

## Gibberellin promotion of physiological leaf spot in detached 'Golden Delicious' apple leaves\*

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Accepted: 3 January 1975

### Summary

Physiological leaf spot development in detached 'Golden Delicious' apple leaves was accelerated when cultured in solutions containing GA<sub>3</sub> or GA<sub>4+7</sub> at 10, 50 and 100 mg/litre for 72 h. The interaction of PBA and GA<sub>4+7</sub> applied simultaneously was highly synergistic in increasing the severity of the disorder over either growth substance alone.

### Introduction

Leaf spot, a non-pathogenic disorder specific to the apple cv. 'Golden Delicious', may appear on mature leaves in the orchard usually in July and August (Jonkers, 1973a). The incidence of the large irregular necrotic spots generally results in premature abscission of the affected leaves. Environmental conditions which tend to provoke the disorder are low light intensity, specific temperature regimes, and vigorous shoot growth (Jonkers, 1973b). Leaf spot severity is also governed by season. The physiology of leaf spot is unclear although we have shown that exogenous GA<sub>4+7</sub> accelerates leaf spot and ABA retards it in intact trees (Kender & Jonkers, 1975).

To understand more of the hormonal implications of leaf spot development these experiments were conducted to study the effects of specific growth substances on detached 'Golden Delicious' apple leaves.

### Material and methods

Fully expanded leaves on terminal shoots were collected from 6-year old fruiting 'Golden Delicious' apple trees on M.9 rootstocks during July, 1974. Average leaf area was 66.3 cm<sup>2</sup>. Approximately 15 % of the foliage exhibited leaf spot during

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GIBBERELLIN PROMOTION OF PHYSIOLOGICAL LEAF SPOT IN APPLE

this period. Leaves (with petioles) were detached from nodes 12 to 16 shoots 40–60 cm in length and brought immediately to the laboratory for treatment. Ten replicate petioles were placed in 200-ml beakers filled with solutions containing the respective growth substances. The petioles were inserted through an aluminium foil cover over each beaker to prevent the blades from contacting the solution. Leaves were maintained at 21 °C under continuous light (33.6 W/m<sup>2</sup>) as suggested from previous data on intact trees (Jonkers, 1973a). Solutions were replaced daily.

*Experiment 1.* Leaves were cultured in solutions containing either 6-(benzylamino)-9-(2-tetrahydropyranyl) 9H-purine (PBA), a mixture of gibberellins 4 and 7 (GA<sub>4+7</sub>), gibberellic acid (GA<sub>3</sub>) or abscisic acid (ABA) at 10, 50 and 100 mg/litre, indoleacetic acid (IAA) at 1, 10 and 25 mg/litre, or water. The experiment was replicated 2 times, first with unspotted leaves and second with leaves exhibiting one spot per leaf prior to detachment. The number and area of new spots were recorded on the same leaves after 24, 48 and 72 h.

*Experiment 2.* Detached unspotted leaves, as described above, were cultured in solutions containing either PBA at 0, 1, 10 and 100 mg/l, GA<sub>4+7</sub> at 0, 1, 10 and

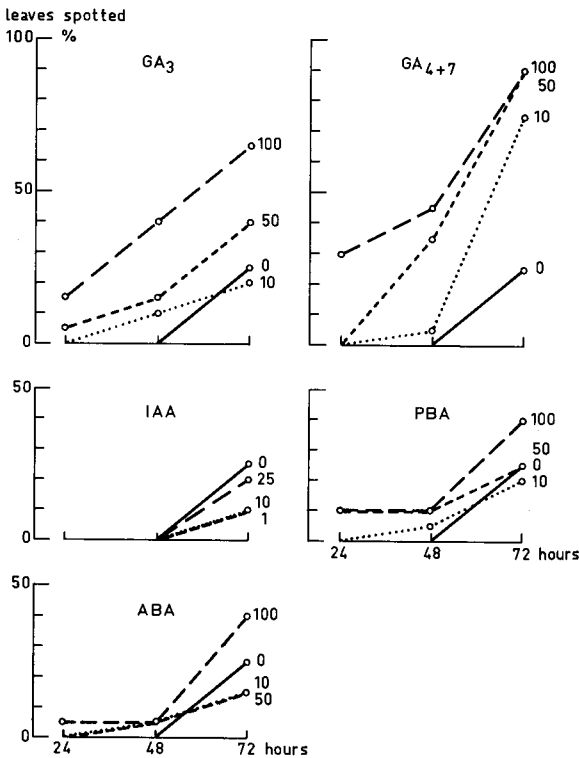


Fig. 1. The effect of GA<sub>3</sub>, GA<sub>4+7</sub>, IAA, PBA and ABA at various concentrations on the percentage of detached 'Golden Delicious' apple leaves developing leaf spots after 24, 48, and 72 h.

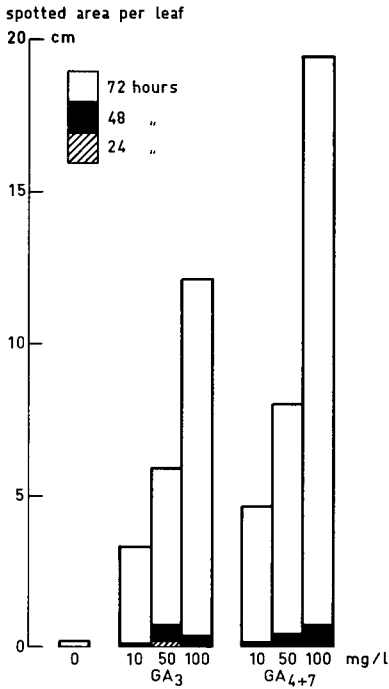


Fig. 2. The effect of GA<sub>3</sub> and GA<sub>4+7</sub> at 10, 50, and 100 mg/litre on the total spotted area per leaf of detached 'Golden Delicious' apple leaves after 24, 48 and 72 h.

100 mg/l, or all combinations of concentrations of the 2 growth substances (Table 1). The number and area of spots per leaf were recorded after 72 h.

**Results**

*Experiment 1.* After 24 h the percentage of leaf spot on detached 'Golden Delicious' apple leaves was minimal in all growth regulator treatments, except GA<sub>4+7</sub> at 100 mg/litre which exhibited 30 % leaf spot within 24 h (Fig. 1). After 48 h, GA<sub>4+7</sub> at 50 and 100 mg/litre and GA<sub>3</sub> at 100 mg/litre increased the percentage of leaves which developed leaf spots. After 72 h, GA<sub>4+7</sub> at all concentrations and GA<sub>3</sub> at 100 mg/litre markedly stimulated the number of leaves with spots. The other growth substance had little effect on the percentage leaves showing spots. Since there was hardly any difference in leaf spotting between previously spotted and unspotted leaves these data have been combined in Fig. 1 and 2.

The total spotted area per leaf was not influenced by growth substance treatment for the first 24 h. After 48 h GA<sub>4+7</sub> and GA<sub>3</sub> treated leaves started to show a slightly larger spotted area than control leaves and after 72 h all concentrations of GA<sub>4+7</sub> and GA<sub>3</sub> markedly increased the magnitude of spot development over all other treatments (Fig. 2). GA<sub>3</sub> was the least severe of the 2 gibberellin treatments in influencing leaf spot. IAA, PBA and ABA had no influence on the size of the spotted area.

## GIBBERELLIN PROMOTION OF PHYSIOLOGICAL LEAF SPOT IN APPLE

Table 1. The effect of GA<sub>4+7</sub> and PBA alone and in combination at various concentrations on the spotting of detached 'Golden Delicious' apple leaves after 72 h.<sup>1</sup>

GA <sub>4+7</sub> (mg/litre)	PBA	Leaves showing spots (%)	Spotted area per leaf (cm <sup>2</sup> )
0	0	20	0.2
0	1	10	0.2
0	10	10	0.3
0	100	30	0.6
1	0	40	1.0
1	1	30	1.1
1	10	60	1.7
1	100	60	1.9
10	0	50	2.8
10	1	60	2.2
10	10	80	4.2
10	100	90	6.5
100	0	90	12.4
100	1	80	15.6
100	10	100	20.8
100	100	100	32.9

<sup>1</sup> Mean of 10 leaves per treatment.

The number of spots per leaf was found to be an unreliable index of leaf spot severity since spots tended to coalesce in advanced stages of the disorder.

Leaves of the cultivar 'Jonathan' detached from adjacent trees did not develop leaf spotting when treated with gibberellins as described above, thus substantiating the cultivar specificity of leaf spot.

*Experiment 2.* PBA alone had no influence on the percentage leaves developing spots at the 3 concentrations tested (Table 1). GA<sub>4+7</sub> alone at 1 mg/litre or together with PBA at 1 mg/litre did not influence leaf spotting but when the PBA concentration of the solution was increased to 10 and 100 mg/litre the severity of the disorder increased somewhat.

GA<sub>4+7</sub> alone at 10 mg/litre slightly provoked leaf spot and was increased further at that concentration by the addition of PBA at 10 and 100 mg/litre to the solution. At the latter combination some synergism existed in the number of leaves developing spots and a highly synergistic effect was found in the total spotted area per leaf.

GA<sub>4+7</sub> alone at 100 mg/litre markedly increased the severity of leaf spot over control leaves. This effect was greatly magnified by the addition of PBA at 10 and 100 mg/litre to the solutions. Application of GA<sub>4+7</sub> and PBA at high concentrations consistently gave a synergistic effect. This effect increased the spotted area per leaf from 1.4 to 2.5 times that expected from an additive effect. The magnitude of the synergism increased as the concentration of the hormones increased.

## Discussion

It was previously reported that the incidence of physiological leaf spot could be hastened and intensified in leaves of intact 'Golden Delicious' apple trees by foliar applications of  $GA_{4+7}$  (Kender & Jonkers, 1975). Although young expanding leaves synthesize considerable amounts of auxins and gibberellins, mature leaves depend upon the import of these hormones. Our premise in repeating similar experiments with detached leaves was that if gibberellins induce physiological leaf spot detached (non-importing) control leaves should produce no spots while spots should appear on GA-treated leaves. We found, as with intact leaves, that spots could be readily produced by culturing leaves in  $GA_3$  or  $GA_{4+7}$  solutions. However, 20 to 30 % of the control leaves also produced spots and we were unable to demonstrate the causal effect of leaf spot. Although caution was exercised in sampling leaves many nodes from the nearest spot, it is possible that some of the test leaves may have been predisposed to leaf spotting prior to detachment. It is evident nevertheless, that both  $GA_3$  and  $GA_{4+7}$  at high concentrations triggered very rapid development of leaf spot suggesting that leaf spot in 'Golden Delicious' apple leaves is influenced by rapid changes in hormone balance. On leaves of intact trees, ABA retarded but did not inhibit the development of leaf spot (Kender & Jonkers, 1975). Such a response was not detected in detached leaves.

The synergistic effect of  $GA_{4+7}$  and PBA in increasing the severity of leaf spot of detached leaves was of interest since a similar observation was made when the same combination was applied to leaves of adjacent intact fruiting trees but not in young non-fruiting trees in the phytotron.

The use of detached apple leaves indicates that gibberellin promoted leaf spot is not necessarily related to rapid shoot growth but rather a localized phenomenon within the affected leaf.

It is concluded from these data and that from previous work on intact trees (Jonkers, 1973a; Kender & Jonkers, 1975), that external conditions affording rapid synthesis of gibberellins promote physiological leaf spot in 'Golden Delicious' apple trees.

## References

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