

## Diagnosis of Various Bacterial Zoonotic Diseases in Human and Bovine populations in Haryana State under the umbrella of One Health

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### Abstract

Zoonotic diseases, also known as zoonoses, are infectious diseases that can be transmitted from animals to humans or vice versa. These diseases are of significant concern to public health as they can cause severe illness, and in some cases, death, so early detection and diagnosis are crucial for effective treatment and prevention. The study was conducted from July 2022 to July 2023 to test suspected samples of both human and animal populations for various common zoonotic diseases at district surveillance laboratories and district veterinary surveillance laboratories, respectively, in Haryana state. Zoonotic diseases such as tuberculosis, paratuberculosis, salmonellosis (typhoid), brucellosis, leptospirosis, and scrub typhus were included in the present study. Out of 25,911 animal samples, 216 were positive for brucellosis as detected through ELISA. None of the samples were found positive for bovine tuberculosis and bovine paratuberculosis. Out of 13,693 suspected human samples, 2,048 were positive for human tuberculosis, and among 22,750 samples, 3884 were positive for salmonellosis. For scrub typhus, 15 out of 99 samples were positive, while 4 out of 70 were positive for leptospirosis. The diagnosis of zoonotic diseases is a complex process that requires a multidisciplinary approach. Early diagnosis and prompt treatment of these diseases are essential for ensuring optimal health outcomes and preventing the spread of these zoonotic diseases to humans and animals.

**Keywords:** Zoonotic disease, Humans, Bovines, Detection, One Health

### Introduction:

Zoonotic infections known for their capacity to transfer between animals and humans (or from humans to animals) pose a substantial worldwide public health issue (Jones et al., 2008). These diseases, originating from various pathogens like bacteria, viruses, parasites, and fungi, hold the potential to induce severe illnesses and fatalities among both human and animal communities (Taylor et al., 2001). The inter linkage between humans, animals, and environmental elements amplifies the possibility of transmitting zoonotic diseases (Hassell et al., 2017). Thus, diverse ways of transmission, infection routes, and clinical manifestations of zoonotic infections pose challenges to healthcare systems and veterinary practices worldwide (Behera et al., 2021). Therefore, increased attention, monitoring, and inter-sectoral efforts are of utmost importance to implement successful measures for the prevention and control of zoonotic diseases.

Among the many zoonotic diseases, tuberculosis (TB) caused by *Mycobacterium tuberculosis*, remains a significant public health concern worldwide, affecting both humans and animals (Anaelom et al., 2010). Paratuberculosis, caused by *Mycobacterium avium* subspecies paratuberculosis, is another zoonotic disease

primarily affecting cattle, sheep, and goats, with potential transmission to humans (Whittington et al., 2005). Salmonellosis, predominantly known for causing typhoid fever and paratyphoid, represent another significant zoonotic infection with various serovars capable of infecting both humans and animals (Majowicz et al., 2010). These pathogens pose risks of infections ranging from gastroenteritis to severe systemic diseases in humans. Brucellosis, caused by *Brucella* species, is a major zoonotic infection posing significant health and economic burdens globally (Seleem et al., 2010). Leptospirosis, caused by spirochetes of the genus *Leptospira*, is another notable zoonosis with a worldwide distribution and potential transmission between animals and humans (Costa et al., 2015). Moreover, scrub typhus, caused by *Orientia tsutsugamushi*, represents a critical zoonosis transmitted by chiggers, posing significant health risks. The complexity and interdependency of zoonotic diseases necessitate comprehensive understanding, effective surveillance, and control measures involving collaboration among public health agencies, veterinarians, and healthcare providers (Taylor et al., 2001; Morse et al., 2012).

To strengthen the surveillance system of zoonotic diseases in Haryana state, ICAR-National Research

Centre on Equines, Hisar was designated as regional coordinator for National One Health Programme for Prevention and Control of Zoonoses (NOHP-PCZ) scheme under NCDC, New Delhi. In our previous attempt for prioritization of zoonotic diseases in Haryana (India) using one health approach, it was observed that bovine tuberculosis and brucellosis ranked 2<sup>nd</sup> and 6<sup>th</sup> in priority list, respectively (Thukral et al., 2023). The objective of the present study was to gather data from both medical and veterinary fields to provide a holistic view of prevalence of bacterial zoonotic diseases in humans and animals and so as to improve surveillance capabilities, enabling early detection and response to disease outbreaks.

## Materials and Methods:

**Study area and period:** Haryana is a state located in the north-western parts of India with 22 districts, about 6848 villages and 154 towns. The state of Haryana is situated between 27.39°N and 30.35°N latitude and 74.27°E and 77.36°E longitude (Shaloo and Kaur, 2016). The present study was conducted from July 2022 to July 2023.

**Targeted zoonotic diseases:** The study was focused on targeted zoonotic diseases such as brucellosis, bovine tuberculosis, paratuberculosis, salmonellosis (typhoid), leptospirosis, and scrub typhus.

**Testing of livestock species for important zoonotic diseases:** The samples were collected from suspected animals at the government veterinary hospitals and were sent to disease diagnostic labs for testing. Samples like faeces, blood, urine, and swabs were collected from suspected animals (irrespective of sex and age) and were transported to the laboratory for further testing. The samples were also collected by the Indian Council of Agricultural Research-National Research Centre on Equines (ICAR-NRCE), Hisar as routine surveillance and tested at NRCE. Tuberculin test, RBPT and ELISA were used for the diagnosis of tuberculosis and brucella, respectively. The test kits like Anigen Rapid Bovine TB Ab Test Kit (Maxanim, USA), RBPT antigen (Innovative Diagnostics, France), Bovine Tuberculin PPD 3000/Avian Tuberculin PPD 2500 kit (Applied Biosystems/ BOVIGAM), Brucella Antibody ELISA Kit (Ringbio) were used in this study. All positive controls were supplied with the kits.

**Testing of Humans for important zoonotic diseases:** Human samples were collected from suspected cases showing symptoms of diseases at government civil hospitals and were sent to district disease diagnostic labs for testing. Widal test and sputum microscopy/CB-NAAT (Widal Test kit by Anamol Lab Pvt. Ltd, Maharashtra) were employed to diagnose salmonellosis and tuberculosis respectively, while IgM ELISA was used to

diagnose both scrub typhus [Human IgM (Sandwich ELISA) ELISA Kit - LS-F10546, LS Bio] and leptospirosis [Human IgM (Competitive EIA) ELISA Kit - LS-F27712, LS Bio].

**Compilation of reports and analysis:** Data regarding abovementioned zoonotic diseases was collected monthly from district surveillance laboratories and district veterinary surveillance laboratories. Further, this data was compiled in Microsoft Excel and analyzed at ICAR-NRCE. Maps were prepared using QGIS version 3.34.1.

## Results and Discussion:

Among 25,911 animal samples, 216 were positive for brucellosis (Table 1). The highest positivity rate (7.25%) was observed in Karnal district followed by Panchkula (2.44%), Kurukshetra (1.89%), Sonapat (1.30%), and Kaithal (1.09%). Other districts like Hisar, Yamuna Nagar, Panipat, and Fatehabad showed less than a 1% positivity rate (Table 1, Figure 1). Previous serological studies showed 12- 29% prevalence rate of bovine brucellosis in Haryana (Chand and Sharma, 2004; Kumar and Chand, 2011; Khurana et al., 2012; Chand and Chhabra, 2013) as Haryana is known as dairy state and promote the dairy farming venture. Surprisingly, none of the 115 faecal samples/animals tested for bovine tuberculosis, and 154 faecal samples/animals for bovine paratuberculosis were found positive. The actual extent of the illness remains unclear as routine surveillance data is absent in many developing nations including India. Despite India's progress in achieving the End TB goal, bovine tuberculosis (bTB) remains mostly concealed (Refaya et al., 2020).

Among the human samples, 2,048 out of 13,693 suspected samples were positive for human tuberculosis (Table 1). For human tuberculosis positivity rate in various districts were 59.90% in Nuh-Mewat, 25.9% in Palwal, 18.44% in Kurukshetra, 14.92% in Sirsa, 10.09% in Rohtak, 13.83% in Mahendergarh, 9.87% in Kaithal and 7.34% in Rohtak (Table 1, Figure 2). Many studies were conducted in Haryana and other part of India to diagnosed its positivity rate among household contacts of newly diagnosed tuberculosis patients as an efficient tool for early diagnosis and treatment of active TB, thus minimizing the severity and decreasing transmission (Chawla et al., 2020; Gupta et al., 2016; Nair et al., 2016; Khaparde et al., 2015). Identifying active TB cases allows for targeted public health interventions such as contact tracing, where individuals who have been in close contact with an infected person can be screened and treated, if necessary, for preventing the spread of the disease.

For salmonellosis in human population, 3,884 out of 22,750 were positive for salmonellosis (Table 1). The highest positivity rate (21.88%) were observed in Hisar,

followed by Kurukshetra (17.94%), Kaithal (17.31%), Palwal (15.63%), Rohtak (14.05%), Rewari (12.71%), Mewat (10.43%), Faridabad (8.62%) and Mahendergarh (5.96%) (Table 1, Figure 2). The Widal slide test, an agglutination-based assay, serves as a valuable tool for quickly diagnosing typhoid or paratyphoid fever during the early stages of the illness (Sherwal et al., 2004). Widal test has been used for over a century in developing countries for diagnosis but it has been reported to have low sensitivity, specificity and positive predictive value (Sherwal et al., 2004; Begum et al., 2009). In different district of Haryana, 9-12% incidence rate were observed for salmonellosis by many researchers (Banerjee et al. 2014; Sharma et al., 2015). The rise in seropositivity might be linked to the population growth and poor hygienic standards. As this disease is mainly preventable, it's crucial to educate individuals about adopting safer hygiene practices and the accessible vaccination methods.

For scrub typhus, 15 out of 99 humans were positive, and 4 out of 70 were positive for leptospirosis. However, 100% positivity rate was observed in Karnal, Kurukshetra and Yamuna Nagar for scrub typhus and similar trend was observed in Karnal and Yamuna Nagar for leptospirosis (Table 1, Figure 1). In rural areas, farmers and agricultural workers are frequently affected, particularly during sowing and harvesting seasons, as

well as meteorological events like monsoons. The transmission of leptospiral infection is closely linked to factors such as the presence of farm animals and rodents, known carriers of the bacteria, in agricultural land. The survival of *Leptospira* causing bacteria in wet and humid environments, coupled with regular human engagement in agricultural and animal-rearing activities, serves as the primary contributors to its transmission (Sethi et al., 2010). Northernmost parts in Karnal and Yamuna Nagar districts in the state received high rainfall during monsoon than other part of Haryana (Singh, 2018) and Karnal is fully irrigated districts, having high cultivated land for rice-wheat crops (Kalra et al., 2001). This might be the reason of high positivity rate in these districts for leptospirosis and scrub typhus.

Targeted surveillance would be helpful for better understanding the prevalence of zoonotic diseases in humans and animals. Such as, testing of humans who regularly come in-contact with animals infected with zoonotic diseases and vice versa will be welcome approach for early detection of potential zoonotic disease transmission. It will also help in understanding the root cause of zoonotic diseases, development of comprehensive control strategies, including the implementation of biosecurity measures and health education programs for both animal handlers and the general public.

**Table 1: Details of zoonotic diseases diagnosed in animals and humans in Haryana**

Diseases	Diagnosis test performed	Suspected case	Positive case	District from which positive cases reported
<b>Bovine brucellosis</b>	RBPT	25,911	216	Karnal, Panchkula, Kurukshetra, Sonapat, Kaithal, Hisar, Yamuna Nagar, Panipat and Fatehabad
<b>Bovine tuberculosis</b>	PPD intradermal/ZN staining	115	0	-
<b>Bovine paratuberculosis</b>	PPD intradermal/ZN staining	154	0	-
<b>Human tuberculosis</b>	Sputum microscopy/ CB-NAAT	13,693	2,048	Mewat, Palwal, Kurukshetra, Sirsa, Rohtak, Mahendergarh, Kaithal and Rohtak
<b>Human salmonellosis</b>	Widal test	22,750	3,884	Hisar, Kurukshetra, Kaithal, Palwal, Rohtak, Rewari, Mewat, Faridabad and Mahendergarh
<b>Human scrub typhus</b>	IgM ELISA	99	15	Karnal, Kurukshetra and Yamuna Nagar
<b>Human leptospirosis</b>	IgM ELISA	70	4	Karnal and Yamuna Nagar

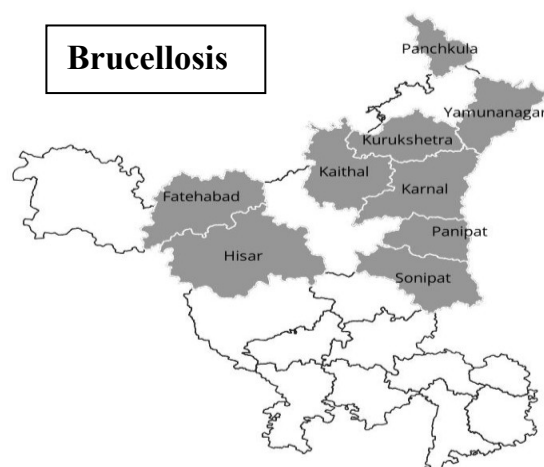


Figure 1: Map depicting the district of Haryana having positive cases of brucellosis in the animal population

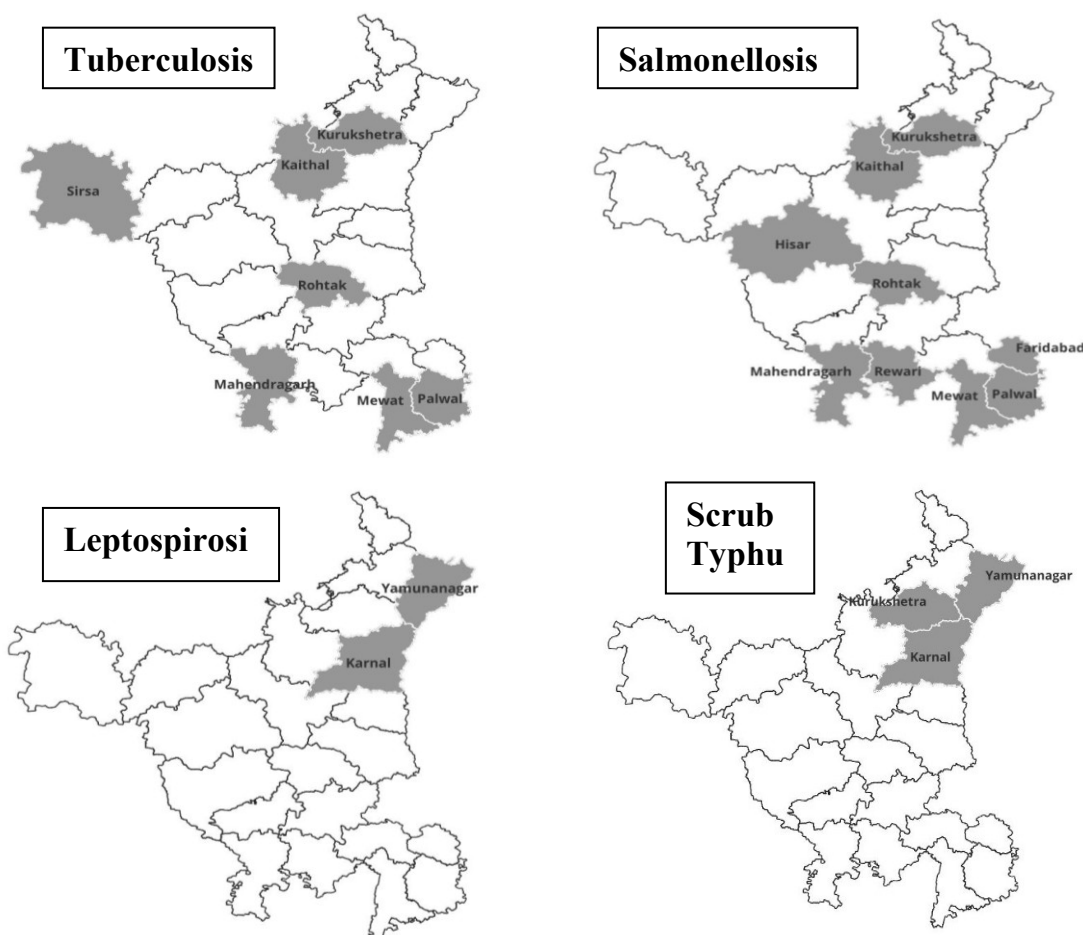


Figure 2: Map depicting the district of Haryana having positive cases for various zoonotic diseases in humans

## Conclusion:

To sum up, the identification of zoonotic diseases demands a multidisciplinary approach among public health agencies, veterinarians, and healthcare practitioners. Early diagnosis and prompt treatment of these diseases is necessary to ensure optimal health

outcomes and prevent the spread of these diseases to animals and humans. The findings underscore the need for enhanced surveillance, targeted preventive strategies, and public health interventions, especially in areas prone to zoonotic diseases. This study lays the groundwork for further research, emphasizing the importance of interdisciplinary collaboration to combat zoonotic diseases in efficient manner.

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### Conflict of Interest:

The authors report no conflict of interest.

### Data availability:

Additional data will be available on request.

### Author's contribution:

Punit Jhandai: Investigation; Conceptualization; Writing – original draft; Formal analysis; Data curation. Shanmugasundaram K: Writing – review and editing; Validation. Dolly Gambhir: Supervision, Monthly reporting. Tushar Nale: Conceptualization, Monitoring and assessment. Ajit Shewale: Conceptualization, Monitoring and assessment. Simmi Tiwari: Supervision; Conceptualization. Harisankar Singha: Supervision; Conceptualization; Writing– review and editing; Validation.

### Ethical statement:

Biological samples from humans or animals were collected at the Out Door Patient Department (OPD) of respective hospitals upon consent of human or animal keeper for disease diagnosis.

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