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## **SUMMARY**

Keywords: inoculation, fertilization, harvest time, productivity, CP (crude protein), NDF (neutral detergent fiber), ADF (acid detergent fiber), calcium/phosphorus ratio

Biomass productivity and quality of alfalfa (*Medicago sativa* L.) has always been in the attention of numerous breeding companies. New cultivars are continuously created with better productivity and quality, following the improvement of the genetic material.

Alfalfa has a high quality potential, the ability in applying the optimal cropping technologies will be found in the volume of production and forage quality. The factors influencing productivity and forage quality of alfalfa are represented by soil fertility, cultivar, weed infestation, pesticides, climate, harvesting conditions (harvesting season, harvesting timing during the day, plant growth stage), preservation and storage methods.

In the agricultural practices, the relationship quantity - quality in forage production is less considered, which is the reason for the failure in estimating the correct harvesting time, with well-known negative consequences.

Therefore, improving the technology of growing alfalfa and thorough analysis of the relationship between biomass productivity and quality, in the stationary conditions from northeast moldavian forest steppe is an activity of topical and high interest.

In the attempt to solve these problems, this doctoral thesis aims to determine the nutritive value of alfalfa (*Medicago sativa* L.) harvested at different growth stages in the moldavian forest steppe conditions.

The objectives and research activities aims to highlight and quantify the elements that influence the nutritive value of alfalfa in different growth stages at harvest and to measure the indicators influencing the biomass productivity on different growth stages at harvest.

The doctoral thesis is structured in two parts and six chapters.

The first part, which contains 63 pages (25% of the doctoral thesis), representing chapters I, II and III, represents a study of the literature in order to understand the current stage of research that relate to the improving of cultivating technology of alfalfa in order to achieve a higher quality of the forage.

**The first chapter** describes the origin, geographic distribution, importance, plant morphology and cultivating technology of alfalfa (*Medicago sativa* L.).

**Chapter II** contains a revision of the latest internal and external research regarding the nutritive value of alfalfa (*Medicago sativa* L.) on different growth stages. It describes the results of the most important scientific papers published, aiming generally the improvement of the technology for growing alfalfa and, in particular, the quality of alfalfa in different growth stages







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at harvest.

Description of the natural conditions of the area where the studies were conducted are exposed in **Chapter III**. The research were conducted in the year 2011 at the Ezareni farm (47°05'-47°10' north latitude and 27°28'-27°33' east longitude) which belongs to the research facility of the University of Agricultural Sciences and Veterinary Medicine from Iasi. Structurally, the territory of Ezăreni farm is located entirely on the old platform unit called "Moldavian Platform", which is an extension of the "Russian Platform" in our country. Ezăreni farm is part of the Dfbx climate province (Koppen's classification), characterized by boreal climate with cold and frosty winters. Average annual rainfall in the farm Ezăreni is about 517.8 mm and the annual average temperature is 9.7°C.

The soil is a cambic chernozem, which is characterized by a neutral - slightly acid pH in the arable layer, with values ranging from 6.68 to 7.01 pH units. The soil is "medium" supplied with humus in the range 0-30 cm (2.40%) and "poor" in the rest of the profile. Analysis of soil total nitrogen (Nt) shows an "medium" supply in the 0-30 cm of soil (1.78%) and "poor" supply in the rest of the profile. Regarding soil P content, the Ap horizon (0-20 cm) has a "medium" supply (26.00 ppm), Atp horizon (20-28 cm) has a "poor" supply (10.43 ppm) the other horizons are "very poor" supplied in P.

The natural vegetation is represented by herbaceous species characteristic to a rather dry steppe climate and totally isolated, with some steppe shrubs.

Part II, containing 75% of the doctoral thesis (191 pages), presents the results of research on the influence of inoculation, fertilization and growth stage at harvest on the productivity and quality of alfalfa.

In Chapter IV are described: thesis purpose, objectives and activities of the research, research methods, cultivating technology and a description of the climatic conditions during experimentations.

In order to achieve thesis purpose and objectives, an experience was organized at the farm Ezăreni affiliated to the University of Agricultural Sciences and Veterinary Medicine from Iasi, in the spring of 2010, which studied three factors, namely: A-innoculation with two graduations ( $a_1$ -without seed innoculation,  $a_2$ -with seed innoculation); factor B-fertilization with four graduations ( $b_1$ -unfertilized,  $b_2$ - $N_{50}P_{50}$ ,  $b_3$ - $N_{75}P_{50}$ ,  $b_4$ -30 t/ha manure); factor C-plant growth stage at harvest with six graduations ( $c_1$ -early bud,  $c_2$ -mid bud,  $c_3$ -late bud;  $c_4$ -early bloom,  $c_5$ -10% bloom and  $c_6$ -full bloom).

The experimental design was a trifactorial split plot design 2x4x6, arranged as subdivided plots in three replicates, with a plot size of 3x6 m  $(18 \text{ m}^2)$  and a harvested area of  $10 \text{ m}^2$  (2x5 m), the total area of the experience:  $2730.425 \text{ m}^2$  (73.30 x37.25 m).

The biological material used was represented by Sandra alfalfa variety (F 660-94) registered in 2003 to I.N.C.D.A. Fundulea, consisting of six families selected from hybrids obtained by crossing local varieties of Adonis, Selena, Sigma and varieties of French origin Concorde, Alize and Vertus.

Research activities were focused in: measurements of plant height, dry matter determination, CP (crude protein), NDF (neutral detergent fiber), ADF (acid detergent fiber),













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magnesium, relative value (RFV), determination of calcium, phosphorus, calcium/phosphorus ratio, calculation of the total amount of dry matter and CP produced per unit area and the leaves/stems ratio.

All observations, measurements and analyzes were in accordance with experimental techniques and standards. The results were interpreted statistically by analysis of variance and limit differences calculation. Also, were calculated the regressions of correlations between growth stages at harvest and analyzed parameters.

Considering the climate, the period 2009-2012 was normal in terms of mean air temperature, which is between 10.4 to 10.8°C, with 0.2 to 0.8°C higher than the annual average, but during the growing season with a positive deviation of 1.3 to 2.2°C above the annual average.

Precipitations variated, depending on the agricultural year. Thus, the agricultural year 2009-2010 was rich in rainfall, the annual amount of rainfall was 648.8 mm, 131.0 mm higher than the annual average and the amount of rainfall recorded during the vegetation period (April-September) was 446.0 mm, 108.5 mm higher than the annual average of 337.5 mm.

The agricultural year 2010-2011 was closer to a normal year. Annual amount of rainfall was 507.6 mm, 10.2 mm lower than the annual average of 517.8 mm. The amount of rainfall recorded during the growing season was 342.8 mm, 5.3 mm higher than the annual average of 337.5 mm.

The agricultural year 2011-2012 was a dry one; the total annual rainfall was only 355.4 mm, 162.4 mm lower than the annual average of 517.8 mm. The amount of rainfall recorded during the vegetation period (April-September) was 245.2 mm by 92.3 mm lower than the annual average of 337.5 mm. In all cases poor distribution of rainfall is the main drawback.

Generally, the agricultural year 2009-2010 was favorable for establishment, growth and development of alfalfa crop, 2010-2011 was a near normal year, and the agricultural year 2011-2012 was less favorable with drought both in winter and summer.

Chapter V presents the results of research on the influence of inoculation, fertilization and growth stage at harvest on plant height, the ratio between leaves and stems of alfalfa and dry matter yield, in the second and third year of vegetation.

The results showed that inoculation influenced positively plant height, but the differences were significant at first and third cut only.

Seed inoculation contributed significantly by raising the DM content in leaves and indirectly by improving feed quality, knowing that the leaf content of CP is at least double than in stems.

Fertilization showed a differential effect, depending on the cut, on plant height of alfalfa and production of DM in the whole plant, leaves and stems. Results indicated a residual effect of manure on the analyzed indicators. Thus, if at the first cut N<sub>75</sub>P<sub>50</sub> fertilization had the greatest influence, at following cuts fertilization with 30 t/ha manure generated the biggest differences.

The factor with the greatest influence on biomass productivity of alfalfa was the plant growth stage at harvesting. Plant height was increasingly higher during plant development but a decreasing trend of the growth rate starting with early flowering. Correlations between age and plant height at the cuts I, II, III year II of vegetation and first cut in the third year of vegetation







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were statistically positive.

From the results it was observed that plant height is positively correlated (very significant) with accumulated temperature during plants development and growth, only at the first cut when sufficient rainfall was provided and the height depended on the amount of heat. Although the overall trend is positive, the amount of heat and plant height does not correlate. Under these conditions, rainfall becomes the limiting factor. Regardless of the cut or phenophase when alfalfa is harvested, there is a very significant correlation between precipitations and plant height. Along with plant development and growth during vegetation, the DM yield of stems and entire plant biomass has an ever continuing growth. The DM yield of leaves grows until the stage of late bud-early flowering, then declines. Effectively, once the first flowers appear, the leaves at the base of stems etiolate, then dry and fall. This phenomenon directly affects the quality of the feed. Hence, as plants are harvested later than the beginning of flowering, leaf production will be incresingly lower as well as the forage quality.

Analyzing the influence of harvest time on the distribution of dry matter production per cut in the second year of vegetation, it is clear that later harvests contribute to the increasing of total DM production. It was observed that the total production of DM, at the first cut is 45-48%, at second and third, 20-27% respectively 26-27%, and 0-9% at fourth cut. Even if DM yields are different depending on the growth stage at harvesting, percentage distribution did not differ very much from one cut to another.

In terms of leaves production, growth stages  $c_2$  (mid bud),  $c_3$  (end bud) and  $c_4$  (early flowering) were the height of this indicator. The situation was similar to the total DM production, but with 4.9% lower at first cut, 2-6% and 2-3% at second and third cut.

Stems production greatly increased with later harvests, from the appearence of buds  $(c_1)$  to complete flowering  $(c_6)$  stems production almost doubles. Regardless of growth stage at harvest, at the first cut was obtained half (48-53%) of the stems. At the next cuts total amount of stems was almost equal.

Chapter VI presents the results of research on the influence of inoculation, fertilization and growth stage at harvest on CP, NDF, ADF, Ca, P, Mg contents, RFV and the Ca/P ratio during the growing season II and III, at the first cut.

The results showed that there was a significant negative correlation between growth stage at harvest and the leaves/stems ratio at the first, second and third cut of the second year of vegetation and first cut in the third year of vegetation.

Analysis of the influence of inoculation on alfalfa quality showed that it influenced significantly the content of NDF in leaves and stems and the content of ADF in stems. Also in the case of other quality indicators (CP content and RFV) there were differences, but statistically not assured. This shows that in the experimental conditions of this study, inoculation did not contributed to the quality of feed.

Crude protein content of plants was negatively influenced by mineral fertilization, even if the leaves CP content was higher, while the stems CP content was less affected, the ratio between leaves/stems influenced this indicator.

Organic fertilization determined high significant differences on the content of NDF and













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ADF in entire plants, leaves and stems. The same trend was manifested at mineral fertilization, except NDF and ADF contents in stems, where the values obtained were negative.

Analyzing the influence of harvesting timing on the content of NDF and ADF in plant leaves and stems we observed that along with ageing of plants, the value of these indicators increased. Correlation between growth stage at harvest and content of plant leaves and stems alfalfa in NDF and ADF is positive, significant and distinctly significant. As plant growth stage passes, NDF and ADF content is increasingly influenced by the content of strains in these compounds as stems have a higher proportion than leaves.

The largest amount of CP per unit area was obtained when alfalfa was harvested in the mid bud stage. Correlation between growth stage at harvest and CP amount in leaves, stems and entire plant shows an increasing trend until the end bud stage, followed by an accentuated decline.

Inoculation had no significant influence on Ca, P, Mg content and Ca/P ratio, virtually the differences were 0.01 g/kg DM or no difference. Instead, within all variants of fertilization, the Ca/P was very significantly reduced, by adding phosphorus through the application of mixed nitrogen and phosphorus fertilizers or manure, which is a complex fertilizer of the highest quality. This outcome is beneficial to the animal health, the natural Ca/P ratio represents a shortcoming of alfalfa, any improvement of this indicator being very important.

Analyzing the influence of harvesting timing on the amount of Ca, P and Mg exported through biomass, we observed that during early bud-end of flowering, total phosphorus exported through biomass has been steadily increasing, along the succession of these stages, while the greatest amount of phosphorus exported through leaves was at the end bud-early flowering growth stage. Between growth stages at harvest and total phosphorus exported through the production of stems, leaves and total DM there was a distinct significant correlation.

Towards the end of the thesis are presented the conclusions and recommendations drawn from the results, as well as a selection of references consulted during the completion of this thesis.