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Selection of Pollens by Honey Bees

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At the Legume Seed Research Laboratory at Logan, Utah, an attempt has been made to determine whether honey bees (Apis mellifera L.) are influenced in their selection of pollen by something other than the structure, color pattern, or perfume of the flower in which the pollen is found. Syrge (1947) found that bees showed a preference for one species of pollen over another when the pollens were offered simultaneously within the hive. Our experiments were designed to find out whether such preferences would show up when pollens were offered outside the hive under conditions somewhat closer to those under which bees ordinarily make their selection.

Materials and Methods

The pollens offered to the bees were alfalfa (Medicago sativa), gumweed (Grindelia squarrosa), greasewood (Sarcobatus vermiculatus), black mustard (Brassica nigra), povertyweed (Iva axillaris), and a mixture of yellow and white sweet clover (Melilotus spp.). In preliminary experiments red clover (Trifolium pratense), corn (Zea mays), and juniper (Juniperus utahensis) pollens were also used, as were also soybean flour with and without dried brewer's yeast.

All these pollens except juniper were obtained from pollen traps attached to beehives. Placed on hive entrances, the traps force incoming bees to crawl through 6-mesh hardware cloth, which knocks the pollen pellets off their legs and into a removable trap. Pellets of the various species of pollen were separated, dried, and then powdered in a small Wiley mill. Preliminary tests in 1950 indicated that particle size influences selection; so in subsequent tests all pollens were put through a 40-mesh sieve.

The alfalfa, greasewood, and povertyweed pollens were collected in 1949; gumweed, black mustard, and red clover in 1950; and sweet clover and additional black mustard in 1953. All the pollens collected in 1949 and 1950 were ground in 1951 and stored in closed jars at room temperature. The pollens collected in 1953 were ground just before use in 1954.

The juniper pollen was obtained by collecting unopened staminate cones of juniper, storing them in paper bags until dry, and then shaking them vigorously and sifting the pollen from the debris.
Measurements were made of some of the physical and nutritive characteristics of the ground pollens used in the 1953 experiments (table 1).

Table 1. Physical and nutritive characteristics of pollens tested.

<table>
<thead>
<tr>
<th>Pollen</th>
<th>Reflectivity (Candle-power)</th>
<th>Per cent Moisture</th>
<th>Per cent Total Sugar</th>
<th>Per cent Crude Protein</th>
</tr>
</thead>
<tbody>
<tr>
<td>Povertyweed</td>
<td>Next to corn</td>
<td>235</td>
<td>21.1</td>
<td>49.0</td>
</tr>
<tr>
<td>Mustard</td>
<td>Wax yellow</td>
<td>180</td>
<td>22.6</td>
<td>35.1</td>
</tr>
<tr>
<td>Greasewood</td>
<td>Pyrethrum yellow</td>
<td>180</td>
<td>24.2</td>
<td>42.9</td>
</tr>
<tr>
<td>Alfalfa</td>
<td>Light stone</td>
<td>170</td>
<td>24.4</td>
<td>37.0</td>
</tr>
<tr>
<td>Mixture</td>
<td>Burnt and yellow stone</td>
<td>150</td>
<td>22.5</td>
<td>39.7</td>
</tr>
<tr>
<td>Gumweed</td>
<td>Centennial brown</td>
<td>100</td>
<td>22.9</td>
<td>40.4</td>
</tr>
</tbody>
</table>

1 As designated in Maerz & Paul (1950).

Intensity of odor was determined by a panel of six persons. Colors were determined for the powdered, dry materials by comparing them with the color chart in Maerz and Paul (1950). Light reflectivity was read from a Weston Master II light meter held over a uniform area of these materials. Moisture readings were made as the pollens were taken from a temperature cabinet at 85°F and 50--60 per cent relative humidity. Conditions in the field were not likely to alter the positions of the pollens on a moisture scale. Total sugar was determined by a modified Shaffer - Hartman - Somogy method, and crude protein by the Kjeldahl method.

The pollens were exposed to the bees in 4-ounce salve tins, 75 mm. in diameter and 25 mm. in depth. To minimize loss of material from wind action and fanning by the bees, each salve tin was placed in a half-pint cardboard container, 95 mm. in diameter and 45 mm. in depth.

The experiments were conducted on bare, level ground about 100 yards from a 30-colony apiary. The containers of pollen were arranged in a 6x6 Latin square with 3 feet between each container. Preliminary tests using other arrangements gave results less satisfactory for statistical analysis. The latin-square design, recommended by the statistical department of the Utah Agricultural Experiment Station, removed possible influence of the relative positions of the pollens.

The experiments were conducted before pollen from natural sources was available to the bees. As soon as natural sources appeared, few bees visited any of the pollens being tested. All trials were conducted on warm, sunny, calm days. The pollen containers were set out while closed, and when all
were distributed they were opened almost simultaneously. Counts were made at intervals of the bees gathering pollen at each container. At the beginning of each test the interval was 10 or more minutes, but as the number of bee visitors increased it became shorter. Trials were discontinued when the bees at any one container became too numerous to be counted accurately.

Preliminary tests in 1951 indicated that when the quantity of material in a container decreased considerably, fewer bees visited that container. Hence, uniform quantities were placed in each dish, and each trial was discontinued before enough material had been removed from any dish to allow the bottom to show.

In each trial five pollens were offered to the bees along with a mixture of all five. In 1954, after the preferences had been established in one trial, an attempt was made in a second trial to mask the odors of the pollens by inserting between the salve tin and the cardboard container a piece of cotton saturated with methyl salicylate.

In 1953 some of the pollens used were a year or more older than others. In 1954, to test the hypothesis that degree of freshness was responsible for some of the differences in attractiveness, mustard pollen collected in 1950 and ground in 1951 was compared with mustard pollen collected in 1953 and ground just before the experiment. Other pollens used in this experiment were gumweed and alfalfa (old material) and sweet clover (fresh material). A mixture of all five served as a check.

Results

In 1951 preliminary tests indicated the bees showed a preference for some pollens, but this preference was not clear-cut or constant. During one trial the total numbers of bees visiting each test substance were as follows: Gumweed 525, corn 370, alfalfa 263, flour plus yeast 142, flour 137, and juniper 136. In another trial when red clover pollen was substituted for juniper it proved to be more attractive than gumweed. The order of attractiveness in this trial was: Red clover 258, gumweed 219, alfalfa 150, flour 46, corn 45, flour plus yeast 29.

<table>
<thead>
<tr>
<th>Pollen</th>
<th>First Trial</th>
<th>Second Trial</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mustard</td>
<td>40.0</td>
<td>31.0</td>
</tr>
<tr>
<td>Mixture</td>
<td>20.3</td>
<td>8.6</td>
</tr>
<tr>
<td>Gumweed</td>
<td>14.0</td>
<td>2.6</td>
</tr>
<tr>
<td>Greasewood</td>
<td>5.3</td>
<td>1.6</td>
</tr>
<tr>
<td>Povertyweed</td>
<td>5.0</td>
<td>1.0</td>
</tr>
<tr>
<td>Alfalfa</td>
<td>4.0</td>
<td>0.6</td>
</tr>
<tr>
<td>LSD (1 per cent)</td>
<td>9.8</td>
<td>3.6</td>
</tr>
</tbody>
</table>
The preferences for pollens offered in the Latin-square design in 1953 are shown in table 2. In both trials mustard was far the most attractive material, and the order of preference for the other pollens remained the same as in 1951.

Table 3 shows that in 1954 mustard pollen collected in 1950 was at least as attractive as that collected in 1953. Also the relative attractiveness is consistent with earlier tests and unaffected by methyl salicylate.

None of the pollen characteristics shows any association with attractiveness to the bees as based on the selection tests.

A panel judging odor intensity agreed that mustard pollen had the strongest odor, but there was little agreement concerning the relative positions of the others. In the attempt to mask pollen odors by permeating the air with methyl salicylate, the order of preference remained the same as in the previous test without this chemical.

Table 3. Average numbers of bees visiting various pollens, 1954.

<table>
<thead>
<tr>
<th>Pollen</th>
<th>Year Collected</th>
<th>Before Addition of Methyl Salicyate</th>
<th>After Addition of Methyl Salicyate</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mustard</td>
<td>1950</td>
<td>23.3</td>
<td>40.1</td>
</tr>
<tr>
<td></td>
<td>1953</td>
<td>18.6</td>
<td>37.0</td>
</tr>
<tr>
<td>Mixture</td>
<td>--</td>
<td>16.0</td>
<td>26.1</td>
</tr>
<tr>
<td>Sweet Clover</td>
<td>1953</td>
<td>14.7</td>
<td>24.1</td>
</tr>
<tr>
<td>Gmweed</td>
<td>1950</td>
<td>10.0</td>
<td>17.8</td>
</tr>
<tr>
<td>Alfalfa</td>
<td>1949</td>
<td>6.0</td>
<td>13.6</td>
</tr>
<tr>
<td>LSD (1 per cent)</td>
<td></td>
<td>9.3</td>
<td>10.4</td>
</tr>
</tbody>
</table>

Discussion

It is concluded from this study that some pollens are more attractive than others to honey bees even when separated from the flower structures, color patterns, and perfumes by which they are normally surrounded. The relatively low attractiveness of alfalfa and povertyweed and the high attractiveness of red clover and mustard are in accord with observations under natural conditions. This suggests that characteristics of the pollen itself affect its selection and may actually outweigh such factors as the difficulties of obtaining it and the influence of the associated flower characteristics.

Once differences in pollen attractiveness were established, we became interested in the factors responsible for these differences. Our experiments were originally designed to eliminate or minimize the effects of particle size, quantity, and position of the pollens in relation to each other. Since freshness of the samples was an unnatural variant introduced by necessity, we were gratified to learn from the results with old and nearly fresh...
mustard pollen that it appears to make no difference. The other factors studied were those that the bees normally encounter in the field and that we believed might influence their choice.

Each species of pollen has its own color, which can change according to its physical condition. Most studies of color have dealt with pollen packed into pellets on the legs of bees (corbicicular pollen). Hodges (1952) has made a study of this subject. Reiter (1947) compared the color of anther and corbicicular pollen. The pollens tested in the experiment were probably not of the same color as either anther or corbicicular pollen, but our tests indicate the probable unimportance of color as a factor in pollen attractiveness.

The conditions of the experiment were not such as to affect the ranking of the pollens in relation to moisture content, and since laboratory determinations of moisture showed no association with attractiveness it appears that moisture content had no influence on selection.

Most of the sugar present in corbicular pollen is due to the nectar added by the bee as she packs it into pellets on her hind legs. Although such high sugar content is not normally encountered in the field by the bee, it did not appear to increase attractiveness since the most attractive pollen, black mustard, had the lowest sugar content.

There is evidence (Brues 1946, Auclair and Maltais 1950) that some insects choose their food on the basis of its nutritive value, but there is no evidence that honey bees do so.

Analyses of the pollens used in the 1953 tests (Table 1) indicate considerable differences in their crude protein content. However, since the least attractive pollen, alfalfa, has a protein content equal to the most attractive, black mustard, it seems safe to assume that some other factor was responsible for the differences in attractiveness observed.

Bethier (1941), Brues (1946), and others have demonstrated with various insects that odor is important in enabling a phytophagous insect to recognize a host plant. Ribbands (1953) cites the work of a number of investigators in pointing out the importance of the sense of smell to honey bees. It is therefore natural to suppose that odor may be important in the relative attractiveness of pollens. Odor intensity was the only factor studied that seemed to have any association with selection, and this was merely on the basis of the more odorous pollen to human sensibilities being also the one most preferred by the bees. This evidence was somewhat weakened by the failure of methyl salicylate to affect the order of preference. Either the chemical did not sufficiently mask the pollen odors or else odor did not influence the selections.

Summary

Experiments were conducted at Logan, Utah, to ascertain whether honey bees (Apis mellifera L.) show preferences for different pollens when their choice is not influenced by the structure, color pattern, or perfume of the flower in which the pollen is normally found.
A number of pure pollens collected in pollen traps, separated, and
ground to fine powders were exposed to honey bees before pollen from
natural sources was available. The bee visitors to each container of pollen
were counted and its relative attractiveness thus determined.

Pollen from black mustard was consistently the most attractive. A
mixture of the pollens used in each test was second in attractiveness.
Sweet clover was third, followed by gumweed, greasewood, povertyweed, and
alfalfa, always in that order.

Freshness of pollen, color and reflectivity, moisture, sugar, and
protein content appeared to have no influence on the attractiveness of the
pollens. Black mustard was chosen by a test panel as being the most odorous
pollen. An attempt to mask the odors of the pollens with methyl salicylate
did not change the order of attractiveness, thus indicating that either the
odors of the pollens were not sufficiently masked or that some factor other
than odor is involved in the attractiveness of pollens.

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