CHEMICAL COMPOSITION AND NUTRITIVE VALUE OF TEFF BIOMASS (*Eragrostis tef* **(Zucc.) Trotter) UNDER THE INFLUENCE OF SOWING RATE AND NITROGEN FERTILIZATION RATE**

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Abstract

In 2021-2022 the influence of three sowing rates (10, 15 and 20 kg ha⁻) and four nitrogen fertilization rates (0, 30, 60 *and 90 kg ha- ¹) on the chemical composition and the nutritive value of teff biomass (Eragrostis tef (Zucc.) Trotter) was* tested for the area of Central South Bulgaria. It was established that nitrogen fertilization increases crude protein content, it being the highest when fertilized with 90 kg ha⁻¹ nitrogen and sowing rate of 15 kg ha⁻¹ in both harvest *phases. Increasing the nitrogen and sowing rates, a negative trend is observed on the content of crude fat, crude fiber and ash, and a positive trend on the content of non-nitrogen extractive substances. The content of feed unit for milk (FUM) and feed unit for growth (FUG) does not change significantly both under the influence of the applied increasing doses of nitrogen fertilization and when the sowing rate is increased. Climatic factors have the strongest influence on the chemical composition of teff biomass. Nitrogen fertilization has a strong influence on the content of crude protein and ash, and harvesting phase - on the content of crude fat and crude fiber.*

Key words: Eragrostis tef (Zucc.) Trotter, chemical composition, nutritive value, sowing rates, nitrogen fertilization rates.

INTRODUCTION

Teff (*Eragrostis tef* (Zucc.) Trotter) is an annual plant of the cereal family. Its grain is an important ingredient for the production of traditional foods and beverages of the population in Ethiopia, Eritrea, South Africa (Ketema, 1997; Bultosa, 2016). Due to the numerous dietary benefits – rich chemical and mineral composition of the biomass as well as adaptability to extreme environmental conditions, this cereal crop is considered promising for cultivation and forage (Saylor, 2017; Vinyard et al., 2018; Kakabouki et al., 2020; Billman et al., 2022).

The biological characteristics of the crop make it possible to use it in compacting crop rotations - it can be successfully sown as a second crop on non-irrigated areas. Teff has high drought tolerance and is presented as a crop that can replace or complement some of the main forage crops (Ketema, 1997; Miller, 2009; Bultosa, 2016).

Studies on the chemical composition of teff biomass worldwide are scarce, especially on its cultivation as bulk forage. In recent years, some authors have reported the potential of the crop to produce silage and hay harvested in different

phenological phases. The quality of teff forage is highly dependent on fertilization, the stage of development at harvest and the number of swaths (Sang-Hoon et al., 2015; Saylor, 2017; Vinyard et al., 2018; Kakabouki et al., 2020; Laca, 2021). Crude protein content in teff biomass according to literature sources varies from 8.5 to 21.5%, fiber content varies from 53 to 73% depending on the development phase (Norberg et al., 2008; Miller, 2009). Development phase at harvest is one of the main factors affecting forage quality and digestibility (Nakata et al., 2018). After the heading phase, photosynthetic products are converted into fibrous structural components, fibers increase, and protein content decreases its values. Lower temperatures slow down the ripening process and the subsequent production of fibrous structural compounds, thereby increasing crude protein content and the overall forage quality. As the development phase progresses, the concentration of lignin increases and the overall digestibility of crude fiber decreases (Vinyard, 2018; Billman et al., 2022).

According to some authors, teff hay has the potential to replace corn silage in cattle rations and become a major forage source in the

nutrition of bulls and dairy heifers (Saylor, 2017; Ream et al., 2020). Currently, there is limited information on the nutritional value of biomass and its potential for use as ruminant feed.

The objective of the study is to determine the influence of the sowing rate, nitrogen fertilization rate and harvesting phase on the chemical composition and nutritional value of teff biomass grown for fodder under the conditions of Central South Bulgaria.

MATERIALS AND METHODS

The research was conducted in the period 2021- 2022 in the area of the village of Tulovo, Stara Zagora county, located in the region of Central South Bulgaria. The field experiment was conducted with a white variety of teff of the Dutch company "Millets place". The experiment was based on the method of fractional plots, with a harvest plot size of 10 m2 , under non-irrigated conditions, after predecessor wheat. The soils in the area are alluvial, slightly to moderately enriched with humus $(1.6\% - 2.6\%)$, with a slightly acidic to neutral reaction, slightly stocked with nitrogen (31.0-35.0 kg/ha) and phosphorus (8.0-27.0 ppm) and slightly to well stocked with potassium (93.0-136.0 ppm).

The influence of sowing rate and nitrogen fertilization on the chemical composition and nutritional value of teff (*Eragrostis tef* (Zucc.) Trotter) biomass in two development phases (milk and dough maturity) were tested.

The studied factors and their levels were as follows: factor A: sowing rate, kg ha⁻¹ $(A1 -$ 10; A2 – 15; A3 – 20); factor B: nitrogen fertilization rates, kg ha⁻¹ (B1 – 0; B2 – 30; B3 -60 ; B4 -90). Variant 1 (A1B1) has been adopted as a control – harvested at the milk maturity phase, with a sowing rate of 10 kg ha^{-1} and without nitrogen fertilization.

With the main tillage, background fertilization with 50 kg ha^{-1} P₂O₅ was made. Fertilization with nitrogen in the specified rates (factor B) was made immediately before sowing.

The following parameters were determined:

dry matter (DM) chemical composition as per Weende (AOAC, 2007); nutritional value of biomass for ruminants was calculated on the basis of the chemical composition - content of crude protein (CP), crude fat or ether extract (EE), crude fibre (CF) and nitrogen-free extract (NFE) using empirical equations (Todorov et al., 2004 and 2007):

 $GE = 0.0242 \text{ CP} + 0.0366 \text{ EE} + 0.0209 \text{ CF} +$ 0.017 NFE;

 $ME = 0.0152 DP + 0.0342 DEE + 0.0128 DCF$ + 0.0159 DNFE;

 $q = ME/GE$;

 $FUM = ME (0.075 + 0.039q);$

 $FUG = ME (0.0382 + 0.104q);$

where: DСF – digestible crude fibre, DEE – digestible ether extract, DNFE – digestible nitrogen free extract, DP – digestible protein, FUM – feed unit for milk, FUG – feed unit for growth, GE – gross energy, MЕ – metabolizable energy. Digestibility coefficients of biomass were obtained from the reference data of Todorov et al. (2007) for common millet, due to lack of data for teff.

For meteorological assessment of the experimental period, the degree of availability of the vegetation precipitation amount and the average vegetation temperature of the air (Р) were calculated. The formula $P = i*100/n + 1$ was used, where: $P - degree of availability, %$; i – sequence number of the individual members in the row (arranged in descending order for the precipitation amounts and in ascending order for the average annual and vegetation temperatures); n – total number of members in the series (Delibaltov, 1962). A representative set of 33 members (1987-2020) was used. Years with an availability of 0 to 25% are considered to be very wet and cool, from 25 to 50% - medium wet and medium cool, from 50 to 75% - medium dry and medium warm and 75 to 100 % - dry and warm.

The statistical processing of the obtained results for the chemical composition has been made with the ANOVA LSD test for statistical significance of the differences. To establish correlation dependencies and factor analysis, the software package for statistical data processing MS Excel software - 2010 was used.

RESULTS AND DISCUSSIONS

Meteorological conditions during the study period are given in Table 1. In terms of precipitation, the studied years have been characterized as relatively unfavourable. On

average for a multi-year period (1987-2020), the amount of precipitation during the vegetation period (May-July) amounts to 210.8 mm. The amount of precipitation during the active teff vegetation in 2021 was 60.3 mm below the norm, in 2022 it was 125.3 mm
below the norm. Regarding vegetation below the norm. Regarding vegetation precipitation, 2021 was characterized as moderately dry with a 60% availability, 2022 as dry with an 80% availability.

The average annual air temperature calculated for a multi-year period of time (1987-2020) was 11.4ºС. The air temperature during the active vegetation period (May-July) was 19.5ºC on average for a multi-year period. In terms of vegetation temperatures, 2021 was characterized as moderately cool $(P = 60\%)$, 2022 as warm ($P = 80\%$).

P% - Degree of vegetation availability of the climatic factors - precipitation and temperature.

The chemical composition of the feed biomass is fundamental for determining its nutritional value.

The protein nutritional value of feed depends primarily on the crude protein (CP) content. In both years of the experiment (Table 2), the CP content was higher in the milk maturity phase - 73.8 and 116.2 g kg⁻¹ dry matter (DM), respectively, compared to the next phase (dough maturity) - 69.1 and 115.1 g kg^{-1} DM, respectively. The conducted studies reveal that CP content in the teff biomass was 95.0 g kg^{-1} DM during the dough maturity phase and decreases to 92.1 g kg^{-1} DM during the dough maturity phase on average for the period 2021- 2022. Higher protein content was found in 2022, which was due to poor moisture availability and higher than normal air temperatures during the vegetation period.

Both by year and on average for the studied period, the lowest CP content in the biomass dry matter was found in the absence of nitrogen fertilization (B1).

It is evident from the obtained data that with an increase in the rate of nitrogen fertilization from 0 kg ha^{-1} to 90 kg ha^{-1} , CP content increases, being the highest at fertilization with 90 kg ha⁻¹ (B4) in both studied biomass harvesting phases. The increase compared to the control reaches up to 26.4% during the milk maturity phase and up to 22.0% during the dough maturity phase, the average differences for the studied period being very well proven $(P<0.001)$.

The average CP content for all three tested sowing rates was higher in the milk maturity phase.

On average for the period of the experiment, in the milk maturity phase at sowing rates of 10 and 15 kg ha⁻¹ and fertilization with 90 kg ha⁻¹ nitrogen, the highest and very well statistically proven (P<0.001) CP content in the teff biomass was reported.

In the dough maturity phase the highest protein content was also found at sowing rate of 15 kg ha⁻¹ and nitrogen fertilization at 90 kg ha⁻¹ (Р<0.001). With an increase in the seeding rate of 20 kg ha-1, CP content was the lowest in both harvest phases and the differences with the control and between the different rates of nitrogen fertilization were not statistically proven.

*A - sowing rates, kg ha⁻¹ (A1 - 10, A2 - 15, A3 - 20); B - nitrogen rates, kg ha⁻¹ (B1 - 0, B2 - 30, B3 - 60, B4 - 90)

*, **, ***- Statistically significant differences of the variants and control at P<0.05; P<0.01; P<0.001

*Different letters indicate statistically significant differences among variants at P<0.05.

The dry matter of teff biomass contains an average of 18.3 g kg^{-1} ether extract (EE) in the milk maturity phase and 20.4 g kg^{-1} EE in the dough maturity phase (Table 3).

The results reflecting the EE content in the biomass reveal that since the share of fat is small, the variation between the individual variants is insignificant. A proven reduction in the EE content was reported only at the highest tested sowing rate $(20 \text{ kg } \text{ha}^{-1})$ in the milk maturity phase. In the dough maturity phase, a decrease in EE content was also observed at a sowing rate of 20 kg ha⁻¹, but this was not statistically proven.

Crude fiber (CF) as the second major component comprises an average of 192.5 to 193.1 g kg^{-1} DM during the studied development phases. With an increase in the nitrogen fertilization rate, a decrease in their content is observed in both harvesting phases. The reduction compared to the control option with nitrogen fertilization at 90 kg ha⁻¹ averaged 13.4% (27.0 g kg⁻¹ DM) in the milk maturity phase and was very well statistically proven $(P<0.001)$ and well proven in the dough maturity phase ($P \le 0.01$). It is known that there is a negative correlation between the crude protein and crude fiber content, which was also confirmed in the present study.

The total Ash content in the teff grass biomass averaged over the study period at the milk maturity phase 54.0 g kg^{-1} DM and decreased to 50.5 $g \text{ kg}^{-1}$ DM at the dough maturity phase. A proven reduction in Ash content between different rates of nitrogen fertilization was observed in both harvest phases at the highest applied sowing rate (20 kg ha^{-1}) .

Nitrogen free extract (NFE) averaged 640.1-163.9 g kg^{-1} DM during the two harvesting phases. Fertilization with increasing nitrogen rates did not significantly affect the amount of NFE. Differences are observed between the individual sowing rates tested. Slightly higher values were obtained at a sowing rate of 20 kg ha^{-1} , and the differences were statistically proven in both phases of teff harvesting.

 $NFE - nitrogen$ free extract

*A - sowing rates, kg ha⁻¹ (A1 - 10, A2 - 15, A3 - 20); B - nitrogen rates, kg ha⁻¹ (B1 - 0, B2 - 30, B3 - 60, B4 - 90)

*Statistically significant differences of the variants and control at P<0.05

*Different letters indicate statistically significant differences among variants at P<0.05.

The total energy value (GE – calorific value of food at complete burning) obtained from teff biomass in the present study varied from 17.88 MJ kg⁻¹ DM in the milk maturity phase to 17.96 MJ kg^{-1} DM in the dough maturity phase (Table 4). Under the influence of fertilization with increasing nitrogen rates, this indicator varied slightly $(17.71-18.07 \text{ MJ kg}^{-1} \text{DM})$.

Animals do not fully utilize the potential energy of feed. A significant part of it is lost with undigested residues during digestion, intermediate exchange, etc.

Teff grass biomass contained an average of 1.20 FUM and 1.28 FUG in kg DM. The FUM and FUG congtent was almost the same and did not change significantly both under the influence of the applied increasing nitrogen fertilization rates and with the increases in the sowing rate from 10 to 20 kg ha⁻¹.

When calculating the correlations between the chemical composition of DM from teff biomass and the studied factors (Table 5), it was found that climatic factors had the strongest influence on CP and CF content and less with a moderate

influence on EE and ash. CP content is strongly positively correlated with vegetation temperatures $(r = 0.91)$ and strongly negatively correlated with vegetation precipitation $(r = -$ 0.91). Contrary to CP, CF content was strongly positively correlated with vegetation precipitation ($r = 0.88$) and strongly negatively

correlated with vegetation temperatures $(r = -$ 0.88).

There is a moderate proven correlation dependence $(r = 0.35)$ between CP content in teff DM and the nitrogen fertilization rate,

A weak negative correlation was observed between CP content and the harvest phase $(r=$ -0.17).

Variant	GE,	ME.	FUM,	FUG,	NE,			
	MJ/kg DM	MJ/kg DM	per kg DM	per kg DM	MJ/kg DM			
Phase of milk maturity								
1. A_1B_1 (control)	17.83	11.80	1.19	1.26	7.14			
2. A_1B_2	17.78	11.74	1.18	1.25	7.10			
$3. A_1B_3$	17.91	11.80	1.19	1.26	7.13			
4. A_1B_4	17.97	11.91	1.20	1.28	7.21			
5. A_2B_1	17.98	11.91	1.20	1.28	7.21			
$6. A_2B_2$	17.88	11.77	1.18	1.26	7.11			
$7. A_2B_3$	17.91	11.91	1.20	1.28	7.22			
$8. A_2B_4$	17.97	11.95	1.21	1.28	7.24			
$9. A_3B_1$	17.72	11.83	1.20	1.27	7.17			
10. A_3B_2	17.90	11.92	1.20	1.28	7.22			
11. A_3B_3	17.97	11.91	1.20	1.28	7.21			
$12. A_3B_4$	17.71	11.81	1.19	1.27	7.16			
Average of phase	17.88	11.86	1.20	1.27	7.17			
Phase of dough maturity								
13. A_1B_1	17.92	11.89	1.20	1.28	7.20			
14. A_1B_2	18.02	11.94	1.20	1.28	7.22			
15. A_1B_3	18.02	11.98	1.21	1.29	7.26			
16. A_1B_4	18.06	11.85	1.19	1.26	7.15			
17. A_2B_1	17.91	11.91	1.20	1.28	7.21			
$18. A_2B_2$	17.87	11.90	1.20	1.28	7.21			
$19. A_2B_3$	17.93	11.88	1.20	1.27	7.18			
$20. A_2B_4$	17.92	11.88	1.20	1.27	7.18			
$21. A_3B_1$	17.89	11.95	1.21	1.29	7.25			
22. A_3B_2	17.94	11.92	1.20	1.28	7.21			
$23. A_3B_3$	17.92	11.97	1.21	1.29	7.26			
$24. A_3B_4$	18.07	11.99	1.21	1.29	7.26			
Average of phase	17.96	11.92	1.20	1.28	7.22			

Table 4. Nutritive value of teff biomass, average for the period 2021-2022

*A - sowing rates, kg ha⁻¹ (A1 – 10, A2 – 15, A3 – 20); B - nitrogen rates, kg ha⁻¹ (B1 – 0, B2 – 30, B3 – 60, B4 - 90); DM - dry matter; GE - Gross energy; ME - Metabolizable energy; FUM – feed unit for milk; FUG – feed unit for growth; NE - Net energy.

The year conditions (Table 6) are the strongest factor (P<0.001), having an effect on CP and CF in teff biomass, showing that they are influenced by temperature and precipitation during the vegetation period.

Nitrogen fertilization has a strong influence $(P<0.001)$ on CP content (7.58%) and a wellproven influence on ash content $(P<0.01)$. The sowing rate has a very well-proven influence on the EE content (P<0.001).

The harvesting phase of teff has a very wellproven effect on CF and EE content.

Table 5. Correlation (r) between chemical composition and factors, $n = 72$

Factors	Crude protein	Ether extract	Crude fiber	Ash
Phase	-0.17	-0.15	-0.01	0.32
Sowing rate	0.02	0.30	-0.07	0.14
Nitrogen rate	$0.35*$	-0.06	0.13	0.01
Temperature V-VII	$0.91*$	0.30	$-0.88*$	0.30
Rainfalls V-VII	$-0.91*$	-0.30	$0.88*$	-0.30

*V-VII – rainfall and temperature during the growing season (May - July);

*Correlation is significant at the P<0.05 level.

No relationship has been established between year sowing rate and the chemical composition of the teff biomass.

Table 6. Influence of factors on the chemical composition of biomass of teff, average for the period $2021 - 2022$, $n = 72$

	Crude	Ether	Crude	Ash
Factors	protein	extract	fiber	
Year	83.01***	9.19*	$77.1***$	$9.2**$
Nitrogen rate	7.58***	6.28	1.88	$9.02**$
Sowing rate	1.16	18.19***	0.2	8.47*
Phase of maturity	0.53	$10.15***$	12.94**	5.85
Year*nitrogen rate	0.47	6.18	0.06	16.65*
Year*sowing rate	1.94	0.02	0.19	5.79
Year*phase	0.14	16.48***	5.93	1.05
Other factors	5.17	33.51	1.7	43.97

*, **, ***Statistically significance at P<0.05, 0.01 and 0.001, respectively.

CONCLUSIONS

Teff biomass contains $73.8-69.1$ g kg^{-1} DM crude protein, $18.3-20.4$ g kg⁻¹ DM crude fat, 192.5-193.1 g kg-1 DM crude fiber, 54.0- 50.5 g kg-1 DM ash and 640.1-643.9 g kg-1 DM NFE.

Nitrogen fertilization affects positively the crude protein content of teff biomass. The crude protein content is higher in the milk maturity phase. The highest crude protein content was obtained at nitrogen fertilization of 90 kg ha⁻¹ and sowing rate of 15 kg ha⁻¹ in both harvest phases.

With an increase in nitrogen and sowing rates, a negative trend was observed on the content of crude fat, crude fiber and ash, and a positive trend on the content of nitrogen-free extract substances in the teff biomass.

The average nutritive value in the teff biomass for ruminants is the following: Gross energy – 17.88-17.96 MJ kg⁻¹ DM, Metabolic energy – 11.86-11.92 MJ kg-1 DM, 1.20 FUM and 1.28 FUG per kg DM. The FUM and FUG content is almost the same and does not change significantly both under the influence of the applied increasing nitrogen fertilization rates and with the increase of the sowing rate.

A strong correlation was found between the crude protein and crude fiber content with the climatic factors during the vegetation period and a good correlation between the crude protein protein and nitrogen fertilization.

Climatic factors have the strongest influence on the chemical composition of teff biomass. Nitrogen fertilization has a strong influence on crude protein and ash content, and the harvesting phase - on the crude fat and crude fiber content.

REFFERENCES

- AOAC international. (2007). Official methods of analysis of AOAC (18 edition, rev. 2), *Association of Official Analytical Chemists Intern*, Gaithersburg, MD, USA.
- Billman, E. D., de Souza, I. A., Smith, R. G., Soder, K. J., Warren, N., & Brito, A. F. (2022). Evaluating warm-season annual forages to fill summer forage gaps in short-season climates. *Crop, Forage & Turfgrass Management*, *8*(1), e20152.
- Bultosa, G. (2016). *Teff: Overview*. In Wrigley, C., Corke, H., Seetharaman, K., & Faubion, J. (Eds.).
- Delibaltov, Y., Hristov, H., & Tsonev, I. (1962). Design Irrigation Scheduling for the Agricultural Crops. *Proc. Bulletin of the Institute of Hydrology and Meteorology*, Sofia, Bulgaria, Vol. 3, 5-56.
- Kakabouki, I., Tzanidaki, A., Folina, A., Roussis, I., Tsiplakou, E., Papastylianou, P., ... & Bilalis, D. J. (2020). Teff (*Eragrostis tef* (Zucc.) Trotter) fodder yield and quality as affected by cutting frequency.
- Ketema, S. (1997). *Tef-Eragrostis tef (Zucc.),* Vol. 12, Bioversity International.
- Laca, M. C. (2021). *Seeding Rate, Nitrogen Fertilzer, and Cutting Timing Effects on Teff Forage Yield and Nutritive Value* (Doctoral dissertation, Utah State University).
- Miller, D. (2009). Teff Grass: A new alternative. In *Proceedings of the 2009 California Alfalfa & Forage Symposium and Western Seed Conference,* Reno, NV, USA, 2-4.
- Nakata, Y., Idota, S., Tobisa, M., & Ishii, Y. (2018). Triple Cropping Systems of Spring Maize, Tropical Grass of Teff (*Eragrostis tef*) and Winter Cereal Crops to Combine Total Digestible Nutrient Yield with Protein Concentration in Southern Kyushu, Japan. *Agricultural Sciences*, *9*(1), 129-140. https://doi.org/10.4236/as.2018.91010
- Norberg, S., Roseberg, R. J., Charlton, B. A., & Shock, C. C. (2008). Teff: a new warm-season annual grass for Oregon.
- Ream, C. N., Stevens, A. V., Hall, J. B., & Chibisa, G. E. (2020). Harvest maturity of *Eragrostis tef* 'Moxie': Effects on ruminal fermentation, total-tract nutrient digestibility, and growth performance in backgrounding beef cattle. *Applied Animal Science*, *36*(5), 600-609.
- Sang-Hoon, L., Dong-Gi, L., & Ki-Won, L. (2015). Evaluation of Forage Production and Tissue Culture Efficiency of Two Teff Grass (*Eragrostis teff*) Cultivars. *Research Journal of Biotechnology Vol*, *10*, 4.
- Saylor, B. A. (2017). *Drought-tolerant teff grass as an alternative forage for dairy cattle* (Doctoral dissertation, Kansas State University).
- Todorov, N., Ilchev, A., Georgieva, V., Gerginov, D., Djuvinov, D., Penkov, D., & Shindarska, Z. (2004). Animal nutrition. *Textbook*, Uniscorp, Sofia, 63-70.
- Todorov, N., Krachunov, I., Dzhuvinov, D., & Aleksandrov, A. (2007). *Guide for Animal Nutrition. Matkom*, Sofia, 399 p.
- Vinyard, J. R., Hall, J. B., Sprinkle, J. E., & Chibisa, G. E. (2018). Effects of maturity at harvest on the nutritive value and ruminal digestion of *Eragrostis tef* (cv. Moxie) when fed to beef cattle. *Journal of Animal Science*, *96*(8), 3420-3432.