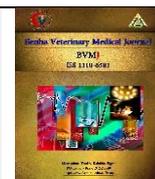




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Ultrasound guided fetometry in Egyptian sheep and goats

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ABSTRACT

The aim of the present study was to build up equation (s) for an intra-uterine fetal aging in Egyptian local sheep and goats breed using ultrasound. This study was carried out on a total number of 110 Baladi sheep and 74 goats under the field condition of Qalyubia governorate, Egypt. Ultrasound examination was conducted through trans-rectal and/or trans-abdominal routes. Throughout the gestation cycle between Day 26 and 112, Crown rump length (CRL) and biparietal diameter (BPD) were measured at least three times per test for each animal for accurate judgment. The correlation coefficient (R^2) of BPD was higher than CRL in sheep and goats (0.9601 and 0.9598 vs. 0.898 and 0.903, respectively) at 39 to 112 days. Statistical analysis showed that no differences ($P=0.258$) between the BPD slopes represent ovine and caprine fetal growth rate. In conclusion, the use of ultrasound estimation in fetal biometry is useful to estimate the gestational age in sheep and goats with particular focus on BPD at all stages of pregnancy after breeding beyond Day 39.

1. INTRODUCTION

An assessment of pregnancy status as well as determination of gestational stage and fetal number is of paramount importance for successful production of sheep and goats. However, poor breeding history of small ruminants reared by small holders represents a great challenge for proper animal rearing and predispose to many health disturbances e.g. ketosis during the periparturient period due to unplanned feeding regimens.

Incorporation of ultrasound diagnosis technology in the field with special attention to breeding and pregnancy determination is helpful for farmers/ livestock producers, veterinarians and researchers. Ultrasonography encourages the development of reproductive management in farms and increases trade in pregnant animals (Reichenbach et al., 2004; Santos et al., 2004). It helps to make decisions on culling/rebreeding, food allocation, clinical and research purposes. Ultrasound is not only used for evaluation of the ovarian functions and/or disorders, but also used to monitor uterine pathophysiology, early pregnancy diagnosis (Beal et al., 1992), embryo/fetal development (Pierson and Ginther, 1987), fetal sexing and prediction of the expected date of parturition (Wright et al., 1988). Moreover, it is applicable for assessing uterine pathology e.g. endometritis, pyometra. Besides, it helps in clarifying the efficacy of treatment regimens (Medan et al., 2003).

Many studies have been conducted on early visualization of the gestational sac. Because of the high incidence of false positive diagnosis due to early embryonic losses, it is better to wait until days 32–34 (Gonzales-Bulnes et al., 2010). Mid-stage pregnancy is characterized by the development of body organs and sexual differentiation of the fetuses. Visualization of fetuses between day 50 and 100 of gestation allows the well separation of the individual

fetuses from one another. Fetal number predictions allowed adequate nutritional management of female in late gestation to avoid metabolic disorders such as hypocalcemia and pregnancy toxemia (Kharache and Kouamo, 2015), minimize pre-lambing feeding costs, improve birth weight, weaning weight and lamb survival, and decrease the incidence of dystocia

To predict the actual fetal gestational age in small ruminants by using ultrasonography, different parameters have been measured e.g. Crown-Rump Length (CRL), Biparietal diameter (BPD), chest diameter, femur length, ...etc. (Kandiel et al., 2015). Accurate information on the gestation stage would be helpful in drying off lactating females at an acceptable time and tracking females in the near term (Suguna et al., 2008).

The present study was aimed to investigate the use of real-time ultrasonography for the determination of pregnancy and/or fetal viability in sheep and goats species. Besides, to assess fetal growth in relation to gestational stage to generate formulae applicable under field conditions in Egyptian balady sheep and goats breed.

2. MATERIAL AND METHODS

2.1. Animals

The present study was carried out on pregnant Baladi sheep (n=110) and goats (n=74) under field condition of Qalyubia governorate, Egypt during the period from December 2018 to December 2019. All animals were admitted to Meet Kenana Veterinary Clinic Tukh, Qalyubia governorate for pregnancy diagnosis.

2.2. Ultrasonographic examination

Ultrasonographic examination of sheep and goats was performed trans-rectally and/or trans-abdominally using a

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real time B- mode scanner equipped with 3-8 MHz transducer (Sonoscape MA5 Vet., China) according to Kandiel et al. (2008).

For trans-rectal examination, animals were restrained in a standing position and the well lubricated ultrasound transducer was preceded through the rectum until the urinary bladder and the uterine horns were imaged. For trans-abdominal examination the animals were restrained horizontally on one side on a table. An area of 20-40 cm around the udder was clipped and both sides of the abdomen have been scanned. The area over which the transducer was moved extended across the width of the abdomen, passing from one side of the udder, across in front of the udder, to the other side (Kandiel et al., 2015).

The stage of pregnancy in the examined animals was determined by examination of crown rump length (CRL) and/or biparietal diameter (BPD) according to Kandiel et al. (2015), and Kandiel et al. (2016).

Crown rump length was measured from 4th week until 6th week of gestation by placing the markers at the upper part of the skull, to the end of the sacrum (end of the spine), when the fetus was fully extended. If the fetus was curved, measurements were taken in two stages: from head to heart, and from heart to sacrum. Biparietal diameter was from the 6th week till end of pregnancy.

From the 6th to the 16th week, biparietal diameter (BPD) was measured as follows: oval shape as possible, closed skull table contour, falx cerebri mid-line dividing the hemispheres into two equidistant sections, and measurements taken from the proximal calvarium outer surface to the distal calvarium inner surface. For an accurate measurement, image symmetry is important.

2.3. Statistical analysis

The relation between gestational period (days) and fetometric parameters (mm) was plotted using a linear regression model (Microsoft Excel) according to the formula $Y = aX + b$, where Y = gestational age, a = constant, x = measured value, and b = dependent variable. Correlation coefficient (R^2) between the changes in fetometric parameters and gestational age was calculated for estimating the reliability of predictability. The significant differences between the slopes of fetal growth in sheep and goat species were calculated using Student's t -test. P value was set at < 0.05 to mark the significance level. The values obtained were presented as mean \pm standard error of mean (SEM).

3. RESULTS

3.1. Ultrasonographic monitoring of the fetal growth rate in sheep and goats

Representative ultrasound images of sheep and goats fetometry are demonstrated in Ultrasonogram 1&2.

3.1.1. Crown rump length

Sheep feti (n=9) examined during early pregnancy from Day 28 to Day 41 of gestation period is presented in fig. 4.1A followed the regression equation $y = 3.635x + 3.8806$ ($R^2 = 0.898$). Goats feti (n=11) examined from Day 26 to Day 41 of gestation followed the regression equation $y = 2.9236x + 4.2582$ ($R^2 = 0.903$) (Fig. 1B).

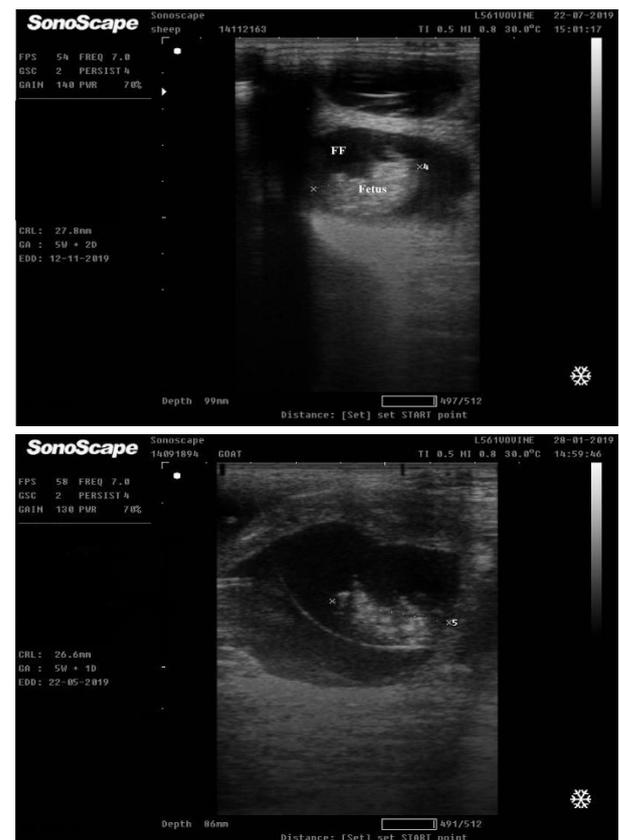
3.1.2. Biparietal diameter

Data regarding the growth rate of sheep and goats feti in relation to gestation period is presented in fig. 1&2 and table 1.

The ovine and caprine feti growth rates showed a steady increase linearly with the progression of gestation period. The regression analysis between fetal age (days) and biparietal diameter (mm) in sheep feti at 39 to 112 days ($R^2=0.9601$; $P<0.0001$) generated the following linear regression equation: BPD (mm) $Y = 0.6581X - 12.922$, $R^2 = 0.9601$. On the other hand, the linear regression equation of caprine feti was $y = 0.62x - 9.701$ ($R^2 = 0.9598$; $P<0.0001$). Statistical analysis verified that the differences between the two slopes (represent ovine and caprine fetal growth rate as indicated by BPD) were not significantly different ($P=0.258$).

4. DISCUSSION

Due to the long interval between births, inability to detect early pregnancy in small ruminants can result in economic losses in milk and newborn production. The determination of gestational age is an important part of the efficient efficacy of reproduction. Ultrasound fetometry serves to assess the average growth rate of fetu during pregnancy, beside its role in the detection of pregnancy and litter size determination. Ultrasonography is the most commonly used approach for early pregnancy diagnosis in veterinary practice since it is a simple, reliable, and non-invasive imaging technique. Crown-rump length measures the fetal length while it is in a curved posture resembling the letter C and is useful for determining the growth rate and gestational stage.



Ultrasonogram 1 Representative ultrasound images of pregnant uterus showed crown rump length in sheep (top) and goats (bottom). Note the presence of hyper echoic fetal mass (white color) surrounded by anechoic fetal fluids (FF; black color).



Ultrasongram 2 Representative ultrasound images of pregnant uterus showed biparietal diameter in sheep (top) and goats (bottom). Note the presence of hyper echic fetal mass (white color) surrounded by anechoic fetal fluids (FF; black color).

Ultrasonography is the most commonly used approach for early pregnancy diagnosis in veterinary practice since it is a

simple, reliable, and non-invasive imaging technique .Crown-rump length measures the fetal length while it is in a curved posture resembling the letter C and is useful for determining the growth rate and gestational stage. In the present study the fetal CRL and the gestational stage were highly correlated in sheep and goats (0.898 and 0.903) during early pregnancy (Day 26-41 post-breeding), indicating its reliability in determining gestational stage at such early period. It is preferable to delay pregnancy until days 32–34 to avoid the false positive diagnosis results from early embryonic losses (Gonzales-Bulnes et al., 2010). Also, beyond the 10th week of breeding CRL becomes inaccessible due to the increase in the fetal size, which is incompatible with the transducer size and its penetration properties (Amer, 2008; Karen et al., 2009). On the other hand, the fast fetal movement during ultrasound examination represent an obstacle for the feasibility of CRL measuring in goats (Abdelghafar et al., 2007). The present findings are in agreement with Martinez et al. (1998), who demonstrated that CRL was best exemplified by a direct relapse ($R^2 = 0.94$) from Days 19 to 38 of gestation in Anglo-Nubian goats. A close correlation ($R = 0.90$) was also obtained in Saanen goats examined between the 5 and 10 week of gestation (Abdelghafar et al., 2011). On the other hand, a higher correlation between CRL and gestational age has been stated by Gonzales-Bulnes et al. (2010) from Days 19 to 48 of gestation ($R^2 = 0.94$), by Lee et al. (2005) from days 19 to 38 days post-mating ($R^2 = 0.94$), and by Kandiel et al. (2015) from 25-70 days ($R^2 = 0.9848$). The differences perhaps due to the frequency of observation, animal breed, type/sensitivity of ultrasound machine.

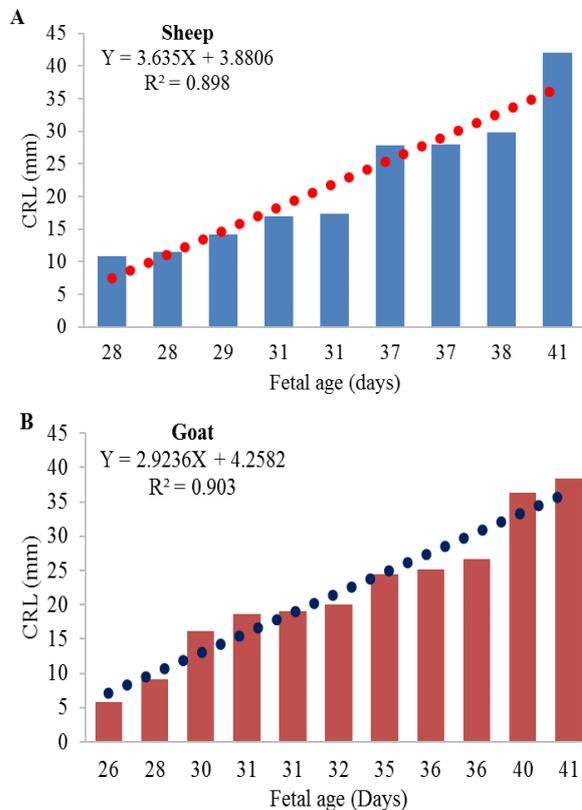


Fig. 1 Changes in fetal growth rate indicated by crown rump length in mm (CRL) in relation to gestational stage (days) in Egyptian balady sheep (A) and she goats (B) species.

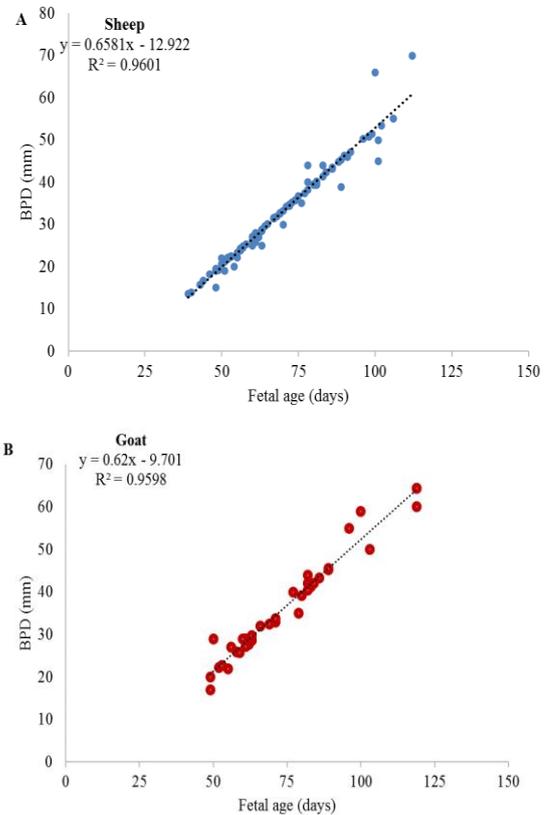


Fig. 2 Changes in fetal growth rate indicated by biparietal diameter in mm (BPD) in relation to gestational stage (days) in Egyptian balady sheep (A) and she goats (B) species.

Table 1 Ovine and caprine fetal biometric measurements (mm) from 6th to 16 weeks of the gestation period

Fetal age (weeks)	BPD (mm) of ovine fetus			BPD (mm) of caprine fetus		
	Mean ± SE	Min.	Max.	Mean ± SE	Min.	Max.
7 w	19.00±0.53	17.00	22.00	20.00±0.05	20	20
8 w	23.00±0.33	21.00	25.00	23.00±0.34	22	24
9 w	27.00±0.32	25.00	29.00	27.00±0.50	26	30
10 w	32.00±0.42	30.00	34.00	33.00±0.78	31	34
11 w	36.00±0.39	34.00	37.00	39.00±0.50	38	39
12 w	40.00±0.44	38.00	43.00	41.00±0.39	39	42
13 w	45.00±0.58	43.00	47.00	45.00±0.69	43	46
14 w	50.00±0.91	47.00	51.00	50.00±0.00	50	50
15 w	52.00±0.35	51.00	53.00	54.00±0.00	54	54
16 w	58.00±2.10	56.00	60.00	64.00±0.00	64	64

The present study showed that the period from day 9 to 112 days characterized by clear anechoic or nearly hypo-echoic brain cavity and hyper-echogenic brain cage (skull bone), which makes an easy access and measurement of the braincase or biparietal diameter. These findings closely agreed with Abdelgafar et al. (2007), Amer (2008 & 2010), Karen et al. (2009), Nwaogu et al. (2010) and Kandiel et al. (2015), who presented a very high correlations between gestation age and BPD in the second trimester of gestation. However, the feasibility of BPD measurement beyond Day 95 post-breeding is also difficult due to the increase in fetal size and compression of the head by other fetal parts (Abdelgafar et al., 2007).

In the current study, a high correlation was found between BPD and gestational stage in sheep and goats (0.9601 and 0.9598, respectively), even high than the recorded correlation with CRL. These findings is similar to that found by Serin et al. (2010) and Gonzales-Bulnes et al. (2010) ($R^2= 0.96$) with fetal age measurement in sheep from day 32-90 days in ewes, and Karen et al. (2009) in Egyptian Baladi goats ($R^2= 0.956$) at 30-105 days of gestation. However, the present findings were higher than that recorded by Segreev et al. (1990) in the ovine species ($R^2=0.94$), but lower than that was recorded in ewes by Haibel and Perkins (1989) ($R^2=98.63\%$), and Kandiel et al. (2015) ($R^2 = 0.9831$) in goats. The differences might be related to the scheme of examination, frequency of examination, fetal numbers and close apposition of fetal organs to the transducer face. Karen et al. (2003) showed that the accuracy of trans-rectal ultrasonography differ with the stage of pregnancy. Amer (2008) reported that the accuracy of pregnancy diagnosis in goats was high for single pregnancies during days 40-60 of gestation, that decrease to 54.3%, with twine pregnancy.

5. CONCLUSIONS

Ultrasound of the fetal biometric variables constitutes an efficient tool to estimate stage of pregnancy in sheep and goats. The practice of BPD in small ruminants presents several advantages over CRL represented by its high correlation ($R^2=0.96$) with gestational period and the extended applicability time (6th to 16th weeks). However, the pros argument of the application of CRL is its applicability at early pregnancy as early as Day 28 post-breeding when the BPD is inaccessible. There was no significant differences in the slope represented the fetal growth curve.

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