

A Detached-leaf Method to Study Pecan Aphid Behavior and Biology¹

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ABSTRACT A detached-leaf method for the study of blackmargined aphid, *Monellia caryella*, yellow pecan aphid, *Monelliopsis pecanis*, and black pecan aphid, *Melanocallis caryaefoliae*, the three species of aphids that infest pecan, *Carya illinoensis*, trees is described. Petri dishes containing 15 ml of 1% water agar were used for the support of 4-cm², surface sterilized, pecan leaf pieces. Compared to previously employed methods, this technique is a low cost, rapid and less labor intense method for the study of aphid biology and behavior, and enables the selective rearing of these aphid species.

KEY WORDS photoperiod, life cycle, development, sexual phase.

The following three species of aphids are severe and potentially production-limiting insect pests of commercial pecan, *Carya illinoensis* (Wangenh.) K. Koch: black margined aphid, *Monellia caryella* (Fitch), yellow pecan aphid, *Monelliopsis pecanis* Bissell, and black pecan aphid, *Melanocallis caryaefoliae* (Davis) (Neel et al. 1985; Tedders 1978). As a result of pesticide overuse and the subsequent development of chemical resistance, growers now have difficulty controlling these pests by chemical methods (Neel et al. 1985). Large numbers of aphids severely injuring the current year's foliage can result in a reduction in nut quality (Tedders and Wood 1985; Tedders et al. 1982; Wood et al. 1987). Severe injury by aphids to pecan foliage also affects tree carbohydrate reserves for the winter, which regulates flowering of the tree the following spring (Tedders and Wood 1985).

Increased information on the behavior and biology of these three aphid species is required if alternative methods of control are to be developed. It is likely that these aphids have unknown vulnerabilities in their life history, behavior or physiology which could be exploited for biological control purposes. However, biological studies of pecan aphids are notoriously difficult due to their small size and delicate structure. These studies have been further complicated by the prolific reproduction rates of aphids, their copious excretion of honeydew, and the tendency of the winged adult forms to jump and fly when handled. Therefore, secure transparent cages that minimize aphid handling while providing containment for easy observation and that can be easily cleaned are required for aphid studies.

In the past, our field and laboratory studies and rearing of pecan aphids utilized pecan seedlings or mature trees as host plants. Large numbers of seedlings were usually required both in biological studies for adequate replication and in

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rearing for adequate aphid numbers. The use of pecan seedlings for these purposes was labor intensive, expensive, and required much space. Also, pecan seedlings could not provide aphids of a selective stage and known age in adequate numbers required for critical biological, behavioral and physiological studies. This paper describes an inexpensive, simple technique which uses detached pecan leaves from seedlings or from mature trees and satisfies the requirements of our aphid studies while enabling the selective rearing of aphid species.

Materials and Methods

Water-agar plates were prepared using deionized water and 1% agar (Difco). After autoclaving, 15 ml of the agar was dispensed into 150-mm plastic petri dishes. Fully expanded mature leaves from greenhouse-grown seedlings or mature trees in the field were collected and washed for 5 min in running deionized water. The leaves were then cut to give ca. 4 cm × 4 cm squares. Squares were subjected to a wash of 1:3 chlorox:water for 2 min, then rinsed four times with sterile deionized water. The leaf sections were subsequently placed on water-agar plates with the adaxial (upper) surface of the leaf in contact with the agar. The petri dishes containing the leaf pieces were maintained at laboratory conditions or in variable bio-climatic cabinets.

Results and Discussion

A detached-leaf method for aphid studied was described by Lakin (1972) which was a modification of Hughes and Woodcock's (1965) previously described method. This method was significantly different than ours in that a nutrient solution was used to support leaf discs which were placed with the lower surface of the leaf in contact with the solution.

In our studies, initial experiments provided evidence that the lower leaf surface was a better habitat for the three aphid species than the upper leaf surface. The aphids, once placed on a leaf segment, would remain and readily establish a feeding site on the lower leaf surface whereas they moved off the upper leaf surface and into the agar. This preferential selection of the lower leaf surface by blackmargined, yellow, and black pecan aphids was also observed in the orchard by Tedders (1978). The petri dish was maintained in an inverted position, orienting the leaf piece in a nearly normal position with the abaxial (lower) surface facing downward. Thus, aphids also were in their natural position. This orientation reduced any problems associated with condensation and water in the petri dish. These water-agar petri dishes allowed aphids to leave the leaflet piece and return without drowning.

This technique proved useful in studying many aspects of aphid biology and life history, including determination of the effects of temperature on their development (Table 1) and the effects of shortened photoperiod on expression of the sexual phase of the life cycle (Table 2). Also, aphids were easily photographed while on the plates (Fig. 1).

We feel this method of maintaining aphids on detached leaves is a useful research tool for the study of these and other aphid species. The leaf tissue remains intact and viable for at least 21 days, which is about 2.5-3 generations of these aphid species (Tedders 1978). This technique not only allows for the study

Table 1. Developmental time, percent survival, and time required for initiation of reproduction by *Melanocallis caryaefoliae*, *Monelliopsis pecanis*, and *Monellia caryella* on detached-leaf pieces in petri dish chambers at photoperiod of 13:11 (L:D) hr and 25, 30, and 35°C.

°C	Species	No. beginning 1st instar aphids	No. days required for maturity	% aphids maturing	No. days required for first reproduction
25	<i>M. caryaefoliae</i>	15	10-11	100	10
	<i>M. pecanis</i>	21	10-11	100	10
	<i>M. caryella</i>	15	7-10	80	10
30	<i>M. caryaefoliae</i>	15	8	93	8
	<i>M. pecanis</i>	21	7-10	100	7
	<i>M. caryella</i>	15	6-8	73	9
35	<i>M. caryaefoliae</i>	15	—	0	—
	<i>M. pecanis</i>	21	—	0	—
	<i>M. caryella</i>	15	—	0	—

Table 2. Effects of photoperiod of the production of sexual forms of the three pecan aphid species. Asexual adults of each species were placed on the leaf, kept for 24 hrs at a 15:9 (L:D) photoperiod to produce the 1st-instar nymphs, then removed. The plates containing 1st-instar nymphs were placed in environmental chambers at 25°C and observed for production of adults at the indicated photoperiods. The symbol (-) denotes no production of sexual forms (+) denotes production of sexual forms.

Species	Photoperiod (hr of light)					
	15	14	13	12	11	10
<i>Monellia caryella</i>	-	-	-	-	+	+
<i>Monelliopsis pecanis</i>	-	-	-	+	+	+
<i>Melanocallis caryaefoliae</i>	-	-	-	+	+	+

of the aphids themselves but of the interaction of the aphid with predators, insecticides, or microbes. Due to the ease of maintaining the aphid colonies, plant material and petri-dish leaf chambers, large numbers of replicates of an observation can be prepared with low labor input and low cost. Furthermore, this technique can provide aphids of a selective stage and known age.

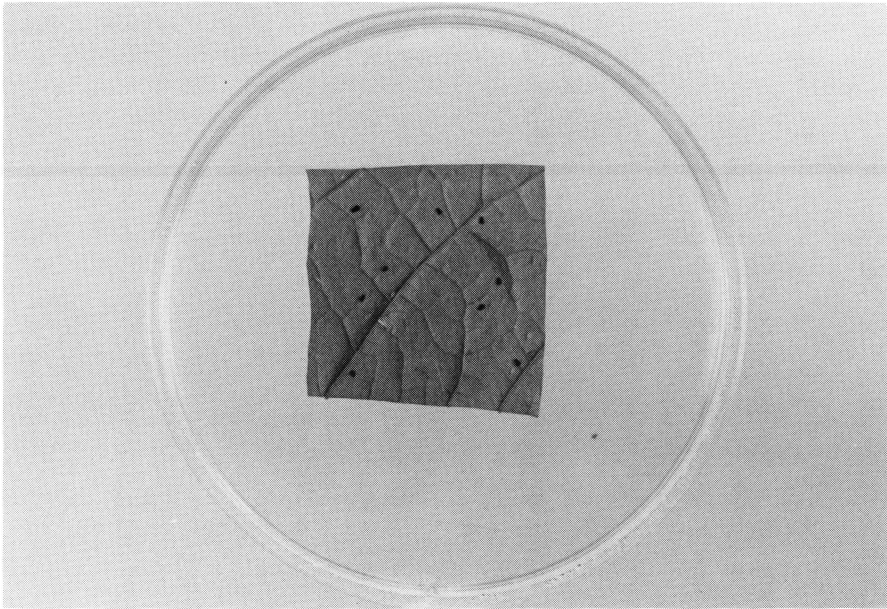


Fig. 1. The detached leaf method for pecan aphid studies.

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