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**ABSTRACT**

**Objective:** Various practical methods have been used for pregnancy diagnosis in sheep. Methods of pregnancy diagnosis depending on visualization of the conceptus or determination of its secretory products in the maternal blood or in the milk are the most accurate and specific methods for pregnancy. In 1980, B-mode ultrasonography was introduced in the veterinary field and used for pregnancy diagnosis. Transrectal ultrasonography has been recommended as a simple, rapid and practical method for early pregnancy diagnosis in sheep. **Methods:** 40 clinically healthy Lori-Bakhtiari ewes aged between 2 to 8 years were used in this study. These ewes were estrus synchronized and inseminated and fertile rams were kept with the ewes for natural breeding. Transrectal ultrasonography was carried out in ewes. **Results:** The sensitivity of the test increased when performed at a more advanced stage of pregnancy. The sensitivity of the test increased in young ewes when scanning took place at a more advanced stage of pregnancy.

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**INTRODUCTION**

The intensive sheep management and the wide spread application of the controlled breeding techniques, such as artificial insemination and out-of season breeding; increase the need for an accurate and practical test for early pregnancy diagnosis. The traditional methods such as non-return to estrus and abdominal ballotment are not satisfactory. In addition, laparotomy, laparoscopy, rosette inhibition test and vaginal biopsy are accurate techniques; however these methods are impractical under farm conditions (Goel and Agrawal, 1992; Gordon, 1999). Various methods have been used to diagnose pregnancy in sheep. These methods can be classified as less practical such as the management method (non-return to estrus), abdominal palpation and ballotment, palpation of the caudal uterine artery, laparotomy, peritoneoscopy and rosette inhibition test reviewed by (Ishwar, 1995), and the most practical methods such as radiography, rectal abdominal palpation, hormonal assays, pregnancy protein assays and ultrasonography. Both pregnancy and fetal numbers are accurately diagnosed by using radiography after Day 70 of the gestation. Rectal-abdominal palpation technique detects pregnancy with an accuracy of 70 to 100% from Days 50 to 100 of gestation; however it has a low accuracy for determining multiple fetuses. Progesterone assays have a high sensitivity and a low specificity at Days 16 to 18. Estrone sulphate assay accurately detects pregnant ewe at Days 30 to 35. Transrectal ultrasonography has been recommended as a simple, rapid and practical method for early pregnancy diagnosis in sheep (Buckrell et al. 1986). Real time B-mode ultrasonography provides a simple, rapid, accurate and non-invasive means for ovine pregnancy diagnosis on the farm (Cunningham and Marsh, 1997). Ovine pregnancy (anechoic intrauterine fluid) can be diagnosed by transrectal ultrasonography (5 MHz) as early as Days 17 to 19 of gestation, while the
embryo proper can be imaged between Days 21 to 34 of gestation (Garcia et al. 1993). However, in many breeds this technique requires the ventral part of the abdomen to be shaved, which may be time-consuming and laborious in large-scale farms. During transrectal scanning with a 5 MHz transducer, early pregnancy can be recognized already at Days 17 to 19 by the presence of anechoic fluid in the uterus (Garcia et al. 1993), while the embryo proper and placentomes can be visualized on Days 26 to 28 of gestation (Buckrell et al. 1986). The age and the breed of the ewes and the experience of the operator are among the main factors responsible for these variable results (Buckrell 1988; Bretzlaff et al. 2001).

2. MATERIALS AND METHODS

40 clinically healthy Lori-Bakhtiari ewes aged between 2 to 8 years were selected for this study. These ewes were selected from a commercial farm around Shahrekord. Estrus was synchronized by means of intravaginal sponges containing 30 mg flurogestone acetate for 14 d at the beginning of the breeding season. At the time of sponge removal the ewes were administered chorionic gonadotrophin, and then fertile rams were introduced to flock for mating. Because the exact dates of breeding of the naturally mated ewes were unknown, gestational ages at the time of scanning of these ewes were calculated retrospectively by subtracting the number of days elapsing between scanning and lambing from the average duration of gestation (150 days) of this breed (Karen et al. 2003). Two weeks after insemination those ewes returned to estrus (n=9) were omitted from study.

A real-time ultrasound scanner equipped with a 5 MHz linear transducer (EX8000 Medison ultrasound system) was used for this study. Transrectal scanning were performed on ewes. At the first and second occasions, the ultrasound scanning was carried out on all ewes. There was an interval of 40 days between the first and second occasions. The ewes which had been diagnosed as pregnant were separated (n=25). The ewes had been fasted for 12 h prior to scanning and were restrained in a standing position during scanning in the milking parlor. In addition, the ventral abdominal wall in front of the udder of the ewes was lifted up by the assistant’s hands while conducting the scanning. If necessary the rectum was cleared of feces and the lubricated transducer was gently inserted into the rectum till the anechoic content of the urinary bladder became visible. Then it was rotated clockwise 90° and anti-clockwise 180° to scan the entire reproductive tract (Schrick and Inskeep, 1993). Depending on the stage of pregnancy, the recognition of the allantoic fluid, embryo proper, placentomes, or a fetus were considered as positive signs of pregnancy in both groups. Based on lambing performance of the tested ewes, the results of ultrasonography examinations were arranged as follows: correct positive diagnosis (a), incorrect positive diagnosis (b), correct negative diagnosis (c), and incorrect negative diagnosis (d). From these values the sensitivity (a/a+d x 100), the specificity (c/c+b x 100), the positive predictive value (a/a+b x 100) and the negative predictive value (c/c+d x 100) of the test in Groups 1 and 2 and their subgroups were calculated (Martin et al. 1987).

3. Results

The sensitivity of the test increased when performed at a more advanced stage of pregnancy, reaching a maximum of 63.3 % at Days 31 to 40. Significant differences in the sensitivity of the test were observed between scan periods Days 25 to 30 and Days 31 to 40 (P= 0.003) and between scan periods Days 18 to 25 and Days 31 to 40 (P< 0.0001) and 41 to 50 (P= 0.002).

A total number of 8 false positive pregnancy diagnoses were made during the examinations performed between Days 18 to 50 of gestation. Regarding the effect of the age of the ewes, the sensitivity of the test was significantly higher in young ewes at scan periods Days 18 to 24 and Days 41 to 50 of gestation. In addition, the sensitivity of the test increased in young ewes when scanning took place at a more advanced stage of pregnancy, reaching the maximum at scan period Days 41 to 50. By contrast, in older ewes the sensitivity of the test decreased after Day 40 of gestation.

Discussion

Early detection of pregnancy and determination of the fetal numbers have economical benefits to sheep producers. Non pregnant ewes could be sold, reducing feed expenses, while non-pregnant lambs could be marketed at higher price than they would bring as mature ewes (Gearhart et al. 1988). Separation of the sheep flocks into pregnant and non-pregnant ewes might reduce reproductive and production losses in form of abortions, stillbirths and production of weak lambs (Wani et al. 1998). Predictions of the number of fetuses would allow appropriate nutritional management of the ewes in late gestation that will prevent pregnancy toxemia (Ford, 1983), minimize pre lambing feeding costs, optimize birth weight, weaning weight and survivability of lambs and reduce the incidence of dystocia (Gearhart et al. 1988). The method used for pregnancy diagnosis should be simple, accurate, rapid, inexpensive, practical and safe for both operators and animals. Methods of pregnancy diagnosis depending on visualization of the conceptus or determination of its secretory products in the maternal blood or in the milk are the most accurate and specific methods for pregnancy. In 1980, B-mode ultrasonography was introduced in the veterinary field and used for pregnancy
diagnosis in mare (Palmer and Driancourt, 1980) and then received large acceptance for diagnosing pregnancy in all domestic animals (Kähn, 1992). Transrectal ultrasonography has been recommended as a simple, rapid and practical method for early pregnancy diagnosis in sheep (Buckrell et al. 1986). However, the accuracy of this technique is greatly variable (Gearhart et al. 1988; Garcia et al. 1993; Kaufluss et al. 1996).

References