EFFECT OF VARIOUS AUXINS ON GROWTH RATE OF STATIC AND SUSPENSION CULTURES OF *PHYSOCHLAINA PRAEALTA* HOOK

TISSUE culture appears to be a promising technique for future production of phytochemicals[1,2]. At this present stage of development tissue culture is not commercially viable because of the slow growth rates of plant cells and low yields of the active constituents. In this paper the authors have studied the effect of auxins on growth rate of static and suspension cultures of *Physochlaina praealta*. Growth is expressed either in terms of fresh tissue per litre per day or as growth index (G.I.), which is a value obtained by dividing the final fresh weight of tissue by its initial weight over a given period of time. Comparative accounts of large scale yields of plant cell cultures are reported in Table I[3-5].

TABLE I

Yield of cell, mass in suspension cultures

Plant	Med.	Vessel	GMS per lit per day Wet	Dry
Ginkgo, Lolium Liek	cm	Carboy	3	..
Rose	cm	Pilot plant	10	..
Carrot	cm	Carboy	6·1	..
,,	,,	Fermentor	..	1·9
Spearmint	Defined	Carboy	4·7	..
,,	,,	Flask	6·3	0·4
Rose	Defined	Phytostat	11	0·4
Hyoscyamus	..	Fermentor	1·6	0·02
Bean	Defined	Flask	..	0·7
,,	Phylone	,,	..	1·1
Kidney Bean	Defined	Fermentor	..	1·2
Apocynum	Defined	Fermentor	5·33	..
Physochlaina	Defined	Flask	19·0	2·2

It appears that *Physochlaina praealta* H. tissue grown in revised Murashige and Skoog's medium[6] with naphthalene acetic acid 0·1 ppm in flask suspension cultures has given growth rates which are faster than any other tissue reported in literature.

Static Cultures

Callus cultures were obtained from hypocotyl regions of sterile seedlings in 100 ml Erlenmeyer flasks containing 40 ml of revised Murashige and Skoog's medium supplemented with coconut water 15% and naphthalene acetic acid 1 ppm. Callus was pale white in colour and friable in nature. The callus cultures thus established and maintained by subculturing at four weeks interval were used as experimental material.

Naphthalene acetic acid, indole acetic acid and 2, 4-dichlorophenoxy acetic acid at 0·25, 0·5 and 1 ppm concentrations were used to study their effects on growth. The cultures were grown in BOD incubator in dark at 25° C. To study growth rates, ten flasks of each treatment were harvested at weekly interval over a period of eight weeks.

Effects of NAA, IAA and 2, 4-D on callus growth are presented in Fig. 1 (*a*, *b* and *c*). Growth index was recorded for each treatment for 6 weeks. A maximum growth yield was obtained in the media with 0·5 ppm NAA (G.I. 14·6). Growth was slightly less at 1 ppm (G.I. 11·6) while it was minimum at 0·25 ppm (G.I. 1·3). IAA from 0·25 ppm to 1 ppm produced almost uniform growth for four weeks, however further growth was maintained at 0·5 (G.I. 10·3) and 1 ppm (G.I. 9·3), while it declined at 0·25 ppm (G.I. 6·1). 2, 4-D produced maximum growth at 0·5 ppm (G.I. 12·4), while at 1 ppm (G.I. 5·5) the growth was considerably less and was minimum at 0·25 ppm (G.I. 1·8).

Suspension Cultures

Four weeks old callus tissue was transferred to 500 ml Erlenmeyer flasks containing 100 ml of liquid medium supplemented with 0·1 ppm NAA. The cultures were incubated in dark at 25° C on a rotary shaker at 120 ppm. The suspension cultures were subcultured in fresh medium at 30 days interval. A study of growth rates was conducted by harvesting four flasks at 10 days interval.

A maximum cellular fresh weight of 57 g per 100 ml medium supplemented with 0·1 ppm NAA was obtained in 30 days. The growth of fresh tissue recorded per litre per day is 19 g which is supposed to be the highest reported value (Fig. 1 *d*).

It is observed that growth of callus in suspension cultures is faster (19 g fresh tissue/l/day, 2·2 g dry tissue/l/day), than that of static cultures (15·0 g fresh tissue/l/day, 0·68 g dry tissue/l/day).

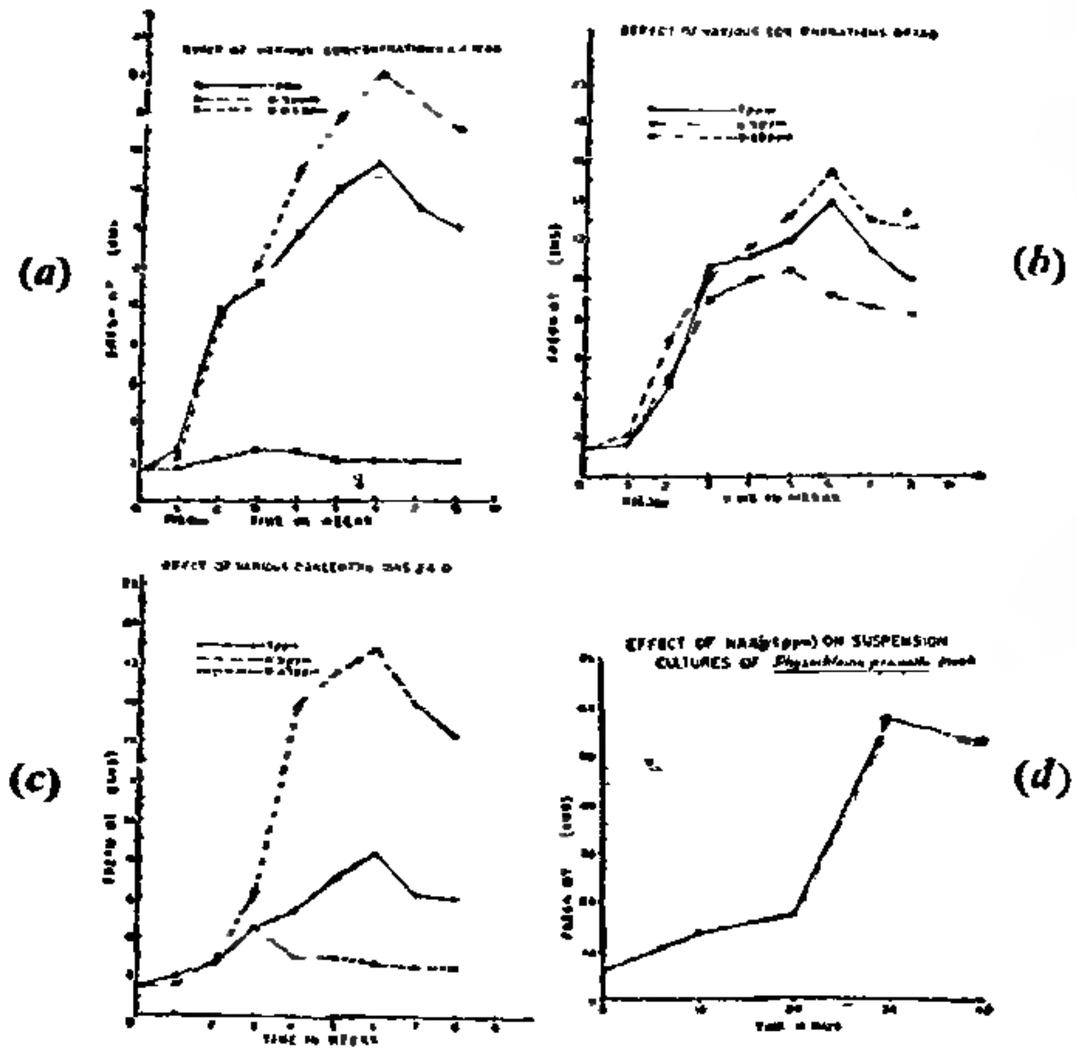

FIG. 1 (*a–d*). Fig. 1 *a–c*. Effect of various concentrations of NAA, IAA and 2, 4-D on static cultures of *Physochlaina praealta*. Fig. 1 *d*. Effect of NAA (0·1 ppm) on suspension cultures of *Physochlaina praealta*.

A large quantity (2·8 kg) of fresh tissue was harvested from cultures growing on Murashige and Skoog's modified medium with NAA concentrations of 1 ppm. The tissue when analysed gave a total alkaloid value of 0·02% on dry weight basis. T.L.C. revealed that the major alkaloid formed was hyoscyamine and two minor unidentified alkaloids were also present. Hyoscine was absent.

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S. GREWAL.
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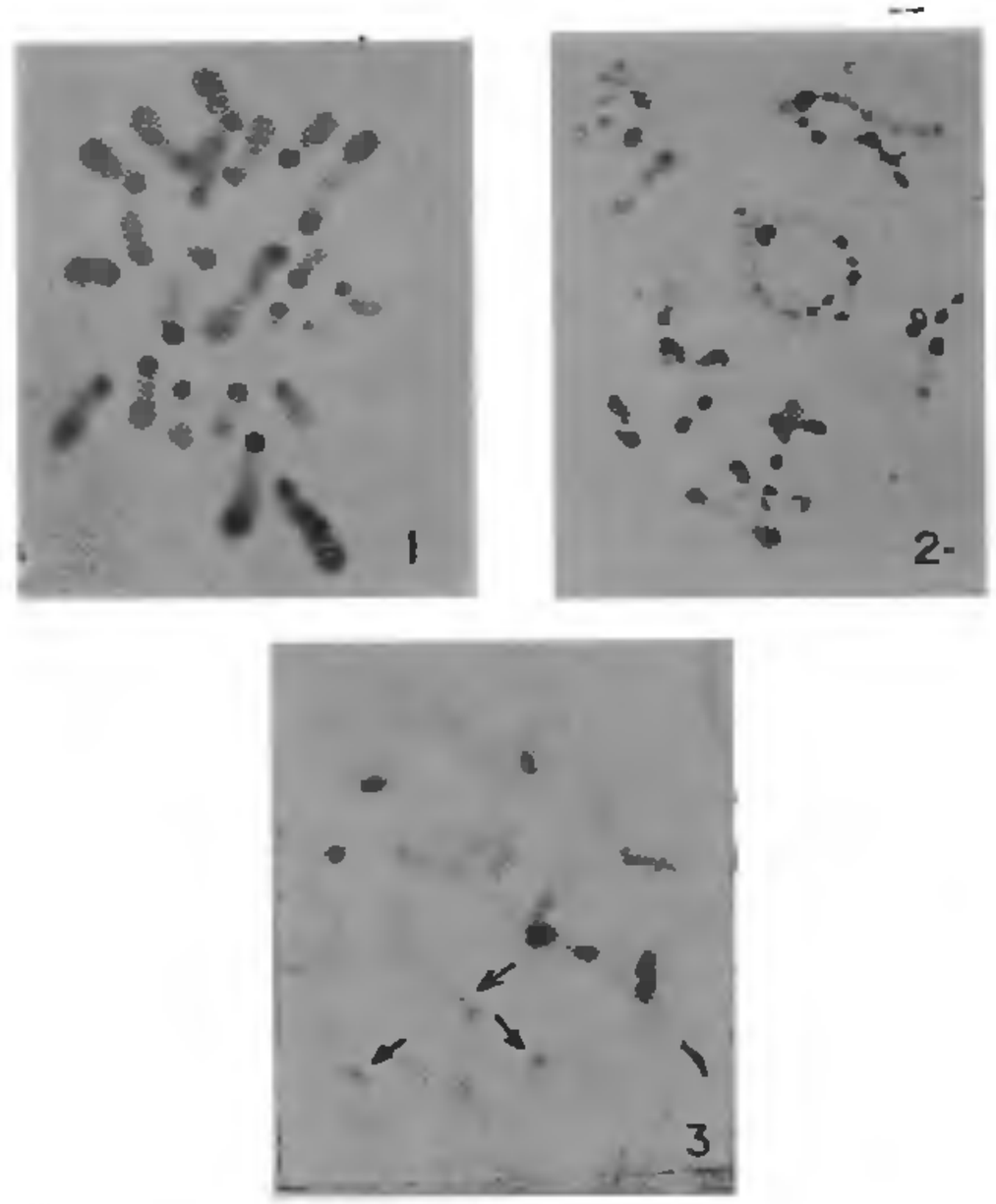
C. K. ATAL.

1. Staba, E. J., *Recent Advances in Phytochem.*, 1969, 75, 106.
2. Steck, W., *Planta Medica*, 1974, 25, 158.
3. Mandels, M, Mugbuire, A. and Hamed, M., *Technical Report*, 68–6–FL. Food Division, FL–58, 1967.
4. Veliky, I. A. and Martin, S. M., *Cand. J. Microbiol.*, 1970, 16, 233.
5. Lee, P. K., Carew, D. P. and Rosazza, J, *Lloydia*, 1972, 35 (2), 150.
6. Murashige, T. and Skoog. F., *Physiol. Plant.*, 1962, 15, 473.

PATTERN OF C-BANDING IN *ACRIDA TURRITA* (ACRIDIDAE—ORTHOPTERA)

THE pattern of distribution of heterochromatin in mammalian chromosomes has been extensively worked out[1·2]. But very little work has been done on the heterochromatin pattern in insect chromosomes. This communication therefore attempts to present the nature of C-banding in *Acrida turrita*.



FIGS. 1–3. Fig. 1. *Acrida turrita* spermatogonial chromosomes showing C-bands. Fig. 2. Diplotene showing distinct centromeric heterochromatin blocks. Fig. 3. Anaphase-I (one pole) showing the double nature (arrows) of the kinetochore.

Males of *Acrida turrita*, a short-horned grasshopper ($2n♂ = 23$) collected from Manasagangotri, Mysore (India) formed the material for study. Testes were dissected out and treated with hypotonic 0·9% sodium citrate solution for 45 minutes. The material was fixed in 1 : 3 acetic/methanol. Flame dried slides were prepared. C-bands were obtained