EVALUATION OF DROUGHT RESISTANCE OF SOME GRASSES AND LEGUMES SPECIES GROWN ALONE OR IN MIXTURES, UNDER CLIMATE CHANGE CONDITIONS

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Abstract

Currently, drought is the factor with the most negative influence on spontaneous flora and fauna. By 2100, air temperatures are expected to rise by 1.8°C to 4.0°C. Continuous research is needed on the drought resistance of perennial grasses and leguminous fodder crops. Legumes can be sown in pure culture, but grasses are most often sown in Romania in mixture with legumes. It is therefore necessary to study the drought resistance of pure crops, but especially of mixtures of perennial grasses and legumes. The experiment was conducted at the Ezăreni Farm and studied 10 simple and complex mixtures of perennial grasses and legumes, with 4 levels of fertilization. Romanian varieties with good adaptability to the cultivation conditions in our country were used to set up the experiment.

Keywords: forage mixtures, fertilization, grassland management, landscape, low input.

INTRODUCTION

The effects of climate change are increasingly being felt through long periods of high temperatures above 30°C associated with a lack of rainfall.

Among the fodder plants most affected by these phenomena of drought and aridity are legumes, which suffer during long periods without rainfall. Their increased sensitivity to drought requires continuous research into increasing drought tolerance (Ilić et al., 2025).

Prolonged drought has negative effects on shoot length, biomass yield, and photosynthesis (a process that is halted at high temperatures to prevent water loss), but the symbiotic capacity of Rhizobium sp. bacteria is also diminished, so inoculation with Synthetic microbial communities (SynComs) has been studied to reduce the negative effects of drought on leguminous plants (Shipar et al., 2025).

It is necessary to understand the mechanisms by which alfalfa responds to stress, as this is an essential characteristic for the creation of new varieties with increased drought resistance. It has therefore been concluded that the experiments carried out in the open field are currently insufficient (Daud et al., 2025). Perennial grasses reacted better to drought conditions than perennial legumes. Grasses managed to create a larger number of roots with a diameter between 0 and 1 mm (Wang et al., 2020). Plants respond differently to soil and climate conditions. In the case of *Bromus inermis* L, which was subjected to a prolonged period of drought, both root mass and stolon number were found to increase (Kroeger and Otfinowski., 2025). It was observed that in terms of plant height and number of shoots, drought-tolerant species were less affected compared to species with lower resistance. The decrease in height and number of shoots represents the adaptation of species to water stress conditions, thus reducing water losses through evapotranspiration (Petcu et al., 2019). Drought also has negative effects on protein content, leading to a decrease in the crude protein content of the feed obtained. At the same time, a decrease in NDF and ADF content was also observed (Kuchenmeister et al., 2013). Recent research shows the importance of using salicylic acid on the positive influences on the growth, productivity, and yield of plants subjected to water stress (Melenciuc., 2015).

Another method studied was the application of chemical or organic fertilizers. Following these experiments, an improvement in drought resistance was observed, as well as a reduction in weeds within the vegetation cover that could damage the vegetation cover through high water consumption compared to cultivated plants. Plants gain a greater ability to use nutrients from the soil. The number of beneficial microbes in grasslands also increases (Lewin et al., 2024).

MATERIAL AND METHOD

The experiment was conducted at the Ezăreni educational farm. It is a two-factor experiment. Factor A: a_1 – *Onobrychis viciifolia* Scop. (100%) (control); a_2 – *Onobrychis viciifolia* Scop. (75%) and *Bromus inermis* Leyss. (25%); a_3 – *Onobrychis viciifolia* Scop. (50%) and *Bromus inermis* Leyss. (50%); a_4 - *Onobrychis viciifolia* Scop. (25%) and *Bromus inermis* Leyss. (75%); a_5 – *Medicago sativa* L. (100%); a_6 – *Medicago sativa* L. (75%) and *Festuca pratensis* (25%); a_7 – *Medicago sativa* L. (50%) and *Festuca pratensis* (50%); a_8 – *Medicago sativa* L. (25%) and *Festuca pratensis* (75%); a_9 – *Medicago sativa* L. (20%), *Lotus corniculatus* L. (15%); *Festuca pratensis* (30%); *Lolium perenne* L. (10%) and *Dactylis glomerata* L. (25%). a_{10} – *Onobrychis viciifolia* Scop. (20%), *Lotus corniculatus* L. (15%); *Agropyron pectiniforme* L. (30%); *Bromus inermis* Leyss. (25%) and *Lolium perenne* L. (10%).

Factor B represents the degree of fertilization applied with the following 4 gradations: b_1 – unfertilized (control);

b_2 – $N_{50}P_{50}K_{50}$; b_3 – $N_{75}P_{75}K_{75}$; b_4 – $N_{100}P_{100}K_{100}$.

The harvestable area is 8m^2, measuring 1x9m. The plants used in the experiment are of Romanian origin, with seeds purchased from Research and Development Station for Meadows Vaslui,, N.A.R.D.I Fundulea, and the Research and Development Institute for Grassland Brașov. The geographical coordinates of the Ezăreni farm, which belongs to the "Ion Ionescu de la Brad" University of Life Sciences in Iași, are: 47°5' - 47°10' north latitude and 27°28' - 27°33' east longitude. To assess the drought resistance and regeneration capacity of plants in the field, the Methodology for examining agronomic and utilization value (VAU Test) published by The State Institute for Testing and Registration of Varieties in 2008 was used. Drought resistance was assessed in an unirrigated system after a period of more than 3 weeks of drought. Grades from 1 to 9 were used. Grade 1 represented high drought resistance/tolerance. A score of 9 was assigned to variants with very poor drought resistance/tolerance.

The scale used was as follows: Note 1 - very good resistance - variants in which plants have very good development after mowing (100%); Note 3 - good resistance, plants with good development after mowing (75%); Note 5 - medium resistance, plants with medium development after mowing (50%); Note 7 - poor resistance, plants with poor development after mowing (25%); Note 9 – very poor resistance, plants that do not regenerate after mowing.

The assessment of regeneration capacity is expressed in grades from 1 to 9, as follows: Note 1 ->135% – very good regeneration capacity, Note 2 (126-135%) – good to very good regeneration capacity; Note 3 (116-125%) – good regeneration capacity; Note 4 (106-115%) – good to medium regeneration capacity; Note 5 (96-105%) – medium regeneration capacity; Note 6 (86-95%) - average to poor regeneration capacity; Note 7 (76-85%) - poor regeneration capacity; Note 8 (65-75%) - poor to very poor regeneration capacity; Note 9 (< 65%) - very poor regeneration capacity. To determine these two indices, measurements were taken in the field on 10 plants from the control variant and 10 plants from each of the other variants.

RESULTS AND DISCUSSIONS

Drought resistance (on a scale of 1 to 9) was assessed after the first mowing, during a period of prolonged drought. In this context, drought resistance scores were assigned simultaneously with the assessment of plant regeneration capacity. Species grown in monoculture or in mixtures that, after the period of intense drought, showed normal growth 2–3 weeks after mowing were considered to have good resistance and received scores close to 1, while species with reduced regeneration were scored closer to 9. Drought resistance was assessed for *Onobrychis viciifolia* and *Medicago sativa*, grown either alone or in simple and complex mixtures, under different conditions of fertilization with complex mineral fertilizers based on nitrogen, phosphorus, and potassium (Table 1).

The interaction between the species or mixture of perennial grasses and legumes and mineral fertilization in the second year of vegetation showed that the scores obtained varied between 3.3 and 6, depending on the species (or mixture) and fertilization. In the control variant (a_1b_1 – *Onobrychis viciifolia* 100%, unfertilized), the species grown alone showed good drought resistance, with approximately 75% of the plants developing properly after mowing, with scores between 3.3 and 4, depending on fertilization (Table 1).

Regarding the drought resistance of the species, the best emergence and development after drought was recorded in variant a_5 – *Medicago sativa* (100%), regardless of fertilization. Thus, the species grown alone on mineral soil or unfertilized received a constant score of 3.3 in all variants studied (Table 1). For this parameter, the differences from the control variant were insignificant, significant, and very significant, depending on the species and mixture (Table 1).

In parallel with the assessment of drought resistance, in the second year of vegetation, the regeneration capacity of plants after the first mowing was also determined, rated on a scale from 1 to 9 (1 – very good, 9 – very poor), 10–15 days after mowing I.

For the species *Onobrychis viciifolia* and *Medicago sativa*, grown either in monoculture or in simple and complex mixtures, under different conditions of fertilization with complex mineral fertilizers based on nitrogen, phosphorus, and potassium, the regeneration capacity of plants after the first mowing was evaluated (Table 2).

Analysis of the interaction between species or mixture and mineral fertilization showed that the scores obtained ranged from 3.3 to 6, depending on the species, type of mixture, and fertilization variant.

In the control variant (a_1b_1 – *Onobrychis viciifolia* 100%, unfertilized), the species *Onobrychis viciifolia* grown alone showed the best regeneration capacity (medium to good), with scores between 3.3 and 4.7, depending on the fertilization treatment (Table 2).

Regarding the regeneration capacity of plants, both in monoculture and in mixtures of two or more species in different proportions, good regeneration was recorded in variant a5 – *Medicago sativa* (100%), regardless of fertilization.

Table 1

The influence of the interaction between the species or mixture of perennial grasses and legumes and mineral fertilization on drought resistance in the second year of vegetation (scores from 1 to 9)

Variant*		**Drought resistance**	**Difference**		**Significance**
		Note	**Note**	**%**	
		Control: a_1b_1 - *O.v.* (100%), unfertilized			
a_1 - *O.v.* (100%) (mt.)	b_1 - unfertilized (mt.)	3,3	Control	100	Control
a_1 - *O.v.* (100%) (mt.)	b_2 - $N_{50}P_{50}K_{50}$	4,0	0,7	120,0	
a_1 - *O.v.* (100%) (mt.)	b_3 - $N_{75}P_{75}K_{75}$	4,0	0,7	120,0	
a_1 - *O.v.* (100%) (mt.)	b_4 - $N_{100}P_{100}K_{100}$	4,0	0,7	120,0	
a_2 - *O.v.* (75%) + *B.i.* (25%)	b_1 - unfertilized	5,7	2,3	170,0	***
a_2 - *O.v.* (75%) + *B.i.* (25%)	b_2 - $N_{50}P_{50}K_{50}$	5,7	2,3	170,0	***
a_2 - *O.v.* (75%) + *B.i.* (25%)	b_3 - $N_{75}P_{75}K_{75}$	5,7	2,3	170,0	***
a_2 - *O.v.* (75%) + *B.i.* (25%)	b_4 - $N_{100}P_{100}K_{100}$	5,7	2,3	170,0	***
a_3 - *O.v.* (50%) + *B.i.* (50%)	b_1 - unfertilized	6,0	2,7	180,0	***
a_3 - *O.v.* (50%) + *B.i.* (50%)	b_2 - $N_{50}P_{50}K_{50}$	6,0	2,7	180,0	***
a_3 - *O.v.* (50%) + *B.i.* (50%)	b_3 - $N_{75}P_{75}K_{75}$	6,0	2,7	180,0	***
a_3 - *O.v.* (50%) + *B.i.* (50%)	b_4 - $N_{100}P_{100}K_{100}$	6,0	2,7	180,0	***
a_4 - *O.v.* (25%) + *B.i.* (75%)	b_1 - unfertilized	4,0	0,7	120,0	
a_4 - *O.v.* (25%) + *B.i.* (75%)	b_2 - $N_{50}P_{50}K_{50}$	4,0	0,7	120,0	
a_4 - *O.v.* (25%) + *B.i.* (75%)	b_3 - $N_{75}P_{75}K_{75}$	4,0	0,7	120,0	
a_4 - *O.v.* (25%) + *B.i.* (75%)	b_4 - $N_{100}P_{100}K_{100}$	4,0	0,7	120,0	
a_5 - *M.s.* (100%)	b_1 - unfertilized	3,3	0,0	100,0	
a_5 - *M.s.* (100%)	b_2 - $N_{50}P_{50}K_{50}$	3,3	0,0	100,0	
a_5 - *M.s.* (100%)	b_3 - $N_{75}P_{75}K_{75}$	3,3	0,0	100,0	
a_5 - *M.s.* (100%)	b_4 - $N_{100}P_{100}K_{100}$	3,3	0,0	100,0	
a_6 - *M.s.* (75%) + *F.p.* (25%)	b_1 - unfertilized	5,0	1,7	150,0	**
a_6 - *M.s.* (75%) + *F.p.* (25%)	b_2 - $N_{50}P_{50}K_{50}$	5,0	1,7	150,0	**
a_6 - *M.s.* (75%) + *F.p.* (25%)	b_3 - $N_{75}P_{75}K_{75}$	5,0	1,7	150,0	**
a_6 - *M.s.* (75%) + *F.p.* (25%)	b_4 - $N_{100}P_{100}K_{100}$	5,0	1,7	150,0	**
a_7 - *M.s.* (50%) + *F.p.* (50%)	b_1 - unfertilized	5,3	2,0	160,0	**
a_7 - *M.s.* (50%) + *F.p.* (50%)	b_2 - $N_{50}P_{50}K_{50}$	5,3	2,0	160,0	**
a_7 - *M.s.* (50%) + *F.p.* (50%)	b_3 - $N_{75}P_{75}K_{75}$	5,3	2,0	160,0	**
a_7 - *M.s.* (50%) + *F.p.* (50%)	b_4 - $N_{100}P_{100}K_{100}$	5,3	2,0	160,0	**
a_8 - *M.s.* (25%) + *F.p.* (75%)	b_1 - unfertilized	4,3	1,0	130,0	
a_8 - *M.s.* (25%) + *F.p.* (75%)	b_2 - $N_{50}P_{50}K_{50}$	4,3	1,0	130,0	
a_8 - *M.s.* (25%) + *F.p.* (75%)	b_3 - $N_{75}P_{75}K_{75}$	4,3	1,0	130,0	
a_8 - *M.s.* (25%) + *F.p.* (75%)	b_4 - $N_{100}P_{100}K_{100}$	4,3	1,0	130,0	
a_9 - *M.s.* (20%) + *L.c.* (15%) + *F.p.* (30%) + *L.p.* (10%) + *D.g.* (25%)	b_1 - unfertilized	5,0	1,7	150,0	**
a_9 - *M.s.* (20%) + *L.c.* (15%) + *F.p.* (30%) + *L.p.* (10%) + *D.g.* (25%)	b_2 - $N_{50}P_{50}K_{50}$	5,0	1,7	150,0	**
a_9 - *M.s.* (20%) + *L.c.* (15%) + *F.p.* (30%) + *L.p.* (10%) + *D.g.* (25%)	b_3 - $N_{75}P_{75}K_{75}$	5,0	1,7	150,0	**
a_9 - *M.s.* (20%) + *L.c.* (15%) + *F.p.* (30%) + *L.p.* (10%) + *D.g.* (25%)	b_4 - $N_{100}P_{100}K_{100}$	5,0	1,7	150,0	**
a_{10} - *O.v.* (20%) + *L.c.* (15%) + *A.p.* (30%) + *B.i.* (25%) + *L.p.* (10%)	b_1 - unfertilized	5,7	2,3	170,0	***
a_{10} - *O.v.* (20%) + *L.c.* (15%) + *A.p.* (30%) + *B.i.* (25%) + *L.p.* (10%)	b_2 - $N_{50}P_{50}K_{50}$	5,7	2,3	170,0	***
a_{10} - *O.v.* (20%) + *L.c.* (15%) + *A.p.* (30%) + *B.i.* (25%) + *L.p.* (10%)	b_3 - $N_{75}P_{75}K_{75}$	5,7	2,3	170,0	***
a_{10} - *O.v.* (20%) + *L.c.* (15%) + *A.p.* (30%) + *B.i.* (25%) + *L.p.* (10%)	b_4 - $N_{100}P_{100}K_{100}$	5,7	2,3	170,0	***
		LSD 5%	1,3		
		LSD 1%	1,7		
		LSD 0,1%	2,2		

* *Onobychis viciifolia (O.V.); Bromus inermis (B.i.); Medicago sativa (M.s*.); *Festuca pratensis (F.p.); Lolium perenne (L.p.); Lotus corniculatus (L.c*.); *Dactylis golomerata (D.g.); Agropyron pectiniforme (A.p.)*

Thus, the species cultivated alone on mineral soil, but also in the unfertilized variant, showed good to medium regeneration, consistently rated at 4.0 in all variants analyzed (Table 2). In the case of the simple mixture of *Onobrychis viciifolia* (50%) + *Bromus inermis* (50%), the plants showed the lowest regeneration capacity (medium to poor), with scores between 5 and 6. For this parameter, the differences from the control variant were, in most cases, negatively significant, but in some variants there were positively significant and distinctly significant differences for all species and mixtures analyzed (Table 2).

Table 2

The influence of the interaction between the species or mixture of perennial grasses and legumes and mineral fertilization on the regeneration capacity of plants after the first cut, in the second year of vegetation (scores from 1 to 9)

Variant*		**Regenerative capacity**	**Difference**		**Significance**
		Note	**Note**	**Meaning**	
		Control: a_1b_1 - *O.v.* (100%), unfertilized			
a_1 - *O.v.* (100%) (mt.)	b_1 - unfertilized (mt.)	4,7	Control	100	Control
	b_2 - $N_{50}P_{50}K_{50}$	3,3	-1,3	71,4	o
	b_3 - $N_{75}P_{75}K_{75}$	3,3	-1,3	71,4	o
	b_4 - $N_{100}P_{100}K_{100}$	3,3	-1,3	71,4	o
a_2 - *O.v.* (75%) + *B.i.* (25%)	b_1 - unfertilized	5,3	0,7	114,3	
	b_2 - $N_{50}P_{50}K_{50}$	4,7	0,0	100,0	
	b_3 - $N_{75}P_{75}K_{75}$	4,7	0,0	100,0	
	b_4 - $N_{100}P_{100}K_{100}$	4,7	0,0	100,0	
a_3 - *O.v.* (50%) + *B.i.* (50%)	b_1 - unfertilized	6,0	1,3	128,6	*
	b_2 - $N_{50}P_{50}K_{50}$	5,0	0,3	107,1	
	b_3 - $N_{75}P_{75}K_{75}$	5,0	0,3	107,1	
	b_4 - $N_{100}P_{100}K_{100}$	5,0	0,3	107,1	
a_4 - *O.v.* (25%) + *B.i.* (75%)	b_1 - unfertilized	5,3	0,7	114,3	
	b_2 - $N_{50}P_{50}K_{50}$	3,7	-1,0	78,6	
	b_3 - $N_{75}P_{75}K_{75}$	3,7	-1,0	78,6	
	b_4 - $N_{100}P_{100}K_{100}$	3,7	-1,0	78,6	
a_5 - *M.s.* (100%)	b_1 - unfertilized	4,0	-0,7	85,7	
	b_2 - $N_{50}P_{50}K_{50}$	4,0	-0,7	85,7	
	b_3 - $N_{75}P_{75}K_{75}$	4,0	-0,7	85,7	
	b_4 - $N_{100}P_{100}K_{100}$	4,0	-0,7	85,7	
a_6 - *M.s.* (75%) + *F.p.* (25%)	b_1 - unfertilized	6,0	1,3	128,6	*
	b_2 - $N_{50}P_{50}K_{50}$	4,3	-0,3	92,9	
	b_3 - $N_{75}P_{75}K_{75}$	4,3	-0,3	92,9	
	b_4 - $N_{100}P_{100}K_{100}$	4,3	-0,3	92,9	
a_7 - *M.s.* (50%) + *F.p.* (50%)	b_1 - unfertilized	6,3	1,7	135,7	**
	b_2 - $N_{50}P_{50}K_{50}$	4,7	0,0	100,0	
	b_3 - $N_{75}P_{75}K_{75}$	5,0	0,3	107,1	
	b_4 - $N_{100}P_{100}K_{100}$	5,0	0,3	107,1	
a_8 - *M.s.* (25%) + *F.p.* (75%)	b_1 - unfertilized	5,0	0,3	107,1	
	b_2 - $N_{50}P_{50}K_{50}$	4,0	-0,7	85,7	

	b_3 - $N_{75}P_{75}K_{75}$	4,0	-0,7	85,7	
	b_4 - $N_{100}P_{100}K_{100}$	4,7	0,0	100,0	
a_9 - *M.s.* (20%) + *L.c.* (15%) + *F.p.* (30%) + *L.p.* (10%) + *D.g.* (25%)	b_1 - unfertilized	4,3	-0,3	92,9	
	b_2 - $N_{50}P_{50}K_{50}$	4,0	-0,7	85,7	
	b_3 - $N_{75}P_{75}K_{75}$	4,0	-0,7	85,7	
	b_4 - $N_{100}P_{100}K_{100}$	4,0	-0,7	85,7	
a_{10} - *O.v.* (20%) + *L.c.* (15%) + *A.p.* (30%) + *B.i.* (25%) + *L.p.* (10%)	b_1 - unfertilized	5,7	1,0	121,4	
	b_2 - $N_{50}P_{50}K_{50}$	4,7	0,0	100,0	
	b_3 - $N_{75}P_{75}K_{75}$	4,7	0,0	100,0	
	b_4 - $N_{100}P_{100}K_{100}$	4,7	0,0	100,0	
		DL 5%	1,2		
		DL 1%	1,5		
		DL 0,1%	2,0		

* *Onobychis viciifolia (O.V.); Bromus inermis (B.i.); Medicago sativa (M.s.); Festuca pratensis (F.p.); Lolium perenne (L.p.); Lotus corniculatus (L.c.); Dactylis golomerata (D.g.); Agropyron pectiniforme (A.p.)*

CONCLUSIONS

In terms of species resistance to drought, both in pure culture and in mixtures of two or more species in different proportions, the highest tolerance to water deficit was recorded in variant a_5– *Medicago sativa* (100%), regardless of the fertilization treatment applied. Thus, for the *Medicago sativa* species grown in monoculture, both on mineral soil and in the unfertilized variant, drought resistance was consistently rated 3.3 in all variants analyzed.

According to the results obtained, in the second year of vegetation, good drought resistance, accompanied by satisfactory plant regeneration after mowing (75%), was recorded in the single-cultured species *Onobrychis viciifolia* (100%) and *Medicago sativa* (100%). Along with the assessment of drought resistance, in the second year of vegetation, the capacity of plants to regenerate after the first mowing was also determined, rated on a scale from 1 to 9 (1 – very good, 9 – very poor), 10–15 days after mowing I.

The analysis of the influence of the interaction between the species or mixture of perennial grasses and legumes and mineral fertilization on this parameter in the second year of vegetation showed that the values obtained varied between 3.3 and 6, depending on the species, type of mixture, and fertilization. In the control variant (a1b1 – *Onobrychis viciifolia* 100%, unfertilized), the species *Onobrychis viciifolia* grown alone showed the best regeneration capacity (medium to good), with values between 3.3 and 4.7, depending on the fertilization variant.

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