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Synergistic effect of combining ionizing radiation and oxidizing agents on controlling degradation of Na-alginate for enhancing growth performance and increasing productivity of zea maize plants

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ABSTRACT

Radiation is a very effective tool for controlling the degradation of natural occurring polymers like Na-alginate which may be used in agricultural purposes. One of the principle factors for reducing the cost is achieving the degradation at low irradiation doses. The addition of some additives such as potassium per-sulfate (KPS), ammonium per-sulfate (APS), or H_2O_2 to Na-alginate polymers during irradiation process enhanced, accelerated, reduced the dose required for their degradation process and also improved the quality of the end use products. The highest degradation rate of Na-alginate was obtained when the APS was used. Molecular weight and structural changes of the degraded alginates were determined. The possible practical use of such degraded Na-alginate as a growth promoter for zea maize plant was investigated. The use of radiation degraded alginate not only increases the productivity of zea maize plant but also improve its quality. The results obtained showed that end product of irradiated Na-alginate may be benefited in agricultural purposes as growth promoter for some plants.

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1. Introduction

Polymers become an important part in everyday life. They have been used in different fields such as medicine, industry and agriculture. Moreover, products made from polymers range from sophisticated articles, as biopolymers (knee joints prosthetic hip, etc.) to aerospace and shuttle materials (Scott, 2003). Recently, there is a tremendous potential for using polymers in agriculture. In the last decades functionalized polymers revolutionized the agricultural, horticultural and food industry with new tools for the molecular treatment of diseases, rapid disease detection, enhancing the ability of plants to absorb nutrients etc. (Smith, 2005). Smart polymeric materials and smart delivery systems helped the agricultural industry to combat viruses and other crop pathogens, functionalized polymers were used to increase the efficiency of hormones, pesticides and herbicides, allowing lower doses to be used and to protect the environment and clean-up existing pollutants (Abd El-Rehim, Hegazy, & Abd El-Mohdy, 2004a; Abd El-Rehim, Hegazy, & Abd El-Mohdy, 2006a; Akelah, Kenawy, & Sherrington, 1995; Bastioli, 1998; Petruzzelli, Volpe, Di Pinto, & Passino, 2000). Water absorbing polymers or hydrogels were introduced for agricultural use. The need for improving the physical properties of soils to increase productivity in the agricultural led to the development of water-soluble polymeric soil conditioners. The use of synthetic or natural materials with good water absorption and retention capacities even under high pressure or temperature solves the problem of the water lack and the desertification that compromise agriculture development. Their use for agricultural applications has shown encouraging results; they have been observed to help reduce irrigation water consumption and the death rate of plants, improve fertilizer retention in the soil, and increase plant growth rate (Abd El-Rehim, 2005, 2006; Abd El-Rehim, Hegazy, & Abd El-Mohdy, 2004b; Abd El-Rehim, Hegazy, & Abd El-Mohdy, 2006b).

During the last decade, there is a great demand for agro-chemical residue-free fresh agricultural products. There is a worldwide trend to explore new natural products that act as growth promoters for plants and that control post-harvest pathogenic diseases, giving priority to that enhance the plant productivity, reduce disease incidence and avoid negative and side effects on human health as a result of the excessive application of synthetic agro-chemicals. The degradable natural compounds derived from animal and plants have interested plant growth regulators and pathologists. Among them chitosan, Na-alginate or carageenan, a high molecular polymer, nontoxic, bioactive agent has become a useful appreciated compounds due to its bio-fertilizer, fungicidal effects or elicitation of defense mechanisms in plant tissues (Aftab et al.,

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2011; Hu, Jiang, Hwang, Liu, & Guan, 2004; Khan, Khan, Aftab, Idrees, & Naeem, 2011; Leon & Daryl, 2004).

Degradation is a very important reaction in the chemistry of high-molecular-weight compounds. It is used for determining the structure of polymeric compounds, and obtaining valuable low molecular weight substances from natural polymers (Li et al., 2010). Radiation induced degradation technology is a new and promising application of ionizing radiation to develop viscose, pulp, paper, food preservation, pharmaceutical production, and natural bioactive agents industries. Polysaccharides and their derivatives exposed to ionizing radiation had been long recognized as degradable type of polymers (Delides, Panagiotalidis, & Lega-Panagiotalisdis, 1981; Yoshii et al., 2003; El-Sawy, Abd El-Rehim, Elbarbary, & Hegazy, 2010).

Controlling the degree of degradation, uniform molecular weight distribution, saving achieved in the chemicals (used in conventional methods) on a cost basis, and environmentally friendly process are the beneficial effects of using radiation technology in these industries. However, such technology is not economic because it needs high irradiation dose (hundreds kGy) to degrade natural polymers when exist in solid form. Therefore, a great effort should be done to reduce the cost required for such technologies. One of the principle factors for reducing the cost is achieving the degradation at low irradiation doses. The process for production of oligosaccharide is carried out by irradiation or by chemical methods (using oxidizing agents). The progress, and challenges in the application of irradiated natural polymers as plant growth promoter includes production of oligosaccharide by combination of irradiation and oxidizing agents to lower the dose required for degradation. Depending on the kind of polysaccharide, the concentration of oxidizing agent can be selected to enhance the effect of degradation thus giving a more economical way of producing oligosaccharides.

In this respect, the present work is dealing with studying the effect of ionizing radiation on the degradation of some natural polymers such as Na-alginate. Trials were made to control and reduce the irradiation dose required for the Na-alginate degradation by adding some additives and controlling the irradiation conditions. Field test on Zea maize plants was conducted. The possible practical use of such degraded natural polymers in agricultural field as a growth promotion was also investigated.

2. Materials and methods

2.1. Materials

Na-alginate, of high molecular weight was supplied from Nice lab, India. Hydrogen peroxide (H_2O_2) 30% Loba chemie, potassium per-sulfate and ammonium per-sulfate BDH limited 98% England, were used as received. Zea maize seeds from Elwady Company for production of seeds and agricultural crops, Egypt.

2.2. Preparation of degraded Na-alginate

Degraded Na-alginate was prepared by mixing Na-alginate (powder) with 10 (wt.%) oxidizing agent dissolved in 10 (wt.%) distilled water to obtain Na-alginate in the paste form. The Na-alginate in the paste form was packaged in polyethylene bags then subjected to gamma irradiation from ^{60}Co source at different doses ranged from 20 to 200 kGy.

2.3. Determination the molecular weight of degraded Na-alginate

The weight-average molecular weights of the degradable polymers were determined by two methods:

2.3.1. Viscometric method

The weight-average molecular weights of the polymers were determined on the basis of the Mark - Houwink equation:

$$[\eta] = KM^{\alpha}$$

where $[\eta]$ is the intrinsic viscosity of the polymers, K and α are constants and M is the weight-average molecular weight.

The molecular weight of Na-alginate can be determined by measuring the intrinsic viscosity of polymer solution in 0.01 M NaCl solution taking $K = 8.1 \times 10^{-3}$ ml/g and $\alpha = 0.92$ (Liu, Zhai, Li, Peng, & Wu, 2002).

2.3.2. Gel permeation chromatography method

Gel permeation chromatography (GPC) of irradiated samples was performed on an 1100 Agilant instrument equipped with organic and aqueous GPC-SEC start up kits with a flow rate of 2 ml/min, maximum pressure 150 bar, minimum pressure 5 bar, injection volume 50 μL and column temperature thermostat 25 °C. The GPC elution curve was monitored by a refractive index detector of optical unit temperature 25 °C and peak width 0.1 min polymer concentration was 0.1 (w/%). The molecular weights were determined from a calibration curve using polyethylene oxide standard for aqueous systems.

2.4. FT-IR spectroscopy

FT-IR study was carried using JASCO FT-IR 6300, Japan in the range of 400–4000 cm^{-1}.

2.5. UV–vis spectroscopy

UV–vis study was carried out using Jasco V-560, Japan, in the range from 190 to 900 nm.

2.6. X-ray diffraction (XRD) studies

X-ray diffraction patterns were obtained with a XRD-DI series, Shimadzu apparatus using nickel-filter and Cu–K a target.

2.7. Plantation

To investigate the effect of degraded sodium alginate on Zea maize plants, the concentration of 100 ppm was used. The field can be divided into different separate lines. After plantation of ages 30, 60 and 90 days, each three lines of plants were sprayed by 100 ppm solution of untreated and treated Na-alginates with different irradiation doses. The growth rate of the zea maize plants treated by Na-alginates is measured and compared with that planted in control lines (untreated plant).

3. Results and discussion

3.1. Synergistic effect of combining ionizing radiation and oxidizing agents on degradation of Na-alginate

Studying the effect of oxidizing agents as hydrogen peroxide, potassium per-sulfate (KPS), ammonium per-sulfate (APS), and/or gamma irradiation on the degradation process of Na-alginate was investigated and shown in Fig. 1A. It was found that the molecular weight of the Na-alginate decreased using gamma radiation or oxidizing agents alone. Meanwhile, combining both gamma radiation and oxidizing agents accelerated the degradation rate and dramatically decreased Na-alginate molecular weight.

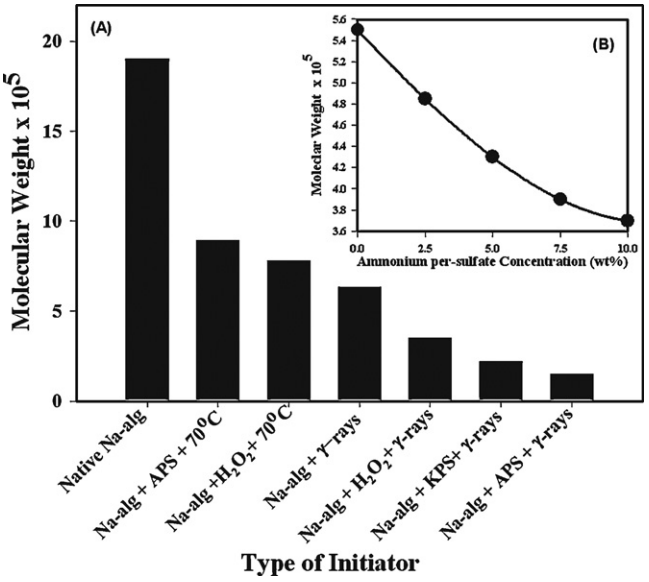


Fig. 1. (A) Effect of some oxidizing agents and ionizing radiation on degradation process of Na-alginate. (B) Effect of ammonium per-sulfate concentration on degradation and the change in molecular weight of Na-alginate irradiated at 80 kGy, the dose rate 6.7 kGy/h.

3.2. Effect of oxidizing agent concentration

Fig. 1B also shows the effect of ammonium per-sulfate concentration on degradation of Na-alginate. It is observed that as the concentration of ammonium per-sulfate increases, the degradation rate sharply increases. Combining 10% APS (wt./wt.) with ionizing radiation, reduced the Na-alginate molecular weight from 1.9×10^6 to 9.2×10^4.

3.3. Effect of irradiation dose on Na-alginate molecular weight

The number average molecular weight and weight average molecular weights of Na-alginate irradiated at different doses were measured by gel permeation chromatography (GPC). Fig. 2 shows the GPC elution curves of native Na-alginate, 120 kGy irradiated Na-alginates, 120 kGy irradiated Na-alginates treated with 10% H_2O_2 and 120 kGy irradiated Na-alginate treated with 10 (wt.%) APS as a function of retention time (min). Low retention time was noted for large molecular weight native sodium alginate. Meanwhile, as the alginate molecule weight decreases, retention time for curve appearance increases. Retention time of 120 kGy irradiated Na-alginate sample treated with 10 (wt.%) APS is longer than that of 120 kGy irradiated alginate sample and 120 kGy alginates samples irradiated in the presence of H_2O_2. The retention time of the irradiated Na-alginate sample is shorter than that of sample irradiated in the presence of 10 (wt.%) APS or H_2O_2. This result indicated that the presence of APS or H_2O_2 during the irradiation process of Na-alginate accelerated its degradation rate.

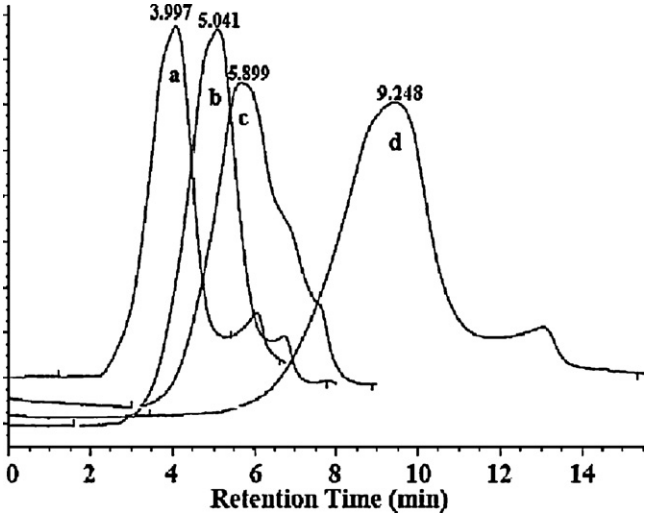


Fig. 2. GPC elution curves of Na-alginates as a function of retention time (a) native, (b) irradiated at 120 kGy, (c) containing 10 (wt.%) APS and irradiated at 120 kGy and (d) containing 10 (wt.%) H_2O_2 and irradiated at 120 kGy, the dose rate 6.7 kGy/h.

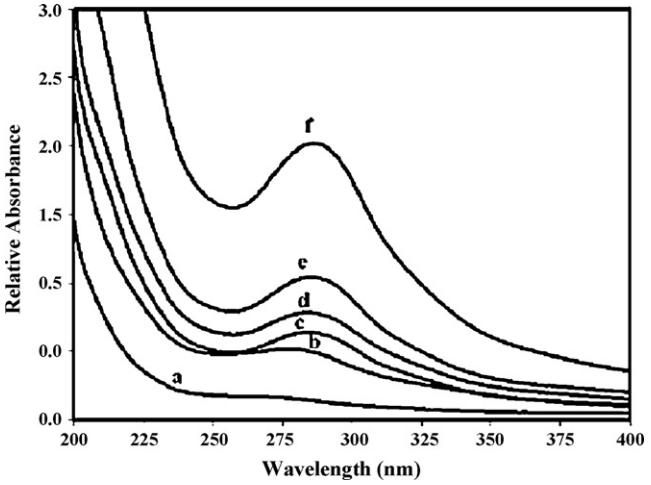


Fig. 3. UV–Vis spectra of (a) unirradiated and irradiated Na-alginate at different irradiation doses of (b) 40 kGy, (c) 80 kGy, (d) 120 kGy, (e) 160 kGy and (f) 200 kGy, the dose rate is 6.7 kGy/h.

Table 1 shows the changes in the number-average molecular weights for Na-alginate measured by GPC as a function of the irradiation dose in absence and presence of 10 (wt.%) APS or H_2O_2. It is observed that as the irradiation dose increases the average molecular weight of Na-alginate decreases. Also, the addition of oxidizing agent to the Na-alginate during the irradiation process accelerates the degradation process. The degradation rate depends on the type of oxidizing agent used. Combining ionizing radiation and oxidizing agents accelerates the rate of polymer scission and reduces the dose required for Na-alginate degradation.

3.4. Structural changes of irradiated Na-alginates

3.4.1. UV-vis spectroscopy

Fig. 3 shows the UV-vis spectra of native and irradiated Na alginate at different doses. There is a new absorption band around 280 nm and the band intensity increases with increasing the irradiation dose. This can be assigned to unsaturated bonds of Na-alginate formed after main chain scission and/or hydrogen abstraction reaction occurred during irradiation (Nagasawa, Mitomoa, Yoshii, & Kume, 2000). The aqueous solution Na-alginate pall yellowish color that changed to brown by radiation confirmed the formation of the unsaturated bonds. As the exposure dose increases, the brown color intensity changes to deep ones.

3.4.2. FT-IR spectroscopy

FT-IR spectra of Na-alginates before and after degradation are shown in Fig. 4. The native Na-alginate showed the polysaccharide structure characteristic absorption bands at 1095 and 1037 cm^{-1} for (C–O stretching). The asymmetric and symmetric stretching of carboxylate vibrations appeared at 1619 and 1420 cm^{-1}, respectively (Leal, Matsuhiro, Rossi, & Caruso, 2008). The spec-

Table 1
The different number average molecular weights of gamma irradiated Na-alginate in absence and the presence of different chemical initiators measured by GPC.

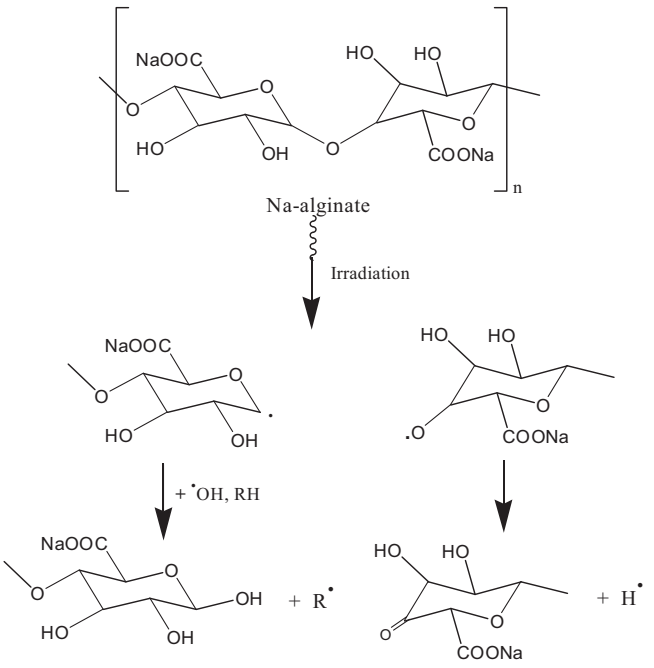
Irradiation Dose (kGy)	Mwt of Irrad. Na-alg	Mwt of Irrad. Na-alg with 10% H_2O_2 (v/wt)	Mwt of Irrad. Na-alg with 10 (wt.%) APS
Mwt of native Na-alg	1.9×10^6	–	–
0			
20	14×10^5	11.2×10^5	7×10^5
40	9.7×10^5	8.7×10^5	4.5×10^5
80	5.5×10^5	4.6×10^5	2.9×10^5
120	4.3×10^5	3.3×10^5	1×10^5
160	4.1×10^5	3×10^5	7.8×10^4
200	3.3×10^5	1.1×10^5	2.9×10^4

trum of oxidized Na-alginate exhibited most of the characteristic adsorption peaks of native alginate but with some differences. For instance, the bands at 1619 cm^{-1} for carboxylate groups, at 3341 for OH groups and at 1035 cm^{-1} and 1095 for (C–O stretching) which became broader and shift to another wave numbers. The spectrum also showed the new bands appeared at 1725 cm^{-1}; The spectra indicated that the formation of carboxyl groups suggesting that ionizing radiation treatment under extreme conditions broke the glucoside bonds with the change of the structure of reducing end residue and formation of –C=O groups.

The data obtained from UV-vis and FT-IR studies indicated that Na-alginate irradiated in solid state suffers scission of acetal linkages in main chains. The scission of the Na-alginate glycosidic bond and formation of unsaturated double bonds during the radiation degradation process occurred. From these finding, the proposed mechanism of degradation after irradiation is illustrated in Scheme 1.

3.4.3. XRD studies

XRD study was made to illustrated the structural changes on Na-alginate occurred by radiation. Native Na-alginate exhibited a characteristic peaks at $2\theta = 21$ as shown in Fig. 5. The intensity of this peak decreases with increasing the irradiation dose. This means that irradiation process caused degradation and destruction on Na-alginate crystal structure. The degradation was probably due to the direct effect of radiation and the indirect effect due to oxidation process. Irradiation of polysaccharides leads to breakdown of the ordered system of intermolecular as well as intermolecular hydrogen bonds. Consequently, the rigidity of chains is influenced by intra-molecular hydrogen bonding and the degree of crystallinity of the material decreases. It was assumed that the degradation first took place preferentially in the amorphous region and then proceeded very moderately from the edge to the inside of the



Scheme 1. Proposed mechanism of radiation degradation of Na-alginate.

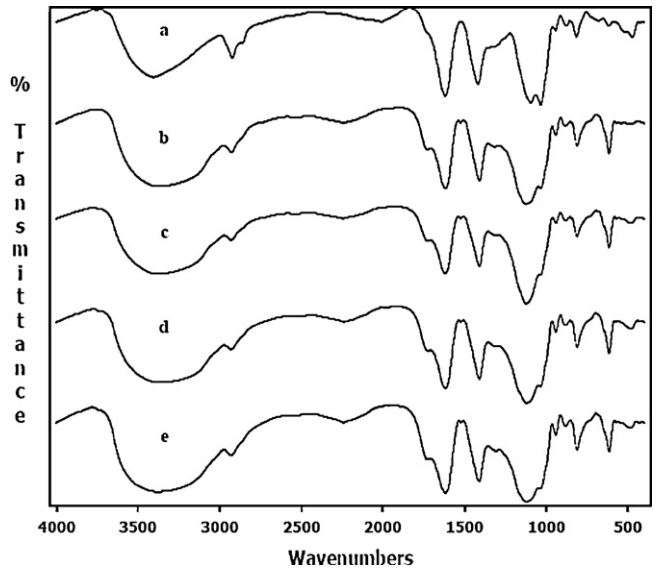


Fig. 4. FT-IR spectra of (a) unirradiated Na-alginate and that treated with 10 (wt.%) ammonium per-sulfate at different irradiation doses of (b) 40, (c) 80, (d) 120, (e) 200 kGy.

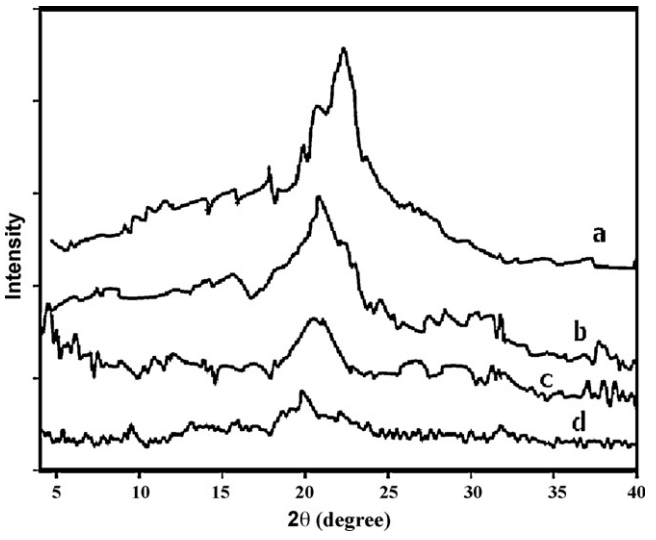


Fig. 5. XRD spectra of (a) Native Na-alginate and that mixed with 10 (wt.%) ammonium per-sulfate at different irradiation doses of (b) 40 kGy, (c) 120 kGy and (d) 200 kGy, the dose rate is 6.7 kGy/h.

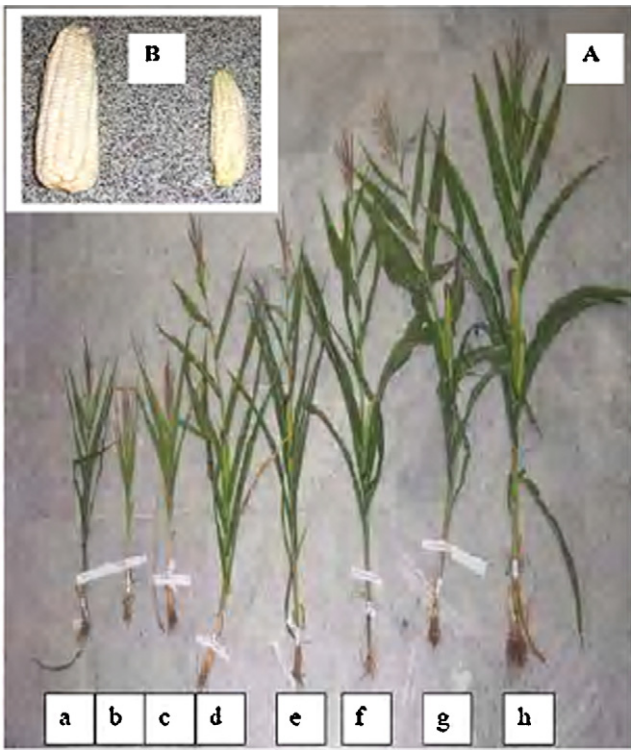


Fig. 6. (A) Effect of degraded Na-alginate (100 ppm of Na-alginate solution in water), on growth of zea maize plant: (a) control (b) native Na-alginate, (c) irradiated Na-alginate at 200 kGy (d) and (e) 80 kGy Na-alginate + (wt.%) APS, (f) 120 kGy Na-alginate + 10 (wt.%) APS, (g) 160 kGy Na-alginate + 10 (wt.%) APS and (h) 200 kGy Na-alginate + 10 (wt.%) APS. (B) Growth of zea maize cob yield size (a) control (b) Na-alginate irradiated at 200 kGy in the presence of 10 (wt.%) APS.

crystalline at higher doses (Mitomo, Watanabe, Ishigaki, & Saito, 1994).

3.5. Applications of degraded alginate polymer in agricultural purposes as growth promoters

Trails were made for using irradiated Na-alginate in agricultural purposes as zea maize growth promoters. The effects of different molecular weight Na-alginates on the growth and other responses of Zea maize plant were investigated. The test field results in Fig. 6A shows that the treatment of the zea maize plant with the 120, 160, 200 kGy irradiated Na-alginate in the presence of 10 (wt.%) APS results in increasing in plant growth. The irradiated Na-alginate not only enhances the plant growth performance but also increases its productivity as seen in Fig. 6B which shows the effect of Na-alginate on the corncob. It is clear that the zea maize cob size of plant treated with irradiated Na-alginate at 200 kGy in the presence of 10 (wt.%) APS is the better than the control one.

Table 2 describes plant height (cm), ear height (cm), ear length (cm), 100 grain weight (g), grain weight/ear (g), biological yield fad. (ardab), straw yield fad. (ardab), grain yield fad. (ardab), total protein (%) and total oil (%) of zea maize plants, spraying with 100 ppm irradiated Na-alginate. It is obvious from Table 2 that different molecular weight Na-alginates investigated here enhances the growth and productive yield of zea maize plant. Low molecular weight alginates prepared at high irradiation doses in the presence of APS has a great effect on quality and quantity of Zea maize plant. 200 kGy irradiated Na-alginate in the presence of APS gives plant height (cm), ear height (cm), ear length (cm), 100 grain wt (g), grain wt/ear (g), biological yield fad. (ardab), straw yield fad. (ardab), grain yield/fad. (ardab), total protein% and total oil % of 229.6, 100.03, 21.49, 34.85, 199.1, 63.52, 33.11, 30.41, 9.47 and

Table 2
Effect of 100 ppm of irradiated Na-alginates in the presence of 10 (wt%) APS on crop yield of zea maize plants.

Treatment	Factor									
	Plant height (cm)	Ear height (cm)	Ear length (cm)	100 (cm) wt (g)	Grain wt/ear (g)	Biological yield fad. (ardab)	Straw yield fad. (ardab)	Grain yield/ fad. (ardab)	Total protein (%)	Total oil (%)
Control	116.5	79.03	17.71	27.38	139.7	46.24	24.47	21.77	8.85	4.48
Blank	116.3	78.92	17.69	27.31	139.1	45.61	24.33	21.78	8.73	4.22
With APS at 40 kGy	221.4	94.26	19.89	29.42	163.8	52.18	26.97	25.21	9.06	4.58
With APS at 80 kGy	222.4	95.47	20.17	31.11	164.9	53.08	27.43	25.65	9.17	4.61
With APS at 120 kGy	223.5	96.47	20.51	32.33	166.7	55.94	29.17	26.77	9.39	4.67
With APS at 160 kGy	227.6	99.28	21.89	33.94	178.4	61.43	32.51	29.92	9.43	4.75
With APS at 200 kGy	229.6	100.03	21.49	34.85	199.1	63.52	33.11	30.41	9.47	4.87

4.87, respectively compared with control plant which gives 116.3, 78.92, 17.69, 27.31, 139.1, 45.61, 24.33, 21.78, 8.73 and 4.22, respectively. The use of irradiated Na-alginate improved the quality of maize plant. The grain size, grain weight, total protein and total oil (%) increased if compared with the control one. The alginate oligomers generated by de-polymerization of Na-alginates have been reported to stimulate the plant growth, seed germination and shoot elongation in plants. They act as signal molecules that regulate plant growth and development as well as the defense reactions in plants by regulating gene expression. The results suggest that alginate-derived oligosaccharide probably applied as leaf-sprays improved the growth attributes, enhanced the acceleration of the metabolic activities, photosynthetic capability, enzyme activities, and artemisinin content of the plant significantly (Hu et al., 2004; Aftab et al., 2011; Khan et al., 2011; Li et al., 2010).

4. Conclusion

There is a synergistic effect on the degradation rate of Na-alginate when the radiation process was carried out in the presence of some oxidizing agent KPS, APS or H_2O_2. The highest degradation rate of Na-alginate obtained when APS was used. To elucidate the structural changes as a result of Na-alginate degradations, UV-vis, FTIR, XRD and molecular weight determination was investigated. The end product of irradiated Na-alginate could be used in agricultural purposes as a growth promoter for plants. The growth and other responses of zea maize plant that treated with irradiated Na-alginate of different molecular weights were investigated. The test field results showed that the treatment of the Zea maize plant with Na-alginate irradiated at doses 160 and 200 kGy in the presence of APS results in increasing in plant growth. The use of Na alginate enhances not only the plant growth and performance but also the productivity.

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