EFFECT OF STREPTOMYCIN BLIGHT SPRAYS ON NUT DROP

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ABSTRACT

We investigated the basis for walnut drop following applications of streptomycin (Agrimycin) as blight control measure. Flowers and fruitlets were collect from five treatments, including an unsprayed control, from prebloom through late post-bloom. Samples were collected regularly beginning at the time of the first pre-bloom spray and extending through the period of nut drop. These samples were fixed and prepared for histological examination. In treatments with a high incidence of nut drop no embryo developed in the young fruit. Examination of the stigma and style showed that pollen tube growth was inhibited in flowers from these treatments. The results indicate that streptomycin inhibits pollen tube growth which precludes fertilization. The pattern of development and timing of nut drop following streptomycin treatment at full bloom is similar in all ways to that of unpollinated walnut flowers where growth appears normal through 3 to 5 weeks when nut drop occurs. If streptomycin were to become available as a walnut blight control measure, sprays timed to coincide with the period of pistillate bloom and pistillate flower receptivity should be avoided.

OBJECTIVES

Streptomycin can be an effective control measure to reduce losses to walnut blight disease. Previous experiments have shown that streptomycin sprays can result in high incidence of nut drop several weeks after the spray treatment. Our objective in this project was to determine the mechanism by which streptomycin causes nut drop.

PROCEDURES

Streptomycin sprays (200 ppm) were made in a ‘Vina’ orchard in Durham as shown in Table 1.

Each treatment had four replicates. Four flower pairs were collected from each replicate for each treatment as appropriate on 21, 28 April and 6, 13, 20 May. By 20 May, one month after full bloom, extensive nut drop had occurred in treatment 2. Flowers were separated into ovary and stigma-style portions with each fixed separately in FAA. For the ovaries and the ovaries of older material, the tissue was embedded in Paraplast Plus, sectioned at a nominal thickness of 8μm, stained with safranin-fast green. The stigma-style portions were softened, gently squashed on slides, stained with alkaline aniline blue and observed with a fluorescence microscope. Pollen tube growth at the stigma and at the base of the style was recorded for 20 stigmas from treatments 1, 2 and 5.

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Table 1. Streptomycin Treatments

<table>
<thead>
<tr>
<th>SPRAY STAGE</th>
<th>SPRAY DATE</th>
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<tr>
<td>1  Bud Break</td>
<td>6 April</td>
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<tr>
<td>2  Pre-, Full- and Post-Bloom</td>
<td>14, 21, 28 April</td>
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<tr>
<td>3  Post-postbloom</td>
<td>6 May</td>
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<tr>
<td>4  Bud Break and Post-postbloom</td>
<td>6 April, 6 May</td>
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<td>5  Untreated control</td>
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RESULTS

All nuts appeared to develop normally though the period of nut drop. Examination of the microscope sections revealed that this normal appearance was deceptive as internal tissues of fruitless from the number 2 treatment failed to develop normally. Representative samples from 13 May are shown in Fig. 1-3. Ovules from this material were approximately 2/3 the size of normal ovules, tissue necrosis was evident at the apical region of the integument, nucellar tissue had grown beyond the micropyle, lacunae developed at the juncture of the chalaza and the funiculus, and nucellar tissue had begun to degenerate.

By 13 May, ovules from untreated controls (Fig. 2) had early stages of embryo growth evident and extensive endosperm development; treatment 2 samples had neither.

Numbers of pollen tubes growing in the stigma and at the base of the style is shown in Fig 4. Pollen tube numbers were reduced somewhat in the prebloom treatment and reduced extensively in the full bloom spray where pollen tubes were present at the style bases in only four of the 20 samples examined.
Fig. 1-3. Ovules from fruitlets collected on 13 May. 1. Apical region of ovule from treatment 2. 2. Apical region of ovule from untreated control. 3. Basal region of ovule from treatment 2. Arrow and insert in Fig. 2 illustrates the embryo. Arrow in Fig. 3 shows lacuna at the funiculus-chalaza juncture. F, Funiculus; I, Integument; N, Nucellus.

Fig. 4. Mean number of pollen tubes growing in the stigma and to the base of the style for treatments 1 (prebloom spray), 2 (full bloom spray) and 5 (unsprayed control). N=20 for each.
Pollen tubes from treatments 5 and 2 are shown in Figs 5-9. Pollen tube growth from the controls (Fig. 5) was normal; long pollen tubes with regular callose plugs grew unbranched through the stigmatic tissue to the style base. Pollen tubes from the full bloom spray treatment showed extensive aberrant growth. These tubes were generally short, branching and irregular tube wall structure was common (Fig. 6, 9), and tubes became occluded by callose deposition at their tips (Fig. 7, 8).

Figs 5-9. 5. Pollen tubes from treatments 5 (control). Note that pollen tubes are long, straight and have regularly spaced callose plugs. 6-9. Pollen tubes from treatment 2 (full bloom spray). Tube growth is aberrant with branching (6), short tubes with callose plugs at their tips (7,8) and irregular cell wall growth evident.

DISCUSSION

Our results show that streptomycin acts to inhibit pollen germination and to disrupt growth of pollen tubes that had germinated. In most species, failure of pollination would result in a drop of flowers shortly after bloom. In walnut the drop associated with streptomycin spray treatments does not occur until 3 to 5 weeks after bloom. This is consistent with the growth of unpollinated walnut flowers which are also retained on the tree for 3 to 5 weeks.

Streptomycin sprays that avoid the pollination period, i.e. before pistillate flowers emerge or after
the bloom period have minimal fruit drop associated with them. This is consistent with our findings where no aberrant growth or tissue damage was associated with these spray treatments. There does not appear to be any adverse effect on the flowers or developing fruitlets attributable to streptomycin other than the inhibition of normal pollen tube growth that prevents fertilization and subsequent fruit set.