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Productivity and quality of biomass of self-sown and reseeded legume species in burnt grass stands

Galina Naydenova* **and Tatyana Bozhanska**

Agricultural Academy, Research Institute of Mountain Stockbreeding and Agriculture, 5600 Troyan, Bulgaria
**Corresponding author:* gmvg@abv.bg

Abstract

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Fire is a significant ecological factor, negatively affecting the mechanical composition, organic matter and water regime of the soil, which in turn determine the productivity, quality and ecological function of grass stands. Because of their symbiotic nitrogen fixation, root structure and macromineral content, legume grasses perform a significant ecological function in the restoration of burnt areas.

The present study aimed to determine the productivity and qualitative parameters of the biomass of reseeded and self-sown legume grasses in burnt grass stands in relation to their potential role in the restoration of meadow and forest ecosystems. In the autumn of 2020, direct reseeding of a burnt semi-natural grass stand was conducted, in a hilly area in the Central Northern Bulgaria, with the following types and genotypes of legume grasses: *Vicia sativa* (Vitan cultivar); *Melilotus albus* (Adela cultivar) and *Trifolium pratense* (Nika 11 cultivar). In the first vegetation after the fire (2021), the types of reseeded and self-regenerating legume grasses, their productivity (fresh and dry biomass) and the main chemical composition were determined. According to the results, the highest amount of biomass with a high content of crude protein, crude fiber and the macronutrients N, P and Ca was formed by the reseeded species, such as *Melilotus albus*, as well as by the self-regenerating rhizome species *Coronilla varia*. According to the monitored qualitative indicators, populations of self-sown species, such as *Bituminaria bituminosa* and *Vicia grandiflora* have potential value as adapted genotypes suitable for regenerative reseeding of burnt grass stands. There is a high self-recovery capacity of the grass stands located in the hilly regions of the Northern Bulgaria, assessed by the productivity and biomass quality of the local legume grasses in the post-fire succession.

Keywords: yield; fires; chemical composition of grass vegetation

Introduction

According to national statistics, the average area of burnt forest territories for the last decade has been 100 000 decares, of which 10% are meadows and rangelands. There are no statistics on burnt semi-natural forage grass stands when the fire does not affect forest areas, which limits estimates of the economic and ecological damages. According to Drach et al. (2020), in Ukraine, burnt grass areas accounted for half (48.3%) of the burnt areas in 2016.

The effect of fires on the dynamics of ecological factors of forest and rangelands ecosystems is well known. First of all, burning adversely affects soil texture and moisture-holding capacity, reduces the content of nitrogen (N) and organic matter by retarding soil microbial processes and causes soil acidification (Ojima et al., 1994; Bogdanov, 2012; 2013a; Yang et al., 2020). In this way, fires significantly limit the productivity of meadow ecosystems, change the structure of grass communities and reduce biodiversity (Ilyina et al., 2021). The same authors indicate periods of self-recovery of burnt grass stands from 6 to 12 years, and re-burning during this period leads to their rapid degradation.

Fires in non-forested, grassy areas in Europe contribute nearly as much as forest fires to pyrogenic carbon emissions, which in turn influence climate (Ostroukhov et al., 2022). High-frequency fires reduce soil carbon stock, thereby limiting the most significant ecological function of grass stands today, namely carbon sequestration (Neary & Leonard, 2020; Abdalla et al., 2022).

It is believed that after forest fires, soil N losses can potentially be compensated by symbiotic nitrogen fixation by native and introduced legume grasses (Kaeser & Kirkman, 2012). Legumes are indicated as the first colonizers of burnt forest and meadow areas in a number of studies (Carbone, 2017; Carbone & Aguilar, 2021; Naydenova et al., 2022). Burning causes legume establishment through heat-induced scarification of the hard seed coats, which stimulates germination (Cushwa & Martin, 1968; Cushwa, 1970). Studies indicate that in burnt areas, legumes gain a nutritional advantage by increasing soil acidity (pH) and thermal mineralization of phosphorus (P) (Sprent, 1999). At the same time, fires reduce the competitive ability of species, which are not adaptable to burning, thus providing greater availability of light, nutrients and water for legumes that emerge from seed or from buds from deeper root necks (Wills & Read, 2007).

Fire is a significant ecological factor that influences the changes in the composition of the formed biomass in the areas after a fire, as the changes in the mechanical composition and organic matter, as well as the water regime of the soil, determine the amount (productivity) of the grass stand (Papanastasis, 2004; Bogdanov, 2013b).

The direct reseeding of degraded grass stands, including as a result of fire, is a rational method for preventing the erosion of improving and restoring pastures and meadows (Graber, 2020). Legume species diversity in an ecosystem can also be maintained through the temporary propagation of perennial seed banks (Kaeser & Kirkman, 2012). There is a lack of research on how reseeding affects the natural recovery and re-establishment of local legume species in burnt grass stands.

Due to the root structure and the process of symbiotic nitrogen fixation, legume grasses make a significant contribution to improving edaphic conditions, limiting erosion processes and restoring biodiversity after fire (Morgan, 1999). The productivity and basic chemical composition of legume biomass in post-fire grass communities is interesting for research. They are indicative of the levels of nitrogen fixation, the potential rate of decomposition and mineralization of this biomass, as well as the amount of macromineral elements that are introduced in the surface soil layer. The obtained data are significant for evaluating the opportunities and ways to restore burnt meadow ecosystems by direct reseeding with legume grasses.

The aim of the present study was to determine the accumulation of crude protein, crude fiber and the macronutrients N, P and Ca in the biomass of reseeded and self-sown legume grasses in a burned grassland in relation to their potential role in the restoration of meadow and forest ecosystems.

Materials and Methods

An experiment was conducted with direct reseeding of a semi-natural pasture grass stand that was burnt in the autumn of 2020, in a hilly area in Central Northern Bulgaria with coordinates 43º15´ and 25º12´. The experiment was started on March 15, 2021, using the method of long plots with an experimental plot area of 30 m^2. Reseeding was done with the following types and genotypes of legumes:

- *Vicia sativa* (Vitan variety);
- *Melilotus albus* (Adela variety);
- *Trifolium pratense* (Nika 11 variety).

For all variants, a sowing rate of 1000 Nb/m^2 germinated seeds was used.

Transects were determined on which phytocenotic observations were conducted during the first vegetation after the fire (2021) and the types of reseeded and self-regenerating legume grasses were determined. Using a ¼ meter, samples were taken to obtain fresh (FMY, kg 0.25 m^{-2}) and dry vegetative yield (DMY, kg 0.25 m^{-2}) of each established species, in the bud-formation phenophase (in triplicate).

Plant samples for chemical analysis were dried in a laboratory dryer at a temperature of 60°C and ground in a laboratory mill to a particle size of 1.0 mm. The chemical composition of dry fodder was analyzed according to Weende analysis: Crude protein (CP, % = 6.25 x N) according to Kieldahl (according to BDS/ISO-5983); Crude fiber (CFr, %); Crude fat (CF, %) (according to BDS/ISO-6492) – by extraction into a Soxhlet extractor; Ash (%) – (according to BDS/ISO-5984) degradation of the organic matter by gradual burning of the sample in a muffle furnace at 550°C; Dry matter (DM, %) – empirically calculated from % moisture; Nitrogen-free extractable substances (NFE, %) = 100 – (CP, % + CFr, % + CF, % + Ash, % + Moisture, %); Calcium (Ca, %) – Stotz (complexometric) and Phosphorus (P, %) – with vanadate-molybdate reagent – spectophotometer (*Agilent 8453 UV – visible Spectroscopy System*) that measure in the sphere of 425 ηm.

Statistical data processing included analysis of variance (ANOVA) and the Analysis Toolpak for Microsoft Excel 2010 software product.

Results and Discussion

Establishment of post-fire legume species and fresh and dry matter productivity

Soil properties were determined by soil organic matter created by vegetation. In this regard, the study of grass species in post-fire successions is essential for clarifying the recovery process of soils and ecosystems after fire. In the first vegetation after the fire, besides the sown species (*Vicia sativa, Melilotus albus* and *Trifolium pratense*), four other types of legume grasses were also established in the grass stand. Three of them are perennials (*Bituminaria bituminosa*, *Coronilla varia* and *Medicago falcata*). They were observed as typical participants in the post-fire successions in the hilly conditions of Central Northern Bulgaria and in another previous study of the present authors (Naydenova et al., 2022).

The presence of anatomical and morphological adaptations is important for the recovery of grass species after fire (Ilyina et al., 2021). In the present experiment, the self-recovery of *Coronilla varia* and *Medicago falcata* in the burnt terrain could be related to the formation of underground secondary stems, from the nodes of which new stems and roots are formed, characteristic of rhizomatous species. The emergence and longevity of perennial legumes in the post-fire area are closely related to the amount of nutrients (including carbohydrates) accumulated in the plant root system (Hainds et al., 1999). *Bituminaria bituminosa* is a tufted grass with a root neck under the soil surface, so it is also not affected by the fire. The only self-sown annual species is *Vicia grandiflora* (Figure 2). This species is superior in productivity of fresh and dry matter and in percentage share in the grass stand, the reseeded vetch of *Vicia sativa* (Figure 1). *Vicia grandiflora* is an autogamous, early maturing large-seeded member of the genus *Vicia*. Early ripening, respectively better penetration/ distribution of seeds in the soil, as well as its large seed size ensure preservation of germination after exposure to high temperatures and a relatively stable residual seed bank after burning. On the other hand, the likely reason for the lower share and, accordingly, productivity of the surface-reseeded cultivated species of vetch – *Vicia sativa* (Figure 4) is due to poor germination be-

Fig. 1. ***Vicia sativa***

Fig. 2. ***Vicia grandiflora***

Fig. 3. ***Melilotus albus***

Fig. 4. ***Trifolium pratense***

cause of disturbed hydrological properties of the soil after the fire. The significantly better establishment of the same vetch genotype can be pointed out as evidence when conducting an experiment with the direct reseeding of degraded pasture grass stand (Naydenova & Vasileva, 2016).

The other two reseeded species, the annual *Melilotus albus* and the perennial *Trifolium pratense*, emerged in good density after reseeding in the burnt grass stand (Figures 3 and 4).

The differences in the productivity of biomass in the first vegetation after the fire of the reseeded and self-sown legume grasses are significant (Table 1). Of the reseeded species, *Melilotus albus* registered a significantly higher yield of fresh matter (from 135.9% to 148.6%) and dry matter (with from 111.1% to 137.5%) compared to that of *Vicia sativa* and *Trifolium pratense*. The significantly lower yield of *Trifolium pratense* is because of the fact that the readings were conducted in the year of reseeding, and in the first vegetation the genotype used formed only leaf rosettes with single generative stems.

Table 1. Productivity of legume species established in the monitored burnt location in the first vegetation after the fire

Legume species	FMY, kg 0.25 m^{-2}	DMY, kg 0.25 m^{-2}
Reseeded		
Vicia sativa	0.195[c]	0.045[c]
Melilotus albus	0.460[ab]	0.095[ab]
Trifolium pratense	0.185[c]	0.040[c]
Self-sown and self-generated		
Bituminaria bituminosa	0.500[ab]	0.123[a]
Coronilla varia	0.550[a]	0.120[a]
Medicago falcata	0.255[c]	0.060[bc]
Vicia grandiflora	0.405[b]	0.085[ab]
$LSD_{0.05}$	0.128	0.04

The highest and almost identical values for fresh and dry biomass productivity were found in the self-regenerating legume grasses, the perennial species *Coronilla varia* (FMY – 0.550 kg 0.25 m^{-2} and DMY – 0.120 kg 0.25 m^{-2}) and *Bituminaria bituminosa* (FMY – 0.500 kg 0.25 m^{-2} and DMY – 0.123 kg 0.25 m^{-2}), followed by the only self-sown annual species *Vicia grandiflora* (FMY – 0.405 kg 0.25 m^{-2} and DMY – 0.085 kg 0.25 m^{-2}).

The productivity of *Medicago falcata* is significantly lower compared to other self-sown and self-generated species in the burnt grass stand, but the share of this species in the grass stand is indicated as a significant element to enhance the long-term sustainability of the ecosystem (Mortenson et al., 2004).

Chemical composition of legume grass species after fire

Significant differences were found in the values of the main chemical composition of the dry matter of the studied reseeded, self-sown and self-generated legume species (Table 2). Among the reseeded species, the data indicated that the dry matter of *Melilotus albus* (the highest yielding species) had the highest crude protein, mineral substances, crude fat, calcium, phosphorus and nitrogen content. The values of the indicators exceeded the averages by 6.2% (CP), 1.9% (Ash), 1.3% (CF), 0.52% (Ca), 0.02% (P) and 1.0% (N). The biomass of *Melilotus albus* also has the lowest content of crude fiber (22.03%) and non-nitrogen extractive substances (27.63%) in dry matter.

Similar results were obtained for self-sown and self-generated species in the burnt grass stand. The biomass of the perennial rhizome species *Coronilla varia* (the species with the highest fresh mass productivity) also had the highest crude protein and nitrogen content. The excess in the values

Table 2. Basic chemical composition (%) of legume species established in the monitored burnt location in the first vegetation after the fire

Legume species	DM	CP	CFr	Ach	CF	NFE	Ca	P	N
Reseeded									
Vicia sativa	22.90	23.99	25.81	7.37	0.84	28.39	2.12	0.43	3.84
*Melilotus albu*s	20.70	30.35	22.03	10.11	3.07	27.63	2.92	0.45	4.86
Trifolium pratense	22.20	18.04	28.68	7.26	1.43	34.79	2.15	0.43	2.89
Mean	21.93	24.13	25.51	8.25	1.78	30.27	2.40	0.43	3.86
Self-sown and self-generated									
Bituminaria bituminosa	24.40	23.95	37.62	10.48	4.96	15.73	3.24	0.36	3.83
Coronilla varia	21.80	35.20	20.16	8.92	2.92	25.03	2.08	0.48	5.63
Medicago falcata	23.10	24.83	29.53	9.08	3.97	32.01	3.45	0.51	3.97
Vicia grandiflora	21.20	28.90	16.15	10.65	1.64	34.88	2.30	0.51	4.62
Mean	22.63	28.22	25.87	9.78	3.37	26.91	2.77	0.47	4.52

compared to the average is by 6.3% (for CP) and 1.1% (for N), respectively. The mineralization of organic matter from high-protein legume species leads to the release of nitrogen in the form of ammonium salts easily absorbed by plants. *Coronilla varia* plants have moderate growth and development, a high content of leaves (63-68%) and a low content of dry matter (Ţîţei et al., 2016). The same research indicates that the self-sowing and self-regeneration of the species in degraded, polluted and eroded soils leads to an increase in the ecological stability of natural grass stands.

The high percentage of nitrogen in the dry matter content was found to accelerate the rate of mineralization in legume grasses in the post-fire succession. Compared to grass species, they also have a lower lignin content and a higher polyphenol content, which stimulates the nitrogen mineralization process (Brandsaeter et al., 2008). The morphological characteristics of individual legume species are also significant in determining the degree of nitrogen absorption from the air and its gradual release into the soil, during which the development of other herbaceous species with high fodder and ecological value can be observed. According to Hendricks et al. (2019), legume leaf tissues developed in the burnt zone had high N concentrations, low lignin concentrations, and a correspondingly low lignin: N ratio (mean 6.7 ± 0.9), indicative of tissues with rapid weight loss and N in mineralization processes. The process of N mineralization is positively correlated with total N concentration (Watthier et al., 2020). The best chemical index of N mineralization was found to be the lignin + polyphenols:N ratio, which accounted for 84% of the variation in the percent of mineralized nitrogen by legumes (Singh & Kumar, 1996).When evaluating the studied species in this aspect, it should be pointed out that among the reseeded species, the white melilot is potentially the fastest mineralization of the biomass, and from the self-sown and self-generated species – *Coronilla varia* and *Vicia grandiflora*. The highest fibrous (37.62%) and the lowest content of crude protein (23.95%), nitrogen (3.83%) and nitrogen-free extractable substances (15.73%) were registered in the dry matter of *Bituminaria bituminosa*, which suggests a delay in the mineralization processes of the biomass of this species that is typical of post-fire successions.

In a study of the cycling of elements, such as N, P, K, Na, Ca and Mg, in four grass species (including *Chrysopogon gryllus*), Pandey et al. (1993) found that their content in the roots (except for Ca) was higher than in the stems. The total annual extraction of N, P, K, Na, Ca and Mg was 137.3, respectively; 10.4; 51.1; 5.5; 8.7 and 18.2 kg/ha. In the annual cycle, 98% N, 77% P, 49% K, 109% Na, 87% Ca and 65% Mg are returned to the soil through detritus. In the present study, the self-sown species *Vicia grandiflora* had the highest concentration of phosphorus (0.51%), minerals (10.65%) and nitrogen-free extractable substances (34.88%) in dry matter. According to Mikić et al. (2013), the species has great potential for cultivation as a high-protein fodder grass, and the biomass quality we found in terms of mineral content makes it suitable for regenerative reseeding of burnt grass stands.

The chemical composition of the biomass of individual grass species affects the flammability and fire resistance. The self-generated species, such as *Bituminaria bituminosa* and *Vicia grandiflora* have a higher ash content, which according to Drach et al. (2020) is associated with a higher resistance to fire and can be considered as a significant ecological adaptation of local plant resources. According to the authors, the maximum exothermic effect for each plant was characterized by different temperature values, as well as ash residue, which impedes the combustion process. Such results were also observed for the reseeded species *Melilotus albus*. In this regard, reseeding with mixtures of local genotypes of the specified legume grasses should be investigated for the recovery of burnt grass stands.

Conclusions

The highest amount of biomass with a high content of crude protein, crude fiber and the macronutrients N, P and Ca was formed in the first vegetation after fire by the reseeded species *Melilotus albus*, as well as by the self-generated rhizome species *Coronilla varia*. According to the monitored qualitative indicators, populations of the self-sown species *Bituminaria bituminos*a and *Vicia grandiflora* have potential value as adapted genotypes suitable for regenerative reseeding of burnt grass stands. The self-recovery capacity of the grass stands located in the hilly regions of Northern Bulgaria, assessed by the productivity and biomass quality of the local legume grasses in the post-fire succession, is high.

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