
CROP PRODUCTION

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Zarianova Z. A.¹, Bekuzarova S. A.^{2,*}

¹ Federal Research Center for legumes and cereal crops, Streletsky, Russia;

² North Caucasus Research Institute of Mountain and Foothill Agriculture, Mikhaylovskoe, Russia

* Corresponding author (bekos37[at]mail.ru)

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THE PHENOTYPIC FEATURES OF RED CLOVER (*TRIFOLIUM PRATENSE* L.) THAT CORRELATE WITH SEED PRODUCTIVITY

Research article

Abstract

To reduce the time of the selection process when creating varieties of red clover (*Trifolium pratense* L.) with a high seed yield, it is necessary to use the source material with the features that positively correlate with an increased seed productivity. It is estimated that in the Northern part of the Central Black Earth Economic region of the Russian Federation the seed productivity of red clover has a high correlation with the weight of the dried mass, a number of stems, inflorescences, seeds in the flowers, seed setability, inflorescence dissemination and a short corolla tube. The average correlation coefficient was found between the seed yield productivity (1000 seeds), the length of the stem and the full visibility of the leaf pattern. A lesser degree positive correlation was found between seed productivity and a number of beans in the flower, two-seeded beans, a number of internodes, the length of time from the beginning of the spring regrowth and up to the anthesis.

The research has revealed the correlations between the seed productivity and economic, biological, and morphological characteristics of red clover in the Northern part of the Black Earth Economic region of the Russian Federation, and the North Caucasus region, in particular in the Oryol Oblast and North Ossetia-Alania, where this crop occupies a leading position among the cultivated perennial herbs. It is used in biological farming at the hayfields and pastures.

It was found that the seed productivity of the red clover as an individual plant is largely determined by the capacity of its development – there was a high correlation between the seed yield and the dried mass of the plant ($r = 0.72$). The plants with the largest number of stems were characterized by the highest seed yield, which was inherent in both diploid and tetraploid cultivars ($r = 0.61-0.79$ and $r = 0.55-0.83$, respectively). The seed productivity of plants depended on the number of productive flowers on the plant ($r = 0.57-0.78$ for diploids and $r = 0.76-0.89$ for tetraploids) and their mass ($r = 0.84$).

There is a lesser degree correlation between seed productivity and the number of beans in the flower ($r = 0.12-0.18$ for diploids and $r = 0.04-0.16$ for tetraploids). To a much greater extent, seed productivity depended on the number of seeds in the flowers ($r = 0.79-0.81$ for diploids and $r = 0.63-0.86$ for tetraploids). For the yield of tetraploid seeds, not only the total content of seeds in the flowers is important, but also the number of developed seeds ($r = 0.84-0.89$), since a significant part of the set seeds dies at various stages of ontogenesis and is removed during the sorting process.

Seed productivity of red clover was largely determined by seed setability ($r = 0.66-0.77$ for diploids and $r = 0.49-0.76$ for tetraploids) and inflorescence dissemination ($r = 0.71-0.80$ for diploids and $r = 0.70-0.75$ for tetraploids).

The seed productivity of red clover was associated with a mass of 1000 seeds. For diploid cultivars, this connection was mostly weak ($r = 0.24-0.32$), for tetraploids - average, which is due to larger seeds ($r = 0.39-0.45$). As for diploid cultivars, the presence of a weak correlation between seed productivity of the two-seeded beans ($r = 0.09-0.18$) and the number of stem internodes ($r = 0.14$) was found.

The presence of an average correlation of the seed productivity with the full visibility of the leaf pattern ($r = 0.30$) was also noted.

Keywords: selection process, selection, phenotypic features, correlations, seed productivity.

ФЕНОТИПИЧЕСКИЕ ПРИЗНАКИ КЛЕВЕРА ЛУГОВОГО (*TRIFOLIUM PRATENSE* L.), КОРРЕЛИРУЮЩИЕ С СЕМЕННОЙ ПРОДУКТИВНОСТЬЮ

Научная статья

Аннотация

Для сокращения времени селекционного процесса при создании сортов клевера лугового (*Trifolium pratense* L.) с высокой урожайностью семян необходимо использовать исходный материал с наличием признаков, положительно коррелирующих с повышенной семенной продуктивностью. Установлено, что в условиях северной части Центрально-Чернозёмного региона Российской Федерации семенная продуктивность клевера лугового имеет высокую сопряжённость с весом сухой массы, количеством стеблей, соцветий, семян в головках, завязываемостью семян, обсеменённостью соцветий, короткой трубочкой венчика. Средний коэффициент корреляции выявлен между урожайностью семян и длиной стебля, полной выраженностью рисунка листа, массой 1000 семян. Установлена положительная корреляционная связь, хотя и в слабой степени, между семенной продуктивностью и количеством бобов в головке, двусемянностью боба, количеством междоузлий, продолжительностью периода от начала весеннего отрастания до начала цветения.

Проведённые нами исследования позволили выявить корреляционные связи между семенной продуктивностью и хозяйственными, биологическими, морфологическими признаками клевера лугового в условиях северной части Центрально-Чернозёмного региона Российской Федерации, и Северо-Кавказском регионе, в частности в Орловской области и в Северной Осетии-Алания, где эта культура занимает лидирующее место среди возделываемых многолетних трав в биологическом земледелии и при подсева на сенокосах и пастбищах.

Установлено, что семенная продуктивность клевера лугового при индивидуальном стоянии растений в значительной степени определяется мощностью их развития – наблюдалась высокая корреляционная связь урожайности семян с сухой массой растения ($r = 0,72$). Высокой урожайностью семян характеризовались растения, имеющие наибольшее количество стеблей, что было присуще как диплоидным, так и тетраплоидным сортообразцам ($r = 0,61-0,79$ и $r = 0,55-0,83$ соответственно). Семенная продуктивность растений зависела от количества продуктивных головок на растении ($r = 0,57-0,78$ для диплоидов и $r = 0,76-0,89$ для тетраплоидов) и их массы ($r = 0,84$).

Отмечено наличие корреляционной связи между семенной продуктивностью и количеством бобов в головках, хотя и в слабой степени ($r = 0,12-0,18$ для диплоидов и $r = 0,04-0,16$ для тетраплоидов). Значительно в большей степени семенная продуктивность определялась числом семян в головках ($r = 0,79-0,81$ для диплоидов и $r = 0,63-0,86$ для тетраплоидов). Для урожайности семян тетраплоидов значение имеет не только общее содержание семян в головках, но и количество выполненных семян ($r = 0,84-0,89$), так как у них значительная часть завязавшихся семян отмирает на различных этапах онтогенеза и удаляется в виде шуплой фракции во время сортировки.

Семенная продуктивность клевера лугового в значительной степени определялась завязываемостью семян ($r = 0,66-0,77$ для диплоидов и $r = 0,49-0,76$ для тетраплоидов) и обсеменённостью соцветий ($r = 0,71-0,80$ для диплоидов и $r = 0,70-0,75$ для тетраплоидов).

Семенная продуктивность клевера лугового имела сопряжённость с массой 1000 семян. Для диплоидных сортообразцов эта связь являлась в основном слабой ($r = 0,24-0,32$), для тетраплоидов - средней в связи с более крупными семенами ($r = 0,39-0,45$). Для диплоидных сортообразцов определено наличие слабой корреляционной связи семенной продуктивности с двусемянностью боба ($r = 0,09-0,18$), количеством междоузлий стебля ($r = 0,14$).

Отмечено наличие средней связи семенной продуктивности с наличием полной выраженностью рисунка листа ($r = 0,30$).

Ключевые слова: селекция, отбор, фенотипические признаки, корреляции, семенная продуктивность.

1. Introduction

Because of the current ecologically unstable situation, there is an urgent need to restore soil fertility in biological agriculture and enrich crop rotations with valuable species of legumes, including red clover. Besides that, this crop is a bioindicator of polluted soils, high-protein animal food and a valuable raw material for the pharmaceutical industry.

To expand the crops of this important plant species, the varieties with high seed productivity are needed.

A number of scientists [1], [2], [3], [4], [5] found out that the formation of seeds in clover plants in most cases correlates with the phenotypic features: the number of generative stems, inflorescences and the number of flowers. The connection of the seed formation with pollen fertility [5], [6], seed weight [7], [8], cotyledon size [9], the length of the period of growth, the size of the root system and glucose content in the root neck [10] was revealed. During the recent years, the crop breeders have been selecting on the basis of the formation of two-seeded beans [11], [12]. However, the above mentioned method takes quite a lot time, since its results appear only in 3-4 generations. Moreover, it is unstable over the years depending on weather conditions. Numerous other researchers do not take into account the pollination factor that affects the number of seeds produced by such

an entomophilic crop as red clover. It was found that only bees with a long proboscis are able to pollinate this clover. Therefore, it is necessary to justify the selection of plants with a short corolla tube.

The researches conducted by other authors have shown that the seed productivity of red clover has different correlations with a number of its economic, biological and morphological characteristics. The values of some indicators for different authors are significantly different, which is probably due to the characteristics of the studied material, climatic conditions and the location of the experiments. Our experiments in various soil and climatic conditions allowed us to determine the phenotypic characteristics that indicate the maximum yield index and the rational ways of selection.

The purpose of the research was to study the conjugacy of seed productivity of the initial and prospective breeding material of red clover with its economic, biological and morphological characteristics in the conditions of the Northern part of the Central Black Earth Economic region of the Russian Federation and the North Caucasus region in connection with the necessity to accelerate the selection process when creating varieties with high seed yield.

2. The methods and principles of the research

The research was conducted in 1991-2017 in the Federal Research Center for Legumes and Cereal Crops in collaboration with the North Caucasus Research Institute of Mountain and Foothill Agriculture of the Vladikavkaz scientific center of the Russian Academy of Sciences in 2010-2019. The soil of at the testing site is a dark gray forest soil of medium loam composition, that is slightly acidic (pH 5.5-5.8) and there is a mountain-meadow soil in the mountainous region of the Republic of North Ossetia-Alania (altitude 1560 m above sea level). The humus content (using Tyurin's method) is 4.8-5.1 % and in the mountainous region it is 7-10 %.

During the experiments different varieties of red clover were studied (both domestic and foreign origin), as well as new varieties created by hybridization and formation of complex hybrid populations based on wild varieties and selected on the base of promising breeding samples [14], [15], [16].

During the establishment of the breeding nurseries, there were used the plant seeds with individual standing placed according to the scheme 0.7 m x 0.7 m. The repeatability number is 20-50 plants of each number. In the breeding nurseries, the growth and the development of the plants were observed. The seed yield index and morphological data were recorded by each plant's individual analysis. In the selection breeding nursery, the plants with high seed productivity with the number of internodes more than 7 (as a trait of longevity and high winter resistance) and generative stems more than 5 (as a trait of high seed productivity) were selected. The selected plants were divided into four variants at the beginning of the anthesis period. During the research of the first variant, the plants were studied without taking into account the length of the corolla tube. In the rest of the variants, the color of flowers was taken into account and the plants with a corolla tube's length of less than 7-8 mm were rejected. In the laboratory conditions, the plant capacity was estimated by weighing the entire plant. The plant's height was measured according to the the longest elongated stems and the number of stems, internodes, flowers, seed setability, dissemination of flowers, two-seeded beans, the total weight of flowers seeds (1000 seeds for each studied variant) was calculated. After threshing the flowers, the weight of the seeds was also taken into account.

Seed setability was determined by the analysis of 5 flowers of the middle tier of each plant and by the ratio calculation of all set seeds to the number of beans. The inflorescence dissemination was calculated at the ratio of developed seeds in the same flowers to the number of beans in them. Two-seeded ones were found at the ratio of the number of beans with two seeds to the total number of beans in 5 flowers of each plant.

The experimental data were processed by the correlation analysis method [17] using Microsoft Office 2007 software package.

3. The results of the research

Our research has revealed certain correlations between the seed productivity and economic, biological, and morphological characteristics of red clover in the Northern part of the Central Black Earth Economic region of the Russian Federation, in the Oryol Oblast and in the mountains of the North Caucasus in particular, where this crop occupies the first place (35-37 %) among cultivated perennial herbs in biological agriculture.

It has been estimated that the seed productivity of the red clover as an individual plant is largely determined by the capacity of its development. There was a high correlation between the productivity of the seeds and the dried mass of the plant ($r = 0.72$). The plants with the largest number of stems were characterized by the highest seed yield, which was inherent in both diploid and tetraploid cultivars ($r = 0.61-0.79$ and $r = 0.55-0.83$, respectively). The seed productivity of plants depended on the number of productive flowers on the plant ($r = 0.57-0.78$ for diploids and $r = 0.76-0.89$ for tetraploids) and their mass ($r = 0.84$). Table 1 shows the data on the correlations with phenotypic features of diploid and tetraploid forms of red clover

Table 1 – The correlation of seed productivity of red clover with its economic, biological and morphological characteristics

The name of a characteristic	Seed productivity correlation coefficient	
	diploids (2x)	tetraploids (4x)
The number of stems, pcs / plant	r = 0,61-0,79	r = 0,55-0,83
The number of productive flowers, pcs / plant	r = 0,57-0,78	r = 0,76-0,89
The number of beans in the flower, pcs	r = 0,12-0,18	r = 0,04-0,16
The total number of seeds in the flower, pcs	r = 0,79-0,81	r = 0,63-0,86
The number of developed seeds in the flower, pcs	r = 0,72-0,76	r = 0,84-0,89
Seed setability, %	r = 0,66-0,77	r = 0,49-0,76
Inflorescence dissemination, %	r = 0,71-0,80	r = 0,70-0,75
The weight of 1000 seeds, g	r = 0,24-0,32	r = 0,39-0,45
A two-seeded bean, %	r = 0,09-0,18	
Stem length, cm	r = 0,46	
The number of internodes, pcs	r = 0,14	
The weight of the dried plant mass, g	r = 0,72	
The weight of flowers, g / plant	r = 0,84	
The full visibility of leaf spotting, score	r = 0,30	
The duration of the period from the beginning of spring growth up to the beginning of anthesis period, days	r = 0,09-0,24	

There is a lesser degree correlation between seed productivity and the number of beans in the flower ($r = 0.12-0.18$ for diploids and $r = 0.04-0.16$ for tetraploids). To a much greater extent, seed productivity depended on the number of seeds in the flowers ($r = 0.79-0.81$ for diploids and $r = 0.63-0.86$ for tetraploids). For the yield of tetraploid seeds, not only the total content of seeds in the flowers is important, but also the number of developed seeds ($r = 0.84-0.89$), since a significant part of the set seeds dies at various stages of ontogenesis and is removed during the sorting process.

The seed productivity of red clover was largely dependant on the seed setability, calculated as the ratio of the sum of developed and underdeveloped seeds to the number of beans in the flower ($r = 0.66-0.77$ for diploids and $r = 0.49-0.76$ for tetraploids). The seed productivity had an even higher correlation with inflorescence dissemination, defined as the ratio of the number of developed seeds to the number of the beans in the flower ($r = 0.71-0.80$ for diploids and $r = 0.70-0.75$ for tetraploids).

The seed productivity of red clover was associated with a mass of 1000 seeds. For diploid cultivars, this connection was mostly weak ($r = 0.24-0.32$), for tetraploids - average, which is due to larger seeds ($r = 0.39-0.45$). As for diploid cultivars, there is a positive correlation between seed productivity of the two-seeded beans ($r = 0.09-0.18$) and the number of stem internodes ($r = 0.14$).

The correlation between the seed productivity with and the full visibility of the leaf pattern ($r = 0,30$) was average.

It has been estimated that in the Northern part of the Central Black Earth Economic region, particularly in the Oryol Oblast, there is a positive correlation between seed yield and the duration of the spring growth before anthesis period ($r = 0,09-0,24$), in contrast to the negative correlation coefficient between these characteristics in Moscow Oblast. This confirms the fact that the Oryol Oblast and the North Caucasus are the most favorable regions for the cultivation of not only early-maturing varieties of clover, but also medium-and late-maturing varieties, which is confirmed by the obtained seed yield data

The obtained research data on the principle of selecting plants with a shortened corolla tube and a bright flower color showed the advantage of the method and a positive correlation dependence (table 2)

Table 2 – The correlation dependence between the shortened corolla tube and the number of seeds (inflorescence dissemination, %)

The experiment's options	Light pink color	Bright pink	Bright red	Dark-red	The correlation coefficient r. =
Without corolla tube selection process-control	43.2	48.8	47.9	43.8	0.56
The selection of flowers with a long corolla tube 9-10 mm	35.6	36.2	38.1	40.2	0.58
The flowers with a corolla tube 8-8.5 mm	44.4	50.8	53.2	46.5	0.64
The flowers with a corolla tube 7-7.5 mm	68.2	72.4	73.6	68.7	0.76

As it follows from table 2 data, the combination of a bright color and a short corolla tube (7-7.5 mm) contributes to the active pollination by insects of various species and thus the correlation coefficient reaches 0,76, which is significantly higher than the coefficient with other phenotypic features.

Based on the obtained results, it can be assumed that the selection of a shortened corolla tube and a bright color of the clover plants contribute to an increase in seed yield. The correlation indicates positive relationships between phenotypic traits and the selection process based on these indicators and also creates favorable conditions for pollination of the selected plants according to other external indicators in the whole complex.

4. The conclusion

According to the conditions in the Northern part of the Central Black Earth Economic region of the Russian Federation and the North Caucasus region, when creating new varieties of red clover with high seed yield, it is necessary to use the source material characterized by developed plants with high dried mass weight, a large number of stems and many flowers. Other characteristics include: high seed setability, inflorescence dissemination and a large number of seeds in the flowers, because the combination of these features leads to a higher seed yield ($r = 0,55-0,89$). The breeding samples with a short corolla tube ($r=0,56-0,76$) have a high correlation dependence. It is also necessary to pay attention to the length of the stems, the weight of 1000 seeds, and the visibility level of the leaves pattern, which have an average correlation with seed productivity ($r = 0.30-0.45$). The authors suggest having a source material characterized by a large number of the beans in the flowers. Also there should be two-seeded beans and a large number of internodes. Mid-ripeness or late ripeness is also an important characteristic, since there is a positive correlation with seed productivity, although to a lesser degree ($r = 0.09-0.24$). The estimated correlations can be used in the selection process of red clover for increased seed productivity while selecting the source material and removing the worst breeding variants.

A lesser degree positive correlation was found between the seed yield and the length of vegetation period from the beginning of the spring growth and up to the red clover's anthesis. It demonstrates that the conditions of the Oryol Oblast and the conditions in the mountains of the North Caucasus are favourable for seed cultivating of not only early maturing but also mid and late varieties of this crop.

Conflict of Interest

None declared.

Конфликт интересов

Не указан.

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