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ERGOT ALKALOID PRODUCTION IN SUSPENSION CULTURES OF *IPOMOEA BATATAS* POIR.

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DIFFERENT aspects of ergot alkaloid biosynthesis in *Claviceps* species, *Aspergillus fumigatus* and other microorganisms have been carried out by earlier workers[1–3]. Some plants belonging to the family Convolvulaceae also contain ergot alkaloids[4,5]. But very little work has been done in higher plants regarding the production of ergot alkaloids *in vitro*. The present communication deals with the influence of various growth regulators on the growth and alkaloid production in suspension cultures of *Ipomoea batatas* Poir.

Tuber callus of *I. batatas* Poir. was initiated on Murashige and Skoog's (MS) medium[6] containing 2 mg/l 2,4-dichlorophenoxyacetic acid (2,4-D) and 0.4 mg/l kinetin (KN). The culture vessels were incubated at a constant temperature (26 ± 2°C) and light (500 lux) conditions. Callus cultures were later maintained on the same medium subculturing every 30 days. Cell suspensions were initiated by inoculating 300 ± 30 mg of fresh tissue into 40 ml of MS basal medium with additives as above (but without agar-agar). Culture vessels were continuously agitated on a horizontal rotary shaker at 26 ± 2°C under constant illumination (500 lux). Growth of cell suspensions was measured as an increase in fresh and dry weights. Five replicate cultures were harvested at the end of 30 days for growth measurements and alkaloid production.

Dry tissue (100 mg) was macerated with 0.4 mg of ammonium hydroxide, 0.5 ml of ethanol and 0.5 ml of diethyl ether overnight, mixed in 25 ml of chloroform and boiled for about 15 min. The extract was then passed through a specially prepared glass column. The filtrate was evaporated to dryness and spotted on thin layer chromatographic plates (Kieselgel G Type 60). The plates were run in chloroform:ethanol (9:1) solvent system and sprayed with modified Van Urk's[7] reagent. The spots were eluted, dissolved in ethanol and total ergot alkaloids were measured in a colorimeter at 580 nm using Ergocrystine as standard.

Different concentrations (0.2, 1.0, 2.0 and 5.0 mg/l) of indole-3-acetic acid (IAA), indole-3-butyric acid (IBA), naphthaleneacetic acid (NAA) and 2,4-D were tested on suspension cultures of sweet potato for their influence on growth and ergot alkaloid production. Increasing concentrations of IAA increased the growth slightly but the alkaloid production was not much influenced (table 1). While maximum growth was obtained with the incorporation of IBA and NAA in the medium, highest production of total alkaloid was noticed on 2,4-D containing medium (on μg/100 mg dry weight basis). NAA is known to enhance RNA content[8] by as much as 50% which would result in the increased synthesis of structural and functional proteins. NAA is also found to accumulate rapidly in plant tissues and is then metabolized slowly to a series of unidentified derivatives[9]. The optimum level of NAA in the system can thus be maintained resulting in the rapid growth of tissues. Though higher concentrations of 2,4-D suppressed the growth, alkaloid production was maximum at 1 mg/l level (table 1) as was also reported by Nambiar[10] in callus cultures of *Evolvulus alsinoides* L. The total alkaloid content on 1 mg/l 2,4-D medium at the end of 30 days was almost equal to that of intact tubers. *In vivo* studies showed the maximum quantity of alkaloids in tubers (17 μg/100 mg of dry tissue) compared to leaves and stems. Hence, suspension cultures derived from tuber callus of sweet potato can be used further for commercial exploitation of these ergot alkaloids.

Low concentrations of KN and 6-benzylaminopurine (BAP) promoted the growth of the tissues considerably but higher concentrations decreased both fresh and dry weight of the tissues (table 2).