

IAA reversible growth inhibition of grape shoots (*Vitis vinifera*) by maleic hydrazide compared to gibberellic acid induced growth

by

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Die Hemmung des Triebwachstums von Reben (*Vitis vinifera*) durch Maleinsäurehydrazid und ihre Aufhebung durch IES im Vergleich mit der Wachstumsförderung durch Gibberellinsäure

Zusammenfassung. — Bei bewässerten vollentwickelten Reben der Sorte Madlene wurde die Wachstumsgeschwindigkeit der Triebe untersucht. Zwischen dem 20. und 55. d nach dem Austrieb wurde eine Periode linearen Längenwachstums festgestellt. Maleinsäurehydrazid (MH) verringerte die Wachstumsgeschwindigkeit, beeinflusste aber die Linearität des Wachstumsverlaufs nicht. Das Ausmaß der Wachstumshemmung war konzentrationsabhängig. Die Wachstumsgeschwindigkeit war erst 5 d nach der Applikation meßbar verändert. IES in Konzentrationen bis zu 80 mg/l hatte keinen Einfluß auf die Wachstumsgeschwindigkeit, wenn sie während der linearen Wachstumsphase angewandt wurde. Dies war auch der Fall, wenn Triebe behandelt wurden, deren Wachstumsgeschwindigkeit durch MH-Vorbehandlung verringert war. IES-Applikation während der 5tägigen Zwischenphase nach der MH-Vorbehandlung konnte — in Abhängigkeit von der IES-Konzentration — die MH-bedingte Hemmung jedoch aufheben. Zwischen der MH-induzierten Wachstumsverringerng und der GS₃-induzierten Förderung wurde keine unmittelbare Wechselwirkung gefunden.

Introduction

The annual growth of grape shoots could be divided into three phases: I. Slow growth during the first 2—3 weeks after bud opening in the spring, II. rapid growth during early summer, and III. slow growth from late summer to fall. Inflorescence differentiation in the buds occurs during the rapid growth period (7). A temporary reduction of the growth rate at that time was thought to allow a better and more uniform initiation of inflorescence primordia in the buds as shown in some other species (10, 20). Maleic hydrazide (MH) was described as one of the first plant growth inhibitors (9, 13). It was widely accepted to be a temporary inhibitor of vegetative growth (5, 10, 14, 20). This was explained by increased IAA oxidase activity induced by the MH (4, 11). LEOPOLD and KLEIN (8) working with pea plants suggested MH to be a potent antiauxin. Others (6), however, working with *Avena* coleoptiles have not found such an antagonism between IAA and MH as the IAA could not overcome the inhibition induced by MH. Also, no change in endogenous IAA level due to MH treatment could be found (3). Long term growth inhibitions induced by MH have clearly been shown, particularly in bulbs (19) where sprouting inhibition results. Using 500 mg MH/l (2) some reduction in internode length of grape shoots was noted.

The effect of IAA and exogenous applied synthetic auxins on various stages of grapevine development are well established (i.e. indolebutyric acid (IBA) on rooting (15), NAA on young fruitlets enabling thinning (12), 4-chlorophenoxyacetic acid (4-CPA) on seedless fruit growth (16), and generally a high sensitivity of grapevines to di- and tri-chlorophenoxy auxins as 2,4-D and 2,4,5-T). It should be noted, however, that non-phenoxy auxins such as IAA and NAA do not show any effect on vegetative shoot growth of grapevines even at rather high concentrations. On the other hand, gibberellic acid (GA_3) was found to be very active in inducing grape shoot growth (17, 18). This growth was usually thin and in our work (unpublished) showed a temporary chlorosis. Controlled growth is needed for studies on the effect of growth on inflorescence induction, increase of fruit set, and prevention of shot berry formation. A possible effect of MH and its interaction with auxin in grapevine was suggested. In this study an attempt was made to determine the growth curve of grape shoots, their response to MH, and the interactions of MH with IAA and GA_3 in the grape shoot system.

Materials and methods

A well-developed vineyard of the Madlene cultivar was chosen in the southern central coastal plain of Israel. The growth of 300 shoots (10/vine) equally positioned

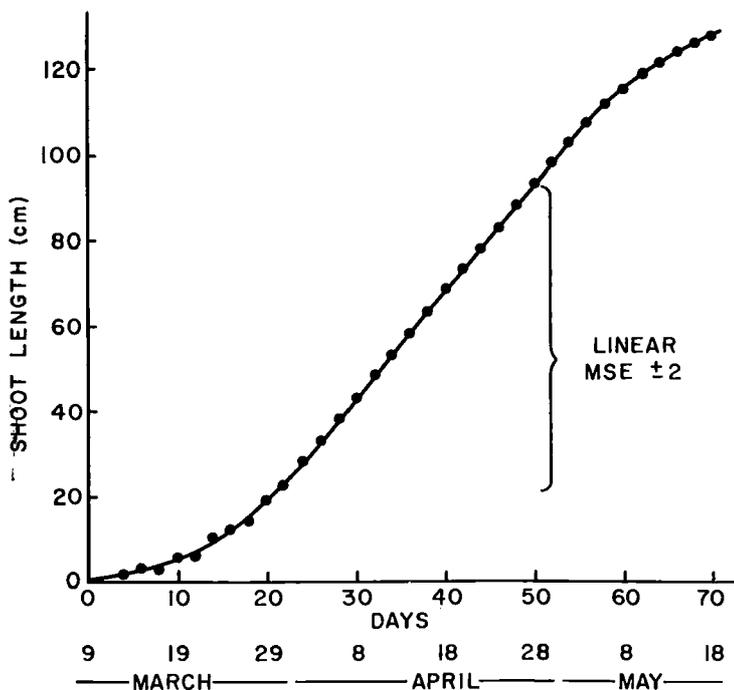


Fig. 1: The vegetative growth of Madlene shoots on irrigated vines during 70 d from bud burst.

Das Triebwachstum bewässerter Madlene-Reben im Verlauf von 70 d nach dem Austrieb.

on the plants was measured. Growth measurements were taken every second day from bud opening for a period of 70 d (Fig. 1). The growth curve achieved showed three growth periods of the canes; a slow growth for the first 20 d, a linear growth for the following 35 d, and a slowing growth thereafter. The linear growth started when the shoots were about 20 cm long and continued until they reached a length of 110 cm. The growth rate during this period was 2.6 cm/d. For further studies, shoots in the linear growth phase, starting when the shoots reached a length of 25 cm, were used (during April).

For regulator application 10 uniform shoots per vine (30 cm long) originating from similarly-positioned buds were chosen. 80 such shoots (on 8 vines) were used per treatment. The growth regulators were applied with a pressurized napsack sprayer and growth measurements were taken for 20–24 d every 2–4 d according to experiment. Both MH and IAA were dissolved in NaOH neutralized with HCl and applied as a sodium salt. In later experiments we used a commercial preparation of a MH-Na salt (MH-40, by Nangatuck Chemical Division, U. S. Rubber Company). Gibberellic acid used was "Giberel", a commercial preparation by ICI England. Similar experiments were conducted in two separate years. The standard error (SE) for each treatment or the mean standard error (MSE) per experiment, when uniform, were used for the analysis of results.

Results

Grape shoots 30 cm long were spray-treated with three concentrations of MH (500, 1000, 2000 mg/l) and the effect on the linear shoot growth determined. All three

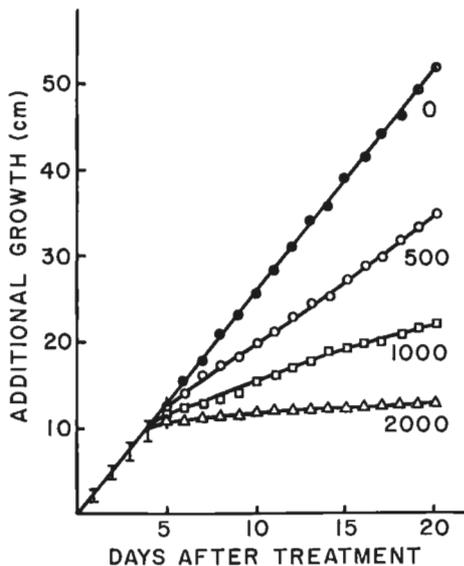


Fig. 2: The effect of MH on the vegetative growth of Madlene vine shoots. Application when shoots were 30 cm long, growth measured every 2–4 d.

Der Einfluß von MH auf das Triebwachstum der Sorte Madlene. Die Applikation erfolgte bei einer Trieblänge von 30 cm, das Wachstum wurde in Abständen von 2–4 d gemessen.

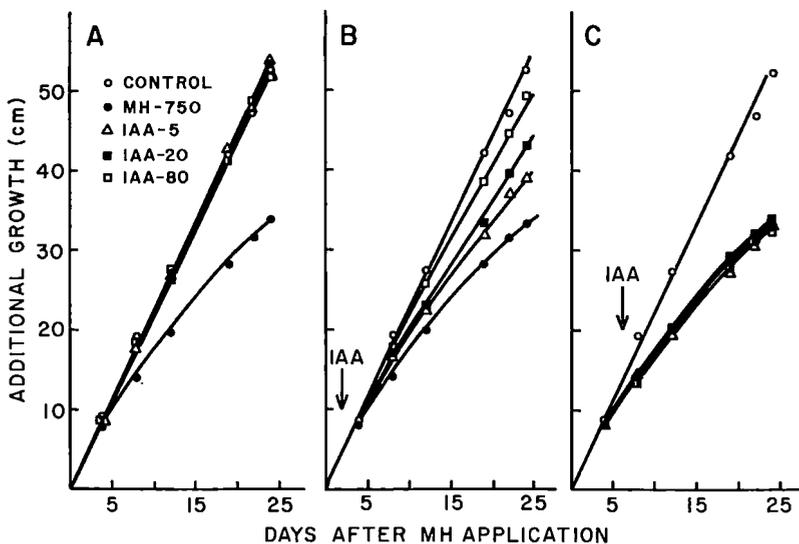


Fig. 3: The effect of MH and IAA on the vegetative growth of growing Madlene vine shoots. — A: IAA or MH applied on control shoots. — B: IAA applied on MH treated shoots 2 d after MH application. — C: IAA applied on 6-d MH pretreated shoots. — MH concentration 750 mg/l; IAA, 5, 20 and 80 mg/l.

Der Einfluß von MH und IES auf das Wachstum junger Triebe der Sorte Madlene. — A: IES- oder MH-Applikation bei Kontrolltrieben. B: IES-Applikation bei MH-behandelten Trieben 2 d nach der MH-Anwendung. — C: IES-Applikation 6 d nach der MH-Vorbehandlung. — Konzentration von MH: 750 mg/l; von IES: 5, 20 und 80 mg/l.

MH concentrations caused a reduction in the growth rate of the shoots. The degree of inhibition was concentration dependent. At the lower concentration (500 mg/l) the inhibition was partial, while at 2000 mg/l inhibition was nearly complete (Fig. 2). The growth inhibition started to be notable only 5 d after application of the MH regardless of the concentration of the inhibitor. Furthermore, following the initial 5 d of uniform growth three new growth rates developed which were linear as was that of the control, but slower. These new growth rates were constant for 20–25 d and then declined as did the control. No reversion to the original growth rate ever occurred.

When spray treatments of three concentrations of IAA (5, 20, 80 mg/l) were given to 30 cm long grape shoots no effect on growth was noted while 750 mg MH/l given at the same time reduced the growth rate by 34 % (Fig. 3 A). When the IAA concentrations were applied on MH pretreated shoots, 2 d after application of the inhibitor, the IAA caused a concentration-dependent reversion of the inhibition (Fig. 3 B). The shoot growth rates resulting from these auxin treatments were also linear and were apparent 5 d following MH application as for the MH-treated control vines. The high IAA concentration (80 mg/l) reversed most of the inhibiting effect of the MH. Higher concentrations (120 mg/l) tested on a limited scale did not change the growth rate further.

Later applications of IAA, 6 d after the MH (750 mg/l) pretreatment, had no effect on the growth rate of the grape shoots. The MH inhibition of the growth rate was unaltered (Fig. 3 C). The effect of MH, IAA and consecutive MH-IAA treat-

ments on the growing points, tendrils, and young leaves of growing Madlene grape shoots is shown in Fig. 4. The lack of a direct IAA effect on the growth was clear in contrast to the stunted, growth inhibited, tendrils and small leaves due to increasing concentrations of MH. The antagonistic effect of IAA applied 2 d after the MH is also demonstrated (Fig. 4 F) showing normal large leaves and only very slight apex inhibition.

A similar experiment using GA_3 at the same three concentrations (on mg/l basis) caused an enhanced growth of the grape shoots when applied to untreated 30 cm long shoots (Fig. 5 A). Later application of the GA_3 on MH-pretreated shoots resulted in similar enhanced growth whether treated 2 or 6 d after MH application although the later applications resulted generally in a somewhat weaker enhancement of growth (Fig. 4 B, C).

The high concentration of GA_3 (80 mg/l) induced a growth rate greater than that of the controls whether applied to untreated control shoots or to MH pretreated ones. This effect of GA_3 is demonstrated in Fig. 6 where the growth response to IAA, GA_3 and MH in the second experimental year is expressed as promotion or inhibition diversion from the control growth. The results from all three application times for MH, IAA and GA_3 were identical with those of the previous season. Only the two higher IAA and GA_3 concentrations were used in the second repeated set of experiments (Fig. 5 A, B, C).

Discussion

The response of grape shoots to MH applications was of growth rate inhibition. MH induced a new, constant growth rate that was proportional to MH concentration. Thus, no temporary inhibition, as shown for tomato (13), occurred in grape shoots. A lag period of 5 d for the beginning of inhibition was found for all MH treatments regardless of concentration. This suggests that a transition period is needed for the development of the new metabolism leading to the slower, but still linear, growth.

As it has been suggested (1, 11) that the MH mode of action is via increased IAA oxidase activity, a possible response of the treated tissue to IAA was speculated, in spite of the lack of response of normal grape shoot growth to IAA. The data presented here (Fig. 3 B) support this contention assuming that during the transition period the added IAA is needed to overcome the MH-induced IAA oxidation activity. Our experiments with grape shoots show that during the transition period from one vigor to another induced by MH, IAA treatments could antagonize this effect and prevent the onset of the reduced growing rate. It seems clear, however, that an established metabolism, resulting in linear growth, whether rapid or inhibited, could not be affected by IAA (Fig. 3 C). Thus, although antagonism between MH and IAA can occur in some systems (8) or conditions, it is not a general phenomenon. A direct competitive antagonism would also be difficult to explain on molecule structural basis. MH has been reported to antagonize GA_3 -induced growth of dwarf peas (4) and could reverse the GA_3 effect when applied to GA_3 -treated dwarf pea plants. In grapes, GA_3 showed a marked promoting effect on shoot growth (17, 18). From this study it seems, however, that there is no direct relationship between the GA_3 promoting activity and the MH inhibiting one. The GA_3 -induced growth on MH-pretreated grape shoots was related to the vigor at time of application and less to the future growth potential of the shoot. Thus, the GA_3 response is not related to the

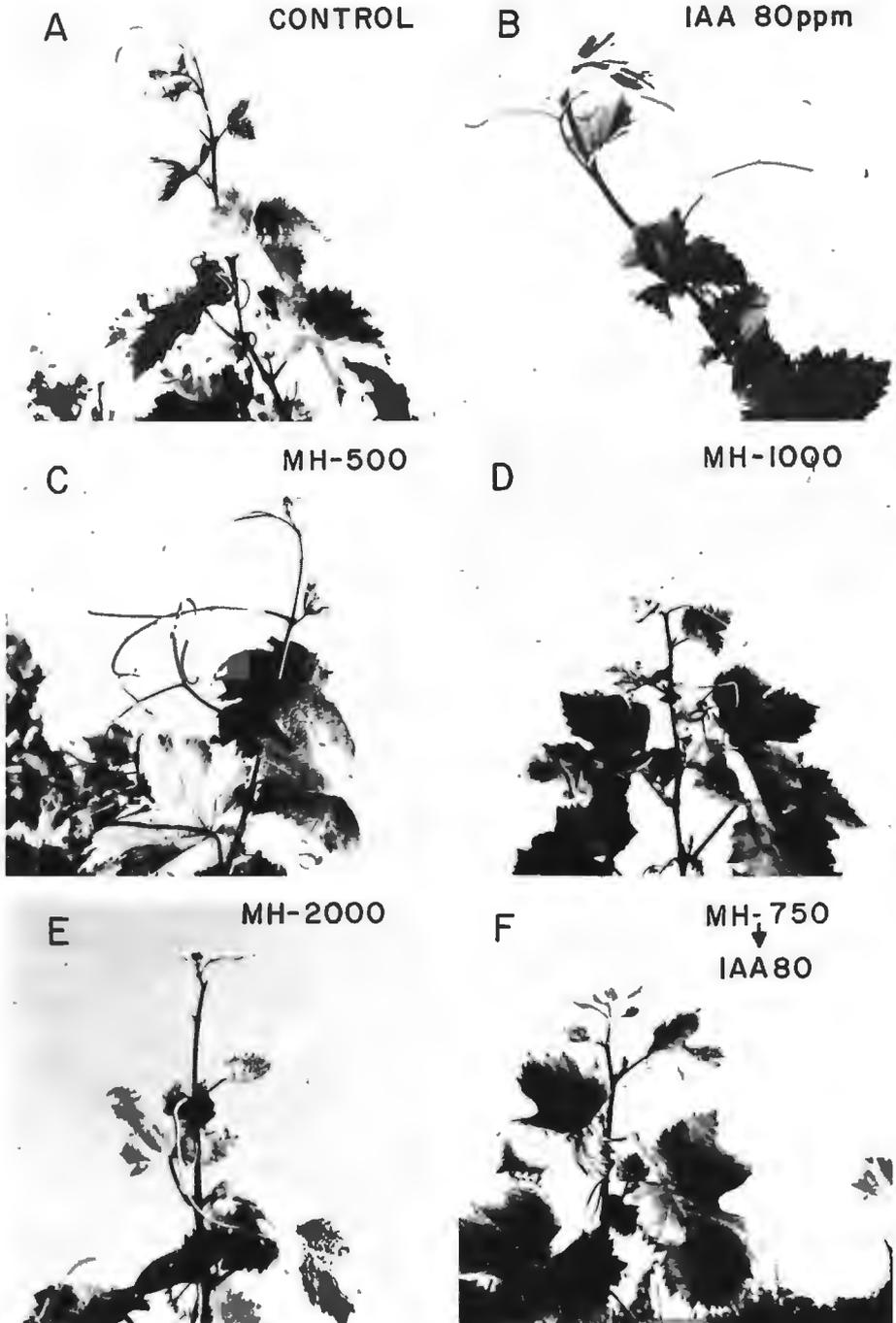


Fig. 4: The effect of MH and MH + IAA on the growing part of Madlene vine shoots. — A: Control. — B-E: MH concentrations in mg/l. — F: IAA application 2 d after MH pretreatment.

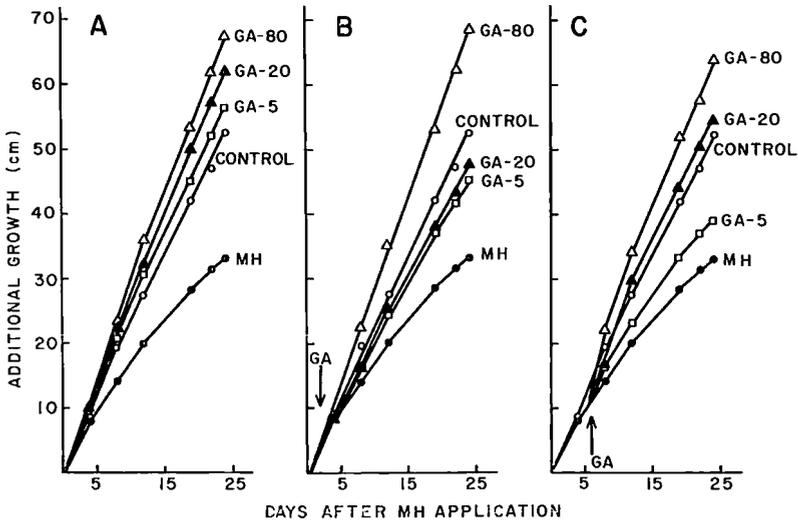


Fig. 5: The effect of GA_3 on control and MH-treated growing Madlene vine shoots. — A: GA_3 applied on control shoots. B: GA_3 applied on 2-d MH-pretreated shoots. — C: GA_3 applied on 6-d MH-pretreated shoots. — MH concentration 750 mg/l; GA_3 concentrations 5, 20 and 80 mg/l.

Der Einfluß von GS_3 auf junge Kontroll- und MH-bekandelte Triebe der Sorte Madlene — A: GS_3 -Applikation bei Kontrolltrieben. — B: GS_3 -Applikation 2 d nach der MH-Vorbehandlung. — C: GS_3 -Applikation 6 d nach der MH-Vorbehandlung. — Konzentration von MH: 750 mg/l; von GS_3 : 5, 20 und 80 mg/l.

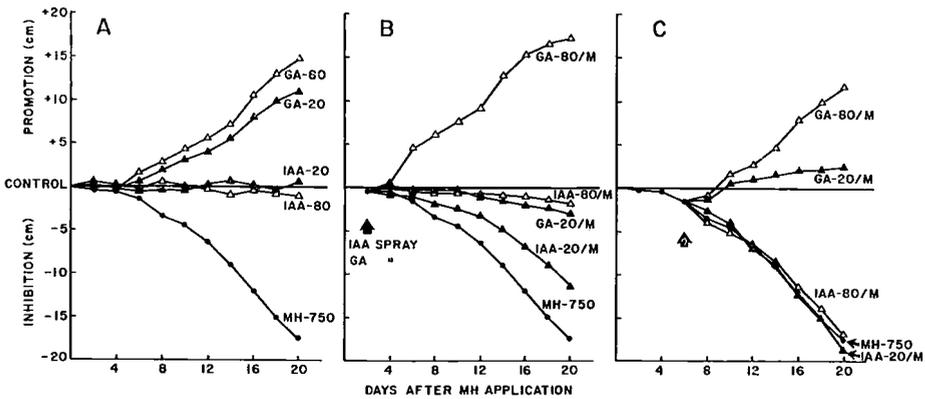


Fig. 6: The relative promotion and inhibiting effects of IAA, GA_3 and MH on growing Madlene vine shoots. — A, B, C: Application of IAA and GA_3 , 20 and 80 mg/l, at three different times as in Figs. 3 and 5 (.../M = MH-pretreated shoots.)

Relative Wachstumsförderung oder -hemmung junger Triebe der Sorte Madlene durch IES, GS_3 und MH. — A, B, C: Anwendung von IES und GS_3 , jeweils 20 und 80 mg/l, zu drei verschiedenen Terminen wie in Abb. 3 und 5. (.../M = MH-Vorbehandlung.)

Der Einfluß von MH und MH + IES auf die jungen Triebteile der Sorte Madlene. — A: Kontrolle. — B-E: MH-Konzentrationen in mg/l. — F: IES-Applikation 2 d nach der MH-Vorbehandlung.

transition stage induced by MH but with the state of growth of the grape shoot at any given time.

Summary

The growth rate of irrigated grape shoots on mature vines was determined. A linear growing period was found between 20 and 55 d after bud opening. Maleic hydrazide (MH) was found to reduce this growth rate without affecting its linearity. The degree of inhibition is concentration dependent. A lag period of 5 d was found from application to a measurable change of the growth rate. IAA had no effect on grape shoot growth rate when applied in the linear growth phase at concentrations up to 80 mg/l. This is true also when applied to shoots with a reduced growth rate induced by MH. IAA applied during the 5-d lag period after MH treatment could, however, reverse the MH inhibition. The degree of reversion is concentration dependent. Gibberellic acid (GA_3) has a pronounced promoting effect on grape shoot growth. No direct interaction between MH-induced growth reduction and GA_3 -induced growth stimulation was found.

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