FLOWERING AND POLLEN STERILITY RESPONSES OF PEANUT PLANTS TO FOLIAR APPLICATIONS OF MALEIC HYDRAZIDE[1]

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To determine its possible use as a male gametocide and, thus, eliminate hand-emasculation in controlled crosses, maleic hydrazide, in concentrations ranging from 50 to 1,000 ppm, was applied to foliage of the peanut, *Arachis hypogaea* L., in the greenhouse before flowering. Little effect on early flowering or pollen fertility was noted at 50, 100, and 200 ppm. Initial flower production was increased and occasional, small reductions in pollen fertility were noted at 250 and 500 ppm. Much reduced early flowering and maximum pollen sterility (80%), for a brief period, occurred at the 1,000 ppm level.

In plant breeding, the time and expense of careful emasculations for controlled hybridizations may become a limiting factor. This is the case in the highly self-pollinated peanut plant, *Arachis hypogaea* L. Use of an effective chemical male gametocide would be of great advantage in peanut breeding because emasculations are tedious, time-consuming, and usually have to be accomplished at inconvenient "after" hours, *i.e.*, 8:00 to 11:00 p.m.

Chopra, Jain, and Swaminathan (1) reported that foliar applications of maleic hydrazide (MH) caused pollen sterility in wheat, onions, and tomatoes. In tomatoes there was high selectivity for pollen sterility with little reduction in egg fertility.

This study was conducted to determine if maleic hydrazide could cause pollen sterility in peanuts.

MATERIALS AND METHODS

In experiment 1, seeds of Starr variety peanuts (*Arachis hypogaea* L.) were planted, in the greenhouse, on October 11, 1966, in a flat in a mixture of equal parts of peat and perlite. The emerged seedlings were transplanted, one per pot, to sandy loam soil in 6 inch standard clay pots on October 31. The plants were arranged on a bench in a randomized block design, watered, and fertilized to maintain good growth. They were grown in a fiberglass greenhouse at 21 to 29 C. The following foliar treatments were applied to each of two plants on one day, *i.e.*, November 7, 10, or 21: a) water spray (control), b) 50 ppm of maleic hydrazide (MH), c) 100 ppm of MH, or d) 200 ppm of MH. Applications were made with a small, hand sprayer until the foliage was wet. Approximately 5 ml of solution was applied per plant. A wetting agent was not used. The source of MH (MH-30) was the United States Rubber Company.[2] Dilutions were made with distilled water. Flowers were removed, beginning with their first appearance (Nov. 30) and counted daily through Dec. 9. Whole flowers were stored dry in envelopes until pollen analyses could be made. To ascertain pollen sterility, pollen staining tests, utilizing aniline blue-lactophenol or acetocarmine, were made by counting the number of unstained grains from a random sample of 500 grains collected from 1 to 3 flowers per plant. When fewer than 500 grains were available, all of the grains were used for the counts. A microscope fitted with an eyepiece net reticle was used for pollen counts from flowers collected on Dec. 1, 4, 7, and 12.

In experiment 2, the technique was similar to the above except that single seeds were planted directly in 6 inch clay pots, on Dec. 7, 1966, and the seedlings were

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sprayed on Dec. 23 with one of the following: a) water (control), b) 250 ppm MH, c) 500 ppm MH, or d) 1,000 ppm MH. Flowers were removed and counted daily from Dec. 27 (first flower) through Feb. 6. Pollen staining counts were made from flowers collected on Jan. 1, 5, and every 3rd day thereafter through Feb. 4. In cases where both of the similarly treated plants failed to flower on a pollen sampling day, flowers from the preceding or following day were used. After the flower collection periods ended, plants were grown until harvested for seed on April 14.

RESULTS AND DISCUSSION

Data from the experiments are presented in Tables 1 and 2 and Figure 1. In Experiment 1, flowering commenced on one or more of the plants receiving each of the treatments on Nov. 30. The MH treatment appeared to have little effect on flower production and no statistically significant differences could be assessed to the amount of MH used or the time of spraying. An occasional instance of small decrease in pollen stainability was observed after plants were treated with 100 and 200 ppm MH (Table 2), but no reduction was great enough to suggest effectiveness of the treatment in breeding techniques. Therefore, higher concentrations of MH were tried in Experiment 2.

TABLE 1. *Total flowers per plant. Experiment 1*[a].

Treatment	Spray date Nov. 7	Nov. 10	Nov. 21
Water (Control)	22	26	18
Maleic hydrazide			
50 ppm	28	24	24
100 ppm	22	17	15
200 ppm	20	13	20

[a] Average of two similarly treated plants (Nov. 30-Dec. 9).

TABLE 2. *Percentage of stainable pollen. Experiment 1.*

Treatment	Spray date	Pollen sample date Dec. 1	Dec. 4	Dec. 7	Dec. 12
Water (control)	Nov. 7	96.4	97.9	96.3	96.4
Maleic hydrazide					
50 ppm		94.7	96.5	95.1	97.2
100 ppm		94.3	95.8	96.6	90.1
200 ppm		94.0	89.9	90.8	95.7
Water (control)	Nov. 10	96.0	95.9	96.5	97.0
Maleic hydrazide					
50 ppm		95.5	95.8	95.1	97.0
100 ppm		94.4	95.0	92.4	93.5
200 ppm		93.1	91.4	92.4	86.7
Water (control)	Nov. 21	97.2	97.4	96.3	95.5
Maleic hydrazide					
50 ppm		95.9	94.6	97.0	97.0
100 ppm		97.8	97.6	97.0	92.9
200 ppm		97.2	97.2	96.4	93.0

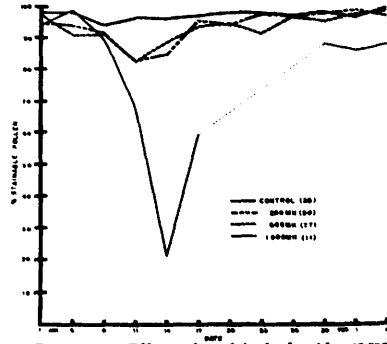

FIGURE 1. Effect of maleic hydrazide (MH) on pollen stainability. Numbers in parentheses (in key) are total numbers of flowers per plant through Feb. 4. No flowers were produced on 1,000 ppm MH-treated plants from Jan. 19 through Jan. 28.

Greater differences in flower number and pollen stainability were associated with the MH treatments in Experiment 2 (Fig. 1). Dates of the appearance of the first flower according to treatment were: 250 and 500 ppm MH, Dec. 27; 1,000 ppm MH, Jan. 1; control, Jan. 3. Flower production was greatest in the 250 and 500 ppm treated plants and lowest where 1,000 ppm had been applied. Differences in total flower number were statistically significant (LSD = 17) for all treatments except that those resulting from use of 250 and 500 ppm MH were not significant from each other. Significant reductions in pollen staining occurred at the 250, 500, and 1,000 ppm MH treatment levels on Jan. 11 and 14. However, stainability of the pollen from plants treated with 250 and 500 ppm MH returned to normal by Jan. 17. Only one of the two plants among those receiving 1,000 ppm MH bloomed during the test period, and its pollen stainability reached a minimum (20.8%) on Jan. 14 and remained below that resulting from other treatments throughout the sampling period.

The effects, if any, that MH may have had on egg fertility were not determined in these experiments. Apparently the plants recovered from all MH treatments; there were no statistically significant differences in seed yield at harvest. Mean seed number and weight (in grams) per plant were, respectively: 17 and 5.2 g for controls; 18

and 6.1 g for 250 ppm MH plants; 14 and 5.2 g for 500 ppm MH plants; 15 and 4.1 for 1,000 ppm MH plants.

Naylor and Davis (2) reported poor flowering of peanuts in a greenhouse experiment where a concentration of 500 ppm MH was applied to seedling plants and complete inhibition of flowering following treatments with 1,000, 2,000, and 4,000 ppm MH. In my experiments, plants receiving 500 ppm MH showed increased flowering. Differences in results may be due to differences in time of application, purity of the MH (Naylor and Davis recrystallized theirs), or environmental conditions, *e.g.*, their greenhouse was cool (21 C). Admittedly, the number of replicate plants treated in my experiments were smaller than desired to give conclusive results.

In peanut breeding, hand pollinations are tedious and seed yields for each successful hand-pollination are low (1 or 2 per fruit). To be useful in peanut breeding, a male gametocide should approach 100% effectiveness on the pollen and have little, if any, effect on the egg. Otherwise, the extra pollinations, which would be required if some female sterility were induced, or the confusion created if a large amount of unexpected selfing occurred, could outweigh the advantage of using a gametocide.

Not to be overlooked, however, is the possibility that moderately low decreases in pollen fertility might be useful in promoting significantly greater natural crossing of peanuts by bees under field conditions. Advantage might be taken of increases in natural crossing by using Hammons' pedigreed natural crossing scheme (3). Proper spacing of plants in the field could allow for male gametocide treatment of the seed parent, while use of an untreated pollen parent with a dominant genetic marker would permit the detection of hybrid progeny.

Of interest is the observation that, in my experiments, MH appeared to increase or decrease initial flowering, depending upon the amount applied. It might be possible, with the proper chemical and application procedure, to promote abundant peanut flowering early in the season, and to "turn off" flowering after sufficient pods were initiated without affecting the seed maturation processes. Such a procedure for insuring uniformly mature seed would be of considerable value in peanuts where the long flowering period contributes to undesirable differences in maturity ranges of harvested seed.

From the data presented here it appears that it may be difficult, if not impossible, to produce adequate numbers of "male-sterile" flowers, for acceptable periods of time, with maleic hydrazide and, thus, to eliminate emasculations in hand-crossed peanuts. Future experiments utilizing maleic hydrazide (and other chemicals) for control of flowering and pollen fertility in peanuts, should be aimed at determining the critical concentration of the chemical and timing of application.

REFERENCES

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