THE EFFECT OF CERTAIN DISEASES AND INSECTS ON SEED SETTING IN ALFALFA

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INTRODUCTION

A problem of great importance to the alfalfa growing industry in Canada and the U.S.A. is the production of alfalfa seed. Production is limited to certain areas in the Western U.S.A. and the three Prairie Provinces of Canada. Scarcity of alfalfa seed and the consequent high price have limited the forage acreage of alfalfa, (9),(35).

Genetic factors are known to affect the seed yield of individual alfalfa plants. Variations in seed yields from year to year have been attributed to such factors as weather, insects and disease. The investigations reported here were begun in 1949 to provide additional information concerning the factors affecting seed production in alfalfa.

REVIEW OF LITERATURE

Factors affecting seed setting in alfalfa have been a subject of investigation ever since the crop attained economic importance. Increasing attention in recent years has been prompted by the continual decrease in alfalfa seed yields in the major seed producing areas. Genetic and environmental factors have been dealt with extensively. Numerous investigations have been concerned with insects (beneficial and injurious) and diseases.

Genetic and environmental factors

Kirk (17) found a pronounced and progressive reduction in vigor of the alfalfa plants with each generation of selfing. The mean yield of seed for second generation selfed lines showed a marked decrease as compared with the seed yield of the corresponding open-pollinated strains. He indicated that controlled pollination with selection in self-fertilized lines which did not show decreases in yield may be utilized in a systematic breeding program.

Clarke and Fryer (6) found that many plants had a hereditary trait of producing a high percentage of sterile pollen under varied environmental conditions. Their investigations indicated that plants had to be tripped to set seed.

Bolton and Fryer (2) found that number of flowers per raceme and number of seeds per pod were heritable characters which affeeted seed yield. High temperatures were found to promote greater pollen growth and better seed set.

Grandfield (14) found that moderate air temperatures, low humidity and low soil moisture were conducive to the storage of high organic reserves, resulting in physiological conditions favourable to seed setting.

Tysdal (33) mentions that high soil moisture is not an inhibiting factor in seed production, unless lodging results.

Jones and Olson (16) found that cross pollination of hand tripped flowers markedly increased seed setting.

Silversides and Olson (26) showed that the mechanical devices they used for tripping alfalfa flowers were not effective in increasing seed set. The increased tripping of the alfalfa blossoms by these devices was offset by the damage caused to the plants.

Southworth (27) outlined a method of producing a high seed setting alfalfa which was both self-tripping and highly fertile. He crossed Medicago sativa with M. lupulina, a self-fertile seed setter. The variety Macsel originated from this cross.

Fryer (12) outlined a method of breeding for increased seed setting by maternal line selection involving a four-year cycle. Plants were scored in the second and third year for desirable vegetative characters and density of seed pods. Plants with a poor score were discarded. The seed obtained in the fourth year was used for another cycle. The variety Ferax was developed by this method.

Tysdal et al. (31) reviewing the literature on the different methods of alfalfa breeding stated that the principles of breeding alfalfa, with some modifications were essentially the same as those which have been established for corn.

Tysdal and Kiesselbach (32) suggested the use of selected

clones of high combining ability and high self-sterility for the production of hybrid alfalfa. The best double cross seed would be made available for commercial utilization. Natural intercrossing of selected clones would result in the formation of synthetic varieties.

Stevenson and Bolton (28) stated that self-tripping alfalfa plants have low forage yields and should not be used in a breeding program.

Bolton (1) indicated that there is a wide range in the combining ability of different alfalfa plants. He outlined a method of alfalfa breeding using disease resistant stock. Self-tripping plants would be discarded. The remaining plants would be rated for self- and cross-fertility. Progeny of both selfed and crossed seed would be grown, and in the selfed lines, plants with undesirable genes would be eliminated. Combining ability of each plant is determined from the progeny of the polycross seed. Progenies grown from the seed of plants crossed in all possible combinations would be used for the final selection.

Tysdal and Crandall (34) outlined a slightly different method. Clones selected for such desirable characters as high seed yield, high forage yield, bacterial wilt resistance, cold resistance and potato leaf hopper resistance, are allowed to intercross at random, polycross seed being obtained. Progenies are then tested for low self-fertility, high seed yield and high forage productivity.

Beneficial and injurious insects

Tysdal (30) mentions that Megachile and Nomia bees are the most efficient trippers of alfalfa, while honey bees are rather poor trippers. Weather conditions and insect visitations determine the seed set of alfalfa.

Lejeune and Olson (20) noted that bumble bees were efficient pollinators of alfalfa, tripping most of the blossoms visited. Honey bees, however tripped very few flowers.

Knowles (18) found a correlation between the presence and number of Megachile and alfalfa seed set. He states that temperature was found to be the most important factor influencing insect activity and tripping of alfalfa blossoms.

Bolton and Peck (2) indicated that, in Northern Saskatchewan, burning of alfalfa fields on which appreciable amounts of lygus bugs were present increased the seed yield.

Hare and Vansell (15) stated that honey bees are important trippers in alfalfa seed fields in the Delta tract of Utah.

Vansell and Todd (36) noted that honey bees would work alfalfa for pollen if competing sources (clovers, mustard, thistles) were absent. Otherwise they were more apt to collect only nectar from the alfalfa.

Carlson (4) found that the use of insecticides increased alfalfa seed yields where lygus bugs were a factor. He obtained a yield of 175 pounds per acre from treated plots compared to 66 pounds per acre from untreated plots. He found that genotypic differences were related to alfalfa seed production. Yields of clonal lines ranged from 37 to 441 pounds per acre.

Drake (8) noted that wild bees were more efficient pollinators of alfalfa than are honey bees. However honey bees were more numerous. He found that the efficiency of pollinating insects was doubled and sometimes trebled by improved cultural practices. Highest yields were obtained where the cutting of the first crop of hay was staggered at 8 to 10 day intervals

to extend the blooming period. A ten percent DDT dust was applied to kill injurious insects.

Wilson (37) found that one pound of DDT and eight ounces of chlordane per acre gave very good control of lygus bugs. Significant increases in yields of alfalfa seed were found in the insecticide treated plots. The insecticide did not affect the population of the pollinators.

McMahon (22) states excellent control of the common injurious insects of alfalfa is obtained in western Canada by the application of three quarters of a pound of DDT per acre, before flowering becomes general.

Diseases

McDonald (21) stated that bacterial wilt is found in Manitoba but so far has not resulted in significant crop losses. Pseudopeziza medicaginis is not very important. Black stem is prevalent in the Province especially in the seed producing areas.

Cherewick (5) reports that crown rot, a strain of Rhizoctonia solani, which attacks both alfalfa and sweet clover, inflicts a heavy loss on alfalfa in Manitoba. He further states that blossom drop in alfalfa, prevalent in Manitoba and frequently resulting in a complete failure of seed set, may be caused by a species of Alternaria.

Peterson and Melchers (24) found that Black stem causes destructive defoliation and discoloration of hay crops. The disease was found to be most severe during cool moist weather. Their investigations showed that the optimum temperature for growth of the pathogen in culture was 21 C with a range of 9 C to 30 C.Of 4 Medicago species tested, (M. sativa, M. lupulina, M. falcata and M. ruthenica) M. falcata was found to be the most resistant to Blackstem.

Koepper (19) tested several varieties and species of alfalfa

for resistance to Uromyces striatus. A plant selection of Ladak which was resistant to rust also exhibited high resistance to Blackstem.

Cormack (7) in studies on Ascochyta imperfecta Peck, stated that the pathogen was both seed and soil borne. Cool moist conditions were most favorable for disease development. New improved Ceresan and Arasan were most effective in seed treatment of alfalfa. The fungus in the soil disappeared two years after the soil was plowed. No blackstem was found in the soil of virgin prairies, virgin woods or fields with cereal crop rotations. The fungus persisted on dry stems and leaves for 5 years and on alfalfa seed for three years. He found that it lowered the forage yield. He reported cases where seed yields may have been decreased because of Black stem.

Reitz, (25) tested 10 varieties and species in both field and greenhouse for resistance to Black stem. Medicago ruthenica was found to be the most resistant. The commercial varieties, Ladak and Grimm were third and fourth in order of resistance.

Stevenson (28) stated that immunity to Black stem has not been found and it seemed probable that a high degree of resistance may be difficult to obtain.

Ellis (10) in preliminary spray tests with chemicals to control Ascochyta abelmoschi, Harter, on a variety of okra (Hibiscus esculentis L.) found that Fermate gave higher yields of marketable pods and lower defoliation ratings than Spergon, Lime sulfur, tribasic copper sulfate and Bordeaux mixture. However, even on the Fermate treated plots the disease was severe and control was not considered satisfactory.

Mitchell et al. (23) experimenting with sulfanilamide derivatives to eradicate leaf rust, found that sulfadiazine as a spray was the most effective. The minimum effective concentrations was 0.1 percent sulfadiazine. A concentration of 0.4 percent sulfadiazine was toxic to the wheat plants.

MATERIALS AND METHODS

1949

Various experiments were conducted, and observations made concerning factors affecting seed setting in alfalfa. Twenty-three lines were tested for seed and forage yield. A caging experiment was conducted in which bees were used as pollinators. The effect of Black stem on seed yield and total plant weight was determined. Observations were made on the numbers and efficiency of pollinating insects present and on the extent of damage caused by injurious insects.

Testing of lines for seed and forage yield

The lines used for this study originated from a nursery established in 1945 consisting of 25 lines each of 25 plants. Plants were rated for seed set in 1946 and 1947. In 1947 twenty-three superior seed yielding plants were selected. Two racemes of each of these plants were self-pollinated. Both open-pollinated and self-pollinated seed was harvested from these plants.

An individual plant nursery was established from this seed in 1948. The nursery was a split-plot randomized design with four replicates. Three plants of open-pollinated origin and three plants of self-pollinated origin of each line formed the main plots of each replicate. Spacing of plants was three feet by three feet.

Plants were harvested individually at maturity and total dry weight per plant taken. Plants were threshed and weight of clean seed was recorded.

Caging experiment with honey bees

Determination of the effectiveness of honey bees in pollination of alfalfa was one of the objectives of this experiment. Evaluation of lines as to selective pollination by honey bees was to be considered. For this purpose a cage, constructed of hardware cloth over a wooden frame, was placed across one replicate in such a position that it enclosed one open-pollinated and one self-pollinated progeny plant of each line. The size of the cage was 72 feet by 6 feet by 6 feet. This cage also provided space for a bee hive at each end.

Visitation by pollinators began about 7 days after the first sign of alfalfa bloom in the nursery. On this date (June 18) two colonies of bees were placed inside the cage. Each colony was provided with two entrances one to the outside and one to the inside of the cage.

One of the colonies dwindled and was removed on August 11. The second colony was normal and was retained till September 16, at which time harvesting operations commenced.

Effect of Black stem on seed yield and total plant weight

Black stem symptoms were first noted on some plants in the nursery at the beginning of June. The disease was prevalent throughout the nursery by the end of July. Individual plants were rated for amount of disease on a percentage basis. Plants were rated for Black stem on both stems and leaves on August 14-16 and on stems alone on September 7.

Two hundred plants of open-pollinated origin were used to determine correlation coefficients to show the effect of Black stem on seed yield and total plant weight. In calculating the correlation coefficients the percentage ratings of the amount of disease on each plant were transformed to degrees. ie. percentage (p) = $\sin^2 \phi$. (Fisher 11).

Pollinating insects

Observations were made on the relative populations and effectiveness of the insect pollinators in the nursery.

Injurious insects

Visual observations were made on the relative number of the different kinds of injurious insects. To evaluate the damage caused by clover seed chalcid, four racemes, from each of six plants taken at random were collected on August 18, and counts of damaged and undamaged seed were taken.

Analysis of data

Correlation coefficients and analysis of variance were determined according to Goulden (13), and transformation of frequencies according to Fisher (11).

1950

Experiments in 1950 dealt mainly with the use of fungicides in the control of Black stem. An experiment to determine the effect of DDT on insects and seed set was conducted. Observations were made on pollinating insects and their habitat.

Effect of fungicides on Black stem and seed yield

Three fungicide dusts were tested as to their effectiveness in controlling Black stem and increasing seed yield. Their active ingredients and composition are as follows:

Trade name	Percentage of Active ingfedients	Composition of active ingredients
Fermate	10	Ferric dimethyl dithio carbamate
Trox	7	Tri-basic copper sulphate
Mulsoid sulfur	95	sulfur

The Red River flood of 1950 necessitated the transfer of the experiment from the University of Manitoba field plots to Teulon, Manitoba, 45 miles N.N.W. of Winnipeg. A 4 acre field of a 3 year old stand of Grimm alfalfa, which had yielded 200 pounds of seed per acre in 1949, was selected. The effect of additional inoculum in the form of old alfalfa straw on the incidence of Black stem was also tested. A split-plot design was used with fungicide treatments comprising the main plots. Additional inoculum (10 pounds of alfalfa straw) was spread over one-half of each main plot. The experiment was replicated 4 times and occupied an area of 2 acres.

Heavy dew every night and occasional rains made weekly application of the fungicides necessary. Applications commenced on June 30th and ended August 16. Trox and sulfur were applied at the rate of 30 pounds and 10 pounds per acre respectively. The first application of Fermate at the rate of 30 pounds per acre resulted in slight burning of the leaves so the rate was subsequently reduced to 20 pounds per acre.

Samples of plants from the different treatments were taken on July 15, July 27, August 15, August 24, and September 6.

A 4 square rod sample of each sub-plot was harvested at maturity. The alfalfa was threshed and weight of cleaned seed recorded.

Effect of fungicides on insect populations

Ten sweeps with a 12 inch insect net were made on each treatment in all replicates on the following dates: July 3, July 5, July 25, August 2, August 10 and August 24. Counts were taken on weevils, alfalfa plant bugs, aphids and pollinators.

Effect of DDT on insect populations and alfalfa seed yield

To test the effect of DDT on insect populations and seed yield of alfalfa a simple randomized block design with 3 replications of 4 square rods each, was set up. One half of each replicate was left as a check.

The other half of each replicate was sprayed on July 5 with a DDT solution (2 pounds of 50% wettable DDT powder to 100 gallons of water) at the rate of three-quarters of a pound of DDT per acre. Five percent of the alfalfa was in bloom, but there was very little pollinating insect activity at this date.

Ten sweeps of each of the six plots were made with a 12 inch insect net on the following dates: July 5, July 7, July 10, July 14, July 25, August 2, August 10 and August 24. Counts were made of weevils, alfalfa plant bugs, aphids and pollinators. The weight of cleaned seed of each plot was recorded.

Study of wild bees

An attempt was made to find the nesting places of the wild bees that pollinate alfalfa. Observations were made on their activities in relation to the environment.

Greenhouse tests on susceptibility of certain alfalfa varieties to Black stem

One plant of Medicago falcata and two plants each of the varieties, Du Puits, Ferax, Ladak, Grimm and Wisconsin Synthetic A, were sprayed with a suspension of Black stem pycnidiospores obtained from old alfalfa straw. The sprayed plants were placed in a moisture chamber for three days. Comparisons of susceptibility were made two weeks after inoculation.

Effect of certain fungicides on the germination of Black stem spores and growth of mycelium on potato-dextrose agar medium

1. Fungicides: Streak cultures of pycnidiospores of Black stem were made on potato dextrose agar medium which contained the following percentages of active ingredients of different fungicides: check; 0.1% and 1.0% of Trox and Fermate: 0.5% and 5% of sulfur. Plates were incubated

at room temperature of 70 degrees Fahrenheit for seven days after which the petri plates were photographed, and comparisons made.

2. Sulfadiazine: Cultures of pycnidiospores and surface sterilized infected leaves were grown in potato dextrose agar medium, which contained the following percentages of sulfadiazine 0.0, 0.05, 0.1, 0.5, 1.0, and 5.0. The petri plates were incubated at a temperature of 70 degrees Fahrenheit for seven days. Observations of germination of spores and growth of mycelium were recorded at the end of this period.

Effect of Sulfadiazine on control of Black stem on alfalfa in the greenhouse.

One test was made to determine the effect of single doses of sulfadiazine at varying concentrations. The test consisted of 2 replicates with 7 treatments. Fourteen plants of a Grimm clone and 14 plants of a Du Puit clone were used. The concentrations used were: 0.0, 0.1, 0.2, 0.4, 0.5, 1.0, and 2.0 percent. Each plant was sprayed with 15 c.c. of one of the concentrations of sulfadiazine. The plants were sprayed 24 hours later with a suspension of Ascochyta imperfecta spores and placed in a moisture chamber for 3 days. Two weeks after inoculation observations on the control of Black stem disease and toxic effects on the plants were recorded.

The second test was made to determine the effect of a varying number of doses of a non-toxic concentration of sulfadiazine. The experiment consisted of 2 replicates of 4 treatments, using 8 plants of a Du Puit clone and 8 plants of a Grimm clone.

Treatments were 0, 1, 2, and 3 doses of 0.4% sulfadiazine with 6 day intervals between each dose. Twenty four hours after the last application of sulfadiazine, each plant was inoculated with a spore suspension of Ascochyta imperfecta and placed in a moisture chamber for three days. Observations were recorded two weeks after inoculation.