

EFFECTS OF TEMPERATURE AND HUMIDITY ON THE BIOLOGY OF  
THE SPOTTED ALFALFA APHID, TERIOAPHIS MACULATA  
(DUCKTON), AND THE PEA APHID, MACROSIPHUM PISI  
(HARRIS), FEEDING ON SELECTED ALFALFA CLONES

by

ALBERT ISAAC

A. B., Tabor College, 1959

---

A THESIS

submitted in partial fulfillment of the

requirements for the degree

MASTER OF SCIENCE

Department of Entomology

KANSAS STATE UNIVERSITY  
Manhattan, Kansas

1961

LD  
2668  
T4  
1961  
ITB  
C.2

11

# TABLE OF CONTENTS

INTRODUCTION . . . . .	1
LITERATURE REVIEW . . . . .	1
Spotted Alfalfa Aphid . . . . .	1
Pea Aphid . . . . .	6
MATERIALS AND METHODS . . . . .	10
RESULTS AND DISCUSSION . . . . .	23
Spotted Alfalfa Aphid . . . . .	23
Biology of the Spotted Alfalfa Aphid as Influenced by Humidity . . . . .	23
Biology of the Spotted Alfalfa Aphid as Influenced by Temperature . . . . .	36
Biology of the Spotted Alfalfa Aphid as Influenced by Temperature and Humidity . . . . .	33
Pea Aphid . . . . .	45
Biology of the Pea Aphid as Influenced by Humidity . . . . .	53
Biology of the Pea Aphid as Influenced by Temperature . . . . .	53
Biology of the Pea Aphid as In fluenced by Temperature and Humidity . . . . .	55
Comparison of Effects of Feeding on Excised and Attached Trifoliolates . . . . .	64
Biology of the Spotted Alfalfa Aphid as Influenced by Excised and Attached Trifoliolates . . . . .	65
Biology of the Pea Aphid as Influenced by Excised and Attached Trifoliolates . . . . .	76
SUMMARY . . . . .	77
ACKNOWLEDGMENT . . . . .	81
LITERATURE CITED . . . . .	82

## INTRODUCTION

The purpose of this investigation was to compare the influence of constant temperature and humidity on the biology of the spotted alfalfa aphid, Therioaphis maculata (Buckton), and the pea aphid, Macrosiphum pisi (Harris), on three selected alfalfa clones. The biology of both aphids was also studied on excised and attached trifoliolates of these three clones at three constant temperatures. These experiments were conducted to determine if aphid reproduction was influenced by excised and attached trifoliolates at a given temperature.

## LITERATURE REVIEW

### Spotted Alfalfa Aphid

The original description of the spotted alfalfa aphid, Therioaphis maculata, was made by Buckton in 1899 (Bodenheimer and Swirski, 1957). Its first appearance in the United States caused considerable confusion as to its identity. It was thought that the yellow clover aphid, Therioaphis trifolii (Monell), had adapted itself to a new host, and at first this name was used (Keynolds and Anderson, 1955, and Dickson et al., 1955). The common name, spotted alfalfa aphid, was proposed by Dickson et al. (1955) to distinguish it from the yellow clover aphid, when it was shown to be a different species.

The native home of the spotted alfalfa aphid was apparently central Asia and southern Eurasia (Bodenheimer and Swirski, 1957). It was probably the same species that had been reported on

alfalfa in Europe, India, and the Mediterranean area (Dickson et al., 1955). Its presence in the United States was first discovered in New Mexico in the fall of 1953 (Dobson and Watts, 1957), but it was not recognized as a new pest until the spring of 1954 (Dobson and Watts, 1957, and Stern and Reynolds, 1957). Dickson et al. (1955) reported that it was first collected in California in early February 1954, and about the same time in Arizona (Tuttle, 1955). In Kansas the aphid was first noted in the southwest corner in August, 1954, by L. J. DePew (Harvey and Hackerott, 1956), and by October 1955, it had spread to all areas of the state.

Although the most important host plant of the spotted alfalfa aphid from an economic standpoint is alfalfa, several other species of leguminous plants are also favorable hosts. Peters and Painter (1957) found 23 species, in the genera Medicago, Melilotus, Trifolium, and Trigonella, to be favorable hosts for its reproduction and development. Bodenheimer and Swirski (1957) reported that this aphid feeds on alfalfa in Israel, Iraq, and Turkey, and on berseem clover, Trifolium alexandrinum L., in Egypt and Israel.

The bionomics of the spotted alfalfa aphid has been studied by Harpaz (1955) in Israel, Dickson et al. (1955) in California, and Nielson and Barnes (1957) in Arizona. Simpson and Burkhardt (1960) found that this aphid could survive during the winter months from the southern to the northern boundary of eastern Kansas. Diehl and Chatters (1956) showed that the spotted alfalfa aphid inserts its stylets intercellu-

larly and feeds within the phloem and mesophyl parenchyma. Considerable quantities of saliva or toxic substances are injected into the tissues with the most evident damage appearing in the mesophyl. Nickel and Sylvester (1959) found that the percentage of plants showing yellow-banding symptoms increased with an increase in the length of feeding time of individual aphids. The systemic symptoms were produced sooner by third and fourth instar than by first and second instar nymphs. Variation in plant susceptibility was the primary cause of the differences in symptom incidence. Paschke and Sylvester (1957), and Diehl and Chatters (1956) presented evidence indicating that the vein-banding was actually an insect-induced plant toxemia, and not due to a virus. McMurtry and Stanford (1960) observed that the setal tubes entered the phloem less frequently in resistant than in susceptible plants.

The alfalfa variety, Lahontan, was one of the first observed to be highly resistant to the spotted alfalfa aphid (Dobson and Watts, 1957, Howe and Smith, 1957). A number of resistant alfalfa varieties were subsequently developed by entomologists and agronomists in several states. Cody, a variety of alfalfa resistant to the spotted alfalfa aphid, was developed in Kansas from the combination of 22 selected Buffalo clones (Harvey et al., 1960, and Sorenson et al., 1961).

Howe and Smith (1957) observed that spotted alfalfa aphid nymphs reproduced on leaves of resistant plants were restless and tended to scatter over the leaflets, while those reproduced on susceptible plants often remained near the mother. Nymphs



placed on leaflets of resistant plants frequently preferred to feed on the midrib, petiole, or stem rather than on the leaf tissues. McMurtry and Stanford (1960) concluded that mortality of the spotted alfalfa aphid on highly resistant plants was caused by starvation or desiccation resulting from a failure to ingest a sufficient quantity of plant sap. Maxwell and Painter (1959) showed that honeydew deposition was reduced when spotted alfalfa aphids fed on resistant plants.

Paschke (1959) found that the primary factor responsible for production of alatae was population density. His experiments indicated that the first instar was the stage in which determination of wing production occurred.

There have been indications that resistance in alfalfa plants to the spotted alfalfa aphid may be less effective at low than at high temperatures. Harvey and Hackett (1956) reported a low rate of aphid survival at an average temperature of 75° F. on resistant compared with susceptible plants selected from Buffalo alfalfa. However, subsequent tests at lower temperatures showed increased survival of aphids on resistant plants. Howe and Smith (1957) observed that spotted alfalfa aphid nymphs were unable to survive on resistant plants in the greenhouse, but were able to complete development on the same plants in the field during cool fall temperatures. The effects of seasonal temperatures on the aphid's reaction on susceptible plants have been reported by Dickson et al. (1955) and Harpaz (1955). They found that development and reproduction were reduced at lower temperatures.

Using selections from the susceptible variety, Buffalo, Hackerott and Harvey (1959) reported that a resistant plant on which an aphid population could not be maintained at 80° would support a limited population at 60° F. The optimum temperature for the reproductive period, total reproduction, and numbers of nymphs which matured and reproduced decreased with an increase in plant resistance. The time required for nymphs to mature and reproduce was longer at 60° than at 80° F. and appeared to increase in length with an increase in plant resistance.

Graham (1959) was apparently the first to study the effects of constant temperature and humidity on the biology of the spotted alfalfa aphid. He found that the optimum temperature for nymphal development was 30° C. (86° F.), under which conditions the aphids were able to complete their development at 5.5 to 6.5 days. He also noted that the duration of reproduction and longevity varied inversely with temperature, and that the low humidity gave a longer reproductive period than the higher humidity levels at all temperatures studied.

McMurtry and Stanford (1960) found that aphids on resistant plants at 77° and 95° F. died in nearly the same period of time as aphids confined in empty cages. They concluded that aphids on highly resistant plants die because of starvation or desiccation. They also reported that variations in the expression of resistance was caused by a change in the plant itself, and that this change took place within 2 days after plants were moved from one temperature to another.

## Pea Aphid

Macrosiphum pisi (Harris) is commonly known as the pea aphid. In 1782, Moses Harris of England described this insect, and named it Aphis pisum. The specific epithet was changed to pisi by Kaltenbach of Germany in 1843. In 1915, Davis of the United States placed it in the genus Macrosiphum (Palmer, 1953).

The earliest record of this aphid was from Europe by Kirby and Spence (1843). According to Davis (1915), the pea aphid was of exotic origin, having been known in European countries as early as 1815. It had also been reported from South Africa and India (Campbell, 1926). In 1899, it first appeared in destructive numbers in America (Davis, 1915), but had been reported by Cyrus Thomas in Illinois in 1878. It first appeared as a pest on alfalfa in Kansas in 1921 (Smith, 1956).

The primary host plants of the pea aphid are in the family Leguminosae. Patch (1938) listed 64 plant species on which this aphid has been observed. Hottes and Frison (1931) reported that pea aphids were collected from 14 plant species in Illinois. Two of these species had not been recorded by Patch. In California, it was observed on 17 species of plants, 15 of which were legumes (Campbell, 1926). One of the non-legumes was not recorded by Patch. Its primary hosts in Kansas are alfalfa, sweet clover, white clover, crimson clover, and vetch (Smith, 1956).

The first comprehensive studies of its biology in the United States were conducted by Davis (1915) and Smith (1916). Campbell (1926) reported that the longer-lived aphids averaged



fewer young per day than the shorter-lived ones, and that the number per day of the former remained high to the last day, whereas the short-lived showed a considerable decline in number toward the end of the reproductive period. He also stated that the average total number of young increased almost in direct proportion to the length of the reproductive period.

Searls (1932) presented the first recorded evidence that resistance to the pea aphid was present in some pea varieties. Searls (1932) and Maltais (1937) tested a large number of pea varieties for aphid resistance and found marked differences in resistance. Resistance in alfalfa was first observed by Blanchard and Dudley (1934) in fields in California and greenhouse experiments in Wisconsin. During a severe pea aphid outbreak in eastern Kansas in 1934, Painter and Grandfield (1935) observed wide variations in the amount of damage and number of aphids swept from different varieties of alfalfa in test plots. Ladak was found to be most resistant. This was later confirmed by Eichman and Webster (1940) in Washington. Albrecht and Chamberlain (1941) were able to differentiate  $F_2$  hybrid progenies of resistant and susceptible plants on the basis of per cent seedlings killed. However, a year later, little difference in reaction between the progenies of resistant and susceptible parents was observed. They concluded that resistance to pea aphids in these plants was not a stable character, but that further study should be made to determine the effect of environment on the expression of resistance. Blanchard (1943) reported that resistant lines which lost their green color because

of poor illumination also tended to lose their resistance to the pea aphid.

Emery (1946) concluded that resistance of alfalfa to the pea aphid was correlated primarily with an acid condition and a low level of sucrose, rather than with a scarcity of proteins in the plants. He attributed the acidity to (1) a deficiency of water, (2) a deficiency of light for photosynthesis, (3) temperatures too low for rapid growth of alfalfa, and (4) temperatures sufficiently high to cause the formation of lignin.

Barker and Tauber (1954) found that a pea plant grown on a high level of nitrogen fertilization was more tolerant to pea aphids. Pea plants deficient in nitrogen or phosphorous cause a marked reduction of the aphid's longevity, because the plants are severely injured in a short time.

Dahms and Painter (1940) showed that aphids confined to the flowering branches reproduced much faster than those confined to vegetative branches of the same plant. Blanchard and Dudley (1934) reported that the longevity of the pea aphid was inversely proportional to the temperature, and that this was true for all periods of the aphid's life. Campbell (1926) found that a rise or fall of  $10^{\circ}$  F. in the mean temperature was found to shorten or lengthen the aphid's life by 15 to 20 days. He also stated that a mean winter temperature of  $48^{\circ}$  to  $55^{\circ}$  F. resulted in a total life span of 35 to 60 days, while a summer mean temperature decreased the life span to 13 to 18 days. Schaefer (1933) made a detailed study of the effect of temperature on the biology of the pea aphid. It was not published in a

technical journal because the results were considered too closely parallel to those of Campbell (1926). Schaefer's investigations were actually carried a step further because he used the degree-hour concept.

Smith and Davis (1926) found that the greatest number of nymphs were produced at an average temperature of 65° F. and a relative humidity of 80 per cent. As the average temperature increased the number of nymphs produced decreased. Temperatures of 90° and 100° F. were definitely unfavorable and a temperature of 100° or higher was usually quickly fatal in temperature-controlled cages. The optimum temperature for this aphid was stated to be between 60° and 70° F. No definite conclusion could be drawn, however, because the temperature could not be held below 60° F. for any appreciable time. Higher reproductive and longevity averages were obtained at constant temperatures than at fluctuating temperatures.

Dahms and Painter (1940) tested individual alfalfa plants selected for various levels of resistance or susceptibility to pea aphids in field plots. At 61° F. an average of 7.4 aphids per female were produced on 16 resistant plants, and 46.2 per cent were dead at the end of 10 days, as compared with 27.3 aphids per female and 2.2 per cent mortality on 10 susceptible plants. At 44° F., 8.9 nymphs were produced per female and a mortality of 1.0 per cent on the resistant plants as compared with 9.9 nymphs per female and only 0.1 per cent mortality on the susceptible plants.

Schaefer (1933) found that under field conditions, in Wisconsin, the peak fecundity of more than 90 progeny per

female was attained at a mean temperature of  $59^{\circ}$  to  $64.4^{\circ}$  F., with a rapid decline below and above this temperature range. He also found that the duration of reproduction was inversely proportional to the temperature, with a range of 8.2 days at a mean of  $80.6^{\circ}$  to 35 days at  $48.4^{\circ}$  F.

Painter (1951, 1953) divides resistance as seen in the field into three bases or mechanisms, preference, antibiosis, and tolerance, which are interrelated. The present experiments were concerned with antibiosis or preference, or both.

#### MATERIALS AND METHODS

The materials and methods used in the experiments were those necessary for determining the effects of various controlled environmental conditions on the aphids while feeding on alfalfa, and for controlling, as much as possible, the known variables.

All pea aphids used in the experiments were selected from a stock culture in the Entomology greenhouse. This culture was formed in 1959 from field collections near Manhattan, Kansas, and has been maintained on broad bean plants (*Vicia fabae*) and alfalfa. Since the original formation of the culture, pea aphids, collected periodically from fields near Manhattan, were added.

The broad bean plants used for rearing the pea aphids were grown in  $10\frac{1}{2}$ " X 8" claypots. Soil of the following mixture was used: four parts black loam, one part sand, and one part sheep manure. To inhibit damping off, the bean seeds were soaked in a Captan solution (1 teaspoon/gal.), and the

seeded pots were then soaked with this solution. The bean seeds were covered with one inch of sand. Bean seeds were planted regularly so that vigorous young plants were usually available for rapid increase of the aphid population. The bean plants were supported by 24" aluminum rods and string.

Aphids were obtained by tilting the potted plant and shaking the aphids on to newspaper. They were transferred to a white enamel pan from which they were selected with an aspirator. Only apterous adults were used. These aphids were of unknown age but only those of healthy appearance and with eyes of the young visible through the abdomen were selected. A braconid parasite, Praon simulans (Prov.), frequently slowed the increase in aphid population. Aphids showing symptoms of being parasitized were not used in the experiments.

The spotted alfalfa aphids used in the experiments were selected from a stock culture maintained in the Entomology greenhouse. Peters and Painter (1958) reported that this culture was originally formed from a composite of collections from seven counties in different regions of Kansas. Since the original formation of the culture, spotted alfalfa aphids collected from fields near Manhattan, Kansas have been added periodically.

All spotted alfalfa aphids used in the experiments were reared on susceptible alfalfa grown in metal flats (4" X 15 3/4" X 22"). The soil used consisted of fine black loam, sand and peat moss. Two flats were placed in metal trays (1 1/2" X 26" X 50 1/2") and covered with large 32-mesh Lumite cages (47" X 26"



X 20") to exclude contamination and parasites. The alfalfa was watered from below.

The spotted alfalfa aphids were removed from the alfalfa flats with a modified vacuum cleaner. A piece of nylon cloth was placed between the end of the hose and metal cone for collection of the aphids. The aphids were then shaken into a white enamel pan from which they were picked up by an aspirator. Healthy appearing apterous adults of unknown age were used for all the experiments. A Bazor Magnifier lamp aided in selecting the apterous adults of both the spotted alfalfa aphid and the pea aphid.

Three clones of the alfalfa variety, Buffalo, were used in all the experiments. These were selected on the basis of previous reaction of the spotted alfalfa aphid and the pea aphid to the plants, and consisted of a resistant, intermediate and semi-susceptible clone. A first cycle selection (50-1266), served as the semi-susceptible clone in the present experiments. This clone has been used as a susceptible check for antibiosis tests of the spotted alfalfa aphid since 1954 and of the pea aphid since 1958 in the greenhouse. Clone 50-1266 is referred to as semi-susceptible because other alfalfa clones in the greenhouse have shown a greater degree of susceptibility to both aphids. Aphids feeding on highly susceptible clones frequently kill the host plant in a short time and thus curtail aphid reproduction. This was undesirable in the present experiments, so clone 50-1266 was used. Serving as the intermediate clone was a second cycle selection (215-9), which has frequently

been used as an intermediate check in pea aphid studies in the greenhouse. A third cycle selection (92-1-1), selected as a resistant clone, has been used as a resistant check in pea aphid studies in the greenhouse. In preliminary experiments in the greenhouse, the level of resistance of all three clones to the spotted alfalfa aphid was similar to that against the pea aphid. The above clones were transferred from the field to the greenhouse in June, 1960, and planted in 9" X 9" clay pots using the same soil mixture as was used for the broad bean plants. During the time the experiments were conducted, these clones were frequently cut back to stimulate new growth, and were given periodic nutrient solutions.

In all the experiments, newly formed trifoliolates, the third or fourth from the apex, were used. Trifoliolates injured or infested by mites or thrips were not used. Though pea aphids apparently prefer to feed on plant terminals, single trifoliolates were used in each replicate because it was difficult to make accurate counts of pea aphid reproduction on alfalfa terminals. Initial aphid infestations in all experiments consisted of three apterous adults, unless stated otherwise.

In the experiments involving humidity studies with the pea aphid and the spotted alfalfa aphid, a constant relative humidity was desired. A review of the literature showed that there were several methods of controlling the humidity but that it was difficult to obtain accurate measurements, especially in small spaces. Buxton (1931) and Buxton and Mellanby (1934) give excellent information on methods of controlling and measuring humidity. Additional information was given by Wilson (1921),

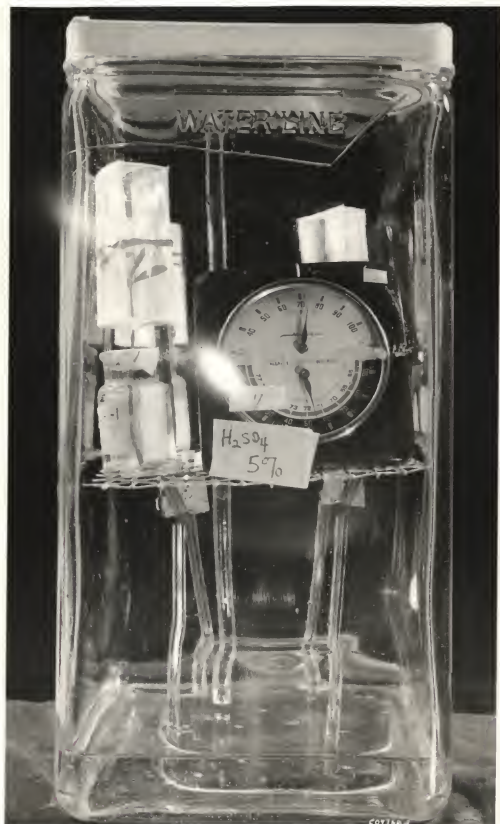
Shelford (1929), Smith (1931), Solomon (1951), and Peterson (1953).

In the present experiments, appropriate concentrations of sulfuric acid established the desired humidities in battery jars. Acid concentrations of 95, 40, and 5 per cent gave relative humidity readings of approximately 25, 50, and 90 per cent, respectively. The two extreme humidities varied  $\pm 5$  per cent while the intermediate varied  $\pm 3$  per cent. Eight hundred milliliters of sulfuric acid solution were used in each battery jar. The apparatus used for determining the effect of constant temperature and humidity on both species of aphids is shown in Plate 1. The battery jars used were all of uniform size ( $7\frac{1}{2}$ " X  $7\frac{1}{2}$ " X  $14\frac{1}{2}$ "). The wire gauze platform in each jar was supported by four  $\frac{1}{2}$  inch glass rods inserted through corks, and fastened to the platform with "Twist-ems". The floor of the platform in each jar was approximately six inches above the acid solution. The  $\frac{5}{8}$  inch glass tube, inserted through masking tape in the center of the platform, was used for pouring the acid solution into the jar. Transparent cellulose nitrate served as a jar lid, and was held securely with masking tape.

The excised alfalfa terminals (hereafter referred to as excises), inserted through an opening in the masking tape, were cut so the stems would extend to near the bottom of the small bottles (Plate 2). In preliminary experiments, excises of the three clones appeared to survive various constant temperatures and humidities equally well in tap water as in nutrient solutions and, therefore tap water was used. One trifoliolate of

EXPLANATION FOR PLATE I

Arrangement of apparatus used for determining the interaction of constant temperature and humidity on the biology of both species of aphids on alfalfa.





EXPLANATION FOR PLATE II

Method used for confining aphids to an excised alfalfa trifoliolate when studying the interaction of temperature and humidity on the biology of both species of aphids.

## PLATE II



each clone was tested in each jar; hence the three small bottles in each jar. Three apterous adult aphids were confined to each trifoliate by means of a transparent plastic box ( $1\frac{1}{4}$ " X  $1\frac{1}{4}$ " X 3"). Two sides of the box were cut out and replaced with nylon cloth in order to provide maximum ventilation. A v-shaped groove was made in one end of the cage to insert a petiole and a cotton plug to protect the petiole. Before using the cages in the experiments, they were tested and found to have no toxic effect on alfalfa plants or aphids. A split bamboo stick, fastened to the small bottle with masking tape, served as a support for the aphid cage. The cage was held in place with a rubber band. The small bottles were placed on the platform near the corner of the jar in such a way that it was possible to look through the cages from both plastic sides when counting the aphids. An Airguide desk type hygrometer-thermometer was placed on the platform.

The effects of the three constant humidities on the aphids were studied at 65° and 85° F. At 85° F., some of the excises appeared yellowish on the fourth day, so at that time all excises with live aphids were replaced by fresh excises. A camel's hair brush was used to transfer the aphids. Three replications of each clone were tested at each humidity. All three humidities were studied concurrently at each temperature; thus, nine jars were used for studying the effect of three constant humidities at a constant temperature on one species of aphid. The jars were rotated each day to minimize unknown variables.

The method employed for measuring humidity is illustrated

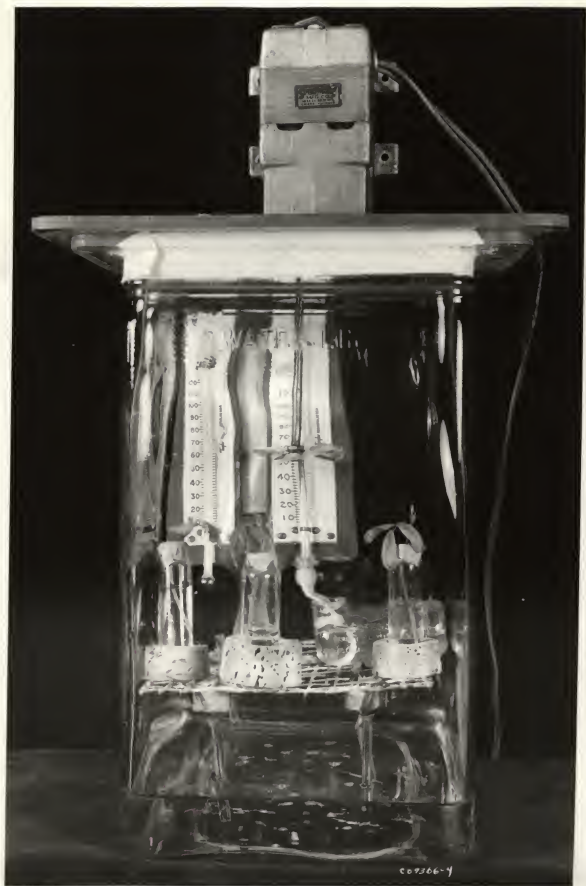
by Plate 3. This method was used because the sensing element of the humidity indicator in the hygrometer-thermometer was not accurate at the extreme humidities. The platform was constructed of the same material as the platform in the jar in Plate 1. A wet and dry bulb thermometer was fitted in the battery jar. Distilled water was used in the glass container to dampen the wick and to obtain a more accurate wet bulb reading. The 110 volt, 1/20 horsepower phonograph motor, mounted on the transparent cellulose nitrate lid, produced revolutions of the 2 inch diameter fan and thus, created an air current in the jar. Only one such apparatus was available, so the other two "control" jars had lids as the jar in Plate 1. One alfalfa excise of each clone was placed in each control jar so that the amount of water vapor in the jar due to transpiration would be similar to that in the experimental jars. Vials, supported by corks and filled with tap water, were used to maintain the alfalfa excises. The excises, inserted through an opening in the masking tape, were not infested with aphids since it was assumed that the aphids would not significantly modify the humidity.

The air current created by the fan was adequate for obtaining approximate relative humidity readings. The apparatus had originally been designed to measure the relative humidity in a jar with aphids on the excises; therefore, the revolutions per minute and the diameter of the fan were kept at a minimum. According to Peterson (1953), an air current of three miles per hour is required for accurate depression of the wet bulb. It is assumed that the air current produced by the apparatus used was somewhat less.

EXPLANATION FOR PLATE III

Arrangement of the apparatus employed for measuring  
relative humidity inside the battery jars.





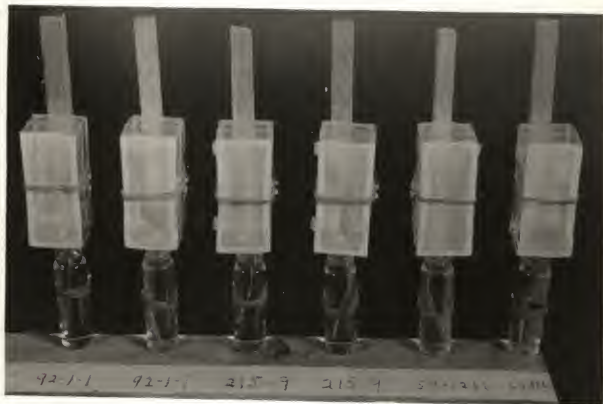
The relative humidity in each control jar was measured soon after each experiment was set up, and again at the end of the experiment on the seventh day. After measuring the relative humidity in one jar the apparatus was transferred to another jar. This transfer required only a few seconds and relatively little humidity disturbance occurred. The motor was operated until a constant reading was obtained on the wet bulb. In the jar with 95 percent sulfuric acid, this required approximately thirty minutes. Constant wet bulb readings were obtained sooner in the other two control jars.

Experiments were also conducted to study the biology of both species of aphids on excised in comparison with attached trifoliolates at several constant temperatures. Methods are illustrated by Plate 4. The cages and methods of infestation were the same as those employed in the jars. The vials, filled with tap water and containing the excises were the same size as those used in the control jars in the humidity and temperature experiments. Wooden slats nailed to one side of a 2" X 4" board supported the vials and cages containing the excises. The cages in the attached experiment were supported by split bamboo sticks. These excised and attached experiments were conducted at constant temperatures of 40°, 55°, and 85° F. At the highest temperature, some of the excises began to appear yellowish on the fifth day, therefore all excises with live aphids were replaced. Aphids were transferred to fresh excises with a camel's hair brush.

All experiments requiring constant humidities or constant

EXPLANATION FOR PLATE IV

Method used to study the biology of both species of aphids on excised and attached alfalfa trifoliolates at several constant temperatures.



temperatures or both were conducted in a Percival walk-in plant growth chamber with inside dimensions of 98 3/4" x 62 3/4" x 65 5/8". The bench in the chamber, constructed of sturdy wood grillwork to permit air flow, could be raised or lowered 15" to 79" from the fluorescent lamps. The bench was set at 36" from the lamps for all the experiments conducted in the chamber. At this level, a constant light intensity of approximately 1500 foot candles, supplied by fourteen 96" Sylvania slim-line fluorescent tubes, was used in all experiments conducted in the chamber.

All aphids used in experiments conducted in the chamber were confined to attached terminals of the susceptible clone, 50-1266, for 24 hours in the chamber, and exposed to the same light intensity and constant temperature as they would be in each subsequent experiment. This method of pre-conditioning was found to be most satisfactory because in this way all the aphids would have a more equal "start". Materials and methods used in pre-conditioning were similar to those shown in the bottom illustration of Plate 4 but the alfalfa terminals were infested instead of a single trifoliolate. Generally, fifteen adult aphids were confined in each cage. To be sure of sufficient numbers of aphids in good condition, approximately twice as many aphids were pre-conditioned as were required for each experiment.

Experiments involving excised and attached trifoliolates were also conducted on both species of aphids in the greenhouse where temperature and humidity fluctuated. The method of infestation and materials were similar to those of the excised

and attached experiments conducted in the chamber except five apterous adult aphids were used instead of three. Two replications of the excised and attached experiments were made in both the greenhouse and in the chamber.

Daily counts of nymphs and adults were made in all experiments. All experiments were continued for seven days. On the seventh day the battery jars were opened and the aphid cages, containing aphids and excises, were removed. Aphids were counted with the aid of the Dazor Magnifier lamp. The cages were likewise removed after severing the petioles in the excised and attached experiments, and the aphids counted. This final count was obviously the most accurate. This was especially true in the experiments where jars were used, because the counts had to be made by looking through the glass and plastic cage which caused some distortion.

The counts of both aphid species made at the third and seventh day of each experiment were used for analysis of significant differences in rate of reproduction and adult survival as influenced by temperature or humidity, or both. The nymphs per adult day, as used in the analysis, was obtained by dividing the cumulative totals of live nymphs by the total number of days all adults lived during each experiment.

The F test (Snedecor, 1959) was used for all statistical analysis of the data. The disadvantage of this method is that though certain individual differences may actually be significant they will not necessarily all be detected.



## RESULTS AND DISCUSSION

## Spotted Alfalfa Aphid

The interaction of temperature and humidity on the biology of the spotted alfalfa aphid is summarized by Tables 1 and 2. This data is also presented by Tables 4-7, where aphid reproduction and survival are compared among temperatures and among humidities. The results of statistical analysis of all variables studied are shown by Table 3.

Biology of the Spotted Alfalfa Aphid as Influenced by Humidity. It was found that the average number of nymphs per adult day and total number adult days were not significantly different at the three humidity levels, irrespective of temperature and clones, at both the third and seventh days (Tables 4 and 5). The numbers represent an average of 27 replications, summed across temperatures and clones. Probable individual differences were thus masked by differences between clones.

However, clone by humidity analysis revealed several significant differences which are shown by Tables 4 and 6. The numbers of nymphs per adult day reared on clone 50-1266 at all humidity levels were significantly greater than on the other clones after both the third and seventh days (Table 4). This was caused by high reproductive rates on clone 50-1266 at all humidity levels at 70° and 85° F. (Table 1). A significantly greater number of nymphs was produced on clone 50-1266 at the high humidity on the third day than at the low humidity. This difference was partly the result of the reproductive rates on

Table 1. The average number of live spotted alfalfa aphid nymphs per adult day on three selected alfalfa clones at three constant temperatures and humidity levels. Each trifoliate was infested with three apterous adult aphids and replicated three times; therefore, each number in the table represents an average of three replications.

Clone number	Number of live nymphs per adult day							
	25%	50% F.	50%	70% F.	25%	50%	90%	85% F.
3rd day count <sup>1</sup>								
50-1266	1.11	1.17	1.63	4.52	6.61	7.41	9.23	9.30
215-9	1.18	.43	.78	3.50	4.33	2.76	5.64	9.00
92-1-1	1.14	1.06	1.57	3.67	4.92	4.22	4.88	5.49
7th day count <sup>2</sup>								
50-1266	2.29	2.75	3.63	11.98	12.41	15.52	17.45	17.19
215-9	2.21	1.79	2.73	7.35	8.83	6.68	10.23	14.09
92-1-1	2.72	2.91	2.04	8.32	10.43	8.92	9.48	9.59

<sup>1</sup> L.S.D. at 5% level between clones and temperature and humidities = 1.80.

<sup>2</sup> L.S.D. at 5% level between clones and temperature and humidities = 3.74.

Table 2. The average total number of adult days of the spotted alfalfa aphid on three selected alfalfa clones at three constant temperatures and humidity levels. Each trifoliate was infested with three apterous adult aphids and replicated three times; therefore, each number in the table represents an average of three replications.

Clone number	Number of total adult days					
	55° F.		70° F.		85° F.	
	25%	50%	25%	50%	25%	50%
3rd day count <sup>1</sup>						
50-1266	9	6.3	9	8.3	9	8.3
215-9	7	7	9	8.3	9	7.7
92-1-1	9	9	8.7	9	9	6.3
7th day count <sup>2</sup>						
50-1266	17.3	13	21	18.7	21	19
215-9	15	15.7	19	15.3	13.3	12.3
92-1-1	21	20.3	19.3	19.3	21	20.7
					9	7

<sup>1</sup> L.S.D. at 5% level between clones and temperatures and humidities = 2.4.

<sup>2</sup> L.S.D. at 5% level between clones and temperatures and humidities = 7.5.

Table 3. The results of statistical analysis of all variables studied as affecting reproduction, development, and survival of the spotted alfalfa aphid, *Therioaphis maculata* (Buckton), on three selected alfalfa clones at three constant temperatures and humidities.

Variable(s)	Live nymphs per adult day			Total adult days		
	F	Sig.	7th day count	F	Sig.	7th day count
Humidity	.54	ns	.36	.93	ns	.16
Temperature	35.60	***	30.90	6.72	**	9.59
H x T	.26	ns	.79	1.32	ns	.76
Clones	4.94	*	5.69	2.14	ns	3.00
C x H	1.37	ns	.79	1.48	ns	1.87
C x T	14.91	***	10.94	8.14	***	6.48
C x T x H	3.44	**	2.10	1.49	ns	.58

\* Significant at the 5% level.

\*\* Significant at the 1% level.

\*\*\* Significant at the .1% level.

ns Non-significant difference.

Table 4. The average number of live spotted alfalfa aphid nymphs per adult day on three selected alfalfa clones at three constant humidity levels. Each trifoliate was infested with three apterous adult aphids and replicated three times. The effects of three constant temperatures were studied at each humidity level; therefore, each number in the table represents an average of nine replications.

Clone number	Number of live nymphs per adult day		
	25%	50%	90%
3rd day count <sup>1</sup>			
50-1266	4.95	5.69	6.16
215-9	3.47	4.27	4.42
92-1-1	3.32	3.83	3.00
Avg. <sup>2</sup>	3.91	4.60	4.53
7th day count <sup>3</sup>			
50-1266	10.57	10.78	10.51
215-9	6.61	8.17	7.65
92-1-1	6.84	7.64	5.53
Avg. <sup>4</sup>	8.01	8.86	7.91

<sup>1</sup> L.S.D. at 5% level between clones and humidity = 1.04.

<sup>2</sup> L.S.D. at 5% level between averages are non-significant.

<sup>3</sup> L.S.D. at 5% level between clones and humidity = 2.15.

<sup>4</sup> L.S.D. at 5% level between averages are non-significant.

Table 5. The average total number of adult days of the spotted alfalfa aphid on three selected alfalfa clones at three constant humidity levels. Each trifoliate was infested with three apterous adult aphids and replicated three times. The effects of three constant temperatures were studied at each humidity level; therefore, each number in the table represents an average of nine replications.

Clone number	Number of total adult days		
	25%	50%	90%
3rd day count <sup>1</sup>			
50-1266	9	7.7	9
215-9	7.9	7.2	6.4
92-1-1	8.1	7.8	6.8
Avg. <sup>2</sup>	8.3	7.6	7.4
7th day count <sup>3</sup>			
50-1266	19.1	18.9	20.3
215-9	14.2	15.2	12.0
92-1-1	16.1	16.1	13.9
Avg. <sup>4</sup>	16.5	15.7	15.4

- <sup>1</sup> L.S.D. at 5% level between clones and humidity = 1.4.  
<sup>2</sup> L.S.D. at 5% level between averages are non-significant.  
<sup>3</sup> L.S.D. at 5% level between clones and humidity = 4.3.  
<sup>4</sup> L.S.D. at 5% level between averages are non-significant.



Table 6. The average number of live spotted alfalfa aphid nymphs per adult day on three selected alfalfa clones at three constant temperatures. Each trifoliolate was infested with three apterous adult aphids and replicated three times. The effects of three constant humidity levels were studied at each temperature; therefore, each number in the table represents an average of nine replications.

Clone number	Number of live nymphs per adult day		
	55° F.	70° F.	85° F.
3rd day count <sup>1</sup>			
50-1266	1.30	6.18	9.29
215-9	.01	3.57	7.79
92-1-1	1.35	4.27	4.53
Avg. <sup>2</sup>	1.16	4.67	7.20
7th day count <sup>3</sup>			
50-1266	2.89	13.31	15.63
215-9	2.24	7.55	12.64
92-1-1	3.22	9.23	7.62
Avg. <sup>4</sup>	2.79	10.03	11.98

<sup>1</sup> L.S.D. at 5% level between clones and temperatures = 1.04.

<sup>2</sup> L.S.D. at 5% level between averages = 1.53.

<sup>3</sup> L.S.D. at 5% level between clones and temperatures = 2.15.

<sup>4</sup> L.S.D. at 5% level between averages = 2.62.

Table 7. The average total number of adult days of the spotted alfalfa aphid on three selected alfalfa clones at three constant temperatures. Each trifoliate was infested with three apterous adult aphids and replicated three times. The effects of three constant humidity levels were studied at each temperature; therefore, each number in the table represents an average of nine replications.

Clone number	Number of total adult days		
	55° F.	70° F.	85° F.
3rd day count <sup>1</sup>			
50-1266	8.1	8.8	8.8
215-9	7.7	8.3	8.4
92-1-1	8.9	9.0	4.4
Avg. <sup>2</sup>	8.2	8.7	6.2
7th day count <sup>3</sup>			
50-1266	17.1	20.2	19.0
215-9	16.6	16.0	8.9
92-1-1	19.9	20.0	6.2
Avg. <sup>4</sup>	17.9	18.7	11.0

<sup>1</sup> L.S.D. at 5% level between clones and temperatures = 1.4.

<sup>2</sup> L.S.D. at 5% level between averages = 1.5.

<sup>3</sup> L.S.D. at 5% level between clones and temperatures = 4.3.

<sup>4</sup> L.S.D. at 5% level between averages = 4.1.

clone 50-1266 at 70° F. where the average number of nymphs per adult day at the high humidity was 7.41 compared to 4.52 at the low humidity (Table 1). The average number of nymphs per adult day on clone 92-1-1 was distinctly smaller than on the other clones. It is interesting to note that the number of nymphs on clone 50-1266 at the seventh day was similar at all three humidity levels (Table 4). Aphids on the resistant clone appeared to reproduce at a faster rate at the intermediate humidity.

Adult survival on clone 50-1266 at the high humidity was significantly greater than on the other clones on the third and seventh days (Table 5). Survival of adults on clone 50-1266 was 100 per cent at the low and high humidity on the third day, and was also high on the seventh day at the high humidity. Adult survival on clones 215-9 and 92-1-1 was apparently inhibited by high humidity, and was significantly lower than survival on clone 50-1266. It is difficult to make conclusive statements about long time adult survival from Table 5 since the experiments were conducted for a relatively short period of time. Experiments conducted during a longer period of time would probably result in more definite conclusions.

Biology of the Spotted Alfalfa Aphid as Influenced by Temperature. A significantly greater number of nymphs per adult day was produced on each clone at 70° than at 55° F. on both the third and seventh days (Table 6). The number of nymphs per adult day on clones 50-1266 and 215-9 was likewise significantly different from clone 92-1-1.

The number of nymphs per adult day on clone 215-9 at 85° F.

was significantly greater than on clone 92-1-1 on the third and seventh day. No significant differences existed between clones at 55° on either day. Also, no significant differences were found between clones 215-9 and 92-1-1 at 70° on the third and seventh day.

Increased aphid reproduction and nymphal survival on clones 50-1266 and 215-9 was observed at 85° F. on the third and seventh day (Table 6). The average number of nymphs per adult day on clone 92-1-1 on the third day was greatest at 85°, but on the seventh day it was greatest at 70°.

Significant differences were found between the average number of nymphs per adult day at each temperature on the third day, irrespective of humidity levels and clones (Table 6). On the seventh day, however, the number of nymphs at 55° was significantly less than that at 70° and 85°.

The results shown in Table 6 are comparable to those of Hackerott and Harvey (1950). They found that the number of live nymphs per adult day increased with temperature at 10° intervals from 60° to 80° F. However, at 90° the rate of reproduction was inhibited on the resistant and susceptible clones, but actually increased on the intermediate clone. Accordingly, the optimum temperature for maximum reproduction on the resistant and susceptible clones should be between 80° and 90°. From the present study, the optimum temperature for maximum reproduction on clone 92-1-1 would be between 70° and 80° F. Since aphid reproduction on clones 215-9 and 50-1266 increased as temperature increased, the optimum temperature for maximum

reproduction on these clones could not be approximated. The relatively smaller number of nymphs per adult day on clone 215-9 at 55° and 70° on both days is somewhat difficult to explain. In preliminary experiments in the greenhouse where the temperature fluctuated, aphid reproduction on excised clone 215-9 was found to be higher than on clone 92-1-1, also in the excised condition. The trifoliolates of clone 215-9 used for the experiments at 55° and 70° were cut from the same potted plants as those used at 85° F. However, it is probable that the spotted alfalfa aphid resistance level of clone 215-9 is greater than clone 92-1-1 at lower temperatures.

Adult survival was higher on clone 92-1-1 at 55° and 70° on the third day than on the other two clones, but the difference was not significant (Table 7). At 85° on the third and seventh day, adult survival on clone 50-1266 was significantly greater than on clones 215-9 and 92-1-1 at 85° was significantly less than at 70° on either day.

Adult survival at 85°, irrespective of clones and humidity levels, was significantly less than at 70° and 55° on both days. Aphid reproduction and nymphal survival was found to be greatest at 85°, while adult survival was highest at 70°.

Biology of the Spotted Alfalfa Aphid as Influenced by Temperature and Humidity. The interaction of temperature and humidity on the number of nymphs per adult day is shown by Table 1 and Fig. 1-3. At 55°, there were no significant differences between humidity levels or clones.

At 70°, the number of nymphs per adult day on clone 50-1266

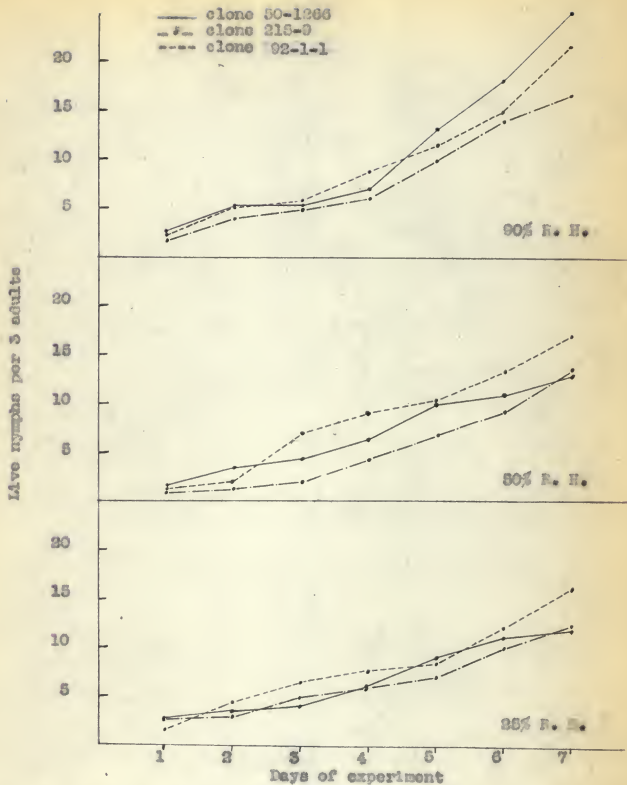


Fig. 1. Average number of live nymphs produced by three apterous spotted alfalfa aphid adults on three selected alfalfa clones as influenced by three humidity levels at 86° F. This graph represents an average of three replications.



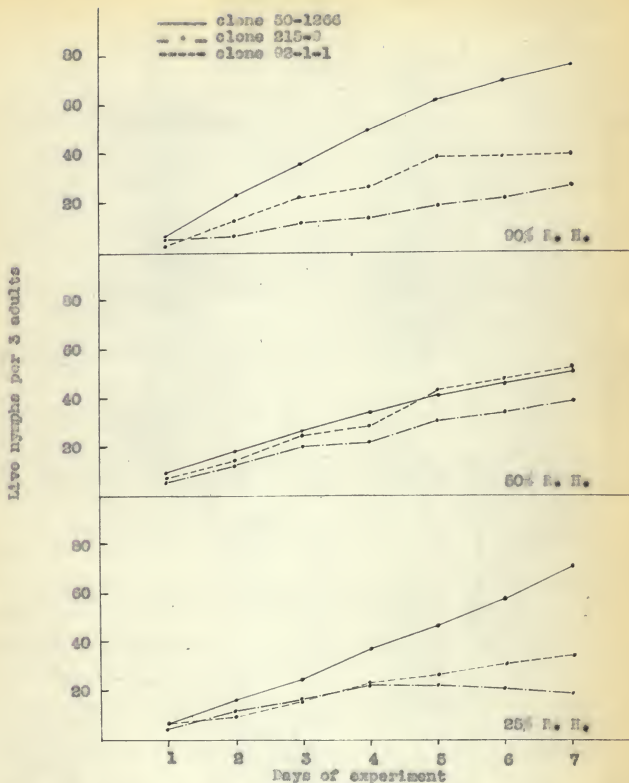


Fig. 2. Average number of live nymphs produced by three apterous spotted alfalfa aphid adults on three selected alfalfa clones as influenced by three humidity levels at 70° F. This graph represents an average of three replications.

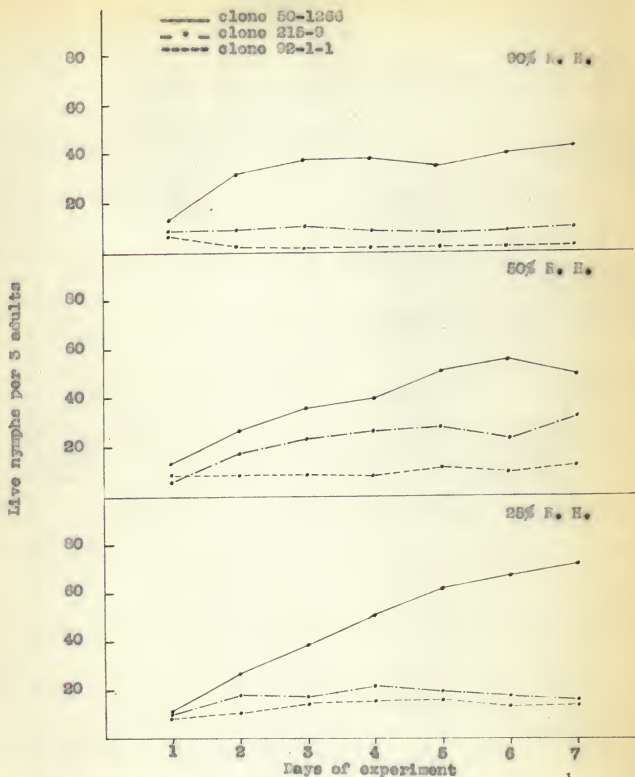


Fig. 5. Average number of live nymphs produced by three apterous spotted alfalfa aphid adults on three selected alfalfa clones as influenced by three humidity levels at 85° F. This graph represents an average of three replications.

was significantly greater at the intermediate and high humidity than on clone 215-9 on the third day, and at all humidity levels on the seventh day. Clone 50-1266 was also significantly higher than clone 92-1-1 at the high humidity at 70° on both days. The number of nymphs per adult day on clone 50-1266 at the low humidity at 70° was significantly less than at the intermediate and high humidity on the third day, and at the high humidity on the seventh day.

At 85°, the number of nymphs per adult day on clone 50-1266 was significantly higher than on the other clones at the low humidity, while on 92-1-1 the number of nymphs was significantly less than on the other clones at the intermediate and high humidity. A significantly smaller number of nymphs was found on clone 50-1266 at 85° at the high humidity on the seventh day than at the lower humidity levels. At 85°, the number of nymphs on clone 215-9 was significantly lower at the low humidity compared with both higher humidity levels on the third day, and the intermediate humidity on the seventh day. On clone 92-1-1, the number of nymphs at 85° at the high humidity was significantly less than at the intermediate humidity on the third day, and both lower humidity levels on the seventh day.

The least significant differences on Table 6 also apply to differences between temperatures at a given humidity for each clone. By the third day, the differences in number of nymphs on clones 50-1266 and 215-9 were significantly greater for each humidity level as the temperature increased. On clone 92-1-1, significant differences existed for each humidity between 55°

and 70° by the third day but not between 70° and 85°.

On the seventh day, the number of nymphs on each clone was significantly greater at each humidity level at 70° than at 55° (Table 1). At 85°, the number of nymphs on clone 50-1266 at the low and intermediate humidity was significantly greater than at the corresponding humidity levels at 70°. However, the number of nymphs on clone 50-1266 at the high humidity at 70° was significantly more than at the same humidity level at 85°. Significant differences also existed between 70° and 85° on clone 215-9 at the intermediate and high humidity. The only significant difference involving 82-1-1 for 70° and 85° was at the high humidity.

The interaction of temperature and humidity on the survival of spotted alfalfa aphid adults is shown by Table 7. At 55°, the one significant difference found was between clones 92-1-1 and 50-1266 at the intermediate humidity on the third day. No significant differences were found between clones and humidity levels at 55° on the seventh day.

No significant differences were found in adult survival at 70° between clones and humidity levels on the third day. On the seventh day there was a significant difference between clones 50-1266 and 215-9 at the high humidity. No significant differences were found between humidity levels for each clone at 70°.

At 85°, adult survival was significantly higher on clone 50-1266 at all humidity levels than on clone 92-1-1 at the end of both days. Adult survival on clone 50-1266 was also significantly higher than on clone 215-9 at the intermediate and high humidity on the third day, and at the high humidity on the

seventh day. Adult survival on clone 215-9 at the high humidity was significantly lower than at the lower humidity levels at the end of both days. On clone 92-1-1, survival of adults at the high humidity was significantly lower than at the lower humidity levels.

Tables 1 and 2 show that humidity is a minor factor in a spotted alfalfa aphid population at 85° and 70°. However, at 85°, the influence of the high humidity on adult survival was especially noticeable on clones 215-9 and 92-1-1. Graham (1959), studying the effects of temperature and humidity on the spotted alfalfa aphid on susceptible alfalfa, found that the low humidities were more favorable for reproduction of this aphid, especially at the higher temperatures. He found this to be most evident at 30° C. (86° F.). This may seem to be contradictory to the results shown in Table 1, but Graham studied the fecundity rates of the aphids during their entire reproductive period, instead of for just seven days. Comparison of the number of nymphs per adult day on clone 50-1266 at 70° and 85° F. in the present study with Graham's results on Caliverde alfalfa for the first seven days was roughly similar at 20° C. (68° F.) and 30° C. (86° F.). Studies by McMurtry and Stanford (1960) indicated that spotted alfalfa aphids on resistant plants die by starvation and desiccation. These workers also found that the physiology of alfalfa plants changed in about two days when the plants were moved from one temperature to another; thus attributing the expression of resistance primarily to a change in the host plant itself. This may be an explanation for the

responses of the aphid on a given clone at different temperatures in the present study.

Maxwell and Painter (1959) found that the rate of honeydew excretion of the spotted alfalfa aphid increased at temperatures up to 85°. In the present study, this was also observed, especially on clone 50-1266.

The trend of reproduction and survival of spotted alfalfa aphid nymphs during the seven days of the experiments is graphically presented by Figures 1-3. The graphs are necessary for a better understanding of the interaction of temperature and humidity on the reproductive rate of the spotted alfalfa aphid.

The above three figures indicate that the expression of resistance to the spotted alfalfa aphid is more evident at 85° F. than at the two lower temperatures. It is interesting to note that at 70°, resistance to the aphid is greater in clone 215-9 than in clone 92-1-1. This statement may be confirmed by records of numerous antibiosis tests, in the Entomology greenhouse at Kansas State University. Thus, the resistance of a given clone to this aphid should be defined in terms of temperature, humidity, and time of year.

#### Pea Aphid

The interaction of temperature and humidity on the biology of the pea aphid is summarized by Tables 8 and 9. This data is also presented by Tables 11-14, where aphid reproduction and survival are compared among temperatures and among humidities. The results of statistical analysis of all variables studied are shown by Table 10.



Table 8. The average number of live pea aphid nymphs per adult day of the pea aphid on three selected alfalfa clones at three constant temperatures and humidity levels. Each trifoliate was infested with three apterous adult aphids and replicated three times; therefore, each number in the table represents an average of three replications.

Clone number	Number of nymphs per adult day					55° F.		70° F.		85° F.	
	25%	50%	90%	25%	50%	25%	50%	25%	50%	25%	50%
<b>3rd day count<sup>1</sup></b>											
50-1266	5.07	5.92	5.34	10.26	11.70	11.11	16.63	14.29	14.34		
215-9	5.67	5.34	6.37	12.52	14.18	12.15	15.78	15.95	11.14		
92-1-1	5.85	5.39	4.80	7.07	12.44	7.26	11.26	12.58	6.46		
<b>7th day count<sup>2</sup></b>											
50-1266	9.14	9.50	9.03	19.13	19.70	19.58	27.31	22.62	15.87		
215-9	8.76	9.22	10.93	20.02	22.03	20.44	21.84	22.05	13.05		
92-1-1	9.60	9.00	9.51	12.48	19.36	13.94	17.95	16.92	5.36		

<sup>1</sup> L.S.D. at 5% level between clones and temperatures and humidities = 3.46.

<sup>2</sup> L.S.D. at 5% level between clones and temperatures and humidities = 5.65.

Table 9. The average total number of adult days of the pea aphid on three selected alfalfa clones at three constant temperatures and humidity levels. Each trifoliate was infested with three apterous adult aphids and replicated three times; therefore, each number in the table represents an average of three replications.

Clone number	55° F.			70° F.			85° F.		
	25%	50%	90%	25%	50%	90%	25%	50%	90%
3rd day count <sup>1</sup>									
50-1266	8.7	9	9	9	9	9	9	8.7	8.7
215-9	8.3	9	8	9	9	9	8	7.3	8.7
92-1-1	9	8	7	8.7	9	9	7.7	5.3	4.7
7th day count <sup>2</sup>									
50-1266	19.3	20.3	19	21	20.3	20	14.7	17	18.3
215-9	19	21	18.7	20.7	19.7	19.7	12.7	14.7	19.3
92-1-1	21	18.7	18.7	19.3	20.3	20.7	12.3	7.7	7.3

<sup>1</sup> L.S.D. at 5% level between clones and temperatures and humidities = 1.27.

<sup>2</sup> L.S.D. at 5% level between clones and temperatures and humidities = 4.87.

Table 10. The results of statistical analysis of all variables studied as affecting reproduction, development, and survival of the pea aphid, *Macrosiphum pisii* (Harris), on three selected alfalfa clones at three constant temperatures and humidities.

Variable(s)	Live nymphs per adult day			Total adult days		
	3rd day count F	7th day count F	Sig.	3rd day count F	7th day count F	Sig.
Humidity	3.61	ns	ns	.71	ns	.001 ns
Temperature	50.37	***	***	5.92	*	17.37 ***
H x T	2.12	ns	ns	.61	ns	.27 ns
Clones	7.78	**	**	5.13	*	2.70 ns
C x H	1.70	ns	ns	2.13	ns	1.08 ns
C x T	4.20	**	**	5.36	**	5.29 **
C x T x H	.79	ns	ns	1.17	ns	1.25 ns

\* Significant at the 5% level.

\*\* Significant at the 1% level.

\*\*\* Significant at the .1% level.

ns Non-significant difference.

1 Actual number is .0041.

Table 11. The average number of live pea aphid nymphs per adult day on three selected alfalfa clones at three constant humidity levels. Each trifoliolate was infested with three apterous adult aphids and replicated three times. The effects of three constant temperatures were studied at each humidity level; therefore, each number in the table represents an average of nine replications.

Clone number	Number of live nymphs per adult day		
	25%	50%	90%
3rd day count <sup>1</sup>			
50-1266	10.65	10.64	10.26
215-9	11.32	11.82	9.89
92-1-1	8.06	10.14	6.17
Avg. <sup>2</sup>	10.01	10.87	8.77
7th day count <sup>3</sup>			
50-1266	18.53	17.27	14.83
215-9	16.87	17.78	14.82
92-1-1	13.34	15.53	9.60
Avg. <sup>4</sup>	16.25	16.86	13.08

<sup>1</sup> L.S.D. at 5% level between clones and humidities = 1.99.

<sup>2</sup> L.S.D. at 5% level between averages = 1.68.

<sup>3</sup> L.S.D. at 5% level between clones and humidities = 3.25.

<sup>4</sup> L.S.D. at 5% level between averages = 2.47.

Table 12. The average total number of adult days of the pea aphid on three selected alfalfa clones at three constant humidity levels. Each trifoliolate was infested with three apterous adult aphids and replicated three times. The effects of three constant temperatures were studied at each humidity level; therefore, each number in the table represents an average on nine replications.

Clone number	Number of total adult days		
	25%	50%	90%
3rd day count <sup>1</sup>			
50-1266	8.9	8.9	8.9
215-9	8.4	8.4	8.6
92-1-1	8.4	7.4	6.9
Avg. <sup>2</sup>	8.6	8.3	8.1
7th day count <sup>3</sup>			
50-1266	18.3	19.2	19.1
215-9	17.5	18.5	18.9
92-1-1	17.6	15.6	15.6
Avg. <sup>4</sup>	17.8	17.7	17.9

<sup>1</sup> L.S.D. at 5% level between clones and humidities = .9.

<sup>2</sup> L.S.D. at 5% level between averages = .9.

<sup>3</sup> L.S.D. at 5% level between clones and humidities = 2.8.

<sup>4</sup> L.S.D. at 5% level between averages = 2.5.

Table 13. The average number of live pea aphid nymphs per adult day on three selected alfalfa clones at three constant temperatures. Each trifoliolate was infested with three apterous adult aphids and replicated three times. The effects of three constant humidity levels were studied at each temperature; therefore, each number in the table represents an average of nine replications.

Clone number	Number of live nymphs per adult day		
	55° F.	70° F.	85° F.
3rd day count <sup>1</sup>			
50-1266	5.44	11.02	15.09
215-9	5.79	12.95	14.29
92-1-1	5.35	8.93	10.10
Avg. <sup>2</sup>	5.53	10.97	13.16
7th day count <sup>3</sup>			
50-1266	9.22	19.47	21.93
215-9	9.65	20.83	18.98
92-1-1	9.64	15.43	13.41
Avg. <sup>4</sup>	9.51	18.58	18.11

<sup>1</sup> L.S.D. at 5% level between clones and temperatures = 1.99.

<sup>2</sup> L.S.D. at 5% level between averages = 1.66.

<sup>3</sup> L.S.D. at 5% level between clones and temperatures = 3.25.

<sup>4</sup> L.S.D. at 5% level between averages = 2.47.



Table 14. The average total number of adult days of the pea aphid on three selected alfalfa clones at three constant temperatures. Each trifoliate was infested with three apterous adult aphids and replicated three times. The effects of three constant humidity levels were studied at each temperature; therefore, each number in the table represents an average of nine replications.

Clone number	Number of total adult days		
	55° F.	70° F.	85° F.
3rd day count <sup>1</sup>			
50-1266	8.9	9.0	8.8
215-9	8.4	9.0	8.0
92-1-1	8.0	8.9	5.9
Avg. <sup>2</sup>	8.4	9.0	7.6
7th day count <sup>3</sup>			
50-1266	19.6	20.4	16.7
215-9	19.6	20.0	15.2
92-1-1	19.4	20.1	9.1
Avg. <sup>4</sup>	19.5	20.2	13.7

<sup>1</sup> L.S.D. at 5% level between clones and temperatures = .9.

<sup>2</sup> L.S.D. at 5% level between averages = .9.

<sup>3</sup> L.S.D. at 5% level between clones and temperatures = 2.8.

<sup>4</sup> L.S.D. at 5% level between averages = 2.8.

Biology of the Pea Aphid as Influenced by Humidity. A comparison of the average number of nymphs per adult on all clones at each humidity level (Table 11), showed that the high humidity had a depressing effect. This was especially evident on the seventh day. However, it is interesting to note that the number of nymphs on clone 92-1-1 at the low humidity was also significantly lower than on the other clones. The number of nymphs, on clone 92-1-1 at the intermediate humidity, was significantly greater than at the high humidity. Aphids on clone 50-1266 reproduced at an increased rate at the lower humidities as shown by the decreased number of nymphs on clone 50-1266 at the high humidity.

The only apparent influence of humidity on survival of pea aphid adults (Table 12) was on clone 92-1-1 at the intermediate and high humidities. At these humidity levels the survival of adults was significantly less than on clone 215-9 and 50-1266.

An overall comparison of the influence of humidity, irrespective of temperature and clones, showed that reproduction and survival of nymphs increased at the lower humidity levels (Table 11) while humidity had less effect on the survival of adults (Table 12). Schaefer (1933) found that humidity has little influence on pea aphid populations on susceptible alfalfa.

Biology of the Pea Aphid as Influenced by Temperature. Significant differences in number of nymphs per adult day and total adult days were found between clones and temperatures on the third and seventh days (Table 13 and 14). At 55°, the average number of nymphs per adult day on all clones was significantly less than at 70° and 85° on both days. However, differences between

clones at 85° were not significant.

At 70°, the number of nymphs on clone 50-1266 on the third day was significantly less than at 80°. The number of nymphs on clone 92-1-1 was significantly less than on clone 215-3 on the third day. Also fewer nymphs were produced on clone 92-1-1 than on clones 50-1266 and 15-2 on the seventh day.

The average number of nymphs per adult day on clone 92-1-1 at 85° was significantly less than on the other clones on both days.

There were no significant differences among clones in the ability of adults to survive at 55° and 70° (Table 14). Also, there were no significant differences in adult survival on the same clone or between clones at 85° and 70° on either days, possibly because the time considered was too short for effects to show.

At 85°, adult survival was significantly decreased on clone 92-1-1 compared with clones 215-3 and 50-1266 on both days (Table 14). Adult survival on all clones on the seventh day was significantly reduced at 85° when compared with the lower temperatures. On the third day, adult survival on clones 215-3 and 92-1-1 at 85° were significantly less than at 70° F.

When only temperature was considered, the average number of nymphs per adult day on the third day was significantly greatest at 85° (Table 13). By the seventh day, the average number of nymphs at 70° was significantly smaller than at the two higher temperatures on both days. Adult survival (Table 14) was most favorable at the two lower temperatures, while a temperature of

85° had a significantly adverse effect.

The present study indicated that, in general, reproduction, development and survival of pea aphids was more favorable at 70° than at either 55° or 85°. However, it did not demonstrate an exact optimum temperature for the aphid since 15° temperature intervals were used in this study. Smith and Davis (1926) stated that the rate of reproduction on susceptible alfalfa was higher at 65° F. than at either 60° and 70°. These temperatures were averages, so it is difficult to compare their results with the present work. Schaefer (1933) concluded that the optimum effective temperature for pea aphid reproduction and development lay between 60° F. and 65° F.

Biology of the Pea Aphid as Influenced by Temperature and Humidity. Differences in aphid reproduction and survival due to the interaction of temperature and humidity (Tables 8 and 9 and Fig. 4-6) were most evident at the high temperature and higher humidity levels on the more resistant clones. Aphids on clone 92-1-1 were adversely affected by the high humidity at 85° when compared with the aphids on the other clones at the same humidity and temperature. At 55°, there were no significant differences in the average number of nymphs per adult day between clones and humidity levels (Table 8).

At 70°, the number of nymphs on clone 92-1-1 was significantly less than on clone 218-9 at the low and high humidity on the third and seventh days. The number of nymphs on clone 92-1-1 was also significantly smaller than on clone 50-1266 at the high humidity on the third day, and at low, and almost significant at the high humidity on the seventh day.

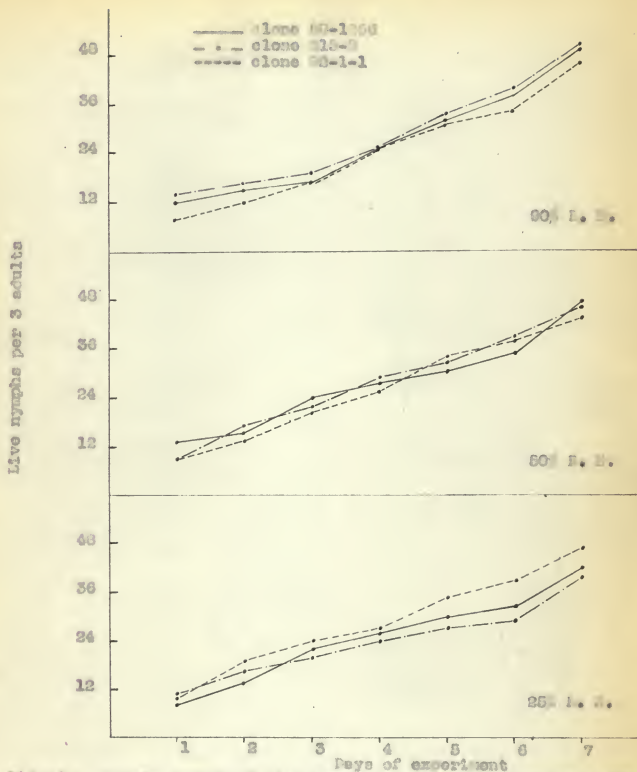


FIG. 4. Average number of live nymphs produced by three apterous pea aphid adults on three selected alfalfa clones as influenced by three humidity levels at 55° F. This graph represents an average of three replications.

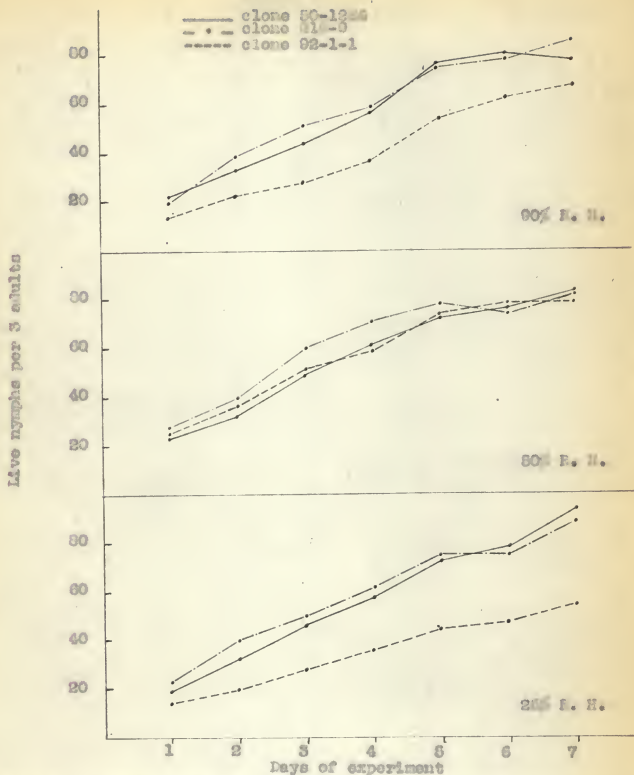


Fig. 5. Average number of live nymphs produced by three apterous pea aphid adults on three selected alfalfa clones as influenced by three humidity levels at 70° F. This graph represents an average of three replications.



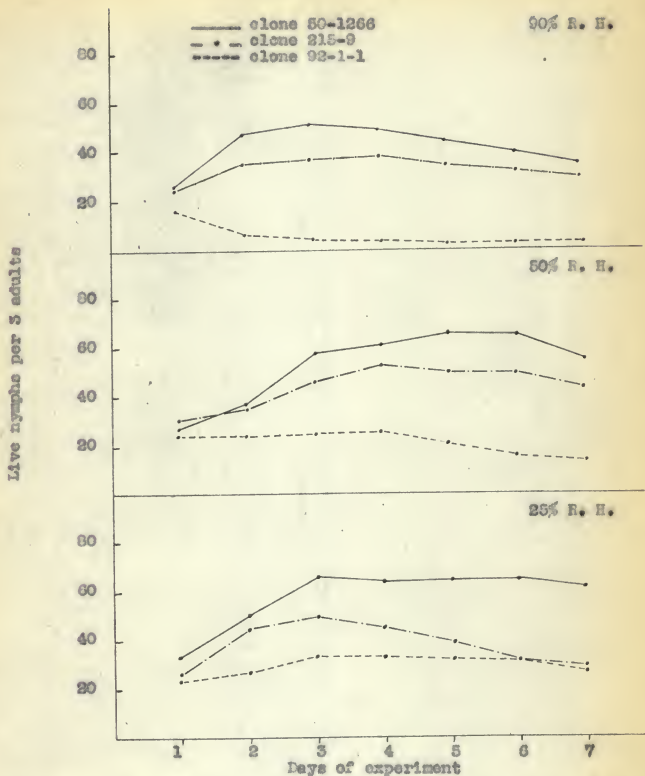


Fig. 6. Average of live nymphs produced by three apterous pea aphid adults on three selected alfalfa clones as influenced by three humidity levels at 85° F. This graph represents an average of three replications.

At 85°, there were significant differences in number of nymphs at the high humidity and the lower humidity levels on clones 215-9 and 92-1-1 on both days. The same significant difference involving clone 50-1266 was found at 85° on the seventh day. The number of nymphs, at the end of both days, on clone 92-1-1 at 85° was significantly smaller at the high humidity than for the other humidity levels at that temperature.

The effects of temperature and humidity on adult survival were not very marked (Table 9). The important difference was at 85° in conjunction with the high humidity where adult survival on clone 92-1-1 was significantly less than on the other clones at both days. Adult survival on clone 92-1-1 at 85° was also significantly less than on the other clones at the intermediate temperature on the seventh day.

Figures 4-6 give a graphical presentation of the trend of reproduction and survival of pea aphid nymphs during the seven days of the experiments. These graphs were made to give a more complete picture of the results in the present study.

It is interesting to note that the expression of resistance is clearly evident at 85° F. (Fig. 6) but much less clearly or not at all at 70° or 55° F. At 55° F., the number of live pea aphid nymphs was approximately the same on all clones throughout the seven days (Fig. 4). At 70°, resistance became apparent at the low and high humidity where the number of live nymphs on clone 92-1-1 was distinctly smaller than on the other clones.

A review of the literature showed that no workers had studied the effects of controlled temperature and humidity on

the pea aphid on alfalfa. The studies by Smith and Davis (1926), Campbell (1928) and Schaefer (1933) were conducted in the greenhouse or field, or both, where temperature and humidity fluctuated. Therefore, it was difficult to correlate the results of the present study with their work. Smith and Davis (1926) were able partially to control the temperature in some of their studies, and found that aphid populations, increased more rapidly under partially controlled temperatures than uncontrolled conditions.

Smith and Davis (1926) concluded that pea aphid reproduction was greatest at 65° F. and 90 per cent relative humidity. Schaefer (1933) stated that a temperature between 60° and 65° F. and a relative humidity of 62 to 70 per cent were optimum for greatest reproduction. As stated earlier, the present experiments were conducted for seven days, therefore, comparisons with results of full life history studies may not be valid.

An interesting phenomenon was observed with both aphid species at 85° F. at the high humidity. At that temperature and humidity a number of progeny were deposited dead and were still contained within the embryonic membrane. This was not observed to occur at any other temperature or humidity. This observation is confirmed by Graham (1959). The higher temperatures apparently increased the metabolic rate, causing premature parturition of progeny developing in the adults as evidenced by the reaction at the high humidity at 85° F. The number of nymphs of both aphid species which died during the experiments increased as the temperature increased (Tables 15 and 16).

Table 15. The total number of dead and live spotted alfalfa aphid nymphs on three selected alfalfa clones at three constant temperatures. Each trifoliate was infested with three apterous adults, and replicated three times. The effects of three constant humidity levels was studied at each temperature; therefore, each number in the table represents a sum of nine replications.

Clone number	55° F.		70° F.		85° F.	
	Dead	Live	Dead	Live	Dead	Live
7th day count						
50-1266	30	145	40	599	215	493
215-3	18	126	142	251	166	179
92-1-1	19	165	132	384	168	83

Table 16. The total number of live and dead pea aphid nymphs on three selected alfalfa clones at three selected alfalfa clones at three constant temperatures. Each trifoliate was infested with three apterous adults, and replicated three times. The effects of three constant humidity levels was studied at each temperature; therefore, each number in the table represents a sum of nine replications.

Clone number	55° F.		Total number of nymphs 70° F.		85° F.	
	Dead	Live	Dead	Live	Dead	Live
7th day count						
50-1266	4	415	89	767	281	443
215-9	2	411	77	764	313	294
92-1-1	1	409	60	612	329	123

Tables 15 and 16 indicate that 70° F. was more favorable for reproduction and survival of both aphids than was 55° or 85° F. This seems to contradict the results as shown by Tables 1 and 8, where reproduction of both aphids seems to be most favorable at 85° F. The greater number of nymphs per adult day of both aphid species at 85° was partially caused by a lower adult survival rate of both aphids at 85°.

A comparison of the interaction of temperature and humidity on the biology of the pea aphid with that of the spotted alfalfa aphid yielded several striking differences. At 55° (Fig. 1 and 4), the number of live pea aphid nymphs produced on all three clones at all three humidities is about twice as great as the number of live spotted alfalfa aphid nymphs. This distinct difference is also shown by Tables 15 and 16.

The number of live pea aphid nymphs at 70° F. was also greater than spotted alfalfa aphid nymphs on all clones and at all humidities, except clone 50-1266 at the high humidity. It was interesting to note that at the intermediate humidity aphid reproduction rate on the three clones was similar for each species.

At 85°, the number of live spotted alfalfa aphid nymphs was again smaller than that of the pea aphid on all clones and at all humidities. However, the level of resistance between clones was similarly expressed by both aphids, especially at the intermediate humidity. At all temperatures, the number of live pea aphid nymphs on all clones and at all humidities was two to three times as great as the number of spotted alfalfa aphid nymphs. This difference is also shown by Tables 1 and 8.



The relatively small influence of humidity on the biology of the spotted alfalfa and pea aphid found in the present study was also shown with the greenbug, Toxoptera graminum (Rond.) by Headlee (1914) and Wadley (1931). Wadley (1931) concludes that sap-feeding insects apparently are not affected directly by humidity. Headlee (1917) suggested that for each stage of each insect species there is a definite internal water optimum--an amount of body fluid which will permit necessary chemical and physical changes to take place with the greatest efficiency. He concludes that humidity acts on an insect directly by (1) the removal of water or (2) the prevention of the loss of body fluid. Emery (1946) found that the water content of alfalfa plants was a primary factor in temporary resistance to the pea aphid. Even though this factor may not significantly alter the atmospheric humidity, it may influence aphid feeding by variations in the turgor pressure. Certainly, humidity should be considered in host plant resistance studies with aphids, but temperature is definitely a more important factor.

#### Comparison of Effects of Feeding on Excised and Attached Trifoliolates

Several workers have observed that the reproductive rate of the spotted alfalfa and the pea aphid differs on excised and attached plant parts of a number of alfalfa varieties. Data obtained regarding several alfalfa varieties, in the Entomology greenhouse at Kansas State University and at the Fort Hays Branch Experiment Station at Hays, Kansas during the past two years, indicates that both aphid species reproduce at an

increased rate on excised plant parts when compared with reproduction on attached parts.

The data obtained in the present comparison of effects of feeding on excised and attached trifoliolates is presented in Tables 17-20 and Fig. 7-12. The primary reason for these studies was to determine the difference, if any, between aphid reproduction on excised and attached trifoliolates at constant temperatures and uncontrolled humidities. Another reason was to observe the biology of both aphids on alfalfa as influenced by a constant temperature of 40° F. A review of the literature indicated that no workers had conducted such studies. Experiments were conducted at 55° and 85° F. to determine the influences of extreme temperatures since the effects may be more critical at these temperatures.

Biology of the Spotted Alfalfa Aphid as Influenced by Excised and Attached Trifoliolates. The average number of nymphs per adult day on both excised and attached trifoliolates of clone 50-1266 increased with an increase in temperature (Table 17). This was also observed on excised trifoliolates of clones 215-9 and 92-1-1. However, attached trifoliolates of the same clones supported less nymphs at 85° F. than at 55° on both the third and seventh day.

The survival of spotted alfalfa aphid adults on clone 50-1266 remained high at all temperatures on both excised and attached trifoliolates (Table 18). Adult survival was inhibited on clones 215-9 and 92-1-1 at 85° F., and on attached trifoliolates of clone 92-1-1 at 55° F.

Figures 7-9 show the average number of live nymphs per

Table 17. The average number of live spotted alfalfa aphid nymphs per adult day on excised and attached trifoliolates of three selected alfalfa clones at three constant temperatures. Each trifoliolate was infested with three apterous adult aphids, and replicated twice.

Clone number	40° F.		55° F.		85° F.	
	Excised	Attached	Excised	Attached	Excised	Attached
3rd day count <sup>1</sup>						
50-1266	.83	1.06	2.50	4.73	12.84	13.17
215-9	.95	.83	4.23	1.67	14.13	2.63
92-1-1	.61	.72	1.72	3.75	13.75	.75
AVG. 2	.81	.72	2.82	3.38	13.57	5.52
7th day count <sup>3</sup>						
50-1266	2.14	2.34	6.91	9.91	21.86	22.80
215-9	2.03	.72	9.36	4.05	24.11	2.57
92-1-1	1.53	1.35	3.27	7.93	24.75	.75
AVG. 4	1.90	1.47	6.51	7.30	23.57	8.71

<sup>1</sup> L.S.D. at 5% level between clones and temperatures and trifoliolates = 9.49.

<sup>2</sup> L.S.D. at 5% level between averages = 9.43.

<sup>3</sup> L.S.D. at 5% level between clones and temperatures and trifoliolates = 17.75.

<sup>4</sup> L.S.D. at 5% level between averages = 17.38.

Table 13. The average number of total adult days of the spotted alfalfa aphid on excised and attached trifoliate leaves of three selected alfalfa clones at three constant temperatures. Each trifoliate was infested with three apterous adult aphids, and replicated twice.

Clone number	40° F.		55° F.		85° F.	
	Excised	Attached	Excised	Attached	Excised	Attached
3rd day count <sup>1</sup>						
50-1266	9.5	9	9	9	9	9
215-9	9	8.5	9	9	6.5	2
92-1-1	9	9	9	4.5	1.5	2.5
AVG. <sup>2</sup>	8.8	8.8	9	7.5	5.8	4.5
7th day count <sup>3</sup>						
50-1266	18.5	19.5	21	21	21	19.5
215-9	21	18	21	20	11	4
92-1-1	21	20	21	6.5	1.5	2.5
AVG. <sup>4</sup>	20.2	19.2	21	15.8	11.2	8.7

<sup>1</sup> L.S.D. at 5% level between clones and temperatures and trifoliate leaves = 2.93.

<sup>2</sup> L.S.D. at 5% level between averages = 6.38.

<sup>3</sup> L.S.D. at 5% level between clones and temperatures and trifoliate leaves = 7.26.

<sup>4</sup> L.S.D. at 5% level between averages = 16.53.

Table 19. The average number of live pea aphid nymphs per adult day on excised and attached trifoliolates of three selected alfalfa clones at three constant temperatures. Each trifoliolate was infested with three apterous adult aphids, and replicated twice.

Clone number	Number of nymphs per adult day			
	40° F.		55° F.	
	Excised	Attached	Excised	Attached
3rd day count <sup>1</sup>				
50-1266	5.06	4.56	12.06	10.25
215-9	3.61	5.73	9.94	9.23
92-1-1	4.83	5.11	7.86	6.50
Avg. 2	4.50	5.13	9.95	8.68
7th day count <sup>3</sup>				
50-1266	9.33	9.08	19.03	17.53
215-9	6.75	10.17	17.44	15.36
92-1-1	8.64	10.14	14.79	11.22
Avg. 4	8.24	9.90	17.09	14.70

<sup>1</sup> L.S.D. at 5% level between clones and temperatures and trifoliolates = 3.86.

<sup>2</sup> L.S.D. at 5% level between averages = 5.40.

<sup>3</sup> L.S.D. at 5% level between clones and temperatures and trifoliolates = 6.24.

<sup>4</sup> L.S.D. at 5% level between averages = 7.04.



Table 20. The average number of total adult days of the pea aphid on excised and attached trifoliolates of three selected alfalfa clones at three constant temperatures. Each trifoliolate was infested with three apterous adult aphids, and replicated twice.

Clone number	40° F.				55° F.				85° F.			
	Excised	Attached	Excised	Attached	Excised	Attached	Excised	Attached	Excised	Attached	Excised	Attached
3rd day count <sup>1</sup>												
50-1266	9	9	9	9	7.5	9	9	9	8.5	9	9	8.5
216-9	9	9	9	9	8.5	9	9	9	6	9	9	6
92-1-1	9	9	9	9	6.5	9	9	9	5	9	9	2.5
AVG. <sup>2</sup>	9	9	9	9	8	8.5	9	9	7.7	9	9	5.8
7th day count <sup>3</sup>												
50-1266	21	21	21	21	15.5	21	21	21	13.5	21	21	13.5
216-9	19	21	21	21	21	18.5	21	21	6	21	21	6
92-1-1	21	21	21	21	21	15	21	21	2.5	21	21	2.5
AVG. <sup>4</sup>	20	21	21	21	19.2	19.2	21	21	10.7	21	21	10.7

<sup>1</sup> L.S.D. at 5% level between clones and temperatures and trifoliolates = 1.93.

<sup>2</sup> L.S.D. at 5% level between averages = 4.28.

<sup>3</sup> L.S.D. at 5% level between clones and temperatures and trifoliolates = 4.40.

<sup>4</sup> L.S.D. at 5% level between averages = 11.13.



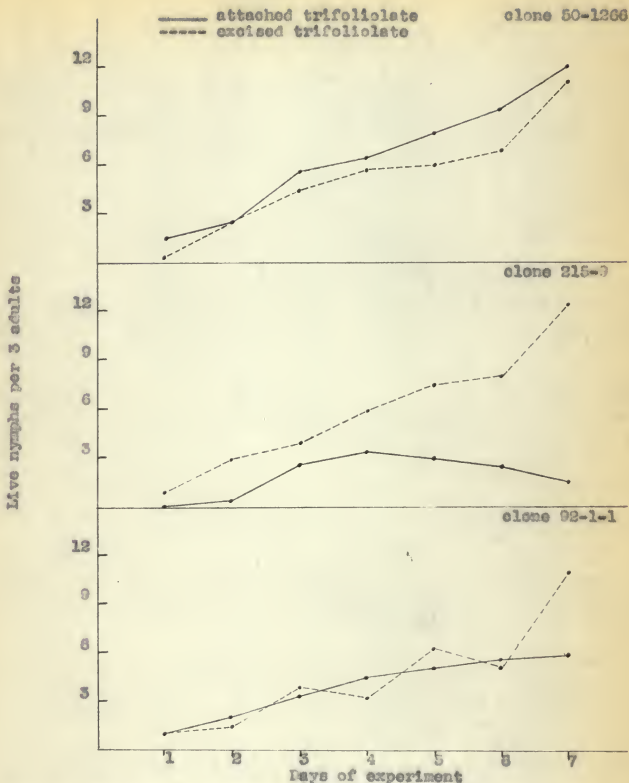


Fig. 7. The average number of live spotted alfalfa aphid nymphs produced by three apterous adults on excised and attached trifoliolates of three selected alfalfa clones at 40° F. This graph represents an average of two replications.

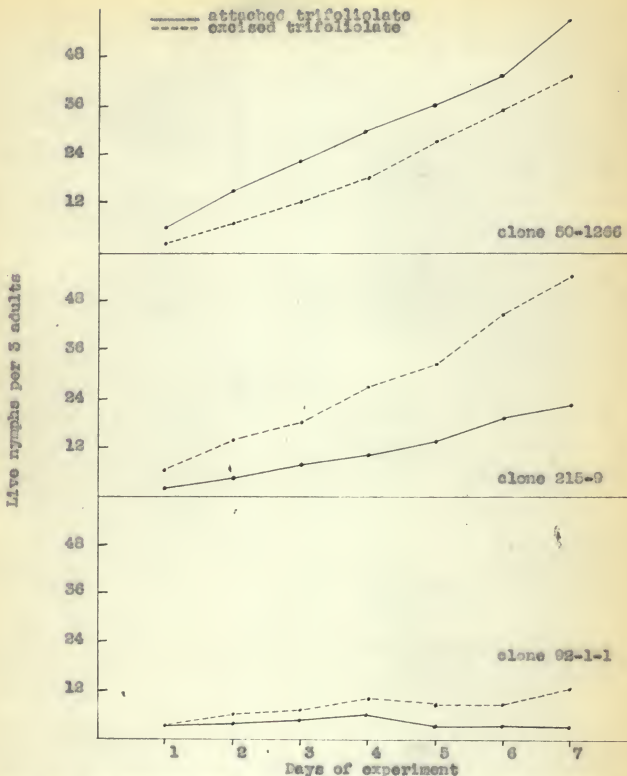


Fig. 8. The average number of live spotted alfalfa aphid nymphs produced by three apterous adults on excised and attached trifoliolates of three selected alfalfa clones at 65° F. This graph represents an average of two replications.

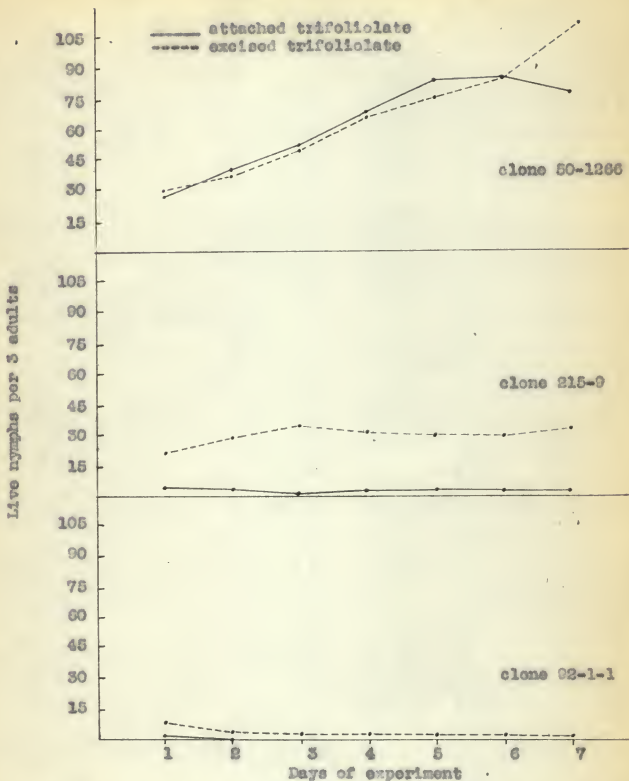


Fig. 9. The average number of live spotted alfalfa aphid nymphs produced by three apterous adults on excised and attached trifoliolates of three selected alfalfa clones at 85° F. This graph represents an average of two replications.

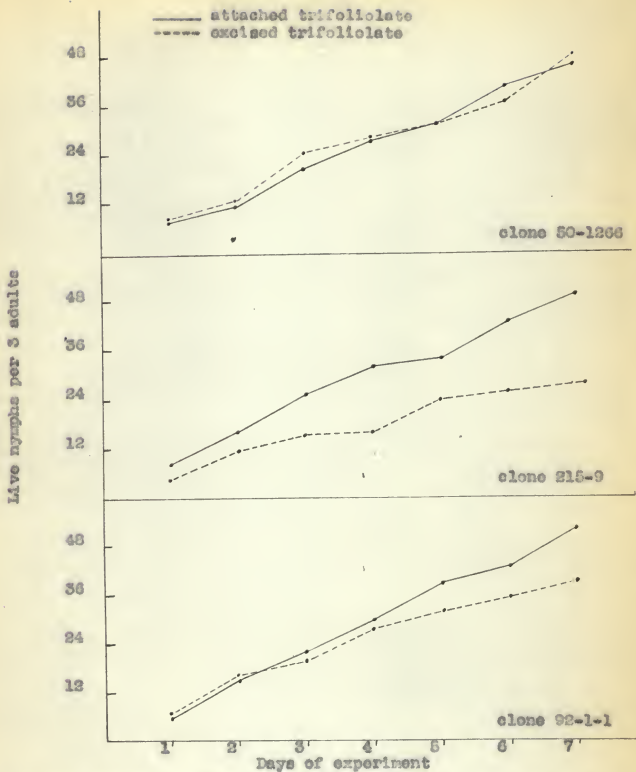


Fig. 10. The average number of live pea aphid nymphs produced by three apterous adults on excised and attached trifoliate leaves of three selected alfalfa clones at 40° F. This graph represents an average of two replications.

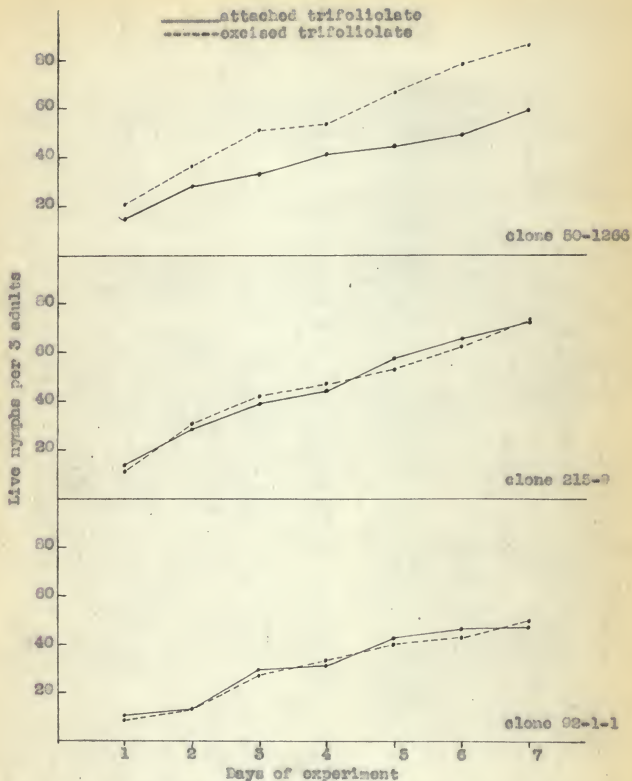


Fig. 11. The average number of live pea aphid nymphs produced by three apterous adults on excised and attached trifoliolates of three selected alfalfa clones at 55° F. This graph represents an average of two replications.

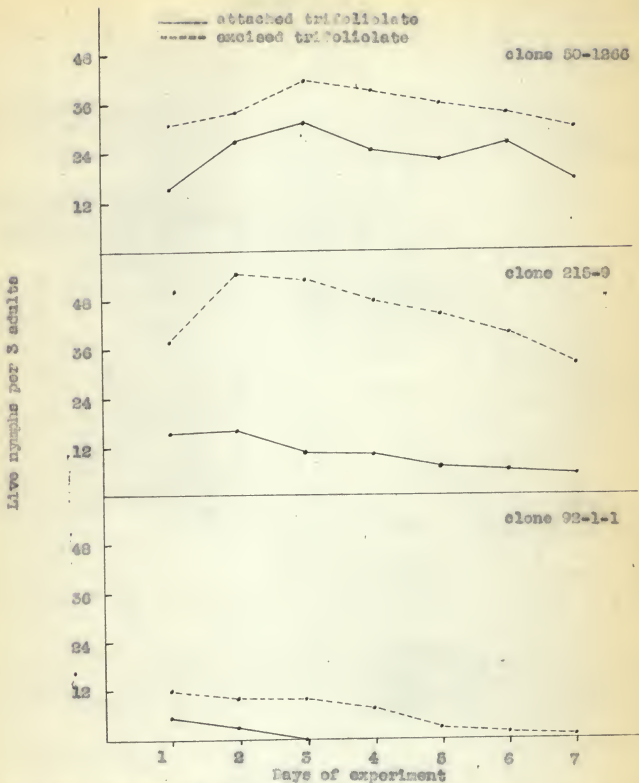


Fig. 12. The average number of live pea aphid nymphs produced by three apterous adults on excised and attached trifoliate leaves of three selected alfalfa clones at 85° F. This graph represents two replications.



trifoliolate on each day of the experiment. Note the wide range in number of nymphs between the clones at 85° F. (Fig. 9).

Biology of the Pea Aphid as Influenced by Excised and Attached Trifoliolates. The average number of pea aphid nymphs per adult day as shown by Table 19 indicated that reproduction of this aphid on excised and attached trifoliolates of all clones was more uniform at 55° F. than at 40° and 85° F. Reproduction was generally higher on all trifoliolates at 85° than at 40°, with the exception of the attached trifoliolates of clone 92-1-1. At 85° F., adult survival was low on all trifoliolates of clone 92-1-1, and on the attached trifoliolates of clone 215-9 and, to a much less extent, 50-1266 (Table 20). The average number of live pea aphid nymphs on the trifoliolates at each day of the experiment is shown by Figures 10-12. From these figures, it is evident that reproduction and survival of the pea aphid was higher at 55° F. than at 40° and 85°. At 85°, aphid reproduction was higher on the excised trifoliolate of all clones than on attached trifoliolates.

The experiments involving excised and attached trifoliolates show that both aphids were able to reproduce and develop at a faster rate on excised trifoliolates at 85° F. than on attached. At 40° and 55° F., differences between numbers of aphids on excised and attached were generally small. As in the temperature and humidity studies discussed previously, the number of nymphs that died increased with temperature during the seven day period.

Graham (1959) indicated that 8.5° C. (47.3° F.) was approaching the minimum temperature at which development of the spotted

alfalfa aphid would be inhibited. The present experiments demonstrated that the spotted alfalfa aphid would reproduce on alfalfa at constant temperatures below 8.5° C. However, it is not known whether the nymphs would develop to maturity since the experiments at 40° F. were conducted for only seven days. Graham (1953) found that the mean nymphal period for the spotted alfalfa aphid at 11° C. (47.3° F.) was 37.62 ± 3.49 days. This would indicate that, providing the nymphs could reach reproductive maturity, the nymphal period at 40° F. might be materially longer.

Pea aphid development at 40° F. was more rapid than development of the spotted alfalfa aphid at the same temperature. The reproductive rate of the pea aphid was quite similar on excised and attached trifoliolates of all clones at 40° F., while there were more variations with the spotted alfalfa aphid.

#### SUMMARY

The purpose of this problem was to compare the influence of temperature and humidity on the biology of the spotted alfalfa aphid, Therioaphis maculata (Duckton), and pea aphid, Macrosiphum pisi (Harris), while feeding on selected alfalfa clones of differing resistance. This problem was initiated because it had been observed in the greenhouse that the results of antibiosis tests on alfalfa clones varied at different times of the year and at different temperatures.

All experiments were conducted in a Percival walk-in plant growth chamber in which the temperature was controlled. A constant light intensity of approximately 1500 foot candles was used.

Varied concentrations of sulfuric acid solutions in battery jars were used to establish the desired relative humidity.

Three clones of the alfalfa variety, Buffalo, were used in all the experiments. These were selected on the basis of the previously recorded reaction of the spotted alfalfa aphid and pea aphid to the plants, and consisted of a resistant (92-1-1), an intermediate (215-9) and a semi-susceptible clone (50-1266). In all experiments, newly formed trifoliolates, the third or fourth from the apex, were used. The trifoliolates were cut off, inserted into small bottles filled with tap water and the bottles placed on a platform inside the battery jar. At the beginning of each experiment three apterous adult aphids were confined in a transparent plastic box attached to each trifoliolate. Two sides of the plastic box were replaced with nylon cloth to permit maximum ventilation. Adult aphids used in the experiments were subjected to a 24 hour pre-conditioning period on the semi-susceptible clone at the experimental temperature.

Experiments were conducted at relative humidities of 25, 50, and 90 per cent at each temperature, 55°, 70°, and 85° F., to study the influence of these conditions on the biology of both aphid species on the three alfalfa clones. Experiments comparing the biologies of both aphids on excised and attached trifoliolates were conducted at 40°, 55°, and 85° F.

Temperature was found to have a greater influence on the biology of both aphid species than humidity. At 55° F., there were no significant differences in average number of nymphs per adult day and total adult days between clones or humidity levels.

An inhibitory influence of high humidity on populations of both aphid species was most evident at 85°, especially on clones 215-9 and 92-1-1.

A temperature of 70° F. was more favorable for pea aphid populations than 85° F. The reproductive rate was similar at both 70° and 85° F., but survival of adults was higher at 70°. The mortality rate increased on all clones as the temperature increased from 55° to 85°.

The number of live spotted alfalfa aphid nymphs per adult day increased on clones 50-1266 and 215-8 as the temperature increased from 55° to 85° F., even though adult survival on all clones was lower at 85° than at 55° or 70°. The rate of reproduction on clone 92-1-1 was generally higher at 70° F. than at 55° or 85°. The mortality rate of this aphid increased on clones 215-9 and 92-1-1 as the temperature increased from 55° to 85°, but on clone 50-1266 the mortality rate was lowest at 70° and highest at 85° F.

The excised and attached trifoliolate studies indicated that populations of both aphid species at 40° and 55° F. were essentially similar in number on excised and attached trifoliolates of all clones. At 85°, however, both aphid populations were distinctly greater on the excised than attached trifoliolates. A temperature of 55° F. appeared to be more favorable for reproduction of the pea aphid than either 40° or 85° F. While reproduction of spotted alfalfa aphids was highest on the excised trifoliolates of all clones at 85° F., adult survival was greater on excised and attached trifoliolates at 40° and 55° F.

The physiology of the trifoliolates is evidently affected as readily by temperature and humidity as is the physiology of the aphids. Cutting off the trifoliolates must also affect their physiology and evidently the level of resistance. Altering the expression of resistance, by exposing aphids and their host plants to various temperatures and humidities, may be caused mainly by a change in the plant itself.



## ACKNOWLEDGMENT

The author wishes to express his sincere appreciation to Dr. E. L. Sorenson of the Department of Agronomy, and Dr. Reginald H. Painter for suggesting the problem and for their guidance and assistance in carrying out the research. Their knowledge and experience in the field of biological research has been of great help while working on the problem.

Appreciation is also expressed to Eldon E. Ortman for invaluable advice and encouragement while making this study and in the preparation of the thesis. Sincere appreciation is due Dr. Herbert Knutson, Head of the Department of Entomology, for making available the facilities and equipment needed, and for reading this thesis.

Thanks are due Floyd J. Hanna, University Photographer, for his services; to Dr. L. Dean Bark and Leo Phillips of the Department of Physics for the construction of apparatus used in measuring relative humidity; and to Dr. Stanley Wearden for statistical evaluation of the data.



## LITERATURE CITED

- Albrecht, H. R., and T. R. Chamberlain. 1941. Instability of resistance to aphids of some strains of alfalfa. Jour. Econ. Ent. 34:551-554.
- Barker, John S., and Oscar E. Tauber. 1954. Fecundity of the pea aphid on garden pea under various combinations of light, moisture and nutrients. Jour. Econ. Ent. 47:113-116.
- Blanchard, Ralph A. 1943. Insect resistance in crop plants. Agron. Jour. 35:718-723.
- Blanchard, R. A., and John E. Dudley, Jr. 1934. Alfalfa plants resistant to the pea aphid. Jour. Econ. Ent. 27:262-264.
- Bodenheimer, F. S., and E. Swirski. 1957. The Aphidoidea of the Middle East. Jerusalem. The Weizmann Science Press of Israel. 378 pp.
- Buxton, Patrick A. 1931. The measurement and control of atmospheric humidity in relation to entomological problems. Bul. Ent. Res. 22:431-447.
- Buxton, P. A., and K. Mellanby. 1934. The measurement and control of humidity. Bul. Ent. Res. 25:171-175.
- Campbell, Roy E. 1926. The pea aphid in California. Jour. Agric. Res. 32:861-881.
- Dehms, F. G., and Reginald H. Painter. 1940. Rate of reproduction of the pea aphid on different alfalfa plants. Jour. Econ. Ent. 33:482-485.
- Davis, J. J. 1915. The pea aphid with relation to forage crops. U. S. D. A. Bul. 276. 67 pp.
- Dickson, R. C., Edward F. Laird, Jr., and George R. Pesho. 1955. The spotted alfalfa aphid. Hilgardia 24:93-118.
- Diehl, R. C., and R. M. Chatters. 1956. Studies on the mechanics of feeding of the spotted alfalfa aphid on alfalfa. Jour. Econ. Ent. 49:589-591.
- Dobson, R. C., and J. G. Watts. 1957. Spotted alfalfa aphid occurrence on seedling alfalfa as influenced by systemic insecticides and varieties. Jour. Econ. Ent. 50:132-135.
- Eichmann, R. D., and E. L. Webster. 1940. The influence of alfalfa on the abundance of the pea aphid on peas grown for canning in southeastern Washington. Wash. Agric. Expt. Sta. Bul. 389. 36 pp.

- Emery, W. T. 1946. Temporary immunity in alfalfa ordinarily susceptible to attack by the pea aphid. Jour. Agric. Res. 73:33-43.
- Graham, H. M. 1959. Effects of temperature and humidity on the biology of Therioaphis maculata (Buckton). Calif. Univ. Pub. in Ent. 16:47-50.
- Hackerott, E. L., and T. L. Harvey. 1959. Effects of temperature on spotted alfalfa aphid reaction to resistance in alfalfa. Jour. Econ. Ent. 52:942-953.
- Harpaz, I. 1955. Bionomics of Therioaphis maculata (Buckton) in Israel. Jour. Econ. Ent. 48:568-571.
- Harvey, T. L., and E. L. Hackerott. 1959. Apparent resistance to the spotted alfalfa aphid selected from seedlings of susceptible alfalfa varieties. Jour. Econ. Ent. 49:289-291.
- Harvey, T. L., E. L. Hackerott, E. L. Sorenson, Reginald H. Painter, E. E. Ortman and D. C. Peters. 1960. The development and performance of Cody alfalfa, a spotted alfalfa aphid resistant variety. Kans. Agric. Expt. Sta. Bul. 114. 26 pp.
- Headlee, T. J. 1914. Some data on the effect of temperature and moisture on the rate of insect metabolism. Jour. Econ. Ent. 7:413-417.
- \_\_\_\_\_. 1917. Some facts relative to the influence of atmospheric humidity on insect metabolism. Jour. Econ. Ent. 10:31-38.
- Hottes, G. C., and T. H. Frison. 1931. The plant lice or Aphidae of Illinois. Ill. Nat. Hist. Sur. Bul. 19:121-447.
- Howe, W. L., and Oliver F. Smith. 1957. Resistance to the spotted alfalfa aphid in Lahontan alfalfa. Jour. Econ. Ent. 50:320-324.
- Kirby, W., and W. Spence. 1843. An introduction to entomology. 6th ed vol. 2. London. 325 pp.
- Maltais, J. B. 1937. Resistance of some varieties of peas to the pea aphid, Illinoia pisi Kalt. Ont. Ent. Soc. Ann. Rpt. 67:40-45.
- Maxwell, Fowden G., and Reginald H. Painter. 1959. Factors affecting rate of honey dew deposition by Therioaphis maculata (Buck.) and Toxoptera graminum (Bond.). Jour. Econ. Ent. 52:368-373.
- McMurtry, J. A., and E. H. Stanford. 1960. Observations of feeding habits of the spotted alfalfa aphid on resistant and susceptible alfalfa plants. Jour. Econ. Ent. 53:714-717.

- Nickel, John L., and Edward S. Sylvester. 1959. Influence of feeding time, stylet penetration, and developmental instar on the toxic effect of the spotted alfalfa aphid. Jour. Econ. Ent. 52:249-254.
- Painter, Reginald H. 1931. Insect resistance in crop plants. New York. The Macmillan Co. 520 pp.
- \_\_\_\_\_. 1958. The study of resistance to aphids in crop plants. Int. Con. Ent. 3, Proc. 3:451-458.
- \_\_\_\_\_, and C. O. Grandfield. 1935. A preliminary report on resistance of alfalfa varieties to pea aphids, Illinois pisi (Kalt). Jour. Amer. Soc. Agro. 27:671-674.
- Palmer, M. A. 1953. Aphids of the Rocky Mountain Region. Lafayette, Ind. Thomas Say Foundation. 452 pp.
- Paschke, John R. 1959. Production of agamic alate forms of the spotted alfalfa aphid, Therioaphis maculata (Buckton). Calif. Univ. Pub. in Ent. 18:125-130.
- Patch, Edith M. 1933. Food-plant catalogue of the aphids of the world including Phylloxeridae. Maine Agric. Expt. Sta. Bul. 393. 431 pp.
- Peters, Don C., and Reginald H. Painter. 1957. A general classification of available small seeded legumes as hosts for three aphids of the "yellow clover complex". Jour. Econ. Ent. 50:231-235.
- \_\_\_\_\_. 1958. Studies on the biologies of three related legume aphids in relation to their host plants. Kans. Agric. Expt. Sta. Tech. Bul. 93. 44 pp.
- Peterson, Alvah. 1953. A manual of entomological techniques. 7th ed. Ann Arbor, Mich. J. W. Edwards. 367 pp.
- Reynolds, H. T., and L. J. Anderson. 1955. Control of the spotted alfalfa aphid on alfalfa in southern California. Jour. Econ. Ent. 48:671-675.
- Schaefer, C. W. 1933. The relation of temperature to the development and reproduction of the pea aphid, Illinois pisi (Kalt.). Unpublished Master's Thesis, Univ. of Wisc. (microfilm copy) 44 pp.
- Searls, E. M. 1932. A preliminary report on the resistance of certain legumes to certain Homopterous insects. Jour. Econ. Ent. 25:46-49.
- Shelford, Victor E. 1949. Laboratory and field ecology. Baltimore. The Williams and Wilkins, Co. 608 pp.

- Simpson, Robert C., and C. W. Burkhardt. 1960. A three-year overwintering study of the spotted alfalfa aphid. Jour. Econ. Ent. 53:220-222.
- Smith, L. B. 1916. The green pea aphid. Va. State Ent. and Plant Path. Rpt. 10:32-63.
- Smith, Roger C. 1931. A study of temperature and humidity conditions in common types of insect rearing cages. Jour. Agric. Res. 43:547-557.
- . 1956. Summary of the unpublished notes in the Department of Entomology on insects affecting alfalfa in Kansas from 1904 to 1956. (original manuscript) 538 pp.
- , and Edgar W. Davis. 1928. The pea aphid as an alfalfa pest in Kansas. Jour. Agric. Res. 33:47-57.
- Snedecor, George W. 1959. Statistical methods. 5th ed. Ames, Iowa. The Iowa State College Press. 534 pp.
- Solomon, M. E. 1951. Control of humidity with potassium hydroxide, sulfuric acid, or other solutions. Bul. Ent. Res. 42:543-554.
- Sorenson, E. L., Reginald H. Painter, E. E. Ortman, H. L. Haeckerott, and T. L. Harvey. 1961. Cody alfalfa. Kans. Agric. Expt. Sta. Circ. 381. 10 pp.
- Stern, Vernon W., and Harold T. Reynolds. 1957. Developments in chemical control of the spotted alfalfa aphid in California, 1955-56. Jour. Econ. Ent. 50:817-821.
- Tuttle, Donald W. 1955. The yellow clover aphid, Univ. of Arizona. Mimeograph Circ.
- Wadley, F. W. 1931. Ecology of *Toxoptera graminum* (Rond.) especially as to the factors affecting importance in the northern United States. Ent. Soc. Amer. Ann. 24:328-395.
- Wilson, Robert E. 1921. Humidity control by means of  $H_2SO_4$  solutions with critical compilation of vapor pressure data. Jour. Indus. and Eng. Chem. 13:328-331.

EFFECTS OF TEMPERATURE AND HUMIDITY ON THE BIOLOGY OF  
THE SPOTTED ALFALFA APHID, THEBIACAPHIS MACULATA  
(BUCKTON), AND THE PEA APHID, MACROSLIPHUM PISI  
(HARRIS), FEEDING ON SELECTED ALFALFA CLONES

by

ALBERT ISAAC

A. B., Tabor College, 1959

---

AN ABSTRACT  
OF A THESIS

submitted in partial fulfillment of the

requirements for the degree

MASTER OF SCIENCE

Department of Entomology

KANSAS STATE UNIVERSITY  
Manhattan, Kansas

1961



The purpose of this problem was to compare the influence of temperature and humidity on the biology of the spotted alfalfa aphid, Therioaphis maculata (Buckton), and pea aphid, Macrosiphum pisi (Harris), while feeding on selected alfalfa clones of differing resistance. This problem was initiated because it had been observed in the greenhouse that the results of antibiosis tests on alfalfa clones varied at different times of the year and at different temperatures.

All experiments were conducted in a Percival walk-in plant growth chamber in which the temperature was controlled. A constant light intensity of approximately 1500 foot candles was used. Varied concentrations of sulfuric acid solutions in battery jars were used to establish the desired relative humidity.

Three clones of the alfalfa variety, Buffalo, were used in all the experiments. These were selected on the basis of the previously recorded reaction of the spotted alfalfa aphid and pea aphid to the plants, and consisted of a resistant (92-1-1), an intermediate (215-3) and a semi-susceptible clone (50-1266). In all experiments, newly formed trifoliolates, the third or fourth from the apex, were used. The trifoliolates were cut off, inserted into small bottles filled with tap water and the bottles placed on a platform inside the battery jar. At the beginning of each experiment three apterous adult aphids were confined in a transparent plastic box attached to each trifoliolate. Two sides of the plastic box were replaced with nylon cloth to permit maximum ventilation. Adult aphids used in the experiments were subjected to a 24 hour pre-conditioning period on the semi-



susceptible clone at the experimental temperature.

Experiments were conducted at relative humidities of 25, 50, and 90 per cent at each temperature, 55°, 70°, and 85° F., to study the influence of these conditions on the biology of both aphid species on the three alfalfa clones. Comparison of the biologies of both aphids on excised and attached trifoliolates were conducted at 40°, 55°, and 85° F.

Temperature was found to have a greater influence on the biology of both aphid species than humidity. At 55° F., there were no significant differences in average number of nymphs per adult day and total adult days between clones or humidity levels. An inhibitory influence of high humidity on populations of both aphid species was most evident at 85°, especially on clones 215-9 and 92-1-1.

A temperature of 70° F. was more favorable for pea aphid populations than 85° F. The reproductive rate was similar at both 70° and 85° F., but survival of adults was higher at 70°. The mortality rate increased on all clones as the temperature increased from 55° to 85°.

The number of live spotted alfalfa aphid nymphs per adult day increased on clones 50-126 and 215-9 as the temperature increased from 55° to 85° F., even though adult survival on all clones was lower at 85° than at 55° or 70°. The rate of reproduction on clone 92-1-1 was generally higher at 70° F. than at 55° or 85°. The mortality rate of this aphid increased on clones 215-9 and 92-1-1 as the temperature increased from 55° to 85°, but on clone 50-126 the mortality rate was lowest at 70° and

highest at 85° F.

The excised and attached trifoliolate studies indicated that populations of both aphid species at 40° and 85° F. were essentially similar in number on excised and attached trifoliolates of all clones. At 85°, however, both aphid populations were distinctly greater on the excised than attached trifoliolates. A temperature of 55° F. appeared to be more favorable for reproduction of the pea aphid than either 40° or 85° F. While reproduction of spotted alfalfa aphids was highest on the excised trifoliolates of all clones at 85° F., adult survival was greater on excised and attached trifoliolates at 40° and 55° F.

The physiology of the trifoliolates is evidently affected as readily by temperature and humidity as is the physiology of the aphids. Cutting off the trifoliolates must also affect their physiology and evidently the level of resistance. Altering the expression of resistance, by exposing aphids and their host plants to various temperatures and humidities, may be caused mainly be a change in the plant itself.