A protocol for a systematic review and meta-analysis

Title:

Is vaccination efficacious to prevent or control colibacillosis in broiler production? A protocol for a systematic review and meta-analysis.

Authors and their affiliations

Surya Paudel[1,2,*], Ilias Apostolakos[3,*], Giuditta Tilli[4], Ronald Vougat ngom[5,6], Helena C. de Carvalho Ferreira[7], Alessandra Piccirillo[4,#]

[1]Clinic for Poultry and Fish Medicine, Department for Farm Animals and Veterinary Public Health, University of Veterinary Medicine Vienna, Austria
[2]Department of Infectious Diseases and Public Health, Jockey Club College of Veterinary Medicine and Life Sciences, City University of Hong Kong, Kowloon, Hong Kong
[3]Dairy Research Institute, Hellenic Agricultural Organization "DIMITRA", Ioannina, Greece
[4]Department of Comparative Biomedicine and Food Science, University of Padua, Legnaro, Italy
[5]Department of Animal Production, School of Veterinary Medicine and Sciences, University of Ngaoundéré, Ngaoundéré, Cameroon
[6]Veterinary Public Health Institute, Vetsuisse Faculty, University of Bern, Bern, Switzerland
[7]Flanders Research Institute for Agriculture, Fisheries and Food, Merelbeke, Belgium

[*] both authors contributed equally
[#] corresponding author. E-mail address: alessandra.piccirillo@unipd.it

Author contributions

The review (PICO) question and protocol described in this document were developed with the contribution and final approval of all co-authors. Surya Paudel and Ilias Apostolakos drafted the protocol and all authors provided their input.

Registration

This protocol is archived at Padua Research Archive (handle code: 11577/3439974) and published online with Systematic Reviews for Animals and Food (SYREAF) available at: http://www.syreaf.org/. This protocol is reported using the items (headings) recommended in the PRISMA-P guidelines (Moher et al., 2015).

Support
This project is funded by the COST Action CA18217 - European Network for Optimization of Veterinary Antimicrobial Treatment (ENOVAT).

Amendments
This review is not an amendment of a previously completed or published protocol. In case any amendments are made to this protocol after its registration, they will be adequately documented in the systematic review as Protocol Deviations.

Acknowledgements
Authors are thankful to Dr. Lisbeth Rem Jessen, Dr. Marnie Brennan, Dr. Luis Pedro Carmo and Dr. Jeroen Dewulf for their expert suggestions while drafting the protocol. We also thank all the members of the ENOVAT Drafting Group "Veterinary guidelines on antimicrobial use in poultry colibacillosis" for their inputs and critical evaluation of the PICO questions.

1. Introduction

1.1. Rationale

Avian pathogenic *Escherichia coli* (APEC) is the causative agent of colibacillosis, a disease with significant economic losses for the broiler industry, and can act as a primary or secondary pathogen when the host immune system is compromised (Nolan et al., 2020). Colibacillosis is manifested as a localised or systemic infection resulting in various disease syndromes that affect all stages of the broiler production. In broiler breeders, increased mortality and decreased egg production due to the salpingitis-peritonitis syndrome can reach the cost of 1.87 euros per housed hen (Landman et al., 2015). At the slaughterhouse, condemnations as a result of cellulitis lead to losses of 0.1%–1.4% of poultry meat and increased labour costs for the process of affected carcasses (Barbieri et al., 2013; Nolan et al., 2020).

E. coli infection of broilers is a global challenge with potential threats to human health. It is probably one of the major reasons for the use of antibiotics in poultry industry. Importantly, *E. coli* is identified as one of the most relevant antimicrobial resistant bacterial pathogens from poultry in a recent report by the European Union (EFSA, 2021). Growing demand to reduce the antibiotic use in poultry has set a clear emphasis on the development of effective vaccine and vaccination strategies against colibacillosis in poultry. In this regard, numerous efforts have been made to identify potential killed, sub-unit or live vaccine candidates, which are summarized in published reviews and book chapters (Ghunaim et al., 2014; Christensen et al., 2020; Nolan et al., 2020; Kathayat et al., 2021; Swelum et al., 2021). However, concerns regarding the strain coverage and feasibility of application still exists and the number of licensed products is very low.

Autogenous vaccines are commonly used in the field that are not required to be tested for the regulatory quality control procedures and might induce negative consequences on performance of birds (Paudel et al., 2021). Thus, *E. coli* is one of the prioritized pathogens in poultry listed by OIE for the significant need to expand the portfolio of vaccine considering applicability in the field and heterologous protection (OIE, 2015).

The protocol is established as an essential and basic tool to perform a systematic review on the efficacy of vaccination to prevent or control colibacillosis in broilers.

1.2. Objectives

This protocol defines the methodology of the systematic review and meta-analysis to address the following PICO question: "*In broilers at risk of colibacillosis, does vaccination versus no vaccination result in higher FCR/fewer condemnations/lower mortality?*" The specific PICO elements are:

1. **P**opulation: Broilers (including the whole production chain).
2. **I**ntervention: Any vaccine (autogenous, inactivated, sub-unit or live) for use in broilers to prevent colibacillosis.
3. **C**omparator: Placebo or untreated control group.
4. **O**utcomes: Mortality, Feed Conversion Ratio (FCR) and condemnations due to colibacillosis at the slaughterhouse.

2. Methods

2.1 Eligibility criteria:

1. Criteria related with the elements of the PICO question (Population, Intervention, Comparator and Outcomes).
2. Language: Publications in English and/or Spanish.
3. Publication types: Journal articles and any other form of research publication that provides results of original research, fulfills the study design eligibility criteria and has a full text of more than [e.g. 500] words.
4. Publication date: No limits.
5. Geographical location of studies: No limits.
6. Studies reporting controlled trials with natural disease exposure will be the primary type of study for inclusion. Disease challenge studies and observational studies will be documented as well and assessed during full-text screening for the reported intervention (administration protocol of licensed vaccine) and measured outcomes of interest.

2.3. Information sources

Bibliographic databases that provide a high level of article recall across biomedical articles (Bramer et al., 2017) will be used. Table 1 lists the databases to be searched. CAB abstract and Agricola will be searched via the University of Bern (Switzerland), Pubmed via the Dairy Research Institute of the Hellenic Agricultural Organization "DIMITRA" (Greece) and Web of Sciences (WOS) via the University of Padova (Italy). All the databases of WOS will be used (Web of science core collection, BIOSIS Citation Index, KCI-Korean Journal Database, Medline, Russian Science Citation Index and SciELO Citation Index). However, we will exclude the following editions: Arts & Humanities Citation Index (A&HCI), Conference Proceedings Citation Index-Science (CPCI-S), Conference Proceedings Citation Index-Social Science & Humanities (CPCI-SSH) and Social Sciences Citation Index (SSCI).

Table 1: List of databases to be searched.

Database	**Interface**	**URL**
MEDLINE	PubMed	https://pubmed.ncbi.nlm.nih.gov/
CAB abstracts	Ovid	https://www.wolterskluwer.com/en/solutions/ovid/cab-abstracts-31
Web of science	Web of Science	http://webofknowledge.com/
Agricola	Proquest	https://www.proquest.com/

2.4. Search strategy

The search strategy will involve a multi-stranded approach that uses a series of searches, with different combinations of concepts to gather all possibly related research and thus achieve high sensitivity (Higgins et al., 2021). If only few papers are found to be relevant to the review, in addition to the database, citations will be extracted from a selection of important papers and reviews. In the event of using search reviews, Scopus database will be used for backward searching. Alerts (also known as literature surveillance services) will be set up in the databases (when available) to monitor published studies relevant to the review question after the original search has been conducted.

The concept of the search strategy will be the following:

[**Broilers**] AND [**Vaccination**] AND [**Colibacillosis**].

Search terms will be amended appropriately to reflect the functionality differences in each database. The general search strategy to identify studies relevant to the PICO of this review will be the following:

#1 (chicken* or poultry* or gallus or broiler* or flock*)

#2 (vaccination* or vaccin* or bacterin* or "sub-unit*" or "killed vaccine*" or "live vaccine*" or "autogenous vaccine")

#3 (colibacillosis or colisepticaemia or peritonitis or coli or Escherichia or coliform or colisepticemia or coligranuloma or "Hjarre's" or "air sac disease" or cellulitis or osteomyelitis or "brittle bone disease" or salpingitis or synovitis or omphalitis or enteritis or "hemorrhagic septicemia" or "chronic respiratory disease" or "swollen head syndrome" or "venereal colibacillosis" or "coliform cellulitis" or "yolk sac infection" or APEC or "pathogenic E. coli" or "primary infection" or "secondary infection" or multifactorial or multicausal)

#1 AND #2 AND #3

2.5. Study Records

Data management
Database records of the articles recovered will be imported into Rayyan or Zotero and duplicates will be deleted. Abstract and full screening will be recorded in Rayyan. Data extraction and risk of bias assessment will be done in Revman. Summary of findings table will be done in GradePro.

Selection process
The citations will be screened in two independent stages.
The first stage of the selection process will consist of titles and abstract screening. Two independent reviewers (Surya Paudel and Ilias Apostolakos) will carry out this task using Rayyan. Conflict will be resolved with a third reviewer (Alessandra Piccirillo) if consensus between the two reviewers cannot be reached. Eligibility of studies will be assessed with the following questions, as suggested by Sargeant et al. (2019):

1. Is the study an original research assessing the use of vaccine(s) to prevent or control colibacillosis in broilers? YES [PASS], NO [EXCLUDE], UNCLEAR [PASS]
2. Does the study include an eligible comparator via a controlled trial, disease challenge study or observational study? YES [PASS], NO [EXCLUDE], UNCLEAR [PASS]

The studies that meet inclusion criteria will pass to the next phase. The concordance among the reviewers will be evaluated by selecting the first 25 citations entering each stage of the process prior to screening all papers. This calibration study will enable discussion and solve disagreement before carrying out the full selection process by the two reviewers (Sanguinetti et al., 2021).

The second stage will involve the full-text screening. Two independent reviewers (Surya Paudel and Ilias Apostolakos) will carry out this task using Rayyan. Conflict will be resolved with a third reviewer (Alessandra Piccirillo) if consensus between the two reviewers cannot be reached. Eligibility of studies will be assessed with the following questions:

1. Is a full text of more than [500] words available? YES [PASS], NO [EXCLUDE] UNCLEAR [PASS]
2. Is a full text available in English and/or Spanish? YES [PASS], NO [EXCLUDE]
3. Is the **Population** of the study broilers? YES [PASS], NO [EXCLUDE], UNCLEAR [PASS]
4. Is the **Intervention** of the study the use of vaccine(s) to prevent or control colibacillosis in broilers? YES [PASS], NO [EXCLUDE], UNCLEAR [PASS]
5. Is at least one of mortality, FCR, or condemnations due to colibacillosis the **Outcome(s)** described? YES [PASS], NO [EXCLUDE], UNCLEAR [PASS]
6. Is the study design a controlled trial with natural disease exposure? YES [PASS to data extraction process], NO [this is a disease challenge study, indicate the vaccine(s) assessed and extract data]
7. Is the study design a controlled trial with natural disease exposure? YES [PASS to data extraction process], NO [this is an observational study, indicate the vaccine(s) assessed and extract data]

Data extraction

Two independent reviewers (Surya Paudel and Ilias Apostolakos) will carry out this task using Revman. Conflict will be resolved with a third reviewer (Alessandra Piccirillo) if consensus between the two reviewers cannot be reached. Data to be extracted from eligible studies will include the following items as (partly) suggested by Sargeant et al. (2019):

General information:

1. Country (where the trial study was conducted). If not stated, use country affiliation of corresponding author.
2. Number and type of flocks (commercial broilers or experimental flocks)
3. Breed
4. Sex
5. Production type (conventional, organic, antibiotic-free)
6. Duration and year(s) of study
7. Production stage/age of birds when intervention was applied
8. Production stage/age of birds when outcome(s) were measured

Intervention data:

1. Commercial name and type of the vaccine
2. Route and dose of administration?
3. Unit of population participants (e.g., flock, house/barn/pen)

4. Description of the comparator group (non-treated or placebo-treated)
5. Number of birds enrolled in the participating unit
6. Number of flocks/house/barns/pens enrolled
7. Number of flocks/house/barns/pens enrolled lost until the end of trial study
8. Number of flocks/house/barns/pens enrolled analyzed
9. Method to account for non-independent observations

Outcome data:

1. Mortality
    a. Level at which mortality was measured (e.g., flock, house/barn/pen)
    b. Time period of measured outcome
2. Feed conversion ratio (FCR)
    a. Feed conversion ratio
    b. Age and/or weight of slaughtered participant birds
3. Condemnations due to colibacillosis
    a. Age and/or weight of slaughtered participant birds

For all relevant outcomes, measures of association (e.g., risk ratio, odds ratio, mean differences for continuous outcomes) will be extracted only if variance measures are available or if they can be calculated from the study's outcome data.

2.6. Risk of Bias Assessment

Risk of bias will be assessed only for controlled trials for each of the measured outcomes and according to the Cochrane risk of bias instrument (Higgins et al., 2021). Details on the risk of bias assessment follow below:

Selection bias is caused by factors affecting the selection of study subjects (Dohoo et al., 2009). The selection bias associated with external validity will not be taken into account.

Information bias is caused by factors relating to attaining precise information on the exposure, outcome, and covariates (Dohoo et al., 2009). This domain will be approached using the following questions:

· Have the definitions of cases of colibacillosis been clearly defined?

· Have the methods used to determine colibacillosis been carried out in such a way that assure truthfulness in the diagnosis?

Low risk of information bias example:

· The diagnosis has been carried out by the combination of clinical disease and laboratory methods.

Examples of low risk of confounding:

· Treatment was randomly assigned to broilers;

· Characteristics such as management practices were matched between control and treatment groups;

· The statistical approaches used adjusted for potential confounding.

Confounding bias is caused by the effects of factors other than the exposure of interest on the observed association (Dohoo et al., 2009). The question that will address this type of bias is the following: Were measures taken into account to reduce potential confounding?

2.7. Data synthesis

The intention of this review is to conduct a quantitative synthesis of results via a (network) meta-analysis if an adequate number of eligible studies are captured with the literature search. If quantitative analysis is not possible, qualitative summary will be made. Furthermore, publication bias will be evaluated using previous approaches (Mavridis *et al*., 2013; Marvridis *et al*., 2014).

Conclusions

The overall objective of this systematic review is to examine the efficacy of vaccination in the prevention/control of colibacillosis in broilers. This will help the decision-making process when applying interventions in broilers by producers and field veterinarians and the suggestions made by policymakers. Moreover, the systematic review will suggest gaps in knowledge that require more research in the future.

References

1. Barbieri, N.L., A.L. de Oliveira, T.M. Tejkowski, D.B. Pavanelo, D.A. Rocha, L.B. Matter, S.M. Callegari-Jacques, B.G. de Brito, F. Horn. 2013. Genotypes and pathogenicity of cellulitis isolates reveal traits that modulate APEC virulence. PLoS One 8: e72322.
2. Bramer, W.M., M.L. Rethlefsen, J. Kleijnen, O.H. Franco. 2017. Optimal database combinations for literature searches in systematic reviews: a prospective exploratory study. Syst. Rev. 6: 245.
3. Christensen, H., J. Bachmeier, M. Bisgaard. 2021. New strategies to prevent and control avian pathogenic *Escherichia coli* (APEC). Avian Pathol. 50: 370-381.
4. Dohoo, I.R., S.W. Martin, H. Stryhn. 2009. Veterinary epidemiologic research. Charlotte, P.E.I.: VER, Inc.
5. EFSA AHAW Panel (EFSA Panel on Animal Health and Welfare), S.S. Nielsen, D.J. Bicout, P. Calistri, E. Canali, J.A. Drewe, B. Garin-Bastuji, J.L. Gonzales Rojas, C. Gortazar Schmidt, M. Herskin, V. Michel, M.A. Miranda Chueca, B. Padalino, P. Pasquali, H.C. Roberts, H. Spoolder, K. Stahl, A. Velarde, A. Viltrop, C. Winckler, J. Dewulf, L. Guardabassi, F. Hilbert, R. Mader, F. Baldinelli, J. Alvarez. 2021. Scientific Opinion on the assessment of animal diseases caused by bacteria resistant to antimicrobials: Poultry. EFSA Journal 19: 7114.

6. Ghunaim, H., M.A. Abu-Madi., S. Kariyawasam. 2014. Advances in vaccination against avian pathogenic *Escherichia coli* respiratory disease: potentials and limitations. Vet. Microbiol. 172: 13-22.
7. GRADE. 2015. A GRADE Working Group approach for rating the quality of treatment effect estimates from network meta-analysis. BMJ 350: h3326.
8. Higgins, J.P.T., J. Thomas, J. Chandler, M. Cumpston, T. Li, M.J. Page, V.A. Welch (Eds). 2021. Cochrane Handbook for Systematic Reviews of Interventions, version 6.2 (updated February 2021). Available from www.training.cochrane.org/handbook.
9. Kathayat, D., D. Lokesh, S. Ranjit, G. Rajashekara. 2021. Avian Pathogenic *Escherichia coli* (APEC): An overview of virulence and pathogenesis factors, zoonotic potential, and control strategies. Pathogens 10: 467.
10. Landman, W.J.M., J.H.H. van Eck. 2015. The incidence and economic impact of the *Escherichia coli* peritonitis syndrome in Dutch poultry farming. Avian Pathol. 44: 370-378.
11. Mavridis, D., A. Sutton, A. Cipriani, G. Salanti. 2013. A fully Bayesian application of the Copas selection model for publication bias extended to network meta-analysis. Stat. Med. 32: 51-66.
12. Mavridis, D., N.J. Welton, A. Sutton, G. Salanti. 2014. A selection model for accounting for publication bias in a full network meta-analysis. Stat. Med. 33: 5399-5412.
13. Moher, D., L. Shamseer, M. Clarke, D. Ghersi, A. Liberati, M. Petticrew, L.A. Stewart. 2015. Preferred reporting items for systematic review and meta-analysis protocols (PRISMA-P) 2015 statement. Syst. Rev. 4: 1-9.
14. Nolan, L. K., J. Vaillancourt, N.L. Barbieri, C.M. Logue. 2020. Colibacillosis. In *Diseases of Poultry* 770-830 (Wiley).
15. OIE. 2015. Report of the meeting of the OIE *ad hoc* group on prioritization of diseases for which vaccines could reduce antimicrobial use in animals. Paris, 21-23 April 2015.
16. Paudel, S., M. Hess, C. Hess. 2021. Advances in understanding bacterial diseases in poultry: challenges and perspectives, in optimising poultry flock health. S. De Wit (Ed). *In press*
17. Sanguinetti, V. M., H. Ganshorn, S. Agbese, M.C. Windeyer. 2021. Protocol for a systematic review of disease control strategies used to prevent infectious mortality and morbidity in pre-weaned beef calves. PRISM Repository. Available from https://prism.ucalgary.ca/handle/1880/113381.
18. Sargeant, J.M., M.D. Bergevin, K. Churchill, K. Dawkins, B. Deb, J. Dunn, C.M. Logue, A. Novy, A.M. O'Connor, M. Reist, C.B. Winder. 2019. The efficacy of antibiotics to control colibacillosis in broiler poultry: a systematic review. Anim. Health Res. Rev. 20: 263-273.
19. Swelum, A.A., A.R. Elbestawy, M.T. El-Saadony, E.O.S. Hussein, R. Alhotan, G.M. Suliman, A.E. Taha, B. Ba-Awadh, K.A. El-Tarabily, M.E.A. El-Hack. 2021. Ways to minimize bacterial infections, with special reference to *Escherichia coli*, to cope with the first-week mortality in chicks: an updated overview. Poult. Sci. 100: 101039.