

## Research Article

## Biochemical Components and Hormonal Levels in Blood of Male Buffaloes Calves as Response to Increase of Age from Birthing to Puberty

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**Abstract: Background and Objective:** 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. The objective of the study was to determine the changes in blood biochemical components and hormonal levels in relation to the change in each of age, live body weight and body weight gain of Egyptian male buffalo's calves from birthing to puberty age. **Materials and Methods:** The present study was carried out on 10 male buffalo calves from at birthing until 24 months of age. The animals were weighed at birth, 3, 6, 12, 18 and 24 months and daily body weight gain (DBWG) was calculated. During weighing the calves, One blood sample from the Jugular vein of each calf was withdrawn with anticoagulant and plasma was separated to estimate thyroxin (T<sub>4</sub>), triiodothyronine (T<sub>3</sub>), cortisol, testosterone and aldosterone hormonal level as well as glucose and protein fractions. **Results and Discussion:** The highest level of T<sub>4</sub> was at birthing (120.2 nmol/l) and decreased to 95.9 nmol/l at weaning. The highest level of T<sub>3</sub> was found at birthing (8.4nmol/l) and decreased to 5.9 nmol/l at weaning age. The lowest cortisol level was at birthing (7.3 ng/dl) while the highest cortisol value was at weaning (23.2 ng/dl) and after 24 months of age (22.6 ng/dl). The highest level of aldosterone was 7.6 ng/ml at birth and 7.3 ng/ml at weaning. Testosterone level increased progressively with increase the age of buffalo calves. T<sub>4</sub>, T<sub>3</sub> and aldosterone hormones have negative significant correlations with each of age, LBW and DBG of buffalo calves. While testosterone level has a highly positive correlation with each of age, LBW and DBG of buffalo calve. Glucose and globulin levels have a negative significant correlation with each of age, LBW and DBG of buffalo calves. **Conclusion:** The changes in blood hormonal levels, especially, testosterone, T<sub>4</sub>, T<sub>3</sub> 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 birthing to puberty age.

**Keywords:** Buffalo, daily gain, hormones, glucose, total proteins, correlations.

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## 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 (FAO, 2000). The water buffalo represents an important part of animal production in Egypt. The estimated herd number exceeds 3.6 million heads (FAO, 2002). 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 (El-Nahas *et al.*, 1998). Buffaloes are the most important and popular livestock

for milk production in Egypt. Buffalo produce 2,300,000 tons milk/year (FAO, 2008).

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 (FAO, 1996). In addition, Buffalo meat

production is superior to bovine meat in quantity, amounting to 180,000 and 155,000 metric tons per year, respectively.

Growth, the increase in live body mass or cell multiplication, is controlled genetically and environmentally. 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 (Habeeb *et al.*, 1992). 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 their levels in blood of animals. But, there is a little information about the changes of plasma hormones concentrations during different ages of Egyptian buffaloes (Garg *et al.*, 2002 and Habeb *et al.*, 2001).

In animals, normal growth and development occur 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 (Ingole *et al.*, 2012). In this study, variations in the plasma T4 and T3 during different ages in male calves were investigated.

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 (Habeeb *et al.* 1992). In this study, variations in the both two hormones during different ages in male buffalo calves were also investigated.

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 (Al-Qarawi *et al.*, 2000 and Al-Qarawi *et al.*, 2004). Testosterone in males is a prerequisite for normal spermatogenesis and normal function of the reproductive tract (Luke and Coffey, 1994). Testosterone also influences the size and function of epididymis with a consequence on maturation and survival of spermatozoa during epididymal transit and testosterone also is a regulator of activities in the testis (Goeritz *et al.*, 2003). In addition, testosterone hormone influences the synthesis of a number of caput and cauda epididymal proteins and some of these proteins could be important for improving spermatozoa maturation,

storage and their acquisition of fertilizing ability (De Pauw *et al.*, 2003).

The objective of the present study was to determine the changes in blood hormonal levels and blood profile included glucose, total protein, albumin and globulin 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.

## MATERIALS AND METHODS

### Location and ethics:

The experimental work was carried out in the El-Khaer and El-Baraka, farm in El-Salhya desert area, El-Sharkia Governorate, Egypt during the year of 2017. Hormonal levels and blood biochemical components were carried out in Biological Application Department, Radioisotopes Applications Division, Nuclear Research Centre, Atomic Energy Authority, at Inshas, Cairo, Egypt (latitude 31° 12' N to 22° 2' N, longitude 25° 53' E to 35° 53' E).

Experimental animals were cared using husbandry guidelines derived from Egyptian Atomic Energy Authority standard operating procedures. This work was reviewed and approved by the Animal Care and Welfare Committee of Egyptian Atomic Energy Authority. These ethics contain relevant information on the Endeavour to reduce animal suffering and adherence to best practices in veterinary care according to the International Council for Laboratory Animal Science guidelines.

### Animals and nutritional practice:

The present 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%), un-decorticated cotton seed meal (25%), solvent-extracted soybean meal (7%), Di-calcium phosphate(1.0%), iodized salt (1.0%) and trace mineral mixture (0.50%) and vitamin AD<sub>3</sub>E (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 AOAC (1990).

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 and E/kg of mixture, respectively. The nutritive values of the feed stuffs were calculated and assuming that the net energy for starch was 80 MJ/kg DM are 4.00 MJ Net energy/kg DM as follow: 60.82 Total digestible nutrients (TDN%); 115 Digestible crude protein (DCP g/kg DM) and 0.50 as Starch equivalent. 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.

**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, One 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 12.00AM by jugular venipuncture 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. All determinations were carried out in the tracer bioclimatology unit, Department of Biological Applications, Nuclear research center, Atomic Energy Authority, Inshas, Cairo, Egypt. Thyroxin (T<sub>4</sub>), triiodothyronine (T<sub>3</sub>), 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 (<sup>125</sup>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.

**Statistical analysis:**

The obtained data were analyzed statistically using SAS (1996) procedures of personal computer. The least significant difference among means was carried out according to Duncan (1955).

**RESULTS AND DISCUSSION**

**Changes in body weight gain in relation to change in age of Egyptian male buffaloes calves:**

Live body weight (LBW) of male buffalo calves, normally, increased gradually with increasing the age of animals. LBW of calves was 31.5 kg at birth and increased gradually to reach to 625.9 kg after 24 months of age (Table 1). Concerning boy weight gain, the results showed that the lowest daily body weight gain (DBWG) of buffalo calves was through the first period of age. DBWG values were 661.8 and 738.3 g during the period from birthing to weaning (0-3 months) and after weaning (3-6 months), respectively. The highest DBWG value was during the fattening period and was nearly the same DBWG during the period from 6 to 24 month (Table 1).

**Table 1:** Live body weight and daily body weight gain of male buffalo's calves from birthing to 24 months of age (X±SE)

Item	At Birth	At weaning, 3 months	After 6 months	After 12 months	After 18 months	After 24 months
<b>Live body weight, kg</b>	31.5±0.4	91.1±1.6	157.5±2.2	316.8±3.7	472.8±3.3	625.9±4.6
<b>Daily body gain, g</b>	0-3 Mon. 661.8 <sup>d</sup> ±16.9	3-6.Mon. 738.3 <sup>c</sup> ±33.1	6-12 Mon. 884.6 <sup>a</sup> ±25.5	12-18 Mon. 869.6 <sup>a</sup> ±18.6	18-24 Mon. 850.3 <sup>ab</sup> ±19.1	0-24Mon. 825.6 <sup>b</sup> ±6.4

a, b ...Means in the same row having different superscripts per each item differ significantly (P<0.05), Mon. = month.

**Changes in blood hormonal levels in relation to change in age of Egyptian male buffaloes calves:**

**Thyroid hormones:**

**Thyroxin (T<sub>4</sub>):**

Results in Table (2) showed that the highest level of T<sub>4</sub> was at birthing (120.2 nmol/l) and decreased to 95.9 nmol/l at weaning (3 months of age). After 6 months of age, T<sub>4</sub> concentration was not affected significantly with advancing of buffalo age up to 24 months and the values were ranged between 73.6 to 79.1 nmol/l. T<sub>4</sub> values remained almost similar with minor fluctuations up to 24 months and did not differ significantly throughout growing and fattening periods until puberty (24 months). The higher concentration of T<sub>4</sub> at birth in the present study is in agreement with the earlier reported findings in buffalo calves (Sharma *et al.*, 1985 and Sharma, 1996). Thyroxin concentrations

of buffalo calves were 5.5, 5.3 and 5.6 µg/dl at 7, 8 and 9 months of age, respectively (Yousef, 1992). In Iranian buffaloes Sarabi calves, Eshratkhah *et al.* (2010) found that the highest levels of thyroid hormone were seen during the first two weeks after birth and concluded that age and season changes have the highest effect on the plasma thyroid hormones concentrations in Sarabi calves. The same authors reported that the highest concentrations of thyroid hormones and their percentages were observed during the early two weeks of life in the Iranian Sarabi calves and concluded that age had significant effect on plasma T<sub>4</sub> and free T<sub>4</sub> concentrations in the Sarabi calves, with values being lower in 1-2 months-old calves. Ingole *et al.* (2012) reported also that T4 concentration decreased with advancing age in buffalo calves. The authors found that the highest T4 concentration was recorded in 0-7 days old buffalo calves and the T<sub>4</sub> concentration decreased significantly to a lowest value at 2-4 months of age and

T4 values remained almost similar up to 24-30 months period until puberty. without significantly difference throughout growing

**Table 2-**Thyroid hormonal levels in male buffalo’s calves as affected by age

Thyroid hormones	At Birth	At weaning, 3 months	After 6 months	After 12 months	After 18 months	After 24 months
<b>T<sub>4</sub></b> (nmol/l)	120.2 <sup>a±</sup> 0.4	95.9 <sup>b±</sup> 3.9	79.1 <sup>c±</sup> 1.1	75.0 <sup>c±</sup> 1.5	73.6 <sup>c±</sup> 2.7	75.3 <sup>c±</sup> 2.5
<b>T<sub>3</sub></b> (nmol/l)	8.4 <sup>a±</sup> 0.3	5.9 <sup>b±</sup> 0.2	4.9 <sup>c±</sup> 0.1	4.4 <sup>c±</sup> 0.2	4.6 <sup>c±</sup> 0.4	4.9 <sup>c±</sup> 0.2

a, b ...Means in the same row having different superscripts per each item differ significantly (P<0.05).

The high levels of T<sub>3</sub> and T<sub>4</sub> at birth is necessary for the calf to get adapted to the external environment and to decrease the stress (El-Masry and Habeeb, 1989). The significantly higher level of T<sub>4</sub> in early life is required for the adjustment to the external environment. T<sub>4</sub> is a calorogenic hormone and the thyroid activity is enhanced for maintaining constant body temperature by increasing the metabolic rate when exposed to cold environment. Therefore the level of T<sub>4</sub> is high during first week of life (Ingole *et al.*, 2012). In addition, thyroid stimulating hormone (TSH) concentration did not differ and remained almost the same from birth to 4-6 months and then gradually increased and reached a highest value at 24-30 months (Ingole *et al.*, 2012).

**Triiodothyronine (T<sub>3</sub>):**

The highest level of T<sub>3</sub> was found at birth (8.4nmol/l) and decreased to 5.9 nmol/l at weaning age. After 6 months of age, T<sub>3</sub> level was not affected with advancing of buffalo age up to 24 months and the values were ranged from 4.4 to 4.9 nmol/l (Table 2). Similar results obtained by Ingole *et al.* (2012) who found that the highest concentration of T<sub>3</sub> was (1.47ng/ml) at 0-7 days and then decreasing with advancing age in buffalo calves. The authors reported that the concentration of T<sub>3</sub> decreased to 1.32 ng/ml at 8-15 days and further declined and reached to a lowest value of 0.39 ng/ml at 15-18 months of age. Hugi and Blum (1997) studied the changes of blood metabolites

and hormones in female breeding calves before, during and after weaning from 4 to 18 weeks of age. The authors found that postprandial concentrations of T<sub>3</sub> hormone gradually decreased significantly with age. The higher concentration of T<sub>3</sub> 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<sub>3</sub> in blood. In addition, this increase in T<sub>3</sub> secretion may also be due to higher thyroid stimulating hormone concentration or decreased T<sub>3</sub> metabolic clearance due to low capability of T<sub>3</sub> degrading enzymatic system in new born calves (Ingole *et al.*, 2012).

**Cortisol:**

The lowest cortisol level was at birth (7.3 ng/dl) while the highest cortisol value was at weaning (23.2 ng/dl) and after 24 months of age (22.6 ng/dl). However, cortisol level was not changed between 6 (12.1 ng/dl) to 12 months (12.3 ng/dl) of age but increased significantly to 16.4 ng/dl after 18 months of age (Table 3). Alvarez and Johnson (1973) found those three peaks in cortisol levels in buffalo calves, the first immediately after birth, the second at puberty and the third at sexual maturity. The change in the concentrations of cortisol was studied in 14 calves between birth and 83 days of age by Knowles *et al.* (2000) and found similar result.

**Table 3** Corisol, aldosterone and testosterone hormonal levels in male buffalo’s calves as affected by age

Hormones	At Birth	At weaning, 3 months	After 6 months	After 12 months	After 18 months	After 24 months
<b>Cortisol</b> ( ng /dl)	7.3 <sup>d±</sup> 0.4	23.2 <sup>a±</sup> 1.2	12.1 <sup>c±</sup> 0.7	12.3 <sup>c±</sup> 0.6	16.4 <sup>b±</sup> 0.4	22.6 <sup>a±</sup> 0.5
<b>Aldosterone</b> (ng/ml)	7.6 <sup>a±</sup> 0.2	7.3 <sup>a±</sup> 0.3	4.6 <sup>b±</sup> 0.2	4.7 <sup>b±</sup> 0.2	4.2 <sup>b±</sup> 0.2	4.1 <sup>b±</sup> 0.1
<b>Testosterone</b> (ng/dl)	5.50 <sup>f±</sup> 1.1	23.63 <sup>e±</sup> 1.6	118.6 <sup>d±</sup> 5.9	170.0 <sup>c±</sup> 9.3	237.5 <sup>b±</sup> 8.0	288.1 <sup>a±</sup> 6.9

a, b ...Means in the same row having different superscripts per each item differ significantly (P<0.05)

**Aldosterone:**

Concerning with aldosterone changes with change in age of buffalo calves, the results showed that the highest level of aldosterone was 7.6 ng/ml at birth and 7.3 ng/ml at weaning. However, from 6 months of age up to 24 months, aldosterone level was not affected

significantly due to the change in age of calves and ranged between 4.1 to 4.7 ng/ml (Table 3).

**Testosterone:**

Testosterone level increased progressively with increase the age of buffalo calves. The lowest level of testosterone was at birth (5.5 ng/dl) while the highest

level was after 24 months of age (288.1 ng/dl) (Table 3). Similar results obtained by Malfatti *et al.* (2006) in Italian Mediterranean buffalo bulls managed in two different breeding conditions. Body weight and plasma testosterone concentrations were measured in 35 Nili-Ravi water buffalo bulls at 5, 15, 17, 21, 25 and 38 months of age (Manzoor *et al.*, 1984). The authors found that body weight and plasma testosterone levels 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. Matsuzaki *et al.* (2000) found that 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 of calves age from 12 to 48 months. In male dromedary camel, El-Harairy and Attia (2010) found that testosterone concentration in

blood plasma showed marked increase with advancing age to be the highest ( $2.13 \pm 0.74$  ng/ml) at 6 years of age during non-breeding season and concluded that testosterone concentrations were closely related with seasonal changes, stage of puberty and advancing age of humped male camels.

**Correlations coefficient between hormonal levels and age, LBW and DBG of buffalo calves:**

Results in Table (4) showed that level each of T<sub>4</sub>, T<sub>3</sub> 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 buffalo calves. However, no significant correlation between cortisol level and age, LBW and DBG of buffalo calves.

**Table 4** Correlations coefficient between hormonal levels with age, LBW and DBG of buffalo calves

Items	Correlations coefficient				
	T4	T3	Cortisol	Aldosterone	Testosterone
Age	-0.769*	-0.680*	0.484 <sup>NS</sup>	-0.832**	0.983**
LBW	-0.747*	-0.654*	0.478 <sup>NS</sup>	-0.817**	0.978**
DBG	-0.945**	-0.937**	0.135 <sup>NS</sup>	-0.910**	0.702*

\*\*=significant at P<0.01, \*=significant at P<0.05 and NS=not significant.

Similar results obtained by Khalil *et al.* (1996), Habeeb *et al.* (1997) and Habeeb *et al.* (2001) in Ossimi and crossbred (Ossimi x Rahmani) lambs, male buffalo calves and Friesian calves, respectively. Habeeb *et al.* (1997) found that the heat-induced change in T<sub>3</sub> hormone in male buffaloes was significantly correlated with DBWG in animals (DBWG = 997.8 – 12.5 x ng / dl decrease in T<sub>3</sub> [r = - 0.881, P< 0.003) and concluded that the heat induced change in T<sub>3</sub> hormone can be used as heat adaptability index for predicting the growth rate of animals under heat stress conditions. Sharma *et al.* (1985) reported that non- significant positive correlation between T<sub>4</sub> and LBW in male buffalo calves. In addition, non-significant negative correlation of serum T<sub>4</sub> with age in buffalo calves was reported by Sharma *et al.* (1985) and Ingole *et al.* (2012). Non- significant positive or negative correlation of serum T<sub>3</sub> with age in male buffalo calves was reported by Sharma *et al.* (1985) and Ingole *et al.* (2012). With regards to the relationship of thyroid hormones with body weight, Ingole *et al.* (2012) reported that T<sub>3</sub> was no significant positively correlated with LBW of buffalo calves and heifers while serum T<sub>4</sub> was no significant negatively correlated with LBW. Similar non-significant positively correlation was reported in relationship between T<sub>3</sub> and LBW in Carora heifers (Leyva-Ocariz, 1997). Dvorak *et al.* (1986) observed inverse relationship of thyroxin with live DBG in lactating cows. Habeeb *et al.* (1997) found that the heat-induced change in cortisol hormone in male buffaloes was significantly correlated with DBWG in animals (DBWG = 978.5 – 88.3 x ng/ ml

increase in cortisol [r = 0.7945, P < 0.01 ] and concluded that the heat induced change in cortisol hormone can be used as heat adaptability index for predicting the growth rate of animals under heat stress conditions.

**Changes in levels of blood glucose and protein fractions in relation to change in age of male buffaloes calves:**

Glucose level decreased significantly with increasing the age of calves. The highest glucose concentration was at birth (88.9 mg/dl) while the lowest level was after 24 months of age (41.5 mg/dl) (Table 5). 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. Concerning protein fractions, Table (5) 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 (8.3 g/dl), at weaning (8.3 g/dl) and after 24 months of age (8.2 g/dl). Albumin level was 3.7 g/dl at birth and increased gradually with increasing the age of calves at 18 months. However, after 24 months of age, albumin decreased significantly to reach the albumin level at weaning. The highest globulin level was at birth (4.6 g/dl) and globulin level from weaning to 24 months of age not changed as shown in Table (5).

**Table 5** 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	After 24 months
Glucose (mg/dl)	88.9 <sup>a±</sup> 1.8	64.8 <sup>b±</sup> 1.5	49.7 <sup>c±</sup> 0.6	48.1 <sup>c±</sup> 0.8	48.7 <sup>c±</sup> 2.0	41.5 <sup>d±</sup> 0.7
T. protein (g/dl)	8.3 <sup>b±</sup> 0.1	8.3 <sup>b±</sup> 0.1	8.9 <sup>a±</sup> 0.2	8.9 <sup>a±</sup> 0.2	8.8 <sup>a±</sup> 0.2	8.2 <sup>b±</sup> 0.1
Albumin, A (g/dl)	3.7 <sup>c±</sup> 0.1	4.1 <sup>b±</sup> 0.1	4.6 <sup>a±</sup> 0.1	4.7 <sup>a±</sup> 0.1	4.6 <sup>a±</sup> 0.1	4.0 <sup>b±</sup> 0.1
Globulin, G (g/dl)	4.6 <sup>a±</sup> 0.1	4.2 <sup>b±</sup> 0.1	4.3 <sup>b±</sup> 0.1	4.2 <sup>b±</sup> 0.1	4.2 <sup>b±</sup> 0.1	4.2 <sup>b±</sup> 0.1

a, b ...Means in the same row having different superscripts per each item differ significantly (P<0.05).

The change in the concentration of glucose was studied in 14 calves between birth and 83 days of age by Knowles *et al.* (2000) and reported similar result. Mohri *et al.* (2007) found that concentration of glucose markedly decreased at day 14 and then remained relatively stable up to day 84 of age. The higher glucose concentration at first hours of calf life may be related to increased levels of corticosteroids during parturition and/or colostrum intake. The changes in the concentrations of total protein and albumin were studied in 14 calves between birth and 83 days of age by Knowles *et al.* (2000) and reported similar result. 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 colostrum.

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

Results in Table (6) showed that glucose and globulin levels have negative significant correlations 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. Early, age of female dairy cattle had a significant negative relationship with glucose level (Tumbleson *et al.* 1971) and a significant positive relationship with serum protein concentration (Tumbleson *et al.* 1973). Age of animal was related linearly to glucose, total protein and globulin in four breeds of dairy cattle (Holstein, Guernsey, Jersey and Brown Swiss) (Shaffer *et al.*, 1981). The authors found that total protein and globulin increased with increasing age while glucose decreased with increasing age. The authors found also that the change per day of age had significant negative relationship (regression coefficient) with glucose and significant positive relationship with total protein as well as globulin.

**Table 6:** 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 <sup>**</sup>	0.039 <sup>NS</sup>	0.279 <sup>NS</sup>	-0.628 <sup>*</sup>
LBW	-0.779 <sup>**</sup>	0.016 <sup>NS</sup>	0.252 <sup>NS</sup>	-0.606 <sup>*</sup>
DBG	-0.779 <sup>**</sup>	0.016 <sup>NS</sup>	0.252 <sup>NS</sup>	-0.606 <sup>*</sup>

\*\*=significant at P<0.01, \*=significant at P<0.05 and NS=not significant.

There were significant age related changes for most hematological and biochemical parameters in blood sample which had taken from 18 male and 14 female Holstein calves within 24–48 h following birth and at 14, 28, 42, 56, 70 and 84 days of age (Mohri *et al.*, 2007). The same authors reported that 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.

Hugi and Blum (1997) studied the changes of blood metabolites and hormones in female breeding calves before, during and after weaning from 4 to 18 weeks of age. The authors found that postprandial concentrations of protein and albumin gradually increased significantly with age. However, the

difference between these reports may be attributed to formulation of animal diets, time of weaning and environmental conditions and many blood hormonal values vary with the age of the animal, with major changes occurring before puberty, especially, testosterone hormone.

**CONCLUSION**

Testosterone level increased progressively while glucose level decreased significantly with increasing age of calves. During the period from 6 to 18 months of age, no significant differences in blood hormones and blood components studied except testosterone and glucose levels. It can be concluded from this study that the changes in blood hormonal

levels, especially, testosterone, T<sub>4</sub>, T<sub>3</sub> 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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