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Changes in Body Weight Gain and Blood Hormonal Levels in Relation to Change in Age of Egyptian Male Buffaloes Calves from Birthing to Puberty

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Abstract: The objective of the present study was to determine the changes in blood hormonal levels and blood profile in relation to the change in each of age, live body weight and body weight gain of Egyptian male buffaloes calves from birthing to 24 months of age. The present study was carried out on 10 male buffalo calves from birthing until 24 months of age. The animals were weighed at birth, 3, 6, 12, 18 and 24 months before the morning feed and daily body weight gain (DBWG) was calculated for 0-3, 3-6, 6-12, 12-18, 18-24 and 0-24 months. During weighing the calves, 0ne blood sample from the Jugular vein of each calf was withdrawn to estimate thyroxin (T_4), triiodothyronine (T_3), cortisol, testosterone and aldosterone hormonal levels as well as glucose and protein fractions. Results showed that live body weight (LBW) of calves was increased gradually from birthing to reach to 592.8 kg after 24 months of age. The lowest DBWG of buffalo calves values were through the first 3 months after birthing and the period from 18-24 months of age while the highest DBWG values were during the period from 6 to 18 months of age. The highest level of T_4 and T_3 were at birth and decreased significantly at weaning. The lowest cortisol level was at birth while the highest cortisol value was at weaning and after 24 months of age. The highest level of aldoserone was at birth and at weaning. From 6 months of age up to 24 months, T_4, T_3 and aldosterone level was not affected significantly due to the change in age of calves. Testosterone level increased progressively with increase the age of buffalo calves. T_4, T_3 and aldosterone hormones have negative significant correlations while testosterone level has a highly positive correlation with each of age, LBW and DBG of buffalo calves. Glucose level decreased significantly with increasing the age of calves. Glucose and globulin levels have a negative significant correlation with each of age, LBW and DBG of buffalo calves. It can be concluded from this study that the changes in blood hormonal levels, especially, testosterone, T_4, T_3 and aldosterone as well as glucose and globulin concentrations are in relation to the change in each of age, live body weight and body weight gain of Egyptian male buffaloes calves from birth to 24 months of age.

Keywords: Buffalo, Daily Gain, Hormones, Glucose, Total Protein, Correlations

1. Introduction

In 2000, the United Nations Food and Agriculture Organization estimated that there were approximately 158 million water buffalo in the world and that 97% of them (approximately 153 million animals) were in Asia [1]. The water buffalo represents an important part of animal production in Egypt. The estimated herd number exceeds 3.6 million heads [2]. It is economically a very important farm animal and genetic improvement of these animals is of economic importance, especially, in reproductive performance and quantity of meat and milk as well as diseases and parasite resistance [3]. Buffaloes are the most important and popular livestock for milk production in Egypt. Buffalo produce 2,300,000 tons milk/year [4]. Milk composition analysis, per 100 grams showed that Buffalo milk has higher fat (8%), protein (4.5%), calcium (0.18-0.23%) and energy (110 kcal) and lower water (81.1%) and cholesterol (8 mg/dl) than cow, goat and sheep milk. Buffalo also has peculiar biological and economical properties e.g. white milk colour and high milk fat and solids not fat contents as compared to local and imported cattle in Egypt. Egyptian water buffalo is the main dairy animal in Egypt; in addition, it serves as an economically important source of meat. It produces about 66% and 43% of the national milk and meat production, respectively. In addition, Buffalo meat production is superior to bovine meat in quantity, amounting to 180,000 and 155,000 metric tons per year, respectively [4].

Growth, the increase in live body mass or cell multiplication, is controlled genetically and environmenally. In mammals, growth is the change in live weight during the different stages of life, as well as, elevated ambient temperatures are considered as some of the environmental factors that can influence average daily gain [5]. Growth is considered outcome of interactions among several factors, where thyroid hormones, cortisol, aldosterone and testosterone plays a key role in coordination of these factors and the information of these hormones in relation to age, growth and live body weight in buffalo calves is not adequate. Many factors i.e., gender, age and other physiological status have complex effect on hormonal levels in blood of animals [6].

Normal growth and development occur in animals only in the presence of thyroid hormone, indicating that thyroid hormones are necessary for normal growth and play a permissive role in growth regulation. Thyroid hormones directly influence growth by altering biochemical reactions; many of them influence size of specific tissues and organs. These hormones affect body mass and dimensions primarily by altering skeletal and or nitrogen metabolism. In addition, thyroid hormones play a permissive role in growth regulation and are essential for maintenance of the basal metabolic rate [7]. Cortisol plays a role in bone growth, immune system function, metabolism of fats, carbohydrates, and protein, nervous system function and stress response and aldosterone hormone regulate sodium and potassium balance [5]. Testosterone in males is a prerequisite for normal spermatogenesis, normal function of the reproductive tract, influences the size and function of epididymis with a consequence on maturation and survival of spermatozoa during epididymal transit and regulator the activities of testis [8]. Testosterone levels are useful in the selection of young sires and to characterize sexual maturity in different breeds and testosterone is directly involved in the onset of puberty and consequently in the onset of spermatogenesis and play an important role in penis detachment [8].

But, there is a little information about the changes in hormonal concentrations during different ages of Egyptian buffaloes. The objective of the present study was to determine the changes in blood hormonal levels and blood profile in relation to the change in each of age, live body weight and body weight gain of Egyptian male buffaloes calves from birthing to 24 months of age.

2. Materials and Methods

2.1. Animals and Nutritional Practice

The present study was carried out in the El-Khaer and El-Baraka, farm in El-Salhya desert area, El-Sharkia Governorate, Egypt. The study was carried out on 10 male buffalo calves from at birth until 24 months of age. During suckling period, calves depend on its mothering milk. After weaning, calves fed the concentrate feed mixture (CFM) and rice straw. Each calves was provided with CFM ration consisting of cracked yellow corn (40%), wheat bran (25%), undecorticated cotton seed meal (25%), solvent-extracted soybean meal (7%), Dicalcium phosphate (1.0%), iodized salt (1.0%) and trace mineral mixture (0.50%) and vitamin AD_3E (0.50%). The chemical composition (on dry matter basis %) of the concentrate feed mixture used in the feeding calves during the experimental period was carried according to [9]. The values of crude protein, crude fat, NDF and ADF are 15.2, 3.0, 20.5 and 13.5%, respectively. In addition, the concentrate feed mixture contains 0.8 % calcium, 0.6% phosphorus, 0.07% magnesium and 0.65% potassium as well as 13500, 4500 and 36 IU vitamins A, D_3 and E/kg of mixture, respectively. CFM ration offered twice daily at morning and evening night at the rate of 2.5 kg CFM /100 kg live body weight (LBW) while rice straw was offered *ad libitum*. Source of fresh drinking water was available automatically all time from underground source to the calves.

2.2. Experimental Design

The animals were weighed at birth, 3, 6, 12, 18 and 24 months to the nearest 1 kg LBW before the morning feed and daily body weight gain (DBWG) was calculated for 0-3, 3-6, 6-12, 12-18, 18-24 and 0-24 months. The calves from weaning to the end of experiment were left loose in doors day and night and raised under wood roofed shed in one yard. During weighing the calves, 0ne blood sample was withdrawn from the Jugular vein of each calf at birth, 3, 6, 12, 18 and 24 months before the morning feed between 10.00 and 12.00 a.m. by jugular vein puncture using disposable syringes. Blood (10 ml) ant-coagulated with disodium-EDTA was used and all tubes were placed immediately on ice-box and were transferred to the laboratory. Blood plasma was separated by centrifugation (2000x g for 30 min.) and stored at -20°C until the hormones and blood components determinations. Thyroxin (T_4), triiodothyronine (T_3), cortisol, testosterone and aldosterone hormones level were estimated by RIA using coated tubes kit; DSL, Inc. Webster, Texas, USA and counting using computerize Gamma Counter. The tracer in the hormones was labeled with iodine-125 (^{125}I). Total protein, albumin and glucose were measured by commercial kits and the concentration of globulin calculated as the difference between total protein and albumin.

2.3. Statistical Analysis

The obtained data were analyzed statistically using [10] procedures of personal computer. The least significant difference among means was carried out according to [11]

3. Results

3.1. Changes in Body Weight Gain in Relation to Change in Age of Male Buffaloes Calves

LBW of male buffalo calves, normally, increased gradually with increasing the age of animals. The lowest DBWG of buffalo calves values were through the first 3 months after birthing and the period from 18-24 months of age. The highest DBWG value was during the growing period and was nearly the same DBWG during the period from 6 to 12 and from 12 to 18 months of age (Table 1).

Table 1. *Live body weight (LBW) and daily body weight gain (DBWG) of male buffalo's calves from birthing to 24 months of age.*

Calves gain	At Birth	At weaning, 3 months	After 6 months	After 12 months	After 18 months	After 24 months
LBW, kg	31.5±0.4	91.1±1.6	157.5±2.2	316.8±3.7	472.8±3.3	592.8±4.6
Period, mo	0-3	3-6	6-12	12-18	18-24	0-24
DBWG, g	662.2^{c}±16.9	737.7^{b}±33.1	885.0^{a}±25.5	866.7^{a}±18.6	666.7^{c}±19.1	779.6^{b}±16.4

3.2. Changes in Blood Hormonal Levels in Relation to Change in Age of Male Buffaloes Calves

Thyroxin (T4):

The highest level of T_4 was at birth and decreased significantly at weaning. After 6 months of age, T_4 concentration was not affected significantly with advancing of buffalo age up to 24 months and the values remained almost similar with minor fluctuations up to 24 months (Table 2 and figure 1).

Table 2. *Hormonal levels in male buffaloes calves as affected by age.*

Hormones	At Birth	At weaning, 3 months	After 6 months	After12 months	After 18 months	After24 months
T_4(nmol/l)	120.2^{a}±0.4	95.9^{b}±3.9	79.1^{c}±1.1	75.0^{c}±1.5	73.6^{c}±2.7	75.3^{c}±2.5
T_3(nmol/l)	8.4^{a}±0.3	5.9^{b}±0.2	4.9^{c}±0.1	4.4^{c}±0.2	4.6^{c}±0.4	4.9^{c}±0.2
Cortisol (ng /dl)	7.3^{d}±0.4	23.2^{a}±1.2	12.1^{c}±0.7	12.3^{c}±0.6	16.4^{b}±0.4	22.6^{a}±0.5
Aldosterone (ng/ml)	7.6^{a}±0.2	7.3^{a}±0.3	4.6^{b}±0.2	4.7^{b}±0.2	4.2^{b}±0.2	4.1^{b}±0.1
Testosterone (ng/dl)	5.50^{f}±1.1	23.63^{e}±1.6	118.6^{d}±5.9	170.0^{c}±9.3	237.5^{b}±8.0	288.1^{a}±6.9

Triiodothyronine (T_3): The highest level of T_3 was found at birth (8.4nmo/l) and decreased to 5.9 nmol/l at weaning. After 6 months of age, T_3 level was not affected with advancing of buffalo age up to 24 months and the values were ranged between 4.4 to 4.9 nmol/l (Table 2 and Figure 2).

Cortisol: The lowest cortisol level was at birth while the highest cortisol value was at weaning and after 24 months of age. Cortisol level was not changed between 6 to 12 months of age but increased significantly after 18 months of age (Table 2).

Aldosterone: The highest level of aldosterone at birth and at weaning. From 6 months of age up to 24 months, aldosterone level was not affected significantly due to the change in age of calves (Table 2 and Figure 2)

Testosterone: Testosterone level increased progressively with increase the age of buffalo calves. The lowest level of testosterone was at birth while the highest level was after 24 months of age (Table 2 and Figure 1).

3.3. Correlations Coefficient Between Hormonal Levels and Age, LBW and DBG of Buffalo Calves

Results in Table (3) showed that level each of T_4, T_3 and aldosterone hormones have a negative significant correlation with age, LBW and DBG of buffalo calves. While level of testosterone has a highly positive correlation with each of age, LBW and DBG of calves. No significant correlation between cortisol level and age, LBW and DBG of buffalo calves.

Table 3. *Correlations coefficient between hormonal levels with age, LBW and DBG of buffalo calves.*

Items	Correlations coefficient				
	T_4	T_3	Cortisol	Aldosterone	Testosterone
Age	-0.769^{*}	-0.680^{*}	0.484	-0.832^{**}	0.983^{**}
LBW	-0.747^{*}	-0.654^{*}	0.478	-0.817^{**}	0.978^{**}
DBG	-0.945^{**}	-0.937^{**}	0.135	-0.910^{**}	0.702^{*}

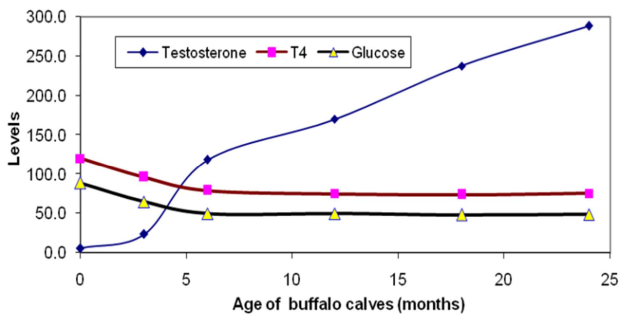

Figure 1. *Effect of age on testosterone (ng/dl), T_4 (nmol/l) and glucose (mg/dl) levels in buffalo calves.*

3.4. Changes in Concentrations of Blood Glucose and Protein Fractions in Relation to Change in Age of Egyptian Male Buffaloes Calves

Glucose level decreased significantly with increasing the age of calves. The highest glucose concentration was at birth while the lowest level was after 24 months of age (Table 4 and Figure 1). These results may be due to that born calves from birthing to weaning depend on glucose from lactose of milk of their mothers as a source for energy requirements. While after weaning, calves essentially depend on FFA from concentrates and roughages as a source for requirements from energy.

Table 4. *Glucose and protein fractions concentrations in male buffalo's calves as affected by age.*

Glucose and proteins	At Birth	At weaning,3 months	After 6 months	After 12 months	After 18 months	After24 months
Glucose(mg/dl)	$88.9^{a}\pm1.8$	$64.8^{b}\pm1.5$	$49.7^{c}\pm0.6$	$48.1^{c}\pm0.8$	$48.7^{c}\pm2.0$	$41.5^{d}\pm0.7$
Total protein (g/dl)	$8.3^{b}\pm0.1$	$8.3^{b}\pm0.1$	$8.9^{a}\pm0.2$	$8.9^{a}\pm0.2$	$8.8^{a}\pm0.2$	$8.2^{b}\pm0.1$
Albumin, A (g/dl)	$3.7^{c}\pm0.1$	$4.1^{b}\pm0.1$	$4.6^{a}\pm0.1$	$4.7^{a}\pm0.1$	$4.6^{a}\pm0.1$	$4.0^{b}\pm0.1$
Globulin, G (g/dl)	$4.6^{a}\pm0.1$	$4.2^{b}\pm0.1$	$4.3^{b}\pm0.1$	$4.2^{b}\pm0.1$	$4.2^{b}\pm0.1$	$4.2^{b}\pm0.1$

Table (4) showed that no significant differences in total protein, albumin and globulin concentrations during the period from 6 to 18 months of age. Total protein values were nearly the same at birth, at weaning and after 24 months of age. Albumin level increased from 6 to 18 months and then decreased significantly at 24 months to reach the albumin level at weaning. The highest globulin level was at birth and globulin level from weaning to 24 months of age was not changed (Table 4 and Figure 2).

3.5. Correlations Coefficient Between Glucose, Total Protein, Albumin and Globulin Levels with Age, LBW and DBG of Buffalo Calves

Results in Table (5) showed that glucose and globulin levels have negative significant correlations and DBG of buffalo calves with each of age, LBW and DBG of buffalo calves. However, no significant correlation between total protein and albumin levels and each of age, LBW and DBG of buffalo calves.

Table 5. *Correlations coefficient between glucose, total protein, albumin and globulin levels with age, LBW and DBG of buffalo calves.*

Item	Correlations coefficient			
	Glucose	Total protein	Albumin	Globulin
Age	-0.801**	0.039	0.279	-0.628*
LBW	-0.779**	0.016	0.252	-0.606*
DBG	-0.779**	0.016	0.252	-0.606*

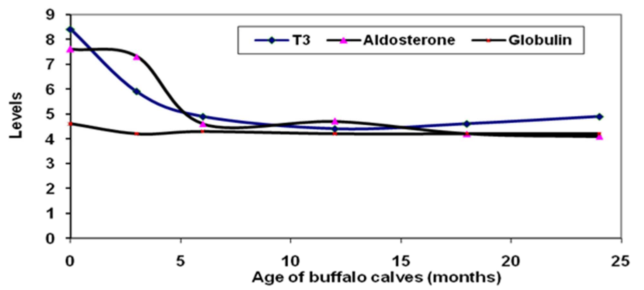

Figure 2. *Effect of age on T_3 (nmol/l), aldosterone (ng/ml) and globulin (g/dl) in buffalo calves.*

4. Discussion

Average DBG of female buffalo calves were varied significantly during the experimental period and the lowest values were at 6 and 12 months old while the highest values were at 8 and 10 months [12].

Age had significant effect on plasma T4 and free T4 concentrations in the Sarabi calves, with values being lower in 1-2 months-old calves and the highest levels of thyroid hormone in Iranian buffaloes Sarabi calves were seen during the first two weeks after birth [13]. T_4 concentration decreased with advancing age in buffalo calves and the highest T_4 concentration was recorded in 0-7 days old buffalo calves and the T_4 concentration decreased significantly to a lowest value at 2-4 months of age and T_4 values remained almost similar up to 24 -30 months without significantly difference throughout growing period until puberty and the highest concentration of T_3 was at 0-7 days and then decreasing with advancing age in buffalo calves and the concentration of T_3 decreased at 8-15 days and further declined and reached to a lowest value at 15-18 months of age [14].

The higher concentration of T_3 in new born calves could be one of the adaptive mechanisms to overcome the stressful period after birth and subsequent declining trend could be attributed to the negative feedback mechanism exerted by already higher concentrations of T_3 in blood. In addition, this increase in T_3 secretion may also be due to higher thyroid stimulating hormone concentration or decreased T_3 metabolic clearance due to low capability of T_3 degrading enzymatic system in new born calves [14]. The high levels of T_3 and T_4 at birth is necessary for the calf to get adapted to the external environment and to decrease the stress and the significantly higher level of T_4 in early life is required for the adjustment to the external environment [14].

Three peaks are in cortisol levels in buffalo calves, the first immediately after birth, the second at puberty and the third at sexual maturity [15]. The difference in the level of cortisol due to the change in body weight may be the bulls that reach puberty and maturity become more stressed and accordingly their cortisol level in the blood reaches the peak value as a result to the increase in the sexual neurogenic stress [5].

Body weight and plasma testosterone levels in Italian Mediterranean buffalo bulls were low between 5 and 21 months and a significant rise in plasma concentration of testosterone was observed at 25 months reaching peak levels at 38 months [16]. Serum level of testosterone in Holstein bulls was low in young animal and then increased with advancing age, since serum level of testosterone increased from 200 to 580 ng/dl with advancing age from 12 to 48 months [17].

The highest T_3 level in male buffaloes was observed in the animals aged 2-4 mo and the lowest T_3 concentration was in the bulls aged 8-18 mo and 42-48 mo. although the fluctuations in the T_3 levels in relation to change in body weight and age of bulls, it was found significantly negative correlation between T_3 and LBW(r = -0.400). At the same time, the regression coefficient of body weight (w) on T3 quadratic no linear as follows: T_3 = 1.38 - 0.003 W + 0.000005 W2 and concluded that each 100 kg increase in LBW resulted in 3% decrease in T_4 level [18]. The heat-induced change in T_3 hormone in male buffaloes was significantly correlated with DBWG in animals (DBWG = 997.8 – 12.5 x ng/dl decrease in T_3 [r = - 0.881, P< 0.003) and the negative correlation between T3 and body weight may be attributed to the increase in tissues utilization of T_3 (i.e. its concentration in blood decreases) as growth rate increased [19]. When the bulls reach puberty and become mature, T_3 utilization by the tissues may decline, i.e. its concentration in blood increases and as bulls grew older (i.e. with body weights more 500 kg) their capacity to produce T_3 may decrease or suffer from increased rate of T_3 destruction [18].

Concentration of glucose markedly decreased at day 14 and then remained relatively stable up to day 84 of age and the higher glucose concentration at first hours of calf life may be related to increased levels of corticosteroids during parturition and/or colostrum intake [20].

Albumin concentrations partially reflect hepatic synthesis and its increasing could be related to compensation of decreasing serum osmotic pressure due to globulin levels decline. The amount and time of colostrum intake has direct effect on the amount of serum total protein and globulin in neonatal calves. Decreasing of serum total protein and globulin amounts after birthing has been attributed to degradation of absorbed immunoglobulin in colostrums [15]. Age related changes had significant effect on total proteins, albumin and globulin levels and approximately similar changes were seen for serum total protein and globulin levels in male and female Holstein calves [20].

No significant positive correlation between T_4 and LBW in male buffalo calves and non significant negative correlation of serum T_4 or T_3 with age LBW of buffalo calves and heifers [14].

Testosterone concentration is related to age [21]. Testosterone plays an important role in sex drive, energy, and behavior, so a significant change in testosterone levels may be alarming with change of age [22]. There was a positive correlation between testosterone concentration and body weight [23] and it is important to emphasize the close relationship between testosterone concentrations with body weight [24]. Testosterone concentration was significantly increased as a function of age or body weight increases and significant positive correlation between body weight and testosterone hormone (r =0.0 974) and the quadratic equation of body weight (w) categories on testosterone hormone was as follows: Testosterone - 0.12 + 0.05 W - 0.00003 W and concluded that each 100 kg increase in LBW resulted in 5.0% increase in testosterone level [18].

The testes in youngest animals are stimulated by chorionic gonadotropin from the placenta to produce a small quantity of testosterone, and then testosterone production increases rapidly to reach the peak at the onset of puberty and lasts throughout most of the remainder of life. When the bulls reach senility, the testosterone production dwindles rapidly beyond. The increase in the animals live body weights as a result of increases age was accompanied by an increase in the level of testosterone in the serum of the male buffaloes [18], [24].

5. Conclusion

During the period from 6 to 18 months of age, no significant differences in blood hormones and blood components except testosterone and glucose levels, being testosterone level increased progressively while glucose level decreased significantly with increasing age of calves. The changes in testosterone, T_4, T_3 and aldosterone as well as glucose and globulin concentrations are in relation to the change in each of age, live body weight and body weight gain of Egyptian male buffaloes calves from birth to 24 months of age.

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