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RESEARCH ARTICLE

May the fetal kidney measurements be collateral criteria on the prediction of gestational age in cattle?

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Özet

Erdoğan S, Kılınç M. Fötal böbrek ölçümleri sığırlarda gestasyonel yaşın tahmin edilmesinde yardımcı bir kriter olabilir mi? **Eurasian J Vet Sci, 2012, 28, 2, 69-76**

Amaç: Bu çalışma gestasyonel yaş ile böbreklerin gelişimsel morfometrik ölçüleri arasındaki ilişkiyi araştırmayı amaçlamaktadır.

Gereç ve Yöntem: Bu çalışmada, 30 adet Holstein ırkı sığır fötusu kullanıldı. Gebeliğin erken, orta ve geç dönemi olmak üzere her bir döneme ait 10 fötus değerlendirildi. Buna göre, toplamda 60 böbrek bilateral disseksiyon sonrası üç farklı renal parametre (uzunluk, genişlik, kalınlık) bakımından değerlendirmeye alındı. Böbreklerden elde edilen morfometrik verilerin istatistiksel analizleri yapıldı.

Bulgular: Genel olarak gebelik süresince tüm renal ölçümler gestasyonel yaş ile önemli derecede korelasyon gösteriyordu. Sağ ve sol böbreğin uzunluk ve genişliğinin gestasyonel yaş ile yüksek derecede ($R^2 \geq 0.90$) korelasyonlu olmasına karşın, her iki böbreğin kalınlığı gestasyonel yaş ile daha az ($R^2 \leq 0.86$) korelasyon göstermekteydi. Tüm renal parametreler gebeliğin erken, orta ve geç dönemindeki alın-sağrı uzunlukları ile de önemli derecede korelasyonluydu, ancak gebeliğin geç döneminde sol böbreğin aksine sağ böbrek kalınlığı ile alın-sağrı uzunluğu arasındaki korelasyon önemsizdi. Ayrıca sol böbreğin cranial genişliği ile alın-sağrı uzunluğu arasındaki korelasyon da önemsiz olarak bulundu.

Öneri: Organ gelişimi ve gestasyonel yaş arasındaki korelasyona anatomik bir yaklaşımda bulunan bu çalışmanın sonuçlarına göre, böbrek ölçümleri gestasyonel yaşın tahmin edilmesinde yardımcı bir kriter olarak kullanılabilir.

Abstract

Erdogan S, Kilinc M. May the fetal kidney measurements be collateral criteria on the prediction of gestational age in cattle? **Eurasian J Vet Sci, 2012, 28, 2, 69-76**

Aim: This study aims to investigate the relationship between gestational age and developmental morphometric measurements of the kidneys.

Materials and Methods: In the present study, 30 Holstein cattle fetuses were used. Ten fetuses, belonging to each of the early, mid- and late gestational stages were evaluated. Accordingly, in total, 60 kidneys were assessed after bilateral dissection for three different renal parameters (length, width, thickness). The morphometric data obtained from the kidneys was subjected to statistical analysis.

Results: In general, throughout gestation, all renal measurements were significantly correlated with gestational age. The length and width of the right and left kidneys were highly ($R^2 \geq 0.90$) correlated with gestational age but, in comparison, the thickness of both kidneys was less correlated ($R^2 \leq 0.86$) with gestational age. All of the renal parameters were significantly correlated with the crown-rump length in the early, mid- and late stages of gestation but, the correlation between the thickness of the right kidney and crown-rump length was not significant in the late stage of gestation contrary to the left kidney. Moreover, the correlation between the cranial width of the left kidney and the crown-rump length was also shown to be insignificant.

Conclusion: According to the results of this study, which was based on an anatomical approach to the correlation between organ development and gestational age; kidney measurements can be used as collateral criteria for the prediction of gestational age.

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Keywords: Kidney measurements, gestational age, cattle

► Introduction

The precise diagnosis of pregnancy and embryonic/fetal viability in domestic animals is important in terms of economics and for the conduct of scientific investigation. Embryonic and fetal deaths in cattle cause substantial economic loss to the producer (Sheldon 1997). The reasons for such mortality are many and include fetal and placental abnormalities, environmental insult, deficient maternal support and inappropriate feto-maternal interactions (Wilmut et al 1986). Considering the high input costs involved, the effective diagnosis of embryonic/fetal viability is particularly valuable in the assessment of assisted reproductive technology (ART) derived pregnancies (Riding et al 2008).

Many methods based on the measurement of the crown-rump length (CRL) (Revol and Wilson 1991, Karen et al 2009), biparietal diameter (Abdelghafar et al 2007, Innocent et al 2010), trunk diameter (Aiumlamai et al 1992, Karen et al 2009), placentome size (Karen et al 2009, Innocent et al 2010), umbilical cord diameter (Revol and Wilson 1991, Innocent et al 2010), fetal heart rate (Curran and Ginther 1995, Vahtiala et al 2004), fetal weight, intercostal space (Gonzalez de Bulnes et al 1998), fetal heart diameter (Yanagawa et al 2009), amniotic sac dimensions and amniotic fluid volume (Riding et al 2008) have been used for the prediction of gestational age in humans and livestock for many years. Besides, some cephalic index standards such as the fronto-occipital length, occipito-nasal length and orbital diameter (Innocent et al 2010) have also been used for this purpose in both humans and animals. Furthermore, the correlation between gestational age and the length of the femur, tibia, humerus, metacarpus and heel has been assessed (Greenwood et al 2002, Herrera et al 2002, Ziylan and Murshid 2003, Karen et al 2009). While most of these studies were conducted in small ruminants (Gonzalez de Bulnes et al 1998, Ali and Hayder 2007) and humans (Suranyi et al 2003, Ziylan and Murshid 2003) owing to both manipulation facility and ease of procurement, the majority of studies in large ruminants were conducted at certain gestational stages (Vahtiala et al 2004, Rosiles et al 2005).

To date, real-time B-mode ultrasound scanning has been used for the regular production of fetometric data. To a less extent, direct fetal measurements have been used to determine the correlation between gestational age and fetal structures. Genital organs and fetal development have been studied in both exotic (Place et al 2002, Yanagawa et al 2009) and laboratory animals (Brown et al 2006) as well as in domestic animals (Lee et al 2005, Ali and Fahmy 2008, Innocent et al 2010) by transabdominal and transrectal ultrasound techniques, but fetal organs have been evaluated less frequently in comparison to the above mentioned traditional criteria. However, many researchers (Garcia et al 1993, Gonzalez de Bulnes et al 1998, Lee et al 2005, Amer 2007) have reported that fetal organ parameters such as the long and short axes of the heart, width of the ribs, weight of the internal organs, stomach and kidney diameters at the long axis have become indispensable and reliable tools in estimating the age of fetuses in animals. Moreover, Doize et al (1997) emphasized the importance of gestational age prediction for maximizing the survival rates of newborns, as well as for preparing appropriate rations based on their nutritional needs and determining the appropriate time of drying off. Herrera et al (2002) reported that it was of great significance to follow these approaches in order to determine correctly the fetal age in obstetric examinations, and in cases of abortion and parturition where the breeding date of flocks was unknown or mating programmes of herds were absent.

It is considered that obtaining morphometric data pertaining to fetal organs and determining correlation levels between organ sizes and gestational age would contribute to the establishment of alternative and collateral criteria for determining the order of development of fetal organs and gestational age in animals subjected to caesarean section or abortion any time during gestation, as well as for monitoring prenatal and perinatal development, distinguishing between normal and abnormal organ development in aborted fetuses, and evaluating pathologically macerated and mummified fetuses.

In the present study was aimed to investigate the relationship between gestational age and developmental morphometric measurements of the kidneys, which have not been evaluated previously for the prediction of gestational age.

► Materials and Methods

Holstein cattle fetuses (n=30) of both sexes were supplied from local slaughterhouses in Diyarbakir province, were used. The gestational age of the fetuses was calculated by using the equation y: 54.6+2.46(x), based on the linear relation between the CRL and gestational age, after the CRL was measured (Harris et al 1983) (Figure 1). In this equation, y and x represent the gestational age and CRL, respectively. Ten fetuses, belonging to each of the early (65-90 days), mid- (90-180) and late (180-270) stages of gestation, were examined, based on this analysis.

In total, 60 kidneys not showing any pathological malformation were evaluated after bilateral dissection. Before performing measurements, the adipose tissue surrounding the kidneys was removed to determine precisely the correct size of the kidneys. The measurements of the kidneys were scaled by a digital calliper with 0.01 mm accuracy. The statistical analyses of the morphometric data obtained from the kidneys were performed by SPSS 17.0 Software® (SPSS Inc., 2008, Chicago, IL, USA) and the correlation between the

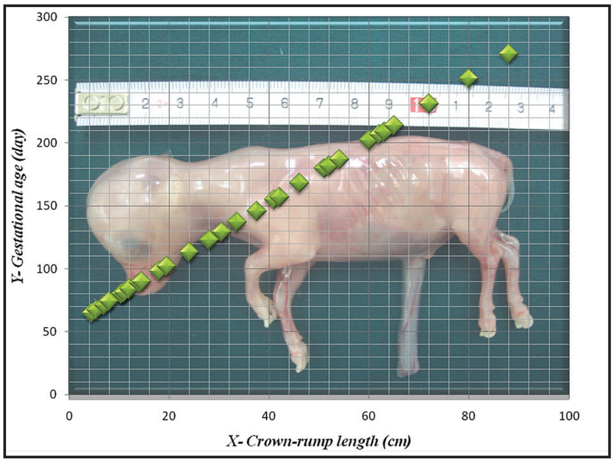

Figure 1. The relationship between Crown-rump length (CRL) and gestational age.

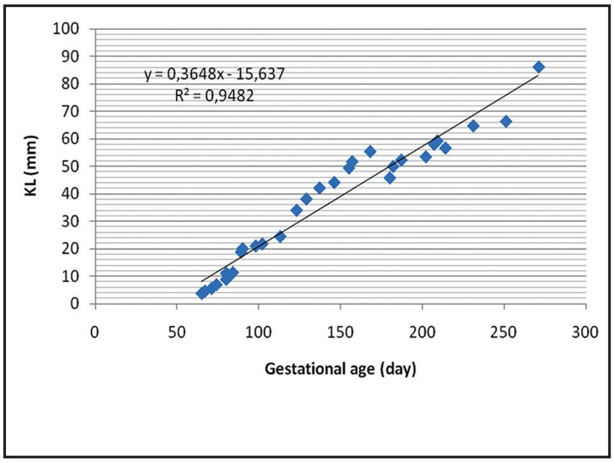

Figure 2. Regression line of the right fetal kidney lengths at different gestational ages (n=30).

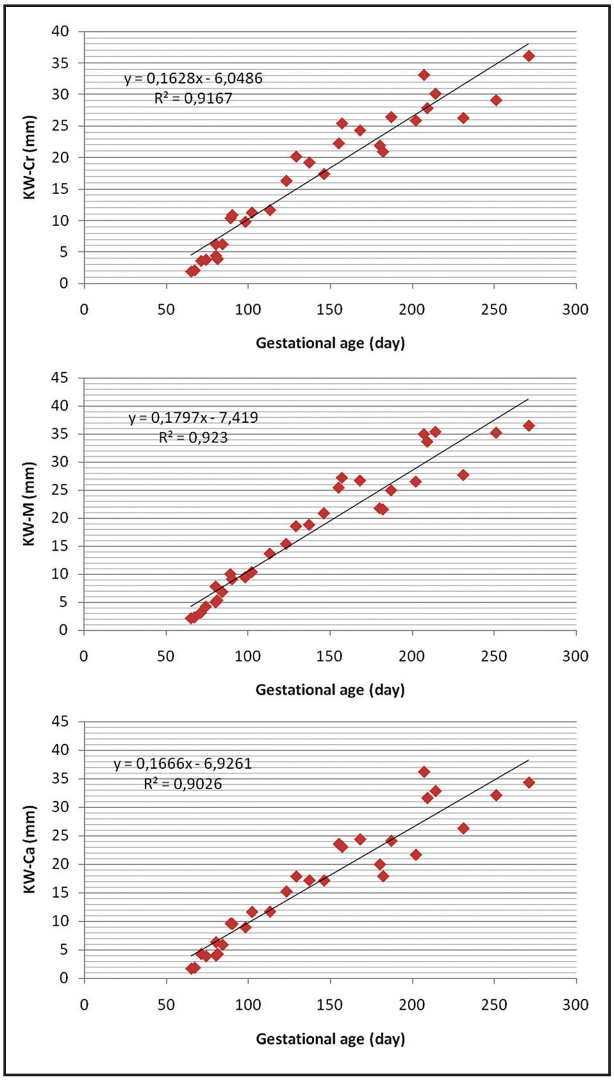

Figure 3. Regression lines of the widths of the right fetal kidneys at different gestational ages (n=30).

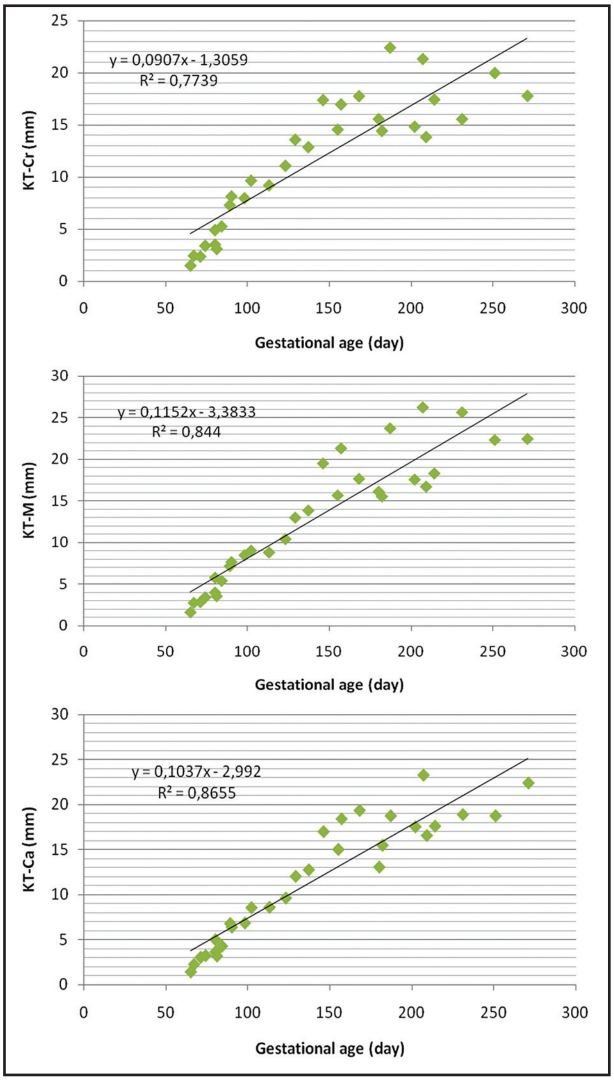

Figure 4. Regression lines of the thicknesses of the right fetal kidneys at different gestational ages (n=30).

variables was determined using Pearson's correlation test. Moreover, regression models were adapted to assess the correlation between the gestational age and each of the renal parameters. After the fetuses were dissected, the following kidney parameters were measured and evaluated (Erdogan and Kilinc 2011);

Kidney Length (KL): The longest distance between the cranial extremity and caudal extremity of each kidney.

Cranial Width of Kidney (KW-Cr): Considering that the kidney is divided into three equal parts, the longest mediolateral distance in the upper one-third of the kidney.

Middle Width of Kidney (KW-M): The longest mediolateral distance between the renal hilus and lateral margin.

Caudal Width of Kidney (KW-Ca): Considering that the kidney is divided into three equal parts, the longest mediolateral distance in the lower one-third of the kidney.

Cranial Thickness of Kidney (KT-Cr): Considering that the kidney is divided into three equal parts, the longest dorsoventral distance in the upper one-third of the kidney.

Middle Thickness of Kidney (KT-M): The longest dorsoventral distance between the renal hilus and lateral margin.

Caudal Thickness of Kidney (KT-Ca): Considering that the kidney is divided into three equal parts, the longest dorsoventral distance in the lower one-third of the kidney.

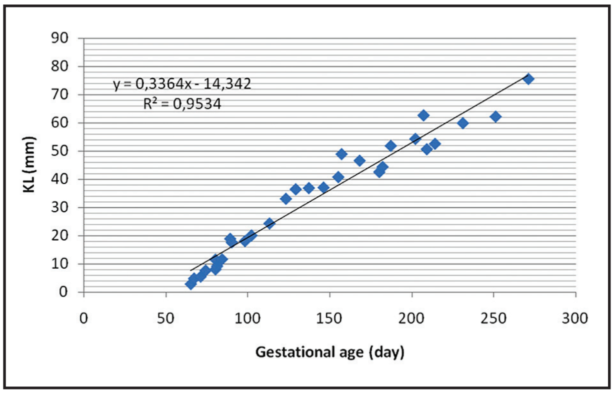

Figure 5. Regression line of the left fetal kidney lengths at different gestational ages (n=30).

► Results

The relationship between kidney measurements, which were obtained from fetuses directly, and gestational age are shown in Figures 2-7. Linear regression curves were generated for all kidney measurements. Based on these regression analyses; in general, all kidney measurements were significantly ($p<0.01$) correlated with gestational age throughout gestation. The coefficients of correlation (R^2) between gestational age and the kidney parameters investigated in the present study ranged between 0.773 (KT-Cr) and 0.953 (KL). The length (Figures 2 and 5) and width (Figures 3 and 6) of the right and left kidneys were highly correlated ($R^2 \geq 0.90$) with gestational age but, the thickness of both kidneys (Figures 4 and 7) was less correlated ($R^2 \leq 0.86$) with gestational age in comparison to the length and width of the kidneys.

Table 1. The correlation between the parameters of the right kidney and CRL in each three groups.

Right Kidney	Group 1 (n=10)	Group 2 (n=10)	Group 3 (n=10)
	CRL	CRL	CRL
KL	0.946**	0.992**	0.952**
KW-Cr	0.907**	0.940**	0.750*
KW- M	0.944**	0.987**	0.749*
KW- Ca	0.912**	0.965**	0.673*
KT-Cr	0.910**	0.943**	0.158
KT-M	0.925**	0.902**	0.459
KT-Ca	0.915**	0.965**	0.615

Group 1; Early stage of gestation, Group 2; Mid-stage of gestation, Group 3; Late stage of gestation, **:$p<0.01$; *: $p<0.05$.

The relationship between kidney parameters and the CRL was calculated separately in each gestational stage (Tables 1 and 2). Accordingly, all of the kidney parameters were significantly ($p<0.01$) correlated with the CRL in the early and mid-stages of gestation but, this situation was different in the right kidney in the late stage of gestation. In the late stage, the correlation between the width measurements of the right kidney and the CRL was significant ($p<0.05$), and the correlation between the length of the right kidney and the CRL was highly significant ($p<0.01$). The correlation between the thickness of the right kidney and the CRL was not significant in the late stage of gestation, contrary to the left kidney. Furthermore; the cranial width measurements of the left kidney was smaller than that of the right kidney. In the late stage of gestation, the cranial pole of the left kidney rotates due to the pressure of the expanding rumen. For this reason, the correlation between the cranial width of the left kidney and the CRL was not significant (Table 2).

Table 2. The correlation between the parameters of the left kidney and CRL in each three groups.

Left Kidney	Group 1 (n=10)	Group 2 (n=10)	Group 3 (n=10)
	CRL	CRL	CRL
KL	0.942**	0.964**	0.895**
KW-Cr	0.933**	0.966**	0.481
KW- M	0.911**	0.957**	0.790**
KW- Ca	0.877**	0.962**	0.768**
KT-Cr	0.904**	0.875**	0.774**
KT-M	0.937**	0.868**	0.838**
KT-Ca	0.889**	0.950**	0.790**

**:$p<0.05$

► Discussion

Studies on the development and growth of some fetal organs (head, eyes, bones, heart, kidney, urinary bladder) became possible with the use of ultrasonography in non-risk pregnancy (Sinclair et al 1998, Noia et al 2002, Ali and Hayder 2007). Gestational diagnosis and estimation of gestational age are essential for the maintenance of high levels of reproductive efficiency (Haibel 1990). Kidney length has been used for the prediction of gestational age in human fetuses (Chitty and Altman 2003, Suranyi et al 2003, Vlajkovic et al 2006) and a significant correlation was detected between kidney parameters and biparietal diameter,

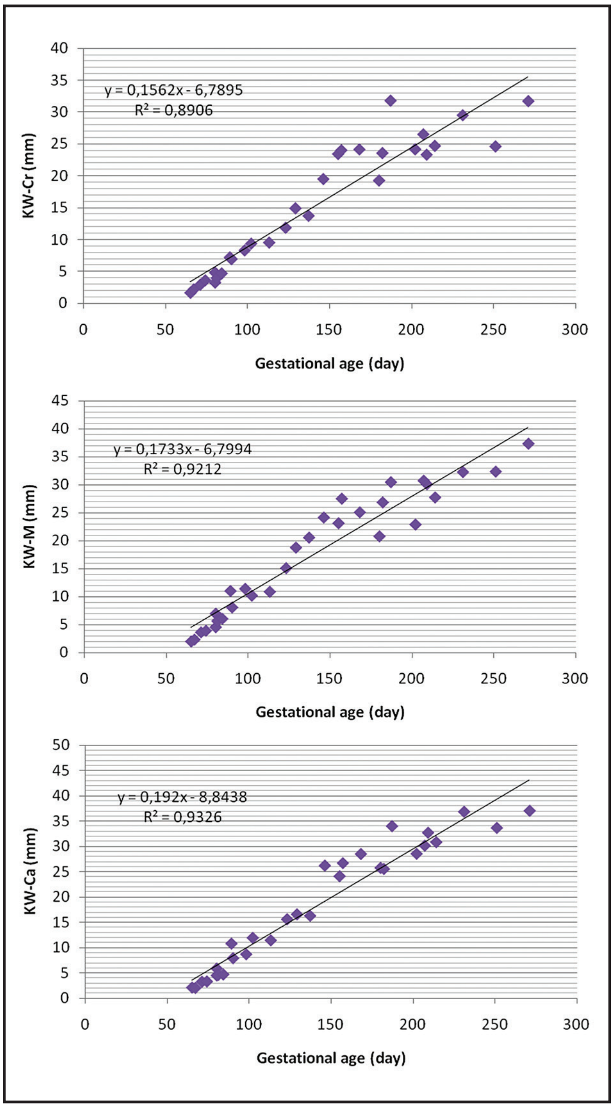

Figure 6. Regression lines of the widths of the left fetal kidneys at different gestational ages (n=30).

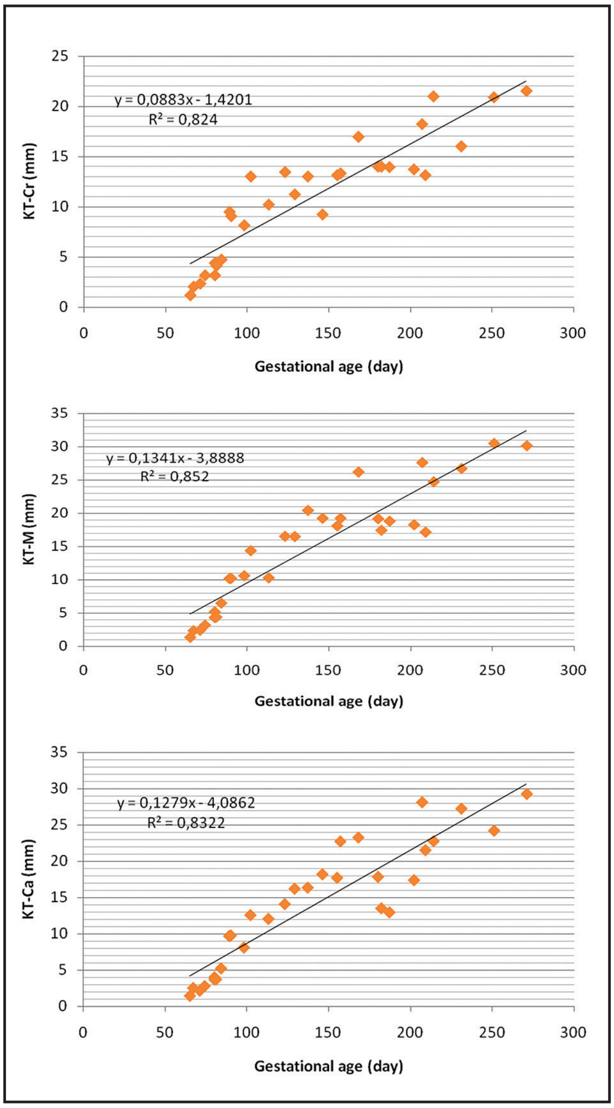

Figure 7. Regression lines of the thicknesses of the left fetal kidneys at different gestational ages (n=30).

CRL, femur length and gestational age. Furthermore, in human fetuses, the relationship between the size of the kidney and renin-angiotensin levels has been evaluated physiologically (Konje et al 1996).

Ali and Hayder (2007) have evaluated the use of ovine kidney length in the estimation of gestational age. They measured the length of the kidneys during the second and third trimesters of gestation and recorded parameters such as the CRL, amniotic vesicle, chest depth, abdominal diameter, ruminal length, eyeball diameter and placentome diameter by ultrasonography. They used only the long axis of the left one to minimize the possible variability between the dimensions of the left and right kidneys. The relationship between all parameters, except for the placentome diameter, and coefficients of regression and correlation was found highly significant ($p<0.001$). In humans, the fetal kidney could be visualized first by day 73.2±6.3 (range 62-76) and the kidney length was measured by high resolution transvaginal ultrasonography between weeks 14 and 17 of gestation (Zalel et al 2002). It was reported that kidney length could be used in the prediction of gestational age as an adjunct in both humans (Zalel et al 2002, Vlajkovic et al 2006) and sheep (Ali and Hayder 2007).

Ali and Fahmy (2008) have used certain parameters of fetal internal organs such as ruminal length, omasal diameter and eyeball diameter as well as external measurements (CRL, biparietal diameter, uterine diameter). In buffaloes (Ali and Fahmy 2008), fetal parts and organs developed differently, with increasing (CRL, biparietal diameter, omasal diameter) or decreasing (uterine diameter, ruminal length, eyeball diameter) growth rate with the progress of gestation. However, a highly significant ($P<0.001$) correlation and regression was detected between gestational age and each of the parameters studied. Coefficients of correlation between the rumen (R^2=0.67), omasum (R^2=0.61) and gestational age was found to be lower in comparison to other organ measurements. Zambelli et al (2004) also detected a highly significant (R^2=0.97) correlation between gastric diameter and gestational age in the second half of gestation in cats. During the first half of gestation, Zambelli et al (2002) compared gross manual measurements (gestational sac and embryonal/fetal length) with ultrasonographic measurements and demonstrated a strong correlation in cats. Gonzales de Bulnes et al (1998) recorded the longitudinal diameters of the stomach, kidneys and urinary bladder, and showed a linear correlation between the measurements of each of the organs, excluding the urinary bladder, and gestational age. Similarly; in this study, a higher correlation (R^2=0.953 for right kidney length, R2=0.954 for left kidney length) was found between the longitudinal length of the kidneys and gestational age.

Gunduz et al (2010) reported that head diameters provided a good index of fetal development because they were highly correlated with gestational age, thus enabling long periods of observation. Haibel (1990) showed that the biparietal diameter was the most representative parameter of gestational age during the second trimester of pregnancy. Kelly and Newnham (1989) considered the occipito-nasal length to be a more accurate measure than the biparietal diameter, showing a linear increase until day 80. However, Gunduz et al (2010) reported that the biparietal diameter was significantly correlated with gestational age between weeks 8 and 20 of gestation. Similar results were reported by Greenwood et al (2002) in Suffolk ewes between days 60 and 120, and by Gonzales de Bulnes et al (1998) in Manchega ewes between days 32 and 90 of gestation. Gonzales de Bulnes et al (1998) reported that the estimation of gestational age was more accurate during early pregnancy since values obtained in late gestation are also affected by the individual characteristics of the fetus.

Gazitua et al (2001) performed biparietal diameter measurements until the sixth month of pregnancy using transrectal ultrasonography in both llamas and alpacas, and reported that the correlation between biparietal diameter and gestational age was highly significant ($p<0.001$). However, these researchers emphasized that although there were some difficulties in obtaining fetal measurements, the resultant functions were reliable predictors of gestational age, as indicated by the highly significant correlation coefficients (ranging between 0.95 and 0.98). In the study, it was seen that most of the renal parameters were highly correlated with gestational age (R2≥0.90), which suggested their use as reliable predictors of gestational age. Zambelli et al (2004) reported that there was a high correlation between gestational age and biparietal diameter in feline fetuses during the second half of pregnancy. Place et al (2002) and Haibel and Fung (1991) stated that the biparietal diameter was a hardly obtainable criterion in late gestation, particularly in their study. Place et al (2002) found a highly significant correlation (R^2=0.987, $p<0.001$) between the femoral length and gestational age after the 65th day of gestation, and they reported that the femoral length could be more advantageous than the biparietal and abdominal diameters. Even though all the measurements obtained from fetuses of Manchega dairy ewes were correlated ($p<0.005$) with gestational age, the highest correlation coefficient (R^2=0.987) was estimated for the biparietal, thoracic diameter and width of the coccygeal vertebra. Gonzales de Bulnes et al (1998) reported that the most appropriate parameters were head diameter, occipito-nasal length and orbit diameter in progressive gestational stages and added that these structures were monitored according to a plan.

The CRL has been frequently used in post-mortem fetal measurements in different species and is considered one of the most typical criteria (Evans and Sacks 1973, Sivachelvan et al 1996). The demonstration of its value in terms of the prediction of gestational age in this research shows similarity to prior data obtained by ultrasonography in animals. Abdelghafar et al (2007) found a highly significant ($p<0.001$) correlation between the CRL and gestational age, similar to the case with the biparietal diameter. Similarly, a high correlation between gestational age and the CRL was detected from day 25 onwards in Mouflon sheep (Santiago-Moreno et al 2005), and between days 36-102 post-mating in ewes (Aiumlamai et al 1992) and the Pygmy goat (Lee et al 2005). Ali and Fahmy (2008) detected that both the correlation between the CRL and biparietal diameter in mid-gestation and the correlation between the CRL and eyeball diameter in late gestation were considerably higher.

In previous studies, fetuses which were obtained as slaughterhouse material were also used and the results of these studies showed that the most satisfactory criterion in the prediction of gestational age was a curved CRL measurement. In these studies, most criteria including head diameter, abdomen circumference, and chest depth were used, but the relationship between the developmental rates of internal organs, such as the kidneys, and gestational age was not assessed (Abdel-Raouf and El-Naggar 1968, 1970). Moreover, measurements of the amniotic compartment and amniotic fluid volume were also evaluated in slaughterhouse materials (Riding et al 2008).

► Conclusions

Measurements of the left kidney were highly correlated with gestational age during pregnancy. On the other hand, the right kidney was strongly correlated with gestational age in both the early and mid-stages of gestation but, in the late stage of gestation, it was detected that this correlation weakened for kidney thickness. It is considered that the data obtained could be helpful in the ultrasonographic evaluation of the correlation between the kidneys and gestational age; and that the length and width of the kidneys give more precise results. Therefore, the difference in the growth of the left and right kidneys in the late stage of gestation should be taken into consideration. According to the results of the present study, which was based on an anatomical approach to the relationship between organ development and gestational age; kidney measurements can be used as collateral criteria in predicting gestational age and can aid practitioners in evaluating fetal organs.

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