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PHYSIOCHEMICAL, BIOCHEMICAL AND PHYTOCHEMICAL PROFILING OF SODIUM ALGINATE AND ITS ROLE AS A SEED COATING POLYMER TO ENHANCING SEED QUALITY IN OKRA (*Abelmoschus esculentus* L.)

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ABSTRACT
Sodium alginate, a naturally occurring polysaccharide extracted from brown marine algae, has emerged as a promising biopolymer for sustainable agricultural applications. This study investigated the extraction, characterization, and functional evaluation of sodium alginate derived from Sargassum myriocystum and assessed its potential as a biodegradable seed-coating polymer for improving seed quality traits in okra (*Abelmoschus esculentus* L.). The extraction yielded 25% sodium alginate. Physicochemical characterization revealed medium viscosity (90 cP), an alkaline pH (7.20), a blackish-brown colour, and high water-absorption capacity. Biochemical analysis indicated a high carbohydrate content (70%) with negligible protein and lipid fractions, while phytochemical screening detected only saponins, indicating high purity. ICP–MS analysis revealed a high sodium content (200,000 ppm), confirming the sodium alginate form, and FTIR analysis verified the structural integrity and purity of the extracted alginate through characteristic O–H and C=O absorption bands with high transmittance (>95%). The extracted alginate was infused with selected biomolecules and applied as a seed-coating polymer. Among the treatments, T_{14} exhibited the best performance, recording 96% germination, 40.58 cm seedling growth, 0.463 g dry matter production per 10 seedlings, higher vigour indices I and II, and only 2% abnormal seedlings. In contrast, the uncoated control (T_0) showed the lowest germination (86%), seedling growth (30.34 cm), dry matter production (0.343 g per 10 seedlings), vigour indices, and the highest proportion of abnormal seedlings (9%). These findings demonstrate that sodium alginate derived from S. *myriocystum* is an effective, eco-friendly alternative to synthetic polymer and microplastic-based seed coatings. As a natural hydrocolloid, it significantly enhances seed quality attributes and holds considerable promise for sustainable and environmentally responsible agricultural practices.
KEYWORDS: Okra, sodium alginate, seed coating, biopolymer, biomolecules, organic agriculture

1. INTRODUCTION
Okra (*Abelmoschu sesculentus* L.), also known as bhendi or lady's finger, is a widely cultivated vegetable across tropical, subtropical, and warm temperate regions. It serves as both an important food source and a source of income for smallholder farmers. The crop's rich composition of fiber, vitamins, minerals, and antioxidants contributes significantly to nutrition and food security (Gemede *et al.*, 2014). In recent years, the industrial demand for okra has increased, primarily due to the valuable mucilage extracted from its pods, which is used in food and pharmaceutical applications (Leng, 2018). Despite its economic and nutritional importance, okra productivity is often constrained by factors such as the limited use of improved varieties, poor seed management, low availability of high-quality seeds, and the presence of hard seeds that hinder uniform germination (Demir, 1997). Moreover, high temperature and humidity conditions during storage adversely affect seed viability and field performance. The relatively high oil content in okra seeds (20-40%) further accelerates lipid peroxidation, damaging cell membranes and resulting in slow or uneven germination (Jarret *et al.*, 2011). The low adoption of high-yielding varieties also reduces the implementation of improved cultivation practices

(Abayneh, 2019). To overcome these challenges, seed invigoration and coating technologies have been developed to enhance germination, emergence, and early plant vigor. Conventional polymer-based coatings improve seed handling, ensure uniform pesticide or nutrient delivery, and facilitate better adhesion of treatment chemicals to seed surfaces (Ekebafe *et al.,* 2011). Such coatings form a thin, uniform layer that that create micro environment around the seed and diffuses easily and is non-toxic to seeds. They act as physical barriers, reducing the leaching of inhibitory substances from seed coats and limiting excess oxygen diffusion to the embryo (Kumar *et al.,* 2014). This controlled microenvironment around the seed improves stress tolerance, delays ageing, and enhances seedling establishment. Additionally, polymers act as temperature-sensitive coatings that regulate water uptake during imbibition, thereby improving germination uniformity under fluctuating environmental conditions (Johnson *et al.,* 1999). However, growing awareness of the environmental impacts associated with synthetic polymers particularly their persistence in soil, disruption of microbial diversity (Mustapha *et al.,* 2025), and contribution to agricultural microplastic pollution (Accinelli *et al.,* 2019) has prompted a shift toward eco-friendly alternatives. Among the biodegradable options, natural polymers such as sodium alginate, derived from brown seaweeds, have gained significant attention. Sodium alginate is a non-toxic, renewable biopolymer composed of β-D-mannuronic and α-L-guluronic acid units, which confer excellent gel-forming ability, adjustable viscosity, and high water-retention capacity, making it particularly suitable for seed-coating applications (Ahmad *et al.,* 1994). Research has shown that alginate-based coatings can enhance germination rate, seedling vigor, and stress tolerance while minimizing ecological risks compared to synthetic coatings (Skrzypczak *et al.,* 2021). Brown seaweeds, especially *Sargassum* species, are rich sources of alginate, although extraction yield and quality vary depending on species, growth stage, and environmental conditions (Kalimuthu, 1980; Dobrincic *et al.,* 2020). In addition to alginate extraction, seaweeds have long been used as organic soil amendments and natural biostimulants to improve seed germination, plant growth, and resilience to environmental stresses (Bawej*aet al.,* 2019; Norrie and Keathley, 2005). Sodium alginate is particularly advantageous in seed-coating due to its high water-uptake capacity, which supports proper seed imbibition and promotes germination (McHugh, 1987). It is widely used in agriculture for coating seeds, fruits, and stem tips (Cisneros-Zevallos and Krochta, 2006). Although most research has focused on its application in synthetic seed production, limited studies have explored its direct effects on germination indices and seedling traits in various crops. For instance, sodium alginate concentrations of 2% and 3% in *Ferulago angulata* L. produced synthetic seeds with maximum germination (Sorbi and Moradi, 2018), while 3.5% sodium alginate improved germination percentage in synthetic seeds of Persian oak. (Fayzi, 2016). Similarly, plant-derived polymers in general offer advantages such as biodegradability, low cost, light weight, and wide availability, further supporting their use in sustainable agricultural practices (Kamath and Park, 1993). When enriched with bioactive substances like humic and fulvic acids, these coatings can stimulate root growth, enhance nutrient uptake, and improve seedling establishment (Chen *et al.,* 2021). Humic substances are commonly applied to mitigate the harmful impacts of chemical fertilizers on soils, and numerous researchers have demonstrated their significant positive influence on plant growth (Ghabbourand Davies, 2001). Fulvic acids, in particular, are effective due to their small molecular size and chelating ability, enabling them to penetrate plant cell membranes and enhance nutrient transport (Bocanegra *et al.,* 2006; Yu *et al.,* 2023). Antioxidants like ascorbic improves the seed physiological performance by reducing oxidative stress during germination (Sivasakthi and Renganayaki 2022). Moreover, the incorporation of plant growth regulators like brassinosteroids (BRs) further strengthens the benefits of biopolymer coatings. BRs interact synergistically with auxins to promote root growth, increase germination rate, and enhance biomass accumulation in both roots and shoots (Zhang *et al.,* 2008; Pravina and Renganayaki 2025). They also regulate gene expression, enzyme activity, and nitrogen fixation, contributing to improved yield potential (Khan *et al.,* 2015). Similarly, salicylic acid (SA) functions as an important signaling molecule that improves water potential, maintains pigment stability, and confers tolerance to abiotic stresses such as drought and salinity (Raskin, 1992; Hayat *et al.,* 2010; Bakry *et al.,* 2012). Glutathione and ascorbic acid are key antioxidants that support seed germination by managing reactive oxygen species (ROS) and maintaining cellular redox balance. During the early stages of germination, glutathione (GSH) typically rises temporarily, contributing to dormancy release and modulating abscisic acid (ABA) signalling both essential for successful germination and seedling establishment. Higher GSH levels are also linked with stronger antioxidant protection and better stress tolerance during this phase (Koramutla *et al.,* 2021).Ascorbic acid is a widespread non-enzymatic antioxidant that not only neutralizes reactive oxygen species (ROS) but also influences several essential physiological processes in plants (Akram *et al.,* 2017).Overall, seed coatings enriched with bioactive compounds, antioxidants, and plant growth regulators can create a beneficial microenvironment around seeds, promoting rapid germination, uniform emergence, and improved seedling establishment. The integration of biodegradable polymers like sodium alginate with bioactive additives represents a sustainable approach to seed enhancement, ensuring high physiological performance while safeguarding soil health and ecological balance (Skrzypczak *et al.,* 2021).The objectives of the present study were:i)To extract and characterize sodium alginate from *Sargassum myriocystum*, examining its physicochemical, biochemical, and phytochemical properties and ii) to evaluate its efficacy as a biodegradable seed-coating polymer, supplemented with biostimulants, growth regulators, and antioxidants, for improving germination, vigor and seedling establishment in okra. By combining seaweed-derived alginate with functional additives, this study aims to develop sustainable seed-coating approaches that enhance productivity while minimizing environmental impact.

2. MATERIALS AND METHODS

2.1 Collection and authentication

The Mandapam coastal region, where the *Sargassum myriocystum* was collected, is located at approximately 9.2886° N latitude and 79.1329° E longitude. This area is part of the Gulf of Mannar, a coastal region known for its rich seaweed diversity. The freshly harvested seeds of Okra var. Arka Anamika (Latitude: 10.12° N or 10° 07' 13" N and Longitude: 77.55° E or 77° 32' 50" E) were procured from Horticultural college and Research Institute, Periyakulam, Theni district. The variety was developed by the Indian Institute of Horticultural Research (IIHR), Bangalore and the laboratory experiment was conducted at the Department of Seed Science and Technology, Tamil Nadu Agricultural University, Madurai, Tamil Nadu, India during 2024-2025.

2.2 Design of the experiment

With four replications, the experiment was done in a completely randomized block design and statistical tool is ANOVA

2.3 Extraction of sodium alginate

Sodium alginate is extracted according to Calumpong *et al.,* 1999 with some modifications. 25 g of dried seaweed (*Sargassum myriocystum*) powder was dissolved in 800 ml of 2 % formaldehyde for 24 hours to remove pigmentation. After such period supernant was decanted and seaweed was collected and washed twice by distilled water. Then seaweed was dissolved in 800 ml of 0.2 M HCl and leaved it for 24 hours to convert alginate into alginic acid, after such period again supernant was discarded and seaweed was collected, then washed by distilled water to remove acid. Collected seaweed was dissolved in 2 % Sodium carbonate solution and heated under water bath for 3 hours at 90-100^{0}c to dissolve the alginic acid into soluble sodium alginate. After that the solution was cooled and centrifuged under 5000 rpm for 30 min. Then supernant was collected carefully and added three volume of ethanol and it precipitate the sodium alginate. It was purified twice with100 ml of acetone and dried @ 65^{0}C. Dried sodium alginate powder was dissolved in distilled water and again precipitate with three volume of ethanol and dried @ 65^{0}C and grind it as fine powder and used for further studies.

2.4. Sodium alginate characteristics

2.4.1 Physiochemical analysis of sodium alginate

2.4.1.1 Yield (g)

Extracted sodium alginate yield was calculated as follows: Weight of alginate/ Weight of seaweed dried biomass *100

2.4.1.2 Estimation of pH and color

About one per cent w/v of sodium alginate polysaccharide powder was prepared in distilled water and the pH of the solution was measured using pH meter. The color was evaluated by visual inspection

2.4.1.3 Moisture content

The moisture of the alginates was determined according to AOAC (1995) through the loss in mass of sodium alginate after heating at 105^{0}C for 24 h.

2.4.1.4 Viscosity

One gram of dried sodium alginate was dissolved in 100 ml of distilled water and the viscosity was measured by using Brookfield viscometer.

2.4.1.5 Water absorbancy

To 25 ml of distilled water 0.5 g of sodium alginate powder was immersed and kept for 24 hours under room condition to reach absorption equilibrium. After specified time, the unabsorbed water was decanted and the swollen sodium alginate was weighed. The relative water absorbency was calculated (Chen *et al.,* 2004),

Water absorbency (g/g) = $(M_2 - M_1) / M_1$

Where,

M_1 – initial weight of sodium alginate

M_2 – final weight of swollen sodium alginate after 24 hrs

2.4.1.5 Solubility behaviour

As per Sivasakthi (2022) the solubility behaviour of the polysaccharide was tested with different inorganic and organic solvents *viz.,* warm water, cold water, ethanol (99.9%), methanol (99.85%), acetone (99.5%) and petroleum ether (99.0%). One gram of sample was taken and added to 25 ml of solvent and stirred with the help of glass rod for 1 min and then solubility behaviour was observed and recorded.

2.4.2 Biochemical analysis of sodium alginate

2.4.2.1 Carbohydrate analysis

The carbohydrate content of the sodium alginate was determined by the phenol sulphuric acid method described by Dubois *et al.,* (1956)

2.4.2.2 Protein analysis

The proteins were tested following Lowry *et al.,* (1951). The protein was calculated using bovine albumin serum as the standard.

2.4.2.3 Lipid's analysis

According to the AOAC methodologies (1950) the total lipids were estimated gravimetrically using chloroform-methanol

2.4.2.4 Ash analysis

The ash content was measured by a gravimetric method using AOAC (1950) by burning. 1 g of the product taken in a silica crucible and kept in a muffle furnace at 575^0C for 24 h. After burning, the content was cooled, weighed, and expressed in terms of percentage.

2.4.3 Phytochemical analysis of sodium alginate

To assess the purity of the extracted alginate, qualitative phytochemical screening was performed for compounds such as flavonoids, alkaloids, tannins, terpenoids, steroids, saponins, glycosides, phenols, and phlorotannins, following the methods of Trease and Evans (1989) and Sofowora (1993). These assays serve as rapid tests to detect the presence of residual secondary metabolites from seaweed, which, if present, indicate possible co-extraction of non-target compounds along with alginate.

2.4.3.1 Test for flavonoids

To determine the presence of flavonoids, a qualitative assay was carried out. For this, 0.5 g of sodium alginate was dissolved in diluted NaOH, followed by the addition of HCl. The formation of a yellow coloration that subsequently disappeared upon acidification indicated the presence of flavonoids.

2.4.3.2 Test for alkaloids

To test for the presence of alkaloids, 0.5 g of sodium alginate was dissolved in 10 ml of 0.1 N HCl and filtered. The filtrate was then used for qualitative analysis. For the assay, 1 ml of 1% HCl was added to 3 ml of the filtrate, followed by a few drops of Mayer's reagent. The formation of a creamy white precipitates confirms the presence of alkaloids.

2.4.3.3 Test for tannins

To analyze the presence of tannins, a qualitative method was used. 0.5 g of sodium alginate was boiled in 10 ml of distilled water and then filtered. A few drops of FeCl3 0.1% were added and observed for a blue-black or brownish green color, indicates the presence of tannins.

2.4.3.4 Test for terpenoids and Steroids

To analyze the presence of terpenoids and steroids, a qualitative method was used. Sodium alginate 0.5 g was added to 2 ml of chloroform. Then, 3 ml of conc. sulfuric acid was added carefully to a layer form. A reddish-brown color for the interface indicated terpenoids presence.

2.4.3.5 Test for saponins

To analyze the presence of saponins, a qualitative method was used. First, 0.5 g of sodium alginate was dissolved in 5 ml of distilled water. The solution was shaken speedly and observed for a stable persistent foam. The foaming was mixed with three drop of olive oil and shaken strongly and the formation of a milky mass was observed, which indicates the existence of saponins.

2.4.3.6 Test for glycosides

The analysis of the presence of glycosides was done with a qualitative method using0.5 g of sodium alginate dissolved in 5 ml of distilled water. Then, 10 ml of 50% sulphuric acid was added to 1 ml of this extract. The mixture was heated in boiling water for 5 min. Then, 10 ml of Fehling's solution (5 ml of each solution A and B) was added and boiled. A brick red precipitate indicated the presence of glycosides.

2.4.3.7 Test for phenolic compounds

To analyze the presence of phenolic compounds, a qualitative method was used. 100 mg of sodium alginate was boiled with 1 ml of distilled water then filtered. Then, 2 ml of filtrate was taken and 2 ml of 1% FeCl3 solution was added. The establishment of bluish black color indicated the existence of phenol.

2.4.3.8 Test for phlorotannins

To analyze the presence of phlorotannins, a qualitative method was used. Sodium alginate 0.5 g was dissolved in 5 ml of distilled water and filtered. Then the filtrate was boiled with 2% HCl solution. A red precipitate indicated the presence of phlorotannins.

2.4.4 ICP-MS analysis

One g of sodium alginate sample was induced into Inductively Coupled Plasma Mass Spectrometry (ICP-MS) quantifies trace elements by ionizing samples in Department of Forage crops, Tamil Nadu Agricultural University, Coimbatore

2.4.5 Identification of functional groups by Fourier Transforms Infra-Red (FTIR)

The functional groups of the isolated sodium alginatewere identified using an FTIR spectrophotometer (FT/IR-4600 Type A) equipped with an Attenuated Total Reflectance (ATR Pro One) sensor at the Central Instrumentation Centre, The American College, Madurai, Tamil Nadu, India. Spectral data were recorded in the range of 650-4000 cm^{-1}.

2.5 Preparation of seed coating polymer

Sodium alginate 2 g was taken and dissolves in 100 ml of distilled water (by magnetic strirrer) and 0.5 ml of glycerol was added to the polymer and different biostimulant, plant growth regulators and antioxidant were added at respective concentration to the polymer. For 1 kg of seed 250 ml of polymer is required for uniform seed coating.

Treatment details:

The Okra seeds var. Arka Anamika were coated with different treatments such as

T_0 - Control (Uncoated seed)

T_1 - Sodium alginate polymer (2%) 250 ml/kg of seed

T_2 - 0.2 g humic acid infused to 100 ml of sodium alginate polymer (2%)
T_3 - 0.3 g humic acid infused to 100 ml of sodium alginate polymer (2%)
T_4 - 0.4 g fulvic acid infused to 100 ml of sSodium alginate polymer (2%)
T_5 - 0.5 g fulvic acid infused to 100 ml of sodium alginate polymer (2%)
T_6 - Salicylic acid 200 ppm infused to 100 ml of sodium alginate polymer (2%)
T_7 - Salicylic acid 250 ppm infused to 100 ml of sodium alginate polymer (2%)
T_8 - Brassinolide 0.5 ppm infused to 100 ml of sodium alginate polymer (2%)
T_9 - Brassinolide 1.5 ppm infused to 100 ml of sodium alginate polymer (2%)
T_{10} - 0.05 g glutothione infused to 100 ml of sodium alginate polymer (2%)
T_{11} - 0.10 g glutothione infused to 100 ml of sodium alginate polymer (2%)
T_{12} - 0.2 g ascorbic acid infused to 100 ml of sodium alginate polymer (2%)
T_{13} - 0.4 g ascorbic acid infused to 100 ml of sodium alginate polymer (2%)
T_{14} - 0.3 g humic acid + 250 ppm salicylic acid + 0.4 g ascorbic acid infused to 100 ml of sodium alginate polymer (2%)

Seeds were coated with respective treatments and cross linked with 5% $CaCl_2$ for 20 min. After that seeds were dried at room temperature for 24 hours.

2.6 Observations on seed quality parameters of okra

2.6.1 Seed germination (%)

The germination test was carried out in roll towel method according to ISTA, 2013 protocol. After 21 days of germination, the seedlings were evaluated and the normal seedlings were counted and germination percentage (GP) was calculated, according to the formula:

Germination percentage (GP**)** = (Total number of normal seedlings germinated /Total number of seeds evaluated) × 100

2.6.2 Abnormal seedlings (%)

The germination test was carried out in roll towel method according to ISTA, 2013 protocol. After 21 days of germination, the seedlings were evaluated and the abnormal seedlings were counted and expressed in percentage Abnormal seedlings (%) = (Total number of abnormal seedlings / Total number of seeds evaluated) × 100

2.6.3 Root length (cm)

After the germination count, ten normal seedlings were randomly chosen from each replication and the length of the roots from the collar area to the tip of the primary root was measured.

2.6.4 Shoot length (cm)

Shoot length was measured on the same seedlings which were used for measuring the length of the roots. The distance along the shoot from the collar to the tip of the primary leaf was measured.

2.6.5 Seedling length (cm)

Seedling length generally refers to the total length of a young plant from the tip of the shoot to the tip of the root. It is usually measured in centimeters (cm) and is obtained by summing:

Seedling length=Shoot length (cm) + Root length (cm)

2.6.6 Dry matter production (g seedlings^{-10})

The seedlings which is used for seedling length measurement same seedlings were placed in a butter paper cover separately and dried in shade for 24 h and then kept in an oven maintained at 85 ± 2℃ for 24 h. Dry weight was recorded and the mean values were expressed in gram. (ISTA, 2013).

2.6.7 Vigour index (Abdul-Baki and Anderson, 1973)

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

Vigour index I= Germination (%) x Root length (cm) + Shoot length (cm)

Vigour index II = Germination (%) x Dry matter production (g seedling^{-10})

3. RESULTS AND DISCUSSION

3.1 Sodium alginate characteristics

3.1.1 Physiochemcial

The physicochemical characteristics of the sodium alginate are presented in Table 1. Sodium alginate extracted from *Sargassum myriocystum* yielded 25%**,** demonstrating efficient recovery of the polysaccharide from the brown seaweed. This yield falls within the range of 14.26–26.07% previously reported for *S. myriocystum* by Kalimuthu (1980), indicating that the extraction procedure employed in the present study was effective. Similar yields have also been reported in other Sargassum species (Dalal *et al.*, 2021). Variations in alginate yield are mainly influenced by the seaweed species, *maturity*, seasonal growth stage, environmental conditions such as light intensity, water temperature, nutrient availability, and extraction methodology (Dobrincic *et al.*, 2020). Therefore, the yield obtained in the present investigation confirms that S. myriocystum is a promising and sustainable source of sodium alginate for agricultural and industrial applications. The extracted sodium alginate exhibited a blackish-brown colour (Fig 1) with a pH of 7.20**,** indicating its near-neutral nature. According to Coppen (1995), these characteristics are consistent with acceptable quality standards for sodium alginate. The darker colour observed in the present study may be attributed to the presence of residual natural pigments and

saponins retained during extraction, whereas Rashedy *et al.,* (2021) reported comparatively lighter alginate preparations obtained using different purification procedures. Although colour variation is expected depending on the extraction process, it does not adversely affect the functional properties of sodium alginate intended for non-food applications such as seed coating. The viscosity of the 1% (w/v) sodium alginate solution was 90 cP**,** indicating a medium-viscosity grade suitable for seed-coating applications. According to the Food Chemicals Codex (FCC), the acceptable viscosity range for commercial sodium alginate varies from 10 to 5000 cP**,** depending on its intended application. The measured viscosity in the present study is therefore well within the recommended range. Viscosity is one of the most important rheological properties governing film formation, adhesion, gel strength, and coating uniformity. It is influenced by several factors, including alginate source, molecular weight, extraction procedure, polymer concentration, pH, temperature, and ionic composition of the solution. Similar observations regarding the importance of rheological behaviour in alginate applications have been reported by Fenoradosoa *et al.,* (2018). The medium viscosity obtained in the present study is particularly advantageous for seed coating because it enables uniform polymer deposition without excessive solution thickness. The extracted sodium alginate contained 10% moisture, which is below the 15% maximum limit recommended by FAO standards (Coppen, 1995), indicating good storage stability and product quality. The polymer dissolved readily in both cold and hot water, whereas it remained insoluble in acetone, ethanol, methanol, and petroleum ether (Fig 2). Similar solubility characteristics were reported by Zhou *et al.,* (2020), who demonstrated that sodium alginate readily dissolves in water owing to its ionic and highly hydrophilic nature. Unlike several conventional polysaccharides that require heating for complete dissolution, sodium alginate forms a stable viscous colloidal solution under normal conditions, thereby facilitating its practical use in agricultural formulations, including seed-coating technologies. A remarkable property of the extracted alginate was its water absorption capacity of 5.5 g**,** with the polymer increasing from1 g to 5.5 g after 24 h**.** This pronounced swelling behaviour reflects the hydrophilic nature of sodium alginate and its ability to retain large quantities of water through hydrogel formation. Ma *et al.,* (2015) reported that sodium alginate hydrogels can absorb 200-300 times their own weight in water because of the abundance of hydroxyl and carboxyl functional groups within the polymer network. Such high water-retention capacity is particularly beneficial for seed-coating applications. Furthermore, Leandro *et al.,* (2020) emphasized that comprehensive physicochemical characterization is essential to ensure the quality, safety and functionality of alginate intended for large-scale industrial applications, particularly in the food sector. The present findings satisfy these quality requirements and demonstrate the suitability of the extracted sodium alginate for further applications.

3.1.2 Biochemical

The biochemical composition of the extracted sodium alginate is presented in Table 1. Carbohydrates were the predominant component, accounting for 70% of the extract, whereas protein (0.95%) and lipid (0.90%) contents were comparatively low. The ash content was determined to be 18.15%, representing the total mineral residue present in the extracted alginate. The predominance of carbohydrates indicates that the extracted product was rich in polysaccharides, whereas proteins and lipids were present only in minor quantities, suggesting effective enrichment of the alginate fraction during extraction. Although the polysaccharide content obtained in the present study was slightly lower than that reported by Osman *et al.,* (2020), it was within the range reported for alginate extracted from various alginophyte species by Rioux *et al.,* (2007), indicating that the extraction efficiency achieved in this study was comparable with previously published reports. The ash content (18.15%) was within the acceptable range reported for commercial sodium alginate. According to the Food Chemicals Codex (1981), sodium alginate should possess ash levels within specified quality standards, while Chapman and Chapman (1980) reported ash values of up to 23% for commercially prepared alginate. Therefore, the ash content obtained in the present investigation is consistent with internationally accepted standards, indicating the satisfactory quality of the extracted sodium alginate.

3.1.3 Phytochemical

Furthermore, qualitative phytochemical screening revealed the absence of flavonoids, alkaloids, tannins, phlorotannins, terpenoids, steroids, glycosides and phenolic compounds (Table 1), indicating that most secondary metabolites present in the seaweed biomass were effectively removed during extraction. Only the saponins were detected in the extract. Since saponins are carbohydrate-based compounds, their presence is consistent with the structural composition of alginate. The absence of other phytochemicals indicates high purity of the extracted sodium alginate, which enhances its suitability for industrial use. Similar findings were reported by Sivagnanavelmurugan *et al.,* (2018) and Kavitha *et al.,* (2019), who also observed the absence of major metabolites in purified alginates, with saponins being the only detectable compound. The detection of only saponins suggests that the purified sodium alginate possesses a high degree of purity, making it suitable for subsequent applications where a clean polysaccharide matrix is required.

3.1.4 ICP-MS elemental analysis

The elemental composition of the extracted sodium alginate, determined by ICP-MS and expressed in parts per million (ppm), is presented in Table 2. Sodium (Na) was the predominant element with a concentration of 200,390.405 ppm. The other major elements detected were calcium (Ca) (3,876.649 ppm), magnesium (Mg) (620.544 ppm), potassium (K) (200.573 ppm), phosphorus (P) (164.078 ppm), and iron (Fe) (158.246 ppm). Trace elements, including aluminium (Al) (28.574 ppm), boron (B) (17.813 ppm), and titanium (Ti) (7.107 ppm), were present in comparatively lower concentrations. Heavy metals such as lead (Pb), arsenic (As),

chromium (Cr), and cadmium (Cd) were detected at 8.685, 3.429, 2.075, and 0.158 ppm, respectively. Other elements including lithium (Li), beryllium (Be), vanadium (V), cobalt (Co), nickel (Ni), molybdenum (Mo), silver (Ag), antimony (Sb), cesium (Cs), and mercury (Hg) were detected only in trace quantities (≤1 ppm in most cases) (Table 2). The ICP-MS analysis demonstrated that sodium was the dominant element, accounting for 200,390.405 ppm, whereas all other elements were present at considerably lower concentrations. The predominance of sodium confirms the successful conversion of alginic acid into sodium alginate during extraction and indicates a high degree of purity of the extracted polymer. Similar elemental compositions have been reported for sodium alginate extracted from brown seaweeds such as *Laminaria* and *Ascophyllum nodosum* using ICP-OES and ICP-MS techniques (Marín *et al.*, 2010). The comparatively low concentrations of heavy metals further support the suitability of the extracted alginate for agricultural and industrial applications.

3.1.5 FTIR spectral analysis

The FTIR spectrum of the extracted sodium alginate showed characteristic absorption bands corresponding to the functional groups of alginate (Table 3; Fig 4). Broad peaks at 3699.764, 3584.056, 3295.750 and 3281.286 cm^{-1} were assigned to O–H stretching vibrations of hydroxyl groups associated with alcohols, phenols and carboxylic acids. The absorption band at 3100.011 cm^{-1} represented aromatic C–H stretching. Strong absorption bands between 1818.544 and 1717.300 cm^{-1} corresponded to C=O stretching vibrations of carboxylic acids, esters and anhydrides, confirming the presence of uronic acid residues that constitute the alginate backbone. In addition, the peaks observed at 1270.860, 1087.655 and 1050.050 cm^{-1} were attributed to C–O stretching vibrations of ester and alcohol groups, confirming the polysaccharide structure of the extracted sodium alginate. Fourier Transform Infrared Spectroscopy (FTIR) is a well-established analytical technique for identifying functional groups and characterizing biomaterials (Li *et al.*, 2011). It has been widely employed for the identification of pure compounds (Stefanowicz *et al.*, 2009), analysis of mixtures (Basiuk, 2001), detection of impurities (Pandey *et al.*, 2016) and determination of material composition (Hakim and Shanks, 2011). The characteristic O–H and C=O absorption bands observed in the present study are in agreement with previously reported spectra of Sargassum-derived sodium alginate (Latifi *et al.*, 2015), confirming the presence of mannuronic and guluronic acid residues that form the alginate polymer. The hydroxyl and carboxyl functional groups contribute to the excellent water absorption, gel-forming ability and calcium-mediated cross-linking properties of sodium alginate, making it an ideal hydrogel-forming biopolymer. These characteristics are particularly advantageous for seed-coating applications, where improved moisture retention and enhanced seed imbibition can promote better germination and seedling establishment under moisture-deficit conditions (Belattmania *et al.*, 2020).

Overall, the physiochemical, biochemical, phytochemical, ICP-MS and FTIR analyses confirmed that the extraction procedure successfully produced high-quality sodium alginate from *Sargassum myriocystum*. The biochemical analysis revealed a carbohydrate-rich composition, phytochemical screening indicated minimal interference from secondary metabolites, ICP-MS confirmed sodium as the predominant element with only trace amounts of other elements, and FTIR verified the characteristic functional groups of sodium alginate. Collectively, these results demonstrate the purity and desirable physicochemical properties of the extracted alginate, supporting its suitability as a natural biopolymer for seed-coating applications. Accordingly, the purified sodium alginate was selected as the coating polymer for evaluating seed quality parameters in okra. After confirming with these parameters sodium alginate further used for seed coating and studied the seed quality parameters on okra.

3.2 Seed coating effect

The graphical representation (Fig 4) illustrates the variation in germination percentage among treatments (T_0-T_{14}). Germination percentage ranged from 86% to 93%, with the highest value recorded in T_{14}, indicating superior germination performance. Treatments T_2, T_3, T_9, and T_{13} also exhibited relatively high germination (89%), whereas the lowest germination percentage (86%) was observed in T_0, T_1, T_{10}, and T_{12}. Table 4 shows that the percentage of abnormal seedlings ranged from 2% to 9%. The lowest proportion of abnormal seedlings was recorded in T_{14} (2%), indicating improved seedling quality, whereas T_0, T_1, T_{10}, and T_{12} exhibited the highest abnormality (9%). Root length differed significantly among the treatments, with the maximum value recorded in T_{14} (22.36 cm) and the minimum in the control (17.23 cm). A similar trend was observed for shoot length, with T_{14} producing the greatest shoot length (18.22 cm) compared with the control (13.11 cm). Consequently, the total seedling length was highest in T_{14} (40.58 cm) and lowest in T_0 (30.34 cm) (Table 4; Fig 5). Dry matter production ranged from 0.343 g in the control to 0.463 g in T_{14}. Treatments T_2, T_3, T_7, and T_{13} produced intermediate dry matter values (0.398 g), although these remained lower than those of T_{14}. Vigour Index I followed a similar trend, with moderate variation among treatments but a pronounced increase in T_{14}, indicating enhanced seedling vigour (Fig 4). Likewise, Vigour Index II reached its highest value in T_{14} (43), whereas the lowest value (29) was recorded in the control treatment. Across all measured parameters, including germination percentage, abnormal seedlings, seedling growth, dry matter production, and vigour indices, T_{14} consistently outperformed all other treatments (Table 4). In the present study, seed coating with humic acid, salicylic acid, and ascorbic acid incorporated into a sodium alginate polymer significantly enhanced the physiological performance of okra seeds. Seed-coating technology improves crop performance by enhancing nutrient uptake, activating antioxidant enzymes, and stimulating the synthesis of secondary metabolites, including phenolics and flavonoids. These biochemical responses strengthen plant metabolism, improve stress

tolerance, and ultimately enhance the nutritional and medicinal quality of crops (Rocha *et al.,* 2019). Sodium alginate, a biodegradable polysaccharide extracted from brown seaweed, is an effective seed-coating material because of its high water-holding capacity, which promotes uniform water imbibition and seed germination (McHugh, 1987). In addition to serving as a biodegradable carrier, sodium alginate provides an additional carbon source owing to its high polysaccharide content, thereby supporting metabolic activity during early seedling growth. The superior seedling vigour and root development observed in T_{14} may be attributed to the synergistic effects of humic acid, salicylic acid, and ascorbic acid incorporated within the sodium alginate matrix. A well-developed root system, including secondary roots, enhances nutrient uptake and facilitates better seedling establishment, which is essential for subsequent plant growth and productivity. Humic substances have been reported to improve nutrient availability and exhibit auxin-like activity, thereby stimulating root elongation (Chen *et al.,* 2004; Mackowiak, 2001). Similar findings were reported by Sivasakthi and Renganayaki (2022), who demonstrated that humic acid-based polymer coatings enhanced germination, seedling growth, and vigour indices. The beneficial effects of humic acid are associated with improved ion transport, increased cell membrane permeability, enhanced respiration, and accelerated enzymatic reactions in the Krebs cycle, leading to increased ATP production and improved plant growth (Zandonadi *et al.,* 2007). Salicylic acid (SA), an important plant signalling molecule, regulates photosynthesis, nitrogen metabolism, osmotic adjustment, water relations, and antioxidant defence mechanisms (Miura and Tada, 2014). It also promotes protein synthesis, primary metabolism, and the activity of antioxidant enzymes, including superoxide dismutase (SOD), peroxidase (POD), and catalase (CAT), which collectively reduce oxidative damage during seed germination and early seedling growth (Sedghi, 2010). Enhanced antioxidant activity and improved stress tolerance following SA application have been reported in Arabidopsis, alfalfa, and ginger (Alonso-Ramírez *et al.,* 2009; Wang *et al.,* 2009; Ghasemzadeh and Jaafar, 2013). Ascorbic acid, a major non-enzymatic antioxidant, further contributes to improved seed performance by scavenging reactive oxygen species and regulating several physiological and metabolic processes (Akram *et al.,* 2017). Its positive influence on seed germination and seedling vigour has also been demonstrated in alfalfa (Chen *et al.,* 2021). The incorporation of humic acid, salicylic acid, and ascorbic acid into a biodegradable sodium alginate coating represents a sustainable and environmentally friendly approach to improving seed physiological quality. This formulation enhances nutrient uptake, metabolic activity, antioxidant defence, and seedling establishment while maintaining soil health and environmental sustainability (Skrzypczak *et al.,* 2021). Overall, the superior physiological performance of okra seeds treated with T_{14} can be attributed to the synergistic interaction of these bioactive compounds within the sodium alginate matrix, resulting in enhanced germination, improved root and shoot growth, greater seedling vigour, and increased early plant establishment.

4. Future Thrust

Future research should focus on optimizing the extraction and purification methods to improve the yield and functional quality of sodium alginate from *Sargassum myriocystum.* The integration of this biopolymer with different bioactive compounds, micronutrients, and beneficial microorganisms can be explored to enhance its efficiency as a multifunctional seed coating material. Further studies under field conditions are essential to validate its performance across various crops and environments. Long-term investigations on soil health, microbial dynamics, and biodegradability will provide insights into its environmental safety and sustainability. Additionally, developing low-cost and scalable production techniques will support its industrial application and promote the replacement of synthetic polymers with natural, eco-friendly alternatives in sustainable agriculture.

5. CONCLUSION

The present investigation demonstrated that sodium alginate extracted from *Sargassum myriocystum* possesses desirable physicochemical and biochemical properties suitable for agricultural use. The moderate viscosity, high carbohydrate content, and excellent water absorption capacity, combined with its biodegradability, make it an efficient and sustainable seed-coating polymer. The biopolymer and additives had a significant effect on the physiological seed performance in okra. Among the treatments, T_{14} (sodium alginate biopolymer 2 % + humic acid 0.3 g + 250 ppm Salicylic acid + ascorbic acid 0.4 g) showed superior performance by improving stand establishment parameters such as germination, root length, shoot length, dry biomass production, vigour index I and II, and by reducing abnormal seedlings compared to the control. These findings highlight that seed coating with S. *myriocystum* derived sodium alginate infused with biomolecules effectively enhances seed quality attributes and seedling growth. Thus, sodium alginate biopolymer represents an eco-friendly and sustainable alternative to synthetic polymers, supporting greener practices in modern seed technology.

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Declarations

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Competing interests

The authors declare that they have no competing interests.

Availability of data and material
The data supporting the findings of this study are available from the corresponding author upon reasonable request.
Code availability
Not applicable.
Authors' contributions
Pravina K. conducted the experiments, analyzed the data, and prepared the manuscript. All authors reviewed and approved the final manuscript.

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Figures

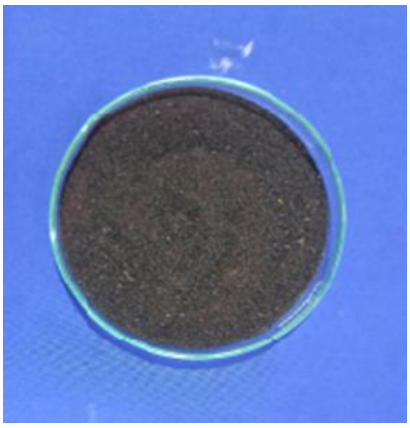
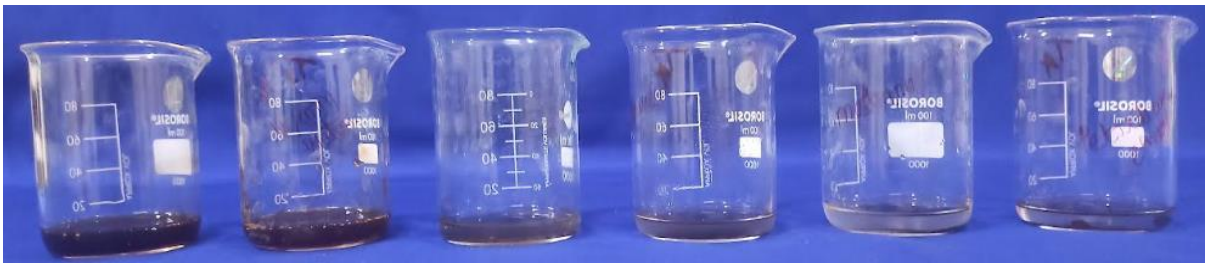


Fig. 1 Color of sodium alginate



Warm water **Cold water** **Acetone** **Ethanol** **Methanol** **Petroleum ether**

Fig. 2 Solubility behaviour of sodium alginate

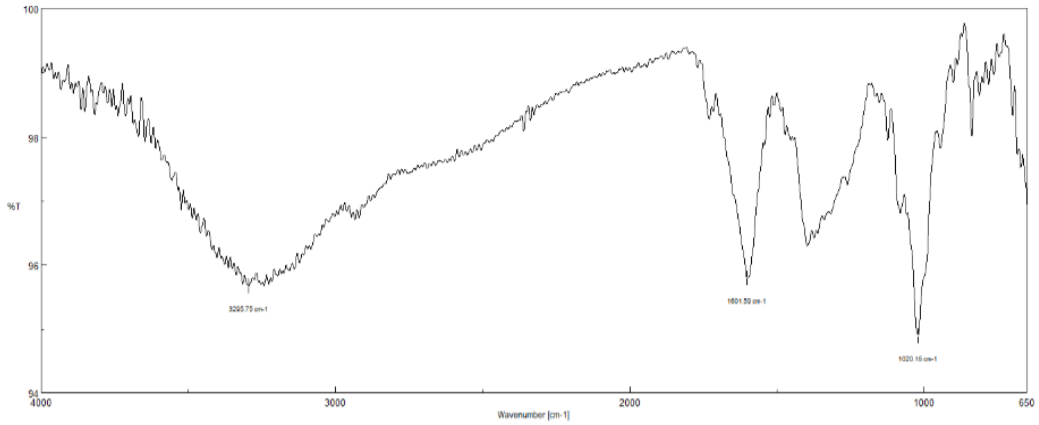


Fig. 3 FTIR spectrum of sodium alginate isolated from *Sargassum myricocystum*

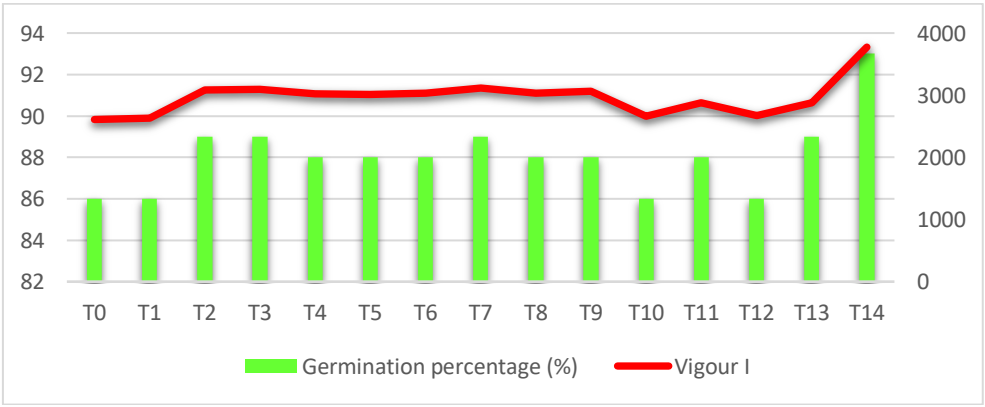
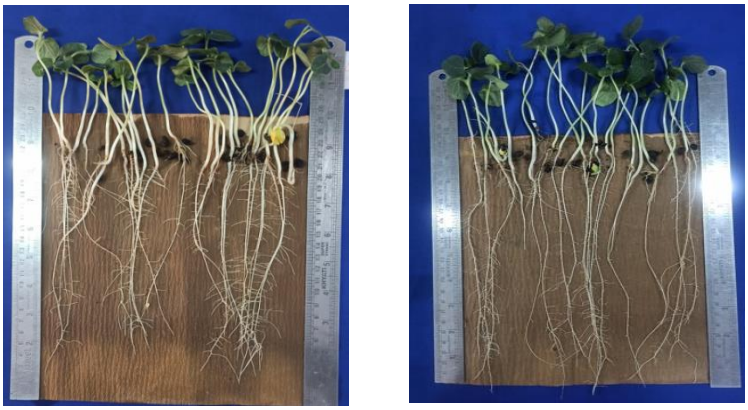


Fig. 4 Impact of biomolecules infused sodium alginate coating on germination percentage (%) and vigour index I on Okra



T_0 T_{14}

Fig. 5 Visualized difference in seedlings

Tables

Table. 1 Represent the physiochemical, biochemical and phytochemical analysis of sodium alginate from seaweed *sargassum myriocystum*

Physiochemical analysis		**Biochemical analysis**		**Phytochemical analysis**	
Yield	25 g	**Carbohydrates**	70 %	**Flavonoids**	–
pH	7.20	**Proteins**	0.95 %	**Alkaloids**	–
Viscocity	90 cp	**Lipid's**	0.90 %	**Tannins**	– (Absent)
Moisture content	10 %	**Ash content**	18.15 %	**Phlorotannins**	–
Water absorbancy	5.5 g			**Terpenoids and Steroids**	–
Solubility Behaviour	Dissolves in both warm and cold water			**Saponins**	+ (Present)
				Glycosides	–
				Phenolic Compounds	–

Table. 2 Represent the ICP-MS analysis

Element (Isotope)	**Element Name**	**Sodium Alginate Sample (ppm)**
7 Li	Lithium	0.022
9 Be	Beryllium	0.009
11 B	Boron	17.813
23 Na	Sodium	200390.405
24 Mg	Magnesium	620.544
27 Al	Aluminium	28.574
31 P	Phosphorus	164.078
39 K	Potassium	200.573
44 Ca	Calcium	3876.649
48 Ti	Titanium	7.107

51 V	Vanadium	0.184
52 Cr	Chromium	2.075
55 Mn	Manganese	4.100
57 Fe	Iron	158.246
59 Co	Cobalt	0.125
60 Ni	Nickel	0.876
63 Cu	Copper	2.013
66 Zn	Zinc	3.237
75 As	Arsenic	3.429
95 Mo	Molybdenum	0.084
107 Ag	Silver	0.021
111 Cd	Cadmium	0.158
118 Sn	Tin	3.413
121 Sb	Antimony	0.010
133 Cs	Cesium	0.001
137 Ba	Barium	9.444
202 Hg	Mercury	0.007
208 Pb	Lead	8.685

Table. 3 FTIR frequency peak (cm^{-1}) and functional groups of the extracted sodium alginate from seaweed *sargassum myriocystum*

S. No.	Wave number (cm^{-1})	% Transmittance	Functional Group (Vibration)	Bond Type / Assignment
1.	3699.764	98.5061	O-H stretching (free)	phenol, alcohol
2.	3584.056	97.6994	O-H stretching (free)	alcohol, phenol
3.	3295.75	95.6635	O-H stretching (hydrogen-bonded)	Alcohol
4.	3281.286	95.7942	O-H stretching	carboxylic acid
5.	3100.011	96.2868	C-H stretching (aromatic)	aromatic hydrocarbon
6.	2906.2	96.9026	O-H stretching	Alcohol
7.	1818.544	99.3767	C=O stretching	Anhydride
8.	1780.939	99.2866	C-H bending	aromatic compound
9.	1775.154	99.1976	C=O stretching	conjugated anhydride
10.	1760.69	99.156	C=O stretching	carboxylic acid
11.	1742.37	98.6035	C=O stretching	Esters
12.	1717.3	98.3983	C=O stretching	Carboxylic acids

13.	1601.592	95.7818	C=C stretching	Aromatic Compounds
14.	1395.246	96.3114	O-H bending	carboxylic acid
15.	1310.393	96.8924	O-H bending	Phenol
16.	1270.86	97.3679	C-O stretching	aromatic ester
17.	1087.655	96.9291	C-O stretching	secondary alcohol
18.	1050.05	96.5043	C-O stretching	primary alcohol
19.	1020.159	94.885	CO-O-CO stretching	Anhydride
20.	900.5941	98.8529	C-H out-of-plane bending	aromatic substitution
21.	830.205	98.8992	C-H bending	1,2,3,4-tetrasubstituted benzene
22.	720.2824	99.4072	aromatic out-of-plane C–H bending	benzene derivative
23.	680.7488	97.7695	C-H out-of-plane bending	aromatic substitution
24.	600.7175	96.2547	M–O stretching	metal–oxygen (inorganic)

Table. 4 Impact of biomolecules infused sodium alginate polymer coating on seed quality parameters of Okra

Treatments	Abnormal seedlings (%)	Root length (cm)	Shoot length (cm)	Seedling length (cm)	Dry matter production (g/ 10 seedlings	Vigour index II
T_0	9	17.23	13.11	30.34	0.343	29
T_1	9	17.59	13.29	30.88	0.351	30
T_2	6	19.46	15.21	34.67	0.398	36
T_3	6	19.46	15.23	34.69	0.393	36
T_4	7	19.21	15.12	34.33	0.394	35
T_5	7	19.17	15.09	34.26	0.392	34
T_6	7	19.37	15.26	34.43	0.391	34
T_7	6	19.68	15.37	35.05	0.398	35
T_8	7	19.29	15.18	34.47	0.394	35
T_9	7	19.49	15.32	34.81	0.396	34
T_{10}	9	17.49	13.53	31.02	0.353	30
T_{11}	7	19.39	15.27	34.66	0.397	32
T_{12}	9	17.78	13.25	31.03	0.351	30
T_{13}	6	19.42	15.31	34.73	0.398	32
T_{14}	2	22.36	18.22	40.58	0.463	43
Mean	7	20.05	15.88	35.93	0.410	35
S.Ed	0.088	0.265	0.203	0.415	0.006	0.388
CD (0.05%)	0.178*	0.535*	0.409*	0.839*	0.013*	0.784*

(**T_0** – control, **T_1**– Sodium alginate Polymer 2 % 250 ml, **T_2** - 0.2 g Humic acid, **T_3**- 0.3 g Humic acid, **T_4** - 0.4 g Fulvic acid, **T_5**- 0.5 g Fulvic acid, **T_6** - Salicylic acid 200 ppm, **T_7** - Salicylic acid 250 ppm, **T_8** – Brassinolide 0.5 ppm, **T_9**- Brassinolide 1.0 ppm, **T_{10}**- 0.05 g Glutothione, **T_{11}** - 0.20 g Glutothione, **T_{12}** - 0.2 g Ascorbic acid, and **T_{13}** - 0.4 g Ascorbic acid **T_{14}** - 0.3 g Humic acid + 250 ppm Salicylic acid + 0.4 g Ascorbic acid)