

Ewes' Milk Urea Concentration methods' optimization, by difference in pH and Mid-Infrared Spectroscopy (MIRS)

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Abstract

Currently, in France, analytical methods used for ewes' Milk Urea Concentration (MUC) rely on cow's methods. However, both matrix are different, as fat and protein content are up to twice higher in ewe milk compared to cow milk. Moreover, MUC variability seems to be higher in ewe milk (from 50 to 1000 mg/l) than in cow milk (from 150 to 500mg/l), hence the question about the analysis methods adequation in ewe milk.

This project aimed at optimizing and or adapting the reference and routine methods used to determine ewe's MUC, currently based and calibrated from cows milk.

The reference method was validated as such; calibration matched for cow milk as well as for sheep milk. Concerning the routine method, creating a new specific predictive equation based on individual and bulk tank ewes' milks neatly improved the performance compared to the existing cows' milk predictive MUC equation.

Keywords: Milk Urea Content, ewes, analytical methods, MIRS, predictive equation.

Milk Urea Concentration (MUC) is a simple indicator reflecting ruminants diet balance, protein concentration and energy in the sheep diet (Bocquier et Caja, 1999, Cannas *et al.*, 1998, Gholi Ramin *et al.*, 2010). MUC also varies according to the fibers or non-fibers carbohydrate sources provided (Giovanetti *et al.*, 2019). Its monitoring helps optimizing the feed cost by limiting waste, hence the economical stake, knowing that the feeding expenses represent from 62% to 72% of the operational expenses in the Atlantic-Pyrenees and North Occitany French sheep dairy areas (Inosys Réseaux d'élevage, 2021). Furthermore, MUC excess exerts negative impacts on reproductive efficiency (Giovanetti *et al.*, 2019). Lastly, according to the type of dairy products, MUC excess might be associated to weaker milk technological ability and final products defects. Thus, MUC is a composite, costless indicator that breeders can easily use in herd management.

Methods currently used to measure Milk Urea Concentration (MUC) in ewe's milk rely on cow's milk methods. However the dairy matrices between those species differ. Thus, this project aimed at:

Introduction

1. Verifying the reference method adequation (enzymatic method using difference in pH ISO 14637 / IDF 195:2004),
2. Verifying the routine method, based on a prediction from cow milk Mid Infra-Red Spectra (MIRS) to determine ewe's milk urea concentration,
3. Optimising, the routine method by developing a specific ewe's milk predictive SMIR equation for MUC.

Material and methods

Concerning the reference method, the repeatability, reproducibility and accuracy were evaluated on 25 samples of individual ewe milk, from Lacaune and Basco-béarnaise breeds, analysed by Actalia Cécalait (Poligny – France) from December 2021 to January 2022.

Concerning the routine method, the analyses were conducted by the interprofessional lab Agrolab's (Aurillac, France), every month from January to June 2022. The data included 2 datasets:

- 260 samples from individual ewe milk (a single flock, for each area: Corse, Nouvelle-Aquitaine and Occitanie, representing around 20 animals per month and per flock),
- 401 samples from bulk tank milk (around 20 flocks respectively for each area).

This original protocol enabled to maximize the existing ewe milk variability, as recommended by De Marchi *et al.* (2014). It was meant to optimize the ewes' MUC predictive model, as the seasonal, geographical, breeds (Lacaune, Basco-béarnaise, Manech tête rousse et noire, Corse), intra and inter-flock variability were taken into account. Every Verimilk was measured by infrared on Foss electric analysers and compared to the reference method (ISO 14637 / IDF 195:2004), by Agrolab's Aurillac. Then, the specific ewe milk predictive MUC equation was established by Partial Least Square regression as described by El Jabri *et al.* (2019).

Results

Reference method

The reference method was validated as such; calibration matched for cow milk as well as for sheep milk, regarding the performances of repeatability, reproducibility and accuracy.

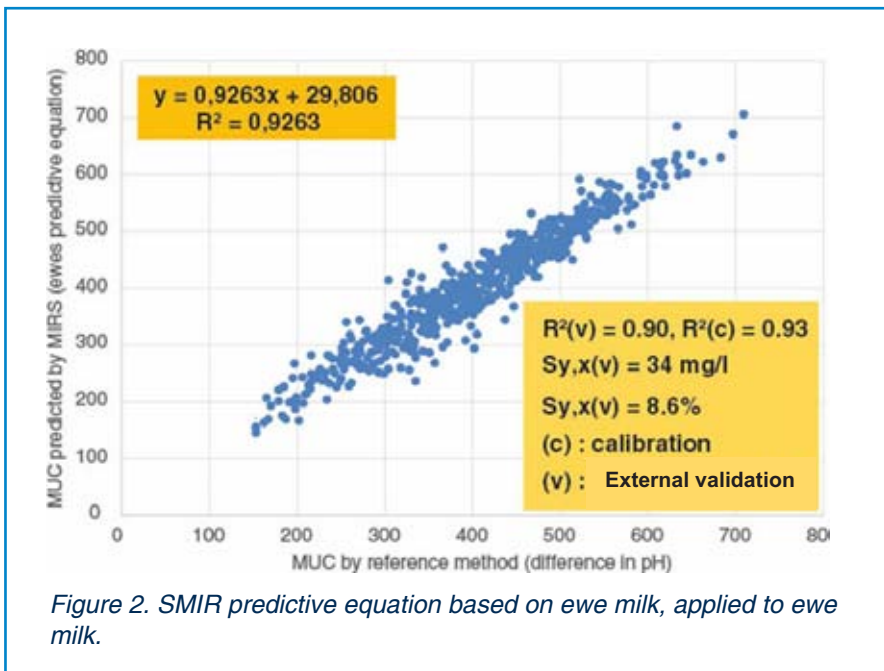
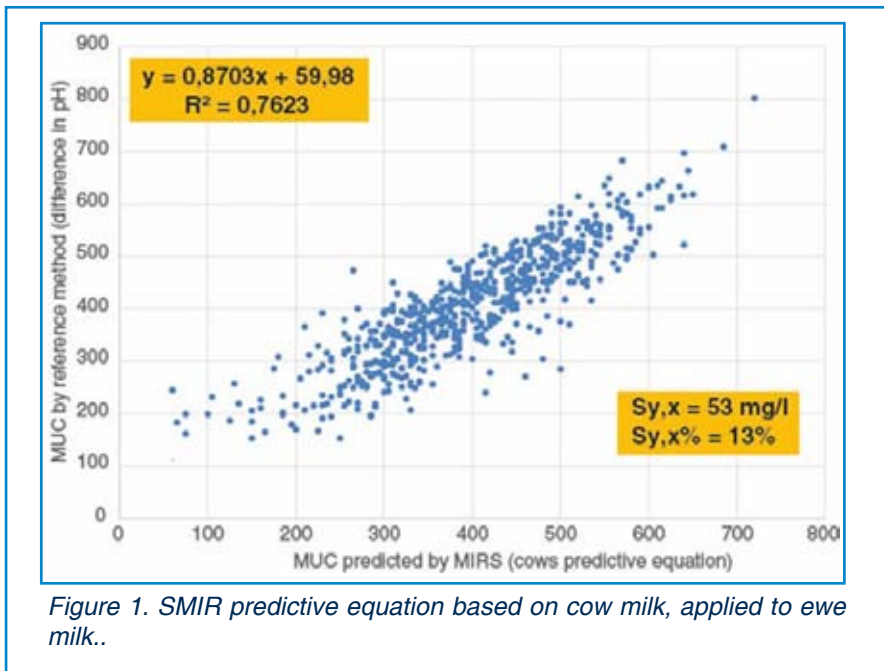
Routin method

The cow's MUC predictive equation did not give a good precision for ewe's milk, as it only accounted for 76% of the ewe's milk variability (coefficient of determination, $R^2= 0.76$). The Residual Standard Deviation RSD ($S_{y,x}$) was then of 53 mg/l MUC vs. 35 mg/l for the cow's MUC predictive equation applied to cow milk.

Table 1. Comparison of Residual Standard Deviation (RSS) on MUC, according to the predictive models and the ruminant species

| Model (type of predictive SMIR equation) | Predictive equation based on cow milk | Predictive equation based on cow milk | Predictive equation based on cow milk | Predictive equation based on ewe milk |
|--|---------------------------------------|---------------------------------------|---------------------------------------|---------------------------------------|
| Analysed milk | cow | goat | ewe | ewe |
| RSD ($S_{y,x}$) | 35 mg/l ¹ | 40 to 59 mg/l ¹ | 53 mg/l | 34 mg/l |

¹ Actalia Cécailait, Trossat P., 2014, MUC evaluation in goats' milk by MIRS method (internal report).



With the specific ewe's milk model, applied to individual and bulk tank milk samples, the performance was equivalent to the cow's milk model applied to cow milk samples. A greater variability was included in the ewe's milk predictive SMIR equation (coefficient of determination of external validation, $R^2(v) = 0.90$), applied to ewe's milk samples. The RSD in external validation ($S_{y,x(v)}$) was improved 34 mg/l, with the specific ewe's predictive SMIR equation vs. 53 mg/l with the cow's milk predictive equation applied to ewe's milk samples.

Conclusion

MUC management can closely be related to ewe's feed optimization, animal health, and final dairy products quality.

Thus, developing specie-specific MUC predictive model by SMIR would neatly improve urea's precision in routine analysis for ewe's milk, may it be individual milk or bulk tank milk samples.

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