

A protocol for a systematic review and meta-analysis

Title:

Prevalence of leptospirosis in cats through different diagnostic methods: a systematic review and meta-analysis

Registration

This protocol is available online in the Systematic Review for Animals and Food (SYREAF) and Open Science Framework (OSF) (<https://osf.io/b3u8j/>). PRISMA-P guidelines are used to compile the content and items in this Protocol (Moher et al., 2019). Preferred Reporting Items for Systematic Review and Meta-analyses (PRISMA) will be used to report this study's systematic review and meta-analysis (Liberati et al., 2009).

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Author contributions

Morsid Andityas (MA) and Dian Meididewi Nuraini (DMN) are two investigators who have contributed: to the design study, searching, screening for inclusion, data extraction, risk of bias assessment, data analysis, and writing of the paper. Sirikachorn Tangkawattana (ST) is the third reviewer who has contributed to providing advice in resolving differences of opinion between the two reviewers. Additionally, all authors contributed to providing suggestions, designing studies, and writing papers.

Amendments

In case there is a deviation from the protocol during the review, the deviation will be reported in this systematic review.

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INTRODUCTION

Rationale

Cats can act as pets or feral cats that live around human life. These conditions provide an opportunity for potential zoonotic diseases, such as *Toxoplasma gondii*, tularemia, cutaneous larval migrans, rabies, leptospirosis, plague and others (Alashraf et al., 2020; Gerhold & Jessup, 2013). Moreover, Current information regarding cat leptospirosis is still controversial. Cats have a strong resistance to leptospirosis (Hartmann et al., 2013; Mwachui et al., 2015). Pathogenic leptospire were found from urine and kidney samples of naturally infected cats (Alashraf et al., 2020; Chan et al., 2014; Rodriguez et al., 2015; Weis & Hartmann, 2017). In addition, the isolation results were also found in healthy cats, which provides evidence that healthy cats can serve as subclinical and subserological reservoirs of leptospirosis (Alashraf et al., 2020; Rodriguez et al., 2014). Clinical signs of feline leptospirosis are usually asymptomatic or mild, although there is evidence of leptospiuria, leptospiremia, and signs pathology of inflammation in kidney and liver (Azócar et al., 2014).

Detecting feline leptospirosis is challenging due to serological, molecular, and other diagnostic limitations. Diagnostic examination of feline leptospirosis in the laboratory is usually based on the Microscopic Agglutination Test (MAT) and Polymerase Chain Reaction (PCR). Serological tests for diagnosing leptospirosis in cats, either by rapid test technique or ELISA, have not been developed (Murillo et al., 2020). To our knowledge, no commercial diagnostic kits are explicitly sold for detecting cat leptospirosis. In epidemiological studies, the limitation of diagnostic methods for detecting cat leptospirosis is a major challenge. Some researchers faced difficulty in studying cases of cat leptospirosis in an area. This situation still raises big questions regarding the role of cats in zoonotic diseases caused by *Leptospira* bacteria. The lack of knowledge regarding the diagnostic techniques used to check the prevalence of leptospirosis in cats is a severe issue in epidemiological studies.

Objectives

The aims of this study is to determine the prevalence of cat leptospirosis through various diagnostic methods, followed by further analysis based on subgroup characteristics (seroprevalence, infection prevalence, continent, sample type, and cut-off). This protocol is designed to answer questions: "What is the prevalence of leptospirosis in cats through different diagnostic methods?"

METHODS

Eligibility criteria

The specific PICO items, which define the eligibility criteria, are as follows:

- Specify the **P**opulation : Cats, Feline, domestic cats and feral cats
- Specify the **I**ntervention : Leptospirosis, Leptospira sp.
- Specify the **C**omparison: Not applicable
- Specify the **O**utcome : Prevalence (seroprevalence & infection prevalence), Continents, diagnostic methods Sample type, cut off value

Study designs to be included.

- Inclusion criteria: case study, survey, cross sectional
- Exclusion criteria: experimental study, cohort, case control

Intervention(s), exposure(s).

- Inclusion criteria: Seroprevalence and infection prevalence of leptospirosis in cats through different diagnostic methods
- Exclusion criteria: Diagnostic methods used for experimental study, case control, and cohort

Animals/population.

- Inclusion criteria: Cats, feline, domestic cats, feral cats, all breed, both sexes, adult cats
- Exclusion criteria: Kitten, human, and animals other than cats

Language: Publications in English

Publication date: No limits.

Geographical location of studies: No limits.

Information sources

Five medical databases; including PubMed, Scopus, Web of Science, ScienceDirect and Google Scholar will be used for the present study (Table 1). In this systematic review and meta-analysis, articles that focus on the diagnostic methods used to case study, survey, and cross sectional of the prevalence of leptospirosis in cats will be searched. Search is only limited to articles using the English language.

Table 1: List of databases to be searched

| Database | URL |
|-----------------|---|
| PubMed | https://pubmed.ncbi.nlm.nih.gov/ |
| Scopus | https://www.scopus.com/ |
| Web of science | http://webofknowledge.com/ |
| ScienceDirect | https://www.sciencedirect.com/ |
| Google Scholar | https://scholar.google.com/ |

Search strategy

The details of the search strategies for each database are provided in the search strategy (the search will be based on keywords containing names and synonyms): seroprevalence, infection prevalence, incidence, epidemiology, serologic, serological, molecular, diagnostic, diagnostic method, diagnostic tool, detection, leptospirosis,

Leptospira, cats and feline. All experimental studies, reviews, systematic reviews, and meta-analyses will be excluded. Medical Subject Heading (MeSH) terms and keywords to be used in the search strategy can be seen in table 2.

Table 2: Search strategies in this study

| Search | Query | Results |
|-----------------------|---|---------|
| PubMed | | |
| #1 | leptospirosis OR leptospira* OR "leptospira serovar*" OR serovar* OR "Weil's syndrome" OR "pretibial fever" | |
| #2 | cat* OR feline | |
| #3 | # 1 AND #2 | |
| Web of Science | | |
| #1 | leptospirosis OR leptospira OR "leptospira serovar" OR serovar OR "Weil's syndrome" OR "pretibial fever" | |
| #2 | seroprevalence OR prevalence OR incidence OR epidemiology OR serologic OR serological OR molecular OR diagnostic OR "diagnostic method" OR "diagnostic tool" OR detection | |
| # 3 | cat OR feline | |
| # 4 | # 1 AND #2 AND #3 | |
| SCOPUS | | |
| #1 | leptospirosis OR leptospira OR "leptospira serovar" OR serovar OR "Weil's syndrome" OR "pretibial fever" | |
| #2 | seroprevalence OR prevalence OR incidence OR epidemiology OR serologic OR serological OR molecular OR diagnostic OR "diagnostic method" OR "diagnostic tool" OR detection | |
| #3 | cat OR feline | |
| #4 | # 1 AND #2 AND #3 | |
| Science Direct | | |
| #1 | leptospirosis OR leptospira | |
| #2 | seroprevalence OR prevalence OR incidence OR epidemiology OR diagnostic | |
| #3 | cat OR feline | |
| #4 | # 1 AND #2 AND #3 | |
| Google Scholar | | |
| #1 | leptospirosis OR leptospira | |
| #2 | cat OR feline | |
| Limit to | allintitle: leptospirosis OR leptospira "cat OR feline" | |

Study Records

Data management

All article databases will be imported into Mendeley for duplication filtering and duplication removal. Then, all articles were entered into Rayyan's library (online software) for screening of each journal. Two investigators will screen all journals independently (MA and DMN). Disagreement between the two reviewers will be resolved through a third reviewer (ST) discussion. Define screening phases (pre-screening based on title and abstract, and full text screening).

Selection process

Data will be extracted from eligible studies independently by two investigators (MA and DMN). Rayyan is used as filtering and data extraction for each journal. The exclude and include process follows the question-statement guidelines in determining relevance. The results of the answers from the questions are divided into three answers according to Rayyan's features: YES (neutral response), NO (not included), MAYBE (neutral response). the decision of an article cannot be continued to the next stage if a question is answered "no" by two investigators (MA and DMN). if found disagreements between the two reviewers from the results of the answers will then be resolved through discussion with the third reviewer (ST). The following questions are used as guidelines:

- 1) Does the primary study report the prevalence of cat leptospirosis?
- 2) Do the title and abstract of an article describe the PICO items that are relevant to this study?
- 3) Is the full text available in the article?
- 4) Is the full text available in English?
- 5) Is this an original research study with a qualified design?

Data collection process

Methods for data extraction Use two steps of screening: 1. Screen the titles and abstracts of retrieved citations use; 2. Screen the relevant full-text articles for eligible study. The titles and abstracts of the studies will be screened independently by two investigators (MA and MDN). The full-text articles will be reviewed by the same two investigators. Discrepancies between the two reviewers will be resolved through discussion with a third reviewer (ST). The author or corresponding author of an article will be contacted by email when additional information is required or the article is not accessible.

Data items

General information for inclusion:

- 1) the first author
- 2) years
- 3) study design (case study, survey, cross sectional study)
- 4) gene target in molecular test
- 5) Location samples
- 6) Animal or populations (feline, domestic, pets, stray, feral, and sheltered)
- 7) Language
- 8) gene target in molecular diagnostic

Prioritise the exclusion criteria:

- 1) type of population (kitten, human, and animals other than cat)
- 2) Intervention (diagnostic methods used for experimental study, case control, and cohort)
- 3) Study designs (experimental study, cohort, case control)

- 4) Article review (a review, systematic review, meta-analysis and others)
- 5) Serovar
- 6) risk factor

Outcomes and prioritization

Data to be extracted:

- 1) Seroprevalence
- 2) infection prevalence
- 3) cut off
- 4) Diagnostic method
- 5) sample type
- 6) continent and country

Risk of Bias Assessment

The risk of bias will be assessed by two authors independently (MA and DMN) using "The National Institutes of Health (NIH) quality assessment tool for observational cohort and cross-sectional studies" (Website: <https://www.nhlbi.nih.gov/health-topics/study-quality-assessment-tools>). Risks will be categorized into domains and assessed as low, high or unclear risk.

Data synthesis

The characteristics of the included studies and the prevalence of cat leptospirosis through different diagnostic methods will be analyzed using a random effect model with effect measure proportions and calculated 95% confidence intervals. The statistical methods to assess heterogeneity are I^2 , Q , and p -value. In Subgroup analyses, we will analyze the seroprevalence and infection prevalence subgroup approach in all prevalence, diagnostic methods, sample types, and continents. While at the cut-off, subgroups will be analyzed based on values $> 1:20$ and $\geq 1:100$. In the sensitivity test, we will rerun the analysis excluding studies with a higher risk of bias. Risks of bias will be categorized into domains and assessed as low, high, or unclear risk. Funnel plot symmetry also will be used to further evaluate the publication bias risk.

Conclusions

The overall objective of this systematic review is to determine the prevalence of feline leptospirosis through various diagnostic methods so that it can be used as a reference and decision in the use of diagnostic methods in epidemiological studies. Information on various characteristics through meta-analysis is expected to add information about the role of cats in zoonotic diseases caused by leptospire.

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