Effect of Exogenous Salicylic Acid on the Antioxidant System of Safflower Plant Exposed to Gamma Irradiation

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***Abstract*—**The present investigation was embraced to look at the impacts of exogenous utilization of Salicylic acid (SA) on non-antioxidant enzyme activity in safflower Carthamas tinctorius L. exposed to γ- irradiation. In a pot trial, SA was applied foliar at the concentrations (0, 100 and 200)µm with gamma ray at the doses (0,50,100 and 150) Gy, antioxidant system activities Ascorbic acid (AsA),α-Tocopherol(α-TOC),Glutathione(GSH), Proline (Pro) and Carotenoid(Car) acitivity of plants were recorded to project the impacts of these treatments.. The outcomes demonstrated that increment in Salicylic acid (SA) concentricity from (0 to 200)µm initiated critical increment in Glutathione, Proline and Carotenoid contents except Ascorbic acid , α- Tocopherol were decreased . Under all levels of gamma ray (0,50,100 and 150) Gy may increase AsA, α-TOC, GSH, Pro and Car activity. the influence of the interaction between gamma ray and Salicylic acid at 150 Gy and 200µm . respectively, was positive givin the highest values in antioxidant system activity.

***Keywords*—** Salicylic acid, gamma ray, antioxidant system, Ascorbic acid (AsA), α- Tocopherol(α-TOC), Glutathione(GSH), Proline (Pro) and Carotenoid(Car).

I. INTRODUCTION

Safflower (Carthamus tinctorius L.) is an organ from the family Compositae or Asteraceae (Weiss, 2000), developed mostly for its seed, which is utilized as palatable oil, birdseed or for its blooms, utilized as color sources and curative purposes (Dordas and Sioulas, 2008; Ekin, 2005; Istanbulluoglu, 2009; Emongor, 2010; Emongor and Kedikanetswe, 2015). safflower oil as a rich origin of monounsaturated unsaturated fat oleic acid 80% and polyunsaturated basic unsaturated fat linoleic acid 87% (Aghamohammadreza et al., 2013; Murthy and Anjani, 2008).

Linoleic acid has been appeared offer wholesome and restorative advantages, for example, anticipation of arteriosclerosis , coronary illness , hyper lipaemia and hypertension (Cosge et al., 2007; Wang and Li, 1985). The seeds of safflower are additionally a rich wellspring of vitamins (β-carotene , thiamine, tocopherols α, β and γ) and minerals (Mn , Zn, Cu and Fe) (Velasco et al., 2005).

Salicylic acid (SA) is an endogenous development controller of phenolic nature and furthermore a significant signaling particle which directs physiological operations in plants, for example, improve development and photosynthesis rate, advance ethylene creation, nitrate reduction, heat generation and Stimulate blossoming (Nazar et al. 2011, Hayat et al., 2010, Syeed et al. 2011, Khan et al. 2014, Hasanuzzaman et al. 2017). SA directs both proline, glycinebetaine , N assimilation, regulate antioxidant system just as glyoxalase compounds and plant-water associations in environmental stress (Alam et al. 2013 , Nazar et al. 2011, Miura and Tada. 2014). (SA) activity is firmly identified with the production of different free radical (Kawano and Muto. 2000). It is also assumes a major job in improving the endogenous cell reinforcement levels in plants which brought about better insurance again oxidative pressure (Hayat et al. 2010; Kadioglu et al. 2011). The cell antioxidant systems comprise of non-enzymatic parts including vit C, E and glutathione (GSH) and antioxidant enzymes creating or searching H2O2, for example, catalase, superoxide dismutase, ascorbate peroxidase, guaiacol glutathione reductase , peroxidase (Asada, 1999; Foyer and Noctor, 2011).. In an later (Hasanuzzaman et al. 2014) watched exogenous utilization of (SA) is a compelling protectant in improving the exercises of both glyoxalase and antioxidant defense and furthermore expanded GSH and AsA content by 39% and 48 %, respectively and upgraded GSH/GSSG ratio by 47% contrasted with salt stress exclusive. Accordingly, MDA and H2O2 substance diminished by 31% and 39 % respectively. Gamma irradiation was caused to vitalize oxidative stress with overproduction of reactive oxygen species ROS for instance, superoxide radicals (O2-), hydrogen peroxides (H2O2) and hydroxyl radicals (OH-) . Xienia et al (2000). γ-ray are in charge of causing various adjustments in physiology and biochemical properties of plants at different dosages, and irritate hormonal equalization, , lipids, proteins, enzymatic action , nucleic acids and leaf alternate (Kiong et al. 2008; Salter and Hewitt. 1992) .In an ongoing report (Ivanova. 2017) set up that seeds of which were pre-sowing treated with various dosages of γ-radiation (50, 100 and 150)Gy, had higher antioxidant activity than the concentrates of safflower leaves got from untreated seeds. As far as we could possibly know, there are insufficient information about the impact of (SA) treatment on non- antioxidantenzyem activity. In this way the target of our examination was to watch the role of (SA) in actuality the exercises of ascorbic acid ,α-tocopherol , glutathione , proline and carotenoid under Gamma irradiation stress condition in safflower leaves.

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II. Materials And Methods

during the developing periods of 2018 . A pot examination was completed at the green house. In silt loam soil and put in a plastic pots limit of 5 Kg. Safflower seeds, were acquired from the general specialist for Agriculture Research Abu Ghraib, Baghdad , Iraq. Seeds were illuminated with various doses of gamma rays (0, 50 , 100 and 150) Gy . γ- irradiation was performed utilizing a (gamma chamber 900) device prepared with a Co- 6o γ source at Department of Physics , College Science , University of Baghdad. Tests without gamma rays filled in as the control. Each pot was treated with various salicylic acid (0 ,50 and 100) µm. A foliar of (SA) multiple times (at10-days interims) was connected to the plants. The last reap was performed 40 days after the beginning of treatments. Every single plastic pot were flooded with faucet water till 66% of field limit week by week till the finish of experiment.

A. Ascorbate extraction and determinatins:

Ascorbic acid content is determined by the method adopted by Hussain et al (2010). Precisely weighted (1g) of each sample in a(25 ml) conical flask include (10 m)l of oxalic acid (50mM) solution and the samples was placed under shade for 24 h for extraction of Vit C contents. After 24 h,the tests were separated throught a (0.45µm) channel paper. At that point 2.5 ml of each example was moved to a separate (25 ml) volumetric darker cup, 2.5 ml of oxalic acid (50 mM) solution. At that point included independently meta phosphoric acid with acetic acid (0.5 ml), sulphuric acid (v/v 5%) solution. (1 ml) and ammonium molybate solution (2 ml) in each volumetric darker flask and make up the volume 25 ml with distilled water. Each sample was then analysed for Vit C at (760 nm) compared with the standard L-ascorbic acid.

B. Determination of proline:

proline was measured after the record of Bates et al. (1973). Around (500 mg) of plant tissue was homogenized in3% (5 ml) of fluid sulphosalicylic acid.. (2 ml) of the supernatant was heated with (2 ml) of glacial acetic acid and (2 ml) of acid ninhydrin and in a water bath at 100∘C for 1 h, and the reaction was ceased in an ice bath. For extraction (4 ml) of toluene was included, and tests were blended overwhelmingly for (15-20) s. Tests were then put aside to permit partition of the natural and watery phases. The absorbance was perused at (520 nm) in spectrophotometer utilizing toluene as a clear. Proline content was resolved from a standard curve and determined dependent on fresh weight.

C. Estimation of Carotenoids:

The example extricates with (Dimethyl sulphoxid) DMSO strategy (Hiscox and Israelstam.1979). Notwithstanding estimation of absorbance of the concentrate at (663) and (645) nm, spectrophotometer readings are likewise recorded at (480) nm wavelength. Carotenoid substance is determined after the formula (Price and Hendry. 1991) : given below:

Total carodenoids (mg.g fw-1)= [A480 + (0.114 ×A663) – (0.638 × A645)] × V / 1000 ×W

D. Estimation of glutathione:

Tests powder were separated in (6%) m-phosphoric acid pH 2.8 containing (1 mM) EDTA.The GSH substance of test concentrates was estimated by response with 5, 5'dithiobis - 2-nitrobenzoic (DTNB) reagent to give an intensify that absorbed at (412 nm).The glutathione concentration was resolved from a standard curve as indicated by Silber et al. (1992).

E. Estimation of α- Tocopherol

vit E were assessed on leaves by spectrophometry as recently depicted with certain adjustments (Leenheer et al. 1988) In this regard, 100 mg of leave removed from each powdered example as portrayed above, was weakened in an acetone/hexane mixture (70/30; v/v) , and complete vit E assurance did on an aliquot by estimating absorbance at 270 nm .in a spectrophotometer against a clear example (dissolvable). Standard curves made with unadulterated vit E were utilized for this reason and the outcomes communicated as mg vit E comparable per 100 g dried powder test.

F. Statistical Analysis

The statistical analyses were carried out using Genstat release (10.3 DE) Data obtained were analyzed statistically to determine using least significant difference (LSD) using one-way analysis of variance (ANOVA) at $p \leq 0.05$.

III. Results

Figure (1) show the effect of gamma radiation doses and salicylic acid in the activity of Ascorbic acid content The results showed significant increase over the control up to 188.63% at 150 Gy . as the results showed that ascorbic acid decreased from 27.57% at 200 µM SA pre-treated samples compared to control.Whil The interaction between dose 150 Gy and 200 µM SA was significant. The Vitamin E has the same behavior by givin a significant increase at 150 Gy. While significantly decreased at 200 µM SA pre- treated Whil The interaction between dose 150 Gy and 200 µM SA was significant (fig.2).

glutathione induction was significantly and positively correlated with the dose of irradiation. Asignificant ($P < 0.05$) increase in glutathione content was observed in safflower leaves of at all exposures of 150 Gy gamma was given the highest mean of content with an increase by 318.26% as compared with un-irradiated control (fig.3). the plants treated with SA increased the content of the glutathione compared with no treatment.The highest value was observed with 200 µM SA treatment an increase rate reached 105.86% compared to the non- salicylic acid treatment. The interaction between dose 150 Gy and 200 µM SA was high significant ($P < 0.05$) for glutathione.

Data related to Proline accumulation is given in (fig.4) Results showed that irradiation led to asignificant increase in Proline accumulation at 150 Gy gamma (180.63%) Compared with the non-exposure treatment of gamma rays . Proline was greatly affected by SA treatment and its content was the highest treatment with 200 µM SA an increase rate reached 22.50 %

compared to the non- salicylic acid treatment The interaction between dose 150 Gy and 200 μM SA was significant for Proline.

Exposure of safflower to 150 Gy significantly enhanced its carotenoid content by 85.49% over the control (fig.5). Application of SA especially 200 μM most accumulation which was especially significant by 26.48% of non- salicylic acid treatment. Gamma irradiation and salicylic acid enhanced carotenoid with maximum value of 0.395 mg.g fw-1at 150 Gy and 200 μM SA .

IV. Discussion

Gamma ray is add to prompt oxidative worry with formed of reactive oxygen species ROS for example superoxide radicals (O2–),hydroxyl radicals (OH–) and H2O2 (Apel and Hirt 2004). To stay away from oxidative harm, plants have advanced different defensive mechanisms to balance the impacts of ROS in cell plant (Foyer et al. 1994). One of the defensive components was the non-enzymatic system which works with the successive and synchronous activities of various non-enzymes, The outcomes demonstrated that non antioxidant enzymes exercises ascorbic acid,α-tocopherol , glutathione , proline and carotenoid altogether expanded in safflower leaves with expanding gamma dose (0-150 Gy).AsA has been appeared to have a fundamental job in a few physiological procedures in plants, including development and differentiation. (Foyer, 1993). Increment Ascorbic acid with Increment gamma ray appeared (fig.1). AsA is one of the most grounded antioxidants a wellspring of electrons for some enzymatic and non-enzymatic responses (Blokhina et al., 2003). Firmly associated with this antioxidative capacity of AsA is GSH which serves for the recovery of diminished AsA in the AsA-GSH cycle (Noctor and Foyer, 1998). This finding was in understanding with (Abdel Haleem et al. 2012) the consequences of (fig.2) outlined an Increment in α-tocopherol content when the safflower plant was presented to gamma ray. α-tocopherol as a an antioxidant deactivates photosynthesis inferred (ROS) and avoids the expansion in lipid peroxidation by rummaging lipid peroxyl radicals in thylakoid layers. The content of α-tocopherol change differentially because of ecological confinements, contingent upon the greatness of the stress and species affectability to stress. α-tocopherol considers a significant part of the plant protection hardware, which keeps up the respectability and typical capacity of the photosynthetic mechanical assembly (Liu et al., 2008). A similar outcome got in cowpea plants (Abdel Haleem et al.2012). On account of glutathione content, there were noteworthy increments (fig.3) by gamma ray at portion levels 0,50,100 and 150 Gy. glutathione is associated with the upkeep of the redox status in plant cells and organs (Horemans et al., 2000). A similar pattern likewise was accounted for by (Marchenko et al. 1996 ; Chakravarty and Sen 2001; Amina and Hossam 2010) The increment glutathione substance is a stress ensuring mechanism of plants uncovered gamma ray. The development of the reactive oxygen species is forestalled by a an antioxidant system including the action of low molecular mass antioxidants glutathione, α-tocopherols and ascorbic acid (Amina and Hossam 2010). Proline was considerble solutes that go about as osmoregulators by contributing in stress resistance, insurance, hydrophobicity, dynamic oxygen searching, and keeping up cell sap pH (Kuznetsov and Shevyakova 2007). Proline capacities to scavenge the ROS and goes about as a cytosolic osmoticum that aides in directing and balancing out different structure and capacities, for example membranes protein and DNA (Kishor et al. 2005). The increase in the substance of proline reflects its positive job in scavenge free radicals delivered by the gamma ray. Comparable discoveries were introduced in different plants uncovered gamma irratiation (Borzouei et al.2013; Wang et al.2017; Hamideldin et al.2017)..Gamma irratiation improved the contents of carotenoids, which were typically improved under stresses conditions to shield chlorophyll from photooxidative harm. Carotenoids secure the photosystemII by deactivating triplet chlorophyll and statemate the impact of singlet oxygen.(Jan et al. 2013;Abdel Haleem et al.2012; Abomohra et al.2016), they found that the Gamma ray improved the carotenoids content.

Salicylic acid lightens abiotic stress incited harm by eliciting oxidative stress, which improves the expression and activity of redox controlled antioxidant system (Ananieva et al. 2004; Li et al., 2013; Csiszar et al., 2014). In our study exogenous SA played significant role in enhancing non-enzymatic components. Our study indicated that AsA content significantly decreased (Fig. 1) with spray SA. The decreased activities of AsA regenerating enzymes (MDHAR and DHAR) impaired the production and regeneration of AsA in gamma stressed plants. The decrease in AsA content was also reported in our previous study (Hasanuzzaman et al. 2014 ; Umebese and Bankole. 2013) ASA and tocopherol, two of the most productive natural cell reinforcements realized they can powerful quencher of singlet oxygen (Bilski et al., 2000). In the present examination, the exogenous utilization of SA apparently diminished the α-tocopherol content under gamma ray. These outcomes are in concurrence with some prior discoveries (Noreen and Ashraf. 2010). In the present examination, activities of antioxidant glutathione in safflower plants were increased under gamma stress just as after SA application. activity Glutathione is a significant solvent antioxidant, since it ensures numerous cell components under oxidative stress. Glutathione assumes an pivotal job in the antioxidant defense system just as the glyoxalase by going about as a substrate or cofactor for specific enzymes (Fig. 3). The study also showed that the exogenous SA significantly increased the content of proline and carotenoid under Gamma irradiation(fig 4.5). Similar finding were presented in other plant treated with SA (Keshavarz et al.2016; Alam et al.2013; Umebese and Bankole.2013; Dong et al.2015; Shaki et al.2018).

References

[1] Abdel Haleem M.A. Mohammed, Heba I. Mohamed, Laila M. Zaki, Asmaa M. Mogazy. 2012. Pre-exposure to gamma rays alleviates the harmful effect of salinity on cowpea plants. Journal of Stress Physiology & Biochemistry, Vol. 8 No. 4 2012, pp. 199-217 ISSN 1997-0838.

[2] Abomohra AEF, ; El-Shouny W; Sharaf M; Abo-Eleneen M.2016. Effect of Gamma Radiation on Growth and Metabolic Activities of Arthrospira platensis. Braz. Arch. Biol. Technol. v.59: e16150476. https://doi.org/10.1590/1678-4324-2016150476

[3] Aghamohammadreza, M., Mirhadi, M. J., Delkhosh, B. and Omidi, A. (2013).Evaluation of native and exotic safflower (Carthamus tinctorius L.) genotypes for some important agronomic traits and fatty acid composition. Annals of Biological Research, 4(6): 200-204.

[4] Alam MM, Hasanuzzaman M, Nahar K, Fujita M. Exogenous salicylic acid ameliorates short-term drought stress in mustard (Brassica juncea L.) seedlings by upregulating the antioxidant defense and glyoxalase system. Australian Journal of Crop Science. 2013;7:1053–1063

[5] Amina A. Aly and Hossam E. S. El-Beltagi.2010. Influence of ionizing irradiation on the antioxidant enzymes of Vicia faba L. GRASAS Y ACEITES, 61 (3), JULIO-SEPTIEMBRE, 288-294, 2010, ISSN: 0017-3495, DOI: 10.3989/gya.111509.

[6] Ananieva EA, Christov KN, and Popova LP. 2004. Exogenous treatment with salicylic acid leads to increased antioxidant capacity in leaves of barley plants exposed to paraquat. Journal of Plant Physiology 161: 319–328. https://doi.org/10.1078/0176-1617-01022

[7] Apel, K. and H. Hirt, 2004. Reactive oxygen species: metabolism, oxidative stress and signal transduction. Annual Review of Plant Biology, 55: 373–399. https://doi.org/10.1146/annurev.arplant.55.031903.141701

[8] Asada, K. (1999). The water-water cycle in chloroplast, scavenging of active oxygen and dissipation of excess photons.Ann. Rev. Plant Mol. Biol., 50: 601-639. https://doi.org/10.1146/annurev.arplant.50.1.601

[9] Bates LS, Waldern RP, Teave ID (1973). Rapid determination of free proline for water stress studies. Plant Soil 39:205-207. https://doi.org/10.1007/BF00018060

[10] Bilski P, Li MY, Ehrenshaft M, Daub ME, Chignell CF (2000). Vitamin B6 and its derivatives are efficient singlet oxygen quencher and potential fungal antioxidants. Photochem. Photobiol. 71: 129-134. https://doi.org/10.1562/0031-8655(2000)071<0129:SIPVBP>2.0.CO;2

[11] Blokhina, O., E. Virolaineu and K. Fagerstedt,2003. antioxidant oxidative damage and oxygen deprivation stress. Ann. Bot., 91: 179-194. https://doi.org/10.1093/aob/mcf118

[12] Borzouei A, Kafi M, Sayahi R, Rabiei E, Amin PS (2013) Biochemical response of two wheat cultivars (Triticum aestivum L.) to gamma radiation. Pak J Bot 45(2):473–477.

[13] Chakravarty, B., and Sen, S. 2001. Enhancement of regeneration potential and variability by g-irradiation in cultured cells of Scilla indica. Biol. Plant. 44(2): 189–193. doi:10.1023/A:1010282805522.

[14] Cosge, B., Gurbuz, B. and Kiralan, M. (2007). Oil content and fatty acid composition of some safflower (Carthamus tinctorius L.) varieties sown in spring and winter. International Journal of Natural and Engineering Sciences, 1: 11-15.

[15] Csiszar J, Horvath E, Vary Z, Gall, Bela K, Brunner S, and Tarii. 2014. Glutathione transferase supergene family in tomato: Salt stress-regulated expression of representative genes from distinct GST classes in plants primed with salicylic acid. Plant Physiology and Biochemistry 78: 15–26. https://doi.org/10.1016/j.plaphy.2014.02.010

[16] Dong Y.J. ,. Wang Z.L, Zhang J.W., Liu S., He Z.L. , He M.R.2015. Interaction effects of nitric oxide and salicylic acid in alleviating salt stress of Gossypium hirsutum L. Journal of Soil Science and Plant Nutrition, 15 (3), 561-573.

[17] Dordas, C. A. and Sioulas, C. (2008). Safflower yield, chlorophyll content,photosynthesis, and water use efficiency response to nitrogen fertilization under rainfed conditions. Industrial Crops and Products, 27: 75-85. https://doi.org/10.1016/j.indcrop.2007.07.020

[18] Ekin, Z. (2005). Resurgence of safflower (Carthamus tinctorius L.) utilization: A global view. Journal of Agronomy, 4: 83-87. https://doi.org/10.3923/ja.2005.83.87

[19] Emongor, V. E. (2010). Safflower (Carthamus tinctorius L.) the underutilized and neglected crop: A review. Asian Journal of Plant Sciences, 9 (6): 299- 306. https://doi.org/10.3923/ajps.2010.299.306

[20] Emongor, V. E., Oagile, O. and Kedikanetswe, B. (2015). Effects of plant population and season on growth and development of safflower (Carthamus tinctorius L.) as an ornamental plant. Acta Horticulturae, 1077: 35-45. https://doi.org/10.17660/ActaHortic.2015.1077.3

[21] Foyer CH, and Noctor G. 2011. Ascorbate and glutathione: the heart of the redox hub. Plant Physiology 155: 2–18. https://doi.org/10.1104/pp.110.167569

[22] Foyer, C.H. 1993. Ascorbic acid. In Antioxidants in higher plants. Edited by R.G. Alscher, J.L. Hess. Boca Raton: CRC Press pp. 31–58. https://doi.org/10.1201/9781315149899-2

[23] Foyer, C.H., P. Decourvieres and K.J. Kunerk,1994. Protection against oxygen radicals: an important defense mechanism studied in transgenic plants. Plant Cell Environ., 17: 507-523. https://doi.org/10.1111/j.1365-3040.1994.tb00146.x

[24] Hamideldin N, Eliwa NE, and Hussein OS.2017. Role of Jasmonic Acid and Gamma Radiation in Alleviating Salt Stress in Moringa. Int. J. Agric. Biol., Vol. 19, No. 1, 2017. https://doi.org/10.17957/IJAB/15.0245

[25] Hasanuzzaman M, Alam MM, Nahar K, Mahmud JA, Ahamed KU, Fujita M. Exogenous salicylic acid alleviates salt stress-induced oxidative damage in Brassica napus by enhancing the antioxidant defense and glyoxalase systems. Australian Journal of Crop Science. 2014;8:631–639. https://doi.org/10.1155/2014/757219

[26] Hasanuzzaman M, Nahar K, Bhuiyan TF, Anee TA, Inafuku M, Oku H and Fujita M.2017. Salicylic Acid: An All-Rounder in Regulating Abiotic Stress Responses in Plants. http://dx.doi.org/10.5772/intechopen.68213.

[27] Hayat Q , S. Hayat, M. Irfan, A. Ahmad, Effect of exogenous salicylic acid under changing environment: a review, Environ. Exp. Bot. 68 (2010) 14–25. https://doi.org/10.1016/j.envexpbot.2009.08.005

[28] Hiscox, J.D. and israelstam, G.F. (1979). A method for extraction of chlorophyII from leaf tissue without maceration. Can. J. Bot. 57: 1332-1334. https://doi.org/10.1139/b79-163

[29] Horemans N, Foyer CH, Asard H. 2000. Transport and action of ascorbate at the plant plasma embrane. Trends in Plant Science 5, 263-267. https://doi.org/10.1016/S1360-1385(00)01649-6

[30] Hussain I, khan L, Khan M.A, Khan F.U, Ayaz S, Khan F.U . 2010 UV Spectrophotometric Analysis Profile of Ascorbic Acid in Medicinal Plants of Pakistan .World Applied Sciences Journal 9 (7):800-803.

[31] Istanbulluoglu, A. (2009). Effects of irrigation regimes on yield and water productivity of safflower (Carthamus tinctorius L.) under Mediterranean climatic condition.Agricultural Water Management, 96: 1792-1798. https://doi.org/10.1016/j.agwat.2009.07.017

[32] Ivanova Raisa.(2017). Antioxidant activity of safflower leaves and its modification by abiotic factors. Lucrări Ştiinţifice- vol.60(2)/2017, seria Agronomie.

[33] Jan S, Parween T, Hameed R, Tariq O. Siddiqi, and Mahmooduzzafar.2013. Effects of presowing gamma irradiation on the photosynthetic pigments, sugar content and carbon gain of Cullen corylifolium (L.) Medik. CHILEAN JOURNAL OF AGRICULTURAL RESEARCH 73(4). https://doi.org/10.4067/S0718-58392013000400003

[34] Kadioglu A, Saruhan N, Sağlam A, Terzi R, Acet T (2011) Exogenous salicylic acid alleviates effects of long term drought stress and delays leaf rolling by inducing antioxidant system. Plant Growth Regul. 64:27–37. https://doi.org/10.1007/s10725-010-9532-3

[35] Kawano T, Muto S (2000). Mechanism of peroxi-dase actions for salicylic acid-induced generation of active oxygen species and an increase in cytosolic cal-cium in tobacco suspension culture. J. Exper. Bot. 51: 685-693. https://doi.org/10.1093/jexbot/51.345.685

[36] Keshavarz H , Sanavy SAMM, Moghaddm RSG.2016. Impact of Foliar Application with Salicylic Acid on Biochemical Characters of Canola Plants under Cold Stress Condition. Not Sci Biol, 8(1):98-105. https://doi.org/10.15835/nsb819766

[37] Khan MIR, Asgher M, Khan NA. Alleviation of salt induced photosynthesis and growth inhibition by salicylic acid involves glycinebetaine and ethylene in mungbean (Vigna radiata L.). Plant Physiology and Biochemistry. 2014;80:67–74 https://doi.org/10.1016/j.plaphy.2014.03.026

[38] Kiong P, Ling A, Lai AG, Hussein S, Harun AR (2008) Physiological responses of Orthosiphon stamineus plantles to gamma irradiation. Am-Euras J Sust Agricult 2(2).

[39] Kishor PK, Sangam S, Amrutha R, Laxmi PS, Naidu K, Rao K, Rao S, Reddy K, Theriappan P, Sreenivasulu N (2005) Regulation of proline biosynthesis, degradation, uptake and transport in higher plants: its

implications in plant growth and abiotic stress tolerance. Curr Sci 88(3):424–438.
[40] Kuznetsov VV, Shevyakova NI (2007) Polyamines and stress tolerance of plants. Plant Stress 1(1):50–71.
[41] Leenheer A. P. D,. Nelis H. J ,. Lambert W. E and R. M. Bauwen, "Chromatography of Fat-Soluble Vitamins in Clinical Chemistry," Journal of Chromatography, Vol. 429, 1988, pp. 3-58. https://doi.org/10.1016/S0378-4347(00)83866-9
[42] Li G, Peng X, Wei L, Kang G. Salicylic acid increases the contents of glutathione and ascorbate and temporally regulates the related gene expression in salt-stressed wheat seedlings. Gene. 2013; 529:321_325. https://doi.org/10.1016/j.gene.2013.07.093
[43] Liu, H., Wang, Y., Xu, J., Su, T., Liu, G., and Ren, D. 2008. Ethylene signaling is required for the acceleration of cell death induced by the activation of AtMEK5 in Arabidopsis. Cell Res. 18(3): 422–432. oi:10.1038/cr.2008.29. PMID:18268539.
[44] Marchenko, M.M., Bloshko, M.M., and Kostyshin, S.S. 1996. The action of low doses of gamma irradiation on the function of the glutathione system in corn (Zea mays L.). Ukr. Biokhim. Zh 68(2): 94–98. [in Russian] PMID:9005670.
[45] Miura K, TadaY. Regulation of water, salinity, and cold stress responses by salicylic acid.Frontiers in Plant Science. 2014;5:4. doi: 10.3389/fpls.2014.00004.
[46] Murthy, I. Y. L. N. and Anjani, K. (2008). Fatty acid composition in Carthamus species.Proceedings of the 7th International Safflower Conference, 3rd – 6th November, Wagga Wagga, NSW, Australia, 1-3.
[47] Nazar R, Iqbal N, Sayeed S, Khan NA. Salicylic acid alleviates decreases in photosynthesis under salt stress by enhancing nitrogen and sulfur assimilation and antioxidant metabolism differentially in two mungbean cultivars. Journal of Plant Physiology. 2011;168:807–815 https://doi.org/10.1016/j.jplph.2010.11.001
[48] Noctor G and Foyer CH. 1998. Ascorbate and glutathione:keeping active oxygen under control. Annual Reviewof Plant Physiology and Plant Molecular Biology 49,249-279. https://doi.org/10.1146/annurev.arplant.49.1.249
[49] Noreen,S and Ashraf,M. 2010. Modulation of salt (NaCl)-induced effects on oil composition and fatty acid profile of sunflower (Helianthus annuus L.) by exogenous application of salicylic acid. J Sci Food Agric 2010; 90: 2608–2616. https://doi.org/10.1002/jsfa.4129
[50] Price, A.H. and Hendry , G.A.F. (1991). Iron catalyzed oxygen radical formation and its possible contribution to drought damage in nne native grasses and three cereals . plant Cell Environ. 14:477-484. https://doi.org/10.1111/j.1365-3040.1991.tb01517.x
[51] Salter, L. and C.N. Hewitt, 1992. Ozonehydrocarbon interactions in plants. Phytochem.,31: 4045-4050. https://doi.org/10.1016/0031-9422(92)80411-7
[52] Shaki F, Hasan E M, Vahid N. 2018. Growth enhancement and salt tolerance of Safflower (Carthamus tinctorius L.), by salicylic acid. Current Plant Biology 13 (2018) 16–22. https://doi.org/10.1016/j.cpb.2018.04.001
[53] Silber R, Farber M, Papopoulos E, Nervla D, Liebes L, Bruch M, Bron R. 1992. Glutathione depletion in chronic lymphocytic leukemia • -lymphocytes. Blood 80, 2038-2040.
[54] Syeed S, Anjum NA, Nazar R, Iqbal N, Masood A, Khan NA (2011) Salicylic acid-mediated changes in photosynthesis, nutrients content and antioxidant metabolism in two mustard (Brassica junceaL.) cultivars differing in salt tolerance. Acta PhysiolPlant 33:877–886. https://doi.org/10.1007/s11738-010-0614-7
[55] Umebese, C. E. and Bankole, A. E.2013. Impact of salicylic acid on antioxidants, biomass and osmotic adjustments in Vigna unguiculata L. walp. during water deficit stress. Afr. J. Biotechnol.Vol.12(33),pp.5200-5207. https://doi.org/10.5897/AJB2013.12087
[56] Velasco, L., Perez-Vich, B. and Fernandez-Martinez, J. M. (2005). Identification and genetic characterization of a safflower mutant with modified tocopherol profile. Plant Breeding, 124: 459-473. https://doi.org/10.1111/j.1439-0523.2005.01150.x
[57] Wang X, Ruonan Ma, Dongjie Cui, Qing Cao, Zhe Shan & Zhen Jiao.(2017). Physio-biochemical and molecular mechanism underlying the enhanced heavy metal tolerance in highland barley seedlings pre-treated with low-dose gamma irradiation. Scientific Reportsvolume 7, Article number: 14233 (2017). https://doi.org/10.1038/s41598-017-14601-8
[58] Wang, G. and Li, Y. (1985). Clinical application of safflower (Carthamus tinctoriusL.). Zhejiang Tradational Chinese Medical Science Journal, 20: 42-43.
[59] Weiss, E. A. (2000). Oilseed Crops. Second Edition, Blackwell cience, Oxford,Chapter 4: 93-129.
[60] Xienia, U., G.C. Foote, S. Van, P.N. Devreotes, S. Alexander and H. Alexander,(2000). Differential developmental expression and cell type specificity of dictystelium catalases and their response to oxidative stress and UV light. Biochem. Biophys. Acta, 149: 295-310. https://doi.org/10.1016/S0167-4781(00)00063-4

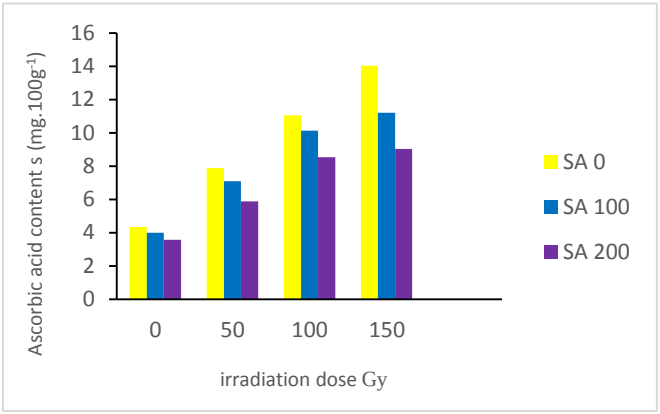

Fig. (1) Effect of γ-ray doses and salicylic acid treatments and their interaction on the contents of Ascorbic acid (mg. 100 g -1) in safflower leaves. L.S.D dose=0.85 L.S.D SA = 0.74 L.S.D dose×SA = 1.48

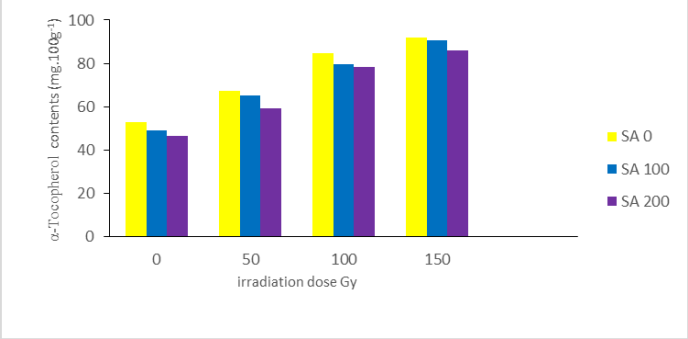

Fig. (2) Effect of γ-ray doses and salicylic acid treatments and their interaction on the contents of α-Tocopherol (mg.100g-1) in safflower leaves. L.S.D dose=1.89 L.S.D SA = 1.64 L.S.D dose×SA = 3.27

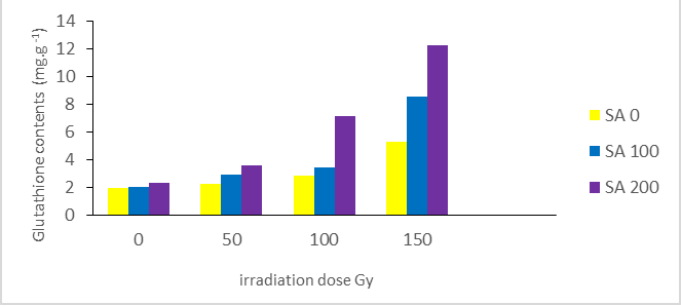

Fig. (3) Effect of γ-ray doses and salicylic acid treatments and their interaction on the contents of glutathione (mg. g-1dw) in safflower leaves. L.S.D dose=1.12 L.S.D SA = 0.97 L.S.D dose×SA = 1.94

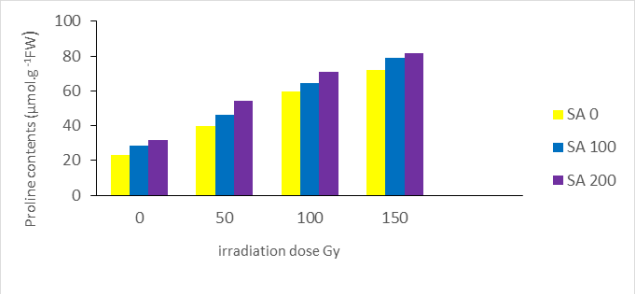

Fig. (4) Effect of γ-ray doses and salicylic acid treatments and their interaction on the Proline contents (µmol.g-1 FW) in safflower leaves. L.S.D dose=2.21 L.S.D SA = 1.91 L.S.D dose×SA = 3.82

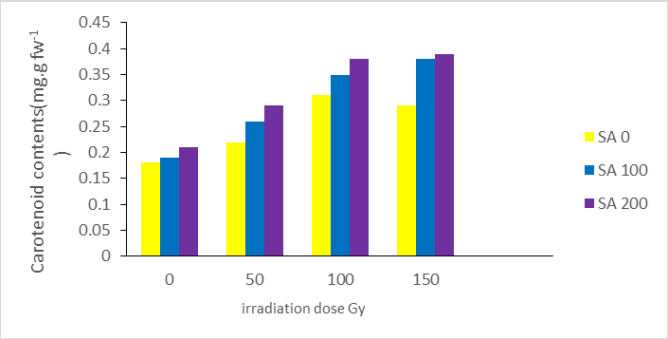

Fig. (5) Effect of γ-ray doses and salicylic acid treatments and their interaction on the contents carotenoid (mg.g fw-1) in safflower leaves. L.S.D dose=0.017 L.S.D SA = 0.015 L.S.D dose×SA = 0.030