CERTAIN FACTORS AFFECTING THE RATE OF HONEYDEW DEPOSITION FROM TWO SPECIES OF APHIDS, *TOXOPTERA GRAMINUM* (ROND.), AND *THERIOAPHIS MACULATA* (BUCK.), WHILE FEEDING ON HOST PLANTS WHEAT, BARLEY AND ALFALFA

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INTRODUCTION

The excretion of honeydew by aphids and other plant-sucking insects has been widely known for centuries. Honeydew is unique in that (1) the water soluble carbohydrates, mainly sugars, may exceed 80 per cent of the total weight of the fresh material, (2) the composition is very complex, containing not only unchanged plant-sap materials, but synthetic components as well, and (3) honeydews usually occur as sticky syrups, only rarely forming crystals. Honeydew when deposited in large quantities becomes a serious detriment to agriculture, especially in the harvesting of certain specialized crops like alfalfa and sorghums where cutting or threshing is necessary. The honeydew deposited on the plant surface also furnishes a substrate for fungus growth, interfering with photosynthesis and normal pollination of the flowers. It also may attract other insects both harmful and useful.

Considerable technical biochemical analysis has been undertaken in recent years in an effort to determine the composition of honeydew as a tool in a study of the nutritional needs of aphids. Virtually no information is available concerning the factors affecting the rate of honeydew deposition or of methods for determining the rate of excretion.

It is the purpose of this research to study certain factors which affect the rate of honeydew deposition by the greenbug, Toxoptera graminum (Rond.), and the spotted alfalfa aphid, Therioaphis maculata (Buck.). Variety of host, temperature and light intensity constituted three of the more important factors considered. The varieties studied were selected because of known variations in degrees of resistance to the greenbug or spotted alfalfa aphid. It was therefore of interest to compare the rate of honeydew deposition by the aphids while feeding on these varieties with their resistance rating.

The methods used in evaluating the rate of honeydew deposition were entirely new; therefore, much of the background experimental work involved the perfecting of procedures that would eliminate as many sources of variability as possible.

Rate of honeydew deposition as used herein is defined as the number of honeydew droplets excreted by an individual aphid during a 24-hour period.

REVIEW OF LITERATURE

Probably the most remarkable feature of honeydew production is the amount produced in relation to the diminutive size of the insects involved. Michel (1942) in studies with Lachmus robaris (L.) on oak measured an average of 5.0 and up to 8.6 milligrams of honeydew per aphid during a twenty-four hour period. The maximum amount is considered to be more than the weight of the insect producing it. Day and Irzykiewicz (1953) estimated from studies with radio-phosphorus that Myzus persica (Sulz.) may ingest 35 per cent of its weight in one hour. Smith (1937) used the rate of deposition of honeydew to measure the rate of feeding of Hyalopterus pruni (Geoff.). The drops of honeydew were caught on sheets of paper coated with bromo-cresol green and mounted on revolving clocks below the feeding aphids. Smith found that the rate of excretion increased with increasing temperatures. Schaefer (1938) found that more honeydew was produced by uncrowded rather than crowded aphids. The effect of evaporation on rate of excretion of honeydew was studied by Broadbent (1951). He found that the rate of excretion was higher in still air than in wind. Broadbent also stated that in twenty-two species of aphids observed none excreted honeydew except when feeding; feeding and excretion continued in darkness, nymphs did not excrete during ecdysis and adults continued at their usual rate when producing young.

The Bionomics of the Greenbug

Rondani first described the greenbug in 1852. Passerini placed it in the genus Toxoptera in 1863. Triticum, Hordeum, Avena, Zea, and Sorghum were mentioned as its food plants by Passerini. Webster and Phillips (1912) listed approximately sixty host species, almost all from the grass family. The greenbug was first recorded in the United States by Pergande (1902) in June, 1882. Dahms (1951) indicated that there have been fourteen major outbreaks in the United States. Rietz (1954) listed the greenbug as one of the five major insects attacking wheat.

Webster and Phillips (1912) described the biology of the greenbug. Luginbill and Beyer (1918) studied the seasonal cycle and life history of the greenbug. Wadley (1931) made extensive ecological studies. A thorough study of the effects of temperature on the greenbug was made by Wadley (1935). Peters (1955) studied the reaction of the greenbugs to small grains, particularly the principal wheat and barley varieties recommended for Kansas. Ortman (1957) made an extensive study of greenbug damage to the root systems and other plant parts of several varieties of wheat.

The Bionomics of the Spotted Alfalfa Aphid

Dickson et al. (1955) reported that the spotted alfalfa aphid has been in the United States at least since February of 1954. Smith (1956) reported its presence in fourteen states. Before 1957, sixteen additional states had reported infestations (U. S. D. A. News Release). Harvey and Hackerott (1956) first reported the spotted alfalfa aphid in Kansas in August, 1954. By late 1955 it had spread to all areas of the state.

The biology of the spotted alfalfa aphid in relation to its host plants has been studied by Peters and Painter (1958).

The Greenbug and Resistance

Painter (1951) compiled and summarized the studies of insect resistance. He listed three main categories of resistance and reports that the greenbug does more damage in proportion to numbers than any other aphid on wheat. Sajo (1894) was first to note possible differences in tolerance among oat varieties to the greenbug. The reactions of greenbugs to several varieties of common durum, and emmer wheat were reported by Wadley (1931). Fenton and Fisher (1940) found that winter barley was more susceptible than winter wheat and that spring oat varieties were injured more severely than the winter wheat and barley. Blizzard (1948) found that Omugi barley from Korea showed exceptional resistance to greenbugs. In Uruguay, Silveirie and Conde (1946) reported differences in susceptibility among varieties of wheat and rye. Dahms (1948) has compared fifteen small grains and noted varietal difference. Fenton and Dahms (1940) observed differences in susceptibility between wheat, barley and oats during the 1939 outbreak. Atkins and Dahms (1945) reported observations on varietal resistance in wheat, oats and barley gathered during the outbreak of 1942. Dahms et al. (1955) reported a selection of the Dickinson wheat variety to be resistant.

The Spotted Alfalfa Aphid and Resistance

Differences among alfalfa varieties in reaction to the spotted alfalfa aphid were reported by Harpaz (1955). Stanford (1955) reported that the spotted alfalfa aphid could not survive on the variety Lahontan. It was reported exceedingly injurious to all commercial varieties of alfalfa in California except Lahontan by Reynolds and Anderson (1955). Howe and Smith (1957) reported on the apparent resistance to the spotted alfalfa aphid in Lahontan alfalfa. Harvey and Hackerott (1956) reported apparent resistance to the aphids of some seedlings selected from susceptible varieties.

Biology of Honeydew Deposition

There are few references in the literature to methods by which honeydew is excreted. Kirby and Spence (1843) stated: "When no ants attend them, by a certain jerk of the body, which takes place at regular intervals, they ejaculate to a distance". Buckton (1876) described honeydew excretion as the forcible ejection of the liquid from the anus. He also reported that Lachnus saligna (Gmelin) erects the terminal rings of their bodies, upon the apex of which a clear drop of fluid will appear. If this is not quickly withdrawn by an attendant ant, it is projected by a peculiar jerk to a considerable distance. Weber (1930) found that in Homoptera very diverse structures have been developed which facilitate excretion and which allows the stream of excrement to take a safe direction away from the insect preventing entanglement with the liquid. Weber indicated that in aphids the anus forms a posteriorly directed valve, the dorsal region usually being drawn out into a tail or cauda. He said that the cauda is raised by special muscles, the anus opened, and the honeydew driven out by the musculature of the rectum. Smith (1937) described the method of excretion of honeydew by Hyalopterus prunis (Geoff.) in these words: "The tip of the abdomen is raised, a globule of transparent colourless liquid is secreted and held for an instant on the tip of the abdomen. The left posterior leg is then swung upward and outward. The drop of excrement is caught by the tip of the tibia and the base of the tarsus and thrown with considerable force. From a height of six inches females frequently throw excrement laterally for a distance of 6 to 8 inches. The mass of each droplet and force and direction of the throw are remarkably uniform". Broadbent (1951) indicated that Smith may have observed only the nymphs.

In studies with 22 species of aphids he indicated that the adult aphids, "raised their abdomen in the air and that the cauda and cornicles rose until they were almost at right angles to the body; though they occasionally waved their posterior legs a few times before the globule appeared, they kept them still while the globule remained poised on the anus for one or two seconds, prior to being flicked off by the cauda. The cauda was flexed back dorsally and then suddenly returned to its normal position, striking the globule as it did so."

Observations of the adult greenbug and spotted alfalfa aphid while excreting honeydew indicate that reactions during deposition were very similar to Broadbent's descriptions.

Some ideas have been advanced in an attempt to explain why such copious amounts of honeydew are excreted by the aphids and by what means the sap is moved into and through the aphids' alimentary tract in such large quantities. Interesting theories have been advanced, but perhaps the most interesting explanation at present is that advanced by Kennedy and Mittler (1953). They found that if the mouthtube of an actively feeding aphid was carefully severed, a considerable amount of fluid continued to ooze from the cut end of the tube remaining in the plant tissue. Similar rates of honeydew excretion were recorded from adjacent intact aphids. It was therefore suggested that during the feeding process, plant turgor pressure forces large amounts of sap, in the form of honeydew, through the insect's alimentary system. Mittler (1957) in studies with Tuberolachnus salignus (Gmelin) elaborated further on this theory. Leonardt (1940) also reported an apparent relationship between osmotic pressure variations in fir trees and the rate of production of honeydew. Waterhouse (1957) reports that if Kennedy and Mittler's theory proves to be of general applicability the function of the filter chamber in aphids may be to detour much of the water of this continuous flow of fluid, thereby reducing not only the volume, but also the rate of flow of material through the region of the midgut where digestive enzymes have to function.

The possibility of using the rate of honeydew excretion by aphids as a measure of metabolism has not been investigated but presents interesting possibilities. Pathak and Painter (1958) found that the corn leaf aphid, *Rhopalosiphum maidis* (Fitch), lost weight on resistant Piper Sudan 428-1 due to a reduction in the amount of plant material ingested. Cartier and Painter (1956) indicated that the fecundity of the corn leaf aphid was reduced significantly while feeding on resistant Piper Sudan 428-1. Peters and Painter (1958) found that the weights of the spotted alfalfa aphid, *Therioaphis maculata* (Buck.) varied considerably with the host plants. Peters (1955) indicated that the greenbug, *Toxoptera graminum* (Rond.), lost weight while feeding on resistant Dicktoo barley and Dickinson wheat. He also noted a decline in the fecundity rate of the greenbug while feeding on Dicktoo barley.

The apparent relation of host resistance to the amount of plant material ingested suggests that resistance depresses the feeding rate and should in theory reduce the rate of honeydew deposition.

Preliminary studies conducted indicated that not only resistance but many other factors as well may significantly reduce the rate of honeydew deposition, conceivably through a reduction in the rate of feeding. If the rate of honeydew deposition could be used as a plausible measure of the rate of feeding, it could also be used as a possible measure of metabolic activity of the aphid. This possibility is further enhanced by indications that the optimum conditions for reproduction and development are also the optimum for honeydew production and that differences from this optimum condition can be measured by the increase or reduction in the rate of honeydew deposition. The optimum conditions for growth and reproduction are frequently used as a measure of metabolic activity. The deviations in metabolic activity as brought about by various factors could be detected by the corresponding fluctuations in the rate of honeydew production thus providing a crude, but simple means of measuring metabolic activity.

MATERIALS AND METHODS

The materials used were those necessary for collecting the honeydew droplets excreted by the aphids while feeding on wheat, barley and alfalfa plants and for controlling, as much as possible, the known variables related to the several experiments.

The greenbugs used in experiments with wheat and barley plants were from a stock culture grown under artificial light in the basement of Insectary No. 2. All of the greenbugs used were progeny of one female collected in September, 1954 from a stock culture in the Entomology greenhouse (Peters, 1955). There were no known sources from which this culture might have been contaminated by other aphids. The aphids used for honeydew studies were usually chosen from a single flat in order to insure homogeneity of environment.

The spotted alfalfa aphids used in the experiments with alfalfa 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 from different regions of Kansas. Since the original formation of the culture, spotted alfalfa aphids collected from fields near Manhattan, Kansas have been periodically added during the summer months.

Apterous adults of both the greenbug and spotted alfalfa aphid were used in all of the experiments except in the study of the effect of age on the rate of honeydew deposition by the greenbug. In this particular experiment nymphs of all ages were used. Apterous adults were used because they could be distinguished from large nymphs by the exsertion of the cauda which occurs at the fourth molt. A Dazor Magnifier lamp was used to select the apterous nymphs.

Four varieties of wheat, _Triticum vulgare_ L., and one group of F_4, Dickinson x Pawnee hybrids, representing several test lines carrying varying degrees of resistance, were used in the experiments with the greenbug. The seed of the varieties Pawnee, Ponca, Bison, Dickinson, and F_4 Dickinson x Pawnee hybrids were obtained from the Agronomy Department.

Three varieties of barley, Hordeum vulgare L., were used in several of the experiments with the greenbug. Seed of the varieties Reno and Dicktoo was obtained from the Agronomy Department. Seed of the variety MO-B475, developed in Missouri, was obtained from the Fort Hays Experiment Station.

Wheat and barley seedlings were grown either in sixteen by twenty-two inch greenhouse flats, two rows to the flat, or in 4-inch clay pots. The soil used was a mixture of six parts black loam, one part sheep manure, and one part sand. The plants were generally two to three weeks old when the tests were begun.

Clones of nine different alfalfa varieties, Medicago sativa L., used in the honeydew experiments with the spotted alfalfa aphid were obtained from the Agronomy Department. Cuttings were made from plants in the field during November, 1957 and potted in 4-inch clay pots. The potted plants were maintained during the period of the experiments in the Entomology greenhouse. No alfalfa seedlings were used in any of the experiments. Clones differ from seedlings in that they are individual plants descended by asexual reproduction from sexually produced plants.

All experiments involving wheat and barley, except the ones on effect of temperature, were conducted in the basement of Insectary No. 2 using 40 watt standard fluorescent tubes placed approximately eighteen inches above the table with four such tubes per eight foot square area. The lights were on 16 hours a day. A Friez Hygrothermograph was used to record the temperature and relative humidity. Average temperature for the period of the experiments was approximately 75°F. Relative humidity averaged approximately 45 per cent. It was comparatively easy to control and maintain the temperature and relative humidity due to the nature of the environment. In the cold months an electric coil heater with thermostat control was used.

All honeydew experiments with the spotted alfalfa aphid, except effect of temperature on the rate of honeydew deposition, were conducted in the Entomology greenhouse. Temperature in the greenhouse during period of the experiments averaged approximately 74°F. with a relative humidity average of approximately 35 per cent. Temperature was maintained by a thermostatically controlled steam circulating heater. During the summer months the greenhouse was cooled with evaporative type equipment. Air was pulled through the greenhouse by a large window fan. In experiments where additional light was required, 40 watt standard white fluorescent tubes were used.

The experiments involving temperature studies with the greenbug and spotted alfalfa aphid were conducted in a 4 x 6 foot cabinet with thermostatically controlled temperatures located in the basement of the Agronomy Plant Research Laboratory. Constant 40 watt standard fluorescent lighting was used in the experiments. Temperatures ranged within 2°F. of preset temperatures. Humidity varied from 40 to 80 per cent due to the different temperatures used. In the experiments with wheat and barley, eleven different temperatures representing five degree intervals from 35°F. to 85°F. were used. In the experiments with alfalfa, six different temperatures representing 10°F. intervals from 35°F. to 85°F. were used.

Most of the experiments with wheat, barley and alfalfa followed a general pattern. Where this general method was modified to a certain degree to fit the situation, the modified methods will be given under a separate heading, Special Experiments.

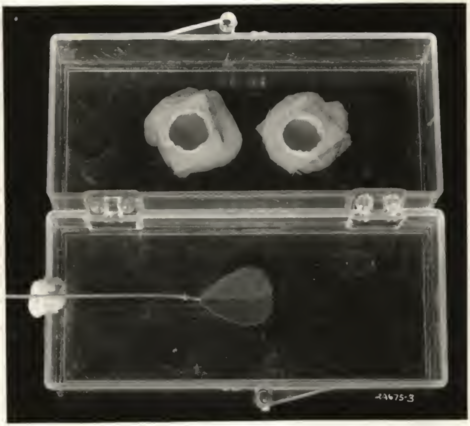
Plastic boxes were adapted to make aphid cages and to provide an efficient means of collecting the droplets of honeydew. Plate I illustrates the 1 x 1¼ x 3 inch cages which had two hinges on the back and one snap type fastener on the front to hold the cage closed. Two holes drilled in the tops of these cages were covered with fine mesh nylon cloth. Boxes used for caging the spotted alfalfa aphids on alfalfa were notched with a v-shaped groove in one end of the box large enough to insert the leaf petiole with a damp cellulose plug for protection of the petiole. Two leaflets of the trifoliate leaf were removed to eliminate the possibility of the honeydew droplets landing on them instead of the plastic cage (Plate I). The plug was put into the groove and the lid closed with the petiole in the center of the cellulose pad. The clear plastic sides made it possible to see the aphids and the deposited honeydew droplets in the cage at all times. The nylon mesh windows allowed for some transpiration from the leaves without condensation of water inside the cage. In adapting the boxes for caging greenbugs on wheat and barley, a groove was cut in each end of the cage, cellulose plugs placed therein and the leaf passed completely through the cage. Plate II illustrates this method.

The cages were held in place while in use by fastening them to ½-inch-square wooden stakes of convenient lengths with No. 32

EXPLANATION OF PLATE I

Illustration of the arrangement of the petiole and leaflet in the 1 x $1\frac{1}{4}$ x 3 inch plastic box used for caging the spotted alfalfa aphid and providing a method for collecting the droplets of honeydew.

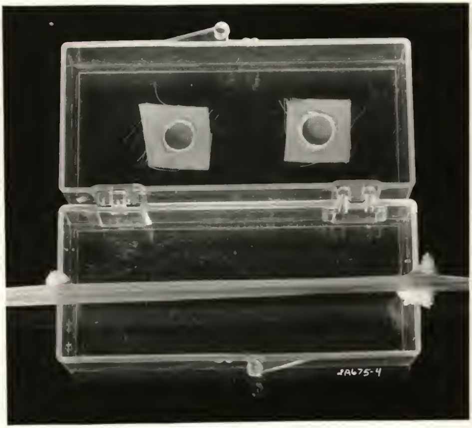
PLATE I



EXPLANATION OF PLATE II

Illustration of the arrangement of the leaf blade of barley and wheat in the 1 x 1½ x 3 inch plastic box used for caging the greenbug and collecting the droplets of honeydew.

PLATE II



rubber bands. In all of the studies with alfalfa, except the experiments on the effect of flowering and effect of distance of leaves from growing point of host plant, the second or third trifoliate leaf from the terminal bud was selected in order to maintain as much uniformity as possible (Plate III). In the experiments with wheat and barley the second leaf was used.

Considerable preliminary investigations were made prior to the adoption of the plastic cage method of collecting honeydew. Experiments were conducted using twenty-five different types of paper. Several chemically treated papers, bromo-cresole green, potassium iodide, and aniline hydrogen phthalate, were investigated for possible use. All of these papers proved to be unsatisfactory primarily because of the inability to distinguish individual droplets after coalescence with neighboring droplets. The possibility of measuring the excreted honeydew by the use of minute pipettes was discarded because of the highly viscous nature of the excrement. Dry weight as a measure of honeydew excretion was attempted but results were erratic making an accurate measurement impossible.

The plastic cage method was developed as a technique after honeydew droplets were seen to appear very distinctly against the clear plastic of the cage. The honeydew droplets retained their convexity and very little coalescence occurred. The attraction for the droplets was undoubtedly due to the physical and chemical composition of the plastic cages because the honeydew droplets would spread and be indistinguishable when collected on glass slides. Plates IV and V show cages with typical honeydew droplet patterns.

EXPLANATION OF PLATE III

Illustration of the arrangement and method of attachment of the plastic cages on the alfalfa plants.

PLATE III

EXPLANATION OF PLATE IV

Illustrations of typical honeydew patterns occurring on plastic cages. Magnified 10X.

PLATE IV

EXPLANATION OF PLATE V

Illustrations of typical honeydew patterns occurring on plastic cages. Magnified 10X.

PLATE V

The different variations in the sizes of the droplets are clearly illustrated in these plates.

All experiments with alfalfa, barley, and wheat were conducted for a 24-hour period; at the end of this interval of time the cages were removed and the honeydew droplets counted with the aid of a Dazor Magnifier lamp.

The aphids were selected from the cultures with an aspirator or a camel's hair brush. Usually one aphid was used per cage, but on several resistant clones of alfalfa as many as ten aphids per cage were used in order to obtain one live aphid that settled in feeding position. An adequate interval of time was allowed in all of the experiments for the aphids to adjust to the plant and begin feeding. This procedure eliminated a possible "carry over" effect from other varieties.

The statistical analysis of variance used in computing Least Significant Difference for all data was in accordance with Snedecor (1938).

SPECIAL EXPERIMENTS

<u>Area of Feeding on Reno Barley Leaf as a Factor Affecting the Rate of Honeydew Deposition by the Greenbug</u>. In this experiment the methods were modified in order to restrict the aphids to particular regions of the barley leaf. The plastic cages were notched on each side rather than on the ends, and the leaf passed across through the cage. Four cages were spaced evenly along the 8-inch barley leaf at approximately 2-inch intervals. The

cages were removed at the end of each 24-hour period and the droplets of honeydew recorded. Nymphs were removed at this time and the cages cleaned and reinserted on the leaf. Plate VI shows the arrangement of the cages on a barley leaf. The cages were not suspended by rubber bands but were placed on a small wooden platform which was at the same level as the leaf.

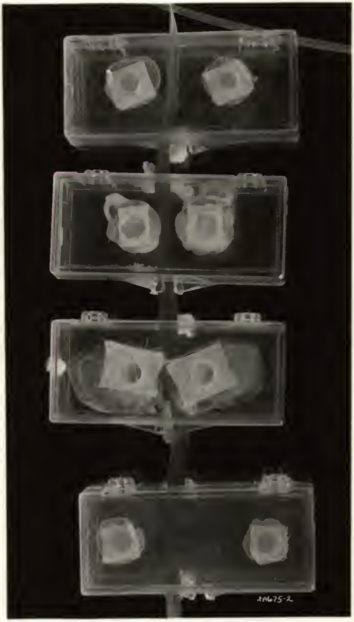
Age of Aphid as a Factor Affecting the Rate of Honeydew Deposition by the Greenbug. Three potted Reno Barley plants were used as the hosts during this experiment. Several apterous greenbug females were isolated in a plastic cage, allowed to feed on a Reno barley plant and reproduce. The cage was checked every three hours during one day and the newly born nymphs removed singly to cages which were then labeled with the time and date, and placed on the second leaf of each potted Reno barley plant. The cages were examined at twenty-four hour intervals throughout the seven days of the experiment. The rate of honeydew deposition for each nymph was recorded until reproduction was observed to occur. Molts were recorded when noticed.

Reduction in Light Intensity as a Factor Affecting the Rate of Honeydew Deposition by the Spotted Alfalfa Aphid. In the first portion of the experiment a large 2½ x 3 foot cardboard box was used to prevent part of the light from reaching the alfalfa plants. This was accomplished by cutting the end out of the box and then preparing a sliding mechanism out of this cut end to regulate the amount of light reaching the plants. The source of light for both the plant in full light and that in restricted light was from two regular 40 watt daylight fluorescent tubes.

EXPLANATION OF PLATE VI

The arrangement of the cages on a Reno barley leaf to restrict the greenbugs to definite areas of feeding.

PLATE VI



In the second portion of the experiment aluminum foil was used to restrict the amount of light entering the individual small cages by wrapping the cages in the foil, allowing light to enter only through the two small nylon mesh covered windows.

The Distance of Leaves from Growing Point of Host Plant as a Factor Affecting the Rate of Honeydew Deposition by the Spotted Alfalfa Aphid. A branch of B16 alfalfa clone, 24 inches in height, was used in this experiment as the host plant. Ten cages were placed on trifoliate leaves occurring at varying intervals going from the first trifoliate leaf below the terminal bud downward to the base of the plant. Other procedures were as previously described for other experiments.

Leaf, Stem, and Petiole Feeding as a Factor Affecting the Rate of Honeydew Deposition by the Spotted Alfalfa Aphid. Methods were identical to other experiments except that the leaves were removed from the petiole thereby forcing the aphids to feed on this specified area. The stems were prepared by removing the leaves and petioles and placing the cages on the bare stems.

Reduced Moisture as a Factor Affecting the Rate of Honeydew Deposition by the Spotted Alfalfa Aphid. The only modification in procedure involved in this experiment was the amount of water allowed the host plants. Three potted B16 alfalfa plants, 18 inches in height, were used. One plant was held at the point of saturation, a second with adequate moisture for normal growth, and the third was maintained as nearly as possible at the incipient wilting stage. These conditions were maintained for one twenty-four hour period only because of the rapid senescence of the third plant.

RESULTS

Greenbugs on Wheat and Barley

Temperature as a Factor Affecting the Rate of Honeydew Deposition of the Greenbug while Feeding on Pawnee, and Dickinson Wheats, and Reno and Dicktoo Barleys. Highly significant increases in the rate of honeydew deposition by the greenbug while feeding on Pawnee were found to occur between all 5°F. temperature intervals from 40°F. to 80°F. No significant increase in the rate of honeydew deposition was found to occur between 35°F. and 40°F. A highly significant decrease in the rate of honeydew deposition occurred between 80°F. and 85°F. The lowest rate of honeydew deposition occurred at 35°F. (4.3 droplets), the highest at 80°F. (28.1 droplets). Table 1 summarizes the results obtained with this variety. Figure 1 gives a graphic comparison with Dickinson wheat, Reno and Dicktoo barleys.

In Dickinson wheat the rate of honeydew deposition was much lower than Pawnee at all temperatures. Significant differences were apparent in Pawnee between 5°F. temperature intervals from 40°F. upward. In Dickinson, non-significant increases in the rate of honeydew deposition between the 5°F. temperature intervals were apparent from 35°F. to 60°F.; however, highly significant differences were recorded in 10°F. intervals between 35°F.

and 65°F. Highly significant differences were found to exist between the 5°F. intervals from 60°F. to 80°F. The rate of honeydew deposition by the greenbug was found to drop very significantly as the temperature increased from 80°F. to 85°F. The highest rate of honeydew excretion was recorded at 80°F. (Table 2 and Fig. 1).

The rate of honeydew deposition by the greenbug was higher on Reno barley than on any other cereal variety tested. Average droplet rate varied from a low of 9.3 at 35°F. to a high of 40.3 at 75°F. No significant difference at the .001 per cent level was observed between 35°F. and 40°F. Highly significant differences were obtained between all 5°F. temperature intervals from 45°F. to 75°F., except at the 50°F. to 55°F. interval. In this one exception the difference approached significance. A peak in the rate of honeydew deposition was reached at 75°F. in this variety. A highly significant decline in the rate of honeydew deposition occurred at temperatures of 80°F. to 85°F. (Table 3 and Fig. 1).

In Dicktoo barley there was a non-significant difference between the average rate of honeydew deposition at temperatures of 35°F. and 40°F. Significant differences at the .001 per cent level occurred between all 5°F. temperature intervals from 40°F. to 75°F. In Dicktoo, as in Reno, a peak in the rate of honeydew deposition occurred at 75°F. A significant decline was recorded at temperatures of 80°F. to 85°F. (Table 4 and Fig. 1).

Table 1. Temperature as a factor affecting the rate of honeydew deposition by the greenbug, *Toxoptera graminum* (Rond.), while feeding for a 24 hour period on Pawnee wheat under constant fluorescent lighting.

Replication Number	Number of honeydew droplets at 35°F.	40°F.	45°F.	50°F.	55°F.	60°F.	65°F.	70°F.	75°F.	80°F.	85°F.
1	6	7	8	12	16	18	19	25	26	28	25
2	8	6	10	14	18	15	17	24	25	26	28
3	3	5	9	11	14	19	26	26	27	27	26
4	5	7	8	13	16	16	24	21	25	27	20
5	2	5	7	11	13	14	19	24	28	32	28
6	2	4	8	13	12	18	21	19	26	34	27
7	3	5	11	10	15	19	20	24	29	30	25
8	6	6	9	14	12	21	20	26	27	26	25
9	5	8	6	12	17	16	21	28	28	29	29
10	4	5	10	9	16	17	25	26	25	28	27
11	5	4	12	12	18	17	28	26	29	27	24
12	3	3	11	15	14	16	22	28	27	24	21
Tot.	52	65	109	146	181	206	262	297	322	338	305
Avg.	4.30	5.41	9.08	12.16	15.10	17.10	21.80	24.70	26.80	28.10	25.40

***L.S.D. = 1.804 at .001 per cent level.

Table 2. Temperature as a factor affecting the rate of honeydew deposition by the greenbug, _Toxoptera graminum_ (Rond.), while feeding for a 24 hour period on Dickinson wheat under constant fluorescent lighting.

Replication Number	Number of honeydew droplets at 35°F.	40°F.	45°F.	50°F.	55°F.	60°F.	65°F.	70°F.	75°F.	80°F.	85°F.
1	2	4	10	11	11	10	11	14	16	17	16
2	5	8	7	8	9	10	13	12	17	16	15
3	6	5	11	9	5	8	12	13	15	23	23
4	7	6	8	7	10	9	13	14	18	19	20
5	3	6	6	12	10	10	10	13	16	16	14
6	5	8	7	10	12	9	11	15	15	18	27
7	4	5	6	7	9	7	12	14	17	20	17
8	5	7	4	6	8	12	10	16	15	22	20
9	5	7	7	6	8	10	15	13	16	17	19
10	3	4	6	7	10	11	11	12	18	19	13
11	4	5	8	6	8	9	11	12	16	24	15
12	4	4	5	9	7	8	10	16	15	20	16
Tot.	53	69	85	98	107	113	139	164	194	231	215
Avg.	4.40	5.70	7.08	8.10	8.90	9.40	11.50	13.60	16.10	19.20	17.90

***L.S.D. = 1.668 at .001 per cent level.

Table 3. Temperature as a factor affecting the rate of honeydew deposition by the greenbug, Toxoptera graminum (Rond.), while feeding on Reno barley for a 24 hour period under constant fluorescent lighting.

Replication Number	Number of honeydew droplets at 35°F.	40°F.	45°F.	50°F.	55°F.	60°F.	65°F.	70°F.	75°F.	80°F.	85°F.
1	10	12	16	17	17	20	30	40	41	36	27
2	8	10	15	19	20	29	29	36	39	38	31
3	9	10	14	15	19	21	26	33	40	33	30
4	13	8	12	14	18	19	25	36	42	35	40
5	10	9	13	18	20	18	27	38	44	38	35
6	7	7	12	16	14	15	35	41	38	42	26
7	9	8	9	13	16	21	36	37	36	45	33
8	10	10	11	15	20	19	30	35	38	40	28
9	11	8	10	13	15	23	25	36	45	43	31
10	7	12	13	15	19	25	26	38	42	45	37
11	8	11	11	19	18	24	25	34	40	39	34
12	10	10	12	20	22	23	28	40	39	39	36
Tot.	112	115	148	194	218	257	342	444	484	473	388
Avg.	9.30	9.58	12.30	16.10	18.10	21.40	28.50	37.00	40.30	39.40	32.30

***L.S.D. = 2.365 at .001 per cent level.

Table 4. Temperature as a factor affecting the rate of honeydew deposition by the greenbug, <u>Toxoptera graminum</u> (Rond.), while feeding on Dicktoo barley for a 24 hour period under constant fluorescent lighting.

Replication Number	Number of honeydew droplets at 35°F.	40°F.	45°F.	50°F.	55°F.	60°F.	65°F.	70°F.	75°F.	80°F.	85°F.
1	4	8	9	9	10	13	15	22	15	21	14
2	5	5	11	7	11	14	14	14	14	15	15
3	2	6	6	12	10	14	16	21	21	13	13
4	4	9	4	8	12	13	14	15	17	20	12
5	9	8	6	10	11	10	11	17	16	12	16
6	4	7	10	10	13	11	15	13	19	23	13
7	7	5	7	9	12	9	14	21	15	14	12
8	5	4	10	14	7	13	12	15	16	10	10
9	7	6	7	8	10	11	14	18	14	15	14
10	4	5	7	7	9	10	15	14	18	11	11
11	5	8	5	9	10	12	13	16	17	15	15
12	3	4	6	8	11	14	12	17	15	14	14
Tot.	59	75	88	111	126	144	165	203	197	183	159
Avg.	4.90	6.20	7.30	9.25	10.50	12.00	13.70	16.90	16.40	15.20	13.20

***L.S.D. = 1.833 at .001 per cent level.

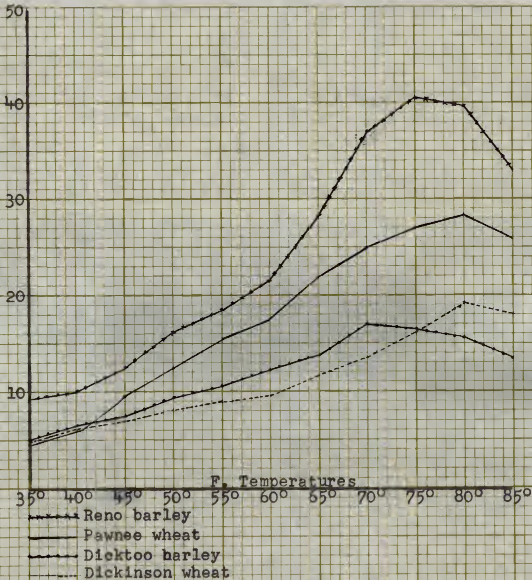


Fig. 1. A comparison of the effect of various temperatures on the rate of honeydew deposition by the greenbug while feeding on Reno and Dicktoo barley and Pawnee and Dickinson wheat for a 24 hour period under constant fluorescent lighting.

Area of Feeding on Reno Barley Leaf as a Factor Affecting the Rate of Honeydew Deposition by the Greenbug. The basal leaves of two Reno barley plants were used. The leaves were divided into four separate feeding areas, approximately two inches apart. There was no significant difference in the rate of honeydew deposition by the greenbug between area 1 (basal portion) and area 2 of the leaf. Significant differences in rate of honeydew deposition occurred at the .05 per cent level between area 2 and area 3. A highly significant difference was apparent in the rate of honeydew deposition by the greenbugs feeding on area 4 (apical region) in contrast with areas 1, 2 and 3 (Table 5).

Feeding on Yellowed vs. Green Leaves as a Factor Affecting the Rate of Honeydew Deposition by the Greenbug. Replication numbers 4, 5 and 6 of this experiment were omitted from the statistical analysis because of the presence of green streaks and spots in one of the yellowed leaves used in the test. The greenbugs apparently preferred these green areas of the leaf to yellowed portions, and were found to excrete at almost the same rate on these green areas as they did on green leaves (Table 6).

An average honeydew droplet rate of 5.71 was recorded for aphids feeding on the yellowed leaves for a twenty-four hour period. In contrast, 16.71 droplets were excreted by the greenbugs feeding on the green leaves. A statistical "t" test showed this difference of 11 droplets to be highly significant at the .001 per cent level.

Table 5. Area of feeding on Reno barley leaf as a factor affecting the rate of honeydew deposition by the greenbug, Toxoptera graminum (Rond.), for a 24 hour period under fluorescent lighting and at an average temperature of 65°F.

Replication Number	Number of droplets deposited on area indicated			
	4 (tip)	3	2	1 (base)
1	9	32	22	22
2	10	28	23	17
3	13	23	12	19
4	12	27	11	22
5	12	20	15	15
6	7	19	10	14
7	8	17	24	20
8	7	18	23	25
9	13	17	18	19
10	16	9	15	20
Tot.	107	210	173	193
Avg.	10.7	21.0	17.3	19.3

***L.S.D. = 4.394 at .001 per cent level
= 3.45 at .05 per cent level

Table 6. The effect of feeding on yellowed vs. green leaves of MO-B475 barley as a factor affecting the rate of honeydew deposition by the greenbug, _Toxoptera graminum_ (Rond.) for a 24 hour period under fluorescent lighting at an average temperature of 75°F.

	Number of droplets on	
Replications	Yellow Leaf	Green Leaf
1	6	14
2	5	20
3	6	16
4*	15	18
5*	16	19
6*	13	12
7	7	15
8	5	17
9	7	17
10	4	18
Tot.	40	117
Avg.	5.71	16.7

*Aphids feeding on green area in yellow leaf. These replications were omitted from the total number of honeydew droplets excreted, average number of droplets and from the statistical analysis.

***L.S.D. = 1.867 at .001 per cent level

Age of Aphid as a Factor Affecting the Rate of Honeydew Deposition. The period of the test constituted seven days. Reproduction was observed to occur on the sixth and seventh days. Due to the adverse effect of reproduction on the rate of honeydew deposition, the data from the sixth and seventh days were omitted from the statistical analysis.

No significant differences were found to occur in the rate of honeydew deposition during the first five days. A statistical analysis did show significant differences occurring between the deposition of honeydew by aphids in cages for the first five days even though there were no significant differences between the rate of deposition by days (Table 7). Molting was observed to have a slight depressive effect on the rate of honeydew deposition. Reproduction was found to significantly retard the rate of honeydew deposition. Figure 2 gives a graphic representation of the average rate of honeydew deposition by the greenbugs by days from birth to time of reproduction.

The Varietal Effect of Pawnee, Ponca, Bison and Dickinson Wheat as a Factor Affecting the Rate of Honeydew Deposition by the Greenbug. The rate of honeydew deposition by the greenbug was highest on Pawnee with an average of 34.0 droplets while an average of 19.0 droplets was excreted by the greenbug while feeding on Dickinson wheat. The latter rate of honeydew deposition was significantly lower than the rate of deposition on the other three varieties. The average rate of honeydew deposition while feeding on Bison and Ponca wheat was 30.5 and 25.9

Table 7. Age of aphid by days from birth to reproduction as a factor affecting the rate of honeydew deposition by the greenbug, Toxoptera graminum (Rond.), while feeding on Reno barley under fluorescent lighting and at an average temperature of 75°F.

Cage	Number of honeydew droplets excreted by days							
	1	2	3	4	5	Total 1st 5 days	6	7
1	32	38	38	30m*	21	159	23m*	12R**
2	40	62	53	38	55	248	9m*	9R**
3	44	49	40	24m*	17	174	8m*	10R**
4	31	27m*	27	24m*	18	127	10m*	8R**
5	37	28	24	45	30	164	19m*	11R**
6	33	31	14m*	29	22	129	9m*	17R**
7	30	30	21m*	54	11m*	146	21	9R**
8	59	31	46	58	46	240	12m*	15R**
9	30	33m*	57	20m*	15m*	155	23	18R**
10	36	31	50	33	31	181	14m*	12R**
11	25	47m*	39	20m*	35	166	18R**	8
12	18	29m*	30	21	38m*	136	9R**	10
13	27	24m*	30	18m*	16	115	11m*	18R**
Tot.	442	460	469	414	355	2140	186	157
Avg.	34	35.4	36.1	31.85	27.3		14.3	12.1

m* = molt.
R** = Reproduction.
L.S.D. 8.085 for first 5 days only at .01 per cent level (between cages only).

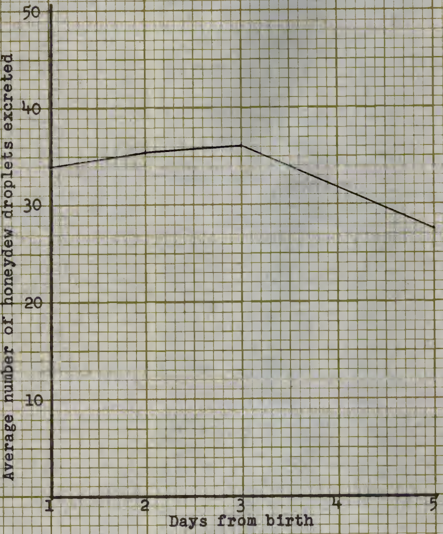


Fig. 2. The rate of honeydew deposition by the greenbug by days from birth until time of reproduction.

Table 8. Average rate of honeydew deposition by fourth instar greenbugs, *Toxoptera graminum* (Rond.), while feeding on Pawnee, Bison, Ponca, and Dickinson wheat varieties for a 24 hour period under fluorescent lighting at an average temperature of 75°F.

Replication Number	Number of droplets excreted by greenbug on			
	Pawnee	Bison	Ponca	Dickinson
1	22	16	26	9
2	43	36	20	14
3	29	15	40	6
4	35	12	17	22
5	48	16	21	27
6	26	20	24	26
7	31	39	21	22
8	49	42	43	27
9	30	36	24	18
10	27	73	23	19
Tot.	340	305	259	190
Avg.	34.0	30.5	25.9	19.0

*L.S.D. = 10.8566 at 5 per cent level

droplets respectively (Table 8). Significant differences were found to exist between Dickinson and the other three varieties. Ponca was almost significantly better than Pawnee. Figure 3 gives a graphic comparison of the rate of honeydew deposition by the greenbug while feeding on the four different wheat varieties.

The Varietal Effect of Dicktoo, MO-B475 and Reno Barley as a Factor Affecting the Rate of Honeydew Deposition by the Greenbug. The average rate of honeydew deposition by the greenbug was found to be generally higher on barley than on wheat. This was especially true on susceptible Reno and MO-B475 barley. Greenbugs feeding on Reno excreted at an average rate of 45.6 droplets over a twenty-four hour period. This was significantly higher than the 19.3 average droplet rate of the greenbug while feeding on Dicktoo. No significant difference was found in the rate of honeydew deposition between MO-B475 and Reno, even though there was a 7.1 droplet difference in the average. There was a definite significant difference in the rate of honeydew deposition between varieties MO-B475 and Dicktoo. Table 9 summarizes the results of the experiment and Fig. 3 shows a graphic comparison of the rate of honeydew deposition by the greenbug while feeding on Reno, Dicktoo and MO-B475 barley.

The rate of honeydew deposition by the greenbug on barley is also compared graphically in Fig. 3 to the rate deposited while feeding on Ponca, Bison, Dickinson and Pawnee wheat.

Table 9. Rate of honeydew deposition by adult greenbugs, *Toxoptera graminum* (Rond.), while feeding on Reno, MO-B475, and Dicktoo barley varieties for a 24 hour period under fluorescent lighting and at an average temperature of 75°F.

Replication Number	Number of droplets excreted by greenbug on: Reno	MO-B475	Dicktoo
1	56	55	40
2	60	35	18
3	50	40	21
4	22	30	15
5	35	54	10
6	65	48	6
7	37	24	40
8	30	48	7
9	61	16	15
10	40	35	21
Tot.	456	385	193
Avg.	45.6	38.5	19.3

***L.S.D. = 12.224 at .001 per cent level

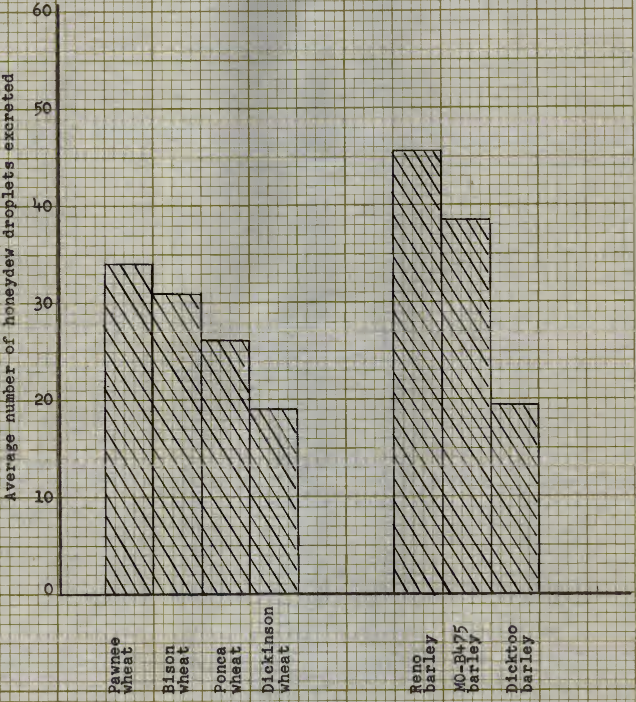


Fig. 3. Comparison of the rate of honeydew deposition by the greenbug while feeding on several wheat and barley varieties for a 24 hour period under fluorescent lighting.

The Effect of F_4, Pawnee x Dickinson Hybrids Carrying Different Degrees of Resistance as a Factor Affecting the Rate of Honeydew Deposition by the Greenbug. The five hybrid lines used in this experiment had been assigned the following per cent plant survival rating on the basis of a previous greenbug resistance test: 13245 - 30 per cent, 13208 - 52 per cent, 13242 - 13 per cent, 13209 - 72 per cent, and 13205 - 85 per cent. The checks used had the following rating: Dickinson - 100 per cent and Pawnee - 0 per cent.

The rate of honeydew deposition by the greenbug was found to be highest from test lines 13245, 13208 and 13242. There were no significant differences in the rate of honeydew deposition among these three hybrid lines. The lowest rate of honeydew deposition occurred from test lines 13209 and 13205. Significant differences at the 5 per cent level were found to exist between 13205 and 13242, 13208 and 13242. There was no significant difference between 13209 and 13242; however, significant differences existed between 13209 as compared to 13245 and 13208 (Table 10).

Spotted Alfalfa Aphid on Alfalfa

Effect of Various Temperatures on the Rate of Honeydew Deposition by the Spotted Alfalfa Aphid while Feeding on Alfalfa Clones B16, B105 and C89. The rate of honeydew deposition by the spotted alfalfa aphid on alfalfa clone B105 increased from a low of 4.66 droplets at 35°F. to a high of 38.25 at 85°F. The

Table 10. The effect of F_1 Pawnee x Dickinson hybrids carrying different degrees of resistance as a factor affecting the rate of honeydew deposition by the greenbug, <u>Toxoptera graminum</u> (Rond.), for a 24 hour period under fluorescent lighting and at an average temperature of 74° F.

Replications	Number of honeydew droplets excreted by greenbug on hybrid number				
Number	13245	13208	13242	13209	13205
1	20	14	18	13	7
2	66	27	55	19	6
3	29	35	21	17	14
4	35	25	15	15	12
5	19	25	23	20	16
6	13	21	29	17	20
7	20	14	15	16	30
8	35	25	14	7	11
9	15	27	10	8	11
10	29	26	20	19	15
Tot.	281	239	220	151	142
Avg.	28.1	23.9	22.0	15.1	14.2
Avg. per cent plant survival in previous greenbug test	30%	52%	13%	72%	85%

*L.S.D. = 9.098 at 5 per cent level

rate of honeydew deposition was found to increase as temperature increased. When averages at the various temperatures were placed on a graph, they formed an almost straight line (Fig. 4). Highly significant differences were found to exist among the rates of honeydew deposition at all the six 10°F. temperature intervals used (Table 11).

The rate of honeydew deposition by the spotted alfalfa aphid feeding on alfalfa clone B16 increased from an average droplet rate of 1.66 at 35°F. to a high of 21.25 at 85°F. The rate of honeydew deposition, as with clone B105, increased rapidly with increased temperatures. When the rate of honeydew deposition was plotted on a graph, it appeared slightly irregular as compared with the rate on clone B105. Highly significant differences were found between all 10°F. intervals from 35°F. to 85°F. No optimum temperature for the rate of honeydew deposition on clones B16 and B105 was found between the temperatures 35°F. to 85°F. (Table 12 and Fig. 4).

Much difficulty was encountered with feeding aphids on alfalfa clone C89. The resistant nature of this plant apparently had a highly detrimental effect upon the insects. The aphids usually died within 24 hours after being caged. Occasionally an aphid would remain alive but would seldom excrete honeydew. Table 13 summarizes the rate of honeydew deposition by the spotted alfalfa aphid while feeding on clone C89. There is a slight indication that honeydew production occurs more often within the temperature range of 45°F. to 65°F. while feeding on this

Table 11. The effect of various temperatures on the honeydew deposition rate by the spotted alfalfa aphid, Therioaphis maculata (Buckton), for a 24 hour period while feeding on alfalfa clone B105 under constant fluorescent lighting.

Replications Number	Number of honeydew droplets at 35°F.	45°F.	55°F.	65°F.	75°F.	85°F.
1	5	7	17	31	37	35
2	4	13	18	22	31	36
3	6	14	18	33	34	40
4	8	12	16	28	28	38
5	6	11	19	32	46	35
6	5	10	20	22	30	43
7	3	10	20	30	31	34
8	4	12	21	28	26	37
9	2	11	18	20	35	40
10	6	14	20	25	38	39
11	3	13	20	28	30	42
12	4	12	17	27	32	40
Tot.	56	139	224	326	398	459
Avg.	4.66	11.6	18.66	27.16	33.16	38.25

***L.S.D. = 2.656 at .001 per cent level.

Table 12. The effect of various temperatures on the honeydew deposition rate of the spotted alfalfa aphid, Therioaphis maculata (Buckton), for a 24 hour period while feeding on alfalfa clone B16 under constant fluorescent lighting.

Replications	Number of honeydew droplets at					
Number	35°F.	45°F.	55°F.	65°F.	75°F.	85°F.
1	2	5	8	13	18	18
2	1	6	10	16	24	23
3	1	4	9	17	17	17
4	2	5	9	14	13	26
5	1	4	8	15	15	25
6	2	3	11	21	21	26
7	2	6	9	23	20	20
8	1	5	8	17	22	19
9	3	4	10	18	14	22
10	2	8	7	12	16	14
11	1	6	14	13	24	24
12	2	7	11	18	17	21
Tot.	20	63	114	197	221	255
Avg.	1.66	5.25	9.50	16.41	18.40	21.25

***L.S.D. = 2.246 at .001 per cent level.

Table 13. The effect of various temperatures on the rate of honeydew deposition by the spotted alfalfa aphid, Therioaphis maculata (Buck.), for a 24 hour period while feeding on alfalfa clone C89 under constant fluorescent lighting.

Replications Number	Number of honeydew droplets excreted at 35°F.	45°F.	55°F.	65°F.	75°F.	85°F.
1	0	0	0	0	0	0
2	0	0	2	0	0	0
3	0	1	0	1	0	0
4	0	0	0	0	0	0
5	0	0	0	0	0	0
6	0	0	0	0	0	0
7	0	0	1	0	0	0
8	0	0	0	2	0	0
9	0	0	0	0	0	1
10	0	0	0	0	0	0
11	0	0	1	0	0	0
12	0	0	0	0	0	0
Tot.	0	1	4	3	0	1
Avg.	0	.0833	.333	.25	0	.0833

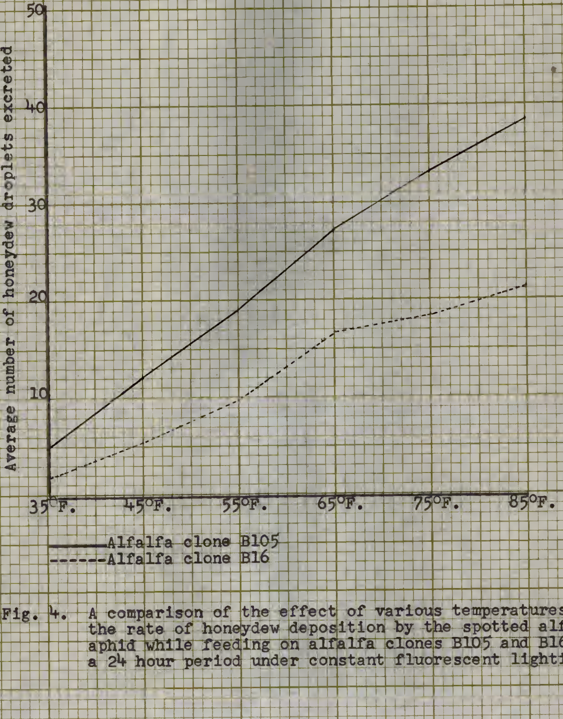


Fig. 4. A comparison of the effect of various temperatures on the rate of honeydew deposition by the spotted alfalfa aphid while feeding on alfalfa clones B105 and B16 for a 24 hour period under constant fluorescent lighting.

clone but the rate of honeydew deposition was found to be negligible.

Light Intensity as a Factor Affecting the Rate of Honeydew Deposition by the Spotted Alfalfa Aphid Feeding on Several Different Clones of Alfalfa. The depressing effect of reduced light on the rate of honeydew deposition by the spotted alfalfa aphid was first noticed during a cloudy interval in March, 1958 while records were being taken on alfalfa clones B105, B8, and C89. Later, additional readings were taken on these same clones during a normal sunny period. Tables 15 and 16 summarize the rate of honeydew deposition by the spotted alfalfa aphid feeding on B105, B8, and C89 clones under the two different light conditions. The reduction in light intensity by the cloudy weather was found to significantly reduce the rate of honeydew deposition by the spotted alfalfa aphid on clone B105 from an average of 31.47 droplets to an average of 11.2 droplets. The rate of honeydew deposition on clone B8 was reduced significantly from an average of 11.8 droplets to a low of 2 droplets. The rate of honeydew deposition on clone C89 was affected only slightly by the reduction in light.

Alfalfa clones B16, 50-1266, and C89 were tested under restricted light conditions using a modified light-proof cardboard box to restrict light from all of the plants except the checks (Table 17). Highly significant differences were noted between the rates of honeydew deposition by the spotted alfalfa aphid feeding on alfalfa clones 50-1266 and B16 when compared

Table 15. Reduced natural light as a factor affecting the rate of honeydew deposition by the spotted alfalfa aphid, Therioaphis maculata (Buck.), feeding for a 24 hour period on C89 and B8 alfalfa clones at an average temperature of 72°F.

Replications Number	Number of droplets: Reduced light C89*	Normal sunshine C89	Reduced light B8*	Normal sunshine B8
1	0	0	0	12
2	0	0	0	13
3	0	0	0	10
4	0	0	0	10
5	0	1	11	12
6	0	0	5	10
7	0	0	0	15
8	0	0	0	6
9	0	5	0	13
10	0	0	4	17
Tot.	0	6	20	118
Avg.	0	0.6	2.0	11.8

*Records were taken during an exceptionally cloudy period during March, 1958. Tests were conducted in the Entomology greenhouse. Normal sunshine records were taken at a later date during good weather.

Table 16. Reduced natural light as a factor affecting the rate of honeydew deposition by the spotted alfalfa aphid, *Therioaphis maculata* (Buck.), feeding on B105 alfalfa clones for a 24 hour period at an average temperature of 74°F.

Replications Number	Number of honeydew droplets excreted under: Reduced light*	Normal light
1	10	38
2	6	25
3	19	28
4	13	54
5	10	32
6	7	28
7	5	15
8	10	52
9	9	47
10	13	24
11	7	32
12	8	26
13	16	23
14	9	36
15	12	29
16	11	32
17	15	24
18	7	30
19	16	28
20	18	35
21	15	23
Tot.	236	661
Avg.	11.20	31.47

[1] *Records taken during an exceptionally cloudy period, March 1958. Tests were conducted in the Entomology greenhouse at Kansas State College. The check was taken later during normal weather.

[2] ***L.S.D. = 8.2 at .001 per cent level.

Table 17. Light intensity as a factor affecting the rate of honeydew deposition by the spotted alfalfa aphid, *Therioaphis maculata* (Buck.), while feeding on alfalfa clones, C89, B16, and 50-1266 for a 24 hour period under constant fluorescent lighting and at an average temperature of 75°F.

Replications Number	Number of honeydew droplets excreted under conditions of					
	Reduced light C89	Normal lighting C89	Reduced light B16*	Normal lighting B16	Reduced light 50-1266*	Normal lighting 50-1266
1	0	0	0	11	21	38
2	0	0	4	14	22	45
3	0	0	4	15	18	35
4	0	1	3	24	20	42
5	0	0	0	22	17	40
6	0	0	0	21	26	40
7	0	0	2	25	5	37
8	0	0	0	20	18	35
9	0	0	3	21	20	45
10	4	0	0	25	22	40
11	0	5	6	15	15	32
12	0	0	5	13	17	35
13	0	0	8	17	23	60
14	1	0	7	24	26	43
15	0	0	4	18	20	42
Tot.	5	6	46	285	290	609
Avg.	.33	.4	3.08	19.0	19.3	40.6

*All natural light was restricted from these plants by a large light proof cardboard box, 2½' x 3', in dimension.
***L.S.D. = 4.52 at .001 per cent level (B16 clone)
***L.S.D. = 9.12 at .001 per cent level (50-1266 clone)

with the rate of honeydew deposition while feeding on the same clones under constant fluorescent lighting. The rate of honeydew deposition by the spotted alfalfa aphid feeding on clone 50-1266 was reduced from an average droplet rate of 40.6 under fluorescent lighting to 19.3 under the reduced light. This represented slightly more than a fifty per cent reduction in the rate of honeydew deposition while feeding on this particular clone. The rate of honeydew deposition by the spotted alfalfa aphid feeding on B16 clone was reduced from an average droplet rate of 19.0 under fluorescent lighting to a low of 3.08 under reduced light. This represented more than a 75 per cent reduction in the average droplet rate on this clone. Reduction in the rate of honeydew deposition brought about by the reduced light conditions appeared to be greater in clones B16 and B8 than in B105 and 50-1266.

The restriction of light from the alfalfa clones and aphids caused a peculiar change in the color and consistency of some of the honeydew droplets. After approximately one day under the restricted light conditions, the aphids began excreting milky white honeydew droplets instead of the usual clear and viscous type. The white droplets were observed to be watery in consistency and after drying had a granular appearance.

Table 18 summarizes the results of the effect of the restriction of light from the immediate area of feeding. The cages were wrapped in aluminum foil to restrict the light only from the leaves where the aphids were feeding. The rate of honeydew deposition by the spotted alfalfa aphid while feeding on check clone

Table 18. The effect of reduced light on the rate of honeydew deposition by the spotted alfalfa aphid, Therioaphis maculata (Buck.), while feeding on plants or alfalfa clone 50-1266 using covered and uncovered cages under constant fluorescent lighting and at an average temperature of 75°F.

Replications: Number	Number of honeydew droplets excreted in Covered: Cages	Uncovered: Cages	Remarks
1	11	30	
2	15	25	
3	20	29	
4	12	30	
5	10	35	
6	20	28	
7	15	30	
8	15	34	
9	14*	34	Droplets white in covered cage
10	9*	30	
11	21*	35	
12	22*	29	
13	16*	32	Droplets white in covered cage
14	37*	38	Droplets white in covered cage
15	19*	25	Droplets white in covered cage
16	20*	31	Droplets white in covered cage
Tot.	276	495	
Avg.	17.25	30.93	

Light was not restricted from plant other than in area of feeding.
*Constitutes readings taken during a second day of the test.
**L.S.D. = 4.78 at 1 per cent level.

50-1266 averaged 30.93 droplets. In contrast, an average rate of 17.25 droplets was excreted by the spotted alfalfa aphid while feeding on clone 50-1266 where light was restricted from the area of feeding by the aluminum foil. This difference of 13.7 droplets was significant at the .001 per cent level.

The honeydew droplets were again observed to be white in color in several replications of this experiment. This phenomenon occurred during the second day of the test but was not as apparent as in the previous experiment where the light was restricted from all the plant.

Tables 16, 17, and 18 are summarized in Table 19. A better understanding of the effect of light intensity as a factor affecting the rate of honeydew deposition can be visualized by reference to this table.

<u>The Effect of Reduced Moisture to Host Plants as a Factor Affecting the Rate of Honeydew Deposition by the Spotted Alfalfa Aphid</u>. Kennedy and Mittler (1953) indicated that turgor pressure may be responsible for forcing plant sap up into and through the aphid's alimentary tract. This study was conducted in order to see if this theory proved to be applicable with the spotted alfalfa aphid, *Therioaphis maculata* (Buck.). Three plants from the B16 alfalfa clone were used as hosts in this experiment. The soil of plant number one was kept at the point of saturation with water, plant number 2 moist, and number 3 dry enough to induce incipient wilting.

Table 19. Reduction in light intensity as a factor affecting the rate of deposition of honeydew by the spotted alfalfa aphid, *Therioaphis maculata* (Buck.), feeding on several different clones of alfalfa for a 24 hour period.

Hosts	Average number of honeydew droplets deposited under				
	Light intensity reduced by cloudy weather (Avg. 72° F.)	Normal sunshine in greenhouse (Avg. 72° F.)	Light restriction from all of plant by large box*.	Light restriction by covering cage with rest of plant exposed to constant fluorescent lighting.	Constant fluorescent lighting
Alfalfa clone C89	0	.6	.33	0	.40
Alfalfa clone B16	-	-	3.08	10.2	19.00
Alfalfa clone B8	2.0	11.8	-	-	-
Alfalfa clone 50-1266	-	-	19.30	-	40.60
Alfalfa clone 50-1266	-	-	-	17.25	30.90
Alfalfa clone B105	11.2	31.4	-	-	-

*Lighting was restricted from these plants by a large light-proof cardboard box, 2½' x 3' in dimension.

Table 20 summarizes the results of this experiment. The highest rate of honeydew deposition by the aphids occurred while feeding on plant number one whose soil was at the point of saturation. The next highest rate of honeydew deposition was found to occur on plant number 2. The average rate of 16.4 droplets obtained on this plant was only 1.1 droplets higher than the rate of deposition on the plant maintained at the incipient wilting stage. No significant differences occurred in the rates of honeydew deposition among the B16 clones under the three different moisture conditions.

A number of milky white honeydew droplets similar to those occurring under reduced light and on yellowed leaves of barley were excreted by the aphids while feeding on the B16 alfalfa clone during incipient wilting.

The Distance of Leaves from Growing Point as a Factor Affecting the Rate of Honeydew Deposition by the Spotted Alfalfa Aphid. Table 21 summarizes the different rates of honeydew deposition by the spotted alfalfa aphids while restricted to ten different areas below the growing point of a twenty-four inch B16 alfalfa plant. It was observed that the rate of honeydew deposition by the spotted alfalfa aphid was lowest while feeding on leaves occurring eight inches below the growing point and highest while feeding on the leaves attached to the basal four inches of the plant. Although the lowest rate was at eight inches and the highest at twenty inches, distance of the leaves from the growing point of the B16 alfalfa plant was found to have no significant affect on

Table 20. The effect of reduced moisture to host plants* as a factor affecting the rate of honeydew deposition by the spotted alfalfa aphid, <u>Therioaphis</u> <u>maculata</u> (Buck.), over a 24 hour period under natural green-[3] house lighting and at an average temperature of 76°F.

Replications Number	Number of honeydew droplets excreted with: Soil saturated	Soil damp	Soil dry, plant at incipient wilting stage**
1	9	11	20
2	13	10	18
3	5	37	17
4	33	9	5
5	21	17	17
6	22	23	14
7	40	9	17
Tot.	143	116	108
Avg.	20.4	16.5	15.4

1 *Average heights of plants of B16 clone were 1½'.
2 **Numerous milky white droplets were noticed at this stage in place of normal clear droplets.
3 All readings were taken on second and third leaves from top of plant.
4 L.S.D. = non-significant.

Table 21. The distance of leaves from growing point as a factor affecting the rate of honeydew deposition by the spotted alfalfa aphid, <u>Therioaphis maculata</u> (Buck.), for a 24 hour period from plant B16 alfalfa, under natural greenhouse lighting and average temperature of 74°F.

Replications: Number	Distance from growing point 1"	3"	6"	8"	12"	15"	17"	20"	22"	24"
1	12	9	11	13	22	30	33	26	22	21
2	14	21	21	15	16	9	30	21	27	26
3	14	15	14	10	16	19	10	20	16	25
4	16	16	11	13	21	13	9	18	15	10
5	14	14	9	7	20	10	12	21	19	21
6	17	17	18	14	17	16	13	17	18	9
7	16	16	19	12	16	14	11	18	17	15
8	18	13	18	15	25	15	13	16	19	14
Tot.	121	121	121	99	153	126	131	157	153	141
Avg.	15.12	15.12	15.12	12.37	19.12	15.25	16.37	19.62	19.12	17.62

L.S.D. = Not significant.

the rate of honeydew deposition by the spotted alfalfa aphid.

Leaf, Stem and Petiole Feeding as a Factor Affecting the Rate of Honeydew Deposition by the Spotted Alfalfa Aphid. The effect of leaf, stem and petiole feeding as a factor affecting the rate of honeydew deposition by the spotted alfalfa aphid is summarized in Table 22. An average rate of 15.6 droplets was excreted by the spotted alfalfa aphids while feeding on the leaves of the B16 alfalfa plant. In contrast, averages of 9.73 and 11.46 droplets were excreted by the aphids while feeding on the petiole and stem. The difference of 5.9 droplets between the rates of honeydew deposition on the petiole and leaf proved to be highly significant at the .001 per cent level. The difference of 4.2 droplets between the rates of deposition by the aphids while feeding on the leaf and stem was also highly significant. The difference of 1.73 droplets occurring between the rate of deposition on the petiole and stem was not found to be significant.

Restricted Feeding on Buds and Flowers or Leaves of Alfalfa Clone B16 as a Factor Affecting the Rate of Honeydew Deposition by the Spotted Alfalfa Aphid. The average rate of honeydew deposition by the spotted alfalfa aphid while feeding on the buds and flowers of a B16 alfalfa plant was not significantly different from the deposition when feeding on the leaves of a non-flowering B16 alfalfa plant (Table 23).

The effect of flowering as a factor affecting the rate of honeydew deposition by the spotted alfalfa aphid is set forth in Table 23. A difference of 2.1 droplets in the average rate of

Table 22. Leaf, stem and petiole feeding as a factor affecting the rate of honeydew deposition by the spotted alfalfa aphid, Therioaphis maculata (Buck.), while feeding on alfalfa plant B16 for a 24 hour period under constant fluorescent lighting and at an average temperature of 72°F.

Replications Number	Number of honeydew droplets excreted while feeding on: Stem	Petiole	Leaf
1	14	10	17
2	9	14	10
3	16	17	9
4	14	12	16
5	9	8	15
6	7	7	12
7	20	9	17
8	14	7	15
9	11	6	16
10	12	9	20
11	6	7	15
12	9	4	16
13	10	10	17
14	13	14	14
15	8	12	25
Tot.	172	146	234
Avg.	11.46	9.73	15.6

***L.S.D. = 2.731 at .001 per cent level

Table 23. Restricted feeding on buds, flowers and leaves of alfalfa clone B16 as a factor affecting the rate of honeydew deposition by the spotted alfalfa aphid, Therioaphis maculata (Buck.), for a 24 hour period under constant fluorescent lighting.

Replications Number	Number of honeydew droplets excreted from aphids feeding at 71°F.	on buds and flowers at 72°F.	on non flowering branch at 71°F.	on non flowering branch at 72°F.
1	9	27	18	17
2	12	16	24	22
3	13	21	17	18
4	7	15	13	15
5	14	17	15	18
6	18	15	25	21
7	15	11	21	28
8	22	26	20	15
9	8	20	25	20
10	13	20	21	27
11	22	15	22	14
12	12	16	24	28
13	25	20	15	22
14	14	13	14	16
15	33	12	10	14
16	15	15	16	16
17	19	11	15	12
18	22	12	18	20
Tot.	293	333	331	343
Avg.	16.27	18.50	18.30	19.0

L.S.D. = non-significant.

deposition was obtained between the flowering B16 plant and the unflowering B16 plant. No significant difference in the rate of honeydew deposition by the spotted alfalfa aphid was detected on the leaves of an unflowering B16 plant.

The Effect of Different Alfalfa Clones Carrying Various Degrees of Resistance as a Factor Affecting the Rate of Honeydew Deposition by the Spotted Alfalfa Aphid. Table 25 summarizes the results obtained from individual experiments with the different clones represented. The rate of honeydew deposition by the spotted alfalfa aphid was found to be highest on the susceptible alfalfa clones B105 and 50-1266. There were no significant differences between the two clones in the rate of honeydew deposition. The rate of honeydew deposition by the spotted alfalfa aphid was approximately the same while feeding on alfalfa clones B4, B8, B16, and 247. Difficulty in getting aphids to settle on clone 247 was experienced. No significant differences in the rate of honeydew deposition were found on alfalfa clones B4, B8, B16 and 247. Highly significant differences were apparent between the rate of honeydew deposition on B4, B8, B16 and 247 as compared to the rate of deposition on clones B105 and 50-1266.

Extreme difficulty in getting spotted alfalfa aphids to survive and excrete on clones C89, C84 and 78 was experienced. Due to this difficulty the number of replications used in the average is necessarily low. The rate of honeydew deposition was approximately the same on these three clones and no significant differences were detected. Highly significant differences were appar-

Table 25. The effect of different alfalfa clones carrying various degrees of resistance as a factor affecting the rate of honeydew deposition by the spotted alfalfa aphid, <u>Therioaphis maculata</u> (Buck.), feeding for a 24 hour period at an average temperature of 67°F.

Clone	Number of replications	Average number of droplets per aphid
B105	15	30.73
50-1266	24	28.54
B4	33	13.21
B8	38	12.22 [3]
B16	27	12.07 [3]
247	7 [1]	10.43
C89	3 [1]	2.88
C84	6 [1]	1.66
78	4 [1]	1.00

[1] The low number of replications used to obtain an average number of honeydew droplets excreted per aphid is due to the highly resistant nature of these plants to spotted alfalfa aphid feeding. Aphids were caged on 247, C89, C84 and 78 but excretion of honeydew occurred only in the number of cages indicated. If zeros had been included averages would have been much lower.

[2] Three L.S.D.'s were taken at random. ***L.S.D. = 5.074, 11.242, and 9.910.

[3] The lower average deposition rates for B8 and B16 in this experiment as compared to others is attributable to the lower temperature and shorter day periods during February, 1958.

Table 26. Rate of honeydew deposition by the spotted alfalfa aphid, Therioaphis maculata (Buck.), for a 24 hour period while feeding on tree alfalfa, Medicago arborea, in comparison with clone 50-1266, Medicago sativa, under natural greenhouse lighting at an average temperature of 74°F.

Replications Number	Number of droplets excreted on M. arborea	Number of droplets excreted on Clone 50-1266, M. sativa
1	18	31
2	26	45
3	35	38
4	22	32
5	19	42
6	32	40
7	29	30
8	22	34
9	27	34
10	15	37
11	20	40
12	32	45
13	40	35
14	44	42
15	20	38
16	19	45
17	29	35
18	38	42
19	23	40
20	20	35
Tot.	530	760
Avg.	26.5	38.0

*L.S.D. = 4.46 at 5 per cent level

ent between the rate of honeydew deposition by the spotted alfalfa aphid while feeding on the resistant clones C89, C84 and 78 as compared to the rate of honeydew excretion on the other clones used in the experiment. Fig. 5 gives a graphic comparison of the different rates of honeydew deposition as occurring on the nine different clones tested.

Comparison of Spotted Alfalfa Aphid Rate of Honeydew Deposition on Tree Alfalfa, *Medicago arborea*, with Alfalfa Clone B105, *Medicago sativa*. Table 26 summarizes the differences in the rate of honeydew deposition by the spotted alfalfa aphid while feeding on two different species of *Medicago*.

An average rate of 38.0 droplets was excreted by the spotted alfalfa aphid while feeding on clone 50-1266. The average droplet rate on tree alfalfa was 26.5. This 11.5 droplet difference in the rate of honeydew deposition between *M. sativa* and *M. arborea* was found to be significant.

DISCUSSION

The effect of temperature on the development and reproduction rates of the greenbug has been thoroughly studied by Headlee (1914) and Wadley (1935). Reproduction and development of the greenbug were found to be most rapid at 80°F. Results indicate that the optimum temperature for honeydew deposition by the greenbug may be near this temperature. Studies with the rate of honeydew deposition at various temperatures on several wheat and barley varieties (Tables 1, 2, 3 and 4) indicate that

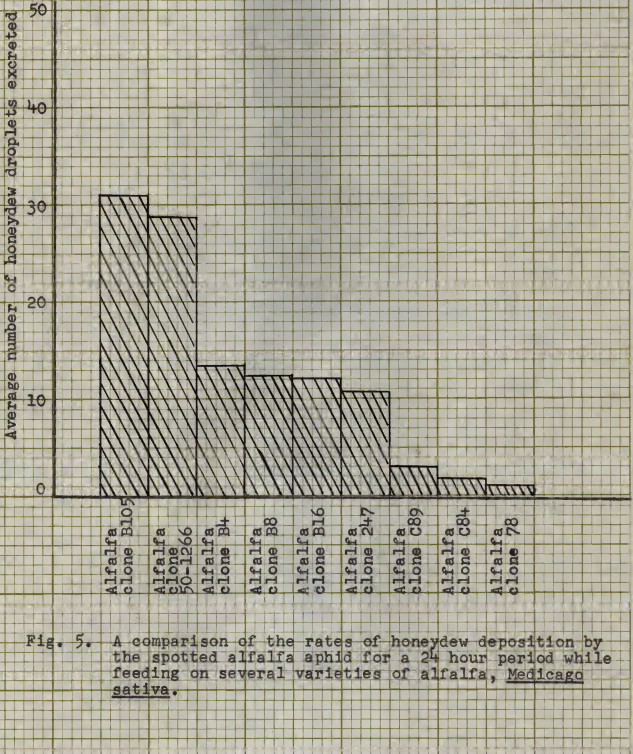


Fig. 5. A comparison of the rates of honeydew deposition by the spotted alfalfa aphid for a 24 hour period while feeding on several varieties of alfalfa, *Medicago sativa*.

the rate of honeydew deposition was highest on wheat at 80°F. The highest rate of excretion on barley was at 75°F. This apparent difference of 5°F. in the optimum temperature for honeydew excretion between wheat and barley may be within experimental error.

The decline in the rate of honeydew deposition occurring above 80°F. in wheat and 75°F. in barley varieties except Dicktoo indicates that the higher temperatures tend to depress significantly the rate of honeydew deposition. This depressing effect at higher temperatures may possibly be attributed to the adverse effect of the higher temperatures on the biology of the aphid which tends to slow down metabolic activity. Another possible explanation might be that higher temperatures promote physiological changes in the plant which might reduce the amount of plant sap ingested by the aphid.

The effect of temperature on the rate of honeydew deposition by the spotted alfalfa aphid was to some extent similar to the rates recorded on wheat and barley. No optimum temperature was found for honeydew production and results indicated that the optimum temperature would be somewhere above 85°F. Howe and Smith (1957) indicated that the spotted alfalfa aphid caused severe injury to susceptible plants in fourteen to sixteen days when day temperature reached 95°F. A possible reason for the difference in optimum temperatures for honeydew production between the two species of aphids might be due in part to the interrelations of the aphids with the host plant. Wheat and barley are cool season plants. The greenbug, being adapted to wheat and barley, would be expected to have a considerably lower optimum temperature for honeydew production than would be expected from the spotted alfalfa aphid which feeds chiefly on alfalfa, a warm season crop.

The fluctuations in the rate of honeydew deposition with changes in temperature may indicate a relationship to the metabolic activity and rate of ingestion of plant material by the insect. The temperature at which the rate of honeydew deposition is highest has also been found to be the temperature of optimum growth and reproduction in the greenbug (Wadley, 1935). A thorough investigation of the possibility of the relationship between rate of ingestion, metabolism and deposition of honeydew needs to be conducted in order to gain additional knowledge on this subject.

Results obtained from studying the effect of area of feeding on a Reno barley leaf as a factor affecting the rate of honeydew deposition by the greenbug showed that significant differences occurred in the rate of honeydew deposition by the greenbug between the apical portion and the rest of the leaf. The reason for the reduction in the rate of honeydew deposition on the apical portion of the leaf is not presently known but may be related to the presence of pectic substances (Gortner, 1938, Gisvold and Rogers, 1941, Miller, 1938) which are present in the apical portions of the leaves and buds during the period of maximum growth of plants.

Yellowed leaves were found to depress significantly the rate of honeydew excretion over the rate when greenbugs fed on green leaves (Table 6). Green areas in yellowed leaves were preferred and the aphids were found to excrete at approximately the same rate on these areas as on green leaves. The reduction in the rate of honeydew deposition is probably due to the absence of available plant sap containing the necessary nutrients. The probable reduction in photosynthesis and the production of lignin would retard transformation of starches to available sugars. The preference of the aphids for the green areas of the leaf may possibly indicate that sugar was still being produced in these areas of the leaf.

The age of the greenbug from birth through the first five days was found to have no significant affect on the rate of honeydew deposition. This suggests that as far as the rate of honeydew deposition is concerned, the age of the nymphs used in experiments would make very little difference. The size of honeydew droplets increased with age indicating that the total volume excreted may increase with age even though the average rate of droplet deposition is approximately the same. The significant differences found between rates of deposition by individual aphids during the period of the test is probably indicative of the effect of molting. Molting and reproduction were observed to depress the rate of honeydew deposition. A clear picture of the effect of molting was not obtained because of incomplete observations.

The significant effect of several varieties of wheat and barley was apparent on the rate of honeydew deposition by the greenbug. On wheat, the rate of honeydew deposition was highest on

Pawnee, with Bison, Ponca and Dickinson following in order. The rate of honeydew deposition on Reno barley was considerably higher than the rate of deposition on MO-B475, Dicktoo, and the four varieties of wheat. The difference found in the rate of honeydew deposition by the greenbug while feeding on wheat and barley varieties may be due in part to the susceptible or resistant nature of these varieties. Reno and Pawnee are well known for their susceptibility to greenbug attack. Ponca and Bison have been rated near Pawnee in susceptibility to greenbugs. MO-B475 barley is more tolerant than Reno but is still considered to be highly susceptible. Dicktoo and Dickinson, which had the lowest rates of honeydew deposition, have considerable tolerance to greenbug attack. Peters (1955) found that the greenbugs lost weight while feeding on Dicktoo barley and Dickinson wheat.

Results indicate that a correlation may exist between resistance and the rate of honeydew deposition. The varieties of wheat and barley used in the experiments may be ranked in the following order from high to low on the rate of honeydew deposition: Reno barley, MO-B475 barley, Pawnee wheat, Bison wheat, Ponca wheat, Dicktoo barley and Dickinson wheat.

The correlation between resistance and the rate of honeydew deposition was further emphasized in results obtained using several F_4, Pawnee x Dickinson hybrids carrying different degrees of resistance (Table 10). The rate of honeydew deposition was found to be lowest on test lines that had the greatest per cent survival in previous greenbug tests. The highest rate of honeydew deposition occurred on test lines that had low percentage survival. The fact that the rate of honeydew deposition on F_4 hybrid 13242 was out of line in correlation with resistance may be due to experimental error or else to the incorrect rating of resistance and per cent survival in previous test or to differences in resistance mechanisms being measured. A thorough study of F_1, F_2 and later generations in crosses is deemed necessary for a clear understanding of the apparent correlation between rate of honeydew deposition and resistance.

This theory is further verified by experiments measuring the rates of honeydew deposition by the spotted alfalfa aphid while feeding on several clones of alfalfa containing different degrees of resistance (Table 25). The rate of honeydew deposition was found to be highest on clones B105 and 50-1266 which are highly susceptible to the spotted alfalfa aphid. The rate of honeydew deposition was significantly depressed on clones B4, B8, B16 and 247 which were selected because of the presence of known degrees of resistance to the spotted alfalfa aphid. The rate of honeydew deposition on highly resistant clones C84, C89 and 78 was negligible. This low rate is undoubtedly attributable to the highly resistant character of these plants which either repels the aphids or causes an antibiotic effect resulting in death.

Light intensity, next to temperature, appears to be an important factor affecting the rate of honeydew deposition. At low light intensity rates of honeydew deposition were reduced from 50 to 75 per cent below normal. These acute reductions in the rate of honeydew deposition may be attributable either to the effect of the reduced light on the biology of the aphid or the effect of the reduced light on the physiology of the host plant which produces chemical changes that are repugnant to the aphid. The excretion of white honeydew droplets tends to indicate that the aphids are probably ingesting plant material with a high starch content. Meyer and Anderson (1952) indicate that a reduction in the rate of photosynthesis would reduce the starch-to-sugar conversion and would promote a high starch content in the plant.

The reduction of moisture given the host plant was found to have no significant effect on the rate of honeydew deposition (Table 20). The rate of honeydew deposition by the spotted alfalfa aphid while feeding on a B16 plant at incipient wilting stage was not significantly different from the rate of honeydew deposition obtained while the spotted alfalfa aphid was feeding on a turgid B16 plant. Kennedy and Mittler (1953) and Mittler (1957), working with Tuberolachnus salignus (Gmelin), advanced the theory that turgor pressure forces the plant sap through the aphid's alimentary system and that a specialized pump was not necessary for the ingestion of plant sap. Results of this experiment, on the contrary, indicate that the spotted alfalfa aphid is able to ingest plant sap at the incipient wilting stage and is capable of excreting honeydew at approximately the same rate as aphids feeding on turgid plants. Turgor pressure is near zero at the incipient wilting stage (Miller, 1938) and, therefore, it seems probable that the spotted alfalfa aphid must apply some type of suction to ingest the plant sap.

The excretion of milky white droplets of honeydew by the spotted alfalfa aphid also occurred during the incipient wilting stage of the plant. The reduction in photosynthesis which occurs during incipient wilting (Thoday, 1910, Iljin, 1923, Brilliant, 1924) raises the starch level in the plant and could be a possible explanation for this phenomenon.

No significant differences were recorded in the rates of honeydew deposition by the spotted alfalfa aphid while feeding on buds, flowers and leaves of a B16 alfalfa clone. No significance was found in the rate of honeydew deposition by the spotted alfalfa aphid while feeding on leaves of a flowering plant as opposed to leaves of a nonflowering plant. Cartier and Painter (1956) found the opposite true in the fecundity rate of the corn leaf aphid, Rhopalosiphum maidis (Fitch), Their studies indicated that the aphids reproduced two to five times faster on the heads than on the leaves of sorghums, and that the weights of the apterous adults reared on the heads were double or triple those reared on the lower leaves of the same plants. These negative results strongly indicate that the changes in the physiology of the plant before and during the period of flowering has no significant effect on the rate of honeydew deposition by the spotted alfalfa aphid.

The highly significant differences obtained in the rate of honeydew deposition by the spotted alfalfa aphid between clone 50-1266, Medicago sativa, and tree alfalfa, Medicago arborea, indicate that the rate of honeydew deposition not only differs significantly between varieties of the same species but may differ significantly between host species of _Medicago_, _Hordeum_ and _Triticum_ as well. A comparison of the rate of honeydew deposition of the greenbug and the spotted alfalfa aphid while feeding on several varieties of wheat, barley and alfalfa are compared at six different temperatures (Table 27). Rate of honeydew deposition was highest on Reno barley at 75°F. with an average droplet rate of 40.3, but at 85°F. the highest rate of honeydew deposition occurred on alfalfa clone B105. The lowest rate of honeydew deposition occurred on alfalfa clone C89. More honeydew was produced on alfalfa clone B16 than on Dickinson wheat and Dicktoo barley.

Results showed that the rate of honeydew deposition on Reno and Dicktoo barley and Pawnee wheat exceeded that found on many of the alfalfa varieties studied. This implies that damage due to honeydew may be as great on barley and wheat as on alfalfa but is not as noticeable to the grower due to the different methods of cultivation and time of harvesting.

The rate of honeydew deposition may provide a new method for measuring resistance. The advantages of using this method lies in the ability to combine this method with the procedures now in use for measuring resistance. Another advantage would be that this method would provide a crude measure of the rate of ingestion and metabolic activity of the insect while feeding on host plants.

The main disadvantage to incorporating this method into the existing procedure for testing for resistance in plants would be

Table 27. A comparison of the average rate of honeydew deposition for 24 hours by two species of aphids, Toxoptera graminum (Rond.) and Therioaphis maculata (Buck.), under similar temperature conditions on different host plants under fluorescent lighting.

Variety, crop and insect	Average number of honeydew droplets at 35°F.	45°F.	55°F.	65°F.	75°F.	85°F.
Therioaphis maculata on						
B105 alfalfa clone	4.66	11.6	18.6	27.1	33.1	38.2
B16 alfalfa clone	1.66	5.25	9.5	16.4	18.4	21.2
C89 alfalfa clone	0	.08	.3	.25	0	.08
Toxoptera graminum on						
Reno barley	9.3	12.3	18.1	28.5	40.3	32.3
Pawnee wheat	4.3	9.08	15.1	21.8	26.8	25.4
Dickinson wheat	4.4	7.08	8.9	11.5	16.1	17.9
Dicktoo barley	4.9	7.3	10.5	13.7	16.4	13.2

the large amount of time and labor required in computing the rate of deposition where many plants are involved. The variation in sizes of many of the droplets might prove to be a problem if a large degree of accuracy is required in tests. Unless a method can be developed for measuring either the volume or the weight of the honeydew droplets it is probable that the rate of deposition would be unsuitable as a measure of resistance by itself but combined with methods now employed it would certainly provide a relative measurement of resistance and in addition contribute valuable pertinent information on the biology of the aphid.

Resistance to aphids found in several varieties of alfalfa and small grains presents to agriculture a most desirable means of control. The presence of resistance in crops not only provides an inexpensive method of control of aphids but also prevents the deposits of honeydew which may retard normal pollination, promote fungus growth, attract other insects, and give rise to difficult problems in harvesting.

SUMMARY

The purpose of this study was to compare the effect of various factors on the rate of honeydew deposition by the greenbug, *Toxoptera graminum* (Rond.), and the spotted alfalfa aphid, *Therioaphis maculata* (Buck.), while feeding on host plants wheat, barley and alfalfa.

Since no previous work of this particular type of study had been reported, considerable time was spent in developing a method that would give reproduceable results. In the method finally developed, honeydew from one aphid feeding on a plant part was caught on the bottom or sides of plastic cages. Usually only one aphid was placed in each cage and was allowed to feed for a twenty-four hour period after which the cages were removed and the honeydew droplets counted. A total of 1,192 cages usually containing a single aphid were placed on plants of sixteen varieties of wheat, barley and alfalfa.

The rate of honeydew deposition was affected significantly in both the greenbug and spotted alfalfa aphid by changes in temperatures and by different varieties and hybrids of plants; the latter may be attributable to the presence of resistance in variable quantities.

The following factors were found to affect significantly the rate of honeydew deposition by the greenbug: (1) the area of feeding on Reno barley leaf, (2) feeding on leaves yellowed by age, and (3) reproduction and molting.

The rate of honeydew deposition by the spotted alfalfa aphid was affected significantly by the following factors or conditions: (1) reduction in light intensity reaching the plant, (2) differences in rates between petiole, leaf, and stem as feeding sites; and (3) feeding on two different species of Medicago.

The age of the aphid through the first five days had no significant affect on the rate of honeydew deposition by the greenbug.

Changes in the following factors or conditions did not affect significantly the rate of honeydew deposition by the spotted alfalfa aphid: (1) reduced moisture to host plant; and (2) distance of leaves from growing point.

Milky white honeydew droplets, excreted by the greenbug feeding on yellowed leaves and by the spotted alfalfa aphid feeding under reduced light intensity and incipient wilting, were noted. These conditions are believed to be related to the decrease in the rate of photosynthesis of the plants under these conditions which is known to promote a high level of starch content in the leaf.

The varieties of wheat, barley and alfalfa can be ranked in the following descending order on the basis of the rates of honeydew deposition by the greenbug and spotted alfalfa aphid: Reno barley, MO-B475 barley, Pawnee wheat, B105 alfalfa clone, Bison wheat, 50-1266 alfalfa clone, Ponca wheat, Dicktoo barley, Dickinson wheat, alfalfa clones B4, B8, B16, 247, C89, C84, and 78.

This comparison clearly indicates that the rate of honeydew deposition on Reno and Dicktoo barley and Pawnee wheat exceeds that found on the alfalfa varieties studied. This implies that damage due to honeydew may be as great on barley and wheat as on alfalfa but is not as noticeable due to the different methods in cultivation and harvesting time.

The rate of honeydew deposition was found to be influenced almost directly in proportion to the known amount of resistance found in the host plants used. This condition suggests the possibility that the rate of honeydew deposition by aphids may be used to measure the degree of resistance of host plants to aphids, rate of ingestion of plant material and serve as a crude measure of the metabolic activity of the insect.

Host resistance not only provide an inexpensive method of control of aphids but also presents the most desirable method of retarding the large deposit of honeydew which interferes with normal pollination, promotes fungus growth, attracts other insects and gives rise to difficult problems in harvesting crops.

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LITERATURE CITED

Atkins, I. M. and R. G. Dahms.
Reaction of small grain varieties to greenbug attack. U. S. Dept. Agr. Tech. Bul. 901. 30 p. 1945.

Blizzard, W. L.
"Omugi barley resistant to greenbugs" in Science serving agriculture. Okla. Agr. Expt. Sta. Bien. Rept. 1946-48. p.27. 1948.

Brilliant, B.
Le teneur en eau dans les feuilles et l' energie assimilatrue, C. R. Acad. Sci. Paris. 178:2122-2125. 1924.

Broadbent, L.
Aphid excretion. Proc. R. Ent. Soc. London (A) 26: Pts. 7-9: 97-103. Sept. 1951.

Buckton, G. B.
Monograph of the British Aphides. Vol. 1. Ray Society. 350 p. London. 1876.

Cartier, J. J. and Reginald H. Painter.
Differential reactions of the two biotypes of corn leaf aphids to resistant and susceptible varieties, hybrids and selections of sorghums. Jour. Econ. Ent. 49:498-508. 1956.

Dahms, R. G.
Comparative tolerance of small grains to greenbugs from Oklahoma and Mississippi. Jour. Econ. Ent. 41:825-826. 1948.

__________.
Preventing greenbug outbreaks. U. S. Dept. Agr. Farmer's Bul. 309. 8 p. 1951.

Dahms, R. G., T. H. Johnston, A. M. Schlehuber, and E. A. Wood, Jr.
Reaction of small grain varieties and hybrids to greenbug attack. Okla. Agr. Expt. Sta. Tech. Bul. T-55. Sept. 1955.

Day, M. F. and H. Irzykiewicz.
Feeding behavior of the aphids _Myzus persica_ and _Brevicoryne brassicae_, studies with radio-phosphorus. Australian Jour. Biol. Science 6:98-108. 1953.

Dickson, R. C., Edward F. Laird, Jr., and George R. Pesho.
The spotted alfalfa aphid. Hilgardia 24:93-118. 1955.

Fenton, F. A., and R. G. Dahms
The 1939 greenbug outbreak in Oklahoma. Jour. Econ. Ent. 33:628-634. 1940.

Gisvold, O. and C. H. Rogers.
The chemistry of plant constituents. Minneapolis, Minn. 392 p. 1941.

Gortner, R. A.
Outlines of Biochemistry. New York: McGraw-Hill. 1017 p. 1938.

Harpaz, I.
Bionomics of Therioaphis maculata (Buck.) in Israel. 2 Jour. Econ. Ent. 48:668-671. 1955.

Harvey, T. L. and H. L. Hackerott.
Apparent resistance to the spotted alfalfa aphid selected from seedlings of susceptible alfalfa varieties. Jour. Econ. Ent. 49:289-291. 1956.

Headlee, T. J.
Some data on the effect of temperature and moisture on the rate of insect metabolism. Jour. Econ. Ent. 7:413-417. 1914.

Howe, W. L. and Oliver F. Smith.
Resistance to the spotted alfalfa aphid in Lahontan alfalfa. Jour. Econ. Ent. 50(3):320-324. 1957.

Iljin, W. S.
Der Einfluss des Wassermongels auf die Kohlenstoffassimilation durch die Pflanzen. Flora 116:360-378. 1923.

Kennedy, J. S. and T. E. Mittler.
A method of obtaining phloem sap via the mouthparts of aphids. Nature 171:528. 1953.

Kirby, W. and W. Spence.
An introduction to Entomology. 6th ed. vol. 2. London. 325 p. 1843.

Leonardt. H.
Beitrage zur Kenntnis der Lachniden der wichtigsten Tannenhonigtauerzenger Zeitschr. Angew Ent. 27:208-272. 1940.

Luginbill, B. and A. H. Beyer.
Contribution to the knowledge of Toxoptera graminum (Rond.) in the south. Jour. Agr. Res. 14:96-119. 1918.

Meyer, B. S. and D. B. Anderson.
Plant physiology. New York: D. Von Nostrand Co. Inc., 1952. 784 p.

Michel, F.
Beitrage zur Kenntnis von Lachmus (Pterochloris) robaris L., einer wichtigen Honegtonezengerin on der Eiche. Zeitschr. Angew. Ent. 29:243-281. 1942.

Miller, E. C.
Plant physiology. New York: McGraw-Hill. 1201 p. 1938.

Mittler, T. E.
Studies on the feeding and nutrition of Tuberolachnus salignus (Gmelin) (Homoptera, Aphididae). Jour. Exp. Biol. 34:334-341. Sept. 1957.

Ortman, Eldon E.
A measurement of greenbug (Toxoptera graminum Rond.) damage to the root systems and other plant parts of several varieties of wheat. Unpublished Master's Thesis, Kansas State College. 54 p. 1957.

Painter, Reginald H.
Insect resistance in crop plants. New York: MacMillan Co. 520 p. 1951.

Passerini, J.
Aphididae Italicae. Genoa, Italy. 1863. p. 28-29.

Pathak, M. D. and Reginald H. Painter.
Differential amounts of material taken up by four biotypes of corn leaf aphids from resistant and susceptible sorghums. Ann. Ent. Soc. Amer. 51:250-254. 1958.

Pergande, Theodore.
The Southern grain louse. U. S. Dept. Agr. Div. Ent. Bul. 38:1-19. 1902.

Peters, Don C.
A comparison of certain features of the biologies of greenbugs, Toxoptera graminum (Rond.), on the recommended Kansas winter wheat and barley varieties. Unpublished master's thesis. Kansas State College. 1955.

Peters, Don C. and R. H. Painter.
Studies on the biologies of three related legume aphids in relation to their host plants. Kan. Agr. Expt. Sta. Tech. Bul. 93. May 1958.

Reitz, L. P.
Wheat breeding and our food supply. Econ. Bot. 8:251-268. 1954.

Reynolds, H. T., and L. D. Anderson.
Control of the spotted alfalfa aphid on alfalfa in Southern California. Jour. Econ. Ent. 48:671-675. 1955.

Roeder, K. D.
Insect physiology. New York: John Wiley and Sons. 1100 p. 1953.

Rondani, C.
Lettera al L. Prof. G. Bertolon: Nota Sopra Una Specie di Afie, volante in numerosa torma nella citta' di Parma. Nuova Ann. Sci. Nat. Bologna, Ser. III. 6:9-12. 1852.

Sajo, Karl
Beiträge zur landwirtschaftlichen Insektenkunde. Ztschr. Pflanzenkrank. 4:41. 1894.

Schaefer, C. W.
Physiological conditions which produce wing development in the pea aphid. Jour. Agr. Res. 57:825-841. 1938.

Silviera, G. A., and J. E. Conde.
(The green aphid (Toxoptera (Schizaphis) graminum Rond.) of cereals in Uruguay.) Rev. Fac. Agron. Univ. Montevideo 41. 54 p. Montevideo. 1946.

Smith, L. M.
Growth, reproduction, feeding and wing development of the mealy plum aphid in relation to climatic factors. Jour. Agr. Res. 54:345-367. 1937.

Snedecor, George W.
Statistical methods. Ames, Iowa: Collegiate Press Inc. 388p. 1938.

Stanford, E. H.
Resistant plants. Calif. Agr. 9(7):5. 1955.

Thoday, D.
Experimental researches on vegetable assimilation and respiration. Proc. Roy. Soc., B. 82:421-450. 1910.

Wadley, F. M.
Ecology of Toxoptera graminum, especially as to factors affecting importance in the northern United States. Ent. Soc. Amer. Ann. 24:325-395. 1931.

____
Development-temperature correlation in the greenbug, Toxoptera graminum. Jour. Agr. Res. 53:259-266. 1935.

Waterhouse, D. F.
Digestion in insects. In Annual Review of Entomology. Stanford Calif: Annual Rev. Inc. 2:1-17. 1957.

Weber, H.
Biologie der Hemipteren. Berlin. 225p. 1930.

Webster, F. M., and W. J. Phillips.
The spring grain-aphis or "green bug". U. S. Dept. Agr. Bur. Ent. Bul. 110:1-153. 1912.

CERTAIN FACTORS AFFECTING THE RATE OF HONEYDEW DEPOSITION IN TWO SPECIES OF APHIDS, <u>TOXOPTERA GRAMINUM</u> (ROND.) AND <u>THERIOAPHIS MACULATA</u> (BUCK.) FEEDING ON HOST PLANTS WHEAT, BARLEY AND ALFALFA

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