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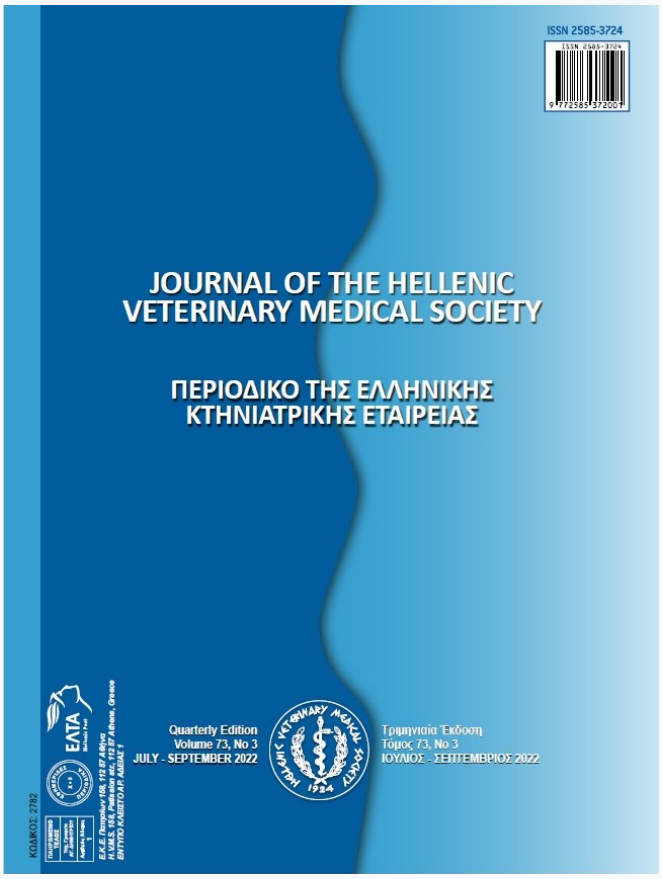

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Research article
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Community based assessment on knowledge, attitude and practices (KAP), risk factors and One Health perspective of brucellosis in rural and urban settings of Pakistan: A Cross-Sectional Study

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ABSTRACT: Bovine brucellosis is an endemic zoonotic infection of ruminants in Pakistan with detrimental impact on livestock economy. The major losses caused by brucellosis in animals include abortions, week calves, infertility, and reduced milk production. Regardless of the prevalence of the disease, limited data is available about brucellosis in Pakistan. The present study aimed to access the one health concept in prospective of brucellosis among small-scale dairy farmers by examining knowledge, attitude and practices at the farms and household level that might pose a risk for humans contracting brucellosis. A community-based cross-sectional survey was conducted among small household dairy farms (n=333) in rural and urban areas of district Rawalpindi and Islamabad. The results revealed almost 74.17% of the participants knew about animal brucellosis and 69.97% of the participants were able to identify the symptoms of animal brucellosis. 46.25% of participants showed knowledge about the transmission of disease from animals to humans. A majority (70.87%) of participants used the raw milk for making other dairy products. Most household farmers are unaware of routes of transmission and major risk factors of brucellosis. Almost all the participants, except veterinarians, including butchers, dairy farm owners and workers, slaughterhouse owners and workers and farmers were involved in at least one risky practice. The moderate knowledge and poor understanding of the disease emphasize the need to initiate awareness programs to educate the farmers who are at high risk along with the improvement of vaccination programs for animals and strict implementation of brucellosis eradication policy which should be devised by government.

Keywords: Brucellosis; Knowledge Attitude; Practice; Risk factors; One health

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INTRODUCTION

Brucellosis is one of the most neglected zoonotic diseases of the world which is caused by the bacteria of genus *Brucella*. This zoonotic infection of livestock has a huge socio-economic impact (O'Callaghan, 2020). Transmission of brucellosis occurs in ruminants through the excretion of contaminated materials from the female genital tract, which constitutes the main form of transmission to other animals and humans. It is also common that excretion occurs through milk or semen (Nejad et al., 2020). Human brucellosis is mainly caused by the contact with infected animal or use of contaminated dairy products (Nejad et al., 2020). Control of brucellosis requires one health approach where animal and human health authorities must work together with livestock holders and awareness programs should be conducted to educate the people at risk (Iabal et al., 2020; O'Callaghan, 2020).

Brucellosis is an endemic zoonotic infection of global significance which has deleterious impact on public health and animal food production (Peng et al., 2020 and Hou et al., 2019). This zoonotic infection of *Brucella,* which infect domestic livestock include *Brucella abortus* in bovines, camels and yaks, *Brucella ovis* in rams, *Brucella meltitensis* in ovines and caprines, *Brucella suis* in swine (Primary host) (Ko & Splitter, 2003) and reindeer. Another two species of *Brucella* including *Brucella ceti* and *pinni pedialis* have also been reported in marine mammals (Cao et al., 2020 and O'Callaghan, 2020a, Shome et al., 2020 and Poester et al., 2014). *B. abortus* is the causative agent of bovine brucellosis however cross-species transmission can also occur when bovines that are kept in close contact with small ruminants, the causative agent of which is *B. melitensis* (Ali et al., 2014; Neta et al., 2010 and Olsen & Tatum, 2010). The species *B. melitensis* biovars 1-3 have been reported in sheep and goats, and *B. abortus* biovars 1-6 and 9 in cattle (Khan & Zahoor, 2018). The *B. abortus* biovars have been reported from different regions of the world; biovar 1 from Trinidad (Fosgate et al., 2002), biovars 1, 2, 3, 4 and 6 from Brazil (Poester et al., 2002 and Minharro et al., 2013), biovar 3 from Tanzania, Kenya and Bangladesh (Mathew et al., 2015; Muendo et al., 2012; Islam et al., 2019), biovar 4 from Colombia (Higuera et al., 2019), biovars 5, 6 and 9 from Spain (Ocampo-Sosa et al., 2005). Globally brucellosis has been reported in 86 different countries (Khan & Zahoor, 2018) and is reported to be eradicated from Central and North Europe, New Zealand, Australia, Canada and Japan (*Brucellosis: OIE*, 2020). But it is still present in South and Central America, Middle East, Mediterranean and Caribbean Basin, Africa and Asia (Adetunji et al., 2019 and Kolo et al., 2019).

The clinical manifestation caused by bovine brucellosis in female animals includes abortion, metritis, vaginal secretions, weak calves, retention of placenta, still birth, low rate of fertility, reduced milk yield and neonatal and embryonic death (Bifo et al., 2020; Pal et al., 2017; Megid et al., 2014; Mekonnen et al., 2010). In male animals bovine brucellosis is characterized by epididymitis, uni- or bilateral testicular atrophy, orchitis, infertility and sperm abnormalities (Megid et al., 2014; Poester et al., 2014; Abubakar et al., 2012). The animals infected by brucellosis shed bacteria in the environment through milk and vaginal secretions and other animals get infected by direct contact with the infected animal or by consuming the contaminated water or feed (Jamil et al., 2020). The laboratory diagnosis of brucellosis can be based on direct and indirect methods and should be performed whenever clinical signs or epidemiological evidences suggestive of the disease are observed (Jamil et al., 2020). Several biological samples can be used for monitoring and laboratory confirmation of the *Brucella spp*. Infection, for example, milk or serum (Jamil et al., 2020). Different immunological diagnostic tests used for the diagnosis of brucellosis include Rose Bengal Plate Test (RBPT) (Abdelbaset et al., 2018), Serum Agglutination Test (SAT) (Kazak et al., 2016), Enzyme-Linked Immunosorbant Assay (ELISA) (Vatankhah et al., 2019), Complement Fixation Test (CFT) (Moti et al., 2013), Milk Ring Test (MRT) (Ali et al., 2015). In addition to the culture and isolation of the infectious agent, the presence of *Brucella spp* can be detected by detecting the genetic material by molecular techniques such as Polymerase Chain Reaction (PCR) (Mol et al., 2020). Molecular techniques are supplemented by phylogenetic techniques such as Multilocus Sequence Typing (MLST) (Piao et al., 2018), Multi Locus Variable number tandem repeat Analysis (MLVA) (Ma et al., 2016) and Single Nucleotide Polymorphism (SNP) (Zafari et al., 2020).

Pakistan, being an agricultural country, relies greatly on livestock and almost 8 million families earn their >35% income from raising livestock. Livestock is the backbone of rural economy and it has been reported that in 2017-18, the livestock products has contributed 58.92% to the gross domestic product

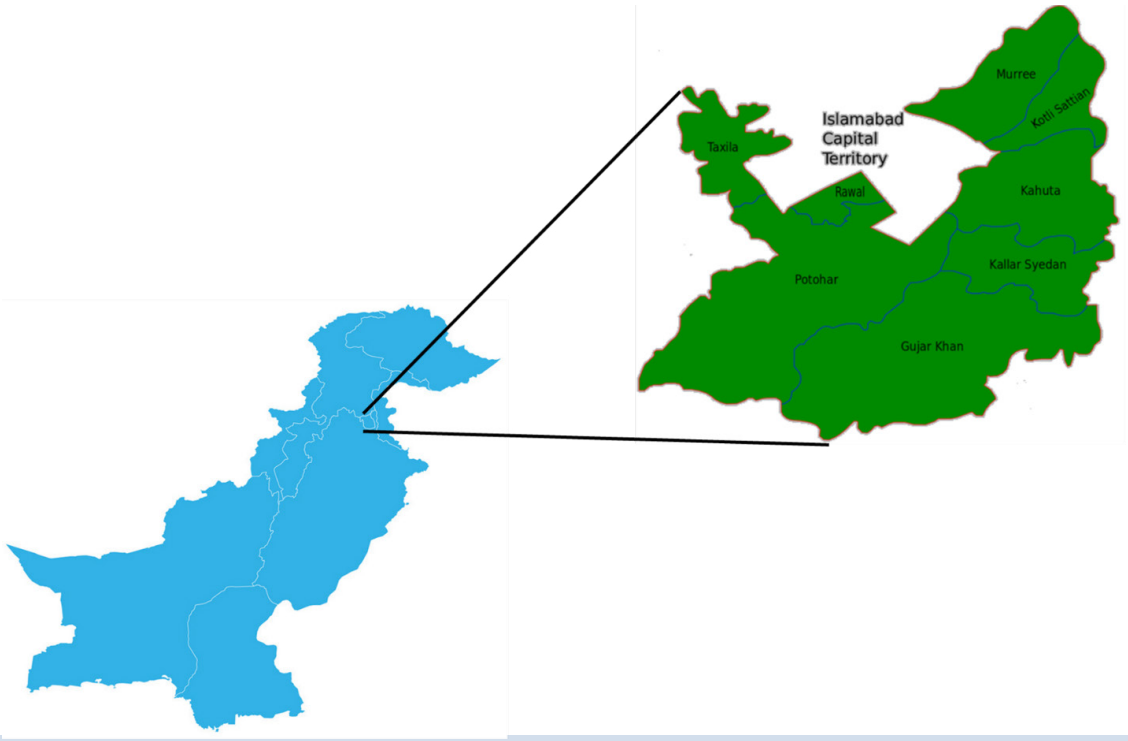

Figure 1 Map showing study area

of Pakistan (Saeed et al., 2020b). Any disease affecting the livestock has a long-lasting and devastating effect on economy of Pakistan. Brucellosis is responsible for adverse effects on animal health consequently causing major economic impact on animal industry. In Pakistan *B. abortus* biovar 1 has been reported to cause bovine brucellosis (Ali et al., 2019; Ali et al., 2014). In Pakistan bovine brucellosis is endemic (Dong et al., 2020; Ali et al., 2016, 2017, 2019, 2020) with the estimated prevalence of 6.5% in different regions of Pakistan (Saeed et al., 2020a).

In spite of having such devastating effects, very few studies have been conducted regarding livestock brucellosis in Pakistan. Pakistan has also developed a five years (2018-2023) national action plan based on One Health Concept to control the brucellosis. According to the draft brucellosis in livestock will be controlled by maintaining animal health cards and registers along with implementation of farm biosecurity principles, awareness programs, training sessions and research will be conducted, strengthening of diagnostic capacity and surveillance against brucellosis at regional, provincial and central level and development of networking between animal and human laboratories for better coordination and sharing of data (Iqbal et al., 2020).

However previous studies from endemic countries have reported significant knowledge about brucellosis in urban and rural populations, with 40-100% of population reporting the awareness about disease (Njenga et al., 2020). The formal education of farmers along with necessary training of persons associated with livestock would greatly help to reduce the burden of disease (Khan & Zahoor, 2018).The availability of limited data is one of the major hurdles towards the eradication of disease. This study was aimed to assess the Knowledge, Attitude and Practices (KAP) regarding the brucellosis in population of Rawalpindi district Pakistan. The information gathered will help to answer some of the major questions to define the conceptual framework for "One Health" to combat brucellosis in Pakistan.

MATERIAL AND METHODS

Study area

A community based cross-sectional analysis was conducted in Rawalpindi district of Punjab and Islamabad, Pakistan. From Rawalpindi district the data was collected from the rural and urban population of Gujar Khan, Kallar Sayaddan, Kahuta, Murree, Rawalpindi and Kotli Sattian (Figure 1). These areas are situated between longitudes of 33.5651° N and latitudes of 73.0169° E. The climate ranges from drizzly warm to

cool dry wintry with the characteristics of the semi-arid region. These districts were selected because of the practical convenience, where human and animals are living in close proximity and many small-scale dairy farmers in these areas who possess less than 10 cattle and buffaloes (Afzal, 2009). The demographic characteristics of the study area are described in Table 1.

Ethics statement

The study was approved by the Ethics Review Board, Department of Biosciences, COMSATS University Islamabad under reference number CUI/BIO/ERB/21/07. The names of participants were not recorded, and participants were also ensured about the privacy of the data.

Participation and sample size

The participants were randomly selected from the rural and urban areas of the study area. People associated with livestock includes farmers who have household livestock, dairy farm owners, veterinarians, butchers, slaughterhouse owners, workers at livestock facilities, general animal health care providers and co-workers participated in the study. The participants who do not have a direct contact with livestock or are below the age of 15 were excluded from this study. Simple random sampling was considered throughout this study and the permanent residents of the Rawalpindi were considered the population, for the categorical data, the margin of error is set to 0.05, the estimate of variance is set at 0.50, and the t-value is fixed at 1.65 by following the Kotrlik and Higgins (2001). Accordingly, sample size of a total of 333 participants were identified and randomly interviewed from the study areas identified based on exclusion and inclusion criteria. The data was collected from August to December 2020.

Data collection

A structured questionnaire was developed to collect the data based on existing literature and contained questions that focused on various topics. Interview method was used to collect the data after the informed consent was obtained from all the participants. A team was trained for interview, data collection and keeping records. The interview process was routinely coordinated with the supervisor to ensure data collection and record maintenance. The purpose of study was explained to the participants. First, the veterinarians and general animal health care providers were asked to fill the questionnaire. The farmers, butchers, dairy farms owners and workers and slaughterhouse owners

Table 1: Demographics of Study Area

Study Area	Tehsil	Region	Population	Area (Sqkm)
Islamabad	Islamabad	Rural	991,747	906
		Urban	1,009,832	
		Grand Total	2,001579	
Rawalpindi District	Rawalpindi	Rural	2,530117*	5285*
		Urban	2,875,516*	
		Grand Total	5,405,633*	
	Gujar Khan	Urban	566,415	1457
		Grand Total	112,0888	
		Grand Total	678,503	
	Kahuta	Rural	160,019	1096**
		Urban	60,557	
		Grand Total	220,576	
	KallarSyedan	Rural	162,938	421
		Urban	54,335	
		Grand Total	217,273	
	KotliSattian	Rural	96,878	304
		Urban	22,434	
		Grand Total	119,312	
	Murree	Rural	207,655	434
		Urban	25,816	
		Grand Total	233,471	
		Urban	2,098231	
		Grand Total	3,258,547	

Source: Pakistan Bureau of Statistics; *Including the statistics of Tehsil Taxila as well; **Including the area of Kallar Seyeddan)

and workers were interviewed. The participants from rural areas were interviewed in their native language. The female participants were interviewed where they are most involved with livestock.

The questionnaire consisting of 84 questions compromising of six parts. The first part included questions about demographic characteristics, second part include knowledge about common symptoms of brucellosis in animals and humans, and potential tests used for diagnosis and identification of brucellosis in animals. The third part include attitude of the participants towards brucellosis and forth part include practices towards brucellosis in animals and suspected humans. The fifth part of the questionnaire focused response of the participants against potential risk factors associated with brucellosis and sixth part consist of the response of the participants towards One Health concept regarding brucellosis.

Statistical analysis

The database was established using MS Excel (Microsoft, Redmond, WA, USA) and basic frequencies were derived. Descriptive statistics was used to initially analyze the data and the variables were classified into independent and dependent variables. The independent variables include age, gender, ethnicity, qualification, occupation, residence, and farming type. The dependent variables include the knowledge about brucellosis, attitude towards brucellosis, practices regarding brucellosis, risk factors associated with brucellosis and questions about One Health. Chi-Square (χ^2) test was used to analyze the significant association between independent and dependent variables. The Spearmen's rank correlation was used to analyze the correlation between knowledge, attitude, practices and risk factors. The *P* values less than 0.05 were considered significant. Statistical analysis was performed using SPSS version 23 (SPSS, Inc., Chicago, IL).

RESULTS

Sociodemographic characteristics of study population

In the current study, a total of 333 participants were included. The sociodemographic characteristics of study population are shown in Table 2. The study population was dominated by males (68.47%) included farmers (44.44%), dairy farm owners (15.01%), veterinarians (6.31%), workers at livestock facility (8.11%) butchers (15.92%), slaughterhouse owners and workers (2.40%) and general animal health care provider (2.40%). Overall,74.47% respondents were Punjabi and 28.53% of the respondents were educated up to the matriculation and 12.61% had no formal education. About 55.26% of the participants belonged to the rural areas and 65.17% of the participants reported the presence of animal health care facility in their area.

Table 2: Sociodemographic characteristics of study population

Variables	Characteristics	Number	Percentage (%
Location	Islamabad	60	18.02
	Rawalpindi	273	81.98
Tehsil	Islamabad	60	18.02
	Rawalpindi	54	16.22
	Gujar Khan	60	18.02
	Kallar Sayyedaan	60	18.02
	Kahuta	60	18.02
	Kotli Sattian	17	5.10
	Murree	22	6.60
Age (in Years)	Below 15	0	0
	15-25	32	9.61
	26-35	57	17.12
	36-45	73	21.92
	46-55	74	22.22
	56-65	62	18.62
	66-75	20	6.01
	76-85	11	3.30
	86-95	4	1.20

Gender	Male	228	68.47
	Female	105	31.53
Ethnicity	Punjabi	248	74.47
	Sindhi	13	3.90
	Pathan	23	6.91
	Urdu speaking	18	5.41
	Kashmeri	18	5.41
	Other/ non-reported	13	3.90
Religion	Muslim	333	100.0
	Non-Muslim	0	0
Marital Status	Married	272	81.68
	Single	61	18.32
Qualification	No formal education	42	12.61
	Primary	41	12.31
	Middle / Elementary	74	22.22
	Matriculation	95	28.53
	Intermediate	31	9.31
	Bachelors	28	8.41
	Masters	22	6.61
	PhD	0	0
Occupation	Farmer (Household livestock)	148	44.44
	Dairy farm owner	50	15.01
	Veterinarian	21	6.31
	Worker at livestock facility	27	8.11
	Butcher	53	15.92
	Worker at slaughterhouse	8	2.40
	General animal health care provider	8	2.40
	Worker at dairy farm	18	5.40
Residence	Rural	184	55.26
	Urban	149	44.74
No of Family members	Less than 5	70	21.02
	5-10	250	75.08
	11-15	9	2.70
	More than 15	4	1.20
Income per Month	Below 10,000 PKR	12	3.60
	11,0000-20,000 PKR	45	13.51
	20,000-30,000 PKR	126	37.84
	Above 30,000 PKR	150	45.05
Time spend with animals (Hours per day)	3-5	140	42.04
	6-10	175	52.55
	11-15	16	4.80
	More than 15	2	0.60
Livestock species	Sheep	3	0.90
	Goat	30	9.01
	Cattle	89	26.73
	Mixed	146	43.84
	Not applicable	65	19.52
Approximate No of animals owned	Less than 5	77	23.12
	6-15	104	31.23
	16-25	41	12.31
	More than 25	31	9.31
	Not applicable	80	24.02

Farming type	Pastoral	64	19.22
	Agro-pastoral	186	55.86
	Not applicable	83	24.92
Presence of animal health care facility in the area	Yes	217	65.17
	No	69	20.72
	Don't know	47	14.11

Knowledge about brucellosis

Most (74.17%) of the participants had already heard about brucellosis and 61.86% of participants did not know that bacteria can infect animals. About 69.97% of the participants were able to identify symptom (one or all) of animal brucellosis and majority (58.26%) of participants did not know about the symptoms of the brucellosis in humans. Brucellosis is preventable in animals by proper vaccination was known by 34.53% of the participants and 55.55% have knowledge that it can be diagnosed in animals. Most (71.17%) of the participants confirmed the presence of brucellosis infected animal in their area. 82.58% of participants reported that they have not heard of any scheme or initiative or plan to control brucellosis. Majority (69.67%) of the respondents had no knowledge about the tests used to diagnose brucellosis. The participants were less (27.93%) knowledgeable about transmission of brucellosis through blood transfusion (Table 3).

Table 3: Knowledge of participants about Brucellosis

Variable	Characteristics	N	Percentage (%)
How are you in contact with animals?	Owner (a)	70	21.02
	Herding (b)	22	6.61
	Milking (c)	13	3.90
	Dung cleaner	7	2.10
	Health provider	27	8.12
	Butcher / Slaughter the animals	57	17.11
	a, b, c and d	126	37.84
Do you know that bacteria can infect animals?	Yes	127	38.14
	No	206	61.86
Have you ever heard of Brucellosis?	Yes	247	74.17
	No	86	25.83
How long have you known about Brucellosis?	1-5 Years	100	30.03
	6-10 Years	113	33.93
	11-15 Years	37	11.11
	Not applicable	83	24.92
From where did you get the information about Brucellosis?	TV / Radio	8	2.40
	Social media (FB/Twitter and other social media platforms)	12	3.60
	From a training session	22	6.61
	From an awareness program about animal health	44	13.21
	Veterinary staff	37	11.11
	Community health worker	39	11.71
	Relatives / Family / Friends	93	27.93
	Other sources	10	3.00
	Not applicable	68	20.42
What are the common symptoms of Brucellosis in animals?	Hygroma/ neonatal mortality/ epididymitis/ orchitis	8	2.40
	Abortion/ still birth/ weak calves	114	34.23
	Infertility	54	16.22
	Reduced milk production	23	6.91
	All of above	34	10.21
	Don't know	100	30.03

Which livestock species do you think can be infected with Brucellosis?	Cattle	100	30.03
	Sheep and Goat	28	8.41
	Camel	6	1.80
	Swine	6	1.80
	All of above	120	36.04
	None of above	26	7.81
	Don't know	47	14.11
What are the common symptoms of Brucellosis in humans?	Undulant fever in; night sweats with peculiar odor, chills and weakness	28	8.41
	Malaise, insomnia, anorexia, headache, arthralgia, constipation, sexual impotence, nervousness and depression	44	13.21
	Encephalitis, meningitis, spondylitis, arthritis, endocarditis, orchitis, and prostatitis	29	8.71
	Spontaneous abortion	28	8.41
	All of above	10	3.00
	Don't know	194	58.26
How Brucellosis can be prevented in animals?	Proper vaccination	115	34.53
	Isolating infected animals	85	25.53
	Minimizing risk factors	33	9.91
	All of above	33	9.91
	Don't know	67	20.12
Can Brucellosis be diagnosed in animals?	Yes	185	55.55
	No	76	22.82
	Don't know	72	21.62
Commonly used sample for diagnostic test of animal brucellosis	Blood	20	6.01
	Urine	18	5.40
	Milk	69	20.72
	All of above	20	6.01
	a and c	33	9.91
	Don't know	173	51.95
Which diagnostic test of brucellosis have you heard about?	Rose Bengal Plate Test (RBPT)	35	10.51
	Brucella ring test	10	3.00
	ELISA	9	2.70
	PCR	12	3.60
	Buffered brucella antigen test (BBAT)	11	3.30
	Other	0	0
	All of above	24	7.21
	None of above	232	69.67
How do you identify Brucellosis infected animal?	By decrease milk production	16	4.80
	By general weakness	60	18.02
	By history of successive abortions	94	28.23
	All of above	35	10.51
	None of above	25	7.51
	Don't know	40	12.01
	By loss of appetite	63	18.92
Have you seen any Brucellosis infected animal in your area?	Yes	237	71.17
	No	96	28.83

Do you know that brucellosis can be transmitted through blood transfusion?	Yes	93	27.93
	No	240	72.07
Have you ever heard of any scheme, initiative or plan to control Brucellosis?	Yes	58	17.42
	No	275	82.58

Attitude towards brucellosis

The attitude of the participants was relatively positive towards brucellosis and 77.18% of the participants got informed when animals get sick. Most (51.65%) of the participant knew brucellosis is serious animal disease but 7.51% of the respondents did not consider it a threat for humans and 21.02% had no perception about the disease. 66.07% of the participants vaccinate their animals and 74.77% of participants have responded that they will go to doctor or clinic if they have symptoms of brucellosis. Majority (84.98%) of the participants did not believe that last trimester abortion in cattle and buffalo is caused by if an aborted woman is in proximity of the herd and majority (51.35%) of them identified any type of abortion in cattle and buffalo as bovine brucellosis. About 40.24% of the participants believed that brucellosis is preventable in animals by vaccination and 18.02% said it can be prevented by contacting veterinary office. 58.26% of the participants thought brucellosis in the suspected humans can be treated by visiting health facilities (Table 4).

Table 4: Attitude of participants towards Brucellosis

Variable	Characteristics	N	Percentage (%)
Did you get informed when animal get sick?	Yes	257	77.18
	No	17	5.10
	Sometimes Yes sometimes No	59	17.72
What is your perception about brucellosis?	Serious animal disease	172	51.65
	Serious human disease	25	7.51
	Both of above	66	19.82
	None of above	70	21.02
Do you vaccinate your animals?	Yes	220	66.07
	No	33	9.91
	Not applicable	80	24.02
Have you attended any training, awareness session or workshop related to livestock Brucellosis?	Yes	37	11.11
	No	296	88.89
Will you support any initiative taken to control Brucellosis?	Yes	312	93.69
	No	21	6.31
Do you think that the disease like brucellosis effect the production of livestock?	Yes	264	79.28
	No	69	20.72
Which measures have you taken or will you take to treat an animal infected with brucellosis?	Seek help from animal health professional	103	30.93
	Vaccination / Treatment	144	43.24
	Isolate the infected animal	41	12.31
	By traditional methods	41	12.31
	Will do nothing	4	1.20
Whom have you sold your animals most regularly?	Local market	93	27.93
	Slaughter house	18	5.40
	General community	146	43.84
	Don't sell animal	13	3.90
	Not applicable	63	18.92

Attitude towards Aborting animals	Sell the Animal	107	32.13
	Inform veterinary officer	103	30.93
	Isolate the animal	48	14.41
	Slaughter the animal	39	11.71
	None of above	36	10.81
Have you ever treat the aborting animal with antibodies?	Yes	122	36.64
	No	211	63.36
How health is ensured when buying or receiving new cattle?	Seek veterinary advice	35	10.51
	Rely on own experience	145	43.54
	Buy from known and/or trusted people	128	38.44
	None of above	13	3.90
	Not applicable	12	3.60
What will you do if a person at your livestock facility has symptoms generally associated with brucellosis?	Go to the doctor or clinic	249	74.77
	Stay home and self-medicate	38	11.41
	Go to the traditional healer	35	10.51
	None of above	11	3.30
Seek Brucellosis treatment from shrines	Yes	96	28.83
	No	237	71.17
Do you believe last trimester abortion in cattle and buffalo is caused by if an aborted woman is in proximity of the herd?	Yes	50	15.02
	No	283	84.98
Do you identify any type of abortion in cattle and buffalo as bovine brucellosis?	Yes	171	51.35
	No	162	48.65
What are your preferred methods of communication to get information?	Day meeting with veterinary services	38	11.41
	Community meeting with veterinary services	127	38.14
	Information pamphlet	41	12.31
	Radio and/or television	46	13.81
	Talks with friends, family and relatives	71	21.32
	None of above	10	3.00
Attitude towards Brucellosis prevention in Animals	Brucellosis can be prevented in animals	49	14.71
	Prevention by vaccination	134	40.24
	Prevention by contacting veterinary office	60	18.02
	Prevention by Isolation of Sick and Aborting animals	36	10.81
	Don’t know	54	16.22
Attitude towards suspected human Brucellosis	Brucellosis can be cured in humans	31	9.31
	Seek prayers	29	8.71
	Visit health facility	194	58.26
	Consuming herbal medicines	25	7.51
	Visit Local Chemist and Purchase Medicine	22	6.61
	Don’t know	32	9.60

Practices regarding brucellosis

Out of 333 participants, 43.84% participants used traditional methods to treat their diseased animals and 30.39% sought help from animal health provider. Majority (60.06%) of the participants responded that they did not ever visit to animal clinic or took any animal-to-animal clinic and 78.68% of the respondents had never attended any training on handling livestock. 58.26% participants responded that they separated the sick animals from healthy ones. 39.64% participants used glove while disposing aborted calves and other discharges and 36.64% had never used gloves while handling them. Only 33.63% participants responded that they disinfected space after parturition and 44.14% responded they did not practice it. Majority (63.06%) of the respondents washed their hands before and after milking and mostly (73.27%) used to clean feeding and water troughs. About 33.93% of the respondents did not store dung piles while 18.02% stored it for three to six months (Table 5).

Table 5: Practices of participants regarding Brucellosis

Variable	Characteristics	N	Percentage (%)
Ever visit to animal clinic or took any animal-to-animal clinic?	Yes	133	39.94
	No	200	60.06
How do you treat any diseased animal?	Do not treat	7	2.10
	Use traditional methods	146	43.84
	Seek help from animal health provider	103	30.93
	Take animal to animal clinic	77	23.12
Have you attended any particular training of handling livestock?	Yes	71	21.32
	No	262	78.68
Do you separate the sick animals from healthy?	Yes	194	58.26
	No	139	41.74
Do you use gloves while disposing aborted calves or other discharges?	Yes	132	39.64
	No	122	36.64
	Not applicable	79	23.72
Do you disinfect the space after parturition?	Yes	112	33.63
	No	147	44.14
	Not applicable	74	22.22
Do you wash your hands before and after milking?	Yes	210	63.06
	No	11	3.30
	Not applicable	112	33.63
How long do you store dung piles?	1-3 months	27	8.11
	3-6 months	60	18.02
	More than 6 months	49	14.71
	Don’t store	113	33.93
	Not applicable	84	25.23
Do you slaughter animals at your livestock facility?	Yes	146	43.84
	No	173	51.95
	Not applicable	14	4.20
Cleaning of feeding and water troughs	Yes	244	73.27
	No	8	2.40
	Not applicable	81	24.32

Risk factors associated with brucellosis

Regarding the risk factors associated with brucellosis, 19.22% of participants considered husbandry practices, 23.12% said residence conditions and feeding and 13.21% responded geography as most important risk factors are all brucellosis. 40.84% of participants did not keep their newly purchased animals in quarantine for some time while 39.34% kept them in quarantine. Most (90.39%) of the participants boiled milk before consumption and 70.87% of the respondents used raw milk to make other dairy products (lasi, butter, ghee etc.,). 46.25% participants lend their male animals to other herds and majority (66.07%) kept mixed species of livestock animals. 55.85% participants did not have any specified delivery rooms at your livestock facility and 53.15% sent their animals

to common grazing areas. Most of the participants (60.96%) responded that they have shared spaced for calves and other animals. 67.87% of the participates responded that they did not use any kind of protective clothing while handling animals and 61.56% used to slaughter animals inside the slaughterhouse while 15.62% used to do home slaughtering methods (Table 6).

Table 6: Response of participants against risk factors associated with Brucellosis

Variable	Characteristics	N	Percentage (%)
Type of herd raised	Single breed	166	49.85
	Mixed breed	135	40.54
	Not applicable	82	24.62
Type of livestock breeding	Inbreeding	124	37.24
	Outbreeding	126	37.84
	Not applicable	83	24.92
Method of animal slaughtering	Home slaughtering	52	15.62
	Slaughtering in slaughter houses	205	61.56
	Do not Slaughter the animal	4	1.20
	Not applicable	72	21.62
What are the important Risk factors for Brucellosis in animals?	Climatic conditions	7	2.10
	Species	15	4.50
	Herd size	17	5.11
	Husbandry practices	64	19.22
	Geography	44	13.21
	Age and Sex	34	10.21
	Residence conditions and feeding	77	23.12
	All of above	9	2.70
Do you keep the newly purchased animal in quarantine for some time?	Yes	131	39.34
	No	136	40.84
	Not applicable	66	19.82
Do you boil milk before consumption?	Yes	301	90.39
	No	32	9.61
How do you keep your livestock animals?	Mixed	220	66.07
	Specie separated	27	8.11
	Age separated	1	0.30
	Sex separated	4	1.20
	Not applicable	81	24.32
Do you use any kind of protective clothing while handling animals?	Yes	79	23.72
	No	226	67.87
	Not applicable	28	8.41
Do you lend the male animals of your herd to other herds?	Yes	154	46.25
	No	98	29.43
	Not applicable	81	24.32
Do you use raw milk to make other dairy products (lasi, butter, ghee etc,)?	Yes	236	70.87
	No	16	4.80
	Not applicable	81	24.32
Do you have any specified delivery rooms at your livestock facility?	Yes	73	21.92
	No	186	55.86
	Not applicable	74	22.22
Do you send your animals to common grazing areas?	Yes	177	53.15
	No	75	22.52
	Not applicable	81	24.32

Do animals have access to stored dung piles?	Yes	54	16.22
	No	189	56.76
	Not applicable	90	27.02
Shared calving space with other animals	Yes	203	60.96
	No	50	15.02
	Not applicable	80	24.02

One Health

The response of the participants was positive towards the one health concept, 61.26% participants already knew about zoonosis and 46.25% of participants knew that brucellosis can be transmitted from animals to humans. 24.92% participants reported the route of transmission of brucellosis is through contact with infected animal, 17.12% responded by consuming infected dairy products and 20.12% had no idea about it. 63.66% were aware about the threats imposed by contaminated dairy products and 58.56% respondents said that they consult a veterinarian in case of an abortion 40.54% but 14.41% reported to dispose abortion material carelessly. Majority (34.53%) of the respondents said they dumped the aborted material anywhere and 18.62% buried them. The participants who reported to live in shared places with animals were 40.54% (Table 7).

Table 7: Response of participants towards One Health questions regarding Brucellosis

Variable	Characteristics	N	Percentage (%)
Any previous knowledge about zoonosis	Yes	204	61.26
	No	129	38.74
Can brucellosis be transmitted from animals to humans?	Yes	154	46.25
	No	119	35.73
	Don't know	60	18.02
How Brucellosis is transmitted?	Through contact with infected animal	83	24.92
	Consuming infected dairy products	57	17.12
	Both of above	54	16.22
	None of above	72	21.62
	Don't know	67	20.12
Do you know about the threats imposed by contaminated dairy products?	Yes	212	63.66
	No	121	36.34
In case of abortion	Consult a veterinarian	195	58.56
	Disposing abortion material carelessly	48	14.41
	Take abortion material to Remote area	47	14.11
	None of above	43	12.91
How did you dispose the aborted material?	Dump it anywhere	115	34.53
	Burning	9	2.70
	Burying	62	18.62
	Throw it in dung pile	57	17.12
	Throw it in canal water	18	5.41
	Other	12	3.60
	Not applicable	60	18.02
Do you live in shared places with animals?	Yes	135	40.54
	No	113	33.93
	Not applicable	85	25.53
Have you ever been subjected to blood donation?	Yes	108	32.43
	No	225	67.57
Have you ever received blood?	Yes	66	19.82
	No	267	80.18

Socio-demographic factors associated with knowledge, attitude, practices, risk factors and one health

Factors found to be significantly associated with knowledge include Tehsil, age group, qualification, occupation, residence, and farming type. A significant association of attitude with gender, ethnicity, qualification, and occupation were found. The factors significantly associated with practices, risk factors and one health also include tehsil, age group, gender, marital status, qualification, occupation (Table 8).

Table 8: Univariate analyses of Socio-demographic characteristics with Knowledge, Attitude, Practices, Risk Factors and One Health

Socio-demographic characteristic	Knowledge	Attitude	Practice	Risk factors	One Health
Location	$\chi^2 = 9037$	$\chi^2 = 32057$	$\chi^2 = 5998$	$\chi^2 = 18391$	$\chi^2 = 3561$
	df = 13	df = 23	df = 8	df = 10	df = 5
	P = 0.770	P = 0.099	P = 0.648	P = 0.049*	P = 0.614
Tehsil	$\chi^2 = 106048$	$\chi^2 = 232896$	$\chi^2 = 106907$	$\chi^2 = 165598$	$\chi^2 = 94342$
	df = 78	df = 138	df = 48	df = 60	df = 30
	P = 0.019*	P <0.001*	P <0.001*	P <0.001*	P < 0.001*
Age group	$\chi^2 = 126484$	$\chi^2 = 197452$	$\chi^2 = 94239$	$\chi^2 = 114945$	$\chi^2 = 47840$
	df = 91	df = 161	df = 56	df = 70	df = 35
	P = 0.008*	P = 0.027*	P = 0.001*	P < 0.001*	P = 0.073
Gender	$\chi^2 = 9252$	$\chi^2 = 53057$	$\chi^2 = 24643$	$\chi^2 = 33457$	$\chi^2 = 4313$
	df = 13	df = 23	df = 8	df = 10	df = 5
	P = 0.754	P <0.001*	P = 0.002*	P <0.001*	P = 0.505
Ethnicity	$\chi^2 = 79300$	$\chi^2 = 144792$	$\chi^2 = 37593$	$\chi^2 = 99395$	$\chi^2 = 34014$
	df = 65	df = 115	df = 40	df = 50	df = 25
	P = 0.109	P = 0.031*	P = 0.579	P <0.001*	P = 0.108
Marital status	$\chi^2 = 13000$	$\chi^2 = 33760$	$\chi^2 = 19944$	$\chi^2 = 17406$	$\chi^2 = 8630$
	df = 13	df = 23	df = 8	df = 10	df = 5
	P = 0.448	P = 0.069	P = 0.011*	P = 0.066	P = 0.125
Qualification	$\chi^2 = 165396$	$\chi^2 = 221181$	$\chi^2 = 171985$	$\chi^2 = 156505$	$\chi^2 = 69625$
	df = 78	df = 138	df = 48	df = 60	df = 30
	P <0.001*	P <0.001*	P <0.001*	P <0.001*	P <0.001*
Occupation	$\chi^2 = 248970$	$\chi^2 = 390737$	$\chi^2 = 263790$	$\chi^2 = 569276$	$\chi^2 = 104005$
	df = 91	df = 161	df = 56	df = 70	df = 35
	P <0.001*	P <0.001*	P <0.001*	P <0.001*	P <0.001*
Residence	$\chi^2 = 26902$	$\chi^2 = 41634$	$\chi^2 = 33532$	$\chi^2 = 35281$	$\chi^2 = 19896$
	df = 13	df = 23	df = 8	df = 10	df = 5
	P = 0.013*	P = 0.010*	P <0.001*	P <0.001*	P <0.001*
Farming type	$\chi^2 = 57830$	$\chi^2 = 166289$	$\chi^2 = 88292$	$\chi^2 = 335340$	$\chi^2 = 69331$
	df = 26	df = 46	df = 16	df = 20	df = 10
	P <0.001*	P <0.001*	P <0.001*	P <0.001*	P <0.001*

*Significant association

Correlation between knowledge, attitude, practices, and risk factors

A significant positive correlation was observed between knowledge and attitude ($r_s = 0.646$, $P < 0.001$). A weak correlation was found between knowledge and practices ($r_s = 0.424$, $P < 0.001$). The correlation between knowledge and risk factors ($r_s = 0.234$, $P < 0.001$) was very weak. Positive correlation was observed between attitude and practices ($r_s = 0.553$, $P < 0.001$). Weak correlation was observed between attitude and risk factors ($r_s = 0.332$, $P < 0.001$). The correlation between practices and risk factors was weak ($r_s = 0.420$, $P < 0.001$) Table 8.

DISCUSSION

The present study was conducted to measure the

level of KAPS among the livestock holders and people directly involved with the livestock of district Rawalpindi and Islamabad. Previously a similar study was carried out, in five districts of Punjab and two district of Sindh provinces, Pakistan, where the levels of KAPS were measured in smallholder dairy farmers (Arif et al., 2017). In the present study, participants were farmers who had household livestock, dairy farm owners, slaughterhouse owners, veterinarians, butchers and general animal health care providers. A majority of participants (74.17%) knew about animal brucellosis. The similar results were reported by previous study where 70% of participants had heard about animal brucellosis (Arif et al., 2017). According to the present study 69.97% of the participants were able to identify the symptoms of animal brucellosis. A previous study from Khyber Pakhtunkhwa (KPK) also reported that only 6.25% participants knew about the symptoms of the disease (Khan et al., 2020). The difference in results might be due differences in the level of education and awareness of the people in two areas.

In the present study only 46.25% participants knew that they can get the disease from animals as reported in the previous study which reported that 23% of the participants were aware of getting the disease from animals whereas only 5% participants showed the knowledge about the transmission of disease from animals to humans in KPK (Khan et al., 2020; Arif et al., 2017). The present study also reported that 46.25% of participants showed the knowledge about the transmission of disease from animals to humans while previous study reported this percentage to be 3% (Arif et al., 2017). The reason for this increased percentage of knowledge might be due that the population of Sindh has low levels of education as compared to the Punjab. Additionally, our study also includes the veterinarians which have shown excellent knowledge about the disease which were not included in the previous study. A slight difference has been observed in the practice of disposing aborted material properly where in present study only 18.62% participants disposed the aborted material properly by burying it opposite to the previous study which reported 24% (Arif et al., 2017).

About 90.39% of the participants did not consume the raw milk but a majority (70.87%) used the raw milk for making other dairy products. The similar findings were reported previously where 60% of farmers and their families used raw dairy products (Arif et al., 2017). 40.54% of the participants claimed to live in shared places with animal as compared to 74% reported previously (Arif et al., 2017). In the current era of urbanization, the population has separated their living space from livestock. For questions regarding One Health, 61.26% of the participants knew about zoonosis as compared to the number (25%) reported previously from KPK (Khan et al., 2020).

In univariable analysis significant difference was observed in practices across tehsils ($P < 0.001$) with similar findings were reported by previous study where herd management practices were found to be significantly different among districts (Arif et al., 2017).In our study veterinarians showed the good knowledge about brucellosis similar to the study conducted in KPK, where they reported sufficient knowledge about brucellosis in human and animal health care professionals (Khan et al., 2020).

Although a considerable progress has been made in gaining the knowledge about the brucellosis and improved practices are being adopted by the livestock owners, the disease is still endemic in Pakistan. In Pakistan, some of the major challenges to control brucellosis includes the absence of regular surveillance system for the zoonotic infections as the investigations are event based, data is not properly shared between agriculture, livestock, public health and the environment departments. The absence of diagnostic systems at primary and secondary level and lack of awareness in health care professionals often leading to misdiagnosis are the major contributing factors of disease endemicity in Pakistan (Iqbal et al., 2020).

CONCLUSION

This study evaluated the knowledge, attitude, and practices of the livestock owners in district Rawalpindi and Islamabad, Pakistan. We have also analyzed the risk factors practiced by livestock owners and One Health aspect regarding brucellosis. Our study concluded that majority of household farmers are unaware of routes of transmission and major risk factors of brucellosis. Almost all the participants except veterinarians including butchers, dairy farm owners, slaughterhouse owners, farmers and co-workers were involved in at least one risky practice. We suggest that the awareness programs should be conducted to educate the people at high risk along with the improvement of vaccination programs for animals and strict implementation of brucellosis eradication policy which should be devised by government.

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CONFLICT OF INTEREST

The authors declare that they have no conflict of interest regarding this paper.

ETHICAL APPROVAL

Approval for this research was granted by the Ethics Review Board, Department of Biosciences, COMSATS University Islamabad under reference number CUI/BIO/ERB/21/07.

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