg/ head vitamin E (dl-alpha-tocopherol acetate, Pharco Pharmaceuticals, Alexandria). The third

group (8 calves), was treated with the same doses of Se+Vit E. The Se and vitamin E values were split into two equal doses, administered with one week interval. The treatments started after weaning (15 weeks-old) and repeated at three months intervals till the calves reached 45 weeks-old. The fourth group (7 calves) was kept without treatment as control.

The calves were fed daily on calves starter (1.5 kg/head) consisted of 40% yellow maize, 22% wheat bran, 20% linseed meal, 5% soybean meal, 5% maize glutin, 5% molasses, 2% calcium carbonate and 1% sodium chloride. Protein content (CP) of the starter was 17.5%. Also, the calves were fed on 1.0 kg/head clover hay (12.3% CP) or green clover and wheat straw (1.0 kg/head). At six months of age, the starter was replaced by concentrate mixture (2.0 kg/head), consisted of 22% yellow maize, 32% cotton seed meal, 26% wheat bran, 12% rice bran, 5% linseed meal, 2% molasses, 0.5% limestone and 0.5% sodium chloride. Protein content of the concentrate mixture was 16%. The wheat straw was replaced by rice straw (1.5 kg/head). The calves were group fed and the level of feeding was changed according to their age change (feeding system practices on the farm). The calves were watered twice daily and body weights were recorded at biweekly intervals. Daily weight gain (kg) for the entire experimental period (30 weeks) was calculated.

Blood samples from jagular vein were collected just before treatment, at 1 and 2 weeks after treatment and repeated at three months intervals (at 14, 15, 16, 28, 29 and 30 weeks after the initiation of the experiment). Serum was separated and stored at -20°C till analysis. Blood serum constituents were determined according to the following: Se (fluorometrically, just before treatment, at 2, 14, 16, 28 and 30 weeks after the initiation of the experiment); total protein (kits "bio Merieux", France); Albumin and transaminase enzymes (kits "Biochemical Trade Inc." U.S.A.); total lipids (kits "bio Merieux", France); cholesterol (kits "Stanbio" U. S. A.) and globulin (calculated by subtracting albumin concentration from total protein concentration).

The data of daily gain were analyzed using GLM procedure in SAS® program and the sex of the calf was included and the following model was used.

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 \begin{array}{lll} y_{ijk} = & \mu + T_i + S_j + T_i^*S_j + e_{ijk} \\ y_{ijk} = & \text{The observation on the calf;} \\ \mu = & \text{Common mean;} \\ T_i = & \text{Fixed effect of the i}^{th} \text{ treatment where is I=(1, 2, 3, and 4);} \\ S_j = & \text{Fixed effect of the j}^{th} \text{ sex of calf where is j=(1 and 2);} \\ T_i^*S_j = & \text{Interaction between the i}^{th} \text{ treatment and j}^{th} \text{ sex;} \\ e_{ijk} = & \text{The random error.} \end{array}
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The data of metabolic profile and selenium concentration in blood serum were analyzed using model 3 in Harvey program (1987).

 $\begin{array}{ll} y_{ijkl} = & \mu + T_i + C_j \left(T_i\right) + I_k + I_k * T_i + e_{ijkl} \\ y_{ijkl} = & \text{The observation on the calf;} \end{array}$

μ= Common mean;

T_i= Fixed effect of the ith treatment where is i=(1, 2, 3, and 4);

 $C_{j}(T_{i})$ = The random effect of the j^{th} calf within the i^{th} treatment where is j=(1 to 31); Fixed effect of the k^{th} interval where is k=(1 to 6) for selenium concentration, while

k=(1 to 9) for metabolic profile;

 $I_k^*T_i^=$ Interaction between the k^{th} interval and the i^{th} treatment;

e_{ijkl}= The random error.

RESULTS AND DISCUSSION

The least squares means of serum Se concentration arranged by treatment are in Table 1. Initial mean Se concentration in serum for the Se, Vit E, Se+Vit E and control groups ranged from 0.026 to 0.030 μ g/ml (Figure 1). This range was higher than initial concentration of 0.020 to 0.025 μ g/ml reported by Stowe *et al.* (1988) for the Holstein cows during dry period. Selenium injection with or without Vit E supplementation increased (P<0.05) serum Se level (0.034 and 0.033 μ g/ml, respectively, vs. 0.028 μ g/ml in control). This is in accordance with that reported by Droke and Loerch (1989); Stowe *et al.* (1992); Van Saun (1990) and El-Ayouty *et al.* (1991) who found that Se alone or in combination with vitamin E increased serum Se level. While, mean serum Se concentrations of Se group were consistently lower than in Se+Vit E group (Figure 1). It appears that both Se and Vit E are acting synergistically (Horton and Jenkins, 1978). On the other hand, Vit E supplementation alone increased serum Se level nonsignificantly compared with control group (0.029 vs. 0.028 μ g/ml). The same results obtained in sheep by Whanger *et al.* (1977).

Least squares means for daily gain (kg) of male and female calves in different treatments during the entire experiment period (30 weeks) are presented in Table 2 and Figure 2. The highest daily gain (0.409 kg) was observed for Se+Vit E followed by Se, Vit E and control groups (0.386, 0.371 and 0.344 Kg, respectively). The Se, Vit Eand Se+Vit E treatments improved significantly (P<0.05) daily gain during the entire experimental period by 42, 27 and 65 gm than those of control group. At the end of experiment the increase in body weights were 8.82, 5.67 and 13.65 kg for the respective groups. These results are in parallel with results recorded by Stuart (1987) after supplementation of calves with intraruminal Se pellets. Wichtel et al. (1996) mentioned that Se in combination with

Table 1. Least squares means for serum Se level (µg/ml) of male and female buffalo calves injected with sodium selenite with or without vitamin E orally, supplemented with vitamin E orally and control groups

	Ireatmen	ent			Intervals	als (week	2			
Item	Se	×. E	Se+ Vit E	10	0	2	14	16	28	30
No. of observation	32	32	32	29	8	23	23	23	23	23
Serum Se (µg/ml)	.033ª	.029 ^b	.034ª	.028 ^b	.029 ^b	.034ª	.030ab	.032 ^{ab}	.031 ^{ab}	.029 ^b
= 1	+1	-1-1	+1	+1	+1	+1	+1	+1	+1	+1
Standard error	.001	.001	.001	.002	.003	.002	.002	.002	.002	.002

Table 2. Least squares means for daily gain (kg) of male and female buffalo calves injected with sodium

	Ireatment				Treatment Sex of calves	lves
Item	Se	N M M	Se+ Vit E	Control	Male Fe	Female
No. of animals	8	80	80	7	15	16
Daily gain (kg)	0.386 ^{ab}	0.371ab	0.409 ^a	0.344b	0.396	0.359 ^b
**	1 1	+1	+1	+1	- y 1	+1
Standard error	0.01	0.01	0.01	0.02	0.01	0.01

Vit E improved daily gain in calves. Droke and Loerch (1989) found that sick steers injected with Se and (or) Vit E tended to have slightly higher daily gains, but in healthy steers, the average daily gain was not affected. On the contrary, several investigators didn't demonstrate any effect of oral Se supplementation on body weight gain of calves (Abdelrahman & Kincaid 1995, and El-Ayouty et al., 1996) and in pigs (Kornegay et al., 1993).

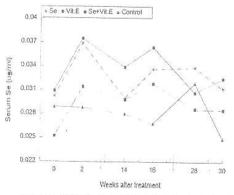


Figure 1. Serum Se concentration by intervals for treated and control groups.

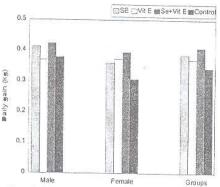


Figure 2. Daily gain (kg) for male and female buffalo calves during the whole experiment as affected by treatments.

Sex of calves (Table 2) affected significantly (P<0.05) daily gain (0.396 vs. 0.359 kg for male and female calves, respectively). Also, Se+Vit E treatment improved body weight gain in male and female calves (0.423 and 0.395 kg, respectively) compared with .379 and .308 kg for the respective sex in the control group (Figure 2).

Serum proteinogram in different treated groups and according to the intervals of experiment was postulated in Table 3 and Figure 3. The effect of treatment was more obvious significantly (P<0.01) on albumin fraction. It might be reflected on A/G ratio significantly (P<0.01) particularly in Vit E group compared with control one (1.57 vs. 1.24). Morozov et al., (1984) demonstrated that there was no effect of sodium selenite treatment on calves 2-4 weeks old on total protein and its fractions. However, it indicated that after selenium absorption, selenium associated with plasma protein (McConnell and Levy, 1962). Shamberger (1983) mentioned that albumin is immediate acceptor of injected selenium which intensified its synthesis. Moreover, the higher albumin concentration obtained in the present study might be indicated the normal health status of calves (Jagos, et al., 1981). Serum protein levels and its fractions increased significantly (P<0.01) with lowering of A/G ratio by successive intervals of the experament

(Figure 3). Jensen (1978) reported that serum protein and its fractions are related to age. Furthermore, El-Ashry et al. (1994) found that plasma protein increased gradually after 6 months old Egyptian buffalo hefiers and reached the maximum at 15 months-old. However, Katunguka-Rwakishaya et al. (1985) cited that plasma protein in calves decreased consistantly within age between 4 and 22 weeks. They added that A\G ratio decreased within the same period, similar with our results.

Table 3. Least squares means ±SE of serum proteinogram of Se (injection), vitamin E (orally), Se plus Vit E treatments and control groups of buffalo calves

Item	Obs	protein gm/100 ml	tments and co albumin gm/100 ml	globulin gm/100 ml	A/G ratio
Overall mean	279	6.96±.04	4.12±.01	2.40±.05	1.34
Treatment:		NS	**	NS	**
Se	72	6.98±.08 ^a	4.10±.03 ^b	2.41±.10 ^a	1.31 ^b
Vit E	72	6.86±.08°	4.20±.03 ^a	2.27±.10 ^a	1.57 ^a
Se+Vit E	72	7.02±.08 ^a	4.09±.03 ^b	2.44±.10 ^a	1.24 ^b
Control	63	6.97±.09 ^a	4.08±.03 ^b	2.49±.11 ^a	1.22 ^b
Intervals:		**	**	**	**
Initiation of treatment	31	6.79±.09 ^{cd}	4.03±.04 ^b	2.49±.11 ^b	1.05°
week-1	31	6.28±.09 ^d	4.10±.04 ^b	1.60±.11°	2.02 ^a
week-2	31	6.35±.09 ^d	4.11±.04 ^{ab}	1.86±,11°	1.78 ^{ab}
week-14	31	7.20±.09 ^{ac}	4.21±.04 ^a	2.38±.11 ^b	1.39 ^{bc}
week-15	31	6.82±:09 ^{bc}	4.11±.04 ^{ab}	2.39±.11 ^b	1.24 ^{bc}
week-16	31	7.34±.09 ^a .	4.21±.04 ^a	2.66±.11°	1.20°
week-28	31	7.11±.09 ^{ab}	4.09±.04 ^b	2.49±.11 ^b	1.21 ^c
week-29	31	7.24±.09 ^{ab}	4.18±.04 ^a	2.54±.11 ^b	1.12°
week-30	31	7.47±.09 ^a	4.00±.04 ^b	3.20±.11 ^a	1.02 ^c

NS Non significant. Significant (P<0.01).

a,b,c,d Values with different superscript in the same column within item are differ significantly (p<0.01).

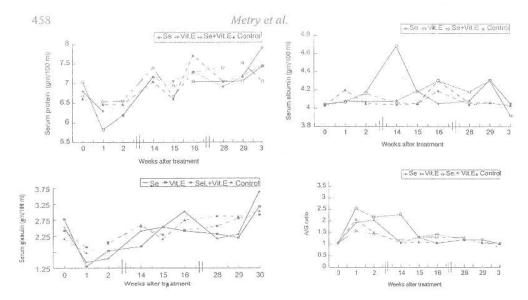


Figure 3. Blood serum protein, albumin, globulin and A/G ratio of buffalo calves injected sodium selenite (0.125 mg/kg BW) with or without vitamin E orally (200-400 mg/head), vitamin E and Control groups on three occasions 0-2 wk, 14-16 wk and 28-30 wk after the initiation of the experiment.

Interaction of treatment with intervals was significant (P<0.01) in serum proteinogram (Figure 3). It may be due to the effect after the successive treatment (at wk-2, wk-16 and wk-30). Serum globulin level i.e. in control and Se groups was 2.49 and 2.44 gm/100 ml, respectively. These values were less than 4.30 mg/ ml plasma globulin level during the last 8 weeks of gestation in Egyptian buffalo (Metry et al., 1994).

Serum GOT and GPT (Table 4 and Figure 4) were not affected by Se and/or Vit E supplementation. The overall means were 44.30 and 18.70 μ I, respectively, which are within the normal ranges (22-71and 5-29 μ I) recorded in normal Holstein-Freisian calves reported by Vagher et al. (1973). However, serum level of GOT greatly increased in hepatic damage in cow (Holtenius and Jacobsson, 1964). Furthermore, Dotta and Robutti (1972) and Kursa (1975) detected that calves with clinical muscular dystrophy showed increases in GPT. While, the effects on serum GOT and GPT levels were prominent (P<0.01) by the intervals of the experiment and its interaction with treatment.

Table 4. Least squares means ±SE for serum GOT, GPT, cholesterol and total lipids as affected by Se (injection) , vitamin E (orally) and Se plus Vit E treatments for buffalo calves

Item	Obs	Serum GOT μ/Ι	Serum GPT μ / Ι	Total lipids mg/100 ml	Cholestero
Overall mean	279	44.33±0.67	18.72±.34	443.4±5.9	85.1±1.5
Treatment:		NS	NS	NS	NS
Se	72	45.61±1.32 ^a	19.50±.66°	438.3±11.6ª	83.3±2.9ª
Vit E	72	45.09±1.32a	18.05±.66 ^a	438.1±11.6°	84.6±2.9 ^a
Se+Vit E	72	42.34±1.32 ^a	18.56±.66ª	447.0±11.6 ^a	87.3±2.9ª
Control	63	44.29±1.41a	18.76±.71°	449.9±12.4°	
Intervals:		**	**	**	85.2±3.1 ^a
Initiation of treatment	31	43.19±2.0 ^b	19.70±.93bc	474.0±12.0ab	80.7±2.8 ^{bc}
week-1	31	41.54±2.0b	14.02±.93°	365,5±12.0°	73.5±2.8°
week-2	31	40.57±2.0b	16.11±.93°	421.0±12.0b	79.7±2.8°
week-14	31	43,00±2.0 ^b	18.02+.93bc	427.3±12.0 ^b	91.6±2.8 ^b
week-15	31	51.89±2.0a	17.08±.93°	434.9+12.0b	91.0±2.8°
week-16	31	56.31±2.0a	18.09±.93bc	451.8±12.0 ^b	A STATE OF S
week-28	31	42.45±2.0b	23.33±.93 ^{ab}	453.0±12.0 ^b	86.1±2.8 ^{bc}
week-29	31	31.20±2.0°	14.39±.93°	459.2±12.0 ^b	84.2±2.8 ^{bc}
week-30	31	48.80±2.0 ^{ab}	27.69±.93°	503.5±12.0°	70.1±2.8° 108.6±2.8°

NS Non significant. ** Significant (P<0.01). **Significant (p<0.01). **Signifi

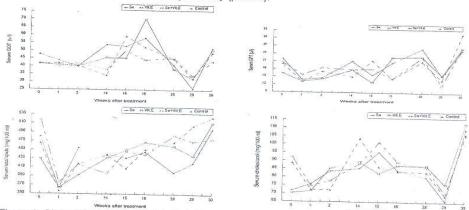


Figure 4. Blood serum GOT, GPT, total lipids and cholesterol of buffalo calves injected sodium selenite (0.125 mg/kg BW) with or without vitamin E orally (200-400 mg/head), vitamin E and Control groups on three occasions 0-2 wk, 14-16 wk and 28-30 wk after the initiation of the experiment.

Temporary decrease of their level may be due to the repeated supplementation of Se and/or Vit E. El-Ayouty et al. (1991) cited that the activity of GOT differed from time to time during the experiment without significant differences between the Se-treated and control calves. The overall means of serum total lipids and cholesterol concentrations (Table 4 and Figure 4) were 443.0 and 85.0 mg/100 ml, respectively. Serum cholesterol level is lower in comparable with that in cows (93-172 mg/ 100 ml) as recorded by Arave et al. (1975) and Baumgatner (1979), but higher than the range of 39-45 mg/ml in lactating buffalo (Metry et al., 1994). Effect of treatment during the experiment was not obvious. While, serum total lipids and cholesterol levels were significantly increased (P<0.01) by successive intervals. In the same pattern (Arave et al., 1975) proved that serum cholesterol increased with increasing age of calves.

CONCLUSION

One of the most surprising features of this experiment was the absence of clinical disease among even the unsupplemented animals, inspite of their low blood serum concentration of Se. Dispite this absence of disease, the evidence suggested that supplementing the buffalo calves with Se and/or vitamin E significantly improved daily gain. Additional studies are needed in different geographical locations to establish the effects with higher doses and shorter intervals between injections.

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دراسات على معاملة عجول الجاموس المصرى بالسيلينيوم مع أو بدون فيتامين هـ ١- التأثير على مستوى السيلينيوم في سيرم الدم ومعدل النمو اليومي وبعض مكونات الدم.

جميل حبيب مترى ' - رؤوف حلمي يوسف ' - رضا محمد خطاب ا

1- معهد بحوث الانتاج الحيواني - مركز البحوث الزراعية - وزارة الزراعة، ٢- معهد بحوث صحة الحيوان - مركز البحوث الزراعية - وزارة الزراعة

أجريت هذه الدراسة لتقدير تأثير حقن السيلينيوم أو كبسولات فيتامبين هـ المعطى عن طريق القم أو المعاملة بالسيلينيوم + فيتامين هـ على مستوى السيلينيوم في سيرم الدم وكذلك على معدل النمو اليومي وبعض مكونات الدم (البروتين الكلى و الألبيومين و الجلوبيولين و نسبة الألبيومين إلى الجلوبيولين و الكوليمتيرول الكلى والليبيدات الكلية والنشاط الانزيمي GOT - GPT) في عجول وعجلات الجاموس المصدي

تم تتشئة ٢١ عجل جاموسى (١٥ ذكر ، ١٦ أنثى) بمحطة البحوث بمحله موسى بمحافظة كفر الشيخ. حيث قسمت الحيوانات عشوائيا عقب الفطام (عمر ١٥ أسبوع) الى أربعة مجاميع.

المجموعة الاولى: (٨ عجول وعجلات) حقنت في العضل بالسيلينيوم في صورة (Sodium)
 ابجرعة قدرها ١٢٥٠، ملليجرام /كجم من وزن الجسم.

٢. المجموعة الثانية : (٨ عجول و عجلات) تناولت كبسولات فيتامين هـ عن طريق الفح (٢٠٠ ٢. المجموعة الثانية : (٨ عجول و عجلات) تناولت كبسولات فيتامين هـ عن طريق الفح (٢٠٠-

٣. المجموعة الثالثة : (٨ عجول وعجلات) عوملت بالسيلينيوم + فيتامين هـ .

جرعات السيلينيوم وفيتامين هدتم تقسيمها الى جرعتين وتم استخدامها في خلال أسبوعين متتاليين وتم تكرار المعاملة كل ٣ شهور (حتى عمر ٤٥ أسبوع).

المجموعة الرابعة: (٧ عجول وعجلات) وكانت مجموعة المقارنة (بدون معاملة).

أدت المعاملة بالسيلينيوم + فيتامين هـ الى رفع تركيز عنصر السيلينيوم فى سيرم دم العجول والعجلات معنويا بعد المعاملة باسبوعين ، ١٤ ، ٢٠، ٣٠ أسبوع. تحسن معدل النمو اليومى للعجول

والعجلات في المجاميع المعاملة بالسيلينيوم ، فيتامين هـ ، سيلينيوم + فيتامين هـ بمقدار ٢٢ ، ٢٧ ، ٦٥ جرام/يوم على التوالى عن مجموعة المقارنة . لم نتأثر مكونات الدم بالمعاملات المختلفة ، بينما كان تأثير التداخل بين المعاملة والفترات الزمنية للتجربة تأثيرا معنويا على مكونات الدم التي درست فيما عدا الليبيدات الكلية.