Auctores summam litterarum suarum ipsi praestant!

INSTITUTUM AGRONOMICUM "DR.P.GROZA" CLUJ (ROMANIA)
NOTULAE BOTANICAE HORTI AGROBOTANICI CLUJENSIS
VI (1971/72)

SEED GERMINATION AND SEEDLING GROWTH OF CITRULLUS LANATUS (THUNB.) MANSF. IN RELATION TO CERTAIN GROWTH REGULATORS

Germinaţia seminţelor şi creşterea plantulelor de Citrullus lanatus (Thunb.) Mansf. în legătură cu anumiţi regulatori de creştere

by M.C.BHANDARI and D.N.SEN

INTRODUCTION. Growth retardants such as cycocel or (2-chloroethyl) trimethyl ammonium chloride has a remarkable effect on seed germination,growth and developmental behaviour of the plants. It behaves as a growth retarding substance in higher concentrations affecting the height of the plants without affecting the flowering and fruiting (Cathey and Stuart, 1961) or inhibit flower formation and stem elongation (Baldev and Lang, 1965). In lower concentrations it may stimulate the growth (Wittwer and Tolbert, 1960; Kelley and Postlethwait, 1962). Generally the response of growth retardants is extremely specific and the same effect must not necessarily be exerted upon plants even of the same family (Cathey,1964).

In several plants cycocel inhibition is overcome by gibberellins and to a greater extent by auxins (Baldev and Lang, 1965).

Gibberellin is an important plant growth hormone (Brian, 1959). The physiological action of applied gibberellins is important in influencing the processes of seed germination, plant growth and development. The response of gibberellin varies in different plants with different gibberellins(Brian and Hemming, 1961). Gibberellins are known to stimulate the dark germination of several light sensitive seeds (Toole and Cathey, 1959; Mohan Ram and Kamini, 1965). It appeared to be more effective on the growth of hypocotyl than the radicle in Sesamum indicum (Chatterji et.al., 1966).

Kinetin or 6-furfuryl aminopurine is a highly growth potent substance having the ability to bring about cytokinesis in cell and to promote cell enlargement (Miller, 1961). The exogenously applied kinetin plays an important role in nucleic acid metabolism, organ formation, apical dominance, flowering and seed germination (Letham, 1967; Helgeson, 1968). Kinetin is known to retard degradation of chlorophyl in isolated mature leaves (Richmond and Lang, 1957) and to promote expansion and synthesis of chlorophyll of isolated pumpkin cotyledons in light (Banerji and Laloraya, 1967 a).

Citrullus lanatus commonly known as "Matira" (watermelon) is cultivated throughout arid regions of India in the fields of pearlmillet in the rainy season. The fruits are supposed to quench the thirst of human beings in this arid zone more than the one cultivated in spring/summer. This taxon

Citrullus confined to Indian arid zone appears to be different from the one grown in spring which bears fruit in early summer all over India. In the present paper the effect of these three different growth regulators were examined on the seed germination and seedling growth behaviour of C.lanatus (syn.C.vulgaris).

MATERIALS AND METHODS. The seeds of C.lanatus were decoated because they show an extremely poor germination due to hard seed coatedness. Each set of ten seeds were kept in sterilized petridishes lined with filter paper and moistened with different concentrations of cycocel, sodium salt of gibberellic acid and kinetin or in distilled water as control. The dishes were kept at $28^{o} \pm 2^{o}$C in total darkness as well as in diffuse light (diffuse light of the day and darkness of the night). The linear growth of the radicle and hypocotyl was measured at the end of the fourth day. The data obtained were statistically analysed.

OBSERVATIONS. In case of cycocel the percentage of germination was percent in total darkness whereas in diffuse light there was very poor germination at higher concentrations at the end of the fourth day. Only the imbibition of cotyledons and slight germination took place in 5o ppm solution but 4o % and 7o % germination occured in 25 ppm and lo ppm solutions, respectively. In total darkness germination was quicker in higher concentration of cycocel although the percentage remained nearly equal in almost all the concentrations at the end of the experimental period (Table 1). The linear growth of the radicle and hypocotyl was promoted only in 1o ppm solution in total darkness (Table 2).

A cent percent germination occured in almost all the concentrations of gibberellic acid both in total darkness as well as in diffuse light. Gibberellic acid accelerated the pace of seed germination in total darkness as well as in diffuse light when compared with control (Tabel 1). The linear growth of the radicle as well the hypocotyl was more in all the concentrations of gibberellic acid on diffuse light, whereas the radicle showed an enhanced growth in total darkness in 2 ppm solution when compared with control (Tabel 3).

In kinetin cent percent germination was obtained in total darkness which was not so in diffuse light at the end of the experimental period. The total percentage of germination was cent percent in 5 ppm solution of kinetin in diffuse light also. Kinetin enhanced the pace of germination in total darkness in the first 24 hours when compared with control (Table 1). It appeared that the requirement of total darkness for germination were not met by kinetin in diffuse light in the initial stages. The seedling growth was promoted in diffuse light in almost all the concentrations of kinetin while it was inhibited in total darkness (Table 4).

The growth in terms of expansion of cotyledons was enhanced in almost all the chemicals used when compared with controls. In diffuse light the high concentration of kinetin was more effective on

expansion of cotyledons as compared to cycocel and gibberellic acid. The increase in the size of cotyledons only in diffuse light may be due to efficient photosynthetic activity. This may result in increased amount of protein and carbohydrates content necessary for regulating the normal growth of the plant or plant parts.

RESULTS AND DISCUSSION. The effect of the three growth regulators were observed on the seed germination and seedling growth of C.lanatus. Cycocel accelerated germination inception in total darkness and inhibited it in diffuse light.

The growth of the seedlings was found to be more in 1o ppm solution of cycocel while it was less in 25 ppm and 5o ppm in total darkness. Hypocotyl growth was mainly suppressed in diffuse light in almost all the concentrations as compared to control (Table 2). The seedlings did not indicate any growth in diffuse light except for some in 1o ppm solution. In majority of the cases cycocel was found to inhibit the growth of the plants (Baldev and Lang, 1965), but in lower concentrations it might promote the growth (Wittwer and Tolbert, 196o). The latter view was found to be supported in the present study where growth was more in lower as compared to higher concentrations (Fig.1).

The presence of gibberellic acid in total darkness accelerated the pace of germination more than in diffuse light in the first 24 hours in all the concentrations when compared with control. Hayashi (194o) showed that barley and rice germinated more rapidly as gibberellin concentration was increased but final germination percentage was nearly the same in all the concentrations.

As regards its effect on seedling growth, an interesting aspect of gibberellic acid on hypocotyl was that it slightly decreased the growth in total darkness and indicated a slight increase in diffuse light (Fig,2.). The known influence of gibberellic acid on the shoot elongation largely remained uneffective in this case. The record of such an inhibitory effect on the shoot elongation by gibberellic acid is noteworthy. Lockhart (1956) reported that the application of gibberellic acid reversed the low intensity light inhibition of pea stem growth. Chatterji et.al., (1966) reported a distinct increase in hypocotyl of *Sesamum indicum* with a slight increase of root in lower concentrations. Halvey and Cathey (1960) indicated that the radicle elongation is a measureable aspect of growth in cucumber seedlings. This has been also observed in the present study. The response of cucumber seedlings to exogenous auxin indicated no growth promotion and the exogenously applied gibberellins could not be effective in the absence of endogenous auxin (Lockhart, 1958). It would be interesting to know whether auxin action in turn is also dependent upon the presence of endogenous or exogenuos gibberellins (Galston and Purvey, 1960). Katsumi et al., (1965) observed that both gibberellin and auxin are required for the normal growth of the cucumber hypocotyl.

The seedlings indicated an enhanced growth in almost all the concentrations of kinetin

in diffuse light but in total darkness it had an inhibitory effect as compared to the control (Fig.3.). The size of the cotyledons increased in all the concentrations which might be due to greater amount of chlorophyll, protein and other unknown contents.This is contradictory to what had been reported by Narain and Laloraya (1970) where the kinetin induced expansion of cotyledons of *Cucumis* increased the inhibitory effect on chlorophyll synthesis and which became more marked with time. Banerji and Laloraya (1967 b) found the retention of protein level and expansion of cotyledons by kinetin in *Cucurbita pepo.*

Gibberellic acid caused marked expansion of cotyledons in *Ipomoea pentaphylla* but inhibited somewhat in growth retardant like cycocel (Sankhla,1969) This was not found so in the present study but the observations agree as regards kinetin induced expansion of cotyledons in *Cucumis* (Narain and Laloraya, 1970).

SUMMARY. The effect of cycocel, gibberellic acid and kinetin was investigated on the seed germination and seedling growth of *C.lanatus*. Cycocel favoured germination in total darkness and largely inhibited it in diffuse light. The growth of radicle and hypocotyl was promoted only in total darkness.

Gibberellic acid accelerated the pace of germination in total darkness more than diffuse light in the first 24 hours. The growth of the radicle as well as the hypocotyl was more in diffuse light, whereas only the radicle indicated an enhanced growth in total darkness in low concentration. The known in-

fluence of this chemical on the shoot elongation largely remained uneffective.

In kinetin cent percent germination was obtained in total darkness which was not so in diffuse light, except for high concentration. It enhanced the pace of germination in total darkness in the first 24 hours. The seedling growth was promoted in diffuse light, while it was inhibited in total darkness. The increase in the size of the cotyledons took place only in diffuse light. It became apparent that kinetin and gibberellic acid could partly substitute the reguirements of total darkness for seed germination in this species.

ACKNOWLEDGEMENTS. Generous gift from Cynamid India Ltd., of Cycocel and facilities provided by the Head, Botany Department of this University are gratefully acknowledged. Thanks are also due to Drs. D.D. Chawan and K.D. Sharma for their help during the progress of this study. The assistance received by Mr. Paras Ram in making photographs is also thankfully acknowledged.

Table 1. Effect of cycocel, gibberellic acid and kinetin in different concentrations (in ppm) on the pace and percentage of seed germination in *C. lanatus* for four days+.

Days	Cycocel						Gibberellic acid						Kinetin						Control	
	TL			DL			TL			DL			TL			DL				
	10	25	50	10	25	50	2	5	10	2	5	10	1	2.5	5	1	2.5	5	TL	DL
1	20	40	40	0		0	50	60	70	30	30	30	60	80	60	0	0	0	0	0
2	60	70	70	30	10	10	80	90	80	80	90	80	80	90	90	60	80	60	50	40
3	80	80	90	60	30	10	90	90	90	80	90	90	90	90	100	60	90	90	60	60
4	100	100	100	70	40	20	100	100	100	100	100	100	100	100	-	90	90	100	90	60

+ Average of three observations.

TL = Total darkness; DL = Diffuse Light

Table 2. Effect of different concentrations of cycocel on the linear growth (in cm)of radicle and hypocotyl in four days old seedlings of C.lanatus

Conc.in ppm.	Total darkness				Diffuse light			
			Cotyledons				Cotyledons	
	Radicle	Hypocotyl	Length	Breadth	Radicle	Hypocotyl	Length	Breadth
Control	4.0 ± 1.0	3.5 ± 1.9	1.0 ± 0.0	0.5 ± 0.0	1.3 ± 0.1	0.5 ± 0.0	0.9 ± 0.0	0.5 ± 0.0
10	9.7 ± 2.0	4.8 ± 0.7	1.1 ± 0.0	0.6 ± 0.0	1.4 ± 0.1	0.3 ± 0.0	1.0 ± 0.0	0.6 ± 0.0
25	4.0 + 0.8	2.2 + 0.9	0.9 + 0,1	0.6 + 0.1	0.4 + 0.0	-	-	-
50	2.8 ± 1.3	0.9 ± 0.5	1.0 ± 0.1	0.6 ± 0.0	-	-	-	-

Table 3. Effect of different concentrations of gibberellic acid on the linear growth (in cm) of radicle and hypocotyl in four days old seedlings of C.lanatus

Conc.in ppm.	Total darkness				Diffuse light			
			Cotyledons				Cotyledons	
	Radicle	Hypocotyl	Length	Breadth	Radicle	Hypocotyl	Length	Breadth
Control	4.0 ± 1.0	3.5 ± 1.9	1.0 ± 0.0	0.5 ± 0.0	1.3 ± 0.1	0.5 ± 0.0	0.9 ± 0.0	0.5 ± 0.0
2	4.7 ± 0.7	3.2 ± 0.8	1.0 ± 0.1	0.6 ± 0.1	2.1 ± 0.5	0.6 ± 0.3	1.0 ± 0.1	0.6 ± 0.1
	3.7 ± 0.8	3.3 ± 1.0	1.0 ± 0.1	0.5 ± 0.1	2.0 ± 0.5	0.6 ± 0.0	1.0 ± 0.0	0.6 ± 0.0
10	3.4 ± 0.9	3.0 ± 0.9	1.0 ± 0.0	0.5 ± 0.0	1.6 ± 0.9	0.7 ± 0.0	1.0 ± 0.0	0.6 ± 0.0

Table 4. Effect of different concentrations of kinetin on the linear growth (in cm) of radicle and hypocotyl in four days old seedlings of C.lanatus

Conc. in ppm.	Total darkness				Diffuse light			
			Cotyledons				Cotyledons	
	Radicle	Hypocotyl	Length	Breadth	Radicle	Hypocotyl	Length	Breadth
Control	4.0 ± 1.0	3.5 ± 1.9	1.0 ± 0.0	0.5 ± 0.0	1.3 ± 0.1	0.5 ± 0.0	0.9 ± 0.0	0.5 ± 0.0
1	3.9 ± 1.1	2.6 ± 1.0	1.0 ± 0.1	0.6 ± 0.1	2.3 ± 0.2	0.6 ± 0.1	1.1 ± 0.1	0.6 ± 0.1
2.5	2.7 ± 1.1	2.5 ± 0.1	1.0 ± 0.1	0.6 ± 0.1	1.7 ± 0.4	0.5 ± 0.1	1.1 ± 0.1	0.7 ± 0.1
5	2.1 ± 0.6	2.4 ± 0.5	1.0 ± 0.1	0.6 ± 0.1	1.4 ± 0.2	0.5 ± 0.1	1.1 ± 0.1	0.8 ± 0.1

REFERENCES

BALDEV, B., and A. LANG (1965): Control of flower formation by growth retardants and gibberellin in Samolus parviflorus, a long day plant. Amer. J. Bot. 52, 408.

BANERJI, D., and M.M. LALORAYA (1967): Chlorophyll promotion in isolated pumpkin cotyledons in the presence of kinetin and chloramphenicol. Plant and Cell Physiol, 8, 263-268.

BANERJI, D., and M.M. LALORAYA (1967 b): Biochemical changes accompanying kinetin induced expansion of isolated Cucurbita pepo cotyledons. Intern. Sympos.on plant growth and development, Calcutta, India, p. 139-146.

BRIAN, P.W. (1959): Effect of gibberellins on plant growth and development. Biol.Rev. 34, 37-84.

BRIAN, P.W., and H.H. HEMMING (1961): Promotion of cucumber hypocotyl growth by two new gibberellins. Nature 189, 74.

CATHEY, H.M. (1964): Physiology of growth retarding chemicals. Ann. Rev. P. Physiol. 15, 271-302.

CATHEY, H.M. and N.W. STUART, (1961): Comparative growth retarding activity of AMO-1618, Phosfon and CCC. Bot. Gaz. 123: 51-57.

CHATTERJI, U.N., N. SANKHLA and D. BAXI (1967): Preliminary studies on the effect of certain growth substances on germination of Sesamum indicum Linn. Indian Agri. 7:46-51.

GALSTON, A.W. and W.K. PURVEY (1960): The mechanism of action of auxin. Ann.Rev.Pl.Physiol. 11: 239-276.

+HAYASHI, T. (1940): J.Agr.Che.Soci.Japan 16: 531-538.

HALVEY, A.H. and H.M. CATHEY (1960): Effect of structure and concentration of gibberellins on the growth of cucumber seedlings. Bot.Gaz. 122: 63-67.

HELGESON, J.P. (1968): The cytokinins. Science 161: 974-981.

KATSUMI, M., B.O. PHINNEY and W.K. PURVEY (1965): The role of gibberellin and auxin in cucumber hypocotyl growth. Physiol. Plant. 18: 462-473.

KELLEY, A.G. and S.N. POSTLETHWAIT (1962): Effect of (2-chloroethyl) trimethyl ammonium chloride on fern gametophyte. Amer.J.Bot. 49: 778-786.

KUSE, G. (1958): Necessity of auxin for the growth effect of gibberellin. Bot. Mag. Tokyo 71: 151-159.

LETHAM, D.S. (1967): Chemistry and physiology of kinetin like compounds. Ann. Rev. Pl. Physiol. 18: 349-364.

LOCKHART, J.A. (1956): Reversal of light inhibition of pea stem growth by the gibberellins. Proc.Nat.Acad.Sci.Us. 42: 841-848.

LOCKHART, J.A. (1958): The response of various species of higher plants to light and gibberellic acid. Physiol.Plant.11: 478-486.

MILLER, C.O. (1961): Kinetin and its related compound in plant growth. Ann. Rev.Pl.Physiol. 12: 395-408.

MOHAN RAM, H.Y. and ISHA KAMINI (1965): Studies on the germination of Withania somnifera seeds. Phytomorphology 15: 31-36.

NARAIN, A., LALORAYA, M.M. (1970): Inhibition of chlorophyll synthesis by kinetin in Cucumis cotyledons.-Plant and Cell Physiol. 11: 173-175.

RICHMOND, A.E., Lang,A. (1970): Effect of kinetin on protein content and survival of detached Xanthium leaves.-Science 125: 650.

SANKHLA, N. (1969): Comparative effect of gibberellin, growth retardants and morphactins on expansion of cotyledons of Ipomoea pentaphylla.-Curr. Sci. 38: 522-523, 1969.

TOOLE, V.K., Cathey, H.M.(1959): Germination of grand rapid lettuce and Lepidium virginiacum seeds as affected by gibberellin. - Pl. Physiol. 34:16.

WITTWER, S.H., Tolbert, N.E. (1968): (2-chloroethyl) trimethyl ammonium chloride and related

compounds as plant growth substances.
V. Growth, flowering and fruiting responses as related to those induced by auxin and gibberellins.-Pl. Physiol. 35: 871-877.

+ not seen in original.

Botany Departament, University of Jodhpur,
Jodhpur, (Raj.)India

Manuscript received at 1 March 1972

DESCRIPTION FOR FIGURES

Fig. 1. Effect of different concentrations of cycocel (R) on the linear growth of radicle and hypocotyl in four days old seedlings of C.lanatus.

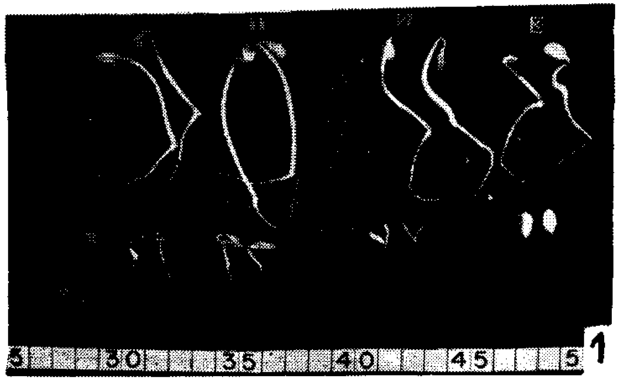
O = Control; 1=lo ppm; 2=25 ppm; 3=5o ppm;
D = Total darkness; L = Diffuse light.

Fig. 2. Effect of different concentrations of gibberellic acid (S) on the linear growth of radicle and hypocotyl in four days old seedling of C.lanatus.

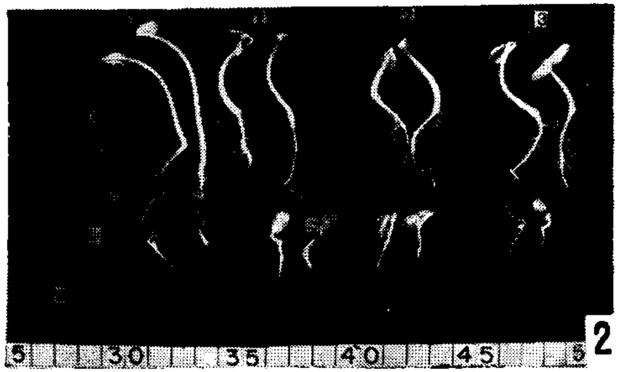
O = Control; 1 = 2 ppm; 2 = 5 ppm; 3 = lo ppm;
D = Total darkness; L = Diffuse light.

Fig. 3. Effect of different concentrations of kinetin (T) on the linear growth of radicle and hypocotyl in four days old seedlings of C.lanatus.

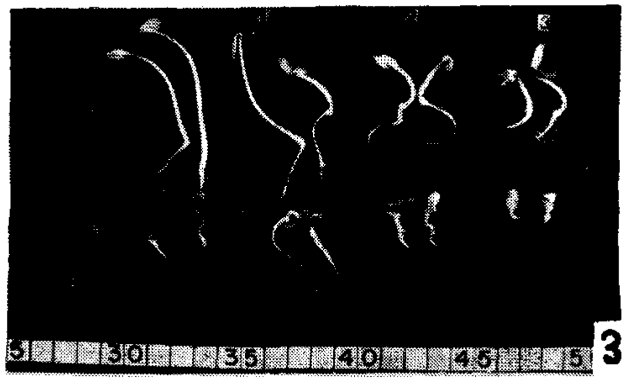
O = Control; 1 = 1 ppm; 2 = 2.5 ppm; 3 = 5 ppm;
D = Total darkness; L = Diffuse light.



1



2



3