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## Meeting the future demands for grassland production

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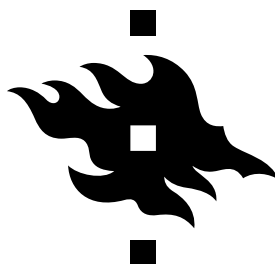
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# Application of the new German protein evaluation system for horses in forage from species-rich meadows

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## Abstract

The German protein evaluation system for horses is based on the pre-caecal digestible crude protein (pcdCP). Forage contents of pcdCP are available for grass-based swards, while those from species-rich swards are scarce. Grass species are low in tannin contents and interactions during protein digestion are unlikely. However, this may be relevant in forage from species-rich grasslands containing tanniferous forage species. Therefore, contents of pcdCP were quantified in fresh, wilted or ensiled forages from species-rich mountain swards and correlations with the content of condensed tannins (CT) were determined. The forages were obtained from three long-term fertiliser experiments (unfertilised or fertilised with PK or NPK) located at different mountainous sites. Forages were characterized for crude nutrient contents and CT. Ammonia content was additionally analysed in ensiled forages. Average contents of pcdCP (per kg dry matter, DM) accounted for 51.3 g, 80.5 and 52.9 g for fresh, wilted and ensiled forage, respectively. Contents of pcdCP were positively correlated with CT contents; however, this correlation was moderate for forages of the first harvest ( $R^2=0.344$ ) and weak ( $R^2=0.085$ ) for forages of the subsequent harvests. The new German protein evaluation system for horses is promising for the prediction of contents of pcdCP adequately in forage from species-rich swards.

**Keywords:** condensed tannins, equine nutrition, mountain grassland, pre-caecal digestible crude protein, species-rich swards

## Introduction

The German protein evaluation system determines the pcdCP content of feeds based on neutral detergent soluble crude protein (NDSCP) with corrections made for silages considering the ammonia-nitrogen (N) content (Kirchhof and Rodehutschord, 2011). This calculation method is primarily based on swards composed of grass or legume species (Meyer and Coenen, 2014), rather than forages from species-rich swards. The latter may contain substantial amounts of CT, which may interact in protein digestion and therefore influence the content of pcdCP (Waghorn, 2008). It is therefore unclear whether the German protein evaluation system can also adequately predict pcdCP contents in forages with moderate CT contents, as in the case of forage from species-rich mountain grasslands.

## Materials and methods

The forage samples originated from three different long-term mineral-fertilisation field experiments located at elevations of 930 m a.s.l. (Thomet and Koch, 1993), 1,190 m a.s.l. (Carlen *et al.*, 1998) and 13,40 m a.s.l. (Baumberger *et al.*, 1996) in the Swiss mountains. Swards were either unfertilised (O) or fertilised with phosphorus (P) and potassium (K) PK or NPK and sampled in three or four plots at each site. Due to long-term mineral fertilisation of the mountain grasslands, species-rich swards had been established (Ineichen, 2018). Forage samples including gross nutrient composition, contents of CT and, in the case of silage samples, ammonia ( $\text{NH}_3$ ) concentrations were also available from previous investigations for fresh forage (Ineichen, 2018), wilted and ensiled forage (Seiler, 2018). To determine contents of pcdCP in forages, neutral detergent insoluble crude protein (NDICP) was analysed according to Licitra *et al.* (1996) using an ANKOM fibre analyzer. Prior to analysis, samples were milled on a Retsch

mill (MM440) for 35 sec at a frequency of 30/sec and a sample of 0.5 g was weighed in an ANKOM fibre bags in duplicates (F57). Chemical analysis of NDICP followed the protocol by the ‘Gesellschaft für Analysetechnik’ HLS (2016). Nitrogen content in the NDICP fraction was determined using a C/N-analyser. Contents of pcdCP (g kg<sup>-1</sup> DM) forage were determined using the following calculations (Kirchhof and Rodehutsord, 2011):

- pcdCP = [CP – NDICP] × 0.9 in fresh and wilted forage
- pcdCP = [CP – NDICP – NH<sub>3</sub>-N × 6.25] × 0.9 in ensiled forage.

Analysis of variance was conducted including ‘type of fertilisation, *f*’ within the first and subsequent harvest using the software (NSCC V9, 2013). Tables display arithmetic means in relation to type of fertilisation and harvest and differences are considered significant at *P*<0.05. Correlations between the content of pcdCP and CT were made for pooled date off all forage samples of the first harvest and the subsequent harvests separately, irrespective of type of conservation of fertilisation.

## Results and discussion

On average, contents of pcdCP were 61.6 g kg<sup>-1</sup> DM for swards of species-rich mountain grasslands (Table 1) and were similar to those reported by Meyer and Coenen (2014) for grass-based forage. From the first to the second harvest, contents of pcdCP increased by +13.2 g kg<sup>-1</sup> DM in fresh and +5.0 g kg<sup>-1</sup> DM in wilted foraged and decreased by -1.8 g kg<sup>-1</sup> DM in ensiled forage. Contents of CP were moderate and ranged from 119 to 133 g kg<sup>-1</sup> DM, which was also reflected by rather high fibre contents. Within each harvest and type of forage conservation, sward long-term fertilisation for >30 years at each site had little influence on pcdCP, CP or NDF contents.

Contents of CT ranged from 0 g to 19 g kg<sup>-1</sup> DM with, on average, lower concentrations in the first (6.26±4.01 g kg<sup>-1</sup> DM; Figure 1A) than in the subsequent harvests (7.38±5.34 g kg<sup>-1</sup> DM; Figure 1B). The correlations between CT (g kg<sup>-1</sup> DM) and pcdCP (g kg<sup>-1</sup> DM) were weak in both the first and subsequent harvests, although positively correlated (*P*<0.05).

- pcdCP = 41.7 + 2.38 3 CT, *R*<sup>2</sup> = 0.344, n=69 (first harvest, Figure 1A)
- pcdCP = 56.1 + 0.96 3 CT, *R*<sup>2</sup> = 0.085, n=83 (subsequent harvests, Figure 1B)

Table 1. Contents of pre-caecal digestible crude protein (pcdCP), crude protein (CP) and neutral detergent fibre (NDF) from forage of species-rich mountain grassland swards.<sup>1</sup>

		First harvest					Following harvests				
		0	PK	NPK	SEM	<i>f</i>	0	PK	NPK	SEM	<i>f</i>
Fresh forage, g kg <sup>-1</sup> DM	pcdCP	44.3	45.0	44.9	1.86	ns	51.2	53.5	68.2	2.91	ns
	CP	111	108	94.7	3.31	*	130	136	133	2.3	ns
	NDF	488	498	531	5.8	ns	380	385	405	7.5	ns
Wilted forage, g kg <sup>-1</sup> DM	pcdCP	72.3	82.6	78.9	1.86	ns	74.0	84.2	90.7	3.57	ns
	CP	124	126	115	1.8	ns	125	134	136	3.5	ns
	NDF	505	478	507	12.0	ns	477	425	441	10.5	ns
Ensiled forage, g kg <sup>-1</sup> DM	pcdCP	58.1	52.8	50.6	2.18	ns	50.2	56.1	49.7	1.98	ns
	CP	129	128	120	2.0	ns	121	131	127	4.0	ns
	NDF	542	501	517	11.1	ns	481	427	428	11.0	ns

<sup>1</sup> Type of fertilisation: unfertilised swards (0), PK or NPK; *f* = fertilisation; SEM = standard error of means; ns = not significant; \* *P*<0.05.

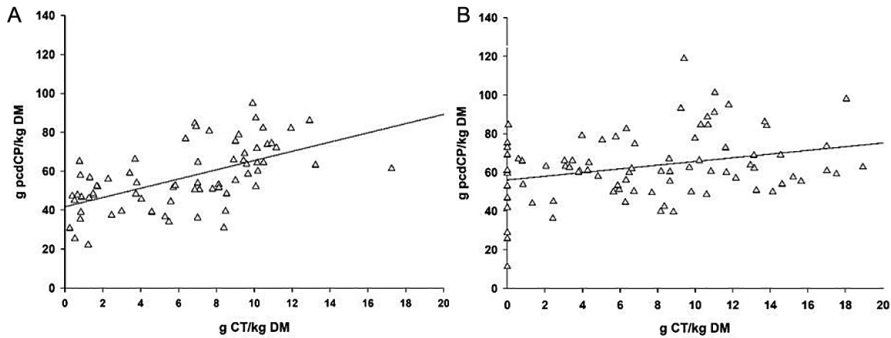


Figure 1. Correlations ( $P < 0.05$ ) of pre-caecal digestible crude protein (pcdCP) and contents of condensed tannins (CT) ( $\text{g kg}^{-1}$  dry matter, DM) in forage samples from species-rich mountain grasslands for the first harvest (A) or the subsequent harvests (B).

## Conclusions

Correlations between CT and pcdCP were moderate in the forage investigated with CT contents below  $20 \text{ g kg}^{-1}$  DM across harvests. A consideration of the CT content in the calculation of pcdCP contents is therefore unlikely. The new German protein evaluation system for horses appears therefore appropriate for predicting contents of pcdCP adequately in forage from swards rich in herbs and also legume species.

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