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RESEARCH ARTICLE

Distribution of Bacterial Agents and Diagnosed Diseases in Samples Sent to the Jockey Club of Türkiye İstanbul Equine Hospital Laboratory: A Retrospective Study (2015-2019)

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ABSTRACT

The categorization of diseases identified in Turkish race and breeding horses, as well as the bacterial agents derived from these equines, have not been thoroughly investigated. This study aimed to examine the bacterial agents in samples taken from sick horses brought to the covering stations and animal hospitals operating under the Jockey Club of Türkiye (TJK) between January 1, 2015, and December 31, 2019, and the retrospective distribution of diseases in the horses from which these samples were taken. It was determined that the most frequently sent sample to the TJK Istanbul Equine Hospital Laboratory was tracheal lavage fluid (47.3%) and that the samples were mostly sent in the spring (33.8%) and autumn (30.8%) seasons. It was determined that the most common respiratory system infections (49.37%), followed by genital system infections (29.11%) and gastrointestinal system infections (18.57%) were seen in the horses from which the samples were taken. *Escherichia coli* (15.6%) was the most commonly detected strain in all submitted samples. In respiratory system infections, the most identified pathogens were *Streptococcus equi* subsp. *zooepidemicus* (14.53%), followed by *Streptococcus equi* subsp. *equi* (10.26%). As a result, it is expected that the findings obtained from this study will contribute to taking specific measures against diseases or bacterial agents with high prevalence in enterprises operating within the TJK and minimizing losses due to horse diseases.

Keywords: Escherichia coli, Horse, Respiratory system diseases, Streptococcus equi subsp. zooepidemicus, Tracheal lavage

Türkiye Jokey Kulübü İstanbul At Hastanesi Laboratuvarına Gönderilen Numunelerdeki Bakteriyel Etkenlerin ve Teşhis Edilen Hastalıkların Dağılımı: Retrospektif Bir Çalışma (2015-2019)

ÖΖ

Türkiye'de damızlık ve yarış atlarında teşhisi yapılmış hastalıkların ve bu atlardan elde edilen bakteriyel etkenlerin sınıflandırılmasına dair kapsamlı bir çalışma bulunmamaktadır. Bu çalışmada, 1 Ocak 2015-31 Aralık 2019 tarihleri arasında, Türkiye Jokey Kulübü (TJK) bünyesinde faaliyet gösteren aşım istasyonu ve hayvan hastanelerine getirilen hasta atlardan alınan numenelerdeki bakteriyel etkenlerin ve bu örneklerin alındığı atlardaki hastalıkların retrospektif olarak dağılımının araştırılması amaçlandı. TJK İstanbul At Hastanesi Laboratuvarı'na en çok gönderilen numunenin trakeal lavaj sıvısı (%47.3) olduğu ve numunelerinin en çok ilkbahar (%33.8) ve sonbahar (%30.8) mevsimlerinde gönderildiği tespit edildi. Gönderilen numunelerin alındığı atlarda en çok solunum sistemi enfeksiyonu (%49.37), daha sonra genital sistem enfeksiyonları (%29.11) ve gastrointestinal sistem enfeksiyonları (%18.57) görüldüğü belirlendi. Gönderilen bütün numunelerde en çok *Escherichia coli* (%15.6) tespit edildi. Solunum sistemi enfeksiyonlarında, en çok *Streptococcus equi* subsp. *zooepidemicus* (%14.53), daha sonra sırasıyla *Streptococcus dysgalactiae* subsp. *equisimilis* (%11.11) ve *Streptococcus equi* subsp. *equi* (%10.26) identifiye edildiği belirlendi. Sonuç olarak, bu çalışmadan elde edilen bulguların, TJK bünyesinde faaliyet gösteren işletmelerde prevalansı yüksek hastalık veya bakteriyel etkenlere karşı spesifik tedbirlerin alınmasına ve at hastalıklarına bağlı kayıpların minimalize edilmesine katkı sağlaması beklenmektedir.

Anahtar Kelimeler: At, Escherichia coli, Streptococcus equi subsp. zooepidemicus, Solunum sistemi hastalıkları, Trakeal lavaj

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INTRODUCTION

Respiratory diseases are a significant cause of morbidity and performance loss in horses of all ages, breeds, and disciplines (Morris and Seeherman 1991; Burrell et al. 1996). Infectious (Strangles, bacterial pneumonia, viral infections) and non-infectious diseases (asthma, laryngeal hemiplegia, recurrent airway obstruction [RAO], etc.) affecting both the upper and lower respiratory tracts are major respiratory system disorders in horses (Burrell et al. 1996; Ainsworth and Hackett 2004; Ekinci et al. 2024; Deniz et al. 2024). Bacterial pneumonia is a significant cause of morbidity and mortality in adult horses (Sweeney 1991). Secondary bacterial infections of the lower respiratory tract occur when opportunistic bacteria, typically part of the normal flora of the oral cavity or upper respiratory tract, gain access to the lower airways and disrupt natural mechanisms, respiratory defense leading to bronchopneumonia or pleuropneumonia (Burrell et al. 1996; Ainsworth and Hackett 2004). Respiratory disease has been detected in 40% to 42% of racehorses presented for poor performance evaluation (Martin et al. 2000).

In India, a study was conducted on 88 healthy and 53 horses with respiratory diseases to isolate and identify aerobic bacteria from the upper respiratory tract. Of the 321 isolates from both groups, 84.1% were identified as gram positive and 15.9% as gram negative bacteria. The most frequently isolated bacteria were Streptococcus equi subsp. zooepidemicus (17.44%), followed by Micrococcus spp. (9.96%), Corynebacterium spp. (9.65%), Staphylococcus intermedius (9.65%), Staphylococcus aureus (8.72%), Bacillus spp. (7.16%), Streptococcus pneumoniae (5.60%), Staphylococcus chromogenes (5.60%), Streptococcus equisimilis (5.29%), Pseudomonas aeruginosa (5.29%), Rhodococcus equi (3.73%), Escherichia coli (3.73%), Klebsiella pneumoniae (3.42%), Proteus vulgaris (3.42%), and Streptococcus equi subsp. equi (1.24%) (Mir et al. 2013). In a retrospective study analyzing antibiotic susceptibility test results of bacterial pathogens cultured from horses at the Zurich University Equine Clinic between 2012 and 2015, Escherichia coli (20%, 60/303) and Staphylococcus aureus (13%, 40/303) were commonly isolated. The highest rate of multidrug resistance was found in Acinetobacter baumannii isolates (96%, 23/24), followed by Enterobacter cloacae (86%, 24/28) and Escherichia coli (80%, 48/60) (van Spijk et al. 2016). In a retrospective study by Erol et al. (2012) investigating the tissue/organ distributions and β-hemolytic antimicrobial susceptibilities of streptococci in horses, a total of 2,497 β-hemolytic streptococci were isolated from 2,391 cases. Among these, Streptococcus equi subsp. zooepidemicus was the most frequently isolated species (72.0%). Other isolated species included Streptococcus dysgalactiae subsp. equisimilis (21.3%), Streptococcus equi subsp. equi (5.8%), and unidentified β -hemolytic streptococci (0.9%). *Escherichia coli* (*E. coli*) is a bacterium commonly found in the intestines of most

mammals. It can be found almost everywhere in the environment due to its spread through feces, water, and soil, its ability to attach to plants, and its capacity to colonize plants (van Duijkeren et al. 2000). Streptococcus equi subsp. equi, Streptococcus dysgalactiae subsp. equisimilis, and Streptococcus equi subsp. zooepidemicus are the three main β -hemolytic Streptococcus species that cause serious and economically significant diseases in horses. Streptococcus equi subsp. equi is the causative agent of Strangles (also known as Lymphadenitis equorum), a highly contagious infection affecting the upper respiratory tract and specific to horses that is associated with their lymph nodes (Timoney 2004; Holden et al. 2009). Streptococcus equi subsp. zooepidemicus is considered a mucosal commensal in the oral cavity, pharynx, and respiratory tract of horses and acts as an opportunistic pathogen causing respiratory and urinary tract diseases (endometritis) in horses and has been reported as the most common bacterium causing placentitis in mares (Erol et al. 2012; Rasmussen et al. 2015; Díaz-Bertrana et al. 2021). The causative agent of Strangles, Streptococcus equi subsp. equi, is a primary bacterial pathogen of the upper respiratory tract and can cause mucosal invasion without predisposing factors (Libardoni et al. 2016; Jaramillo-Morales et al. 2023).

In Türkiye, there are limited studies on the classification of bacterial agents and diseases identified in breeding and racehorses (Yildirim et al. 2015; Çalışkan and Tel 2021; Diri et al. 2022; Baydar et al. 2023; Deniz et al. 2024). Indeed, the lack of classification of diseases observed in horses, which have very high monetary value, contributes to difficulties in tracking equine diseases in Türkiye, resulting in a scarcity of epidemiological data and inadequate hospital management. Additionally, the inability to document diseases leads to insufficient preventive measures against some significant illnesses and consequently results in economic losses. Moreover, taking preventive measures against prevalent diseases or bacterial agents is crucial for minimizing losses related to these agents or diseases and ensuring the effective and efficient use of resources in horse farms. This study aims to retrospectively investigate the distribution of bacterial agents identified from samples sent to the TJK İstanbul Equine Hospital laboratory between 2015 and 2019 and the diseases of the horses from which these samples were obtained.

MATERIALS and METHODS

Animal Material

The animal material for this study comprised resident and guest horses from the TJK (Jockey Club of 424 Türkiye) facilities, including animal hospitals and breeding stations, between January 1, 2015, and December 31, 2019.

Sample Collection

Fecal, nasal swab, abscess content, clitoral swab, uterine swab and tracheal lavage fluid samples taken from horses showing signs of disease to confirm the suspected diseases were sent to the laboratory (TJK İstanbul Equine Hospital Laboratory, Bakırköy, İstanbul, Türkiye) under appropriate storage conditions.

Bacteriologic analyses

Tracheal aspirate, nasal swab, endometrial swab, endometrial washing fluid and abscess content samples were cultured onto two 5% sheep blood agar (Laborlar, Türkiye) and mac conkey agar (Laborlar, Türkiye) for bacteriological examination. One blood agar was incubated in aerobic atmosphere and the other in microaerophilic atmosphere at 37 °C for 24-72 hours. After 72 hours, cultures with no growth were considered negative. In positive cultures, growths were checked for purity/presence of contaminant colonies. Cultures with 2< number of different types of growth were considered as contamination. Suspicious colonies in positive cultures were identified by conventional biochemical tests (Diagnostics i.n.c., Slovakia). After determining the morphological characteristics of the isolated colonies and the hemolysis characteristics of the colonies showing growth on blood agar, Gram staining (GBL, Türkiye) was performed on suspicious colonies to determine the Gram characteristics and morphology of the agents. Then catalase (GBL, Türkiye) and oxidase tests (Diagnostics i.n.c., Slovakia) were performed. Subsequently, а commercial kit (Diagnostics i.n.c., Slovakia) was used for conventional biochemical tests (Quinn et al. 1998). All bacteriological analyses were performed by a veterinarian specialized in microbiology and the results were recorded.

Data Collection

Data on the types of samples and identified bacterial agents sent to the TJK İstanbul Equine Hospital Laboratory between January 1, 2015, and December 31, 2019, were retrospectively obtained from laboratory records. Subsequently, information about the diseases of the horses from which these samples were collected, including age, breed, sex, and season, was retrieved from hospital and laboratory records.

Statistical Analysis

Statistical analyses were conducted using IBM-SPSS for Windows Release 25.0 (SPSS Inc., Chicago, IL, USA). Descriptive statistics were expressed as frequencies and percentages. The relationship between categorical variables was analyzed using the Chi-Square (χ^2) test (alternatively, Fisher's exact test). Graphs were generated using GraphPad Prism 9.0 (GraphPad Software, Inc., San Diego, CA, USA). A p-value of <0.05 was considered statistically significant.

RESULTS

In this retrospective study, samples sent to the TJK İstanbul Equine Hospital laboratory were identified as coming from the following facilities: İstanbul Equine Hospital, Silivri Breeding Station, İzmit Equine Hospital, Kartepe Equine Hospital, Bursa Equine Hospital, Karacabey Equine Hospital, İzmir Equine Hospital, Torbalı Breeding Station, Mahmudiye Equine Hospital, Ankara Equine Hospital, Elazig Equine Hospital, Divarbakir Equine Hospital, Sanliurfa Equine Hospital, Sanliurfa Breeding Station, Adana Equine Hospital, and Seyhan Breeding Station. It was determined that 83.1% of the samples sent for bacterial culture were from Thoroughbred horses, while 16.9% were from Arabian horses. Among these horses, 56.5% were female and 43.5% were male, with an average age of 6.93±4.78 years.

It was determined that 81.4% of the samples were collected in 2019, 13.9% in 2018, 3.8% in 2016, 0.4% in 2015, and 0.4% in 2017. No statistically significant association was observed between the year category and the categories of identified bacterial agents (χ^2 : 130.846, P= 0.997).

A total of 1,847 samples were sent to the İstanbul Equine Hospital Laboratory for bacteriological examination from horses showing signs of disease suspected of originating from specific systems or regions. Of these 1,847 samples, 237 (12.83%) were identified with one or more bacteria. Among the submitted samples, 47.3% were from tracheal lavage fluid, 28.3% from the uterus, 17.7% from the rectum, 1.69% from the clitoris, 1.27% from the kidneys and urethra, 0.84% from the air sacs, 0.84% from internal organs (liver, spleen), 0.42% from lung tissue, 0.42% from the placenta, and 0.42% from synovial fluid (Figure 1).

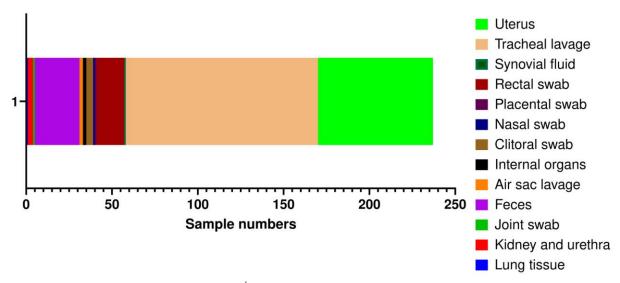


Figure 1. Distribution of samples sent to TJK İstanbul Equine Hospital Laboratory according to the regions where they were taken.

Samples sent to the TJK İstanbul Equine Hospital Laboratory were most frequently collected in March (15.6%), followed by September (12.7%), April (10.5%), June (10.1%), October (9.3%), November (8.9%), May (7.6%), January (6.3%), February (5.9%), August (5.9%), and December (0.4%). When examining the distribution of samples by season, the highest number were collected in spring (33.8%), followed by autumn (30.8%), summer (22.8%), and winter (12.7%) (Figure 2). However, no statistically significant association was observed between the seasonal categories of sample collection and the identified bacterial agents (χ^2 : 159.990, P= 0.171).

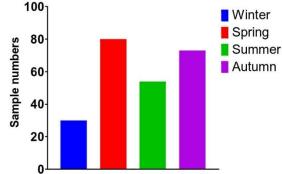


Figure 2. Distribution of samples sent to the TJK Istanbul Horse Hospital laboratory and from which bacterial agents were isolated, according to the seasons in which they were taken.

Among all the samples sent to the TJK İstanbul Equine Hospital Laboratory, *Escherichia coli* was the most frequently isolated bacterium (15.6%). The following bacteria were isolated in decreasing order: *Streptococcus equi* (11.0%), *S. zooepidemicus* (9.7%), *Streptococcus agalactiae* (7.6%), *Streptococcus dysgalactiae* subsp. *equisimilis* (7.6%), *Corynebacterium jeikeium* (5.9%), *Enterobacter cloacae* (4.6%), *Klebsiella pneumoniae* (4.6%), *Salmonella* spp. (2.5%), *Pseudomonas aeruginosa* (2.1%), *Aeromonas hydrophila* (1.7%), *Stenotrophomonas* Among the horses from which samples were sent to the TJK İstanbul Equine Hospital Laboratory, the most frequently diagnosed condition was respiratory system infection (pneumonia) (49.37%, 117/237), followed by genital system infection (metritis, endometritis) (29.96%, 71/237), gastrointestinal system infections (enteritis, enterocolitis, colic) (18.57%, 44/237), urinary system infection (cystitis) (1.27%, 3/237), and arthritis (0.84%, 2/237) (Figure 3).

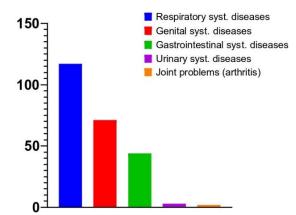


Figure 3. Distribution of diseases in horses from which samples were taken and bacterial agents were isolated, sent to the laboratory of TJK İstanbul Equine Hospital.

maltophilia (1.7%), Streptococcus equisimilis (1.7%), Yersinia pseudotuberculosis (1.7%), Pantoea agglomerans (1.3%), Proteus mirabilis (1.3%), Rhodococcus equi (1.3%), S. aureus (1.3%), and Sphingomonas paucimobilis (1.3%) (Table 1).

In respiratory system infections, the most frequently isolated bacteria were *Streptococcus equi* subsp. *zooepidemicus* (14.53%, 17/117), followed by *Streptococcus dysgalactiae* subsp. *equisimilis* (11.11%, 13/117), *Streptococcus equi* subsp. *equi* (10.26%,

12/117), Escherichia coli (10.26%, 12/117), Klebsiella pneumoniae (9.40%, 11/117), and Streptococcus agalactiae (9.40%, 11/117) (Table 2).

In horses diagnosed with genital system infections (metritis/endometritis/placentitis, urovagina),

the most frequently isolated bacteria were *Streptococcus* equi (18.06%, 13/72), followed by *Escherichia coli* (12.5%, 9/72), *Streptococcus agalactiae* (9.72%, 7/72), *Corynebacterium jeikeium* (8.33%, 6/72), *Streptococcus equi* subsp. zooepidemicus (8.33%, 6/72), and *Streptococcus*

dysgalactiae subsp. equisimilis (6.94%, 5/72), among others.

In horses with gastrointestinal system infections, the most frequently isolated bacteria were *E. coli* (37.21%, 16/43), followed by *Corynebacterium jeikeium* (18.60%, 8/43), *Salmonella* spp. (13.95%, 6/43), and *Enterobacter cloacae* (11.63%, 5/43) (Table 3).

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Bacterial agents	%	n/total
Escherichia coli	15.6	37/237
Streptococcus equi subsp. equi	11.0	26/237
Streptococcus equi subsp. zooepidemicus	9.7	23/237
Streptococcus agalactiae	7.6	18/237
Streptococcus dysgalactiae subsp. equisimilis	7.6	18/237
Corynebacterium jeikeium	5.9	14/237
Enterobacter cloacae	4.6	11/237
Klebsiella pneumoniae	4.6	11/237
Salmonella spp	2.5	6/237
Pseudomonas aeruginosa	2.1	5/237
Aeromonas hydrophila	1.7	4/237
Stenotrophomonas maltophilia	1.7	4/237
Streptococcus equisimilis	1.7	4/237
Yersinia pseudotiiberculosis	1.7	4/237
Pantoea agglomerans	1.3	3/237
Proteus mirabilis	1.3	3/237
Rhodococcus equi	1.3	3/237
Staphylococcus aureus	1.3	3/237
Sphingomonas paucimobilis	1.3	3/237

Others; Serratia rubidaea (0.4%, 1/237), Shigella spp. (0.8%, 2/237), Staphylococcus haemolyticus (0.4%, 1/237), Staphylococcus lugdunensis (0.4%, 1/237), Staphylococcus scheiferi (0.4%, 1/237), Staphylococcus scheiferi (0.4%, 1/237), Staphylococcus vitulinus (0.4%, 1/237), Streptococcus porcinus (0.4%, 1/237), Streptococcus sanguis (0.4%, 1/237), Streptococcus scheiferi (0.4%, 1/237), Streptococcus uberis (0.4%, 1/237), Streptococcus sedentarius (0.8%, 2/237), Gardneralla vaginalis (0.4%, 1/237), Enterococcus facalis (0.4%, 1/237), Enterobacter spp. (0.4%, 1/237), Enterobacter spp. (0.4%, 1/237), Enterobacter spp. (0.4%, 1/237), Enterobacter spp. (0.4%, 1/237), Enterobacter gergoviae (0.4%, 1/237), Enterobacter spp. (0.4%, 1/237), Enterobacter support (0.4%, 1/237), Enterobacter spp. (0.4%, 1/237), Corynebacterium ulcerans (0.4%, 1/237), Corynebacterium spp. (0.4%, 1/237), Corynebacterium spp. (0.4%, 1/237), Catrobacter freundii (0.4%, 1/237), Bacillus circulans (0.4%, 1/237), Aerococcus urinae (0.4%, 1/237), Data were expressed as % (n/total).

Table 2. Distribution of bacterial agents isolated from respiratory system infections

Bacterial agents	0/0	n/total
Streptococcus equi subsp zooepidemicus	14.53	17/117
Streptococcus dysgalactiae subsp. equisimilis	11.11	13/117
Streptococcus equi subsp equi	11.11	13/117
Escherichia coli	10.26	12/117
Klebsiella pneumoniae	9.40	11/117
Streptococcus agalactiae	9.40	11/117
Enterobacter cloacae	4.27	5/117
Yersinia pseudotüberculosis	3.42	4/117
Aeromonas hydrophila	3.42	4/117
Stenotrophomonas maltophilia	2.56	3/117
Rhodococcus equi	2.56	3/117
Pseudomonas aeruginosa	2.56	3/117
Streptococcus equisimilis	1.71	2/117
Corynebacterium pseudotüberculosis	0.85	1/117
Corynebacterium bovis	0.85	1/117
Citrobacter freundii	0.85	1/117
Enterobacter cancerogenus	0.85	1/117
Enterobacter sakazakii	0.85	1/117
Enterococcus faecium	0.85	1/117
Micrococcus sedentarius	0.85	1/117
Pantoea agglomerans	0.85	1/117
Serratia rubidaea	0.85	1/117
Shigella spp.	0.85	1/117
Sphingomonas paucimobilis	0.85	1/117
Staphylococcus lugdunensis	0.85	1/117
Staphylococcus vitulinus	0.85	1/117
Streptococcus porcinus	0.85	1/117
Streptococcus scheiferi	0.85	1/117
Vibrio metschnikovii	0.85	1/117

Table 3 Distribution of bacterial agents isolated from horses diagnosed with gastrointestinal system infection

Bacterial agents	%	n/total
Escherichia coli	37.21	16/43
Corynebacterium jeikeium	18.60	8/43
Salmonella spp.	13.95	6/43
Enterobacter cloacae	11.63	5/43
Enterobacter spp.	4.65	2/43
Pseudomonas aeruginosa	4.65	2/43
Bacillus circulans	2.33	1/43
Enterobacter gergoviae	2.33	1/43
Enterococcus faecium	2.33	1/43
Proteus mirabilis	2.33	1/43

Data were expressed as % (n/total).

DISCUSSION

This study reveals that among the samples sent to the TJK İstanbul Equine Hospital Laboratory for bacteriological analysis, tracheal lavage fluid was the most frequently collected type of sample (47.3%). The majority of samples were submitted in the spring (33.8%) and autumn (30.8%) seasons. The most commonly isolated bacterium from all samples was Escherichia coli (15.6%), followed by Streptococcus equi subsp. equi (11.0%) and Streptococcus equi subsp. zooepidemicus (9.7%). The horses from which the samples were collected were most commonly diagnosed with respiratory system diseases (49.37%), followed by genital system diseases (29.11%) and gastrointestinal system diseases (18.57%). The high incidence of respiratory system infections in the present study may be related to the fact that the majority of the horses from which the samples were taken were racehorses or had a racing history and that there was high horse mobility in some facilities. Respiratory system diseases are common in racehorses as in other young domestic animal species (Burrell et al., 1996). The start of the breeding season at TJK facilities in February and the subsequent transfer of horses from various sources may increase the incidence of respiratory infections. Another possible reason is that the immunity of foals born in spring (March, April, May) may be suppressed due to the stress caused by the separation of their mothers in the fall season. In addition, the increase in the pathogen load in the environment associated with the cooling of the weather and the onset of the rainy season in the fall may explain the increase in the number of respiratory infections.

Escherichia coli is a bacterium commonly found in the intestines of most mammals. It is widely distributed in the environment due to its presence in feces, its spread through water and soil, its ability to adhere to plants, and its potential to colonize plant surfaces (van Duijkeren et al., 2000). E. coli is a prevalent commensal organism in the intestines of horses (Feary et al., 2003). This bacterium is often cultured from the feces of both healthy and diarrheal horses (Maddox et al., 2011; Johns et al., 2012). In the current study, E. coli was the most frequently isolated bacterium from the samples, accounting for 15.6%. The primary concern regarding the presence of E. coli in horse feces relates to human health. This is because antimicrobial-resistant and multidrug-resistant strains of E. coli can be isolated from the feces of hospitalized horses, particularly those treated with oral antibiotics (Maddox et al., 2015).

In the current study, *E. coli* was the most frequently isolated bacterium in horses diagnosed with gastrointestinal system diseases (37.21%, 16/43), followed by *Corynebacterium jeikeium* (18.60%, 8/43) and *Salmonella* spp. (13.95%, 6/43). Enteritis, colitis, and/or enterocolitis are among the most common

causes of disease and mortality in horses (Macías-Rioseco et al., 2020). Diagnosing the etiology of enteritis, colitis, and enterocolitis is challenging, and the cause remains undetermined in approximately 50% of cases (Uzal et al., 2015). Enteropathogens such as Clostridium difficile, Clostridium perfringens, E. coli, and Salmonella spp. have been associated with acute enteritis in horses (Browning et al., 1991; Mallicote et al., 2012). Salmonella spp., Clostridium perfringens type A NetF-positive, C. perfringens type C, Clostridioides difficile, Clostridium piliforme, Paeniclostridium sordellii, Rhodococcus equi, and Neorickettsia risticii are among the primary bacterial causes of enterocolitis in horses (Uzal et al., 2022). Salmonella spp. can be found in the intestines of clinically healthy horses. Stress and antibiotic therapy are considered major factors that predispose to clinical salmonellosis (Alinovi et al., 2003). Horses infected with Salmonella spp. may appear clinically healthy or may show mild to severe clinical signs. Factors contributing to this variability include host-related factors (e.g., stress, immune status, concurrent gastrointestinal diseases) and pathogen-related factors (e.g., serotype, infection dose). Horses with mild disease may exhibit slight fever, soft stools, and a temporary decrease in feed intake. Those with severe disease may show symptoms such as watery diarrhea, fever, toxemia, anorexia, and colic. Serious dehydration, electrolyte imbalances, acid-base disturbances, and protein-losing enteropathy may result from malabsorptive and hypersecretory diarrhea (Shaw & Stämpfli, 2018). Corynebacterium jeikeium has increasingly been identified in various clinical conditions, particularly in immunocompromised individuals, and is recognized as a significant nosocomial pathogen. C. jeikeium is one of the most frequently isolated medically significant corynebacterial species in intensive care unit patients. C. jeikeium is a non-motile, Grampositive rod that appears as a cocobacillus. Other infections attributed to C. jeikeium include skin and wound infections, catheter-associated infections, enteritis, meningitis, osteomyelitis, peritonitis, pneumonia, and pyelonephritis (Denise, 2018).

Respiratory system diseases are common in racehorses, much like in other young companion animal species (Burrell et al., 1996). Respiratory infections can occur in horses of all ages, with affected horses typically exhibiting clinical signs such as exercise intolerance, coughing, nasal discharge, fever, dyspnea, tachypnea, general depression, and loss of appetite. Diagnostic techniques such as bronchoalveolar lavage, transtracheal wash, thoracic ultrasonography, or thoracic radiography are often used to confirm a suspected diagnosis (Ainsworth & Hackett, 2004). Respiratory tract diseases are particularly significant in horses, especially in young Thoroughbreds. Inflammation detected in the trachea and bronchi is referred to as inflammatory airway disease, and it is more impactful and frequent compared to other respiratory symptoms in this population (Wood et al., 2005). Respiratory disorders are second in importance only to musculoskeletal disorders in limiting an equine athlete's performance (Hewson & Arroyo, 2015). In the current study, respiratory system infections were found to be the most prevalent among infections at facilities operating under TJK, accounting for 49.37%. A study conducted in Ethiopia similarly reported that cough and nasal discharge associated with respiratory infections were commonly observed in horses (Laing et al., 2021).

Streptococcus equi subsp. zooepidemicus is a part of the normal bacterial flora in horses and is responsible for a range of diseases including pneumonia, abortion, upper respiratory tract infections, wound infections, testicular infections, and neonatal infections (Newton et al. 2003; Lindahl et al. 2013). In the present study, it was found that the most commonly used sample type was tracheal lavage fluid (47.3%). Furthermore, Streptococcus equi subsp. zooepidemicus was the most frequently isolated pathogen in respiratory infections, accounting for 14.53% (17/117). Similarly, a Erol et retrospective study by al. (2012)comprehensively examined the tissue/organ distribution and antimicrobial susceptibility patterns of β -hemolytic streptococci in horses between January 1, 2000, and December 31, 2010. In this study, a total of 2,497 β -hemolytic streptococci were isolated from 2,391 cases, with S. equi subsp. zooepidemicus being the most frequently isolated type (72.0%). S. zooepidemicus is associated with S. equi subsp. equi, which is the causative agent of Strangles (germ, watering can disease). In the current study, S. equi subsp. equi (11.0%) and S. equi subsp. zooepidemicus (9.7%) were the most frequently isolated pathogens after E. coli (15.6%) among the samples sent to the TJK İstanbul Equine Hospital Laboratory. Jaramillo-Morales et al. (2022) reported that young horses with a recent history of transport have a higher likelihood of testing positive for S. equi in guttural pouch swabs, with prevalences of 13.5% for S. equi subsp. equi and 1.5% for S. equi subsp. zooepidemicus. In the USA, S. equi was found in 715 out of 9,409 horses with upper respiratory infections (7.6%) (Jaramillo-Morales et al. 2023). Another study reported a prevalence of S. equi subsp. equi in herds at 5.86% (Libardoni et al. 2016). Çalışkan and Tel (2021) examined a total of 60 samples, including 32 nasal swabs and 28 tracheal aspirates, for S. equi and S. zooepidemicus. They reported isolating Streptococcus spp. from 22 out of 60 samples (36%), of which 3 strains (19.1%) were S. equi and 19 strains (20.6%) were S. zooepidemicus. In another study conducted in Türkiye, Streptococcus spp. was isolated from 2 out of 133 samples (1.2%), with one being S. equi subsp. zooepidemicus and the other S. pneumoniae (Diri et al. 2022). Additionally, S. equi subsp. zooepidemicus was isolated from 6 out of 93 horses (6.5%) in another study (Acke et al. 2015).

In the present study, after Streptococcus equi subsp. zooepidemicus (14.53%), the most commonly isolated pathogens in respiratory system diseases were Streptococcus dysgalactiae subsp. equisimilis (11.11%, 13/117), Streptococcus equi subsp. equi (11.11%, 13/117), Escherichia coli (10.26%, 12/117), Streptococcus agalactiae (9.40%, 11/117), and Klebsiella pneumoniae (9.40%, 11/117). S. equisimilis has rarely been isolated from the placentas of aborted, stillborn, and premature foals (Hong et al. 1993). Streptococcus dysgalactiae subsp. equisimilis is commonly found as a commensal in skin and mucosal surfaces and is an opportunistic pathogen for various animal species, including humans, horses, dogs, and pigs (Timoney 2004; İncili et al. 2023). It has recently been isolated from horses with a history of respiratory disease or Strangles-like illness (Laus et al. 2007). S. equisimilis is the second most frequently identified bacterial agent in pneumonia cases in horses, following S. equi subsp. zooepidemicus (Erol et al. 2012). In one study, Streptococcus dysgalactiae subsp. equisimilis was detected in 29 out of 99 nasal swabs (29.3%) (Preziuso et al. 2010). Another study isolated S. dysgalactiae subsp. equisimilis from 22 out of 93 horses (23.7%) (Acke et al. 2015).

In this study, it was determined that the samples sent to the TJK İstanbul Equine Hospital Laboratory were most frequently collected in the spring (33.8%) and then in the fall (30.8%). These samples were primarily from horses with respiratory infections. Analyzing the monthly distribution of the samples, it was found that most samples were sent in March. This can be explained by the start of the breeding season at the TJK facilities in February and the subsequent transfer of horses from various sources, which could lead to the transmission of diseases to healthy horses. The prevalence of recurrent airway obstruction (RAO) in horses, which is associated with exposure to hay and straw, may increase due to horses being kept in stables during the winter months (Bracher et al. 1991). Inflammatory airway disease (IAD) tends to peak at the beginning of spring and then decrease throughout the year, with significant increases often observed during seasonal transitions (Wood et al. 2005). Kutasi et al. (2011) reported that the prevalence of recurrent airway obstruction (RAO) is higher in the spring and summer compared to the winter months. It was noted that many horses began to show symptoms during this period, or the clinical signs became more severe. Another study observed a trend of three main peaks for RAO and IAD throughout the year. These diseases were reported to peak in early spring, with a smaller peak in midsummer and another peak at the end of summer (Kutasi et al. 2011; Couëtil and Ward 2003). The study indicated that the likelihood of RAO in horses is 1.6 times higher in winter and 1.5 times higher in spring compared to summer.

Bacterial species involved in infectious equine endometritis are generally residents of the mare's

normal microbiota. The most common bacterium triggering endometritis in mares is Streptococcus equi subsp. zooepidemicus (Purswell et al. 1989; Langoni et al. 1997; Benko et al. 2016). However, other seen microorganisms commonly in equine Escherichia endometritis include coli, Klebsiella pneumoniae, Pseudomonas spp., Staphylococcus aureus, Corynebacterium spp., Bacillus spp., and Actinomyces (Riddle et al. 2007; Leblanc et al. 2009; Davis et al. 2013). In this study, the most frequently isolated bacteria from mares diagnosed with genital system diseases (metritis, endometritis) were Streptococcus equi subsp. equi (18.06%, 13/72), followed by E. coli (12.5%, 9/72) and Streptococcus agalactiae (9.72%, 7/72). Postpartum metritis is typically associated with trauma during parturition the retention of fetal membranes within 10 days after birth (more commonly 2-4 days after birth) and contamination of the urogenital system (Morris et al. 2020). An increase of one unit in the endometrial edema score in early postpartum mares has been reported to increase the likelihood of diagnosing subclinical endometritis caused by Streptococcus equi subsp. zooepidemicus by 5.5 times (Rasmussen et al. 2015). Díaz-Bertrana et al. (2021) isolated Staphylococcus (25.1%), Streptococcus (18.2%), Escherichia (17.3%), and Pseudomonas (12.1%) from samples collected from 363 mares with a history of repeated infertility, positive endometrial cytology, and/or vaginal discharge.

Anaerobic cultures were only performed on samples obtained from abscess content. However, the fact that anaerobic and *Mycoplasma* spp. cultures were not conducted on the other samples is one of the limitations of this study.

CONCLUSION

In conclusion, the findings from this study can contribute to the implementation of specific measures against prevalent diseases or bacterial agents in animal hospitals, stud farms, boarding facilities, and breeding stations operated under the TJK. This, in turn, may help minimize losses related to equine diseases.

Conflict of interest: The authors have no conflicts of interest to report.

Authors' Contributions: MCT and GE contributed to the project idea, design and execution of the study. MCT contributed to the acquisition of data. MCT and GE analysed the data. MCT and GE drafted and wrote the manuscript. MCT and GE reviewed the manuscript critically. All authors have read and approved the finalized manuscript.

Ethical approval:This study was carried out in the Turkish Jockey Club, İstanbul Equine Hospital Laboratory. "This study is not subject to the permission of HADYEK in accordance with the "Regulation on Working Procedures and Principles of

Animal Experiments Ethics Committees" 8 (k1). The data, information and documents presented in this article were obtained within the framework of academic and ethical rules."

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