

## EFFECT OF SOME PHENOLS AND GA<sub>3</sub> ON, GERMINATION, EARLY SEEDLING GROWTH AND $\alpha$ -AMYLASE AND PROTEASE ACTIVITY OF *CICER ARIETINUM* L.<sup>1</sup>

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### ABSTRACT

The effect of  $\beta$ -naphthol, resorcinol phloroglucinol and GA<sub>3</sub> was studied on seed germination and seedling growth of *Cicer arietinum* L. Germination percentage remained unaffected under all the treatments. Phenols and GA<sub>3</sub> promoted the plumule and radicle growth at 10 and 100 mg/l concentration while 500 mg/l retarded it. GA<sub>3</sub> antagonised the inhibitory effect of higher concentration of phenols. Lateral root development was retarded by GA<sub>3</sub> and was enhanced by low concentration of phenols.  $\alpha$ -Amylase and protease activity were promoted at low and retarded at high concentration of both GA<sub>3</sub> and phenols.

### INTRODUCTION

Kumar and Tayal (1982) found enhancement of radicle and plumule growth by low concentrations and inhibition by higher concentrations of various phenols in lentil and mungbean. GA<sub>3</sub> was found to antagonize the inhibition caused by phenols in *Gajanus cajan* (Tayal and Sharma, 1981).  $\alpha$ -Amylase activity was also affected by GA<sub>3</sub> *in vivo* (Chrispeels and Varner, 1967). In order to confirm the above findings and to have a better understanding of seed germination and seedling growth in legumes, the present work was undertaken to study the effect of phenols, alone and in combination with GA<sub>3</sub> on seed germination seedling growth,  $\alpha$ -amylase and protease activities, in *Cicer arietinum*.

### MATERIAL AND METHODS

Seeds of *Cicer arietinum* cv. C-235,

procured from local seed store were properly sterilized with calcium hypochlorite and sown in petridishes (12 cm dia), cushioned with double layers of Whatman filter paper. Ten seeds were placed equidistantly in each petridish and three replicates were taken for each treatment. Solutions of  $\beta$ -naphthol, resorcinol, phloroglucinol and GA<sub>3</sub> were prepared in 10, 100 and 500 mg/l concentrations. Phenolic solutions alone and in combination with GA<sub>3</sub> were used as shown in Table I. 1 ml of different solutions was added to respective petridishes in which 5 ml of distilled water had already been added, 6 ml of distilled water was added in one set of petridishes, to serve as control.

All extractions were done at 4°C temperature, 35 grams seeds (Table I) from each sample were separately crushed and homogenized in prechilled mortar and pestle in ice cold acetate buffer (pH 5.2). Homogenates were

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filtered through a double layer of cheese cloth and centrifuged at 4000 rpm for 10 min. The supernatant was precipitated with ammonium sulphate between zero and 80% saturation. Precipitates, collected by centrifugation at 16000 rpm for 15 min were dissolved in 10 ml of acetate buffer for each sample. These solutions were used to determine amylase and protease activity by the method described by Mahadavan (1975).

Three days after germination the length of plumules radicles and the number of lateral roots was noted.  $\alpha$ -Amylase and protease activity was studied *in vivo*.

### OBSERVATIONS

None of the concentrations of used chemicals affect the germination percentage. Phenols and  $GA_3$  both in 10 and 100 mg/l concentrations enha-

TABLE I

PERCENTAGE PROMOTION/INHIBITION OF PLUMULE, RADICLE AND NUMBER OF LATERAL ROOT OF *Cicer Arietinum* BY THE APPLICATION OF PHENOLS AND  $GA_3$  ALONE AND INCOMBINATION.

Treatment	% Germination	% Plumule growth promotion/inhibition	% Radicle Promotion/inhibition	% Lateral roots promotion/inhibition
Control	100	—	—	—
$GA_3$				
10	+100	+140.0	+20.0	—26.6
100	+200	+220.0	+12.6	—42.9
500	—97	—20.0	+21.2	—56.9
$\beta$ -naphthol				
10	100	+26.6	+3.6	—2.4
100	97	+61.2	+1.6	—5.4
500	97	—10.6	—12.6	—6.0
Resorcinol				
10	100	+80.6	+18.6	+6.4
100	99	+100.0	+52.8	+8.4
500	98	—20.6	—8.7	—1.6
Phloroglucinol				
10	97	+142.0	+18.6	+3.6
100	98	+162.0	+52.8	+30.8
500	97	—20.6	—8.7	—26.3
$\beta$ -naphthol + $GA_3$				
100 + 100	98	+140.6	+10.5	—40.3
500 + 100	97	+80.6	—10.6	—50.0
Resorcinol + $GA_3$				
100 + 100	100	+160.2	+20.6	—46.4
500 + 100	97	+90.6	+1.26	—52.0
Phloroglucinol + $GA_3$				
100 + 100	100	+230.0	+20.9	—40.0
500 + 100	100	+130.0	+1.23	—47.0



nce the plumule extension, the latter being more promotory. 500 mg/l of all used chemicals have retardation effect on plumule growth. In combinations the retardation caused by 500 mg/l of phenol is antagonized by 100 mg/l of  $GA_3$ . 10 and 100 mg/l of all used chemicals also enhance radicle growth while 500 mg/l retard it. The inhibitory effect of 500 mg/l phenols is found to be antagonized by 100 mg/l of  $GA_3$ . The lateral root development is retarded more with the increase in concentration of  $GA_3$ . 10 and 100 mg/l of phenols except  $\beta$ -naphthol enhance lateral root development while 500 mg/l retard it. All combinations show remarkable retardation in this process (Table I).

Figure 1 shows  $\alpha$ -amylase activity *in vivo* as affected by  $GA_3$  and phenols.

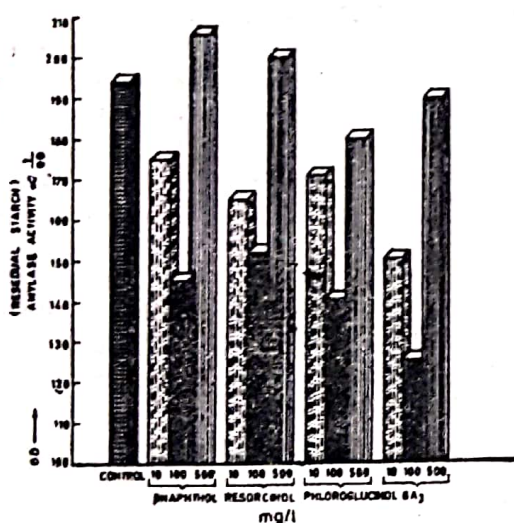


Fig. 1. EFFECT OF PHENOLS AND  $GA_3$  ON  $\alpha$  AMYLASE ACTIVITY

$GA_3$  enhances its activity both at 10 and 100 mg/l but retard it at 500 mg/l. Phenols except phloroglucinol retard the activity at 500 mg/l. 10 and 100 mg/l of all being promotory.

All used phenols increased the protease activity at 10 and 100 mg/l.

Phloroglucinol in particular do not retard protease activity even at 500 mg/l. This is against  $\beta$ -naphthol and resorcinol where 500 mg/l showed reduction in protease activity (Fig. 2).

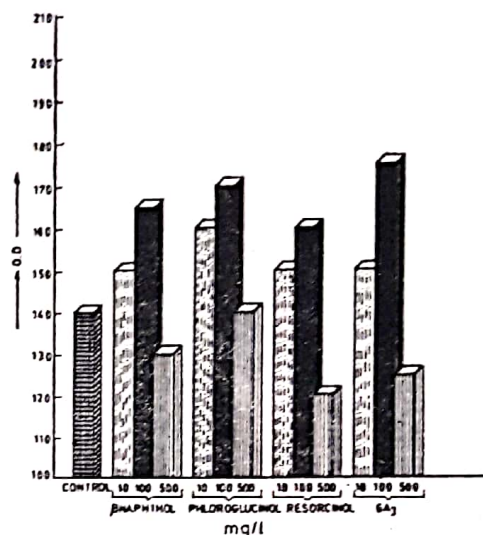


Fig. 2. EFFECT OF PHENOLS AND  $GA_3$  ON PROTEASE ACTIVITY

## DISCUSSION

Phenols in the present studies do not affect germination percentage, as is also shown by Tayal and Sharma (1981) and Kumar and Tayal (1982) in other leguminous seeds. These results, however, differ from the early findings of Phillips (1961) and Thakur (1977) where exogenous application of phenols, was shown to cause inhibition of germination in lettuce and sugar mapple.

Enhancement of growth by  $GA_3$  in this case is similar to the findings of Singh and Nanda (1981) in *Callistemon viminalis* L. Retardation of plumule and radicle growth by 500 mg/l is probably because its being supraoptimal. Frankland and Wareing (1960) had also shown promotion of hypocotyl growth by 10 and 100 mg/l concentrations. The retardation of lateral root development by higher concentration of phenols is similar

to one obtained by Bendixen and Peterson (1962).

The growth promotion by phenols at low concentration get support from Nanda *et al.* (1977) and Datta and Nanda (1978) who have shown them to increase the number of branches and tillers in *Setaria italica* and in triticale. Retardation of growth by higher concentrations of phenols get support from Miminoshvilli (1973) who found inhibition of carrot tissue growth by phenolic concentration higher than 100 mg/l. The promotion of lateral root development by phenols are according to the results of Dhawan and Nanda (1981).

Interaction studies reveal that promotive concentrations of phenolics behave synergistically with  $GA_3$  as is also shown by Nanda *et al.* (1977) and Kumar *et al.* (1978). The antagonization of inhibitory actions of higher phenolic concentrations by  $GA_3$  in the present studies is similar to one shown earlier by Phillips (1961), Tizio and Paupardin (1971) and Corcoran *et al.* (1972).

The growth promoting effects of  $GA_3$  and phenols may be assigned to facilitated mobilization of stored carbohydrates and proteins. This assumption gets support from our experiments on  $\alpha$ -amylase and protease (Fig 1 and 2) and also from Kumar and Nanda (1981) who have shown increase in the amount of RNA in  $GA_3$  treated meristematic cells of *Impatiens balsamina*, that can indirectly affect enzyme synthesis. Shastri and Muir (1963) have also shown higher amylase and protease levels in  $GA_3$  treated seeds. Reduction in seedling growth at 500 mg/l of  $GA_3$  and phenols may be because of it's being supra optimal. The inhibition of growth by phenols, might be because of its inhibition of over all metabolism

through non-specific metabolic block of ATP synthesis (Khan, 1968).

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