

Section 7. Mating Disruption/SIR

MICROSCOPIC ANALYSIS OF TREE LEAVES SPRAYED WITH MICROCAPSULE FORMULATIONS

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The application of foliar sprays, including sprays that contain microcapsules (MECs), is recognized as a complex process. Microcapsules applied as foliar sprays are intended to impact leaves, stick to the leaves and remain there until they no longer function. Leaves are intended to act as a support platform for MECs while they release their actives by vaporization into the surrounding environment or until they are consumed by insects feeding on the leaf. Which mode of action is used in a specific case depends upon the nature of the actives carried by a MEC.

Significantly, MECs sprayed as a foliar spray do not necessarily stick or adhere to a leaf surface. There are several reasons for this. For example, they may not strike a leaf during the spray process. If they strike a leaf, they may immediately rebound off the leaf due to their kinetic energy. If they strike a leaf and remain on the leaf as part of a water droplet, they may drain off the leaf along with excess water droplets. Even if a MEC initially adheres to a leaf surface, it may fall off the leaf later due to leaf agitation caused by wind, washing off caused by rain, or a combination of wind and rain that occurs in a storm. These combined factors may result in loss of a large fraction of sprayed MECs from leaves, and thereby affect the degree of pest control they provide. Accordingly, defining the distribution of MECs on leaves sprayed with MEC formulations as a function of time after spraying should provide valuable insight into the fate of sprayed MECs. Due to the small size of MECs, this requires microscopic analysis of leaf surfaces.

Scanning electron microscopy (SEM) was used in this study. Samples (1 cm x 0.5 to 0.8 cm) of the top and bottom surface of each sprayed leaf examined were cut and placed directly on standard SEM stubs of 1.2 cm diameter. The surface area of each sample was then examined systematically in the SEM. Leaves from sprayed walnut, almond, sweet gum, pear, peach and apple trees were examined. Sprayed peach and apple leaves were provided by Dr. L. Gut (Michigan State University). Number of leaves examined that were field sprayed with MECs: 18 (top and bottom surface); 4 (top or bottom only). Control surfaces sprayed in the laboratory with MECs (5 leaves and 2 non-leaf) or never sprayed with MECs (3 leaves top and bottom) were examined for reference purposes.

The surface of all leaves examined contained particles. The number of particles varied dramatically and randomly from sample to sample. There also was great variation in the nature of the particles present. Many were irregularly shaped and obviously not microcapsules or microcapsule debris. They appear to be inorganic particles. Many

spherical or sphere-like particles observed had a complex surface texture. This clearly indicates that such particles were of biological origin and not MECs. Other spherical particles appear to be MECs based on comparison with the SEM of capsules located on reference leaf surfaces sprayed in the laboratory. However, such comparisons do not provide absolute confirmation. All particles located on the surface of a field sprayed leaves that appear to be MECs have a diameter of 5-20 μm . Larger capsules have not been observed, although they may be present in the MEC formulation being sprayed. In general, it is not easy to decipher if a specific particle located on a field sprayed leaf surface is a MEC. This is due in part to the infrequent appearance of particles that have the general shape of a MEC and uncertainty of what geometrical changes a MEC can experience when it is field sprayed onto a leaf or after it has been on a leaf for a finite period. In order to confirm that a specific particle is a MEC, it will be necessary to tag each MEC with a characteristic feature not carried by inorganic or organic particles found in orchards.

It is logical to question whether or not it is reasonable to expect to find a MEC in the small area (0.5-0.8 cm^2) examined in a SEM or any other microscopic analysis. The problem with any microscopic assay procedure is that the area evaluated decreases as the degree of magnification increases. Thus, one is always faced with the question of whether or not the area examined by reasonable effort is sufficient to give a representative picture of the total surface. The answer is determined by the number of MECs that should be present per unit area of leaf surface. This number will be a function of the size of the MECs sprayed, the dose of MECs sprayed, the amount of leaf surface area available to a sprayed MEC and, of course, the fraction of sprayed MECs that stick or adhere to leaf surface. Mean MEC size and dose sprayed are numbers that are readily available. Since the mass of a microcapsule decreases with capsule diameter cubed, the number of capsules required to carry a fixed weight of actives increase greatly as capsule size decreases. For capsules of 10 μm diameter, a spray dose of 1 gm of actives requires approximately 2×10^9 capsules (capsule density: $1 \text{g}/\text{cm}^3$; 80 wt. % actives). This is a large number of particles. Rough estimates of leaf surface area for specific orchards sprayed can be made. Available leaf area will vary greatly from orchard to orchard depending upon tree size, planting density and leaf area per tree. In this study, variations in leaf size were very apparent. For example one sample of sprayed peach leaves was small ($3 \times 0.9 \text{ cm}$) while others taken from the same orchard later in the growing season were much larger ($13 \times 3 \text{ cm}$). The former were sprayed early in the growing season and were not fully developed whereas the latter were sprayed after they had fully developed. Thus, the total leaf area available for MEC adhesion in the same orchard was much smaller in the former case than the latter case. Of course, the fraction of sprayed capsules that adhere to a leaf is unknown and is a parameter that microscopic analysis of sprayed leaf surfaces may provide some insight.