chlorina, respectively. The results are explained accordingly in figure 1. The results clearly indicate that independent single recessive genes are responsible for *albina* and *chlorina* lethal chlorophyll mutations. Our results agree with similar results earlier obtained with six lethal chlorophyll mutations in pearl millet[2] and two non-lethal chlorophyll mutations in soybeans and pearl millet.[3,4]

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1. Bahl, J. R. and Gupta, P. K., *Theor. Appl. Genet.*, 1982, **63**, 23.
2. Burton, G. W. and Powell, J. B., *Crop Sci.*, 1965, **5**, 1.
3. Hanna, W. W., Burton, G. W. and Powell, J. B., *J. Hered.*, 1978, **69**, 273.
4. Nissly, C. R., Bernard, R. L. and Hittle, C. N., *J. Hered.*, 1981, **72**, 141.

PROMOTORY EFFECT OF GLUTAMINE ON ROOT REGENERATION FROM CALLUS CULTURES OF MUNG

R. P. SINGH and B. D. SINGH

Department of Genetics and Plant Breeding, Institute of Agricultural Sciences, Banaras Hindu University, Varanasi 221 005, India

CALLUS cultures of legumes generally require organic supplements[1] for profuse growth and, sometimes, bean seed extract[2], or glutamine[3] for growth and maintenance. Growth regulator combination with sugar concentrations is reported to determine bud and root regeneration from callus cultures[4], and absence of reduced nitrogen favours root regeneration while the presence favours embryo regeneration[5]. The present study reports the promotory effect of glutamine, a source of reduced nitrogen, on root regeneration from callus cultures of mungbean (*Vigna radiata* L. Wilczek).

Mungbean calli were initiated from shoot explants from 3-day old seedlings on B_5 medium[6] containing 0.1 μg ml^{-1} kinetin and 0.5 μg ml^{-1} 2,4-D. The calli were transferred to B_5 medium containing 0.05 and 0.1 μg ml^{-1} 2,4-D, 0.01 and 0.1 μg ml^{-1} kinetin and 0, 0.5, 1, 2 and 4 g l^{-1} glutamine in all possible combinations. Fresh and dry weights of calli, and the number of roots regenerated were recorded after 30 and 40 days, respectively. Data were analyzed according to the completely randomized design and means were compared using CD[7].

Glutamine significantly promoted fresh and dry weights of mungbean calli; interaction of glutamine with kinetin and or 2,4-D was significant. The promotory effect of glutamine was more pronounced at 0.1 μg ml^{-1} each of 2,4-D and kinetin. The glutamine containing media showed 17–77% more fresh and dry weights (table 1). A similar result has been reported in soybean[8]. Generally amino acids are inhibitory to the growth of callus and cell cultures in the presence of nitrate and ammonium salts[9]. But glutamine enhanced callus growth in mungbean in the presence of nitrate and ammonium salts of B_5 medium. In fact, calli of *Cajanus cajan* require glutamine for maintenance on B_5 medium[3].

The presence of glutamine in B_5 medium at low 2,4-D and kinetin concentrations promoted root regeneration; the promotory effect was evident particularly at 1, 2 and 4 g l^{-1} glutamine (table 1). These observations do not agree with those of Halperin[5], who reported that reduced nitrogen in the form of ammonium chloride or amino acids including glutamine

Table 1. *Effect of glutamine on callus fresh and dry weights of, and root regeneration from mungbean calli. Each value is mean of six replicates.*

Growth regulator (μg ml^{-1})		Glutamine	Fresh weight	Dry weight	Number of roots/ culture tube
2,4-D	Kinetin	(g l^{-1})	(mg)	(mg)	
0.05	0.01	0	889 6	87.8	2.3
		0.5	1189.6	112.0	2.7
		1 0	1295.0	124.6	3 5
		2.0	1307.8	126.6	6.1
		4.0	1361.2	131.4	7.3
0.05	0.1	0	904.8	89.0	1.5
		0.5	1389.4	134.0	2.7
		1.0	1429.2	142.4	6 0
		2.0	1478.9	144.6	10 0
		4 0	1599.2	148.6	6.3
0.1	0.01	0	877.0	84.0	2.5
		0.5	1234.4	120.4	2.7
		1 0	1289.0	126.0	8.0
		2 0	1357.6	131.0	7.6
		4.0	1370 8	133.6	7.0
0.1	0.1	0	1158.4	117.0	1.7
		0.5	1436 6	137.4	3 3
		1.0	1537.0	150.6	5.5
		2.0	1558 1	151.6	8.7
		4.0	1667 2	163 0	6.7
CD ($P = 0.05$)			108 2	9.1	0.5

suppressed rhizogenesis and promoted embryogenesis in carrot (*Daucus carota* L.) cells *in vitro*. The concentration of reduced nitrogen (30 mM) used by Halperin[5] was higher than those used in the present study (4.38 g l^{-1} as compared to up to 4 g l^{-1} glutamine in the present study) but this difference appears to be rather small to account for the differences in the findings of the two studies. Another possible explanation may be that different species may respond differently reduce to nitrogen in the culture medium.

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1. Scowcroft, W. R., *Adv. Agron.*, 1977, **29,** 38.
2. Crocomo, O. J., Sharp, W. R. and Peters, J. E., *Z. Pflanzenphysiol.*, 1976, **78,** 456.
3. Singh, R. P., *Tissue culture studies on Mung* (*Vigna radiata L. Wilczek.*) Ph. D. Thesis, Banaras Hindu University, Varanasi, 1981.
4. Tran Thanh Van, K., *Plant tissue culture and its biotechnological application* (eds) W. Barz, E. Reinhard and M. H. Zenk, Springer-Verlag, Berlin Heildelberg, New York, 1977, p. 365.
5. Halperin, W., *Am. J. Bot.*, 1966, **53,** 443.
6. Gamborg, O. L., Miller, R. A. and Ojima, K., *Exp. Cell Res.*, 1968, **50,** 151.
7. Steel, R. G. D. and Torrie, J. H., *Principles and procedures of statistics.* McGraw Hill, New York, 1960, p. 120.
8. Ojima, K. and Gamborg, O. L., *Biochemistry and physiology of plant growth substances* (eds.) F. Wightman and G. Setterfield, Runge Press, Ottawa, 1968, p. 857.
9. Gamborg, O. L. and Shyluk, J. P., *Plant Physiol.*, 1970, **45,** 598.

LONGEVITY OF GUARD CELL CHLOROPLASTS UNDER TOXIC ACTION

K. GEORGE

Department of Botany, University of Kerala, Kariavattom, Trivandrum 695 581, India.

THE unique property of the herbicidal chemical, 3-amino 1,2,4 triazole (AT) selectively degrading chloroplasts in green plant cells is reported[1-4]. At higher (lethal) concentrations, both mesophyll and guard cell chloroplasts of the treated leaves are destroyed. When applied in sublethal concentrations to growing buds it prevents chloroplast development and the leaves produced after this chemical treatment turn white. The whitened leaves when examined showed all the mesophyll cells devoid of chloroplasts. Among the mesophyll cell chloroplasts, those in the cells adjoining to the stomatal openings were the last to be degraded. Nevertheless, the chloroplasts of the stomatal guard cells were fully green, intact, functional and appeared to be least affected by the chemical treatment. The stomata in the whitened area were found to open during day and close at night just like those of normal untreated leaves. Their stomatal pore in the open condition was in no case smaller indicating that their guard cell chloroplasts were neither defunct nor sluggish. Further, examination of AT treated whitened areas of leaves from several plants, both dicot and monocot showed similar results.

The striking conservation of the guard cell chloroplasts of the AT treated leaves in contrast with the complete degradation of their mesophyll counterparts is interesting and a consideration of how the guard cell chloroplasts survive longer even under the toxic action of the chemical would be profitable because it has implications for the understanding of cellular aging and stomatal function. Longevity of guard cell chloroplasts in senescing leaves where the mesophyll chloroplasts get degraded and defunct[5] is a case comparable to the present observations after AT treatment. Accumulation of toxic waste materials in cells either by the absorption of an externally applied chemical as in the present case or by their formation inside the tissues on account of degenerative processes during senescence could be one of the important ultimate reasons for chloroplast degradation. In the guard cells of the stomatal opening such accumulation of toxic materials is prevented by their immediate removal through oxidation or detoxification due to sufficient aeration since the guard cells are most exposed to or are in immediate contact with fresh air. On the other hand, in the case of mesophyll chloroplasts as they are situated away from the stomatal opening and thus lacking in sufficient ventilation, immediate removal of toxic materials through oxidation naturally is not possible. This should account for the survival and longevity of guard cell chloroplasts in both AT treated (figures 1 and 2) and senescing leaves.

The fact that at lethal concentrations AT degrades both mesophyll and guard cell chloroplasts alike shows that both mesophyll and guard cells are permeable to AT. This aspect of differential permeability of mesophyll and guard cells to AT though not extensively verified with higher plant cells, experi-