

ANNOTATION

for dissertation of Kenebaev Amankeldi Turgambekovich on the theme «Screening of *M.sativa L.* and *M.varia Mart.* alfalfa germplasm collections for breeding» submitted for degree of philosophy doctor (PhD) on speciality 6D080100 – «Agronomy»

Relevance of the research topic. Alfalfa is a widely used fodder crop in global agriculture. Sowing alfalfa in Kazakhstan is about 1.5 million hectares. The increase in alfalfa sowings is one of the most important measures in the formation of the fodder base for livestock. At the same time, it is very important to obtain high-yielding varieties of alfalfa and introduce them into production.

Efficient use of the gene pool in breeding is one of the main goals, and the selection of source material adapted to local environmental conditions is the most important stage of breeding.

The culture of alfalfa is diverse. According to the latest scientific data, 21 species of alfalfa occur in nature and in cultivated species, and in Kazakhstan there are 7 species of alfalfa: *M. Sativa L.*, *M. varia Mart.*, *M. falcate L.*, *M. trianschanica Vass.*, *M. coerulea Less.*, *M. trautvetteri Summ.* and *M. difalcata Sinsk.* Wild alfalfa species are diploid ($2n=16$), cultivated species are tetraploid ($2n=32$). In Kazakhstan, two species of *M. sativa L.* are mainly cultivated, and *M. varia mart.* Therefore, 134 alfalfa varieties belonging to these two species originating from 18 countries were taken for research. The variety samples were characterized by economically valuable traits and properties, the most perfect samples were selected and presented as a source material for the development of breeding work. At the same time, the relationship of traits was studied and their influence on productivity (green mass and seed yield) was determined.

Research on this topic, dissertation work was carried out in accordance with the thematic plan of the Kazakh Research Institute of Agriculture and Crop Production and is one of the stages in the creation of new high-yielding varieties for the southern and southeastern regions of the Republic of Kazakhstan.

New varieties provide an opportunity to increase the yield of alfalfa by 15-20% without additional costs. At the same time, the effective use of the gene pool in breeding confirms the relevance of this topic.

The purpose of the dissertation research.

Selection of promising varieties of alfalfa required for breeding in terms of economically valuable traits.

The main objectives of the study:

To achieve the goal of the study in the framework of the dissertation work, the following tasks were set:

- selection of the most valuable variety samples of the alfalfa collection with breeding traits in terms of properties and productivity;
- analysis of the evaluation of breeding traits for each cutting, depending on the year of alfalfa growth;
- determination of resistance to diseases of varieties of alfalfa;
- selection of the best varieties in terms of seed productivity from the alfalfa collection;
- selection of source material from the studied collection for effective use in alfalfa breeding, depending on the purpose of the selection.

Research methods.

To determine the most valuable specimens of the alfalfa collection, the following selection traits and properties were carefully studied and evaluated.

1) Phenological observations: year of sowing, date of sowing, germination and full germination of seeds, appearance of the first, second and third true leaves, budding phase: beginning of budding and flowering; spring regrowth and regrowth after each mowing, monitoring the formation and formation of the brush. According to the date of the beginning of the alfalfa flowering phase, the studied varieties were divided into early-ripening, mid-ripening and late-ripening groups.

2) Plant height. Basically, they were determined before each mowing and when observing the dynamics of phase growth. Measurements were made in 3-fold repetition on each studied variety sample.

3) Determined before each mowing by counting the number of shoots extending from the tillering node.

4) Leafiness of plants. To determine it, we used trial sheaves, which were divided into two fractions: stems and leaves.

The "leaves" fraction included inflorescences and petioles of leaves. foliage expressed as a percentage of leaves to the total weight of the test sheaf.

5) Accounting for winter hardiness and thinning in the summer period was carried out by counting each plant on each variety sample (per 1 m² in triplicate) during the period of spring regrowth and before going into winter (autumn).

6) Accounting for vegetative organs was carried out at the first, second and third cuttings, and in the fourth cutting it was limited only to counting the growing stems. In all samples, the number of growing stems (bush) and the number of branches per plant, leaf size, plant height, number and length of internode, and stem thickness were studied. For this, 10 plants were selected from each variety.

7) Accounting for the yield was carried out in the phase of the beginning of flowering, by direct weighing of the green mass. And to determine the yield of dry matter (hay), a trial sheaf of 0.5 kg was taken from each studied sample. The yield of hay was determined as a percentage, then the yield of green mass was recalculated for the corresponding yield of air-dry mass (hay).

8) Infections with fungal diseases were detected at the first and second stages of mowing. The degree of infection with fungal diseases was noted at the beginning of the flowering phase (before cutting) and was determined by a five-point system:

0 - no spots on the leaves

1 - the surface of the sheet is covered with spots up to 10% of their total area:

2 - from 15 to 25%

3 - from 30 to 50 %

4 - above 50%

9) In the Laboratory of Biotechnology, Biochemistry, Plant Physiology and Product Quality Evaluation of the Institute, an analysis was carried out for protein content in the most valuable studied samples of alfalfa.

10) The nature of the formation of the generative part of the variety samples and the structure of the seed productivity of alfalfa have been studied. The studies were carried out in the 3rd year of alfalfa vegetation, at the second mowing. To study the generative organs and the structure of their development, as well as the structure of the seed yield, 10 plants from each sample were studied. Accounting was made by counting the inflorescence of the main stem and the number of flowers that appeared on the first and subsequent branches, as well as the duration of flowering, inflorescences and their density. The number of inflorescences was determined on average from 10 plants.

Self-pollination was determined in alfalfa varieties by isolating 10 medium inflorescences from the main stem using parchment insulators. Under each insulator was placed (up to three inflorescences before flowering).

11) Accounting for the seed yield from the accounting plot was carried out.

12) The R and Rstudio programs were used for statistical processing of the experimental results. Link to R and Rstudio below.

Citing R and RStudio - <https://ropensci.org/blog/2021/11/16/how-to-cite-r-and-r-packages/>

The main provisions submitted for defense (proven scientific hypotheses and other conclusions that are new knowledge).

- From two species of alfalfa *M. Sativa L.*, and *M. varia mart.* 134 varieties were classified according to breeding values and traits, the best specimens were selected for each trait;

- Carrying out a correlation analysis of the relationship between signs of green mass and seed productivity;
- Selection of source materials for selection and proposal for their use.

Description of the main results of the study:

Based on the studied 134 variety samples from the world gene pool, the source material with economically valuable traits for various breeding areas was selected:

1. The earliest maturing samples were taken from Italy (k-5975), Turkmenistan (k-8883).
2. According to winter hardiness, the following varieties were distinguished: Canada (k-33299), Russia (k-45860), Ukraine (k-21826).
3. Plant height of alfalfa (*M. Sativa L.*) stands out samples: from Italy (k-5677), Pakistan (k-41985), Italy (k-27065), Kazakhstan (k-6021), Ukraine (k-1721), Russia (k-11417), and among the samples of variable alfalfa (*M. varia Mart.*): from Ukraine (k-20002), Russia (k-31885), Ukraine (k-21787), Estonia (k-38914).
4. In terms of productive tillering among alfalfa (*M. Sativa L.*) the following varieties distinguished themselves: from the USA (k-14), Italy (k-5677), France (k-315), Uzbekistan (k-267) and among samples of variable alfalfa (*M. varia Mart.*): from Canada (k-39932), Ukraine (k-26713), Kazakhstan (k-47492), Ukraine (k-23206), Kazakhstan (k-34627).
5. By the number of branches among alfalfa (*M. Sativa L.*): from Italy (k-5677), Pakistan (k-41985), China (k-11), France (k-315), Kazakhstan (k-36049), among variable alfalfa (*M. varia Mart.*) the highest indicator was: from Kazakhstan (k-34627), Ukraine (k-20002), Russia (k-31885), Ukraine (k-26713), Estonia (k-38914).
6. According to the parameter of high foliage among the varieties of alfalfa (*M. Sativa L.*) the best results were shown by: Azerbaijan (k-45905), USA (k-46451), Egypt (k-5143), Armenia (k-45036). and according to samples of alfalfa (*M. varia Mart.*): Russia (k-31885), Canada (k-33299), Canada (k-39932) Kazakhstan.
7. Among the studied varieties, the most resistant to fungal diseases were identified. The best complex resistance to fungal diseases (yellow leaf spot, brown leaf spot and rust) was shown by varieties from Kyrgyzstan (k-6238), Uzbekistan (k-21634), Italy (k-5975).
8. The yield of green mass among alfalfa (*M. Sativa L.*) exceeded the standard from 32.5 - 51.1% samples: from Uzbekistan (k-267), France (k-315), Russia (k-9), China (k-11), Italy (k-5677), Kazakhstan (k-191). For alfalfa (*M. varia Mart.*) samples: from Ukraine (k-446), Russia (k-538), Russia (k-406), Ukraine (k-454), which respectively exceeded the standard from 23.2 - 36,5 %.
9. The yield of dry mass (hay) of varieties of alfalfa (*M. Sativa L.*) was: from France (k-315), Russia (k-9), Italy (k-5677), Uzbekistan (k-267) which is higher than the standard by 25.9 - 41 %. Among the variable alfalfa (*M. varia Mart.*), the best results were shown by varieties: from Russia (k-538), Russia (k-406), Ukraine (k-454), these samples were higher than the standard by 21.0-25.6 %.
10. The amount of crude protein in the first cut varied from 17.0 to 20.1 %, the highest rates were noted in samples from Armenia (k - 313) - 20.1 %, Kazakhstan (k - 246) - 18.5 %. In the second cut with a high protein content, samples from Armenia (k-313) - 21.1%, Italy (k-5677) - 22.1 %, Kazakhstan (k-246) - 22.6 % and Russia (k-322) - 22.3 % differed.
11. In the collection nursery of alfalfa (*M. Sativa L.*), the largest number of racemes was formed in varieties: from Russia (k-473), Turkmenistan (k-253), Uzbekistan (k-226), Italy (k-5677), France (k-315) in variable alfalfa (*M. varia Mart.*): Ukraine (k-446), Estonia (k-404), Kazakhstan (k-34627), Ukraine (k-450).
12. In cross-pollination, the ability to set seeds between alfalfa varieties varied from 38.4 to 62.3%. Most alfalfa accessions originate from North America and the Eurasian continent, and some countries in Western Europe and Central Asia have a high percentage of cross-pollination. In these samples, seed setting ranged from 62.3 to 69.8 %. Samples with high fertility were: from Ukraine (k-450), Turkmenistan (k-253) and France (k-315). Their ability to bind seeds was at the level of 67.3 - 72.1 %.

13. Self-fertility of varieties from China (k-33740), Kazakhstan (k-36119) was at a high level - 13.5-14.2 %.

14. According to the largest number of seeds in beans, the best indicators are for samples from Sweden (k-356), Ukraine (k-450), Turkmenistan (k-253) and France (k-315).

15. Seed yield of alfalfa (*M. Sativa L.*) is high had varieties: from the USA (k-365), Turkmenistan (k-253), Russia (k-473), Italy (k-5677), France (k-315) and accordingly, exceeded the standard by 15.2 - 21, 9 %. And for alfalfa (*M. varia Mart.*) samples: from Kazakhstan (k-34627), Ukraine (k-21826), Estonia (k-404), Ukraine (k-450) which also exceeded the standard from 12.9 - 18.7 %.

Substantiation of novelty and significance of the obtained results. As a scientific novelty of the study:

The scientific novelty of the study lies in the fact that through their complex isolation, not only valuable traits and properties of valuable initial samples for alfalfa breeding were obtained, but also specific recommendations for breeding were given. For the first time, the important properties of generative organs were described in the initial samples of alfalfa: self-pollination, the ability to bind seeds due to fertilization, participation in the production of a new variety using isolated samples.

Depending on these indicators, the potential productivity of seeds and the level of its implementation were theoretically determined.

The practical significance of research: alfalfa variety "Kokshalgyn" was created with the participation of a researcher (author's certificate No. 1011 dated 12/30/2022). Based on the data obtained, promising samples of alfalfa are recommended for use in breeding work as a source of valuable traits. Valuable source material isolated as a result of research was transferred to the laboratory of fodder crops of the Kazakh Research Institute of Agriculture and Plant Growing to supplement the breeding technology and used in breeding new varieties.

Compliance with the directions of development of science or government programs.

The dissertation work was carried out according to the following programs of "KazNII of Agriculture and Plant Growing" LLP - "Creation of highly productive varieties of perennial grasses with high fodder qualities: multi-cutting alfalfa, rapidly growing under irrigation conditions and sainfoin, drought-resistant sweet clover with increased nitrogen fixation capacity, wheatgrass adapted for the steppe and semi-desert zone » No. state. 0118RK01209 (2018-2021);

- " Research and ensuring the conservation, replenishment, restoration and efficient use of agricultural plant genetic resources to ensure the breeding process " No. state. BR10765017 (2021-2023);

Description of the contribution of the doctoral student to the preparation of each publication.

During the work on the dissertation, the doctoral student took part in the development of the program and research methodology, set up and conducted experiments, and showed great responsibility. He carried out his research tasks with great interest. Significant results have been achieved through the correct use of specific methods of control, accounting and analysis in solving the tasks. The author personally participated in experimental studies and mastered field and laboratory methods, as well as methodological requirements for research on problems of agricultural science. All the results and conclusions given in the dissertation were obtained and formulated based on the results of the study with the direct participation of the researcher. Based on the materials of the dissertation research, 8 scientific articles were published.

In the data of international scientific conferences:

- Abaev S.S., Meirman G.T., Erzhanova S.T., Kenebaev A.T. Genetic resources of wild species of alfalfa (*Medicago L*) // Mater. Int. scientific pract . Conf . "Innovative approaches to the use of agrobiodiversity in the context of agricultural development". – Tashkent. – 2019. – P. 44-48.

- Kalibaev B. B., Bekturganov A., Kenebaev A. T. Yield of green and dry mass of hybrid alfalfa populations // Collection of materials of the international scientific and practical conference

of young scientists "innovative ideas aimed at the development of agriculture in regions with a sharply changing climate". - Ashutas: 2020. - P. 61-65.

- Kenebaev A. T. Selection of highly productive alfalfa samples for breeding purposes in the conditions of south-east Kazakhstan // Seifullin Readings – 18: Youth and Science – a look into the future materials of the international scientific conference. - Astana: 2022.- № 1(2). - P. 294-298.

- Kenebaev A.T., Kaskabaev N.B. The study of the alfalfa collection in the conditions of the southeast of Kazakhstan according to the main economic and valuable features // Collection of materials of the international scientific and practical conference "Adaptation of crop production to the conditions of global climate change: problems and solutions". – Almalybak. – 2022. - P. 109-112

"Committee for Quality Assurance in Science and Higher Education" of the Ministry of Science and Higher Education of the Republic of Kazakhstan:

- Kenebaev A.T., Meyirman G.T., Erzhanova S.T., Abaev S.S. a comprehensive assessment of the collection of sowing (*M. Sativa L.*) and (*M. varia Mart.*) alfalfa species as a source material for breeding. Bulletin of the Korkyt Ata Kyzylorda University. - Kyzylorda, 2022. - №2(62).- P. 261-273. <https://doi.org/10.52081/bkaku.2022.v62.i3.101>

- Kenebayev A.T., Yerzhanova S.T., Yesimbekova, M.A., Abayev S.S., Kalibayev B.B. Fertility of alfalfa varieties in self-pollination and cross pollination // bulletin of the Korkyt Ata Kyzylorda university. - Kyzylorda, 2022. - №4(63).- P. 160-169.

<https://doi.org/10.52081/bkaku.2022.v63.i4.140>

In foreign journals and scientific journals with a high percentile in accordance with the requirements of a peer-reviewed dissertation:

- Kenebayev A.T, Meiirman G.T, Yerzhanova S.T, Yesimbekova M.A, Abayev S.S. Manifestation of Valuable Selective Traits in Alfalfa Collection Samples // OnLine Journal of Biological Sciences. – 2022. Vol. 22, №2. - P. 237-246

DOI: <https://doi.org/10.3844/obscl.2022.237.246>

- Kalibayev B.B., Meiirman G.T., Yerzhanova S.T., Abaev S.S., Kenebaev A.T. Genetic diversity of perennial wild species of alfalfa subgenus *falcago* (*reichb*) *grossh* . In Kazakhstan and their contributions in the breeding // AGRIVITA Journal of Agricultural Science. - 2021. Vol. 43(2). – P. 300–309

DOI : <http://doi.org/10.17503/agrivita.v43i2.2894>

About volume And dissertation structure .

The dissertation consists of an introduction and main part, conclusion, references and applications. The main part is divided into 7 chapters. The total volume of the dissertation is 132 pages, including 25 tables and 28 figures. List of used literature - 162, including 70 foreign publications.

I express my gratitude scientific adviser who provided scientific and methodological assistance in carrying out research work: doctor of agricultural sciences. Sciences Professor, Academician of the National Academy of Sciences of the Republic of Kazakhstan Meiirman Galiolla Tolendiuly and Head of the Laboratory of the Genetic Fund of Agricultural Crops, Doctor of Biol. Sciences Professor Yessimbekova Minura Akhmetovna, as well as the scientific team of the laboratory of fodder crops.