

## EPIDEMIOLOGY OF HUMAN BRUCELLOSIS IN ANIMAL-EXPOSED INDIVIDUALS IN DISTRICTS DIR (DIR UPPER AND DIR LOWER), KHYBER PAKHTUNKHWA, PAKISTAN

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### Article Info



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### Abstract

**Background:** Brucellosis, a zoonotic contagious disease caused by the bacterium of the genus *Brucella*, is a significant public health concern. The infection can cause damage to various organs and systems in the body, both in acute and chronic phases. Understanding the disease's epidemiology and risk factors is crucial for control and prevention. Therefore, this study aims to provide insights into the recent infection status and risk factors for brucellosis in Districts Dir of KP-Pakistan.

**Methods:** This study sample includes 261 individuals exposed to animals in domestic rearing or farms; the study period was from February 2021 to August 2021. Blood samples were collected by a trained person, and a pre-designed questionnaire was used to collect demographic information. The blood samples were subjected to ELISA(IgM), and statistical software STATA (Version-14) was used for data analysis.

**Results:** The result indicated an overall seroprevalence of 36.40% in District Dir, higher in Dir Lower (39.62%) than in Dir Upper (34.10%). The disease was more prevalent in women (39.05%) than in men (31.52%). Individuals with higher age (age  $\geq 41$ , 51.35%), being illiterate (47.9%), and farm workers (53.33%) had the highest prevalence of the disease ( $P < 0.05$ ).

**Conclusion:** This preliminary study has revealed an alarming prevalence of Brucellosis among District Dir people involved in domestic animal rearing and farm workers. To mitigate the spread and impact of this disease, urgent and comprehensive measures are needed. These measures should include vaccination, public awareness and the use of protective equipment when handling animals and their products.

### Keywords:

*Brucellosis, epidemiology, zoonosis, risk factors, Pakistan.*

## Background

Brucellosis is an infectious disease caused by bacteria of the genus *Brucella*. This is the most prevalent zoonotic disease worldwide (1,2) and is associated with multiple brucella species (3). The causative agent of bovine brucellosis is *B. abortus*, a gram-negative coccobacillus, *B. melitensis*, *B. suis* (4, 5), while in goats and sheep *B. melitensis* produces the disease, while in rams *B. ovis* causes epididymitis (6). Bovine brucellosis is sometimes called a contagious abortion or Bang's disease. Human brucellosis manifestations occur in the form of undulant fever, Mediterranean fever, Crimean fever, remitting fever, goat fever, maltase fever and Gibraltar fever (7).

The species in the genus *Brucella* has the property to adapt to the new host upon transmission. They can transmit to their primary host by direct and indirect contact and occasionally transmit to other susceptible hosts (8). Its risk increases with the mix-farming practices (9).

The disease is primarily endemic in low- and middle-income countries across the Mediterranean region, the Arabian Peninsula, Asia, Africa, and certain areas of Central and South America (10, 11). Animal brucellosis exhibits the highest prevalence in the Middle East, sub-Saharan Africa, China, India, Peru, and Mexico (12). Few countries are free from brucellosis, predominantly located in developed regions such as Western and Northern Europe, Canada, Japan, Australia, and New Zealand (13). In the Middle East, Yemen, Iran, Syria, Turkey, and more recently Saudi Arabia, report significant numbers of human cases (14). Yemen has the highest annual incidence rate, at 88.6 per 100,000. In endemic regions, the disease is transmitted through direct contact or exposure to environments contaminated with birthing materials, such as infected placentas, foetuses, or uterine fluids.

In humans, in addition to direct contact with animals, the disease is also acquired by consuming animal products such as unpasteurised milk and cheese, etc. Human brucellosis affects people of any age and sex; it appears in many signs and symptoms, including intermittent fever, chill, excessive sweating, headache, body pain, and unidentified weight loss. These non-specific symptoms make it difficult to distinguish between brucellosis and other febrile conditions. However, timely diagnosis and treatment of the disease are crucial; otherwise, the disease becomes chronic and can potentially lead to other serious complications, including osteoarticular, hepatobiliary, cardiovascular, and central nervous system disorders (15). Additionally, brucellosis poses a significant occupational risk to those working with livestock, such as veterinarians, farmers, and slaughterhouse workers, who are at higher risk due to their exposure to infected animals and aborted foetuses or placentas (16).

In Pakistan, Brucellosis is also regarded as an endemic that affects domestic ruminants (e.g., cattle, buffalo, sheep, goats, and camels), non-ruminants (e.g., horses and dogs), wildlife, and humans. As an agricultural country, the livestock sub-sector is essential to Pakistan's economy. According to agricultural sector survey's 2022-2023 estimations, there were 55.5 million cattle and 45 buffaloes 32.3 million sheep and 84.7 goats, 6.4 million equines, and 1.1 million camels (17). In Pakistan, there is a strong bond between animals and humans not only because animals are their source of income but also because they symbolise their financial status for individuals and institutions.

Pakistan faces a double burden of disease: economic loss to the animals and their decreased production, as well as human morbidity associated with it. Therefore, this study aims to investigate the prevalence of Brucellosis in the exposed population in District Dir Lower.

Methodology

Study Area

This study, the first of its kind in the area, was conducted in district Dir, Khyber Pakhtunkhwa (KP), Pakistan. No previous study about this zoonotic disease has been conducted in this location, making our research particularly significant.

Dir is in the north-western belt of KP province of Pakistan. It is divided into two regions, i.e. District Upper Dir and Lower Dir. The area is comprised of the mountains of the Hindukush range. The area of Dir is 5282 km<sup>2</sup>. It has summer and winter seasons. In many areas, there is heavy snowfall, especially in Upper Dir, which is considered a cold area of Pakistan. The temperature ranges from 34.46C<sup>o</sup> in summer to -2.3C<sup>o</sup> in winter. Geographically, Dir is bordered by Swat in the east, Malakand in the south, Bajaur in the southwest, Afghanistan in the west, and Chitral in the northwest (18). Due to substantial hilly grazing grounds and fertile agricultural land, people in rural areas rear animals, especially cows, buffaloes, goats, and sheep, for milk and meat purposes. Also, there are many animal farms for commercial purposes in the area from which milk and dairy products are supplied to the markets.

Data Collection

The blood sampling was performed using a stratified random sampling technique. A trained person collected a total of 261 blood samples after obtaining informed consent from the participants who were occupationally or domestically exposed to animals, veterinary professionals, farm workers, butchers, and women rearing animals at home. Veterinary professionals are the people who are trained to practice veterinary medicine. They may diagnose, treat, and prevent illness in animals or work in veterinary laboratories. Animal farm workers are the people responsible for the care and management of livestock. Their duties include feeding and watering animals, milking, cleaning and maintaining animal's houses, monitoring animal health, herding and moving animals, assisting with breeding and birthing, and keeping a record of the farm. Butchers are professional people who work in slaughterhouses and prepare and sell meat products. Females are not involved professionally but domestically in caring for animals at home. They water and feed animals, perform milking, and clean and maintain their animal's houses or rooms. Individuals who had no direct contact with animals were excluded from this study (Table 1).

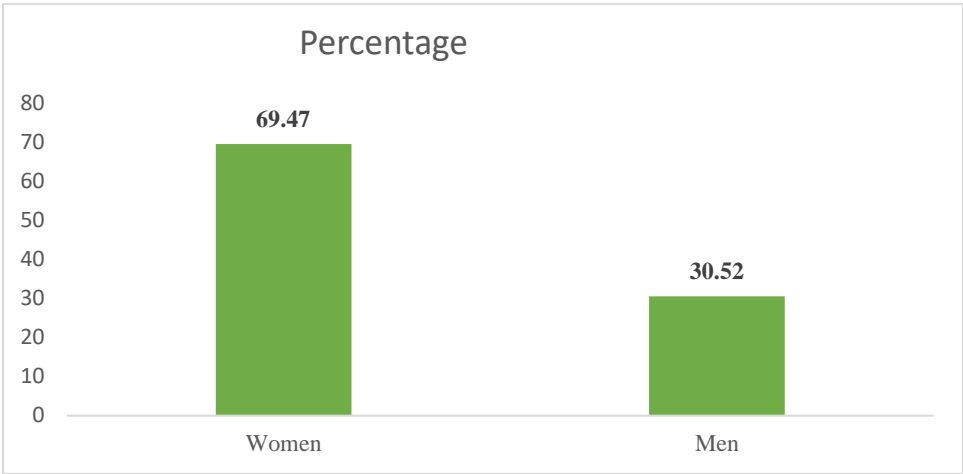
Table 1: Socio-demographic factors of study sample district Dir (Upper and Lower).KP, Pakistan

S#	Variable	Number	Percentage
a.	Area		
	Upper Dir	102	(39.08%)
	Lower Dir	159	(60.92%)

	Total	261	(100%)
<b>b.</b>	<b>Gender</b>		
	Male	92	(35.25%)
	Female	169	(64.75%)
<b>c.</b>	<b>Age</b>		
	Up to 20	31	(11.88%)
	21 – 40	156	(59.77%)
	41-above	74	(28.35%)
<b>d.</b>	<b>Education</b>		
	Illiterate	98	(37.55%)
	Secondary	142	(54.41%)
	Graduated	21	(8.05%)
<b>e.</b>	<b>Occupation</b>		
	Housewives	169	(64.75%)
	Form workers	30	(11.49%)
	Butchers	45	(17.24%)
	Veterinary doctors	17	(6.51%)

**Results**

The overall positive cases of brucellosis were 95/261 (36.40%) by ELISA in people of Dir (Dir upper and Dir lower). Of the affected individuals, 69.47% were women, and 30.53% were men (Figure 1). The prevalence in Dir Upper and Dir Lower was found to be 34.10% and 39.62%, respectively. The gender-wise prevalence showed 31.52% in male samples and 39.05% in female samples, higher in females than males. However, statistically significant differences were found in the age-wise prevalence of 29.03%, 30.77%, and 51.35% in the three age groups, i.e. Up to 20, 20-40, and 40 & above, respectively. The education level was also significantly associated with the prevalence (47.96%, 33.10%, and 4.76% in the three education-based categories, i.e. Illiterate, secondary education, and graduation, respectively) ( $P < 0.001$ ). In the occupation-based categories, the prevalence was found to be 39.05%, 53.33%, 22.22%, and 17.65% in the four categories of Housewives, Farmworkers, Butchers, and veterinarians respectively being highest in the farm workers ( $P = 0.013$ ) (Table 2).



**Figure: Percentage of men and women affected by Brucellosis in Dir Upper & Lower**

**Table 2. Seroprevalence of brucellosis in district Dir (Upper and Lower), KP Pakistan**

S#	Variable	ELISA (positive) N* (%)	P*-value
a.	Area		
	Upper Dir	32 (34.10%)	0.176
	Lower Dir	63 (39.62%)	
	Total	95 (36.40%)	
b.	Gender		
	Male	29 (31.52%)	0.227
	Female	66 (39.05%)	
c.	Age		
	Up to 20	9 (29.03%)	0.007
	21 – 40	48 (30.77%)	
	41-above	38 (51.35%)	
d.	Education		
	Illiterate	47 (47.96%)	<0.001
	Secondary	47 (33.10%)	
	Graduated	1 (4.76%)	
e.	Occupation		
	Housewives	66 (39.05%)	0.013
	Form workers	16 (53.33%)	

	Butchers	10 (22.22%)	
	Veterinary doctors	3 (17.65%)	

n = number of positive cases.

P = significant value by the chi-square test for differences between groups.

**Discussion**

The main objective of this study was to find seroprevalence in the population of Dir with a specific focus on the people exposed to animals by rearing them for domestic or commercial use. To the best of our knowledge, this is the first study on Brucellosis's seroprevalence in the population of district Dir Lower and Upper. The study showed an overall seroprevalence of 36.40% in the exposed people of this study area, being the highest of all results reported previously from other locations in KP province. The previous studies in other districts of KP province showed 10% by SPAT (Serum Plate Agglutination Test) in Charsadda (19), 24.6% by SPAT in occupationally exposed veterinary professionals in District Bannu (20), and 27.47% by ELISA in the female residents of Malakand, district (21).

One reason for this high seroprevalence rate is that only those individuals who were in close contact with animals, occupationally or domestically, were sampled for this study. Secondly, the geographic location of District Dir is favourable for Brucella's growth. District Dir, located in the northern region of Khyber Pakhtunkhwa (KP), Pakistan, experiences relatively lower temperatures than the previously mentioned areas. This cooler climate provides an ideal environment for the survival of Brucella bacteria, which thrive in moderately cold conditions. Given that geographical factors play a significant role in the prevalence of diseases, the unique climate of District Dir may contribute to the persistence and transmission of brucellosis in the area. The study showed that seroprevalence was 39.62% in Lower Dir and 31.37% in Upper Dir. The seroprevalence in Lower Dir was higher than in Upper Dir. This high prevalence in Lower Dir might be due to its urban setting, which has numerous dairy farms and slaughterhouses, resulting in more frequent animal exchanges. On the other hand, Upper Dir is mainly rural, where animals are kept domestically in homes, and there is almost no farming.

The current study showed that the seroprevalence is 31.52% in males and 39.05% in females.

Similar results were reported from other districts of KP-Pakistan in other studies, as 37.06% in women, 24.2% in men from various hospitals in Peshawar by SPAT (Shahid M 2014), 14.6% in women, and 12.1% in men by SAT from selected hospitals in district Abbottabad (22). In contrast, studies from other countries such as India (23), Bangladesh (24) and Egypt (25) showed that the incidence is higher in men than women.

This higher prevalence in females of Pakistan is attributed to their increased time spent in activities involving animals, including milking, handling their waists, and animal husbandry at home. Moreover, women in these rural areas of the District Dir were mostly illiterate and were unaware of the risks associated with zoonotic diseases with rearing animals. In comparison, there were more educated men in the male sample and some veterinary professionals and butchers who were more knowledgeable regarding

zoonotic diseases. However, male farm workers were also found to be careless due to being largely uneducated and unaware of zoonotic diseases.

Brucellosis affects people irrespective of their age. The study showed that a high prevalence exists in the age group of 41 years and above, i.e. 51.35% in the area. Mainly, people from this age group are involved in the rearing, milking, and slaughtering of animals. High prevalence in the age group 40-60 in district Charsadda was also reported by Parveen et al., 2015, which shows similarity with the present findings (19). Similarly, Adam and Hassan, 2010, found in their study that the average age of a Brucellosis patient was 43.9 years. (26)

The prevalence of brucellosis found in the group based on education was highest in the category of “Illiterate”, i.e., 47.96%. The high prevalence rate in the uneducated group of people is due to unawareness of taking protective measures during animals’ caregiving practices.

The study showed the highest prevalence rate among farm workers, compared to the other categories based on occupation, which is 53.33%. Previously, Ali et al., 2018, reported a 20.40% prevalence (27), Tumwine et al., 2015 reported a 20.5% prevalence (28), and Shahid, 2014 reported a 33% prevalence in farm workers higher than other occupational categories (29). Most of the farmworkers in the area were using raw milk, a risk factor for infection with brucellosis.

This study has highlighted the zoonotic disease Brucellosis, indicating the high prevalence and transmission from animals to humans. The inappropriate methods of animal rearing and managing livestock in the Dir district have led to this alarming situation of increased health risks for the people. The intensified human-animal interactions, lack of awareness of the health risks associated with animal rearing, and ignorance of the protective measures while handling animal products and waste materials are vital contributors to the rise in brucellosis. These findings underscore the need for enhanced surveillance systems, stricter laws on animal health, and improvement of the public health system. Regular monitoring and immunisation of animals and caretakers are recommended. Public awareness campaigns, combined with One Health approaches integrating animals and public and environmental health, require the collaboration of all stakeholders to reduce the incidence of zoonotic diseases.

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### **Ethical Statement**

This study proposal was evaluated and Approved by the Advanced Study Research Board (ASRB /Dir/A&R/AWKUM/2021/ 5635) committee members of Abdul Wali Khan University Mardan, Khyber Pakhtunkhwa, Pakistan. All ethical standards regarding human subject use, informed consent from participants and following the ethical guidelines outlined in the Declaration of Helsinki to protect human rights and welfare are ensured.



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