

GAMMA RADIATION EFFECTS ON SOME BIOCHEMICAL CHANGES IN POTATO TUBERS AT DIFFERENT STORAGE PERIODS.

Abdel-Raheem, E.A.¹; M.A. Shalan²; I.A. Orabi² and O.I. Saleh²

¹ **Biochemistry Dept., Faculty of Agriculture, Cairo Univeristy, Egypt.**

² **National Center for Radiation Research and Technology, Atomic Energy Authority, P.O. Box 29- Nasr City, Cairo 113701-Egypt.**

ABSTRACT

The effect of gamma irradiation on some biochemical compounds of potato tubers "alph"; during different periods of storage; was studied. Irradiation doses 0, 10, 20 and 40 Gy before planting and 0, 80, 160 and 320 Gy after planting were used as two different storage methods. Also, the effect of periodic time storage at 0, 30, 60, 90 and 120 days on biochemical compounds was investigated. The changes in biochemical compounds such as; total phenolic compounds, chlorogenic acid and peroxidase enzyme activity by colorimetric spectroscopic analysis were followed. Glycoalkaloids were determined by HPLC technique. The results proved that the storage was much better and more predominant in the first than the second method. The remarkable results of the two parameter effects radiation doses and storage time were affected on safety preservative of potato tubers for long time about 120 days and the effective radiation dose was found to be between 10-20 Gy. HPLC represented that it is better to use potato varieties with high β -solanine content more than that of β -chaconine content. So, in this work potato tubers can be stored for more expanded time > 60 days up to 120 days; with using 20 Gy gamma radiation dose before planting. Under these conditions; β -solanine / β -chaconine ratio was 87/13 % with less content of β -chaconine and therefore; less toxic effects on the human liver.

Keywords: Gamma radiation; Total phenolic compounds; Glycoalkaloids; β -solanine; β -chaconine; Chlorogenic acid; Peroxidase enzyme; Planting; Storage.

INTRODUCTION

The potato (*Solanum tuberosum* L.) is an important food crop grown throughout the world. Suberization is a tissue-specific process where by cell walls become impregnated with a poly(phenolic) matrix coincident with the deposition of a poly(aliphatic) matrix between the plasmalemma and carbohydrate cell wall (Bernards and Lewis, 1998). While the nature of the phenolic matrix remains incompletely defined, it has recently been shown that in potato tubers, it comprises primarily hydroxycinnamic acids, and their derivatives (Bernards, et al. 1995 and Negrel, et al. 1996). It has also been shown indirectly that in the suberized tissues of *Quercus suber* (Gil, et al. 1997) and *Clivia miniata* (Schreiber, 1996; Zeier and Schreiber, 1997), there is a significant amount of hydroxycinnamic acid present in the cell walls. Kim, et al. (2002) refere that Peroxidase (POD) activity of potatoes during growth was increased by 7-16% at 1, 8 and 16 Gy irradiation doses compared to the control. Ramamurthy, et al. (2000) showed no effects of gamma-irradiation on lignin biosynthesis during healing of wounds in potato (*Solanum tuberosum* cv. Kufri Chandramuki) tubers. Activities of peroxidase and its isozyme patterns were not significantly changed by irradiation. It is suggested that

lignin biosynthesis may be decreased in irradiated potatoes due to differential regulation of enzymes involved in lignin biosynthesis in response to exposure to gamma-irradiation.

Potato tubers, on exposure to light in the field, store, shop or home, develop greening which is indication of chlorophyll production. This is attributed to elevated levels of the glycoalkaloids β -solanine and β -chaconine; a maximum level of 20 mg per 100 g fresh weight is generally regarded as an acceptable upper limit for total glycoalkaloid content (Sinden and Webb 1972). Higher levels produce varying degrees of food poisoning and associated symptoms (Van Gelder, 1990). A number of researchers investigated the effect of gamma irradiation on the production of chlorophyll and glycoalkaloids with conflicting results. Gamma irradiation of tubers generally reduces the rates of chlorophyll formation (Hetherington and MacQueen 1963; Winchester, et al. 1976). Glycoalkaloid concentration in all cv. were higher than the recommended food safety level, this was reached after 8 days in cv. Kerrs Pink and Desiree and 13 days in Pentland Hawk.

Chlorogenic acid; ester of caffeic acid and quinic acid; has an important influence on defence mechanisms and quality in potato tubers. Tissues from gamma-irradiated tubers have been shown to contain less chlorogenic acid than the non-irradiated control tubers (Pendharkar and Nair, 1987 and Ramamurthy, *et al.* 1992) where the biochemical events responsible for this change. Friedman, (1997) conducted that gamma-radiation affect on the stability of the major potato polyphenol, chlorogenic acid. Also beneficial effects of phenolic compounds in the diet as antioxidants, antimutagens, anticarcinogens, antiglycemic, and hypocholesterolemic agents. Understanding the biochemical basis of stress-induced formation of polyphenols in plants, the chemistry of their transformations in the plant and in foods, and their functions in plant physiology, food science, nutrition, and health should stimulate interest in maximizing beneficial sensory, nutritional, and health effects of polyphenols in the diet. Such efforts should lead to better foods and improved human health. Orsak, *et al.* (2001) indicates the effects of irradiation on total polyphenols and phenolic acids in potatoes. Total polyphenol content in potato were identified by HPLC. L-Tyrosine (35-70%) was the main phenolic compound, followed by chlorogenic acid, caffeic acid and ferulic acid. Gamma-irradiation at lower doses caused an increase in total polyphenols, but had the opposite effect at higher doses. During storage after harvesting, potatoes tend to sprout but this problem is generally controlled by the use of low doses (75-150 Gy) of gamma radiation which are very effective in inhibiting sprouting during post-harvest storage (Thomas, 1984). Sharabash, et al. (1995) found that chlorogenic acid was increased by prolonging the storage time and/or increasing the exposure dose. During a post-irradiation period of 21 days a depletion in chlorogenic acid was observed. This is a result of its impaired synthesis as well as an accelerated conversion of chlorogenic acid to ferulic and sinapic acids and their deposition in lignin. The results offer an explanation for the lower level of chlorogenic acid invariably observed in irradiated potato tubers. The aim of this work was focused on determination of total phenolic compounds,

chlorogenic acid, peroxidase enzyme and glycoalkaloids (α -solanine and α -chaconine) of irradiated and non-irradiated potato tubers before planting and after harvesting and study the effect of different storage periods on such compounds.

MATERIALS AND METHODS

(1) Materials:-

1.1- Potato:

One cultivar of potatoes namely alpha (*Solanum tuberosum* L.) was obtained as the summer seed potatoes, Holland, at 1999 – 2000 season, Ministry of Agriculture of Egypt.

1.2- Radiation source:

The potato samples were irradiated by Co⁶⁰ Canada Cell 220 Research Irradiator. The irradiator dose rate 7.3×10^{-3} Gy/ min.

(2) Experimental:-

2.1- planting:

The planting experimental field was prepared in National Center for Radiation Research and Technology (NCRRT), located in Nasr City, Cairo, Egypt. The soil of the experimental field was clay loam with pH 7.7, as open field. Tuber potatoes were irradiated with gamma rays at doses of 0, 10, 20 and 40 Gy then cultivated on February 21th, 2000 season, at depth 12-15 cm and space with 30 cm among the cultivated seed potatoes. The treated seed tubers were planted in soil with adequate moisture content at planting time. After 7-10 days from planting time, the planting ratio reach to higher than 95%. The different growth periods; 25, 50, 75 and 100 days from start growth was considered.

2.2- Storage:

Storage experimental started from the potatoes after 100 days (Jun 2^{ed}, 2000) from planting and divided into two storage experiments. The first was the potato tubers resulted from irradiated potato tubers with 0, 10, 20 and 40 Gy before planting and the second was the potato tubers; resulted from the non-irradiated (control) planting; were irradiated with 0, 80, 160 and 320 Gy. The samples were taken from both experiments at different storage periods 0, 30, 60, 90 and 120 days where the chemical analysis of each sample was determined.

(3)-Chemical analysis:-

Total phenolic compounds, Glycoalkaloids (α -solanine and α -chaconine), Chlorogenic acid, and Peroxidase activity were determined in different potato tubers.

3.1- Total phenolic compounds:

Total soluble phenolic compounds extracted by the method of Mendel, *et al.* (1989) were determined using the folin dennis colorimetric method (A.O.A.C., 1965) at wave length 730 nm. The concentration was calculated from the standard curve of pyrogalllic acid as mg. per gram dry weight.

3.2- Glycoalkaloids:

The extraction spiking experiments and determination by H.P.L.C. were observed by Mendel and Lan (1992). A Beckman 1090 HPLC system with a 427 integrator and a 165 UV-Vis. variable-wavelength detector was used (Friedman and Levin, 1989). The column was ODS C18 Perkin-Elmer column (5 micron), 250x4.6 mm i.d. spherisorb ODS-2 microguard column. The mobile phase consisted of 50% acetonitrile containing 5 mM sodium lauryl sulfate (SDS) and 5 mM anhydrous sodium sulfate. The pH was adjusted to 4.5 with 1% sulfuric acid. The solvent flow rate was 1ml/min, and the UV absorbance was monitored at 200 nm.

A glycoalkaloid stock solution (1 mg/ml) was prepared by dissolving α -solanine and α -chaconine (sigma chemical Co.). Working standard solution were prepared by dilution with methanol at 4 concentration (0.5, 0.25, 0.125 and 0.0625 mg/ml of α -solanine and α -chaconine. All solvents were of HPLC grade.

3.3- Chlorogenic acid:

The quantitative colorimetric assay for chlorogenic acid was based on the Hoepfner reaction (Hoepfner, 1932). The absorbance of prepared samples was measured after 15 min incubation at wave length 520 nm against a reagent blank. Total chlorogenic acid was calculated from a standard curve prepared with authentic chlorogenic acid (Mendel, *et al.* 1989).

3.4- Peroxidase enzyme:

The extraction and determination colorimetric method were described by Laurie and Carl (2000). Peroxidase activity was determined in enzyme extract after the extraction method by Galeazzi and Sgarbieri, (1981) and Galeazzi, *et al.* (1981) spectrophotometry based on the procedure originally developed by George, (1953). The reaction was based on the oxidation of hydrogen peroxide in the presence of the enzyme, with a colour change from clear to orange/pink, and measured using an absorbance of 436 nm with a Shimadzu 120-UV spectrophotometer.

RESULTS AND DISSCUTION

1- Effect of different storage periods and gamma irradiation doses on total phenolic compounds of potato tubers before and after planting:

Table (1) showed that total phenolic compounds mg/100g d.wt. increased with prolongating storage up to 120 days for both non-irradiated and irradiated samples due to the activation of synthase enzyme which leads to more production of sucrose and glucose. On the other hand; total phenolic compounds content slightly increased at lower irradiation doses 10 Gy and after that; its content decreased with increasing gamma irradiation doses on potato tubers before planting. This can be attributed for that; gamma irradiation at lower doses caused an increase in total phenols due to the activation of phenolic synthase enzyme, but had the opposite effect at higher doses; more than 10 Gy due to the degradation effects on the phenolic

compounds (Spinks, *et al.*, 1976). From fig. (1) it was noticed that the best conditions for the storage was for irradiated potato tubers before planting at 10 Gy; and storage time between 90 and 120 days. The higher irradiation dose more than 10 Gy had no significant effect on total phenolic compounds.

Table (2) and fig. (2) showed the effect of gamma irradiation on total phenolic compounds of irradiated potato tubers during different storage periods. It was found from the results that the total phenolic compounds content increased with increasing the period of storage at ambient temperature and the less amount recorded at 120 days was the irradiated tuber potato at 160 Gy after planting. These results referred that; there was no remarkable change between non-irradiated and irradiated potato tubers in the total phenolic compounds content through all the time of storage. This is because phenolic compounds had high resistance to gamma radiation which can not easily be degraded due to its high resonance structure by the electronic delocalization in their aromatic rings. These results agree with many authors such as Hespels, (1978) who stated that phenolic compounds of potato tubers were increased with gamma irradiation at 50, 80, 120, 140, 150 Gy, due to the enzymatic activation. The active species due to the radiolysis products by gamma irradiation e^- , H^+ , OH^+ , O_2^+ , and HO_2^+ , play an important role which accelerates the enzymatic activity and therefore, their influence as tubers preservative by the clean method using ionizing gamma radiation. Bergers, (1981) showed that the phenolic compounds in potatoes irradiated at the highest dose level of 3 kGy undergo a time dependent change during storage. This could be partly ascribed to the β -glycoside of scopoletin (7-hydroxy 6-methoxy, coumarin) and also accompanied with a general decrease of chlorogenic acid (Mondy and Gosselin (1988) and Nell and Barry (1989)).

Table (1): Effect of different storage periods on total phenolic compounds of potato tubers before planting at different gamma irradiation doses.

Treatment	Total phenolic compounds (mg /100 g d.wt.)				
	Storage periods (day)				
	0	30	60	90	120
Control	15.16 ^a ±0.906 [0.00]	15.66 ^a ±0.478 [3.29]	19.04 ^a ±1.005 [25.59]	22.34 ^a ±1.605 [47.36]	25.36 ^a ±2.816 [67.28]
10 Gy	15.85 ^a ±0.115 [4.55]	16.08 ^a ±.188 [6.07]	18.43 ^a ±.517 [21.57]	23.51 ^a ±.376 [55.08]	24.95 ^a ±0.312 [64.58]
20 Gy	15.48 ^a ±0.376 [2.11]	15.73 ^a ±.004 [3.76]	16.89 ^b ±.674 [11.41]	23.07 ^a ±.992 [52.17]	18.56 ^b ±0.875 [22.43]
40 Gy	13.51 ^b ±0.549 [10.88]	15.86 ^a ±.919 [4.62]	16.68 ^b ±.523 [10.02]	17.89 ^b ±.460 [18.01]	16.04 ^b ±0.536 [5.80]
L.S.D 0.05	1.064	1.370	1.333	2.241	2.837
P	** 0.0044	ns 0.8982	** 0.0086	** 0.0014	*** 0.0001

[] : Values between brackets reflect the percentages from the initial values.

a, b, c, d : Significant among doses.

± : Standard deviation.

2- Effect of different storage periods and gamma irradiation doses on glycoalkaloids of potato tubers irradiated before and after planting:

Tables (3) and (4) showed the effect of different storage periods on total glycoalkaloids (β -solanine and β -chaconine) of potato tubers resulted from irradiation before planting (0, 10, 20 and 40 Gy) and irradiation after planting (0, 80, 160 and 320 Gy). The storage of both cases represented that total glycoalkaloids increased with the prolongation time of storage for non-irradiated and irradiated samples. In general, the longer the storage time, the higher the glycoalkaloid levels. Olsson and Roslund (1994) in a 9-month storage study of several clones kept at 4°C found that glycoalkaloid levels fluctuated over time, sometimes rising and sometimes dropping in a few instances even falling below original levels. Wunsch and Munzert (1994) studied the effects of 6-month storage at 4°C on glycoalkaloid distribution in five different cultivars. Although there was some variation in the cultivars, the general trend was a slight reduction in glycoalkaloids over time. The results follow up of such increasing in the total glycoalkaloids, accompanying with raising greenish chlorophyll. Moreover; it was found that the rate of total glycoalkaloids production increased by increasing gamma irradiation by approximately 2-folds of the control. While in the second method of storage, (irradiation after planting); the rate of increasing total glycoalkaloids was much higher than the first method. Therefore; the first method of storage (irradiation before planting) is more suitable and predominant than the second method due to the lower content of total glycoalkaloids in potato tubers irradiated before planting. The optimum conditions in this work were gamma irradiation at 20 Gy and storage at 60 days for potato tubers irradiated before planting, where the glycoalkaloid level was less amount. The glycoalkaloids concentration remained lower than the recommended food safety level 200 mg/kg⁻¹ fresh tubers (Liliana, *et al.* 2000).

Table (3): Effect of different storage periods on glycoalkaloids (β -solanine and β -chaconine) [mg/100 g F.wt.] of potato tubers irradiated (0, 10, 20 and 40 Gy) as seed potatoes before planting.

Treatment	Storage Periods								
	0 days			60 days			120 days		
	β -s	β -c	Total	β -s	β -c	Total	β -s	β -c	Total
Control	5.40	2.24	7.636	14.81	8.89	23.699	28.95	17.79	46.727
(s : c)			71 : 29			62 : 38			62 : 38
20 Gy	5.19	7.05	12.231	19.77	14.06	33.825	80.40	12.38	92.785
(s : c)			42 : 58			58 : 42			87 : 13
40 Gy	21.86	9.86	31.716	58.85	28.81	87.667	22.88	174.50	197.402
(s : c)			69 : 31			67 : 33			12 : 88

- (s : c): Ratio β -solanine to β -chaconine in total glycoalkaloids.

- β -s : β -solanine. - β -c : β -chaconine. - Total : Total glycoalkaloids.

Table (4): Effect of different storage periods and gamma irradiation doses on glycoalkaloids (α -solanine and α -chaconine) [mg/100 g F.wt.] of potato tubers irradiated (0, 80, 160 and 320 Gy) after planting.

Treatment	Storage Periods								
	0 days			60 days			120 days		
	α -s	α -c	Total	α -s	α -c	Total	α -s	α -c	Total
Control	5.40	2.24	7.636	14.81	8.89	23.699	28.95	17.79	46.727
(s : c)			71 : 29			62 : 38			62 : 38
80 Gy	30.56	17.31	47.874	65.00	71.30	136.303	230.60	107.55	338.138
(s : c)			64 : 36			48 : 52			68 : 32
160 Gy	160.90	16.545	177.445	202.95	191.8	394.733	189.00	211.90	400.937
(s : c)			91 : 9			51 : 49			47 : 53
320 Gy	173.00	40.43	213.451	293.8	105.4	399.221	289.00	217.15	506.115
(s : c)			81 : 19			74 : 26			57 : 43

- (s : c): Ratio α -solanine to α -chaconine in total glycoalkaloids.
 - α -s : α -solanine. - α -c : α -chaconine. - Total : Total glycoalkaloids.

Figure (3) showed that in most cases the improved varieties had a significantly higher percentage of α -solanine compared to the controls. It is important to note that α -chaconine is more embryo toxic and has a greater ability to induce liver enzymes than α -solanine (Caldwell, *et al.* 1991 and Friedman, *et al.* 1991). The toxicological potency of α -chaconine has been evaluated to be higher than that of α -solanine, which means that α -chaconine may have more effect in cases of potato poisoning (Toyoda, *et al.* 1991). It may therefore; be better to use potato varieties with high α -solanine content more than that of α -chaconine content to enhance food safety (Friedman, M. and Dao, L. 1992), so, in this work potato tubers can be stored for more expanded time > 60 days up to 120 days; using gamma irradiation dose effects at 20 Gy before planting, where α -solanine / α -chaconine ratio was 87:13 with less content of α -chaconine percentage

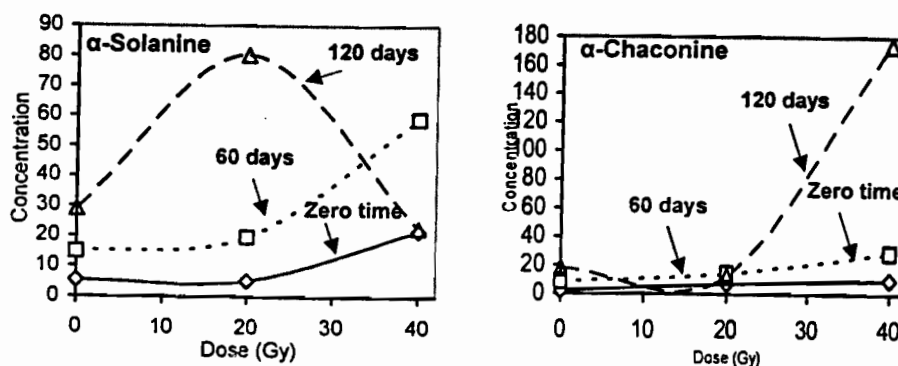


Fig. (3): Effect of gamma irradiation doses on glycoalkaloids α -solanine and α -chaconine mg/100 g F.wt. of irradiated potato tubers (0, 10, 20 and 40 Gy) at different storage periods.

The results in fig. (4) showed the reverse behaviour, where the percentage of α -chaconine is much higher content than α -solanine, which influence on the quality of potato tubers due to the hazard effects on the man healthy. There are many factors affected on glycoalkaloid compounds by different ratios from α -solanine and α -chaconine such as time, light energy, temperature and gamma irradiation doses (Dale, *et al.* 1997). Increasing or decreasing the ratio percentages of α -solanine and α -chaconine depends on their chemical structure by the above affecting factors (Olsson and Roslund, 1994).

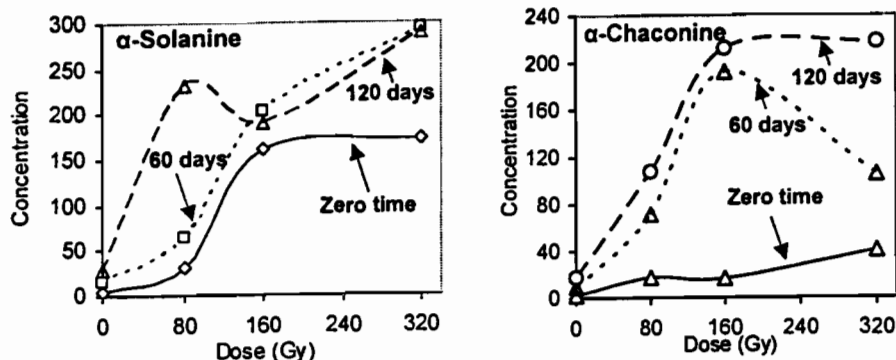


Fig. (4): Effect of gamma irradiation doses on glycoalkaloids α -solanine and α -chaconine mg/100 g F.wt. of irradiated potato tubers (0, 80, 160 and 320 Gy) at different storage periods.

- Effect of different storage periods and gamma irradiation doses on chlorogenic acid of potato tubers irradiated before and after planting:

Table (5) and fig. (5) showed the effect of storage on non-irradiated and irradiated tuber potatoes (0, 10, 20 and 40 Gy) at different storage periods 0, 30, 60, 90 and 120 days. It was found that; chlorogenic acid content increased with increasing the storage time for non-irradiated and irradiated tuber potatoes at fixed ambient temperature. These results can be explained by the fact that; increasing in chlorogenic acid levels during storage, may be linked to sugar accumulation that is known to occur at storage ambient temperature (Dale and Mackay 1994).

On the other hand; it was also observed that; the increase in chlorogenic acid levels during storage with increasing the irradiation dose done for seeds potatoes before planting was up to 20 Gy. This result due to the activation of phenolase enzyme which is responsible for transformation phenolic compounds to chlorogenic acid. While; at 40 Gy, chlorogenic acid was caused by a reduction in synthesis and also, as a result of accelerated conversion of chlorogenic acid to lignin (Pendhankar and Nair 1987 and Ramamurthy, *et al.* 1992).

Table (5): Effect of different storage periods on chlorogenic acid of potato tubers before planting at different gamma irradiation doses.

Treatment	Chlorogenic acid (mg /100 g d.wt.)				
	Storage periods (day)				
	0	30	60	90	120
Control	8.8 ^b ±1.312 [0.00]	12.4 ^a ±1.153 [40.91]	13.6 ^b ±0.889 [45.55]	19.7 ^b ±0.3 [123.9]	28 ^b ±0.656 [218.2]
10 Gy	9.3 ^a ±0.2 [5.68]	10.8 ^a ±1.058 [22.73]	11.6 ^c ±0.625 [31.82]	21.4 ^a ±0.2 [143.2]	32 ^a ±0.2 [263.6]
20 Gy	10.4 ^{ab} ±0.557 [18.18]	11.8 ^a ±0.1 [34.09]	15.8 ^a ±0.656 [79.55]	17.8 ^c ±0.173 [102.27]	31.6 ^a ±0.9 [259.1]
40 Gy	8.2 ^b ±0.1 [6.82]	8.6 ^b ±0.819 [2.27]	10.2 ^d ±0.4 [15.91]	12.6 ^d ±0.2 [43.18]	17.4 ^c ±0.7 [97.73]
L.S.D 0.05	1.358	1.666	1.252	0.421	1.252
P	* 0.031	** 0.004	*** 0.000	*** 0.000	*** 0.000

[] : Values between brackets reflect the percentages from the initial values.
a, b, c, d : Significant among doses. ± : Standard deviation.

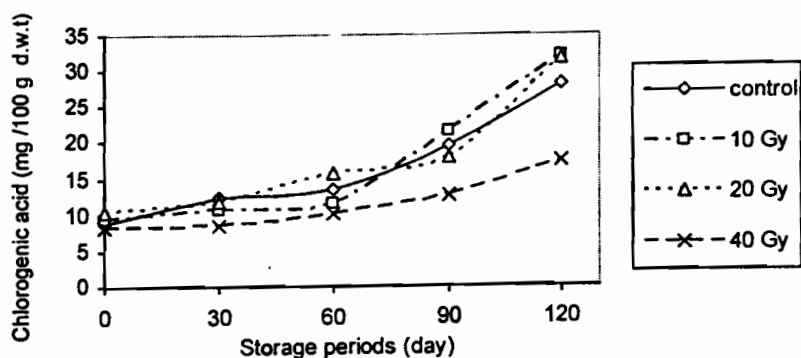


Fig. (5): Effect of different storage periods on chlorogenic acid of potato tubers before planting at different gamma irradiation doses.

Table (6) and fig. (6) showed the effect of gamma irradiation on chlorogenic acid of irradiated potato tubers during different storage periods. At first, one must be recognized that, zero storage periods in days mean the yield at 100 days of growth time; where their samples are non-irradiated tuber potato planting. It was found that chlorogenic acid levels increase with increasing storage time 0, 30, 60, 90 and 120 days and also; with each irradiated sample. These results referred that the storage time factor was more influenced than of gamma irradiation dose factor. This can be attributed to; at the storage factor, the rate of chlorogenic acid synthesis was more than the rate of lignin formation. But in other side; the factor of gamma irradiation effects within storage reduced the chlorogenic acid levels with increasing gamma irradiation doses. The later result can be explained that in non-irradiated tubers anabolism of chlorogenic acid was more active than its catabolism, and hence the chlorogenic acid content was maintained at a

4- Effect of different storage periods and gamma irradiation doses on Peroxidase activity of potato tubers irradiated before and after planting:

Table (7) and figure (7), illustrated the effects of gamma irradiation on peroxidase activity of potato tubers irradiated as seed potatoes during different storage periods. It was found from the results that; with perolongated the storage time, peroxidase activity will reduced and, also its activity decreases with increasing gamma irradiation dose ranged between (10-40 Gy).

Such value ranged from 73.5 to 61.2 U/ml min⁻¹ at zero time storage, compared to non-irradiated (control) at 67.3 U/ml min⁻¹. On the other side; by increasing gamma irradiation on potato tuber seeds followed by storage; the yield of potato tubers, can be stored with a minimum loss of peroxidase activity 20.3% for tubers at 10 Gy and highest period 120 day were the optimum radiation condition to keep the potato tubers for the longest storage period, where the peroxidase activity has a most value which involved with H₂O₂. The purified anionic enzyme readily formed dehydrogenative polymers from ferulic acid in the presence of H₂O₂ that are characterized by a high level of cross-linking (potentially through side chain C-8) and a high degree of retention of side chain unsaturation. The anionic peroxidase isoform of potato in the polymerization of the poly(aromatic) domain during suberization, (Mark, et al. 1999 and Converso and Fernandes, 1996). potato tubers have irradiation dose more than 10 Gy are less resistant under storage; because inactivation of peroxidase function due to decrease the amount of H₂O₂.

Table (7): Effect of different storage periods on peroxidase activity of potato tubers before planting at different gamma irradiation doses.

Treatment	Peroxidase activity (U / ml. min ⁻¹)				
	Storage periods (day)				
	0	30	60	90	120
Control	67.3 ^{ab} ±0.2 [0.00]	49.5 ^c ±3.559 [26.45]	40.3 ^d ±0.265 [40.11]	32.1 ^d ±3.551 [52.30]	25.6 ^d ±1.808 [61.96]
10 Gy	73.5 ^a ±4.694 [9.21]	70.2 ^a ±1.114 [4.41]	65.8 ^a ±1.778 [2.23]	59.7 ^a ±0.964 [11.29]	53.6 ^a ±0.2 [20.36]
20 Gy	69.7 ^a ±5.102 [3.57]	62.9 ^b ±0.265 [6.54]	54.3 ^b ±3.315 [19.32]	49.1 ^b ±2.007 [27.04]	43.8 ^b ±0.1 [34.92]
40 Gy	61.2 ^b ±0.1 [9.06]	54.7 ^c ±4.851 [18.72]	44.7 ^c ±0.557 [33.58]	38.5 ^c ±0.2 [42.79]	32.6 ^c ±0.2 [51.56]
L.S.D 0.05	6.531	5.766	3.589	3.951	1.726
P	* 0.0146	*** 0.0002	*** 0.0000	*** 0.0000	*** 0.0000

[] : Values between brackets reflect the percentages from the initial values.

a, b, c, d : Significant among doses.

± : Standard deviation.

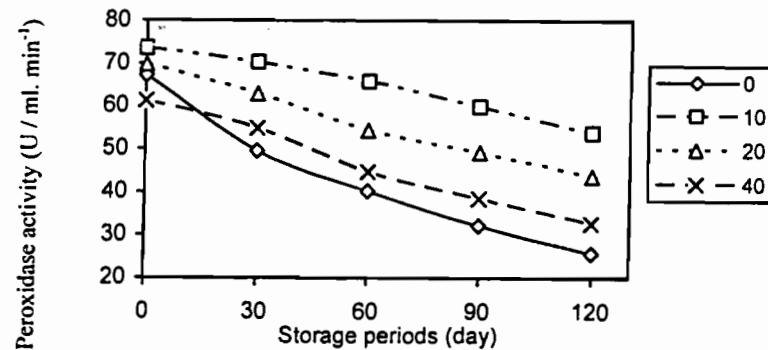


Figure (7): Effect of different storage periods on peroxidase activity of potato tubers before planting at different gamma irradiation doses.

The Effect of gamma radiation and different storage periods (0, 30, 60, 90 and 120 days) on Peroxidase activity of irradiated potato tubers (0, 80, 160 and 320 Gy) were shown in table (8) and figure (8). It was found that the inactive peroxidase enzyme increased to about 62% with increasing the storage time at 120 days, with respect to the non-irradiated tuber (control). Also, on the other hand, the same effect of gamma radiation on the inactive peroxidase; where it reached to about 74.4% inactivation at 120 days and irradiation doses 320 Gy. These results have shown that the peroxidase enzyme responsible for browning colour and tuber lignification, can be selectively inactivated by combined increasing gamma irradiation doses and storage periods. The results proved that first method was more predominant than the second one. The inactivation percentage of peroxidase enzyme in the first method was about 20.3% at radiation dose 10 Gy, while the other method showed much higher inactivation at the same period of storage 120 days which is 74.4% and much costs of gamma radiation dose 320 Gy. To understand the mechanism involved in this paradoxical situation; the activity of cinnamyl alcohol dehydrogenase (CAD), a specific enzyme involved in lignification, showed a positive correlation with phenylalanine ammonia-lyase (PAL) activity and lignin biosynthesis in potatoes. One possible reason for this could be the ineffective utilization of transcinnamic acid by gamma-irradiated tuber tissues, a view also expressed in a study on biochemical aspects of gamma-irradiated potatoes by Pendharkar and Nair (1987). Another possibility could be the preferential channelling of transcinnamic acid by secondary pathways to flavonoids and anthocyanins. Peroxidase has been implicated in lignification and suberisation during potato tuber storage. Reduced accumulation of lignin, accompanied as a result of gamma irradiation, may affect the long term storage stability of irradiated potatoes Ramamurthy, *et al.* 2000. Generally, the extent of the inactivation increased as the gamma radiation, temperature and holding time were collectively increased. For both enzymes, temperature appeared to have had the most marked effect, resulting in the loss of activities. These enzymes responsible for colour degradation and flavour loss can be selectively inactivated by

combined gamma radiation, temperature and holding time treatment. This could be of significance for commercial processing aiming to produce a minimally processed product with a nearly fresh quality and extended storage (Laurie and Carl (2000)).

Table (8): Effect of different storage periods on peroxidase activity of potato tubers after planting at different gamma irradiation doses.

Treatment	Peroxidase activity (U / ml. min ⁻¹)				
	Storage periods (day)				
	0	30	60	90	120
Control	67.3 ^a ±0.2 [0.00]	49.5 ^a ±3.559 26.45]	40.3 ^a ±0.264 40.11]	32.1 ^a ±3.55 52.30]	25.6 ^a ±1.808 [61.96]
80 Gy	53.2 ^b ±3.251 [20.95]	41.7 ^b ±0.4 [38.04]	34.3 ^b ±0.264 49.03]	28.1 ^b ±0.173 58.25]	23.5 ^b ±2.951 65.08]
160 Gy	45.7 ^c ±0.2 32.09]	38.8 ^c ±5.384 42.35]	31.2 ^c ±2.931 53.64]	25.3 ^c ±0.889 62.41]	21.1 ^c ±0.458 [68.65]
320 Gy	38.4 ^d ±0.5292 [42.94]	32.9 ^d ±3.345 [51.11]	26.5 ^d ±2.571 [60.62]	20.7 ^d ±0.3 [69.24]	17.2 ^d ±1.212 74.44]
L.S.D 0.05	3.112	6.854	3.687	3.462	3.479
P	*** 0.0000	** 0.0035	*** 0.0002	*** 0.0004	** 0.0029

[] : Values between brackets reflect the percentages from the initial values.

a, b, c, d : Significant among doses.

± : Standard deviation.

REFERENCES

- A.O.A.C. (1965). Official Method of Analysis of the Association of Official Agriculture Chemists, 10th Edition, Washington, D.C., 200: 44.
- Bergers, W.W.A. (1981). Investigation of the contents of phenolic and alkaloid compounds of gamma irradiated potatoes during storage. *Food Chem*, 6: 47.
- Bernards, M.A. and N.G. Lewis (1998). The macromolecular aromatic domain in suberized tissue: a changing paradigm. *Phytochemistry*, 47: 915-933.
- Bernards, M.A.; M.L. Lopez; J. Zajicek and N.G. Lewis (1995). Hydroxycinnamic acid-derived polymers constitute the polyaromatic domain of suberin. *J. Biol Chem.*, 270: 7382-7386.
- Caldwell, K.A.; O.K. Grosjean; P.R. Henika and M. Friedman (1991). Hepatic ornithine decarboxylase induction by potato glycoalkaloids in rats. *Food Chem. Toxicol*, 29: 531-535.
- Converso, D.A. and M.E. Fernandez (1996). Ca² activation of wheat peroxidase: a possible physiological mechanism of control. *Arch Biochem Biophys*, 333: 59-65.
- Dale, M.F.B.; D.W. Griffiths; H. Bain and B.A. Goodman (1997). The effect of gamma irradiation on glycoalkaloid and chlorophyll synthesis in seven potato cultivars. *J. Sci. Food Agric.*, 75 (2): 141-147.

- Dale, M.F.B. and G.R. Mackay (1994). Inheritance of table and processing quality. Potato Genetics, eds Bradshaw J E and Mackay G R. CAB International, Oxon, UK, pp 285-315.
- Friedman, M. (1997). Chemistry, biochemistry, and dietary role of potato polyphenols. A review. *J. Agric. Food Chem.*, 45 (5): 1523-1540.
- Friedman, M. and L. Dao (1992). Distribution of glycoalkaloids in potato plants and commercial potato products. *J. Agric. Food Chem.*, 40: 419-423.
- Friedman, M. and C.E. Levin (1989). Composition of Jimsonweed (*Datura stramonium*) seeds. *J. Agric. Food Chem.*, 37: 998-1005.
- Friedman, M.; J.R. Raybum and J.A. Bantle (1991). Developmental toxicology of potato alkaloids in the frog embryo teratogenesis assay-Xenopus (FETAX). *Food Chem. Toxicol*, 29: 537-547.
- Galeazzi, A.A.M. and V.C. Sgarbieri (1981). Substrate specificity and inhibition of polyphenoloxidase (PPO) from a dwarf variety of banana (*Musa cavendishii*. L.). *J. Food Sci.*, 46: 1404-1406.
- Galeazzi, A.A.M., V.C. Sgarbieri and S.M. Constantinides (1981). Isolation, purification and physicochemical characterisation of polyphenoloxidase (PPO) from a dwarf variety of banana (*Musa cavendishii*. L.). *J. Food Sci.*, 46: 150-155.
- George, P. (1953). Intermediate compound formation with peroxidase and strong oxidising agents. *J. Biol. Chem.*, 201: 423-426.
- Gil, A.M.; M. Lopes; J. Rocha and C.P. Neto (1997). A ¹³C solid state nuclear magnetic resonance spectroscopic study of cork cell wall structure: the effect of suberin removal. *Int. J. Biol. Macromol*, 20: 293-305.
- Hespeels, L. (1978). Progress in food irradiation (Belgium). *In Food irradiation information, No.9.*
- Hetherington, C.H. and K.F. MacQueen (1963). Results of the Canadian potato irradiation programme. *Proc. Japan Conf. Radio-isotopes*, 5: 132.
- Hoepfner, W. (1932). Zwei neue Reaktionen für Kaffesäure und Chlorogensäure. *Chem. Ztg.*, 59: 991.
- Kim-JaeSung; Lee-HaeYoun; Baek-MyungHwa; Kim-JaeHo; Kim-SungYeul; Kim-JS; Lee-HY; Baek-MH; Kim-JH; Kim-SY (2002). Effects of low dose gamma radiation on the dormancy, growth and physiological activity of seed potato (*Solanum tuberosum* L.). *J. Korean Society for Hort.Sci.*, 43 (5): 596-602.
- Laurie, M.D. and J.S. Carl (2000). Combined effect of high pressure, temperature and holding time on polyphenoloxidase and peroxidase activity in banana (*Musa acuminata*). *J. Sci Food Agric.*, 80: 719-724.
- Liliana, B.; M. Eric; D. André; E. Nelly; C. Enrique; L. Yvan; C. Enrique and L. Yvan (2000). Glycoalkaloids in potato tubers: the effect of variety and drought stress on the β -solanine and β -chaconine contents of potatoes. *J. Sci. Food Agric.*, 80: 2096-2100.
- Mark, A.B.; D.F. Warren; B.L. David; P. Ronny; Y. Xiaolong; S.Anita and L.P. Guy (1999). Biochemical characterization of the suberization-

- associated anionic peroxidase of potato. *Plant Physiology*, 121: 135-145.
- Mendel, F. and D. Lan (1992). Distribution of glycoalkaloids in potato plants and commercial potato products. *J. Agric. Food Chem.*, 40 (3): 419-423.
- Mendel, F.; D. Lan and R.G. Michael (1989). Ergot alkaloid and chlorogenic acid content in different varieties of morning-glory (*Ipomoea spp.*) seeds. *J. Agric. Food Chem.*, 37: 708-712.
- Mondy, N. I. and B. Gosselin (1988). Effect of peeling on total phenols total glycoalkaloids, discoloration and flavor of cooked potatoes. *J. Food Sci.*, 53: 756-759.
- Negrel, J.; B. Pollet and C. Lapierre (1996). Ether-linked ferulic acid amides in natural and wound periderms of potato tuber. *Phytochemistry*, 43: 1195-1199.
- Nell, I.M. and G. Barry (1989). Effect of irradiation on discoloration, phenols and lipids of potatoes. *J. Food Sci.*, 54 (4): 982-984.
- Olsson, K. and C.A. Roslund (1994). Changes in glycoalkaloid content of potato during long-term storage. In: Abstracts from the joint meeting of the Agronomy and Utilization sections of the *European Association of Potato Research*, 41: 24-30.
- Orabi, A.I. (1985). Effect of gamma rays on some properties of stored potatoes. *Ph.D. Thesis. Fac. of Agric., Ain Shams, Univ. Cairo, Egypt.*
- Orsak, M.; J. Lachman; M. Vejdova; V. Pivec and K. Hamouz (2001). Changes of selected secondary metabolites in potatoes and buckwheat caused by UV, gamma- and microwave irradiation. *Rostlinna Vyroba*, 47 (11): 493-500.
- Pendharkar, M.B. and P.M. Nair (1987). Alteration in phenylpropanoid metabolism in gamma irradiated potatoes. *Potato Research*, 30: 589-601.
- Pendharkar, M.B. and P.M. Nair (1995). A comparative study of phenylpropanoid metabolism in gamma irradiated and non-irradiated potato tubers. *Potato Research*, 38 (2): 187-198.
- Ramamurthy, M.S.; B. Maiti; P. Thomas and P.M. Nair (1992). High-performance liquid chromatography determination of phenolic acids in potato tubers (*Solanum tuberosum*) during wound healing. *J. Agric. Food Chem.*, 40 (4): 569-572.
- Ramamurthy, M.S.; K.K. Ussuf; P.M. Nair and P. Thomas (2000). Lignin biosynthesis during wound healing of potato tubers in response to gamma irradiation. *Postharvest Biology and Technology*, 18: 267-272.
- Schreiber, L. (1996). Chemical composition of Casparian strips isolated from *Clivia miniata* Reg. roots: evidence for lignin. *Planta*, 199: 596-601.
- Sharabash, M.; I.O. Orabi; I.I. Eloksh; M.A. Abd-Ali; M.F. Barakat and S.M. El-Mashri (1995). Studies on the storage of irradiated potatoes. *Proceedings of the second Arab conference on the peaceful uses of atomic energy. Part II: A and B. Oct 1995*, pp. 969.
- Sinden, S.L. and R.E. Webb (1972). Effect of variety and location on the glycoalkaloid content of potatoes. *Am. Potato J.*, 49: 334-338.

- Spinks, J.W.T.; R.J. Woods; W.E. John and N.Y. Sons (1976). "An Introduction to Radiation Chemistry." J. Wiley, New York, Illus., Xiv, 504 p.
- Thomas, P. (1984). Radiation preservation of foods of plant origin. Part 1. Potatoes and other tuber crops. *CRC Crit Rev. Food Sci. Nut.*, 19: (4), 327-379.
- Toyoda, M.; W.D. Rausch; K. Inoue; Y. Ohno; Y. Fujiyama; K. Takagi and Y. Saito (1991). Comparison of solanaeous glycoalkaloids evoked Ca influx in different types of cultured cells. *Toxicol. In Vitro*, 5: 347-351.
- Van Gelder, W.M.J. (1990). Chemistry, toxicology and occurrence of steroidal glycoalkaloids: potential contaminants of the potato. In: *Poisonous Plants Containing Edible Plants*, ed Rizk A-F. CRC Press, Boca Raton, Florida, USA, pp 117-156.
- Winchester, R.V.; A.C. Thomas and H.T. Brodrick (1976). Reduction of greening in potatoes by gamma irradiation. *Agroplantae*, 8: 95-96.
- Wunsch, A. and M. Munzert (1994). Effect of storage and cultivar on the distribution of glycoalkaloids in potato tuber. *Potato Res.*, 37: 3-10.
- Zeier, J. and L. Schreiber (1997). Chemical composition of hypodermal and endodermal cell walls and xylem vessels isolated from *Clivia miniata*: identification of the biopolymers lignin and suberin. *Plant Physiol.*, 113: 1223-1231.

تأثيرات أشعة جاما على بعض التغيرات البيوكيميائية لدرنات البطاطس على فترات تخزين مختلفة.

- أمام عبد المبدى عبد الرحيم* - مجدى عبد العليم محمد شعلان* - إبراهيم عرابي احمد** -
عمر إبراهيم صالح
* قسم الكيمياء الحيوية - كلية الزراعة جامعة القاهرة.
** المركز القومي للبحوث وتكنولوجيا الأشعاع - هيئة الطاقة الذرية.

تم دراسة تأثير أشعة جاما على درنات البطاطس صنف " ألفا " قبل وبعد الزراعة بطريقتى تخزين مختلفة، عند جرعات إشعاعية (صفر، ١٠، ٢٠، ٤٠ جراى) بالطريقة الأولى قبل الزراعة وكذلك (صفر، ٨٠، ١٦٠، ٣٢٠ جراى) بالطريقة الثانية بعد الزراعة، وكذلك تأثير فترات التخزين (صفر، ٣٠، ٦٠، ٩٠، ١٢٠ يوم) على بعض المركبات البيوكيميائية مثل المركبات الفينولية الكلية وحامض الكلورجنيك والإنزيم البيروكسيدى ومركبات الجليكوكالويد (ألفا-سولانين و ألفا-شاكونين). وقد تم تقدير تركيزات تلك المركبات باستخدام أجهزة التقنين الكيميائية مثل جهاز التحليل الطيفى للمركبات الثلاثة الأولى وإستخدام جهاز التحليل الكروماتوجرافى السائل على الفصل (HPLC) فى تقدير تركيزات مركبات الجليكوكالويد (ألفا-سولانين و ألفا-شاكونين). وقد تبين من النتائج بأن طريقة التخزين الأولى أفضل من الثانية نتيجة لأنخفاض تركيز تلك المركبات. ومن ناحية أخرى أتضح أن أفضل جرعة إشعاعية لدرنات البطاطس المعرضة للإشعاع قبل الزراعة، هى تلك التى تتراوح ما بين ١٠ إلى ٢٠ جراى والتسى أدت لأطالسة فترة تخزين البطاطس لأطول فترة ممكنة تقترب من ١٢٠ يوم، ويرجع ذلك لتأثير أشعة جاما على النشاط الأنزيمى المؤثر فى تكوين تلك المركبات. وأتضح أيضا من نتائج التحليل الكروماتوجرافى HPLC لمركبات الجليكوكالويد (ألفا-سولانين و ألفا-شاكونين) بأنه يمكن تخزين درنات البطاطس بالطريقة الأولى لأكثر من ٦٠ يوم وتمتد إلى ١٢٠ يوم، وذلك بإستخدام جرعة أشعاعية ٢٠ جراى، حيث ينخفض تركيز ألفا-شاكونين إلى أقل مايمكن (١٣%) وأيضاً أقل تأثير سمي على صحة كبد الأتسان.