INTERRELATIONS AMONG THE QUALITY FEATURES OF THE FORAGE IN WHITE TREFOIL (*TRIFOLIUM REPENS* L.) UNDER THE INFLUENCE OF THE YEAR)

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Abstract

White trefoil (Trifolium repens L.) is a very important species in the permanent grasslands and cultivated on seeded pastures due to the great nutritional value for the grazing animals reflected in animal production. The investigation had in view the assessment of the interrelations among fodder quality features and their contribution in the formation of the. The considered variables were dry matter, crude protein, crude fat, cellulose, ash, nitrogen and non-nitrogenous extractive substances. There was also investigated the differences between the two experimental years, respectively year 2021 and year 2022. The statistical methods use rely on the correlation analysis and principal component analysis. The main conclusion obtained show the change of the contribution in the production of the analysed chemical features from a year to the other probably to the climatic differences between the years.

Keywords: *Trifolium repens* L., white trefoil, crude protein, crude fat, cellulose, ash, nitrogen and non-nitrogenous extractive substances.

INTRODUCTION

Nowadays is a great demand for the obtaining of healthier and safer food, for the protection of the direct consumers, respectively human population. This fact supposes the improvement of the nutritive value and food safety both in the case of vegetal and animal products. Having in view the obtaining of the animal products there is very important the relationship among soil – plant – animal, and implicitly the obtained production. That is why the safety of the food products of animal origin is in very strong relationship with the quality of the feeds and forages used for animal feeding. A proper feeding of the animals is essential to obtain the estimated primary production and income (Georgescu et al., 2007).

White trefoil (*Trifolium repens* L.) is common the temperate permanent grasslands, but also in the composition of the seeded pastures (Turkington and Burdon, 1983; Sackville Hamilton and Harper, 1989) being highly appreciated for the forager value in Europe (Frame and Laidlaw, 2005; Abberton and Marshall, 2005; Annicchiarico et al., 2015).

White trefoil grows and develops in a creeping system, and the vegetative dissemination is possible due to the stolon's system found at the soil surface. This growing type allows a great tolerance to grazing, because the herbivore animals are eating only the leaves and flowers of the plant, the damage being minimal, favouring their regeneration in a reasonable time interval for the next grazing cycle (Dragomir, 2005; Dragomir and Dragomir 2012).

The above-soil biomass production is represented by the fresh fodder that is the first noticeable parameter in the context of production, this being assessed by analysing the dry mass yield (DM). White trefoil is used as forage due to the high production of high-quality forage in grazing system, from this reason being considered one of the most valuable forage species (Olifirovych, 2018).

An important indicator for the fresh fodder yield is represented by the leaves of white trefoil. As big the leaf is, as its' contribution in the fresh fodder yield is higher. Leaves are the most valuable component in the qualitative forager production, influencing the forage quality, because usually it is containing 2-3 times more proteins in comparison with stems (Marcuvitz and Turkington, 2000; Perehrym, 2021).

The phyto-chemical profile of the white trefoil shows that is containing numerous substances that have a very important role for the grazing animals' health fed with it. Thus, the have indirectly a positive role on the animal production due to the presence in the animals feed (Ahmad and Zeb, 2021).

The purpose of the work was to assess the relationship among some production and quality features (dry matter, crude protein, crude fat, cellulose, ash, nitrogen and non-nitrogenous extractive substances) and the difference between the experimental years, respectively 2021 and 2022.

MATERIAL AND METHOD

For the assessment of the seasonal growth variation of the white clover (*Trifolium repens* L.), in the conditions of sheep grazing in rotational system, the experimental field was located at the Research and Development Station for the Breeding of Sheep and Goats Caransebeș (SCDCOC Caransebeș). The experiment was set on a soil albic luvisoil type, pseudogleic, slightly acid and moderately supplied with phosphorus and potassium.

The experimental field had 18 variants, consisting in six replicates with three graduations, resulting 72 experimental plots.

The assessment of the fresh fodder was using the direct method of cutting and weighting of the sampled biomass was the starting point for the determining of the dry matter (DM).

Dry matter yield was assessed by the drying method in oven at a temperature of 105-110 °C. the obtained results were extrapolated at hectare, being expressed in t/ha DM.

Determinarea masei verzi prin metoda directă a cosirii cântăririi stă la baza determinării producției de substanță uscată (SU).

For the analysis of the chemical composition of the forrage production from the analysed experimental variants, the samples were analysed in the laboratory of Animal Nutrition and Feeding Discipline from the Faculty of Bioengineering of the Animal Resources from the University of Life Sciences "King Mihai I" from Timisoara, as follows.

The quality forage parameters analysed in laboratory were:

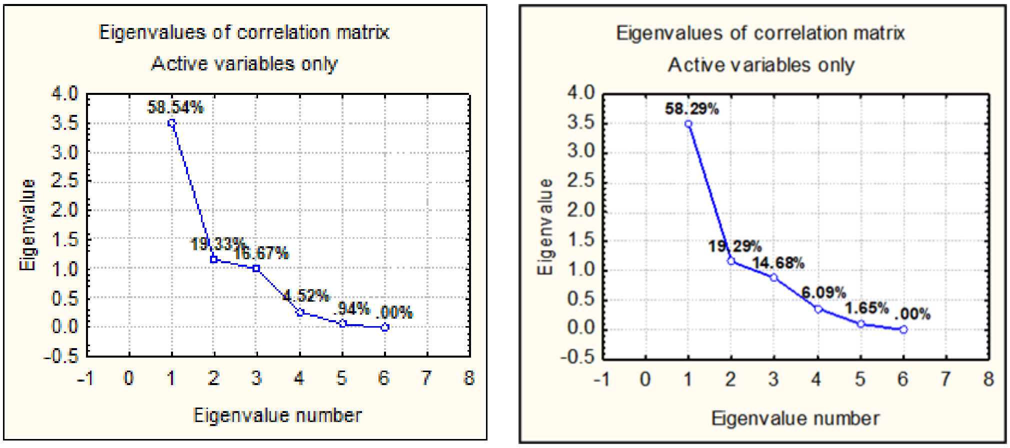
- the dry matter content (DM%) – by gravimetric method, respectively by drying the fresh fodder in oven at 105 ^{o}C until the sample mass is constant after two successive weighing;
- the crude protein content (CP%) – was measured using Kjeldahl method, respectively the block mineralization and steam distillation;
- the crude fat content (%) – was assessed using Soxhlet method of extraction with organic solvent;
- the cellulose content (%) – was measured by applying intermediate filtration;
- the ash content (%) – was measured using the gravimetric method of the residue left after sample burning in the calcination furnace;
- the nitrogen content (N%) – NFE% N = 100 – (CP% +ash% + cellulose% +fat% +U%);
- the non-nitrogenous extractive substances content (NFE%) -NFE% DM =DM –(CP% +ash% +cellulose% +fat%).

The data were processed with current statistical methods, respectively variation coefficients (Cv) and correlation coefficients (r) (Hair et al., 1995; Săvoiu, 2010; Dinca et al., 2019) and principal components analysis (Gruijters, 2019). For the statistical processing was used the statistical software SPSS (IBM SPSS Statistics for Windows, 2020).

RESULTS AND DISCUSSIONS

Comparing the two years of research, respectively 2021 and 2022, from the point of view of the principal components, both for the variables (CP%, fat%, cellulose%, ash%, N% and NFE%) and both for variants.

Analysing the graphical representation of the eigenvalues and following the Evrard criterion for obtaining the number of principal components, we can decide to choose 3 components (Figure 1).



2021 **2022**

Figure 1. Eigenvalues chart for the two years of production 2021 and 2022

If we look at the division of the amount of information and the fact that only the first 3 components add information to a variable in its original form, then we will only keep the latter ones. We can also note that a proportion of 94.54% of the original information for 2021 and 92.25% for the year 2022 [Table 1] is extracted by the new variables.

For the year 2021, the first component brings a variance of 58.54%, respectively a proportion of 58.54% is explained by component 1 [CP1]. The second component brings a variance of 19.33%. Thus, a proportion of 19.33% is explained by component 2 [CP2]. The third component brings a variance of 16.67%, therefore in this case a proportion of 16.67% is explained by the component 3 [CP3]. Thus, the shift from the 6 variables to only three was achieved with minimal information loss of about 5.46%.

For the year 2022, the first component brought a variance of 58.29%, respectively a proportion of 58.29% was explained by component 1 [CP1]. The second component brought a variance of 19.29%, respectively a proportion of 19.29% was explained by component 2 [CP2]. The third component brought a variance of 14.68%, and in this case a proportion of 14.68% was explained by the component 3 [CP3]. Thus, the shift from 6 variables to only three was achieved with minimal information loss of about 7.75%.

The general conclusion that is referring to the first three retained components according with Evrard criterion, or Benzecri (the percentage coverage criterion) or Cattell criterion (scree test) for the year 2021 was 94.54 %, with a minimum information loss of about 5.46 %, respectively for the year 2022 was 92.25 %, with a minimum information loss of about 7,74% (Table 1).

Table 1

Eigenvalues of correlation matrix, and related statistics

Component	year 2021			year 2022		
	Eigenvalue	% Total variance	Cumulative %	Eigenvalue	% Total variance	Cumulative %
1	3.512418	58.5403	58.54	3.497113	58.28522	58.29
2	1.159507	19.32512	**77.87**	1.157531	19.29219	**77.58**
3	1.000355	16.67258	94.54	0.880645	14.67741	92.25
4	0.271321	4.52202	99.06	0.365519	6.09198	98.35
5	0.056351	0.93918	100.00	0.099123	1.65204	100.00
6	0.000048	0.0008	100	0.00007	0.00116	100

The factor matrix or principal component matrix (Table 2) is very important in our analysis, since its elements, also known as factor intensities, are the correlation coefficients between the original variables (CP%, fat%, cellulose%, ash%, N% and NFE%) and the respective component (in our case CP1/Factor 1, CP2/Factor 2, CP3/Factor 3). The correlation coefficients obtained were both positive and negative. In both 2021 and 2022, three main components (Factor 1, Factor 2 and Factor 3) were obtained.

Analysing the principal components matrix (Table 2) that were noticed the followings:

- For the component PC1: In the year 2021 is a strong correlation with CP%, N%, NFE% and fat%, but there isn't any correlation with cellulose% (r = 0.14) and ash% (r = 0.04), the correlation coefficient for the last two being very small. In the year 2022 is a strong correlation with CP%, N%, NFE% ash% and with CP%, N%, NFE% and fat%, but there is a low correlation with fat% and isn't any correlation with the cellulose%.
- For the component PC2: In the year 2021 is a strong correlation with ash%, low with fat%, but there isn't any correlation with the other considered variables because the correlation coefficients were very small, respectively comprised between 0.15 and 0.02. In the year 2022 is a strong correlation with cellulose%, low correlation with

fat%, the other variables having very low correlation coefficients comprised between 0.26 and 0.06.

- For the component PC3: In the year 2021 is a strong correlation with cellulose%, the other variables have very small correlation coefficients comprised between 0.15 and 0.02. In the year 2022 is a strong correlation with fat%, the other variables having very low correlation coefficients comprised between 0.34 and 0.12.

Table 2

Principal components matrix (factor-variable correlations, based on correlations)

year 2021				year 2022			
variable	Factor 1 [CP1]	Factor 2 [CP2]	Factor 3 [CP3]	variable	Factor 1 [CP1]	Factor 2 [CP2]	Factor 3 [CP3]
CP%	**-0.975639 [1]**	-0.148542[4]	-0.001559[6]	**CP%**	**-0.975638 [1]**	-0.111480[5]	-0.123595[6]
fat%	**0.798376 [4]**	-0.418883[2]	0.107915[3]	fat%	**-0.401731 [5]**	**0.460680[2]**	**0.768805[1]**
cellulose%	-0.144528 [5]	**-0.118477[5]**	**0.979297[1]**	cellulose%	**0.141385 [6]**	**0.919229[1]**	-0.301012[3]
ash%	**-0.042682 [6]**	**-0.961484[1]**	-0.167142[2]	**ash%**	**-0.848197 [3]**	-0.064783[6]	0.216343[4]
N%	**-0.974890 [2]**	-0.152259[3]	-0.006668[5]	**N%**	**-0.974492 [2]**	-0.115453[4]	-0.128345[5]
NFE%	**0.974689 [3]**	-0.017537[6]	0.041269[4]	**NFE%**	**0.833544 [4]**	-0.265273[3]	0.347021[2]

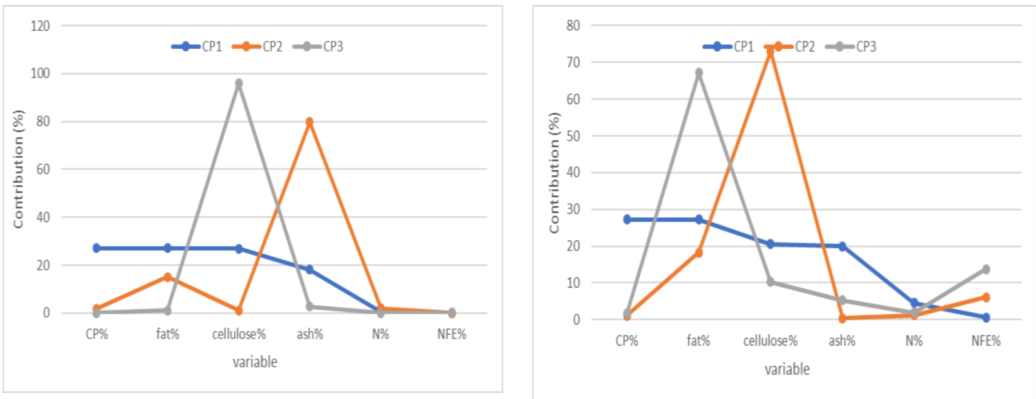
Analysing the position occupied by each variable (Table 2), according to the correlation coefficient we can highlight the followings:

- In the principal component 1 (PC1): the variables CP% and N% are maintaining the same position in both years of research, respectively position 1st and 2nd, variable NFE% is on the 3rd position in 2021 and descends to the position 4th in 2022, fat% descends from the 4th position in 2021 to the 5th position in 2022, cellulose% descends from the position 5th in 2021 to position 6th in in 2022, ash% climbs from 6th position in 2021 to 3rd position in 2022.
- In the principal component 2 (PC2): variable fat% is maintaining the same position in both years of research respectively 2nd position, ash% descends from the position 1st position in 2021 to the 6th position in 2022, N% descends from the position 3rd in 2021 to the position 4th in 2022, cellulose% climbs from 5th position in 2021 to 1st position in 2022 and NFE% also climbs from 6th position in 2021 to 3rd position in 2022.

- In the principal component 3 (PC3): the variables CP% and N% are maintaining the same position in both years of research, respectively position 6th and respectively 5th, NFE% climbs from 5th position in 2021 to 2nd position in 2022, fat% climbs from 3rd position in 2021 to 1st position in 2022, cellulose descends from the 1st position in 2021 to the position 3rd in 2022 and ash descends from the 2nd position in 2022 to the position 4th in 2022.

In the following it was analysed the values of the correlation coefficients between each variables analysed, respectively CP%, fat%, cellulose%, ash%, N% and NFE% in relationship with the corresponding component. The fact that most of the variables are changing the position from a year to other can be interpreted as being influenced of the climatic conditions different form a year to other, this aspect being necessary to be investigated in future research.

In Figure 2 is presented the contribution of the variabiles in the 3 principal components as contribution of the variables CP%, fat%, cellulose%, ash%, N% and NFE% in the years 2021-2022.



2021 **2022**

Figure 2. Contribution of the variables in the 3 principal components (contribution of the variables CP%, fat%, cellulose%, ash%, N% and NFE%) in the years 2021-2022

Analysing the correlation coefficients (Table 2, Figure 2) and the contributions (Figure), the results obtaines can be sumarized as it follows:

- In the principal component 1 (PC1): CP% had r = -0.976, and a contribution of 27%; N% had r = -0.975, and a contribution of 27%; NFE% had r = 0.975 with 27% contribution in 2021 and r = 0.833 and 20% contribution in 2022; fat% had r = 0.798 with 18.3% contribution in 2021 and r

= -0.402 and 4.6% contribution in 2022; cellulose% had r = -0.144 with 0.6% contribution in 2021 and r = 0.141 and 0.6% contribution in 2022; and as% had r = -0.043 with 0.1% contribution in 2021 and r = -0.848 and 20.6% contribution in 2022.

- In the principal component 2 (PC2): ash% had r = -0.962 with 79.7% contribution in 2021 and r = -0.065 and 0.4% contribution in 2022; fat% had r = -0.42 with 15.3% contribution in 2021 and r = 0.46 and 18.3% contribution in 2022; cellulose% had r = -0.12 with 1.2% contribution in 2021 and r = 0.92 and 73% contribution in 2022; the other considered variables had a very low contribution.
- In the principal component 3 (PC3): cellulose% had r = 0.98 with 95.9% contribution in 2021 and r = -0.3 and 10.3% contribution in 2022; fat% had r = 0.11 with 1.16% contribution in 2021 and r = 0.77 and 67.1% contribution in 2022; the other considered variables had a very low contribution.

CONCLUSIONS

White trefoil quality and yield chemical structure has a very interesting manifestation from a year to other, having in view the variable considered in this research, respectively CP%, fat%, cellulose%, ash%, N% and NFE%. The most important finding is represented by great change in the contribution of some variables from a year to other as it was. This fact that it can be interpreted as being under the influence of the climatic conditions that differ strongly form a year to other, this being one of the aspects that can be investigated by future research.

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