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Research Article

Investigation of immunoglobulin G, lactoferrin and zinc levels in blood sera of calves fed fresh and frozen colostrum

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ABSTRACT

Colostrum has high nutritional values, is more easily digestible than milk, and has a comparatively higher concentration of dry matter, fat and non-fat dry matter, protein and most importantly, immunoglobulin (Ig). The most important task of immunoglobulins is to neutralize pathogens and toxins through neuttalization. Lactoferrin is a protein product of the transferrin gene family with iron binding ability. Lactoferrin serves as a major component of the secondary granules of polymorphonuclear neutrophils and is produced by epithelial cells, including those in the mammary gland. Zinc acts as a cofactor and activator of more than 300 enzymes in different metabolic pathways and is known as a biologically important trace mineral. The aim of this study is to examine whether the immunoglobulin G, lactoferrin and zinc concentrations in colostrum, which is vital for newborn calves, show a decrease tendency by freezing. Fresh colostrum was given to one group (n=12) and frozen (-20 °C) colostrum (n=12) was given to one group and blood samples were collected after 32 hours. According to the results obtained, there was no statistical difference between the groups in the initial measurements of IgG, lactoferrin and zinc values in group 1 (fresh colostrum) and group 2 (frozen colostrum). The differences between the first and second measurements among themselves in both groups were found to be statistically significant. In the second measurements between the two groups, no statistical difference was found between the values of immunoglobulin G (p= 0.996), lactoferrin (p = 0.513), zinc (p = 0.605).

INTRODUCTION

A large part of the dry matter in the composition of colostrum consists of immunoglobulins, and their most important task is to contain maternal antibodies that help protect the newborn against disease pathogens in the first days after birth. Colostrum helps to provide the energy required for the offspring to maintain body temperature and to expel meconium from the digestive system (Koyuncu and Karaca, 2018).

Immunoglobulin G is the main immune component in bovine colostrum and milk, but low concentrations of IgA and IgM are also present (Leyton et al., 2007). IgG is the smallest immunoglobulin group with the highest concentration (70-80 %) in the blood (Diker, 2011).

IgG protects the intestinal mucosa against pathogenic microorganisms and gives passive immunity to the newborn ruminant until his own immune system develops with colostrum. IgG antibodies express multifunctional activities, including complement activation, bacterial opsonization, agglutination, and act by binding to specific sites on the surface of most infectious agents or products by inactivating or reducing infection (Leyton et al., 2007). It is produced from B lymphocytes and plasma cells in secondary lymphoid organs. It is produced most intensively during secondary immune response (Diker, 2011).

Lactoferrin is a member of the transferrin family, including serum transferrin, melanotransferrin, and ovotransferrin. Lactoferrin is a glycoprotein capable of binding iron found in the milk of many species such as cow, human, goat, mare, and mouse. Lactoferrin plays a role in natural immunity, as well as antibacterial, antiviral, antifungal, antiprotozoal, anticarcinogenic, antioxidant, improving bone health, regulating iron absorption in the intestine, immunomodulation, anti-inflammatory, and cell growth control. In addition, it can exhibit quite a lot of biological activity, including the ability to bind and inhibit some bioactive compounds such as glycosaminoglycan and lipopolysaccharide (Yıldırım et al., 2011). Lactoferrin also plays an important role in the immune system defense mechanism. It prevents microorganisms from adhering to the host cell and prevent them from multiplying by colonization or kills them (Legrand et al., 2005).

Zinc (Zn), which has important physiological effects on plants and animals, is one of the essential trace elements (Önder and Yıldız, 2002). Zn is essential for the immune system. It acts by interacting specifically with the components of the immune system, which is a highly proliferative system

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(Chasapis et al., 2012).

The aim of this study was to examine whether the immunoglobulin G, lactoferrin and Zn concentrations in colostrum, which is vital for newborn calves, show a decrease tendency by freezing.

MATERIAL and METHODS

The dams used in this study were Simmental cows in the 3rd lactation, with the same feeding and vaccination program from the same farm. The colostrum given to both groups were measured with a refractometer immediately after collection. Only colostrum with a dry matter of 24-26% were included in the study. The colostrum to be frozen were placed in storage containers and stored at -20 °C. The colostrum was thawed, and the temperature was raised to 37 °C (by dissolving in bain-marie) before feeding the calves from frozen colostrum group. It was provided to the calf within half an hour after birth. Calves in the fresh colostrum group also received colostrum within half an hour after birth. The calves in the study were divided into two groups; 1: those fed fresh colostrum (n=12), 2: those fed frozen colostrum (n=12). Blood samples were taken from all animals in both groups twice, the first before colostrum feeding immediately after birth, and the second after feeding 7.5 liters of colostrum 32 hours after birth. Blood samples from all calves were taken from *Vena jugularis externa* into vacutainers using a disposable sterile syringe. Silicone-based plastic tubes (9 ml) with clot activator were used for serum samples. The collected blood samples were first kept in a portube for 30 minutes to clot. Then, the sera were removed by centrifugation at 4000 rpm for 10 minutes. Serum samples were transferred to Eppendorf tubes (1.5 ml) with the help of automatic pipette. The tubes were stored at -20 °C until serological IgG, lactoferrin and Zn analyses were performed. IgG and lactoferrin values in blood serum were measured by Enzyme-Linked Immunosorbent Assay (ELISA) method. Bovine specific IgG and bovine specific lactoferrin ELISA kits (Biox®, Belgium) were used in the study. In biochemical analysis, Zn values were measured with Gesan Chem 200-1102422® (Italy) autoanalyzer device.

Statistical analysis: The findings obtained were evaluated using IBM SPSS 22.0 for Windows package program. Shapiro-Wilk test was used for the normal distribution of the data. Because of the normal distribution of data, in-group and between-group comparisons were made using two-way analysis of variance in repeated measurements. Multiple comparison tests with benferoni correction were used. Pearson Correlation coefficient was used to analyze the relationship between variables.

RESULTS

In the comparison of the first measurement of lactoferrin value in Group 1 (fresh colostrum) and Group 2 (frozen colostrum), there was no statistically significant difference ($p = 0.914$). In the comparison of the second measurement of lactoferrin value between the groups in Group 1 and Group 2, there was no statistical difference ($p = 0.513$). In group 1, a significant statistical ($p < 0.001$) difference was found in the comparison of the first measurement value of the lactoferrin value and the second measurement value. In Group 2, a significant statistical ($p < 0.001$) difference was found in the comparison of the first measurement value of the lactoferrin value and the second measurement value. (Table 1).

Table 1. Lactoferrin values in calves fed fresh and frozen colostrum.

	Group 1 (n=12)	**Group 2** (n=12)
	$\bar{x} \pm ss$	$\bar{x} \pm ss$
Lactoferrin 1	$328.33196.81^{Aa}$	319.93179^{Aa}
(ug/ml)	209.49 (146.81-664.90)	239.18 (168.16-754.46)
Lactoferrin 2	2375.551392^{Ab}	2525.121181^{Ab}
(ug/ml)	1719.65 (1146.30-4775.78)	1779.79 (558.87-4724.03)

There is statistical difference between columns with different lower-case superscripts ($p < 0.05$).
There is a statistical difference between rows with different upper-case letters ($p < 0.05$).

In the comparison of the first measurement of Zn value between groups, there was no statistical ($p = 0.762$) difference between group 1 and group 2. There was no statistically significant difference ($p = 0.605$) in the comparison of the second measurement of Zn value between groups in Group 1 and Group 2. In group 1, a significant statistical ($p < 0.001$) difference was found in the comparison of the first measurement value of the zinc value and the second measurement value. In Group 2, a significant statistical ($p < 0.001$) difference was found in the comparison of the first measurement value of the zinc value and the second measurement value (Table 2).

In the comparison of the first measurement of immunoglobulin value between the groups in group 1 and group 2, there was no statistically significant difference ($p=0,833$). In Group 1 and Group 2, there was no statistically significant difference ($p = 0.996$) in the comparison of the second measurement of immunoglobulin value between groups. In Group 1,

Table 2. Zinc values in calves fed fresh and frozen colostrum

	Group 1 (n=12)	**Group 2** (n=12)
	$\bar{x}$ ±ss	$\bar{x}$ ±ss
Zinc 1	130.4135.4^{Aa}	125.5841.5^{Aa}
(µg/dL)	138.50 (73.00-184.00)	129.50 (58.00-199.00)
Zinc 2	190.8337.34^{Ab}	182.6638.82^{Ab}
(µg/dL)	199.00 (135.00-257.00)	182.00 (121.00-235.00)

There is statistical difference between columns with different lower-case superscripts ($p < 0.05$).
There is a statistical difference between rows with different upper-case letters ($p < 0.05$).

a significant statistical ($p < 0.001$) difference was found in the comparison of the first measurement value of the Immunoglobulin value and the second measurement value. In Group 2, a significant statistical ($p < 0.001$) difference was found in the comparison of the first measurement value of the Immunoglobulin value and the second measurement value. (Table 3)

A positive correlation was found between lactoferrin and Zn (rho = 0.411; p = 0.046). No relationship was found between immunoglobulin and Zn (rho = -0.315; p =0.134). No relationship was found between lactoferrin and IgG (rho = -0.350; p = 0.094).

Table 3. Immunoglobulin values in calves fed fresh and frozen colostrum

	Group 1(n=12)	**Group 2**(n=12)
Immunoglobulin1	4.891.91^{Aa}	5.082.39 ^{Aa}
(ngr/mL)	4.30 (2.96-8.36)	4.98 (1.56-8.34)
Immunoglobulin2	64.2710.69^{Ab}	64.2415.10^{Ab}
(ngr/mL)	64.02 (44.35-82.96)	70.57 (25.03-80.21)

There is statistical difference between columns with different lower-case superscripts ($p < 0.05$).
There is a statistical difference between rows with different upper-case letters ($p < 0.05$).

DISCUSSION

A quality colostrum allows the development of the defense system against pathogens that newborn calves may encounter in the first days of life. The immune systems of the offspring of animals that do not have high quality colostrum or whose colostrum cannot be used on the grounds that they have disease cannot develop sufficiently. The resulting weakness in the immune system increases the risk of disease and death in calves (Aydoğdu, 2014). Immunoglobulins provide passive immunity until the newborn's own immune system develops and protects the intestinal mucosa against pathogens (Leyton et al., 2007). Immunoglobulins are absorbed by the process of some specialized cells in the small intestine called "pinocytosis". These cells leave their place to basal cells over time (Erdem and Atasever, 2005). This period is limited to the first 24-48 hours after birth (Aydoğdu, 2014).

In a study conducted, serum IgG concentrations of calves receiving fresh colostrum and receiving frozen colostrum were examined and no statistical difference was found between IgG concentrations according to the results of the study (Costa et al., 2017). According to the results of this study, no statistical difference was found between IgG concentrations. In this respect, it is in parallel with our study. However, unlike our study, it has been reported that calves receiving frozen colostrum for 30 days experience severe diarrhea compared to calves receiving fresh colostrum. No diarrhea was found in the calves in both groups in our study up to one month old. In another study, a group of calves were fed colostrum stored at +4 °C and another group was fed with colostrum frozen at -20°C (Holloway et al., 2001). In this study, no statistically significant difference was found between the IgG concentrations in the blood serum of calves fed fresh or frozen colostrum from the same dam. As a result of the study, the suggestion that frozen colostrum can be used as a source of IgG for calves was supported. The results of this study are similar to the results we obtained with IgG values.

In another study, serum IgG values were compared in calves fed fresh colostrum and a commercial colostrum supplement (Holloway et al., 2002). Significantly higher serum IgG concentrations were detected in calves fed fresh colostrum compared to calves fed colostrum supplement. This study showed

that IgG in fresh colostrum is absorbed more efficiently than IgG found in colostrum supplements.

In addition, according to the results of our study, no statistically significant difference was observed in the IgG values in the blood serum of calves fed with fresh and frozen colostrum at the 32nd hour after birth.

Lactoferrin is an iron-binding protein that forms the first defense mechanism against infections and inflammation. Lactoferrin, a multifunctional and important immunoregulatory protein, is a component of breast and lacrimal secretions, seminal and synovial fluids, and plasma and neutrophil granules. Although lactoferrin is present in plasma, its amount is significantly lower than in milk (Lönnerdal and Iyer, 1995). Tsuji et al. (1990) compared the amount of colostrum lactoferrin in cattle with different yield directions in their study. While the average amount of colostrum lactoferrin in dairy cattle was 2 mg/mL, it was found to be 0.5 mg/mL in beef cattle. While the amount of lactoferrin in colostrum is affected by the lactation number of dairy cattle, it has not been reported in beef cattle. Lakritz et al., (2000) compared the IgG and lactoferrin values in the blood serum of calves in two groups fed with pasteurized colostrum and frozen colostrum in their study. As a result of the study, it was revealed that the lactoferrin value in the blood serum of calves receiving pasteurized colostrum was lower than that of calves receiving frozen colostrum. In this study, it was determined that pasteurization method at 76 °C destroyed colostrum proteins. According to the results of our study, no statistically significant difference was observed in the lactoferrin values in the blood serum of calves fed fresh and frozen colostrum 32 hours after birth.

Zn, which is in the structure of many enzymes, is a necessary micro element for normal growth, calf development, and reproductive functions in mature animals (Elmasoğlu, 2008). Any microelement deficiency is seen during the pregnancy of cattle negatively affects the development of the fetus and calf health. It has been proven that microelements cross the placenta and breast barrier. Adequate micro element saturation in pregnant animals has been found to be important for the needs of the young during intrauterine and postnatal development. In addition, microelements also affect the milk and colostrum quality. The concentration of zinc in the blood of newborn calves is significantly higher than that of their dams, which means that the calf organism can accumulate Zn during intrauterine development (Pavlata et al., 2004). Arcagök et al., (2013) investigated the relationship between iron deficiency and blood Zn level in childhood. A statistically significant positive correlation was found between Zn levels and iron levels. This study explains the positive relationship between Zn, which has a high iron affinity, and lactoferrin in our study. It was determined that serum Cu and Zn concentrations were significantly lower in calves with diarrhea compared to healthy calves (Elmasoğlu, 2008).

CONCLUSION

In this study, blood IgG, lactoferrin, and Zn levels in calves fed fresh and frozen colostrum were compared. In the light of the results obtained, the IgG concentration (ngr / ml) in fresh colostrum (64.27 ± 10.69), frozen colostrum (64.24 ± 15.10) (p = 0.996), lactoferrin concentration (ug / ml) in fresh colostrum (2375.55 ± 1392.81), frozen colostrum (2525.12 ± 1181.34) (p = 0.513), Zn concentrations (µg / dL) in fresh colostrum (190.83 ± 37.34), frozen colostrum (182.66 ± 38.82) (p = 0.605) were observed. When the three parameters between the two groups were compared separately, the differences were not found to be statistically significant in any of them. This study demonstrated the potential applicability of colostrum taken from dams who have high quality colostrum and who do not have any health problems in calves of cows that do not have high quality colostrum, or calves that cannot receive their dam's colostrum due to health problems in their mothers, and stored by freezing.

DECLARATIONS

Ethics Approval

This research was carried out on the basis of the permission of Mehmet Akif Ersoy University Local Animal Ethics Committee dated 16.10.2019 and numbered 571.

Conflict of Interest

The authors declare that there have no conflict of interests.

Consent for Publication

Does not need a publication consent.

Author Contributions

In all sections of the final article, each author contributed equally.

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Research Article

A study on behavior and welfare characteristics of Holstein cattle in manure and sand beddings in free-stall barns

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ABSTRACT

The aim of the present study was to investigate the effects of manure and sand bedding on animal welfare and behavioral characteristics in Holstein cattle. A total of 110 Holstein dairy cattle were divided into two groups (manure and sand bedding) with a stall length of 125x250 cm and a walking area. At the end of the study, right and left carpal and tarsal joint lesions were scored. In addition, blood serum glucose, cholesterol, triglyceride and total protein levels were measured. During the study, behavioral characteristics of the animals (duration of standing, duration of lying/resting, number of steps, frequency of standing and lying) were recorded by NEDAP tracking system. Carpal and tarsal lesions of the joints, blood serum glucose, total protein, and triglyceride showed no-significant differences statistically between the groups. However, in terms of the standing time ($P<0.05$), lying/resting time ($P<0.001$), the number of steps ($P<0.001$) and standing/lying ($P<0.001$) frequency behavioural characteristics, neutrophil-lymphocyte ratio ($P<0.01$), and cholesterol levels ($P<0.05$) showed statistically significant differences between the groups. Lying/resting time was determined as 689 minutes in the sand bedding group and 638 minutes in the manure bedding group ($P<0.001$). In conclusion, sand bedding has beneficial effects to increase cow's welfare by increasing the lying/resting time. Lying/resting behavior is important to Holstein dairy cattle, but caution and a full understanding of the context and the variation of the blood parameters in question is needed before drawing firm conclusions about animal welfare from measures of biochemical blood parameters.

INTRODUCTION

Animal behaviors are taken as important criteria when determining the health level of animals. Therefore, changes in behavior and the presence/absence of particular behaviors can provide a valuable indication of the animal's welfare state (Yakan et al., 2007; Akbaş, 2013). The type of bedding used in loose-housing of dairy cows has a considerable effect on the animal comfort and well-being provided by the stalls. According to Endres (2022), Ozbeyaz & Unal (2018), while choosing the-barn floor, it is important to prefer materials which help the control of indoor temperature, and protect the animals from injuries due to ground friction, easy to clean and resistant but free of toxic substances. Additionally, a bedding material should be chosen that will not adversely affect the shelter air quality and the humidity level of the environment (Dimov & Marinov, 2021). According to Kumar Singh (2022), organic bedding materials as manure absorb moisture well, they are compatible with the manure processing systems, easily accessible, available in sufficient quantities in different regions of the world, and are not expensive. Dimov and Marinov (2021) reported that the sand bedding generally doesn't contain nitrogen or carbon required for growth of microorganisms.

In cattle farming, the bedding type should be chosen as dry, safe, comfortable, and durable. In the cases where the bedding is not chosen appropriately the dairy cows undergo stress, and lower productivity leading the cow to be affected negatively in terms of health and behavioral characteristics. According to Leach et al. (2014) the dry, soft, and with consistent quality and quantity bedding resembles the best floor characteristics. Also, for the udder health of the cows that spend most of the day resting, it is essential that the bedding is dry, clean, and soft. Demirci (2005) reported that sand bedding is more advantageous compared to others in terms of being easy to access, having low microorganism load and being the most preferable bedding of cows in hot regions. As cows lay on the ground from a 25-30 cm distance during lying/resting positions, the bedding used in the shelter ground should be soft considering the height of animals' live weight. Otherwise, knee

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abrasions as well as wounds and lesions due to these abrasions may occur on animals (Weary & Tucker, 2003). Kocyigit (2014) reported that the most important reason for laminitis among animals is the behavior of remaining standing for a long time. Tucker et al. (2003) indicated that the lying/resting time for the cows is lower in sand beddings than to those which are provided sawdust beddings. Neutrophile-lymphocyte (N-L) ratio is accepted as an important indicator of stress in small and large ruminants (Broom, 2014). Kayar (2013) stated that it's an important indicator of stress when glucocorticoids increase the blood glucose level. Other blood parameters that are used to evaluate stress in animals are triglyceride, total protein and cholesterol levels (Bedanova et al., 2007).

Rushen et al. (2008) indicated that an increase in a number of the hoof diseases and lameness when there is absence of bedding. The housing of dairy cows only on a concrete floor is one of the main causes of lameness and deterioration of their welfare in general. Rushen et al. (2007) reported that the use of rubber mats contributes to the dairy cows' hooves health and reduces the lameness percentage in the cow herd. Dual chamber mattresses water beds reduce the leg diseases incidence, in particular the appearance of hock joints wounds due to pressure (Kour, 2022). Cows with a compost bedding show reduced incidence of lameness as compared to cows housed in free-stall barns with a sand bedding (Lobeck et al., 2011). Well-managed free-stall production systems farms with a bedding of manure and sand do not differ significantly in terms of hock joint health in cows (Eckelkamp et al., 2016).

As a topic that arises in the last decade, animal wealth has also brought some legislative regulations and applications along with it. Therefore, researches about indoor conditions of shelters, the impact of these conditions over animals, and the reactions the animals show towards these conditions have become very important. This study is carried out with the aim of providing a perspective on effects of manure and sand bedding on health and behavioral features of Holstein cattle. In this manner, it has aimed to measure the effect of manure and sand bedding on Holstein cows' behavioral characteristics such as carpal and tarsal joint lesions, standing and lying/resting time, and number of steps, frequency of standing/lying down and biochemical blood parameters, and N-L ratio.

MATERIALS and METHOD

Animals and Experimental design

The study was performed with the permission of the Aydin Adnan Menderes University Animal Experimentation Local Ethics Committee (No: 13.06.2017-64583101/048). The research was carried out in a 6-month-period between November 2017 and April 2018 at a semi-open free-stall dairy cattle business operating in Foca, İzmir. The sample of the research was derived from 110 dairy cows of the Holstein breed (Black Pied) between the ages of 2-6 being raised in this-farm. The animals were randomly divided into two groups (n=55). The research was carried out in two separate groups which have 50 station stall areas each. The size of the stall dimensions were 125x250 cm for each group. Manure was used on the first sample group as bedding whereas sand was used for the second group. One of the organic beddings, manure, was obtained by drying the stool after passing it through separators. Acquired dry and solid manure was used as dry manure bedding. On the other hand, fine sand with 40-50 cm depth was used in the sand bedding system. With this purpose, the edges of the soil floor were raised and sand was filled into it. The sand used as bedding material was observed daily when the cows were out for milking and lacking places of sand were filled in if there are any. The barn floor on the service road and all other surfaces of the farm is concrete. The manure scrapers only used in sand bedding group. The crawl spaces are made of aggregate insulating concrete at all stalls.

Husbandry

During the research, the animals had exactly the same care and feeding conditions under the care management conditions of the-farm. The animals were fed twice a day at the same time, once at 09:00 and secondly at 16:00. Animals were fed with mixed roughage and concentrate feeds. Feeding was performed as total mixed ration (TMR). The crude protein ratio of the feed was 18% and energy value vas 2900 kcal/kg. Feed and water was offered ad libitum to all cows in both groups during the study. The minimum temperature of the region where the study was performed was between 7.2-14.1 °C and the maximum temperature was 13.1-22.5 °C during the periods of November 2017-April 2018 (Anonyms, 2021).

Sampling and measurements

At the end of the study, carpal and tarsal joint lesions, neutrophil-lymphocyte (N-L) ratio and some biochemical blood parameters (glucose, cholesterol, triglyceride and total protein) were determined in all cows in the groups using manure and sand bedding. Right and left carpal and tarsal joint lesions of all animal groups were determined by scoring method. The scoring system between 0-5 was used for cows' carpal and tarsal joint lesion scoring (Demirci, 2005). In the description: score (0) no lesion observed, (1) bare, pale areas, (2) bare, red areas, (3) occurrence of serum and/or sore scabs, (4) open, infected wounds, (5) disorders/swellings (edema on knees or feet) on carpal/tarsal joints.

During the research, behavioral characteristics of the animals (duration of standing, duration of lying/resting, number of steps, frequency of standing and lying) were followed and recorded on an individual and daily basis using a wristband tracking system, NEDAP Smarttag Leg - Cow Control System, placed on their ankles (Nedap Livestock Management, DC Groenlo, The Netherlands). NEDAP smart wristband system identify each cow and registers the standing, lying, walking and inactive behaviors of all cows 24/7. In this study, data was received from the NEDAP system. The standing and lying/resting time as a minute, number of steps data as a number were recorded separately for two separate groups. And, standing/lying frequency data as a how many times the cow lying down and stands up daily was recorded. Neutrophile-lymphocyte (N-L) ratio and the biochemical blood parameters (glucose, cholesterol, triglyceride and total protein) were evaluated for all animals in both groups. For the N-L ratio, 1 ml of blood sample was taken from cows' jugular vein with EDTA tubes.

After blood smears made of these blood samples were printed with May-Grünwald-Giemsa, there were 100 cells counted on each preparation (O'Loughlin et al., 2011). In order to determine biochemical parameters, 5 ml of blood sample was taken into serum tubes without anticoagulant from animals' jugular veins. After taken blood samples were centrifuged in 4 ºC, 2000x g for 15 minutes, serum parts were separated and were put in Eppendorf tubes while the rest was stored in a –80 ºC fridge after the emergent analysis were completed for further evaluations. Blood glucose, cholesterol, triglyceride and total protein levels were determined using appropriate commercial kits (Archem Diagnostic Ind. Ltd., Istanbul, Turkey) with autoanalyzer (Sinnowa D280, Sinnowa Elec. Technology Co., Nanjing, China).

Statistical analysis

Statistical analysis of the collected data was made using SPSS 22.0 statistical package program (Inc., Chicago, II, USA). In the statistical analysis, significance test of the difference between two means (Student's t-test) was used to evaluate N-L ratio and biochemical blood parameters, and behavioral characters of Holstein cattle. Carpal and tarsal joint lesions score were classified into six scales: score 0, no lesion in the carpal/tarsal joint; good; score 5, disorders/swellings in the carpal/tarsal joint; poor. As the data of carpal and tarsal joint lesions did not meet the assumptions of normality data were analyzed with non-parametric tests. The Mann–Whitney U test was used to determine the effect of manure and sand bedding groups on carpal and tarsal joint lesions data. Mann–Whitney U test is based on the median, not mean (Tai et al., 2022). Thus, the data are presented as the median, minimum, maximum, rank average and rank total. Paired Samples t-Test was used to determine the differences in behavior characteristics among months depending on manure and sand bedding groups.

RESULTS

Carpal and tarsal joint lesions scoring in the manure and sand bedding groups, rank average and total values are provided in Table 1 and Figure 1a, b. Although it is determined that the carpal joint lesion scoring rank average for animals in the sand bedding group was higher than those in manure bedding group with a rank average of 58.39; the effect of bedding on carpal joint lesion was found statistically insignificant. Tarsal joint lesion rank average was determined as 54.00 in the manure bedding group and 57.00 in the sand bedding group.

Table 1. Effects of manure and sand bedding on the carpal and tarsal lesions scores of the joints in Holstein cows

Examined parameters	Group of manure bedding					Group of sand bedding					
	n	Median	Min - Max	Rank average	Rank total	n	Median	Min - Max	Rank average	Rank total	P
Carpal joint	55	0.00	0.00-2.00	52.61	2893.50	55	0.00	0.00-2.00	58.39	3211.50	–
Tarsal joint	55	0.00	0.00-1.00	54.00	3970.00	55	0.00	0.00-1.00	57.00	3135.00	–

n: Number of cows. -: P>0.05.

Average time of standing, time of lying/resting, number of steps and frequency of standing/lying related to manure and bedding groups and standard errors were presented in Table 2. In December, time of standing was identified higher for manure beddings with 288 minutes than in sand bedding group with 285 minutes (P<0.05). In the study, the highest average lying/resting time in bedding groups was identified as 692 minutes in March, in manner groups it was identified as 642 in April. Between November-April period, general average value for time of lying/resting was identified respectively as 638 and 689 minutes in manner and sand bedding groups (P<0.001). During research period, the lowest average number of steps was identified in manure group as 2365 in January, the highest average for number of steps was identified as 2762 in sand group in March. Overall, the average of standing/lying frequency within the study period of November-April was identified as 9.40 and 7.97 in manure and sand bedding groups respectively (P<0.001).

The average N-L ratio for manure group was identified as 1.07 while it was identified as 1.47 in the sand bedding groups (P<0.01). Serum cholesterol level among manure and bedding groups was identified respectively, as 108.04 and 123.62 mg/dL (P<0.05) (Table 3).

DISCUSSION

Carpal and tarsal joint lesions are important indicators for evaluating cow health (Endres, 2022). In the study, the effect of bedding type was found statistically insignificant on carpal and tarsal lesion scores. The highest carpal joint lesions score was found in manure and sand groups as "2", the highest tarsal joint lesion score was found as "1" in manure and sand groups. Similarly, Chaplin et al. (2000) indicated that the relation between bedding groups was not significant in terms of carpal and tarsal joint lesions as well as laminitis. Also, they indicated that having no severe recorded lesions -no carpal and tarsal joint lesions of greater than score "2". The manure and sand bedding reduce the incidence of the tarsal joint lesions in cows (Van Gastelen et al., 2011; Eckelkamp et al., 2016; Dimov & Marinov, 2021). Due to the low carpal and tarsal joint scores in bedding groups, it can be said that the use of sand and manure bedding underlay does not cause a problem in terms of carpal and tarsal joint lesions in Holstein dairy cattle.

Livesely et al. (2002) founded that the incidence of carpal and tarsal joint lesions in Holstein heifers in the mat bedding group was significantly higher than in heifers on straw and mattress bedding. Rushen et al. (2007) stated that the incidence of carpal and tarsal joint lesions was significantly high on dairy cows which were housed on concrete bedding.

Although the overall average standing time values within November-April periods were found significantly close to each other (respectively 287 and 286 minutes in manure and sand bedding groups), the difference between bedding groups

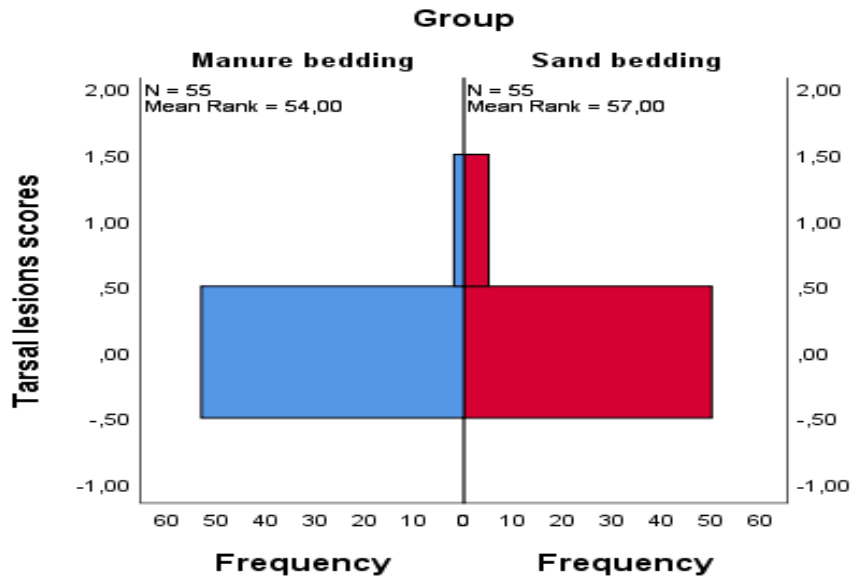

Figure 1. Effects of manure and sand bedding on the tarsal lesions scores of the joints in Holstein cows.

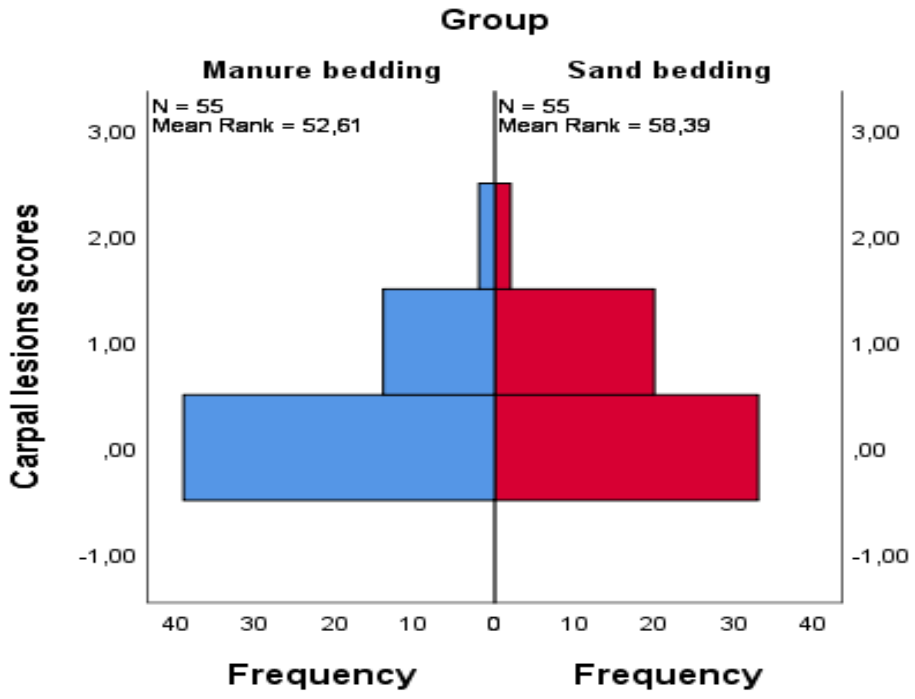

Figure 2. Effects of manure and sand bedding on the carpal lesions scores of the joints in Holstein cows.

were found statistically significant. Although they were small differences, this can only be explained by the fact that the large number of data set in the system obtained may have caused the value differences to be important between the groups which were measured with the NEDAP wristband system that had been following each animal 24/7 for six months. Kocyigit (2014) stated that one of the most important reasons for laminitis in dairy cows is long time of standing. According to Endres (2022), time of standing is used to evaluate cow health. Buyukok (2017) stated that in cases where concrete bedding is

used in dairy cow shelters, animals do not prefer to lie down on these types of beddings and spend most of their time standing. The fact that the overall average standing time in the manure and sand bedding groups were very close to each other in the study which suggests that the bedding was comfortable and the shelter is healthy in terms of animal welfare. The study findings (286 and 287 minutes) are in line with the results of other studies that reports the average standing time being between 0.3-13.0 hours/day (h/d) (Cook et al., 2004; Cook, 2008). The average free-stall cow spends 2.4 h/d standing in an alley socializing, moving between the feed bunk and stalls and returning from the parlor. Once in the stall, the average cow spends 2.9 h/d standing in the stall (Cook et al., 2004). Values determined by this study were found to be higher than the standing time (respectively 3.30 and 2.67 hours in free stall and free system shelters) that Grant (2007) stated in the study,

Table 2. Effects of manure and sand bedding on the carpal and tarsal lesions scores of the joints in Holstein cows

Bedding Groups- Standing Time (minute)						
		Manure Bedding		Sand Bedding		
Period (Month)	n	$\bar{x} \pm S^{\bar{x}}$	n	$\bar{x} \pm S^{\bar{x}}$	t value	P
November	55	287 ± 0.78	55	288 ± 0.80^{x}	-0.153	–
December	55	288 ± 0.80^{a}	55	$285 \pm 0.77^{b,y}$	2.402	*
January	55	287 ± 0.79	55	285 ± 0.79^{y}	1.449	–
February	55	287 ± 0.81	55	$286 \pm 0.80^{x,y}$	1.199	–
March	55	287 ± 0.79	55	$287 \pm 0.80^{x,y}$	-0.055	–
April	55	287 ± 0.79	55	$286 \pm 0.79^{x,y}$	0.876	–
Mean		287 ± 0.32^{a}		286 ± 0.01^{b}	2.324	*
P		-		*	–: P>0.05, *:P<0.05	
Bedding Groups- Lying/Resting Time (minute)						
		Manure Bedding		Sand Bedding		
Period (Month)	n	$\bar{x} \pm S^{\bar{x}}$	n	$\bar{x} \pm S^{\bar{x}}$	t value	P
November	55	634 ± 3.73^{b}	55	691 ± 4.09^{a}	-10.322	***
December	55	639 ± 3.56^{b}	55	684 ± 3.97^{a}	-8.528	***
January	55	638 ± 3.55^{b}	55	687 ± 3.83^{a}	-9.247	***
February	55	635 ± 3.62^{b}	55	690 ± 3.95^{a}	-10.332	***
March	55	638 ± 3.56^{b}	55	692 ± 3.90^{a}	-10.142	***
April	55	642 ± 3.55^{b}	55	689 ± 3.84^{a}	-9.054	***
Mean		638 ± 1.47^{b}		689 ± 1.60^{a}	-23.516	***
P		–		–	–: P>0.05, ***:P<0.001	
Bedding Groups- The Number of Steps						
		Manure Bedding		Sand Bedding		
Period (Month)	n	$\bar{x} \pm S^{\bar{x}}$	n	$\bar{x} \pm S^{\bar{x}}$	t value	P
November	55	2379 ± 15.82^{b}	55	$2746 \pm 10.98^{a,x,y}$	-19.038	***
December	55	2407 ± 20.16^{b}	55	$2744 \pm 10.74^{a,x,}y$	-14.738	***
January	55	2365 ± 8.84^{b}	55	$2741 \pm 10.52^{a,x,y}$	-27.326	***
February	55	2369 ± 9.55^{b}	55	$2737 \pm 10.58^{a,x,y}$	-25.788	***
March	55	2381 ± 9.06^{b}	55	$2762 \pm 10.66^{a,x}$	-27.195	***
April	55	2379 ± 8.98^{b}	55	$2724 \pm 10.61^{a,y}$	-24.866	***
Mean		2380 ± 5.27^{b}		2742 ± 4.36^{a}	-52.921	***
P		–		*	–: P>0.05, ***:P<0.001	

Table 2. cont. Effects of manure and sand bedding on the carpal and tarsal lesions scores of the joints in Holstein cows

	Bedding Groups- Standing/Lying Frequency					
		Manure Bedding		Sand Bedding		
Period (Month)	n	$\bar{x} \pm S^{\bar{x}}$	n	$\bar{x} \pm S^{\bar{x}}$	t value	P
November	55	9.44 ± 0.05^{a}	55	7.93 ± 0.03^{b}	26.555	***
December	55	9.41 ± 0.05^{a}	55	7.96 ± 0.04^{b}	25.441	***
January	55	9.38 ± 0.05^{a}	55	7.99 ± 0.03^{b}	24.394	***
February	55	9.39 ± 0.05^{a}	55	7.98 ± 0.0^{3}b	24.011	***
March	55	9.44 ± 0.05^{a}	55	7.96 ± 0.03^{b}	25.771	***
April	55	9.37 ± 0.04^{a}	55	7.98 ± 0.03^{b}	24.438	***
Mean		9.40 ± 0.02^{a}		7.97 ± 0.02^{b}	61.508	***
P		–		–	–: P>0.05, ***:P<0.001	

Table 3. Neutrophil-to-lymphocyte ratio and serum biochemical levels between groups and standart errors.

	Bedding groups					
		Manure bedding		Sand bedding	t value	P
Examined parameters	n	$\bar{x} \pm S^{\bar{x}}$	n	$\bar{x} \pm S^{\bar{x}}$		
N-L ratio	55	1.07 ± 0.06^{b}	55	1.47 ± 0.10^{a}	-3.217	**
Glucose (mg/dL)	55	45.78 ± 0.70	55	45.49 ± 0.78	0.279	–
Total protein (g/dL)	55	7.39 ± 0.06	55	7.30 ± 0.07	1.012	–
Cholesterol (mg/dL)	55	108.04 ± 4.77^{b}	55	123.62 ± 4.72^{a}	-2.322	*
Triglyceride (mg/dL)	55	24.09 ± 1.15	55	24.36 ± 1.07	-0.173	–

n: Number of animals in the groups. [a,b]: The difference between groups with different letters on the same line in terms of N-L ratio and biochemical blood parameters is statistically significant (P<0.05). *: P<0.05, **: P<0.01, -:P>0.05.

which analyzed the behavioral characteristics of cattle using the same approach. The result of the study for overall average being determined statistically significant among bedding groups in case of standing time during the study period (November-April), is compatible with other researches. According to the Rushen et al. (2007) in the study that was carried out in order to reveal the positive aspects of the soft shelter floor in terms of animal welfare, it was determined that animals had been spending much more time standing on concrete beddings than on rubber beddings. Haley et al. (2001) stated that animals had longer time of standing on concrete beddings while Calamari et al. (2009) stated they had longer time of standing on rubber mat and mattress beddings compared to sand and straw beddings.

In the months of November, December, January, February, March and April, it was determined that the animals in manure bedding group had tended to show longer lying/resting behavior than those in sand bedding group in terms of overall average value. This result can be explained by the fact that the frequency of standing up/lying down in the manure bedding group was higher than that of in the sand bedding group, which may have reduced time to lying/resting in cows. Lying/resting behavior was an important behavior for dairy cows which spend most of their lives in the semi-open barn system, and they spend about 45% of their time lying/resting on a daily basis (Haley et al., 2001). Longer time spent lying/resting behavior prolongs the rumination time, increases blood flow to the mammary gland, and lowers tension on the hoof (Rao et al., 2014). Cows tend to lying/resting, and if for some reason they are obstructed, they subsequently compensate the lying time at the expense of other activities, such as reducing social contact time, faster feeding, and more (Munksgaard et al., 2005).

Average lying/resting time found in study is (respectively 638 and 689 minutes in manure and sand bedding groups), and was compatible with the average values found by the studies of Haley et al. (2001), Cook et al. (2004), Tucker et al. (2021) while it was found a little less than the findings of Calamari et al. (2009) that state dairy cows spend approximately 50% of their days on lying/resting. The time the cow spends lying/resting behavior has been used in a number of studies as a measure of the comfort the resting area provides. Haley et al. (2001), reported that the lying time of lactating dairy cows which use mattress bedding in their beddings were much higher (12.3 h/d) than those with concrete (10.4 h/d). Once in the stall, the average cow spends 11.3 h/day lying in the stall (range 2.8-17.6) on average (Cook et al., 2004). Tucker et al. (2021) reported that the mean daily duration of time spent

lying down is around 11 h/d, but varies among cows from less than 6 h/d to more than 16 h/d. In study, the fact that the 45-50 cm thick sand layer on the suitably compacted soil floor served as a good cushion and provided drainage might have an impact on the result that the time of lying/resting was founded longer in terms of overall average in cows belonging to sand bedding group in all months examined within November-April period. In this case, it can be considered as the sand bedding being more comfortable for animals than manure bedding material. The results were in line with those reported previously by different researchers (Halet et al., 2000; 2001; Manninen et al., 2002; Tucker et al., 2003; Rushen et al., 2007; Koçyiğit, 2014). Tucker et al. (2003) determined the lying/resting time of dairy cows which use sand bedding in their beddings were much longer than those with sawdust. Manninen et al. (2002) found the time of lying/resting on cows which had 2-3 mm sand bedding without straw was much shorter than those with rubber bedding covered with straw and concrete bedding covered with lots of straw. Rushen et al. (2007) reported that the lying/resting time for dairy cows in mat bedding group was much higher than those in concrete bedding. Kocyigit (2014) reported that lying is an important behavior for dairy cows, and they spend respectively 32.57±0.67%, 36.79±0.91% and 37.66±0.68% in concrete, rubber mat and mattress bedding groups of their time lying down on a daily basis. According to the results of the study, it was expected that these differences in bedding groups in terms of lying down/lying time may be affected by conducting the studies under different environmental conditions and analyzing the effects of the diversity and different factors in the bedding types used in the study.

The NEDAP smart bracelet system based on the correct evaluation of the parameters monitored such as health, welfare and aggressive behavior. According to NEDAP-Livestock Management herd records, number of steps for healthy animals is 2500-3000 daily. In the study, it was determined that the number of steps in sand bedding groups were higher than those in manure bedding group in the months of November, December, January, February, March and April. The study results were found in line with the data obtained from NEDAP system average results and they provide information on number of steps which is one of the most important criteria of animal health level. Telezhenko & Bergsten (2005) examined the behavioral characteristics of dairy cows on five different bedding types: grid floor with or without 20 mm thick elastic rubber mattress cow, concrete floor with or without rubber and compacted sand. They found that the cows walk more slowly on the concrete grid floor with much shorter steps than on the sand bedding. It was determined that the cows on the concrete grid bedding took shorter steps than the cows on the sand bedding, however; they did not indicate a significant change in their walking speed. They found that the step lengths of the cows on the rubber bedding were longer than on the concrete grid bedding. As a result, it has been stated that the rubber bedding had a positive effect on the behavior of the cows.

In the study, it was determined that the animals in the group using manure bedding showed more standing/lying frequency compared to those in sand bedding group in the months. Tucker et al. (2003) found the lying frequency in cows which use sand bedding was much lower than those which use sawdust bedding. In the study, the fact that the standing/lying frequency being found higher in the manure group compared to the sand bedding group. This can be explained by the fact that the use of manure bedding increased the restlessness of the cows and the animals show frequent standing and lying behaviors during the day.

Davis et al. (2008) determined that the decrease in lymphocyte level and increase in neutrophil level causes N-L ratio to increase in animals under stress. In the study, N-L ratio in cows belonging to the ones in manure bedding group was found lower than those in sand bedding group. There are no researches found in literature review which evaluates the effect of bedding usage factor on N-L ratio. According to a study carried out by Hong et al. (2019) by examining the effects of stressed Holstein cows in terms of N-L ratio, they found the N-L ratio to be respectively as 2.32 and 1.27 in transported and non-transported cows. The study findings were in line with the results of other studies that reports the N-L ratio being above 1.00 (Hong et al., 2019) and between 0.6-1.4 (Kulberg et al., 2002). In the study, it can be said that administrative factors such as the diameter, dryness, thickness of the sand and how often it is changed should be reviewed considering that the high N-L ratio in the sand bedding group was affected by the stress of the cows in the sand bedding. In addition, the difference in the bedding groups may have been caused by the fact that the N-L ratio of cows was highly affected by some physiological factors such as estrus, pregnancy, lactation period, nutrition and health status, and the treatment of the animal before blood collection. Since biochemical blood parameters were not affected by a single factor, the variation in their measurements was quite wide as they have quantitative properties that can be observed under the influence of multiple factors. According to the studies that were conducted to determine biochemical blood reference levels in cattle, and the values for the range of change were reported as 40-80 mg/dL for glucose level, 80-120 mg/dL for cholesterol level, 15-45 mg/dL for triglyceride level and 6.7-7.5 g/dL for total protein level (Kayar, 2013). It has been observed that the biochemical blood values obtained as a result of the study were compatible with the results reported by Kayar (2013). In this study, bedding type had no significant affect, except for cholesterol level, on all blood parameters. Similarly, Kocyigit (2014) found no influence of bedding type groups (concrete, rubber and mattress) on serum glucose level in Brown Breed Cattle. Besides, he found that the effect of lactation order on glucose level was statistically significant ($P<0.05$). It was determined that serum cholesterol level was higher for the cows reared in sand bedding than those reared in manure bedding, although the difference between bedding groups in terms of serum cholesterol level was not statistically significant. In the light of these findings, it can be said that the biggest problem encountered in terms of blood parameters used to measure stress is the variation seen among animals in reactions to stress.

CONCLUSION

In conclusion, the type of bedding used in loose-housing of dairy cows has a considerable effect on the animal comfort provided by the free-stalls. When choosing bedding, first the comfort that it will provide to the animals must be considered. Results indicated that the use of sand and manure beddings in Holstein dairy cattle breeding is suitable in terms of protection against adverse effects on carpal and tarsal joint lesions. Cow behavior control provides the most precise and complete information about the health, welfare, and location status of individual cows and groups. The duration of lying is a good indicator of the comfort and bedding quality. The cows use sand bedding as compared with the manure bedding, which leads to increasing the length of the lying/resting time up to 689 minutes a day. Sand bedding have been shown to increase cow welfare by increasing the lying/resting time. Also, decreases time of standing and frequency of standing up/lying down behaviors with the sand bedding determine an improve in Holstein dairy cattle comfort. However, the manure bedding resulted in lower stress as indicated by the decreasing N-L ratio in cows. And, the use of manure bedding increased the blood cholesterol level, which is a biochemical parameter. In future studies, stress-related clues can be obtained when working with Elisa kits (such as total antioxidant capacity (TAS), total oxidant status (TOS), Superoxide dismutase (SOD)) directly related to stress. In order to reveal the effects of bedding type on some behavioral and welfare characteristics of cows, it is necessary to increase the knowledge on this subject by conducting new research.

DECLARATION

Ethics Approval

The Animal Care and Practice, Committee of Aydın Adnan Menderes University approved all procedures involved in this experimental study (13.06.2017-64583101/048).

Conflict of Interest

The authors declared that there are no conflicts of interest.

Consent for Publication

Not applicable

Author Contribution

Idea, concept and design: EDF, MA

Data collection and analysis: MA, EDF

Drafting of the manuscript: EDF, MA

Critical review: EDF, MA

Data Availability

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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Research Article

The effect of activity on some milking parameters in Holstein cows

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ABSTRACT

The study was conducted on 41-second lactation Holstein cows of German origin. The shelter type is a semi-open field type and the research period is 12 months. The activities in the first 100 days of lactation per day were 451.4 ± 133.5, and in the second 100 days, it was determined at 420.78 ± 118.0. The activities are divided into 3 parts within 24 hours (at night, during the day between two milkings, and in the evening). While there was no statistical difference between days 100 and 200 of lactation, the lowest activity was recorded at night and the highest activity during the day. Mean daily milk yield was 28.28 ± 3.86 kg for the first 100 days and 25.15 ± 3.61 kg for the following 100 days, and the difference was found to be significant ($P<0.001$). In order to investigate the influence of the daily activity numbers taken into account as factors on the milking parameters, 4 groups were formed for the 100th and 200th days. Accordingly, the effect of activity on milk conductivity, milking duration, and milk yield could not be determined in the first 100 days; its effect on milk flow rate proved significant ($P<0.05$). No effect of activity on milking parameters was observed in 200 days of lactation. It is assumed that the inclusion of the housing type, lactation number, and lactation period as well as the seasonal effect in activity studies in cows will contribute positively to animal welfare in herd management.

INTRODUCTION

The monitoring and control of milk yield, fertility, diseases, and preventive medical services in modern cattle dairy farms can now be carried out with herd management systems integrated into the milking system. Monitoring of cows individually or on a herd basis; provides meaningful and appropriate zootechnical parameters by interpreting the data coming to the system with artificial intelligence. Some activities of a cow during the day are fixed and immutable. The daily time cows spend eating (4.5-5 hours) drinking (0.5 hours) and milking (2.5 hours) is around eight hours. Of these mandatory activities, fewer than 17 hours a day are spent on recreation, social activities, and waiting at stops. The cows spend 2.4 hours returning from milking, going back and forth between feeders and stalls, and social activities. They wait at standing for about 2 hours (Grant, 2006; Cook, 2008). López-Gatiusa et al., (2005) stated that an increase in the animal's movement is one of the signs of estrus and that there is a remarkable change in the physical activity of animals in estrus. In an animal in estrus, the number of steps, head and neck movements increases from 69% to 170% compared to the normal period (Mayo et al., 2019), and success rates between 51% and 81% have been obtained using various mathematical algorithms to estimate heat based on pedometer data (Rolefs et al., 2005). Increase or decrease in activity in cows, lying periods, when live weight is assessed with performance trait data; at the same time, it also contributes to the assessment of the animal welfare level in the herd (Graf, 2017). It has been stated that besides detecting estrus by activity, it is possible to detect metabolic and digestive disorders of animals by monitoring the changes in the number of daily steps (Edwards & Tozer, 2004). Wangler et al., (2005) reported that the activity was higher in the first 60 days of lactation compared to the following 200th day, and the reason for this was due to integration activities when the cow that left the birthplace started to fight again. The same researchers found that activity was related to milk yield; They reported that although activity decreased in cows with a daily milk yield of 15-35 kg, it increased when they produced more than 35 kg. In the study, which examined the influence of the number of steps on heat and rumination behavior in Holstein cows at the University of Bologna, it was stated that the average number of daily steps in the normal period was 589 (Strapak et al., 2021). Shepley et al., (2020) determined the number of daily steps as 1062.2 in summer and 1179.4 in winter in a free roam area with 11.8 m^2 o crawl space per cow. In a study comparing the behavior of cows that go to pasture and cannot go; it has been reported that cows that go to pasture take 2.7 times more steps per day (1506 or 4064) (Dohme-Meier et al., 2014). In a study investigating activities during estrus, the number of steps in 3 days before and after estrus was determined as 891 and 849 (Zebari et al., 2018). In milking systems with modern herd management; Different milking parameters and mechanical parameters are required concerning udder health and milk hygiene. The electrical conductivity, which expresses the measure of the solution that transfers the current between two

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electrolytes, is given as mS/cm (millisiemens per centimeter) (Hillerton & Walton, 1991). Grabowski (2000) stated that the electrical conductivity reference values in cow's milk at 25°C were between 4.83-5.23 mS cm-1. The average milk flow rate value expresses the amount of milk in kilograms (kg) passing through the milking system per minute. The values are used to monitor the milking preparation of the udder and the machine. Sharipov et al., (2020) the study, in which they determined the functional properties and the milk yield of the udder with a milking interval of 10.5-11.99 hours, the milk yield of 24.1 kg, the milking time of 5.68 min, the average milk flow at 1.80 kg/min and the maximum milk flow at 3.14 kg/min. In a study on Italian Holstein cows, the average milk flow rate was found to be 2.48 kg/min in the first 150 days of lactation, 2.36 kg/min in the second 150 days of lactation, and the difference between them was significant. In the same study, milking time was 7.36 minutes in the first 150 days and 6.31 minutes in the following 150 days; the peak electrical conductivities were found to be 6.33 mS/cm and 6.56 mS/cm, emphasizing that the difference is significant (Sandrucci et al., 2020).

In this study, the connection between the activity data of the cows, which are continuously recorded in the herd management system, and the milking data from the milking system (lactation time milk yield, daily milk volume, milking time, milk flow from the udder per minute, conductivity rate of the milk, which is one of the indicators for the Milk quality) evaluations are carried out. In this context; It is aimed to take retrospectively the data stored on the herd management system servers (GEA Systems, Dairy Plan) available in the aforementioned enterprise, and to contribute to the herd health, herd management, and thus to the sustainability of the enterprise more efficiently with possible correlations between these data.

MATERIAL and METHODS

The place where the study was carried out: The study was carried out in January-December 2021 at The study was carried out in January-December 2021 at Burdur Mehmet Akif Ersoy University Agriculture, Food and Livestock Research Application, and Research Center Dairy Farm. The facility is located at an altitude of 1300 meters and has the coordinates 370N'54"N 30019'00"E. The facility is a half-open free-range stall and the milking unit is located in the stall. There is at least one free stall and a feeding table for each animal. The stall bed floors are of two-layer sponge+rubber type, and the feeding and walking areas are made of rubber over concrete. The walking areas are open and the floor is concrete. Manure cleaning is done with a scraper system. There is 7.7 m^2 closed and 9.23 m^2 open area per cow.

Data sources

For the study data set; Information on 41 Holstein cows in the second lactation, all of which were imported from Germany in the same age group (second lactation at the age of 36-46 months), was used. Cows were subjected to the same nutritional conditions in the same shelter in all their physiological periods, and group feeding was not done for dairy cows. The type of diet is ad libitum. The ration contents were prepared as (corn silage: 18 kg, alfalfa 3.75 kg, hay 2 kg, concentrate feed: 8.5 kg, cottonseed meal with 28% protein: 1.7 kg, corn flake: 3.75 kg, vitamin premix: 0.5 kg). Data were collected between January and December 2021. In determining the activity behaviors of cows; Foot type Cow Scout brand pedometers (GEA Systems) are used, which are transmitted to the database with a radio frequency antenna. Activation information is transmitted to the database every 15 minutes, and 12 sets of information are created daily, at 2-hour intervals. Milking data is taken from the Dairy Plan database and integrated into the same system. Milk yields were taken as daily morning and evening (kg), daily average milk flow rate (kg/min), milking duration (min) as daily total, and the value per milking was obtained by dividing it into two. The milk conductivity values were recorded according to weekly averages (mS cm^{-1}). Cows were milked with a 2x6 fishbone milking system. The vacuum pressure of the milking system is between 40-42 kPa. Since all the cows in the farm are in the same age group, they were milked at the same time. Milk conductivity values are formed according to the averages of pre-milking, main milking, and last milking. Data from 41 cows were used for the first 100 days of lactation. The number of cows decreased to 31 in the second 100 days as the data were inadequate as some cows left the farm and were born in autumn.

Statistical analysis

The movement data of the cows were taken as the first 100 and the second 100 days, consisting of 12 data daily, and the data recorded in the MS Excell program were evaluated one by one. In an animal in estrus, the number of steps and head and neck movements increases from 69% to 170% compared to the normal period (Mayo et al., 2019). For this reason, unusual increases and subsequent 18-21-day increases are not included in the data set so that they do not affect the average values. "Minitab 19" program was used for statistical analysis (Minitab, 2019). Descriptive statistics were made for the averages of the activity data, and they were divided into 4 groups as quartiles and classified as a factor. For movement behavior during the day; was divided into 3 slices, between 00:01 night and morning milking, between two milkings, between evening milking and 24:00 o'clock, divided. The significance test of the examined factors was done with a single factor analysis of variance and the Tukey test was applied to compare the subgroups with each other. The Pearson method was used to determine the phenotypic correlations of milking data (Tekin, 2010).

It was approved in accordance with the decision of Burdur Mehmet Akif Ersoy University non-interventional ethics committee no. GO 2021/389.

RESULTS

The number of activities in the first 100 and 200 days of lactation, the average values of the milking data, and the statistical evaluation results are given in Table 1.

As a result of the research, the number of daily activities was determined as 451.4 in the first 100 days of lactation and 420.70 in the second 100 days (Table 1). It was observed that there was a decrease of 30.62 on the 200th day of lactation, but the difference between them was statistically insignificant.

Table 1. The average and statistical evaluation results of activity and milking data in the first 100 and, second 100 days of lactation ($\bar{x} \pm S\bar{x}$).

Lactation 0-100 days			Lactation 100-200 days			
Characteristics	**n**	$\bar{x}$ $S\bar{x}$	**Characteristics**	**n**	$\bar{x}$ $S\bar{x}$	**P**
Activity (hour)	41	451.4±133.5	Activity (hour)	31	420.70±118	**NS**
00:01-08:30	41	80.93±18.54	00:01-08:30	31	74.82±17.21	**NS**
08:31-18:00	41	234.68±88.06	08:31-18:00	31	212.97±7.7	**NS**
18:01-23:59	41	135.84±34.66	18:01-23:59	31	132.93±39.4	**NS**
Conductivity (mS/cm)	41	512.53±32.19	Conductivity (mS/cm)	31	501.19±25.25	**NS**
Milk flow rate (kg/min)	41	2.63±0.51	Milk flow rate (kg/min)	31	2.55±0.56	**NS**
Milking duration (min)	41	6:34±1:37	Milking duration (min)	31	6:09±1:37	**NS**
Daily milk (kg)	41	28.28±3.86 [a]	Daily milk (kg)	31	25.16±3.61 [b]	**0.001 *****

***: P<0.001 NS: Non significant
a,b: Differences between groups, indicated by different letters on the same line, are important

In the activity behaviors during the day, it was determined that the least activity was between 00:01 at night and morning milking (74.28- 80.93), and the highest activity was between two milkings (212.97-234.68). Milk conductivity values were found as 512.53 mS/cm in the first 100 days and 501.19 mS/cm in the second 100 days, and the difference was not statistically significant. The average milk flow rate was determined to be 2.63 kg/min in the first 100 days of lactation and 2.55 kg/min on the 200th day of lactation, the difference between them not being statistically significant. It was found that the milking time was 6:34 minutes in the first 100 days and decreased to 6:09 minutes in the following 100 days, and the difference between them was not statistically significant. The daily milk yields in the first 100 days were found to be 28.28 kg and in the second 100 days, 25.16 kg, and the difference between them was significant (P<0.001).

Statistical assessment results for determining the effect of activity on milking data in the first 100 and 200 days of lactation are given in Table 2 and Table 3.

per ranges of conductivity (mS/cm), flow rate (kg/min), milking duration (min), and daily milk yield (kg) as 497.54–526.29; 2.32-2.94; 5:59-7:33; 26.17-29.35 was determined. In the second 100 days, the values were recorded as 493.56–513.00; 2.46-2.69; 5:59-6:32 and 23,76-27,32 determined. The number of steps in the first 100 and 200 days of lactation for the given parameters was not found to be significant, except for the flow rate in the first 100 days (p<0.05).

The phenotypic correlation coefficients (rp) between milking parameters examined on the first 200th day of lactation are given in Table 4.

Phenotypic correlations performed for the first 200 days of lactation showed a significantly high negative correlation between milking duration and flow rate (P < 0.001). A moderate positive correlation was found between daily milk yield and milking duration (P<0.001).

As a result, it was concluded that the effects of different fa-

Table 2. Statistical evaluation results of determining the effect of activity on milking data on the 100th day of lactation ($\bar{x} \pm S\bar{x}$).

		Lactation 100 days			
Activity (days)	**n**	**Conductivity (mS/cm)**	**Milk flow rate (kg/min)**	**Milking duration (min)**	**Daily milk (kg)**
352,0≤	10	497.54±24.88	2.32±0.42 [b]	7:33±2:07	29.14±3.31
352,0-434,2	11	513.04±20.78	2.58±0.30 [ab]	6:02±0:58	26.17±3.31
434,2-549,0	10	513.22±21.67	2.94±0.51 [a]	5:59±0:49	28.68±4.33
≥549,0	10	526.29±50.76	2.70±0.65 [ab]	6:44±1:52	29.35±4.08
P		NS	*	NS	NS

*: P<0.05 NS: Non significant
a,b: Differences between groups, indicated by different letters on the same line, are important

Table 2 and Table 3 show the effect of activity levels on milking parameters. The first 100 days showed the lower and up-

ctors such as barn type, resting and crawling areas in the barn, herd management preferences, seasonal effects, and lactation

Table 3. Statistical evaluation results to determine the effect of activity on milking data between 100-200 days of lactation.

Lactation 100-200 days					
Activity (days)	**n**	**Conductivity (mS/cm)**	**Milk flow rate (kg/min)**	**Milking duration (min)**	**Daily milk (kg)**
338,1≤	8	499.81±17.47	2.69±0.44	6:03±1:14	27.32±4.33
338,1,-394,7	8	513.00±39.42	2.46±0.58	6:05±1:52	23.76±2.38
394,7-482,4	7	493.56±21.85	2.54±0.73	6:32±2:09	25.51±2.66
≥482,4	8	497.43±15.10	2.50±0.58	5:59±1:24	24.08±4.03
P		NS	NS	NS	NS

NS: Non significant

Table 4. Phenotypic correlation coefficients (rp) between milk conductivity, milk flow rate, milking duration, and daily milk yield in the first 200 days of lactation.

	Conductivity (mS/cm)	**Milk flow rate (kg/min)**	**Milking duration (min)**
Milk flow rate (kg/min)	0.082		
Milking duration (min)	0.064	- 0.759***	
Daily milk (kg)	0.257	0.172	0.422***

***: P<0.001

periods should also be evaluated in studies to determine the effect of activity on milking parameters in dairy cows.

DISCUSSION

The increase in activity during the first 100 days compared to 200 days seen in Table 1 agrees with the data reported by Wangler et al., (2005) reported data. They expressed the efforts of the cow that left the barn to be involved in the class struggle in the first 60 days and increased activity through integration activities. In this study, it was found that the daily activity numbers found (849-1506) were generally lower than those reported in the literature (Dohme-Meier et al., 2014; Zebari et al., 2018; Shepley et al., 2020). These findings are consistent with the findings reporting that the lowest activity during the day in dairy cows occurs between 04-08 in the morning (Rolefs et al., 2005). In a doctoral thesis investigating the effects of manure and sand litters on welfare and behavioral characteristics, the number of daily steps was found to be 2380 in holsteins with manure bases and 2742 in sand litters, and it was stated that the difference was significant (Akdeniz, 2020). The difference in activity numbers between our research and other studies; It is possible to attribute this to the significant impact of housing type and conditions on the number of activities reported (Demir, 2010). Because in this study, shelter conditions and the effects of shelter conditions on animal behavior were not examined. It has been observed that the electrical conductivity and flow rate values of the milk are within the specified reference range (Grabowski, 2000; Sandrucci et al., 2020; Sharipov et al., 2020). In our study it was even higher in the first 100 days of lactation; As reported by Bruckmaier et al., (1995); milking duration at the beginning of lactation was high, but decreased towards the middle and end of lactation. In addition, it was observed that the maximum machine milking time (7.0 kg/min) in Holstein cattle was less than stated by the same researchers. In addition, the maximum machine milking time observed in Holstein cattle was lower (7.0 kg/min) than reported by the same investigators (Table 1). From the beginning of lactation milk production increases and usually peaks between 6-8 weeks and then tends to decrease (Huth, 1995). Similarly, in the second 100 days of lactation in this study, a decrease in daily milk yield was observed that was statistically significant (P<0.001). It has been reported that there is an increase in milk flow rate depending on the increase in milk yield but decreases in milk flow rate in the later stages of lactation (Tilki et al., 2005). Similarly, milk flow rate and, although not significantly, milking time decreased in the second 100 days in this study. It has been reported that there are positive correlations between milk yield and flow rate, and there will be a decrease in milk yield due to a decrease in milk flow rate (Bruckmaier et al., 1995; Huth, 1995; Tilki et al., 2005). In this study, the positive correlation (rp=0.172) between milk flow rate and milk yield indicated in Table 4 was considered as one of the reasons for the decrease in milk yield in the 200 days of lactation.

In Table 2 and Table 3, where the effect of activity on milk parameters is shown, the conductivity of the milk was not affected by the number of steps. We can say that it is compatible with Rossing et al.'s (1987) finding that the variation shows little variation according to the way of breeding, but a strong variation on the individual animal basis and from day to day. The milk flow rate was affected by the number of steps in the first 100 days. The cows included in this study were divided into four different groups according to their activity numbers. It is seen that the cows showing the highest activity among these groups actually reach near-normal activities. In the literature, it is reported that the number of daily activities in the shelter is between 589-1179.4 (Dohme – Meier et al., 2014; Strapak et al., 2021). In Table 2, it is seen that the highest milk yielding group is 549 and above daily activity, and at the same time, the flow rate increases as it approaches the normal activity limits. Although in our study there is a difference depending on the

activity; The seasonal effect can be regarded as the determining influence on the results, because the seasonal influence on the discharge rate was higher, especially for cows calving in the summer between May and August (Rossing et al., 1987). Because in our study, the effect of the month and season was not examined and some cows gave birth in the summer period. It is also possible to attribute this to the individual differences of the cows in the study group. Because Firk et al., (2002) noted that the reproducibility of activity traits varies greatly between and within cows. While the repetition rate of the activity was found 27.4%, the degree in milk parameters was determined as 70-78.7% (Firk et al., 2002). One reason why flow rate is important in the first 100 days is to relate it to milking time. While the highest milking time was detected in the activity range where the flow rate was the lowest, the lowest milking time was determined when the flow rate was the highest. We can say that this is in accordance with the emphasis by Jarshaji and Zülkadir (2019) that the individual differences in milking speed between cows affect the milking duration.

In the first 100 and 200 days, the effect of activity on milking duration and daily milk yield could not be determined. However, although it was not determined as important, it is seen that the milk yields of the groups with less than 352 and more than 549 activities in the first 100 days of lactation were higher. This can be attributed to the fact that low-activity cows engage in less class fighting and integration activities after calving (Wangler et al., 2005). Accordingly, we can say that the cow uses its energy mostly for milk production. Likewise, it was observed that the activity had a positive effect on milk yield in cows whose activity was close to the normal limits and with more than 549 activity (Table 2). Although the milk yield was not significant in the second 100 days of lactation, the highest milk yield was observed in the cows with the least activity (Table 3). We can state that these cows direct their energy to milk yield by moving less as in the first 100 days.

The high negative correlation (-0.759) between milking time and flow rate given in Table 4 was consistent with the literature data (Bruckmaier et al., 1995; Tilki et al., 2005; Edwards et al., 2014). Edwards et al., (2014) found a slightly lower positive correlation between daily milk yield and milking time than our study.

CONCLUSION

As a result, it was determined that the most activity in dairy cows occurred between the bright hours of the day (08:31-18:00), while the least activity was between midnight and morning hours (00:01-08:30). It was observed that milk yield was higher in the first 100 days of lactation compared to the following 100 days. It has been observed that increasing the number of steps in the first 100 days to 549 and above has a positive effect on milk yield. It has been observed that the cows with the least activity in the first 200 days of lactation direct their energy to milk production rather than movement, and therefore milk yields are higher. Studies to be carried out in combination with animal behavior and preferences such as feeding, drinking water, standing, and walking times during the day; It is thought that it will contribute to the development of herd management and sustainable animal husbandry as well as improving animal welfare in farms.

DECLARATIONS

Ethics Approval

It was approved in accordance with the decision of Burdur Mehmet Akif Ersoy University non-interventional ethics committee no. GO 2021/389.

Conflict of Interest

There is no conflict of interest.

Consent for Publication

Does not need a publication consent.

Author contribution

Idea, concept and design: OA, EA

Data collection and analysis: EA, OA

Drafting of the manuscript: OA, EA

Critical review: OA, EA

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Araştırma Makalesi

Farklı işletme koşullarında ineklerin meme başlarında Bovine papillomavirus kaynaklı gelişen siğillerin tedavisinde uygulanan yöntemlerin (otolog aşı ve Newcastle disease virus La Sota suşu) değerlendirilmesi

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Bu makale, Burdur Mehmet Akif Ersoy Üniversitesi Bilimsel Araştırma Projeleri Koordinatörlüğü Doktora Tez Projeleri kapsamında 0556-DR-18 proje numarası ile desteklenmiştir. Makale, Doktora Tez Projesinin bir bölümünden özetlenmiştir.

ÖZ

Burdur-Merkez ve ilçelerinde halk elinde, farklı yapıdaki işletmelerde bulunan 106 adet dişi ineğin bovine papillomavirus (BPV) kaynaklı gelişen meme siğillerinden doku örnekleri toplandı. BPV tip spesifik primerleri açısından PCR testi ile 106 örneğin hepsinde BPV pozitiflik belirlendi. Çalışmada; işletme tipleri, bireysel hayvan bakım ve besleme koşulları, sağım koşulları ve ahır temizliği parametreler yönünden incelenerek değerlendirildi. Ayrıca, meme siğillerinin makroskobik görünümlerine göre (düz ve yuvarlak, pirinç tanesi, saplı-sivri) sınıflandırılmış, BPV tip spesifik primerleri ile pozitiflik tespit edilmiş 30 örnekte iki farklı tedavi uygulama yöntemleri çalışıldı. Birinci grupta tasnif edilmiş 15 hayvana otojen aşı, ikinci grupta tasnif edilmiş 15 hayvana Newcastle disease virus (NDV) La Sota suş aşı uygulamaları yapıldı. Otojen aşı uygulamalarının yapıldığı, farklı makroskobik görünümlere sahip meme siğillerinde %100 gerileme ve iyileşme sağlandı. Newcastle disease virus La Sota aşı uygulamalarının yapıldığı farklı makroskobik görünümlere sahip meme siğillerinde %53.3 gerileme ve iyileşme sağlandı. İneklerde meme siğillerinin tedavisinde otojen aşı uygulamalarının daha etkin sonuçlar verdiği tespit edildi. Ayrıca, çalışmada meme siğillerinin oluşumunda; işletme, bakım, besleme, sağım, ahır temizliği ve diğer faktörlerin de etkili olabileceği sonucuna varıldı.

Evaluation of the methods (autologous vaccine and Newcastle disease virus La sota strain) applied in the treatment of warts caused by bovine papillomavirus on the teats of cows in different management conditions

ABSTRACT

Tissue samples were collected from teat warts caused by Bovine papillomavirus (BPV) from 106 female cows in the the farmers and in different management in Burdur and its county. BPV positivity was determined in all 106 samples by PCR test for BPV type-specific primers. In the study; management types, individual animal care and feeding conditions, milking conditions and cattle-shed cleaning were examined and evaluated in terms of parameters. In addition, two different treatment methods were studied in 30 samples that were classified according to the macroscopic appearance of teat warts (flat and round, rice grain, filiform) and were positive for BPV type-specific primers. Autogenous vaccine was administered to 15 animals classified in the first group and Newcastle disease virus (NDV) La Sota strain vaccine was administered to 15 animals classified in the second group. While 100% regression and recovery was achiev in teat warts with different macroscopic appearances in which autogenous vaccines were applied, 53.3% regression and improvement was achieved in teat warts with different macroscopic appearances in which Newcastle disease virus La Sota vaccine was applied. It was determined that autogenous vaccine applications gave more effective results in the treatment of teat warts in cows. In addition, in the formation of teat warts in the study; It was interpreted that barn, care, feeding, milking, cowshed cleaning and other factors may also be effective.

GİRİŞ

Papillomatozis; tüm memelilerde ve kuşlarda görülen Papillomavirus tarafından oluşturulan bir hastalıktır. Sığırlar evcil hayvanlar içinde en çok etkilenen hayvanlardır (Campo, 2006). PV'ler tarafından oluşturulan doğal enfeksiyonlar genellikle virüsün doku affinitesine bağlı olarak, konak türlerinin deri ya da mukozal membranlarına yerleşerek, deride sıyrık ve açıklıklardan vücuda girmektedir (Lange & Favrot, 2011). Hayvanlar arasında enfeksiyon direkt veya indirekt temas ile gerçekleşir (Emre & Alaçam, 2009). Hayvanın bulunduğu ortamda yaralanmaya neden olan her türlü aletler, dikenli teller, cerrahi yaralar, yularlar, burun halkaları ve yemlikler gibi yaralayıcı etmenler kontaminasyon meydana gelmesine yol açmaktadırlar (Blood ve ark., 1988). BPV kulak küpeleme ve boynuz koter islemleri ya da damar içi enjeksiyon sonrasında kontamine ekipmanlar ile bütünlüğü bozulmuş deriden vücuda girmektedir (Olson, 1986). BPV'ye bağlı papillom lezyonlarının şekillenmesinden sineklerin, ekolojik değişikliklerin, hormonal dengesizliğin beslenme şartlarının (vitamin yetersizlikleri vb.), genetik yapı ve mutasyonların, coğrafi koşullarının ve işletme koşullarının etkili olduğu bildirilmiştir (Lindsey ve ark., 2009; Hamad ve ark., 2016). BPV bulaşmasında, atlar ve sığırlar ara-

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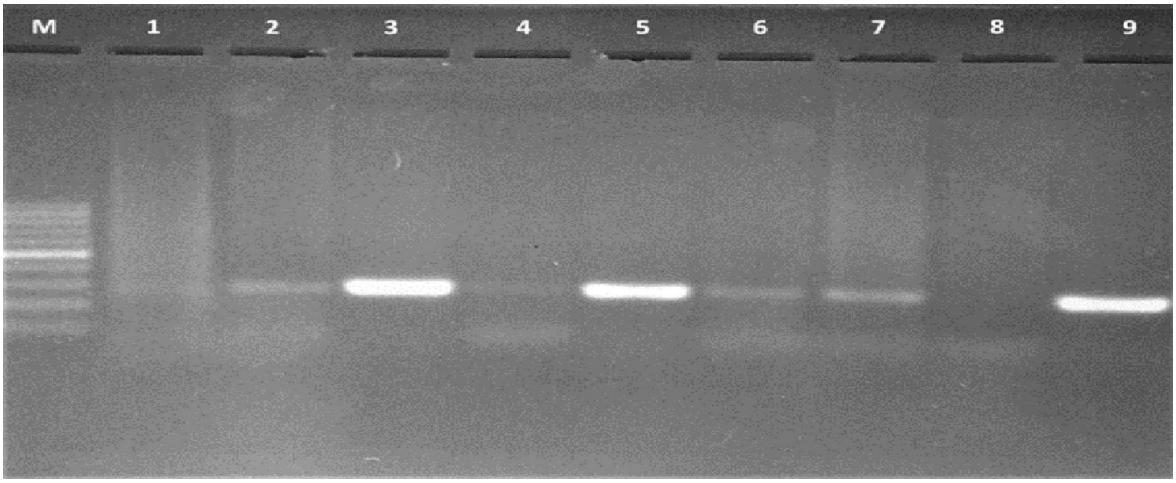

Şekil 1. BPV-9 pozitif örnekler (M: 100 bp; NK: 8; 1, 2, 3, 4, 5, 6, 7, 9: Pozitif örnekler, 264 bp)
Figure 1. BPV-9 positive samples (M: 100 bp; NC: 8; 1, 2, 3, 4, 5, 6, 7, 9: Positive samples, 264 bp)

sında arthropodların vektör olabileceği bildirilmektedir (Nasir & Campo, 2008; Finlay ve ark., 2009). PV lezyonları, süt sığırcılığı yapılan işletmelerde meme ve meme uçlarında meydana gelen papillomlar nedeniyle; mastitis, süt veriminin azalması, meme uçlarının körleşmesi, şekil bozuklukları nedeniyle meme başlarının süt sağım makinalarına girememesi, ağrı nedeniyle sağım yapılamaması ve tüm bunların etkisiyle dolayısıyla büyük ekonomik kayıplara neden olmaktadır. Sığırlarda meme ve meme uçlarında görülen papillomlara, bölgenin hassasiyeti ve papillomların küçük olması nedeniyle veteriner hekimlerce etkin olarak müdahale edilememektedir. Oluşan papillomlar süt verimini etkilemediği, fonksiyon bozukluğuna yol açmadığı, sürüde yaygınlaşmadığı, hayvanların satışının yapılmadığı durumlarda yetiştiriciler ve veteriner hekimler tarafından göz ardı edilmekte ve tedavi yapılmamaktadır. Ancak, her yetiştirici bu siğillerden kurtulmak da istemektedir. BPV kaynaklı enfeksiyonların tedavisinde pek çok yöntem de uygulanmaktadır.

Bu çalışmada, farklı işletme koşullarında ineklerin meme başlarında Bovine papillomavirus kaynaklı gelişen siğillerin tedavisinde kullanılan iki yöntemin (otojen aşı ve NDV La sota aşı suşu) karşılaştırılması ve işletme koşullarının değerlendirilmesi amaçlanmıştır.

GEREÇ ve YÖNTEM

Hayvanlar ve İşletmeler

Burdur-Merkez ve ilçelerinde halk elinde bulunan çeşitli işletmelerden meme başlarında farklı görünümde lezyon bulunan 106 adet dişi inekten siğil doku örnekleri toplandı. Bovine papillomavirus kaynaklı gelişen siğillerin tedavi yöntemlerinin uygulanacağı her biri 15 adet olan iki grup (toplam 30 adet) oluşturuldu. Her grupta bulunan hayvanlarda; 5 adedi düz ve yuvarlak (flat and round), 5 adedi pirinç tanesi (ricegrain, cauliflower) ve 5 adedi saplı-sivri (filiform) olarak sınıflandırıldı. Otojen aşı hazırlanması için de memelerdeki farklı makroskobik görünüme sahip siğillerden örnek alındı.

PCR

DNeasy Blood & Tissue Kit (Qiagen, Germany) kullanılarak meme başı lezyonlarından virus DNA ekstrakte edildi. Silva ve ark. (2016)'nın bildirdiği BPV tip spesifik primerleri (BPV tip 1-14, tip 7 hariç) kullanıldı. Her primer (tipler) Silva ve ark. (2016)'nın bildirmiş olduğu prosedürde kombine edilerek uygulandı. %2 Tris asetat buffer (TAE) agaroz jel elektroforezis ve etidium bromid boyası kullanımı ile amplifikasyon ürünleri gösterildi (Şekil 1).

İşletme Tipleri, Bireysel Hayvan Bakım ve Besleme Koşulları, Sağım Koşulları ve Ahır Temizliği Değerlendirilmesi

Örneklemelerin yapıldığı işletmelerde değerlendirme ve analizlerin yapılabilmesi için anket uygulaması yapıldı. Ankette örneklemelerin yapıldığı İşletmeler Tipine göre: Küçük (1-49 arası hayvan sayısı), Orta (50-100 hayvan sayısı), Büyük (100 üstü hayvan sayısı) ve Açık, Yarı Açık, Kapalı; Hayvan bakım ve besleme koşullarına göre: Kötü, Orta, İyi; Sağım Koşullarına göre: Elle Sağım, Makine ile Sağım; Sağım Temizlik/Dezenfektan Kullanımına göre: Var, Yok; Ahır Temizliğine göre: Kötü, Orta, İyi olarak sınıflandırma gerçekleştirildi.

Otojen Aşı Hazırlama ve Uygulaması

Uygulamanın yapılacağı hayvanlardan 5 gr kadar meme lezyonları alındı. Hunt (1984) adlı araştırmacının belirlemiş olduğu hazırlama protokülüne uygun olarak otojen aşı hazırlandı. Buna göre, papillom örnekleri ilk olarak ılık sabunlu suda yıkandı. Ardından dış ortamda steril bistüri ile petri kaplarında ufak parçalara bölündüler, porselen havana aktarılarak üstleri kaplanacak kadar (1-2 gr) steril kum döküldü. İyice porselende ezilerek, daha sonra fizyolojik tuzlu su ilavesi yapıldı. Elde edilen karışım daha önceden hazırlanmış steril tülbentli beherden süzüldü. Tülbentli bezden alınan süzüntüler, 50 ml'lik santrifüj tüplerine aktarılarak 3000 devirde 30 dakika santrifüj edildiler. Santrifügasyon işlemi sonrasında elde edilen süpernatantlar, bir başka tüpe aktarıldı. Bu tüplerde bulunan süpernatantların üstlerine %40'lık formaldehit solüsyonundan 6 ml aktarılıp, kapakları kapatılarak ters yüz edildiler. Bu son karışım, kapaklar açılarak 37 ˚C'lik etüvde 24 saat bekletildi. Ardından işlem görmüş süpernatantlar, sırasıyla 0.45 nm ve 0.22 nm çaplı enjektör filtrelerden geçirilerek steril aşı viallerine (şişelerine) süzüldüler ve en son bu karışıma antibiyotik ilavesi yapılarak alimunyum vial kapakları flakon crimper yardımıyla kapatıldı. Aşılar vialle-

re konulmadan önce filtrelerden süzülmüş ana stoktan bakteri ve mantar agarlara ekimi yapıldı. Süpernatantların elde edilmesi için 1 hafta inkübasyona bırakıldılar. Kapaklanmış aşı vialleri +4 °C'lik buzdolabında bekletildiler. Uygun sonuç alınan aşı, meme papillomu olan hayvanın ön omuz bölgesinden deri altı 10 ml uygulandı. Uygulama 3 doz olarak 10 gün aralıklarla aynı yerden ve aynı dozda tekrarlandı. Aşı uygulaması yapılan tüm hayvanlarda immun destekleyici preparat kullanılmadı.

NDV La Sota Aşı Suşunun Uygulanması

Uygulamanın yapılacağı hayvanlara ticari bir firmanın üretmiş olduğu NDV La sota aşı suşundan (Hipraviar® S, liyofilize aşı+sulandırma solüsyonu karışımı) hayvanın ön omuz bölgesinden deri altı 1 doz uygulandı. Uygulama 4 doz olarak 7 gün aralıklarla aynı yerden ve aynı dozda tekrarlandı. Aşı uygulaması yapılan tüm hayvanlarda immun destekleyici preparat kullanılmadı.

İstatistiki Analizler

İşletme tipleri, bireysel hayvan bakım ve besleme koşulları, sağım koşulları, ahır temizliği ve tedavi uygulama sonuçları matematiksel (%) olarak değerlendirildi. İşletme tipleri, bireysel hayvan bakım ve besleme koşulları, sağım koşulları ve ahır temizliği karşılaştırmalarında Chi-square testi kullanıldı.

BULGULAR

PCR

BPV tip spesifik primerleri (Tablo 1) olan BPV-1 (L1), 2 (L1), 3 (L1), 4 (E7), 5 (L1), 6 (L1), 8 (L1), 9 (L1), 10 (L1), 11 (L1), 12 (L1), 13 (L1), 13 (E5) ve 14 (L1) kullanıldı. Buna göre, genel olarak 106 örneğin hepsinde BPV pozitiflik bulunurken, hiçbir örnekte BPV-14 pozitiflik tespit edilmedi.

İşletme Tipleri, Bireysel Hayvan Bakım ve Besleme Koşulları, Sağım Koşulları ve Ahır Temizliği Değerlendirme Sonuçları

İşletmeler Tipine göre; en fazla enfekte hayvan Küçük işletmelerde (90 adet; %84.91), en az enfekte hayvan Büyük işletmelerde (6 adet; %5.66), en fazla enfekte hayvan Yarı Açık sistem yetiştirme yapan işletmelerde (56 adet; %52.83), en az enfekte hayvan Kapalı sistem yetiştirme yapan işletmelerde (6 adet; %5.66) belirlendi. Hayvan Bakım ve Besleme Koşullarına göre; en fazla enfekte hayvan Orta seviyedeki işletmelerde (62

Tablo 1. BPV tip spesifik primerler
Table 1. BPV type spesific primers

Primerler	Sekanslar	Fragment Boyutu (bp)	Amplifiye bölge
BPV-1	F-5' GGA GCG CCT GCT AAC TAT AGG 3' R-5' ATC TGT TGT TTG GGT GGT GAC 3'	301	L1 gen
BPV-2	F-5' GTT ATA CCA CCC AAA GAA GAC CCT 3' R-5' CTG GTT GCA ACA GCT CTC TTT CTC 3'	164	L1 gen
BPV-3	F-5' CAG TCA ATT GCA ACT AGA TGC C 3' R-5' GGC TGC TAC TTT CAA AAG TGA 3'	216	L1 gen
BPV-4	F-5' GCT GAC CTT CCA GTC TTA AT 3' R-5' CAG TTT CAA TCT CCT CTT CA 3'	170	E7 gen
BPV-5	F-5' GGC ATG TAG AGG AAT ATA AGC 3' R-5' TTC TCT GAG ATC AAT ATT CC 3'	262	L1 gen
BPV-6	F-5' TTA GAG ACC TGG AAC TTG GG 3' R-5' TAC GCT TTG GCG CTT TTT TGC 3'	294	L1 gen
BPV-8	F-5' TAG AGG ACA CAT ACC GCT TCC AAA GC 3' R-5' TTT GCG AGC ACT GCA GGT GAT CCC 3'	196	L1 gen
BPV-9	F-5' AAA GAG CAA ATC GGG AGC ACC 3' R-5' AAC TAA TGA CCC ACT AGG GCT CC 3'	264	L1 gen
BPV-10	F-5'AAG GCA TTT GTG GTC TCG AGG 3' R-5' CTA AAG AAC CAC TTG GAG TGC C3'	148	L1 gen
BPV-11	F-5' TGC AGA CAC TCA ACC AGG AG 3' R-5' CCA TAA GGG TCG TTG CTC AT 3'	197	L1 gen
BPV-12	F-5' AAA GCT GAA CCA TGC AAA CC 3' R-5' TAA CAA TGT CAA GGG GCA CA 3'	159	L1 gen
BPV-13	F-5' CCA ACC CCA GTA AGC AAG GT 3' R-5' AAG AGG TTG ACC TCG GGA GA 3'	288	L1 gen
BPV-13	F-5' CAC TGC CAT TTG GTG TTC TT 3' R-5'AGC AGT CAA AAT GAT CCC AA 3'	153	E5 gen
BPV-14	F-5'GGA ACA AAC CTC ACA ATC AC 3' R-5'CCA GTT CTC TAA TAC TGA GG 3'	195	L1 gen

Tablo 2. İşletme tipleri, bireysel hayvan bakım ve besleme koşulları, sağım koşulları ve ahır temizliği değerlendirme sonuçları
Table 2. Results of evaluation of barn types, individual animal care and feeding conditions, milking, cowshed cleaning

İşletme Tipi	Küçük	Orta	Büyük
	(1-49 hayvan sayısı)	(50-100 hayvan sayısı)	(>100 hayvan sayısı)
	90 (%84.91)	10 (%9.43)	6 (%5.66)
	Açık	Yarı açık	Kapalı
	44 (%41.51)	56 (%52.83)	6 (%5.66)
Hayvan Bakım- Besleme Koşulları	Kötü	Orta	İyi
	40 (%37.74)	62 (%58.49)	4 (%3.77)
Sağım Koşulları	Elle sağım		Makine Sağım
	15 (%14.15)		91 (%85.85)
Sağım temizlik/ dezenfektan kullanımı	Var		Yok
	76 (%71.70)		30 (%28.30)
Ahır temizliği	Kötü	Orta	İyi
	20 (%18.87)	80 (%75.47)	6 (%5.66)

Şekil 2. Otojen aşı uygulama öncesi ve sonrası (düz ve yuvarlak form)
Figure 2. Before and later of autovaccine (flat and round form)

Şekil 3. Otojen aşı uygulama öncesi ve sonrası, (pirinç tanesi form)
Figure 3. Before and later of autovaccine (ricegrain form

Şekil 4. Otojen aşı uygulama öncesi ve sonrası, (saplı-sivri form)
Figure 4. Before and later of autovaccine (filiform)

Şekil 5. NDV La sota aşı suşu uygulama öncesi ve sonrası, (düz ve yuvarlak form)
Figure 5. Before and later of NDV La sota strain (flat and round form)

Şekil 6. NDV La sota aşı suşu uygulama öncesi ve sonrası, (saplı-sivri form)
Figure 6. Before and later of NDV La sota strain (filiform)

Şekil 7. NDV La sota aşı uygulama öncesi ve sonrası, (pirinç tanesi form)
Figure 7. Before and later of NDV La sota strain (ricegrain form)

adet; %58.49), en az enfekte hayvan İyi seviyedeki işletmelerde (4 adet; %3.77) bulundu. Sağım Koşullarına göre; en fazla enfekte hayvan Makine ile Sağım yapan işletmelerde (91 adet; %85.85) tespit edildi. Sağım Temizlik/Dezenfektan Kullanımına göre; en fazla enfekte hayvan Sağım öncesi-sonrası Temizliğini/Dezenfeksiyonunu yapan işletmelerde (76 adet; %71.70) görüldü. Ahır Temizliğine göre: en fazla enfekte hayvan Orta seviyedeki işletmelerde (80 adet; %75.47), en az enfekte hayvan İyi seviyedeki işletmelerde (6 adet; %5.66) belirlendi (Tablo 2). Örneklemenin yapıldığı işletmelerde çevre koşullarının BPV tiplerinin yayılımında etkisi incelendiğinde; sağım koşullarının (elle veya makine) ve ahırda temizlik/dezenfektan kullanımının (var veya yok) etkili olmadığı belirlendi ($p>0.05$).

Otojen Aşı Uygulama Sonuçları

Meme papillomlarının makroskobik görünümlerine göre; 5 adet düz ve yuvarlak form (BPV-1, BPV-2, BPV-9, BPV-10, BPV-11), 5 adet pirinç tanesi form (BPV-6, BPV-10, BPV-12, BPV-8+BPV-9, BPV-3+BPV-6+BPV-8+BPV-11), 5 adet saplı ve sivri form (BPV-2, BPV-6, BPV-8, BPV-2+BPV-4+-BPV-6, BPV-2+BPV-5+BPV-8+BPV-9+BPV-10+BPV-13) olmak üzere toplam 15 hayvanda otojen aşı uygulamaları yapıldı. Meme lezyonlarından tespit edilen tüm tiplerde otolog aşı uygulaması sonucunda %100 (15/15) başarı sağlandı. Meme papillomlarının makroskobik görünümlerine göre; 5 adet düz ve yuvarlak formda (Şekil 2), 5 adet pirinç tanesi formda (Şekil 3) ve 5 adet saplı-sivri formda (Şekil 4) otojen aşı uygulamaları sonucu %100 gerileme ve iyileşme sağlandı.

NDV La sota aşı suşunun uygulama sonuçları

Meme papillomlarının makroskobik görünümlerine göre; 5 adet düz ve yuvarlak form (BPV-1, BPV-2, BPV-9, BPV-10, BPV-11), 5 adet pirinç tanesi form (BPV-6, BPV-10, BPV-12, BPV-8+BPV-9, BPV-3+BPV-6+BPV-8+BPV-11), 5 adet saplı ve sivri form (BPV-2, BPV-6, BPV-8, BPV-2+BPV-4+BPV-6, BPV-2+BPV-5+BPV-8+BPV-9+BPV-10+BPV-13) olmak üzere toplam 15 hayvanda NDV La sota suşu uygulamaları yapıldı. Meme papillomlarının makroskobik görünümlerine göre; 5 adet düz ve yuvarlak formda %100 (5/5) (Şekil 5), 5 adet saplı-sivri formda %60 (3/5) (Şekil 6) başarı sağlanırken, 5 adet pirinç tanesi formda hiç iyileşme (Şekil 7) tespit edilmedi. Bu uygulama sonucu %53.3 (8/15) iyileşme ve gerileme sağlandı.

TARTIŞMA

Meme bezi, alınan besinleri değerlendirerek süte çeviren ve aynı zamanda onu depo etme özelliğine sahip bir dokudur. İneklerde dört adet meme başı bulunmaktadır. Meme başının en önemli fonksiyonu, üretilen sütün boşaltılmasını sağlamaktır. Buzağıların süt emdiği yerlerdir (Baştan, 2013). Meme papillomatozis olguları sığır sağlığını etkileyen en önemli hastalıklardan biri olup, özellikle meme ve meme başında meydana gelen lezyonlar süt endüstrisinde önemli ekonomik kayıplara yol açmaktadır (Campo, 2003).

Araştırmamızda meme lezyonlarında BPV varlığı en yüksek düzeyde, 1-49 adet hayvan sayısı bulunduran küçük (%84.91) ve yarı-açık (%52.83) işletmelerde belirlendi. Bu tür işletmeler aile tipi olup, çoğunlukla karasinek ve sivrisinek populasyonunun yoğun bulunduğu köy yerleşimlerinde yer almaktadır. Etrafında hayvan dışkıları gübre amaçlı kurutulmaya bırakılmıştır. Ayrıca, göl etrafında konumlanmış köylerde sivrisinek uçuşları oldukça yoğundur. Bu yerleşim bölgelerinde karasinek ve sivrisinek ile kontrol ve mücadele yapılmamaktadır. Karasinek ve sivrisinekler BPV enfeksiyonunun bulaştırılmasında önemli vektörler olarak yer almaktadır (Nasir & Campo, 2008; Finlay ve ark., 2009). Küçük ve orta ölçekli hayvancılık işletmelerinde, sığırların yaşam alanlarında biriken dışkılar çoğunlukla az sıklıkta temizlenmekte, hatta yarı-açık gezinme alanlarında hayvanların dizlerini geçecek boya ulaşabilmektedir. Küçük ve orta düzeyde hayvancılık işletmelerinde ahır tavanları tozlu, örümcek kaplı, duvarlar hayvan dışkısı ile bulaşıktır. Bu araştırmada yer alan işletmelerin genelinde (büyük ticari işletmeler hariç) bilinçli hayvan besleme (dönemsel rasyon hazırlama) yöntemi kullanılmamaktaydı. Çoğunlukla, süt satışı yaptıkları firmalara ait fenni yem tüketimi yapılmaktaydı. Yeterli olamayan beslenme ve hormonal dengesizliklerde immün sistemin yetersizliği sonucu BPV enfeksiyon riskinin artabileceği ifade edilmektedir (Lindsey ve ark. 2009; Hamad ve ark., 2016). Araştırmamızda BPV varlığı hayvan bakım-besleme koşullarının orta düzeyde (%58.49) ve ahır temizliğinin orta düzeyde (%75.47) olduğu işletmelerde en yüksek düzeyde bulundu. BPV enfeksiyonun işletme koşullarında süt sağım makineleri gibi kontamine materyallerle kontakt bulaşma sağlanabileceği bildirilmiştir (Chambers ve ark., 2003). Süt sığırcılığının yapıldığı işletmelerde meme yaralanmaları, meme körleşmesi, mastitis, meme örümceği gibi meme hastalıkları yaygın olarak gelişmektedir. Genellikle küçük ve orta ölçekli işletmelerde süt sağım makineleri kullanılmaktadır. İşletmelerde, sağım öncesi ve sonrası sağım makinelerinde yıkama, temizlik ve dezenfektan uygulaması yapılmaktadır. Memelerde ise genelde sadece yıkama yapılmaktadır. Ancak, en yaygın problem süt sağım makinelerinin düzenli vakum ayarlarının kalibrasyonunun yapılmamasıdır. Sağım makinasındaki problemler (aşırı veya düzensiz vakum seviyeleri, vakum regulatörünün kirliliği, yetersiz vakum pompası kapasitesi, hatalı meme başı kadehi iç lastikleri, hatalı pulzasyon ve sağım makinalarının bakımsızlığı), sağım sürecine bağlı olarak meme başı ve derisinde renk değişiklikleri, ödem, konjesyon, peteşi, hemoraji ve hiperkeratoz gibi sorunlara yol açabilmektedir (Mein ve ark., 2001). Sağım makineleri meme başı travmalarına neden olabilmektedir. Sağım makinesinin çeşitli çalışma sorunları ve uygun olmayan sağım başlıkları meme başı deliği ile meme başı duvarında berelenme ve lezyonların oluşmasına neden olmaktadır. Danimarka'da yapılan çalışmalarda tüm meme enfeksiyonlarının %6.6'sının sağım makinesine bağlı sorunlardan kaynaklanabileceği ifade edilmiştir (Balık, 1998; Özenç, 2000). Araştırmamızda BPV enfeksiyonunun sağımın makine ile yapıldığı (%85.85) ve sağım öncesi ve sonrası temizlik ve dezenfektan kullanımının yapıldığı (%71.70) işletmelerde en yüksek düzeyde olduğu belirlendi. Araştırmacılarında (Lindsey ve ark., 2009) bildirdiği gibi kan, süt, idrar, semen, uterus lavajları ile de BPV enfeksiyonunun bulaşabildiği düşünülürse, işletmelerde enfekte hayvanların yukarıda bahsedilen durumlar dışında da buzdağının görünmeyen kısmının bu yönleri ile de detaylı olarak incelenmesi gerekmektedir.

Çalışmamızda, meme papillomlarının makroskobik görünümlerine göre; 5 adet düz ve yuvarlak formda, 5 adet pirinç tanesi formda ve 5 adet saplı ve sivri formda otojen aşı uygulamaları sonucu %100 gerileme ve iyileşme sağlandı. Oto-

log aşılamalar sonrasında %100 iyileşmenin olduğu bazı olgu raporlarında bildirilmiştir (Ranjan ve ark., 2013; Mayilkumar ve ark., 2014). Ayrıca, Lesnik ve ark. (1999), sığırlarda papillom olgularında otojen aşı uygulaması ile %93.5 ve Sadasiva ve ark. (2000), %80 tedavi başarısı elde etmişlerdir. 1-2 hafta ara ile yapılan iki aşılama uygulama sonrası %80-85 başarı, memelerde düz ve sapsız papillomalarda %33 düzeyinde iyileşme oranı, deri papillomlarında %71 başarı yapılan çalışmalarda bildirilmiştir (Radostits ve ark., 1994; Wadhwa ve ark., 1995; Smith, 1996). Bu durumun tersine, otojen aşıların başarılı olmadığı çalışmalarda rapor edilmiştir (Ndarathi & Mbuthia, 1994). PV enfeksiyonlarının birçok hayvan modelinde nötralizan antikorların yeni enfeksiyon oluşumunu engellediği gösterilmiştir (Galloway, 2003). PV enfeksiyonlarına karşı aşılar Virus Like Partikülleri (VLPs L1 kapsit proteini veya L1+L2) temelinde geliştirilmiştir (Leder ve ark., 2001). VLPs temelinde aşılar devamlı kalıcı oldukları, enfeksiyöz virionların yüzeyinde yüksek immunojenik epitoplar bulundurdukları için tercih edilmektedirler. Araştırmacılar (Prasad ve ark., 1980) otojen aşılamanın sığırların hem kutan hem de meme papillomlarında etkili olduğunu bildirmelerine rağmen, meme papillomlarına yönelik hazırlanan otojen aşılarda sağlanan tedavi başarısının, deri papillomlarında sağlanan başarılardan düşük olduğunu ifade etmişlerdir. Bunun nedeni, immunitenin oluşmasından sorumlu viral kapsit antijenlerin atipik meme papillomlarında, tipik fibropapillomlara göre daha düşük konsantrasyonda bulunmasına bağlanmıştır (Jana, 2015). Keza, memelerde histopatolojik bulgularda papillom olgusu, vücut papillomlarında fibropapillom olguları sıkça görülmektedir. Ticari amaçla hazırlanmış papillom aşılarının nadiren papillom lezyonlarını gerilettiği veya yeni lezyonlardan korumada etkili olabileceği bildirilmiştir. Ancak bu durumun, dokularda suş benzerliği var olduğu durumlarda gerçekleşebileceğini açıklamışlardır (Campo, 1991; Scott & Anderson, 1992; Smith, 1996). Bir tek hayvandan alınarak hazırlanan aşının tüm enfekte hayvanları iyileştiremediği, tüm hayvanlardan alınan ve miks hazırlanan aşıların daha iyi sonuçlar verdiği birçok çalışma ile ortaya konmuştur (Ndarathi & Mbuthia, 1994; Thaiya ve ark., 2009). BPV aşılarının etkinliği; virus tipi, papillomun gelişim dönemi, papillom dokusunun toplama şekli, miktarı, aşı hazırlama tekniği, aşı uygulama takvimi ve hayvanın immunitesine bağlı değişkenlik gösterebilmektedir (Ranjan ve ark., 2013; Jana, 2015).

Bu çalışmada, NDV La Sota suşu uygulamaları sonucunda meme papillomlarının makroskobik görünümlerine göre; 5 adet düz ve yuvarlak formda %100 (5/5), 5 adet saplı ve sivri formda %60 (3/5) başarı sağlanırken, 5 adet pirinç tanesi formda tam iyileşme ve gerileme tespit edilmedi. Bu çalışma sonucunda toplamda %53.3 iyileşme ve gerileme sağlandı. Avki ve ark. (2003), NDV'nin immunstimulan etkilerini analiz etmek için 14 ineğe subkutan NDV'nin La Sota suşu uygulamışlardır. Araştırmanın 60. gününde papillomlarda; 8 inekte (%57) lezyonlarda gerileme, 5 inekte tam iyileşme (%36) ve 1(%7) inekte hiçbir değişim olmadığı bildirilmiştir. La Sota suşu NDV aşılarının antikor artışı sağladığı ve sınırlı TNF-alfa artışı gösterdiği belirtilmiştir. Puri ve ark. (2011), 34 adet deri papillom olgusu gösteren sığırda NDV La Sota suşu uygulamasında, papillomlarda %75'ten daha düşük oranda gerileme ve %8.8 düzeyinde tam iyileşme gözlemişlerdir. Hamad ve ark. (2012), 3 hayvan grubunda; birinci grupta 15 hayvana otojen aşılama, ikinci grupta 10 hayvana hücre kültür aşısı ve üçüncü gruptaki 10 hayvana virülent NDV aşısı ile tedavi uygulamaları yapmışlardır. Her üç grupta da başarılı sonuçlar alındığını rapor etmişlerdir.

SONUÇ

BPV sonucu gelişen meme papillomlarının tedavisinde otojen aşı uygulamaları daha başarılı sonuçlar verdi. BPV tip dağılımının işletme ve diğer koşullar incelendiğinde; karasinek ve sivrisinek popülasyonunun yoğun olduğu, hayvan yaşam alanlarında dışkı temizliğinin az yapıldığı ve dışkı birikiminin (gübre amaçlı) fazla olduğu, fenni yem tüketimi yoğun yapılan, sağım makinası ile süt sağımı yapan, sağım öncesi ve sonrası meme temizlik ve dezenfeksiyonuna dikkat eden, sağım makina bakımlarını periyodik olarak düzenli yapmayan işletmelerde enfeksiyonun daha yoğun olduğu belirlendi.

BPV enfeksiyonunun meme lezyonlarında şekillenmesine kaynak olabilecek diğer faktörler üzerinde de detaylı çalışma yapılmasını, bulaşmaya yardımcı faktörlerin önlenmesini, tedavide öncelikle otojen aşı uygulamalarının yaygın olarak kullanılmasının, alternatif tedavi metotlarının geliştirilmesinin ve BPV tiplerine yönelik meme papillomu için standart ticari aşılar geliştirilmesinin faydalı olacağı kanısındayız.

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Araştırma Makalesi

Kırsalda girişimcilik örneği: uzman eller projesi ile desteklenen hayvancılık işletmelerinin sosyo-ekonomik analizi

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ÖZ

Bu çalışma, Türkiye'de 4 ilde pilot olarak uygulanan uzman eller projesi kapsamında büyükbaş ve küçükbaş hayvan yetiştiriciliği projesi uygulayan işletmelerinin sosyo-ekonomik yapısı, proje uygulamasına bakış açıları ve projelerin sürdürebilirliklerine ilişkin tespitlerin ortaya konulması amacıyla yapılmıştır. Uzman eller projesi kapsamında hayvancılık projesinden 50 girişimci faydalanmıştır. Araştırma kapsamında Türkiye genelinde 18 küçükbaş ve 18 büyükbaş toplamda 36 proje girişimcisinden çevrimiçi anket uygulamaları kullanılarak veriler toplanmıştır. Girişimcilerin %47,2'si daha önce başka bir işte çalıştığı ve %61,1'inin hayvancılık faaliyetine devam etmeyi planladığı anlaşılmıştır. İşletme sahiplerinin %24'ü finansman ihtiyacını kredi temini ile karşılamaktadır. Girişimcilerin 24 tanesinin hayvancılık dışında başka bir geliri olmadığı tespit edilmiştir. Kapasite kullanım oranı ortalama %58,6 olarak hesaplanmıştır. Girişimcilerin %61,1'inin projeye devam etmek istemesi, gençlerin sektöre olan ilgisinin olduğunu göstermektedir. Proje kapsamında alanında yeterli eğitimi almış ve de kırsal alanda kendi işletmesini kurmak isteyen genç girişimciler açısından bir fırsat olduğu düşünülmektedir. Projenin bütün illerde uygulanması sektörde eğitim almış girişimcilerle kırsal alana yeni bir dinamizm kazandıracak ve hayvansal üretimin sürdürülebilirliğine katkı sağlayabileceği öngörülmektedir.

An example of entrepreneurship in rural areas: socio-economic analysis of livestock enterprises supported by the expert hands project

ABSTRACT

This study was carried out in order to reveal the socio-economic structure of the enterprises that implement the cattle and small cattle breeding project within the scope of the expert hands project implemented as a pilot in 4 provinces in Turkey, their perspectives on the project implementation and the sustainability of the projects. Within the expert hands project scope, 50 entrepreneurs benefited from the livestock project. Within the area of the research, data were collected from the entrepreneurs of 36 projects, 18 small ruminant, and 18 cattle, in Turkey, using online survey applications. It has been determined that 47.2% of the entrepreneurs have worked in another job before. 61.1% of the entrepreneurs stated that they will continue their livestock activities. 24%of business owners meet their financing needs by providing loans. It has been determined that 24 of the entrepreneurs do not have any income other than animal husbandry. The capacity utilization rate was calculated as 58.6%on average. The fact that 61.1 % of the entrepreneurs want to continue the project shows that the interest of young people in the sector is still high. It is thought that there is an opportunity for those who want to start their own business in rural areas despite having received sufficient training in the field within the scope of the project. Implementation of the project in all provinces will bring a new dynamism to the rural area and contribute to the sustainability of animal production with entrepreneurs trained in the sector.

GİRİŞ

Son zamanlarda dünyayı etkisine alan pandemi ve iklim değişikliği etkisi ile kuraklığın yaşanması; tarım ve hayvancılık ürünlerinin arzında daralmalar meydana getirmektedir. Bundan dolayı da emtia fiyatlarında artışa bağlı olarak gıda enflasyonu sorunu karşımıza çıkmaktadır. Bir yandan hayvansal ürünlerin pazarlama yapısı içindeki aracıların etkisi (Aral ve ark., 2016), diğer yandan hayvansal ürün piyasasına yapılan müdehaleler (Arıkan ve ark., 2019; Akın ve ark., 2020) tüketici fiyatlarının artmasına neden olmuştur. Diğer tarftan sermaye hasıla katsayısı diğer sektörlere oranla daha düşük olan tarım ve hayvancılık sektöründe sürdürülebilirliğin önemi, bu son gelişmelerden sonra daha da artmıştır.

Toplumun beslenmesi, ilgili sanayi kollarına hammadde sağlama, gelir dağılımındaki dengesizliğin giderilmesi, ülkelerin kendi kendine yeterliliğini sağlaması, katma değer yaratma, mirasın ve doğanın korunması, kaliteli üretimle yaşam standardının arttırılması, kırsal alanda istihdam gibi fonksiyonları olan tarım ve hayvancılık sektörlerinde sürdürülebilirliğin sağlanması adına sektörün desteklenmesi farklı yöntemlerle uygulanmaktadır (Eraktan, 1989).

Konunun sosyolojik boyutları da göz önüne alındığında, dezavantajlı konuma sahip kırsal alanda yaşayan insanların

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yaşam standartlarının yükseltilmesinin yanı sıra gelirlerinin de arttırılmasına yönelik tedbirler 2000'li yıllara doğru artış göstermiş olup, söz konusu gelir getirici tedbirlerin bütün dünya ülkelerinde genel bir tarım politikası haline geldiği gözlenmektedir (Gökhan, 2017).

Özellikle 2005 yılında AB'ye tam üyelik müzakerelerinin başlamasından sonra, AB'de uygulanan tarım ve hayvancılık politikalarının etkisi ile Türkiye'de bir dizi değişim ve dönüşüm süreci başlamıştır (Eker, 2015). Çıkarılan kanunlar ile yasal uyumlaştırma yapılmasının yanı sıra, uygulamalarda da revizyonlara gidilmiştir. Bu kapsamda Türkiye'de de kırsal kalkınma desteklemeleri 2006 yılından itibaren çeşitli yöntemlerle uygulanmaya başlanmış olup günümüzde de devam etmektedir (TRGM, 2021). Bu yöntemlerden birisi Türkiye'de 2016-2018 yılları arasında tarım ve hayvancılık sektöründeki yaş ortalamasının düşürülmesi, genç nüfusunda sektörden uzaklaşmasını önlemek, genç işgücünün tarım ve hayvancılık sektöründe istihdamını arttırıcı girişimlerde bulunmalarının önünü açmak amacıyla uygulanan Genç Çiftçi Projesidir (RG, 2016).

Tarım ve Orman Bakanlığı tarafından girişimcilik faaliyeti adı altında uygulanan bir diğer proje 2020 yılında pilot olarak 4 ilde uygulanan uzman eller projesidir. Meslek yüksekokulu veya üniversitelerin tarım, hayvancılık, ormancılık, gıda ve su ürünleri alanlarında eğitim veren bölümlerinden mezun genç nüfusun kırsal alanda istihdamına katkı sağlamak; tarım, hayvancılık, ormancılık, gıda ve su ürünleri sektörlerinde girişimciliği desteklemek, bu faaliyetlerin uzman kişiler tarafından yapılmasını teşvik etmek; tarımsal üretimin miktarını, kalitesini ve verimliliğini arttırmak; kırsal alanda tarımsal üretim yapan mevcut işletmelere örnek ve önderlik oluşturacak sürdürülebilir yatırımların desteklenmesi amacıyla 2020 yılında 98 proje uygulanmıştır (RG, 2019).

Proje kısaca, kırsal alanda yaşan ve proje uygulama süresince kırsal alanda yaşamayı taahhüt eden, az çok doğal kaynaklara erişim imkanı olan, kendi işgücünü kullanabilecek, girişimci ruha sahip ve konusunda uzman kişilerin üretime başlaması için sermaye sağlanması olarak değerlendirilebilir.

Gençlere, kadınlara, kırsal alanda ikamet edenlere, eğitim düzeyini yüksek olanlara, diploma haricinde uygulayacağı projeye ilişkin eğitim belgesi olanlara, evlilere, şehit yakını-gazi, engelli yada engelliye bakmakla yükümlü olanlara öncelik saplanmıştır. Aynı zamanda mezun olduğu bölüm ile uygulayacağı proje arasında paralellik olanlara, iş planı içeriğine göre proje sürdürülebilirliği daha yüksek olanlara, projeye öz sermaye katkısı sağlayanlara değerlendirmede daha fazla puan verilmiş ve proje uygulamada öncelik sağlanmıştır (TRGM, 2021). Bu kriterler belirlenirken, projenin sürdürülebilirlik potansiyelinin yanı sıra, sosyal devlet ilkesi de göz önünde bulundurulmuştur.

Bu çalışma ile Türkiye'de pilot olarak 4 ilde uygulanan uzman eller projesi ile büyükbaş ve küçükbaş hayvan yetiştiriciliği projesi uygulayan işletmelerinin sosyo-ekonomik yapısı, girişimciliğe ve proje uygulamasına bakış açıları ve projelerin sürdürebilirliklerine ilişkin tespitlerin ortaya konulması amaçlanmıştır.

GEREÇ ve YÖNTEM

Araştırma kapsamında tam sayım yöntemi ile belirlenen Türkiye genelinde 4 il ve 12 ilçede uzman eller projesinden yararlanan 29'u büyükbaş ve 21'i küçükbaş hayvan yetiştiricisi olmak üzere 50 girişimciye ulaşılmıştır. Çalışmaya katılmayı kabul eden 18 küçükbaş ve 18 büyükbaş olmak üzere, toplam 36 projenin hak sahibi ile çevrimiçi anket uygulamaları kullanılarak araştırmanın verileri toplanmıştır.

Çalışmanın gerecini, uzman ellerde büyükbaş ve küçükbaş hayvan yetiştiriciliği projeleri uygulamış girişimciler ile elektronik ortamda veri temin yoluyla sağlanan 2021 yılı üretim dönemine ait veriler oluşturmuştur. Girişimcilerin sektöre bakışa açıları, hayvancılığa devam etme konusundaki kararlılıkları, araziye ve finansmana erişim durumları ile ürünlerini pazarlama yöntemlerine ilişkin veriler değerlendirilmiştir. Araştırma kapsamında elde edilen verilerle veri tabanı oluşturulmuş ve çıkan sonuçlar ortalama±standart sapma, yüzde ve frekans değerler üzerinden değerlendirilmiştir. Basit, tanımlayıcı ve yer gösterici istatistiki analizler SPSS 25 version paket programı ile gerçekleştirilmiştir (IBM Corp. Released, 2017).

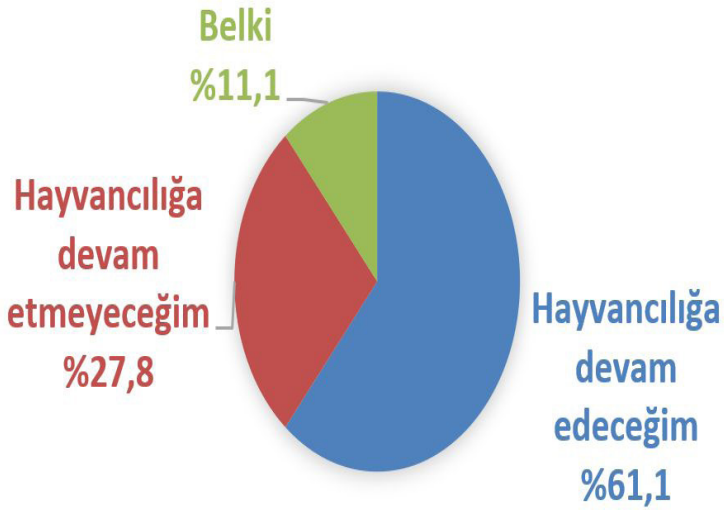
BULGULAR

Çalışma kapsamında; işletme sahiplerinin yaşı en küçük 22 ve en büyük 51 olmak üzere genel yaş ortalaması 30,77 olarak tespit edilmiştir. Veri temin edilen 36 işletmeden, 13 işletme sahibinin (%36,1) kadın ve 23 işletme sahibinin ise (%63,9) erkek girişimcilerden oluştuğu tespit edilmiştir. İşletme sahiplerinin 8'i (%22,2) anne babasıyla, 11'i (%30,6) geniş aile olarak, 14'ü (%38,9) eş ve çocuklarıyla ve 3'ü (%8,3) ise yalnız yaşadığını bildirmiştir. Girişimcilerin eğitim durumları incelendiğinde, 14 ön lisans (%38,9), 17 lisans (%47,2) ve 5 girişimcinin ise yüksek lisans (%13,9) mezunu olduğu gözlenmiştir.

Hayvancılık sektöründe girişimcilik faaliyetine başlayan bu kişilerin 17'si (%47,2) daha önce başka bir işte SGK'lı çalıştığını, 10'u (%27,8) ise daha önce herhangi bir işte girişimcilik faaliyetinde bulunduğunu beyan etmiştir. Söz konusu faaliyetleri bırakma sebebi olarak ise, yeterince para kazanamadıkları fikri birinci sırada yer alırken, uzman eller projesinden faydalanmak ikinci sırada yer almıştır. Sosyal sebepler üçüncü sırada, bırakılan sektörün kendisine uygun olmadığı düşüncesi dördüncü sırada yer almıştır.

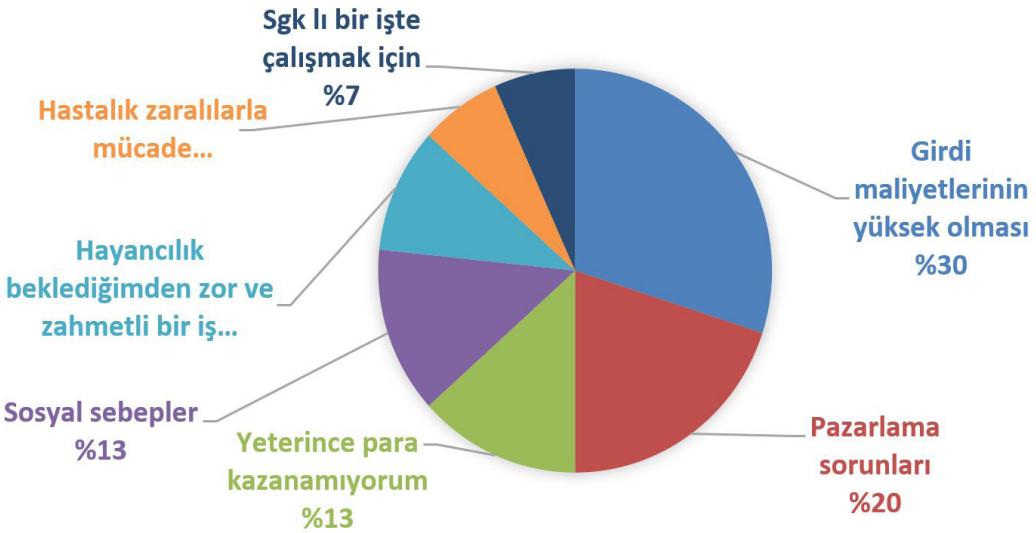
Uzman eller projesi kapsamında 2020 yılına ait hayvan alımları Nisan ayı ile Ekim ayı arasında yapılmış ve projeler hayata geçirilmeye başlanmıştır. Söz konusu süreçten 12-15 ay sonra yapılan araştırmada bütün işletmelerin faaliyetlerine devam ettiği anlaşılmaktadır.

Araştırma kapsamında incelenen 36 işletmenin, proje zorunluluğu nedeniyle kontrol ve takip süresi olan ikinci yılın sonunda hayvancılık faaliyetlerini sürdürmekteki kararlılıkları Grafik 1 de gösterilmiştir. Buna göre, 22 girişimci (% 61,1'i) hayvancılığa devam etmeyi düşündüğünü, 10 girişimci (% 27,8'i) hayvancılığa devam etmeyi düşünmediğini ve 4 girişimcinin (%11,1) ise çekimser kaldığı gözlenmiştir.



Şekil 1. Girişimcilerin işletmelerin faaliyetini sürdürme kararlılığı
Figure 1. The determination of entrepreneurs to continue the operation of enterprises

Çalışma kapsamında hayvancılığa devam etmeyi düşünmeyen girişimcilere, devam etmek istememe sebepleri sorulmuş olup, varsa birden fazla seçenek işaretleyebilecekleri bildirilmiştir. Grafik 2 de görüldüğü üzere, hayvancılığı bırakma gerekçesi olarak; girdi maliyetlerinin yüksek olması ve pazarlama sorunları % 50'lik bir yer tutmaktadır. Yeterince para kazanamamak, sosyal sebepler, yeni başlayanlar için sektörün zor ve zahmetli olması diğer önemli sebepler olarak karşımıza çıkmaktadır.



Şekil 2. Hayvancılık faaliyetini bırakmaya ilişkin sebepler
Figure 2. Reasons for quitting livestock activity

İşletmelerde arazi mülkiyet durumuna göre ortalama kullanılan arazi miktarları ve oranları Tablo 1'de sunulmuştur.

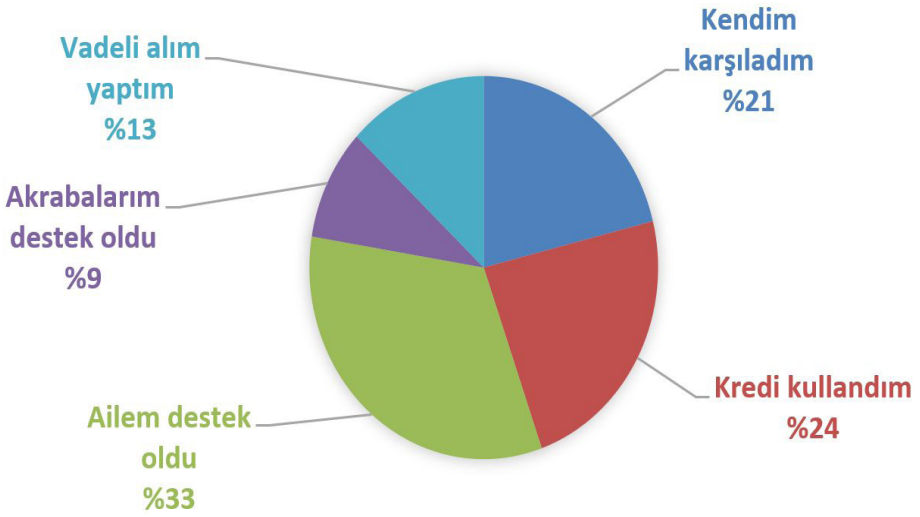
Tablo 1 incelendiğinde girişimcilerin hayvancılık faaliyetleri için işletme başına ortalama 29,6 da. arazi kullandığı; bu arazilerin %60,7'i (18,41 da./işletme) aileye ait ve ücret ödenmeden kullanılan arazilerden; %34,3 'ü (10,25 da./işletme) kiralanan arazilerden; %5'inin (0,94 da./işletme) ise kendine ait arazilerden oluştuğu anlaşılmıştır. Kullanılan arazilerin %28,4'ünün (8,42 da./işletme) ise yem bitkisi üretimi için kullanıldığı bildirilmiştir.

Grafik 3 de, girişimcilerin üretimi devam ettirmek için kullandıkları finansman kaynakları gösterilmiştir. Grafik incelendiğinde, ailesinden destek alanların oranı %33 ile ilk sırada yer alırken, kredi kullanımı %23,5 ile ikinci sırada yer almıştır. Üretim masraflarını kendilerinin karşıladığını bildirenler %21,1, vadeli alım yaptım diyenler %13 ve akrabalarından destek alanların oranı %9,4 olarak tespit edilmiştir.

Araştırmada işletmede üretilen hayvansal ürünlerin pazarlanma yöntemlerine ilişkin ilişkin bulgular Grafik 4 de gösterilmiştir. Buna göre ürünlerini kendi imkânlarıyla yöresel ürün pazarları veya hayvan pazarlarında pazarlayanlar %43,1 ile ilk sırada yer almış; toptancılar, tüccarlar, celepler vasıtasıyla

Tablo 1. Arazi mülkiyet durumuna göre işletme başına düşen ortalama kullanılan arazi miktarları ve oranları (da, %)
Table 1. Average amount and proportions of land used per enterprise by land ownership (da, %)

Yetiştiricilik konusu	İşletme sayısı	Kendine ait arazi	Aileye ait arazi	Kiralanan arazi	Toplam arazi	Yem bitkisi için kullanılan
Büyükbaş	18	1,5 daa. %7,8	11,22 daa. %58,1	6,58 daa. %34,1	19,3 daa. %100	9,27 daa. %48
Küçükbaş	18	0,95 daa %2.3	25,61 daa %63,3	13,91 daa %34,4	40,47 daa %100	7,56 daa %18,6
Ortalama	36	0,94 daa %5	18,41 daa %60,7	10,25 daa %34,3	29,6 daa. %100	8,42 daa. %28,4



Şekil 3. Girişimcilerin işletme finansmanını karşılama yöntemleri
Figure 3. Ways of entrepreneus to meet business finance

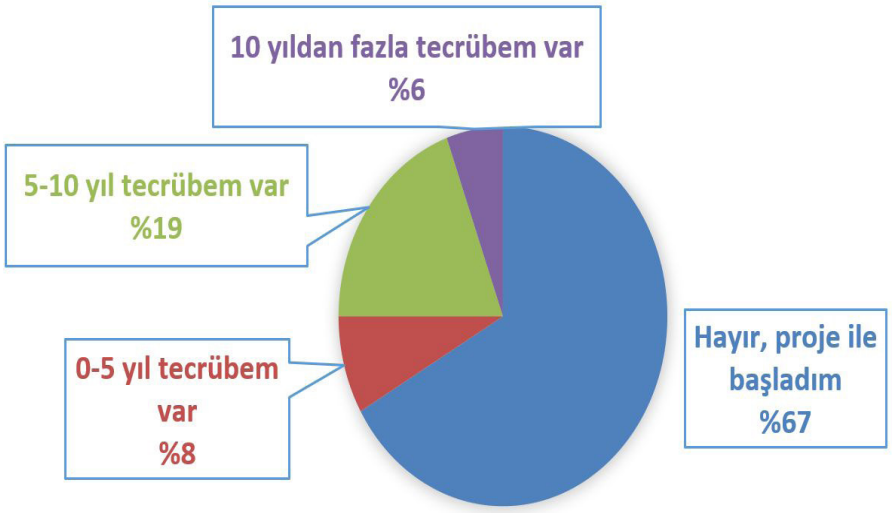
Şekil 4. İşletmede üretilen hayvansal ürünlerin pazarlanma yöntemleri
Figure 4. Marketing methods of animal products produced in the enterprise

pazarlayanlar %30,8, çiftliklerinde veya köylerinde pazarlayanlar %24,6 ve sözleşmeli olarak pazarlayanları oranı %1,5 olarak tespit edilmiştir.

Grafik 5'de veri temin edilen işletmelerin 24'ü (%66,6'sı) hayvancılık haricinde herhangi bir gelirinin olmadığını bildirmiştir. Hayvancılık harici geliri olan 12 işletme içerisinde ise, toplam geliri içerisinde hayvancılığın oranı yarıdan az olan 9 kişi (%25), gelirinin yarıdan fazlası hayvancılık harici olan ise 3 (%8,4) kişi olarak bildirilmiştir.

Hayvancılık tecrübelerinin belirlenmesi için alınan veriler Grafik 6'da sunulmuştur. Daha önce başka bir işletmede hayvancılık yapmayıp ilk defa uzman eller projesi ile hayvancılığa

Şekil 5. Girişimcilerin hayvancılık haricinde gelir durumu
Figure 5. Income status of entrepreneurs excluding livestock enterprise



Şekil 6. Proje öncesinde hayvancılık faaliyetlerine ilişkin durum
Figure 6. The situation regarding livestock activities before the project

başlayan 24 girişimci (%66,6) olduğu tespit edilmiştir. Daha önce hayvancılık yapanların 2'si (%5,5) 10 yıldan fazla süredir, 7'si (%19,4) ise 5-10 yıldır hayvancılık tecrübesinin olduğunu bildirmiştir.

Hem küçükbaş (n=16) hem de büyükbaş (n=16) işletmelerin %89'u sadece kendi alanında hayvancılık faaliyeti sürdürmekte olduğu, 2 işletmenin (%5,5) hem büyükbaş hem küçükbaş hayvancılık yaptığı, kalan 2 işletmenin (%5,5) ise hem hayvancılık hem de tarım faaliyeti yürüten miks işletme olduğu bildirilmiştir.

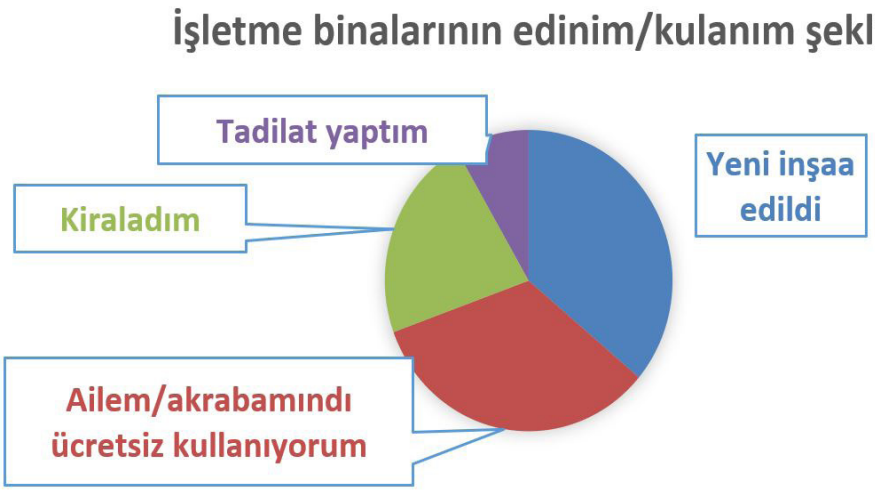
İşletme binaları üzerinde yapılan araştırmada, 3 işletmenin (%8,3) açık, 11 işletmenin (%30,6) kapalı ve 22 işletmenin (%61,1) ise yarı açık olduğu bildirilmiştir. İşletme binalarında kullanılan malzemeye göre 16 işletmenin (%44,4) kargır betonarme, 9 işletmenin (%25) çadır/sundurma, 6 işletmenin (%16,7) modern işletme şeklinde ve 5 işletmenin (%13,9) de kerpiç/taş olarak inşa edildiği beyan edilmiştir.

İşletme binalarının edinim şekline ilişkin yapılan araştırma sonuçları Grafik 7 de gösterilmiştir. Buna göre, 13'ünün (%36,1) yeni inşa edildiği, 12'sinin (%33,3) ailem veya akrabaların olduğu ve herhangi bir masraf edilmediği ve ücretsiz kullanıldığı, 8'inin (%22,2) kiralandığı, 3'ünün de (%8,3) tadilat yaparak az masrafla faal hale getirildiği bildirilmiştir.

Kapasite kullanım oranları incelendiğinde ise, büyükbaş hayvancılık işletmelerinde %53,2, küçükbaş hayvancılık işletmelerinde %64 olmak üzere ortalama kapasite kullanım oranı %58,6 olarak hesaplanmıştır.

Proje uygulamaya başlayan girişimcilerin işletmelerinde meydana gelen değişimler Tablo 2'de sunulmuştur.

Tablo 2 incelendiğinde, en çok artış gösterenlerin hayvan sayısı, hayvancılık konusunda bilgi ve tecrübe, resmi kurumlarla olan iletişim, desteklemeler konusundaki farkındalık, özgüven ve insanlarla olan iletişim olduğu; buna karşın en çok azalan



Şekil 7. İşletme binalarının edinim/kulanım şekli
Figure7. Acquisition/use of enterprise buildings

Tablo 2. Proje uygulayan işletmelerde meydana gelen başlıca değişimler
Table 2. Major changes in the companies implementing the project

Değişim	Artış gösteren işletme sayısı	Azalış gösteren işletme sayısı	Değişiklik göstermeyen işletme sayısı
Hayvan sayısı	29	1	6
Hayvancılıkla ilgili makine-ekipman	27	0	9
Hayvancılık konusunda bilgi ve tecrübe	32	0	4
Desteklemeler konusundaki farkındalık	29	4	3
Resmi kurumlarla olan iletişim	32	1	3
Teknoloji kulanım düzeyi	24	2	10
Sermaye/birikim/tasarruf	20	12	4
Borçlar	19	11	6
Özgüven/ insanlar ile olan iletişim	30	2	4
Aileye ekonomik olarak bağımlılık	13	20	3

aileye olan ekonomik bağımlılık, borçlar, sermaye ve tasarruflar olduğu, en çok değişmeyen kavramın ise teknoloji kullanım düzeyi olduğu tespit edilmiştir.

TARTIŞMA

Hayvancılık işletmelerinde yürütülen çalışmalarda işletme sahiplerinin yaş ortalamasının yüksek olduğu tespit edilmiştir (Ayvazoğlu Demir ve ark., 2015; Arıkan ve Gökhan, 2019). Bunun yanında yapılan başka bir araştırmada, aktif tarım ve hayvancılık işletme sahiplerinin genel yaş ortalaması 45,8 olarak tespit edilmiştir (KKB, 2021).

Uzman eller projesinde başvuru şartlarında yaş aralığına ilişkin herhangi bir sınırlama konulmamış olmasına rağmen, yaşı genç olan girişimcilere puanlamada öncelik verilmiştir. Araştırmamızın konusu hayvancılık işletmelerindeki yaş ortalaması 30,77 olarak tespit edilmiş olup, 22 yaşında proje uygulayan olduğu gibi 51 yaşında proje uygulayan da vardır. Söz konusu yaş ortalaması Tarım ve Orman Bakanlığı tarafından uygulanan genç çiftçi projelerindeki yaş ortalamalarında olduğu gibi Türkiye'deki genel yaş ortalamasının altındadır.

Veri temin edilen 36 işletmeden 13 işletme sahibinin (%36,1) kadın, 23 işletme sahibinin (%63,9) ise erkek girişimcilerden oluştuğu tespit edilmiştir. Söz konusu proje uygulayan kadınlara puanlamada pozitif ayrımcılık yapılmış olsa da, genç çiftçi projesi uygulayan kadın girişimcilerden (%74) oldukça düşüktür (TRGM, 2021; Satar & Sakarya, 2021). Cinsiyete ilişkin farkın bu kadar yüksek olmasındaki başlıca sebebinin, uzman eller projesinde belirli okul mezunlarının başvuru yapması ve muhtemelen bu bölümlerden mezun olan erkek oranının yüksek olması olarak değerlendirilmektedir. Diğer taraftan kırsal alanda yürütülen çalışmalarda kadın girişimcilerin eğitim seviyesinin düşük düzeyde olduğu bildirilmiştir (Karaturan ve ark., 2017).

Girişimcilerin eğitim durumları incelendiğinde, 14 ön lisans (%38,9), 17 lisans (%47,2) ve 5 girişimcinin ise yüksek lisans (%13,9) mezunu olduğu gözlenmiştir. Türkiye'de yapılan birçok araştırmada hayvancılık işletme sahiplerinin büyük kısmının ilkokul ve/veya ortaokul mezunu olduğu gözlenmektedir. Uzman eller projesinin başvuru şartlarında, en az yüksekokul mezunu olma zorunluluğu olduğundan, yapılan diğer çalışmalara göre eğitim durumunun yüksek olması ve eğitim-

li girişimcilerle hayvancılığın yapılması kabul edilebilir olarak değerlendirilmektedir.

Tarım ve Orman Bakanlığı tarafından uzman eller projesine benzer şekilde uygulanan genç çiftçi projesine ilişkin Yalova ilinde yapılan araştırmada (Güder, 2019) katılımcıların %91,9'unun, Isparta İlinde yapılan araştırmada (Başaranoğlu, 2019) ise %67,27'sinin kontrol ve denetim süresi bittikten sonra da proje konusu üretime devam edeceğini beyan ettikleri bildirilmiştir. Genç çiftçi projesine ilişkin yapılan başka bir çalışmada (Satar & Sakarya, 2021) kamu tarafından yapılan kontrol ve yaptırım süresinden yaklaşık bir yıl geçmesine rağmen faaliyetine devam eden hayvancılık işletme oranı %85,1 oranında olduğu tespit edilmiştir.

İşletme faaliyetlerinin sürdürülebilirliği göz önüne alındığında (Şekil-1), genç çiftçi projesine kıyasla uzman eller projesine devam eden hayvancılık işletmelerinin oranının görece daha düşük olacağı öngörülmektedir. Bunun sebebi olarak ise son zamanlarda KOVİD-19 salgınının da etkisi ile dünya genelinde yaşanan fiyat dalgalanmaları, girdi maliyetlerindeki artışların ürün fiyatlarına görece daha az yansıması ve üretici rantındaki azalmalar olduğu söylenebilir. Nitekim pandemi döneminde hayvansal ürün fiyatları artmış ancak girdi fiyatlarında artış daha fazla olmuştur (Sarıözkan ve ark., 2021).

Araştırmamızda, yeterince para kazanamamak, sosyal sebepler, yeni başlayanlar için sektörün zor ve zahmetli gelmesi hayvancılığın bırakılmasında diğer önemli sebepler olarak karşımıza çıkmaktadır (Şekil-2). Genç çiftçi projelerine ilişkin yapılan çalışmalarda da (Alkan, 2019; Unakıtan & Başaran, 2018), girişimcilerin faaliyetlerini sürdürme sebepleri arasında ekonomik sebeplerden ziyade sosyal ve kültürel sebepler dikkat çekicidir. Bu sebeple, tarım ve hayvancılığın, ekonomik anlamda gelir sağlamasının yanı sıra kırsal alanda sosyal bir yaşam tarzı ve kültür olduğu söylenebilir.

Hayvancılık işletmelerindeki gider kaleminin büyük kısmını oluşturan yem maliyetlerinin minimize edilmesinde, arazi varlığı ve arazinin verimli kullanılması önem arz etmektedir. Yerebakan (2017) tarafından yapılan bir araştırmada koyunculuk işletmelerinin %76,4'ünün kendisine ait arazilerinin olduğu; Özsayın ve Everets (2018) tarafından yapılan başka bir araştırmada ise sahip olunan mülk arazi miktarının 33,2 daa olduğu bildirilmiştir. Ancak yapılan bu araştırmalarda işletme sahibinin yaş ortalamasının 50 civarında olduğundan ortalama arazi büyüklüklerinin yüksek olduğu, daha düşük yaş ortalaması olan işletmelerde yapılan araştırmalarda (Satar & Sakarya, 2021), bu çalışmada olduğu gibi ciddi bir oranda arazi varlığının olmadığı anlaşılmaktadır.

Araştırma kapsamında yaş ortalamasının 30 civarında olması ve genç girişimcilerin arazi varlığının sınırlı olması krediye erişimi kısıtlamaktadır. İşletmelerde kullanılan arazilerin sadece %5'i üreticilerin kendilerine ait arazilerden oluşurken, %60,7'i aileye ait ve ücret ödenmeden kullanılan arazilerden oluşmaktadır. Türkiye'de kültürel öğelere bağlı olarak tarımsal araziler ailenin gençleri tarafından işlense de, resmi olarak genellikle aile büyüklerinin üzerinde kayıtlıdır. Yapılan araştırmada bunu kanıtlar niteliktedir. Her ne kadar Türkiye'deki gençlerin arazi edinimleri düşük ve kısıtlı olsa da, fiili duruma bakıldığında ailelerin kolektif bir şekilde üretime devam ettiği arazinin asıl sahibine bakılmaksızın hane içerisindeki üretim desenine göre işlendiği gözlenmektedir.

Araştırma yapılan işletmelerle paralel olarak, ABD ve Avrupa Birliğindeki Young Farmer Survey gibi birçok çalışmada, gençlerin finansmana, krediye ve araziye erişimlerinin zor olduğu bildirilmektedir (NYFC, 2021).

Hayvancılık işletmelerinde, üretim faaliyetlerinin yürütülmesi için gereken finansman ekonomik sürdürülebilirlik için önem arz etmektedir. Genç çiftçilerle yapılan araştırmada, tarımsal faaliyeti bırakma gerekçesi olarak finansmana erişimde yaşanan zorluklar da bildirilmiştir (Berk & Armağan, 2019). Genç çiftçiler arasında yapılan bir araştırmada, %23,3'ü ise hibenin yanı sıra kredi de çekerek tarımsal faaliyetini sürdürüldüğü bildirilmiştir (Akkaya & Gülçubuk, 2018). AB'de, 40 yaşın altındaki 2.000'den fazla çiftçi arasında yapılan bir araştırmada, satın alma veya kiralamayla araziye edindikten sonra en büyük ihtiyacın mali destek (sübvansiyonlar), krediye erişim olduğu bildirilmiştir. Satar ve Sakarya (2021) tarafından genç çiftçiler arasında yapılan araştırmada, üreticilerin %41'i üretim masraflarını kendilerinin karşıladığını bildirirken, %53'ü aile ve akrabaların destek olduğunu, %4'ünün vadeli alım yaptığını ve %2'sinin kredi kullandığını bildirmiştir.

Araştırmamızda, girişimciler hayvancılık faaliyetlerini sürdürmek için her ne kadar aile ve akrabalarından destek alsalar da, yüksekokul ve üniversite eğitimi görmüş oldukları için belirli birikimlerini finansman olarak kullandıkları ve krediye erişimlerinin de daha önce yapılan araştırmalardaki genç girişimcilerden daha fazla olmasını sağladığını düşündürmektedir. Nitekim son yıllarda yapılan çalışmalarda çiftlik sahibi/yöneticilerinin büyükbir kısmının üniversite mezunu olduğu bildirilmiştir (Akın ve ark., 2020).

Yapılan bazı araştırmalarda aile işletmelerinin pazarlama yöntemi olarak %50'lere yaklaşan geleneksel pazarlama yöntemi tüccar, celep ve toptancılar olduğu karşımıza çıkmaktadır (Satar, 2021). Araştırma kapsamındaki işletmelerde pazarlama yöntemlerine ilişkin bulgular değerlendirildiğinde, geleneksel pazarlama yöntemi olan "toptancılar, tüccarlar, celepler vasıtasıyla" seçeneği %30,8 ile ikinci sırada yer almıştır. Bunun yanı sıra "kendi imkânlarımla, yöresel ürün pazarı veya hayvan pazarı" ve "çiftliğimde/köyümde" seçeneğinin toplamı %68 gibi bir orana sahip olması sevindirici olup, konusunda uzman bu girişimcilerin, aldıkları eğitimin, kullandıkları iletişim araçlarının da etkisi ile pazarlama konusunda inovatif davrandıkları ve ürünlerini çeşitlendirerek kendi yöntemleriyle pazarladıkları gözlemlenmiştir.

Yapılan araştırmada her 3 kişiden ikisinin gelirleri sadece hayvancılık olduğu bildirilmiştir. Hayvancılık işletmelerindeki en büyük gelirin asıl yapılan iş olan hayvan ve hayvansal üründen elde edilesi, kişinin başka faaliyetlerle meşgul olmadığı ve uzmanlaşma anlamında olumlu bir durum olduğu şeklinde değerlendirilmektedir. Buna karşın, yıl boyu faaliyeti

devam eden ve bu yönü ile de sanayi işletmelerine benzeyen hayvancılık işletmelerinde, her dönem girdi (masraf) ve çıktılar (gelir) devam etmektedir. Sadece hayvancılıktan gelir elde eden işletmeler, gerek mevsimsel ve gerekse yapısal olarak oluşan arzın çok olduğu dönemlerden, kuraklıklardan, girdi ve satılan ürünlerdeki fiyat dalgalanmalarından çok etkilenmekte; çeşitli finansman temininde zorlanabilmektedir. Oysa kısmen geliri olan ve bunun yanı sıra hayvancılık faaliyetlerini sürdüren işletmelerin piyasa şartlarında söz konusu değişkenlere karşı kırılganlıklarının azaldığını düşündürmektedir. Bu sebeple, yukarıda bildirilen ve finansmanını kendinin karşıladığını bildiren işletmeler genellikle hayvancılık dışında gelir elde eden işletmelerden oluşmaktadır.

Hayvancılığın doğa koşullarına bağımlı, ekonomik, biyolojik, çevresel, sosyolojik zorlukları içeren bir sektör olduğu söylenebilir. Hayvancılık işletmelerindeki başarı, teknik bilginin yanı sıra, fiili olarak yapılacak uygulamalarla becerikli olmaya da dayanmaktadır. Araştırmada proje uygulayanlardan üçte birinin az da olsa daha önce hayvancılık tecrübesi olduğu, büyük oranının ise uzman eller projesi ile hayvancılık faaliyetlerine başladığı bildirilmiştir. Söz konusu tecrübeye sahip olan girişimcilerin sektörü ve sektörün zorlularını bildiğinden sürdürülebilirliklerinin daha yüksek olduğunu düşündürmektedir. Zira kontrol ve denetim süresi sonunda hayvancılığı bırakmayı düşünen girişimcilerin %10'u hayvancılığın beklenilenden daha zor ve zahmetli bir iş olduğu düşüncesini bildirmişlerdir.

İşletmelerin büyük kısmının (%89) sadece bir hayvancılık faaliyetini yürütmesi, işletmelerde uzmanlaşma olduğunu ve hayvancılık faaliyetlerinde artan bilgi, birikim ve uzmanlıkla zaman içerisinde daha verimli ve sürdürülebilir bir üretimle devam edeceği fikrini güçlendirmektedir.

İşletmelerin büyük kısmının açık veya yarı açık olduğu, kerpiç/taş işletme binasının ise diğer çalışmalara göre (Satar & Sakarya, 2021) az olduğu gözlenmiştir. Konusunda uzman olan bu kişilerin uyguladığı projelerdeki işletme binalarının coğrafi konuma göre değişmekle birlikte mümkün mertebe hem hijyen hem de verimlilik açısında açık ve yarı açık işletmelerde yürütülmesi önemli bir tespittir. Böylelikle projenin amacında belirtilen, uzman ellerin proje uyguladıkları yerlerde diğer yetiştiricilere örneklik/önderlik teşkil etmesi hedefinin zaman içerisinde gerçekleşeceğini düşündürmektedir.

İşletme binalarının sadece %36'sının yeni inşa edilmesi, geri kalan binaların aile/akrabalardan ücretsiz kullanılması, küçük tadilatlarla üretime kazandırılması veya boş olan binaların kiralanması, atıl durumda olan veya kullanılmayan binaların ekonomiye kazandırılmasını sağladığı söylenebilir. Özellikle aile işletmeleri kurulurken, kırsal alanın birçok yerinde daha önceden yapılmış ve atıl durumda olan binaların kiralanması veya bu şekilde ücretsiz kullanması, özellikle işletme kurulurken büyük bir gideri oluşturan sabit yatırımlara gerek kalmadan hem zamandan hem de maliyetlerden tasarruf sağlanmış olacaktır.

Mülga Gıda, Tarım ve Hayvancılık Bakanlığının yayınlamış olduğu Kırmızı Et Stratejisinde (GTHB, 2021), Türkiye'deki besi işletmelerinin yeterli kapasite ile çalışamadığı, mevcut işletmelerin atıl durumda bulunan üretim kapasitelerini artırabilmeleri ve piyasa düzenlemelerinde başarılı olabilmeleri için önlemler alınması zorunluluğu gündeme gelmiştir. Bu şekilde girdilerin azalacağı, verimlilik ve gelirlerin artış sağlayacağı, böylece işletmelerin rekabet edebilir ve sürdürülebilir olabilecekleri vurgulanmıştır. Tutar ve Eryüzlü (2015) tarafından Sakarya ilindeki hayvancılık işletmelerinde kapasite kullanımına ilişkin yapılan araştırmada, işletmelerin %60'ından fazlasının kapasite kullanım oranının %50'nin altında olduğu, bunun en büyük sebebinin ise, hammadde kalite ve tedariğinde yaşanan sorunlar olduğu tespit edilmiştir. Araştıra kapsamında kapasite kullanım oranının küçükbaş hayvancılık işletmelerinde %64, büyükbaş hayvancılık işletmelerinde %53,2 çıkması istenilen düzeyde olmasa da, diğer araştırmalara göre yüksek tespit edilmiştir. Bunun en büyük sebeplerinden birisinin, yeni inşaat giderlerinden ziyade, hali hazırda bulunan, ücretsiz veya düşük ücretle girişimcilerin kullandıkları işletme binalarından olduğunu düşündürmektedir.

SONUÇ

Girişimcilerin ortalama yaşı 30 civarında olduğundan gerek araziye ve gerekse finansmana erişimde dezavantajlı olmalarına rağmen, arazi ve finansman kullanımında aile bireylerinden ve akrabalarından destek alması projelerin sürdürülebilirliklerinin yüksek olacağı fikrini güçlendirmektedir. Bunun yanı sıra, işletme binalarının da büyük kısının aile bireylerinden temin edilmesi veya küçük tadilatlarla üretime başlanması kırsal alanda atıl duran binaların ekonomiye kazandırılması açısından değerli bir durumdur.

Girişimcilerin, daha önce başka sektörlerde girişimcilik faaliyetinde bulunmasına rağmen projeye başvurması ve kırsal alanda hayvancılık faaliyetine başlaması, gençlerin sektöre olan ilgisinin hala yüksek olduğunu göstermektedir. Proje kontrol ve denetim süresi sonunda %60'dan fazlasının devam etme eğiliminde olması, işletme faaliyetlerini bırakmayı düşünenlerin ise en büyük gerekçe olarak yapısal sorunlar göstermesi bu durumu kanıtlar niteliktedir.

Geleneksel olarak kullanılan pazarlama yapısının (toptancılar, tüccar, celep vb.) aksine, girişimcilerin büyük oranda kendi imkânlarıyla pazarlama yapmaları ve aracılar olmadan direkt tüketiciye ulaşmaları, ürünlerinin daha uygun fiyattan satmalarını ve daha çok gelir elde etmelerini sağlamaktadır.

Teknoloji kullanım düzeyi yüksek ve yenilikçi fikir/uygulamalara açık olan söz konusu girişimci kitlenin, hayvancılık konusunda, ekonomik anlamda, sosyal ve iletişim alanlarında olumlu değişimler göstermesiyle, faaliyet yürüttükleri kırsal alanda yaşayan diğer yetiştiricilere dolaylı katkılar sağlayacaktır.

Tarım ve Orman Bakanlığının uygulamış olduğu uzman eller projesi, alanında yeterli eğitimi almış olmasına karşın kırsal alanda kendi işini kurmak isteyen girişimciler açısından bir fırsat olduğu düşünülmektedir. Bu sebeple, 2022-2024 yılları arasında Türkiye'nin bütün illerinde uygulanması planlanan uzman eller projesinin, tarım, hayvancılık, gıda, su ürünleri ve

ormancılık sektöründe eğitim almış girişimcilerle kırsal alana yeni bir dinamizm kazandıracağı beklenmektedir.

Bu dinamizmin oluşmasında, hayvancılık alt üretim dalları itibariyle Türkiye'de bölge koşulları göz önünde bulundurularak hem doğal kaynakları korumak hemde bu kaynakların sürdürülebilir kullanımını sağlamak adına üretimin planlanlanması hayvancılık alanında rekabet gücünün artırılmasına katkı sağlayacaktır.

BEYANNAMELER

Etik onayı ve katılma onayı

Uygulanamaz

Çıkar çatışması

Yazarlar, herhangi bir çıkar çatışması beyan etmemektedir.

Yazar katkıları

MS ve MSA çalışmayı tasarladı.

MS çalışmanın verilerini topladı ve analiz etti.

MS, MSA ve AG analizleri yorumlayarak makaleyi yazdı.

MS ve AG makalenin eleştirel incelemesini yaptı.

Veri kullanılabilirliği

Bu çalışmanın verileri yazarlara aittir.

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Research Article

Intercondylar width index of the tibia in the dogs: A morphological study

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ABSTRACT

Morphological characteristics of the proximal tibia affect the orthopedic balance of the knee region; such as the width of the condyles, tibial plateau angle, position of the patella; as well as the intercondylar width. The objectives of this study are: (1) to calculate the intercondylar width index of the tibia in dogs, (2) to determine if there are gender-related differences in these indexes, and (3) to compare them among the six breeds of dog. For this study, the tibial bones of 84 dogs from 26 different breeds were used. The radiographs of the tibias were taken, and the tibial plateau and intercondylar widths were measured. The intercondylar width index was calculated by using the following formula: (intercondylar width/tibial plateau width) *100. The mean intercondylar width index of the dogs was 19.2±2.72. The index values of male and female dogs were 19.1±3.32 and 19.3±2.02, respectively. The significant differences in the index were not determined between male and female dogs, but among dogs' breeds. The dogs in the high-risk breeds had significantly smaller eminence width index values than the dogs in the low-risk breeds related to the non-traumatic cranial cruciate ligament rupture. The results of this study suggested performing further clinical studies to evaluate whether the difference in intercondylar width index among breeds in dogs is associated with cranial cruciate ligament deficiency.

INTRODUCTION

The femorotibial joint forms between the condyles of the femur and the tibia. The bone shapes of the femoral and tibial articular surfaces allow movement of 180°, but this large capacity of motion is restricted by soft tissues such as ligaments, joints, and muscles (Dye, 1987; Gupte, 2007). The femorotibial joint acts seamlessly as long as the congruency of all components of the joint. One of these components is the cranial cruciate ligament (CrCL) that arises from the caudomedial part of the lateral condyle of the femur and attaches to the cranial intercondyloid area of the tibia (Dyce, 2002; De Rooster, 2006). This ligament is an important joint stabilizer, primarily limiting anterior translation and medial rotation of the tibia relative to the femur as well as limiting hyperextension of the stifle joint (Arnoczky, 1977; Slocum, 1983; Lopez, 2003; Tashman, 2004). The cranial cruciate ligament rupture is one of the most common problems in dogs. Furthermore, it has increased over time, all relevant information as the underlying cause of the cranial cruciate ligament rupture is of utmost importance for breeding strategies, for identifying dogs that are below risk as well as for developing treatment strategies. The non-traumatic CrCL rupture causes hind limb lameness in dogs and its pathogenesis is believed to be multifactorial. Some anatomical characteristics are considered to play a role in this rupture despite lacking a consensus. These anatomic characteristics are the trochlear notch size of the femur (Lewis, 2008; Griffon 2010; Kara, 2011), the distal femoral geometry of the femur (Kara, 2018), the tibial plateau slope (Macias, 2002; Osmond, 2006; Talaat, 2006; Mostafa, 2009; Sabanci, 2014) alignment of the proximal shaft of the tibia (Osmond, 2006; Mostafa, 2009), and the tibial tuberosity size (Guerrer, 2007; Boudrieau, 2009; Inauen 2009; Renwick, 2009; Griffon, 2010). These studies provide important data in assessing the orthopaedical potential risk factors as well as the treatment strategies for the CrCL rupture in dogs. In addition to above mentioned characteristics, the CrCL size may be important because a smaller material in size has less strength than a larger one, assuming that their material properties are similar. Based on this basic biomechanical principle and the assumption that the diameter of the anterior cruciate ligament (corresponding CrCL in dogs) is approximately the same as the tibial intercondylar width, the eminence width index from the tibia was developed in humans (Uhorchak, 2003). This index is calculated by dividing intercondylar width into tibial plateau width and the difference is determined between the groups with and without the nontraumatic CrCL injury in both men and women (Uhorchak, 2003). It is thought that the term intercondylar is more suitable than the term eminence, therefore in the manuscript, the intercondylar width index (ICWI) was used instead of the eminence width index in humans. To the best of our knowledge, the intercondylar width index has not been documented previously in dogs. We

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hypothesized that this index, like human beings, would be an additional variable for the evaluation of CrCL deficiency in dogs. With these in mind, the objectives of the study are to calculate the ICWI in dogs, to determine if there are gender-related differences in ICWI, to compare the ICWI among the six breeds of dogs.

MATERIAL and METHODS

Materials

A total number of 168 tibias were procured from 84 mature dogs, including 41 males and 43 females. The bone materials were obtained from the Veterinary Anatomy Department Collections of Adnan Menderes University and Istanbul University in Turkey. Two criteria, the lack of gross pathological changes, and the fusion of proximal tibial growth plates; were used for inclusion in the study. Twenty-six different breeds were used in this study as follows; the Anatolian shepherd (n=11), German shepherd (n=11), mixed breed (n=11), Boxer (n=6), Rottweiler (n=6), white Terrier (n=6), Doberman (n=4), Pointer (n=4), Cocker (n=3), Collie (n=2), Pekingese (n=2), Setter (n=2), Siberian Husky (n=2), St. Bernard (n=2), and one sample for the following dog breeds: American Staffordshire Terrier, Bulldog, Caucasian shepherd, Chow Chow, Clumber, Dachshund, Great Dane, Golden Retriever, Malamute, Mastiff, Pitbull, and Shar-Pei.

Measurements

The craniocaudal radiographs of the tibias were taken by positioning the caudal surface of the tibias on the film cassette as the caudal ends of both condyles touched the cassette and centering the beam on the middle of two intercondylar tubercles (Comed Medical System, EVA-HF750, Korea). For the measurements of the length and midshaft width of tibias, the radiographic images were transferred to a computer in JPEG format. The JPEG images were transferred to 'ImageJ software' (Wisconsin - USA), with fixed scale and resolution (3456x2304 pixels) for calibrating and measuring. Tibial plateau and intercondylar widths were measured by using a modified method, described in humans (Uhorchak, 2003). A line was drawn between the most proximal parts of the medial and lateral condyles of the tibia, and the width of this line was chosen as the tibial plateau width. Having marked the peaks of the intercondylar tubercles, two lines were drawn perpendicularly from these points to the line of tibial plateau width. The distance between the later lines was designated as the intercondylar width (Figure 1).

Statistical Analysis

After repeating the measurements three times by the same investigator at an interval of 15 days, the three results were compared by repeated ANOVA. Once it was determined that there was no statistical significance among the measurements, the average of the triplicate measures was taken for the statistical analysis. Mean values for the left and right sides were compared by using paired t-test to assess the differences between the right and left sides. After determining that there was no significance between the two sides, the mean of each measurement was then used to calculate the intercondylar width index for each dog. As a result, one value per dog from each variable was obtained for further analysis. The intercondylar width index (ICWI) was calculated, using the formula; (intercondylar width/tibial plateau width) * 100. Mann-Whitney U test was used to determine whether gender-related differences in the index exist or not. The tibial length was also used as a covariate in addition to gender by using ANCOVA. The ICWI was compared among the six breeds consisting of six or more dogs by the Kruskal-Wallis test. Mann-Whitney with Bonferroni correction test was used as post hoc if the differences were significant. Statistical analysis was performed using SPSS statistical package program (SPSS for Windows, version 19.0, California, USA). Summary statistics were reported as the mean, standard deviation (±SD), 95% confidence interval, and percent variation coefficient. The level of significance was set at p values <0.05 for all the analyses.

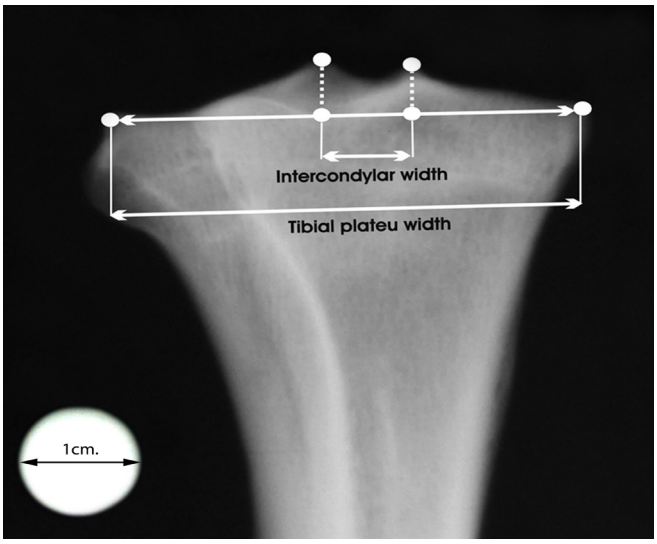

Figure 1. The tibial plateau and intercondylar widths measurements in craniocaudal radiograph of the proximal tibia.

RESULTS

Twenty-six different breeds were included in the study, the majority of the dogs being from the Anatolian shepherd, the German shepherd, and mixed breed. All samples were obtained from adult dogs and the proximal tibial growth plate was fused. The proportion of males and females was almost equal, 49% and 51%, respectively. There was no significant difference in tibial length (p= .186), mid-shaft width (p= .189), tibial plateau width (p= .878) and intercondylar width (p= .541) between the right and left sides. The mean tibial length and mid-shaft width were 197±47.2 mm and 14.5±2.91 mm, respectively. The tibial plateau width was 35.3±6.75 mm and the intercondylar width was 6.73±1.41 mm. The ICWI was calculated as 19.2±2.72 and the 95% confidence interval was from 18.6 to 19.8 for all dogs. The mean ICWI of male and female dogs were 19.1±3.32 and 19.3±2.02, respectively (Figure 2). The difference in ICWI between male and female dogs was not significant (p= .534). When tibial length was

used as a covariate, the gender-based differences were also not determined (p= .727).

Based on the comparison of the ICWI among the six breeds, the lowest ICWI was seen in Rottweiler. There were significant differences between the Rottweiler and Anatolian shepherd as well as between the Rottweiler and Terrier (Table 1). Dot plots for the ICWI in six dog breeds and 95% confidence interval for 84 dogs were presented in figure 3. The ICWI values were below the lower bound of the 95% confidence interval in all Rottweiler and Boxer, except one Boxer

The degree of variation for tibial plateau width, intercondylar width, and ICWI was calculated as 19.1%, 21.0%, and 14.1%, respectively. Among these, the intercondylar width showed the highest variation while the ICWI showed the lowest variation.

Table 1. The intercondylar width index of tibia in six breeds of the dogs.

	Rottweiler (n=6)	Boxer (n=6)	Terrier (n=6)	German shepherd (n=11)	Mixed breed (n=11)	Anatolian shepherd (n=11)	p
	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD	
Intercondylar width index	15.8 ± 0.72^{a}	17.2 ± 2.46^{ab}	19.8 ± 4.96^{b}	17.7 ± 2.01^{ab}	19.2 ± 2.01^{ab}	19.9 ± 1.76^{b}	0.009

Different superscript letters in the same line are significantly different ($p<0.05$).

DISCUSSION

The pathogenesis of nontraumatic CrCL rupture is believed to be multifactorial and its prevalence among dogs has increased over time (Witsberger, 2008). Skeletal factors are suggested to be evaluated first in the assessment of the CrCL rupture (Tillman, 2002) because any conformational differences between sexes or individuals are considered as the underlying cause for the susceptibility to stifle instability (Hashemi, 2011). Therefore, conformational characteristics remain an active area of research (Griffin, 2006). However, the great variation in size among breeds and individuals within the same breed causes difficulty in practical usage of the absolute morphometric values in dogs. In this regard, the indices are important for the documentation and comparison of these values. Percent variation coefficients obtained in the present study confirm these observations as the ICWI showed relatively less variation than the tibial plateau and the intercondylar widths as 14.1%, 19.1%, and 21.0%, respectively.

Women have a greater chance of developing the anterior cruciate ligament rupture than men (Uhorchak, 2003; Chandrashekar, 2005; 2006; Dienst, 2007; Hashemi, 2011). Furthermore, there are reports demonstrating that there exists a relationship between the nontraumatic rupture and the weight and height in humans (Bennet, 1988; Charlton, 2002; Wolters, 2011). However, there is no consensus on the effect of gender to the incidence of the CrCL rupture in dogs. Male (Grierson, 2011), female (Harasen, 1995; Zeltzman, 2005; Wilke, 2006), or neutered dogs (Whitehair, 1993; Duval, 1999; Witsberger, 2008) are reported to be more susceptible to developing the CrCL rupture. In the present study, weight records of dogs were not available, but the ICWI did not differ significantly between male and female dogs. Also, a gender-based difference was not determined when the tibial length was used as a covariate. It would appear from these observations that the tibial length and gender differences may not have any effect on ICWI in dogs.

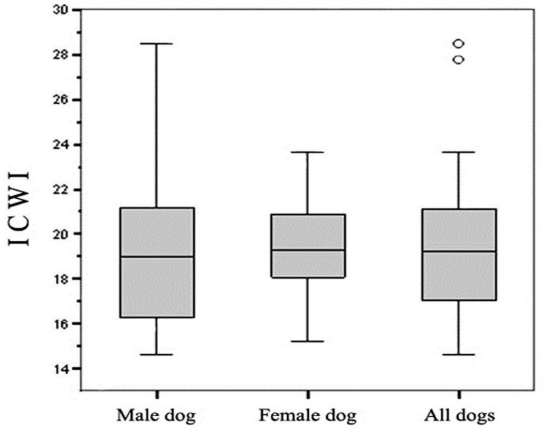

Figure 2. Box plots of the index values in male, female, and all dogs. The median and mean values of the index are the same, and "o" indicates the extreme values. (ICWI: Intercondylar Width Index)

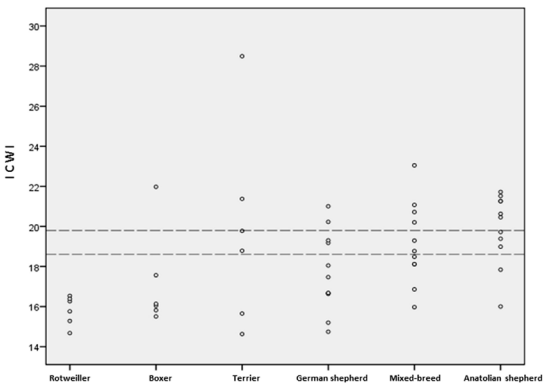

Figure 3. Dot plots of the index values in six breeds of dogs. The interval between two broken lines indicates the 95% confidence interval for 84 dogs. (ICWI: Intercondylar Width Index)

The ICWI developed in humans is based on the assumption that the diameter of the CrCL is approximately the same as

tibial intercondylar width (Uhorchak, 2003). Although there has not been a study that compares the CrCl diameter and the intercondylar width in dogs, the significant differences in ICWI were determined between the Rottweilers and Anatolian shepherd as well as between the Rottweilers and Terriers. In this study design, it was impossible to find out the underlying cause of this difference.

It should be noted that the present study had limitations. We had no records related to the soft tissue injuries of the stifle joint, although only tibias without gross pathological changes of mature dogs were included in the study. However, it is not unlikely that the soft tissue injuries lead to any changes in the intercondylar width lacking gross pathological changes of the tibial plateau.

CONCLUSION

In conclusion, in light of the fact that the prevalence of the CrCL rupture has increased over time in dogs (Witsberger, 2008), all relevant information as for the underlying cause of the CrCL rupture is of utmost importance for breeding strategies, for identifying dogs that are under risk as well as for developing treatment strategies in cases of the CrCL rupture. This study introduces, to the best of our knowledge, the ICWI for the first time in dogs. The lowest ICWI was determined in the Rottweilers and Boxers, but retrospective studies are needed using an adequate number of dogs in each breed with and without risk for nontraumatic CrCL rupture to extend and clarify the results of the present study.

DECLARATIONS

Ethics Approval

Not applicable

Conflict of Interest

The author declares that he has no conflict of interest.

Consent for Publication

Not applicable

Author contribution

Idea, concept, and design: SSS

Data collection and analysis: SSS

Drafting of the manuscript: SSS

Critical review: SSS

Data Availability

The data used to support the findings of the study are included within the article.

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Araştırma Makalesi

Oksfendazol, oksiklozanid ve oksfendazol-oksiklozanid kombinasyonunun koyun ve keçilerde biyokimyasal ve hematolojik parametreler üzerine etkileri

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Anahtar Kelimeler:
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ÖZ

Mevcut araştırmada sağlıklı koyun ve keçilere oral yolla oksfendazol, oksiklozanid ve oksfendazol-oksiklozanid kombinasyon uygulamasının, farklı zamanlarda biyokimyasal ve hematolojik parametreler üzerine etkisinin belirlenmesi amaçlanmıştır. Çalışma, ilaç uygulamaları arası 21 gün olmak üzere üç periyotta çapraz dizayna göre tasarlanmıştır. İlaç uygulamaları klinik olarak sağlıklı oldukları belirlenen, 1-3 yaştaki, dişi, İvesi koyun (n=6) ve Alpin keçi (n=6) ırkı üzerinde gerçekleştirilmiştir. Koyun ve keçilere oral yolla 7.5 mg/kg oksfendazol, 15 mg/kg oksiklozanid ve oksfendazol-oksiklozanid (7.5 mg/kg-15 mg/kg) kombinasyonu uygulanmıştır. Kan örnekleri ilaç uygulamalarından önce (0.saat, kontrol) ve sonraki 8., 24., 72. saat ile 7. günde alınmıştır. Biyokimyasal parametrelerden albümin, alkalen fosfataz, alanin aminotransferaz, aspartat aminotransferaz, kolesterol, trigliserid, total protein, kan üre nitrojen, kreatinin değerleri otoanalizatör cihazında belirlenmiştir. Hematolojik parametrelerden alyuvar sayısı, hemoglobin, hematokrit, ortalama hücresel hacim, ortalama korpüsküler hemoglobin, ortalama korpüsküler hemoglobin konsantrasyonu, akyuvar sayısı, lenfosit, monosit, % granülosit, % lenfosit, % monosit ve % eritrosit dağılım genişliği değerleri kan hücresi sayım cihazında ölçülmüştür. İlaç grupları ve zamana bağlı olarak hematolojik ve serum biyokimya parametrelerinde anlamlı bir fark bulunamamıştır. Bu sonuçlar koyun ve keçilere oral yolla, 7.5 mg/kg oksfendazol ve 15 mg/kg oksiklozanid tek veya kombine olarak uygulandığında hematolojik ve biyokimyasal parametrelerin fizyolojik sınırlarda olduğunu ve klinik olarak anlamlı bir etkisinin olmadığını göstermiştir.

Effects of oxfendazole, oxyclozanide and oxfendazole-oxyclozanide combination on biochemical and hematological parameters in sheep and goats

ABSTRACT

In the present study, it was aimed to determine the effect on biochemical and hematological parameters in blood samples taken at different times after oral administration of oxfendazole, oxyclozanide and oxfendazole-oxyclozanide combination to healthy sheep and goats. The study was designed according to the crossover design in three periods with 21 days between drug administrations. Drug administrations were carried out on healthy females, 1-3 years old Awassi sheep (n=6) and Alpine goats (n=6). Sheep and goats were administered orally, 7.5 mg/kg oxfendazole, 15 mg/kg oxyclozanide and oxfendazole-oxyclozanide (7.5 mg/kg-15 mg/kg) combination. Blood samples were taken before drug administration at 0 (pre-treatment) and 8, 24, 72. hours and 7 days post dosing. Among the biochemical parameters, albumin, alkaline phosphatase, alanine aminotransferase, aspartate aminotransferase, cholesterol, triglyceride, total protein, blood urea nitrogen, creatinine values were determined in an autoanalyzer. Among the hematological parameters, red blood cells, hemoglobin, hematocrit, mean cellular volume, mean corpuscular hemoglobin, mean corpuscular hemoglobin concentration, white blood cells, lymphocyte, monocytes, granulocyte %, lymphocyte %, monocyte % and red cell distribution width % values were measured in the blood cell counter. No significant difference was found in hematological and serum biochemical parameters depending on drug groups and time. These results show that when 7.5 mg/kg oxfendazole and 15 mg/kg oxyclozanide are administered orally to sheep and goats alone or in combination, hematological and biochemical parameters are within physiological limits and have no clinically significant effect.

GİRİŞ

Benzimidazol grubu antelmentikler, veteriner ve beşerî hekimlikte yaygın olarak kullanılmaktadır. Benzimidazol bileşiklerin terapötik indeksinin geniş olması, dünya çapında başarılı ve yaygın kullanımlarında önemli bir faktördür (Campbell, 1990). Oksfendazol (OFZ), antelmentik etkinliği yüksek benzimidazollerdendir. Özellikle koyun ve keçilerde akciğer ve mide-bağırsak kurtlarına karşı etkilidir, ayrıca birçok gastrointestinal nematodun larval dönemlerini de inhibe eder (Lanusse & Pri-

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chard, 1993). Genel olarak benzimidazol türevi antelmentikler, tüm memeli türlerinde geniş ölçüde metabolize edilir. Birincil metabolitler genellikle oksidasyon ve hidroliz reaksiyonlarının ürünleridir ve tümü ana ilaca göre daha polar ve hidrofiliktir. Ayrıca benzimidazol antelmentiklerin detoksifikasyonunda faz II konjugasyon reaksiyonları oldukça önemlidir; oksidize ve hidrolize metabolitler polaritelerini artırmak için glukuronid ve/veya sülfat ile konjuge edilir; bu da idrar veya safra yoluyla atılımı kolaylaştırır (Lanusse & Prichard, 1993). OFZ'nin fenbendazole dönüşümü tersine çevrilebilir. OFZ'nin fenbendazole indirgenmesi gastrointestinal sistemde meydana gelirken, fenbendazolün OFZ'ye S-oksidasyonu esas olarak karaciğerde

Tablo 1. Koyunlara oral yolla, tek doz oksfendazol (OFZ, 7.5 mg/kg), oksiklozanid (OCZ, 15 mg/kg) ve oksfendazol-oksiklozanid kombinasyonu (OFZ-OCZ, 7.5 mg/kg-15 mg/kg) uygulandıktan sonra hematolojik parametreler üzerine etkileri (Ortalama ± SS)
Table 1. Effects on hematological parameters after oral administration of a single dose of oxfendazole (OFZ, 7.5 mg/kg), oxyclozanide (OCZ, 15 mg/kg) and oxfendazole-oxyclozanide combination (OFZ-OCZ, 7.5 mg/kg-15 mg/kg) to sheep (Mean ± SD)

Parametre	İlaç Grupları	0.saat	8.saat	24.saat	72.saat	7.gün	RD*
RBC	OFZ	8.75±0.81	8.68±0.78	8.64±0.78	8.59±0.79	8.50±0.75	8-18 ($\times10^6$/µL)
	OCZ	8.77±0.67	8.69±0.70	8.60±0.69	8.52±0.73	8.46±0.68	
	OFZ-OCZ	8.74±0.82	8.66±0.78	8.59±0.73	8.51±0.59	8.48±0.58	
HGB	OFZ	10.25±0.77	10.24±0.77	10.23±0.78	10.22±0.79	10.14±0.79	9-15 (g/dL)
	OCZ	10.40±0.83	10.34±0.87	10.28±0.89	10.22±0.95	10.19±0.95	
	OFZ-OCZ	10.18±0.66	10.13±0.63	10.09±0.61	10.05±0.53	10.09±0.54	
HCT	OFZ	25.86±2.02	25.69±1.96	25.66±1.97	25.63±1.98	25.45±1.96	24-49 (%)
	OCZ	26.47±3.27	26.34±3.36	26.25±3.42	26.15±3.53	26.13±3.53	
	OFZ-OCZ	25.56±1.38	25.53±1.36	25.53±1.36	25.55±1.39	25.72±1.40	
MCV	OFZ	29.63±2.06	29.71±2.05	29.80±2.02	29.93±1.97	30.03±1.93	28-40 (fL)
	OCZ	30.25±3.59	30.38±3.52	30.55±3.38	30.70±3.26	30.87±3.13	
	OFZ-OCZ	29.42±2.37	29.66±2.37	29.89±2.28	30.14±2.23	30.41±2.10	
MCH	OFZ	11.81±0.60	11.86±0.57	11.91±0.53	11.92±0.53	11.95±0.50	8-12 (pg)
	OCZ	11.88±0.73	11.92±0.70	11.95±0.68	12.00±0.63	12.04±0.60	
	OFZ-OCZ	11.58±0.57	11.63±0.56	11.69±0.55	11.74±0.54	11.81±0.53	
MCHC	OFZ	39.82±1.12	39.91±1.06	39.93±1.06	39.91±1.06	39.91±1.06	30-40 (g/dL)
	OCZ	39.58±2.35	39.56±2.34	39.45±2.24	39.41±2.21	39.31±2.15	
	OFZ-OCZ	39.88±1.56	39.73±1.60	39.58±1.54	39.43±1.53	39.29±1.46	
WBC	OFZ	6.39±2.27	6.34±2.31	6.17±2.33	5.99±2.37	5.75±2.33	4-12 ($\times10^3$/µL)
	OCZ	6.04±1.98	6.00±1.99	5.92±2.00	5.83±2.02	5.80±2.01	
	OFZ-OCZ	5.86±2.03	5.82±2.02	5.95±2.23	5.88±2.18	5.84±2.17	
Lenfosit	OFZ	3.13±1.24	3.11±1.25	3.04±1.27	2.94±1.32	2.82±1.27	2-9 ($\times10^3$/µL)
	OCZ	2.82±1.05	2.78±1.05	2.74±1.05	2.66±1.00	2.64±1.00	
	OFZ-OCZ	2.82±1.08	2.75±1.09	2.74±1.08	2.69±1.05	2.65±1.01	
Monosit	OFZ	0.51±0.17	0.51±0.18	0.49±0.18	0.48±0.18	0.46±0.18	0-0.75 ($\times10^3$/µL)
	OCZ	0.43±0.15	0.43±0.15	0.43±0.15	0.43±0.15	0.43±0.15	
	OFZ-OCZ	0.40±0.14	0.40±0.14	0.40±0.14	0.40±0.14	0.41±0.15	
% Granülosit	OFZ	43.50±11.53	43.48±11.52	43.23±11.52	43.94±12.03	43.70±12.17^{a}	11-60 (%)
	OCZ	46.39±8.97	46.69±8.76	46.73±8.76	47.21±8.15	47.11±8.14^{ab}	
	OFZ-OCZ	46.94±14.65	47.68±14.86	48.34±15.34	48.56±15.33	48.88±15.11^{b}	
% Lenfosit	OFZ	49.21±12.40	49.31±12.39	49.51±12.38	48.80±12.87	49.08±13.05	40-75 (%)
	OCZ	47.22±9.28	46.93±9.06	46.87±9.04	46.30±8.32	46.37±8.31	
	OFZ-OCZ	46.85±14.60	46.08±14.80	45.43±15.20	45.20±15.18	44.83±14.94	
% Monosit	OFZ	7.29±1.36	7.21±1.46	7.26±1.48	7.26±1.48	7.21±1.52	0-7.5 (%)
	OCZ	6.38±1.90	6.38±1.90	6.38±1.90	6.48±1.89	6.51±1.88	
	OFZ-OCZ	6.21±1.37	6.24±1.37	6.24±1.37	6.24±1.37	6.29±1.38	
% RDW	OFZ	23.83±0.91	23.75±0.92	23.66±0.93	23.58±1.02	23.47±1.02	
	OCZ	23.93±0.96	23.80±0.96	23.70±0.97	23.57±0.96	23.50±0.90	
	OFZ-OCZ	23.57±1.00	23.43±1.03	23.30±1.04	23.15±1.01	23.01±1.03	

Aynı sütundaki farklı harfler $^{(a,b)}$ istatistiki açıdan önemlidir ($p<0.05$). RD: Referans değer, *(Bülbül, 2013; Aiello & Moses, 2016).

Tablo 2. Koyunlara oral yolla, tek doz oksfendazol (OFZ, 7.5 mg/kg), oksiklozanid (OCZ, 15 mg/kg) ve oksfendazol-oksiklozanid kombinasyonu (OFZ-OCZ, 7.5 mg/kg-15 mg/kg) uygulandıktan sonra biyokimyasal parametreler üzerine etkileri (Ortalama ± SS)
Table 2. Effects on biochemical parameters after oral administration of a single dose of oxfendazole (OFZ, 7.5 mg/kg), oxyclozanide (OCZ, 15 mg/kg) and oxfendazole-oxyclozanide combination (OFZ-OCZ, 7.5 mg/kg-15 mg/kg) to sheep (Mean ± SD)

Parametre	İlaç Grupları	0.saat	8.saat	24.saat	72.saat	7.gün	RD*
ALB	OFZ	2.17±0.29	2.15±0.27	2.15±0.27	2.16±0.27	2.15±0.27	2.7-3.7 (g/dL)
	OCZ	2.12±0.26	2.12±0.27	2.12±0.27	2.13±0.28	2.14±0.28	
	OFZ-OCZ	2.25±0.27	2.22±0.27	2.20±0.27	2.19±0.26	2.19±0.26	
ALP	OFZ	229.31±88.60	224.42±93.53	218.85±97.49	213.50±101.11	206.50±104.23	68-387 (u/L)
	OCZ	311.42±165.42	293.92±166.86	274.65±162.94	258.65±160.33	241.77±142.15	
	OFZ-OCZ	280.50±127.99	268.31±133.88	258.04±138.95	246.96±141.62	237.00±143.54	
ALT	OFZ	14.54±2.53	14.23±2.45	14.08±2.59	13.88±2.63	13.69±2.57	15-44 (u/L)
	OCZ	15.12±5.41	14.85±5.17	14.42±4.73	14.12±4.40	13.38±3.54	
	OFZ-OCZ	16.04±4.38	15.58±4.46	15.23±4.54	17.77±4.54	14.42±4.54	
AST	OFZ	72.85±12.77	71.81±11.69	71.58±11.73	71.54±11.73	71.54±11.73	49-123 (u/L)
	OCZ	71.81±12.45	71.38±12.52	70.42±12.23	69.69±12.14	69.73±12.76	
	OFZ-OCZ	75.31±13.59	74.77±13.91	74.58±14.05	73.85±14.46	73.73±14.52	
BUN	OFZ	14.00±8.40	14.31±8.12	14.62±7.95	14.92±7.75	14.96±7.72	10-26 (mg/dL)
	OCZ	13.00±7.13	13.54±7.50	14.12±8.09	14.62±8.35	14.69±8.35	
	OFZ-OCZ	16.19±8.87	15.38±8.71	14.62±8.33	13.85±7.86	13.31±7.76	
CH	OFZ	51.12±6.68	51.23±6.77	51.92±6.71	52.46±6.53	52.65±6.64	44-90 (mg/dL)
	OCZ	53.15±12.34	53.19±12.34	52.96±12.36	53.04±12.35	53.38±12.99	
	OFZ-OCZ	55.15±11.34	54.35±11.63	54.04±11.80	53.96±11.82	54.12±11.76	
CR	OFZ	0.59±0.07	0.59±0.07	0.60±0.07	0.60±0.07	0.60±0.07	0.5-1.9 (mg/dL)
	OCZ	0.56±0.07	0.57±0.08	0.58±0.08	0.58±0.08	0.59±0.08	
	OFZ-OCZ	0.59±0.07	0.58±0.05	0.59±0.05	0.59±0.05	0.60±0.06	
TP	OFZ	5.82±0.72	5.82±0.73	5.88±0.71	5.93±0.69	5.95±0.69	5.5-7.8 (g/dL)
	OCZ	5.61±0.63	5.68±0.71	5.72±0.75	5.78±0.80	5.86±0.82	
	OFZ-OCZ	5.86±0.56	5.84±0.56	5.87±0.56	5.89±0.56	5.96±0.55	
TRIG	OFZ	25.65±23.43	25.31±23.37	25.00±23.26	24.81±23.13	24.08±22.93	15-40 (mg/dL)
	OCZ	16.96±8.80	16.38±8.81	15.19±7.32	15.12±7.25	14.92±6.85	
	OFZ-OCZ	18.73±7.63	17.62±6.94	16.96±6.08	16.35±5.72	16.19±5.76	

RD: Referans değer, *(Bülbül, 2013; Aiello & Moses, 2016).

meydana gelir (Lanusse & Prichard, 1993). OFZ ayrıca daha az aktif bir metabolit olan fenbendazol sülfona karaciğerde oksidize olur. Bu oksidatif adım, daha yavaş ve dönüşümsüzdür; bu olaya sitokrom P-450 sistemi aracılık eder (Oukessou & Chkounda, 1997).

Oksiklozanid (OCZ) parazitlerde oksidatif fosforilasyon zincirini kırarak etki eden salisilanid türevi antelmentik bir ilaçtır (Veenendaal & De Waal, 1974). Büyük ve küçük ruminant türlerinde karaciğer kelebeklerinin ergin dönemlerinin tedavisi ve kontrolünde kullanılır (Paraud ve ark., 2009; Walley, 1966).

Veteriner sahada antelmentik kombinasyonları evcil ve çiftlik hayvanlarında farklı iç parazit türlerine karşı etki spektrumunu genişletmek ve dirençli nematodlara karşı etkinlik sağlamak amacıyla yaygın olarak kullanılmaktadır (Miller & Craig, 1996; Waller ve ark., 1990). OFZ-OCZ kombinasyonu ile trematod, cestod ve nematodlara karşı etkinlik sağlayacak şekilde spektrum genişlediğinden büyük ve küçük ruminantlarda parazit infestasyonlarının tedavisi ve kontrolünde sıklıkla tercih edilmektedir. Bununla birlikte veteriner sahada ruhsatlandırılmış OFZ-OCZ müstahzarlarında hedef tür olarak küçük ruminantlar için sadece koyun belirtildiğinden; keçilere uygulanan ilaç dozu, koyunlar için belirlenen doz üzerinden hesaplanmaktadır. İstenmeyen ilaç reaksiyonları dozaj rejimi ve tür farklılığı ile ilişkili olarak ortaya çıkabilir ve klinik semptomlar, biyokimyasal ve hematolojik laboratuvar testleri ile belirlenebilir.

Biyokimyasal ve hematolojik parametreler yapısal toksik etkilerin göstergesi olarak kabul edilir. OFZ, OCZ ve OFZ-OCZ kombinasyonunun koyun ve keçi kanında hematolojik ve biyokimyasal parametrelere olan etkisi bugüne kadar bildirilmemiştir. Bu araştırmanın amacı oral yolla, tek doz uygulanan OFZ, OCZ ve OFZ-OCZ kombinasyonunun farklı saatlerde koyun ve keçilerde hematolojik ve biyokimyasal parametreler üzerinde herhangi bir değişikliğe neden olup olmadığını değerlendirmektir.

Tablo 3. Keçilere oral yolla, tek doz oksfendazol (OFZ, 7.5 mg/kg), oksiklozanid (OCZ, 15 mg/kg) ve oksfendazol-oksiklozanid kombinasyonu (OFZ-OCZ, 7.5 mg/kg-15 mg/kg) uygulandıktan sonra hematolojik parametreler üzerine etkileri (Ortalama ± SS)
Table 3. Effects on hematological parameters after oral administration of a single dose of oxfendazole (OFZ, 7.5 mg/kg), oxyclozanide (OCZ, 15 mg/kg) and oxfendazole-oxyclozanide combination (OFZ-OCZ, 7.5 mg/kg-15 mg/kg) to goats (Mean ± SD)

Parametre	İlaç Grupları	0.saat	8.saat	24.saat	72.saat	7.gün	RD*
RBC	OFZ	14.28±2.03	14.01±1.91	13.92±1.85	13.87±1.85	13.78±1.85	8-18 ($\times10^6$/µL)
	OCZ	14.43±1.75	14.30±1.95	14.27±1.98	14.20±2.06	14.19±2.07	
	OFZ-OCZ	14.07±2.21	14.15±2.20	14.21±2.16	14.26±2.14	14.28±2.12	
HGB	OFZ	9.88±1.46	9.83±1.45	9.75±1.40	9.75±1.40	9.71±1.38	8-12 (g/dL)
	OCZ	9.93±0.95	9.86±1.04	9.88±1.01	9.88±1.02	9.93±0.98	
	OFZ-OCZ	9.70±1.43	9.85±1.56	9.97±1.55	10.06±1.56	10.07±1.55	
HCT	OFZ	22.99±7.50	23.92±5.90	23.71±5.77	23.70±5.77	23.60±5.72	21-38 (%)
	OCZ	22.28±8.51	22.13±8.51	22.20±8.51	22.23±8.51	22.40±8.54	
	OFZ-OCZ	21.43±8.34	21.88±8.70	22.26±8.87	22.59±9.01	22.63±9.01	
MCV	OFZ	17.90±1.37	18.05±1.51	18.01±1.51	18.08±1.56	18.13±1.60	16-25 (fL)
	OCZ	17.94±1.33	18.03±1.41	18.13±1.50	18.27±1.54	18.42±1.57	
	OFZ-OCZ	17.97±1.25	18.17±1.55	18.37±1.76	18.55±1.91	18.55±1.91	
MCH	OFZ	6.90±0.59	6.99±0.57	6.98±0.57	7.01±0.60	7.04±0.61	5.2-8 (pg)
	OCZ	6.92±0.68	6.95±0.70	6.98±0.72	7.03±0.74	7.07±0.76	
	OFZ-OCZ	6.88±0.63	6.94±0.71	7.00±0.76	7.04±0.81	7.03±0.81	
MCHC	OFZ	38.80±2.17	38.98±1.84	39.01±1.84	39.01±1.84	39.03±1.84	30-39 (g/dL)
	OCZ	38.65±2.71	38.60±2.71	38.58±2.70	38.51±2.67	38.47±2.66	
	OFZ-OCZ	38.61±2.37	38.50±2.35	38.39±2.33	38.26±2.30	38.21±2.26	
WBC	OFZ	13.32±3.45	13.31±3.46	12.91±2.67	12.66±2.38	12.40±1.98^{ab}	4-13.5 ($\times10^3$/µL)
	OCZ	12.73±2.59	12.58±2.63	12.50±2.61	12.36±2.63	12.16±2.70^{a}	
	OFZ-OCZ	13.49±1.49	13.49±1.49	13.54±1.55	13.44±1.54	13.42±1.55^{b}	
Lenfosit	OFZ	8.82±1.66	8.85±1.66	8.69±1.37	8.62±1.28	8.51±1.03	2-9 ($\times10^3$/µL)
	OCZ	8.26±1.82	8.23±1.81	8.23±1.81	8.19±1.81	8.13±1.81	
	OFZ-OCZ	8.63±1.23	8.72±1.29	8.92±1.39	8.98±1.38	9.00±1.30	
Monosit	OFZ	0.68±0.21	0.67±0.22	0.65±0.18	0.63±0.18	0.61±0.17^{ab}	0-0.65 ($\times10^3$/µL)
	OCZ	0.57±0.16	0.55±0.16	0.55±0.16	0.53±0.16	0.51±0.17^{a}	
	OFZ-OCZ	0.69±0.16	0.69±0.16	0.68±0.15	0.66±0.15	0.66±0.15^{b}	
% Granülosit	OFZ	26.73±6.95	26.45±7.25	25.99±6.82	25.43±6.60	24.94±6.75^{a}	%11-60
	OCZ	29.05±7.16	28.46±7.45	28.14±7.53	27.73±7.60	27.07±7.95^{a}	
	OFZ-OCZ	32.79±6.34	32.13±6.74	31.16±6.49	30.35±6.50	29.89±5.81^{b}	
% Lenfosit	OFZ	68.40±7.35	68.73±7.75	69.25±7.31	69.90±7.18	70.47±7.43^{a}	%50-70
	OCZ	66.68±7.23	67.38±7.66	67.73±7.75	68.23±7.84	68.94±8.27^{a}	
	OFZ-OCZ	62.40±6.71	63.06±7.06	64.14±6.62	65.05±6.68	65.50±5.99^{b}	
% Monosit	OFZ	4.87±1.05	4.82±1.13	4.80±1.12	4.72±1.22	4.64±1.28	%0-5
	OCZ	4.28±0.91	4.17±1.03	4.14±1.03	4.05±1.04	3.99±1.08	
	OFZ-OCZ	4.82±0.88	4.81±0.87	4.70±0.72	4.60±0.77	4.60±0.77	
% RDW	OFZ	29.71±1.38	29.63±1.47	29.67±1.46	29.71±1.44	29.76±1.43	%17.8-24.4
	OCZ	29.55±1.16	29.60±1.14	29.64±1.11	29.59±1.09	29.52±1.08	
	OFZ-OCZ	29.75±1.05	29.60±1.21	29.43±1.33	29.28±1.44	29.29±1.44	

Aynı sütundaki farklı harfler (^{a,b}) istatistiki açıdan önemlidir ($p<0.05$). RD: Referans değer, *(Bülbül, 2013; Aiello & Moses, 2016).

Tablo 4. Keçilere oral yolla, tek doz oksfendazol (OFZ, 7.5 mg/kg), oksiklozanid (OCZ, 15 mg/kg) ve oksfendazol-oksiklozanid kombinasyonu (OFZ-OCZ, 7.5 mg/kg-15 mg/kg) uygulandıktan sonra biyokimyasal parametreler üzerine etkileri (Ortalama ± SS)
Table 4. Effects on biochemical parameters after oral administration of a single dose of oxfendazole (OFZ, 7.5 mg/kg), oxyclozanide (OCZ, 15 mg/kg) and oxfendazole-oxyclozanide combination (OFZ-OCZ, 7.5 mg/kg-15 mg/kg) to goats (Mean ± SD)

Parametre	İlaç Grupları	0.saat	8.saat	24.saat	72.saat	7.gün	RD*
ALB	OFZ	2.37±0.57	2.32±0.50	2.3±0.45	2.31±0.45	2.32±0.45	2.3-3.6 (g/dL)
	OCZ	2.39±0.33	2.39±0.33	2.42±0.31	2.43±0.30	2.46±0.30	
	OFZ-OCZ	2.34±0.35	2.36±0.38	2.36±0.38	2.39±0.38	2.40±0.39	
ALP	OFZ	340.42±374.10	293.46±257.0	299.19±261.51	273.81±195.75	266.88±185.92	93-387 (u/L)
	OCZ	323.90±236.63	305.05±203.83	318.25±203.89	297.78±158.84	292.98±149.86	
	OFZ-OCZ	342.27±309.53	335.77±300.23	323.04±282.22	316.96±276.34	307.69±264.06	
ALT	OFZ	19.85±7.56	19.92±7.53	19.31±6.49	19.73±6.21	20.19±5.73	15-52 (u/L)
	OCZ	20.30±5.85	20.53±5.54	20.42±5.58	20.65±5.30	21.03±4.75	
	OFZ-OCZ	17.50±5.69	18.00±5.48	18.35±5.23	18.77±4.79	19.08±4.37	
AST	OFZ	72.35±16.44	71.04±14.48	70.81±14.01	71.23±14.61	72.35±15.05	66-230 (u/L)
	OCZ	77.50±13.52	77.42±13.53	77.65±13.45	77.31±13.47	78.0±13.22	
	OFZ-OCZ	74.12±13.32	75.42±14.51	75.92±14.86	77.35±15.05	78.27±14.80	
BUN	OFZ	13.54±6.94	12.77±6.09	12.85±6.14	13.12±6.85	13.27±7.03	10-28 (mg/dL)
	OCZ	14.45±6.42	14.34±6.57	13.99±6.77	14.49±6.79	14.38±6.71	
	OFZ-OCZ	13.42±6.86	14.77±8.23	15.73±8.50	16.42±8.24	16.54±8.15	
CH	OFZ	70.85±31.66	66.77±24.71	65.38±22.96	63.96±20.37	62.58±18.70	62-136 (mg/dL)
	OCZ	66.76±17.44	65.30±16.73	66.26±15.47	65.34±14.89	64.92±14.73	
	OFZ-OCZ	67.73±22.12	68.19±22.85	68.35±23.02	69.62±24.06	68.92±23.80	
CH	OFZ	0.62±0.11	0.61±0.10	0.60±0.09	0.61±0.10	0.61±0.10	0.6-1.5 (mg/dL)
	OCZ	0.61±0.07	0.61±0.07	0.62±0.06	0.62±0.06	0.62±0.07	
	OFZ-OCZ	0.62±0.06	0.62±0.07	0.62±0.07	0.62±0.07	0.62±0.07	
TP	OFZ	6.27±1.39	6.10±1.23	6.0±1.02	6.01±1.01	6.01±1.01	6.1-7.5 (g/dL)
	OCZ	6.07±0.84	6.08±0.84	6.13±0.81	6.15±0.81	6.21±0.78	
	OFZ-OCZ	6.10±0.91	6.14±0.94	6.15±0.94	6.20±0.92	6.22±0.92	
TRIG	OFZ	13.42±8.0	12.42±6.87	11.92±6.61	11.92±6.61	12.0±6.66	5.0-52.5 (mg/dL)
	OCZ	11.63±5.66	10.90±4.91	10.98±4.86	11.25±4.83	11.32±4.80	
	OFZ-OCZ	10.15±5.73	11.00±6.78	11.65±6.79	12.00±6.72	12.38±7.0	

RD: Referans değer, *(Bülbül, 2013; Aiello & Moses, 2016).

GEREÇ ve YÖNTEM

Araştırma, öncesinde klinik olarak sağlıklı oldukları belirlenen, 1-3 yaş aralığında, dişi, İvesi koyun (56±14 kg, n=6) ile Alpin keçi (38±2 kg, n=6) ırkı üzerinde gerçekleştirildi. Deneme süresince koyun ve keçiler ayrı padoklarda tutuldu. Hayvanlar yonca otu ve konsantre yemle beslendi ve su ad libitum olarak verildi.

Araştırma, koyun ve keçilerde uygulamalar arası 21 gün olmak üzere, üç periyotta, çapraz dizaynda gerçekleştirildi. Hayvanlara OFZ (375 mg oksfendazol, Anadolu İlaç ve Kimya San. A.Ş., Konya, Türkiye) 7.5 mg/kg dozda (Köse ve ark., 2007), OCZ (750 mg oksiklozanid, Anadolu İlaç ve Kimya San. A.Ş., Konya, Türkiye) 15 mg/kg dozda (Gokbulut ve ark., 2014), OFZ (7.5 mg/kg) ve OCZ (15 mg/kg) kombinasyonunun tablet formu (375 mg oksfendazol, 750 mg oksiklozanid, Anadolu İlaç ve Kimya San. A.Ş., Konya, Türkiye) oral yolla, tek doz uygulandı. Uygulamadan önce (0.saat, kontrol) ve sonrası 8., 24., 72. saat ve 7. günde kan örnekleri hematolojik analizler için K3EDTA'lı ve biyokimyasal analizler için jelli tüplere alındı. Hematolojik parametreler için alyuvar sayıları (RBC), hemoglobin (HGB), hematokrit (HCT), ortala-ma korpüsküler hacim (MCV), ortalama korpüsküler hemog-lobin (MCH), ortalama korpüsküler hemoglobin konsantras-yonu (MCHC), akyuvar sayıları (WBC), lenfosit, monosit, % granülosit, % lenfosit, % monosit ve % eritrosit dağılım geniş-liği (% RDW) değerleri kanların alındığı süreler içinde otoma-tik hematoloji analizatöründe (Exigo,Boule Medical AB, Span-ga, İsveç) ölçüldü (Kandir & Keskin, 2016). Antikoagülan içermeyen jelli tüpler ise serum elde etmek için 3000 g'de 10 dakika santrifüj edildi ve analize kadar -70 °C'de saklandı. Bi-yokimyasal parametrelerden albümin (ALB), alkalen fosfataz (ALP), alanin aminotransferaz (ALT), aspartat aminotransfe-raz (AST), kolesterol

(CH), trigliserid (TRIG), total protein (TP), kan üre nitrojen (BUN), kreatinin (CR) parametreleri otoanalizatör cihazında (Zybio ZS 400, Çin) belirlendi..

Çalışmada tüm değerler ortalama±standart sapma (SS) olarak gösterildi. Parametreler arasındaki istatistiksel farklılıklar tek yönlü varyans analizi (One-Way ANOVA) testi ile belirlendi. Posthoc test olarak ise Tukey testi kullanıldı (IBM SPSS 23.0). P değerlerinin <0.05 olması, istatistiki açıdan önemli kabul edildi.

BULGULAR

Koyun ve keçilere OFZ, OCZ ve OFZ-OCZ kombinasyonu uygulandıktan sonra hayvanların genel durumlarında hiçbir istenmeyen reaksiyon gözlemlenmemiştir. Koyunlardan elde edilen hematolojik ve biyokimyasal parametreler sırasıyla Tablo 1 ve 2, keçilerden elde edilen hematolojik ve biyokimyasal parametreler sırasıyla Tablo 3 ve 4'de sunulmuştur. Bu çalışmada koyun ve keçilere sırasıyla 7.5 ve 15 mg/kg dozlarda OFZ, OCZ ve OFZ-OCZ kombinasyonu uygulandığında hematolojik ve biyokimyasal parametreler üzerinde klinik olarak anlamlı bir değişikliğe neden olmadığı belirlenmiştir (P>0.05).

Koyunlarda hematolojik parametreler değerlendirildiğinde zamana ve ilaç gruplarına bağlı olarak istatistiki olarak anlam ifade etmeyen ve fizyolojik sınırlar içerisinde kalan dalgalanmalar izlenmiştir (Tablo 1).

Keçilerde ise referans aralık içerisinde WBC ve monosit (OCZ ile OFZ-OCZ grubu arasında), % granülosit ve % lenfosit (OFZ ile OFZ-OCZ grubu ve OCZ ile OFZ-OCZ grubu arasında) düzeylerinde gruplar arası 7. günde (Tablo 3) istatistiksel olarak farklılık belirlenmiştir (P<0.05).

Koyun ve keçilerde zamana ve ilaç gruplarına bağlı olarak biyokimyasal parametrelerde (Tablo 2 ve Tablo 4) referans aralık içerisinde istatistiksel farklılık gözlenmemiştir (P>0.05).

TARTIŞMA

Antelmentik ilaçlar, veteriner hekimlikte kemoterapötikler içerisinde en sık kullanılan ilaçlar arasında yer almaktadır. Antelmentik ilaçlar içerisinde benzimidazoller, çiftlik hayvanlarında iç parazit infestasyonlarının kontrolü ve tedavisinde yaygın olarak kullanılmaktadır. Oksfendazol, metil-5 (6)-fenilsulfinil-2-benzimidazol karbamat, benzimidazol antelmentik ailesinin bir üyesidir. Fenbendazolün sülfoksit metabolitidir ve fenbendazolün etki spektrumunda oksfendazol önemli rol oynar. Aynı zamanda ön-benzimidazollerden febantel metabolize edildiğinde fenbendazol ve son olarak da oksfendazole dönüşür (El-Makawy ve ark., 2006). Oksfendazol gruptaki diğer benzimidazollerle karşılaştırıldığında gastrointestinal sistemden emilimini takiben maksimum kan konsantrasyonuna daha yavaş ulaşır; böylece hem serum da hem de intestinal sistemde etkili konsantrasyon düzeyini daha uzun süre koruyarak etkinliğini artırır (Prichard & Ranjan, 1993).

Yapılan literatür taramalarında çiftlik hayvanlarında yaygın olarak kullanılan OFZ ve OCZ'nin hematolojik ve serum biyokimya düzeyleri üzerine etkisi ile ilgili yeterli sayıda çalışma bulunmadığı değerlendirilmiştir. Bu sebeple, araştırmamızda koyun ve keçilerde OFZ, OCZ ve kombinasyonunun önerilen dozlarda uygulandığında, farklı örnekleme zamanlarında hematolojik ve biyokimyasal parametrelerde meydana getirdiği değişimler incelenmiştir. Elde edilen bulgulara göre OFZ, OCZ ve OFZ-OCZ'nin koyun ve keçilerde farklı zaman aralıklarında özellikle WBC, monosit, % granülosit, % lenfosit, % monosit değerlerinde dalgalanmalara sebep olduğu ancak bu durumun fizyolojik sınırlar içerisinde kaldığı izlenmiştir.

OFZ, 14 gün süreyle 33 mg/kg dozda Sprague-Dawley ratlarına uygulandığında hematolojik parametrelerde değişikliklere neden olmuştur. Long-Evans ratlarında yapılan üç aylık bir çalışmada, 600 mg/kg dozda yem içerisinde verildiğinde hepatik hipertrofi, vakuolasyon ve hepatik nekroz gözlenmiştir. Diğer patolojik değişiklikler arasında testiküler atrofi, splenik nekroz, kemik iliği hiperplazisi kaydedilmiştir. Yeme 200 mg/kg dozda katıldığında ise sadece hafif hepatik hipertrofi gözlenmiştir (EMEA, 2004). OFZ bir ay süreyle 3000 mg/kg, üç ay süreyle 75 mg/kg dozda diyete katılarak farelere uygulandığında, farelerin karaciğer ağırlıklarında artış, biyokimyasal değerlerde ve yağ vakuolasyonunda artış tespit edilmiştir (EMEA, 2004). El-Makawy ve ark. (2006) yaptıkları çalışmada; erkek Swiss farelerine sekiz hafta süreyle, dişi Swiss farelerine ise dört hafta süreyle oral yolla (gavaj), günlük 0.01 mL dozda OFZ uygulamışlardır. OFZ uygulanan fareler kontrol grubuyla karşılaştırıldığında karaciğer, böbrek ve testislerin total protein içeriğinde istatistiksel olarak anlamlı bir artış tespit edilmiştir. Ayrıca OFZ uygulanan hem erkek hem de dişi farelerde ALT ve AST düzeyleri önemli ölçüde yüksek belirlenmiştir. Codd ve ark. (2015) Sprague-Dawley ratlara (üç dişi, üç erkek) oral yolla 5, 25, 100 ve 200 mg/kg dozda OFZ'yi 14 gün süreyle uygulamışlardır. Dişilerde 15. günde WBC, lenfosit, nötrofil, monosit, eozinofil ve bazofil sayısında doza bağlı olarak düşüş belirlenmiştir. Dişilerde WBC düzeyindeki düşüş doza bağlı olarak artmıştır. Erkek ratlarda ilaca bağlı olarak görülen etkiler çok daha az şiddetli olmuştur ve sadece 200 mg/kg doz grubunda WBC düzeyinde düşüş belirlenmiştir. RBC üzerinde de etkiler kaydedilmiştir ancak kaydedilen bulgu, WBC düzeyine göre daha az önemli bulunmuştur. Elde edilen veriler araştırmacılar tarafından toksikolojik nitelikte kabul edilmemiştir. Bir başka çalışmada OFZ oral yolla, 0.5, 1, 3, 7.5, 15, 30 ve 60 mg/kg dozlarda, tek sefer, 14 gün süreyle sağlıklı bireylere uygulanmıştır. OFZ uygulanan 70 bireyin 40'ında anormal klinik laboratuvar değerleri kaydedilmiştir. Bu bulgular arasında orta dereceli lökositoz veya lökopeni, nötropeni, eozinofili, bikarbonat ve AST düzeyinde değişiklikler kaydedilmiştir. Kaydedilen bu veriler normal aralığın dışında olmasına karşın, uygulanan OFZ dozlarının toksisite oluşturacak düzeyde olmadığı bildirilmiştir (An ve ark., 2019). OFZ'nin toksik ve kardiyovasküler sistem üzerine istenmeyen etkilerini ortaya koymak amacıyla, Beagle ırkı köpeklere oral yolla 0, 5, 25 ve 100 mg/kg dozda tek sefer OFZ uygulanmıştır. Hematolojik, biyokimyasal ve koagülasyon parametrelerini içeren klinik laboratuvar değerlendirmelerinde herhangi bir değişiklik kaydedilmediği bildirilmiştir (Lead & Deye, 2015). İki hafta süreyle 0-200 mg/kg dozda, oral yolla OFZ uygulanan ratlarda; WBC değerinde doza bağlı olarak düşüş belirlenmiştir. OFZ uygulaması sonlandırıldığında ise değerler normale dönmüştür. Ayrıca biyokimya sonuçları değerlendirildiğinde OFZ'nin karaciğer enzimleri üzerinde hiçbir etkisi olmadığı görülmüştür (Lead & Deye, 2015). Rat

ve köpeklere iki hafta süreyle 11-200 mg/kg arasında değişen dozlarda oral yolla OFZ uygulandığında, yüksek dozlarda nötrofil sayısı, hemoglobin ve hematokritte düşüş tespit edilmiştir (Lead & Deye, 2015).

OCZ, salisilanid grubunda yer alan sığır, koyun ve keçilerde fasciolosis tedavisi ve kontrolünde kullanılan bir antelmentiktir (EMEA, 1998). OCZ'nin glukuronid metaboliti belirlenmiş olmasına rağmen salisilanilidlerin çoğunlukla metabolize olmadığı varsayılmaktadır (Broome & Jones, 1966). Bu metabolit safrada konsantre olmasına rağmen antelmentik olarak aktif değildir (McKellar & Kinabo, 1991).

OCZ, dişi ve erkek ratlara 74, 185 ve 370 mg/kg dozlarda, 28 gün süreyle günde bir defa oral yolla (gavaj) uygulanmıştır. Yapılan bu çalışmada hem erkek hem de dişi ratlarda tüm doz seviyelerinde hematolojik parametrelerin düzeyleri, referans aralıklar içerisinde belirlenmiştir. Erkek ratlarda 185 mg/kg doz düzeyinde AST, 370 mg/kg doz düzeyinde ise hem AST hem de ALP yüksek tespit edilmiştir. Dişi ratlarda ise 185 mg/kg doz düzeyinde ALP, 370 mg/kg doz düzeyinde ise AST ve ALP yükselirken, TRIG düzeyinde azalma kaydedilmiştir. Ayrıca 370 mg/kg doz düzeyinde BUN referans aralık içerisinde kontrol grubuna kıyasla yüksek kaydedilmiştir (Wang ve ark., 2019). AST ve ALT'nin ilgili hücre ve dokular zarar gördükten sonra seruma salındığı bilinmektedir (Wallig ve ark., 2017). Çoğu toksikolojik çalışmada ALT artışı, genellikle hepatosit hasarının spesifik bir göstergesidir. Hepatosit hasarı sırasında serum AST miktarı artabilir. Bu artış, AST'nin kastaki yüksek aktivitesi nedeniyle spesifik değildir. Karaciğer hasarında AST genellikle serum ALT aktivitesine paraleldir. Genel olarak AST konsantrasyonundaki yükselme, etkilenen hepatositlerin sayısı ile ilişkilidir ve patolojik temelde lezyonun ciddiyetini veya geri dönüşümlülüğünü yansıtmaz. OCZ'nin ratlarda ALP artışına ve karaciğerde toksik etkilere neden olduğu bildirilmiştir (Walley, 1966). ALP aktivitesinin artması kolestazın bir göstergesidir. İlaca bağlı hepatosit nekrozu da ALP'nin aktivitesini artırabilir (Foster, 2005). BUN, klinik tanıda böbrek fonksiyonunu değerlendirmek için kullanılan bir belirteçtir (Wallig ve ark., 2017). Wang ve ark. (2019)'nın yaptıkları çalışmada yüksek doz OCZ grubunda (hem erkek hem dişi) BUN düzeyinin yükselmesi, böbrekte inflamasyon veya toksin üretimi nedeniyle nefronlarda hasar meydana geldiğini gösterebilir (21). Bir başka çalışmada Simental ırkı dişi ve erkek sığırlara 0, 10, 30 ve 50 mg/kg dozda, oral yolla oksiklozanid uygulanmıştır (Dong ve ark., 2019). Araştırmacılar hematoloji ve serum biyokimya testlerinde tedavi ile kontrol grupları arasında WBC, lenfosit, RBC, HGB, MCHC, TP, ALB, AST, ALP değerlerinde bazı önemli farklılıklar kaydetmişlerdir. Ancak bu farklılıkların doza veya zamana bağlı olmadığını, referans aralıkta olduğunu ve/veya farklılıkların ihmal edilebilecek düzeyde olduğunu belirtmişlerdir.

Deney hayvanlarında yapılan çalışmalar incelendiğinde OFZ ve OCZ'nin yüksek dozlarda hematoloji ve biyokimya sonuçlarını etkilediği, bu çalışmaların çoğunda da bulguların referans aralıklar içerisinde olduğu görülmüştür. Karaciğer ve böbrek, ilaç etkilerine karşı önemli ve hassas organlardır (Lin ve ark., 2018). Serum biyokimya düzeylerinde farklılık belirlenmemesi, uygulanan dozlarda karaciğer ve böbrek hasarının oluşmadığını göstermektedir.

SONUÇ

Sonuç olarak, koyun ve keçilere oral yolla 7.5 mg/kg OFZ ve 15 mg/kg OCZ, tek ve kombine olarak uygulandığında hemogram ve biyokimyasal parametreler üzerine belirgin olumsuz etkilerinin gözlenmediği ifade edilebilir. Sunulan araştırmada elde edilen bulgular koyun ve keçiler için OFZ, OCZ ve OFZ-OCZ'nin önerilen dozlarda kullanımının hematolojik ve biyokimyasal parametreler üzerinde literatür veride bahsedilen değerlerde özellikle 24. saat ve 7. gün aralığında çeşitli dalgalanmalara yol açtığı gözlense de bu değerlerin fizyolojik sınırlar içerisinde kalması sebebiyle göz ardı edilebilir düzeyde olduğu belirtilebilir.

BEYANNAMELER

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Çıkar çatışması

Yazarlar arasında herhangi bir çıkar çatışması yoktur.

Yazar katkıları

Fikir, Kavram ve Tasarım: ZÖK, SK

Veri Toplama ve Analiz: ZÖK, SK, ÇA

Makalenin Yazımı: ZÖK

Eleştirel İnceleme: SK

Veri kullanılabilirliği

Bu çalışmanın bulgularını destekleyen veriler makul talep üzerine sorumlu yazardan temin edilebilir.

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Research Article

The seroprevalence of Francisella tularensis in horse herds in Turkey

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ABSTRACT

Tularemia, caused by Francisella tularensis, can exist in nature over a long period of time. The disease can be transmitted by ticks, biting flies, contaminated food and water, and inhalation. F. tularensis infections are particularly common in North America, Europe, and Asia. Tularemia is often water-associated and affects humans and an array of animals, including domestic animals, small wild mammals and fish. The literature about tularemia in horses is limited; however, fever, dyspnea, incoordination and depression have been reported. This study aimed to estimate the seroprevalence of F. tularensis in horse herds in Turkey. A total of 109 horses, aged 36 months and older, were randomly sampled from different regions of Turkey. The serum samples were tested for the presence of antibodies to F. tularensis, using the Microagglutination Test (MAT), which has 51% sensitivity and 99% specificity. The overall (animal), within-herd and between-herd apparent seroprevalence values were calculated as 40.4% (95% CI = 31.6 to 49.8%), 41.1% (95 CI = 32.3 to 50.6%) and 81.8% (95% CI = 52.3 to 94.9%), respectively. The Rogan-Gladen estimator was then used to correct the apparent seroprevalence values to true seroprevalence values of 78.7% overall (95% CI = 61.3 to 97.5%), 80.2% within-herd (95 CI = 62.5 to 99.2%), and 161.6% between-herd (95 CI = 103 to 187.7%). The results provide useful information regarding the prevalence of tularemia in horse herds in Kafkas University Faculty of Veterinary Medicine Department of Microbiology, which it is hoped will attract the particular attention of veterinarians, enabling the establishment of an efficient control program.

INTRODUCTION

Tularemia is caused by a nonmotile, pleomorphic, Gram-negative coccobacilli bacteria called Francisella tularensis (F. tularensis), and is a common zoonotic infectious disease predominantly seen in the Northern Hemisphere (WHO, 2007; Mead, 2008). The disease is known by various names, such as Francis disease, Ohara disease, Rabbit fever-plague, Horsefly fever, Siberian ulcer and Hunter's disease (Kubelkova, 2015). F. tularensis is a resistant bacterium that can survive in cold and humid environments for weeks; however, it is not resistant to sunlight, high temperature or chlorination (Dikici, 2012). Transmission may occur by inhalation, ingestion, contact with infected animals, or via the bites of arthropod vectors (Arslanyılmaz,2014; CDC, 2018). Rodents (rats, mice and squirrels) and rabbits (Lagomorpha) are the most important reservoirs for tularemia (CDC, 2018). The existence of tularemia in sheep, cattle, pigs and horses was confirmed for the first time in the USSR (Pollitzer, 1963). Observation of tularemia in these animals led to the conclusion that Ixodes ticks, which were prevalent from 1939 to 1941 played an interepidemic role in the infection (Pollitzer, 1963). Tularemia is rarely reported in horses, but its occurrence is often accompanied by a severe tick infestation. Signs of tularemia in horses apparently include fever, shortness of breath (dyspnea), incoordination, depression, ataxia, and edema of the legs. Intense tick infestation and seroconversion have generally been observed in horses (Otlu, 2009). Death can occur within a day. In some cases, tularemia may be present in asymptomatic horses. Generally, animals with the disease are treated with agent-specific antibiotics such as streptomycin. Although it is difficult to control the disease in horses, this can be achieved by reducing tick infestation and rapid diagnosis and treatment. Animals that have recovered from the disease develop long-term immunity (Foley, 2019).

The diagnosis of tularemia can be performed with serological tests, such as the widely-used Microagglutination test (MAT), Hemagglutination test (HA) and the Enzyme-Linked Immuno-Sorbent Assay (ELISA). Among these, the MAT, using a stained F. tularensis antigen, is the most common tool for diagnosis (Arslanyılmaz, 2014).

The aim of this study was to determine the presence of antibodies to F. tularensis in horses by MAT and thus estimate the seroprevalence of tularemia in Turkey.

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MATERIAL and METHODS

Study design and Sample Collection

This study was approved by the local ethical committee. (Protocol no: 2021/17 Date: 05.11.2021 VKMAE). Blood samples for the study were collected from 11 different regions of Turkey, between 2021 and 2022, from ≥36 month-old horses, randomly selected from farms that implemented an extensive rearing system (stock farming mainly based on pasture and meadows) (Table 1).

A total of 109 blood samples, taken from horses with no history of vaccination against tularemia, were submitted to the laboratory. Serum samples were obtained by centrifugation at 3000 rpm for 10 min and stored at -20 °C, pending analysis.

Microagglutination Test (MAT)

A Microagglutination Test (MAT) was used for the detection of antibodies to F. tularensis in horse blood sera. MAT was performed with an antigen prepared from a standard strain of F. tularensis strain (NCTC 10857) (Arslanyılmaz, 2014). First, 40 µl saline buffer was put into the first well of the U-bottomed plate. Twenty-five µl of saline was put into the next 6 wells for sample sub-dilutions. Twenty-five µl of positive serum (1:160 titer) and 25 µl of saline were put into the 8th and 9th wells, respectively, for positive and negative control. Ten µl of test serum was introduced into the first well, and 25 µl of liquid content was transferred from the first well to the next, continuing to the 6th sub-dilution. Then, all of the wells, including the positive and negative controls, received 25 µl of stained antigen and, thus, 1:10 to 1:640 sub-dilutions were obtained. The test plate was put into a humidified box and incubated at 37°C overnight. Agglutination of the antigen-antibody complex in a net-like form, leaving a completely clear supernatant, was considered a positive reaction. Agglutination in a small, centrally-gathered smooth-edged form, surrounded by light red diluents, was evaluated as a negative reaction (Karataş Yeni, 2015; Kılıç, 2013).

The MAT test for F. tularensis has been shown to cross react with Brucella spp. at titers of up to 1:20 (Karataş yeni, 2015; Kılıç, 2013). Therefore, in this study, horse blood serum samples with a titer of 1:20, were subjected to the Brucella Microagglutination Test (Kılıç, 2013).

Statistical analysis

The data were statistically analyzed with the SPSS® Version 20. MAT results were evaluated by the Chi-square test (Preacher, 2001) and p-values smaller than 0.05 were assumed significant. The cut-off values established by Maurin (Maurin, 2020) were used for MAT sensitivity and specificity. The case definition and subsequent serial calculations of the apparent individual and mass prevalences (within-herd and between-herd) were carried out by the method reported by Buyuk et al. (2014). The true seroprevalence values for the animals overall, within-herd, and between-herd were calculated using the Rogan-Gladen estimator (Rogan, 1978).

RESULTS

Of the 109 horse blood serum examined for tularemia, 44 (40.4%) were found positive for F. tularensis with a titer of ≥1:20. When evaluated in terms of F.tularensis seropositivity, the p value was determined as >0.05. No statistical significance was observed among 11 different regions.The antibody titer distribution of the serum was 1:20 in 34 sera, 1:40 in 8 sera and 1:80 in 2 sera. In the Brucella Micro-Agglutination test results, a 1:10 titer was found in 4 samples. According to these results, the cross-reaction cut-off values were insignificant. The overall (animal), within-herd, and between-herd apparent seroprevalence values were calculated as 40.4% (95% CI = 31.6 to

Table 1. Sample distribution among the location and the results of the MAT survey for *F. tularensis*

Location	Number of samples	Number of seropositive samples	Apparent prevalence		True prevalence	
			Estimate, %	95% CI	Estimate, %	95% CI
Location 1	1	-	-	-	-	-
Location 2	1	1	100	20.7-100	100.7	-88-108
Location 3	1	-	-	-	-	-
Location 4	2	2	100	34.2-100	198	66.5-198
Location 5	2	2	100	34.2-100	198	66.5-198
Location 6	3	2	66.7	20.8-93.9	131.3	39.5-194.6
Location 7	7	1	14.3	2.6-51.3	26.6	-0.5-100.6
Location 8	10	2	20	5.7-51	38	9.3-100
Location 9	15	12	80	54.8-93	158	107.6-184
Location 10	25	12	48	30-67	94	58.1-131
Location 11	42	10	23.8	13.5-38.5	45.6	25-75.1
Total	109	44	40.4	31.6-49.8	78.7	61.3-97.5

49.8%), 41.1% (95 CI = 32.3 to 50.6%) and 81.8% (95% CI = 52.3 to 94.9%), respectively (Table 1).

True seroprevalence values were estimated by conversion from the apparent seroprevalence values using the Rogan-Gladen estimator. The true overall (animal), within-herd, and between-herd seroprevalence values were calculated as 78.7% (95% CI = 61.3 to 97.5%), 80.2% (95 CI = 62.5 to 99.2%) and 161.6% (95 CI = 103 to 187.7%), respectively (Table 2).

Table 2. Prevalence estimates of *F. tularensis* among animals, within-herds and between-herds

Prevalence type	Number tested	Number positive for F. tularensis	Apparent prevalence		True prevalence	
			Estimate, %	95% CI	Estimate, %	95% CI
Overall (Animal)	109	44	40.4	31.6-49.8	78.7	61.3-97.5
Within-herd	107	44	41.1	32.3-50.6	80.2	62.5-99.2
Between-herd	11	9	81.8	52.3-94.9	161.6	103-187.7

DISCUSSION

Tularemia is a zoonotic disease caused by F. tularensis, which is found worldwide, including in Turkey. It is also a potentially significant biological weapon. Amongst domestic animals, sheep are the primary host; however, tularemia has also been reported in dogs, cats, pigs, and horses (Otlu, 2009; Gese, 1997; Mörner, 1983). Some animals are highly susceptible to tularemia and, if infected, usually die before F. tularensis antibodies have even formed. However, antibodies are detectable in species such as cattle, sheep, dogs, pigs and horses by agglutination (MAT and tube agglutination tests) techniques (OIE, 2009; Bevanger, 1988; Arata,1973, Celebi, 2013). There are very few studies reporting clinical and pathological presentations of tularemia in horses infected experimentally or naturally. Generally, natural infection follows a heavy tick infestation and courses fever, dyspnea, incoordination, depression, and sudden death in horses (Tokgöz, 1938; Jellison, 1958; Claus, 1959; Cino, 2021). At autopsy, swelling and numerous necrotic foci in the lung, the liver and the spleen and diffuse necrosis in the intestinal lymph nodes are observed (Jellison, 1958; Cino, 2021). The existence of Tularemia in horses indicated by these clinical and pathological symptoms was made absolute by both the agent isolation and PCR or immunohistochemical methods (Jellison, 1958; Claus,1959; Cino, 2021).

Studies about the seroprevalence of tularemia in horses are very limited (Celebi, 2013; Tokgöz, 1938; Jellison, 1958, Stark, 1979). Horses, which are relatively more resistant to infection than other livestock, can develop a detectable antibody response to F. tularensis and can be detected for diagnostic purposes over a period. As a matter of fact, antibody titers decrease over time in horses, as in sheep cases, and turn negative within months (Jellison, 1958). The antibody titers have been reported in horses exposed to the agent or surviving between 1:10 and 1:640 (Celebi, 2013; Tokgöz, 1938; Jellison, 1958). A comprehensive study of the presence of F. tularensis antibodies in livestock, including horses, was conducted by Celebi et al. (2013) in the Kars Region of Turkey and a 50% (15/30 horses) seropositivity was obtained with antibody titer ranging from 1:10 to 1:40. In a study conducted in the USA, the presence of the causative agent in wild horses was demonstrated serologically with a rate of less than 11% (Stark, 1979). In the present study, the overall apparent and true seroprevalence was 40.4% and 78.7%, respectively. Although the cut-off value was taken as 1:20, the seropositivity rate was quite close to the rate reported by Celebi et al. (2013). However, the prevalence differences between the farm horses and the wild horses may have emerged from the geographical differences and the species and population intensity of ticks in these habitats. Considering the highly contagious nature of tularemia in both humans and animals, the different positivity rates of within-herd and between-herds, which are higher than the individual prevalence, are remarkable in this respect (Table 2).

Although serological methods, especially MAT, are widely used in the diagnosis of infections caused by such bacterial agents, which are difficult to culture, they have some disadvantages in terms of both cross-reaction possibilities with bacteria of close antigenic structure similarities and low diagnostic capabilities (OIE, 2018). Therefore, the corrected prevalence values calculated with the Rogan-Gladen estimator provide realistic diagnostic values that will enable us to accurately estimate the prevalence of the causative agent by eliminating such handicaps of the MAT. By its corrected version, the prevalence estimates of F. tularensis were detected at higher rates, revealing the common carriage of the causative agent in horses and their potential roles in possible transmissions.

CONCLUSION

As a result, a large-scale study representing the whole of Turkey was conducted for the first time in horses and F. tularensis seropositivity was found to be high. More comprehensive studies are needed to fully establish the degree of roles of horses in the eco-epidemiology of the disease.

DECLARATIONS

Ethics Approval

This study was approved by the local ethical committee (Protocol no: 2021/17 Date: 05.11.2021 VKMAE). Conflict of Interest

Authors declare that there are no conflicts of interest for this study.

Consent for Publication

Not applicable

Author Contribution

In all sections of the final article, each author contributed equally.

Data Availability

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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Research Article

The acute effect of thiamine on serum insulin levels and some biochemical parameters in excessive alcohol-consuming rats

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ABSTRACT

In the present study, it was aimed to investigate the effectiveness of thiamine in rats with a binge drinking model. For this purpose, a total of 21 Sprague Dawley rats were divided into three equal groups; control, alcohol, and thiamine+alcohol groups. The thiamine+ alcohol group was given thiamine at a daily dose of 100 mg/kg by oral gavage, starting 2 days before the alcohol administration. Alcohol and thiamine+alcohol groups were given 3.45g/kg/day alcohol as 20% for 3 consecutive days. At the end of the study, while serum total bile acid, total bilirubin, and insulin levels increased in rats in the alcohol group compared to the rats in the control group; total protein and albumin levels decreased ($p<0.05$). In the thiamine + alcohol group, LDL-cholesterol, total cholesterol, bile acid levels, and AST enzyme activity increased, while ALT enzyme activity and total protein levels decreased compared to the control group ($p<0.05$). There was no statistically significant result in the values in the thiamine + alcohol group compared to the alcohol group. It might be concluded that acutely administered thiamine supplementation had no effect on alcohol-induced biochemical parameter changes in binge drinking animals. In this positive effect, studies with longer-term thiamine use may be needed.

INTRODUCTION

It is reported that excessive alcohol consumption, which is among the highest risk factors for mortality, is an important predisposing agent for more than 200 diseases (O'Keefe et al, 2014). Although most of the studies focused on chronic alcohol consumption, the most common type of drinking especially in the young population is excessive alcohol consumption (Ngandeu et al, 2018). Excessive alcohol consumption is a concern especially in young individuals, particularly due to neurotoxic effects (Bajac et al 2016). Binge drinking, is a type of excessive alcohol consumption; has been defined as the consumption of five or more units of alcoholic beverage in a period of approximately two hours (Wechsler et al, 1994). Binge (excessive) drinking is considered a public health problem associated with metabolic diseases such as diabetes and obesity. In addition, the influence of binge drinking on glucose metabolism has not yet been fully elucidated (Naimi et al 2003).

The effects of alcohol on glucose metabolism and insulin secretion have been variously reported, depending on the amount of alcohol consumed, type of meal, and duration of drinking. Alcohol is metabolized in the body to acetaldehyde and acetic acid, and these metabolites and reactive oxygen species formed in the metabolic process can directly affect glucose metabolism. It is thought that the mechanism by which excessive drinking causes hyperglycemia is that alcohol directly inhibits glucose-stimulated insulin secretion and inhibits glycogen synthesis in the liver, preventing glucose utilization by oxidative and non-oxidative pathways. Also, although alcohol does not inhibit the binding of insulin itself, it can cause insulin resistance by causing abnormalities in intracellular signalling after insulin binds to insulin receptors in peripheral tissues. On the other hand, an increase in insulin secretion is also observed due to the intracellular signalling effect of alcohol (Jang and Eun, 2012).

Thiamine (Vitamin B1) is a necessary vitamin for all tissues for the continuity of the activities of different enzymes involved in the metabolism of carbohydrates. Thiamine is converted to thiamine pyrophosphate (the active form of thiamine) by the enzyme thiamine diphosphokinase in the liver (Bettendorff et al., 1996). Conversion of thiamine to its active form is decreased, especially in liver damage as a result of chronic alcohol consumption. In cases of thiamine deficiency, disorders occur in the pathways in the synthesis of nucleic acids, fatty acids, etc. in the body because thiamine is needed for the functions of various enzymes necessary for the metabolism of cells (Chandrakumar et al. 2018). Three of these enzymes; the alpha-keto glutamate dehydrogenase, the pyruvate dehydrogenase complexes and transketolase play important roles in glucose and lipid metabolism. Alcohol damages the intestinal lining, causing disrupted thiamine absorption from the gut. Therefore, a deficiency of thiamine may be seen in the use of alcohol (Gastaldi et al. 1989). In addition, thiamine needs to

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be transformed into an active form in order to act as a cofactor of enzymes involved in lipid and glucose metabolism. The enzyme thiamine pyrophosphpkinase converts thiamine to thiamine pyrophosphate (the active form of thiamine). In excessive alcohol intake, this enzyme activity is impaired, therefore, there is a decrease in thiamine activity (Langlais, 1995). As a result of the above-mentioned mechanisms, it has been revealed that alcohol use can be caused thiamine deficiency and, as a result, deterioration in metabolic pathways.

This study was aimed to investigate the effect of thiamine supplementation on changes of some serum biochemical parameters levels and serum insulin hormone levels of high alcohol intake 1-2 times a month, which is very common in young population.

MATERIAL and METHODS

This study was carried out in Mehmet Akif Ersoy University's experimental animal breeding and experimental research center. (Ethical approval no: 688/2020)

Animals

Totally 21 adult female Spraque Dawley rats were used in the study. During the study, 7 rats were housed in each cage. Rats in plastic rat cages, at 23±2 °C of room temperature, in a 50±10% relative humidity environment, in a 12 h light/12 h dark cycle were fed ad-libitum. At the end of the one-week adaptation period, the rats were divided into three equal groups as control, alcohol, and thiamine+alcohol groups. The rats were given standard rat food and regular tap water ad libitum during the experiment. The thiamine+alcohol group was given thiamine hydrochloride (Sigma Aldrich, Germany) at a daily dose of 100 mg/kg by oral gavage, starting 2 days before the alcohol administration. Alcohol and thiamine+alcohol groups were given 3.45 g/kg/day of ethanol intraperitoneally. Ethanol administration to both groups continued for 3 days, and thiamine was continued to be given to the thiamine+alcohol group these days. In the control group, saline was administered intraperitoneally on the days given alcohol. At the end of the 5th day, blood samples were collected from the animals for biochemical analysis, and euthanasia procedures were performed.

Collection of Samples

At the end of the experiment, all rats in the study were collected blood by cardiac puncture under general anesthesia (thiopental anesthesia, 40 mg/kg) and the rats were sacrificed by cervical dislocation. Blood samples were centrifuged at 3000 rpm for 15 minutes for obtaining serum samples. Serum samples were stored at -20 °C until the analysis.

Biochemical Analysis

The analysis of biochemical parameters (Glucose, triglycerides, total-cholesterol (C), high density lipoprotein-cholesterol (HDL-C), low density lipoprotein-cholesterol (LDL-C), alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase (ALP), bile acid, total protein, total bilirubin, albumin) of blood serum samples were done with an autoanalyzer (Randox-Monaco, England) and were carried out with the spectrophotometric measurement method.

Serum Insulin Hormone Analysis

Insulin hormone levels in blood serum samples were measured with a microplate reader (Biotek-Epoch, USA) at 450 nm wavelength using the ELISA method using a commercial kit (BT-Lab, China) according to kit procedures.

Statistical Analysis

Firstly, a normality test was applied to determine whether the data were parametric or nonparametric. Since the number of data in each group was <30, the results were interpreted according to the Shapiro-Wishlk test in the normality test. In this case; since $p>0.05$ for glucose, triglycerides, total cholesterol (C), HDL-C, ALT, AST, bile acid, total protein, albumin, and insulin variables, these variables show normal distribution within the three groups. Since $p<0.05$ in the thiamine group for the LDL-C variable, the thiamine group for the ALP variable, and the thiamine group for the total bilirubin variable, it does not show a normal distribution for these thiamine groups but control and alcohol groups show normal distribution. In this case, one-way anova was applied to the normally distributed variables. While the thiamine group did not show normal distribution, the nonparametric kruskal wallis test was applied for 3 variables.

RESULTS

At the end of the study, while serum total bile acid, total bilirubin and insulin hormone levels increased in rats in the alcohol group compared to the rats in the control group; total protein and albumin values decreased ($P<0.05$). In the thiamine + alcohol group, LDL-C, total-C, bile acid levels and AST enzyme activity increased, while ALT enzyme activity and total protein levels decreased compared to the control group ($P<0.05$) (Table 1). In addition, there was no significant difference between the groups in serum glucose, triglycerides, HDL-C values and ALP enzyme activity (Table 1).

DISCUSSION

Chronic or acute alcohol consumption is known to damage various organs (Dguzeh et al., 2018). Since 95% of the alcohol consumed is metabolized in the liver, it has been proven that liver damage due to chronic and acute alcohol consumption is caused by the change in oxidative and energy metabolism in the liver (Lieber, 1997). These free radicals can damage numerous cellular components, primarily nucleic acids, lipids, and proteins. In this way, it contributes to the formation of many diseases, including alcoholic liver disease, by causing oxidative damage (Ward et al., 1989). There are agents that eliminate the harmful effects of oxidants and are described as antioxidants. While some of these agents are produced in the body endogenously, some of them must be taken exogenously from the diet (Bouayed et al., 2010). Vitamin B1, named as thiamine, belongs to the class of water-soluble vitamins that acts a significant role in energy metabolism, especially from carbohydrates (Portari et al., 2016). On the other hand, thiamine interacts with free radicals and hydroperoxides, preventing lipid perox-

Table 1. Serum biochemical parameters and serum insulin hormone levels.

Parameters	Control	Alcohol	Thiamine+Alcohol
Glucose (mg/dL)	272.55 ± 11.93 [a]	279.17 ± 19.96 [a]	281.73 ± 21.18 [a]
Triglycerides (mg/dL)	63.12 ± 2.95 [a]	58.71 ± 7.06 [a]	66.37 ± 7.02 [a]
Total Cholesterol (mg/dL)	64.86 ± 2.73 [a]	69.43 ± 1.98 [ab]	75.59 ± 2.36 [b]
HDL-C (mg/dL)	25.17 ± 0.70 [a]	21.83 ± 1.51 [a]	23.17 ± 0.98 [a]
LDL-C (mg/dL)	27.06 ± 2.29 [a]	35.85 ± 2.47 [ab]	33.80 ± 2.13 [b]
ALT (U/L)	95.60 ± 7.25 [a]	78.75 ± 5.89 [ab]	71.64 ± 5.36 [b]
AST (U/L)	127.85 ± 26.51 [a]	221.35 ± 14.43 [ab]	252.63 ± 36.58 [b]
ALP (U/L)	318.33 ± 6.28 [a]	204.83 ± 19.68 [a]	257.33 ± 60.51 [a]
Bile Acid (umol/L)	109.36 ± 6.91 [a]	131.50 ± 5.69 [b]	132.18 ± 5.12 [b]
Total Protein (g/dL)	7.23 ± 0.12 [a]	6.59 ± 0.20 [b]	6.25 ± 0.08 [b]
Total Bilirubin (mg/dL)	0.04 ± 0.01 [a]	0.09 ± 0.02 [b]	0.09 ± 0.01 [ab]
Albumin (g/dL)	3.83 ± 0.18 [a]	3.08 ± 0.05 [b]	2.93 ± 0.06 [b]
Insulin (umol/L)	2.05 ± 0.27[a]	2.54 ± 0.29[b]	2.44 ± 0.26[b]

idation and free radical oxidation in the liver (Lukienko et al 2000). Thiamine supplementation is used in thiamine deficiency, which is seen especially in alcoholics, due to poor eating habits and decreased absorption in the intestines (Lemos et al., 2005, Tallaksen et al., 1993).

In this study, we aimed to research the effects of thiamine treatment against the deterioration in some serum biochemical parameters and insulin secretion caused by acute alcohol administration in rats, an experimental model that produces effects similar to the effects caused by excessive alcohol ingestion in humans.

There are studies showing that acute alcohol consumption has no effect on blood glucose levels in humans (Schrieks et al., 2015; Steiner et al 2015). Indeed, a meta-analysis study showing alcohol consumption (10-70 kg/day) has no effect of in healthy individuals also supports this result (Schrieks et al., 2015). A cohort study shows that binge drinking causes higher fasting plasma glucose levels in men compared to women. Again, in the same study, it was reported that the highest blood glucose level associated with alcohol consumption among women aged 16-43 was in women aged 43 years (Nygren et al., 2017). On the other hand, in a study with rats, healthy and diabetic rats were given alcohol for 30 days. It has been reported that alcohol administration significantly increased blood glucose levels in diabetic rats, but there was no significant change in the healthy group (Shanmugam et al., 2011). In the current study, no significant change was observed in the blood glucose levels of healthy rats in which the binge drinking model was created (Table 1).

In one study, mice were administered with a chronic and binge drinking protocol, and compared with the control group, the alcohol group had a significant increase in serum activities of alanine aminotransferase (ALT), triglycerides, and LDL-C (Liu et al., 2014). It is well established that binge drinking is associated with an increase in LDL-C of approximately 10%, with an increased risk of atherosclerosis (Wood et al., 1998). In the current study, serum total cholesterol and LDL cholesterol increased after the binge drinking protocol. In a cohort study conducted by Al-attas et al (2014), it was reported that total cholesterol values decreased in individuals with and without diabetes who were given thiamine supplementation for 3 months. In a study on rats with diabetes, it was reported that thiamine supplementation reduced plasma cholesterol and triglyceride levels. Thiamine had no effect on HDL-C in these rats (Babaei et al, 2004). In the current study, while total cholesterol levels increased, LDL-C decreased in animals given thiamine (Table 1).

In a study, serum protein and albumin levels were significantly higher in non-drinkers compared to moderate or heavy drinkers. Heavy drinkers had significantly lower serum protein and albumin levels than moderate drinkers. In addition, heavy and moderate drinkers have significantly higher total and direct bilirubin levels than non-drinkers (Ebuehi and Asonya, 2007). Another study also reported that acute consumption of alcohol increases serum bilirubin levels in humans (O'Malley et al., 2015).

Indeed, in our study, serum protein and albumin levels were decreased and the total bilirubin levels were increased in the alcohol group according to control group. However, in the current study, thiamine administration did not significantly affect total protein, albumin, and total bilirubin values (Table 1).

In Ebuehi and Asonya (2007)'s study, serum AST, ALT, and ALP activities were significantly lower in control group than in moderate or heavy alcohol drinkers. The AST, ALT, and ALP enzyme activities of heavy drinkers were significantly higher than those of moderate drinkers. The predominance of AST

enzyme activity over ALT enzyme activity in alcoholic liver disorder was first studied in 1967 and the importance of a high AST/ALT ratio for the alcoholic liver disorder was reported. (Harinasuta, 1967). It has been reported that a decrease in hepatic ALT activity is among the causes of the high AST/ALT ratio in alcoholic liver disease (Maltoff et al., 1980). In the current study, ALT enzyme activity decreased and AST enzyme activity increased, and the AST/ALT ratio increased in the alcohol group (Table 1). In the study of Portari et al. (2013), administration of thiamine to rats administered alcohol caused a decrease in serum ALT and AST enzyme activities. In the current study, while increased AST enzyme activity was observed in the thiamine group, ALT enzyme activity decreased (Table 1).

On the other hand, it has been reported that alcohol intake increases serum bile acid levels in some studies (Donepudi et al., 2017; Manley and Ding, 2015). In the current study, it is seen that elevated bile acid levels in the alcohol and alcohol+thiamine group compared to the control group (Table 1). The role of bile acids in glucose metabolism also have been supported by a correlation between altered serum/plasma bile acid levels in metabolic changes such as obesity. Studies have shown that increased serum/plasma bile acids levels impair insulin sensitivity (Syring et al, 2019). As a matter of fact, it has been reported that serum bile acid levels are increased in insulin-resistant rats and mice (Shapiro et al, 2018).

Many studies show that acute alcohol consumption increases plasma insulin concentration, while alcohol acutely decreases insulin-stimulated glucose uptake in the whole body (Yki et al., 1988; Avogaro et al., 1996). There are studies showing that acute overdose of alcohol also causes insulin resistance in rats (Spolarics et al., 1994; Dhillon et al., 1996). Indeed, in our study, it was observed that an overdose of alcohol had higher insulin levels than healthy animals, but it did not affect the serum glucose level, and there was insufficient insulin activity (Table 1). On the other hand, in order to investigate the protective effect of thiamine, insulin levels decreased in the alcohol group to which thiamine was administered, although it was not statistically significant compared to the alcohol group.

CONCLUSION

In rodents with a binge drinking pattern, serum biochemical values are adversely affected, as in humans who drink heavily. In addition, it has been observed that the administration of thiamine vitamin, which is deficient in alcohol use and metabolic disorders occur due to this deficiency, during and before the acute use of alcohol has no effect on the changing biochemical parameters. Therefore, longer-term studies may be needed to determine the effects of thiamine on alcohol use.

DECLARATIONS

Ethics Approval

This study was approved by Burdur Mehmet Akif Ersoy University Rectorate, Animal Experiments Local Ethics Committee (Decision No: 688/2020).

Conflict of Interest

There is no conflict of interest.

Consent for Publication

Not applicable

Author contribution

Idea, concept, and design: HEE

Data collection and analysis: HEE

Drafting of the manuscript: HEE

Critical review: HEE

Data Availability

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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Research Article

Three pioneer academicians from the field of veterinary surgery in Türkiye

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ABSTRACT

The first examples of biography studies carried out within the scope of research on the history of veterinary medicine in Türkiye were given by Muzaffer Bekman, and these examples were continued with the efforts of Prof. Nihal Erk and Prof. Ferruh Dinçer. These studies, which are also given importance by later academicians in the field of veterinary medicine, are carried out both in the style of necrology and in the style of scientific biography, after the retirement or death of the professionals serving their profession. In this study, the scientific life stories of Ord. Prof. Salih Zeki Berker, Prof. M. Tevfik Başer and Prof. Burhanettin Öktem are discussed. The main material of the study consisted of the personnel files in the Archives of the Dean's Office of the Ankara University Faculty of Veterinary Medicine, and the first-hand sources obtained from the Biography Archive of the History of Veterinary Medicine and Deontology Department. In addition, various books and journals in the Library of the Department were also used. The data obtained were evaluated within the methodology of history and transferred to the text in chronological order. Ord. Prof. Salih Zeki Berker, Prof. M. Tevfik Başer and Prof. Burhanettin Öktem's academic stages, contributions to education, professional services, publishing activities and the traces they left on their colleagues are revealed. Thus, it is aimed both to contribute to the veterinary medicine biography archive and to introduce these valuable professionals to new generations.

Türkiye'de veteriner cerrahi alanından öncü üç akademisyen

ÖZ

Türkiye'de veteriner hekimliği tarihi araştırmaları kapsamında gerçekleştirilen biyografi çalışmalarının ilk örnekleri Muzaffer Bekman tarafından verilmiş, bu çalışmalar Prof. Dr. Nihal Erk ve Prof. Dr. Ferruh Dinçer'in çabalarıyla devam ettirilmiştir. Veteriner hekimliği tarihi alanının sonraki akademisyenlerince de önemsenen bu çalışmalar, mesleğine hizmet veren meslek büyüklerinin emekliliklerinin ya da vefatlarının ardından hem nekroloji tarzında hem de bilimsel biyografi yazımı tarzında sürdürülmektedir. Bu çalışmada Türkiye'de veteriner hekimliğinde cerrahi alanının önde gelen isimlerinden olan Ord. Prof. Salih Zeki Berker, Prof. Dr. M. Tevfik Başer ve Prof. Dr. Burhanettin Öktem'in bilimsel yaşam öyküleri ele alınmıştır. Çalışmanın ana materyalini, Ankara Üniversitesi Veteriner Fakültesi Dekanlık Arşivinde yer alan Personel Özlük Dosyaları ve Veteriner Hekimliği Tarihi ve Deontoloji Anabilim Dalı Biyografi Arşivinden sağlanan ilk elden kaynaklar oluşturmuştur. Bunun yanı sıra Anabilim Dalı Kütüphanesinde yer alan çeşitli kitap ve dergilerden de yararlanılmıştır. Elde edilen veriler tarih metodolojisi içerisinde değerlendirilerek kronolojik sıra ile yazıya aktarılmıştır. Ord. Prof. Salih Zeki Berker, Prof. Dr. M. Tevfik Başer ve Prof. Dr. Burhanettin Öktem'in akademik aşamaları, eğitim-öğretime yaptıkları katkılar, mesleki hizmetleri, yayın faaliyetleri ve meslektaşlarında bıraktıkları izler ortaya koyulmuştur. Böylece hem veteriner hekimliği biyografi arşivine katkı sağlamak hem de bu değerli meslek büyüklerimizin yeni nesillere tanıtılması amaçlanmıştır.

INTRODUCTION

In the Ottoman period, treatments and practices related to veterinary medicine were applied with folkloric methods within the framework of master-apprentice relationship before 1842, and after the establishment of the Military Veterinary School, the field of surgery, which is one of the most important elements of veterinary medicine, began to develop in parallel with this process (Godlewsky et al., 1846). Godlewsky, Dubroca, Lewis, Hüseyin Hüsnü, Dezutter, Yusuf Ziya, Necip Rıza Zobo and Hayreddin at the Military Veterinary School; Mehmed Ali, Yusuf Ziya, Martel, Abdullah, Hayreddin and Salih Zeki at the Civil Veterinary School (1889); Yusuf Ziya, Mustafa Santur, Salih Zeki, M. Tevfik and Burhanettin at the Higher Veterinary School (1921) which was established with the merge of Military and Civil Veterinary Schools, made ef-

[1]Additional Law to the Universities Law No. 5234, Official Gazette dated 07.07. 1948 and numbered 6951.
[2]Universities Law No. 4936, Official Gazette No. 6336, dated 18.06.1946.

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forts to develop this field both with the works they prepared and the courses they taught (Bekman, 1940, Tüzdil, 1955, Erk & Dinçer, 1970).

After the establishment of the Republic of Türkiye in 1923, the education, which was interrupted during the war years, was among the most prominent development plans of the new Türkiye, and the Higher Agricultural Institute (HAI) was established with modern facilities in Ankara in 1933 upon the principle that veterinary medicine and agriculture are the cornerstones of the country's economy (Tüzdil 1955). With this breakthrough, the Higher Veterinary School in İstanbul, with all its staff and equipment, was transferred to Ankara as a "faculty" (Bekman 1940, Tuzdil 1955). "Institute of External Diseases Clinic" within the institute system in HAI continued its activities with Max Gebhardt, Ludwig Lutz, Salih Zeki (Berker), M. Tevfik (Başer), Burhanettin (Öktem), Bekir Sıtkı (Artun) (Erk, 1961).

In 1948[1], after HAI Veterinary Faculty was engaged to Ankara University (AU), which was established in 1946[2], the unit continued its activities with the academic staff including Ord. Prof. Salih Zeki Berker, Prof. Tevfik Başer, Prof. Burhanettin Öktem, Prof. Bekir Sıtkı Artun, Asst. Hüseyin Erk and Asst. Selim Tolkun. From 1933 to May 2022, a total of 30 faculty members took part in the Ankara University Faculty of Veterinary Medicine (AUFVM) Department of Surgery (Anonymous, 2022).

Biography studies have a special place and importance in the field of veterinary medicine history (Küçükaslan &Yerlikaya, 2016). The first examples of biographical studies carried out within the scope of research on the history of veterinary medicine in Türkiye were given by Muzaffer Bekman, one of the leading names in the field (Başağaç&Öztürk, 2003). These studies were continued with the efforts of Prof. Nihal Erk and Prof. Ferruh Dinçer and gained momentum with the works of other academicians who joined the field in the following years. Various articles are published both in the style of necrology and in the style of scientific biography, after the retirement or death of the professionals serving their profession. The tags of these articles are reported in the bibliography source prepared by Küçükaslan and Yerlikaya (Küçükaslan &Yerlikaya, 2016). This study was carried out to contribute to the veterinary medicine biography archive by examining the scientific lives of Ord. Prof. Salih Zeki Berker, Prof. M. Tevfik Başer and Prof. Burhanettin Öktem.

MATERIAL and METHODS

The main material of the study was the Personnel Files of the AUFVM Dean's Archive. In addition to the first-hand sources from the Biography Archive of the AUFVM Veterinary History and Deontology Department, various books and journals in the Department's Library were also used. The data obtained were evaluated within the methodology of history and transferred to the text in chronological order.

RESULTS

Ord. Prof. Salih Zeki Berker

Salih Zeki Berker (Figure 1) was born in Trabzon in 1886. His father is Ahmet Şaban, who is engaged in trade, and his mother is Zehra. He completed his primary, secondary and high school education there. He graduated from İstanbul Higher Civil Veterinary School (İstanbul Mülkiye Baytar Mekteb-i Âlisi) in 1908, which he entered with an exam in 1904 (Nebioğlu, 1962). After working as a veterinarian for a while in the districts of the Rumeli-Kosovo province, he passed the exam set by the Ministry of Agriculture in 1909 and was among the 10 veterinarians sent to Europe for surgery specialization. After two years of specialization training in the field of surgery at the Alfort Veterinary School in France, he returned to his homeland in 1911 and joined the school staff. The "*fenn-i vilade*" (reproduction science) courses, which were taught within surgery courses, were assigned to Salih Zeki in 1911 (Tüzdil, 1955, Öktem, 1963). Salih Zeki , who joined the Balkan War in 1912, was discharged at the end of the war and was appointed as a surgical assistant at the Civil Veterinary School in 1913, he also participated in World War I in 1914. Salih Zeki was awarded the War Medal and Order of the Medjidie for his military services (Anonymous, 1969).

After the Military and Civil Veterinary Schools were merged into the Higher Veterinary School in 1921, Salih Zeki became the school principal by election for the first time in 1923 and

Figure 1. Ord. Prof. Salih Zeki Berker

continued this duty until 1928 (Anonymous, 1969). In this school, Salih Zeki was appointed as a lecturer of *fenn-i vilade-i baytari* (veterinary obstetrics), *emrazı hariciye-i umumiye ve hususiye* (general and special surgery), *ameliyat-ı cerrahiye* (operational knowledge), *fennî nalbandi* (orthopedics), *emrazı mevaşi* (ruminant diseases) courses (Öktem, 1963).

With the establishment of the Higher Agricultural Institute in Ankara in 1933[1], the Higher Veterinary School in İstanbul was transferred to Ankara as a "faculty". Salih Zeki was transferred to the HAI Veterinary Faculty "*Serîriyyât-ı Cerrahiye Enstitüsü*" (Institute of External Diseases Clinic) as an associate professor (29.10.1933) (Anonymous, 1934). Salih Zeki Berker

Table 1. Articles of Salih Zeki Berker

İneklerde Romatizmanın Seri Tedavisi. Baytarî Mecmua, 1(1): 17-19, 1339 (1923).
Osteomalasi ve Şeraiti Husulü. Baytarî Mecmua, 1(2): 59-63, 1339 (1923).
Dâ-i Distom-u Kebedî'nin (Kelebek Hastalığı) Tedavi-i Şafi-si. Baytarî Mecmua, 1(7): 219-224, 1339 (1923).
Gebelik Esnasında ve Doğurma Zamanında Dişi Hayvanlara Yapılması Lâzım Gelen Muamele ve İhtimam. Baytarî Mecmua, 2(3): 77-83, 1340 (1924).
Vaz'ı Hamil Etmiş Olan Dişi Hayvanlara ve Nevzata Yapılması Lâzım Gelen İhtimâm. Baytarî Mecmua, 2(4): 110-117, 1340 (1924).
İneklerde Akametin Esbabı ve İzalesi Çareleri. Baytarî Mecmua, 2(2): 33-42, 1340 (1924).
Yeni Doğan Hayvanat Yavrularına Karşı Yapılması İcab Eden İhtimâmat ve Emzirme Usûlleri. Baytarî Mecmua, 2(5): 144-151, 1340 (1924).
Hayvanat Yavrularına Yapılması Lâzım Gelen İhtimâmattan Sun'i Emzirme Usûlleri. Baytarî Mecmua, 2(6): 173-180, 1340 (1924).
Hayvanat Yavrularına Yapılması Lazım Gelen İhtimâmattan Sütten Kesme Yahut Fetam. Baytarî Mecmua, 2(8): 243-248, 1341 (1925).
(M. Halit ve Samuel ile) Distomatozun Şafî Devasının Memleketimizde İmâl ve İstihzarı Hakkında Mektebimizde Yapılan Tecrübeler ve Netaici. Baytarî Mecmua, 2(10): 306-317, 1341 (1925).
Kuzularda Dâ-i Felc-i Hâlfi. Baytarî Mecmua, 2(12): 359-362, 1341 (1925).
Koyunlarda Müvellid-i Kayh-ı İntânat. Baytarî Mecmua, 3(1): 8-13, 1341 (1925).
Humma-i Acul'un Esbab ve Mihanikiyet-i Husûlü. Baytarî Mecmua, 3(6):168-176, 1926.
Hayvanatta Gebeliğin Alâimi ve Tanılması. Baytarî Mecmua, 3(11): 335-342, 1926 ve 3(12): 364-372, 1926.
Adem-i Tahallüs'ün Tedavisi. Baytarî Mecmua, 4(4): 97-106, 1926.
Vaz'ı Hamilden Evvelki Felc-i Hâlfi. Baytarî Mecmua, 4(5-6): 146-149, 1926.
Burun-Meri Sondasıyla Beygirlere İlâç İçirmek. Baytarî Mecmua, 7(4): 97-101, 1929.
Hayvanlarda İhsâ. Baytarî Mecmua, 7(8): 225-232, 1930.
Kısrak ve İnekte Gebeliğin Erkenden Teşhisi (Çeviri). Baytarî Mecmua, 7(10): 297-305, 1930.
İhsâ'ya Dair Bir Müşahade. Baytarî Mecmua, 7(10): 310-311, 1930.
Kısrak ve İneklerde Akametin Esbabı ve Tedavisi. T. Vet. Hekiml. Dern. Derg., 5: 13-17, 1931 ve 6: 74-79, 1931 ve 7: 28-34, 1932.
Epidüral İptal-i His. T. Vet. Hekiml. Dern. Derg., 9: 7-17, 1932.
Harp Cerrahisi. T. Vet. Hekiml. Dern. Derg., 11(11): 9-15, 1942.
Kastrasyon Arızalarından Beygirin Kastrasyon Hemophilie'si. T. Vet. Hekiml. Dern. Derg., 16(11): 21-24, 1946.
Kısrakta Kısırlık. T. Vet. Hekiml. Dern. Derg., 18(25):14-21 1948.
Tolkun S ile), Yeni Bir Anestezik "Anovenol-K". T. Vet. Hekiml. Dern. Derg., 22, 66-67, 67-71, 1952

Apart from his duties at the school, Salih Zeki founded the *"Ettıba-yı Baytariye Muhadenet Cemiyeti"* in 1927, which forms the basis of today's Turkish Veterinary Medical Society. Berker was an active member of the Turkish Veterinary Medical Society and had many professional articles in the society's journal. In addition, Salih Zeki undertook important duties as a manager and specialist in Fatih Animal Hospital[3] until 1931, which was established in İstanbul in 1927 (Anonymous, 1933, Kural, 1938).

was promoted to professoriate in 1936 and was awarded the title of ordinary professor (distinguished professor) in 1944 (Erk & Dinçer, 1970).

The divisions that were in the form of "institutes" since the establishment of HAI were converted into "departments" (*kürsü*) at the beginning of 1952. In this context, Ord. Prof. Salih Zeki Berker was brought to the management of the First Surgery Department and Clinic[4,5]. Berker, who speaks French and Ottoman languages, conducted many studies in his field in Italy, France, Switzerland, Netherlands, Germany, England,

Table 2. Books of Salih Zeki Berker

Emraz-ı Mevaşi, I.cilt, Hilâl Matbaası, İstanbul, 304 s, 1342 (1926)
Emraz-ı Mevaşi, II. cilt, Hilâl Matbaası, İstanbul, 304 s, 1342 (1926).
Fenn-i Vilâde-i Baytarî, Hilâl Matbaası, İstanbul, 396 s, 1928.
Gebe Hayvanlara Henüz Doğurmuş Olanlara ve Doğan Yavrulara Yapılması Lazım Gelen Tedbirler, Sanayii Nefise Matbaası, İstanbul, 32 s, 1928.
Ehlî Hayvanların Cerrahî Hastalıkları, I. cilt, Sanayii Nefise Matbaası, İstanbul, 712 s, 1931
Ehlî Hayvanların Cerrahî Hastalıkları, II. cilt, Necmi İstiklal Matbaası, İstanbul, 877 s, 1932.
Nalbantlık, Maarif Basımevi İstanbul, 209 s, 1939.
Ehlî Hayvanların Cerrahî Hastalıkları, I. cilt, YZE, Ankara, 248 s, 1939.
Ehlî Hayvanların Cerrahî Hastalıkları, II. cilt, YZE, Ankara, 211 s, 1940.
Ehlî Hayvanların Doğum Bilgisi, YZE, Ankara, 265 s, 1943.
(Artun BS ile) Ehlî Hayvanların Dış Hastalıkları Klinik Teşhisi, YZE, Ankara, 91 s, 1945.
(Öktem B ile) Evcil Hayvanların Göz Hastalıkları, Ders Kitabı No: 36, YZE, Ankara, 111 s, 1946.
(Öktem B ile) Genel Şirurji, I. baskı, AÜ Basımevi, Ankara, 396 s, 1956.
(Öktem B ile) Genel Şirurji, II. baskı, AÜ Basımevi, Ankara, 429 s, 1956.

Hungary and Bulgaria (Nebioğlu, 1962). The list of Salih Zeki's works is presented in Table 1 and Table 2.

Ord. Prof. Salih Zeki Berker retired from the age limit in 1956, as he turned 70 years old. Berker, who served the profession of veterinary medicine for 53 years and nine months, died on March 30, 1970 in İstanbul due to a set of ailments.

Prof. M. Tevfik Başer

Mehmet Tevfik Başer (Figure 2) was born in İzmir in 1893[6]. He completed his primary and secondary education in İzmir in 1911 and enrolled in the *Mülkiye Baytar Mektebi Alisi* (Higher Civil Veterinary School) in the same year. While he was still a student, he was drafted into the military as a veterinary reserve officer candidate in 1914 due to the outbreak of World War I[7]. Having served in the Thrace, Syria, Anatolia, Iran and Iraq fronts, M. Tevfik was discharged from the military in 1918 with the rank of lieutenant. M.Tevfik returned to veterinary school after completing his military service and graduated in 1920. Afterward, he worked as a Municipal Veterinarian in İstanbul for two years (17.08.1923-14.03.1925). M.Tevfik went to France Alfort Veterinary School in 1925 under the Ministry of Agriculture initiative and completed his Ph.D. in surgery in 1928[6]. After returning home, he was appointed as the Chief of Surgical Diseases at İstanbul Higher Veterinary School (07.11.1928-25.06.1937)[8]. Başer completed his habilitation work on surgical hernias[9] in 1937 and was awarded the title of second-class associate professor in 1937 and first-class associate professor in 1939[10,11,12]. Continuing his academic studies, Başer was promoted to second-class professoriate in 1941 and first-class professoriate in 1945[13,14].

Prof. M. Tevfik Başer provided many services related to his profession both inside and outside the faculty throughout his academic life. He worked in Karacabey Stud Farm about surgical diseases seen in mares[15] and was among the members of the committee formed for the race determination of Thoroughbred Arabian horses[16]. Prof. M. Tevfik Başer was in the

Figure 2. Prof. M. Tevfik Başer

[3]Fatih Animal Hospital, which was established on 22.04.1927 as a subordinate of İstanbul Fatih Municipality, was established to treat the animals of the municipality and the sick animals of the people free of charge.
[4]Decision of the AU Senate dated 15.01.1952 and numbered 661.
[5]Decision of the AU Senate dated 15.01.1952 and numbered 661.
[6]Personnel Files of Prof. M. Tevfik Başer, AUFVM Dean's Archive.
[7]Letter dated 19.04.1945 and numbered 193 sent to HAI Rectorate by HAI Veterinary Faculty Surgery Institute.
[8]The letter dated 18. 11.1948 and numbered 1231 sent by the AU Rectorate Personal Affairs Directorate to the Deanery of AUFVM.

Table 3. Articles of M.Tevfik Başer

Trakya'da İhsâ. Baytarî Mecmua, 7(1): 2-14, 1929 ve 7(2): 47-54, 1929 ve 7(3): 77-78, 1929 ve 7(4): 118-128, 1929.
Yaraların "Lactoserum" la Tedavisi. Baytarî Mecmua, 7(3): 72-77, 1929.
Laypzik Cerrahi Enstitüsünde Meslektaşlarımızdan Saip Ali Beye. Baytarî Mecmua, 7(6): 171-178, 1930.
(Aysoy S ile), Autohaemotherapie'nin Göz Hastalıklarına Tatbiki. Baytarî Mecmua, 7(9): 262-266, 1930 ve 7(10): 311-313, 1930 ve 7(11): 346-351, 1930.
Tektırnaklılarda Üçüncü Falanksın İstihsali. YZE Dergisi, 1(1): 239-246, 1943.

Table 4. Books of M. Tevfik Başer

Ehlî Hayvanlarda Karın Fıtkı. YZE, Ankara, 43 s, 1939.
Fennî Nalbantlık kısım 1, Talebe Ders Kılavuzu (Roto), Sayı 89, 1940.
Fennî Nalbantlık kısım 2, Talebe Ders Kılavuzu (Roto), Sayı 61, 1940.
Ayak Ameliyeleri, Talebe Ders Kılavuzu (Roto), Sayı 104, 1941.
Hayvanlarda Koterizasyon, YZE, Ankara, 85s, 1943

management of the Second Surgery Department[6] (1952) and Obstetrics and Gynecology Department[17] (1957). He also managed various administrative duties such as senate, commission and jury memberships[18,19,20]. Başer, in addition to his assignments in homeland, went to Italy, Switzerland, France, England and Germany and conducted numerous studies in his field[21,22,23]. The list of works by M. Tevfik Başer is presented in Table 3 and Table 4. Başer, who has a special interest in equestrian sports, especially horse racing, and is knowledgeable about the special treatment methods of racehorses as well as the administration of horse races, served as a Principal Member of the Jockey Club of Türkiye in 1959. He served in this field for more than 30 years and became a member of the Supreme Council of Commissars (Anonymous, 2022, Tolkun, 1960, Tolkun, 1969). Prof. M. Tevfik Başer passed away on 26.09.1960 in the hospital where he was admitted with the diagnosis of myocardial infarction and heart failure in İstanbul after 42 years and 6 months of professional life[24,25].

Prof. Burhanettin Öktem

Burhanettin Öktem (Figure 3) was born in İstanbul in 1904. His father is Police Captain İbrahim. He completed his primary education in İstanbul and İzmit, his secondary and high school education in Harbiye Military High School, and graduated from the Higher Veterinary School with the first degree in 1927. After graduation, he was appointed as a veterinarian for the fight against rinderpest in Samsun (28.08.1927-05.03.1928). He did his internship for one year with the rank of lieutenant at the İstanbul Military Veterinarian Practice School *(İstanbul Askeri Baytar Tatbikat Mektebi)* (26.05.1928-26.05.1929). Later, he worked as a government veterinarian for four years in Erzurum's Hınıs and Pasinler and Gümüşhane's Torul and Kelkit counties (06.09.1929-14.01.1933). Öktem, who started as an assistant in general medicine at İstanbul Higher Veterinary School in 1933, was appointed as an assistant for external diseases on 28.10.1933 after this school was abolished and trans-

Figure 3. Prof. Burhanettin Öktem

[9]Baser M.T. Ehli Hayvanlarda Karın Fıtkı (Habilitation Thesis), HAI, Ankara, 1939.
[10]The letter dated 19.04.1945 and numbered 193 sent by HAI Veterinary Faculty Surgery Institute to the HAI Rectorate.
[11]The letter dated 30.12.1939 and numbered 2356 sent by the T.C. Ministry of Agriculture, Department of Agriculture, HAI Administrative Consultancy to the HAI Specialist Consultancy.
[12]The letter dated 23.10.1933 and numbered 1848 sent by the T.C. Ministry of Agriculture HAI Rectorate to M. Tevfik Başer.
[13]The letter dated 14.04.1941 and numbered 2470 sent by the T.C. Ministry of Agriculture HAI Rectorate to the Higher Deputation Authority.
[14]The letter dated 22.12.1945 and numbered 21262 sent by the T.C. Ministry of Agriculture Personal Affairs Directorate to the HAI Rectorate.

Table 5. Articles of Burhanettin Öktem

Baytar Fakültesinde Poliklinik Çalışmaları. T. Vet. Hekiml. Dern. Derg., 5(4): 527-532, 1935.
Evcil hayvanlarda ve bilhassa beygirlerde alt çene kemiği (mandibula) kırıkları. T. Vet. Hekiml. Dern. Derg., 12(3-4): 3-31, 1942.
Pisipisi otunun husule getirdiği travmasal bozukluklar ve hastalıklar. T. Vet. Hekiml. Dern. Derg., 13(3): 44-58, 1945.
Dişi bir köpekte bütün bir pisipisi otu (Hordeum bulbosum) başağından ileri gelme gayri tam göğüs fistülü ve myositis olayı. YZE Dergisi, 3(2): 500-504, 1945.
Veteriner şirurjide sulfonamid preparatlarının lokal kullanılışı ve bilhassa ultraseptylurée ile marfanil-prontalbin yara tozlarından elde olunan faydalı sonuçlar. YZE Dergisi, 5(2): 514-536, 1946.
Klinik muayene usullerile kesin olarak teşhis edilemeyen iki karın fıtkı (hernia abdominalis) olayı. YZE Dergisi, 7(13): 231-239, 1946.
Türkiye'de zehirli yılanlar, zararları ve bunların sokmalarından ileri gelen arızaların tedavisi. T. Vet. Hekiml. Dern. Derg., 16(7): 10-19, 1946.
Karşılaştığımız enteresan bir olay dolayısiyle coxae (kalça) kırıkları üzerinde incelemeler. YZE Derg., 9(1): 96-111, 1947.
(Erk H ile), Damar içine trypaflavin eriyikleri şırıngasından sonra şekillenen phlebitis ve periphlebitis gangraenosa olayları. T. Vet. Hekiml. Dern. Derg., 19(28-29): 23-39, 1949.
Gebelik nasıl anlaşılır? Gebelere nasıl bakılır? Üretim, 1(10): 11, 1950.
Doğuran hayvanlar ve yavrularına yapılacak yardımlar. Üretim, 1(12): 16, 1950 ve 2(13-14): 10-11, 1950.
Hayvanlarımızın değeri ve bunların sağlığını korumadaki ödevlerimiz. Üretim, 2(20): 4-5, 1950.
Lokal olarak kullanılan sulfonamidlerin veteriner şirurjide ve tercihan karın operasyonlarındaki proflaktik değerleri. T. Vet. Hekiml. Dern. Derg., 20(43-44): 138-166, 1950.
Rahmetli Prof. Dr. Tevfik Başer'e son vazifemizi yaparken. T. Vet. Hekiml. Dern. Derg., 30(168-169): 933-938, 1960.
Fakültemiz otuz yaşında. T. Vet. Hekiml. Dern. Dern., 33(11-12): 562-572, 1963.
(Anteplioğlu H ile), Tendogen ve arthrogen hydrops'ların çeşitli cortison preparatları ile tedavisi üzerinde araştırmalar. A.Ü. Vet. Fak. Derg., 14(1): 25-54, 1967.
(Anteplioğlu H ile), Evcil hayvanların arthritis prulenta'larının lokal olarak penicillin enjeksiyonlarıyla tedavisi üzerinde araştırmalar. A.Ü. Vet. Fak. Derg., 14(3): 321-341, 1967.
(Finci A ile), Evcil hayvanlarda çok seyrek raslanan çene kilitlenmesi olayları. A.Ü. Vet. Fak. Derg., 16(1): 17-26, 1969.
Avrupa Veteriner Şirurjiyenleri Birliğinin 8. Kongresinden izlenimler. T. Vet. Hekiml. Dern. Derg., 39(8): 14-16, 1969.

ferred to Ankara HAI. He was first promoted to second-degree assistantship, and in 1935 to first-degree assistantship[26,27]. Öktem went to Germany in 1935 to conduct research in his field and stayed at the Hannover and Berlin Veterinary Schools for three months and participated in the works of surgery and small animal clinics. Returning to Türkiye after his education abroad and continuing his doctoral thesis, Öktem was promoted first as the chief assistant and two years later as the branch

[15]Decision of the AUFVM Professors Board dated 07.05.1958.
[16]The letter dated 28.05.1948 and numbered 126 sent by the T.C. Ministry of Agriculture, Department of Veterinary Affairs, General Directorate of Veterinary Affairs to the HAI Rectorate.
[17]Decision of the AUFVM Professors Board dated 29.06.1957 and numbered 84.
[18]Letter from the AUFVM Deanery to Prof. M. Tevfik Başer, dated 29.03.1956 and numbered 826.
[19]The letter dated 21.12.1950 and numbered 5680 sent by the HAI Veterinary Faculty Deanery to HAI Rectorate.
[20]Decision of the Professors Board of AUFVM dated 27.06.1955.
[21]The letter dated 27.08.1954 and numbered 2008 sent by the T.C. AU Rectorate Personal Affairs Directorate to the Veterinary Faculty Deanery.
[22]The letter dated 25.08.1954 and numbered 1983 sent by the T.C. AU Rectorate Personal Affairs Directorate to the Veterinary Faculty Deanery.
[23]Letter 107 dated 14.07.1955 sent by the Department of Surgery of the Faculty of Veterinary Medicine to the Veterinary Faculty Deanery
[24]UFVM Deanery's letter dated 29.09.1960 and numbered 4827.
[25]The letter dated 06.10.1960 and numbered 1198 sent by Ord. Prof. Ekrem Şerif Egeli from the Internal Medicine Clinic of İstanbul University Faculty of Medicine to the Veterinary Faculty Deanery.
[26]Personnel files of Prof. Burhanettin Öktem, AUFVM Deanery Archive.
[27]AUFVM Veterinary History and Deontology Department Biography Archive. Burhanettin Öktem File No: 563.
[28]The letter dated 09.07.1943 and numbered 594 sent by the Directorate of the Institute of Surgery to the Deanery of the Faculty of Veterinary Medicine at HAI.
[29]Öktem B. Türkiye'de Nalbantlık (Ph.D. Thesis), HAI, Ankara, 1939.
[30]The letter dated 17.08.1937 and numbered 4114 sent by the T.C. Ministry of Agriculture Personal Affairs Directorate to the HAI Rectorate.
[31]The letter dated 16.07.1937 and numbered 2196 sent by the T.C. Ministry of Agriculture Personal Affairs Directorate to the HAI Rectorate.
[32]Decision of Officers Selection Council, letter dated 16.10.1939 and numbered 41.
[33]Öktem B, Ehli Erkek Hayvanlarda Kastrasyon ve Memleketimiz İçin Müntehap Usuller (Habilitation Thesis), HAI, Ankara, 1943.
[34]Letter 74 dated 12.02.1941 sent by the Deanery of the HAI Faculty of Veterinary Medicine to the HAI Rectorate.
[35]The letter of the Higher Deputation Authority dated 16.07.1943 and numbered 44 and the Council Decision numbered 46.
[36]The letter dated 28.09.1943 and numbered 1772 sent by the T.C. Ministry of Agriculture HAI Administrative Consultancy to the HAI Rectorate.
[37]The letter dated 19.03.1947 and numbered 450 sent by the Deanery of the HAI Faculty of Veterinary Medicine to the HAI Rectorate.
[38]The letter dated 22.02.1946 and numbered 229 sent by the Deanery of the HAI Faculty of Veterinary Medicine to the HAI Rectorate.

Table 6. Books of Burhanettin Öktem

İş Hayvanlarında Tırnak Bakımı. Köylüye Faydalı Bilgiler, YZE Yayınları, No:16, Ankara, 1938.
Yeni Doğan Yavrulara Nasıl Yardım Etmeli ve Nelerine Dikkat Etmeli?. Köylüye Faydalı Bilgiler, YZE Yayınları, No:17, 1938.
Türkiye'de Nalbantlık. YZE, Ankara, 145 s, 1939.
(Berker SZ ile) Evcil Hayvanların Göz Hastalıkları, Ders Kitabı No: 36, YZE, Ankara, 111 s, 1946.
Evcil Erkek Hayvanlarda Kastrasyon ve Memleketimiz İçin En Elverişli Usuller. YZE, Ankara, 151 s, 1948.
(Berker SZ ile) Genel Şirurji, I. baskı, AÜ Basımevi, Ankara, 396 s, 1956.
(Berker SZ ile) Genel Şirurji, II. baskı, AÜ Basımevi, Ankara, 429 s, 1956.
Evcil Hayvanların Göz Hastalıkları. Ders Kitabı No: 50, II. Baskı, Yeni Desen Matbaası, Ankara, 132s, 1959.
Özel Şirurji-Baş ve Boyun Hastalıkları. Ders Kitabı No: 59, I. Fasikül, İstiklâl Matbaası, Ankara, 177 s, 1962.
(Anteplioğlu H ile) Ankara Bölgesi İneklerinde Görülen Mastitis'in Tedavisi Üzerinde Mukayeseli İncelemeler. İstiklâl Matbaası, Ankara, 96 s, 1962.
Özel Şirurji-Göğüs ve Karın Hastalıkları. I. baskı, İstiklâl Matbaası, Ankara, 221s, 1964.
Özel Şirurji- Ön ve Arka Bacak Hastalıkları. AÜ Basımevi, Ankara, 294 s, 1966.
(Samsar E ile) Enteresan Bir Polyabcedation Olayı Dolayısiyle Çeşitli Evcil Hayvanlarda Rasladığımız Apselerin Sebep ve Tedavileri Üzerinde İncelemeler. AÜ Basımevi, Ankara, 58s, 1967.
Özel Şirurji-Göğüs ve Karın Hastalıkları. II. baskı, İstiklâl Matbaası, Ankara, 193s, 1969.

chief after completing his doctoral thesis titled "Türkiye'de Nalbantlık (Horseshoeing in Türkiye)"[29] in 1937[30,31,32].

Öktem prepared his habilitation thesis[33] titled "Ehli erkek hayvanlarda kastrasyon ve memleketimiz için müntehap usuller (Castration in domestic male animals and the appropriate methods for our country)" in 1943, and was awarded the HAI second-degree associate professorship after a German language exam and trial course[34,35,36]. Continuing his academic studies after being an associate professor, he was promoted to professorship in 1947[37]. Throughout his academic life, he conducted research about his profession both inside and outside the faculty. For instance, he worked in Sultansuyu Stud Farm[38] in 1946 and in Karacabey Stud Farm in 1947 and 1954[39,40,41],

[39]The letter dated 31.01.1947 and numbered 6499 sent by the General Directorate of Veterinary Affairs of the Ministry of Agriculture to the HAI Rectorate.
[40]The letter dated 29.01.1954 and numbered 5595 sent by the General Directorate of Veterinary Affairs of the Ministry of Agriculture to the Deanery of AUFVM.
[41]AUFVM Professors Board Decision dated 6.02.1954 and numbered 76.
[42]The letter dated 31.12.1949 and numbered 5909 sent by the Deanery of the HAI Veterinary Faculty to the Presidency of the General Staff Veterinary Department.
[43]The letter dated 08.05.1958 and numbered 1236 sent by the T.C. Ministry of Agriculture Personal Affairs Directorate to the Deanery of the Faculty of Veterinary Medicine.
[44]AUFVM Professors Board Decision No. 42 dated 25.04.1958.
[45]The letter dated 17.07.1973 and numbered 4417 sent by the AUFVM Deanery to the Professoriate of the First Surgery Department.
[46]The letter dated 22.07.1968 and numbered 2701 sent by the AUFVM Deanery to the Professoriate of the First Surgery Department.
[47]AUFVM Professors Board Decision dated 11.07.1968 and numbered 148.
[48]AUFVM Professors Board Decision dated 11.07.1968 and numbered 148.
[49]The letter dated 29.08.1969 and numbered 31-507-820 sent by the T.C. Ministry of Agriculture Personal Affairs Directorate to the Deanery of the Faculty of Veterinary Medicine.
[50]AUFVM Professors Board Decision dated 26.8.1969 and numbered 193.
[51]AUFVM Professors Board Decision dated 14.11.1968 and numbered 180.
[52]The letter dated 29.11.1968 and numbered 4386 sent by the AUFVM Deanery to the Professoriate of the First Surgery Department.
[53]AUFVM Professors Board Decision dated 1.06.1955 and numbered 79.
[54]The letter dated 12.07.1955 and numbered 31-500 sent by the T.C. Ministry of Agriculture Personal Affairs Directorate to the Deanery of the Faculty of Veterinary Medicine.
[55]The letter dated 2.09.1955 and numbered 126 sent by the AUFVM First Surgery Clinic to the Deanery of the Faculty of Veterinary Medicine.
[56]The letter dated 10.10.1961 and numbered 3083 sent by the AUFVM Deanery to the Rectorate.
[57]AUFVM Professors Board Decision dated 28.06.1961 and numbered 106.
[58]The letter dated 16.09.1968 and numbered 169 sent by the Professoriate of the First Surgery Department and Clinic to the Deanery of the Faculty of Veterinary Medicine.
[59]AUFVM Faculty Board Decision dated 9.05.1974 and numbered 111.
[60]AUFVM Professors Board Decision dated 27.11.1956 and numbered 3646.
[61]AUFVM Professors Board Decision dated 25.06.1957 and numbered 81.
[62]AUFVM Professors Board Decision dated 4.11.1968 and numbered 168.
[63]The letter dated 12.11.1968 and numbered 4293 written by the Deanery of AUFVM to Prof. Burhanettin Öktem.
[64]AUFVM Professors Board Decision dated 2.11.1966 and numbered 147.
[65]AUFVM Professors Board Decision dated 24.06.1959 and numbered 83.
[66]AUFVM Professors Board Decision dated 3.11.1960 and numbered 113.
[67]The letter dated 17.09.1964 and numbered 282 sent by the AUFVM Chair of the First Surgery Department and Clinic Professioriate to the Deanery of the Faculty of Veterinary Medicine.
[68]The letter dated 11.04.1967 and numbered 1392 sent by the AUFVM Deanery to the First Surgery Department Professoriate.
[69]The letter dated 17.06.1974 and numbered 4013 sent by the Deanery of the AUFVM to the AU Rectorate.
[70]The letter dated 03.07.1974 and numbered 4604 sent by the Deanery of AUFVM to Prof. Burhanettin Öktem.

gave surgical courses in the Military Veterinary Academy in the 1949-1950 academic year[42], and worked for 20 days at the İstanbul Animal Health Officers School Clinic and Municipality Animal Hospital in 1958[43,44]. Between 1968 and 1973, he worked in the provinces of Balıkesir, Manisa, İstanbul, Bursa, Denizli, Antalya and Mersin[45,46,47,48,49,50,51,52]. Öktem conducted scientific studies in his field in England, Germany, the Netherlands, Sweden, Denmark, Austria and France for 18 months[53,54,55,56,57]. He represented our country at the European Veterinary Surgery Congress held in Italy[58].

Öktem, in addition to his many administrative duties, such as serving as the head of the Surgery and Reproduction Departments, was a member of the Senate, Faculty Board and commissions in various years[59,60,61,62,63,64,65,66]. Öktem, who was actively working in the process of spreading veterinary medicine education and the establishment of new veterinary faculties, contributed to the establishment of Elazığ and İstanbul veterinary faculties by taking part in the related commissions[67,68].

After 47 years of professional life, Öktem retired from the age limit on 14 July 1974[69]. Training hundreds of students throughout his career and making significant contributions to his field with his scientific research (Table 5 and Table 6), Prof. Burhanettin Öktem was not forgotten by his colleagues and he was given a service plaque by AUFVM in 1974[70]. Öktem died on September 20, 1993 (Anonymous, 1993).

DISCUSSION

In the study, Ord. Prof. Salih Zeki Berker, Prof. M. Tevfik Başer and Prof. Burhanettin Öktem's scientific life stories are discussed (Figure 4, Figure 5, Figure 6). Within the scope of this study, it is thought that introducing eminent professionals to academicians and veterinary medicine students is important in terms of historical awareness. Küçükaslan & Yerlikaya (2016) reported that biographies in the style of necrology were written after the death of valuable scientists in the history of veterinary medicine. When these texts are examined, it is seen that there are various texts in which the contributions of the members of the profession to science and the profession are explained, among which there are writings by Berker, Başer and Öktem. For example, while describing Salih Zeki Berker after his death, Kolaylı (1970) explained that he was a teacher who *"loves his profession very much, is jealous and tries to prevent it from falling into the hands of others"*. Tolkun (1970) described his teacher Berker as *"a valuable teacher who corrects a problem every time he comes to the clinic, explains the importance of the white coat every time, is fussy, impatient, has a tough temperament, but explains the subjects to his students over and over again and does not hesitate to appreciate them"*. Hergüner[71] who worked in the printing house for many years during HAI period at AU, stated that Salih Zeki Berker was *"hardworking, steadfast, resilient, respectful, and one of the great teachers who know how to make himself respected"*. These are expressions that reveal Berker's academic personality. It has been determined that there are many works written by Berker (Table 1 and Table 2). Among these, Küçükaslan (2012) reported that the work named *"Fenn-i Vilâde-i Baytarî"* (veterinary reproduction science) written by Berker is the first copyrighted work written in the field of obstetrics and gynecology. It can be argued that with this work of Berker, he set an example of an academic path for his colleagues. Prof. Salih Zeki Berker was given the title of ordinary professor in 1944. It can be argued that giving this title to Berker, who has important services both scientifically and professionally, can be considered an important criterion of value. As a matter of fact, considering that this title is legally[1] only given to those with outstanding scientific research and scientists who made great contributions to the profession, the value of Berker in terms of the profession is better understood.

After Prof. M. Tevfik Başer, his colleagues wrote necrological texts about him. For example, Taşdelen (1960) quoted that Başer was known as an honest and honorable person with the words: *"He accepted that it is one of the qualities of humanity that people not to stay away from honesty and fear of God and who pursue honor and wealth with greed through illegitimate ways are devastated, by mentioning that it is the law of nature"*. It can be argued that, as one of the great professors in the field of surgery, the words of Prof. M. Tevfik Başer are also a piece of advice for today's colleagues. Ord. Prof. Hilmi Dilgimen, expressed his view of Prof. M. Tevfik Başer: *"He is the first founder of modern surgical methods and techniques in our country"* (Sevinç, 1960). Similarly, Öktem's (1960): *"He is the unique operator who brings, teaches and succeeds in modern surgery, the technique and meticulousness of performing operations under asepsis and antisepsis in Turkish Veterinary Medicine."* and Tolkun's: *"He is my late teacher who introduced modern aseptic veterinary surgery to our country"* phrases support the view that he undertook a leading role in this regard. Öktem (1963), regarding his polyclinic and clinical studies in 1928, said: *"Despite the fact that most of the courses on external diseases (surgery), which is my branch, were taught by our teacher Salih Zeki Berker, the Assistant Professor and the Chief of the Clinic Tevfik Başer who has returned to the country in 1928 after completing his Ph.D. in France, Alfort Veterinary School, manages the polyclinic and clinical studies with a meticulousness that we have not seen until then, and he personally performed the operative interventions with great knowledge"*. All these statements in the literature reveal that Başer contributed greatly to the field by bringing a pioneering approach to the field of veterinary surgery.

Being interested in horse races for many years and a member of the Higher Stewards Council of Commissars in Horse Racing, Prof. M. Tevfik Başer was not forgotten by the Jockey Club of Türkiye, many races were organized in his name (Anonymous, 2022). This can be considered as an indication that Başer provided important services in this particular area.

In his associate professorship report about Burhanettin Öktem, Professor of the Institute of Surgery M. Tevfik Başer described Öktem as *"serious, decent and honest, does not spare time, has mature clinical information and is able to manage the surgery clinic independently and performs surgery by taking all responsibility"*[72].

After Prof. Burhanettin Öktem's retirement, his colleague

[71]Personal interview with the printing officer Ramazan Hergüner on 02.03.2010.
[72]The letter dated 19.05.1949 and numbered 3221 sent by the Directorate of the Institute of Surgery to the Deanery of the Faculty of Veterinary Medicine at HAI.

Figure 4. The foot diseases exam that President İsmet İnönü attended during his visit to Ankara University Faculty of Veterinary Medicine (1949)

Figure 5. Surgery instructors with faculty members at Ankara University Faculty of Veterinary Medicine (First row from left to right, 1. M. Tevfik Başer, 4. Salih Zeki Berker, 10. Burhanettin Öktem, 1947)

Figure 6. M.Tevfik Başer, Salih Zeki Berker and Burhanettin Öktem during clinical courses at Ankara University Faculty of Veterinary Medicine

Prof. Hayrettin Anteplioğlu stated the followings about his teacher: *"Our teacher was very meticulous in his work. In the classroom, in practice, in the clinic, in the study of problems, in general, he sought the best and the most complete. In fact, this quality was a necessity of his creation. For a surgeon, this feature is considered a great and necessary factor. In the administrative and scientific stages he continued at the Faculty of Veterinary Medicine for many years, he remained extremely loyal to the laws. His firm belief, his cautiousness in examining events, his high-pitched voice that we hear from time to time, and then his slow return to a smiling softness, his stance against no neediness, his tendency towards doing the right in every job, his behavior that expresses his love of the profession, are good gifts for us to always consider and think about"*. He emphasized that *"despite the tiring and difficult conditions, he always carried out useful and valuable works for the public regarding the economic*

[73]The letter dated 02.08. 1974 and numbered 258 sent by the Chair of First Surgery and Clinic Department to the Deanery of AUFVM.
[74]The letter dated 05.08. 1974 and numbered 5281 sent by the Dean of AUFVM, Prof. Satı Baran, to the Head of the Publication Commission.

problems of the country, and contributed greatly to the acquisition of many facilities and apparatus for the Surgical and Orthopedic units, and the afforestation and greening of the environment" [73,74].

CONCLUSIONS

As the surgery is one of the building blocks of the education process in veterinary medicine, it can be argued that these professionals, who are one of the first lecturers in the field of veterinary surgery, undertook important tasks in education, scientific research, publication activities, training of other lecturers and added significant value to the veterinary medicine profession. It can be said that the results obtained through biography studies can set an example for young profession candidates and have the feature of creating a spiritual effect that they can be proud of.

DECLARATIONS

Ethics Approval

Not aplicable.

Conflict of Interest

Authors declare that there are no conflicts of interest for this study.

Consent for Publication

Not applicable

Author Contribution

Idea, concept, and design: NY

Data collection and analysis: NY, ÖK

Drafting of the manuscript: NY, ÖK

Critical review: NY

Data Availability

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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Research Article

Clinical and radiological evaluation of metacarpal fractures in calves: A retrospective study: 72 cases

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ABSTRACT

Metacarpal fractures are frequently encountered in calves, especially as a result of dystocia. This study, it was aimed to evaluate metacarpus fractures in calves from multiple aspects, especially the fracture's causes, location and classification, and to determine the most appropriate treatment option. Seventy two calves diagnosed with a metacarpus fracture, aged between 1 and 15 days, regardless of gender or breed, were included in the study. Calves were classified according to fracture type and treatment method. In the treatment, external fixation with Steinman pins and polyvinylchloride-fiberglass plaster supported closed or windowed bandage methods were preferred. According to the findings of the radiological examination, the fractures were determined to be distal diaphyseal in 40 (55%) cases, epiphyseal in 20 (27%) cases, and middle diaphyseal in 12 (18%) cases. It was discovered that 46 of 51 cases with closed fractures and bandages healed without complications, while 5 patients died due to comorbidities (calf diarrhea, aspiration pneumonia). Five of seven patients who had external fixation using Steinmann pins recovered without complications, meanwhile 2 patients succumbed to infection. Full recovery was observed in 12 of 14 patients who underwent window bandage, and it was discovered that 2 patients died due to secondary infection. As a result, substantial data that will contribute to clinical practice and literature knowledge regarding the causes, location, classification and treatment of metacarpal fractures, which are commonly encountered in calves and cause both economic and productivity loss, have been uncovered.

INTRODUCTION

Extremity fractures are prevalent in calves. Dystocia, crushing of calf by a cow, trauma from other animals, and faulty manipulations during childbirth are among the most common causes of limb fractures. (Aksoy et al, 2009, Nuss et al, 2011, Arıcan et al, 2014, Belge et al, 2016). Metacarpus fractures constitute approximately half (50%) of all extremity fractures in calves (Nichols et al, 2010, Rodrigues et al, 2012, Arıcan et al, 2014, Öztaş & Avki 2015). Calves exhibit behaviors such as reluctance to move, inability to stand on their feet, and inability to walk, as well as moving in a lying position. Radiograpic views taken from different angles (e.g. dorsopalmar/plantar, mediolateral and anteroposterior) in conjunction with clinical findings are helpful in the diagnosis of fracture (Ewoldt et al, 2003, Mulon, 2014).

Many factors such as the shape of the fracture, its localization, duration, and the economic and breeding value of the animal influence the effectiveness of fracture treatment (Görgül et al, 2004, Aksoy et al, 2009, Öztaş & Avki 2015, Yurtal et al, 2020). External and internal methods could be employed to reduce and stabilize the broken bone. In farm animals, the bandage technique supported by materials such as polyvinylchloride (PVC) and aluminum, alone or in combination with a bandage such as Thomas Splint, has been frequently used as a treatment option for external fixation of closed fractures (Auer et al, 1993, Nuss et al, 2011, Mulon, et al. 2014, Belge et al, 2016). Internal fixation techniques require general anesthesia, expertise, special tools, and equipment. In addition, some issues such as nonunion, angular deformity, and osteomyelitis in comminuted, open or infected fractures reduce the likelihood of success (Arıcan et al, 2014).

The current study was aimed to determine the causes of

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metacarpus fractures in calves, radiological findings, the success of the treatment options, and the most appropriate treatment option in light of this data.

MATERIAL and METHODS

Case Selection

Seventy-two calves with different breeds and genders, aged between 1 and 15 days, were brought to the Kafkas University Veterinary Medicine Faculty Animal Hospital with the complaint of lameness due to dystocia or trauma and diagnosed with metacarpus fracture were included in this study. Systematic and inspection-palpation examinations for fractures were performed on patients who presented with complaints of lameness in the front leg due to dystocia or trauma. Subsequently, mediolateral and anteroposterior radiographs of the corresponding extremity of each calf were performed for a definitive diagnosis. Following the determination of the anatomical location and shape of the fracture, the preparations were planned for the necessary interventions.

Equipments

Considering the different fracture types, the closed fixation technique was preferred for closed fractures and open fractures suited for windowed bandage application. To provide fixation, 2-4 pieces of 10 cm x 10 m roll bandage, 1 - 2 pieces of 250 g hydrophilic cotton, 1 - 2 pieces of polyvinyl chloride (PVC) prepared in appropriate sizes, 2 - 3 pieces of number 3 and/or number 4 fiberglass plasters were used. In infected fractures that are not suitable for open and windowed bandages, 4 Steinman pins (Safir®, Antalya/Turkey) with a diameter of Ø3-5 mm were preferred as external fixation material for placing transversal for 2 pieces of each fracture fragment.

Surgical Procedure

Two hours prior to the surgery, antibiotic (Cefazolin, 30 mg/kg, IM, Cezol, Deva®, Istanbul) was administered to each calf with fractures that were to be externally treated with transversal pinning. Following the shaving and cleaning of the related extremity, the area was prepared for aseptic surgery. The operation was performed with sedation with 0.05 mg/kg IM xylazine HCl (Rompun 2%, Bayer®, İstanbul) and general anesthesia with 4 mg/kg IM ketamine HCl (Ketakontrol®, Doğa İlaç, İstanbul). Following anesthesia, Steinmann pins prepared in appropriate diameters were placed transversally (Bilateral-Uniplanar) as 2 pins each for the proximal and distal fragments. Then, the outsider pin ends were fixed using fiberglass plaster, the pins' roots were cleaned with 10% povidone-iodine and the operation was completed (Figure 1).

Figure 1. Postoperative clinical view of a case that had external fixation with transfixation pinning

In closed fractures, or open fractures suitable for a windowed bandage, xylazine HCl (0.2 mg/kg IM) sedation was followed by fracture fixation using previously prepared supported bandage materials (Figure 2).

In addition to the use of antibiotics on open fractures where a windowed bandage is applied, for wound care; Antibiotic ointment (Furacin, 2% Nitrofurazon, Zentiva, Çorlu) and cicatrizant pomade (Madecassol®%1, Centella asiatica, Bayer, İstanbul) were used locally. After the preparation of bandage materials, the calves were sedated with xylazine HCl (0.2 mg/kg IM), the fragments were corrected by traction, control radiographs were acquired for appropriate reduction, and the extremities were wrapped with hydrophilic cotton, roll bandage, PVC, and fiberglass casts. Rigid stabilization and fixation were obtained (Figure 3).

Postoperative Care and Follow-up Periods

In this study, hospitalization was not performed on the pa-

Figure 2. Post-treatment clinical image of a case in which a PVC-reinforced fiberglass plaster and window bandage was applied

Figure 3. Stages of the bandage technique applied with double PVC reinforced fiberglass plaster in the treatment of metacarpus fractures in calves. A: Placing a lateral and medial PVC B: Completing the bandage process by wrapping the entire leg with fiberglass plaster C: Standing of the calf immediately after the bandaging process is completed

tients. In cases where external fixation with Steinmann pinning was performed, patient owners were instructed to confine the patients in a small area prior to discharge and daily cleaning of the pin holes with povidone-iodine (10%). In addition, postoperatively, antibiotics (Cefazolin, 30 mg/kg, IM, Cezol®, Deva, Istanbul) were administered for 7 days and meloxicam for 3 days to reduce pain (0.2 mg/kg/day, SC, Bavet Meloxicam, Bavet®, Istanbul). Moreover, patient owners were informed that open wound care should be done daily, calves should be kept in a small area, antibiotics and painkillers should be given to patients who have applied a bandage with a window to their fractures. No medical treatment was applied in cases of closed fractures, the owners were instructed to confine patients in a small area. No medical treatment was administered in the cases of closed fractures, and the owners were instructed to confine the patients in a small area. Clinical and radiological evaluations were performed by calling the patients for a follow-up at intervals of 2 or 3 weeks postoperatively.

The bandage material was removed in an average of 4-6 weeks in cases where a windowed bandage was applied, and in 4-5 weeks in cases where a bandage was applied for closed fractures.

RESULTS

Of the patients included in the study, 48 (66%) were aged 1-5 days, and 24 (34%) were aged 5-15 days. 55 (76%) of the cases belonged to the Simmental breed, 12 (16%) to the

Crossbreed Simmental, and 5 (8%) to the Brown Swiss breed. In addition, 56 (77%) of the cases were male and 16 (23%) were female. In light of the anamnesis information obtained during the clinical examination of each case, it was determined that the fracture was caused by excessive strain due to excessive traction during dystocia, traumas from the cows or other calves. In the examination performed by inspection, it was observed that 51 (71%) of the cases had closed fractures

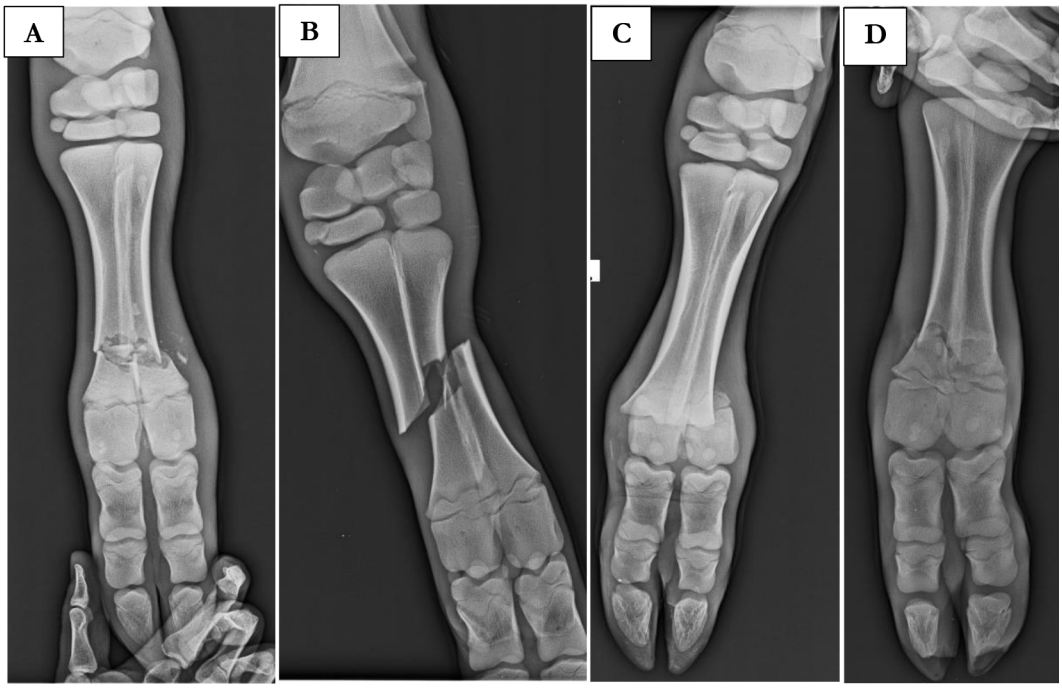

Figure 4. Classification of fractures according to radiological examination findings A: Distal diaphyseal fracture B: Middle diaphyseal fracture C: Distal epiphyseal fracture (Salther Harris Type I) D: Distal epiphyseal fracture (Salther Harris Type II)

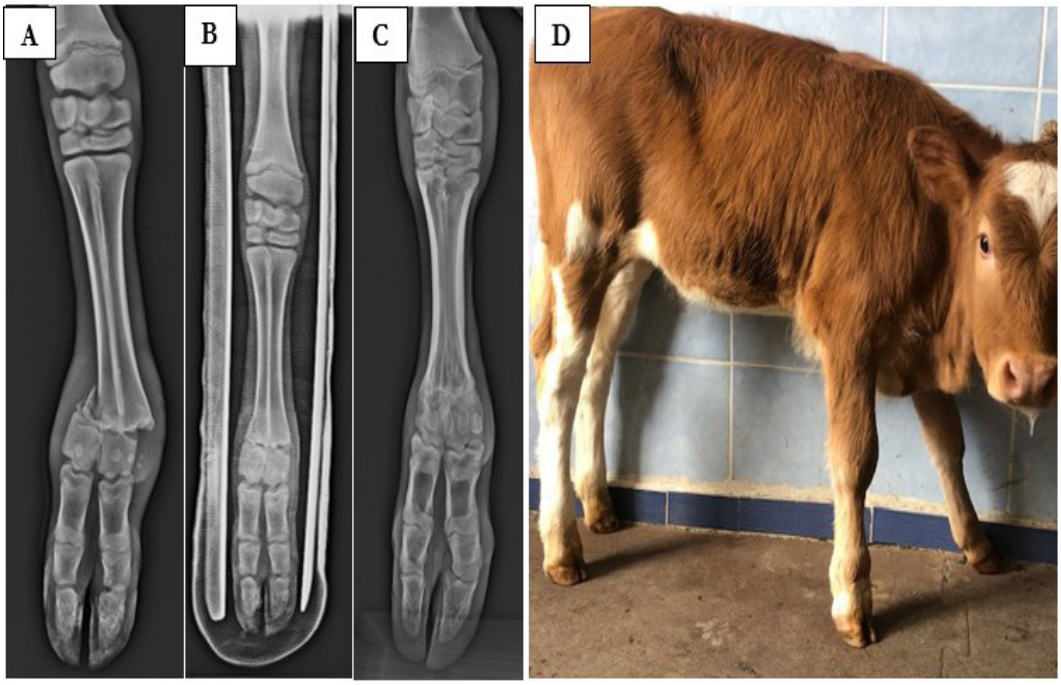

Figure 5. Treatment stages of a distal epiphyseal Salther Harris Type II fracture of a case A: Radiographic image of the fracture before treatment B: Radiograph of the fracture on day 0 after bandage with double PVC reinforced fiberglass plaster C: Radiograph of the fracture on day 35 after treatment D: Post-treatment of the calf 90 days clinical picture

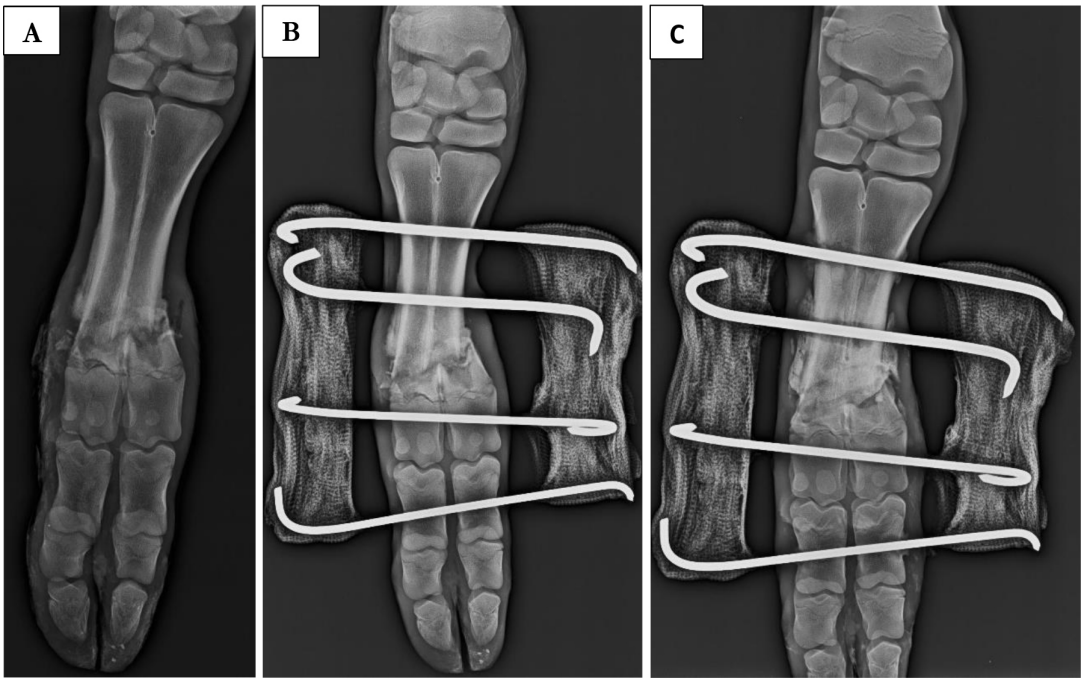

Figure 6. Treatment stages of an infected open distal diaphyseal metacarpus fracture of a case A: Preoperative radiographic image of the fracture B: Postoperative day 0 radiograph C: Postoperative 28th day radiograph

and 21 (29%) had open or infected fractures. Upon palpation examination, routine fracture findings such as crepitation and abnormal mobility were observed in all cases. According to the findings of the radiological examination performed to determine the location and shape of the fracture and to make a definitive diagnosis, it was determined that fractures were located at distal diaphyseal in 40 (55%), epiphyseal in 20 (27%) and mid-diaphyseal in 12 (18%) of the cases. In addition, when epiphyseal fractures were classified within themselves, it was observed that 12 (60%) of 20 cases were Salter-Harris Type I and 8 (40%) were Type II (Figure 4).

In the treatment, external fixation was performed using Steinmann pins in 7 of 21 cases of open fractures, and in the other 14 cases, a windowed bandage was performed with PVC and fiberglass plaster. In closed fractures, while stabilization was provided by using a fiberglass cast and caudally placed PVC in 22 of the 51 cases, fixation was achieved by using two PVC and fiberglass plasters placed medially and laterally in 29 cases. It was observed that all cases, both bandaged and operative, were able to use the related extremity without any problems immediately following the completion of the procedures. In the follow-ups of the patients with closed fractures that were bandaged, it was observed that 43 of 51 patients healed without any complications, recovery was achieved in 3 patients after the bandage was renewed due to wetting, and 5 patients died due to comorbidities (calf diarrhea, aspiration pneumonia) (Figure 5).

Five of seven patients who underwent external fixation using a Steinmann pin recovered successfully, meanwhile, two patients succumbed to infection. Full recovery was observed in 12 of 14 patients who underwent windowed bandage, and 2 patients died due to secondary infection (Figure 6).

DISCUSSION

Metacarpus fractures, which are frequently encountered in calves, are usually the result of excessive traction and incorrect manipulations during birth, and postnatal trauma. Postpartum fractures in calves usually occur within 1-10 days following the birth (Fessler & Adams, 1996, Görgül et al. 2004, Aksoy et al. 2009, Arıcan et al. 2014, Yurdakul, 2018). Male, gigantic, and have a relatively high birth weight calves are at the high-risk group for the second stage of birth (Akın, 2017). Similar to the literature, the metacarpus fractures in our cases occurred from dystocia, incorrect interventions during birth, and postnatal trauma. The calves ranged in age from 1 to 15 days, and all of them were culture breeds (Simmental, Simmental cross, and Brown Swiss). We can attribute the frequent occurrence of metacarpus fractures to the large size of the calves of especially large culture breeds, the lack of selection of seeds suitable for the breed of the animal in artificial insemination, and the larger size of male calves. In addition, the fact that these features make birth more difficult, and the resulting difficulty can be explained by the excessive force applied during the tether or manual pulling on the distal part of the metacarpus during birth.

Metacarpus fractures in calves can be in the form of open or closed fractures (Tulleners, 1986, Aksoy et al. 2009). Studies have reported that metacarpus fractures are generally localized in the distal diaphysis, metaphysis, distal epiphysis, and diaphyseal regions (Elma, 1988, Tulleners, 1986, Görgül et al, 2004, Aksoy et al, 2009, Belge et al, 2016). Metacarpus fractures in calves occur most frequently in the distal epiphysis and metaphysis (Aithal et al 2007). In their study, Sevil and Öcal (2006) reported that the weakest part of the metacarpus is the distal part. Metacarpus fractures in calves can be in the

form of open or closed fractures (Tulleners, 1986, Aksoy et al. 2009). Salter-Harris Type I is the most common type of fracture in calves with distal epiphysis and metaphyseal fractures (Tullers, 1986). Elma (1988) radiologically evaluated a total of 69 metacarpal fractures and reported that there were diaphyseal fractures in 42 of the cases, metaphyseal fractures in 20 cases, distal epiphyseal fractures in 4 cases, and epiphyseal fractures with metaphyseal fracture (Salter-Harris Type II) in 3 cases. In the clinical and radiological evaluation of 72 calves with metacarpus fractures in our study, it was revealed that 51 (71%) were closed and 21 (29%) were open fractures. It was determined that the location and type of the fracture was consistent with some of the literature and inconsistent with others. In our study, distal diaphyseal fractures comprised the majority of cases with 40, epiphyseal fractures accounted for 20 cases and middle diaphyseal fractures for 12 cases. In addition, when epiphyseal fractures were classified within themselves, it was observed that 12 (60%) of 20 cases were Salter-Harris Type I, and 8 (40%) were Type II. In our cases, we can associate the formation of the majority of fractures in the distal region of the bone with the weaker structure of the bone in the transition region from the diaphysis to the metaphysis compared to other regions. Moreover, this region is often affected when owners attempt to assist in birth with a rope and use excessive traction, resulting in fractures.

The age and general health of the animal, its genetic potential, location and type of fracture, fracture site, severity of soft tissue injury, presence of bacterial contamination, and degree of movement of the fracture should be considered in the selection of a treatment technique. In addition, fracture characteristics influence the fracture's healing process (Görgül et al, 2004, Aithal et al. et al, 2004, Gangl et al, 2006). Open fractures with severely traumatized soft tissues commonly become infected, making repair significantly more difficult. If contamination occurs at the fracture site and persistent infection develops, the treatment is probably to fail (Desrochers et al, 2004, Arıcan et al, 2014). Many internal and external methods have been described in the literature for the treatment of metacarpus fractures (Arıcan et al, 2014, Mulon, 2014, Belge, 2016). Bandage (PVC, fiberglass, and Thomas splints) applications are generally preferred in closed metacarpus fractures with good outcomes. On the other hand, some open metacarpus fractures can be treated with windowed bandages if suitable and necessary, whereas some open fractures that are not suitable for a windowed bandage can be treated via many methods such as plate osteosynthesis, intramedullary pinning, and external fixation with transfixation pins (Ferguson, 1982, Mulon 2014, Arıcan et al, 2014, Salcı et al, 2016, Gillespie et al, 2018, Yadav et al, 2020, Yurtal et al. et al, 2020). In our study, while determining the treatment options, the most appropriate treatment option was determined by considering the fracture's location, shape, connection with the external environment (open and closed fracture), the presence of contamination, the economic value of the animal, and the time of fracture. Infection occurred in all 21 cases of open fractures in the current study, external fixation with transfixation pins was applied in 7 of these cases, while a windowed bandage with PVC+fiberglass plaster was applied in the 14 cases. No infection was detected in any of the 51 cases of closed fractures, and in 22 of these cases, a PVC+fiberglass plaster bandage was placed on palmar side, while PVC was placed on both lateral and medial sides of the metacarpus in 29 cases. After obtaining reduction and stabilization of the fracture with a wrap of materials, fiberglass plaster was applied to strengthen the fixation. The uneventful recovery of 46 out of 51 cases in which bandages were applied in closed fractures, 12 out of 14 cases in which a windowed bandage was applied in open fractures, and 5 out of 7 cases in which external fixation was applied with transfixation pins revealed the suitability of the selected treatment options. We may attribute the absence of any adverse events in the cases treated with a bandage with the high recovery rate, to the fact that bandage treatment is more convenient for the treatment of metacarpus fractures. Furthermore, this can be associated with the two PVCs placed on the lateral and medial of the fractured metacarpus. In the cases which resulted in death, it was understood in the controls that the causes of death were not due to the treatment, but rather the result of the negligence of the patients' owners. In open metacarpus fractures, the risk of infection is quite high, which reduces the likelihood of treatment. For this reason, patient owners must be made aware of this matter.

CONCLUSION

Numerous treatment options exist for metacarpus fractures, which are prevalent in calves and cause significant economic and productivity losses if left untreated. When determining the treatment option, the condition of the fracture (closed or open fracture), shape, location, presence of infection, cost of treatment, the economic value of the animal, and care conditions during the recovery period should be considered. Even if post-operative care are carefully monitored, infection becomes inevitable as the calves are housed in a barn environment. For this reason, it has been concluded that non-invasive bandage techniques are less costly and have a higher recovery rate than invasive methods, particularly for the treatment of distal metacarpus fractures.

DECLARATIONS

Ethics Approval

This study was conducted with the approval of Kafkas University Animal Experiments Local Ethics Committee (Approval no: KAÜ-HADYEK/2022-048).

Conflict of Interest

The authors report no conflicts of interest. The authors alone are responsible for the content and writing of paper.

Consent for Publication

Not applicable

Author contribution

Idea, concept and design: UA, İÖ, ÖA and ET

Data collection and analysis: UA, İÖ, ÖA, CŞE, EK, UY, ET, MT and AY

Drafting of the manuscript: UA, UY and ET

Critical review: UA, İÖ, ÖA, CŞE, EK, UY, ET, MT and AY

Data Availability

The datasets analyzed during the current study are available from the corresponding author on reasonable request.

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Research Article

A first look at the history of Turkish veterinary medicine with Mehmet Âkif (Ersoy)

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Mehmet Âkif

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ABSTRACT

The history of the Agricultural and Civil Veterinary School in Halkalı, where Mehmet Âkif was a student first and then served as a lecturer, is in a very important place in terms of teaching veterinary medicine and agriculture in Turkey. The opening of the Halkalı Agricultural College and the opening of the Civil Veterinary College coincided with the same time period. The Civil Veterinary College was the partner of the Agricultural College opened in Halkalı in the first years. The Civilian Veterinary College was later moved to Sultanahmet, and agricultural education was continued in Halkalı. The teaching staff of the Halkalı Higher Agricultural College has published a journal called "Halkalı Ziraat Mekteb-i Âlisi Mecmuası". Mehmet Âkif Bey (Ersoy), who was on the teaching staff of the College at that time, wrote an article containing the history of the School in the first issue of the Magazine. The article provides information about the Halkalı Agricultural College, as well as information about the beginning of the teaching of Civil Veterinary College. In the article, besides the information about Halkalı Higher Agricultural School, information about the beginning of civilian veterinary medicine education is also given. In this study, the relationship of Mehmet Âkif Ersoy's article, which was determined to be related to the history of the profession, with the research conducted on the teaching of veterinary medicine in Turkey and the history of agricultural teaching, was analyzed. The article, which contains information about Âkif's profession, was evaluated in terms of the history of veterinary medicine and presented to the field of science history and research ethics.

Mehmet Âkif (Ersoy) ile Türk Veteriner Hekimliği Tarihine bir ilk bakış

ÖZ

Mehmet Âkif'in, önce öğrencisi oldugu, sonra öğretim elemanı olarak görev yaptığı Halkalı'daki Ziraat ve Sivil Veteriner Okulu tarihi Türkiye'de veteriner hekimliği ve ziraat öğretimi açısından çok önemli bir yerdedir. Halkalı Ziraat Okulunun açılışı ile Sivil Veteriner Okulunun açılışı aynı zaman dilimine rastlamıştır. Sivil Veteriner Okulu ilk yıllarda Halkalıda açılan Ziraat Okulunun paydaşı olmuştur. Sivil Veteriner Okulu daha sonra Sultanahmet'e taşınmış, Halkalıda ziraat öğretimi sürdürülmüştür. Halkalı Yüksek Ziraat Okulunun öğretim kadrosu "Halkalı Ziraat Mekteb-i Âlisi Mecmuası" adında bir dergi yayınlamıştır. O dönem Okulun öğretim kadrosunda yer alan Mehmet Âkif (Ersoy), Derginin ilk sayısında Okulun tarihçesini içeren bir makale yazmıştır. Makalede Halkalı Ziraat Okulu'na ilişkin bilgilerin yanı sıra sivil veteriner hekimlik öğretiminin başlangıcına ait bilgiler de verilmiştir. Âkif'in makalesi, hem veteriner hekimliği öğretimi tarihi hem de ziraat öğretimi tarihi ile ilgili bazı çalışmalarda da kullanılmıştır. Bu çalışmada, Mehmet Âkif Ersoy'un meslek tarihi ile ilgili olduğu belirlenen makalesinin Türkiye'de veteriner hekimliği öğretimi ve ziraat öğretimi tarihi hakkında yapılmış araştırmalarla ilişkisi analiz edilmiştir. Âkif'in veteriner hekimliği öğretimine ilişkin bilgiler içeren makalesi, veteriner hekimliği tarihi açısından değerlendirilerek bilim tarihi ve araştırma etiği alanına sunulmuştur.

INTRODUCTION

Mehmet Âkif Ersoy, a veterinarian, also served as a lecturer in, Literature at the Darülfünun[1]; Literary of Faculty, Literature of Ottoman (Dinçer, 2011), Official Correspondence Language (Çeşme, 2014b) at the Halkalı Agricultural College, and Language of Turkish at the Farm Machinist School (Dinçer, 2011). Âkif, who served on the teaching staff at these institutions, was also appointed as a lecturer of Health Police Code at the Civil Veterinary College, at the Official Correspondence of Veterinary, at the Law of Commercial and at the Law of Medicine, but resigned before starting to work at this School (Dinçer, 2011).

Âkif has also served in the "Ottoman Society of Veterinary Scientific" (Osmanlı Cemiyet-i İlmiyye-i Baytariyesi), the first non-governmental organization of the veterinary medicine profession in Turkey, and in "The Journal of Veterinary Scientific" (Mecmua-i Fünûn-i Baytariye), the publication organ of this Association (Anonim, 1908, Dinçer, 1964, Dinçer, 1965).

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Âkif was also the President of the "Veterinary Medicine College Alumni Association" founded in 1910, and together with Cafer Fahri (Dikmen) and Civani Bey, they published the "Veterinary Science Journal" (Risale-i Fenn-i Baytari) (Dinçer, 1964).

The lack of a civilian veterinarians in the Ottoman geography to provide health services to the animals of the people has been a serious obstacle to preventing epidemics; it was officially established in 1867 that it would be possible to eliminate this problem by expanding the veterinary class within the Military Academy (Tan, 2017). In the following period, according to Dinçer (Dinçer, 1976), the necessity of training civilian veterinarians in Turkey was stated in the journal called "Vasıta-i Servet", which is the first periodical publication about veterinary medicine (Anonim 1880a, Anonim 1880b). In the aforementioned journal, it is stated that permission was requested from the sultan to open the Civil Veterinary School together with the Agricultural School in Ayamama Farm in 1881(Anonim 1880c). Information on the establishment of a commission to determine issues such as the management of the School, the curriculum, and the functioning of a new veterinary school has been given in the Civilian Medical College and the decision has been made and the decision has been submitted to the Sultan (Anonim, 1888a). In the second meeting held afterwards, Alfort Veterinary College was taken as an example, the admission conditions of the students to be admitted to the school, the education period of the school, the curriculum and the need for instructors were determined and decided. It has been stated that this Commission will convene again and that the certificate to be prepared as a result of this meeting will be submitted to the Ministry of Education (Anonim, 1888b). At the meeting of the Commission on April 21, 1888; the conditions set for the students to be admitted to the school, the duration of the school and the courses were determined. It has been stated that the first two classes of the school can take classes at the Civilian Medical College; the last two classes will be appropriate to be held on a farm close to Istanbul and where it will be easy for faculty members to reach. It has been stated that it will be appropriate for the students who will study at the Medical School to appoint 11 teaching staff and 12 assistants for their teaching and management jobs (Anonim 1888b). A commission was established for the opening of a Civil Veterinary College, in which Mehmed Ali Bey was also appointed. The Commission has prepared a protocol and stated that the practice of the Pasteur method vaccination should be taught to veterinarians and municipal doctors (Birinci, 2018a, Birinci, 2018b, Erk&Dinçer, 1970). It has been stated that veterinarians are needed to treat and prevent the spread of animal diseases that occur throughout the country, and for this, a veterinary class should be created at the Civilian Medical College (Birinci, 2018a, Birinci, 2018b, Erk&Dinçer, 1970, Kaya Doğanay, F. 2017). It has been emphasized that the veterinary medicine class to be opened will need four years of education, and it is imperative to purchase a farm where the practice can be done in order for the teaching to be good. Mehmet Ali Bey stated that since the purchase of a farm and the construction of a school will be costly, a part of the Agricultural College, most of which has been completed on the Halkalı Farm, can be allocated to veterinary classes (Birinci, 2018a, Birinci, 2018b, Kaya Doğanay, 2017). This proposal was considered appropriate, and it was decided that the two classes that will learn almost the same lessons given at the Medical School should continue teaching at this School, and the last two classes that should practice on animals every day should continue teaching at the School in Halkalı. It has been decided that the students who will be taught at the Medical College will be boarding during the day and the classes who will be taught on the farm will be boarding (Erk&Dinçer, 1970, Bekman, 1940, Erk, 1966, Subhi Edhem, 1918). And thus, the first students of Veterinary College started teaching in the Medical College in Ahırkapı district in 1889 during the day (Dinçer, 1976, Birinci, 2018a, Birinci, 2018b, Erk&Dinçer, 1970, Kaya Doğanay, 2017, Bekman, 1940, Erk, 1966, Subhi Edhem, 1918, Tüzdil, 1955).

A draft law on the opening of an Agricultural College was prepared and submitted to the Ministry by Amasyan Efendi, a member of the Assembly of the Ministry of Trade and Agriculture (Demirel&Kaya Doğanay, 2011, Yıldız, 2012). The construction of the School was started in 1884 under the supervision of a commission established under the name of the Preparatory Commission within the Mining Council and headed by District Governor Cin Izzet Bey, but it could not be completed (Çeşme, 2014a, Demirel&Kaya Doğanay, 2011, Yıldız, 2012). A Commission including Mehmed Ali Bey and Agricultural Counsellor Ağaton Efendi was established to finish the construction (Birinci, 2018a, Birinci, 2018b, Demirel&Kaya Doğanay, 2011, Yıldız, 2012). After the construction completed by the efforts of this Commission, 19 students in Civilian Veterinary Classes who were opened at the School of Civil Medicine in Halkalı were transferred in 1891 and the first agricultural students were enrolled a year later, and the School began teaching under the name of "Halkalı Agricultural and Veterinary College" (Birinci, 2018a, Birinci, 2018b, Demirel&Kaya Doğanay, 2011, Yıldız, 2012). The school in Halkalı became an independent agricultural school in 1894 and graduated its first graduates in 1896. Foresters were also trained at this School until 1910 (Çeşme, 2014a, Yıldız, 2012).

It is aimed to present Mehmet Âkifs article about the School, which he graduated from and later worked as a lecturer, in the field of veterinary medicine history, by evaluating it in terms of the history of veterinary medicine teaching in Turkey. Akif's article on the history of Turkish veterinary medicine was compared with similar studies and analyzed. The article has been opened for discussion in the fields of history of science and research ethics.

MATERIAL and METHOD

The main material of the study is the title of "School Location, Structures, History", which is located between the second and tenth pages of the first issue of Halkalı Ziraat Mekteb-i Âlisi Mecmuası magazine, which started to be published in April 1917. The research has been supported by research conducted on the history of veterinary medicine teaching and agricultural teaching in Turkey. Transcription and transliteration of the article written and published in Arabic letters were made, and document analysis and content analysis methods were followed from qualitative research methods.

RESULTS

Halkalı Agricultural College Journal; No 1, April 1917

The Location of the School, Buildings, Its History, Scholar Akif Bey (p.2-10) (Figure 1-9)

Âkif started his article by giving the geographical structure of the location where the School is located, its distance from nearby settlements and metric details. Halkalı Agricultural College is located in the north-west of İstanbul on a hill 112 meters above the sea. It is located 16 km from İstanbul, 9 km from the nearest railway station, Ayastefanos[2]. At its location, three basic structures were mentioned, and the existence of the main building of the School, the sample farm and outbuildings belonging to the farm were explained on the land. The width of this rectangular-shaped view is 123.5 m, and the length is 137.5 meters.

The main School building has two floors, along with the ground floor. The left side of the first floor is divided into classrooms and students' rest, in turn, there is a bright corridor with a view of the surroundings, and on the right side there are classrooms with rooms for the principal, teacher and employees. On the second floor there is a large dormitory dedicated to students on the left side, and on the right side there are bedrooms belonging to teachers and employees, as well as rooms for studying botany and plant diseases, zoology and agricultural pests (atelier).

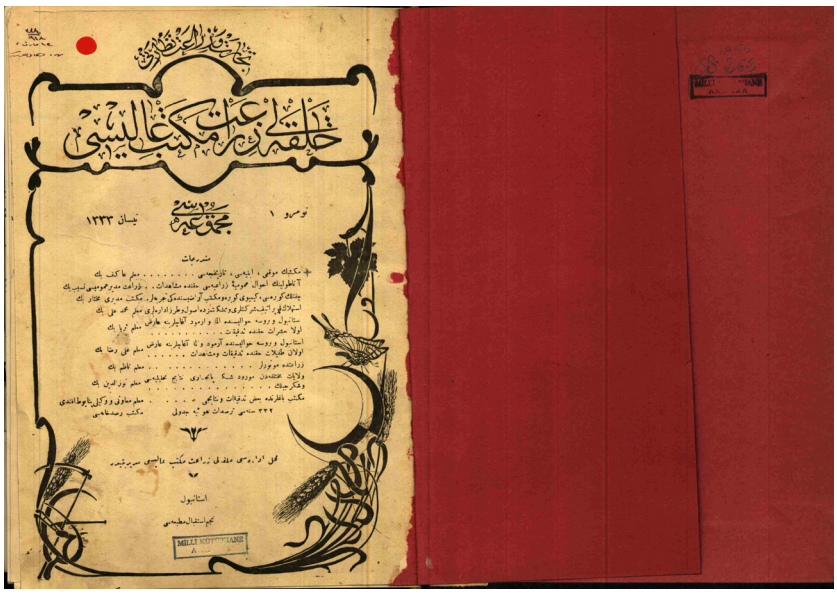

Figure 1. Halkalı Agricultural College Journal; No 1, April 1917 (Cover).

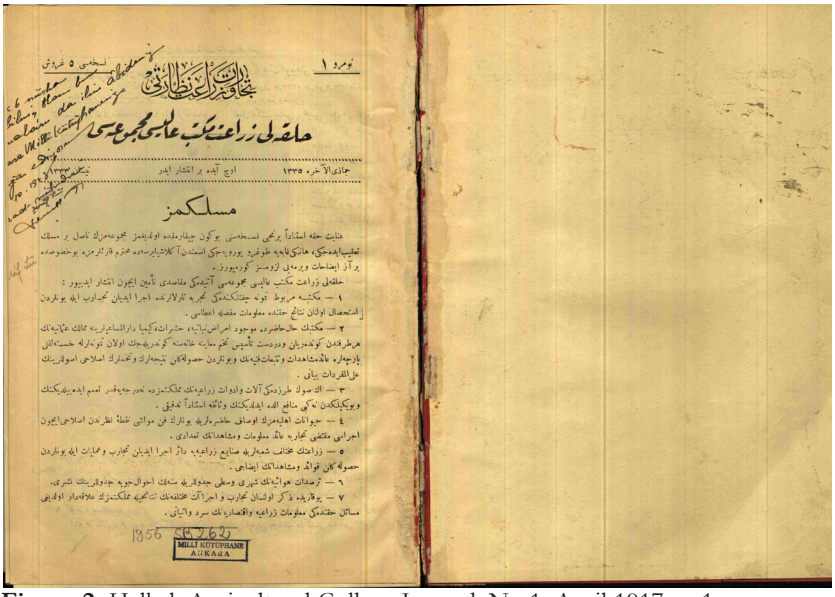

Figure 2. Halkalı Agricultural College Journal; No 1, April 1917, p. 1

In the eastern wing of the building there are two chemical and assay houses, one dedicated to teachers and the other to students, as well as an amphitheater, soil science atelier, carpenter and a spacious leveling workshop organized for the

[2]Nowadays Yeşilköy.

new Farm Machinist Departmant. On the north side there is a dairy house, a machine building, and horse, cow, and ox barns. There is also a warehouse, a haystack, a repair shop, a laundry room, a hammam in the west wing. There is a poultry house

of Cevdet Pasha to the Trade Supervision, before which there was no such institution and institution manager. Seeing that it would not be enough to send students to Europe alone to ensure the development of a scientific, modern farming, gra-

Figure 3. Halkalı Agricultural College Journal; No 1, April 1917, pp. 2-3.

and an observatory 15 meters ahead of this facade. Further on, cattle cowhouse, sheep corral with a barn. The land located on the south side includes gardens of plants and fruit trees, as well as various trees and American vine rods and specimen vineyards. The land facing the northeast facade is covered with a forest of pine nut trees. The fertilizer storage area is on the north side. A kilometer from the southern facade there are vegetable gardens on the southwestern border. The total land area of the farm is shown below*.

Agricultural land and pasture	**3121.5**
Stony land where Agriculture cannot be done	**2508**
Vineyard, Garden, Nursery, Buildings	**128**
Forest, Meadow	**164**
Stone Quarry	**63**
	5984.5

*See Figure 3

It has been stated that the soils of Halkalı are a Miocene Period layer. The soil is clayey-calcareous, clayey, or clayey with dirt. The non-agricultural part is in pasture form. It is curious that this situation was not taken into account when the Halkalı hill was *"absolutely not conducive"*[3] to the establishment of such a higher education institution and a sample farm related to agriculture, both the location and the soil structure of its land, and this is a curious issue. Therefore, we need *"to settle"* this point at the beginning of the work. Amasyan Efendi first expressed the idea of opening an Agricultural School in our country and training specialists in this field. He graduated from the Grignon National Agronomique School of France and was appointed Director of Agriculture at the time of the Ministership

dually abandoning agriculture in the old ways, Amasyan Efendi thought about creating an institution that had not existed in our country until then, and worked hard to turn it from request to action. Finally, it was decided to establish an agricultural school during the Ministry of the late Suphi Pasha.

In addition, all the farms close to İstanbul, such as İncirli, Ayamama, and the land is quite suitable for agriculture, were applied to so that both the teachers who will be assigned to the teaching post can easily go back and forth and be an excellent example for the public. But, unfortunately, none of them could be bought. However, those who were in favor of the establishment of the agricultural school were in a very pitiful minority compared to those who were against it. For this, it was necessary either to abandon this initiative, or to take a lenient attitude to the choice of land. Seeing this situation, Amasyan Efendi bought the school's land from Rukiye Hanım, the wife of the Egyptian Khurshid Pasha, for 2000 liras. A commission was created under the name of the Preparatory Commission under the same Ministry. Its chairman was District Governor Cin Izzet Bey. This person is the General Staff Major General late İzzet Pasha, who later founded the Melinite Institution in Zeytinburnu. The construction was started under the supervision of the Commission mentioned in the plan prepared by Amasyan Efendi. Since the first part of the school and the allowance were also over, the building remained empty for two years.

Two years later, during the Ministry of Zihni Pasha, another Commission consisting of Agricultural Director Nuri, Agricultural Bank Accountant Zuhdi, Veterinary Affairs Consultant Mehmed Ali Bey, Agricultural Inspector Ağaton Efendi was established, and the rest of the buildings were completed

[3]The statements given in italics in quotation marks belong to the author. This important criticism, honestly made and owned by the author, wanted to be especially emphasized for the reader.

through this Commission. The most valuable and hardworking member of this second Commission, Mehmed Ali Bey, was appointed as a Veterinary Affairs Consultant at the suggestion of Amasyan Efendi when he was an instructor at the Military Veterinary College "General Diseases and Preventive

Figure 4. Halkalı Agricultural College Journal; No 1, April 1917, pp. 4-5

Health" at the Ministry of Commerce and then appointed as a Veterinary Affairs Inspectorate.

After the construction work of the Halkalı Agricultural College was completed, the first students of Civil Veterinary

to a serious loss of agricultural wealth during the Ministry of the late Zihni Pasha, and was raised at a Cabinet meeting. As a result, it was decided to open a special class at the Civil Medical College to teach the practice of Pasteur method vaccination. After that, a commission including Mehmed Ali Bey was established within the Medical College to be taken care of by notifying the relevant departments with a notification.

Mehmet Ali Bey stated that it is "unacceptable" to teach those who have nothing to do with human or veterinary me-

Figure 5. Halkalı Agricultural College Journal; No 1, April 1917, p. 6

College were enrolled in the school. As follows: from time to time, the attention of the Government was drawn to animal diseases, which caused great damage to the country by becoming an epidemic, killing thousands of animals and leading

dicine the practice of the Pasteur method vaccination, which was decided to be taught and disseminated by the Council of Ministers, cannot be accepted.

Stating that infectious diseases cannot be prevented by such a method, measures to be taken against infectious animal diseases will only be successful thanks to veterinarians (however

Figure 6. Halkalı Agricultural College Journal; No 1, April 1917, p. **7**

took the teaching of animal health), and a "Civilian Veterinary College" should be opened for this, Mehmed Ali Bey submitted a proposal to the Commission on this issue.

Although the Commission found the proposal very appropriate, it stated that it was impossible to open a four-class school due to the lack of the Ministry's budget.

In addition, Mehmet Ali Bey suggested that courses such as physics, chemistry, botany, zoology could be taken with Medical School students, and courses such as anatomy-physiology could also be taught, The first two classes of the School to be opened could be taught in a Civilian Medical College during the regular school days and the other two classes could be completed in Halkalı Agricultural College that would be completed by then. The Commission has decided to prepare a detailed report on this issue, considering Mehmet Ali Bey's proposal and explanation appropriate. The reasoned opinion

Figure 7. Halkalı Agricultural College Journal; No 1, April 1917, p. 8

written and prepared by Mehmet Ali Bey was submitted to the Central authority and, an order of the Sultan was issued based on the grounds that the legal structure was also deemed

appropriate. After the construction of the Halkalı Agricultural College was completed, the students of the Civil Veterinary College were taken to the School for the first time.

logy courses from their own faculty members.

Of the 25 students enrolled in the Civilian Veterinary Scho-

Figure 8. Halkalı Agricultural College Journal; No 1, April 1917, p. 9

The upper classes of the Civil Medical College were receiving their education in the building located in Ahırkapı, which was insufficient for the physical structure of this building even for Medical College students. However, provided that they are

ol opened in 1889 in the Civilian Medical College building, 19 of them went to the School in Halkalı, which was completed in 1891, two years later, as a boarder in 1891. Agricultural students were admitted to the School a year later, and the official

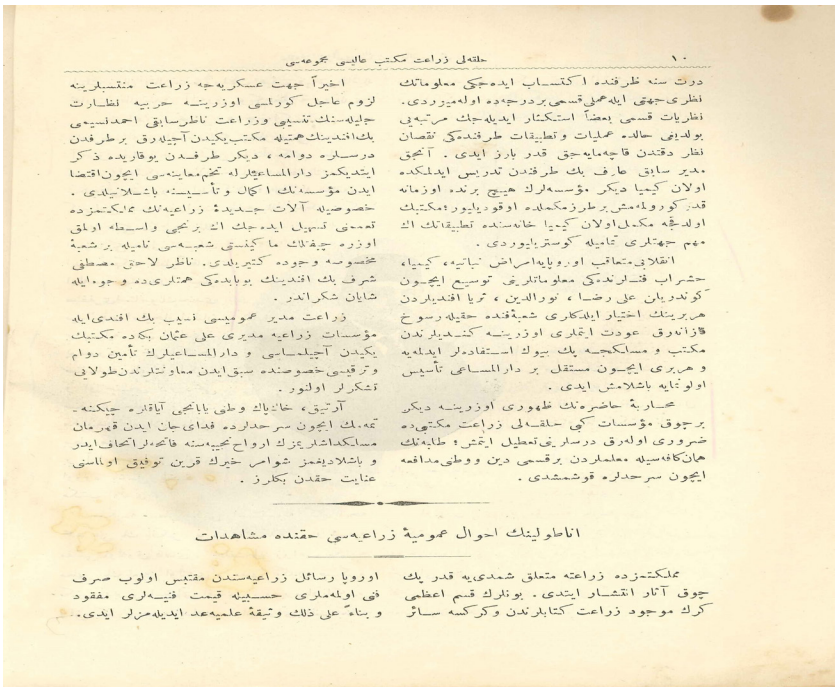

Figure 9. Halkalı Agricultural College Journal; No 1, April 1917, p. 10

graduates of Civil Secondary School, the registered veterinary students continued their courses with the Medical College students in the said building and only took anatomy and physio-

opening of the Agricultural School was also held after agricultural students were enrolled. The school is also called "Halkalı Agricultural and Veterinary College".

Mehmet Âkif stated in the inscription decked on the School that agriculture depends on animals such as cattle and sheep; that the building combines these two professions that are very useful for the country is expressed in a "poetic" language and transferred what is written in the inscription to his article.

Mehmed Ali Bey, who founded the Civil Veterinary College, was also appointed as the first Director of the School, which was first named the "Halkalı Veterinary College", and a year later the "Halkalı Agricultural and Veterinary College". When he becomes a director, his military rank is lieutenant colonel.

Before the agricultural student was admitted to the school, İstrati Efendi was appointed as the Second Director and Zakaryan Efendi was appointed as the farm manager. İstrati Efendi taught "French and Cattle and Sheep Farming" and Zakaryan Efendi taught "General Agriculture" to Veterinary College students.

Nuri Bey was the Director of Agriculture when Halkalı Veterinary College was opened. İstrati Efendi was first appointed Director of Statistics and then retired. After Nuri Bey moved to the General Directorate of Editorial Affairs of the Ministry of Foreign Affairs, the Agricultural Directorate was also assigned to Cemal Bey, the General Director of the Ziraat Bank of the period, with an additional assignment. Cemal Bey was present as the General Director at the official opening of the Halkalı Agricultural and Veterinary College. During the period of Cemal Bey Agricultural Directorate, a five-branch Board was established in the Ministry under the name of "Agricultural Science Board" (Ziraat Heyet-i Fenniyesi); an inspector was appointed at the head of the branches. Aram Efendi has also been made the senior maneger of this Board. Agaton Efendi, Mazhar Bey, Vahan Efendi were the inspectors of each branch. Mehmed Ali Bey was also appointed as the Fifth Branch Inspector, who was engaged only in veterinary affairs.

The Forest, Mining and Agricultural Branches were merged and a Ministry was established in Rumi 1309[4] (1893-1894); Selim Efendi, who was later promoted to the rank of General, was appointed Minister. At the same time, the Scientific Council of Agriculture was completely removed from the Ministry of Commerce and given to the new Ministry.

Veterinary Class students from the School in Halkalı graduated from two semesters in 1893 and 1894 and received diplomas.

The lack of physical capacity of the Medical School required that the daytime veterinary classes in the Medical School be moved to the Halkalı Veterinary College on a mandatory basis and that these classes be transferred to boarding education. Since physical disability is also present in the School building in Halkalı, it has created the problem of housing two eight-class Schools in this building. For this reason, the veterinary classrooms were moved to a rented building on Cündi Square in İstanbul; thus, the Civil Veterinary College became a four-class co-educational and boarding decoupled School.

After the Civil Veterinary College was brought to İstanbul from Halkalı, Mehmed Ali Bey continued his duties as the Director and Branch Inspector of this School. Mazhar Bey was appointed as the Director of Halkalı Agricultural College. After Mazhar Bey, İstrati Efendi was appointed as the Director of the School and Refik Bey was appointed as the Deputy Director. İstrati Efendi was appointed as the General Director of Agriculture, and Refik Bey became the Director, and Su'ad bey was appointed as his deputy. After Refik Bey became the Second Director of Ziraat Bank, Chemist Arif Bey, who has been teaching chemistry since the opening of the school, became the Director. After Arif Bey, this task was given to Muhtar Bey

The first graduates of the Halkalı Agricultural School received their diplomas in 1896. From then on, until the beginning of the war[5], the School graduated twenty, thirty students every year. Forestry education was also provided to the students at this School, and the Halkalı Agricultural College trained the forest officers until the Forestry College was established near Büyükdere.

The level of education of the Halkalı Agricultural College is quite high compared to the education of the period in the country. In addition, it has been stated that although theoretical courses are given at a high level in the school due to the fact that the students enrolled in the School have received different education before higher education and that all the needs of the Agricultural College have not been completed, the applications of the courses remain noticeably incomplete. Besides, it has been stated that the chemistry course given by the previous Principal of the School, Arif Bey, is more advanced than the courses of many institutions. The most important aspects of the application in the excellent chemistry laboratory of the school have been fully demonstrated to the student.

After the revolution[6] Ali Riza, Nureddin and Süreyya Efendi, who were sent to Europe to increase their knowledge in the fields of plant diseases, chemistry and entomology, made quite use of their professions and schools upon their return by gaining expertise in their chosen field of science, and a special working unit was established for each of them.

Like many other institutions, the Halkalı Agricultural College, like the beginning of the war, Decommissioned its classes on a mandatory basis, almost all of its students and some of its teaching staff rushed to the fronts to defend religion and the motherland. Upon the urgent and compulsory need for those who studied Agriculture during the war years, with the assistance of the Ministry of Defense and with the efforts of Ahmed Nesimi Bey, who had previously served as the Director of Agriculture, the School in Halkalı was reopened. New study units have been established at the school and intensive studies have been carried out to eliminate the deficiencies of the existing ones. A new and special branch has been opened under the name of Farm Machinist Department, especially as the first tool that will facilitate the spread of new agricultural tools in our country.

The new Minister Mustafa Şeref Bey was thanked for his efforts in this field, the Director General of Agriculture Nesib Bey and the Director of Agricultural Institutions Ali Osman

[4]Since the month is not specified in the year, Rumi calander shows the dates 1893 – 1894 as Gregorian dates.
[5]In the article, with the expression "war", Âkif refers to the First World War.
[6]II. Constitutional Monarchy.

Bey for the start of the School's re-education, the creation and development of working units and their assistance in the lessons.

Âkif finished his article with the words "The Conquerors dedicate the souls of our heroic colleagues who sacrificed their lives at the borders so as not to trample the unblemished-homeland on foreign feet, and we expect from God to achieve the success of the good deeds we have started".

DISCUSSION

Vasıta-i Servet is the first journal in the field of veterinary medicine and agriculture, published in 1880 (Dinçer, 1976, Demir,2017). July April 1917- July 1918, the Halkalı Agricultural College Journal was published by the faculty members of the Halkalı Agricultural College (Kadıoğlu, 2002). In Dinçer's work (Dinçer, 2011), the statement that Mehmet Âkif was a teacher at the Halkalı Agricultural College "Official Correspondence Language" in 1919 can be considered as a document indicating that the author of the article titled "Location, Buildings, History of the School", which appeared in the first issue of the journal, is Mehmet Âkif Ersoy.

After the decision to open an Agricultural College, the purchase of a farm in İstanbul for the school, information about the physical characteristics of the School and the School farm, information about the construction process of the School (Çeşme, 2014b, Demirel&Kaya Doğanay, 2011, Yıldız, 2012) coincide with the information provided by Âkif in his study. The fact that the unfinished construction of the school was highlighted in Âkif's study due to the expiration of the appropriation allocated for the school should be considered as an example related to the economic crisis (Dumont, 1995, Georgeon, 1995) seen as a result of the impact of foreign capital on the country's economy at that time.

The fact that the photos (Figure 5, Figure 6, Figure 7, Figure 8) of the administrators of the School included in the study at the Halkalı Agricultural College are in line with the names included in the study of the Çeşme (Çeşme, 2014a) can be considered as an example of Âkifs meticulous preparation of his work.

Mehmet Âkifs article also contained information about the opening of the Civil Veterinary College. Âkifs article stated that the elimination of epidemic animal diseases in the country and the fight against epidemic animal diseases will only be possible with veterinarians who have been trained in this field (Tan, 2017) was also included in Tan's study. It is noteworthy that the information in the article about the opening of the Civil Veterinary Medicine School contains close data with the information in other recent articles (Anonim, 1880a, Anonim, 1880b, Anonim, 1880c, Anonim, 1888a, Anonim,1888b, Birinci, 2018a, Birinci, 2018b, Kaya Doğanay, 2017, Demirel&Kaya Doğanay, 2011, Yıldız, 2012).

The fact that the information provided in Mehmet Âkifs publication about forestry teaching is similar to the information in other studies (Yıldız, 2012, Çeşme, 2014b, Gümüş, 2016) can be considered as a sign that Âkifs publication is a meticulously prepared study.

Mehmet Âkifs article is included among the sources of articles and studies (Çeşme, 2014a, Kadıoğlu, 2002, Çeşme, 2014b, Melikoğlu Gölcü, 2019) related to the history of Halkalı Agricultural College and veterinary medicine teaching. It is noteworthy that the information and statements about the civilian veterinary school in the oldest book on the history of veterinary medicine written in the Latin-based Turkish alphabet (Bekman, 1940) overlap with Mehmet Âkifs work.

In Kadıoğlu's study (Kadıoğlu, 2002), it was reported that the article appeared on the two and eighth pages of the First Issue of the Journal in the information of Mehmet Âkifs article and in the bibliographic imprint information. In the bibliography lists of some articles related to veterinary medicine teaching and agricultural teaching (Çeşme, 2014a, Çeşme, 2014b, Melikoğlu Gölcü, 2019), Âkifs article is included, again, as in Kadıoğlu's study (Kadıoğlu, 2002). However; in this study, it was revealed that Âkifs article was included between the two and tenth pages of the first issue of the Halkalı Agricultural College Journal.

CONCLUSION

As a result, Mehmet Âkifs article has become a source for historical studies related to the teaching of veterinary medicine and the teaching of agriculture. The content of this article is in line with studies based on archival research conducted in the following years. It is important for Âkif to be and as a college student, a civil servant, and a teaching staff in Halkalı. Because this indicates the possibility of confirming this information with primary sources through interviews with people who took part in the opening process of Halkalı Agricultural College and Civil Veterinary College.

All these information and data shows that Âkifs article has the feature of a meticulously written, original historical study and that he left an important source for the history of Turkish veterinary medicine.

DECLARATIONS

Ethics Approval

The permission of the ethics committee is not required for this study.

Conflict of Interest

There are no conflicts of interest between the authors.

Consent for Publication

Not applicable.

Author Contributions

The entire workload of the study was fulfilled by AÖ.

Data availability

Data supporting the findings of this study are available from the responsible author upon reasonable request.

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