



Determination of Seed germination ability and metabolic changes on *Zea mays* L. by the treatment of Chemical, Bijamrita and the cyanobacterial biofertilizer extract (Cyanospray).

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Abstract

This research aims to investigate the seed germination at Model Organic Farm, Bharathidasan University, Tiruchirappalli, Tamilnadu, India, to determine the germination ability of NaCl, Bijamrita and cyanospray solution on *Zea mays* L. seed under laboratory conditions. Different treatments of NaCl (20 mM), Bijamrita and cyanospray were applied at various concentrations (0.1% to 0.5%), as a growth medium for seed germination. All the growth attributes such as germination percentage, Epicotyl length, Hypocotyl length, and Number of roots to be increased at 0.3% of cyanospray solution when compared to other concentrations of cyanospray, control and other treatments. During germination process, the *Zea mays* L. seeds protein content reduced but the total soluble sugar content increased at 0.3% of cyanospray when compared to control and other treatments. Hence, it is concluded that the efficiency of cyanospray at 0.3% level showed maximum seed germination ability when compared to other treatments.

Key words: Cyanospray, *Zea mays* L. Epicotyl, Hypocotyl, growth medium.

Introduction

Germination of seeds involves a rise in general metabolic activity and initiates the formation of a seedling from the embryo. The first step in germination is absorption of water, which results in swelling of the seed. This water uptake is accompanied by a rapid increase in the respiratory rate of the embryo. Soon after the absorption of water by the seed, enzymes such as lipases, proteinases,

phosphatases and hydrolases act on the seed thereby helping to break down the storage materials (Bewley and Black, 1985). The breakdown products are later transported from one part of the seed to another and new materials are also synthesized (Arteca, 1997). The production of growth promoting substances by cyanobacteria and their direct growth promoting effect on rice crop has been clearly stated by Kannaiyan *et al.* (1997). Earlier study report indicated that in addition to the supply of N₂ fixing cyanobacteria could also release a variety of substance such as hormones, vitamins amino acid and polysaccharide (Kaushik, 1993) and growth promoting substances (Ravishankar, 2000).

Zea mays L. (corn) is one of the most important crops in India which serves as food and corn oil for human consumption, feed for livestock and poultry, and raw material for agro based industries (Khatoon *et al.* 2010). Maize has become highly polymorphic and is perhaps the cultivated species that contains the greatest amount of genetic variability. Being cross pollinated, salinity tolerance may exist in maize (Paterniani, 1990). Normally during the corn seed germination numerous problems occurred because of initial growth nutrient deficiency and other environmental factors. Hence, this study focused on the effect of cyanospray and chemical solutions on seed germination of *zea mays* L.

Materials and Methods

Organism and culture conditions

A fresh water cyanobacterium belonging to *Oscillatoria annae* was obtained from the germplasm of National Facility for Marine Cyanobacteria, Bharathidasan University, Tiruchirappalli, Tamilnadu, India. The culture was maintained in BG11 medium (Rippka *et. al.*, 1979), at 1500 lux. at 25±20 C with 10/12 hrs light /dark cycle.

Lignocellulosic material

Coir pith was collected from coir industry, near Srirangam, Tiruchirappalli, Tamilnadu, India.

Preparation of Cyanospray solution

Preparation of cyanospray was carried out in which cyanobacteria (*Oscillatoria annae*) and coir pith was inoculated in 1:10 ratio (wet weight: dry weight). After 20-25 days of incubation the degraded pellet and supernatant were separated and used as biofertilizer. Thus, pellet was used as solid fertilizer (cyanopith) and supernatant was called cyanospray for the plant growth solution.

Experimental design

Seeds of *Zea mays* L. (corn) were obtained from Tamil Nadu Agricultural University, Tamil Nadu and India. The seeds were surface sterilized in 5% sodium hypochlorite solution for 10 minutes to prevent fungal contamination, then washed thoroughly with deionized water and air - dried at an ambient temperature of 32 °C. Then, various types of solutions such as NaCl (20 mM) (Maria *et al.* 2010), concentrated Bijamrita (Subhash Palekar, 2006) and different concentration of cyanospray (0.1%, 0.2%, 0.3%, 0.4%, 0.5%) were prepared and applied to seed for treatments. Petri dishes (8cm diam) were washed with deionized water and lined with filter paper (Whatman No.1) for germination study. Each Petri dish received 20 seeds of *Zea mays* L. and 20ml of treatments solution, Treatments comprised of control (deionized water), NaCl (20 mM), different concentration of cyanospray (0.1%, 0.2%, 0.3%, 0.4%, 0.5%) and concentrated Bijamrita solution.

The effect of Cyanospray and Bijamrita in seed germination was determined by measuring the Number of roots and length of Epocotyl and hypocotyl were measured through the use of scale meter and the Biochemical contents like protein (Lowry *et al.* 1951) total soluble sugar (Yemm and Willis, 1954) were estimated. Data were analyzed following analyses of variance (Anova) technique and mean separations were adjusted by the multiple comparison test. Means were compared by using Fisher's LSD test at 5% level of significance.

Result

The result of various treatments of NaCl, bijamrita and cyanospray showed significant effect on germination percent of *Zea mays* L. seed (Table.I). The maximum percent of germination was observed in 0.3% cyanospray treatment. The treatments of NaCl, cyanospray and bijamrita showed significant variations in the percent seed germinations when compared to the control.

The longest epicotyls length was recorded at 0.3% of cyanospray solution treatment when compared to untreated seeds (Fig - 1). The seed treated with 0.3% of cyanospray was maximum in epicotyl length while the control is the least (Fig - 2). The hypocotyls length was not much influenced by various concentrations of cyanospray (0.1%, 0.2%, 0.3%, 0.4%, 0.5%), NaCl (20 mM), Bijamrita. The hypocotyls length were significantly increased ($p = 0.05$) in 0.3% cyanospray treatment when compared to control treatment.

The numbers of roots were less in the control treatment. The maximum number of roots was observed in 0.3% of cyanospray treatment (Fig - 3) which was significantly different from other treatments.

There were marked differences ($P = 0.05$) in soluble sugar content of seeds which was allowed to grow under different concentration of cyanospray (0.1%, 0.2%, 0.3%, 0.4%, 0.5%), NaCl (20 mM), bijamrita treatment compared to control (Fig – 4). The maximum soluble sugar content was recorded in 0.3% of cyanospray treated seeds after 72 hours of incubation when compared to control and all other treatments.

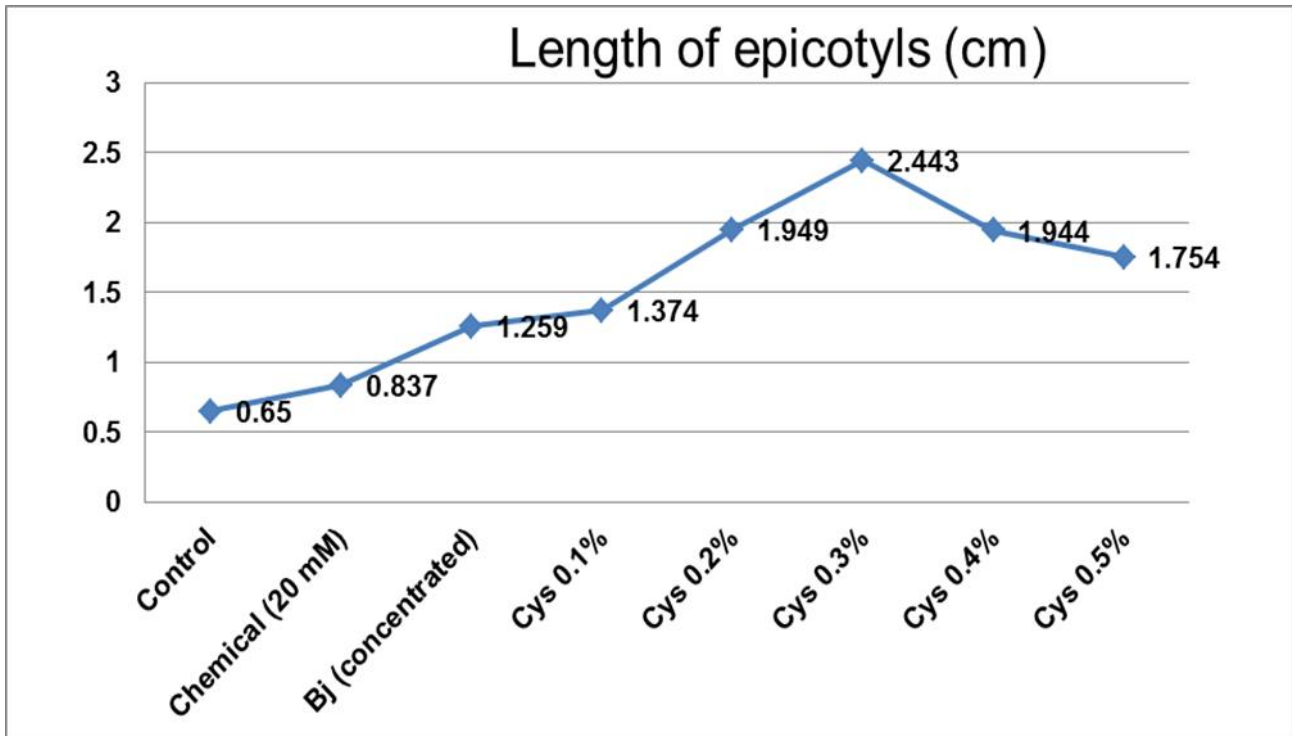
As germination progressed, the amount of protein stored in the endosperm gradually diminished in the control and in all other treatments (Fig – 5). The protein content was significantly ($p = 0.05$) less in 0.3% of cyanospray compared to the control.

Table I: Effect of NaCl, Bijamrita and Cyanospray solution on % of Seed germination of *Zea mays* L.

% of Seed germination									
Hours	0	12	24	36	48	60	72	84	96
Control	0	0	0	30	60	60	65	70	75
Chemical (NaCl)	0	0	5	55	75	85	90	90	90
Bj(Con.)	0	0	0	30	60	65	75	75	75
Cys - 0.1%	0	0	0	35	60	60	75	85	90
Cys - 0.2%	0	0	0	40	65	75	85	85	90
Cys - 0.3%	0	0	10	65	85	95	<u>100</u>	<u>100</u>	<u>100</u>
Cys - 0.4%	0	0	5	40	75	85	90	95	<u>100</u>
Cys - 0.5%	0	0	0	40	70	75	75	80	85

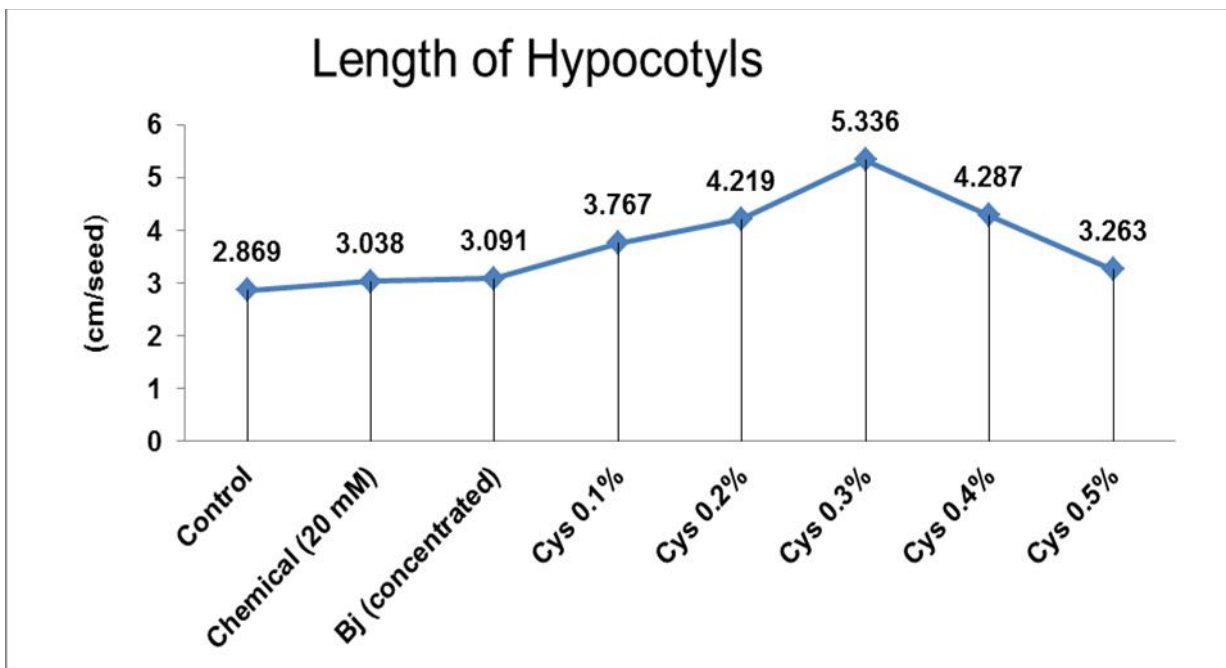
Cys- Cyanospray, Bj- Bijamrita

Fig 1. Effect of NaCl, Bijamrita and Cyanospray solution on Morphological parameters of *Zea mays* L.



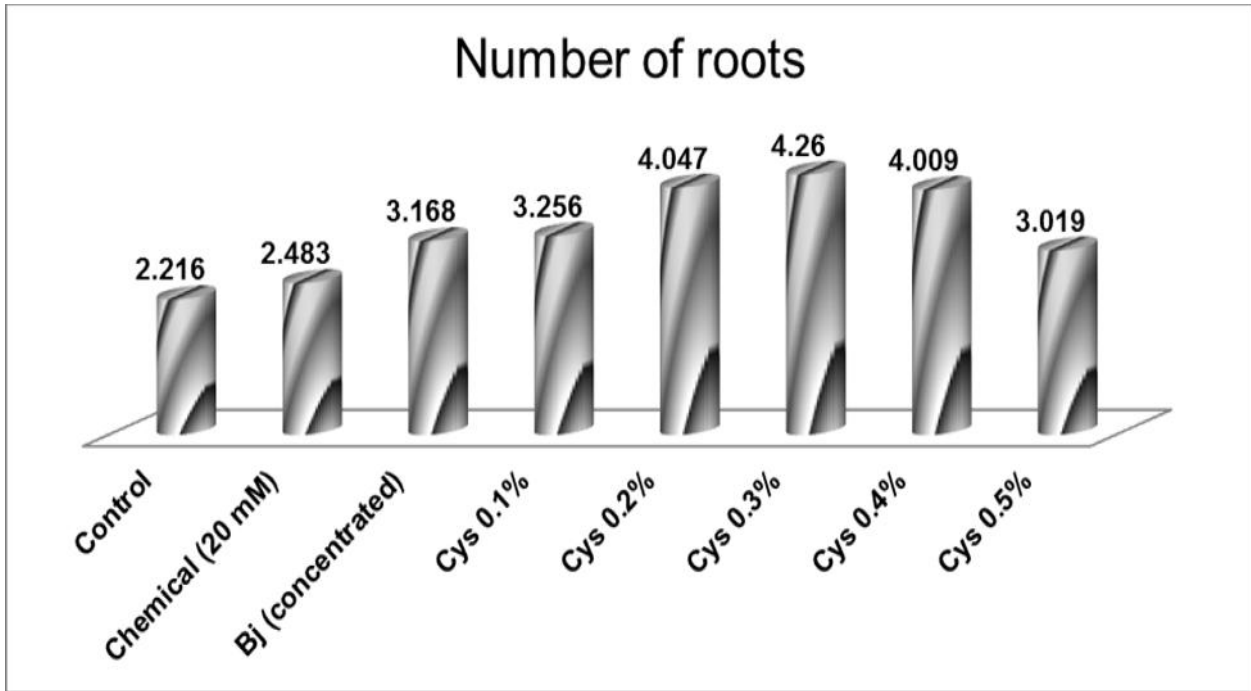
Cys- Cyanospray, Bj- Bijamrita, Chemical - NaCl

Fig 2. Effect of NaCl, Bijamrita and Cyanospray solution on Morphological parameters of *Zea mays* L.



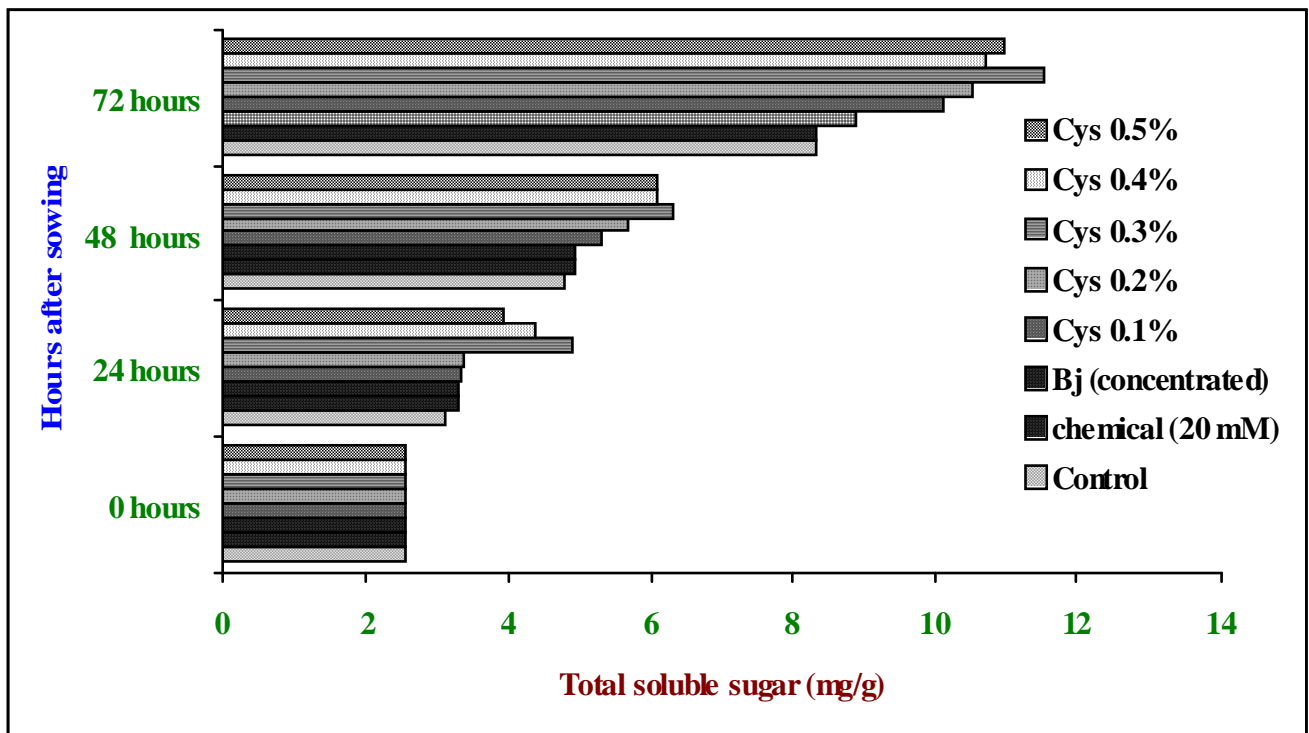
Cys- Cyanospray, Bj- Bijamrita, Chemical - NaCl

Fig 3. Effect of NaCl, Bijamrita and Cyanospray solution on Morphological parameters of *Zea mays* L.

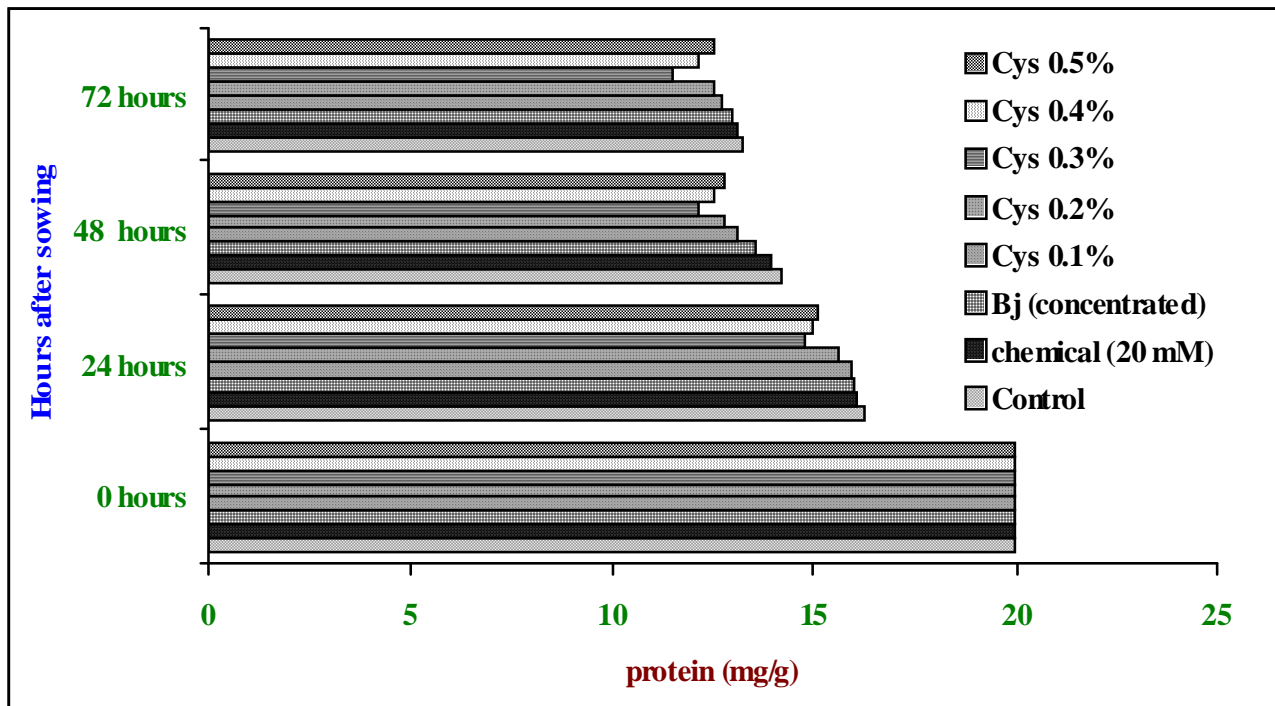


Cys- Cyanospray, Bj- Bijamrita, Chemical - NaCl

Fig 4. Effect of NaCl, Bijamrita and Cyanospray solution on Total soluble sugar content of *Zea mays* L.



Cys- Cyanospray, Bj- Bijamrita, Chemical - NaCl

Fig 5. Effect of NaCl, Bijamrita and Cyanospray solution on protein content of *Zea mays* L.

Cys- Cyanospray, Bj- Bijamrita, Chemical - NaCl

Discussion

The present results showed that the germination of *zea mays* seedling is influenced by the presents of cyanospray solution treatments. The results obtained from this investigation are similar to the reports of **Raghava and Murty (1988)** and their study revealed that lower concentration (100 and 200ppm) of IAA treatment increased the fresh and dry weights of *Physalin peruviana* and *P. angulata* while higher concentrations (500ppm) were slightly inhibitory. **Ravishankar, (2000)** showed the effect of *Phormidium valderianum* extract on germination of *Vigna mungo*- T9 variety and the extract was sprayed in three doses as foliar spray, during young vegetative stage (17th day), flowering stage (33rd day) and pod formation stage (41st day). Percentage of seed germination, number of leaves, length of shoot, root, and number of nodules, flowers, pod number, length, weight, seed number and weight showed better results when compared to untreated plants.

The production of growth promoting substances by cyanobacteria and their direct growth promoting effect on rice crop has been well elucidated by **Kannaiyan et al. (1997)**. Earlier study report indicated that in addition to the supply of N₂ fixing cyanobacteria could also release a variety of substances such as hormones, vitamins, amino acids, polysaccharides (**Kaushik, 1993**) and growth promoting substances (**Ravishankar, 2000; Kanagalatha, 2001**). These reports supported the present results on induction of epicotyl and hypocotyl growth by the cyanobacterial extract of strain no. 6. Thus, among

all 20 cyanobacterial strains only one strain showed maximum percentage of seed germination and also exhibited maximum growth of epicotyl and hypocotyl on three different plant seeds tested. This present study revealed that PGR activity was more in strain no.6 when compared to all other cyanobacteria strains. Hence, in this study the cyanospray treatment solution of 0.3% showed maximum result in percent germination and morphological results. Here the cyanospray might have improved the plant growth hormones such as IAA. It stimulates the germination power and growth. The stimulatory effect of GA3 on seed germination has been reported by many researchers (**Lang, 1965; Stokes, 1965**). GA3 application accelerated the hydrolysis of starch to soluble sugar by enhancing the hydrolytic enzymes such as α -amylase, β -amylase, maltase and invertase. A similar result was also observed by **Salla et al. (1991)** in rice. Decrease in the amount of protein during germination is explained by the fact that the protein degraded into soluble nitrogenous compounds through the action of proteolytic enzymes, which in turn were utilized by various parts of the seedling **Mayer and Mayber, (1982)**.

Conclusion

The germination study proved that 0.3% cyanospray-treated seeds showed the maximum germination percentage and increase in morphological parameters such as epicotyls length, hypocotyls length, and number of radical, and also increase in biochemical contents. Hence, it concluded that cyanospray 0.3% solution enhances the germination ability of *Zea mays*.

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