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Large Scale Multiplication of Ginger (*Zingiber Officinale* Rosc.) From Shoot-tip Culture

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Abstract: The rapid multiplication from the soot-tip of ginger through ***in vitro*** culture has taken. Aseptic shoot tip from rhizome of ginger were cultured on MS medium. Three persent sucrose, 0.5% agar and different concentrations and combinations of hormone were used for the media. Vigorous ginger plantlets were obtained directly from the shoot tip explant supplemented with 2.5 and 0.5 mg/l Kn. 22-25 plantlets from each explants were directly transferred to the field without any acclimatization. 100% plants were successfully survived in the field condition.

Key words: Ginger, ***In vitro***, multiplication, zingiber, multiplication

Introduction

Zingiber officinale Rosc. (Ginger) of the family Zingiberaceae is a moderately sized family of relatively advanced monocotyledonous plant are zhizomatous herbs found throughout tropical and subtropical region with its main distribution in Asia. Several authors have quoted different figures for the total number of genera and species but it is probably appropriate to quote the world record to be at least 51 genera and 1500 species (Chen and Chen, 1988). Ginger is an important crop for a number of countries and its economic importance is reflected in the fact that it is currently the tenth most important spices in world terms. Ginger rhizomes are used as spices, in herbal medicine and as raw material in the food beverage and pharmaceutical industries. It is rich in secondary metabolites such as oleoresin (Bhagyalakshmi and Singh, 1988). The demand for fresh and dry ginger and its essential oil on the world market is high both in domestic and industrial trade. Ginger is vegetatively propagated through underground rhizomes exclusively and farmers usually take planting material from their own product, a practice that tends to spread diseases. Most crop improvement programs of this species are confined to evaluation and selection of naturally occurring clonal variation means of plant propagation and a tool for crop improvement (Vasil,1988). Clonal multiplication of ginger through multiplication has been reported (Hosoki and Sagawa, 1977; Wang, 1989; Balachandran ***et al.***, 1990 and Rout ***et al.***, 1997). There are some early reports on ***in vitro*** culture of ginger (Pillai and Kumar, 1982;

Nadgauda ***et al.***, 1980; Inden ***et al.***, 1988; Bhagyalakshmi and Singh, 1988; Noguchi and Yamakawa, 1988). The potential use of tissue culture in ginger was demonstrated by De Lange ***et al.*** (1997) who reported complete elimination of nematodes by tissue culture. The present communication describes a method for the rapid clonal multiplication.

Materials and Methods

From 1998 to 2000 in the laboratory of plant Tissue Culture Section, BCSIR the research was conducted. Shoot tips were collected from rhizomes derived from field grown locally available variety of ginger used as the source of explants in these investigation.

Preparation of explants

Clean rhizome pieces with shoot tips were surface sterilized in 0.1% mercuric chloride solution for 15 min and were thoroughly washed 5-6 times in sterile distilled water. Then the shoot tips of ginger rhizomes were excised with the help of sharp blade and collected in a sterilized petridish for inoculation. Aseptic shoot tip from rhizome of ginger were cultured on Murashige and Skoog medium, 3% sucrose, 0.5% agar and different concentrations of BAP (Benzylaminopurin), Kn (Kinetin) and 2,4-D (2,4-Dichlorophenoxyacetic acid). pH of the medium was adjusted to 5.8 before autoclaving. After four weeks of inoculation shoots with available roots were found in same medium. Multiple plantlets were separated and subculture onto same fresh medium. From each explant 22-25 plantlets were directly transferred to the field without any acclimatization.

Results and Discussion

These investigations were undertaken with view to optimize ***in vitro*** propagation technique considering various culture aspects for locally available ***Zingiber Officinale*** Rosc.

Fig. 1: Multiple shoots derived from shoot tip explant of ginger on MS+2.5 mg/l BAP+0.5 mg/l Kn

Fig. 2: Shoot-root development in ginger on the same medium, after 45 days of subculture

Different concentrations and combinations of BAP, Kn and 2,4-D were used in MS medium. Shoot tip from the aseptic rhizome were used as explant in this experiment. A total of twenty (all data are not shown) different concentrations of BAP combination with Kn and 2,4-D were tested in MS medium for the initiation and multiplications of shoots. Frequency of shoot proliferation was maximum at 2.5 mg l^{-1} BAP and 0.5 mg l^{-1} Kn and the number of shoot was 22-25 per explant (Fig. 1).The multiplication rate recorded in this study are higher than those reported by Nadgauda ***et al.*** (1978), Kuruvinashetty ***et al.*** (1982), Singh (1988), Hoque ***et al.*** (1999) Sharma and Singh (1997). It took 26 days for shoot induction and 30 days for root induction (Table 1).Balachandran ***et al.*** (1990) also obtained good response towards multiple shoot regeneration of ginger using same hormonal supplements in MS medium. Multiplication rate in the treatment with BAP 0.5 mg l^{-1}, which showed 2 plantlets were lowest. Among the BAP-2,4-D formulations, maximum multiplication was observed at BAP 1.0 and 0.5 mg l^{-1} 2,4-D where the number of shoots was 15. Numerous adventitious shoot primordia were observed near the basal portion of the shoot cluster. Regenerated plantlets produced prolific roots in same hormonal supplements in same medium. Huge plantlets were observed (Fig. 2) after 45 days of shoot initiation in the same medium (MS+2.5 mg l^{-1} BAP+0.5 mg l^{-1} Kn) This result contrast with previous reports (Nadgauda ***et al.***, 1978; Bhagyalakshmi and Singh, 1988; Rout ***et al.***, 1998; Sharma and Singh,1997).

Table 1: Shoot-root regeneration from shoot tip explants of Ginger in MS supplemented with different concentrations and combination with BAP, Kn and 2,4-D (No. of explants inculated= 25)

Growth regulators mg/l			Number of shoots/explants	Days to shoot induction	Days to root induction in same media
BAP	Kn	2,4-D			
0.5	-	-	2	25	-
0.5	0.5	-	7	28	-
1.0	0.5	-	7	20	-
1.5	0.5	-	7-10	30	-
2.0	0.5	-	10-12	25	35
*2.5	0.5	-	22-25	26	30
3.0	0.5	-	15-16	29	30
0.5	-	0.5	5-8	28	-
1.0	-	0.5	15	30	30
1.5	-	1.0	8-12	32	35
2.0	-	1.0	8-12	30	-
2.5	-	1.5	10	28	-
3.0	-	1.5	10	30	-

Fig. 3: Regenerated plantlets of ginger transplanted in plastic bag

Fig. 4: Successful plantation of ginger tissue cultured plant in the field

In vitro produced plantlets were easily established in soil without any acclimatization (Fig. 3, 4) Where as it has not been indicated in previous reports. Hundred percent plants were

successfully survived in the field. Earlier studies (Hosoki and Sagawa, 1977 and Nadgauda ***et al.***, 1978) reported only 60-70% survival. The ***in vitro*** multiplied plants were morphologically uniform and indistinguishable from their respective parent varieties.

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