

# **Influence of contrasting environments on forage quality of ryegrass and four legumes growing in binary swards**

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## **Abstract**

Forage legumes are valuable alternatives for mineral N fertilizers in grassland, due to their N fixing capacity and the assumed increase in forage quality of the sward. However, forage quality of binary swards may differ in response to site and legume species. The objective of the present study, carried out within the frame of the COST Action 852, was to identify the influence of legume species growing in contrasting environments on legume proportion, crude protein (CP) and metabolisable energy (ME) contents. At each of the five participating sites the experiment was carried out as a completely randomized block design with three replications. Four legume species were tested: white clover, red clover, alfalfa and birdsfoot trefoil. The first cut was performed in spring after first node detection in grass and repeated 30±3 days afterwards (5-cut system). Legume proportion varied largely from 5% to 79%, with birdsfoot trefoil showing the lowest legume proportion in the swards across sites. Alfalfa had the highest N contents at all sites. Results show that, especially in ME content, legumes are less responsive to variation due to contrasting environments than the companion grass.

Keywords: forage legumes, companion grass, forage quality, contrasting environments

## **Introduction**

Although grassland based production systems account for half of the animal requirements for feed in Europe, large amounts of protein-rich concentrate feeds are imported and the demand for high quality vegetable protein is still increasing. Forage legumes are valuable protein sources, providing alternatives for ruminant nutrition. Additionally, they may reduce the need for mineral nitrogen fertilizer, and contribute to reduce nitrogen losses on the farm. However, feed quality of forage-legume based swards may differ between sites in Europe due to environmental constraints. The objective of the present study was to characterize the influence of legume species and site on feed quality of forage legumes and companion grass grown in binary swards. The study is part of the EU-COST Action 852.

## Materials and methods

The same cultivars of white clover (*Trifolium repens* L.), red clover (*Trifolium pratense* L.), alfalfa (*Medicago sativa* L.) and birdsfoot trefoil (*Lotus corniculatus* L.) were established at each site as binary swards with perennial ryegrass (*Lolium perenne* L.) in a randomized block design with three replications. White clover, red clover, alfalfa and birdsfoot trefoil were sown at rates of 4, 8, 16, and 8 kg/ha, respectively, with 15 kg/ha of perennial ryegrass, or 20 kg/ha in a pure grass stand. In the year after sward establishment, swards were cut first when the first grass node was detectable. Subsequent cuts took place every 30±3 days yielding a total of 5 cuts. Samples were separated into legume, grass and weed fraction. Feed quality analysis comprises the determination of the contents of crude protein (CP) and metabolisable energy (ME) separately for the grass and legume fraction. The CP was calculated as total N × 6.25 and the ME content was estimated from the cellulase method (De Boever *et al.*, 1988). The equations applied were as follows (Weissbach *et al.*, 1996):

$$ME_{\text{grass}} \text{ (MJ/kg DM)} = 13.98 - 0.0147 \cdot CA - 0.0102 \cdot RE - 0.00000254 \cdot RE^2 + 0.00234 \cdot CP$$

$$ME_{\text{legume}} \text{ (MJ/kg DM)} = 13.98 - 0.0147 \cdot CA - 0.0137 \cdot RE + 0.00234 \cdot CP$$

where CA = crude ash; RE = residue after enzymatic digestion; CP = crude protein

Data on a year basis were used for analysis of variance, and means were compared using Student's T-Test. The probabilities were adjusted using the Bonferroni-Holm test.

## Results

Table 1 shows the average weather data for the participating sites from March 31<sup>st</sup> to October 1<sup>st</sup>. Norway had the lowest global radiation and average temperature.

Table 1. Weather data of the experimental year at the participating sites

Site	Temperature, °C	Precipitation, mm	Global radiation, J/cm <sup>2</sup>
Norway	10.6	551	1274
Switzerland	15.3	722	1546
Netherlands	14.9	396	1568
Germany 2004*	13.8	418	1550
Germany 2005*	14.1	325	1551

\* In Germany the experiment was established separately in two subsequent years.

The results of the ME content of companion grass and legumes are shown in Tables 2 to 4. For the companion grass, the interaction site × sward type was significant (Table 2). The ME content of the companion grass was highest in Norway.

Table 2. Metabolisable energy content of the companion grass (perennial ryegrass)

Site/sward	White clover	Red clover	Alfalfa	Birdsfoot trefoil	Pure stand
Norway	11.8 <sup>a</sup>	11.5 <sup>a</sup>	11.6 <sup>a</sup>	11.9 <sup>a</sup>	11.9 <sup>a</sup>
Switzerland	10.7 <sup>b</sup>	10.4 <sup>b</sup>	10.8 <sup>b</sup>	10.4 <sup>b</sup>	10.6 <sup>bc</sup>
Netherlands	10.8 <sup>b</sup>	10.8 <sup>b</sup>	10.6 <sup>b</sup>	10.5 <sup>b</sup>	10.9 <sup>b</sup>
Germany 04	10.9 <sup>b*</sup>	10.7 <sup>b*</sup>	10.5 <sup>b</sup>	10.4 <sup>b</sup>	10.3 <sup>c</sup>
Germany 05	10.5 <sup>b</sup>	10.8 <sup>b</sup>	10.6 <sup>b</sup>	10.4 <sup>b</sup>	10.4 <sup>bc</sup>

SE=0.15; n=73

\* significantly different from pure grass stand ( $P < 0.05$ ) within sites

<sup>a,b</sup> site means carrying no common superscript are significantly different ( $P < 0.05$ )

For the ME content of forage legumes only the main effects were significant (Tables 3 and 4). White clover with 10.9 MJ ME/kg DM was the legume species with the highest ( $P<0.05$ ) ME content across sites. In Norway birdsfoot trefoil did not perform well.

Table 3. Metabolisable energy content of forage legumes – site effect

Site	ME, MJ/kg DM
Norway	10.5 <sup>c</sup>
Switzerland	10.3 <sup>c</sup>
Netherlands	10.9 <sup>a</sup>
Germany 04	10.4 <sup>c</sup>
Germany 05	10.7 <sup>b</sup>

SE=0.07; n=45;  $P<0.05$

Table 4. Metabolisable energy content of forage legumes – legume species effect

Species*	ME, MJ/kg DM
White clover	10.9 <sup>a</sup>
Red clover	10.5 <sup>b</sup>
Alfalfa	10.3 <sup>b</sup>

SE=0.05; n=45;  $P<0.05$

\* Birdsfoot trefoil was excluded due to missing values in Norway

The CP content of the companion grass is shown in Table 5. The interaction site × sward type was significant. The grass grown in Switzerland had the highest CP contents and that of Germany 05 showed the lowest contents. Grass grown with birdsfoot trefoil showed similar CP contents as the pure grass stand. In Norway, only grass growing with red clover showed higher CP contents compared to the pure stand.

Table 5. Crude protein content of the companion grass (perennial ryegrass)

Site/sward	White clover	Red clover	Alfalfa	Birdsfoot trefoil	Pure stand
Norway	13.2 <sup>c</sup>	14.2 <sup>b*</sup>	12.4 <sup>c</sup>	11.8 <sup>c</sup>	11.8 <sup>b</sup>
Switzerland	18.4 <sup>ab*</sup>	17.7 <sup>a*</sup>	19.4 <sup>a*</sup>	15.9 <sup>a</sup>	15.9 <sup>a</sup>
Netherlands	19.7 <sup>a*</sup>	18.6 <sup>a*</sup>	16.8 <sup>b*</sup>	14.2 <sup>b</sup>	13.2 <sup>b</sup>
Germany 04	17.4 <sup>b*</sup>	15.0 <sup>b*</sup>	15.6 <sup>b*</sup>	13.2 <sup>bc*</sup>	10.2 <sup>c</sup>
Germany 05	10.5 <sup>d*</sup>	11.3 <sup>c*</sup>	10.2 <sup>d*</sup>	8.5 <sup>d</sup>	7.2 <sup>d</sup>

SE=0.60; n=73

\* significantly different from pure grass stand ( $P<0.05$ ) within sites

<sup>a,b</sup> site means carrying no common superscript are significantly different ( $P<0.05$ )

The CP content of the forage legumes is shown in Table 6. The interaction site × sward type was significant. All legume species grown in Norway showed significantly lower CP contents than in the other sites. Considering legume species, alfalfa had the highest CP contents between all sites and red clover the lowest. An exception was Germany, where no differences for legumes species were observed.

Table 6. Crude protein content (% DM) of forage legumes at different sites

Site/species	White clover	Red clover	Alfalfa	Birdsfoot trefoil <sup>1)</sup>
Norway	20.4 <sup>cB</sup>	20.1 <sup>cB</sup>	22.4 <sup>cA</sup>	-
Switzerland	24.5 <sup>bA</sup>	22.3 <sup>bB</sup>	25.6 <sup>bA</sup>	25.2
Netherlands	26.0 <sup>aA</sup>	23.4 <sup>bB</sup>	27.1 <sup>aA</sup>	26.6
Germany 04	26.0 <sup>aA</sup>	25.1 <sup>aA</sup>	25.8 <sup>abA</sup>	26.1
Germany 05	23.4 <sup>bA</sup>	24.4 <sup>abA</sup>	24.5 <sup>bA</sup>	24.3

SE=0.39; n=45; <sup>1)</sup> Birdsfoot trefoil was excluded from calculations due to missing values in Norway

<sup>a,b</sup> site means within legume species carrying no common superscript are significantly different ( $P<0.05$ )

<sup>A,B</sup> legume species means within site carrying no common superscript are significantly different ( $P<0.05$ )

Table 7 shows the legume proportion at each site. The interaction site × sward type was significant. Low proportions of birdsfoot trefoil were observed at most sites, except in Germany 04.

Table 7. Average yearly legume proportion (%) of the swards at different sites on a DM basis

Site/sward	White clover	Red clover	Alfalfa	Birdsfoot trefoil
Norway	29.5 <sup>bB</sup>	44.8 <sup>bA</sup>	23.1 <sup>dB</sup>	4.9 <sup>cC</sup>
Switzerland	16.7 <sup>cB</sup>	46.6 <sup>bA</sup>	57.3 <sup>bA</sup>	8.2 <sup>cB</sup>
Netherlands	43.7 <sup>abAB</sup>	51.8 <sup>abA</sup>	38.5 <sup>cB</sup>	21.9 <sup>bC</sup>
Germany 04	38.6 <sup>bD</sup>	65.3 <sup>aB</sup>	79.0 <sup>aA</sup>	54.0 <sup>aC</sup>
Germany 05	53.4 <sup>aA</sup>	61.7 <sup>aA</sup>	59.6 <sup>bA</sup>	13.0 <sup>bcB</sup>

SE=3.4; n=60

<sup>a,b</sup> site means within legume species carrying no common superscript are significantly different ( $P<0.05$ )

<sup>A,B</sup> legume species means within site carrying no common superscript are significantly different ( $P<0.05$ )

## Discussion

Based on the results presented, the influence of sites on forage quality was most pronounced for the companion grass. The site effect was a result of different environmental conditions, as the grass growing in Norway had the lowest average temperature in the period, resulting in high ME contents. This confirms previous observations, as cell wall constituents deposited at lower temperatures are less lignified and higher in digestibility, and storage carbohydrates tend to accumulate in leaf tissue (Chatterton *et al.*, 1989). Although white clover was the legume species with the highest ME content, differences due to site were of minor magnitude, when compared to the companion grass.

Legume proportion varied largely from 5% to 79% for all species and sites, with birdsfoot trefoil showing the lowest legume proportion. This large variation in legume proportion affected the CP content of the companion grass. However, the differences in the increase in CP content in the companion grass with increasing legume proportion seems to be more pronounced for legume species than for contrasting sites.

The CP contents of forage legumes ranged from 20.4 to 27.1% DM considering sites and legume species. Differences in CP content between species were of minor magnitude. The frequent cutting each 30 days may have been one of the reasons for that, as legumes were always cut in an early development stage.

## Conclusions

In binary swards with legumes, the nutritional quality of the companion grass has to be considered especially in northern Europe as it may influence forage quality positively. The observed differences between sites and legume species for ME and CP contents in legumes are of marginal relevance for ruminants. The results suggest that, although grown in contrasting environments, any legume species could be used in binary swards in the first production year producing forage of similar CP and ME contents when cut every 30 days. The weak establishment of birdsfoot trefoil in most sites may limit its use, though.

## References

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