

Survival Study of an Agriculturally Important Microbial Consortium in a Selected Formulations

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ABSTRACT

A laboratory investigation was carried out to study the survival rate of an agriculturally important microbial consortium (*Rhizobium sp.*, *Bacillus megaterium* and *Pseudomonas fluorescens*) in a selected formulations (alginate based, fluid bed dryer, lignite based and liquid) as a single, dual and triple inoculants. After 180 days of storage, the higher survival rate (per cent) of *Rhizobium sp.*, (95.36 per cent), *Bacillus megaterium* (93.81 per cent) whereas, *Pseudomonas fluorescens* recorded of 95.46 per cent in alginate based formulations. Overall, the maximum viable cells were maintained in the alginate based formulations followed by liquid formulation and lower per cent survival rate of the inoculants were observed in fluid bed dryer based formulations.

Keywords: Alginate, Fluid Bed Dryer, Lignite, Liquid formulations, *Rhizobium sp.* *Bacillus megaterium*, *Pseudomonas fluorescens*

INTRODUCTION

The formulation consists of establishing viable cells in a suitable formulation together with additives that aid in stabilization and protection of microbial cells during storage, transport and at the target. The establishment of a member of plant growth promoting rhizobacteria in the rhizosphere could be achieved either by seed, seedling or soil inoculations with a suitable inoculant formulation. Inoculant formulations are

generally categorized into three groups such as carrier based inoculants, liquid based inoculants, and granular inoculants.

The development of successful microbial inoculants involves a selection of a suitable formulation to support the growth of microorganisms and to a maintain maximum number of viable cells. An ideal formulation should be cost effective, non-toxic, easy to process (Vijaykumar & Brahmaprakash, 2020).

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MATERIALS AND METHODS

The experiment was carried out in the Department of Agricultural Microbiology, University of Agricultural Sciences, Gandhi Krishi Vigyan Kendra, Bengaluru-65

2.1 Preparation of different microbial consortium formulations

Three different agriculturally important microorganisms (*Rhizobium* sp., *Bacillus megaterium* and *Pseudomonas fluorescens*) were prepared in single, dual and triple combination by using four (alginate based, fluid bed dryer, lignite based and liquid) different formulations as suggested by Vijaykumar and Brahmaaprakash (2018)

2.2 Survival rate study

The initial microbial load in each combination (single, dual and triple) of different formulations (alginate based, fluid bed dryer, lignite based and liquid) were considered as one hundred percent population. Further, the per cent survival rate was monitored as suggested by Shilpa and Brahmaaprakash (2016)

2.2 Treatment details

The experiment was consisting of 7 treatments: T₁ – *Rhizobium* sp., T₂ – *Bacillus megaterium*., T₃ – *Pseudomonas fluorescens*., T₄ – *Rhizobium* sp.+ *Bacillus megaterium*., T₅ – *Rhizobium* sp.+ *Pseudomonas fluorescens*., T₆ – *Bacillus megaterium* + *Pseudomonas fluorescens*., T₇ – *Rhizobium* sp.+ *Bacillus megaterium* + *Pseudomonas fluorescens*

RESULTS AND DISCUSSION

3.1 Shelf life of single inoculants in different formulations

3.1.1 *Rhizobium* sp.

After 180 days of storage, the per cent survival of *Rhizobium* sp., reduced to 94.56 per cent in alginate based formulation (Fig. 1). The per cent survival of *Rhizobium* sp. in FBD based formulation after 180 days of storage reduced to 73.96 per cent (Fig. 2). In lignite formulation, per cent survival of *Rhizobium* sp., after 180 days of storage reduced to 89.00 per cent (Fig. 3). Whereas, in liquid formulation after 180 days of storage the

population was reduced to 92.75 per cent (Fig. 4).

3.1.2 *Bacillus megaterium*

After 180 days of storage, the per cent survival of *B. megaterium* in alginate based formulation was reduced to 93.81 per cent (Fig. 1). In case of fluid bed dryer (fbd) based formulation, the per cent survival of *B. megaterium* was reduced to 90.11 per cent (Fig. 2). The per cent survival of *B. megaterium* in lignite formulation after 180 days of storage was reduced to 89.19 per cent (Fig. 3). Whereas, *B. megaterium* got reduced to 92.97 per cent after 180 days of storage in liquid formulation (Fig. 4).

3.1.3 *Pseudomonas fluorescens*

The per cent survival of *P. fluorescens* in alginate based formulation after 180 days of storage was reduced to 95.46 per cent (Fig. 1). After 180 days of storage, The per cent survival of *P. fluorescens* in FBD based formulation was reduced to 89.93 per cent (Fig. 2). In lignite formulation, the per cent survival of *P. fluorescens* after 180 days of storage was reduced to 86.14 per cent (Fig. 3). Whereas, in liquid formulation, The per cent survival of *P. fluorescens* was reduced to 93.81 per cent after 180 days of storage (Fig. 4).

3.2 Shelf life of dual inoculants in different formulations

3.2.1 *Rhizobium* sp. and *Bacillus megaterium*

The per cent survival of *Rhizobium* sp. and *B. megaterium* in alginate based formulation after 180 days of storage were reduced to 95.25 and 91.32 per cent respectively (Fig. 1). In FBD based formulation, the per cent survival of *Rhizobium* sp. and *B. megaterium* were reduced to 78.43 and 85.57 per cent respectively after 180 days of storage (Fig. 2). The per cent survival of *Rhizobium* sp. and *B. megaterium* in lignite formulation after 180 days of storage were 83.00 and 85.16 per cent respectively (Fig. 3). In case of liquid formulation, the per cent survival of *Rhizobium* sp. and *B. megaterium* were reduced to 93.29 and 91.18 per cent respectively after 180 days of storage (Fig. 4).

3.2.2 *Rhizobium* sp. and *Pseudomonas fluorescens*

The per cent survival of *Rhizobium* sp. and *P. fluorescens* in alginate based formulation after 180 days of storage were reduced to 95.36 and 94.67 per cent respectively (Fig. 1). In case of FBD based formulation, the per cent survival of *Rhizobium* sp. and *P. fluorescens* were reduced to 81.45 and 87.68 per cent respectively (Fig. 2). Whereas, in lignite formulation, the per cent survival of *Rhizobium* sp. and *P. fluorescens* were reduced to 84.96 and 88.87 per cent respectively (Fig. 3). In case of liquid formulation, the per cent survival of *Rhizobium* sp. and *P. fluorescens* were reduced to 92.92 and 94.23 per cent respectively (Fig. 4).

3.2.3 *Bacillus megaterium* and *Pseudomonas fluorescens*

After 180 days of storage, the per cent survival of *B. megaterium* and *P. fluorescens* in alginate based formulation after 180 days of storage reduced to 93.06 and 95.06 per cent respectively (Fig. 1). In FBD based formulation, the per cent survival of *B. megaterium* and *P. fluorescens* were reduced to 88.26 and 87.75 per cent respectively (Fig. 2). The per cent survival of *B. megaterium* and *P. fluorescens* in lignite formulation after 180 days of storage reduced to 80.89 and 88.87 per cent respectively (Fig. 3). Whereas, the per cent survival of *B. megaterium* and *P. fluorescens* in liquid formulation after 180 days of storage were reduced to 92.06 and 91.47 per cent respectively (Fig. 4).

Rhizobium sp., in inoculant 5 (*Rhizobium* sp., + *P. fluorescens*) maintained maximum cell density after 180 days of survival studies in alginate based formulation whereas, *P. fluorescens* in inoculant 6 (*B. megaterium* + *P. fluorescens*) of alginate based formulation recorded statistically on par cell density with that of inoculant 5 of liquid formulation after 180 days of survival studies.

3.3 Shelf life of triple inoculants in different formulations

3.3.1 *Rhizobium* sp., *Bacillus megaterium* and *Pseudomonas fluorescens*

After 180 days of storage, the per cent survival of *Rhizobium* sp. (91.11), *B. megaterium* (86.77) and *P. fluorescens* (95.16) were reduced in alginate based formulation after 180 days (Fig.1). in FBD formulation, The per cent survival of *Rhizobium* sp., *B. megaterium* and *P. fluorescens* were reduced to 80.96, 91.36 and 87.80 per cent respectively (Fig. 2). The per cent survival of *Rhizobium* sp., *B. megaterium* and *P. fluorescens* in lignite formulation after 180 days of storage reduced to 89.29, 82.32 and 93.27 per cent respectively (Fig. 3).

Whereas in liquid formulation, the per cent survival of *Rhizobium* sp., *B. megaterium* and *P. fluorescens* were reduced to 90.60, 88.51 and 92.39 per cent respectively after 180 days of storage (Fig. 4).

The population of *Rhizobium* sp., *B. megaterium* and *P. fluorescens* as triple inoculants (*Rhizobium* sp., + *B. megaterium* + *P. fluorescens*) have recorded the significance of differences in all the test formulations.

P. fluorescens in the inoculant 7 (*Rhizobium* sp., + *B. megaterium* + *P. fluorescens*) of alginate based formulation maintained maximum cell density followed by in liquid formulation, lignite whereas, lower cell density was recorded in FBD based formulation.

Among all the test formulations, the higher per cent survival of the inoculants were observed (after 180 days of storage) in the alginate based formulations followed by liquid formulations and least was recorded in the fluid bed dryer based formulations. The higher per cent survival rate in alginate based formulation might be due to the reduced metabolic activity of cells by holding the cells inside the bead. Lower cell density was recorded in the FBD based formulations because of hot air passed through the bed for a prolonged time and hence, possibly, the inocula of inoculants might have come down but interestingly, the inoculant *B. megaterium* could able to maintain optimum cell density may be due to spore formation. The similar studies were reported by Arya et al. (2012), Bashan et al. (2014), Lavanya (2014), Sneha

and BrahmaPrakash (2017), Swapna and BrahmaPrakash (2013), Dayamani and

BrahmaPrakash (2014) and Lavanya (2014).

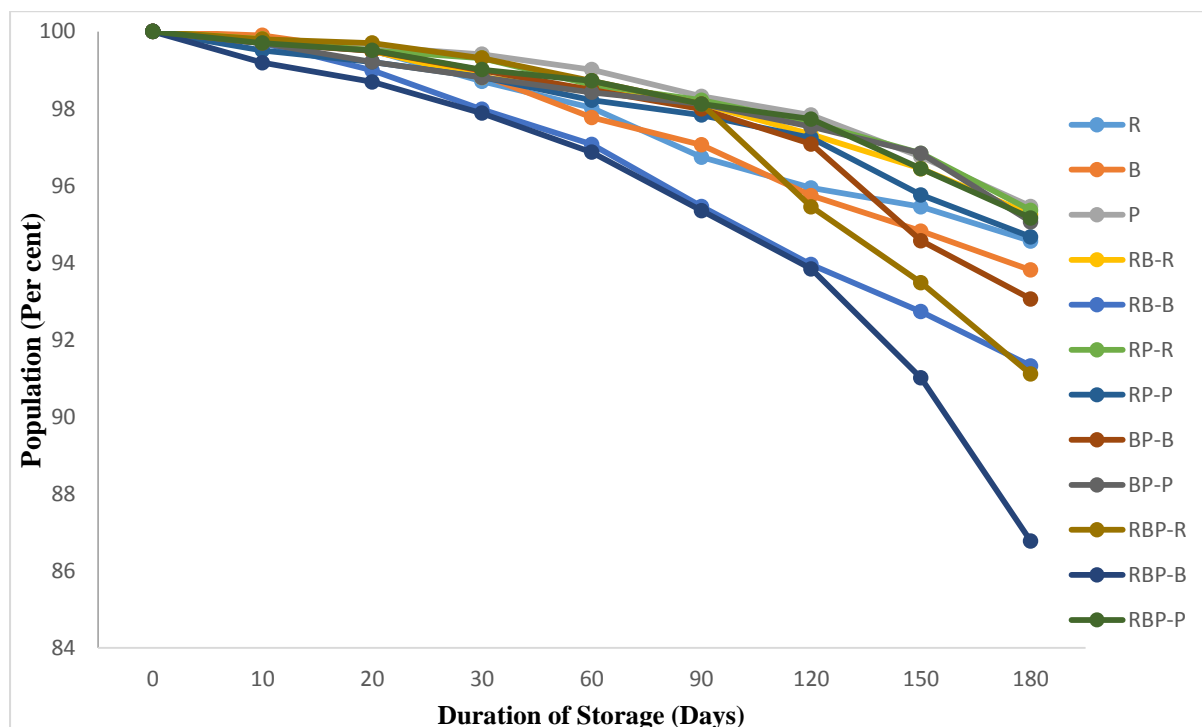


Fig. 1: Per cent survival of microbial inoculants of a consortium in alginate based formulation up to 180 days

R; *Rhizobium* sp., B; *Bacillus megaterium* P; *Pseudomonas fluorescens*

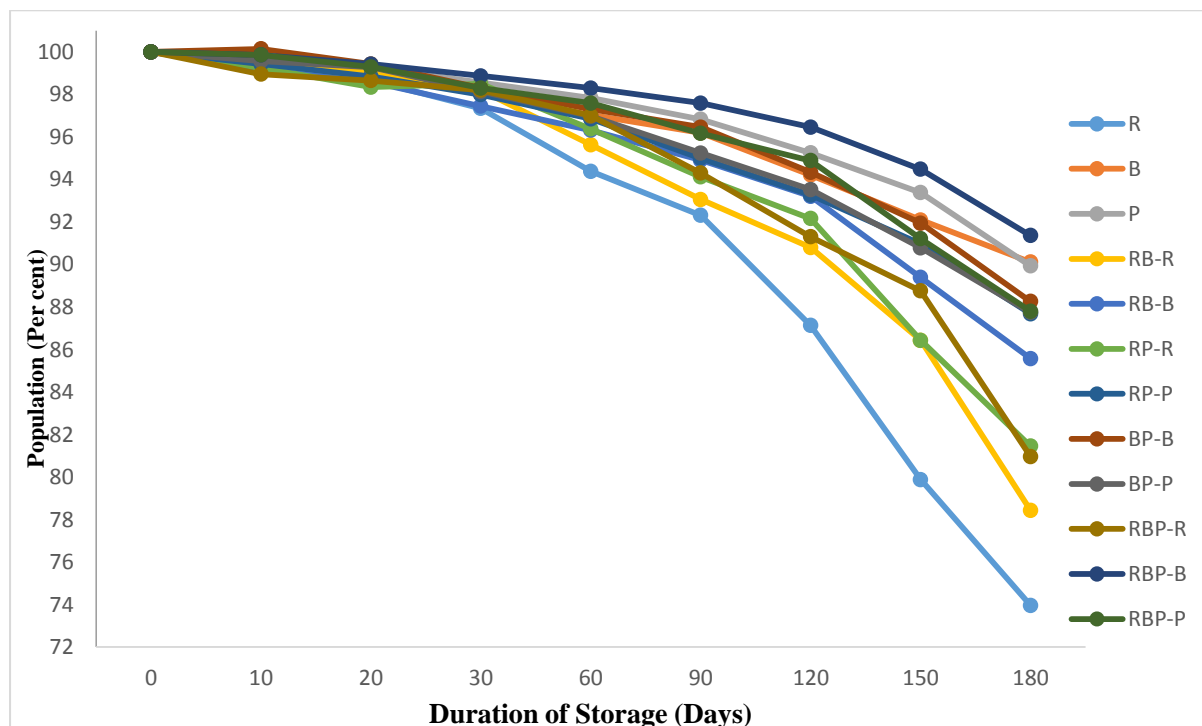


Fig. 2: Per cent survival of microbial inoculants of a consortium in fluid bed dryer based formulation up to 180 days

R; *Rhizobium* sp., B; *Bacillus megaterium* P; *Pseudomonas fluorescens*

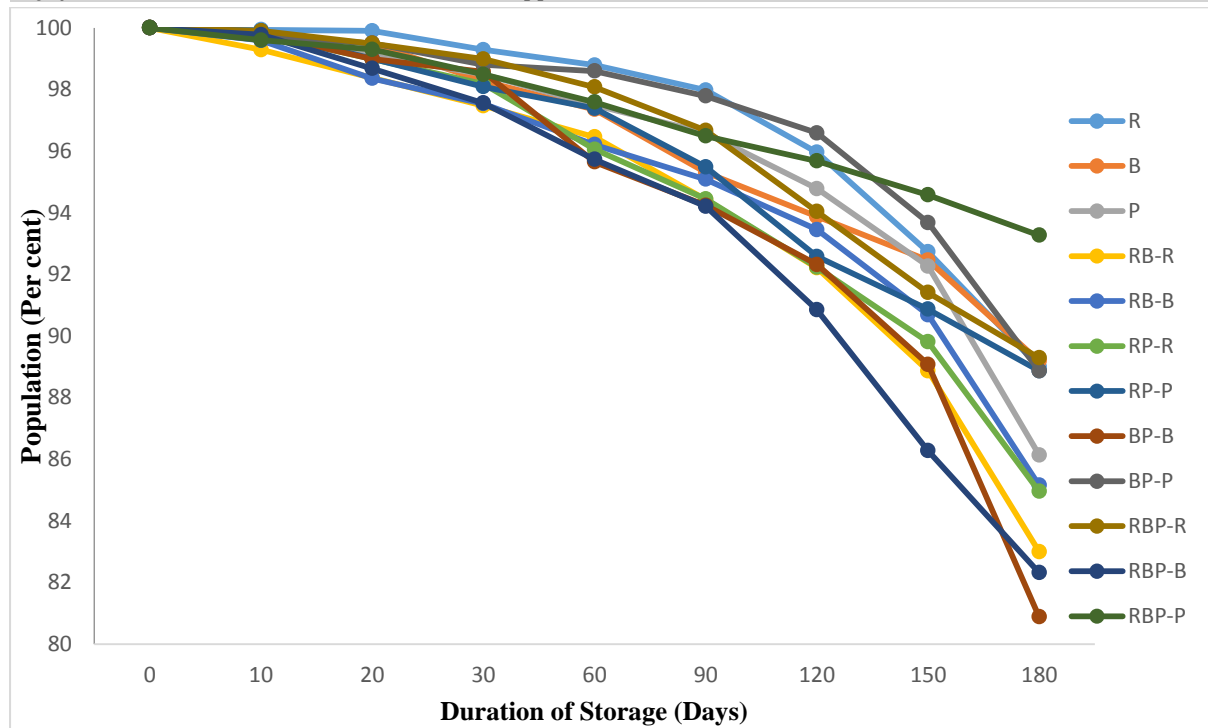


Fig. 3: Per cent survival of microbial inoculants of a consortium in lignite formulation up to 180 days
 R; *Rhizobium* sp., B; *Bacillus megaterium* P; *Pseudomonas fluorescens*

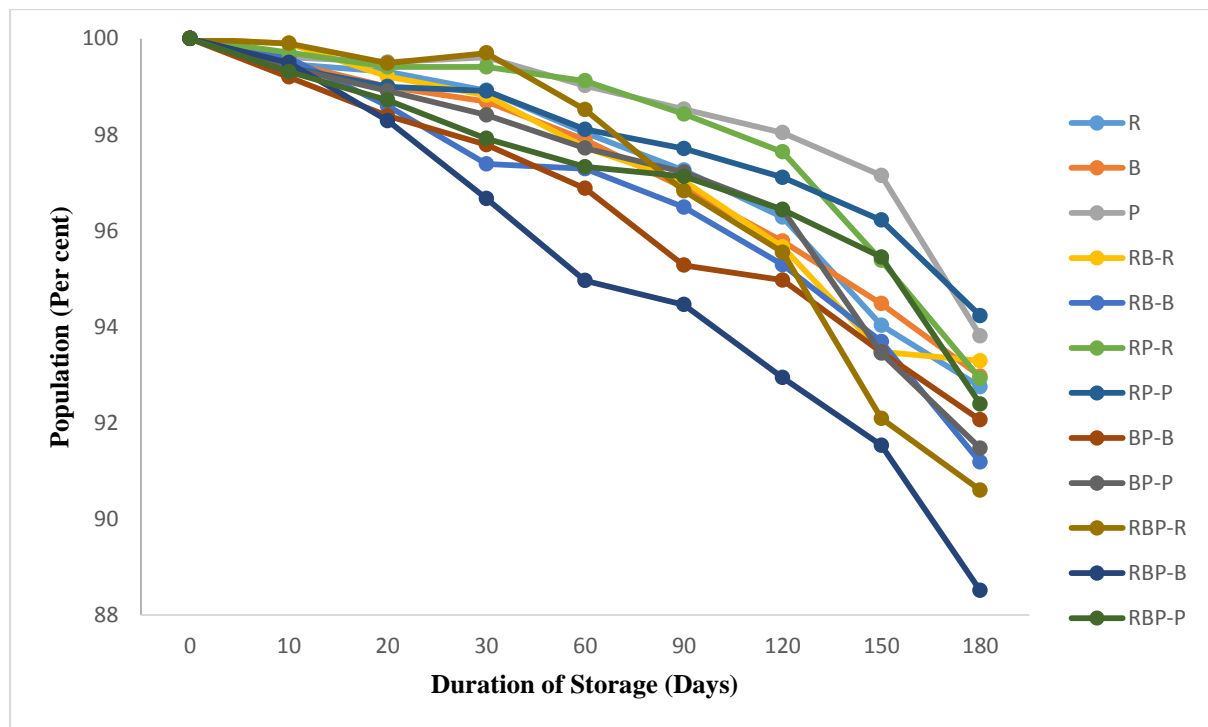


Fig. 4: Per cent survival of microbial inoculants of a consortium in liquid formulation up to 180 days
 R; *Rhizobium* sp., B; *Bacillus megaterium* P; *Pseudomonas fluorescens*

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