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Comparative study on wheat grains produced from plants treated by irradiated sodium alginate and stored in refrigerator with newly planted on vitality, quality, and their resistance to lesser grain borer (*Rhyzopertha dominica*)

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Abstract: *Field experiment was conducted to compare between the wheat grains produced from plants sprayed by 1% solution of irradiated Na- alginate (ISA) stored in refrigerator for 30 months with those planted recently on vitality, quality and resistance to injury by Rhyzopertha dominica invasion. It was observed that untreated stored grains T_1 decreased germination percentage about 15% comparing by newly planted control (T) and the depress in the stored treated grains were 7, 2 & 3% in 80 (T_2), 120 (T_3), and 160 (T_4) kGy, respectively. While insignificant changes for the aforementioned treatments in concern to germination rate observed. The best growth characters were observed in T_4. The highest soluble sugar, total carbohydrates and total amino acids were detected in T_3. In entmology experiment it was noticed that all treatments had a range of resistance to R. dominica injury under laboratory conditions. The resistance cannot be attributed to grain size, but attributed to infestation percentage and frass weight. The most resistance found in stored grains (T_3 and T_2), respectively, in general non-stored seeds was more resistance to R. dominica than those stored in refrigerator. Moreover, (ISA) treatments had an inhibitory effect on insect amylase which varying from 85.6% - 56.1% and invertase which varying from 67.3% - 6.8%. Except pest tested with stored grains (T_4) at low temperature showed an increase 13.5% and 20.6% in amylase and invertase activity, respectively. Irradiated sodium alginate preserved grains survival after long storage period especially T_3 followed by T_4. Also,using ISA before sowing stimulate the growth and yield.*

Key words*: Triticum aestivum, Rhyzopertha dominica, irradiated Na-alginate, storage.*

I. Introduction

Globally wheat, *Triticum aestivum* is the second most important food crop; it contributes about 25% to the total grain production and is part of daily human diet. Stored food grains face severe damage due to infestation by insects. The insect damages are ranging from 5-30% of the world's total agricultural production. Gamma rays belong to ionizing radiation and are the most energetic form of such electromagnetic radiation having the energy level from around ten to several hundred kilo electron volts. Therefore, it is the most penetrating types of radiation (Kovacs, E. and A. Keresztes, 2002). Also, Sodium alginate which is natural resource exposed, either in aqueous solution or in a dry state, to a radiation so that it decomposed to a lower-molecular weight polysaccharide. there was an increase in faba bean plant performance by using PAA m/Na-alginate copolymer and suggested it is possible to use it in the agriculture field as a soil conditioner, providing the plant with water as well as oligoalginate growth promoter (Abd El-Rehim, 2006). Harvested grains suffer enormous damage from insect pests during storage. *Rhyzopertha dominica*, the lesser grain borer, is one of the most important insect pests infesting whole, dried, and sound grain of many cereals and legumes throughout the world (Nighat et al., 2013). Their protection in storage has been the subject of many studies including the search for resistant varieties. Screening of many seed varieties had led to the successful isolation of strains that are resistant to insect pests in some African countries. Also, Knudsen et al., (1990) reported application of alginate in bio-pesticide formulation. The objective of this search compare between stored wheat grains produced from plants sprayed by irradiated Sodium alginate (ISA) in refrigerator and those newly sown and sprayed, on vitality and quality of grains. Also, determination relative susceptibility of the produced grains to attacked by lesser grain borer *R.dominica* under laboratory condition and its effect on certain insect's carbohydrases enzymes activity.

II. Material and Methods

Since, the methods used in this paper were similar to that described in details for wheat in previous paper (Hamideldin and Hussein, 2009) only a brief summary is given here. Wheat plants (Giza 168) that grown

in sand loamy soil under protected wire area were sprinkled by aqueous solution (1%) of sodium alginate irradiated by 0, 80, 120 and 160 kGy (T_1, T_2, T_3, T_4) monthly until harvest date. Plants were harvested and grains stored at 15°C for about 30 months in refrigerator. The stored grains and recent harvested grains were replanted under same field conditions and the new plants were treated as above by the irradiated sodium alginate (ISA). The growth and yield parameters, sugars and amino acids concentration were determined, in addition to test the produced grains as bio-pesticides through insect experiment.

2.1. Grains germination

The stored grains together with recent harvested grains were tested for germination percentage and germination rate before replanting. Equal numbers of seeds were germinated in glass Petri dishes on filter paper each contained fixed amount of water; the dishes were placed in a controlled-temperature incubator at 20 °C in the dark. The germination percentage (GP) was calculated. The germination rate (GR) calculated according to Bewely and Black, 1984.

2.2. Growth and yield characters

The growth and yield characters measured were length of shoot (cm), length of spike (cm), weight of spike, weight of seeds plant^{-1} and 1000 seeds weight in gram were measured at the end of the experiment. The means of ten plants were subjected to the standard analysis of variance procedure.

2.3.Soluble, insoluble sugar and total carbohydrates estimation

Extraction and estimation of soluble sugars A known weight of the dried samples was homogenized with 80% ethanol then put in a boiling water bath for 15 minutes. After cooling the extract was decanted and repeated extraction three times and collect supernatant after filtration and oven dried at 60°C to dryness. The residue left were dried and used for determination of polysaccharide. Dissolve in a known volume of distilled water to be ready for determination of soluble sugars by recommended method of (Homme et al., 1992). The anthrone sulphuric acid method according to Whistler et al., 1962 was used for determination of soluble sugars. The reagent consists of 0.2g anthrone (Rasayan Mumbai prod. No. 37246) and 100ml 95% cooled sulphuric acid. These were carefully mixed in a conical flask under continuous cooling (freshly prepared). 0.1 ml of samples sugar solution was put in a clean Pyrex test tube and mixed with 10 ml of anthrone reagent then heated at 100°C in water bath for exactly 7 minutes, after which directly cooled. The developed colour was read at a wavelength 620 nm against a blank containing only water and anthrone reagent using Schimadzu 120-02 Spectrophotometer. A calibration curve using pure glucose was carried out. The dry residue left after extraction of soluble sugars was used for determination of polysaccharide. A known weight of dried materials was added to 10ml 1.5Nsulphuric acid in tube with air reflux and heated in water bath at 100°C for 6 hours. The hydrolysate was made up to a known volume to be ready for polysaccharide determination as above by anthrone method (Whistler et al., 1962) The data were calculated from a calibration curve as mentioned above.
Total carbohydrates were calculated as the sum of the amounts of soluble sugars and polysaccharides of the same sample. All data were calculated as mg 100g^{-1} dry weight.

2.4. Amino acids analysis

were done in amino acid laboratory at NCRRT, the sample prepared by weighing 50 mg powdered grains in glass tube containing 5 ml of 6 N HCl, the tube was sealed and kept in an oven at 110°C for 24 h for complete digestion (AOAC,1990). Samples were evaporated and dissolved in Sodium Citrate then filtered to be ready for analysis (Baxter, 1996). The System used was High performance Amino Acid Analyzer, Biochroma 20.

2.5. Insect experiment:

2.5.1. Rearing techniqu

The strain used in the present study *R hyzoperthadominica* obtained from Natural control laboratory at (NCRRT), Nasr City, Cairo, Egypt. Wheat was used for larval feeding under laboratory conditions (27±2˚C and 65±5% relative humidity) and away from any internal chemical pressure. Laboratory tests revealed its susceptibility.

2.5.2. Determination the susceptibility of wheat for *R.dominica*

Wheat that produced from non-stored seeds (T) and that produced from stored one (T_1) was used to determine they susceptibility for *R. dominica*. The tests conducted by placing grain into glass cups topped with muslin handkerchief and secured with rubber bands. Fifteen couples of newly emerged adult's beetles of

homogeneous age from laboratory-stocked culture released in the middle of glass containers (15 x 6 cm) contain (10 g) grain samples weight (three replicate) and kept for 3 months. All treatments receiving feeding damage compared using a similar procedure. Occurrence of other criterion such as total grains, healthy and damaged grains, and percentage damage recorded at the end of the experiment. For evaluating frass weight, the quantity of frass material within each treatment separated, weighed, and recorded. Similarly, the mean proportions of losses compared between varieties to calculate significant differences between the treatments.

2.5.3. Determination of enzymatic activity of *R. dominica*

To evaluate the enzymatic activity under laboratory conditions (27±20C and 65 ±5% relative humidity), 300 adults form *R.dominica* were fed on 5g of treated seeds for each replicate, and placed in glass cups topped with muslin handkerchief and secured with rubber bands till 15 days. Adult supernatant for chemical analysis obtained from the survived adult in each treatment according to EL Saidy (1989), the insects homogenized in water (20 mg/ml) and centrifuged in cold at 10.000 g for 15 min. The supernatant used as enzyme solution.

2.5.4. Enzyme assay

Amylase and invertase activity were assayed by the dinitrosalicylic acid (DNS) procedure Ishaaya and Swirski (1970).Using soluble starch as a substrate in case of amylase and sucrose solution in case invertase. Samples (50 µl) were incubated in 50 µL 0.5 M phosphate buffer (pH 6.9)100 µL 1% (w/v) starch for amylase and 100 µL 1% (w/v) for sucrose. The samples incubated at 37°C for exactly 15 min. in case of amylase and 60 min. in invertase determination. Maltose used as a standard. Each data point represents the mean of three independent assays. Calculations of enzymatic activity carried according to the equation denoted by Abou El-Ghar et al., (1996), enzymatic activity (µg glucose/min./ml haemo lymph) equal:

$$\frac{\text{O.D of test- O.D of zero}}{\text{O.D. of standard}} \times \text{Conc.of standard} \times \frac{1}{\text{Incubation time(min.)}} \times \frac{1}{\text{volume(ml.) of sample}}$$

2.6. Statistical analysis: Data obtained were statistically analyzed by using Costat statistical program software (1990) and Duncan's multiple range test (Duncan, 1955) applied at 5% probability level to compare the differences among estimated parameters.

V. Results

1. Effect of storage on germination percentage and germination rate

The longevity of wheat grains storage evaluated for their survival after storage for 30 months.The treated grains (T_1, T_2, T_3 and T_4) together with recent control grains (T) undergo to germination experiment. The mean squares from Figure (1) shows that, storage of untreated grains T_1 for 30 months decreased germination percentage by about 15% comparing by recent control (T) and depressed 7, 2 & 3% for T_2, T_3 and T_4, respectively. It was noticed that germination percentage ranged between 93.3-98.0% for grains produced from plants sprinkeled by ISA (T_2, T_3 and T_4) comparing by (T_1) 85.33%.

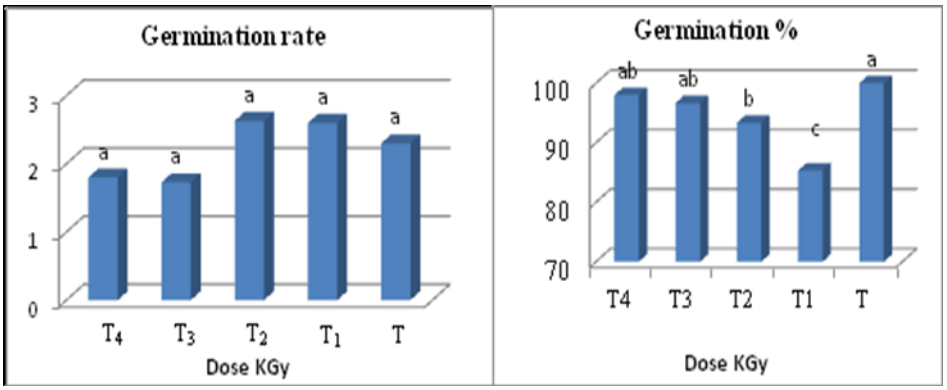

Fig1.Effect of storage on germination rate and percentage of wheat after storage for 30 months. T=non-stored control, T_1= stored control, T_2=80 kGy, T_3=120 k Gy, T_4=160 kGy. Different letters indicate significant variation.

In concern to germination rate, various treatment of (ISA) were not able to reach the level of significant with T.Germination rate for (T_1) and (T_2) were haigher then T as shown in (Fig1). Using of (ISA) can improve vitality of wheat grains for a long time.

2. Effect of storage on plant growth parameters

All grains stored in refregeratore for thirty months produced plants having higher plant length than non-stored control and heavier in weight either for plant or spike. The effect of ISA on shoot heigt in this study was stimulatory as shown from data in (Table 1). Grains exposed to T_2, T_3 and T_4 produced higher plants comparing to T_1 & T. The stimulatory effect of ISA on shoot legnth of plants more pronounced at T_2. Insignificant change noticed in1000 grains weight either before or after storage,. But in concern to weight of grains plant^{-1} , 30 months-old control grains had the heaviest weight plant^{-1} followed by T_2, T_4 and T_3. Same trends was observed regarding to spike weight and legnth. The best growth characters were detected in plants sprinkled by T_4.

Table1. Growth and yield characters of wheat plants, sprayed by (1%) solution of irradiated sodium alginate compared by that produced from stored grains.

Treatment	Na-Alginate (kGy)	Growth characters					
		Plant height (cm)	Plant weight (g)	Spike length (cm)	Spike weight (g)	Weight of seeds/plant (g)	1000 seeds weight (g)
Non-stored	T	80.5^{e}	5.31^{b}	8.00^{c}	1.56^{d}	1.05^{e}	342^{a}
	T_2	91.4^{ab}	5.82^{b}	8.35^{bc}	1.70 ^{cd}	1.03^{cd}	419^{a}
	T_3	84.9^{c}	5.61^{b}	7.90^{c}	1.54^{d}	1.17^{ed}	367^{a}
	T_4	93.8^{a}	6.97^{a}	9.90^{a}	2.22^{ab}	1.59^{b}	368^{a}
Stored	T_1	87.9^{bc}	7.26^{a}	10.40 ^{a}	2.31^{ab}	2.03^{a}	340^{a}
	T_2	86.8^{c}	6.66^{a}	8.94^{b}	1.99^{cb}	1.42^{cb}	381^{a}
	T_3	81.7^{de}	5.25^{b}	8.74^{bc}	1.52^{d}	1.12^{ed}	366^{a}
	T_4	92.4^{a}	6.87^{a}	10.00^{a}	2.39^{a}	1.60^{b}	408^{a}

T=non-stored control, T_1= stored control, T_2=80kGy, T_3=120kGy, T_4=160 kGy. Different letters indicate significant variation.

3. Soluble, insoluble sugar and total carbohydrates in wheat grains

Insignificant change in case of insoluble sugars, but in case of soluble one T_3 followed by T_4 in stored grains had the high significant sugar concentration. Also, in total carbohydrates significantly increase under spraying by ISA (T_3) the same observation were noticed.

Table 2. Soluble, insoluble and total carbohydrates (mg/g) of wheat grains produced from plants, sprayed by (1%) solution of irradiated sodium alginate compared by that produced from stored grains.

Treatment	Na-Alginate (kGy)	Soluble carbohydrate	Insoluble carbohydrate	Total carbohydrates
Non-stored	T	0.83^{c}	10.43^{a}	11.26^{f}
	T_2	0.77^{c}	10.97^{a}	11.74^{e}
	T_3	1.0^{c}	12.92^{a}	13.92^{a}
	T_4	0.80^{c}	11.14^{a}	11.94^{ed}
Stored	T_1	0.80^{c}	11.34^{a}	12.14^{cb}
	T_2	1.16^{bc}	10.92^{a}	12.08^{cbd}
	T_3	1.72^{a}	10.50^{a}	12.22^{b}
	T_4	1.40^{ab}	10.50^{a}	11.90^{ed}

T=non-stored control, T_1= stored control, T_2=80kGy, T_3=120kGy, T_4=160 kGy. Different letters indicate significant variation

4. Amino acids concentration

Total amino acids of non-stored wheat T was 128mg/g become 127.5 after storage for 30 months under low temperature.There is a gradual increase in the total amino acids concentrations with the increase of alginate dose (T_2& T_3) after storage of grains. The plants produced from non-stored grains and sprayed by T_2, T_3 and T_4 had total amino acids 127, 141and 120.5 mg g^{-1}, and while after storage changes to 132, 154 and 113.5 mg g^{-1} for the same aforementioned treatment of (ISA). It was noticed that total amino acids decreased in T_2 and T_4 meanwhile increased in T_3 in which the proline had the highest concentration (9 mg) comparing by the rest of treatment used. Also, marked variations in methionine, isoleucine, leucine and histidine concentrations were observed in stored grains (Table 3).

Table 3. Amino acids concentration (mg/ g) of wheat grains produced from plants, sprayed by (1%) solution of irradiated sodium alginate compared by those produced from stored grains.

Na-Alginate kGy Amino acids	Non-sorted				Stored			
	T	T_2	T_3	T_4	T_1	T_2	T_3	T_4
Aspartic	8	6	7.5	6	7	6	7.5	5
Threonine	7	6	6.5	6	7	6	7.5	5
Serine	8	7.5	7.5	7.5	8.5	7	9	7
Glutamic	11.5	11.5	11.5	11.5	11.5	11.7	11.5	11.5
Proline	5.5	5	5	6	6	5.5	9	5.5
Glycine	5	5.5	5.5	5.5	5.5	5	6	5
Alanine	4.5	4.5	5	4.5	4.5	4.5	5	4
Cystine	4	4.5	6	4	4.5	5.5	6	3.5
Valine	6.5	7	8	6.5	6.5	7.5	8	6.5
Methionine	0.5	4.5	0.5	0.5	0.5	1	8	0
Isoleucine	4.5	6	7	4.5	5	8	7	4.5
Leucine	28	26.5	33	26	28	30.5	33	26.5
Tryrosine	6.5	6	9	6.5	6.5	8	9.5	6
Phenylalanine	8.5	9.5	10.5	9	10	10.5	10.5	9
Histidine	2.5	0.5	1	0.5	0	0.5	0.5	0
Lysine	9	8.5	9	7.5	8	7.5	8.5	7
Arginine	8.5	8	8.5	8.5	8.5	7.5	7.5	7.5
Total mg/ g	128	127	141	120.5	127.5	132	154	113.5

T=non-stored control, T_1= stored control, T_2=80kGy, T_3=120kGy, T_4=160 kGy

5. Susceptibility of *R.dominca*

Seeing that the different treatments exhibited variation in numbers of grain in each 10g sample naturally there existed diversity in number of healthy grains at the end of exposure to infestation. In this respect, the highest healthy grains 291 and 286 occurred in treatment by ISA (T_3 &T_3) of non-stored and stored grains respectively. Number of healthy grains in other treatments varied from 257.7 to 284.7 per sample compared to 255.7 in control. For the parameter of number of grains attacked by pest, responses of all test treatments also varied. The maximum damaged grains observed in plants treated by ISA of stored grains. However, the number of damaged grains decreased in treated non-stored compared to control. The results on % infestation are also presented in the same Table. The lowest % damage and most tolerant was also in treated non-stored T_3 showing 7.3% infestation. The highest infestation was in T_4 stored at 16.9%; consequently, both these treatments behaved most tolerant and susceptible responses, respectively.The other treatments showed moderate differences for this character, where the magnitudes of % damage ranged from 7.9% to 12.6%. Frass weight followed a similar trend in all treatments as was observed in case of % infestation intensity. Maximum frass weight recorded in T_4 stored in low temperature (295mg). The minimum frass weight found in T_3 non-stored (110mg). All the remaining treatments gave an idea that frass weight remained in the order of 140.7mg up to200mg.

Table 4. Susceptibility of *R.dominca* to non-stored and stored wheat plants, sprayed by (1%) solution of irradiated sodium alginate (80, 120 &160 kGy)

Treatment kGy		Mean±S.D.				
		Total Grains	Healthy grains	Damaged grains	Percent infestation	Frass weight (mg)
Non-stored	T	302 ± 7^{b}	255.7 ± 10^{e}	46.3 ± 3^{b}	15.4 ± 1.4^{b}	275 ± 15^{b}
	T_2	280 ± 6^{c}	257.7 ± 5^{de}	22.3 ± 3^{d}	7.9 ± 1^{e}	140.7 ± 10^{d}
	T_3	314 ± 11^{ab}	291 ± 10^{d}	23 ± 3^{d}	7.3 ± 0.7^{e}	110 ± 10^{e}
	T_4	317 ± 17^{ab}	284.7 ± 15^{ab}	32.3 ± 2^{c}	10.2 ± 0.5^{d}	146.7 ± 6^{d}
Stored	T1	324.7 ± 10^{a}	283.7 ± 7^{abc}	41 ± 4^{c}	12.6 ± 1^{c}	217 ± 21^{c}
	T2	310.7 ± 5.9^{ab}	273.7 ± 9^{bc}	37 ± 2^{c}	11.9 ± 0.8^{cd}	200 ± 10^{c}
	T3	322.7 ± 5^{a}	286 ± 2^{ab}	36.7 ± 2^{c}	11.4 ± 0.6^{cd}	153.3 ± 20^{d}

	T4	321.3±10[a]	267±14[cde]	54.3±5[a]	16.9±2[a]	295±13[a]

T=non-stored control, T_1= stored control, T_2=80kGy, T_3=120kGy, T_4=160 kGy. Different letters indicate significant variation.

6. Carbohydrases (amylase and invertase) activity

The changes in carbohydrases (amylase and invertase) activities in the total content of *R. dominca* tissue tested with wheat treated by ISA (T_2, T_3 and T_4) demonstrated in table (5).The results showed a significant decrease ($p<0.05$) in the activity of amylase and invertase as compared with the control in all treatment except insect pest amylase and invertase tested with T_4 stored in low temperature. The highest inhibition effects observed when insect tested with wheat treated by ISA and non-stored T_3 were 85.6% and67.3% for amylase and invertase, respectively. In contrast,the lowest inhibitions found in wheat and stored in low temperature were 34.2 % and 6.8% for amylase, respectively.

Table 5. Changes in carbohydrase (amylase and invertase) activity in the total content of *R.dominca* tested with non-stored and stored wheat plants, sprayed by (1%) solution of irradiated sodium alginate (80, 120 &160 kGy)

Treatment KGy		Amylase Enzymatic activity (µg glucose /min /ml haemolymph)		Invertase Enzymatic activity (µg glucose /min /ml haemolymph)	
		Mean±S.D.	%Inhibition	Mean±S.D.	%inhibition
Non-stored	T	112.8±4.8[b]	-	211.6±3.4[b]	-
	T2	25.5±2.7[g]	77.4	157.6±3[f]	25.5
	T3	16.2±1.5[h]	85.6	126.5±6.7[g]	67.3
	T4	33±1.2[f]	70.7	171±1.3[e]	19.2
Stored	T1	74.2±3.8[c]	34.2	197.3±1.9[c]	6.8
	T2	49.5±1[d]	56.1	190.6±2.5[cd]	9.9
	T3	41.2±1.3[e]	63.5	180.9±2.5[de]	14.5
	T4	128±2.7[a]	+13.5	255.4±14[a]	+20.7

T=non-stored control, T_1= stored control, T_2=80kGy, T_3=120kGy, T_4=160 kGy. Different letters indicate significant variation.

V. Discussion

It was noticed that appliction of (ISA) as foliar spray during planting season preserved the survival of the produced grains after long storage period in refrgerator especially (T_3) followed by (T_4). These results are in consistent with those of Abd El-Rehim et al. (2006) and (2011); they stated that the increase in germination percentage might be due to (ISA) stimulating effects or due to the elimination of germinating bacterial populations,their spores and mould fungi. Also, Rani et al. (2013) can be stored Pinto beans with lower initial moisture contents (12 and 14% w.b.) safely at lower temperatures (10 and 20 °C) maintaining appreciable seed germination, seed coat colour, and microbial stability for 16 weeks. The results obtained referred that plants sprayed by ISA (T_4) of stored and non stored grains induced growth and yelid characters. Similar conclusion were also obtained by Abd El-Rehim et al. (2011) on *Zea maize* plants treatment with 120, 160, 200 kGy ISA in the presence of wt (10%) ammonium per-sulfate not only enhances the plant growth performance but also increases its productivity. Khan et al. (2011) also confirmed the growth promoting effect of ISA in *Papaver somniferum*. IAEA-RCA (2003) reported that after the cucumber was sprayed three times with alginate solution, the seedling quality and major characters of cucumber were better than the blank group, their yield also increased by 15 to 20%. Soluble sugar and total carbohydrates increased unpon spraying plants by (T_3) that may be du to increase in photosynthesis rate.Similar conclusion in rice obtained by Hien et al. (2000); they noticed a significant enhancement in net- photosynthesis and CO_2 assimilation as a result of application of depolymerized SA. Also, Afatab et al. (2011) stated that the application of ISA might improve photosynthetic capacity and photosynthetic pigments. The important role of SA-oligosaccharides has been recognized in inducing cell signaling in various plants, leading to stimulation of various physiological processes (John et al., 1997). Enhancement in - photosynthesis and CO_2 assimilation or improve photosynthetic capacity and photosynthetic pigments as mentioned above perform to increase in sugar content in plants and amino acids content of the plants as a result of treating with (ISA). Total amino acids increased in plants sprayed with ISA (T_3) of stored and non-stored grains. Similar findings were obtained by Hamideldin and Hussein (2009). Cereal grains serve as sources of income to small and large scale farmers in developing countries. Following harvest, these crops often infested by insect pests in store, which cause considerable damage in form of loss of weight loss and conversion of nutrient, reduction in germination capacity or loss of seed vigour (Ahmed and Yusuf,2007). ISA used in the present study are not toxic to vertebrates as well as they are cheap, moreover the ability of Na- alginate to produce grains that maintain better quality and more resistance to lesser grain borer *R.*

dominica. The obtained results may discuss in the following aspects: All treatments tested the susceptibility of *R. dominca* to wheat treated by (ISA T_2, T_3 andT_4) had showen a range of resistance to lesser grain borer injury under laboratory conditions. The resistance cannot be attributed to size of grain, but the resistance can be attributed to percentage of infestation and frass weight. The most resistance seeds found in seeds treated by T_2, T_3 and non-stored, respectively, in general seeds treated by ISA and non-stored was more resistance to *R. dominica* than seeds treated and stored in low temperature. Many authors studied seeds resistance to insect injury also obtained similar findings. Amjad et al., (2011) reported that % infestation of *T. castaneum* ranged from 5.72% to 27.70%. However, in the present study % infestation of *R. dominica* ranged from 7.3% to 16.9%. Bandyopadhyay and Ghosh (1999) investigated the loss of stored wheat ranging from 4.3 to 25.5% damage. Likewise, at present maximum frass material recorded ranging from 110 mg to 295mg. Changes in carbohydrase (amylase and invertase) activity in the total content of *R. dominca* tissue tested with wheat treated by (T_2, T_3 andT_4) non- stored and stored at low temperature discussed in the present study. Many of the natural plant compounds and organic compounds used in the control of insect pests are known to affect digestive enzymes. Secondary organic compounds synthesized by plants have an important role in protecting plants against insect pests (Athanassiou et al., 2005; Pare and Tumlinson, 1999). These compounds affect insects by causing a delay in larval growth and can act as antifeedant (Shekari et al.,2008). The current results showed that all treatments had inhibitory activity on insect amylases varying from nearly 85.6% to 56.1% and varying from 67.3% to 6.8% for invertase. Except insect pest tested T_4 and stored in low temperature showen an increase 13.5% and 20.6% in amylase and invertase activity, respectively.

VI. Conclusion

From the obtained results, using ISA as foliar spray improved crop quantity, quality and increase their resistance for insect infestation. Furthermore it render viable and vital grains after long shelf life at low temperature. So, it can recommended to apply ISA on large scale for increasing crop at harvest, keeping grains vitality on storage and increasing their resistance for insects.

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