

Article

The Effect of Tytanit Foliar Application and Different Nitrogen on Fibre Fraction Content and the Feed Value of *Festulolium braunii*

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Abstract: The aim of the present study was to determine the effect of Tytanit foliar application, also comparing it with that of nitrogen, on the content of *Festulolium braunii* fibre fractions, its nutritional value and feed intake. The experiment showed that forage of better quality could be obtained by reducing the amounts of traditional nitrogen fertilisers, or excluding them and using a product containing titanium instead. The experiment commenced in the spring of 2014 in the field of the University of Natural Sciences and Humanities in Siedlce. The plant used in the experiment was the Felopa variety of *Festulolium braunii*. The effects of Tytanit foliar application at a concentration of 0.2% and 1% and of mineral nitrogen at a dose of 80 and 160 kg ha⁻¹ were studied. *Festulolium braunii* was harvested three times a year in the period 2015–2017. The amounts of NDF, ADF, and ADL in the plant material were determined by near-infrared reflection spectroscopy (NIRS) using the NIRFex N-500. Relative feed value (RFV) and dry matter intake (DMI) were calculated. The higher dose of Tytanit supplied to *Festulolium braunii* contributed to an increase in NDF and ADF fraction content and a reduction in RFV and ADL fraction and DMI.

Keywords: *Festulolium braunii*; Tytanit; nitrogen; feed value



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1. Introduction

Recently, mainly neutral detergent fibre (NDF) and acid detergent fibre (ADF) contents have been taken into account to assess roughage quality [1–3]. NDF content affects forage intake by animals [4,5], but ADF, consisting of the least digestible forage ingredients such as cellulose, lignin, silica and ash, is negatively correlated with the digestibility of other nutrients [5]. Currently, ADF and NDF values are also commonly used to calculate the amount of feedstuff an animal is able to digest, total digestible nutrient content and other energy components. In addition, relative feed value (an indicator for allocating suitable feedstuffs to a given group of animals) is used in contexts such as hay price fixing, feed management verification, cutting and storage control.

In order to ensure their suitable chemical composition, plants need adequate amounts of nutrients. The most decisive aspect of growing methods affecting the size and quality of crop yields is the use of basic fertilisers in appropriate proportions. A new area of interest is the application of products supporting plant immunity, increasing yield and improving soil fertility [6]. They can reduce the use of traditional mineral fertilisers or eliminate them altogether [7]. While some such biostimulants are applied to the soil, like soil conditioners, others, like titanium-based Tytanit, are sprayed on leaves [7–10]. Numerous studies have shown that titanium (Ti) has a beneficial effect on physiological-biochemical processes in plants and contributes to yield growth [11–13]. According to Radkowski and Radkowska [14], titanium increases the chlorophyll content of leaves, accelerates their growth and development [11] and reduces the sensitivity of plants to adverse environmental conditions. Research results confirm its beneficial effects on the

plant yield and its quality, and it is commonly used in the cultivation of various agricultural and horticultural plants [15–18]. However, there are very few reports on the use of titanium as a stimulant in the cultivation of *Festulolium braunii* [19–22]. According to Borowiecki [23], the plant has recently attracted increasing interest among forage producers due to its high yield potential and its good quality [2,24,25]. According to Sosnowski and Jankowski [26] and Staniak [27], *Festulolium braunii* has a good growth and development rate in successive years, and the characteristic feature of this grass is the high share of leaves in green matter in relation to generative shoots [28]. In assessing a grass species, it is important to determine not only its chemical composition but also the content of fibre fractions that affect its nutritional value and feed intake [29].

Taking the above into account, the aim of the study described herein was to determine the effect of Tytanit foliar application, also comparing it with that of nitrogen, on the content of *Festulolium braunii* fibre fractions, its nutritional value and feed intake.

2. Material and Methods

2.1. Experimental Design

The experiment was commenced in the spring of 2014 in the field of the University of Natural Sciences and Humanities in Siedlce, central-eastern Poland, on soil with a granulometric composition of loamy sand with a pH_{KCl} of 6.75. Organic carbon content was 37.0 g kg^{-1} with 1.75 g kg^{-1} of total nitrogen. The organic carbon content of the soil was determined by wet oxidation followed by titration and the content of the nitrogen by the Kjeldahl method. The absorbable forms of phosphorus and potassium, determined by the Egner-Rhiem method, were medium, with 39.9 mg kg^{-1} and 128 mg kg^{-1} , respectively. The macronutrient content of the soil was as follows: (g kg^{-1}): P-1.05; K-1.00; Ca-2.40; Mg-1.25; S-0.508; Na-0.312. It was determined by the ICP-AES method, after prior dry mineralisation of the samples. Before the experiment was set up, soil samples for chemical analysis were collected with a steel tube (a soil sampler). The plant used in the experiment was the Felopa variety of *Festulolium braunii* (K. Richt.) A. Camus. The seeding rate was adopted according to standards developed by IMUZ in Falenty [30].

Every growing season nitrogen was applied in early spring (before the start of the growing period) and after the first and second cuts. Phosphorus and potassium were not used due to the moderate soil content of their available forms. Tytanit spraying was carried out once during stem formation. In the year of sowing (2014), no treatment was used and grass was left to fully develop, with weeds being cut three times. During its full use (2015–2017), *Festulolium braunii* was harvested three times a year. The first cut was carried out in late May, the second in early July and the third in mid-September in each growing period. At the time of cut, 0.5 kg of green matter was collected from each plot and dried to determined dry matter content.

2.2. Treatment Combinations

In a completely randomised design, experimental plots, each of 1.5 m^2 , were replicated three times, with the following treatment combinations (Table 1):

Table 1. Treatment combinations.

Plot.	Treatment
1	control (no treatment);
2	ammonium nitrate (N1) of 80 kg N ha^{-1} , (24 g N to a plot);
3	ammonium nitrate (N2) of 160 kg N ha^{-1} , (47 g N to a plot);
4	Tytanit [®] (Ti ₁) at a concentration of 0.2%, (1 cm^3 per 500 cm^3 of water);
5	Tytanit [®] (Ti ₂) at a concentration of 1%, (5 cm^3 fertiliser per 500 cm^3 of water);
	ammonium nitrate (N1) + Tytanit [®] (Ti ₁);
6	ammonium nitrate (N2) + Tytanit [®] (Ti ₂)

2.3. Forage Quality Determination

The amounts of NFD, ADF, and ADL (acid-detergent lignin) in the plant material were determined by near-infrared reflection spectroscopy (NIRS) using the NIRFex N-500. Relative feed value (RFV) and dry matter intake (DMI) were calculated. For this purpose, the forage quality test of Linn and Martin [31] was used, for which the classification parameter was relative feed value (RFV), calculated according to the following formula:

$$\text{RFV} = (\text{DDM} \times \text{DMI}):1.29$$

where RFV—relative feed value (dimensionless), DDM—digestible dry matter, $\text{DDM} = 88.9 - 0.779 \times \text{ADF} (\%)$, DMI—dry matter intake (% body weight), $\text{DMI} = 120:\text{NDF}$.

Based on the calculated RFV values, the forage was classified according to the quality test of Linn and Martin [31]. Individual quality classes were assigned to animal feeding groups (Table 2).

Table 2. Assessment of forage suitability in animal nutrition on the basis of RFV [31].

Quality Class	RFV Ranges	Feed Consumer
I	>151	Best high-productivity cows
II	125–151	High-productivity cows, young heifer selected for breeding
III	103–124	Good-quality beef cattle, older heifers, dairy cows—to a limited extent
IV	87–102	Beef cattle and dried dairy cows
V	75–86	Dried beef cows—supplemented with high-energy feed

2.4. Meteorological Conditions

Meteorological data from 2015–2017 were obtained from the Hydrological and Meteorological Station in Siedlce. In order to determine the temporary variability of temperatures and precipitation and their effect on plants, Sielianinov's hydrothermal coefficient was calculated [32]. It is based on the monthly amount of precipitation (P) and the monthly sum of daily air temperatures (t). Sielianinov's hydrothermal coefficient (K) was calculated using the following formula [33]:

$$K = P \times 10 / \Sigma t$$

K—Sielianinov's coefficient, P—monthly sum of precipitation, Σt —monthly sum of daily air temperatures.

2.5. Statistical Analysis

The results were processed statistically, with differences between means assessed using variance analysis. Statistica Version 10.0 [34] was used for calculations. The significance of the differences between means was estimated by Tukey's test at a significance level of $\alpha \leq 0.05$. Variance analysis for a two-factor experiment in a split-plot design was used according to mathematical models:

1. Determination of the effect of treatment across growing seasons:

$$y_{ijl} = m + a_i + g_j + e^1_{ij} + b_l + ab_{il} + e^2_{ijl}$$

where m—overall mean, a_i —the effect of growing seasons, g_j —the effect of replications, e^1_{ij} —error 1, b_l —the effect of treatment, ab_{il} —the effect of interaction between growing seasons and treatment, e^2_{ijl} —error 2.

2. Determination of the effect of treatment across *Festulolium braunii* harvests:

$$y_{ijl} = m + a_i + g_j + e^1_{ij} + b_l + ab_{il} + e^2_{ijl}$$

where m —overall mean, a_i —the effect of treatment, g_j —the effect of replications, e^1_{ij} —error 1, b_l —the effect of harvests, ab_{il} —the effect of interaction between harvests and treatment, e^2_{ijl} —error 2.

In order to demonstrate the relationship between the variables, a linear correlation analysis (Pearson correlation analysis) was applied, on the basis of which correlation coefficients were calculated.

3. Results and Discussion

3.1. Weather Conditions

Sielianinov's hydrothermal coefficient (Table 3) indicated that the worst weather conditions were in the second year (2016), with as many as four extremely dry, dry or moderately dry months during the growing season. The most favourable hydrothermal conditions for the growth and development of *Festulolium braunii* were in the first year (2015), when three months were moderately wet. The same year, the last two months (September and October) were affected by droughts, but that did not affect grass growth and development since this was the end of the growing season.

Table 3. The values of Sielianinov's hydrothermal coefficient (K) in the growing season.

Year	Month							Mean
	April	May	June	July	August	September	October	
2015	1.36 (o)	1.87 (mw)	1.64 (mw)	0.59 (sd)	1.92 (mw)	0.64 (sd)	0.12 (ed)	-
2016	1.22 (md)	2.63 (sw)	0.87 (d)	1.08 (md)	0.18 (ed)	1.46 (o)	1.94 (mw)	-
2017	2.88 (sw)	1.15 (md)	1.08 (md)	0.45 (sd)	0.96 (d)	1.92 (mw)	1.90 (mw)	-

K ≤ 0.4 extreme drought (ed), 0.4 < K ≤ 0.7 severe drought (sd), 0.7 < K ≤ 1.0 drought (d), 1.0 < K ≤ 1.3 moderate drought (md), 1.3 < K ≤ 1.6 optimal (o), 1.6 < K ≤ 2.0 moderately wet (mw), 2.0 < K ≤ 2.5 wet (w), 2.5 < K ≤ 3.0 severely wet (sw), K > 3.0 extremely wet (ew).

3.2. NDF Content

According to Brzóska and Śliwiński [5], as a source of energy for rumen microorganisms, NDF is particularly important for ruminants. In addition, it gives the feed a structure and serves as ballast filling the rumen. Its content of feedstuffs makes it possible to determine the amount of forage intake [4]. In the modern animal nutrition system, NDF is widely used instead of raw fibre [35,36]. Its desired content in dairy hay is approximately 400 g kg⁻¹ DM [37].

NDF content varied in the same growing season for both treatment combinations and cuts (Table 4) and averaged 537.6 g kg⁻¹.

Table 4. Neutral detergent fibre (NDF) content in *Festulolium braunii* (g kg⁻¹ DM).

Treatment	Means across Growing Seasons				Means across Cuts			
	Years				Cuts			
	2015	2016	2017	Mean	I	II	III	Mean
0	539.3	562.5	554.5	552.1 A	559.5	530.9	565.9	552.1 A
N ₁	546.2	520.7	531.4	532.8 BC	528.7	552.7	516.8	532.8 BC
N ₂	537.3	534.2	567.5	546.3 AB	544.2	547.9	546.9	546.3 AB
Ti ₁	517.5	527.5	532.5	525.8 BC	539.6	520.9	516.9	525.8 C
Ti ₂	537.3	536.3	575.2	549.6 A	536.5	570.2	542.1	549.6 A
N ₁ + Ti ₁	505.9	527.5	538.3	523.9 C	521.8	522.4	527.5	523.9 C
N ₂ + Ti ₂	516.8	540.3	541.3	532.8 BC	530.4	525.4	542.6	532.8 BC
mean	528.6 c	535.6 b	548.7 a	537.6	537.2 a	538.6 a	537.0 a	537.6

Different lowercase letters in the same row or different uppercase letters in the same column indicate significant differences between treatments.

A similar content of this fraction in the *Festulolium braunii* biomass ($532 \text{ g kg}^{-1} \text{ DM}$) was obtained by Olszewska and Kobyliński [2]. However, the values for meadow plants recorded by Grzelak and Bocian [38] were lower, ranging from 411.8 to $501.0 \text{ g kg}^{-1} \text{ DM}$. Jankowska-Huflejt and Wróbel [39] found significantly higher amounts in meadow and pasture forage of organic farms, ranging from 513.5 to $616.1 \text{ g kg}^{-1} \text{ DM}$. In 2015, the highest NDF content ($546.2 \text{ g kg}^{-1} \text{ DM}$) was in plants treated with a lower dose of nitrogen (80 kg N ha^{-1}), which was not significantly different from the control ($539.3 \text{ g kg}^{-1} \text{ DM}$) or from the effect of the higher doses of nitrogen or Tytanit ($537.3 \text{ g kg}^{-1} \text{ DM}$). Compared to the higher one, the lower Tytanit dose significantly reduced it ($517.3 \text{ g kg}^{-1} \text{ DM}$). Similar trends persisted with Tytanit and nitrogen combined application, although differences were not statistically significant. Compared to Tytanit applied on its own, combined higher and lower doses of Tytanit and nitrogen were found to reduce NDF content. In turn, Sosnowski [40] noted that in *Festulolium braunii*, it significantly increased to $515 \text{ g kg}^{-1} \text{ DM}$ in response to mineral fertilisers, but it did not change significantly after the use of the UGmax soil conditioner. UGmax is a natural, liquid concentrate composed of microorganisms and macro- and micro-elements.

NDF content significantly varied across growing seasons. In the second year (2016), compared to the first (2015), it was higher on average by 7.0 g kg^{-1} and in 2017 by 20.1 g kg^{-1} . Across all treatment combinations, the higher Tytanit dose resulted in the highest NDF content of *Festulolium braunii* (575.2 g kg^{-1}). In the remaining years, no such effect was noted. In both the second and third years, the values depending on the treatment were similar to those in the first year. Additionally, the average results of three years indicated that it increased with the higher separate doses of both nitrogen (from 532.8 to $546.3 \text{ g kg}^{-1} \text{ DM}$) and Tytanit (from 525.8 to $549.6 \text{ g kg}^{-1} \text{ DM}$). When applied together, they reduced NDF content in relation to the same separate doses. Among the averages across treatments, the highest value was recorded in the third year ($548.7 \text{ g kg}^{-1} \text{ DM}$) and the smallest in the first ($528.6 \text{ g kg}^{-1} \text{ DM}$).

As an average over the years of research, the highest NDF content was in control plants with $552.1 \text{ g kg}^{-1} \text{ DM}$, and in those treated with the higher doses of Tytanit ($549.6 \text{ g kg}^{-1} \text{ DM}$) and nitrogen ($546.3 \text{ g kg}^{-1} \text{ DM}$). It should be assumed that *Festulium braunii* treated with Tytanit produced softer leaf blades than control plants, which affected its nutritional value (lower NDF content), with greater sugar-soluble content and greater digestibility of organic matter. The lowest amounts were recorded on plots with the lower doses of nitrogen and Tytanit applied together ($523.9 \text{ g kg}^{-1} \text{ DM}$) and with the lower dose of Tytanit on its own ($525.8 \text{ g kg}^{-1} \text{ DM}$). Differences between the highest and lowest values were statistically significant.

NDF fraction content (Table 4) also varied across cuts. Compared to lower ones, the separate higher doses of nitrogen and Tytanit, or their combined higher dose, resulted in a slight increase of the cut average (by 13.59 , 23.80 and 8.94 g kg^{-1} , respectively). Moreover, compared to control (552.1 g kg^{-1}), for the lower doses of each fertiliser the content was significantly lower. Across cuts, the average content was 537.6 g kg^{-1} , similar to values obtained for *Festulolium* by Wiśniewska-Kadżajan and Stefaniak [41]. Sosnowski [40] also recorded similar results, with $530 \text{ g kg}^{-1} \text{ DM}$ in *Dactylis glomerata*. In studies on the *Festulolium*-alfalfa mixture, the same author [19] recorded slightly lower content (480.0 g kg^{-1}). This relationship was confirmed by Salama and Navar [3], indicating that generally NDF content of grasses was higher by about 8% than that of legume plants. Tomic et al. [42] recorded as much as $636.0 \text{ g kg}^{-1} \text{ DM}$ of NDF fraction in *Dactylis glomerata*. In the studies of Wiśniewska-Kadżajan and Stefaniak [41], the highest *Festulolium braunii* content was recorded in the second cut (532.1 g kg^{-1}) and the smallest in the third ($518.9 \text{ g kg}^{-1} \text{ DM}$). Similar results were obtained by Sosnowski and Jankowski [43], who in a *Festulolium* mixture with alfalfa also recorded the highest concentration of this parameter in the second cut. In the present experiment, NDF content in individual cuts of *Festulolium braunii* did not significantly differ.

3.3. ADF Content

ADF fraction is constituted by the least digestible forage ingredients, such as cellulose, lignin and silica [5]. Its content varied according to treatments, growing seasons and cuts (Table 5). In all years of research the highest was in plants on plots with the higher dose of Tytanit, with 381.7, 374.2 and 367.8 g kg⁻¹ DM, respectively.

Table 5. Acid detergent fibre (ADF) content in *Festulolium braunii* (g kg⁻¹ DM).

Treatment	Means across Growing Seasons				Means across Cuts			
	Years				Cuts			
	2015	2016	2017	Mean	I	II	III	Mean
0	358.7	336.7	329.2	341.5 E	237.0	220.8	318.7	258.8 AB
N ₁	368.6	324.9	362.2	351.9 CD	235.1	218.5	341.9	265.2 A
N ₂	347.8	374.5	352.8	358.4 BC	233.2	215.85	343.1	264.1 AB
Ti ₁	348.0	355.8	319.7	341.2 E	230.8	213.27	322.2	255.4 AB
Ti ₂	381.7	374.2	367.8	374.6 A	228.7	210.68	358.4	265.9 A
N ₁ + Ti ₁	368.5	360.6	364.9	364.7 B	226.0	207.31	349.8	261.1 AB
N ₂ + Ti ₂	323.0	355.9	358.7	345.9 DE	223.4	204.08	317.7	248.4 B
mean	356.6 a	354.7 ab	350.8 b	354.0	230.6 a	212.93 b	336.0 a	259.8

Different lowercase letters in the same row or different uppercase letters in the same column indicate significant differences between treatments.

Lower values were recorded by Wiśniewska-Kadžajan and Stefaniak [41], with the average ADF content in *Festulolium braunii* in the range of 320–350 g kg⁻¹ DM. Similarly, lower ADF fraction content in the *Festulolium braunii* biomass (332.0 g kg⁻¹ DM) was reported by Olszewska and Kobylński [2]. According to Linn and Martin [31], a reduction of its amounts improves forage digestibility. Thus, its content is a good indicator of roughage quality. According to Brzóska and Śliwiński [5], in traditional forage for dairy cows, ADF content in dry matter should be between 190 and 210 g kg⁻¹.

In the first year, it was the lowest in *Festulolium braunii* treated with the higher dose of nitrogen applied together with Tytanit (323.0 g kg⁻¹ DM), in the second (324.9 g kg⁻¹ DM) with the lower dose of nitrogen (80 kg N ha⁻¹) and in the third (319.7 g kg⁻¹ DM) with the lower dose of Tytanit (0.2%). Across growing periods (Table 5) nitrogen doses did not cause significant differences. However, Jankowska [44] reported that increasing nitrogen doses reduced the share of ADF fraction in meadow hay. However, the higher dose of Tytanit of 1%, compared to the lower one, with 0.2%, resulted in its significant increase, from 341.2 to 374.6 g kg⁻¹ DM. In turn, the combined higher dose of nitrogen and Tytanit contributed to a significant decrease from 364.7 to 345.9 g kg⁻¹ DM. Sosnowski [40] reported that in *Festulolium braunii* it was 323 g kg⁻¹ DM, and the UGmax soil conditioner and mineral fertilisers did not significantly change the value. The ADF content increased (up to 333 g kg⁻¹ DM) due to the interaction of UGmax soil fertiliser with mineral fertilization.

Average ADF content in *Festulolium* across treatments was 354.0 g kg⁻¹, slightly higher than the standard recommended by Brzóska and Śliwiński [5]. In similar studies Grzelak and Bocian [38] recorded slightly lower content in hay (277–343 g kg⁻¹ DM) and Jankowska-Huflejt and Wróbel [39] in pasture plants (285–300 g kg⁻¹ DM). In turn, Sosnowski [45] found comparable ADF fraction content in *Lolium perenne* (334 g kg⁻¹ DM) and *Dactylis glomerata* (356 g kg⁻¹ DM). Significantly higher values in both of those grass species were recorded by Tomic et al. [42], with 326 and 374 g kg⁻¹ DM, respectively. Comparing the years of research, as average for treatments, the most ADF was in the first (356.6 g kg⁻¹) and the least in the third (350.8 g kg⁻¹) and only between these years the difference was statistically significant. On the other hand, on average, across growing periods, the lowest content was in the plants on the plot with the lower dose of Tytanit (341.2 g kg⁻¹ DM), differing significantly from the higher dose (374.6 g kg⁻¹). An increase in ADF content was also observed as an effect of nitrogen applied on its own, but these differences were not statistically significant. In turn, the higher dose of nitrogen and Tytanit resulted in a

significant decrease from 364.7 to 345.9 g kg⁻¹ DM. On average, across growing periods ADF content in control plants was lower than in those treated with fertiliser combinations, except for the lower dose of Tytanit. Acid detergent content in *Festulolium* biomass varied significantly between cuts (Table 5). On average, plants from the third cut contained its highest amounts (335.9 g kg⁻¹ DM), significantly more than in the first (230.9 g kg⁻¹ DM) and the second (212.9 g kg⁻¹ DM). Sosnowski and Jankowski [43] recorded very similar ADF content in a mixture of alfalfa with *Festulolium* (322.8 g kg⁻¹ DM) in the third cut. The average values across cuts of all years indicated that the combined higher doses of nitrogen and Tytanit reduced ADF content compared to the effects of other fertiliser treatments. Thus, the interaction of experimental factors affected ADF content. According to Acar et al. [46], increased temperature and limited humidity have a major impact on changes in plant cell wall chemistry, deteriorating feed quality.

3.4. ADL Content

According to Kotlarz et al. [47], the content of ADL is an indicator of how advanced the process of plant lignification is. Lignin makes it difficult to attach bacteria to the cell walls, as a result of which digestion is limited. Therefore, as ADL content in the forage increases, its digestibility deteriorates. In the present experiment (Table 6), its average value was 39.91 g kg⁻¹, but its amounts varied for both treatment combinations, growing seasons and cuts. In the research of Wiśniewska-Kadzajan and Stefaniak [41], ADL content in *Lolium multiflorum* was 39–41 g kg⁻¹ DM, while in *Festulolium braunii* it was slightly higher (40–45 g kg⁻¹) than in the present experiment.

Table 6. Acid detergent lignin (ADL) content in *Festulolium braunii* (g kg⁻¹ DM).

Treatment	Means across Growing Seasons				Means across Cuts			
	Years				Cuts			
	2015	2016	2017	Mean	I	II	III	Mean
0	33.32	35.22	42.33	36.96 D	37.21	37.41	36.24	36.95 D
N ₁	43.92	38.13	43.56	41.87 AB	43.15	40.99	41.45	41.86 AB
N ₂	34.53	41.48	47.69	41.23 B	40.27	40.60	42.82	41.23 B
Ti ₁	45.99	38.12	45.10	43.07 A	42.34	44.37	42.49	43.07 A
Ti ₂	44.75	36.09	35.67	38.84 C	38.57	39.80	38.13	38.83 C
N ₁ + Ti ₁	28.63	35.57	42.10	35.43 D	34.72	35.64	35.92	35.43 D
N ₂ + Ti ₂	44.76	37.03	44.19	41.99 AB	40.31	43.47	42.18	41.99 AB
mean	39.41 b	37.37 c	42.95 a	39.91	39.51 a	40.33 a	39.89 a	31.91

Different lowercase letters in the same row or different uppercase letters in the same column indicate significant differences between treatments.

The content varied greatly for all growing seasons and was highest in the third year (42.95 g kg⁻¹) and the lowest in the second (37.37 g kg⁻¹ DM). The differences between growing seasons were statistically significant. During a growing season with the lowest rainfall, Jankowska [44] found the lowest values in permanent meadow forage. In the present experiment, the greatest drought was in the second year (2016), especially in April, June, July and August. Similar values were recorded for meadow hay by Grzelak [48]. Wróbel et al. [49] recorded a much higher value of about 45 g kg⁻¹ in the dry matter of meadow forage.

In the first year, the most favourable ADL content was found on the plot with the combined lower dose of nitrogen and Tytanit (28.63 g kg⁻¹ DM). Furthermore, the higher dose of nitrogen used on its own resulted in its statistically significant reduction (from 43.92 to 34.53 g kg⁻¹ DM). In the second and third years, however, this trend was reversed, also with a statistically proven difference. By contrast, supplying plants with the higher dose of Tytanit resulted in a reduction in the ADL content in all years of research, with significant differences in the second and third years. The combined application of the higher dose of nitrogen and Tytanit resulted in its significant increase in all years, to 44.76, 37.03 and

44.19 g kg⁻¹ DM. Sosnowski [40] recorded *Festulolium braunii* ADL fraction content of 44.4 g kg⁻¹ DM on average, but a soil conditioner slightly reduced it (to 41.0 g kg⁻¹ DM). On the other hand, studies by Barszczewski et al. [50] found that in meadow plants it ranged from 37.6 to 52.9 g kg⁻¹ DM.

Compared to the lower one, the higher dose of Tytanit significantly reduced ADL content, ranging across growing seasons from 43.07 to 38.84 g kg⁻¹. In contrast, the combined higher dose of nitrogen and Tytanit significantly increased it from 35.43 to 41.99 g kg⁻¹. The values (Table 6) did not differ significantly between three cuts, ranging from 39.51 to 40.33 g kg⁻¹ DM. In turn, Barszczewski et al. [50] found that higher content of ADL in the forage of the second cut and lower in the first and third. The average values across cuts of all years indicated that of all fertiliser treatments the combined lower doses of Tytanit and nitrogen had the most favourable effect on ADL content.

3.5. Dry Matter Intake (DMI)

According to the data presented in Table 7, *Festulolium braunii* dry matter intake (DMI) averaged 2.23%. Similar values for *Festulolium braunii* at the level of 2.30% were obtained by Olszewska and Kobylński [2]. The values recorded in the experiment were similar to those published by Jankowska-Huflejt and Wróbel [39], ranging from 1.96% for hay to 2.42% for pasture plants. Moore and Undersander [51] also reported values for feedstuff to be between 1.93 and 2.17%.

Table 7. Dry matter intake (DMI) of *Festulolium braunii* (%).

Treatment	Means across Growing Seasons				Means across Cuts			
	Years				Cuts			
	2015	2016	2017	Mean	I	II	III	Mean
0	2.23	2.13	2.16	2.17 A	2.19	2.28	2.13	2.22 A
N ₁	2.20	2.30	2.26	2.25 A	2.26	2.15	2.29	2.23 A
N ₂	2.23	2.32	2.11	2.20 A	2.13	2.17	2.23	2.18 A
Ti ₁	2.32	2.21	2.26	2.26 A	2.23	2.31	2.33	2.29 A
Ti ₂	2.23	2.25	2.09	2.19 A	2.24	2.14	2.22	2.20 A
N ₁ + Ti ₁	2.37	2.28	2.23	2.29 A	2.30	2.30	2.28	2.29 A
N ₂ + Ti ₂	2.32	2.23	2.22	2.26 A	2.26	2.27	2.22	2.25 A
mean	2.28 a	2.25 a	2.19 a	2.24	2.23 a	2.23 a	2.24 a	2.24

Different lowercase letters in the same row or different uppercase letters in the same column indicate significant differences between treatments.

On average, across growing periods the largest dry matter intake was recorded for plants treated with both the higher and the lower combined doses of Tytanit and nitrogen, as well as with the lower dose of Tytanit and with the lower dose of nitrogen, applied on their own. Similarly, Sosnowski and Jankowski [43] demonstrated that a higher dose of nitrogen reduced it.

The lowest DMI of *Festulolium braunii* was recorded on the control plot (2.17%) but also on those with the higher doses of nitrogen (2.20%) and Tytanit (2.19%). Slightly higher values of about 2.50% were recorded by Sosnowski and Jankowski [43] and Sosnowski [40] in a *Festulolium*-alfalfa mixtures. Across the cuts, lower DMI were recorded for grass from the first and second harvest (2.23%), slightly higher from cut III (2.24%). Differences in *Festulolium* values across treatments, growing seasons and cuts were generally not higher than a fraction of a percent and did not differ significantly. It can therefore be assumed that they were of little importance in assessing DMI.

3.6. Relative Feed Value (RFV)

Roughage nutrients should cover needs of animals resulting from their productivity and requirements particular to their species. On the basis of ADF and NDF content, a comprehensive assessment of forage quality was made, calculating RFV, which combines

digestibility and feedstuff intake into a single parameter [44]. Based on the RFV classification provided by Linn and Martin [31], *Festulolium braunii* forage was assigned to quality classes adequate for specific animal groups (Table 2). For *Festulolium braunii* RFV varied across treatment combinations, growing seasons and cuts (Table 8). Across growing seasons its average value was 106.0, which, according to Linn and Martin [31], made it Class 3 forage, intended for beef cattle, older heifers, and, to a lesser extent, for dairy cows. As an average effect of fertiliser treatments across growing seasons, the most favourable RFV (106.0) was recorded in plants treated with the lower dose of Tytanit.

Table 8. Relative feed value (RFV) of *Festulolium braunii* dry matter.

Treatment	Means across Growing Seasons				Means across Cuts			
	Years				Cuts			
	2015	2016	2017	Mean	I	II	III	Mean
0	103.1	103.6	106.0	104.2 A	102.8	107.7	105.3	105.3 ABC
N ₁	102.7	114.7	106.3	107.9 A	107.8	101.6	112.4	107.3 AB
N ₂	104.8	106.1	100.6	103.8 A	99.4	101.3	108.7	103.1 BC
Ti ₁	111.1	102.9	112.0	108.7 A	105.8	103.6	111.1	106.8 AB
Ti ₂	102.3	109.2	97.29	102.9 B	102.5	99.7	104.9	102.4 C
N ₁ + Ti ₁	110.7	108.2	101.3	106.7 A	105.9	102.2	106.2	104.8 ABC
N ₂ + Ti ₂	114.6	105.9	103.4	108.0 A	106.2	107.9	109.9	108.1 A
mean	107.0 a	107.2 a	103.8 b	106.0	104.3 a	103.4 a	108.4 a	106.0

Different lowercase letters in the same row or different uppercase letters in the same column indicate significant differences between treatments.

On average over the years of research, only plants from the plot with the higher dose of Tytanit (102.9) should be classified as Class 4, to be fed to beef cattle and dried dairy cows. Similarly, Wiśniewska-Kadzajan and Jankowski [52] pointed out that the RFV of *Lolium multiflorum* ranging from 93.7 to 107.8 and of *Festulolium braunii* ranging from 95.0 to 110.4 was assigned to Class 3 and 4.

Across growing seasons, the highest RFV was recorded in the first year (107.0). In the third year it was by nearly four units lower, with 103.8, making a statistically significant difference. On the other hand, higher values were recorded in the third cut (108.4) than in the first and second, but the differences were not statistically significant. Average values across treatments and cuts qualified *Festulolium braunii* forage as Class 3.

3.7. Correlation Coefficient

Average of the growing seasons, correlation coefficients (Table 9) indicated that when the NDF content of the *Festulolium braunii* increased, RFV and DMI decreased. In addition, RFV decreased as ADF content increased. In turn, when RFV increased, DMI also increased. Those were very strong correlations.

Table 9. Linear correlation coefficients between *Festulolium braunii* NDF and ADF content and other characteristics.

Variable	Means across Growing Seasons				
	NDF	ADF	ADL	RFV	DMI
	for NDF				
NDF	1.00				
ADF	0.087	1.00			
ADL	0.120	−0.184	1.00		
RFV	−0.762 *	−0.540 *	−0.001	1.00	
DMI	−0.951 *	−0.015	−0.106	0.773 *	1.00

Table 9. Cont.

Variable	Means across Growing Seasons				
	NDF	ADF	ADL	RFV	DMI
	for ADF				
ADF	1.00				
NDF	0.087	1.00			
ADL	−0.037	−0.028	1.00		
RFV	−0.539 *	−0.762 *	0.092	1.00	
DMI	0.095	−0.388	0.241	0.165	1.00

$p \leq 0.05$; $n = 189$; * significant difference.

4. Conclusions

The higher dose of Tytanit supplied to *Festulolium braunii* contributed to an increase in NDF and ADF fraction content and to a reduction in RFV and ADL fraction and DMI. The combination of Tytanit and nitrogen, especially at the higher dose, resulted in an increase in the content of NDF, ADL and RFV in *Festulolium braunii*, reducing the amounts of ADF and DMI.

Nitrogen supplied to *Festulolium braunii* on its own, especially at the higher dose, resulted in an increase in NDF and ADF content while reducing RFV and the amounts of ADL and DMI.

Of all the treatments, the lower dose (0.2%) of Tytanit resulted in the most favourable effect on *Festulolium braunii* ADF content and RFV. On the other hand, the other parameters, i.e., the content of NDF, ADL and DMI, were most favourable as a response to the combined lower dose of Tytanit and nitrogen. Fertiliser combinations affected *Festulolium braunii* parameters compared to those of the control, having a positive impact on forage quality. As the average across growing periods, ADF content in control plants was lower than in those treated with fertiliser combinations, except for the lower dose of Tytanit.

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