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# Distribution of Connective Tissue Fibers in the Feline Ovary and Uterus

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Research Article	ABSTRACT
	Connective tissue is significant because it is one of the supporting tissues, it connects organs. In addition, it
History	contains inflammatory response components and provides the environment where metabolic exchange occurs
Received: 25 (10/2022	between cells. Collagen fibers are the most common type of connective tissue component. Type 3 collagen,
Received: 25/10/2022 Accepted: 27/11/2022	glycoproteins, and proteoglycans make up reticulin fibers. They allow the uterus and ovaries to contract and
Accepted. 27/11/2022	stretch since they are situated between the muscles and collagen fibers in these organs. According to the
	requirements of the microenvironment in which they are found, elastic fibers have persistent changeable
	functionalities and are resistant to tensile forces. After regular histological tissue follow-up after washing, tissues
	removed from the ovaries and uterus of cats were fixed in 10% formaldehyde and blocked in the paraplast.
	Sections from the prepared blocks were cut at 5–6 $\mu$ m. The distribution and structure of these fibers were
	studied using the methods of Orsein for the structure of elastic fibers derived from connective tissue fibers, Van Gieson's for the structure of collagen fibers, and Gordon and Sweet's dyeing process for the structure of reticulin
	fibers. It was determined that the ovary and uterus had increased collagen fiber dispersion. Less dispersion was
Constanting	observed in the reticulin and elastic fibers. The collagen fibers in the elastic and reticulin fibers, which were
Copyright	arranged in thick bundles between the follicles in the ovary, were found to have a thinner structure. It was noted
<b>B</b> 08	that the collagen fibers were more thickly distributed in the lamina propria of the uterine endometrium, where
This work is licensed under	the elastic and reticulin fibers were formed from thin filaments.
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International License	Keywords: Cat, Connective tissue fibers, Ovary, Uterine
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# Introduction

Connective tissue has gained significance because it serves as a support tissue, connects organs, preserves their shape, and contains components of the inflammatory response. Connective tissue is the medium through which metabolic exchange between blood and cells occurs. Connective tissue is made up of cells, fibers, and essential substances. (Junqueira and Carneiro 2003). Protein molecules are arranged in a certain way to form the strands of connective tissue. Organelles in the Golgi apparatus and the rough endoplasmic reticulum perform the necessary processes for specialized sequencing (Kirzenbaum, 2006). According to their shape and responses to histological dyes, connective tissue fibers and collagen fibers are categorized as elastic fibers and reticulin fibers, respectively (Ross and Pawlina 2011; Ergün 2016). Elastic fibers are made of elastin protein, while collagen and reticulin fibers are made of collagen protein (Ergün 2016).

The most prevalent component of connective tissue is collagen strands. Collagen, a protein that makes up about 20% of the proteins in the body, is generated from structural elements with stretching and stretching capabilities (Ergün 2016). Depending on the organ and tissue they are in these fibers' density and diameter change (Banks 1986). Under a light microscope, they are visible as wavy structures with varying lengths and widths. This is because they are made up of thinner strands, as revealed by electron microscope tests (Ergün 2016). The diameter of the strands changes in mature and young tissues that have completed their differentiation. They are seen in 15-20 nm in diameter in immature connective tissues and approximately 300 nm in mature connective tissues (Ergün 2016; Junqueira and Carneiro 2003). Collagen fibers have different types due to the collagen molecules' different arrangements (Banks 1986).

Reticulin fibers; They consist of type 3 collagen, glycoproteins and proteoglycans (Kirzenbaum, 2006). They are fibers that are thicker than collagen fibers in diameter and that branch and anastomose. They are located between the muscles and collagen fibers in these organs, giving the uterus and ovaries the ability to contract and stretch (Auersberg et al. 1991). They are demonstrated by special methods originating from the carbohydrate groups in their structure (Kirzenbaum 2006). Fibers with a diameter of 0.5-2  $\mu$ m appear as diffuse webs. Reticulin fibers form flexible networks, especially in organs with periodic volume changes, such as smooth muscle, lymph nodes, endocrine glands, the liver, the uterus, and the small intestine (Kirzenbaum 2006).

Elastic fibers; consist of a specialized arrangement of fine oxyalanine, elaunin, and elastic filaments. This specialized arrangement forms microfibrils and microfiber bundles (Junqueira and Carneiro 2003). Elastic fibers are resistant to tensile and stretching and have durable variable functions according to their microenvironment requirements (Ergün 2016; Junqueira and Carneiro 2003). Therefore, tissues with dense elastic fibers can be called elastic tissues. They are often distinguished in organs such as parts of the penis, elastic arteries, specialized parts of the spine, and auricle (Junqueira and Carneiro, 2003).

Cycles in the ovary and uterus; It is divided into follicular, ovulatory and luteal stages. In the follicular stage, the stages of follicle development are seen in the ovary. In the ovulatory phase, the Graff follicle ovulates. Finally, the luteal stage is the stage in which the corpus luteum and its species are found (Kirzenbaum 2006). Follicular development, ovulation, and corpus luteum developmental stages are controlled by hormones in the pituitary gland, ovary and uterus (Auersberg et al. 1991).

Because of their cyclical characteristics and the fact that cats are accepted as individuals at home, they are frequently desired animals to be researched and their characteristics to be revealed. This study, plans to reveal the distribution of connective tissue fibers of the ovaries and uterus of these animals, which are different due to their cyclic characteristics, and contribute to the literature.

# **Material and Methods**

Ovarian and uterine tissues taken from 6 female cats of different breeds and ages, brought to Sivas Cumhuriyet University Veterinary Faculty Animal Hospital for ovariohisteroctomy in 2022, were used. Ovarian and uterus tissues, obtained with the permission of Sivas Cumhuriyet University Experimental Animals and Local Ethics Committee dated 04.10.2022 and numbered 557, were fixed in 10% formaldehyde, and the tissues were blocked in the paraplast after routine histological tissue follow-up after washing. Sections of 5-6  $\mu$ m thickness were taken from the prepared blocks. Orsein (Bancroft and Stevens 1996, p.55) for the structure of elastic fibers from connective tissue fibers, Van Gieson's (Bancroft and Stevens 1996, p.39) for the structure of collagen fibers, Gordon and Sweet's dyeing technique to determine the structure of reticulin fibers (Bancroft and Stevens 1996, p.49) The distribution and structure of these fibers were determined using.

# Results

When the ovaries taken from cats were evaluated, it was observed that the three types of fibers examined in the ovaries had different densities. It was determined that the collagen fiber distribution was higher (Figure 1 A-B). Elastic fibers and reticulin fibers were less dispersed (Figure 1 C-D). It was observed that the collagen fibers were in the cortex and medulla regions, around the follicles, around the interstitial cells, in the theca externa parts around the follicle, around the corpus luteum and corpus albicans, and around and between the blood vessels in the medulla. It was determined that the elastic fibers were located around the follicles in a thinner structure in the ovary, they were the least in the three types of fibers in terms of distribution of the reticulin fibers, and they were in the form of very thin fibers anastomosing around the follicles and between the follicles.



Figure 1. Representation of collagen, elastic fibers and reticulin fibers in the ovary.
1A: Ovarian medulla collagen fibers, Van Giesson, x10
1B: Ovary, collagen fibers located between follicles, Van Giesson, x20
1C: Elastic fibers between ovaries, follicles and corpus luteum, Orsein, x20
1D: Ovary, follicles between, reticulin fibers, Gordon and Sweet's Silver Stain, x20

When all three layers were evaluated in the uterus in cats, the highest thread distribution was observed in the endometrium due to excess connective tissue. Fibers were seen between the muscles in the myometrium. Collagen fibers were found densely and extensively in the endometrium (Figure 2 A), elastic fibers in the form of thin and small bundles (Figure 2 B), and in fine structures near

crypts and vessels and within the endometrial stroma. It was observed that the reticulin fibers were quite sparse and thin (Figure 2 C), close to the vessels and crypts, and were generally located within the endometrial stroma. It was determined that collagen and elastic fibers were more common in the uterine myometrium between the muscles and around the vessels in the stratum vasculare.



Figure 2. Representation of collagen, elastic fibers and reticulin fibers in uterus
2A: Uterine collagen fibers, Van Giesson, x10
2B: Uterine elastic fibers, Orsein, x20
2C: Uterine reticulin fibers, Gordon and Sweet's Silver Stain, x20

#### Discussion

Ovary in cats; This study was planned because the development of follicles is home to the corpus luteum after ovulation and the connective tissue is responsible for these steps. In our study, it was determined that there were differences in the distribution of connective tissue fibers in different cycle steps of the ovary. It has been observed that collagen fibers are more distributed to elastic fibers and reticulin fibers, and collagen fibers are located between the follicles in the cortex, around the follicles, around the corpus luteum, where they are located in the theca externa layers. The results of the study of Auersberg et al. (1991) and the results of our study were seen to be compatible. Auersberg et al. (1991) reported in their study that in cell culture and ovarian tissue sections, collagen fibers were located around the corpus luteum and interstitial cells. They stated that the increase in the connective tissue and collagen fibers in these regions was higher than the elastic and reticulin fibers (Auersberg et al., 1991). Similar results were seen in the study in cats. In the same study, it was observed that the connective tissue plays an important role in the formation of the corpus luteum in the ovulation region and the formation of a scar tissue in the region after the ovulation process in the transition to the luteal phase. In this study, the fact that the corpus luteum is surrounded by connective tissue at various steps and contains all three types of connective tissue fibers can be explained similarly to the findings of Auersberg et al., (1991). In the regression steps of the corpus boxeum, an increase in connective tissue was also determined in this region. Vielela et al. (2019) emphasized the importance of connective tissue in the ovary of cats and explained that collagen fibers, elastic fibers, and reticulin fibers are located around the follicles at different developmental

stages and in the luteal phase in the ovary. Vielela et al. (2019) stated that connective tissue fibers in cryopreservation and auto-transplantation in the cat ovary are very effective for the success of their procedures. In this study (Vielela et al., 2019) ovarian samples on days 7, 14, 28, 49, 63 were used, and as a result, it was determined that the connective tissue ratio in cryopreservation increased in the cortex region after thawing. It has been determined that the increase in connective tissue can occur as a result of being affected by physiological conditions and changing it with conditions such as cryopreservation, in parallel with the result of our study that ovarian connective tissue fibers are seen at different rates in ovarian follicles at different developmental stages.

Another important organ of the genital system in females is undoubtedly the uterus. Due to the continuity of pregnancy and its physiological and hormonal functions, the uterus was evaluated together with the ovary in cats in this study. Ichijo et al. (1976) reported that the distribution of connective tissue fibers in the uterus changes during pregnancy and non-pregnant period. In this study conducted in cats, it was observed that the distribution of connective tissue fibers in the uterus in different processes varied in the animals examined, and the elastic fibers, where the collagen fibers were more densely located, were located between the muscles and close to the endometrial glands. Ichijo et al. (1976) explained the similarity between the results of the study on connective tissue fibers in the female uterus and the study on the distribution of uterine connective tissue fibers in cats. Comeau and Benhalima (2018) stated in their study on American lobster (Homarus americanus) that the function and distribution ratio of connective tissue fibers in terms of elastic and collagen fibers in the ovary and uterus parts of the fertilization mechanism

changed. In this study, it was thought that the variability of the connective tissue thread distribution in the uterus may be effective in the fertilization mechanism. Ichijo et al. (1976) explained that the reason why collagen fibers make thick bundles and that the uterus is not adversely affected by the effect of ejaculate content. In this study, it was determined that collagen fibers made thick bundles in the uterus and ovary.

### Conclusion

In this study, which was carried out in feline ovary and uterus, the distribution and localization characteristics of connective tissue fibers were tried to be revealed. This histochemical and histological study will contribute to future immunohistochemical studies on this subject.

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