

Original Research Article

Influence Upshot of biopolymer synthesized from Tamarind Seed Polysaccharide (TSP) on seed physiological and biochemical parameters of hybrid maize var. COH(M)8 seeds

ABSTRACT

The experiment was conducted in Department of Seed Science and Technology, Tamil Nadu Agricultural University, Coimbatore, Tamil Nadu during 2021. Biopolymer was synthesized from Tamarind Seed Polysaccharide (TSP) and added with different additives and coated the maize seeds to know how they affect on their physiological and biochemical parameters. The seeds were given with four treatments viz., T₀- control (untreated seed), T₁- biopolymer (B.P) @ 10g kg⁻¹, T₂ = B.P (10g) + Humic acid (0.3g) + Zimmu leaf extract (0.5 ml), and T₃ = T₂+ Ascorbic acid (0.2 g), and evaluated for seed quality parameters. The results of the present investigation revealed that T₃ was found to be significantly superior because the percentage of viz., higher germination percentage (95%), speed of germination (31.64), seed metabolic efficiency (2.78), seedling root length (25.92 cm), seedling shoot length (15.57 cm), total dry biomass matter production (9.41 g), vigour index I (3942), vigour Index-II (94.15). This treatment also recorded highest value of biochemical parameters such as α-amylase (2.23), dehydrogenase (1.87 OD value), catalase catalyses (29.94) and peroxidase (7.00) activities. It reduced the abnormal seedlings (2%), dead seeds (3%), days to 50% germination (2.54), mean emergence time (3.02) and pathogen infection (0.25%). We The results concluded that maize seeds coated with T₃ = T₂ + Ascorbic acid (0.2 g) had performed better seedling establishment and may be recommended as pre sowing seed treatment under organic agriculture.

Key words: Tamarind Seed Polysaccharide, Biopolymer, Humic acid, Zimmu leaf extract, Seed germination and Vigour

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1. INTRODUCTION

One of the most ~~important~~~~significant~~ cereals in the world is maize (*Zea mays* L.). It is used as a human food, animal feed and a raw ingredient in a variety of industrial products ~~all over the world~~. Maize is a high-yielding, ~~an~~ easily digestible crop ~~that is~~ widely utilized in confectioneries. It's a key ingredient in the manufacture of starch, oil, protein, alcoholic drinks, food sweeteners, and more recently in ~~the~~ bio-fuel industry. ~~It is known as the "Queen of Cereals" because of its versatility and magical properties.~~

Maize is a ~~plant~~ species ~~that can~~ adapted to a wide range of environmental ~~circumstances~~~~conditions~~, but pest and disease outbreaks can ~~lead to~~ lower crop yields and quality, ~~or both~~. Moreover, seed ~~q~~uality ~~seed~~ is essential for success ~~in~~ agriculture, ~~because~~ as each seed must germinate ~~promptly~~~~quickly~~ and ~~develop~~erate a healthy seedling, ~~and profitable resulting in higher crop~~ yields. [1]. ~~Seed coating~~ Seed coating technology has advanced ~~rapidly~~~~quickly~~ over the last three decades, and it ~~now~~ offers a cost-effective method of seed improvement. ~~Seed coating~~ is the act of applying a beneficial material directly on a seed to generate a thin, homogeneous coating without changing the seed's shape or size. ~~Seed coating~~ has presented promising results in many crops including cereals. ~~Seed coating~~ with synthetic polymers has gained rapid acceptance by the seed industry. ~~It~~ makes room for including all the required ingredients like inoculants, protectants, nutrients, plant growth promoters, hydrophobic / hydrophilic substances, herbicides, oxygen suppliers etc.

~~Seed coating~~ with polymer enhances chemical adhesion to the seed and allows for dust-free handling of treated seed [2]. The polymer coating is easy to apply, ~~quickly~~ diffuses ~~promptly~~, and is non-toxic to ~~the~~ seed ~~during~~ germination. The polymer coating may operate as a physical barrier, preventing inhibitors leaching from seed coverings and restricting oxygen transport to the embryo [3]. Thereby, it provides protection from the stress imposed by ageing, improves plant stand and emergence of seedlings. Polymer acts ~~as a temperature switch~~ and protective coating by regulating the water uptake and subsequent germination of seed [4]. Coating results in more uniform and accurate seed ~~sowing~~ rate ~~because of~~~~due to~~ the smooth flow of the seed during ~~the~~ mechanical ~~procedures~~~~sowing~~. Increase in germination can also be observed in polymer coated seed. Addition of colourants helps in visual monitoring of placement accuracy, enhance the appearance, marketability and consumer preference.

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~~Despite~~Though the polymer coating has large number of benefits on seed quality parameters and agriculture, the continuous use of synthetic polymer and synthetic colourants may degrade the soil quality and ultimately reduces the crop yield. Because most of the seed coating polymers are synthetic and very slowly degradable in nature. Continuous using of polymers may leads to its accumulation of polymer in the soil profile and they may causes negative effect on the soil microorganisms and soil healthy. Eventually it-they may impacts the crop growth and yield, and may pollute the water bodies and environment conditions. Considering this issue, we synthesized the biopolymer from tamarind seed polysaccharide and studied its influenceeffect on seed physiological and biochemical characteristics in one hybrid of maize.

2. MATERIALS AND METHODS

We conducted a The Laboratory experiment was conducted to study "The Effect of biopolymer coating on the seed quality parameters of the hHybrid mMaize COH(M)8 (*Zea mays* L.) (COH(M)8), at the Department of Seed Science and Technology, Tamil Nadu Agricultural University, Coimbatore, Tamil Nadu during 2021. The treatments used were T₀= Control (untreated seed), T₁= biopolymer (B.P) @ 10g kg⁻¹, T₂= B.P (10g) + Humic acid (0.3g) + Zimmu leaf extract (0.5 ml), and T₃= B.P (10g) + Humic acid (0.3g) + Zimmu leaf extract (0.5 ml) + ascorbic acid (0.2 g).

Observations

2.1. Seed physical characters

2.1.1. One hundred400 seed weight

Seed weight was estimated by weighing 100 seeds from eight replication and the mean values were expressed in gram.

2.1.2. Seed moisture content

Moisture estimation was carried out by high constant temperature method based on the ~~per~~ ISTA protocol [5].

2.2. Seed physiological characters

2.2.1. Percentage of sSeed germination (%)

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The germination test was carried out ~~by using as per~~ the procedure prescribed by ISTA [5] in roll towel ~~paper~~ method. The test conditions were 25±2°C and 95±5% RH maintained in a germination room illuminated with fluorescent light. After seven days, the number of normal seedlings ~~produced~~ was counted and germination percentage (GP) was calculated, according to the formula:

$$\text{Germination percentage (GP)} = (\text{Ng} / \text{Nt}) \times 100$$

Where Ng is a total number of normal seedlings germinated, Nt is a total number of seeds ~~evaluated~~.

2.2.2. Days ~~to for fifty~~ 50 per cent ~~of~~ germination and maximum germination

In ~~the~~ sand media, the number seeds germinated was recorded daily up to final count and number of days required ~~to for~~ 50 per cent germination and number of days required ~~to for~~ maximum germination was computed according to Heydecker & Coolbear [6] and Mauromicale & Cavallaro [7] respectively.

2.2.3. ~~Rate~~ Speed of germination

Numbers of seeds germinated were counted daily up to seven days at the same time ~~of day~~. From the number of seeds germinated on each counting day, the ~~speed~~ of germination was computed adopting the formula given by Maguire [8].

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Comment [JW21]: I recommend the terminology rate because the physical dimension of rate is 1/T

2.2.4. Mean germination time

Mean germination time (MGT) was calculated according to Bailly *et al.* [9] using the formula:

$$\text{MGT} = \sum (\text{Dn}) / \sum n$$

Where, n is the number of seeds germinated on each day and D is the day of counting

2.2.5. Endosperm and embryo degradation (Seed Metabolic Efficiency)

Amount of seed respired (SMR) was calculated as

$$\text{SMR} = \text{SDW} - (\text{SHW} + \text{RTW} + \text{RSW})$$

Comment [JW22]: The Acronym does not fit.

Where,

SDW - Seed dry ~~biomass weight~~ before germination

SHW - Shoot dry ~~biomass weight~~

RTW - Root dry weight

RSW - Remaining seed dry weight

Seed Metabolic Efficiency (SME) was calculated using the formula [10]

$$\text{SME} = (\text{SHW} + \text{RTW}) / \text{SMR}$$

Comment [JW23]: Is there reference for these measurements?

2.2.6. Root length and shoot length (cm)

Ten normal seedlings from the standard germination test were randomly selected and the root and shoot length was measured from the collar region to the tip of the primary root and tip of the shoot respectively. The average value was expressed in centimeters.

2.2.7. Dry biomass production (g seedlings⁻¹⁰) and total dry biomass production (g) [5]

The seedlings used for growth measurement and the remaining normal seedlings were placed in a butter paper cover separately and dried under shade for 24 h and then kept in an oven maintained at $85 \pm 2^\circ\text{C}$ for 24 h. Dry biomass weight was recorded and the mean values were expressed in g. The total dry biomass production was calculated by adding dry weight of ten seedlings and remaining normal seedlings.

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2.2.8. Vigour index [11]

Vigour index values were computed using the following formula and the mean values were expressed in whole number.

$$\text{Vigour index I} = \text{Germination (\%)} \times \text{Total seedling length (cm)}$$

$$\text{Vigour index II} = \text{Germination (\%)} \times \text{Dry matter production (g seedling}^{-10}\text{)}$$

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2.3. Biochemical parameters

2.3.1. Dehydrogenase activity (OD value)

Dehydrogenase activity was estimated by the procedure described by Kittock and Law [12].

2.3.2. α -amylase activity (mg maltose liberated / minute)

α -amylase activity in pre germinated seeds were carried out according to the method suggested by Simpson and Naylor [13].

2.3.3. Peroxidase activity (Δ A436 / min/ g of seed)

Peroxidase activity was estimated as per the procedure described by Singh *et al.* [14].

2.3.4. Catalase activity (mMol H₂O₂ degraded / minute)

Catalase activity was measured by an assay of hydrogen peroxide based on formation of its stable complex with ammonium molybdate [15].

2.4. Seed health

2.4.1. Pathogen infection (%)

Pathogen infection was assessed by the protocol given by ISTA [5].

Data analysis:

Please write here the model of data analysis with the software applied to these data set as well as the test to discriminate the treatments. Remember, the goal is to have the experiment replicated as possible. Test to discriminate the treatments and probability level.

3. RESULTS AND DISCUSSION

The result revealed that, seeds coated with bio-polymer (B.P) and B.P with additives shows non-significant difference for 100 seed weight and moisture content (Table 1). Polymer coating forms a very thin layer around the seeds without obscuring size and shape, hence it did not altered the seed weight. After seed coating, the seeds were dried under shade for one hour, hence the method it did not change the moisture content significantly in the coated-seeds.

According to the results, all the studied traits were affected by the treatments and there was completely significant difference observed among the treatments. Seed physiological characters viz., germination percent (95%), speed of germination (31.64), seed metabolic efficiency (2.78), seedling root length (25.92 cm), seedling shoot length (15.57 cm), drymatter production/10 seedlings (0.991 g), total drymatter production (9.41 g), vigour index-I (3942), and vigor index-II (94.15) were significantly recorded maximum in T₃ whereas found lowest in control (Table 1,2 and Fig. 1,2). It significantly reduced days for 50% germination (2.54), mean emergence time (3.02), abnormal seedling (2%) and dead seeds (3%) compare to other treatments (Table 1).

The improvement in seed physiological parameters in T₃ may be due to additives such as humic acid and ascorbic acid present in the polymer formulation. Humic acid is one of bio-stimulants that are known as the organic substances which promote plant growth [16]. Humic acid improves the nutrient availability especially microelements in soils because it promotes nutrient uptake in the form of chelating agent. Moreover, humic substances may increase root growth in a similar manner to auxins [17, 18]. The present results similar to the findings of Asgharipour and Rafiei [19] and Basalma [20] who reported that seed treated with HA recorded maximum germination, seedling length, seedling fresh weight, seedling dry weight and vigour index in barley and safflower respectively. Likewise, HA seed treatment increased the shoot fresh and dry weight of seedlings in tomato [21], maize [22], pea [23], wheat [24], cucumber, squash and marigold [25].

The activity mechanism of humic acid in promoting plant growth is not fully understood, and the beneficial effects to plants are difficult to comprehend due to its chemical heterogeneity [26,27]. The most established explanations for the beneficial effects of HA are related to their positive influence on ion transport, which improves cell permeability, thereby affects absorption. They also promote increased

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respiration and speed of enzymatic reactions of the Krebs cycle, resulting in increased ATP production, altering directly plant metabolism and consequently may influence growth and development [28,29]. The increase in absorption rates can be explained by the activation of ATPase present in the plasma membrane [30], acting on two mechanisms essential to plant growth, through the energy supply to the secondary systems in the translocation of ions and by increasing plasticity of the cell wall, thus allowing cell growth and division [31].

The reason for improved seed physiological parameters in T₃ may also be due to ascorbic acid present in the polymer formulation. Ascorbic acid (AsA), also known as ascorbate or vitamin C, is a low molecular weight water-soluble antioxidant both in plants and animals. And AsA is a universal non-enzymatic antioxidant having a substantial potential of not only scavenging reactive oxygen species (ROS), but also modulating many fundamental functions in plants both under stress and nonstress conditions [32,33,34]. Burguieres *et al.* [35] and Chen *et al.* [36] reported that seeds treated with AsA increased the germination, seedling length, fresh weight and dry weight in pea and alfalfa respectively.

Comment [JW33]: Other ...

Table 1. Effect of biopolymer and additives on stand establishment traits of maize

Comment [JW34]: I suggest the Anova Table as the first Figure in the Results section.

Treatments	100 seed weight (g)	Seed moisture content (%)	Abnormal seedling (%)	Dead seeds (%)	Days for 50 per cent germination	Speed of germination	Mean emergence time
T ₀	31.27	10.22	5	5	3.16	27.31	3.37
T ₁	31.44	10.24	5	5	3.14	27.66	3.34
T ₂	31.43	10.23	2	5	2.62	28.42	3.08
T ₃	31.44	10.22	2	3	2.54	31.64	3.02
Mean	31.40	10.23	4	5	2.87	28.76	3.20
SEd	0.26	0.06	0.03	0.06	0.04	0.40	0.03
CD (P=0.05)	0.55	0.13	0.06	0.12	0.08	0.85	0.06

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Comment [JW35]: See the suggest above.

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Table 2. Effect of biopolymer and additives on seedling vigour parameters of maize

Treatments	Seed metabolic efficiency	Shoot length (cm)	Root length (cm)	Dry matter production (g/10 seedlings)	Total dry matter production (g)	Vigour index II
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T ₀	2.24	12.85	23.48	0.908	8.17	81.72
T ₁	2.26	13.14	23.85	0.912	8.21	82.08
T ₂	2.65	15.14	25.65	0.956	8.89	88.91
T ₃	2.78	15.57	25.92	0.991	9.41	94.15
Mean	2.48	14.18	24.73	0.942	8.67	86.72
SEd	0.02	0.13	0.39	0.01	0.06	1.03
CD (P=0.05)	0.04	0.28	0.83	0.02	0.13	2.18



T₀



T₃

Fig. 1. Effect of biopolymer with additives on seed germination and seedling growth of maize

Comment [JW38]: This Figure is difficult of interpretation.

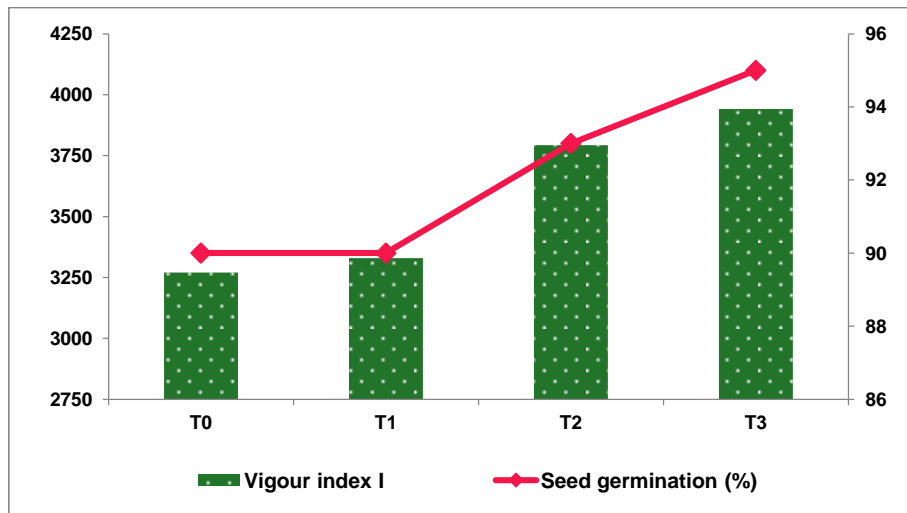


Fig. 2. Effect of biopolymer with additives on seed germination and seedling vigour index I of hybrid maize

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Among the treatments T₃ recorded significantly higher levels of enzymatic activities viz., α-amylase (2.23 mg maltose/min), dehydrogenase (1.87 OD value), catalase (29.94 mMol H₂O₂ / min) and peroxidase (7.00 Δ A436 /min/g) compared to other treatments (Table 3). The result similar to the findings of Burguieres *et al.* [35] who stated that seeds treated with AsA increased the guaiacol peroxidase (GPX), superoxide dismutase (SOD) and catalase (CAT) activities in pea. Similarly, Chen *et al.* [36] reported that seeds treated with AsA increased the α-amylase and protease activities in alfalfa seeds.

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The results shows that the seeds treated with T₂ and T₃ significantly reduced the pathogen infection (0.25%) than compared to T₀ and T₁ (1.25 and 1.50% respectively) (Table 3). This may be the reason for reduced dead seed percentage in T₂ and T₃. Reduced pathogen infection is due to added antimicrobial agent (zimmu leaf extract) in the polymer. Satya *et al.* [37] found that the leaf extract of zimmu showed the maximum antifungal activity against *Rhizoctonia solani* and it also effective in inhibiting the growth of other fungal and bacterial pathogens viz., *Aspergillus flavus*, *Alternaria solani*, *Curvularia lunata*, *Xanthomonas campestris* pv. *Malvacearum*, *X. oryzae* pv. *oryzae*, and *X. oxonopodis* pv. *citri*. Thus the study indicates that the physiological and biochemical improvement in T₃ is due to synergetic effect of humic acid and ascorbic acid present in the polymer.

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Table 3. Effect of biopolymer and additives on biochemical parameters and pathogen infection of maize

Treatments	α -amylase activity (mg maltose min ⁻¹)	Dehydrogenase activity (OD value)	Catalase activity (μ g H ₂ O ₂ /min/mg protein)	Peroxidase activity (Δ OD 430 mg ⁻¹ min ⁻¹)	Pathogen infection (%)
T ₀	2.17	1.76	28.16	6.58	1.25
T ₁	2.17	1.78	28.18	6.59	1.50
T ₂	2.18	1.80	28.53	6.59	0.25
T ₃	2.23	1.87	29.94	7.00	0.25
Mean	2.19	1.80	28.70	6.69	0.81
SEd	0.03	0.02	0.17	0.05	0.009
CD (P=0.05)	0.07	0.03	0.37	0.12	0.020

4. CONCLUSION

The Biopolymer and additives had a significant effect on the physiological and biochemical seed performance quality traits in this hybrid maize. T₃ improved the stand establishment such as seed germination, speed of germination, seed metabolic efficiency, shoot length, root length, drymatter production, vigour index I and vigour index II compared to other treatments. It also increased the dehydrogenase, α - amylase, catalase and peroxidase activities. It reduced the days to 50 % germination, mean germination time, abnormal seedlings, dead seeds and pathogen infection compared to control. Thus, the study highlighted that seeds coated with B.P (10g) + Humic acid (0.3g) + Zimmu leaf extract (0.5 ml) + ascorbic acid (0.2 g) improved the seed performance quality characteristics in this hybrid maize.

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Comment [JW45]: The first affirmative requires investigation on seed lots and more hybrids genotypes.

Comment [JW46]:

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