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IMPACT OF FOLIAR TREATMENT WITH HUMATE FERTILIZER ON THE BIOPRODUCTIVE INDICATORS OF A NATURAL MEADOW OF *Chrysopogon gryllus* TYPE IN CONDITIONS OF THE CENTRAL BALKAN MOUNTAIN

Tatyana BOZHANSKA, Minko ILIEV, Magdalena PETKOVA, Biser BOZHANSKI

Research Institute of Mountain Stockbreeding and Agriculture, 281 Vasil Levski Street, 5600 Troyan, Bulgaria

Corresponding author email: tbozhanska@mail.bg

Abstract

The experiment was conducted in the region of the Central Balkan Mountain (Bulgaria) in the period 2013-2015. Humate fertilizers were the objective for the present research was humate fertilizers (Phosphorus humate, Boron humate and Molybdenum humate) applied individually and in combination (at the stage of active vegetation of grass species) on the natural grass stand of Chrysopogon gryllus type. Grass stands treated with Molybdenum humate alone (160 ml/da) formed the highest yield of fresh (570.0 kg/da) and dry (195.6 kg/da) mass, which was proven (P <0.01). While the variants with foliar treatment by a combination of biofertilizers, such as Phosphorus humate (250 ml/da) + Boron humate (100 ml/da) + Molybdenum humate (100 ml/da), registered the lowest values (470.0 kg/da of fresh mass and 155.7 kg/da of dry mass). The percentage and species share of the main biological groups (grasses, legumes, motley grasses) in the natural grass stand changed in a positive way from the first to the third experimental year. The share of both main species Chrysopogon gryllus (characterizing the grass community) and motley grasses decreased, while there was an increase of Agrostis capillaris and legume meadow grasses (Trifolium campestre and Lotus corniculatus) in the above ground mass. The above data presupposes the formation of biomass with better qualitative and quantitative indicators.

Key words*: Chrysopogon gryllus L., natural grass stand, productivity, botanical composition.*

INTRODUCTION

The treatment of agricultural crops with biofertilizers is one of the main emphasis in the program for building sustainable agriculture and maintaining soil fertility (Kostadinova & Popov, 2012; Saini & Kumar, 2014; Georgiev et al., 2017; Bozhanska et al., 2019; Guilherme et al., 2020). The application of organic products of natural origin has a beneficial effect on plants, development and nutritional regime of plants (Churkova, 2012; Churkova & Bozhanska, 2016). Organic products with complex action and high content of labile humate substances perform a corrective function in the integrated "soil-plant" system. Biofertilizers contain formulations of live microorganisms, enzymes and microelements in an easily accessible form that facilitate seed germination and viability (Han et al., 2007; Bhardwaj et al., 2014; Bozhanska & Naydenova, 2020). The introduction of humate products stimulates the growth of the root system, increases the utilization rate of nutrients (Datta et al., 2011; Churkova, 2013a,b) and improves the photosynthetic activity and carbohydrate metabolism (Sengalevich, 2007) of plants. The realized aboveground mass is of better quality and is used more efficiently as a source of animal feed (Naydenova et al., 2013; Naydenova et al., 2014; Bozhanska et al., 2017; Churkova, 2019). Methodical and regulated fertilization creates conditions for changes in the phytocenological and qualitative profile of natural grass communities (Samuil et al., 2013; Iliev, 2018) by stimulating or suppressing the share of main grass species in meadow and pasture grass stands (Maruşca et al., 2014). As a result, there is degradation in the percentage share of low-quality plant species and an advantage of grasses and legumes, which provide easily digestible, energy and protein balanced feed biomass (Naydenova et al., 2013; Vasileva & Enchev, 2018).

In Bulgaria, the researches related to the application of humate fertilizers on natural grass stands are limited and insufficient. Therefore, the objective of the present study is to observe the impact of annual foliar treatment with humate fertilizer on the bioproductive indicators of a natural meadow of *Chrysopogon gryllus* type in conditions of the Central Balkan Mountain.

MATERIALS AND METHODS

The experiment was conducted in the spring (20th of April) of 2013 on a natural grass stand of *Chrysopogon gryllus* type (the predominant species were bunch grasses and common bent) in the main part of the Central Balkan Mountain. The experimental design was a block method with 4 replications. The plot size was 5 m^2.

In terms of climate, the territory belongs to the Pre-Balkan (Mountain) climate region of the temperate-continental climate subregion (Sabev & Stanev, 1963). Temperatures bear the marks of continental influence. The average annual temperature is 10-11°C and is characterized by territorial differentiation (from north to south) with increasing altitude (Ninov, 1997). The distribution of precipitation is uneven (maximum in summer: 309.0 mm and minimum in winter: 168.0 mm). In the spring and autumn the amount of precipitation is 242.0 mm and 209.0 mm, respectively, and the annual amount of precipitation reaches from 567.0 mm to 1200.0 mm.

The manifestation of climatic indicators is an important factor (Andreeva et al., 2015) determining the impact of humate fertilizers included in the experiment on the development and productivity of natural grass stand. The vegetation precipitation in 2013 was 658.7 mm at an average air temperature of 15.4°C (Figure 1). The lowest average air temperature (14.8°C) during the vegetation period, was registered in the second experimental year (2014), when the amount of precipitation reached maximum values (1045.9 mm) compared to the other experimental years. In 2015, the sum of precipitation for the period March-October was 787.9 mm, and the average air temperature was 15.9°C.

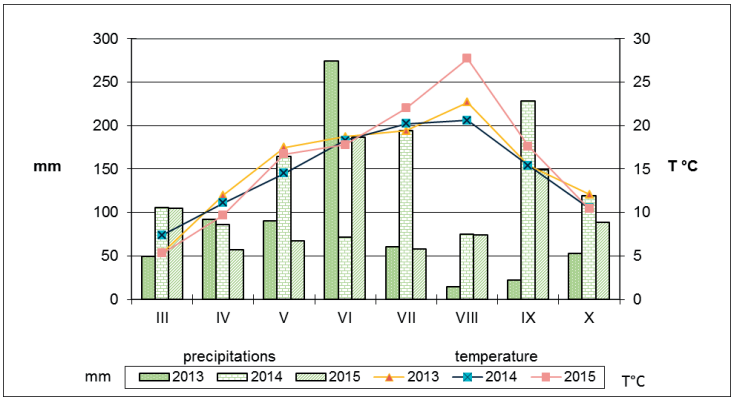

Figure 1. Average monthly temperatures (°C) and monthly precipitation amounts (mm) for the vegetation period (March-October) 2013-2015

Variants in the study were

1. Control (C)
2. Phosphorus humate (300 ml/da) - PH (300 ml/da)
3. Boron humate (160 ml/da) - BH (160 ml/da)
4. Molybdenum humate (160 ml/da) - MH (160 ml/da)
5. Phosphorus humate (250 ml/da) + Boron humate (100 ml/da) + Molybdenum humate (100 ml/da) - PH (250 ml/da) + BH (100 ml/da) + MH (100 ml/da).

Characteristics of the applied high polymeric humate fertilizers

Phosphorus humate contains: N - 6%; P - 18%; Organic carbon - 0.4%; Active substance - 98%; Organic substances; Humate acids;

Fulvic acids; Amino acids; Valine; Glutamine; Methionine; Lysine; Vitamins and Microelements.

Boron humate contains: N - 6%; B - 5%; Organic carbon - 0.4%; Active substance - 98%; Organic substances; Humate acids; Fulvic acids; Amino acids; Valine; Glutamine; Lysine; Methionine; Vitamins and Microelements.

Molybdenum humate contains: N - 6%; B - 5%; Organic carbon - 0.4%; Active substance - 98%; Organic substances; Humate acids; Fulvic acids; Amino acids; Valine; Glutamine; Methionine; Lysine; Vitamins and Microelements.

The foliar treatment with the tested fertilizers was applied once, every year. The working solution was imported with a back sprayer during the period of active vegetation for the grass species. During the three-year study period, three mowings were carried out on the grass cover (one in each of the experimental years), in the phase of hay hay harvesting stage of the grass stand, i.e. when the grasses are in the tasseling/ear formation and the legumes in the beginning of flowering.

Experimental data were statistically processed by analysis of variance (ANOVA).

RESULTS AND DISCUSSIONS

On average for the period, the highest yield of fresh (570.0 kg/da) and dry (195.6 kg/da) mass was registered in the variants with foliar treatment with Molybdenum humate (160 ml/da) - Tables 1 and 2. The amount of fresh mass of that variant exceeded the control by 33.1% (P <0.01). The grass stands treated with the combination of Phosphorus humate (250 ml/da) + Boron humate (100 ml/da) + Molybdenum humate (100 ml/da), registered the lowest values (470.0 kg/da of fresh mass and 155.7 kg/da of dry mass).

Table 1. Fresh mass yield (kg/da) of natural meadow of *Chrysopogon gryllus* type over the years and on average for the period of 2013-2015

Variants	2013		2014		2015		2013-2015	
	kg/da	% to C	kg/da	% to C	kg/da	% to C	kg/da	% to C
Control (C)	440.0	100.0	440.0	100.0	405.0	100.0	428.3	100.0
FH (300 ml/da)	500.0	113.6	540.0	122.7	450.0	111.1	496.7	116.0
BH (160 ml/da)	520.0	118.2	420.0	95.5	640.0	158.0	526.7	123.0
MH (160 ml/da)	550.0	125.0	460.0	104.6	**700.0**	**172.8***	**570.0**	**133.1***
FH (250 ml/da) + BH (100 ml/da) + MH (100 ml/da)	490.0	111.4	450.0	102.3	470.0	116.1	470.0	109.7
$LSD_{0.05}$	***144.9***	***32.9***	***103.8***	***23.6***	***273.7***	***67.6***	***125.6***	***29.6***
$LSD_{0.01}$	***203.4***	***46.2***	***145.7***	***33.1***	***384.2***	***94.9***	***176.3***	***41.5***
$LSD_{0.001}$	***287.2***	***65.3***	***205.7***	***46.7***	***542.4***	***133.9***	***248.9***	***58.6***

Table 2. Dry mass yield (kg/da) of natural meadow of *Chrysopogon gryllus* type over the years and on average for the period of 2013-2015

Variants	2013		2014		2015		2013-2015	
	kg/da	% to C	kg/da	% to C	kg/da	% to C	kg/da	% to C
Control (C)	150.4	100.0	147.4	100.0	191.4	100.0	163.1	100.0
FH (300 ml/da)	158.9	105.7	172.4	116.9	147.4	77.0	159.6	97.9
BH (160 ml/da)	167.0	111.1	144.0	97.7	227.5	118.8	179.5	110.1
MH (160 ml/da)	183.7	122.2	153.1	103.9	250.0	130.6	195.6	119.9
FH (250 ml/da) + BH (100 ml/da) + MH (100 ml/da)	155.4	103.4	145.9	98.9	165.8	86.6	155.7	95.5
$LSD_{0.05}$	***47.1***	***31.4***	***34.2***	***23.3***	***97.9***	***51.1***	***43.4***	***26.6***
$LSD_{0.01}$	***66.2***	***44.1***	***48.1***	***32.7***	***137.5***	***71.8***	***61.0***	***37.4***
$LSD_{0.001}$	***93.4***	***62.3***	***67.9***	***46.2***	***194.1***	***101.3***	***86.0***	***52.8***

In the first experimental year, the yield of **fresh mass** from a natural meadow of *Chrysopogon gryllus* type varied from 440.0 kg/da (Control) to 550.0 kg/da (MX - 160 ml/da). In percentage terms, the excess of the indicator in the treated grass stands compared to the control was from

11.4 to 25.0%. In the year (2014) with the lowest air temperature and the highest amount of vegetation precipitation, fertilization with Phosphorus humate (300 ml/da) had the highest effect on the productivity of the natural grass stand. The values of the indicator exceeded the control by 22.7%. In 2015, the effect of foliar treatment with Molybdenum humate (160 ml/da) led to a proven excess in fresh mass yield by 72.84% (P <0.05).

In the years of the experimental period, the application of the studied humate fertilizers did not significantly affect the yield of **dry mass** from a natural meadow of *Chrysopogon gryllus* type. In the first and third experimental years, the variants treated with Molybdenum humate (160 ml/da) had the highest values. The excess compared to the controls was by 22.2 and 30.6%, respectively. In the second experimental year, the foliar application of Phosphorus humate (300 ml/da) had the highest impact on the dry mass amount.

The treatment with humate fertilizers increased the percentage and species share of useful grasses and legumes in the natural grass stand from the first to the third experimental year, and the amount of motley grasses decreased by from 25.0 to 34.4% (Figure 2, 3 and 4).

In the first year of the experiment, grasses (*Agrostis capillaris*, *Bothriochloa ischaemum, Chrysopogon gryllus, Festuca ovina*), legumes (*Trifolium campestre, Lotus corniculatus*) and motley grasses took 12.4%, 3.0% and 84.6% of the total natural grass stand, respectively.

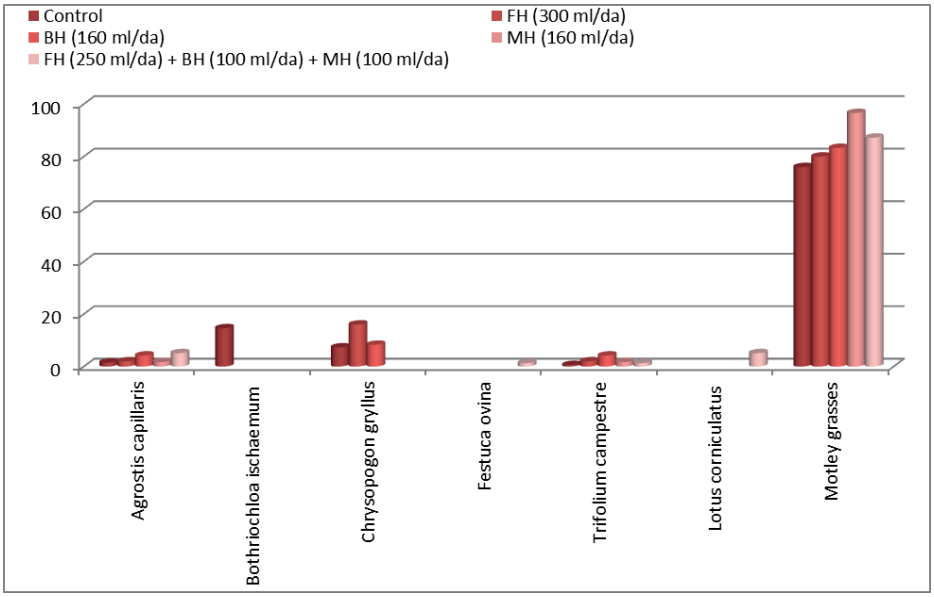

Figure 2. Botanical composition of a natural meadow of *Chrysopogon gryllus* type, treated by humate fertilizers in the first experimental year (2013)

Grasses in the treated variants occupied a smaller share in the composition of the grass mass compared to those in the control.

In contrast, legumes in fertilizer variants occupied from 1.7% (Molybdenum humate 160 ml/da) to 6.4% (Phosphorus humate - 250 ml/da + Boron humate - 100 ml/da + Molybdenum humate -100 ml/da) of harvested from grass stand, at 0.6% - for untreated plots.

In the second experimental year, the percentage of grasses (*Agrostis capillaris, Bothriochloa ischaemum, Chrysopogon gryllus, Festuca ovina, Anthoxanthum ododratum, Holcus lanatus, Cynosurus cristatus*) and legumes (*Trifolium campestre, Lotus corniculatus*) was higher with 11.4 and 23.0%, and the share of motley grasses decreased by 34.4%.

Grasses prevailed by 44.3% compared to the control only in the variants treated with Boron humate (160 ml/da).

The foliar application of Phosphorus humate (300 ml/da) and the combination of humate

fertilizers (Phosphorus humate - 250 ml/da + Boron humate - 100 ml/da + Molybdenum humate - 100 ml/da) positively affected the share of legumes and increased their share in grass stand respectively from 7.0 to 75.9%.

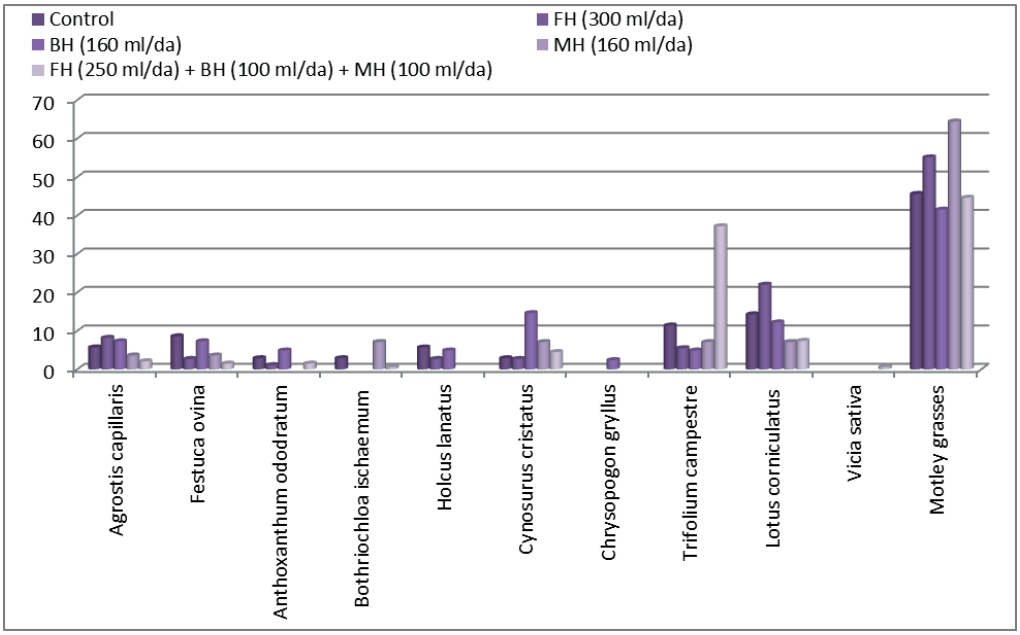

Figure 3. Botanical composition of a natural meadow of *Chrysopogon gryllus* type, treated by humate fertilizers in the second experimental year (2014)

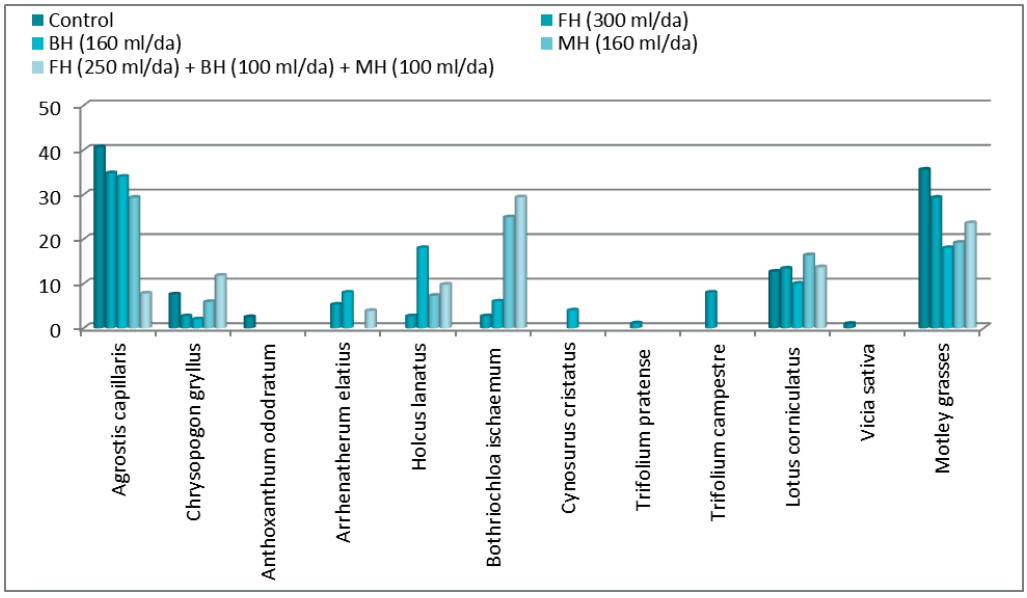

Figure 4. Botanical composition of a natural meadow of *Chrysopogon gryllus* type, treated by humate fertilizers in the third experimental year (2015)

In the third experimental year, positive changes were observed in the grass community. As a result of the imported fertilizers, a species diversity of grasses was found in all test variants. The characteristic species defining the grass stand *Chrysopogon gryllus* was displaced by *Bothriochloa ischaemum* and *Agrostis capillaris,* a trend, which continued from the previous year. The highest percentage share of *Agrostis capillaris* (40.6%) was registered in the non-treated control and the lowest (7.8%) in the variant with combined action of humate fertilizers. When applying Phosphorus humate (300 ml/da) the percentage share of *Agrostis capillaris* - 34.8% of the total species composition of the grass stand. The high presence of *Agrostis capillaris* in natural grasses determines a high pastoral value on grass stand (Andreoiu et al., 2020). *Bothriochloa ischaemum* prevailed most in the variants with Molybdenum humate (24.9%) and the combination of Phosphorus humate - 250 ml/da + Boron humate - 100 ml/da + Molybdenum humate - 100 ml/da (29.4%).

In the variants with application of Boron humate (160 ml/da), Phosphorus humate (300 ml/da), and the variants with a combination of humate fertilizers, the share of grass species, such as *Arrhenatherum elatius* was 8.0%, 5.3% and 3.9% respectively of the total grass stand composition.

The effect of foliar fertilization in all tested variants directly affected the legume component in the grass stand. Typical representatives are *Lotus corniculatus, Trifolium campestre, Trifolium pratens*e and *Vicia sativa*. Their share is particularly pronounced in the variants fertilized with Phosphorus humate (300 ml/da), where their share reached 13.4%. This is the variant with the most balanced floristic composition, conditionally divided into groups (grasses:legumes:motley grasses) in the ratio 42.4:22.5:29.3%, which implies the formation of an above-ground mass with good nutritional value. The percentage of grasses and legumes in natural grass stands affects the quality of the aboveground mass (Gür & Şen, 2016; Francisquini Junior et al., 2020), and the manner and intensity of use, as well as the selective behavior of ruminants, lead to an increase in the density of reproductive and invasive species (respectively reduces the volume of useful grass species preferred by farm animals) in the composition of meadow grasses (Naydenova & Mitev, 2011; Bayraktar, 2012; Tuna et al., 2013). The presences of highly productive species that have stabilizing properties dominate fertilized assemblages and enhance ecosystem stability (Yang et al., 2011). The applied fertilization improved and enriched the botanical composition of the natural grass stand, creating fodder that meets the needs of ruminants. In the third experimental year, the dominant species of legumes was *Lotus corniculatus*. Foliar fertilization with Molybdenum humate (160 ml/da) increased its percentage in the grass stand to 16.4%. In the group of clover, apart from *Trifolium campestre*, no other representatives characterizing this species have been identified (exception is var. 2, where the red clover participated with 1.1% of the total species composition).

CONCLUSIONS

Foliar feeding of natural grass stand of Chrysopogon gryllus, type with organic biofertilizers based on humic acids affects the productivity of fresh and dry mass. Treatment with Molybdenum humate (160 ml/da) resulted in a significant ($P < 0.01$) excess of fresh mass yield (570.0 kg/da) compared to the nontreated control (428.3 kg/da). The influence of other humate products is less pronounced, as the probable reason for this are the specific interactions between the specific climatic conditions during the year, the type of grass stand, the fertilization rate and the method of fertilizer application.

The effect of foliar treatment with biofertilizers had a significant effect on the floristic composition of the grass stand. The percentage and species share of the main biological groups (grasses, legumes, motley grasses) in the natural grass stand changed in a positive way from the first to the third experimental year. There is a reduced share of the main species *Chrysopogon gryllus* (characterizing the grass community) and motley grasses in the formed aboveground mass. The share of *Agrostis capillaris* and legumes (*Trifolium campestre* and *Lotus corniculatus*) significantly increased, which suggests better quality indicators of grass biomass.

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