

# Effects of potential evapotranspiration on condensed tannin and milk production potential in four grassland mixtures

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## Abstract

Dairy farming faces challenges of summer fodder scarcity and methane emissions. To address these issues, one may incorporate plants rich in condensed tannins (CT) into temporary grassland mixtures. However, knowledge regarding the link between CT, milk production and climate remains limited in a field context. We conducted an experiment with four grassland mixtures (grass, grass and legumes, grass and plants rich in essential oils, grass and plants rich in tannins) replicated four times and grazed over six rotations. CT content, dry matter yields, and botanical composition were analysed. We calculated milk production potential (MPP) based on nutrient analysis of the plant material and obtained potential evapotranspiration (PET) data. No differences between mixtures were observed during rotation where PET was low whereas CT content in the tannin mixture was higher in high PET rotation. MPP remained stable over time for the legume, the essential oil and the tannin mixtures but decreased significantly for pure grass. CT content in the tannin mixture was strongly correlated with *Lotus corniculatus* abundance. Our study suggests that plants produce most CT during peak PET in summer, but forage production is highest during lower PET in spring. Thus, tannin-rich mixtures may primarily mitigate methane emissions in summer.

**Keywords:** multi-species grassland, condensed tannins, evapotranspiration, *Lotus corniculatus*

## Introduction

Pastures characterized by a higher diversity of plant species have been proposed to facilitate increased nutrient absorption by ruminants and promote better cattle health (Distel *et al.*, 2020). This effect is due in part to the absorption of plant secondary metabolites such as condensed tannins (CT). These tannins, or proanthocyanidines, are polyphenolic compounds found in various plant species, in which they act as defence mechanisms against biotic (Barbehenn and Constabel, 2011) and abiotic stress such as drought (Gourlay *et al.*, 2022). CT could potentially reduce methane emissions from cattle during digestion by inhibiting certain microorganisms involved in methane production in the rumen (Wang *et al.*, 2015). On-field studies on this topic remain scarce, highlighting the importance of comparing forage production and quality in grassland mixtures with tannin-rich plants versus conventional blends. We anticipated that a grassland mix enriched with tannin-rich plants would consistently display higher CT levels throughout the vegetation season, given the intentional selection of tannin-rich plant species. Concurrently, we expected these CT levels to correlate with potential evapotranspiration (PET). In addition, traditional blends, selected for their productivity, were predicted to sustain elevated productivity across the entire vegetation period.

## Materials and methods

The study was conducted during a single growing season from the end of March to October 2022 in Frick, Switzerland (47°30'51" N 8°1'26" E). The experimental field, covering approximately 1.3 hectares, was divided into 16 plots. In the autumn of 2021, four grassland mixtures were randomly allocated and sown on four plots each: grass, grass and legumes, grass and plants rich in essential oils, grass and tannin-rich plants (in the following referred to as Grass; Legume; Tannin and Oil mixtures). Throughout six grazing rotations by a herd of 23 dairy cows, we closely monitored the botanical composition, the dry matter yield (DM in kg ha<sup>-1</sup>) and the CT content (in g (kg DM)<sup>-1</sup>) of each plot. Furthermore, we calculated the milk

production potential (MPP in kg ha<sup>-1</sup>) of a hypothetical dairy cow for each rotation using the INRA equation (INRA, 2010). PET was assessed (mm/day average over each rotation period, Turc method, [agrometeo.ch](http://agrometeo.ch)). All statistical analyses were conducted using R. Given the non-normal distribution of our data, we employed Spearman tests to assess the correlation between CT levels and PET, between MPP and PET (for each mix) and between the relative abundance of supposedly tannin rich species (only in Tannin mix) and CT levels. Wilcoxon pairwise comparison tests were executed to discern variations in CT and MPP between the mixtures throughout the six rotations.

## Results and discussion

The PET exhibited an increase during the first four rotations, spanning from the end of March to the end of June. Rotations 4 and 5 were conducted during the period when PET reached its peak. Subsequently, there was a decline observed from rotation 5 (August) to rotation 6 (October) (Figure 1). There were significant positive correlations between CT and PET in each of the four mixtures (Table 1). The strongest correlation was observed in the Tannin mix.

Significant differences in CT levels among the four mixtures were observed only at rotation 5, where the Legume mixture exhibited significantly higher CT levels than the Grass mixture, and the Tannin mixture demonstrated significantly higher CT levels than each of the other three (Figure 2a). In the Grass mixture, there was a significant negative correlation between MPP and PET ( $\rho=-0.47$ ,  $S=3370$ ,  $P=0.02197$ ). Significant differences in MPP among the four mixtures were observed only at rotation 1, where the Grass and the Legume mixtures exhibited significantly higher MPP than the Tannin and the Oil mixtures (Figure 2b). These findings suggest that the initial highest yields of the Grass mixture were later negatively impacted by the PET. It also appeared that the MPP of the Grass mixture was lower in

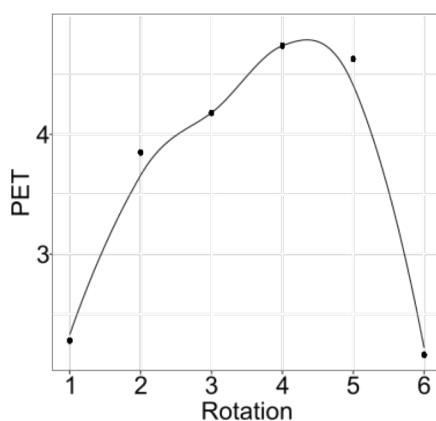


Figure 1. Variation of the PET along the 6 rotations.

Table 1. Results of the Spearman correlation tests between CT and PET in each of the mixtures.

Mix	Spearman correlation coefficient ( $\rho$ )	S	P-value
Grass	0.46	1237.8	0.023*
Legume	0.55	1043.5	0.006**
Tannin	0.84	362.56	<0.001***
Oil	0.41	1359.7	0.047*

\* $P \leq 0.05$ , \*\* $P \leq 0.01$ , \*\*\* $P \leq 0.001$ .

the rotation 5 than for the other mixtures but the correlation was not more than marginally significant. Within the potentially tannin-rich plants of our Tannin mixture, only the relative abundance of *Lotus corniculatus* and the CT levels correlated significantly ( $\rho=0.82$ ,  $S=419.59$ ,  $P=1.074e-06$ ).

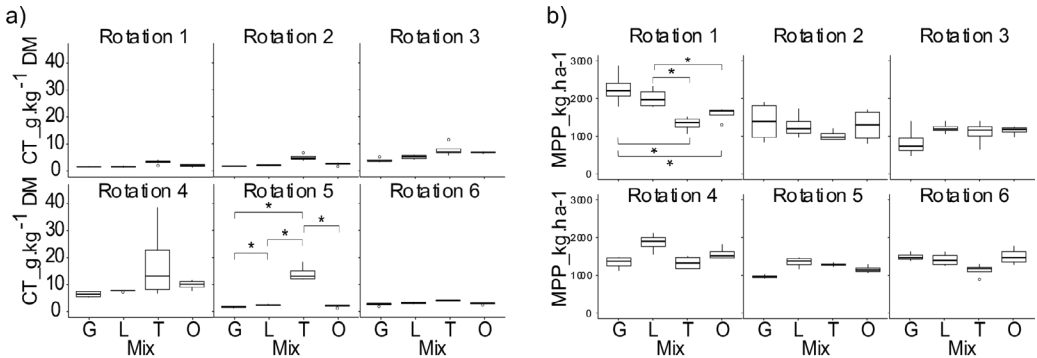


Figure 2. CT levels in g (kg DM)<sup>-1</sup> (a) and MPP (b) in each of the mixtures and rotations. Significant P-values from the Wilcoxon pairwise comparison tests are depicted by the corresponding asterisk, with \* $P\leq 0.05$ , \*\* $P\leq 0.01$ , \*\*\* $P\leq 0.001$ .

## Conclusion

Conforming to our hypothesis, CT were correlated with PET in all mixtures. However, in contradiction with what we expected, the Tannin mixture showed higher CT levels than the other mixture only in rotation 5. This result could be explained by the higher *Lotus corniculatus* abundance in rotation 5 as this species has the ability to sustain higher PET. This implies that using tannin-rich species to modulate digestive processes in the rumen, i.e. reduce methanogenesis or improve protein digestibility, may only be effective during periods with high PET. Surprisingly, the Grass and Legume mixtures displayed higher MPP than the other mixtures only at rotation 1. This could be partially explained by the adverse impact of PET on Grass, along with the Tannin and Oil mixtures' ability to maintain yields despite higher PET. In conclusion, our results underscore the significance of climate and timing in designing grassland systems that offer multiple services.

## Acknowledgements

We express gratitude to Samuel Imboden for his work in the lab and colleagues for their precious work in the field. This project is supported by the Stiftung Edith Maryon.

## References

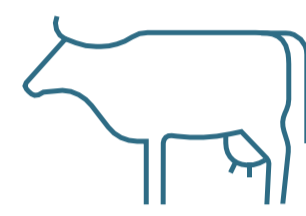
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## Effects of potential evapotranspiration on condensed tannin and milk production potential in four grassland mixes

### Introduction

It is necessary to adapt grassland to changes in climate and consumer expectations in order to :

- Maintain high-quality grass production in summer,
- Reduce methane emissions from Swiss herds.
- Promote the well-being and health of the animals



One approach could be to incorporate plants rich in condensed tannins (CT) into temporary grassland mixes but knowledge remains limited

### Methods

4 mixtures to be sown in autumn 2021 on a 1.3 ha field in Frick (Switzerland):

- Grass - G
- Grass and legumes - L
- Grass and Tannin-rich plants - T
- Grass and essential oil-rich plants - O

Measurements from April to October 2022 :

- Yields
- Potential evapotranspiration (PET)
- Condensed tannin (CT) content
- Milk production potential (MPP)

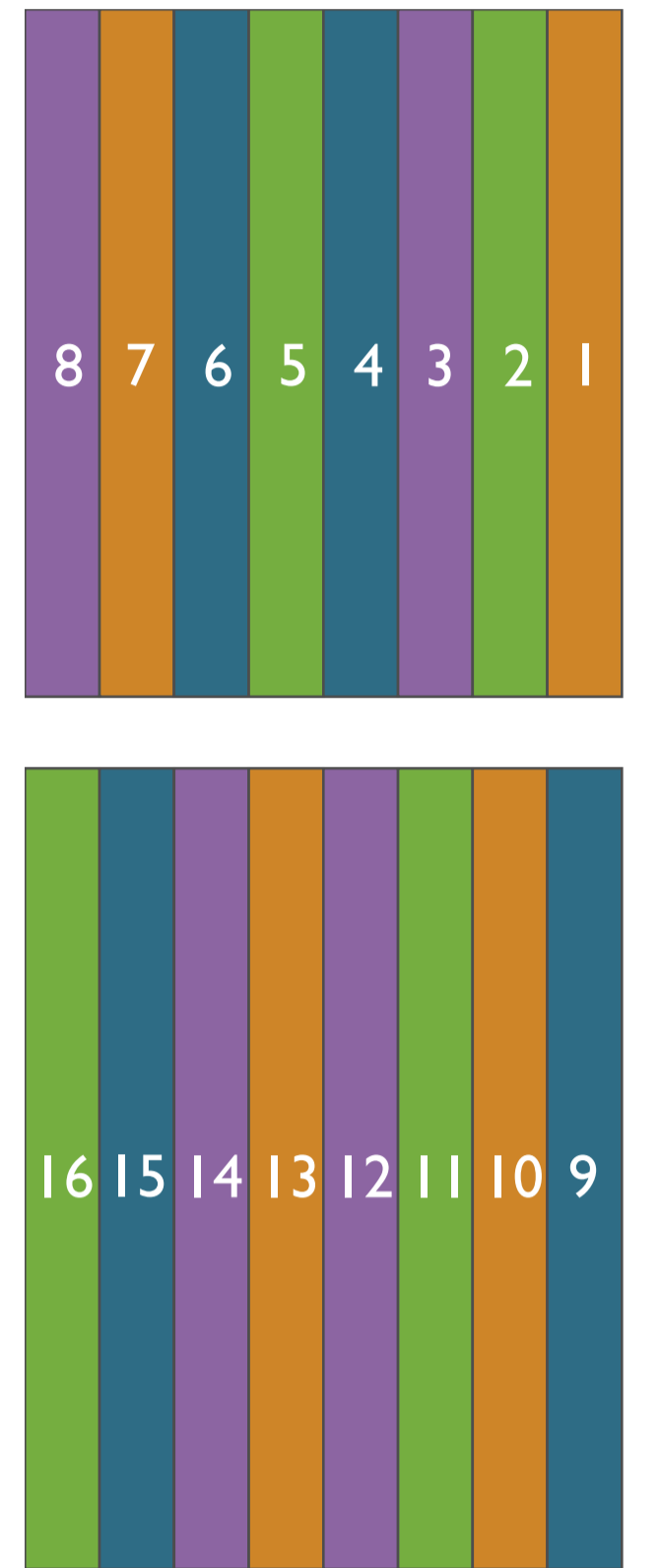


Fig. 1: Design of the experimental field

### Results and Discussion

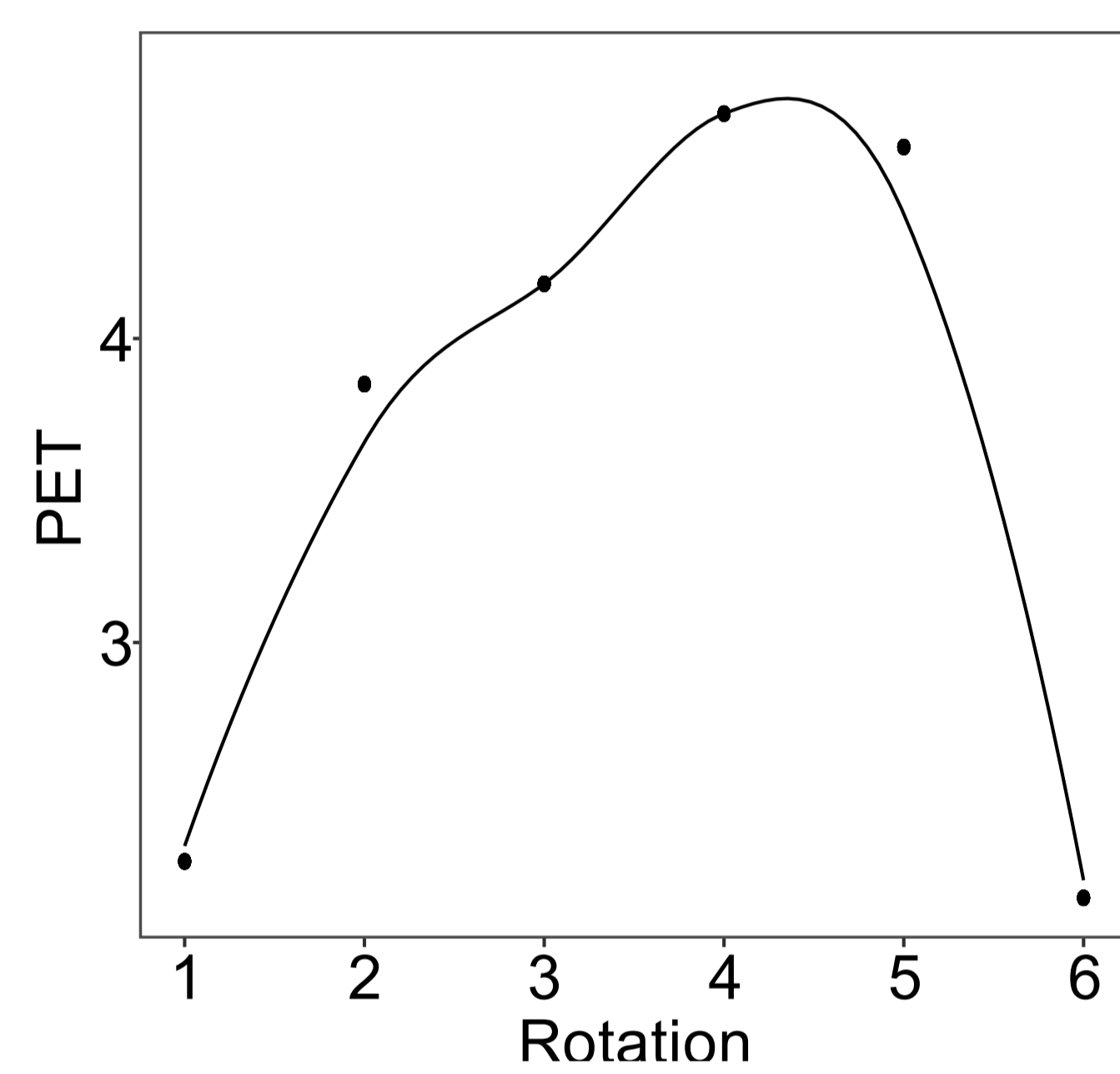


Fig. 2: Variation of the PET along the 6 rotations

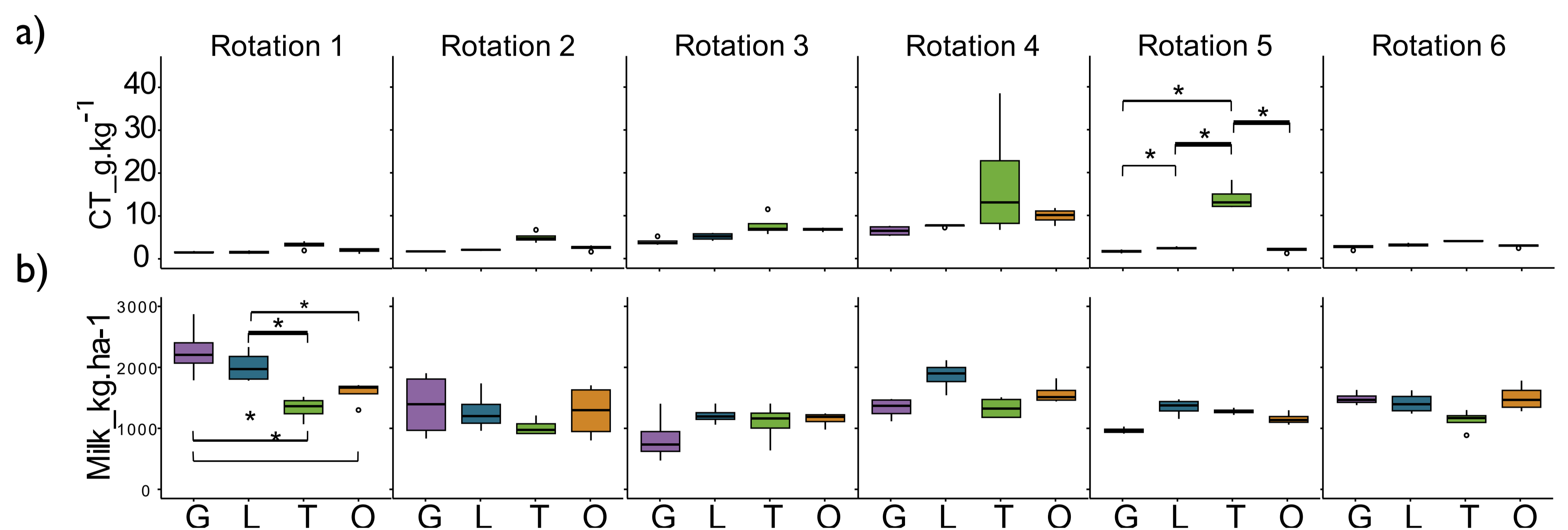


Fig. 3a: CT levels in g.kg<sup>-1</sup> in each of the mixes and rotations

Fig. 3b: MPP in each of the mixes and rotations

Significant P-values from the Wilcoxon pairwise comparison tests are depicted by the corresponding \*, with \*  $P \leq 0.05$ , \*\*  $P \leq 0.01$ , \*\*\*  $P \leq 0.001$ .

- The Grass and legumes mixture enabled the highest MPP on average over the season
- ...even in drier conditions (Rotation 5)
- There were significant positive correlations between CT and PET in each of the four mixes (Table 1). The strongest correlation was observed in the Tannin mix
- The tannin content is highest in summer (higher PET rotation) in the mixture, which is rich in tannin-containing species.
- No significant differences between the CT content of the tannin mix and of the other mixes outside the high PET rotation

Mix	$\rho$	S	P-value
Grass	0.46	1237.8	0.023*
Legume	0.55	1043.5	0.006**
Tannin	0.84	362.56	<0.001***
Oil	0.41	1359.7	0.047*

Tab.1: Results of the Spearman correlation tests between CT and PET in each of the mixes

### Conclusion

Our results suggest that using tannin-rich species to modulate digestive processes in the rumen, i.e. reduce methanogenesis or improve protein digestibility, may only be effective during periods with high PET.

#### Acknowledgements

We warmly thank Samuel Imboden for his help in conducting preparative work in the lab, and all colleagues who promptly helped us in the field. This project received support from the Stiftung Edith Maryon.

#### Projektpartner:

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