

WOAH Reference Laboratory Reports Activities 2025

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LABORATORY INFORMATION

| | |
|---|--|
| *Name of disease (or topic) for which you are a designated WOA Reference Laboratory: | Leptospirosis |
| *Address of laboratory: | ICAR-NIVEDI, Yelahanka, Bengaluru, India |
| *Tel: | +9108023093136 |
| *E-mail address: | b.vinayagamurthy@icar.org.in |
| Website: | https://www.nivedi.res.in/ |
| *Name (including Title) of Head of Laboratory (Responsible Official): | Dr. BALAMURUGAN VINAYAGAMURTHY |
| *Name (including Title and Position) of WOA Reference Expert: | Dr BALAMURUGAN VINAYAGAMURTHY, PRINCIPAL SCIENTIST |
| *Which of the following defines your laboratory? Check all that apply: | Governmental |

TOR1: DIAGNOSTIC METHODS

1. Did your laboratory perform diagnostic tests for the specified disease/topic for purposes such as disease diagnosis, screening of animals for export, surveillance, etc.? (Not for quality control, proficiency testing or staff training)

Yes

| Diagnostic Test | Indicated in WOA Manual (Yes/No) | Total number of test performed last year | |
|---|----------------------------------|--|-----------------|
| | | Nationally | Internationally |
| Indirect diagnostic tests | | | |
| Microscopic Agglutination Test (MAT) for Diagnosis | Yes | 263 | 0 |
| Microscopic Agglutination Test (MAT) for Surveillance | Yes | 1274 | 0 |
| Direct diagnostic tests | | | |
| PCR (LipL32) for Diagnosis | Yes | 112 | 0 |
| PCR (LipL32) for Surveillance | Yes | 31 | 0 |
| Real Time qPCR (LipL32) for Diagnosis | Yes | 23 | 0 |

TOR2: REFERENCE MATERIAL

2. Did your laboratory produce or supply imported standard reference reagents officially recognised by WOA?

No

3. Did your laboratory supply standard reference reagents (nonWOAH-approved) and/or other diagnostic reagents to WOA Members?

Yes

| Type of reagent available | Related diagnostic test | Produced/ provide | Amount supplied nationally (ml, mg) | Amount supplied internationally (ml, mg) | No. of recipient WOA Member Countries | Country of recipients |
|---------------------------|-------------------------|-------------------|--|--|---------------------------------------|-----------------------|
| Leptospira serovars | MAT | Provided | 1ml of Culture each for 7 laboratories (a total of | NA | 1 | INDIA, |

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| | | | | | | |
|---|--|-----------------------|--|----|---|--------|
| | | | 23 serovars supplied) | | | |
| Leptospira staining kit | Identification of Leptospira through silver staining | Produced and Supplied | 4 kits (each capacity to test of 100 samples) supplied to 4 Laboratories | NA | 1 | INDIA, |
| BovineLeptoLAT KIT (in house developed) | Rapid Test -Latex Agglutination Test | Produced and Supplied | 23 kits (each capacity to test of 100 samples) supplied to 13 Laboratories | NA | 1 | INDIA, |
| Leptospira reference serovar DNA | PCR | Provided | 50µl of DNA each for 12 Laboratories | NA | 1 | INDIA, |

4. Did your laboratory produce vaccines?

No

5. Did your laboratory supply vaccines to WOAHA Members?

TOR3: NEW PROCEDURES

6. Did your laboratory develop new diagnostic methods for the designated pathogen or disease?

No

7. Did your laboratory validate diagnostic methods according to WOAHA Standards for the designated pathogen or disease?

No

8. Did your laboratory develop new vaccines for the designated pathogen or disease?

9. Did your laboratory validate vaccines according to WOAHA Standards for the designated pathogen or disease?

TOR4: DIAGNOSTIC TESTING FACILITIES

10. Did your laboratory carry out diagnostic testing for other WOAHA Members?

No

11. Did your laboratory provide expert advice in technical consultancies on the request of an WOAHA Member?

Yes

| Name of the WOAHA Member Country receiving a technical consultancy | Purpose | How the advice was provided |
|--|---|---|
| NEPAL | Leptospirosis Diagnostic Support: Provided technical advice and support for screening samples for leptospirosis using the Microscopic Agglutination Test (MAT). Additionally, shared guidelines for the safe transport of biological materials to facilitate effective testing and international collaboration. | Correspondence for Collaboration: Communication was conducted via email with the concerned officer, Dr. Binod Lekhak, Professor and Head, Central Department of Microbiology, Tribhuvan University, Nepal, to facilitate coordination and information exchange. |
| JAPAN | For Integrated surveillance in Leptospirosis to Dr. Kinzang Dukpa, Regional One Health Coordinator, WOAHA- Regional representation for Asia and the Pacific, Japan. | Provided PPT slides and document /publication materials- on integrated surveillance for control and management of Leptospirosis to WOAHA |
| ERITREA | Establishment of Leptospirosis diagnostic Laboratory | Provided One month hands on training on Leptospira laboratory training sponsored through IAEA , Austria Fellowship programme. |

TOR5: COLLABORATIVE SCIENTIFIC AND TECHNICAL STUDIES

12. Did your laboratory participate in international scientific studies in collaboration with WOAHA Members other than the own?

No

13. In exercising your activities, have you identified any regulatory research needs* relevant for WOAHA?

No

TOR6: EPIZOOLOGICAL DATA

14. Did your Laboratory collect epidemiological data relevant to international disease control?

Yes

If the answer is yes, please provide details of the data collected:

1. Seroprevalence and Serogroup Mapping:

Epidemiological studies were conducted to collect data on host factors (species, age, sex, breed) and farm-level parameters, including population density, history of abortions or reproductive disorders, introduction of new animals, feed and fodder management, health status, breeding methods, hygiene practices, herd characteristics, presence of other animals (dogs, rodents, or other livestock), farm type (individual or mixed), farm location, and GPS-based geographic distribution. This information was used to assess serogroup prevalence and identify risk factors for leptospirosis.

2. Outbreak Investigation:

Collection of clinical samples from animals and fetuses, rodent samples, and environmental samples (water, soil) was carried out. Data on attack rate, morbidity, mortality, susceptible population, vaccination records, feeding methods, source of feed/fodder, animal purchase sources, drainage and waste management, and breeding practices were recorded to investigate outbreaks and identify contributing factors.

3. Rodent and Environmental Surveillance:

Rodent trapping and identification were performed to capture target species, with subsequent screening of rodent samples. Rodent movements in farms and households were monitored. Environmental samples, including soil from burrowing areas, moist soil, water from drainage, stagnant water, household waste drainage, animal drinking troughs, and cleaning water, were collected and screened to understand environmental reservoirs and transmission pathways of leptospirosis.

15. Did your laboratory disseminate epidemiological data that had been processed and analysed?

Yes

If the answer is yes, please provide details of the data collected:

A. Publications in peer-reviewed journals

B. Institute Annual Reports

1. Active Environmental Transmission of *Leptospira* in a Coastal District of Dakshina Kannada, Karnataka, India.2. Molecular Detection and Phylogenetic Characterization of Pathogenic and Intermediate *Leptospira* Species among Febrile Human Cases in Dakshina Kannada, India.

3. Outbreak Investigation Leptospirosis: ICAR-NIVEDI investigated unexplained livestock deaths (130 animals) at a Kanakapura farm on 27/03/2025. Epidemiological interviews, post-mortems, and testing of 165 samples indicated possible silage-associated exposure. MAT showed low-titer antibodies in 5/42 sera, while PCR confirmed active *Leptospira* infection in blood, vaginal swabs, tissues, and urine. Immediate control measures, biosecurity strengthening, and vaccination were advised to prevent recurrence.

4. Disease Outbreak Investigation in Pig farm, Mandya, Karnataka, India ; An abortion outbreak affecting pigs in Mandya, India, revealed co-detection of *Leptospira interrogans* and *Brucella* spp. in placental tissue and serum samples. This first confirmed swine abortion outbreak of its kind highlights environmental transmission risks and underscores the need for integrated pathogen diagnostics and surveillance in endemic, resource-limited livestock production settings.

16. What method of dissemination of information is most often used by your laboratory? (Indicate in the appropriate box the number by category and list the details in the box)

a) Articles published in peer-reviewed journals:

7

1. Kumar KV, Bokade PP, Pal A, Mahadevappa S, Kumar CHB, Gulati BR & Balamurugan V (2025). One Health approach for understanding and managing animal leptospirosis in India. *Epi-Dis-PHERE (Publication of Health Resilience), Quarterly e-Journal of National Centre for Disease Control, Vol. 1 issue 1, January 2025. Publisher: Sh. Prakash Doval, Assistant Director (Admin), PBA, NCDC, MoH&FW, GoI.*

2. Veena RK, Jayashankar M, Kumar KV, Padma MR & Balamurugan V (2025). Geospatial distribution of *Leptospira*-specific antibodies in febrile illness cases from Dakshina Kannada, India (2022–2023). *Trop Med Int Health, 30 (7), 737–748. https://doi.org/10.1111/tmi.14132*

3. Kumar KV, Bokade PP, Pal A, Sowjanya Kumari S, Bharath V, Shome BR & Balamurugan V (2025). Detection of anti-leptospiral antibodies in the serum of animals using recombinant *Leptospira* GroEL-based latex agglutination test. *Microb Pathog, 205, 107658. https://doi.org/10.1016/j.micpath.2025.107658*

4. Hota A, Thankappan S, Biswal S, Sahoo N, Behera SK, Balamurugan V, Tma SK, Nagarajan M & Deneke Y (2025). Detection of ovine leptospirosis in various agro-climatic

zones of Odisha in the aftermath of cyclone Hudhud using a multi-faceted approach. *Indian J Anim Sci*, 95 (5), 387–395. <https://doi.org/10.56093/ijans.v95i5.167456>

5. Nazar M, Ramesh PT, Lathamani VS, Balamurugan V, Rathnamma D & Murthy KMS (2025). Seroprevalence and molecular identification of canine leptospirosis in and around Bangalore. *Int J Adv Biochem Res*, 9 (7S), 901–908. <https://doi.org/10.33545/26174693.2025.v9.i7SI.5026>

6. Murag S, Rathnamma D, Koppad S, Choudapur MK, Suresh KP, Balamurugan V & Patil SS (2025). Seroprevalence and determination of serogroup-specific antibodies of *Leptospira* in cattle and buffaloes in Karnataka, India. *Indian J Anim Sci*, 95 (7), 580–587.

7. Vinod Kumar K, Bokade PP, Lakshman R, Deenadayalan O, Sowjanya Kumari S, Nayak A, Pal A, Suresh KP, Dharmashekar C, Shivamallu C, Balamurugan V. Meta-analysis of bovine leptospirosis prevalence in India. *Arch Razi Inst*. 2025 May 31. [Online published].

b) International conferences:

7

1. Thushar H. C., Arun Y. P., Swathi M., K. Vinod Kumar, V. Balamurugan (2025).

Standardization of multiplex PCR (mPCR) for detection and differentiation of *Leptospira*. Poster presented at the International Symposium on "Advancing One Health: Concept to Action for Promoting Human, Animal and Environmental Health" and the XXI Annual Conference of the Indian Association of Veterinary Public Health Specialists (IAVPHS), College of Veterinary Science, GADVASU, Ludhiana, Punjab, India, 11–12 December 2025 (P61).

2. K. Vinod Kumar, Arun Y. P., Prajakta P. Bokade, Archana Pal, Abhilasha K., Veena R. K., Chethan Kumar H. B., B. R. Gulati, V. Balamurugan (2025).

Molecular detection and phylogenetic characterization of pathogenic and intermediate *Leptospira* species from febrile human cases in Dakshina Kannada, India. Poster presented at the International Symposium on Advancing One Health..., GADVASU, Ludhiana, Punjab, India, 11–12 December 2025 (P65).

3. Archana Pal, K. Vinod Kumar, Arun Y. P., Prajakta P. Bokade, R. K. Veena, Shilpa Devraj, Chethan Kumar H. B., Baldev R. Gulati, V. Balamurugan (2025).

Active environmental transmission of *Leptospira* in a coastal Dakshina Kannada district of Karnataka, India. Poster presented at the International Symposium on Advancing One Health..., GADVASU, Ludhiana, Punjab, India, 11–12 December 2025 (P126).

4. Prajakta P. Bokade, K. Vinod Kumar, M. Sreevidhya, Archana Pal, Arun Y. P., Shweta Priya, Archudhan L., V. Balamurugan (2025). Pathogenicity study of a virulent *Leptospira* isolate from Bandicoot rat in a golden Syrian hamster model: Toward identification of diagnostic and immunogenic biomarkers. Poster presented at the International Symposium on Advancing One Health..., GADVASU, Ludhiana, Punjab, India, 11–12 December 2025 (P128).

5. K. Vinod Kumar, Arun Y. P., Prajakta P. Bokade, Archana Pal, Rakshit Ojha, Swathi M., Baldev R. Gulati, V. Balamurugan (2025). Comparative metabolomics reveals antagonistic metabolic networks underpinning the transition between planktonic and biofilm states of *Leptospira*. Poster presented at the International Symposium on Advancing One Health..., GADVASU, Ludhiana, Punjab, India, 11–12 December 2025 (P140).

6. Archana Pal, K. Vinod Kumar, Prajakta P. Bokade, Arun Y. P., Sreevidhya M., Rakshit Ojha, Swathi M., Abhilasha K., R. K. Veena, Satyamitra Shekhar, Anita Chauhan, Amrutlal Patel, Baldev R. Gulati, V. Balamurugan (2025). Characterization of a virulent *Leptospira interrogans* strain: Advancing challenge models for vaccine development in India. Poster presented at VIBCON 2025 – XXX Annual Convention of ISVIB and the International Conference on "Envisioning Livestock Production and Protection under the One Health Landscape", ICAR–IVRI, Mukteswar Campus, Nainital, Uttarakhand, India, 6–8 November 2025 (P13–14).

7. V. Balamurugan, K. Vinod Kumar, Y. P. Arun, B. R. Gulati (2025). Bridging boundaries: Integrated One Health approach to the control and management of leptospirosis. Poster presented at VIBCON 2025 – XXX Annual Convention of ISVIB and the International Conference on "Envisioning Livestock Production and Protection under the One Health Landscape", ICAR–IVRI, Mukteswar Campus, Nainital, Uttarakhand, India, 6–8 November 2025 (P06).

c) National conferences:

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1. Dr. V. Balamurugan delivered Expert Lecture on title "Veterinary Role in Leptospirosis Control: Diagnosis, Management, and Prevention" at 4th One Health e-Learning Round Leptospirosis "Clinical Management, Diagnosis, Control and Its Public Health Impact" Organized by Centre for One Health National Centre for Disease Control, Ministry of Health & Family Welfare, Directorate General of Health Services Gol on 18th February 2025 (Virtual mode)

2. Dr. V. Balamurugan participated and delivered guest lecture on One Health strategy to control Leptospirosis in India, in Zoonotic Diseases webinar Series (online) conducted by CAHO, New Delhi on 6th September 2025. Over 100 participants attended the lecture and benefited.

3. Dr. V. Balamurugan delivered an expert talk on topic "Leptospirosis at the Human–Animal–Environment Interface: Advancing One Health Strategies" at VIBCON-2025 (XXX Annual Convention of ISVIB and International Conference) organized by ICAR-IVRI, Mukteswar on 5 November, 2025.

d) Other (Provide website address or link to appropriate information):

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1. Dr. Arun Y.P participated and awarded for best poster presentation on title "Molecular detection and phylogenetic characterization of Pathogenic and intermediate *Leptospira* species from Febrile Illness Human cases in Dakshina Kannada, India" at the XXI Annual conference of Indian Association of Veterinary Public Health Specialists (IAVPHS) held at Ludhiana, Punjab, from 11th - 12th December 2025.

2. Ms. Prajakta participated and awarded for best oral presentation on title "Pathogenicity study of a virulent *Leptospira* isolate from Bandicoot rat in a Golden Syrian Hamster model: Towards identification of diagnostic and immunogenic biomarkers" at the XXI Annual conference of Indian Association of Veterinary Public Health Specialists (IAVPHS) held at Ludhiana, Punjab, from 11th - 12th December 2025.

3. Chethan Kumar H.B, Vinod Kumar K, Arun Y.P, M. Nagalingam, Siju Susan Jacob, and Balamurugan V (2025). Training Manual : Training on Techniques for Zoonotic Disease Diagnosis, Surveillance and Mapping. First Edition, Pages xviii+ 124, ICAR-National Institute of Veterinary Epidemiology and Disease Informatics (NIVEDI), Bengaluru, India.

4. V. Balamurugan, Vinod Kumar, Arun Y P, Swathi M., Prajakta P. Bokade, M. Nagalingam, H. B. Chethan Kumar, K. (2025). Training Manual: Laboratory Diagnosis of

Leptospirosis. First Edition, Pages 105, Publisher- ICAR-National Institute of Veterinary Epidemiology and Disease Informatics, Bengaluru-560064, India. ISBN: 9789-360-131-067

5. Balamurugan V, Vinod Kumar K, SreeVidhya M, Arun YP, Chethan Kumar HB, Nagalingam M. *FAQs on Leptospirosis. ICAR-National Institute of Veterinary Epidemiology and Disease Informatics; Bengaluru: 2025. Pp 1-36 ISBN: 978-93-343-7669-2*

6. Dr. Arun Y.P participated in 2-day training programme on Inter-Laboratory Comparison, Proficiency Testing, and Evaluation of Scores (ILCPT), held from 25–26 June 2025 at NITS, BIS, Noida, under the NCDC project, focused on enhancing participants' understanding of quality assurance in laboratory practices. Key topics covered included the principles and objectives of inter-laboratory comparisons, design and implementation of proficiency testing (PT) schemes, statistical analysis and interpretation of PT results using Z-scores, and compliance with ISO/IEC 17043 standards. The training enabled participants to evaluate laboratory performance, ensure test result reliability, and strengthen competency in conducting and participating in national and international PT programmes.

TOR7: SCIENTIFIC AND TECHNICAL TRAINING

17. Did your laboratory provide scientific and technical training to laboratory personnel from other WOAHP Members?

Yes

a) Technical visit : 0

b) Seminars : 0

c) Hands-on training courses: 1

d) Internships (>1 month) 0

| Type of technical training provided (a, b, c or d) | Country of origin of the expert(s) provided with training | No. participants from the corresponding country |
|--|---|---|
| C | ERITREA | 1 |

TOR8: QUALITY ASSURANCE

18. Does your laboratory have a Quality Management System?

Yes

| Quality management system adopted | Certificate scan (PDF, JPG, PNG format) | |
|-----------------------------------|---|--|
| ISO 17025:2017 | PDF | NABL Accreditation certificates NIVEDI.pdf |
| ISO 9001:2015 | PDF | ISO9001 CERTIFICATE NIVEDI.pdf |

19. Is your quality management system accredited?

Yes

| Test for which your laboratory is accredited | Accreditation body |
|--|---|
| Detection of Leptospira antibodies by MAT (Microscopic Agglutination Test) | National Accreditation Board for Testing and Calibration Laboratories (NABL), India |
| Detection of Leptospira genomic DNA by PCR | National Accreditation Board for Testing and Calibration Laboratories (NABL), India |
| Detection of Leptospira genomic DNA by Real time qPCR | National Accreditation Board for Testing and Calibration Laboratories (NABL), India |

20. Does your laboratory maintain a "biorisk management system" for the pathogen and the disease concerned?

Yes

ICAR-NIVEDI's facilities are NABL-accredited for the storage and handling of selected agents, including Leptospira. All laboratory activities are conducted under Biosafety Level 2 (BSL-2) biocontainment conditions.

TOR9: SCIENTIFIC MEETINGS

21. Did your laboratory organise scientific meetings related to the pathogen in question on behalf of WOAHP?

No

22. Did your laboratory participate in scientific meetings related to the pathogen in question on behalf of WOAHP?

No

TOR10: NETWORK WITH WOAHP REFERENCE LABORATORIES

23. Did your laboratory exchange information with other WOAHP Reference Laboratories designated for the same pathogen or disease?

Not applicable (only WOAHP Reference Laboratory designated for the disease)

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24. Are you a member of a network of WOA Reference Laboratories designated for the same pathogen?

Not applicable (only WOA Reference Laboratory designated for the disease)

25. Did you organise or participate in inter-laboratory proficiency tests with WOA Reference Laboratories designated for the same pathogen during the past 2 years?

Yes

| Purpose of the proficiency test: | Role of your Reference Laboratory (organiser/ participant) | No. participating Laboratories | Participating WOA Ref. Labs/ organising WOA Ref Lab |
|--|--|--------------------------------|---|
| Performance of MAT (Reference Serovars) and Leptospirosis diagnosis by MAT | Participant | 77 | International Proficiency Testing Scheme organized by ILS for the Leptospirosis MAT |

26. Did your laboratory collaborate with other WOA Reference Laboratories for the same disease on scientific research projects for the diagnosis or control of the pathogen of interest?

No

TOR11: OTHER INTERLABORATORY PROFICIENCY TESTING

27. Did your laboratory organise or participate in inter-laboratory proficiency tests with laboratories other than WOA Reference Laboratories for the same pathogen during the past 2 years?

Yes

| Purpose for inter-laboratory test comparisons ¹ | Role of your reference laboratory (organizer/participant) | No. participating laboratories | Name of the test | WOAH Member Countries |
|---|---|--------------------------------|------------------|-----------------------|
| Performance of Leptospirosis diagnosis by PCR (Name of the test : Leptospirosis molecular LEPN 435) | Participant (certificate received in January 2025) | 50 | PCR and qPCR | AUSTRALIA, |

TOR12: EXPERT CONSULTANTS

28. Did your laboratory place expert consultants at the disposal of WOA?

No

29. Additional comments regarding your report:

Yes

- As a designated WOA Reference Laboratory for Leptospirosis, ICAR–NIVEDI is fully equipped to undertake sample screening for both WOA member and non-member countries. Assistance from WOA and/or FAO is requested to streamline the receipt of samples from Asian, SAARC, or other regional countries.
- ICAR–NIVEDI has received official permission from the Department of Animal Husbandry and Dairying (DAHD), Government of India, to receive samples from WOA member countries, in accordance with the prescribed guidelines, for Leptospirosis diagnostic testing.
- Laboratory Information Management System (LIMS): The offline, customized LIMS has been developed according to the existing framework for the testing and diagnosis of samples for Leptospirosis.
- ICAR–NIVEDI plans to organize international training programs—both virtual and hands-on—for WOA member and non-member countries in the upcoming reporting year (2026). Support is requested from WOA and/or FAO in terms of organization and funding for these trainings.
- ICAR–NIVEDI's WOA Reference Laboratories actively participate in online meetings, seminars, and conferences organized by FAO, WOA, and the PPR Secretariat, as required, to enhance expertise, facilitate global collaboration, and share technical knowledge.
- ICAR–NIVEDI's WOA Reference Laboratory actively participates in online meetings, seminars, and conferences organized by FAO and WOA at various times, as and when required, to acquire knowledge, enhance expertise, collaborate effectively, and share our expertise with global communities.
- A hands-on training program on laboratory diagnosis of leptospirosis was conducted at ICAR–NIVEDI, Bengaluru, from 11–14 August 2025. The program provided practical exposure to serological, molecular diagnostic techniques and biosafety practices. A total of 34 participants from 11 Indian states, representing veterinary, medical, and public health sectors, attended the training.
- ICAR–NIVEDI expresses its interest in initiating a WOA Reference Laboratory Network for Leptospirosis, aimed at linking the WOA Leptospira Working Group and reference laboratories worldwide. This initiative would strengthen global collaboration, promote harmonization of protocols, facilitate genomic data sharing and analysis, and enhance collective expertise.
- A training program titled "Techniques for Zoonotic Disease Diagnosis, Surveillance and Mapping" was conducted at ICAR–NIVEDI, Bengaluru, from 03–07 March 2025. The training focused on integrated laboratory and field-based approaches for zoonotic disease surveillance. Thirty-two participants from nine Indian states representing veterinary, medical, and public health departments participated.
- An international one-month training program on "Isolation and Identification of Leptospira using Microbiological and Molecular Biology Tools" was organized at ICAR–NIVEDI, Bengaluru, from 04 August to 03 September 2025. The program emphasized advanced culture techniques, molecular diagnostics, and confirmation methods.
- Laboratory developed a New Virulent Leptospira Strain as a Challenge Hamster Model for Vaccine Efficacy and Quality Control Evaluation of Leptospirosis Vaccines to Boost Vaccine Development.