

## TESTING A SELECTION OF ALFALFA VARIETIES FOR ECOLOGICAL PLASTICITY, PRODUCTIVITY AND A NUMBER OF QUALITATIVE PARAMETERS

Niculae DINCA<sup>1</sup>, Daniel DUNEA<sup>2</sup>, Ana-Maria STANCIU<sup>1</sup>, Nicolae PĂTRU<sup>3</sup>

<sup>1</sup>University of Agronomic Sciences and Veterinary Medicine of Bucharest, 59 Marasti Blvd,  
District 1, Bucharest, Romania

<sup>2</sup>Valahia University of Targoviste, Faculty of Environmental Engineering and Food Science,  
Aleea Sinaia 13, Targoviste, Romania

<sup>3</sup>S.C. Patru Agro SRL, 2A Ceahlau Street, District 6, Bucharest, Romania

Corresponding author email: dan.dunea@valahia.ro

### Abstract

The cultivation of alfalfa has an essential role in ensuring the required fodder, but the achievement of cropping performances e.g. proper yield, persistence, winter hardiness, competition capacity, ecological plasticity, depends on a multitude of technological, environmental, and genetic factors. The selection of suitable varieties for cultivation in pure stands or more perennial and productive mixtures with other perennial legumes and grasses is a key element for a successful crop. The objective of the study was to characterize the bio-productive and forage quality performances of 18 alfalfa varieties available for cultivation in Romania, originating from Romania and abroad, in the eco-climatic conditions specific to the Romanian Plain. The screening of these performances that substantiate the appreciation of the biological yield was based on detailed experiences in the field and laboratory. A set of representative indicators was selected for measurements, determinations, and observations, including growth rate, tillering, leaf-stem ratio, forage quality indicator, average crop yield, and canopy height. It was found that Romanian varieties presented improved ecological plasticity compared to the foreign varieties in the eco-climatic conditions of the experimental site.

**Key words:** *Medicago sativa L.*, dry matter, growth rate, leaf-area ratio, tillering, forage quality, yield.

### INTRODUCTION

Alfalfa is a very popular plant worldwide due to the possibility of using it for multiple purposes. The associated environmental impacts for producing the alfalfa for fodder differ depending on the type of production system and the region. It is important to take into account water and land resources together with assessing other impacts of agriculture products when performing a comparison between farming systems and improvement measures (Wang et al., 2021).

Compared to other fodder crops (Dunea et al., 2015), alfalfa is clearly superior through large fodder productions and fodder quality provided by its ability to resist for many years without being sown and optimized irrigation has an important role in achieving superior yields (Dincă et al., 2017). Cavero et al. (2017) found that the maximum alfalfa forage yield was lower in the first cropping year ( $17 \text{ t ha}^{-1}$ ) compared to the two following years (20-22 t

$\text{ha}^{-1}$ ). In the second and third year of cropping of the Aragón variety, the increase of forage yield occurred when the applied irrigation was 115% of crop irrigation requirement compared to the first year when the yield increased linearly with the applied irrigation in a semiarid Mediterranean climate (Zaragoza, Spain). Schitea (2010) reported for Romanian alfalfa cultivars an average production of more than  $17 \text{ t DM ha}^{-1}$  of the three cropping years in irrigated conditions and application of optimal fertilization. For Romania climate, alfalfa yields were found to be responsive to irrigation level, decreasing with reductions in irrigation amount.

Phosphorous fertilization also increases the forage yield in both young and old rainfed alfalfa stands in a semiarid environment (Fan et al., 2016).

Symbiotic rhizobia provide beneficial effects on leguminous plants (Dincă and Dunea, 2017) including alfalfa. A novel effect of rhizobia on forage quality of alfalfa was found i.e., the

nodulated alfalfa showed an increase in lignin content and a decrease in digestibility in comparison with non-nodulated plants due to a potential defensive response of plants to the rhizobial invasion (Zhang et al., 2016).

The forage production obtained from alfalfa cropping can be used in the form of green fodder, hay, silage, semi-hay, and dehydrated fodder (Lloveras et al., 2008).

Alfalfa also plays an important role in the rotation of crops being an excellent precursor plant. It accumulates appreciable amounts of nitrogen in the soil, and organic substances through the root system improving the physical and chemical properties of soils.

During its long history as a cultivated plant, numerous ecotypes, local populations, and alfalfa varieties have been developed under the influence of various pedo-climatic conditions that are adapted to a specific geographical area. For many farmers, the selection of the alfalfa variety means buying the cheapest product on the market, but if we analyze this issue in more detail, we may find that the performance of the variety is more important than the price of seeds.

The price of seeds is important up to a certain level, after which, the gain per kilogram of seed means financial losses per hectare in the productivity of the variety during the exploitation of the crop.

The specialists in the cultivation of fodder plants from Romania and abroad have proven repeatedly that the results obtained in the alfalfa crops are influenced by a series of characteristics related to the yield and quality of the obtained fodder.

A series of indices can provide a ranking of the varieties regarding these performance characteristics such as:

- growth rate;
- average yield (tons dry matter  $\text{ha}^{-1}$ );
- canopy height;
- tillering rate;
- indices of forage quality (crude protein, digestibility, etc.) - Sanderson (1992).

Due to the particularities of technology and economic importance, the alfalfa crop is a long-term investment (Smeal et al., 1991). To make the most of it, these investments must be followed as decisions taken to respect the correct order (e.g., pedochemical analysis of

the soil, planning of the sowing densities depending on the crop destination, the choice of a suitable variety adapted to the growth and development conditions, etc.).

These few details make a difference in the success of alfalfa cultivation. The goal of seeding high-yielding varieties able to provide the required crop performance is sustained by proper researches in breeding new cultivars adapted to the climate variations of the cropping area and global warming. Li and Brummer (2012) suggest several directions to improve selection in alfalfa including diversity selection and paternity testing, introgression of quantitative trait loci, and genomic selection.

An adequate breeding program for alfalfa suitable varieties should consider the combination of high yield and production that is well distributed over the growth season, with high quality of produced forage. Furthermore, they must be hardy and adaptable, relatively with lower demands in terms of agronomic inputs. Consequently, a successful alfalfa variety should present several bio-productive features such as:

- have a medium-tall height of canopy with thin stalks and a proper leaf-stem ratio;
- to be suitable for early cuttings allowing an intensive use;
- providing a compromise between hardiness and quality: good yield in fertile plain soils and in drier hilly areas or where the winters are colder;
- have a positive fiber digestibility and protein content;
- have appropriate radiation use efficiency (Dunea et al., 2019) and water use efficiency (Dincă et al., 2017);
- have a lower environmental impact based on its life cycle assessment (Wang et al., 2021).

In this context, a breeding program is promoted by S.C. Patru Agro S.R.L. (<https://samantalucerna.ro/>) in cooperation with the academic researchers for developing new alfalfa varieties that are suitable for the specific Romanian eco-pedoclimatic conditions. Such varieties should have improved winter hardiness due to the cold winters with a higher incidence and duration of frost days. The program provided two varieties i.e., Dobrogea and Valahia, which have been included in the

national official list of varieties (<https://istis.ro/image/data/download/catalog-oficial/catalog%202020.pdf>), while Dobrogea is included in the OECD list of varieties eligible for seed certification (Table 1).

The paper presents the multiannual results obtained from field-testing of a large number of alfalfa varieties from which 18 have been selected for synthetic presentation of the cropping performance based on previously mentioned indices. The screening of these varieties originating from Romania, Italy, France, and the U.S.A. can provide useful information for the farmers and specialists in fodder production.

## MATERIALS AND METHODS

The screening experiments were carried out on plots in Gherghita Plain, Puchenii Mari village

(N44.824060, E26.092660) – Figure 1. Each of the 18 *alfalfa* varieties had three replicates and several variables have been determined for each harvest cycle: Relative Growth Rate, Tillering Rate and type (vegetative and generative), Leaf/Stem Ratio (g DM leaf m<sup>-2</sup>/g DM stem m<sup>-2</sup>), net yield, Forage quality (based on several parameters), canopy height, RUE, and Leaf Area Index (determined indirectly with a Delta-T Devices SunScan SS1 canopy analyzer system and checked randomly using measurements of leaf area samples with a digital planimeter).

The collection comprises varieties from Romania, other European countries, and the U.S.A.

All plots have been maintained in the same optimal conditions of fertilization and irrigation during this multiannual experiment (2017-2020).

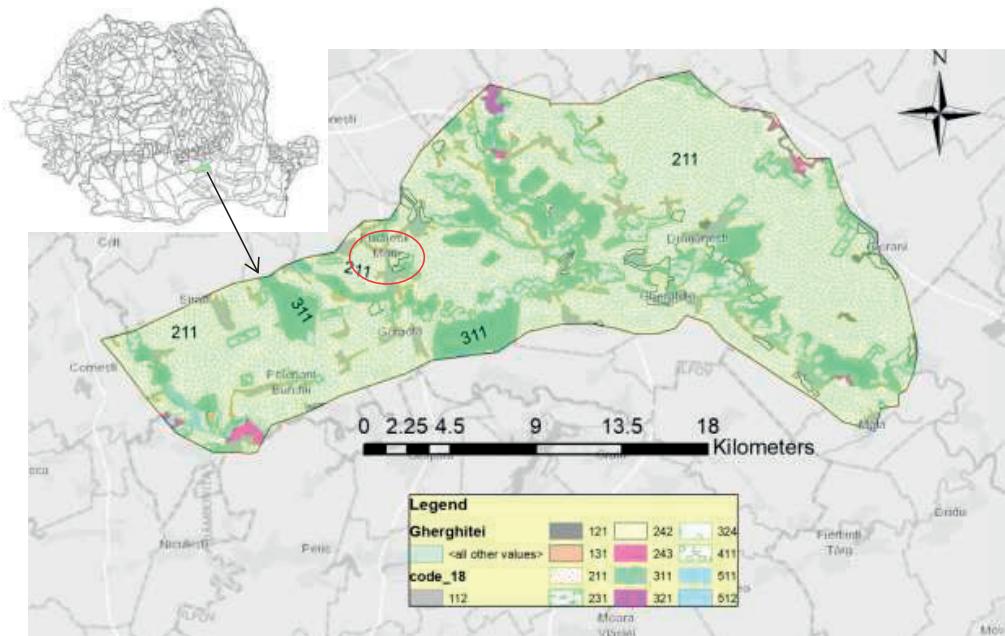


Figure 1. The study area located in Gherghitei Plain, south of Romania, in Puchenii Mari village - Land use/land cover (LULC) showing the 231 INSPIRE category attributed to Pastures; 242: Complex cultivation patterns; 321: Natural grasslands

More details regarding the experimental setup in Gherghitei Plain can be found in Dunea et al. (2019) and in Dincă and Dunea (2018) regarding the screening protocol of alfalfa varieties. Gherghitei Plain is a favorable area for alfalfa cropping.

Meteorological parameters such as net radiation, air temperature, air relative humidity, wind speed, etc. have been monitored with an Adcon Telemetry dedicated system mounted near the experimental field.

Table 1 presents the tested alfalfa varieties.

Table 1. Alfalfa varieties tested in the multiannual ranking experiment (7 Romanian and 11 foreign varieties from U.S.A./Austria, Italy and France)

Variety	Maintainer name	Maintainer code*	Country
Sandra	INCDA Fundulea	1562	Romania
Mădălina	INCDA Fundulea	1562	Romania
Dobrogea	SC Patru Agro SRL	2782	Romania
Mihaela	INCDA Fundulea	1562	Romania
Valahia	SC Patru Agro SRL	2782	Romania
Roxana	INCDA Fundulea	1562	Romania
Cezara	INCDA Fundulea	1562	Romania
Dimitra	Continental Semences	724	Italy
PR55V48	S&W Seed Company/Pioneer Hi-Bred Services GmbH	3133	U.S.A., Austria
Pomposa	Gennari & Schiavi	1241	Italy
Galaxie	GIE Grass	1266	France
PR54V09	S&W Seed Company/Pioneer Hi-Bred Services GmbH	3133	U.S.A., Austria
Gea	Roberto Guarneri/Maro Tarim Insaat ve Turizm	1359, 1988	Italy, Turkey
Harpe	GIE Grass	1266	France
Orca	RAGT 2n	2595	France
Bardine	Barenbrug Holland B.V.	401, 402	The Netherlands
Giulia	Natura Srl	2150	Italy
Letizia	Compagnia Generale Servizi	698	Italy

\*<https://www.oecd.org/agriculture/seeds/documents/codes-and-schemes-list-of-varieties-eligible-for-seed-certification.pdf>

The following indicators were determined to characterize the biological efficiency of the tested varieties and to establish an overall ranking in the eco-pedoclimatic conditions of the experimental field:

- *Growth Rate* - the accumulation of dry matter per unit area during a day (kg DM/m<sup>2</sup>/day) and *Relative Growth Rate* (RGR) - the average growth in length of alfalfa plants in centimeters per day (cm/day);
- *Degree of tillering* (DT) - the average of the number of generative shoots, respectively of the number of vegetative shoots per plant; the indicator is useful in estimating empirically the fodder production yield;

Fodder yield/m<sup>2</sup> = NP × [(NLP × GL) + (NFP × GF)] (NP - number of plants/m<sup>2</sup>, NLP - number of shoots/plant, GL - average weight of a shoot, NFP - number of leaves/plant, GF - average leaf weight);

- *Leaf/Stem Ratio* - the mass ratio between the amount of dry matter of the leaves and the amount of dry matter of the stems per unit area;
- *Forage quality* - a synthetic indicator of fodder quality using a regressive statistical model that uses as variables several indicators determined by the specific laboratory analyzes performed in an accredited laboratory, respectively dry matter content (DM), crude protein content (CP), crude fat content, crude cellulose content, crude ash content and the digestibility coefficient;
- *Estimated average forage yield* - represents the estimated amount of forage per unit area (t DM/ha);
- *Canopy height* - the average height of the canopy from ground to the average height of plants (cm).

All the characteristics were categorized using a five steps rank (1-low; 5-excellent) to screen empirically the performance of each variety in an easier way. Then, the multiannual ranks of the characteristics were used in a weighted regression in which more weight was given to the observations with smaller variance because these observations provide more reliable information about the regression function than those with large variances (Neter et al., 1996). More weights were attributed to forage yield and to forage quality as important parameters for assessing the overall crop performance.

## RESULTS AND DISCUSSIONS

A selection of the most relevant characteristics determined during the multiannual screening trials performed considered yield, forage quality, tillering, and growth rate.

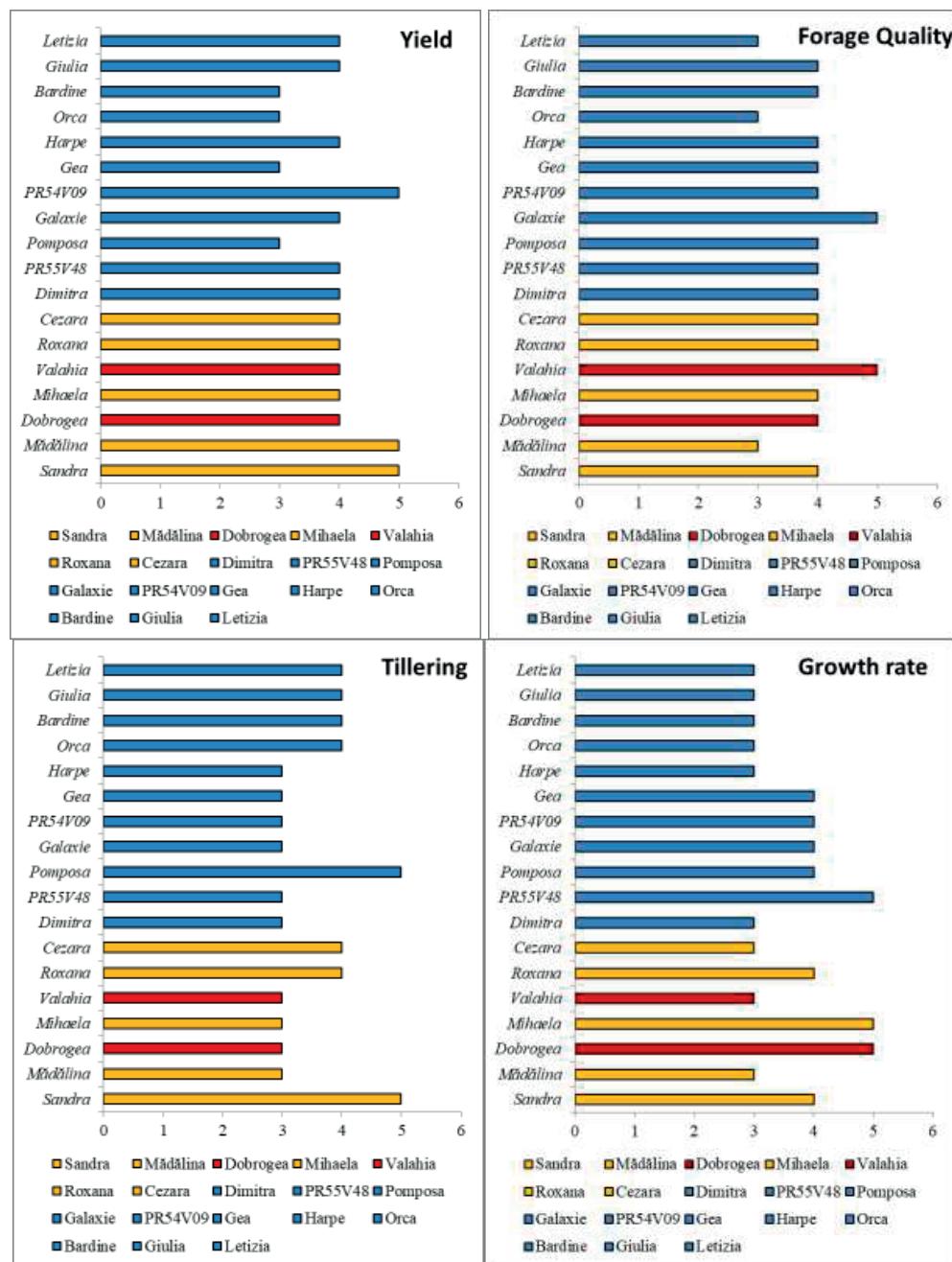


Figure 2. Evaluation of the most important parameters related to the crop performance in 18 alfalfa varieties in the eco-pedoclimatic conditions of Gherghita Plain, Romania using ranks from 1 to 5 (1-low; 5-excellent)

Some indicative values can be retrieved by using the means of all the varieties. Consequently, the RGR showed an average of 2.2 cm/day (Standard deviation - SD = 0.2) at the first cutting, 2.6 cm/day (SD = 0.3) at the

second cutting, and 0.9 (SD = 0.3) at the third cutting. The growth rate had an average of 2.4 g DM m<sup>-2</sup> (SD = 0.2) leading to an overall yield of 14 t ha<sup>-1</sup> DM.

Regarding the tillering capacity, the average of varieties for all cuttings reached 16 generative shoots ( $SD = 6$ ) and 13 vegetative ones ( $SD = 5$ ) per plant. The L/S ratio presented a lower variability having an average of 0.7 ( $SD = 0.1$ ) and the canopy height was 60 cm ( $SD = 6.5$ ).

Regarding the feed value of the resulted fodder, the average of the 18 varieties for the feed unit for milk (UNL - assessed on the basis of the energy effect of barley; 1 UNL = 1700 Kcal = 7.11 MJ) was 0.9 kg/DM ( $SD = 0.03$ ) and for the coefficient of organic substances digestibility was 75 ( $SD = 0.25$ ).

Figure 2 shows synthetically these results. It can be seen that two Romanian varieties (Sandra and Mădălina) and one U.S. variety (PR54V09) reached the highest yields, but in terms of forage quality, Valahia and Galaxie were the most qualitative. Interestingly, most of the varieties reached good qualitative levels by rank 4. There were three exceptions (Letizia, Orca, and Mădălina). Regarding the tillering capacity, Sandra and Pomposa had the highest rank, and generally, the foreign varieties showed a better tillering than the Romanian ones. In terms of growth rate, Dobrogea, Mihaela, and PR55V48 were the most relevant between the tested varieties.

Figure 3 shows the overall results provided by the weighted regression for adjusting a hierarchy that included all the measured characteristics. It can be observed that the selected varieties both Romanian and foreign ones showed good performances in the experimental conditions under optimal irrigation and fertilization regime. The average of the synthetized rank for the Romanian varieties was 3.8 ( $SD = 0.27$ ), and 3.6 ( $SD = 0.25$ ) for foreign varieties.

The maximum rank was obtained by two Romanian varieties i.e. Dobrogea and Sandra followed by PR55V48. All the selected varieties have valuable bio-productive characteristics being a useful selection pool considering that the *variety selection* is the foremost decision growers can make.

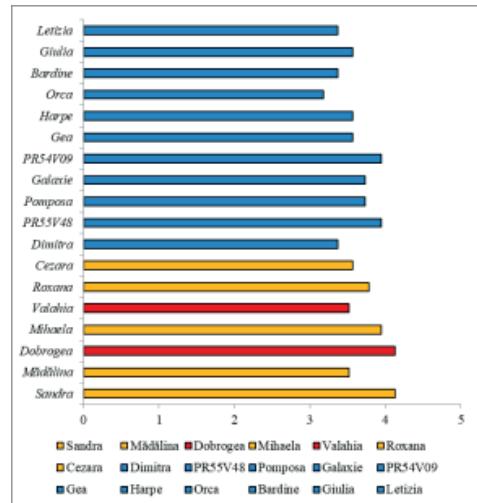


Figure 3. Results of the weighted regression showing the overall performance of the tested alfalfa varieties

Other characteristics that must be taken into account for future assessments are dormancy, resistance to diseases and pests, and the resistance to drought. In Romanian varieties, the dormancy reaches 4-4.5, being considered high, representing an important asset for the success of alfalfa cultivation, considering the frequency of frosty winters in Romania. Foreign varieties have a lower dormancy (6-7), respectively a lower frost resistance. This limitation must be carefully analyzed before considering the selection of a foreign variety. New instruments for grassland and forage crops analysis such as multi- and hyper-spectral detection sensors are available on satellites and on aerial platforms including UAVs increasing the spatiotemporal performance for acquiring ground data from terrestrial ecosystems including forage cropping systems (Noland et al., 2018). This promising technique can provide support in validating the performance of alfalfa varieties on large-scale farms in the context of the forecasted aridity index, which is expected to increase in the south of Romania according to the simulations from the climate models.

## CONCLUSIONS

By presenting the certified cultivated varieties in Romania, we tried to bring in the attention of the alfalfa growers, the main characteristics of the current certified Romanian and foreign varieties and thus providing them the possibility to choose the right variety depending on the particularities and objectives of their farm. This study can be used as a benchmark for the performances of the tested varieties.

Based on the results, the Romanian varieties presented very good ecological plasticity with drought and frost resistance combined with high biological efficiencies. Dobrogea and Sandra are very productive varieties, while the new variety Valahia showed the highest forage quality. In the eco-climatic conditions of South Romania, PR54V09 and PR55V48 have been the most high-yielding varieties from the foreign group of cultivars.

In the next period, we will continue to analyze the behavior of certain alfalfa varieties in various farm conditions by testing a selection of varieties in production fields and we will return with updated information to help the farmers in taking the substantiated decisions in choosing the proper alfalfa variety for their needs (<https://samantalucerna.ro/>).

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