

*Original Scientific Article***A SURVEY REPORT ON MANAGEMENT PRACTICES ON DAIRY DONKEY FARMS IN SERBIA**

Snežana Bulajić¹, Milica Kovačević-Filipović², Jasna Lončina¹, Miroslav Kjosevski³,
Ena Dobrić³, Katarina Nenadović⁴

¹*Department of Food Hygiene and Technology, Faculty of Veterinary Medicine, University of Belgrade, Bulevar Oslobođenja 18, 11000, Belgrade, Serbia*

²*Department of Pathophysiology, Faculty of Veterinary Medicine, University of Belgrade, Bulevar Oslobođenja 18, 11000, Belgrade, Serbia*

³*Department of Animal Hygiene and Environmental Protection, Faculty of Veterinary Medicine-Skopje, Ss. Cyril and Methodius University in Skopje, Lazar Pop-Trajkov 5-7, 1000 Skopje, N. Macedonia*

⁴*Department of Animal Hygiene, Faculty of Veterinary Medicine, University of Belgrade, Bulevar Oslobođenja 18, 11000, Belgrade, Serbia*

Received 29 August 2025; Received in revised form 28 October 2025; Accepted 6 November 2025

ABSTRACT

In Serbia, a growing demand for donkey milk is driving an increase in dairy donkey farms. This paper describes the demography and management practices at dairy donkey farms in Serbia using semi-structured face-to-face interviews with their owners. Additionally, microbiology of raw donkey milk was conducted to assess its hygienic quality and safety. The results show that most owners were male, aged over 51, who had not participated in dairy farming training courses (8/9; 88.89%). Most of the farms were located in North Serbia; they were family-run, with more than 20 animals in the herd. At the farms, straw was predominantly used as bedding material. Inadequate bedding hygiene and shelter dustiness were reported in 22.22% and 55.56% of the farms, respectively. Only two owners reported undertaking disinfection procedures for shelters or stables. All donkeys were fed hay supplemented with corn or oats, while wheat bran was provided at two farms. Healthcare measures such as vaccination and dental care were not carried out at the investigated farms, while deworming had never been performed on four of them (44.44%). The most common reported health problems were sudden death, abortion, stillbirth, and laminitis. Fore-milking was not performed on any of the farms. The post-milking procedure involved filtering through cotton gauze, packing in plastic bottles, and then refrigerating and/or freezing the milk, to be later sold at the farm. Presence of food-borne pathogens in raw donkey milk was not detected. Our results could help identify the key problems in country-specific donkey dairy management.

Key words: dairy donkey farms, management, milk microbiology, Serbia, animal welfare

INTRODUCTION

Donkey husbandry in Serbia is deeply embedded in the country's cultural, economic, and social fabric. Historically, donkeys served as working

animals in rural communities and nowadays they hold a symbolic value in the Serbian folklore where they are portrayed as humble and loyal figures. In addition, raw donkey milk has traditionally been used as a natural remedy, particularly for respiratory conditions such as asthma and bronchitis (1).

Agricultural mechanization significantly reduced the practical value of donkeys, leading to a decline in their population after the Second World War. Today, only a limited number remain, primarily in specialized herds for milk production. According to the data from the most recent Agricultural Census in 2023, a total of 2.380 donkeys, hinnies, and mules were registered in

Corresponding author: Assoc. Prof. Katarina Nenadovic, PhD

E-mail address: katarinar@vet.bg.ac.rs

Present address: Department of Animal Hygiene, Faculty of Veterinary Medicine, University of Belgrade, Bulevar Oslobođenja 18, 11000 Belgrade, Serbia

Phone: +381642314232

Copyright: © 2026 Bulajić S. This is an open-access article published under the terms of the Creative Commons Attribution License which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.

Competing Interests: The authors have declared that no competing interests exist.

Available Online First: 27 February 2026

Published on: 15 March 2026

<https://doi.org/10.2478/macvetrev-2026-0014>

Serbia (2). Conservation programs have established breeding centers in regions such as Sremska Mitrovica, Stara Planina, and Kovilj to preserve genetic diversity and reestablish sustainable management systems (3).

The Balkan Donkey is the predominant breed in Serbia, managed mostly at smallholder farms under semi-extensive conditions. A distinct genetic and morphological population, the Banat Donkey, has also been identified in Serbia, and it may merit classification as a separate breed (4). Its limited numbers emphasize the urgency for targeted conservation measures.

In recent years, the growing interest in donkey milk, particularly due to its nutritional qualities and potential in managing cow-milk protein allergy (5), has led to a gradual resurgence of donkey farming in Serbia (6). Despite this trend, farm management systems differ widely, and concerns have been raised about animal welfare (7) and public health risks associated with the sale of raw milk (8).

The research on donkeys in Serbia has so far been primarily focused on morphological, biochemical, hematological, and genetic characterization (4, 9), as well as on-farm welfare conditions (7). A few studies have addressed the microbiological status and antimicrobial properties of donkey milk (1, 10). However, detailed information on farm-level management practices remains scarce.

In this study, we hypothesized that differences in farm management practices among dairy donkey farms and the demographics of the owners, are associated with variation in milk hygiene, animal welfare, and overall dairy donkey performances. This working hypothesis guided the development of the survey and the analytical framework of our research. Therefore, the aim of this study was to examine the current dairy donkey management practices in Serbia. Considering the fact that in Serbia there are no specific regulatory criteria for donkey milk, we also conducted microbiological analysis to assess hygienic quality and safety of the milk.

MATERIAL AND METHODS

Ethical approval

The participation in this study was voluntary with confidentiality of information. The authors followed the Law on Personal Data Protection of the Republic of Serbia (RS) ensuring that data collected were processed in accordance with legal and ethical guidelines.

The study received approval from the Ethical Committee of the Ethical Committee of the Faculty of Veterinary Medicine, University of Belgrade, RS. In accordance with the Serbian Animal Welfare Law, authorization was obtained from the Ministry of Agriculture, Forestry, and Water Management, RS (Permit No. 323-07-07930/2022-05).

Data source: Farms

Livestock statistics in Serbia do not list donkeys as a separate category; instead, they are grouped with horses, mules, and hinnies in the Register of Agricultural Holdings. As subsidies for donkeys apply only to genetic resource preservation and not milk production, the Register lacks specific data on farms specialized in donkey dairying.

Contact information for dairy donkey farmers was obtained through local authorities, colleagues from the Farm and Field Veterinary Service, and comprehensive internet searches using keywords “dairy donkey farms,” “donkey milk,” “donkey breeding,” and “Serbia”. Therefore, farms were selected using a convenience sampling approach. We visited nine dairy donkey farms, out of which eight were family holdings, and one was company-owned in the period between March and September 2024. Farmers were contacted by phone, informed about the project, and invited to participate by completing a questionnaire, with verbal consent obtained. All the farms were subsequently visited in person by the research team.

Data collection

Semi-structured interviews were used to collect information from the farmers. Responses were recorded in a questionnaire format by the interviewer. The questionnaire comprised 54 questions and contained sections on the demographics of the owners; farm characteristics and general on-farm management practices; dietary management; health management; on-farm milking management, milk processing and storage; milk distribution and marketing. During the farm visit, the authors evaluated bedding hygiene and shelter/stable dustiness. The hygiene of the bedding material was evaluated according to the AWIN welfare assessment (11). Ventilation in the stables was taken into account in the subjective evaluation of barn air quality.

Milk sampling

Milk samples were collected from clinically healthy Domestic Balkan jennies during morning

Table 1. Growth media, incubation temperature and time, and methods used for the enumeration of microorganisms

Species/Group of microorganisms	Growth media	Incubation conditions	Reference method
TBC	PCA (Merck, Darmstadt, Germany)	30.0 °C, 72 h	ISO 4833-1:2013
PBC	PCA (Merck, Darmstadt, Germany)	6.5 °C, 10 days	ISO 17410:2019
CPS	BP agar (Oxoid)	37.0 °C, 24 h	ISO 6888-1:2021
Enterobacteriaceae	VRBG agar (Oxoid, UK)	37.0 °C, 24 h	ISO 21528-2:2017
LAB	MRS agar (Thermo Fisher Scientific, UK)	30.0 °C, 72 h	ISO 15214:1998
Salmonella spp.	XLD agar (Oxoid, UK)	37.0 °C, 24 h	ISO 6579-1:2017
Listeria monocytogenes	Brilliance™ Listeria Agar (Thermo Fisher Scientific, UK)	37.0 °C, 24(48) h	ISO 11290-1:2017
Cronobacter spp.	CCI agar (Merck, Germany)	41.5 °C, 24 h	ISO 22964:2017
Bacillus cereus	MYP agar (Merck, Germany)	30.0 °C, 24(48) h	ISO 7932:2004
Escherichia coli O157:H7	CT-SMAC agar (Thermo Fisher Scientific, UK)	37.0 °C, 24 h	ISO 16654:2001

TBC-total bacterial count; PCA-plate count agar; PBC-psychrotrophic bacterial count; CPS-coagulase positive Staphylococci; BP-Baird Parker; VRBG-violet red bile glucose; LAB-lactic acid bacteria; MRS (De Man, Rogosa and Sharpe); XLD-xylose-lysine-deoxycholate; CCI-chromogenic cronobacter isolation; MYP-mannitol egg yolk polymyxin; CT-SMAC-cefixime tellurite sorbitol MacConkey

manual milking. The animals were 3.5–9 years old, with the mass of 150–200 kg, and were 30–200 days postpartum. Foals were separated 6–12 h before milking, usually with visual contact maintained.

Udder preparation varied: at most farms, the udder was washed with lukewarm water and dried with a shared cloth or paper towel, while on three farms, no preparation was performed; on one farm teats were wiped with a napkin. Fore-milking was not practiced, but the first streams were discarded before hand-milking into clean plastic jugs. Milk was filtered through gauze, transferred into sterile bottles, refrigerated at 4 °C, and transported to the laboratory. Sampling was not possible on one farm with no lactating jennies. In total, 36 samples were collected from eight farms.

Microbiological analyses

The following microbiological analyses were performed: total bacterial count (TBC), psychrotrophic bacterial count (PBC), coagulase-positive staphylococci (CPS), Enterobacteriaceae, lactic acid bacteria (LAB), *Salmonella* spp., *Listeria monocytogenes*, *Cronobacter* spp., *Bacillus cereus*, and *Escherichia coli* O157:H7. The growth media, incubation time/temperature, and specific method used for each group/species of targeted microorganisms are shown in Table 1. All the analyses were done in triplicate.

Statistical analysis

The data were exported and analyzed using descriptive statistics of the statistical package GraphPad Prism Software, Version 9.00 (GraphPad, Inc., Boston, MA, USA). The bacterial counts, determined by the colony count technique and expressed as CFU/mL, were log₁₀ transformed to normalize the distributions before calculating the arithmetic means and standard deviation of these transformed counts. The qualitative data were narrated and explained logically based on the existing condition and literature.

RESULTS

A total of nine donkey owners completed the survey. Table 2 shows that most farm owners were male, over 51 years old, and held a secondary school degree. More than half had over a decade of farming experience, though only a minority reported farming as their primary occupation. Many relied on off-farm income sources such as pensions, ecotourism, or salaries, with donkey milk serving as a complementary income. Few were members of farmer organizations, and most had not attended training courses. The main motivation for keeping donkeys was strong personal affection and the wish to promote their value and improve their public image.

Table 2. Demographic data of donkey owners in the survey

Questions		Number of respondents (total = 9)	
		n	%
Gender	Female	1	11.11
	Male	8	88.89
Age ranges (years)	21–30	1	11.11
	31–40	0	0
	41–50	1	11.11
	51–60	2	22.22
	Over 60	5	55.56
Education	Primary school	2	22.22
	Secondary school	4	44.44
	Tertiary school	3	33.33
Years involved with donkeys	≤5	1	11.11
	6-10	3	33.33
	11-20	3	33.33
	>20	2	22.22
Main occupation	Livestock farmer	3	33.33
	Tourism-related business	1	11.11
	Formal employment	2	22.22
	Pensioner	2	22.22
	Casual labor/Business or service	1	11.11
Primary source of income	Livestock production	4	44.44
	Off-farm wage employment/salary, pension, small business, eco-tourism	5	55.56
Membership in farmer organization/or other organization (non-governmental organization)	Yes	3	33.33
	No	6	66.67
Participation in educations/ attendance to training courses	Yes	1	11.11
	No	8	88.89
Motivation for practicing donkey dairying	Love for donkeys	5	55.56
	Health benefits (Self-convinced of the health-promoting attributes of donkey milk)	1	11.11
	Eco-tourism	1	11.11
	Desire to live in nature/reconnect with the Earth/ embrace a more sustainable and harmonious relationship with the world around us	1	11.11
	Conservation of animal genetic resources	1	11.11

At the investigated farms, 572 donkeys were housed. Over half of the farms were located in northern Serbia, and most were family-run, with medium to large herds ranging from 15 to 250 donkeys, predominantly females. The Domestic Balkan donkey was the principal breed, with a few Banat donkeys. The farms practiced semi-extensive management, often providing seasonal pasture access and housing donkeys in three-sided sheds under natural light. Straw bedding was common, though

hygiene practices were inconsistent; dustiness was frequently observed, and disinfection was rarely carried out. Feeding was not tailored to age, sex, or production stage. Stallions were usually kept with jennies, and harem breeding was the prevailing system, with first foaling after two years and weaning typically after six months. The primary purpose of rearing was milk production and processing, while some farms also sold male foals, offered recreational services, or engaged in meat production (Table 3).

Table 3. The farm characteristics and general on-farm management data

Questions	Number of farms (total = 9)		
	n	%	
Farm location	North Serbia	5	55.56
	Central Serbia	3	33.33
	Southeast Serbia	1	11.11
Farm running type	Family run	6	66.67
	Have employees	3	33.33
Farming system	Extensive	0	0
	Semi-extensive	9	100.00
Herd size*	1-19	2	22.22
	≥20	7	77.78
Type of housing facility	3-sided run-in sheds	6	66.67
	Run-in sheds	1	11.11
	Simple brick stable	2	22.22
Shelter/Stable size (m²)	250-300	3	33.33
	300-500	2	22.22
	Not reported	4	44.44
Shelter/Stable lighting**	Natural light	9	100.00
	Artificial light	2	22.22
Type of bedding**	Straw	7	77.78
	Hay	1	11.11
	Sand	1	11.11
	No bedding	2	22.22
Hygienic quality of bedding	Inadequate	2	22.22
	Adequate	5	55.56
	Not evaluated	2	22.22
Shelter/stable dustiness	Yes	5	55.56
	No	4	44.44
Shelter/Stable disinfection	Yes	2	22.22
	No	7	77.78
Type of turnout area**	Paddock	4	44.44
	Yard	2	22.22
	Pasture	4	44.44
Grouping strategies according to the specific feeding requirements	Yes	0	0
	No	9	100.00
Housing of stallions	Grouped with females	5	55.56
	Tied in the stable with females	3	33.33
	Housed singly	1	11.11
Breeding type	Harem	5	55.56
	In hand mating	4	44.44
Age at first birth	<2 years	0	0
	≥2 years	9	100.00
Weaning age (months)	<6	2	22.22
	≥6	5	55.56
	Not reported	2	22.22
Purpose of animals on the farm**	Donkey dairying	9	100.00
	Eco-tourism	3	33.33
	Donkey meat products	1	11.11
	Selling of male foals	4	44.44

*Reported during the visit; **More than one answer was allowed

Table 4. Feeding regime

Questions	Number of farms (total = 9)		
	n	%	
Water source	Municipal water supply	5	55.56
	Ground water (well)	1	11.11
	Surface water (ponds, lakes, streams)	3	33.33
Waterer type	Automatic drinking bowls	2	22.22
	Bucket	6	66.67
	Others (old, discarded bath tubs)	1	11.11
Frequency of feeding (daily)	Once	1	11.11
	Twice	7	77.78
	Three times	1	11.11
Feed type*	Pasture	4	44.44
	Hay	9	100.00
	Grains (corn, oat)	9	100.00
	Wheat bran	2	22.22
Vitamin and/or mineral supplements	Yes	4	44.44
	No	5	55.56

*More than one answer was allowed

Table 5. Health management

Questions	Number of farms (total = 9)		
	n	%	
Vaccination	Yes	0	0
	No	9	100.00
Deworming	Never	4	44.44
	Once per year	3	33.33
	Twice per year	2	22.22
Clinical examinations, diagnostic tests, blood sampling	Yes	0	0
	No	8	88.89
	Periodically	1	11.11
Dental care	No	9	100.00
	Yes	0	0
Hoof trimming	Never	0	0
	Once per year	3	33.33
	Twice per year	4	44.44
	If necessary	2	22.22
Health problems*	Abortion	2	22.22
	Sinusitis	1	11.11
	Limb deformities	1	11.11
	Laminitis	2	22.22
	Sudden death	3	33.33
	Stillbirth	2	22.22
	Not reported	3	33.33

*More than one answer was allowed

Donkeys had free access to water at all farms (Table 4). Most drank from buckets, while a few farms used automatic drinkers. Water sources included wells, springs, or, most commonly, the public supply system. The diet consisted mainly of hay, predominantly clover, along with grains such as maize, oats, wheat, or wheat bran. Forage was typically provided twice daily, and during the dry season, some farms allowed free grazing on natural pastures. Mineral or vitamin supplements, usually in the form of mineral blocks, were commonly provided in less than half farms.

Routine healthcare practices and common health problems are summarized in Table 5. None of the donkeys had been vaccinated or received dental care. At nearly half of the farms, animals had never been dewormed, while others carried out deworming once or twice a year, occasionally

limited to foals or non-lactating jennies. Hoof trimming was usually performed once or twice a year, though at some farms, it was done only when needed; all owners reported hood trimming on their own. Clinical examinations and diagnostic testing were rarely performed. The reported main health problems included sudden death, abortion, laminitis, and stillbirths.

On-farm milking practices are summarized in Table 6. None of the farms had a designated milking area, and all relied on manual milking, most commonly once per day. Fore-milking was not practiced. Pre-milking hygiene varied: at some farms, no udder preparation was carried out, while others washed the udder with lukewarm water and dried it with a communal cloth, or less commonly, used a dry wipe. Daily milk yield per jenny typically exceeded 500 mL. Jennies and foals were

Table 6. Milking procedure, milk processing and storage

Questions	Number of farms (total = 9)		
	n	%	
Daily milking number	1	5	55.56
	2	2	22.22
	>2	1	11.11
	Not reported	1	11.11
Foal separation before milking (hours)	<6	1	11.11
	6-12	5	55.56
	>12	0	0
	Not reported	3	33.33
Foals housed near their mothers with the possibility of visual and/or tactile contact	Visual	8	88.89
	Not reported	1	11.11
Pre-milking procedures	Wash the udder with lukewarm water and dry with a towel	4	44.44
	Dry wipe with a napkin if necessary	1	11.11
	No	3	33.33
	Not reported	1	11.11
Quantity of milk per jenny /day (mL)	<500	4	44.44
	≥500	5	55.56
Milk filtration	Strainer with cotton gauze	7	77.78
	No	2	22.22
Milk packaging	Plastic bottle	9	100.00
	Glass bottle	0	0
Milk storage type*	Refrigerated	4	44.44
	Frozen	6	66.67
Storage time*	3-5 days if refrigerated	4	44.44
	3-6 months if frozen	6	66.67

*More than one answer was allowed

Table 7. Milk distribution and marketing data

Questions	Number of farms (total = 9)		
	n	%	
Milk distribution	Direct sale at the farm gate	9	100.00
Main motivational factors for milk purchasing	Health problems (bronchitis, asthma, cough, chemotherapy, boost immunity, treat eczema and psoriasis)	9	100.00
Marketing channels*	Word of mouth	8	88.89
	Social network	5	55.56
	Local TV	5	55.56
	Local radio	3	33.33
	Website	1	11.11
Recommended daily intake	100 mL	4	44.44
	200 mL	2	22.22
	250 mL	1	11.11
	Not reported	2	22.22
Price for 1L (euros)	<30	6	66.67
	31-50	3	33.33
Labelling	Yes	6	66.67
	No	3	33.33
Milk use*	Human consumption	9	100.00
	Cosmetics	6	66.67

*More than one answer was allowed

Table 8. Microbiological profile of the donkey milk samples (\log_{10} cfu/mL; N=36)

Parameter	Mean±SD	Min–Max
TBC	2.16±1.10	0.54–4.77
PBC	1.97±1.58	0.54–4.61
Enterobacteriaceae	1.69±1.07	0.60–3.74
CPS	ND	/
LAB	2.30±1.08	0.74–4.27

TBC-total bacterial count; PBC-psychrotrophic bacterial count; CPS-coagulase positive staphylococci; LAB-lactic acid bacteria; SD-standard deviation; ND-not detected

usually separated for 6-12 h before milking, with visual contact maintained at nearly all farms. In post-milking, milk was generally filtered through gauze, though a few farms did not filter. The storage practices also differed: some refrigerated milk for sale within 3–5 days, while others froze it at -20 °C or used rapid freezing at -40 °C, extending shelf life to 3–6 months.

Milk distribution and marketing practices are summarized in Table 7. The main sales channel was direct farm-gate sale, most often to families with young children and infants. Consumers primarily sought donkey milk for its perceived health benefits,

particularly for respiratory, digestive, and skin conditions. Marketing relied almost exclusively on word of mouth. The typical price was around €30 per liter. All farms produced milk for human consumption, and many also collaborated with galenic pharmacies to produce skincare products such as soaps and creams. Product labeling was common, and the recommended daily intake reported by farmers ranged from 100 to 250 mL.

The safety status of raw donkey milk was assessed by monitoring for presence of *Salmonella* spp., *Listeria monocytogenes*, *Cronobacter* spp., *Bacillus cereus* and *Escherichia coli* O157:H7.

None of the targeted pathogenic microorganisms were found in any of the analyzed milk samples. The microbiological results of the 36 donkey milk samples for TBC, PBC, Enterobacteriaceae, CPS, and LAB are presented in Table 8. Total bacterial count was in range 0.54 and 4.77 log cfu/mL. The mean value of PBC and Enterobacteriaceae was 1.97 and 1.69 log cfu/mL, respectively. Coagulase-positive staphylococci were not detected. The average count of lactic acid bacteria was 2.30 log cfu/mL.

DISCUSSION

This survey showed that donkey farm owners in Serbia were motivated by personal enthusiasm rather than formal training in dairy farming. Housing, feeding, healthcare, and milking practices were largely based on experience and intuition rather than standardized guidelines. Nevertheless, microbiological testing revealed no pathogens in raw milk, indicating that these management systems can still ensure satisfactory milk safety.

Most donkey farm owners were male, well-educated, over 51 years old, and had bred donkeys for more than 11 years. Similar gender and age patterns were reported in China (12). These findings are consistent with national statistics, as the latest Agricultural Census (13) showed that 77% of agricultural managers were male and 69.23% were over 56 years old. In Serbia, livestock management has traditionally been a male role. The predominance of older farm holders reflects rural depopulation, aging, and migration, with long-term effects on productivity and innovation. Serbia shares these challenges with the EU, where 57.6% of farm managers are 55 or older (14). The relatively high education level of donkey owners in this study is encouraging, since they are more likely to adopt new technologies and innovative practices, while being generally more risk-averse.

For many smallholders, joining farmers' organizations mainly offers visibility to attract external support. One farm owner was a member of the Cattle Breeders Association, focused on conserving endangered breeds with traditional methods, while two others belonged to a non-governmental organization that has driven major conservation initiatives in Serbia. Although local breeders recognize the potential of dairy donkeys, government support remains limited, leaving husbandry dependent on individual enthusiasts.

At the investigated farms, donkeys were kept semi-extensively, with four farms providing

seasonal pasture (April–November), which enhances welfare by allowing natural behaviors (12). All farms offered shelters with free access during bad weather; it is essential because donkey coat is less waterproof than horses' one (15). Straw was the most common bedding, consistent with previous studies (16, 17), and considered the safest for ingestion though it carries risks of dust and spores (18). Poor bedding hygiene and dusty shelters were noted in 22.22% and 50% of the farms, respectively, posing risks of respiratory and hoof problems (19, 20). Only two farmers reported disinfecting shelters, despite their importance in controlling infectious diseases (21).

Donkey diet should be based on high-fiber, low-energy forages (22). Our study shows that donkeys in Serbia are fed hay with grain supplements (corn, oats), while two farms also used wheat bran. Hay is the primary forage for donkeys in Europe (16, 17), whereas straw predominates in Northeastern China (12).

Most farmers lack education on donkey healthcare, including vaccination, dental care, deworming, and hoof management, highlighting the need for regulation in line with the EU Platform on Animal Welfare (23) and Animal Health Law (24). Donkeys in Serbia are exposed to typical equine pathogens (25), and owners reported sudden death, abortion, laminitis, and stillbirth as major health issues. Hoof trimming was performed only once or twice a year, much less frequently than the recommended interval of 6–8 weeks, likely contributing to laminitis, a globally prevalent and painful welfare concern (20).

Although donkey dairy farming is growing in the EU, no specific legislation exists. Guidelines issued in 2017 (6) recommend hygienic milking procedures, veterinary oversight, and pre-milking steps, udder inspection, fore-milking, washing, and drying, to ensure milk hygiene and detect abnormalities (26). Our survey shows that foals were separated 6–12 hours before milking, allowing visual contact; however, fore-milking was not performed, limiting early detection of mastitis. Communal teat drying was used by nearly half of the farmers, a practice discouraged due to disease risk (27). Daily milk yield per jenny exceeded 500 mL on more than half of the farms, and is influenced by breed, management, feeding, lactation stage, and milking procedures (28).

Milk was filtered, bottled, and sold raw at the farm gate, reflecting common European practice (29), but raising food safety concerns due to inconsistent hygiene and labeling. National regulations require

labels stating: 'Raw milk. Use after cooking' (30), yet compliance was inconsistent, and some farms made unverified health claims such as 'donkey milk strengthens the immune system'. Donkey milk health benefits have not been rigorously verified (31). Consumer perceptions of food risks are often shaped more by emotion than by science (32). Therefore, governments must enforce food safety regulations and provide accurate, reliable information rather than relying solely on individual perceptions.

Good dairy practices, hygiene, and milk handling strongly influence the microbiological quality of donkey milk. While this study found no foodborne pathogens, the target consumers, infants, young children, and immunocompromised adults, warrant stricter official control (8). There is no evidence that raw milk is nutritionally superior to pasteurized milk (33), and currently, no safety standards exist for donkey milk.

The total bacterial count is currently used both as hygienic marker and indicator of product shelf-life. The observed low TBC count in the present study was in accordance with previous studies (34, 35) and in compliance with the European legislation (36). The low bacterial count in donkey milk with low mastitis prevalence (34) is due to several factors: semi-extensive farming reducing pathogen transmission, equid udder's anatomy limits contamination (37), frequent milking with teat washing (38), and antimicrobial components like lysozyme, abundant in donkey milk, contribute to its favorable microbiological quality (28).

Gram-negative psychrotrophic microorganisms represent the dominant microbiota of raw donkey's milk, with *Pseudomonas* spp., being the most abundant (39). Psychrotrophic members of genus *Pseudomonas* are considered to be among the species responsible for donkey milk spoilage and thus for limiting its shelf-life (40). The average count of psychrotrophic bacteria in the present study was found to be 1.97 log cfu/mL. Our results are in the agreement with previous studies (35, 41).

Enterobacteriaceae, as common environmental organisms, are good indicators when monitoring environmental conditions and overall farm hygiene. The Enterobacteriaceae count in the present study (1.69 log cfu/mL) was in line with other literature reports (35, 42).

In the present study, coagulase-positive staphylococci count was below the detection limit (1 log cfu/mL). A particular safety concern has been identified with the isolation of methicillin-

resistant staphylococci from the donkey milk in Italy (43), which can be seen as a potential source of community-acquired MRSA. This finding highlights the need for strict hygiene practices during milking and adequate microbiological monitoring, especially considering that raw donkey milk is increasingly being consumed by sensitive consumer groups.

Donkey milk could provide a novel source of LAB strains with biotechnological potential (44). The present study revealed mean LAB counts of 2.30 log cfu/mL. The donkey milk from Piedmont farms was characterized by a higher overall LAB mean of 4.20 log cfu/mL (45), which is consistent with findings reported by Carminatti et al. (44).

The study limitation: It is important to note that this study relied on self-reported management practices, which may impose a degree of response bias. There is also the possibility that some respondents provided socially desirable answers, potentially affecting the accuracy and objectivity of part of the results. Another limitation of this study pertains to the use of a convenience sampling approach, which may restrict the generalizability of the study results. Nevertheless, this method was considered appropriate given the study's exploratory character and the logistical constraints involved.

CONCLUSION

In conclusion, this study demonstrates that dairy donkey farming in Serbia is characterized by suboptimal hygiene practices, insufficient preventive health care, and a lack of formal farmer training, although no analyzed microbiological hazards were detected in raw donkey milk samples. These findings highlight the need for targeted farmer education and basic farm management and milk hygiene guidelines. Furthermore, the present study offers a valuable evidence base for developing national regulations on donkey milk production and commercialization, supporting informed policymaking.

CONFLICT OF INTEREST

The authors declare that they have no financial or non-financial conflict of interest regarding authorship and publication of this article.

ACKNOWLEDGMENTS

This work was supported by the Ministry of Education, Science and Technological Development of the Republic of Serbia (Contract number 451-03-34/2026-03/200143). Special thanks to Dr. Tijana Ledina (Faculty of Veterinary Medicine, University of Belgrade) for her technical assistance during data collection.

AUTHORS' CONTRIBUTION

SB made the conceptualization, investigation, writing of the original draft, writing-review and editing, data curation and supervision. MKF, MK and ED were involved in writing-review and editing. JL was included in writing-review and editing and formal analysis. KN took part in writing of the original draft, writing-review and editing, data curation and supervision.

REFERENCES

- Šarić, Ć.Lj., Šarić, M.B., Kravić, S.Ž., Plavšić, D.V., Milovanović, I.Lj., Gubić, J.M., Nedeljković, N.M. (2014). Antibacterial activity of Domestic Balkan donkey milk toward *Listeria monocytogenes* and *Staphylococcus aureus*. *Food Feed Res.* 41(1): 47-54. <https://doi.org/10.5937/FFR14010475>
- Statistical Office of the Republic of Serbia-Statistical Yearbook of the Republic of Serbia (2024). [(accessed on 30 March 2025)]; Available online: <https://publikacije.stat.gov.rs/G2024/PdfE/G20242057.pdf>
- Trailović, D., Trailović, R., Urošević, M., Trailović, I. (2021). Breeding, care and diseases of donkeys. Center for Preservation of Indigenous Breeds-CEPIB, Belgrade. <https://onlinelibrary.wiley.com/doi/10.1111/age.12589>
- Stanišić, Lj., Aleksić, J.M., Dimitrijević, V., Kovačević, B., Stevanović, J., Stanimirović, Z. (2020). Banat donkey, a neglected donkey breed from the central Balkans (Serbia). *PeerJ.* 8, e8598. <https://doi.org/10.7717/peerj.8598> PMID:32175186 PMCID:PMC7059758
- Keitshweditse, B., Tsvakirai, C.Z., Mabuza, M.L., Tshela, M. (2024). Towards the expansion of the functional dairy market: Determining donkey milk value propositions and identifying possible consumers. *Future Foods.* 10, 100467. <https://doi.org/10.1016/j.fufo.2024.100467>
- Dai, F., Dalla Costa, E., Burden, F., Judge, A., Minero, M. (2019). The development of guidelines to improve dairy donkey management and welfare. *Ital J Anim Sci.* 18(1): 189-193. <https://doi.org/10.1080/1828051X.2018.1503571>
- Kovandžić, M., Pintarić, Š., Đorđević, J., Ledina, T., Savić Radovanović, R., Čobanović, N. (2024). On farm welfare conditions of dairy donkeys: A case study in Northern Serbia. *Meat Technol.* 65(2): 146-153. <https://doi.org/10.18485/meattech.2024.65.2.8>
- Conte, F., Panebianco, A. (2019). Potential hazards associated with raw donkey milk consumption: A review. *Int Dairy J.* 2019, 5782974. <https://doi.org/10.1155/2019/5782974> PMID:31275956 PMCID:PMC6582899
- Stanišić, Lj., Dimitrijević, V., Simeunović, P., Lakić, N., Radović, I., Ivanković, A., Stevanović, J., Stanimirović, Z. (2015). Morphological, biochemical and hematological characterization of endangered Balkan donkey breed. *Acta Vet-Beograd.* 65(1): 125-136. <https://doi.org/10.1515/acve-2015-0010>
- Šarić, Lj., Premović, T., Šarić, B., Čabarkapa, I., Todorčić, O., Miljanić, J., Lazarević, J., Karabasil, N. (2023). Microbiological quality of raw donkey milk from Serbia and its antibacterial properties at pre-cooling temperature. *Animals.* 13(3): 327. <https://doi.org/10.3390/ani13030327> PMID:36766215 PMCID:PMC9913105
- AWIN. (2015). AWIN welfare assessment protocol for donkeys. <https://air.unimi.it/retrieve/dfa8b992-3aaa-748b-e053-3a05fe0a3a96/AWINProtocolDonkeys.pdf>
- Deng, L., Shi, S., Li, J., Tang, C., Han, Y., Xie, P. (2021). A survey of smallholder farms regarding demographics, health care, and management factors of donkeys in Northeastern China. *Front Vet Sci.* 8, 626622. <https://doi.org/10.3389/fvets.2021.626622> PMID:33937368 PMCID:PMC8079733
- Statistical Office of the Republic of Serbia-Agricultural Census (2023) [(accessed on 30 March 2025)]; Available online: <https://www.stat.gov.rs/en-us/vesti/20240521-popispoljoprivrede2023/>
- Korhecz, R., Vladisavljević, M., Sedlak, O., Marcikić Horvat, A., Eremić Đodić, J. (2025). Demographic analysis of the agricultural population of Serbia. *Econ Agric.* 72(1): 205-224. <https://doi.org/10.59267/ekoPolj2501205K>
- The Donkey Sanctuary (2015). Donkey care handbook. 4th ed. The Donkey Sanctuary, United Kingdom
- Dai, F., Segati, G., Dalla Costa, E., Burden, F., Judge, A., Minero, M. (2017). Management practices and milk production in dairy donkey farms distributed over the Italian territory. *Maced Vet Rev.* 40(2): 131-136. <https://doi.org/10.1515/macvetrev-2017-0016>

17. Schäfer, J., Gerber, V., Hungerbühler, V., Schaefer, S., Unger, L. (2024). Management, health, and veterinary care of donkeys in Switzerland: A cross-sectional study. *Schweiz Arch Tierheilkd.* 166(12): 633-646.
<https://doi.org/10.17236/sat00439>
PMid:39623873
18. Werhahn, H., Hessel, E.F., Bachhausen, I., Van den Weghe, H.F.A. (2010). Effects of different bedding materials on the behavior of horses housed in single stalls. *J Equine Vet Sci.* 30(8): 425-431.
<https://doi.org/10.1016/j.jevs.2010.07.005>
19. Kwiatkowska-Stenzel, A., Witkowska, D., Sowińska, J., Stopyra, A. (2017). The effect of stable bedding materials on dust levels, microbial air contamination and equine respiratory health. *Res Vet Sci.* 115, 523-529.
<https://doi.org/10.1016/j.rvsc.2017.09.022>
PMid:28972942
20. Mandić, A., Matorkić, B., Spariosu, K., Radaković, M., Mitrović, A., Nenadović, K., Filipović, M.K. (2025). Obesity, urea and uric acid: potential indicators of subclinical metabolic imbalance in donkeys (pilot study). *Pak Vet J.* 45(3): 1415-1418.
<https://doi.org/10.22541/au.173920217.77636905/v1>
21. Dwyer, R.M. (2004). Environmental disinfection to control equine infectious diseases, *Vet Clin Equine.* 20(3): 531-542.
<https://doi.org/10.1016/j.cveq.2004.07.001>
PMid:15519816
22. Smith, D.G., Burden, F.A. (2013). Practical donkey and mule nutrition. In: R.J. Geor, M. Coenen, P. Harris (Eds.), *Equine applied and clinical nutrition: health, welfare and performance* (pp.106-140). Amsterdam, The Netherlands: Elsevier Ltd., Saunders
<https://doi.org/10.1016/B978-0-7020-3422-0.00016-X>
23. EU Platform on Animal Welfare (2020). Guide to good animal welfare practice for the keeping, care, training and use of donkeys and donkey hybrids, DOC/11066/2020. Available at: https://food.ec.europa.eu/system/files/2021-03/aw_platform_plat-conc_guide_donkeys_en.pdf
24. The Animal Health Law (Regulation EU/2016/429). Available at: <https://eur-lex.europa.eu/legal-content/EN/ALL/?uri=CELEX%3A32016R0429>
25. Lazić, S., Savić, S., Petrović, T., Lazić, G., Žekić, M., Drobnjak, D., Lupulović, D. (2023). Serological examinations of significant viral infections in domestic donkeys at the special nature reserve “Zasavica”, Serbia. *Animals.* 13(13): 2056.
<https://doi.org/10.3390/ani13132056>
PMid:37443854 PMCID:PMC10340027
26. Dairy Donkeys (2017). Good animal management practices for donkey milk production. Available at: <https://www.energie-cheval.fr/wp-content/uploads/2015/12/Dai-2017-Dairy-Donkeys-Good-practises-Livret.pdf>
27. Ruegg, P.L. (2004). Pre-milking cow preparation-secret methods of producing high quality milk. In: NMC Regional Meeting Proceedings (pp. 34-40).
28. Salimei, E., Fantuz, F. (2013). Horse and donkey milk. In: Y.W. Park, G.F.W. Haenlein (Eds.), *Milk and dairy products in human nutrition: production, composition and health* (pp. 594-613). Hoboken: John Wiley & Sons Ltd.
<https://doi.org/10.1002/9781118534168.ch27>
29. Camillo, F., Rota, A., Biagini, L., Tesi, M., Fanelli, D., Panzani, D. (2018). The current situation and trend of donkey industry in Europe. *J Equine Vet Sci.* 65, 44-49.
<https://doi.org/10.1016/j.jevs.2017.11.008>
30. Official Gazette of the Republic of Serbia (2017). Regulation on small quantities of primary products used to supply consumers, areas for performing of these activities, and derogations related to small entities in the business with food of animal origin. Official Gazette of the Republic of Serbia 111/2017. Available at: <https://faolex.fao.org/docs/pdf/srb172808.pdf>
31. Khan, M.Z., Chen, W., Li, M., Ren, W., Huang, B., Kou, X., Ullah, Q., Wei, L., Wang, T., Khan, A., Zhang, Z., Li, L., Wang, C. (2024). Is there sufficient evidence to support the health benefits of including donkey milk in the diet? *Front Nutr.* 11, 1404998.
<https://doi.org/10.3389/fnut.2024.1404998>
PMid:39385792 PMCID:PMC11462490
32. Rembischevski, P., Caldas, E.D. (2020). Risk perception related to food. *Food Sci Technol.* 40(4): 779-785.
<https://doi.org/10.1590/fst.28219>
33. Lucey, J.A. (2015). Raw milk consumption: risks and benefits. *Nutr Today.* 50(4): 189-193.
<https://doi.org/10.1097/NT.000000000000108>
PMid:27340300 PMCID:PMC4890836
34. Pilla, R., Daprà, V., Zecconi, A., Piccinini, R. (2010). Hygienic and health characteristics of donkey milk during a follow-up study. *J Dairy Res.* 77(4): 392-397.
<https://doi.org/10.1017/S0022029910000221>
PMid:20883563
35. Sarno, E., Santoro, A.M.L., Di Palo, R., Costanzo, N. (2012). Microbiological quality of raw donkey milk from Campania region. *Ital J Anim Sci.* 11, e49.
<https://doi.org/10.4081/ijas.2012.e49>
36. European Union (2004). Regulation No. 853/2004 of the European Parliament and of the Council of 29 April 2004 laying down specific rules for food of animal origin. *Off. J. Eur. Union L* 226: 22-82.

37. Papademas, P., Mousikos, E.P., Aspri, M. (2022). Valorization of donkey milk: technology, functionality, and future prospects. *JDS Commun.* 3(3): 228-233.
<https://doi.org/10.3168/jdsc.2021-0175>
PMid:36338810 PMCID:PMC9623768
38. Kaskous, S., Pfaffl, M.W. (2022). Milk properties and morphological characteristics of the donkey mammary gland for development of an adopted milking machine-a review. *Dairy* 3(2): 233-247.
<https://doi.org/10.3390/dairy3020019>
39. Russo, P., Fiocco, D., Albenzio, M., Spano, G., Capozzi, V. (2020). Microbial populations of fresh and cold stored donkey milk by high-throughput sequencing provide indication for a correct management of this high-value product. *Appl Sci. (Basel)*. 10(7): 2314.
<https://doi.org/10.3390/app10072314>
40. Soto Del Rio, M., Dalmaso, A., Civera, T., Bottero, M.T. (2017). Characterization of bacterial communities of donkey milk by high throughput sequencing. *Int J Food Microbiol.* 251, 67-72.
<https://doi.org/10.1016/j.ijfoodmicro.2017.03.023>
PMid:28431310
41. Coppola, R., Salimei, E., Succi, M., Sorrentino, E., Nanni, M., Ranieri, P., Belliblanes, R., Grazia, L. (2002). Behaviour of *Lactobacillus rhamnosus* strains in ass's milk. *Ann Microbiol.* 52, 55-60.
42. Giacometti, F., Bardasi, L., Merialdi, G., Morbarigazzi, M., Federici, S., Piva, S., Serraino, A. (2016). Shelf life of donkey milk subjected to different treatment and storage conditions. *J Dairy Sci.* 99(6): 4291-4299.
<https://doi.org/10.3168/jds.2015-10741>
PMid:26995124
43. Naccari, F., Foti, M., Giacobello, C., Mariavitale, P., Passantino, A., Russo, C., Conte, F. (2009). Methicillin resistant *Staphylococcus* sp. isolated from donkey's milk in Sicily: preliminary study. In: *Proceed. of 1st Int. Congress of the European Assoc. for Vet. Pharmacol. and Toxicol.* July, 12-16, (pp.150), Leipzig, Germany.
44. Carminati, D., Tidona, F., Fornasari, M.E., Rossetti, L., Meucci, A., Giraffa, G. (2014). Biotyping of cultivable lactic acid bacteria isolated from donkey milk. *Lett Appl Microbiol.* 59(3): 299-305.
<https://doi.org/10.1111/lam.12275>
PMid:24749686
45. Cavallarin, L., Giribaldi, M., de los Dolores Soto-Del Rio, M., Valle, E., Barbarino, G., Gennero, M.S., Civera, T. (2015). A survey on the milk chemical and microbiological quality in dairy donkey farms located in North Western Italy. *Food Control.* 50, 230-235.
<https://doi.org/10.1016/j.foodcont.2014.08.019>