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INFLUENCE OF VITAMINS ON GROWTH OF *IN VITRO* GROWN NORMAL AND NEMATODE INDUCED ROOT GALL TISSUES OF *SOLANUM MELONGENA*

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Effects of various vitamins were observed on the growth of cultured normal and nematode induced root gall tissues of *Solanum melongena*. Different concentrations of individual vitamins showed variable effects on the growth of these tissues. Moderate to poor growth was observed on calcium pantothenate, choline chloride, cyanocobalamin, biotin and riboflavin. Thiamine and folic acid proved better than any other vitamins supporting good growth. Both the tissues tolerated the absence of pyridoxine. Considerable reduction in the growth of both the tissues was observed in absence of m-inositol and nicotinic acid. In all the experiments growth of gall tissue was better as compared to normal tissue.

Keywords : Gall tissue; *In vitro* growth; Nematode; Normal tissue; *Solanum melongena*; Vitamins.

Introduction

The study of role of vitamins in the metabolism of plant tissues grown *in vitro* has gained importance with the discovery of growth factor requirements. As most of the vitamins act as co-enzymes to a number of enzyme systems functioning with in the cell, they suggest the importance of vitamins in culturing plant tissues[1-5]. Various tissues synthesize vitamins in sub-optimal amounts and the addition of vitamins to nutrient media improve tissue growth[6-9]. Most of the studies are confined to normal tissues only[10,11]. In the present report studies have been made to compare the effect of ten vitamins on the growth of normal (hypocotyl in origin) and nematode induced root gall callus of *Solanum melongena*.

Material and Methods

Nematode (*Meloidogyne incognita*) induced root gall and normal tissues were isolated and maintained on Murashige and Skoog's (MS) medium[12] supplemented with 10.0 mg/l NAA and 0.08 mg/l kinetin. For this particular experimentation one tissue piece (about 200 mg) was grown in 100 ml 'Erlenmeyer' flasks containing 40 ml of solidified MS-medium. The cultures were incubated in dark at $26^0 \pm 2^0$C and around 55% relative humidity.

Concentrated stock solution of each of the chemically pure grade vitamins was prepared in distilled water. Various concentrations of vitamins viz. Biotin, Calcium pantothenate, Choline chloride, Cyanocobalamin, Folic acid and Riboflavin (0.5-10.0 mg/l); Thiamine. HCl, Pyridoxine. HCl, Nicotinic Acid (0.1-4.0 mg/l) and m-inositol (50-1,000 mg/l) were incorporated in the medium separately. The media were adjusted to pH 5.8 before autoclaving at 1.06 Kg/cm^2 and for 15 minutes. In control experiments the vitamin in question was eliminated. Each vitamin was replicated six times and each experiment was repeated three times. After 30 days of growth, the tissues were harvested and the fresh weight determined.

Results and Discussion

In controls, the growth of gall tissue was better (9.00 ± 0.20 gm/flask) in comparision to normal tissue (8.60 ± 0.17 gm/flask). On addition of biotin and riboflavin, the growth of normal and gall tissues decreased continuously with increasing levels. Growth was less than in the controls except at 0.5 mg/l of either vitamin (Table 1). Poor growth of the tissues on riboflavin has also been noted in *Lycopersicon esculentum*[13] and *Nigella*[14]. In all the cases the growth of gall tissue was better than normal tissue.

Growth increased very slightly on calcium pantothenate and choline chloride (0.5 - 5.0 mg/l) in comparision to controls.

Table 1. Effect of optimal concentration of different vitamins when incorporated in MS-medium separately, on growth of normal and gall tissue of *Solanum melongena*.

(Values are mean ± SD of 6 replicates)

S. No.	Vitamin incorporated in the MS-medium	TISSUE TYPE			
		NORMAL TISSUE		GALL TISSUE	
		Optimal concentration (mg/l)	Fresh Weight (gm±SD)	Optimal concentration (mg/l)	Fresh Weight (gm±SD)
1.	Biotin	0.5	8.6 ± 0.12	0.5	10.65 ± 0.20
2.	Calcium pantothenate	2.5	8.75 ± 0.15	0.5	10.50 ± 0.14
3.	Choline Chloride	2.5	9.40 ± 0.15	5.0	11.36 ± 0.20
4.	Cyanocobalamin	0.5	8.68 ± 0.16	1.0	10.15 ± 0.18
5.	Folic acid	1.0	9.58 ± 0.20	2.5	12.10 ± 0.22
6.	Riboflavin	0.5	8.45 ± 0.12	0.5	10.36 ± 0.12

Table 2. Effect of optimal concentration of different vitamins when incorporated in MS-medium separately, on growth of normal and gall tissue of *Solanum melongena*.

(Values are mean ± SD of 6 replicates)

S. No.	Vitamin incorporated in the MS-medium	TISSUE TYPE			
		NORMAL TISSUE		GALL TISSUE	
		Optimal concentration (mg/l)	Fresh Weight (gm±SD)	Optimal concentration (mg/l)	Fresh Weight (gm±SD)
1.	Thiamine	0.25	7.1 ± 0.10	0.25	9.0 ± 0.12
2.	Pyridoxine	0.5	6.8 ± 0.10	0.5	8.7 ± 0.10
3.	Nicotinic Acid	0.5	7.0 ± 0.12	0.5	9.0 ± 0.10
4.	m-inositol	100.0	6.7 ± 0.12	50.0	9.5 ± 0.12

On cyanocobalamin, the growth of the normal tissue decreased continuously in comparision to controls, however the growth of gall tissues was slightly higher than the controls on 0.5-1.0 mg/l but on further increase in cyanocobalamin the growth decreased (Table 1).

Folic acid proved better than any other vitamins as the maximum growth of normal tissue (9.58 ± 0.20 gm/flask) and gall tissues (12.10 ± 0.22 gm/flask) was observed on 1.0 mg/l and 2.5 mg/l, respectively (Table 1). Folic acid was found to be most essential for the growth of *L. esculentum*[13] and *Nigella* tissue and interactions of vitamin and hormones resulted in best growth of the tissue[14].

Addition of vitamins viz thiamine, pyridoxine, nicotinic acid and m-inositol increased the growth of normal and gall tissues. Thiamine appeared to be more essential as maximum growth of normal (7.1 ± 0.10 gm/flask) and gall (9.0 ± 0.12 gm/flask) tissue was recorded on 0.25 mg/l of thiamine (Table 2). Its absence in the medium reduced the growth of normal tissue (2.0 ± 0.13 gm/flask) as compared to the gall tissue (3.2 ± 0.10 gm/flask). Both the normal and gall tissues tolerated the absence of pyridoxine and with the increasing levels of pyridoxine the growth of both the type of tissues increased to some extent with an optimum at 0.5 mg/l. However, in *Nigella sativa* Chattopadhyay and Mukherjee[14] and in *L. esculentum* Mathur and Singh[15] recorded pyridoxine to be essential for the growth of callus cultures.

Differences were noted in the optimum amounts of m-inositol needed for the growth of gall and normal tissues, at 50 mg/l and 100 mg/l, respectively (Table 2). However, considerable reduction in the growth of both the tissues was observed in absence of m-inositol and nicotinic acid. Nicotinic acid increased the fresh weight of both normal and gall tissues at 0.5 mg/l concentration in the medium and was next to thiamine in its action.

In all the experiements the growth of gell tissue was better as compared to the normal tissue. This may be because of the fact that gall tissue have more endogenous level of auxins than in the normal tissue. Interactions of vitamins and hormones might be responsible for better growth of gall tissue.

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