TÜBİTAK

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Comparison of placentome diameters in single and twin-pregnant sheep by ultrasonographic method

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Abstract: The aim of this study was to determine the correlation between fetal number and placentome diameters in pregnant sheep. In the present study 25 Kıvırcık mixed breed pregnant ewes were used. Pregnancy of the ewes was diagnosed by transrectal ultrasonography by the 25th day after insemination. A transabdominal probe (6.5 MHz) was used on the pregnant ewes at the 30th day. At this time the placentomes appeared as hypoechogenic areas with a "C"-like shape. Throughout the pregnancies 5 placentomes were measured from each ewe every week. The results showed no significant difference between placentome diameter size in single and twin-pregnant ewes except during the 5th and 12th weeks. The average of placentome diameter size in the single and twin-pregnant ewes was, respectively, 6.60 ± 0.56 mm and 8.20 ± 0.45 mm at the 5th week and 34.02 ± 1.62 mm and 29.47 ± 0.63 mm at the 12th week. The minimum significance degree was $P < 0.05$ in statistical analysis. This study found that measurement of placentome diameter size at the 5th and 12th week of pregnancy is not helpful in determining fetal number in a pregnant ewe. However, further investigations are needed to explain the difference in placentome diameter at the 5th and 12th weeks of pregnancy.

Key words: Pregnant ewe, fetal number, placentome diameter

Tekli ve ikiz gebe koyunlarda plasentom çaplarının ultrasonografik metot ile karşılaştırılması

Özet: Bu çalışmanın amacı, gebe koyunlarda plasentom çaplarıyla fötal sayı arasındaki korelasyonun belirlenmesidir. Bu amaçla çalışmada 25 adet Kıvırcık melezi gebe koyun kullanıldı. Koyunların gebelikleri tohumlamanın 25. gününden itibaren uygulanan transrektal ultrasonografi ile tespit edildi. 30. günde gebelere transabdominal ultrasonografi (6,5 MHz) uygulandı. Bu dönemde plasentomlar "C" şeklinde hipoekojenik alanlar olarak görüntülendi. Gebelikleri boyunca birer hafta arayla her bir koyundan beşer adet plasentom ölçümü yapıldı. Sonuçlar, 5. ve 12. haftalar dışında tekli ve ikiz gebe koyunların plasentom çapları arasında belirgin bir farklılığın olmadığını gösterdi. Tekli ve ikiz gebe koyunların 5. ve 12. hafta ortalama plasentom çapları sırasıyla 6,60 ± 0,56 mm; 8,20 ± 045 mm; 34,02 ± 1,62 mm ve 29,47 ± 0,63 mm olarak belirlendi. İstatistiksel analizlerde minimum önemlilik derecesi $P < 0,05$ olarak kabul edildi. Sonuç olarak, gebe bir koyunda gebeliğin 5. ve 12. haftalarında plasentom çaplarının ölçülmesinin fötal sayının belirlenmesinde yardımcı olabileceği, ancak bu farklılığın açıklanabilmesi için başka çalışmalara da ihtiyaç duyulduğu kanısına varıldı.

Anahtar sözcükler: Gebe koyun, fötal sayı, plasentom çapı

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Introduction

Diagnosing pregnancy and determining fetal number are important tools for organizing appropriate nutritional management of ewes, mating nonpregnant ewes in the same breeding season, reducing the incidence of dystocia, determining fertility problems, and preventing the slaughter of pregnant females (1-4). Various methods, such as radiography (5), abdominal and rectoabdominal palpation (6,7), measurement of plasma progesterone concentration (6,8), assessment of estrone sulfate (9), evaluation of pregnancy proteins (5), and ultrasonography (7) have been used to diagnose pregnancy in ewes. Regarding fetal number, the concentration of serum estrone sulfate (9) and serum progesterone (8,10), radiography (11), Doppler ultrasonography (12), and real time B-mode ultrasonography (4) have been evaluated.

Placentomes are composed of fetal cotyledonary and maternal caruncular microvilli, and they are responsible for maternal-fetal nutrient exchange in ruminants (13). They are easily detected by transrectal ultrasonography on day 32 of gestation and appear as echogenic areas on the surface of the endometrium. On day 42 placentomes are imaged in cross section as cup-shaped hyperechogenic areas (1,3).

Ewes bearing twins require much more energy in late gestation than ewes bearing singletons (14). Considering the placentomes' role in maternal-fetal nutrient exchange, it was postulated that placentome diameters may be different in ewes bearing twins and singletons. The objective of the present study was to investigate the relationship between fetal number and mean placentome diameter in pregnant ewes.

Materials and methods

The experiment was carried out in breeding season. Data were obtained from 25 Kıvırcık mixed breed ewes who had given birth before. They ranged in age from 2.5 to 5 years and weighed between 40 and 60 kg. The ewes became pregnant after synchronization of estrus with intravaginal progestagen-impregnated sponges (30 mg flourogestone acetate, Chronogest, Intervet, Turkey) for 12 days and a single dose i.m. injection of 400 IU PMSG (Chronogest/PMSG, Intervet, Turkey) at sponge withdrawal. The ewes were artificially inseminated with fresh semen 48 h after sponge withdrawal. All the animals were housed at the Faculty of Veterinary Medicine, İstanbul University during their pregnancy. They received routine treatment against internal and external parasites before the study. Ewes were fed with roughage (1 kg/ewe/day) during the pregnancy and concentrate mixture (0.5 kg/ewe/day) was added to the ration during the last 6 weeks of pregnancy and during the lactation period. Trace mineral salt and water were available on an ad libitum basis.

The ultrasonographic scans were carried out using a real time B-mode scanner (Medison SA-600 V, Medison CO, Seoul, Korea) equipped with a 6 MHz linear array transducer and 7.5 MHz micro convex array transducer. On the 25th day after insemination the ewes were checked for pregnancy using the transrectal ultrasonography method, as described by Karen et al. (15). Each ewe was restrained in a standing position, and the transducer was lubricated, introduced into the rectum, and rotated 90° clockwise and 180° anticlockwise in order to image all of the reproductive tract. At the 30th day transabdominal ultrasonography was performed weekly during the remainder of the pregnancy. Diameter sizes of the 5 largest placentomes from each ewe were measured.

The data are expressed as means ± SD. Statistical analysis was performed using independent-sample t test.

Results

A total of 428 ultrasonographic examinations were obtained from 25 ewes between days 25 and 149 after insemination (22 ewes gave birth between days 142 and 146, 3 ewes gave birth at day 149 after insemination). Pregnancy in the ewes was diagnosed by transrectal ultrasonographic imaging of the embryonic vesicles as anechoic areas in front of the urinary bladder (Figure 1). Fetal number can be detected through the same practice when it is possible to detect the embryo and/or placentome as echoic structures in the vesicle (Figure 2). Among the ewes in this study 10 singleton and 15 twin pregnancies were detected, and the accuracy of fetal number detection with transrectal ultrasonography at day 25 after insemination was established as 100% after parturition.

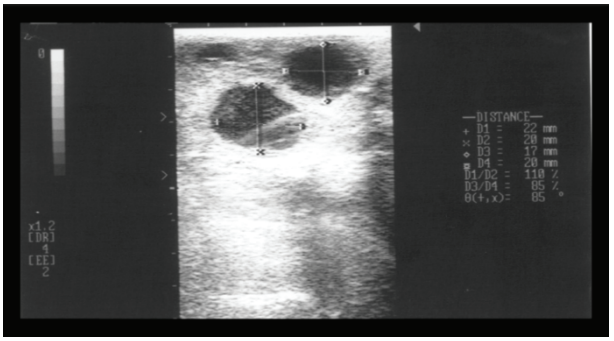

Figure 1. Embrionic vesicles of a twin-bearing ewe at day 25 after insemination.

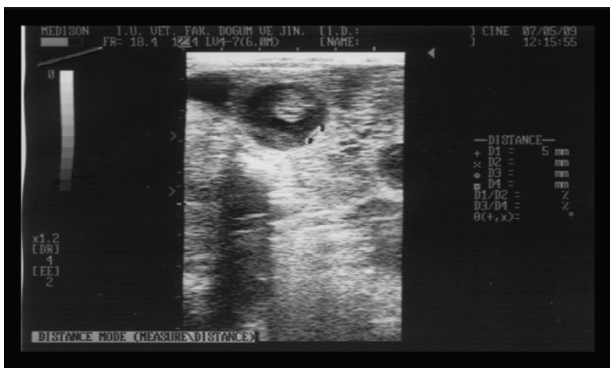

Figure 2. The placentome imaged as an echoic small nodule on the surface of endometrium at day 25 after insemination.

The placentome was easily observed for the first time on day 25 as an echoic structure on the surface of the endometrium. At this term the mean placentome diameter in singleton and twin pregnancies was 5.4 and 5.27 mm, respectively. The mean values of placentome diameters are given in Figure 3. A rapid placentome development was noted between the 5th and 9th weeks of pregnancy in both singleton and twin-bearing ewes. There was no significant change in the placentome size between the 10th and 20th weeks. The maximal placentome size was 34.02 mm in singleton-bearing ewes at the 12th week and 32.48 mm in twin-bearing ewes at the 16th week.

The results showed that there was no significant difference in placentome diameter size in single and twin-pregnant ewes except during the 5th and 12th weeks (Figure 4). The average of placentome diameter size in the singleton and twin-pregnant ewes was, respectively, 6.60 ± 0.56 mm and 8.20 ± 0.45 mm at the 5th week and 34.02 ± 1.62 mm and 29.47 ± 0.63 mm at the 12th week. The minimum

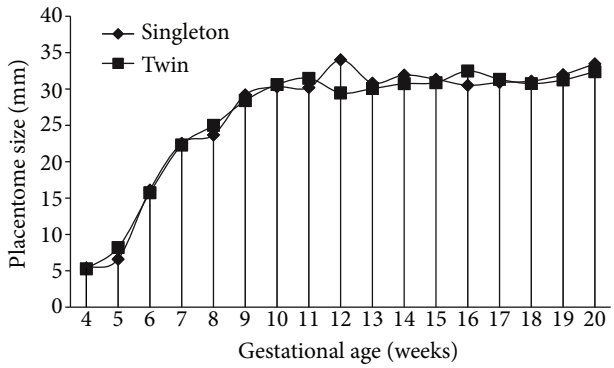

Figure 3. Comparison of placentome size and gestational age in singleton and twin-bearing ewes.

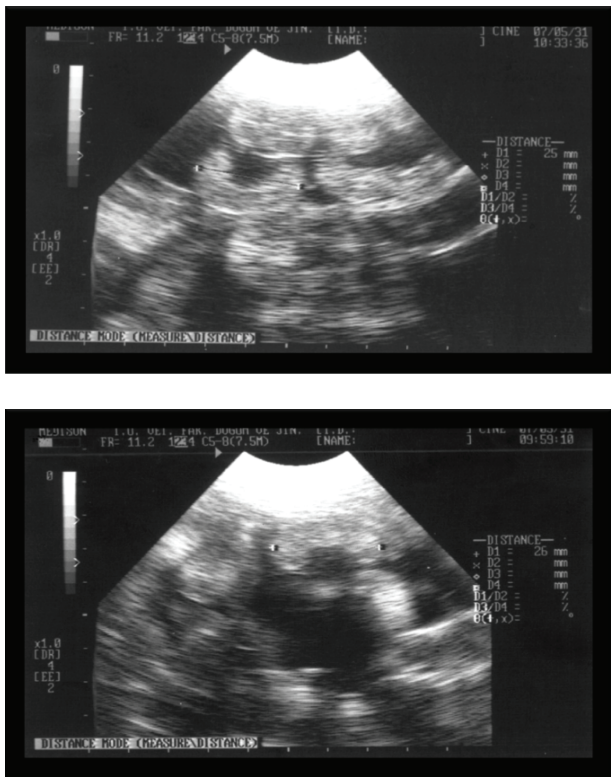

Figure 4. The placentome size of singleton and twin bearing ewes at the 8th week.

significance degree was $P < 0.05$ in statistical analysis, and the significance was $P < 0.05$ and $P < 0.01$ at the 5th and 12th weeks, respectively.

Discussion

There are many studies (1,15-17) about measurement of placentome size, but studies that

evaluate the relationship between the placentome diameter size and fetal number in ewes are rare.

Kaulfuss et al. (18) first detected the placentome on day 27.6 ± 2.4 after mating by transrectal ultrasonography. In the present study the placentome was easily observed for the first time on day 25 as an echoic structure on the surface of the endometrium by using a 6 MHz linear array transducer transrectally. However, Doize et al. (1) reported that the placentomes could be detected at day 32, and Yotov (19) also reported the first detection of placentomes at day 35 by using a 5 MHz transducer. The difference among the studies could be attributed to the effect of the breed and different operators.

A significant increase in the placentome size was noted in both of the groups from the 5th week. Placentomes exhibited slower growth after the 12th week. The maximal size of the placentome was 34.02 ± 0.05 mm in singleton-bearing ewes in the 12th week and 32.48 ± 0.04 mm in twin-bearing ewes in the 16th week. These results are consistent with the results of Doize et al. (1), Aydın et al. (20), and Bhattacharrya et al. (21).

It is known that placental exchange in the ovine placenta is dependent on the number and size of placentomes, which may be influenced by both maternal and fetal factors (22). Thus, larger placentomes are expected in twin pregnancies. The results of the present study, however, do not correspond with this expectation.

Placentome diameter may be a good measure to describe the development of the placenta during pregnancy and the relationship between fetal and placental growth (18). Kaulfuss et al. reported that placentome diameter increased more in twin pregnancies than in singletons during the third month of pregnancy. In the present study the difference between placentome diameter size in singleton and twin-pregnant ewes was significant in the 5th and 12th weeks. This difference was more striking in the 12th week ($P < 0.01$), in line with the findings of Kaulfuss et al. This may be due to the changes associated with increased placental perfusion and tissue permeability during late pregnancy (23). It has been speculated that as placentomes advance in type they differ in vascularity and nutrient transport capacity (24).

In conclusion, ultrasonography is a useful method for determining pregnancy and fetal number. In this study the placentome diameters of twin-pregnant ewes differed from singleton-pregnant ewes in the 5th and 12th weeks of pregnancy. It was determined, however, that placentome diameter size measurement is not a useful method for revealing fetal number in pregnant ewes.

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