tures of *Cucumis melo* var. *utilissimus* Duthie and Fuller, *Cucumis melo* Linn., *Cucumis sativus* Linn., *Citrullus vulgaris* Schrad., *Momordica charantia.* Linn. and *Luffa acutangula* (Linn.) Roxb. However, with passage of time, in old cultures (23 months) of *Momordica* NAA (1.0 mg/l) plus adenine (33.75 mg/l) induced buds and plantlets and in 26-month old *Cucumis melo utilissimus* callus, NAA plus adenine treatment-induced embryoids and benzyl adenine (BA) 1.0 mg/l plus indole 3-butyric acid (IBA) 1.0 mg/l produced buds and plantlets. These treatments were ineffective in early phase of the cultures.

In view of the failure of differentiation in freshly isolated callus tissues used and success in inducing shoot-bud formation in the old ones, effect of Cucurbitacin B and E was tested on the old cultures of *Momordica* & *Cucumis* and also in the *Solanum nigrum* morphogenetic system[9] developed in this laboratory. Explants from stock cultures of *S. nigrum* were transferred to morphogenetic medium comprising basal medium of Murashige and Skoog[10] (BM) containing BA (0.5 mg/l). Authentic samples of Cucurbitacin B & E were added at the concentrations of 0.05, 0.25 and 1.25 mg/l to the respective morphogenetic media of the three cultures. The cultures were grown under a photoperiod of 16 hr light with an intensity of 2000 lux at the culture level and at 25° ± 1°C. Control sets were run simultaneously. Neither suppression nor acceleration of differentiation was observed in all the three materials. The result clearly indicated that failure of bud formation in fresh cultures of cucurbits in the numerous treatments tested was not due to adverse effect of Cucurbitacins, if present.

It has been reported earlier that bud initiation is preceeded by starch accumulation[11]. Induction of α-amylase activity by GA is well known[12]. Gibberellins have been shown to repress shoot-bud formation in callus tissues[13,14]. The presence of GA or GA-like substances in callus tissues has been reported[15]. Thus, GA at an appropriate concentration in the tissues will reduce or inhibit shoot-bud differentiation. Cucurbitacins antagonize GA-induced α-amylase activity[6]. Though not experimentally demonstrated, callus cultures of Cucurbits are likely to contain Cucurbitacins. If failure of bud induction in the early period of growth be considered to be due to the presence of effective levels of GA within the tissues, then it could be expected that the presence of cucurbitacin will counteract the effect of GA and may stimulate bud induction.

Further studies are being carried out to elucidate the regulatory role of Cucurbitacin, if any, in callus growth and differentiation.

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DIRECT AND INDIRECT EFFECTS OF GAMMA RAYS ON STIMULATION OF MORPHOGENESIS IN LONG TERM TISSUE CULTURE OF RICE (*ORYZA SATIVA* L.).

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REGENERATION of plants from various explanted tissues of rice has been reported by several workers[1-4]. However, the calli lose the capability to regenerate shoots during the course of subcultures[2,5]. For exploiting the benefits of tissues culture techniques, methods to maintain calli and induce regeneration of

plantlets in long term cultures need to be worked out. This communication reports the stimulation of morophogenesis in a cell line of rice maintained for 8 months using gamma irradiation. Hulled grains of rice cv. IR-8 were surface-sterilized and aseptically germinated on agar (0.8%)-sucrose (0.5%) medium. About 1 cm long root pieces of 1 week-old seedlings were cultured on the medium of Murashige and Skoog[6] supplemented with 2 mg/l 1,2,4-D (MS_2)and incubated at 27 ± 1° C in the dark. The cultures were maintained by reculturing them on a fresh medium (MS_2) at an interval of one month. For regeneration plants, the calli were transferred to the basal medium of Murashige and Skoog[6], (MS) and incubated under constant illumination of 2500 lux. Eight month-old cultures which had lost the capacity to differentiate shoots were exposed to 0.5, 1 and 2 kR doses of gamma rays in a chamber having an intensity of 23.3 kR/h (source ^{60}Co). The cultures were then transferred to MS medium and incubated under light. In another set, the medium was also exposed to these doses of gamma rays and irradiated as well as non-irradiated calli transferred to these media.

On MS_2 medium, the root explants formed nodular calli within a few weeks of culture. These calli turned green on transfer to MS medium and regenerated whole plants within a month. The number of plants varied from 1–5 per callus piece of about 100 mg fresh weight. However, as the number of passages of the calli on MS_2 medium increased, their shoot formation capability decreased and 8 month-old calli failed to produce shoots on MS medium though roots were readily regenerated. Table 1 shows that irradiating the callus with 1 kR dose had a more pronounced stimulating effect on plant regeneration (80%) than irradiating the medium (66%). Irradiatiation of both the medium and the callus resulted in regeneration of 33% calli only. All these treatments, however, were positive in inducing plant restoration as compared to the control cultures grown on non-irradiated medium where none of the calli produced plantlets. The treated calli differentiated numerous shoots in 4 weeks of culture on MS medium. Exposure to 0.5 kR dose did not induce regeneration which may be due to the threshold requirement for plant regeneration in this cell line between 0.5 and 1 kR. However, 2 kR dose caused blackening of the calli without further growth.

Though the exact mode of action of gamma irradiation on the calli and the media is not understood, low doses have been found to stimulate organogenesis in tissue cultures of other species also[7-9]. Direct effects of irradiation are speculated to be due to lowering of the endogenous levels of auxins in tissues[10] whereas the effects on media are probably due to the changes brought about in sucrose[11], rendering the medium more favourable for inducing plant regeneration.

TABLE 1

Effect of 1 kR dose of gamma irradiation on the differentiation of 8 month-old rice tissue culture.

Treatment	Total No. of cultures	No. of shoot forming cultures	Percent of shoot forming calli
Control	16	0	0
Calli irradiated	15	12	80
Medium irradiated	18	12	66
Calli and medium irradiated	18	6	33

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