ASSESSMENT OF LIVE BODY WEIGHT, BODY FLUIDS AND CONCENTRATIONS OF SODIUM, POTASSIUM AND ALDOSTERONE IN DRY AND PREGNANT EWES DURING SUMMER AND WINTER

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SUMMARY

The aim of this study was to assess live body weight (LBW), percentages of total body fluids (TBF), extracellular fluids (ECF), intracellular fluids (ICF), plasma volume (PV), and concentrations of aldosterone (ALD), sodium (Na) and potassium (K) in dry, early and late pregnant ewes in summer (July-August) and winter (February-March). A total of adult healthy 48 ewes with different physiological statuses (8 dry ewes, 8 early pregnant ewes at 22-35 d of pregnancy, and late pregnant ewes at 144-145 d of pregnancy) were chosen by random way and housed in semi-open pens under the normal environmental conditions, Mallawi Sheep Farm ElMinia Governorate, Upper Egypt. TBW was determined by injecting j.v. 0.5 ml Urea solution/kg LBW, ECF was determined by injecting j.v. 0.25 ml sodium Thiosianate solution (5%) /kg LBW, PV was determined by injecting j.v. Evan's blue solution (0.5%) 0.1 ml/kg LBW, and ICF was calculated by subtract TBW from ECF. Before feed intake, blood samples were collected (6:00 a.m.) from all ewes via j.v. puncture for plasma aldosterone, Na and K determination. Results of this study indicate that LBW and Na concentration of dry and pregnant ewes were greater (P<0.05) in summer than winter. K concentration in dry and early pregnancy was greater (P<0.05) in winter than summer, but it was greater (P<0.05) in late pregnancy in summer than winter. K concentration had a negative correlation with TBF, ICF and PV. Ald. concentrations and PV % in dry and pregnant ewes were greater in winter than summer. TBF and ECF percentages
in dry ewes were similar in summer and winter, while they were ($P<0.05$) greater in early and late pregnancy in summer than winter. ICF % in the dry ewes was ($P<0.05$) greater in winter than summer, while the ICF % in early and late pregnancy was similar of both summer and winter. In conclusion, LBW of dry and pregnant ewes increased with increasing TBF % and Na concentration in summer, Ald. concentrations of dry and pregnant ewes increased with increasing Na and K concentrations and PV % in winter. K concentration had negative correlations with TBF, ICF and PV compartments. These results can be used to maintain body fluids and plasma volume responsible for the optimal cellular functions in sheep, particularly in summer.

Key words: Body weight, body fluids, Aldosterone, sodium, potassium, season and dry and pregnant ewes.

INTRODUCTION

Changes in water volume alone have diluting or concentrating effects on body fluids. This means that the dehydrated animal will lose relatively more water than sodium, hence the osmolarity of body fluids increases and the body must conserve water than sodium. Otherwise, maintenance of plasma volume is essential for preservation of normal cell volume and function where shrinkage or swelling of cells can occur when the cells swim in a low or high serum osmolality. Louden, (2009) reported that regulation of plasma osmolality, which is determined primarily by the serum sodium concentration occurs through changes in water balance, whereas volume regulation is principally determined by changes in sodium excretion. Kidneys have a regulated mechanism for reabsorbing sodium in the distal tubules of nephron, this mechanism is controlled by aldosterone secreted by the adrenal cortex. Luetscher and Axelrad (1954) reported that aldosterone excretion in the urine of normal individuals changed
considerably with the amount of sodium intake. Many investigators (Kumar et al. 1959, Venning et al. 1959, Watanabe et al. 1963) reported that aldosterone secretion increased consistently in the third trimester of normal pregnancy and became more responsiveness to changes in dietary sodium. Jones et al. (1959) postulated the increased urinary excretion of aldosterone in mid-pregnancy was attributed to an alteration in the metabolism of the hormone from that in normal non-pregnants. Davis et al. (1958) found that the stimulation of aldosterone by the loss of fluid and electrolytes from the vascular tree in experimental ascite could not be offset by expansion of the intravascular volume. This might also explain the fact that increased secretion of aldosterone occurs in pregnancy despite increased plasma volume (Tysoe and Lowenstein 1950) and extracellular volume (Chesley 1943). Increasing plasma potassium (k) concentration is a potent stimulus to aldosterone secretion in the non-pregnant sheep (Blair-West et al. 1962; Funder et al. 1969. Furthermore, Na and K intake greatly influences plasma aldosterone concentrations in sheep as evidenced by findings of Blair-West et al. (1963). Chesley, (1944), Venning et al. (1959) indicated that sodium is retained during pregnancy and that this retained sodium is needed to meet the requirements of the fetus variously estimated to be from 7-50m Eq a day. It is obvious of the information mentioned above that water body fluids, aldosterone, sodium and potassium have been studied in some detail in pregnant animals (Hix et al. 1959; Baker et al. 1961; Vincent et al. 1986; Omer and Bayoumi 1980), but the effects of season on these parameters in dry and early and late pregnant ewes have not taken much investigations. Therefore, the aim of
this study was to assess body fluids, live body weight and concentrations of aldosterone, sodium and potassium in dry and early and late pregnant ewes during summer (July-August) and winter (February-March).

**MATERIALS AND METHODS**

**Animals**
A total of 48 adult ewes were housed in semi-open pens under the normal environmental conditions, Animal Production Research Institute, Mallawi sheep barn, ElMinia Governorate, Upper Egypt. All animals were vaccinated against foot and mouth diseases and rift valley fever by using Bacillus Calmette Guerin (BCG) for lambs, Foot and mouth disease (FMD), Rift valley fever, Covexin 8 (for treatment of coccidia and other diseases).

**Live body weight (LBW)**
All animals with their different physiological status were weighed (kg) once per season, where dry, early and late pregnant ewes were weighed in July-August (summer) and February-March (winter).

**Feeding**
Animals were fed maintenance ration according to NRC (1985), the ration contained 12% crude protein, 6 % fat, 12 % fiber and 50 % starch value. Ingredients of this ration are yellow corn (44 %), soyabean (23 %), wheat bran (14 %), rice bran (11.5 %), molasses (4.5 %), limestone (2 %) and salt (1 %). All animals were fed wheat straw *ad libitum* throughout the experiment, while the green fodder was provided to animals in the period...
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from February to March (winter). Water and mineral mixture blocks were freely available throughout the experiment.

**Measuring body fluids**

**Total body water (TBW)**

TBW was determined by injecting j.v. 0.5 ml Urea solution/kg body weight (13% concentration) according to Preston and Kock (1973).

**Extracellular fluid (ECF)**

ECF was determined by injecting j.v. 0.25 ml sodium Thiosianate (NaSCN) solution (5%) /kg body weight according to Hix et al., (1959) and Carlson et al., (1979).

**Plasma volume (PV)**

PV was determined by injecting j.v. Evan's blue solution (0.5%) 0.1 ml/kg body weight according to Lindinger et al., (1995). Before injection, blood samples were collected from each animal and considered as control for the other samples withdrawn after solution injection. The solution was injected into j.v. within 90 seconds of the control sample. After a mixing time, blood samples (12 ml) were collected in the intervals 10, 20, 30, 40, 50 and 60 minutes according to Hodgetts, (1961), centrifuged at $3000 \times g$ for 15 minutes for separating the plasma, and stored at -20 °C for further analyses.
Intracellular fluid (ICF)
ICF was calculated as follows:
ICF = TBW - ECF
Where, TBW = Total body water, ECF = Extracellular fluids.

Blood collection
A total of 48 blood samples (10 ml/each) were collected (6:00 a.m.) from all ewes via j.v. puncture for aldosterone, Na and K plasma determination in summer (July-August) and winter (February-March). Blood samples were centrifuged at 3000 x g for 15 minutes to separate the plasma, then the plasma was preserved in Ependorf tubes (1.5 ml for each) under -20 C for further analyses.

Sodium (Na) and Potassium (k) assay
Plasma sodium and potassium concentrations were determined by method of enzymatic colormetry using kits purchased from the Egyptian Company for Biotechnology (S.A.E.), Egypt.

Aldosterone hormone (Ald) assay
This hormone was assayed by radioimmunoassay (RIA) technique using kits purchased by the Immunotech Beckman Coulter Company (Czech). Ald concentration was determined in the blood plasma according to the method of James and Wilson (1976).

Statistical analysis
All results were subjected to an analysis of variance procedure using general linear model (GLM) using SAS (1999) program. The differences between means were analyzed by least significant difference (L.S.D).
The model used for statistical analysis of LBW (kg), TBF (%), ECF (%), ICF (%), PV (%) and concentrations of Na (mmol/L), K (mmol/L) and Ald. (pg/ml blood plasma) in dry, early- and late-pregnant ewes in summer and winter was:

\[ Y_{ij} = \mu + S_i + p_j + SP_{ij} + E_{ijk} \]

Where,

- \( Y \) = Experimental observation.
- \( \mu \) = General mean.
- \( S_i \) = The effect of season (summer and winter).
- \( p_j \) = The effects of (physiological status, dry, early- and late-pregnant ewes).
- \( SP_{ij} \) = The effect of interaction between season and physiological status.
- \( E_{ij} \) = Experimental error of ij observation.

The correlation coefficients between all studied parameters were also obtained.
RESULTS

Season had a significant influence on LBW in dry and pregnant (early and late pregnancy) ewes, where LBW was greater \( (P<0.05) \) in summer than winter (Fig. 1).

![Bar chart showing LBW in dry and pregnant ewes in summer and winter.](image)

Fig. 1. LBW in dry and pregnant ewes in summer and winter.

Season had also a significant influence on Na concentration in dry and pregnant ewes, where Na concentration in dry and early pregnant ewes was greater \( (P<0.05) \) in summer than winter, whereas Na concentration in late pregnant ewes was not affected by season (Table 1). In general, there was a positive correlation \( (r = 0.56; P<005) \) between Na and K concentrations. Otherwise, K concentration in dry and early pregnancy was greater \( (P<0.05) \) in winter than summer, but it was greater \( (P<0.05) \) in late pregnant during summer than winter (Table 1). Results of this study indicate that K concentration had a negative correlations with TBF \( (r = -0.29; P<005) \), ICF \( (r = -0.26; P<005) \) and PV \( (r = -0.15; P<005) \).
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DISCUSSION

The present study reveals that the LBW increased in summer than winter and that this increase may be attributed to the increase in Na concentration and TBW percentage. Dieckmann and Pottinger (1955) reported that accumulation of 50 mEq Na per kg LBW increased LBW in the last trimester of pregnancy. The current results indicate that Na concentrations in dry and early pregnant ewes were higher in summer than winter, these are similar with findings obtained by Fadare, et al., (2013), where it was reported that Na concentration of adult West African Dwarf sheep was significantly lower in winter (84.17±2.34 mmol/L) than summer (97.19±1.46 mmol/L). In contrast findings of Adedeji (2009) and El-Nouty (1996) showed that goats and cattle had significantly lower values of Na ions during hot season compared with winter. Also, Olsson et al., (1995) showed that increasing ambient temperature from 20° C to 39.5° C for five hours decreased plasma Na concentration of Swedish goats during 5 weeks pre-partum. The no effect of season on Na concentration of late pregnant ewes in the current study is in agreement with findings obtained by Macfarlane et al. (1959) and Abd El-Khalek (1997).

The current results indicate that K concentration in dry and early pregnancy was greater in winter than summer, these are in consistent with that obtained by Adedeji (2009) and El-Nouty (1996) where they reported that goats and cattle had significantly lower K concentrations in hot season than winter. Fadare, et al., (2013) pointed out that the lowered K+ concentration in adult West African Dwarf sheep during the hot weather was due to loss of Na ions in sweat. Otherwise, season had no influence on
plasma K\(^+\) concentrations in adult Merino ewes (Macfarlane et al. 1959). The present findings indicate that K concentration in late pregnancy was greater in summer than winter. Srikandakumar and Johnson (2004) showed that heat stress increased plasma K+ in Holstein and Jersey cows, whereas, it was decreased in Australian milking zebu. 

The present study indicates that aldosterone concentrations in dry, early and late pregnant ewes were greater in winter than summer. These findings are in agreement with findings of Aboul-Naga (1987) and disagreement with that reported by Lund-larsen et al. (1978) and Olsson et al. (1995). However, the increased aldosterone concentration in winter may refer to an increase in potassium and sodium contents of roughage and green fodders compared with their contents in summer roughages. Blair-West et al. (1962); Funder et al. (1969) reported that increasing plasma K concentration led to increase in aldosterone secretion in non-pregnant ewes. Furthermore, increasing Na and K intake greatly influences plasma aldosterone levels in sheep as evidenced by findings of Blair-West et al.(1963.).

Many previous studies (Freyberg et al. 1938; Taylor et al. 1939; Chesley, 1944) reported that Na is retained during pregnancy, and that this retained sodium is required to meet the needs of the fetus.In human, Chesley, (1944) demonstrated that the increased urinary excretion of aldosterone in pregnancy was due partially to a change in the metabolic rate of the hormone compared with that in normal nonpregnant women. Watanabe et
al. (1963) reported that the secretory rate of Ald. was consistently elevated in the third trimester and that this elevation was associated with changes in sodium intake. The indicated also that average secretory rates of Ald. from weeks 28 to 35 was relatively uniform at about 1,000 ug per day, while the further increase in the secretion occurred from week 36 to term. Plentl and Gray (1959) and Davey et al. (1961) indicated that there is a net increase in exchangeable sodium, averaging 510 mEq in the last two trimesters of pregnancy, and that this increment could be responsible for pregnancy outcomes and increase in the maternal blood volume. They also added that the withdrawal of sodium from the mother to the fetus could be responsible for stimulation of Ald. Secretion

Results of this study indicate that TBF was greater in summer than winter this is in agreement with findings of El-Sayed (1988) and Al-Amer, (2011) where it was reported that TBF in dry goats and sheep was significantly greater in summer than winter, this increase may be attributed to escape some fluids from extra vascular tissue space or digestive tract to plasma as evidenced by Chaiyabutr et al., (1990) Otherwise, TBF in Merino sheep was greater under the cold conditions than the hot ones (Macfarlane et al. (1966). ECF in dry ewes was similar in summer and winter which is consistent with findings obtained by Shell et al., (1995) where it was reported that exposure of cows to solar radiation did not change the pattern of extracellular fluids (ECF). In contrast, ECF in dry ewes reared under desert conditions was lower in summer than winter.
However, the current study indicates that ECF in early and late pregnancy increased in summer than winter, this is in agreement with illustration of findings obtained from Silanikove, (1987) where it was found that exposure of German Merino ewes to high temperature (above 38˚C dry bulb) ingested much water than was required for intermediate metabolism and evaporation, allowing plasma and extracellular volume to increase.

The increased ICF percentage in the dry ewes observed in the current results is in agreement with that results obtained by El-Sayed (1988), where it was reported that ICF in dry ewes reared under desert conditions decreased in summer than winter, because in winter, there was return water from the intracellular fluid space to the interstitial fluid space which in turn the volume of blood in sheep. Shell et al., (1995) showed that the exposure of cows to solar radiation led to reduced intracellular fluids (ICF). The current results indicate that ICF percentage was similar in both summer and winter, these results are not similar to findings obtained by Khalifa et al., (2008), where it was reported that exposure of adult pregnant Baladi goats to solar radiation led to the reduction in TBW by 1.76 % and ICF % by 2.52%.

The current results reveal that PV % increase in winter than summer and that this increase was accompanied by the increase in Ald. concentrations in dry, early and late pregnant ewes during winter. Tysoe and Lowenstein 1950 indicated that Ald. concentration increased in pregnancy despite increased plasma volume and extracellular volume (Chesley, 1943). Al-
Amer, (2011) found that the mean PV values was affected significantly by season (summer and winter), species and physiological status of animal.

CONCLUSION

The hypothesis of this study was built on assessments of body fluids compartments and their relationships to aldosterone sodium and potassium concentrations and body weight in dry early and late pregnant ewes during summer and winter. The most revealing results obtained from the current study emphasized that LBW of dry and pregnant ewes was increased with increasing TBF % and Na concentration in summer, Ald. concentrations of dry and pregnant ewes were affected by season and increased with increasing Na and K concentrations and PV % in winter. K concentration had a negative correlations with TBF, ICF and PV compartments, hence these results can be used to maintain body fluids and plasma volume responsible for optimal cellular functions in sheep, particularly in summer.

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