

CONTAMINATION BY ESCHERICHIA COLI IN TOTAL MIXED RATIONS WITH A HIGH PROPORTION OF FORAGE CACTUS



Juliana Silva de Oliveira¹, Edson Mauro Santos¹, Danillo Marte Pereira¹, Gilvânia Avelino da Costa¹ DZ/UFPB Department of Animal Science, Federal University of Paraíba, Areia, PB, Brazil

Introduction

Importance of Forage Cactus

- >>> Highly relevant in arid and semi-arid regions.
- >>> Source of energy and hydration for ruminants due to:
 - ✓ Low dry matter content.
 - ✓ High non-fibrous carbohydrate concentration.

Nutritional Limitations

- >>> Low levels of neutral detergent fiber (NDF).
- >>> Excessive use may cause gastrointestinal disorders in ruminants.

Microbiological Risk & Feed Management

- >>> High forage cactus inclusion may lead to contamination by pathogenic microorganisms (e.g., *E. coli*).
- >>> Contamination can occur during:
 - ✓ Mixing of ingredients.
 - ✓ Distribution of the diet.
- >>> Farm-level feed management directly influences TMR hygienic quality.

Effect of the Ensiling Process

- >>> TMR silage based on forage cactus and *Gliricidia* showed:
 - ✓ Lower microbial diversity.
 - ✓ Dominance of beneficial bacteria (e.g., *Bacillus* genus).
 - ✓ No detection of harmful microbes like enterobacteria.
- Q Thus, we hypothesize that the fermentation process inhibits the growth of potentially pathogenic microorganisms, such as Escherichia coli (E. coli), in diets with high proportions of forage cactus.

(Pereira et al., 2025).

Objectives

The aim of this study was to quantify the *E. coli* population in TMR containing high proportions of forage cactus and subjected to different feeding methods: *in natura* and silage.

Materials and Methods

- Location: Forage Laboratory of the Center of Agricultural Sciences (CCA),
 Federal University of Paraíba (UFPB) − Campus II, Areia, Paraíba, Brazil.
- **→ Design**: Completely randomized, $2 \times 2 \times 2$ factorial
- ✓ Buffel grass (*Cenchrus ciliaris* L.) hay levels: 7.5% and 45%
- ✓ With/without fecal contamination
- ✓ Feed type: fresh TMR and TMR silage
- ✓ Time points: 0, 6, 12, and 24 hours exposure
- → Replicates: 4 per treatment
- → TMR silage pH at opening: average of 4.7

→ Fecal Contamination Procedure

- >>> Feces collected from goat pens to simulate *E. coli* contamination
- >> 10 g of fresh feces diluted in 90 mL sterile distilled water
- >>> Homogenized and filtered through sterile gaze
- >>> Aqueous extract used to inoculate TMR

→ Microbial Quantification

- >>> Method: Based on González and Rodríguez (2003)
- >>> Medium: EMB Levine Agar (Kasvi, K25 1050)
 - ✓ Selective for gram-negative bacteria
 - ✓ E. coli identified by metallic green colonies
- >>> Only plates with 30–300 CFU considered

Complementary Analyses

- ✓ Average pH of silage upon opening: 4.7.
- ✓ Statistical analysis: Tukey's test at 5% probability.

Results

E. coli Population

- >>> Fresh TMR showed an overall increase in *E. coli* counts after 24h exposure, exceeding **6.85** Log CFU/g, regardless of hay level or contamination.
- >>> TMR Silage consistently maintained lower *E. coli* counts, indicating the protective effect of ensiling.
- >> 7.5% Hay Content
- >>> Contaminated Fresh TMR:
 - ✓ Sharp increase after 6h (+1.35 Log CFU/g from time 0)
 - ✓ Peak at 24h: 6.92 Log CFU/g
- >> Silage TMR:
 - ✓ Initial: 3.32 Log CFU/g \rightarrow reduced to 1.97 Log CFU/g at 24h
- >>> Uncontaminated Fresh TMR:
 - ✓ Final count at 24h: 7.35 Log CFU/g
- >>> Uncontaminated Silage TMR:
 - ✓ Final count at 24h: 3.70 Log CFU/g

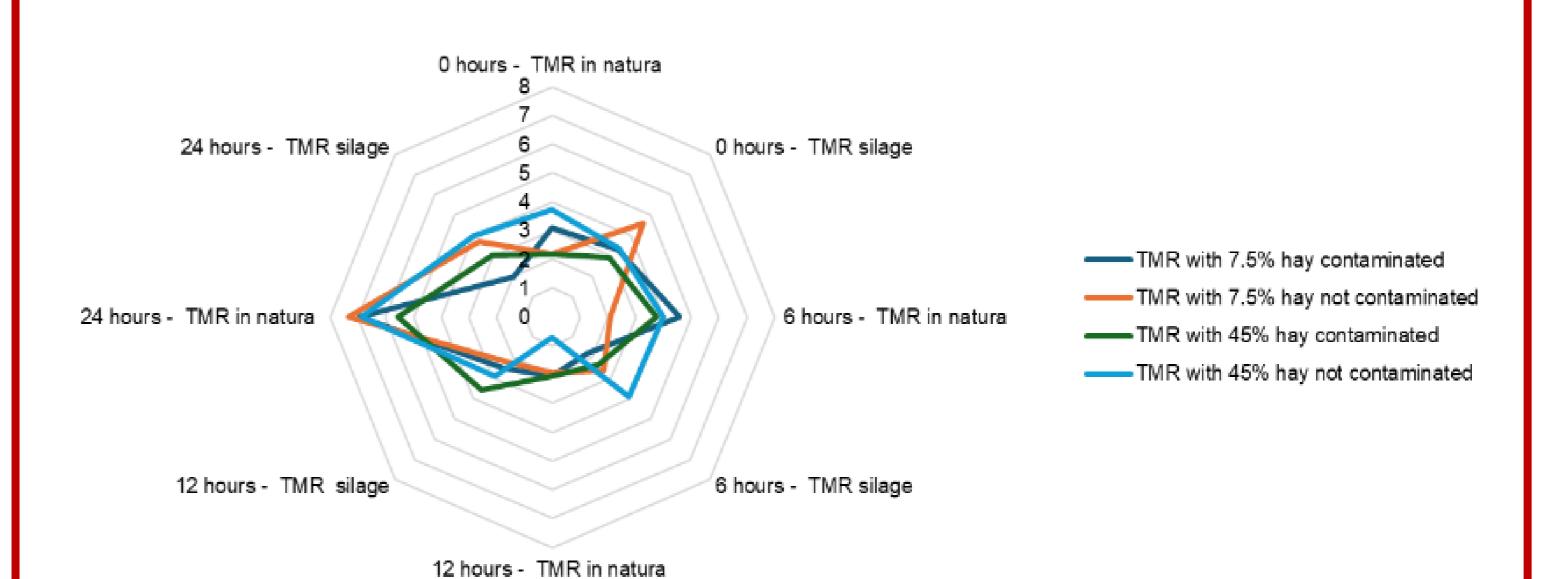


Figure 1. Escherichia coli population (Log CFU/g) in a total mixed ration with high proportions of forage cactus, containing different levels of buffel grass hay (7.5 and 45%), contaminated and non-contaminated, evaluated over the time of exposure to the environment (0, 6, 12 and 24 hours) in two forms of feed management: in natura and silage.

- **45% Hay Content**
- >>> Contaminated Fresh TMR:
 - ✓ Time 0: 2.18 Log CFU/g \rightarrow **5.57 Log CFU/g** at 24h
- >>> Silage TMR: Maintained counts below 3.03 Log CFU/g
- >>> Uncontaminated Samples: Same trend lower counts in silage TMR

Conclusions

✓ The ensiling process plays an important role in reducing microorganisms with pathogenic potential such as *E. coli* in TMR, helping to maintain its hygienic quality. In addition, contamination influences the *E. coli* population, reinforcing the importance of hygiene during the mixing and feeding process. Based on the obtained results, the adoption of TMR silage with 7.5% hay is recommended as the most effective feeding strategy for reducing gastrointestinal tract disorders in ruminants.

Acknowledgements

✓ The authors acknowledge to CNPq, CAPES, FAPESQ, and INCT-CA for financial support.

References

Pereira, D. M., Oliveira, J. S., Santos, F. N. S., Macêdo, Alberto J. S., Gomes, P. G. B., Pereira, L. S., Silva, E. S., Cruz, G. F. L., Perazzo, A. F., Santos, E. M. (2025). Forage cactus as a modulator of forage sorghum silage fermentation: An alternative for animal feed in drylands. *Chilean journal of agricultural research*, 85(1), 47-56.