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Development of an Encapsulated Prill Formulation of *Gliocladium virens* Miller, Giddens & Foster

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Formulation and application methods are often of paramount importance in biological control (Papavizas and Lumsden, 1980; Papavizas and Lewis, 1983; Papavizas *et al.*, 1984). Accordingly several techniques *viz*, antagonist in liquid (Kerr, 1980), organic matter (Wells *et al.*, 1972), as seed treatment (Harmen *et al.*, 1980; Windel, 1981; Fravel *et al.*, 1985) and in vermiculite or in clay such as pyrax (Fravel *et al.*, 1983) have been tested. Reports of the incorporation of mycoherbicides into sodium alginate suggested that this method may have potential for use with biocontrol fungi (Walker and Connick, 1983). The resulting granular preparation is not only lighter in weight than liquid but also the granules are more uniform and less bulky than most organic matter preparation. The reaction between aqueous solution of sodium alginate and metal cation Ca^{++} to form gels (Kondo, 1979) has been used to formulate myco-and chemical herbicide (Barrette, 1978; Connick, 1982; Walker and Connick, 1983). Bentonite is normally added as bulking agent. The method produces bio-degradable prills of relatively uniform size. The prills were found convenient for storage and compatible with commonly used agriculture machinery.

The objectives of the present study were to formulate the biocontrol agent, *Gliocladium virens* Miller, Giddens and Foster in sodium alginate prills, study its suitability for applications and storage and investigate the shelflife of the antagonist in prills.

The method adopted for the preparation of sodium alginate prills was described by Fravel *et al.* (1985). Ten 'g' of sodium alginate was added to a litre of warm distilled water and boiled till complete dissolution. To this was added bentonite clay @ 10 g/litre and 8-10 g of air dried biomass of *G.virens*. The entire sodium alginate - bentonite clay biomass mixture was thoroughly homogenised at room temperature in a waring blender and the homogenate was slowly dropped into 0.2 M solution of dehydrated $CaCl_2$ through a 1.5 mm diameter orifice of burrette to facilitate the formation of spherical prills. The prills were air dried for 24 hour in laminar flow table and stored in polypack at room temperature.

The shelflife of the antagonist in sodium-alginate prills was studied under room condition by determining the c.f.u./ml of suspension of prills periodically. The initial population (c.f.u./ml suspension) was determined immediately after mixing the inoculum with sodium-alginate slurry and the initial population of antagonist in prills was determined just after formation of prills. Thereafter, the c.f.u./ml suspension of prills was determined periodically for 12 weeks. The prills were acerated in a phosphate buffer mixture of 8.7×10^{-2} (M) Na_2HPO_4 and 3.0×10^{-2}(M) KH_2PO_4 (pH 7.7). Twelve formed prills were considered as equivalent to 1 ml suspension of sodium alginate biomass of antagonist - bentonite mixture. Enumeration of c.f.u. was done by dilution plate technique using modified TSM (Saha, 1995).

The results showed that the initial population of the antagonist in sodium alginate-biomass-bentonite suspension, irrespective of isolates ranged between $4.0\text{-}5.0 \times 10^8$ c.f.u./ml of suspension (Table 1). There was a reduction in population during the first 24 h of formation of prills and was considered as an establishment phase of the antagonist in the prills. Thereafter an increase in population of an-

Table 1. Shelf life of *G.virens* isolates in alginate prills

Isolate	Initial population in dry biomass (x 10^8/gm)	Mean population (*C.f.u. x 10^8 per ml)/days after				
		0	1	12	24	48
15 GV1	5.3*	4.6	11.6	30.4	9.4	1.0
26 GV 1	4.5	4.0	8.4	22.5	11.0	0.6
1 AGV 2	4.4	4.25	7.2	20.5	7.8	0.8
1 AGV 6	4.5	4.0	7.8	22.0	9.4	0.6

	S.Em	C.D. (P=0.05)
Isolate	1.26	3.949
Period of incubation	1.54	4.27
Isolate x period of incubation	3.08	8.56

* Average of five replications and each replication is based on three separate determinations.

tagonist (Ca.20.0-30.0 x 10^8 c.f.u./ml) took place for 12 days. Then the population declined steadily and by 72 days there were no viable *G.virens* in the prills. It was also noted that the population of the antagonist within prill went below threshold (Ca.1 x 10^8 c.f.u./g soil) at about 45-50 days and prills became shrivelled, deformed and smaller in size with time. The initial decrease in population of the antagonist during the first 24 h after the formation of prills might be due to drying immediately after they were formed. Fravel *et al.* (1985) observed a loss in viability, even upto 99%, of most of the organisms including *Gliocladium* spp. in Na alginate pellets. In the present experiment, 4-5 fold increase in population upto first 2 weeks was followed by an initial reduction. The Na alginate prills are effective as a point source of living propagules and it possess several advantages including low cost of production, versatility and composition (Fravel *et al.*, 1985)

The basic material sodium alginate has been used for the formulation of different materials (Scher, 1977; Burrelle, 1978; Walker and Connick, 1983). Sodiumalginate and Ca Cl_2 are commonly used as food additives and are considered to be non-toxic to non target organisms (Fravel *et al.*, 1985)

The results of the present experiment, make it quite clear that for commercialization, the methods of prill formulation will have to be modified by incorporating slowly utilizable nutrient source to permit long duration survival and preferably, proliferation of antagonist in storage.

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KEY WORDS : *Gliocladium virens*, formulation, Sodium alginate prill, shelf life

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