Ultrasonographic Monitoring of Follicular Development, Ovulation and Corpora lutea Formation in a Bitch

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Abstract: Follicular development, ovulation and corpora lutea formation in a bitch were monitored daily by ultrasonography using a 8-MHz linear transducer. These findings were compared with vaginal cytology and changes in the peripheral serum concentrations of oestradiol-17ß and progesterone. The follicles were identified as anechoic spherical structures on day 6 of procestrus. The numbers of follicles imaged on day 6 of procestrus were 3 on the left ovary and 3 on the right ovary. The average follicular size was 0.67 ± 0.06 cm on the right ovary and 0.48 ± 0.02 cm on the left ovary during the follicular phase. Apparent ovulation was characterised by rapid disappearance of the anechoic antrum in both ovaries within 24 h. This finding corresponded to progressive obliteration of the anechoic region and was characteristic for the postovulatory corpora lutea in both ovaries. Corpora lutea were seen as structures containing anechoic lumen of 3.5-4 mm and thick walls. In conclusion, ultrasonographic findings related to cyclic changes in the ovaries agreed with hormonal and vaginal cytological data on the bitch.

Key Words: Bitch, ultrasonography, ovary, follicle, ovulation, corpora lutea

Bir Köpekte Folliküler Gelişim, Ovulasyon ve Korpora lutea Formasyonunun Ultrasonografi ile İzlenmesi

Özet: Bir köpekte 8 MHz'lik linear prob kullanılarak folliküler gelişim, ovulasyon ve korpora lutea oluşumu ultrasonografik olarak izlendi. Ultrasonografik bulgular, vaginal sitolojik bulgular yanında serum östradiol ve progesteron konsantrasyonlarında meydana gelen değişimlerle karşılaştırıldı. Folliküller proöstrusun 6. günü yuvarlak şekilli anekoik yapılar olarak görüldü. Sağ ve sol ovaryumda üçer adet follikül sayıldı. Sağ ovaryumda saptanan folliküllerin ortalama çapı 0,67 ± 0,06 cm, sol ovaryumda ise 0,48 ± 0,02 cm olarak ölçüldü. Ovulasyon, 24 saat içerisinde her iki ovaryumdaki anekoik foliküllerin hızlı kaybolması ile karakterize oldu. Ovulasyonu takiben her iki ovaryumda da 3,5-4 mm çapında anekoik lumene sahip ve kalın cidarlı (duvarlı) korpora lutea görüldü. Sonuç olarak ovaryumlardaki siklik değişimlerle ilgili ultrasonografik bulgularla hormonal ve sitolojik veriler birbiriyle uygun bulundu.

Anahtar Sözcükler: Dişi köpek, ultrasonografi, ovaryum, follikül, ovulasyon, korpora lutea

In the bitch, it is possible to view ovaries and ovarian structures during the oestrous cycle and anoestrus periods. The prerequisites are an appropriate examination technique and an ultrasound scanner with good resolution. However, it is difficult to view ovaries during the anoestrous period (1).

The literature reveals several studies on the observation of physiological changes during the oestrous cycle of the bitch (follicle development, ovulation, and corpora lutea formation) by ultrasonography (USG).

Studies have investigated whether ovulation, a very important phase in the determination of optimum mating time, can be detected by USG (2-4). The role of USG, a non-invasive and convenient method for imaging of ovulation, has been investigated since successful artificial insemination has been shown to require the detection of ovulation in bitches (5).

The present study aimed to monitor changes in the canine ovary during the follicular phase, ovulation, and luteal phase by USG compared with vaginal cytology and

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hormonal findings, and to determine whether USG with an 8-MHz linear probe can be used to predict the time of ovulation reliably.

Examinations were carried out on a healthy, normally cycling 1-year-old setter bitch. She was housed in the Department of Obstetrics and Gynaecology, Veterinary

Faculty of Ankara University, and had free exercise outside for 5 h daily. In order to keep the intestinal canal as empty as possible during the ultrasonographic examinations, she was not fed before examinations.

Ultrasonography was performed twice daily beginning from day 6 of procestrus (day 6 from the beginning of



Figure 1. Ultrasonographic image of canine ovaries and corresponding vaginal cytology during the follicular phase, ovulation and luteal phase (follicular phase: day –3 to 0; ovulation: day 0; luteal phase: days 1 to 4. USG: ultrasonography; sb: transversal section of small bowel; K: Kidney; white arrow: fluid accumulation in the Bursa ovarica.

vaginal bleeding) until the beginning of cytological dioestrus and then once a day until late dioestrus. Vaginal smears were taken and blood was collected once daily throughout the study. Serum progesterone and oestradiol-17ß were analysed by RIA (Immunotech RIA progesterone and oestradiol kit, Beckman Company, France). Vaginal cells were stained by Papanicolaou stain (6).

Ultrasonography was performed from the left and right shaved flank using a phase array 8.0-MHz linear scanner with a commercial ultrasound gel but without a standoff pad. The dog was placed in either left or right lateral recumbency, or during some examinations she was positioned in dorsal recumbency. Ovarian size (length and width) as well as the numbers and diameter of follicles and corpora lutea were recorded (4).

Both ovaries were imaged throughout the study. The follicles that were located on the cortex uniformly were identified as anechoic spherical structures on day 6 of procestrus (3 days before ovulation; Figure 1, day –3). Numbers of follicles on day 6 of procestrus was 3 on the left and 3 on the right ovary. Average follicular size was 0.67 ± 0.06 cm on the right ovary and 0.48 ± 0.02 cm on the left ovary during the follicular phase. Apparent ovulation was characterised by rapid disappearance of the anechoic antrum in both ovaries within 24 h. Synchronously to ovulation, irregular and anechoic fluid accumulation was observed in the left bursa ovarica (Figure 1, day 0). This finding corresponded to the disappearance of the anechoic region and was characteristic for the postovulatory corpora lutea in both

ovaries. Corpora lutea appeared as structures with anechoic lumen of 3.5-4 mm and surrounding thick wall. They protruded from the surface of the ovaries (Figure 1, days 1 and 4).

From day 6 of procestrus (3 days before ovulation) until day 1 of dicestrus, the average increase in diameter of emerging corpora lutea was significantly related to the mean increase in progesterone concentration.

As the late follicular phase continues until ovulation, vaginal cytology revealed 80%-90% of the cells to be superficial cells with a nucleus (Figure 1, day –3 and 0). After ovulation, most of these cells had lost their nuclei, underwent keratinisation and remnants of degenerated cells progressively increased. Basophilic changes started on day 4 following ovulation. The number of basophilic stained cells increased and on day 5 of ovulation neutrophils were seen to emerge (day 1 of dioestrus).

The progesterone level increased slightly during the last 3 days before ovulation, rose significantly after ovulation and stayed at high levels during the early luteal phase. High oestrogen concentrations sharply decreased to basal levels from the day of ovulation (Figure 2, Table).

In the present study, ultrasonographic findings of the ovaries corresponded well with hormonal and vaginal cytological data. Therefore, USG can be considered a reliable and useful tool for determining ovulation. Furthermore, the 8-MHz linear probe provided effective resolution for the examination of canine ovaries.



Figure 2. Plasma concentrations of progesterone and oestradiol-17ß from day -3 to day 18 (ovulation = day 0).

Diameter of follicles and corpora lutea (cm) Phases of Cycle Right Ovary Left Ovary Length/width (cm) Length/width (cm) Left Right Follicular phase 2.50 ± 0.01/1.39 ± 0.02 $1.37 \pm 0.02/1.08 \pm 0.02$ 0.67 ± 0.06 0.48 ± 0.02 (days -3 to 0) Ovulation 2.02/1.04 1.41/1.13 (days 0) Luteal phase $1.99 \pm 0.01/1.11 \pm 0.03$ $1.50 \pm 0.05/1.12 \pm 0.03$ 0.59 ± 0.01 0.56 ± 0.02 (days 1 to 18)

Table. Ovarian length and width, diameter of follicles and corpora lutea at different oestrous cycle stages in the bitch. Values given are means ± SD.

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