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Research Paper

The Effects of Different Humic Acid and Seaweed on Some Yield and Yield Components of Ryegrass

Abdullah Özköse[1, *], Ramazan Acar[1] Rana Ertuğrul[1] Havvanur Çobanoğlu[1]

[1]*Selcuk University, Faculty of Agriculture, Department of Field Crop, Konya, 42025, Türkiye*

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Abstract: Livestock farm need forage crops that have high yields and good nutritional value per unit area. One of the plants that can contribute to covering this need is ryegrass. In the present study, the purpose was to determine the effects of biostimulants as organic fertilizer and their doses on some yield and yield components of ryegrass. The study was conducted under Konya conditions in 2017 with 3 replications according to the randomized blocks design. According to the results of the study, stem length was found to be 45.9-51.4 cm, stem diameter was 2.07-3.07 mm, the number of leaves on the main stem was 3.8-4.3, leaf length was 31.3-36.1 cm, leaf width was 5.8-6.8 mm, green fodder yield was 5303-6166 kg/da, and hay yield ranged between 989 and 1157 kg/da. Although a difference was detected between the variable that were observed, it was not found to be statistically significant. Since the yield and nutritional values of ryegrass may vary according to varieties, regions and seasons, the data obtained are preliminary information for researchers working on this subject. It would be useful to conduct more studies on location, year, etc. to confirm the findings of the study.

Keywords: *Humic acid, Lolium multiflorum, organic fertilizer, ryegrass, seaweed, yield*

Introduction

Lolium multiflorum (annual ryegrass or Italian ryegrass) is native to central and southern Europe, north-west Africa and south-west Asia (Hubbard, 1968; Soya et al., 1997). It is also found naturally in Türkiye. Today, it is an important fodder plant which is cultivated almost all over the world. Ryegrass is an annual, sometimes biennial at high altitudes, dense-tillering grass-forming forage crop used for green fodder, hay, silage, and grazing purposes. It has a long growing period and is highly productive when adequate fertilization and maintenance are made under suitable climatic conditions (Aganga et al., 2004). The fact that it can be mowed more than once a year, has a high nutritional value, is eaten by animals with appetite, is suitable for grazing and frequent mowing, and it also can be silaged as a mixture or pure makes ryegrass valuable.

The yield and nutritional values of ryegrass vary among cultivars, regions, and growing techniques. Fertilization, in particular, has the effect of increasing yield and quality. It is necessary to use more fertilizers, especially nitrogen fertilizers for high yields in ryegrass, which has a negative effect on the production costs and the environment. It is necessary to reduce the use of excessive inorganic fertilizers for sustainable production in ryegrass and to increase the use of organic fertilizers.

The deficiency of organic matter is common in the soils of the region with a continental climate. Agricultural techniques and climate are effective in this. Soil organic matter loss is caused by many factors such as wrong tillage techniques, erosion, stubble burning, and overgrazing (Demiray et al., 2023). The low organic matter of the soil is among the most important obstacles to sustainability in agriculture. Organic matter affects the physical, chemical, and biological characteristics of soils positively (Adhikariet al., 2023; Demiray et al., 2023). Inadequate supply of organic fertilizers (i.e., peat, sheep manure, and compost from municipal solid wastes) and high transportation application costs limit their use. Also, the problem with the use of organic fertilizers is the large amount to be used and the difficulties in applying them together with other fertilizers (Asenjo, 2000). However, environmental awareness, rising costs of synthetic fertilizers, and high transportation costs require the use of renewable fertilizers such as seaweed extracts (Hunter, 2004). In our present day, as well as mineral fertilizers, organic matter of different origins, humic substances, seaweed extracts, amino acids, and biostimulants are employed as organic fertilizers. Humic acid and seaweed extracts are also

*Corresponding: E-Mail: aozkose@selcuk.edu.tr; Tel.+ +903322232888

classified as biostimulants, which positively affect plant growth, plant nutrition, product quality, and yield as materials that can be applied to plants from leaves, soil, or seeds to increase the resistance of plants to stress and may contain organic or inorganic compounds, microorganisms, and some of them also have regulating effects of the soil (Külahtaş and Çokuysal, 2016). As well as providing good root development, more nutrients, and water intake in plants, seaweed increases the resistance of the plant to diseases and pests, stress factors such as frost, drought, inadequate sun, excessive water, heat, and extreme cold (Yağmur et al., 2021). The effects of humic acid in agriculture facilitate the uptake of nutrients in the soil by plants, increasing the activities of microorganisms and the water holding capacity and air permeability of the soil as well as the resistance of plants to stress conditions with positive effects on resistance to pests and diseases under stress conditions (Külahtaş and Çokuysal, 2016; Yılmaz and Boz, 2022; Eryiğit and Husamalddin, 2023).

Organic fertilisers are better than chemical fertilisers instead of the use of the environmental pollution and prevent environmental pollution and utilisation of farm waste is very important (Demiray and Özaslan Parlak, 2023). In recent years, efforts to reduce the use of mineral fertilisers by increasing the use of organic fertilisers have accelerated. Therefore, in the present study, the effects of humic acid and seaweed fertilizers applied in different mowings and doses on the yield and yield components of ryegrass were determined.

Materials and Methods

The study was conducted to determine the effects of different organic fertilizers on yield and yield components of ryegrass under Konya conditions at the Research and Application Station of Field Crops Department, Faculty of Agriculture, Selcuk University, Konya, Türkiye, in 2017. Barsmultra II variety of ryegrass (*Lolium italicum* L. Syn. *L. multiflorum* Lam) obtained from a private brand was employed as a plant material.

The trial area where the study was conducted is located at the coordinates 38º02'N dan 31º30'E and at an altitude of about 1016 m above sea level. Konya is located in the southern part of the Central Anatolian Region and has a continental climate with harsh, cold, and snowy winters and hot and dry summers.

The climate data of 2017 and the long-term average of the months in which the study was conducted are given in Table 1. The average monthly temperature was the lowest in April at 10.8 °C, the highest in July and July at 25.2 °C, and the average temperature was 19.8 °C during the trial period. Compared to the long-term average (18.8 °C), it was warmer (19.8 °C) during the 2017 trial period. The average relative humidity was 46.4% in the months of the trial period in 2017, and lower than the long-term average (48.6%). Monthly rainfall was the lowest in July with 0.0 mm, the highest in May with 43.7 mm, and the total amount of precipitation was 131.1 mm in 2017. The total precipitation during the trial in 2017 was slightly higher than the long-term average (129.0 mm).

Table 1. Climate Data of Konya Province for 2017 and Long-term (LT) Average*

Month	Precipitation (mm)		Air Temperature ºC		Relative Humidity (%)	
	2017	LT	2017	LT	2017	LT
Apr	39.3	32.7	10.8	11.1	53.0	58.1
May	43.7	44.4	15.4	15.7	57.9	56.2
Jun	25.4	24.8	20.4	20.1	54.6	49.0
Jul	0.0	6.9	25.2	23.5	35.6	41.3
Aug	19.4	6.7	24.3	23.3	45.3	41.0
Sep	3.3	13.5	22.4	18.8	31.7	46.9
Total	131.1	129.0	-	-	-	-
Mean	-	-	19.8	18.8	46.4	48.6

Konya Climate Data for 2017. T.R. Ministry of Agriculture and Forestry 8th Regional Directorate of Meteorology, Konya.

According to the soil analysis, the area was identified to be clayey-loamy texture and alkaline characteristics (pH: 7.7). Organic matter amount 1.19%, EC (µS / cm): 193, P_2O_5:10.86 ppm, K_2O: 221.16 ppm, Zn: 2.12 ppm, Fe: 1.30 ppm, Cu: 0.82 ppm, Mn: 4.95 ppm, Ca: 5800.00 ppm and Na: 65.49 ppm were determined (Table 2).

Table 2. Some Physical and Chemical Characteristics of Trialal Field Soil*

Soil depth (cm)	pH	EC (µS/cm)	Texture	Organic substance (%)	P_2O_5 (mg/kg)	K_2O (mg/kg)
0-30	7.7	193	clayey-loamy	1.19	10.86	221.16
Soil depth (cm)	Ca (mg/kg)	Na (mg/kg)	Mn (mg/kg)	Zn (mg/kg)	Fe (mg/kg)	Cu (mg/kg)
0-30	5800.00	65.49	4.95	2.12	1.30	0.82

**Soil analyses were made in Konya Commodity Exchange Laboratories (KLD).*

Three different organic fertilizers were employed in the study. Liquid humic acid, granule humic acid, and foliar fertilizer obtained from seaweed were employed as organic fertilizers. Fertilizer contents are given below (the contents declared by the brands).

Liquid humic acid (LH)

Total organic matter	: 15%
Total humic + fulvic acid	: 15%
Water-soluble K_2O	: 2.5%
pH	: 9-11
Raw Material	: Leonardite + KOH

Granule Humic Acid (GH)

Organic matter	: 90%
Humic acid	: 50%
Amino acid	: 10%
Nitrogen	: 16%
Potassium (K_2O)	: 1%
Phosphorus (P_2O_5)	: 2%
Humidity	: 1%
pH	: 3-5

Seaweed (SW)

Organic matter	: 40%
Alginic acid	: 8%
Water soluble K_2O	: 15%
EC (dS/m)	: 36.2%
pH (with 10 distilled water)	: 7-9
Raw material	: Seaweed

The trial was set up according to the Random Block Design with 3 replications. The observations and measurements were made according to TTSM (Variety Registration and Seed Certification Center, Republic of Türkiye Ministry of Agriculture And Forestry) Technical Instructions (Lolium L. species) (Anonymous, 2001). The soil was plowed and then harrowed. During the soil preparation, 20 kg/da of DAP fertilizer was given as base fertilizer and mixed with the soil on October 7, 2017, by hand to a depth of 2-3 cm on the rows opened with a marker. The rows were then closed and pressed with a soil roller.

Plot dimensions were as follows

Row spacing	: 20 cm
Number of rows per plot	: 10 rows
Plot row length	: 4 m
Plot area in sowing	: 0.20 x 10 x 4 = 8 m^2
Plot area at harvest	: 0.20 x 8 x 3 = 4.8 m^2

The cultivation area of each plot was 8 m^2 and the data, yield and samples were taken from 4.8 m^2 areas, taking into account the edge effects at harvest. It is recommended to mow at the beginning of spike emergence in Italian ryegrass (Özköse et al., 2015). The Italian ryegrass variety used in the study did not show a tendency to spike. Therefore, plant height was taken as a basis in determining the harvest. Three doses of 3 different organic fertilizers were tried. In determining the doses, the

manufacturer's recommendations for Italian ryegrass were taken into consideration. The characteristics of the fertilizers and their doses were as follows:

Liquid humic acid : 5 L/da; 10 L/da 15 L/da
Granule humic acid : 5 kg/da; 10 kg/da 15 kg/da
Seaweed : 50 g/da; 100 g/da; 150 g/da

Liquid and granule humic acids were applied to the soil before planting, and seaweed fertilizer was applied to the leaves one week after each mowing. Also, pure nitrogen (N) is given to all plots, including the control, at 5 kg/da after each mowing. Irrigation and weed control were performed as cultural processes. The weeds in the plots were controlled by hand picking or hoeing. Irrigation was done according to the water needs of the plant and rainfall. There was no tendency to spike in the cultivar employed. The plants were mowed three times on 19 June, 27 July, and 21 September 2017. Plant height (cm), stem thickness (mm), number of leaves (pieces), leaf length (cm), leaf width (mm), green fodder yield (kg/da), and hay yield (kg/da) observations and measurements were performed in the study.

The data obtained in the study were subjected to analysis of variance in the MSTAT-C program with 3 replication according to the experimental design in randomized block design. The LSD test was employed to compare the means, and the means were grouped at $p<0.01$ or $p<0.05$ according to the significance level determined as a result of the variance analysis.

Results and Discussion

Three doses of three different organic fertilizers were tried, and three mowings were conducted during the study. Plant height, stem thickness, number of leaves, leaf length and leaf width were determined by taking the average of three mowings, while green fodder yield and hay yield were determined by taking the sum of three mowings. The effects of fertilizers and doses employed in the study on all the varisbles examined were not found to be significant (Table 3).

Table 3. Yield elements of the ryegrass to which different organic fertilizers and doses

Fertilizer	Plant Height (cm)	Stem Thickness (mm)	Number of leaves (pcs)	Leaf Length (cm)	Leaf Width (mm)	Green Fodder Yield (kg/da)	Hay Yield (kg/da)
Control	47.2	3.03	4.3	34.0	6.8	5629	1088
LH1	46.1	2.73	4.1	32.0	6.3	5469	1065
LH2	49.5	2.90	4.2	35.8	6.3	5808	1075
LH3	46.6	2.97	4.1	34.9	6.0	5714	1066
GH1	51.4	2.93	4.3	36.1	6.8	5916	1132
GH2	49.9	3.07	4.0	35.7	6.5	6166	1114
GH3	49.2	2.87	4.0	33.1	6.3	6166	1157
SW1	45.9	2.70	4.0	31.3	5.8	5703	1109
SW2	46.0	3.03	4.1	32.9	6.3	5303	989
SW3	48.2	2.77	3.8	35.1	6.1	5814	1102
LSD	ns	ns	ns	ns	ns	ns	ns

LH= Liquid Humic Acid; LH1: 5 L/da; LH2: 10 L/da; LH3: 15 L/da; GH= Granule Humic Acid; GH1: 5 kg/da; GH2: 10 kgt/da; GH3: 15 kg/da; SW= Seaweed: SW1: 50 g/da; SW2: 100 g/da; SW3: 150 g/da; ns= Not Significant

Plant Height (cm)

The lowest plant height was measured in SW1 treatment at 45.9 cm and the highest in GH1 treatment at 51.4 cm. However, the effect of the fertilizer types and doses employed on the average plant height was not statistically significant. Plant height in ryegrass was determined by some other researchers to be between 48.1 cm (Özdemir, 2017) and 123.8 cm (Sever, 2021). The differences in plant height of ryegrass depended to nitrogen fertiliser doses (Kesiktaş, 2010; Çolak, 2015; Çetin, 2017; Pak Örün, 2019), the number of mowings (Kuşvuran and Taysı 2005), inter-row distance (İnce, 2000), mixture rates with legumes (Özkan, 2017; Sever, 2021), varieties (Aktar, 2019; Acar, 2020) and mowing times (Özköse et al. 2015). The Italian ryegrass cultivar used in the study had a short plant height. Because the Italian ryegrass cultivar used in the study did not show a tendency to spike.

Main Stem Thickness (mm)
The stem diameter varied between 2.70 and 3.07 mm. However, the effect of the fertilizer types and doses employed on the average stem thickness was not statistically significant. This result was similar with several previous study that has been reported. Özköse et al. (2015) reported that the stem diameter of ryegrass ranged from1.69 to 3.75 cm, Çolak (2015) reported results ranged form 2.92 to 3.69 mm, Çetin (2017) reprted results ranged from 3.20 to 3.80 mm, Pak Örün (2019) reported the results ranged from 2.74 to 3.32 mm and Sever (2021) also reported the results ranged from 2.42 to 3.19 mm. Although there were similarities between the results of the present study and the results of the other researchers, there were also some differences. The reason for this may be the different cultivars employed in the studies, the effect of the trials, and the climate, soil, and growing conditions in which the trials were conducted.

Number of Leaves (pcs/stem)
The number of leaves was determined instead of the number of nodes on the main stem since the ryegrass variety used in the study did not have stem emergence. Italian ryegrass cultivar used in the study has vernalisation request. When it is sown as summer in spring, it does not show a tendency to spike. In the above-ground part of the plant, the number of leaves is the same as the number of internodes since one leaf emerges from each node. In the study, the number of leaves on the main stem varied between 3.8 and 4.3. The number of leaves obtained in the study was similar to the number of nodes obtained by Darvishi (2009) (3 – 4), Özköse et al. (2015) (4.2 – 5.9), Anonymous (2015) (3 – 7), and Anonymous (2016) (4 – 7) were close to or slightly lower than the number of nodes.

Leaf Length (cm)
The leaf length obtained in this study was 31.3 - 36.1 cm, higher than Darvishi (2009) (14.0 - 20.0 cm) and similar to Özköse et al. (2015) (10.2 - 37.2 cm), Anonymous (2015) (16.8 - 36.3 cm) and Anonymous (2016) (16.7 - 42.5 cm). In the present study, the variation range of the average leaf length is narrow because of the use of a single variety and the small effect of fertilizer applications. However, the variation ranges were high because of the use and applications of many cultivars in the studies of other researchers.

Leaf Width (mm)
The leaf width varied between 5.8 and 6.8 mm. The data obtained were lower than the flag leaf width of 7.2 – 9.5 mm obtained by Darvishi (2009), 8.0 – 17.0 mm determined by Anonymous (2015) and 8.4 – 15.0 mm determined by Anonymous (2016). The difference between the studies may be due to the genotype, climate and soil conditions of the growing region and differences in agricultural practices.

Green Fodder Yield (kg/da)
Green fodder yield of ryegrass varied between 5303 – 6166 kg/da and the effect of the fertilizers and their doses on yield was statistically insignificant. There may be many reasons for this result. These are; insufficient doses of the fertilisers used, deficiency of other elements in the soil limiting the yield, poor response of the variety used to the fertilisers applied, climatic conditions in the study year, especially high temperature, and many factors may have separate or combined effects. Compared to the results in the literature, the green fodder yield obtained from the study result was similar to the green grass fodder reported by Gültekin (2008) with 3313.7 – 6591.8 kg/da; higher than the results obtained by Darvishi (2009) with 2626.4 – 3439.0 kg/da, Çolak (2015) with 845.4 – 1931.7 kg/da, Kesiktaş (2010) with 1334.6 – 1814.5 kg/da, Kuşvuran and Taysı (2005) with 2984.1 – 3102.0 kg/da, Demiray and Özaslan Parlak (2023) with 2481.7 – 4948.3 kg/da and Rahetlah et al. (2013) with 1468.0 kg/ha; but lower than the results obtained by Anonymous (2015) with 7033.1 – 12758.5 kg/da and Anonymous (2016) with 9620.1 – 11293.5 kg/da.

Hay yield (kg/da)
The hay yield varied between 989 and 1157 kg/da. However, the effect of the fertilizer types and doses employed on the average hay yield was not statistically significant. The fact that the difference

between the hay yields according to the applied fertiliser and doses was not statistically significant may be due to the fact that the applied doses were not sufficient to affect the yield of Italian ryegrass, which has a high yield capacity. Italian ryegrass variety used in the research has vernalisation request. When it was sown as summer in spring, it did not show a tendency to spike. Although the plant formed abundant leaves, the dry matter content of the leaves was low and this affected the herbage yield. The region where the experiment was conducted is hot and low relative humidity during summer months. Even if enough irrigation is done, water loss is high with evapotranspiration. The growth of Italian ryegrass, which is a cool climate plant, slows down in summer. High temperature and humidity is one of the most important factors limiting the yield. This situation limits the effect of applied fertiliser doses on yield. When compared with the results in the literature, the hay yield obtained in the present study was similar to the yield reported in the study by Özköse et al. (2015) (812.2 – 1855.6 kg/da), Gültekin (2008) (781.4 – 1294.2 kg/da), Darvishi (2009) (922.7 – 1643.2 kg/da), and higher than that reported by Çolak (2015) (224.4 – 455.9 kg/da), Kesiktaş (2010) (398.7 – 550.2 kg/da), Kuşvuran and Tansı (2005) (642.2 – 731.0 kg/da), Pavinato et al. (2014) (485 – 525 kg/da), Rahetlah et al. (2013) (303 kg/da), and lower than that reported by Anonymous (2015) (1678.3 – 2902.4 kg/da) and Anonymous (2016) (2219.0 – 2580.8 kg/da). Genetic potential of the variety used; soil characteristics; climatic conditions such as temperature, relative humidity, precipitation; agricultural practices such as irrigation, types and amounts of fertiliser applied, amount of seed sown, weed and pest control are effective on the yield of Italian ryegrass. These factors individually or together may have an effect on the similarity or difference with the results of other researchers.

Conclusion

The study was conducted in Konya conditions for one year in 2017 and three different doses of three different organic fertilizers were used. However, the effects of fertilizers and doses on all yield and yield components were not found to be significant. The applied doses of organic fertilizers did not increase the yield even compared to the control group. It would be useful to conduct more studies on location, year, etc. to confirm the findings of the study. In order to make more reliable conclusions according to these results, further studies should be carried out for a few more years under different climatic and soil conditions, detailed soil analysis of the experimental area according to different depths and supplementing the missing macro and micro element content, and using more ryegrass varieties instead of a single variety.

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