

4. ———. 1967. Identification of prunin (naringenin-7-glucoside) in dormant peach buds as a wheat coleoptile growth inhibitor. *HortScience* 2:105-106.
5. ——— and F. B. WIDMOYER. 1970. The effects of gibberellic acid on flower differentiation, date of bloom, and flower hardness of peach. *J. Amer. Soc. Hort. Sci. (In Press)*.
6. DENNIS, F. G., JR. and L. J. EDGERTON. 1960. The relationship between an inhibitor and rest in peach flower buds. *Proc. Amer. Soc. Hort. Sci.* 77:107-116.
7. EL-MANSY, H. I. and D. R. WALKER. 1969. Seasonal fluctuation of flavanones in 'Elberta' peach flower buds during and after the termination of rest. *J. Amer. Soc. Hort. Sci.* 94:298-301.
8. EREZ, A. and S. LAVEE. 1969. Prunin identification, biological activity and quantitative change in comparison to naringenin in dormant peach buds. *Plant Physiol.* 44:342-346.
9. HENDERSHOTT, C. H. and D. R. WALKER. 1959. Identification of a growth inhibitor from extracts of dormant peach flower buds. *Science* 130:798-800.
10. ——— and ———. 1959. Seasonal fluctuation in quantity of growth substances in resting peach flower buds. *Proc. Amer. Soc. Hort. Sci.* 74:121-129.
11. LIPE, W. N. and J. C. CRANE. 1966. Dormancy regulation in peach seed. *Science* 153:541-542.
12. MARTIN, G. C., M. IONA, R. MASON and H. I. FORDE. 1969. Changes in endogenous growth substances in the embryos of *Juglans regia* during stratification. *J. Amer. Soc. Hort. Sci.* 94:13-17.
13. MACMILLAN, J. and P. J. SUTER. 1963. Thin layer chromatography of the gibberellins. *Nature* 197:790.
14. MILBORROW, B. V. 1967. The identification of (+) abscisin II, [(+) dormin], in plants and measurements of its concentrations. *Planta* 76:93-113.
15. NITSCH, J. P. and C. NITSCH. 1956. Studies on the growth of coleoptile and first internode sections. A new, sensitive, straight growth test for auxins. *Plant Physiol.* 31:94-111.
16. PHILLIPS, I. D. J. and P. F. WAREING. 1959. Studies in dormancy of sycamore. II. The effect of day length on the natural growth inhibitor content of the shoot. *J. Expt. Bot.* 10:504-514.
17. SAMISH, R. M. and S. LAVEE. 1962. The chilling requirement of fruit trees. *Proc. XVI Int. Hort. Cong.* pp. 372-388.
18. SONDHEIMER, E. and EVA C. GALSON. 1968. Abscisic acid levels and seed dormancy. *Plant Physiol.* 13:1443-1449.
19. THOMAS, T. H., P. F. WAREING and P. M. ROBINSON. 1965. Action of the sycamore "dormin" as a gibberellin antagonist. *Nature* 205:1270-1272.
20. WAREING, P. F., J. GOOD and J. MANUEL. 1968. Some possible physiological roles of abscisic acid. pp. 1561-1579. F. Wightman and G. Setterfield (eds.), *Biochemistry and physiology of plant growth substances*. The Runge Press, Ltd., Ottawa.

Phenylmercuric Acetate Effects on Water Loss of the Tomato¹

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Abstract. One of paired tomato plants was sprayed with 100 ppm phenylmercuric acetate (PMA). Transpiration rates were measured gravimetrically. During the initial daylight periods, PMA treatment reduced water losses in 2 tests. Conversely, night water losses were higher for the PMA treated plants in both tests. When moisture stress symptoms occurred, water losses by the treated plant were higher. The results indicate that PMA closes the stomates at some small aperture. This reduces transpiration when plants are not stressed for water. Relative increased water losses occur, however, when untreated plant losses would be minimal (dark, wilted).

RECENTLY, much research has centered on the chemical reduction of plant stomate aperture and resultant reduction in transpiration. The ability of phenylmercuric acetate (PMA) to reduce transpiration has been clearly demonstrated. Zelitch and Waggoner (8) report that PMA closed stomates of tobacco and corn. Slayter and Bierhuizen (3) reported that PMA in concentrations of 10^{-4} and 10^{-5} M caused proportionately greater reduction in transpiration than photosynthesis of cotton leaves. Hence, water-use efficiency, expressed as the transpiration ratio (g of water transpired/g of carbohydrates produced), was improved.

The results of Gale (1) and Slayter and Bierhuizen (3) show that transpiration was reduced by film-type plastic antitranspirants. However, Gale (1) noted that under conditions which lead to a high degree of plant moisture stress (hot, dry, sunny weather), the film-type antitranspirants caused increased transpiration. This increase was thought to be due to delayed stomatal closure of the treated plants. Under field conditions, Gale showed an increase in water-use efficiency with film-type antitranspirants.

Use of PMA could result in improved use of existing water supplies in agriculture. Zelitch (6) described a delay in wilting of PMA-treated plants and stated that

"the longer time that the leaf hydration in the treated plants remained above the controls indicated the benefit that might be derived from the closure of stomates prior to a drought."

Zelitch (7) stated that if an applied substance works specifically to reduce the stomate aperture, transpiration rate will be inhibited to a greater degree than the photosynthetic rate. Also, that PMA is the most widely used material for closing stomates; that the probable mechanism of closure is via the formation of mercaptides with the sulfhydryl groups of proteins and membranes; and that PMA probably will not be translocated from a treated leaf or to newly formed ones.

There is a paucity of information on the effect of PMA on horticultural plants cultured in normal circumstances. Granger and Edgerton (2) observed the closure of stomata and injury to apple leaves at concentrations of PMA from 300 to 1000 ppm. Results obtained on PMA effects in a preliminary study on field grown tomatoes in New Mexico indicated that the PMA-treated plants were under a greater moisture stress; leaflets curled upward, exposing the developing fruit; and vegetative growth appeared to be substantially reduced. Much of the exposed fruit (both green and ripe) scalded in the desert sun.

From these results a further study was suggested to determine the effect of PMA on water losses. This paper summarizes results from a greenhouse study conducted

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to determine the water losses of young tomato plants treated with PMA.

METHODS AND MATERIALS

Two similar tests on transplant tomatoes (cv. Manapal) were conducted. In both tests, 2 plants were selected for similarity of appearance and size; one was treated, the other untreated. The treated plants were sprayed to drip with a 100 ppm solution of PMA dissolved in deionized water with 2 drops of Tween-20 per liter of solution. The check plants were sprayed with deionized water containing Tween-20.

After the foliage dried, each pot was placed in a plastic bag. The bag opening was closed around the stem below the cotyledonary leaves and carefully secured with a paper-covered wire to avoid stem injury and prevent moisture diffusion from the rooting media. Each plant was weighed and the time recorded. Subsequent weighings were made at the end of the first day and at the beginning, mid-day, and at the end of the second day. The water-loss measurements were terminated at the end of the second day.

No provision was made for resupplying the water lost, and wilting of some plants did occur. At the termination of the second test, wilting was severe in most plants. When wilting of at least one member of a pair occurred, results of that pair were not included in the analysis of present or future transpirational periods. In the second test, uniform occurrence of wilting during the final period made it possible to compare moisture loss of treated plants under a moisture stress condition. In this case, those pairs with plants which were not wilted prior to the beginning of the period were omitted from the evaluation.

At the end of the experiment, the plastic bags were removed, pots watered, and the plants were allowed to restore turgidity overnight. Leaflets were removed and the fresh weight was determined for each plant within one hour of sunrise. All leaflet tissue was dried in an oven at 65° C for 3 days, and dry weights were determined. In a few cases, leaflet damage caused by wilting was excessive and turgidity was not restored. These plants were not used in evaluating water loss expressed on a leaflet weight basis. Transpiration was assumed to be equivalent to losses in weight noted between weighings. Transpiration rate was expressed as g of water lost per hour on either a plant or per g of fresh or dry weight of leaflet tissue. Leaf area of treated and normal plants was estimated from leaf disc weights of known areas and computation of the total area of each plant from the fresh weights of the leaflets. Mean differences in leaf area varied less than 4 percent and were not statistically significant. No significant effect of PMA on the fresh or dry weight of leaflets was noted.

Both experiments were conducted in a glass greenhouse with heavy shade. The light was diffuse, thus reducing effects of unequal illumination on moisture loss. In a preliminary study, the hourly water loss rate of a plant in the test section was approximately one-half the water loss rate occurring in a comparable greenhouse section in full sunlight without shading.

Specific procedures for each test were as follows: *Test No. 1.* Fifteen pairs of plants (approximately 7 inches tall) were sprayed at 11:00 AM. The first plant weighing was made at 1:09 PM September 3, 1969. This was recorded as the beginning of the test. The final weighing was made at 4:52 PM on September 4 (test end). The maximum temperature and minimum relative humidity

was 97° F at 23 percent. The vapor pressure deficit from saturation was 46 millibars (5). *Test No. 2.* Twelve pairs of plants (approximately 9 inches tall) were treated at 10:35 AM on September 8, 1969. The initial weighing was made at 11:37 AM of the same day. The final weighing was made at 4:23 PM, September 9. Care was taken to carefully delineate water losses occurring in the light and dark periods. The maximum temperature and minimum relative humidity was 97° F and 32 percent. The vapor pressure deficit from saturation was 41 millibars (5).

The results were analyzed statistically with the t-test for each period according to the method described by Snedecor (4).

RESULTS AND DISCUSSION

Transpiration during the daylight hours was significantly reduced by PMA treatment of the tomato plants (Tables 1 and 2). This is consistent with published results (3, 6, 7, 8). Water loss during the dark period was higher for PMA-treated plants and was proportionately higher in the second experiment (48 versus 171 percent). The night period recorded in the first experiment included a few daylight hours in the evening and morning. Since, in the second experiment, the period was purposely confined to dark hours, the data reported more accurately reflect effects of PMA on water losses in the dark than the first test.

In the second experiment, most of the plants wilted during the final period (11:58 AM to 4:23 PM). Under stress conditions, PMA-treated plants lost significantly more water than untreated plants. It might be reasoned that since the water available to all plants was reasonably uniform and finite, and since the PMA-treated plants lost less water initially, the transpiration rate during this period should be higher. A summation of the plant water loss for the entire test showed that 7 of 11 treated plants lost more water than untreated ones. The mean increase in water loss per treated plant was 4.24 grams (approximately 7 percent increase).

Table 1. The effect of PMA treatment on transpiration rate of transplant tomatoes during day and night periods. (Exp. I).

Treatment	Day	Night	Day	
	September 3	September 3-4	September 4	
	1:09-4:29PM	4:29PM-7:27AM	7:27AM-1:56PM	1:56-4:52PM
	g of water lost per hour per plant			
Untreated.....	5.07 ¹	0.514 ¹	3.41 ²	3.95 ³
PMA treated.....	4.02	0.761	2.23	3.09
PMA induced change (%).....	-23.7	+48.1	-34.6	-21.8
Significance.....	**	**	**	*
	g of water lost per hour per g fresh leaf tissue			
Untreated.....	1.033 ²	0.135 ²	0.731 ⁴	0.866 ⁵
PMA treatment.....	0.813	0.229	0.487	0.760
PMA induced Change (%).....	-21.3	+69.6	-33.4	-12.2
Significance.....	**	**	**	*
	g of water lost per hour per g dry leaf tissue			
Untreated.....	5.749 ²	0.768 ²	3.840 ⁴	4.538 ⁵
PMA treatment.....	4.696	1.317	2.716	4.032
PMA induced change (%).....	-18.3	+71.6	-29.3	-11.2
Significance.....	*	**	**	NS

¹Mean of 15 plants.

²Mean of 12 plants.

³Mean of 8 plants.

⁴Mean of 10 plants.

⁵Mean of 7 plants.

*Significant at the .05 level of probability.

**Significant at the .01 level of probability.

NS = not significant.

Table 2. The effect of PMA treatment on transpiration rate of transplant tomatoes during day and night periods. (Exp. II).

Treatment	Day	Night	Day		
	Septem- ber 8 11:37- 7:00PM ¹	Septem- ber 8-9 7:00PM- 6:33AM ¹	6:33- 8:15AM ¹	8:15- 11:58AM ²	11:58- 4:23PM ³
	g of water lost per hour per plant				
Untreated.....	5.42	0.212	0.600	4.97	1.64
PMA treated....	4.32	0.576	0.429	3.05	3.68
PMA induced change (%)..	-20.3	+171.6	-28.5	-38.6	+124.4
Significance....	**	**	NS	**	*
	g of water lost per hour per one g fresh leaflet tissue				
Untreated.....	0.7168	0.0278	0.0807	0.6541	0.2241
PMA treated....	0.5851	0.0792	0.0580	0.4128	0.4947
PMA induced change (%)..	-18.4	+184.9	-28.1	-36.9	+120.7
Significance....	**	**	NS	**	*
	g of water lost per hour per g dry leaflet tissue				
Untreated.....	4.822	0.184	0.403	4.343	1.397
PMA treated....	2.769	0.518	0.377	2.680	3.243
PMA induced change (%)..	-21.8	+181.5	-6.5	-38.3	+132.1
Significance....	**	**	NS	**	*

¹Mean of 12 plants. *Significant at the .05 level of probability.
²Mean of 6 plants. **Significant at the .01 level of probability.
(stressed plant pairs NS = not significant.
omitted).
³Mean of 11 plants.
(non-stressed plant pair
omitted).

Observations made on the severity of wilting also support the proposal that PMA-treated plants lose more water under severe moisture stress. Wilting symptom ratings recorded at 11:55 AM and 2:15 PM showed that untreated plants were more severely wilted. Earlier wilting of untreated plants is consistent with the results of Zelitch (6). At the conclusion of the experiment (4:23 PM), wilting symptoms were rated as equal in treated and untreated plants. However, when plant pairs were compared individually, the PMA-treated plants were more severely wilted.

After termination of the water loss portion of the experiment, plants were watered and observed for resto-

ration of turgidity the following morning. Recovery of all untreated plants was normal, no permanent injury was noted. Permanent injury to some leaves occurred on one-third of the PMA-treated plants. The remaining plants recovered normally. A similar loss of the ability to recover by some treated plants after subjection to severe moisture stress was noted in subsequent tests unreported here.

The results of the tests with PMA confirm the reports that PMA treatment can result in reduced plant moisture losses. If available water is inadequate, however, water losses can increase with PMA treatment. At times, when stomata would normally be closed (dark, high moisture stress), treated plants lose water at a more rapid rate than normal plants. This suggests that PMA treatment fixes the stomate aperture open at some minimal size which, in turn, limits the ability of the plant to restrict or eliminate a small but critical water loss potential. It is possible that the leaf curling symptoms noted in the preliminary field test and thought to be a symptom of greater moisture stress in the field could have occurred when the PMA-treated plants continued to lose water when available soil water became limited.

LITERATURE CITED

- GALE, J. 1961. Studies on plant antitranspirants. *Physiol. Plantarum* 14:777-786.
- GRANGER, R. L. and L. J. EDGERTON. 1966. Effects of two petroleum spray oils and phenylmercuric acetate on the stomata of apple leaves. *Proc. Amer. Soc. Hort. Sci.* 88:48-51.
- SLAYTER, R. O. and J. F. BIERHUIZEN. 1964. The influence of several transpiration suppressants on transpiration, photosynthesis, and water use efficiency of cotton leaves. *Australian J. Bio. Sci.* 17:131-146.
- SNEDECOR, GEORGE W. 1957. *Statistical Methods*. 5th Edition. Iowa State Univ. Press.
- WILLIAM, G. D. V. and R. LEGER. 1967. Vapor pressure deficit, relative humidity and dew point temperatures conversion tables. *Agr. Nat. Tech. Bul. 12. Plant Res. Inst., Can. Dept. Agr. Ottawa, Ont.*
- ZELITCH, ISRAEL. 1967. Control of leaf stomata—their role in transpiration and photosynthesis. *Amer. Scientist*, 55:472-486.
- . 1969. Stomate control. *Ann. Rev. Plant Phys.* 20:329-350.
- and PAUL E. WAGGONER. 1962. The effect of chemical control of stomata on transpiration and photosynthesis. *Proc. Nat. Acad. Sci.* 48:1101-1108.