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SELECTION FOR DISEASE-RESISTANT CLOVER

A PRELIMINARY REPORT

BY

SAMUEL M. BAIN AND SAMUEL H. ESSARY

An experiment in cooperation with the Plant Breeding Investigations of the Bureau of Plant Industry, U. S. Department of Agriculture

KNOXVILLE, TENNESSEE

The Agricultural Experiment Station

OF THE UNIVERSITY OF TENNESSEE

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SELECTION FOR DISEASE-RESISTANT CLOVER

A PRELIMINARY REPORT

A NEW CLOVER DISEASE*.

The farmers of Tennessee have for a number of years had serious difficulty in raising red clover. The trouble has been gradually increasing in severity until within recent years the crop has had to be almost entirely abandoned in many parts of the State. This condition of affairs has seriously interfered with the general agricultural practice here, for it appears that no other crop can fully replace red clover in our prevalent system of farming.

In 1905 the writers began an investigation of the question, and early in the season found a new and hitherto undescribed fungous disease belonging to a class generally known as anthracnose. This fungus belongs to the genus known to botanists as *Colletotrichum*, and has been given the scientific name *Colletotrichum trifolii*. The Colletotrichums are known to be the cause of a number of other serious plant diseases, whose general behavior is quite similar to the clover anthracnose here considered. Several other diseases were found on red clover here, but none of them appears to do injury to the crop at all comparable to this new anthracnose. The disease was found in almost every clover field visited, and seems to exist in its severest form in the oldest and best farming sections of Tennessee. While beyond a doubt other clover diseases than those thus far found will eventually come to light in the State, and may prove to be of considerable importance, there can now remain little doubt that the one here discussed is far more responsible for the failure of the clover crop than any other disease or soil condition.

While it is not the purpose here to discuss conditions elsewhere, it is nevertheless of interest to know that the same disease has been found also in West Virginia, Kentucky and Arkansas. It is impossible to state at present anything very definite as to its geographical extent. It occurs also on alfalfa, having been found to cause considerable injury to that plant at one point in Virginia, and here at the Experiment Station farm. Its occurrence in these widely separated localities makes it at least probable that the disease is widely disseminated.

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*An announcement of the discovery of the disease here under consideration was made in *Science*, Vol. 17, p. 503, 1905. A technical description of the fungus (*Colletotrichum trifolii*) causing it occurs in the *Journal of Mycology*, Vol. 12, p. 192, 1906. A detailed account of studies on the life history of this fungus will appear in a future bulletin of this Station.

FIG. 1.—PLANT OF RED CLOVER, 48 INCHES HIGH, IN FIELD NEWLY CLEARED

A good crop of clover had perished on this field during the previous summer. This shows that failure of clover crop is not due to lack of fertility of the soil.

EXPERIMENTS LOOKING TOWARD A REMEDY

As soon as it was apparent that the disease had been definitely located the search was begun for a possible remedy. A thorough field survey had shown that clover dies on every variety of soil existing in this State, and under almost every conceivable method of cultivation. While the disease is usually more severe on fields where clover has been grown for a long time, this is by no means always the case. In Fig. 1 is shown a field cleared for only three years and sown to clover for the first time in the spring of 1905, where an excellent stand died out almost entirely during the following summer. The stumps of trees were still abundant, weeds and stubble showed evidence of fertility, and the few surviving clover plants had made a luxuriant growth, one plant, as shown in the figure, measuring four feet in height.

Spraying methods could hardly be considered in the case of a crop like clover. While there can be little doubt that the disease is disseminated partly through clover seed itself, and that treatment of the seed with some disinfecting solution would tend to keep it out of clean land, yet this could hardly avail much with a plant so universally abundant along roadsides and fence-rows, from which the disease might reach an adjoining field at any time. Since, therefore, neither methods of cultivation, spraying, nor disinfecting seed promised much, all available time and resources at hand were directed toward the production of a variety of resistant clover by selection. The first year's test of this select seed has given such striking and encouraging results that it was decided to publish a preliminary report on the subject.

METHOD OF MAKING SELECTIONS

On going into badly infected fields in August and September, at which time most of the clover has died, healthy plants may be found here and there, which have resisted the disease and produced a good crop of seed. Often the disease has destroyed the crop only in patches. In every selection the effort was made to get the isolated healthy plants that were even partly covered by dead ones. In this way there was a greater probability of securing genuine resistance than if one plant had been chosen out of a group of healthy ones. Several weeks were thus spent in the field in August and September, 1905. Over two hundred selections were made, and the seed of individual plants kept separate.

ARRANGEMENT OF THE EXPERIMENTAL PLOT

For the experimental plot was selected an area of land on the Experiment Station farm where the disease existed in 1905. While there was every reason to suspect an abundance of the germs of the disease on this plot, precautions were taken to insure its presence during the season of 1906. In the first place, the ground was not turned, but merely disked thoroughly in the spring of 1906. It was thought that deep plowing might cover up many of the germs of the disease, so that they would not reach the surface as readily as with shallow cultivation. In February

a load of dead clover was raked up from another field and stored in the barn (see Fig. 2). This field of clover, as was stated above (p. 5), was almost totally destroyed in 1905. After the clover in the experimental plot had been up for some weeks (five), this dead clover was scattered over it, so that scarcely a single plant in the whole plot was out of contact with dead clover stems. To insure still further that every clover plant had an equal opportunity to contract the disease, the dead and dying plants from the non-select rows were scattered over the selections on July 15. Therefore, there can scarcely be any doubt that any superiority shown by the plants from select seed is due solely to some inherent resistance to the disease in question. The clover seed was planted on March

FIG. 2.—GATHERING DEAD CLOVER IN SAME FIELD AS SHOWN IN FIG. 1.

There was in many parts of this field a perfect mat of clover plants that had died the previous summer. This dead clover was stored away and later scattered over the plants in the experimental plot so as to insure the presence of the disease among the selections.

10-13, 1906, in rows 18 inches apart. Each selection was duly labeled and its location recorded. Alternating with each select row was sown a row of commercial clover seed purchased in the Knoxville market. The section of country where this commercial seed was obtained could not be learned, but the sample was quite clean and fully up to the standard in purity and germinating power.

The seed of all varieties began germinating by March 20, and all went well with the experiment until April 7, when the greater part of

the plot was suddenly attacked by large numbers of larvae, identified by Mr. Bentley, assistant entomologist in the Station, as the clover weevil (*Phytonomus punctatus*).

In spite of every method resorted to for checking the ravages of this insect, it destroyed the greater part of the clover in the plot, leaving only about fifteen rows at one end practically undisturbed. There were so few plants left over a large portion of the plot that the few selections remaining there were transplanted into rows adjacent to the uninjured ones at the other end of the plot between June 5 and 20.

This portion of the plot was cultivated frequently by hand and wheel-

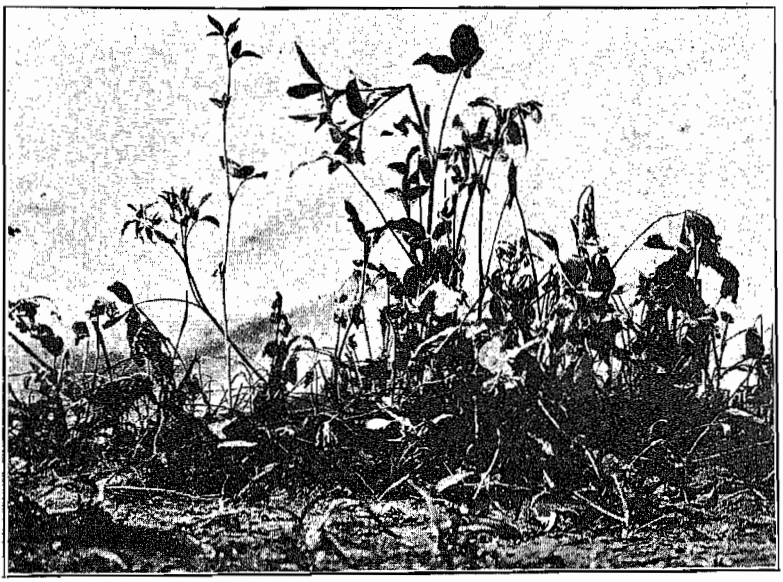

FIG. 3.—A PORTION OF A NON-SELECT ROW OF CLOVER PLANTS IN THE EXPERIMENTAL PLOT.

This photograph was taken July 9. It shows strikingly the effect of the anthracnose on ordinary clover. A few weeks later only an occasional plant in these non-select rows was alive.

hoeing, and all the plants, select and non-select, were growing vigorously until June 1, when the non-select rows began to show evident signs of disease. By June 20 the rows from commercial seed were blackened and dying, while the rows of selections alternating with them were almost perfectly healthy and normal. Several efforts were made to secure photographs of the plot so as to show the contrast between these alternating select and non-select rows, but on account of the close proximity of the rows none of these views were satisfactory, although the contrast between adjacent select and non-select plants was striking on the most casual inspection. Fig. 3 shows some plants from one of the non-select rows, taken on July 9. This may be taken as fairly representative of all

portions of the non-select rows. This shows a striking contrast with Fig. 4, which gives a similar view of an adjacent row of selections taken on Sept. 14. It should also be explained that the entire plot had been mown off in the interval between July 9 and Sept. 14, so as to simulate actual cultural conditions. By this date but few of the non-select plants were living. A very conservative estimate would place the percentage of selections which lived up to Sept. 15 at 95; while not more than 5 per cent of the non-select plants survived. Those few were probably

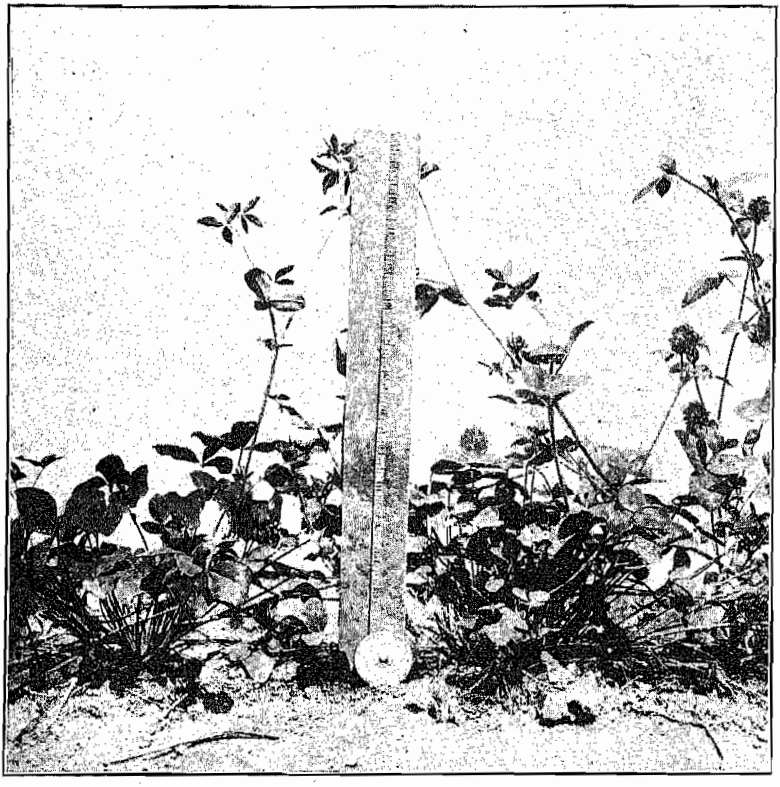

FIG. 4.—A PORTION OF A SELECT ROW OF CLOVER PLANTS IN THE EXPERIMENTAL PLOT.

This photograph was taken Sept. 14, over two months after most of the plants in the adjacent row were dead, as shown in Fig. 3. These plants had been mown once in the interval.

resistant. While these selections nearly all made a fair average growth, it should be stated that many of them showed some of the lesions of the anthracnose at that date (Sept. 15). But in nearly every case the stock was in perfect health at that time, and there was a good crop of stems, leaves and seed. In many instances the plants were exceptionally vigorous and without a trace of the anthracnose. Just before going to press

an examination of the experimental plot showed that a number of plants of both select and non-select parentage had died during the winter. It was then evident that the selections were on the whole decidedly more healthy in appearance. A conservative estimate places the odds in favor of the selections at this date at 50 to 1. This is based on the percentage of surviving plants of each group to the approximate number of that group which germinated.

Attention has been called to the fact that alsike clover is not attacked by this anthracnose. In only one case has the disease been found on alsike, and this was a seedling growing in the greenhouse.

While numerous experiments have been made in working out the life history of the fungus causing this disease, which it is not the purpose to present here, one experiment may be described as bearing on the subject.

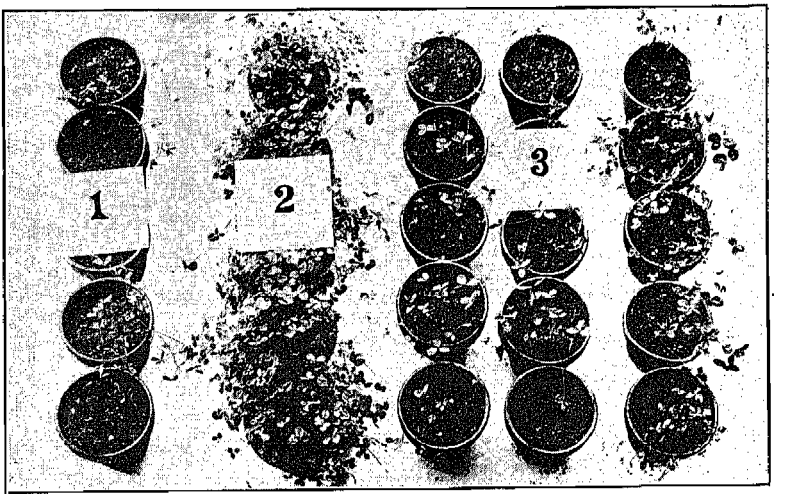

FIG. 5.—THE EFFECT OF THE ANTHRACNOSE ON ALFALFA (1), ALSIKE CLOVER (2), AND RED CLOVER (3).

These pots were planted under exactly similar conditions, on the same date. Nearly all alfalfa and red clover plants are dead, while the alsike plants have resisted the disease and made vigorous growth. Seed were planted July 12; disease made its appearance July 22; photographed Sept. 12.

On July 12 a number of flower pots were seeded with red clover, alsike clover, and alfalfa, with a view to making inoculation tests with spores of *Colletotrichum*. The seeds were thickly sown in each pot and all the plants made a vigorous growth for some days, until accidental infection (July 22) occurred and the red clover and alfalfa seedlings began to die rapidly of anthracnose. The pots were all closely adjacent on the same bench, and were left for some weeks in this position. They were similarly watered at intervals, but not otherwise interfered with, except that the row of alsike pots was placed in contact on both sides with pots of red clover. This entire series of pots is shown in Fig. 5.

Group No. 1 is alfalfa, No. 2 is alsike, No. 3 is red clover. The contrast between the alsike and the other pots is due to its perfect immunity to the anthracnose, which, as shown, has destroyed most of the plants of red clover and alfalfa. The few surviving plants of red clover and alfalfa were doubtless resistant to the disease.

CONCLUSIONS

The most important result of the above-described experiments is the striking indication of disease resistance shown by select individual red clover plants. Since every opportunity existed for the infection of the plants from select seed, and they were subjected to the most rigid test for susceptibility to disease, there is very strong indication, at least, that success will attend the effort being made to produce an anthracnose-resistant clover. The evidence may be briefly stated thus: Certain clover plants growing in 1905, surrounded by great numbers of other plants killed by anthracnose, produced offspring which, in turn, also resisted the disease alongside non-select plants which were killed by the disease. Of course it remains to be seen whether the next and subsequent generations will likewise resist the disease. This matter can only be determined by future experiments.

It is necessary to emphasize that no clover seed is as yet available for distribution. The small quantity of seed now on hand must be planted under proper conditions for continuing the tests. While plants will be propagated as rapidly as practicable, even with the most sanguine expectations realized, it will perhaps require several years before seed are ready for distribution.

SUMMARY

1. The red clover crop of this State has been very uncertain for a number of years.

2. The failure of the crop is due in the great majority of instances to a new fungous disease caused by *Colletotrichum trifolii*.

3. This disease belongs to a class known as anthracnose, whose general character is well known to plant pathologists.

4. So far as known no cultural methods of handling the clover crop will prevent or even appreciably diminish the ravages of this disease, and it appears to exist on every kind of soil in Tennessee.

5. The same disease also attacks alfalfa, but to what extent in this State is not yet known.

6. Alsike clover is almost absolutely immune to this disease.

7. Occasional healthy plants of red clover in badly stricken fields in different parts of Tennessee have produced in the second generation plants which were strikingly resistant to the disease. Whether this resistance will be maintained to future generations can not be foretold with certainty.

8. While the effort is being made to propagate resistant plants rapidly and to secure a supply of seed from such plants, it must be distinctly understood that no such seed are as yet ready for distribution, either by the Tennessee Experiment Station or the U. S. Department of Agriculture.