Prevalence of Bovine and Human Brucellosis in Pakistan–A Review

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Abstract | Brucellosis is one of the neglected widespread zoonoses around the globe affecting animals as well as humans. The purpose of the current review is to illustrate a report on the seroprevalence of bovine and human Brucellosis and associated risk factors in Pakistan. A total of 29 articles on Brucellosis from 2000 to May 2020 were selected and reviewed for the estimation of Brucella incidence in cattle, buffaloes and humans of Pakistan. The current study is based on the research articles in which Brucella prevalence is primarily determined through the following serological tests: Rose Bengal plate test (RBPT), milk ring test (MRT), serum agglutination test (SAT) and enzyme-linked immunosorbent assay (ELISA). Due to diversity in sensitivity and specificity of applied serological tests, it was found a vast divergence in the prevalence of Brucellosis. The seroprevalence of bovine Brucellosis is also varied due to geographical and environmental conditions, different breeds, sexes, and the difference in ages as within cattle varied from 2.05% to 28.90%, and found in buffaloes ranged from 1.6% to 21.73%. The Brucellosis in humans was reported in livestock workers, butchers, veterinary professionals, and those people who have direct contact with animals or using raw milk and Brucellosis has been found in them from 14% to 17.74%. The published reports portray that Brucellosis does affect both humans and animals in Pakistan. A proper surveillance system, effective vaccination plan and execution of segregation and culling strategy regarding Brucella infected animals, are desperately needed for the eradication of Brucellosis in Pakistan.

Keywords | Seroprevalence, Bovine, Human, Brucellosis, Pakistan

INTRODUCTION

Brucellosis is one of the important and widespread zoonoses around the globe except in some counties of high income (Poester et al., 2002; Pappas et al., 2006; OIE, 2009). Brucellosis is the most neglected and unrecognized infection not only in the world but also in Pakistan (Rubah et al., 2013; WHO, 2012). The disease is caused by a gram-negative bacteria Brucella that has so far twelve identified species (Rajala, 2016). Mostly Brucella species can cause disease in multiple species of animals including humans (Godfroid et al., 2010). In bovines, Brucella abortus is predominantly responsible for Brucellosis, while less frequently caused by Brucella melitensis and occasionally by Brucella suis (OIE, 2016). It is an important infection of sexually matured animals that can cause significant reproductive losses, in-terms of last trimester abortion, infertility, stillbirth, orchitis, placentitis and also causes possible excretion of the organism in uterine discharges and milk (Forbes & Tessaro, 1996; Wadood et al., 2009; England et al., 2004; Corbel et al., 2006; Islam et al., 2013). Infected animals mostly abort once but remain infected throughout their entire life (Godfroid et al., 2010). Brucellosis is not only one of the most important diseases in prospective of animals and human health but also it has socio-economic importance in rural areas due to dependence on livestock and dairy products for income generation and livestock is considered as a lifeline for the 95% rural population of
the developing countries (Maadi et al., 2011; Wadood et al., 2009; Hoffmann, 1999). Bovine Brucellosis is also a disease of yak, buffalo, and bison that causes similar signs and symptoms to those seen in cattle (OIE, 2009).

Worldwide, twelve *Brucella* species have been recognized but have two important members, *Brucella melitensis* and *Brucella abortus* that affect mainly sheep, goats, and bovines (cattle & buffalo) respectively. Furthermore, there are other ten species of *Brucella* including *B. suis*, *B. canis*, *B. ovis*, *B. neonatoc*, *B. microti*, *B. pinnipedia*, *B. ceti*, *B. papione- tis*, *B. vulpis*, and *B. inopinata* (Corbel, 1998; Whatmore, 2009). Transmission of the disease occurs in the herd mostly due to the ingestion of bacteria from the aborted fetuses, placenta, and discharges of cattle that contaminate pauster or water. (Park et al., 2005).

Pakistan is a country with more than 207.8 million people with one of the world’s fourth-leading milk-producing countries that produce yearly 54,328 thousand tons of milk (Pasha & Hafiz, 2017; ESOP, 2014, Iqbal & Ashraf, 2017). Pakistan has the sixth-largest livestock-based economy in the world with an animal population of 186.2 million. Furthermore, about 30 million people or 8 million families have their earnings from the livestock sector and the majority of them live in rural areas of Pakistan. Livestock sector productions have a major contribution of approximately 60 % of agriculture and 11.2 % of Pakistan gross domestic product (GDP), (Jan & Akram, 2018; GoP, 2019; Chandio et al., 2017). In Pakistan, cattle and buffaloes have cultural, dietary, and economic impacts that accomplished their worth in our rural society but the spread of infectious diseases especially Brucellosis is an emerging hazard for the livestock of Pakistan. The main causes of *Brucella* spreading are improper surveillance, under-reporting, lack of knowledge, the informal markets of raw milk, and the absence of proper legislation about the *Brucella* prevalent animals. The current review, therefore, focuses on Pakistan, where bovine Brucellosis is common but still under-researched and reported. The objectives of the current review are to summarize the prevalence and zoonotic importance of Brucellosis in Pakistan. This study also provides relevant information and conceptual ideas that will help in controlling and eradication of Brucellosis in Pakistan.

**MATERIAL AND METHODS**

A comprehensive search was made to identify the relevant studies and data published from 2000 to May, 2020 by using major databases of Google Scholar, PubMed and Google search engine. The keywords used were Seroprevalence, bovine, human Brucellosis, risk factors, and suitable combinations of the above to make sure the search for relevant studies.

**INCLUSION AND EXCLUSION CRITERIA**

The basic criteria for the selection of articles was based on the following serological tests: Rose Bengal plate test (RBPT), milk ring test (MRT), serum agglutination test (SAT) and enzyme-linked immunosorbent assay (ELISA). The paper published from January 2000 to May 2020 that are available on Google scholar, PubMed and Google search engine were included in the current review study. The peer reviewed articles on human and bovine Brucellosis that were published in the English language were also considered in the current study. The articles published other than English language, Conference Papers and those without peer reviewed were excluded from the study.

**CHARACTERISTICS OF THE STUDY**

Initially, a total number of 1250 titles of published articles were read and 146 articles were selected for the further consideration. After removing duplicates the abstracts of 119 articles were selected for screening. The abstracts of 72 papers after screening were excluded and the remaining 47 articles were selected for the full paper review. After full screening of the articles another 18 papers were excluded that did not match our inclusion criteria. The remaining 29 articles were included in the study for quality data analysis. In the selected articles three (3) studies were on cattle, buffaloes, and humans, ten (10) on buffaloes and cattle, while Seven (7) Five (5) and four(4) studies were conducted only on cattle buffaloes and human respectively as shown in Figure-1, Table-1.

![Figure 1: Review process adapted for the prevalence of bovine and human Brucellosis in Pakistan.](image-url)

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Compact and precise sampling design is considered basic for any epidemiological study, in the current review, it is found that many studies have no proper criteria for animal selection and applied non-randomization techniques for their studies or surveys. The exact incidence of Brucellosis is still unknown in Pakistan but it varies due to the species’ sex, age, geographical and climate conditions. Moreover, its spread is mainly influenced by the husbandry system as well as environmental conditions (Gul and Khan, 2007; WHO, 1997).

Punjab is the second largest province in terms of land size area and the largest human and animal populated province of Pakistan (Ghori et al., 2017; PBTI). In the current review, a total of fourteen (14) studies were included in cattle and buffaloes of Punjab province and found an overall average frequency of Brucellosis of 8% and 7.26% in cattle and buffaloes respectively. (Table-1). In cattle, the seroprevalence based on 14 different studies were found between the range minimum of 2.05% to a maximum of 28.90% (Nawaz et al., 2016; Ullah et al., 2019) (Table 1). Two studies were conducted on cattle at the Pothohar region, northern salt range areas of province Punjab, and found 3.60% lower Brucellosis incidence through MRT but a similar type of study was performed in the same region and found 5.01% seroprevalence in cattle. (Ali et al., 2014; Shahzad et al., 2015). Studies of cattle at government farms, private farms and in different villages portray a huge Brucellosis incidence variation; a survey study was executed at properly managed government livestock dairy production research institute, Bahadar Nagar, district Okara and reported 14.11% Brucellosis in cattle (Chaudhry, 2000) but later on another study was conducted at all government livestock dairy farms which revealed a low incidence of 2.15% of Brucellosis in cattle (Jamil et al., 2020).

At government livestock farms, better husbandry techniques were practised than that of private farms. In comparative surveillance conducted at private and government farms reported overall 15.40% prevalence in cattle while comparatively a higher frequency of Brucellosis is found at private than that of government livestock farms in cattle (Nasir et al., 2004). Hussain et al. (2008) described in their study the seroprevalence of cattle at different villages and livestock farms and reported a 10.18% seroprevalence of Brucellosis in cattle.

A study on large scale in thirty-six districts of Punjab was performed in which 59665 serum samples were collected from the bovine and found a prevalence of 2.05% in cattle (Nawaz et al., 2016). Later on similar studies were conducted in northern Punjab Rawalpindi area by Ahmad et al. (2017) and southern belt district Rejanpur by Ismail et al. (2018) to determine the seroprevalence of Brucellosis in cattle and it was found 8.1% high prevalence rate in the northern areas Rawalpindi region as compared to 4.4% in cattle at Rajanpur. Meanwhile it was found a low prevalence of 3.8% in cattle in the eastern districts of Punjab Okara, Faisalabad, Kasur and Lahore (Saeed et al., 2019). A study was designed for the diagnosis of Brucella prevalence in cattle and buffaloes at desert Thal area of district Bhakkar and found a prevalence of 11.9% in cattle respectively (Shehzad et al., 2020).

In buffaloes frequency of brucellosis is based on 16 articles in which a high prevalence of 21.73 % was found in districts: Lahore, kushab, Bhakkar, khanewal, Gujranwala of Punjab province while a low prevalence of 1.6% was reported in northern Punjab district Rawalpindi and nearby areas of Islamabad (Nasir et al., 2004; Ahmad et al., 2017). Different studies on seroprevalence of Brucellosis in buffaloes revealed variation in results likewise at government livestock dairy production research institute, Bahadar Nagar, district Okara, at private livestock farms and in different villages, found 11.17%, 21.73% and 9.38% respectively (Chaudhry, 2000; Nasir et al., 2004; Hussain et al., 2008). A comprehensive and large scale study was conducted in all 36 districts of Punjab and found an overall very low prevalence of 7.86% of Brucellosis in buffaloes as compared to all previous studies (Nawaz et al., 2016).

In the northern Punjab Potohar area, the seroprevalence of Brucellosis in buffaloes was reported quite low at 1.6% as compared to 5.2% in south Punjab district Rajanpur (Ahmad et al., 2017; Ismail et al., 2018).

Balochistan is area wise the largest province of Pakistan in terms of land size but 4.9% of the human population lives there. Livestock is a very important source of food and income generation for the rural population of Balochistan province and the livestock population of Balochistan is 20% of the total national population of Pakistan. The buffaloes are at very low numbers of 300000 heads but have a good cattle population of 2.3 million (Kakar et al., 2008). There were conducted two studies on cattle Brucellosis by using blood serum in Baluchistan province and found a maximum prevalence of 3.95% in Quetta area while slightly low 3.5% in the district Lorelai (Shafee et al., 2012; Khan et al., 2016). Only one study was executed on buffaloes by using raw milk for the diagnosis of Brucella incidence at the Quetta area and reported 1.7% Brucellosis (Shafee et al., 2011).

Sindh is the third largest province of Pakistan. The livestock population is an important component of the agriculture sector (Wasim, 2007). A serological study was carried out for the determination of Brucellosis reported a 25% rate of Brucella infection in cattle but another report
Effects of Brucellosis incidence in buffaloes showed(98,80),(408,113) a very low infection of 1.99% (Mangi et al., 2015; Yousaf et al., 2016).

*Brucella* infection causes abortion in both cattle and buffaloes and in a study executed at Hyderabad District of Sindh for the diagnosis of brucellosis in bovines having a history of abortion revealed a very high prevalence rate of Brucellosis of 31.88% and 47.19% in cattle and buffaloes respectively (Soomro et al., 2014).

Khyber Pakhtunkhwa (KPK) is the third largest province of Pakistan in terms of livestock and human population. According to the Livestock Census (2006), it has buffaloes of about 808,068 heads of lactating, 184,229 dry and 97,664 without calving and has cattle of about 605,9041 heads lactating, 605,9041 dry, and 605,9041 without calving. Very high seropositivity of 17.58% was found in cattle at government Livestock District Kohat but a low prevalence of 3.97% was reported in two districts, Banu and Laki Marwat. (Hamidullah et al., 2009; Bakhtullah et al., 2014) while a study of Brucellosis in buffaloes revealed an 11% incidence in the Swat valley of KPK (Khana et al., 2017).

Gilgit Baltistan (GB) is the fifth province of Pakistan with a very low bovine population. Only one study was executed for the determination of *Brucella* infection in cattle at Ghizer, Skardu and Gilgit districts of GB and found 10.93% Brucellosis (Saher et al., 2018) (Table-1).

In milk-based samples, MRT was used and reported a low prevalence of 1.7% in raw milk of buffalo at Quetta while maximum prevalence was found 47.19% in Hyderabad province Sindh. Similarly in cattle prevalence of Brucellosis in raw milk reported 31.88% at Hyderabad while a low prevalence of 2.25% was found in Lorelai Balochistan (Soomro et al., 2014; Khan et al., 2016).

RBPT test was applied in 21 out of 29 studies and was additionally reconfirmed through SAT in 5 studies and by ELISA in 12 studies respectively. The maximum seroprevalence of Brucellosis confirmed with SAT and ELISA was 17.58% and 27.86% respectively while the lowest prevalence 1.75% and 1.2% were found in cattle respectively (Khan et al., 2016; Ismail et al., 2018; Hamidullah et al., 2009; Ullah et al., 2019). Similarly, the higher seroprevalence of 9.51% and 13% were found while the lower of 7.64% and 1.21% by SAT and ELISA were found respectively in buffaloes (Nasir et al., 2004; Khana et al., 2017; Chaudhry, 2000; Shehzad et al., 2020).

**Effect of Age on Brucellosis**

Age is an important factor that affects the prevalence of Brucellosis. Chaudhry, (2000) reported that seroprevalence in adult cattle and buffalo of 2–10 years was found high as compared to the young age of under 2 years of cattle and buffalo. In a similar type of study, Nasir et al. (2014) recorded that Brucellosis is high in buffaloes of 3 years old or more. While Saeed et al. (2019) determined that adult or mature cattle and buffaloes are more prevalent for Brucellosis than young ones. Whereas studies executed by Bakhhtullah et al. (2014) and Mangi et al. (2015) showed cattle having an age group of more than four years is more prevalent for Brucellosis. Similarly, in a recent study regarding seroprevalence of Brucellosis, it has been declared that in mature, pregnant, and lactating cows incidence of *Brucella* is quite higher than in immature, dry, and non-lactating ones (Khan et al., 2020).

**Effect of Sex on Brucellosis**

Nasir et al. (2014) used RBPT and SAT for seroprevalence of Brucellosis in buffaloes and detected seroprevalence of Brucellosis quite higher in female buffaloes than in males, while another study conducted in Okara, Faisal Abad, Kasur, Lahore districts of Punjab, reported a difference in results to the previous ones that males were more prevalent than females (Saeed et al., 2019). Shahjee et al. (2012); Mangi et al. (2015) diagnosed seroprevalence of Brucellosis in cattle and buffaloes and stated a quite higher rate of seroprevalence in female than male.

**Effect of Species on Brucellosis**

Soomro et al. (2014); Nawaz et al. (2016); Saeed et al. (2019); Jamil et al. (2020) reported that Brucellosis seroprevalence is higher in buffaloes than in cattle while in a recent study by Shehzad et al. (2020) reported contrary that Brucellosis in cattle is higher than in buffaloes.

**Effect of Breeds on Brucellosis**

Shahjed et al. (2015); Mangi et al. (2015); Bakhhtullah et al. (2014); Khan et al. (2020) found in their studies that seroprevalence of Brucellosis is higher in exotic cattle and Friesian cross with local breeds than in local breeds of Pakistan, while Khan et al. (2016) reported differently with previous studies that *Brucella* is higher in the local breed in district Lorelai than crossbred cattle.

**Effect of Management Systems on Brucellosis**

Nasir et al. (2004) investigated the seroprevalence of Brucellosis on government and private livestock farms and reported a higher prevalence rate in private livestock farms due to poor management as compared to in government livestock farms having proper management. On the contrary Shafee et al. (2011) reported higher prevalence at government dairy farms in Quetta than in private farms. According to the research of Bakhhtullah et al. (2014), Brucellosis is more prevalent in free-grazing cattle in field areas as compared to stall feeders.
| Province / Region | District/Area | Prevalence % | Source | Species |
|-------------------|--------------|--------------|--------|---------|
| **Punjab** | Okara | 14.11 | 10.0 | - | - | Chaudhry, (2000) |
| Lahore, kushab, Bhakkar, khanewal, Gujranwala. | 15.40 | 7.50 | - | - | - | Nasir et al., 2004 |
| Dairy Farms, Villages of Punjab | 10.18 | - | - | 8 | Hussain et al., 2008 |
| Potohar plateau, Chakwal, Attok, Rawalpindi. | - | - | 3.60 | - | Ali et al., 2014 |
| Potohar region | 5.01 | - | - | - | Shahzad et al., 2015 |
| All 36 district of Punjab | 2.05 | - | - | - | Nawaz et al., 2016 |
| Rajanpur | 4.4 | - | - | 1.2 | Ismail et al., 2018 |
| **Capital/ Punjab** | Islamabad, Rawalpindi | 8.1 | - | - | 6.6 | Ahmad et al., 2017 |
| Okara, Faisalabad, Kasur, Lahore | 3.8 | - | - | - | Saeed et al., 2019 |
| L&DD Dairy Farms | 2.15 | - | - | 1.91 | Jamil et al., 2020 |
| Gujranwala Division | 28.90 | - | - | 27.86 | Ullah et al., 2019 |
| Faisalabad | 12.53 | - | - | 2.40 | Khanet et al., 2020 |
| Bhakkar | 11.9 | - | - | 2.29 | Shehzad et al., 2020 |
| District Okara | 11.17 | 7.64 | - | - | Chaudhry, (2000) |
| Lahore, kushab, Bhakkar, khanewal, Gujranwala | 21.73 | 9.51 | - | - | Nasir et al., 2004 |
| **Punjab** | Dairy Farms, Villages of Punjab | 9.38 | - | - | 6.92 | Hussain et al., 2008 |
| Potohar plateau, Chakwal, Attok, Rawalpindi. | - | - | 3.09 | - | Ali et al., 2014 |
| Dera Chal other private farms | 12.2 | - | - | - | Nasir et al., 2014 |
| All 36 district of Punjab | 7.86 | - | - | - | Nawaz et al., 2016 |
| Rajanpur | 5.2 | - | - | 2.0 | Ismail et al., 2018 |
| Okara, Faisalabad, Kasur, Lahore | 5.1 | - | - | - | Saeed et al., 2019 |
| L&DD Dairy Farms | 5.62 | - | - | 4.64 | Jamil et al., 2020 |
| Bhakkar | 7.2 | - | - | 1.21 | Shehzad et al., 2020 |
| **Capital/ Punjab** | Islamabad, Rawalpindi | 1.6 | - | - | 1.6 | Ahmad et al., 2017 |
Geographical and seasonal effects on distribution of Brucellosis

Nawaz et al. (2016) described a very higher Brucella prevalence in central districts of Punjab than in northern and southern Punjab while Saeed et al. (2019); Bakhtullah et al. (2014); recorded difference in seroprevalence of Brucellosis in different areas. Likewise areas, regions, and the seasons also affect the seroprevalence of Brucellosis as Yousaf et al. (2016) stated a higher rate of Brucellosis in winter than in summer season.

Associated risk factors of Brucellosis in bovines

Ismail et al. (2018) recorded that bovines (buffalo & cattle) with abortion history showed a high rate of seroprevalence of Brucellosis. Furthermore it was found a positive correlation among Brucella positive animals and reproductive disorders like repeat breeding, metritis, and retention of fetal membrane. Saeed et al. (2019) and Shehzad et al. (2020) determined a high seroprevalence rate of Brucellosis in animals having a history of abortion. In another research executed by Ullah et al. (2019) reported a significant association between reproductive disorders (abortion, stillbirth, repeat breeders, and retention of the fetal membrane) and seropositive cattle of Brucellosis.

Isolation and identification of Brucella in bovines

Ali et al. (2014) collected samples of milk, vaginal swab, and aborted fetuses from the cattle and buffaloes and subjected them to the culturing procedures. For identification of Brucella strains, the researchers used the biotyping and...
molecular method and confirmed the *B. abortus* biovar 1 incidence in collected samples. Most of the studies reported incidence of *Brucella abortus* in seropositive buffaloes and cattle by using real-time (RT) PCR (Ahmad et al., 2017; Jamil et al., 2020). Meanwhile, in another study, the researchers reported *Brucella melitensis* in all seropositive samples of cattle and buffaloes by using RT-PCR (Saeed et al., 2019).

**HUMAN BRUCELLOSIS**
The human frequency of Brucellosis was found in 7 out of 29 reviewed articles and all studies reveal occupational risks. There is no study that can explain the incidence of Brucellosis at the national level. The prevalence of Brucellosis was found 5.8% to 17.74% in different areas of the province Punjab, Pakistan (Ali et al., 2016; Shehzad et al., 2020). We have found no article used in the current study, that can reveal the human Brucellosis incidence in other provinces of Pakistan.

Hussain et al. (2008) carried out a study to investigate Brucellosis in persons in close contact with animals like villagers, livestock and slaughterhouse workers. In this study, the researchers recorded overall 14% *Brucella* prevalence in humans and also reported a high prevalence rate in males, village workers, and abattoir associated personnel. Another study reported that Brucellosis is more prevalent in females of rural areas than the in the females of the urban areas and also it was found that seropositive females have symptoms and history of intrauterine fetal death, abortion, and miscarriage (Ali et al., 2016). The frequency of *Brucella* infection was recorded at a high rate in males and females having their ages equal or more than forty (40) years with the history of animal contact (Saddique et al., 2019). In a similar type of research work by Ali et al. (2018) recorded that males are more seropositive than females and described a high rate of prevalence in the age group of 20–30 years and also displayed that people who were resident of rural areas, kept animals in their homes and were uneducated, had a high rate of prevalence. Moreover, the researchers also described some associated potential risk factors like milking, exposure to animals, and slaughtering.

A study was conducted for the diagnosis of Brucellosis in persons in contact with animals like slaughterhouse workers, animal keepers, loaders, veterinary professionals, meat sellers, cleaners, and drivers. In this study it was found overall 21.7% Brucellosis in them, while the important diagnosis of the study was that Brucellosis was found at high rate in animal keepers (37.5%), illiterate workers and persons having old age 50–60 years (Mukhtar et al., 2008). A recent study by Shehzad et al. (2020) described that Brucellosis not only caused a high rate of abortion in spousers of seropositive humans but also is a predisposing factor for many other diseases. In the study, the researchers found a high rate of Brucellosis in married personals and in those people who consumed the raw milk. RBPT was used as a primary diagnostic tool for human Brucellosis in 6 (85.71%) studies but only in one study the researcher applied ELISA as a confirmatory test (Table 1).

**ECONOMIC IMPACTS OF BRUCELLOSIS**
There is found no article regarding the economic impacts of Brucellosis in Pakistan.

**DISCUSSION**
The current review aimed to portray the trends of Brucellosis infection in cattle, buffaloes, and humans and explore the association between seropositivity and potential risks of Brucellosis in Pakistan. The reviewed serological data for the current study reveals strong evidence of Brucellosis spread in Pakistan not only in cattle and buffaloes but also in humans especially those remaining in close contact with animals. However, we found only 29 studies that are considered to have quality data useful for the estimation of the *Brucella* burden in Pakistan. The quantity and quality of research about the epidemiology of Brucellosis are very limited. There is no description found about future incidence estimation and economic losses of disease in published literature. Additionally in the current review found a very limited validity and scope of studies that create hindrance for obtaining a true seroprevalence of Brucellosis. Most of the articles about the prevalence have very limited and unclear diagnostic designs and also found nonrandomized sampling approaches or small sample size which depicts limited inauthentic seroprevalence. There is also found in some articles a lack of standardized techniques for the diagnosis of Brucellosis which leads to a misleading estimation of prevalence. For proper estimation and understanding of Brucellosis epidemiology, it is considered that the isolation of species and characterization of the prevalent biovars of *Brucella* are very essential, because all serological tests used for incidence are useful but have limited specification and sensitivity.

Seven studies revealed the incidence of Brucellosis in humans but there is no study that describes the isolation of *Brucella* species. So the accurate data about prevalence of *Brucella* species in humans is very necessary for the future control and eradication programs for Brucellosis.

In animals, the majority of studies about Brucellosis incidence is carried out with serological tests i.e. RBPT, SAT, MRT based and in RBPT and MRT, locally prepared antigens are used for diagnosis. But only 8 out of 18 studies described the manufacturers of antigens. ELISA has been used as a confirmatory test in 13 studies and has been
proved more sensitive, accurate and specific. No researcher described any Brucella vaccination history in his research about the diagnosis of Brucellosis through serological tests. This review found the highest seroprevalence of 25% and the minimum of 2.05% in cattle through the blood serum samples and similarly found a high rate of 21.73% and minimum of 1.6% in buffaloes (Mangi et al., 2015; Nawaz et al., 2016; Ahmad et al., 2017; Nasir et al., 2004). The seroprevalence of Brucellosis in bovine varied due to geographical and environmental conditions, different breeds, sexes, and the difference in ages as within cattle varied from 2.05% to 28.90%, 7.50% to 11.43%, 3.09% to 43.19%, and 1.91 to 27.86 through the serological tests RBPT, SAT, MRT, and ELISA respectively. Moreover, the prevalence was found in buffaloes 1.6% to 21.73%, 7.64% to 9.51%, 3.09 to 47.19%, and 1.21% to 6.92 through the serological tests RBPT, SAT, MRT, and ELISA respectively (Nawaz et al., 2016; Ullah et al., 2019; Nasir et al., 2004; Chaudhry, (2000); Ali et al., 2014; Jamil et al., 2020; Ahmad et al., 2017; Soomro et al., 2014; Shehzad et al., 2020; Hussain et al., 2008). There is found a huge variation in the results of Brucella prevalence. It may be due to geographical distribution, the difference in husbandry techniques and might be due to seasonal or environmental effects as some articles reported that the rate of prevalence varied area to area and also change under different environmental conditions (Nawaz et al., 2016; Bakhtullah et al., 2014; Yousaf et al., 2016). Similarly in the same region or area, a difference in the incidence of Brucellosis is found. Two studies conducted in pothohar, found variations in Brucella incidence in cattle; it might be due to the difference in applied diagnostic techniques (Ali et al., 2014; Shahzad et al., 2015).

Association between reproductive disorders and seropositivity of Brucellosis in bovines found in 4 out of 29 studies and also especially found a significant relationship among the abortion and seropositive animals (Ismail et al., 2018; Saeed et al., 2019; Shehzad et al., 2020; Ullah et al., 2019; Saddique et al., 2019; Mukhtar et al., 2008).

None of the studies out of the 29 reviewed articles revealed the incidence of Brucella species or biovars in humans and also can explain species based association between animal and human Brucellosis. Most of the studies were conducted about the incidence of Brucellosis in humans who are in close contact with animals or veterinary professionals (Hussain et al., 2008; Ali et al., 2016). But only one study found about the use of unpasteurized milk in seropositive humans and also threw light on the risks associated with human to human and reported miscarriages in the spouse of male seropositive for Brucellosis (Shehzad et al., 2020). There is a lack of a wide range and randomized seroprevalence study of Brucellosis in humans which can explain the true frequency of Brucellosis in humans in Pakistan.

CONCLUSION

In the current review of literature, it is summarized that bovine and human Brucellosis is highly prevalent in Pakistan but there is a need of a large scale systematic and randomized study program and approaches for the accurate and actual prevalence of Brucellosis. Moreover, studies are needed that can estimate the actual economic losses. The veterinary and public health authorities need to collaborate their efforts to develop a comprehensive and compact control program and implement it to minimize financial losses due to Brucellosis. It is found that there is no proper vaccination and culling program or stamping of seropositive animals. Therefore in the context of Pakistan, there is a need of the strengthening the surveillance and disease reporting systems and enhancing the diagnostic capabilities of laboratories in all districts. Awareness campaigns should be launched for the education of farmers regarding the hazards of Brucellosis.

AUTHOR'S CONTRIBUTION

The submitting author is responsible for ensuring that contributions of all authors are correct as authors, Aamir Shehzad, Wiwiek Tyasningsih and Saif ur Rehman have made their contributions in data search, selection and analysis of the quality of data, designing and drafting of the manuscript while the author Fedik Abdul Rantam critically reviewed the current study.

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