

## **Evaluation of Functional Structures in the Ovaries Pre and Post Ovulation by Doppler Ultrasonography in Bitches**

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Abstract: In this article, functional structures on the ovaries of bitches before and after ovulation are evaluated with Doppler ultrasonography and information concerning the interpretations of findings are transferred. Doppler effect is the first step of Doppler imaging. This effect is the shift in the frequency of the transmitted ultrasound. Spectral and/or color Doppler ultrasonography is the easiest and the most important diagnostic technique in assessing ovarian hemodynamic. The Doppler shift and the Doppler angle are used by the instrument for computing blood velocity. Spectral Doppler contains quantitative information. This information includes the peak systolic volume, end-diastolic volume, and time-averaged maximum velocities, Doppler indices (pulsatility index; PI; resistance index, RI). Doppler indices are used to assess vascular perfusion and computed using values PSV, EDV, and TAMV. The presence of blood flow in a color doppler image is shown as colorencoded and superimposed on a real-time B-mode image. Color Doppler imaging provides detailed monitoring of the local blood flow in ovarian follicles and corpus luteum. The discovery of Doppler ultrasonography that includes blood flow information has led to the formation of research issues in veterinary medicine. It is a developing technology in veterinary medicine. This technology is provided an understanding of hemodynamics and the relationship between blood flow and physiological-pathophysiological processes.

Keywords: Bitch, Corpus luteum, Doppler ultrasonography, Follicle.

## Dişi Köpeklerde Ovulasyon Öncesi ve Sonrasında Ovaryumlardaki Fonksiyonel Yapıların Doppler Ultrasonografi ile Değerlendirilmesi

Öz: Bu derlemede dişi köpeklerde ovulasyon öncesi ve sonrasında ovaryumlardaki fonksiyonel yapıların Doppler ultrasonografi ile değerlendirilmesi ve bulguların yorumlanmasına ilişkin bilgiler verilmiştir. Doppler ultrasonografi, Doppler etkisini kullanan tıbbi ultrasonografidir. Doppler etki, Doppler görüntülemenin ilk adımıdır. Bu etki gönderilen ultrasonun frekansındaki kaymadır. Spektral ve/veya renkli Doppler ultrasonografi, ovaryan hemodinamiğin değerlendirilmesinde en kolay ve en önemli tanı yöntemidir. Spektral Doppler kantitatif bilgiler içerir. Bu bilgiler; maksimum sistolik hacim (PSV), son diyastolik hacim (EDV) ve ortalama maksimum hızları (TAMV), Doppler indekslerini (pulsatil indeks, PI; rezistans indeks, RI) içerir. Doppler indeksleri vasküler perfüzyonun değerlendirilmesinde kullanılır ve hesaplanmalarında PSV, EDV ve TAMV değerlerinden yararlanılır. Renkli Doppler görüntülemede kan akımının varlığı, renklerle kodlandırılmış ve gerçek zamanlı B mod görüntü üstüne eklenmiş olarak gösterilir. Renkli Doppler görüntüleme ovaryan foliküllerin ve korpus luteumun lokal kan akımının detaylı olarak takibini sağlar. Kan akım bilgilerini içeren Doppler ultrasonografinin keşfi veteriner hekimlikte yeni araştırma konularının oluşmasına neden olmuştur. Doppler ultrason, veteriner hekimlikte gelişmekte olan bir teknolojidir. Bu teknoloji ile hemodinamiğin ve kan akımı ile fizyolojik-patofizyolojik süreçler arasındaki ilişkinin anlaşılması sağlanmaktadır.

Anahtar Kelimeler: Dişi Köpek, Doppler Ultrasonografi, Folikül, Korpus Luteum

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#### INTRODUCTION

oppler ultrasonography is an ultrasonographic imaging method used to investigate blood flow velocity and flow characteristics. The first step in determining the aforementioned characteristics is the Doppler effect. Physicist Johann Christian Doppler (1942) expressed the basis of the Doppler effect mathematically by studying the motions of stars. As a result of his observations, the researcher determined that when stars move towards the earth, their light shifts towards blue, which is the expression of a small wavelength (high frequency), and towards red when the earth and stars move away from each other (1-3).

The Doppler principle refers to the change in frequency that occurs when the motion of the turbulent or laminar flow in the vascular structure is detected. In the medical application of this principle, the sound wave is directed to the erythrocytes in motion. Doppler shift is the change in the frequency of the reflected wave caused by the relative movement between the sound wave from the probe and the reflector. The higher the transmitted frequency, the greater the shift in frequency of a given reflector velocity. If the erythrocytes come towards the probe, the rotating frequency increases, and if they move away, it decreases. Doppler effect refers to the shift caused by subtracting the transmitted frequency from the reflected frequency. The sound wave must be parallel to the flow to achieve maximum velocity. If the angle of the sound wave to the reflector exceeds 60 degrees, the velocities cannot be obtained correctly (3-5).

In veterinary ultrasonography, information about all arteries and veins in the body can be obtained with the Doppler blood flow method (6-8). Commonly, when examining the peripheral blood circulation, organs (9-11), transplanted organs, and tumors are used in research (12-14).

Local blood flow can be demonstrated by determining the color according to the mean frequency shifts with color Doppler (15). Color

Doppler allows the assessment of blood flow in vessels that cannot be seen by B-mode imaging (9). Taking advantage of this, it converts the average velocities of blood (erythrocytes) moving in the examined area into a color image. The flow information is placed on the B-mode data, colored according to the direction and velocity of the flow relative to the probe to represent the blood flow. The advantage of adding color to the B-mode is that it allows the examination of the morphological structures of different tissues. On the other hand, it does not change the ultrasonographic finding provided by B-mode. This method was named "Color Duplex Doppler Ultrasonography" because of the color display of the flow in the vein (1,16). Spectral, color, and power Doppler imaging has facilitated the physiological interpretation of hemodynamics. The presence of functional structures (corpus luteum, follicle) in the ovary and the ability to easily measure changes in their quality by computer-aided image analysis and mathematical modeling helped to create new explanations and research topics (7,15).

Spectral or color Doppler imaging is required for a complete evaluation of hemodynamics. Analysis of spectral waveform includes qualitative (current presence, direction, property) and quantitative (velocity, the velocity of blood flow, resistance in the vascular bed, state of the vessel). In Color Doppler Ultrasonography (CDUS), a rapid qualitative evaluation can be made about the flow. Quantitative evaluation of blood flow can be done by spectral analysis. For accurate Pulse Wave Spectral Doppler information, only visible vessels should be considered, the sampling volume should be placed correctly, and the Doppler angle should be less than 60 degrees. Compared with spectral Doppler, it is easier to apply the color Doppler method. In color Doppler ultrasonography, anatomical information, mean blood flow velocity, and direction information can be displayed simultaneously (9,16,17).

# Examination of Ovarian Hemodynamics with Doppler Ultrasonography

Conventional B-mode ultrasonography technique used in direct imaging of cyclic changes in ovarian structures may not provide sufficient information about the functional status and activity of tissues. The ovaries of an adult female dog show cyclical changes. At the same time, changing metabolic conditions occur depending on the adaptation of the local blood supply. Therefore, at the stage where the endocrine activity is at its maximum, the ovary is the organ with the most blood supply in the body. The rapid increase in blood flow increases the supply of oxygen (O2) and necessary chemical elements to the pre-ovulatory follicle. These chemical elements are necessary for ovulation to occur and for progesterone (P4) production to begin (11,18).

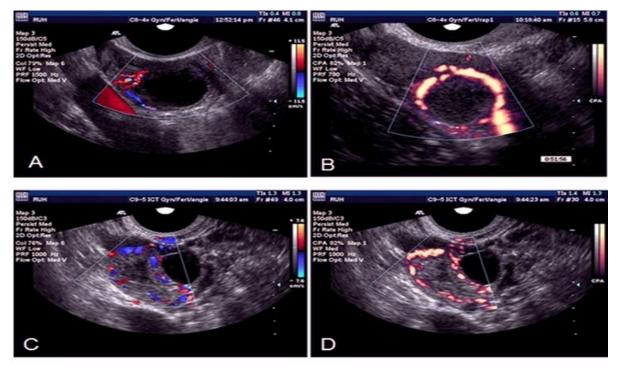
In a study on the oestrus cycle and intraovarian blood flow changes especially during the follicular phase in dogs, it has been shown that ovarian hemodynamic studies are necessary for monitoring the functions of reproductive tissues (18). Following ovulation, angiogenesis and the formation of a normal functional corpus luteum were found important for implantation of the fertile embryo. Continuity of luteal function is associated with the vascular structure (19-22).

Hemodynamic changes in the ovary: They occur during folliculogenesis, ovulation, formation, and regression of the corpus luteum (CL). The dynamic changes that occur during these cyclic events are not fully understood. Physiological and pathological changes in ovarian hemodynamics, the non-invasive nature of Doppler ultrasonography allows direct assessment of the peripheral blood circulation throughout the estrus cycle. Changes in blood flow velocity and direction can be detected with different Other Doppler imaging methods. Doppler parameters such as RI, PI, and systolic/diastolic (S / D) ratios are also used to determine the typical vascular flow. Color Doppler ultrasonography can be used to evaluate ovarian hemodynamics. It is possible to monitor blood flow in the walls of preovulatory follicles or an area restricted within the CL (3,11,15,18,23). In a study, they found that the vascularization in the follicular structures in the ovaries during the oestrus phase is located outside the follicle wall, and a more intense vascularization is observed during the period when the follicle is in the Graaf follicle phase. Also, in the process towards ovulation, after they reported that vascularization and more difficult to detect ovulation the corpus luteum vascularization proceeds inwardly from the wall (24).

Folliculogenesis and ovulation occur due to a functional microcirculatory network (21,23). There may be a gradual increase in intraovarian coloration depending on the number and size of follicles. A rapid increase in intraovarian coloration and an early diastolic notch in the pre-ovulation blood flow curve can be seen in the pre-ovulatory period. High blood flows and low PI and RI values are typical during ovulation and the early luteal period (Table 1). Using Doppler ultrasonography, the vascular ring can be detected around the corpus luteum (Figure 1-C), and when the CL fully develops, vascularity becomes more pronounced, so the resistance to the flow in the vessel decreases. In contrast, during luteal regression, the color image is less and resistance increases, i.e. a gradual decrease in ovarian blood flow can be seen. From the 30<sup>th</sup> day after ovulation, blood flow velocity starts to decrease and Doppler indexes (PI and RI) can increase from the 40<sup>th</sup> day. According to the information obtained from animals that show pregnants and normal cycles, it has been observed that there is a link between ovarian blood flow and progesterone (P<sub>4</sub>) production. Maximum perfusion can be thought to reflect the period in which progesterone is produced in the highest amount. Besides, it is reported that the gradual decrease in progesterone concentration from the 30<sup>th</sup> to the 70<sup>th</sup> day after ovulation is an indicator of physiological luteal regression (15,18,25). It is suggested that these rhythmic changes in blood flow during the estrus cycle occur depending on the temporary estrogen/progesterone ratio in the circulation (26).

**Table 1.** Doppler measurements of left and right intraovarian arteries throughout the estrous cycle (18). **Tablo 1.** Östrus siklusu boyunca sol ve sağ intraovaryan arterlerin Doppler ölçümleri (18).

Days	Ovary	Max. systolic velocity (cm/s)	Max. diastolic velocity (cm / s)	End diastolic velocity (cm / s)	PI	RI
-6	L	13.82±3.00	8.08±2.19	3.86±1.16	1.44±0.27	0.68±0.06
	R	12.34±3.16	7.00±2.12	3.30±1.00	1.48±0.35	0.73±0.07
-2	L	17.32±5.01	11.84±4.07	7.03±3.07	0.99±0.19	0.60±0.06
	R	15.00±1.68	10.33±1.82	5.82±1.62	1.16±0.35	0.64±0.08
0	L	21.09±4.25	15.50±4.07	9.97±2.19	0.85±0.09	0.55±0.03
(ovulation)	R	23.14±6.30	15.87±5.94	10.08±3.68	0.95±0.15	0.57±0.05
2	L	23.89±4.30	16.93±3.74	10.77±1.98	0.87±0.16	0.55±0.06
	R	21.82±3.80	15.56±3.81	9.09±2.12	0.97±0.19	0.58±0.06
5	L	22.03±5.05	15.26±3.57	9.80±2.42	0.85±0.12	0.55±0.05
	R	21.65±6.49	15.06±4.63	8.36±3.49	1.05±0.19	0.62±0.06
10	L	22.70±4.49	17.29±4.53	10.21±2.72	0.82±0.11	0.54±0.05
	R	22.80±6.46	15.05±4.69	9.39±3.77	1.01±0.32	0.59±0.07
30	L	18.15±3.08	12.62±2.21	8.09±1.49	0.89±0.13	0.56±0.04
	R	16.19±5.09	10.59±3.13	6.19±2.23	1.05±0.22	0.61±0.07
50	L	11.99±4.93	7.56±3.15	4.63±2.30	1.10±0.24	0.62±0.06
	R	10.49±4.02	5.99±2.49	3.28±1.47	1.40±0.40	0.69±0.06
70	L	3.80±4.88	2.31±3.03	1.14±1.63	1.37±0.30	0.72±0.10
	R	2.70±4.67	1.69±2.91	0.87±1.63	1.27±0.46	0.68±0.09



**Figure 1.** Color (A) and Power Doppler (B) ultrasound images of preovulatory follicles. Also, sites of ovulation (C, D) demonstrating the blood flow around walls of the collapsed follicle/developing corpus luteum (15). **Şekil 1.** Preovulatör folliküllün Renkli (A) ve Power Doppler (B)'de görüntüleri. Ayrıca ovulasyon alanında (C,D) kollabe olan follikülün veya CL'un etrafında tespit edilen kan akımının görüntüsü (15)

#### CONCLUSION

As a conclusion, Doppler sonography is a useful technique for the non-invasive evaluation of reproductive perfusion in many animals during various stages of the oestrus cycle, pregnancy, and **CDUS** and Pulse puerperium. Doppler Ultrasonography have been proven to be reliable for the detection and diagnosis of physiological and pathophysiological ovarian efficacy. However, the changes in Doppler criteria alone are not sufficient to show the changes in the estrus cycle in female dogs, and it is more accurate to support them with vaginoscopy, cytology, and hormone analysis (especially P<sub>4</sub>). In addition, It may be lead to a new method for the therapy of reproductive disorders.

#### **Conflict of interest**

The author declares that she has no conflict of interest.

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