Received: 16 April 2024 | Accepted: 19 May 2024

DOI: 10.1002/jpr3.12102

ORIGINAL ARTICLE

Nutrition

The impact of storage, handling, and treatment on nutritional quality and safety of animal milk: A protocol for the systematic review and meta-analysis

Roba Alwasila[1] | **Alaina Berg**[2] | **Tarah Colaizy**[3] | **Abigail Smith**[4] | **James Evans**[4] | **Mohammad Hassan Murad**[5] | **Zulfiqar Ahmed Bhutta**[6,7,8,9] | **Aamer Imdad**[10]

[1]College of Public Health, The University of Iowa, Iowa City, Iowa, USA

[2]The University of Iowa Carver College of Medicine, Iowa City, Iowa, USA

[3]Division of Neonatology, Stead Family Department of Pediatrics, University of Iowa Carver College of Medicine, Iowa City, Iowa, USA

[4]Health Sciences Library, State University of New York Upstate Medical University, Syracuse, New York, USA

[5]Division of Preventive, Occupational and Aerospace Medicine, Department of Internal Medicine, Mayo Clinic, Rochester, Minnesota, USA

[6]Centre for Global Child Health, Hospital for Sick Children, Toronto, Ontario, Canada

[7]Centre of Excellence in Women and Child Health, Aga Khan University, Karachi, Pakistan

[8]Institute for Global Health and Development, Aga Khan University, Karachi, Pakistan

[9]Department of Nutrition, Joannah and Brian Lawson Centre for Child Nutrition, University of Toronto, Toronto, Ontario, Canada

[10]Division of Gastroenterology, Hepatology, Pancreatology, and Nutrition, Stead Family Department of Pediatrics, University of Iowa Carver College of Medicine, Iowa City, Iowa, USA

Correspondence

Aamer Imdad, Division of Gastroenterology, Hepatology, Pancreatology, and Nutrition, Stead Family Department of Pediatrics, University of Iowa, 200 Hawkins Dr, Iowa City, IA 52242, USA.
Email: aamer-imdad@uiowa.edu

Funding information

WHO (Performance of Work (APW)), Grant/Award Number: 2024/1445457

Abstract

Objectives: Human milk has been shown to reduce severe morbidity in preterm/low-birth-weight infants and is therefore the recommended nutritional source. When infants cannot receive maternal milk, donor human milk (DHM) is recommended. The use of human milk banking facilities is increasing to meet the need for DHM. DHM is unique compared to maternal milk as it must be processed and stored. The processing and storage of animal milk has been more rigorously studied than human milk and can serve as proxy to create DHM banking guidelines.

Methods and Analysis: We will search electronic databases, grey literature, and the websites of relevant international organizations. We will include studies that evaluated the impact of storage, handling, and treatment on the nutritional quality and safety of animal milk. We will not restrict study date, language, or design. If sufficient homogeneity exists between studies, we will conduct a meta-analysis. We will evaluate the methodological quality of each study using the SYRCLE's (Systematic Review Centre for Laboratory Animal Experimentation) risk of bias tool. (1) We will evaluate the overall certainty of the evidence using the Grading of Recommendations Assessment, Development, and Evaluation approach.

Conclusion: In this systematic review and meta-analysis, commissioned by the World Health Organization, we will synthesize the available literature regarding the impact of various storage, handling, and treatment practices on the nutritional quality and safety of animal milk.

KEYWORDS

cow's milk, dairy milk, pasturization

1 | INTRODUCTION

Globally, around 13 million babies are born prematurely (before 37 weeks of pregnancy),[1] and over 20 million with low birth weight (less than 2.5 kg), each year.[2] The American Academy of Pediatrics, World Health Organization, and European Society of Pediatric Gastroenterology, Hepatology, and Nutrition recommend the use of maternal breast milk for feeding preterm and low-birth-weight infants and support the use of donor human milk (DHM) for feeding preterm infants when maternal milk is not available.[3–5]

Concerns have been raised about DHM's nutritional adequacy, as it has been associated with poorer growth outcomes in preterm infants compared with those fed with preterm formula or maternal milk.[3,6,7] This discrepancy has highlighted a need for comprehensive research due to various factors involved in the process of DHM storage, handling, and treatment which may alter its nutrient content.[8] The process of pasteurization and storage has been shown to decrease some bioactive factors and nutrients.[9]

There is limited experience with the collection, processing, and storage of DHM.[8] The processing and storage of animal milk has been more rigorously studied and can serve as a proxy for understanding the processing and storage of human milk.

In this World Health Organization (WHO)-commissioned systematic review and meta-analysis, we aim to synthesize and evaluate the available literature regarding the impact of the methods of storage, handling, and treatment practices on the nutritional quality and safety of animal milk.

What is Known

- Breastmilk is the recommended nutritional source for infants, and the use of donor human milk (DHM) is increasing worldwide.
- The impact of processing and storage of animal milk on its nutritional quality and safety can help understand how processing and storage practices for DHM could affect nutritional quality and safety.
- There has been no systematic review published to date regarding the effect of storage, handling, and treatment practices on the nutritional quality and safety of animal milk.

What is New

- In this World Health Organization-commissioned systematic review and meta-analysis, we will summarize and synthesize the available literature regarding the impact of storage, handling, and treatment practices on the nutritional quality and safety of animal milk.

2 | METHODS

This systematic review is being commissioned by the WHO, conducted following the standard guidelines of Cochrane Collaboration, and reported following the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) guidelines.[10] This systematic review will be completed in consultation with the WHO. The protocol was registered with the Open Science Framework (Registration ID: https://doi.org/10.17605/OSF.IO/BHPXZ)

3 | ELIGIBILITY CRITERIA

3.1 | Type of population

The population of interest encompasses milk derived from animal milk including cow's milk, buffalo milk, goat milk, camel milk, and donkey's milk. We will exclude the studies on synthetic milk, condensed animal milk, dried milk, or other dairy products such as yogurt or cheese.

3.2 | Type of intervention and comparator

The exposure of interest is the storage, handling, and treatment of animal milk. The storage methods include refrigeration, freezing, length of timing of storage, and containers used for storage. Methods for handling include the following: freeze-thaw number of times and

methods, and exposure to light. The methods of treatment include methods for prevention of contamination such as high-pressure pasteurization, ultraviolet C irradiation, high-temperature/short time, Holder pasteurization, heat treatment (time/temperature), or thermoultrasonic methods.[11,12]

We will consider a comparison group without the procedure of interest for storage, handling, and treatment. However, we will not exclude a study because of the lack of a control group.

3.3 | Types of studies

While randomized controlled trials (RCTs) are typically preferred for assessing interventions, it is anticipated that RCTs might be limited or unavailable in the context of our research on storage, handling, and treatment of animal milk. Consequently, our review will adopt a more inclusive approach, encompassing a range of study designs. We will include observational studies and quasi-experimental studies with a control group. We will exclude case studies and opinion pieces.

3.4 | Types of outcomes measures

To be included in the systematic review, studies must report at least one of the following outcomes:

1. Primary outcomes: Nutritional quality: This includes assessing changes in essential nutrients like proteins, fats, vitamins, and minerals in animal milk.
   a. Macronutrient: Quantity of protein (g/dL) (continuous).
   b. Macronutrient: Quantity of carbohydrate (g/dL) (continuous).
   c. Macronutrient: Quantity of fat (g/dL); Fat oxidation (continuous).
   d. Micronutrient: Quantity of vitamins such as vitamins A, D, and B_{12} (μg/mL) (continuous).
   e. Micronutrients: Quantity of minerals such as iron, zinc, phosphorus, potassium, sodium, magnesium, and calcium (g/kg) (continuous).
   f. Immunoglobulins: Quantity of immunoglobulin (Ig) A, IgG, and IgM (g/L) (continuous).
   g. Lipase: Quantity of lipase in milk (lipoprotein lipase, etc.): (g/L) (continuous).
2. Secondary outcomes: Safety: We will evaluate the safety of animal milk, focusing on the presence of contaminants, pathogens, or any other factors that could pose health risks, influenced by various handling, storage, and treatment practices.
   a. Presence of contaminants (dichotomous).
   b. Presence of pathogens (dichotomous).
   c. Presence of alkaline phosphatase (dichotomous): this is a heat-labile enzyme that serves as a marker of unsuccessful pasteurization as it is completely wiped out with pasteurization.

3.5 | Information sources

Electronic searches will be conducted across multiple databases, including PubMed, EMBASE, the Cochrane Central Register of Controlled Trials, Web of Science, CINHAL, Scopus, and the WHO Global Index Medicus. We will impose no limitations on the searches in terms of outcomes, study design, publication status, date, or language. Our proposed search strategy for PubMed is detailed in File S1.

3.6 | Other sources

We will explore ClinicalTrials.gov for any ongoing or unpublished studies. The search will extend to websites of relevant international organizations like WHO, United Nations Children's Fund (UNICEF), the Global Alliance for Improved Nutrition, and the International Food Policy Research Institute. This includes veterinary-focused databases such as Centre for Agriculture and Biosciences International (CAB) Abstracts and VetMed Resource to cover aspects of veterinary medicine relevant to our study, and milk bank associations like the Human Milk Bank Association of North America and the European Milk Bank Association. We will also meticulously review the reference lists of previously published reviews and recent studies to identify any further eligible studies.

4 | STUDY RECORDS

4.1 | Study selection process

All studies identified in the electronic search will be exported to Covidence for screening.[13] Two authors will independently screen all titles and abstracts to identify studies relevant to the research question. Then, those studies identified as relevant will be screened with a full-text review to determine if they are eligible for inclusion. A third author will be available to resolve any conflicts during screening. We will include a list of studies that were excluded after full-text screening and provide detailed reasons for their exclusion.

4.2 | Data collection and management process

Studies deemed eligible for inclusion during the full-text review step will then proceed with full data extraction. Two authors will independently extract data for each study, and any conflicts will be resolved by discussion

with the help of the senior author on the team as needed. The following information with be extracted for each study: first author, publication date, study site, study year, study population, intervention, comparison, outcomes, and risk of bias. The methodological quality of each study will be assessed using the SYRCLE's (Systematic Review Centre for Laboratory Animal Experimentation) risk of bias tool.[14] We will contact the authors if data for exposure or outcomes is missing from reports. For continuous outcomes, if a study does not report a standard deviation (SD) for a mean and this cannot be calculated from the reported data and information is unavailable from authors, we will use an SD reported from a similar study for the same outcome.

4.3 | Data synthesis

We will report a table of the characteristics of the included studies, and a separate table for the summary of findings for the primary outcomes. We will also report narrative summaries of the results of included studies regarding the outcomes of interest. Studies from low- and middle-income countries will be reported separately from those from high-income countries. We will use the Grading of Recommendations Assessment, Development, and Evaluation (GRADE) approach to evaluate the overall certainty of the evidence, using the GradePro software.[15] We will report the results of the GRADE assessment in the table which summarizes the findings for primary outcomes, with each finding characterized as very low, low, moderate, or high certainty.

We will conduct and report meta-analyses when data is available from more than one study and there is sufficient clinical and methodological homogeneity between studies. Meta-analyses will report dichotomous outcomes with relative risk and corresponding 95% confidence intervals. We will report continuous outcomes with mean effect size estimates and corresponding 95% confidence intervals. We will calculate the statistical heterogeneity of effect sizes using the χ^2 and I^2 statistics. Funnel plots and regression tests for funnel plot asymmetry will be used to assess small studies and publication bias if/when the meta-analysis includes 10 or more studies. We will use the statistical software RevMan and Stata to complete the analyses.[16,17]

5 | SUBGROUP ANALYSES

1. Storage conditions: temperature, duration.
2. Handling practices: transportation methods and hygiene standards.
3. Treatment techniques: various treatment methods (e.g., pasteurization, homogenization).
4. Time: time between milking and processing on milk quality and safety.

6 | SENSITIVITY ANALYSIS

1. Meta-analysis with fixed effect model.

7 | ETHICS

This systematic review and meta-analysis would not directly involve human subjects and was considered a non-human subject research by Institutional Reviw Board of University of Iowa (IRB no 202406271).

8 | DISSEMINATION

The findings will be published in a peer-reviewed journal and intended to inform the WHO recommendations for human milk banking.

9 | CONCLUSION

This systematic review and meta-analysis will synthesize the available literature regarding the effect of storage, handling, and treatment of animal milk on its nutritional quality and safety. This information will be useful to guide the WHO's development of global standards for the storage, handling, and treatment of DHM in human milk banks.

ACKNOWLEDGMENTS

This systematic review is funded by WHO (Performance of Work (APW) Reference No. 2024/1445457).

CONFLICT OF INTEREST STATEMENT

Dr. Colaizy is the research director of the research committee of the Human Milk Banking Association of North America (HMBANA). She contributes to this position on a volunteer basis and has never received any honorarium from HMBANA. This proposal was not discussed in the research committee of HMBANA, and HMBANA did not have any role in the design of this study. The remaining authors declare no conflict of interest.

ORCID

Alaina Berg http://orcid.org/0009-0008-4577-9253
Aamer Imdad http://orcid.org/0000-0002-7026-0006

REFERENCES

1. Ohuma EO, Moller AB, Bradley E, et al. National, regional, and global estimates of preterm birth in 2020, with trends from 2010: a systematic analysis. *Lancet*. 2023;402(10409):1261-1271.
2. Blencowe H, Krasevec J, de Onis M, et al. National, regional, and worldwide estimates of low birthweight in 2015, with trends from 2000: a systematic analysis. *Lancet Global Health*. 2019;7(7):e849-e860.

3. Arslanoglu, S, Corpeleijn W, Moro G, et al. Donor human milk for preterm infants: current evidence and research directions. *J Pediatr Gastroenterol Nutr*. 2013;57(4):535-542.
4. Section on Breastfeeding. Breastfeeding and the use of human milk. *Pediatrics*. 2012;129(3):e827-41.
5. WHO. *WHO Recommendations for Care of the Preterm or Low-Birth-Weight Infant*. WHO; 2022.
6. Grummer-Strawn LM, Reinold C, Krebs NF, Centers for Disease Control and Prevention. Use of World Health Organization and CDC growth charts for children aged 0-59 months in the United States. *MMWR Recomm Rep*. 2010;59(RR-9):1-15.
7. Quigley M, Embleton ND, McGuire W. Formula versus donor breast milk for feeding preterm or low birth weight infants. *Cochrane Database Syst Rev*. 2019;7(7):CD002971.
8. Colaizy TT. Effects of milk banking procedures on nutritional and bioactive components of donor human milk. *Semin Perinatol*. 2021;45(2):151382.
9. Perrin MT, Belfort MB, Hagadorn JI, et al. The nutritional composition and energy content of donor human milk: a systematic review. *Adv Nutr*. 2020;11(4):960-970.
10. Tricco AC, Lillie E, Zarin W, et al. PRISMA Extension for Scoping Reviews (PRISMA-ScR): checklist and explanation. *Ann Intern Med*. 2018;169(7):467-473.
11. Astráin-Redín L, Skipnes D, Cebrián G, Álvarez-Lanzarote I, Rode TM. Effect of the application of ultrasound to homogenize milk and the subsequent pasteurization by pulsed electric field, high hydrostatic pressure, and microwaves. *Foods*. 2023;12(7):1457.
12. Rankin SA, Bradley RL, Miller G, Mildenhall KB. A 100-year review: a century of dairy processing advancements-pasteurization, cleaning and sanitation, and sanitary equipment design. *J Dairy Sci*. 2017;100(12):9903-9915.
13. Covidence systematic review software [Computer Program]. Melbourne: Veritas Health Innovation.
14. Hooijmans CR, Rovers MM, de Vries RB, Leenaars M, Ritskes-Hoitinga M, Langendam MW. SYRCLE's risk of bias tool for animal studies. *BMC Med Res Methodol*. 2014;14(43):43.
15. GRADEpro GDT: Gradepro Guideline Development Tool [Computer Program]. McMaster University (developed by Evidence Prime, Inc.).
16. Review Manager WEb (RevMan Web). [Computer Program]. Version 2020. The Cochrane Collaboration.
17. Stata 14 [Computer Program]. Version 14. StataCorp; 2015.

SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

How to cite this article: Alwasila R, Berg A, Colaizy T, et al. The impact of storage, handling, and treatment on nutritional quality and safety of animal milk: a protocol for the systematic review and meta-analysis. *JPGN Rep*. 2024;5:342-346. doi:10.1002/jpr3.12102