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Physiological Changes Accompanied with Some Herbicides on Peanut Leaves, Arachis hypogaea L.

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ABSTRACT

Herbicides are widely used for weeds control and potential hazards may have negative effects on the environment and human health. The purpose of this study was to determine quantitatively phytotoxicity during the application of the herbicides. Photosynthetic pigments, total protein contents and its separation electrophoretically, plant defensive enzymes and non- enzymes substances in treated Giza 6 (G6) of peanut leaves (Arachis hypogaea L.) plants at different concentrations of the tested herbicides. The obtained results showed significant differences in chl a chl. b and carotenoids because of the treatments of G6 with the tested herbicides. There were side effects on these photosynthetic pigments concentrations, especially when using the double recommended rate of these herbicides. Protein contents and its separation electrophoretically, the results indicated the absence of some protein bands in leaves treated with the double recommended rate. Respecting the plant defensive enzymes, ascorbate peroxidases (APX), peroxidases (PODs), Superoxide dismutase (SOD) and catalase (CAT), it was found that increased significantly in these defensive enzymes when treated G6 peanut leaves with the double recommended rate. Also, it was found that there was a significant increase in non- enzymes substances (H₂O₂, MDA and total phenols) in treated G6 peanut leaves with the double recommended rate. So, this study was recommended that avoid using the double recommended rates of these tested herbicides on peanut leaves because of the interactions between these herbicides and some biological processes in the peanut plants and finally reduced the quantitative and qualitative of the product.

INTRODUCTION

Peanut (*Arachis hypogea* L.) is a major oilseed crop in Egypt and around the world. In Egypt, it is the 13th principal economic food crop (Kabir *et al.*, 2013) and the fourth most important oilseed crop in the world (Taphee and Jongur, 2014). It originated from South America and the Mediterranean region according to (Fayez *et al.*, 2014). Through a symbiotic relationship with specific Rhizobium bacteria, peanuts can produce nitrogenfixing nodules. Under field conditions, weeds compete with peanuts for moisture, nutrients, sunlight, and space.Weeds infestation was an important limiting factor in producing the potential yield of any crops (Patel *et al.*, 2020). The relative abundance of common weeds in groundnut fields were, *Galinsoga parviflora* (25%), *Cynodon dactylon* 28%, *Eleucine*

indica (24%), *Ageratum conyzoides* (9%), *Euphorbia geniculata* (8%), *Amaranthus lividus* (6%) (Korav *et al.*, 2020).

Furthermore, the presence of weeds in the harvested crop reduced the crop quality according to (Gianessi and Carpenter, 2000). Phytotoxicity is caused by many herbicides molecules due to their adverse effects on enzymes, and some plants possess Anderson (1983). Pendimethalin is one of the herbicides that is used directly as a soil treatment. Dinitroanaline and pendimethalin herbicides were registered in Egypt on grass weeds for peanuts, because of their inhibitory effects on growth and cell division. pendimethalin is commonly used as a pre-plant treatment prior to emergence (Olorunmaiye, 2009; Jat et al., 2011 and Saha et al., 2015). After the emergence of both grasses and non-target plants, postemergence herbicides such as cyclohexanedione (clethodim) and (aryloxy phenoxy propionate) fluazifop- P- butyl acetyl were used. _The herbicide Imazapic (Imidazolinone) inhibits the enzyme acetohydroxyacid synthase (AHAS), which is involved in the synthesis of aliphatic amino acids, in selected annual and perennial broadleaves and grasses. (Horbowicz et al., 2013). Weeds treated with imazapic (Imidazolinone), fluazifop-p-butyl and clethodim stop growing within a few hours, show gradual discoloration on newer growth in three days and eventually necrosis, desiccation, and plant death occur within two weeks. Herbicide-treated plants exhibited changes in protein banding patterns as well as a reduction in all plant features measured (Aboulila et al., 2016). Herbicides can cause negative effects on food crops as an unintended consequence of their use. Furthermore, these negative effects were genotoxic and morphological in nature according to (Hammok and Al-mandeel, 2020). As long as conditions are favorable, enzymes will catalyze repeatedly their respective chemical reactions. Herbicide inhibitors of ACCase cause an increase in reactive oxygen species such as superoxide radicals, hydrogen peroxide, hydroxyl radicals, and singlet oxygen (Fayez et al., 2014). Oxidative stress caused by abiotic and biotic factors disrupts membrane integrity as a result of lipid peroxidation, according to (Fayez and Bazaid, 2014). In this study, we determined the changes in some photosynthetic pigments of peanut leaves that were treated with the tested herbicides, namely pendimethalin, fluazifop-p-butyl, imazapic and clethodim herbicides as well as changes in defensive enzymes and nonenzymes.

MATERIALS AND METHODS

Herbicides Used:

The herbicides concentrations were used according to the recommendations of the Egyptian Agriculture Ministry:

Pendimethalin (Stomp Extra 45.5 % CS)^R, [N-(1-ethylpropyl)-2, 6-dinitro-3, 4-xylidine], Application rate (1.5 L/fad.), was procured from Kanza-Group, BASF, Egypt.

Fluazifop-p-butyl (fusillade max 12.5 % EC) ^R, (R)-2-[4-[[5-(trifluoromethyl)-2-pyridinyl [oxy] phenoxy] propanate, Application rate (1.5 L /fad.), was available from Syngenta, Egypt. Clethodim (Select super 12.5 % EC) ^R, ((E, E)- (\pm) - 2- [1- [[(3- chloro- 2-propenyl) oxy] imino] propyl] - 5- [2- (ethylthio)propyl] - 3- hydroxy - 2- cyclohexen - 1- one, Application rate (1.00 L/fad.), was available from Syngenta, Egypt.

Imazapic, (Sheto 24 % SL)^R, 5-methyl-2-[4-methyl-5-oxo-4-(propan-2-yl)-4, 5-dihydro-1Himidazol-2-yl] pyridine-3-carboxylic acid, Application rate (100 cm/fad.), was procured from Shoura Chemicals, Egypt.

The Experiments:

Seeds of peanut (*Arachis hypogaea* L.), *cv*. Giza 6 were obtained from Agriculture Research Center, Ministry of Agriculture, Egypt. Seeds were sown in 15 cm diameter plastic pots filled with a 1500 g mixture of sterilized sandy soil (75 % sand, 14 % clay, 7.1% silt, 65 g

peat moss, and 3 mg urea fertilizer per kg of soil) in June 2021. Experiments were carried out in laboratories and on the experimental farm of the Faculty of Agriculture, Zagazig University, Egypt, to investigate the side effects of the selected herbicides on *A. hypogaea*. The trial was laid out with 15 treatments and replicated thrice; each replicate was one pot containing five plants. Three pots were left as a check treatment. The treatments were three rates (half recommended rate, recommended rate, double the recommended rate) of each herbicide including the application of one pre-emergent herbicide pendimethalin (at the rate of 750, 1500 and 3000 ml. /fad.) at two days after sowing. Twenty-one day of *A. hypogaea* growing, the three post-emergence herbicides i.e, fluazifop-p-butyl (at the rate of 750, 1500 and 3000 ml. /fad.), clethodim (at the rate of 500, 1000 and 2000 ml. /fad.) and imazapic (at the rate of 50, 100 and 200 ml. /fad.) were sprayed as a foliar application. In regard to control, peanut leaves were foliar with water only.

Estimation of Pigment Contents in Peanut Leaves:

After 15 days of treatment, fresh peanut leaves were collected to determine chlorophyll (a), chlorophyll (b) and carotenoids. The pigments were extracted from peanut leaves with 80% acetone according to (Fadeel,1962). Chlorophyll (a), Chlorophyll (b), and carotenoids were quantitatively determined spectrophotometrically (Jenway 6300 UV/VIS, Jenway, UK) at wavelengths 663, 647, and 470 nm, respectively, according to (Lichtenthaler, 1987) :

Chl a = 12.25 A663 – 2.79 A647.

Chl b = 21.50 A647 - 5.10 A663.

 $Car = (1,000 \times A470 - 1.82 \times Chl a - 95.15 \times Chl b)/225.$

The concentrations for Chl a, Chl b, and the sum of leaf Car (Xanthophylls and Carotenes) were expressed as mg per g of fresh matter (FM).

Biochemical Diagnosis:

Protein Profile:

Determination of Proteins:

After 15 days of spraying, samples of the tested peanut leaves were randomly taken from each plot to determine total protein content using a commercial kit according to (Gornal *et al.*,1949).

SDS- PAGE Electrophoresis:

The sodium dodecyl sulphate polyacrylamide gel electrophoresis (SDS-PAGE) was used to study the protein pattern in different peanut treatments compared with the control. SDS-PAGE was performed at room temperature in vertical apparatus as described by (Laemmli, 1970).

Defensive Enzymes Activities:

Ascorbate Peroxidase:

(APX, E.C. 1.11.1.11) the activity was determined according to (Nakano and Asada,1981).

Peroxidase Activities:

Peroxidase activities (E.C. 1.11.1.7) were determined colorimetrically in the treated peanut leaves with the tested herbicides compared with control according to (MacAdam *et al.*, 1992; Zhang, 1992 and Trevisan *et al.*,1997).

Superoxide dismutase (SOD, E.C. 1.15.1.1) activity was assayed according to (Beauchamp and Fridovich,1971).

Catalase (CAT, E.C. 1.11.1.6) activity was measured according to (Chandlee and Scandalios, 1984).

Defensive Non- Enzymatic Substances:

Hydrogen Peroxide Content:

 H_2O_2 was determined in peanut leaves colorimetrically according to (Jana and Choudhuri, 1981).

Lipid Peroxidation:

Malondialdehyde (MDA) content was determined as an indication of leaf lipid peroxidation (Hernández and Almansa, 2002).

Total Phenols:

Determination of total phenols content in peanut leaves treated with the four tested herbicides was done according to the method developed by (Malick and Singh, 1980).

Statistical Analysis:

The data were compiled and tabulated for statistical analysis. Data of photosynthetic pigments and other biochemical constituents (defensive enzymes and non- enzymes substances) in peanut leaves treated with four herbicides compared with the untreated leaves were subjected to one-way analysis of variance (ANOVA) to determine the statistical significance of treatments using the Duncun Test Duncan (1955) at 1%, using the statistical software SPSS 14.00 software (SPSS Inc. Chicago, II, USA).

RESULTS AND DISCUSSION

Effect of Herbicides on the Photosynthetic Pigments:

Photosynthetic pigment contents (chl a, chl b, total chl a+b and carotenoids) were estimated. Data showed that in (Table 1). Peanut leaves untreated were the highest amounts of photosynthetic pigments, followed by Peanut leaves treated with half recommended rates, recommended rates and double of recommended rates of the four tested herbicides. The amounts of chl. a, content were increased in untreated Giza 6 leaves (4.59 ± 0.39) , but it was the lowest content (chl. a 1.32 ± 0.11) in the leaves treated with double recommended rates of herbicide, imazapic, followed by clethodim (1.40 ± 0.07) , pendimethalin (1.81 ± 0.26) and fluazifop-p-butyl (2.95 ± 0.34). Furthermore, in response to four herbicides treatments, it was found that the amounts of chl. b were increased significantly in untreated peanut leaves with the tested herbicides compared with the treated leaves. The chl. b amounts were ascending ordered in peanut leaves treated with double recommended rates $(0.98 \pm 0.02, 1.00 \pm 0.51,$ 1.65 ± 0.68 , 2.33 ± 0.34 and 3.92 ± 0.22 , respectively, in leaves treated with imazapic, pendimethalin, clethodim, fluazifop-p-butyl and control treatments. Also, it was observed that the peanut leaves treated with double recommended rates shown decreased in carotenoid contents descending, $(0.3\pm0.01, 0.32\pm0.09, 0.35\pm0.07, 0.60\pm0.07)$ and $0.96\pm0.07)$ for imazapic, pendimethalin, clethodim, fluazifop-p-butyl and control treatments.

Table 1. Effect of some herbicides on photosynthetic pigments contents of peanut plants G6 cultivar (A. hypogaea L.).

	Photosynthetic pigments contents and herbicides rates											
Treatment	Chl. (a) [mg/g (FM)]			Chl. (b) [mg/g (FM)]			Total Chl. (a/b) [mg/g (FM)]			Car. [mg/g (FM)]		
	Half	R.R.	Double	Half	R.R.	Double	Half	R.R.	Double	Half	R.R.	Double
	R.R.		R.R.	R.R.		R.R.	R.R.		R.R.	R.R.		R.R.
	Pre-emergent herbicide after 15 days from application											
Pendimethalin	3.61± 0.66ª	3.91± 0.30 ^a	1.81± 0.26be	4.08± 0.27*	3.58± 0.32*	1.00± 0.51b	7.69± 0.47 ^{ab}	7.50± 0.62*	2.81± 0.76be	0.87 ± 0.05 ab	0.85±0.07*	0.32±0.09bc
	Post-emergent herbicides after 15 days from application											
Fluazifop-p-butyl	3.74± 0.41*	$3.91{\pm}~0.35^{a}$	2.95 ± 0.34^{b}	$3.17{\pm}~0.38^{ab}$	$3.62{\pm}~0.35{^a}$	$2.33{\pm}~0.34^{ab}$	6.91± 0.77ab	7.54 ± 0.70^{a}	$5.28 \pm 0.66^{\circ}$	0.78± 0.09xb	0.85±0.08 ^a	0.60±0.07 ^b
Imazapic	3.70 ± 0.38^{a}	$2.30{\pm}~0.04{^{\rm a}}$	1.32±0.11°	$3.00{\pm}~0.52^{ab}$	$1.97{\pm}~0.03^{a}$	0.98± 0.02b	6.70± 0.89ab	$4.26{\pm}~0.04{}^{\rm a}$	2.30± 0.10°	0.76± 0.10 ^{ab}	0.48±0.01×	0.30±0.01°
Clethodim	2.48 ± 0.60^{a}	$2.35{\pm}~0.94{}^{\rm a}$	1.40± 0.07°	1.70± 0.70b	$2.02{\pm}~0.97{}^{\rm a}$	1.65 ± 0.68^{b}	4.18± 1.25 ^b	$4.37{\pm}~1.90{}^{\rm a}$	3.05± 0.66bc	$0.47 \pm 0.14^{\text{b}}$	0.49±0.22ª	0.35±0.07bc
Control	4.59± 0.39*	4.59± 0.39*	4.59± 0.39*	3.92± 0.22*	3.92± 0.22*	3.92± 0.22*	8.52± 0.61 ^{ab}	8.52± 0.61*	8.52± 0.61*	0.96± 0.07*	0.96±0.07*	0.96±0.07*
F	2.222	4.284	26.584	4.345	3.736	8.547	3.727	4.032	18.273	3.727	4.032	18.273

Different letters mean significant differences at p 0.01 according to Duncan multiple range tests at each characteristic.

Under unfavourable conditions, photosynthesis is the primary source of oxidative stress in plants (Arora *et al.*, 2002; Murata *et al.*, 2007). Photosynthesis inhibitors, oxidative stress, and increased intracellular creation of reactive oxygen species (ROS) were all generated by herbicides, which can lead to macromolecule damage and an increase in plant

defence levels (Arora *et al.*, 2002; Galhano *et al.*, 2010). After treatments with imazapic, clethodim, pendimethalin, and fluazifop-p-butyl, leaves were usually patched and occasionally bleached. Changes in photosynthetic pigment concentration are primarily responsible for changes in leaf colour or bleaching caused by stress. Previous studies indicated that photosynthetic herbicides produced bleaching in leaf chloroplasts due to severe photo-oxidative damage (Dalla Vecchia *et al.*, 2001), induced leaf senescence or oxidative stress (Galhano *et al.*, 2010).

Chlorophyll concentration loss could be a good sign of plant growth and development harm (Yin *et al.*, 2008). In the current investigation, imazpaic exhibited the most effective herbicide effect on lowering pigment content in the peanut G6 cultivar, depending on the dose administered. Herbicides reduced pigment concentration by causing pigment breakdown or inhibiting chlorophyll or carotene production. Chlorophyll content declines under stress were previously connected to chlorophyll molecule breakdown. (Stroch *et al.*, 2008). Our findings are similar to those of (Galhano et al., 2009), who found that Basagran® interacts with the photosynthetic electron transport chain, resulting in an oxidative reaction.

Basagran[®] blocks electron transfer in photosystem II, which reduces photosynthesis (PS II) (Bagchi *et al.*, 2003). Because Basagran[®] competes with quinone B (QB) for binding sites in the PS II reaction centre, the electron flow rate from H2O to NADP via PS II electron carriers is reduced.

Bentazon inhibits PS II, resulting in the generation of energetic singlet and triplet chlorophyll states, as well as reactive oxygen species (ROS) such as singlet oxygen (1O2) (Macedo *et al.*, 2008), The superoxide anion (O2), hydrogen peroxide (H₂O₂), and hydroxyl radical (_OH) have all been linked to a variety of plant physiological problems (Blokhina *et al.*, 2003; Malencic *et al.*, 2008), and induced oxidative damage in photosynthetic cell proteins and membranes (Pospísil, 2009). SA has been proven in recent studies to govern plant adaptation responses to a wide range of biotic and abiotic stressors (Zhou *et al.*, 2009; Cui *et al.*, 2010), Because of less oxidative stress, SA administration to untreated leaves was able to trigger the synthesis of photosynthetic pigments in both cultivars, as shown in the results.

Effect of Some Herbicides on Protein Profile in Peanut Plants G6 cultivar: Total Proteins:

Data showed that (Table 2) total protein in peanut leaves treated with the four tested herbicides at the recommended, half and double the recommended rates varied from 2.40 ± 0.35 to 5.21 ± 0.58 mg/dL. The highest total protein values were (5.21 ± 0.58 mg/dL) recorded in peanut leaves control treatments. The total protein values in other treatments recommended rates, half recommended and double recommended, Alissa, Fayrouz, Omniya and Tomato- 036 were 5.18 ± 0.15 , 4.18 ± 0.16 , 3.48 ± 0.47 and 2.90 ± 1.06 mg/ dL, respectively. Whereas, the lowest total protein values were recorded in Tomato- GS (0.84 ± 0.55 mg/dL).

Proteins Electrophoretic Separation (SDS- PAGE):

The obtained results from SDS- PAGE technique were similar to the obtained results from total proteins colourmetrically. The absence of some protein bands may due to the toxic effects of these herbicides, especially the double recommended rate on peanut leaves (Fig.1).



Fig 1: SDS- PAGE of proteins in peanut leaves treated with four herbicides, M: protein marker, L₁: PHR, L₂: PRR, L₃: PDR, L₄: FHR, L₅: FRR, L₆: FDR, L₇: CHR and L₈: CRR, L₉: CDR, L₁₀: IHR, L₁₁: IRR, L₁₂: IDR, L₁₃: Cont., L₁₄: Cont. and L₁₅: Cont., respectively.

Antioxidant Enzymes:

Ascorbate Peroxidase and Peroxidase Activities:

Data found that (Table 3) The tested herbicides applications on peanut leaves caused highly increase in Ascorbate peroxidase (APX) and peroxidases (POD) activities of G6 cultivar treated with the double recommended rates of the herbicide, pendimethalin (288.30% and 311.99%) higher compared to their controls treatments, respectively. Regarding fluazifop-p-butyl, imazapic and clethodim treatments, APXs were increased by 250.26, 212.05 and 213.25%, respectively, compared with untreated leaves. On the same trend, it was found that peroxidases (PODs) activities in the peanut leaves treated with the double recommended rate of the same tested herbicides were increased significantly compared with half, recommended and control. The changes in SOD activities were increased by 269.38, 262.15 and 249.61 in G6 cultivar leaves treated with pendimethalin, imazapic and clethodim, respectively, compared with untreated G6 leaves.

Previous research has shown that the elevated POD activity in plant tissues can be used as a biomarker for various contaminant stresses (Song *et al.*, 2007). The increase in POD activity in wheat leaves as a result of the haloxyfop herbicide treatment is most likely due to the peroxidation of the membrane lipids, POD may be a sensitive monitor indicating plant damage caused by herbicides contamination. Obviously, in this study, SOD activity was increased in the G6 cultivar in response to the four herbicides mentioned above, pendimethalin, imazapic and clethodim and fluazifop-p-butyl.

Similar stimulation in SOD has been observed in tomatoes under salt stress (Molina *et al.*, 2002) and in wheat plants under herbicide stress (Agarwal *et al.*, 2005). Enhanced levels of SOD in plants have been correlated with tolerance to oxidative stress (Van Breusegem *et al.*, 1999). Furthermore, it has been discovered that overproduction of SOD in plant chloroplasts increased protection against herbicides treatments (Cui *et al.*, 2010). **Superoxide Dismutase (SOD) and Catalase (CAT) activities**

Data found that in (Table 3) SOD and CAT activities were varied significantly among the four tested herbicides. For example, the application of a double recommended rate of fluzifop-p-butyl caused an increase in activities of both SOD and CAT by 217.95 and 264.46% in G6 cultivar respectively, compared with untreated G6 leaves. While the

activities of these defensive enzymes were reduction in G6 cultivar treated with half and recommended rates of the four tested herbicides.

Hydrogen Peroxide (H2O2) and Malondialdehyde (MDA) Contents:

Data showed that (Table 2) application of fluazifop-p-butyl, imazapic, pendimethalin and clethodim on peanut G6 cultivar leaves showed higher amounts of H_2O_2 , especially when using the double recommended rate of these tested herbicides. Generally, the concentration of H_2O_2 in G6 cultivar were increased significantly as a response to these herbicides. The herbicide clethodim caused an increase in both H_2O_2 and MD activities by 209.23 and 173.13% in G6 cultivar, respectively, compared with untreated G6 leaves cultivar. While it was found that the activities of these defensive enzymes were reduced in G6 treated with half and recommended rates of the four tested herbicides.

Phenolic Compounds Content:

Data showed that (Table 2) application of the four tested herbicides on peanut G6 leaves cultivar showed higher values of total phenols, especially when using the double recommended rate of the tested herbicides. Fluazifop-p-butyl shows an increase in total phenols by 203.70% in G6 cultivar compared with control treatments while the activities of these defensive enzymes were reduced in G6 cultivar treated with half and recommended rates of the four tested herbicides.

The antioxidant enzyme system including POD, SOD, APX and CAT enzymes plays an important role in scavenging ROS providing a balanced redox status of an organism (Fecht-Christoffers *et al.*, 2003). In this study, variable changes in the activities of antioxidant enzymes were observed. The changes were included the induction of the defensive enzymes. These data were in agreement with Radwan *et al.* (2019), who found that Basagran® application to G5 cultivar leaves resulted in APX, SOD activity stimulation and POD and CAT inhibition. The CAT activity was inhibited by other herbicides like atrazine, so the reduction in CAT activity by atrazine was caused by the accumulated levels of H₂O₂. APX and CAT represent the major enzymes of H₂O₂ degradation (Vanacker *et al.*, 1998). Also, POD and APX enzyme had a role in preventing H₂O₂ accumulation (Hassan and Alla, 2005).

	Growth attributes and herbicides rates											
	H ₂ O ₂ concentration Malondialdehyde concentration						Total					
Treatment	[nmol/g (FM)]			[nmol/g (FM)]			Total protein (mg/dL)			phenols		
	Half R.R.	R.R.	Double R.R.	Half R.R.	R.R.	Double R.R.	Half R.R.	R.R.	Double R.R.	Half R.R.	R.R.	Double R.R.
	Pre-emergent herbicide after 15 days from application											
Pendimethalin	23.58± 0.88ª	$22.92{\pm}0.23^{ab}$	36.37± 2.10 ^{ab}	31.10± 3.64ª	32.72 ± 0.24^{b}	$46.92{\pm}1.68^a$	4.24± 0.15a	3.84± 0.11 ^{ab}	$3.39{\scriptstyle\pm}~0.29^{ab}$	21.84± 0.69ª	25.47± 2.66ª	33.91± 0.65 ^b
%	107.53	104.55	165.89	97.72	102.81	147.42	81.45	73.77	65.07	100.57	117.27	156.12
	Post-emergent herbicides after 15 days from application											
Fluazifop-p- butyl	22.84± 1.77ª	23.58± 4.37 ^{ab}	39.23± 3.44ª	34.29± 2.53ª	33.96± 4.22 ^b	$50.24{\scriptstyle\pm}2.92^{a}$	4.48± 0.43a	$3.87{\pm}0.54^{ab}$	$3.94{\pm}0.60^{ab}$	21.25± 0.58ª	23.90± 0.38 ^a	44.24± 0.61 ^a
%	104.17	107.56	178.91	107.73	106.69	157.85	85.92	74.34	75.62	97.84	110.05	203.70
Imazapic	19.24± 0.60ª	$36.21{\pm}2.03^a$	45.27± 2.83ª	29.84± 0.71ª	46.80± 1.99ª	$55.06{\scriptstyle\pm}\;3.22^{a}$	$4.81{\pm}\ 0.95a$	$2.84{\scriptstyle\pm}~0.33^{b}$	$2.40{\pm}~0.35^{b}$	21.88± 0.88 ^a	22.53± 0.23 ^a	35.11± 0.67 ^b
%	87.73	166.14	206.46	93.75	147.03	172.97	92.32	54.57	46.00	100.72	103.74	161.64
Clethodim	26.58± 5.21ª	$32.11{\pm}0.67^{ab}$	45.88± 4.26ª	38.16± 3.94ª	41.63± 0.94 ^{ab}	$55.11{\scriptstyle\pm}~4.62{^a}$	3.86± 0.27a	$3.55{\pm}0.30^{ab}$	$2.54{\pm}~0.41^{b}$	21.84± 0.88 ^a	25.78± 0.30 ^a	34.17± 0.39 ^b
%	121.21	146.43	209.23	119.88	130.80	173.13	74.15	68.20	48.69	100.55	118.68	157.32
Control	21.93± 4.39ª	21.93± 4.39b	21.93± 4.39 ^b	31.83± 1.73ª	31.83± 1.73 ^b	31.83± 1.73 ^b	5.21± 0.58a	5.21± 0.58ª	$5.21{\pm}~0.58^a$	21.72± 0.39 ^a	21.72± 0.39 ^a	21.72± 0.39°
F	0.700	4.783	7.646	1.396	8.307	9.987	0.889	4.414	6.199	0.137	2.091	209.597

Table 2. Effect of some herbicides on biochemical parameters in G6 cultivar of peanut plants (A. hypogaea L.).

Different letters mean significant differences at p 0.01 according to Duncan multiple range tests at each characteristic.

	Growth attributes and herbicides rates											
Treatment	Ascorbate Peroxidase [Unit/g (FM)]			Peroxidase [Unit/g (FM)]			Superoxide dismutase [Unit/g (FM)]			Catalase [Unit/g (FM)]		
	Half R.R.	R.R.	Double R.R.	Half R.R.	R.R.	Double R.R.	Half R.R.	R.R.	Double R.R.	Half R.R.	R.R.	Double R.R.
	Pre-emergent herbicide after 15 days from application											
Pendimethalin	$2.63{\pm}0.42^{a}$	$4.12{\pm}0.25^{\rm a}$	5.58± 1.21*	31.11± 1.10ª	41.70± 1.81=	61.73± 1.81ª	92.10± 4.18=	113.61± 7.18 ^s	145.26± 2.90 ^b	8.49± 0.37ª	13.14± 1.22 ^a	$17.81{\pm}1.26^{a}$
%	135.63	212.91	288.30	135.74	181.98	269.38	123.98	152.93	195.54	103.45	160.11	217.02
	Post-emergent herbicides after 15 days from application											
Fluazifop-p- butyl	2.32± 0.60ª	$3.03{\pm}0.20{}^{\rm a}$	4.85± 0.60ab	32.86± 2.30*	40.17± 0.78*	71.50± 3.69*	92.14± 6.06=	120.40± 2.96*	161.91± 4.38*	6.57± 0.53=	11.27± 0.68ab	21.70± 3.76ª
%	119.62	156.63	250.26	143.37	175.30	311.99	124.04	162.07	217.95	80.10	137.37	264.46
Imazapic	$2.33{\pm}0.48^{\rm a}$	$3.43{\pm}0.90{^{\rm a}}$	4.11± 0.29ab	31.73± 0.78*	39.88± 2.67ª	60.08± 5.63ª	75.73± 5.36=	91.92± 3.95b	116.23± 2.13 ^c	$6.92{\pm}0.77{^a}$	11.45± 0.57ab	$19.29{\scriptstyle\pm}~0.57{\scriptstylea}$
%	120.14	177.28	212.05	138.47	174.04	262.15	101.94	123.73	156.46	84.28	139.56	235.05
Clethodim	2.44± 0.52*	$2.47{\pm}~0.32{}^{\rm a}$	4.13± 0.26ab	31.92± 2.34	42.95± 1.43*	57.20± 1.58*	92.07± 7.54*	118.30± 1.70 ^a	138.77± 1.71 ^b	6.90± 1.67ª	11.18± 0.64ab	$17.50{\pm}0.37^{\rm a}$
%	125.82	127.54	213.25	139.30	187.40	249.61	123.94	159.25	186.81	84.08	136.23	213.28
Control	1.94± 0.15*	1.94± 0.15=	1.94± 0.15b	22.92± 1.46 ^b	22.92±1.46 ^b	22.92± 1.46 ^b	74.29± 3.88*	74.29± 3.88 ^b	74.29± 3.884	8.21± 1.53ª	8.21± 1.53b	8.21± 1.53b
F	0.300	3.424	4.639	5.639	22.446	32.360	2.841	21.256	114.311	0.612	3.154	7.134

Table 3. Effect of some herbicides on biochemical parameters in G6 cultivar of peanut plants (*A. hypogaea* L.).

Different letters mean significant differences at p 0.01 according to Duncan multiple range tests at each characteristic.

CONCLUSION

Management strategies to protect peanuts (*Arachis hypogaea* L.) from weeds damage require multiple applications of herbicides. Herbicides usage came out with both positive and negative results on the tested crops and environment. They led to the decrease of the number of photosynthetic pigments and quality of the agricultural chemicals in the cultivation of crops with various health problems and accumulation in the soil and water. Although peanut, *A. hypogaea* was more tolerant to the tested herbicides, pendimethalin, fluazifop-p-butyl, imazapic and clethodim than the weed species, peanut exhibited cell leakage and necrosis after contact with low concentrations, suggesting that differences in biochemical tolerances are not the basis for selective use.

REFERENCES

- Aboulila, A. A., E. B. Belal, M. M. Metwaly and H. R. El-Ramady (2016). Degenotoxicity of Pendimethalin Contaminated Clay Soil by *Pseudomonas resinovorans* Using Anatomical, Cytogenetic and Biochemical Analysis in *Vicia faba* Plants. *International Journal of Current Research Bioscience Plant Biology*, 3(2): 38-53.
- Agarwal, S., Sairam, R., Srivastava, G., Tyagi, A., Meena, R., (2005). Role of ABA salicylic acid, calcium and hydrogen peroxide on antioxidant enzymes induction in wheat seedlings. *Plant Science*, 169: 559–570.
- Anderson, W. P. (1983). Weed Science: Principles, Second Edition, West Publishing Company.
- Arora, A., Sairam, R., Srivastava, G., (2002). Oxidative stress and antioxidative system in plants. *Current Science Bangalore*, 82: 1227–1238.
- Bagchi, S.N., Pistorius, E.K., Michel, K.-P. (2003). A Synechococcus sp. PCC 7942 mutant with a higher tolerance towards bentazone. *Photosynth. Research*, 75: 171–182.
- Beauchamp, C. and Fridovich I. (1971). Superoxide dismutase: improved assays and an assay applicable to acrylamide gels. *Annual Biochemistry*, 44: 276-287.
- Blokhina, O., Virolainen, E., Fagerstedt, K.V. (2003). Antioxidants, oxidative damage and oxygen deprivation stress: a review. *Annals of Botany*, 91: 179–194.
- Chandlee, J., Scandalios, J. (1984). Analysis of variants affecting the catalase developmental program in maize scutellum. *Theoretical and Applied Genetics*, 69: 71–77.

- Cui, J., Zhang, R., Wu, G.L., Zhu, H.M., Yang, H. (2010). Salicylic acid reduces napropamide toxicity by preventing its accumulation in rapeseed (Brassica napus L.). Archives Environmental Contamination Toxicology, 59: 100–108.
- Dalla Vecchia, F., Barbato, R., La Rocca, N., Moro, I., Rascio, N. (2001). Responses to bleaching herbicides by leaf chloroplasts of maize plants grown at different temperatures. *Journal Experimental Botany*, 52: 811–820.
- Ding, C.-K., Wang, C., Gross, K.C., Smith, D.L. (2002). Jasmonate and salicylate induce the expression of pathogenesis-related-protein genes and increase resistance to chilling injury in tomato fruit. *Planta*, 214, 895–901.
- Duncan, D.B. (1955): Multiple range and multiple F tests. *Biometrics*, 11: 1-42.
- Fayez, K.A. and Bazaid, S.A. (2014). Improving drought and salinity tolerance in barley by application of salicylic acid and potassium nitrate. *Journal of the Saudi Society of Agricultural Sciences*, 13: 45-55.
- Fayez, K.A., D.E.M. Radwan, A.K. Mohamed and A.M. Abdelrahman (2014). Fusilade herbicide causes alterations in chloroplast ultrastructure, pigment content and physiological activities of peanut leaves. *Photosynthetica*, 52 (4): 548-554.
- Fecht-Christoffers, M.M., Maier, P., Horst, W.J. (2003). Apoplastic peroxidases and ascorbate are involved in manganese toxicity and tolerance of Vigna unguiculata. Physiol. *Plantarum*, 117, 237–244.
- Fideel, A.A. (1962). Location and properties of chloroplasts and pigment determination in roots. *Physiology of Plant*, 15: 130-147.
- Galhano, V., Peixoto, F., Gomes-Laranjo, J. (2010). Bentazon triggers the promotion of oxidative damage in the Portuguese ricefield cyanobacterium Anabaena cylindrica: response of the antioxidant system. *Environmental Toxicology*, 25: 517–526.
- Galhano, V., Peixoto, F., Gomes-Laranjo, J., Fern_andez-Valiente, E. (2009). Differential effects of bentazon and molinate on Anabaena cylindrica, an autochthonous cyanobacterium of Portuguese rice field agro-ecosystems. *Water Air Soil Pollution*, 197: 211–222.
- Gianessi, L. P. and Carpenter J. E. (2000). Agricultural biotechnology: benefits of transgenic soybeans. National Center for Food and Agricultural Policy 1616 P Street, NW, First Floor Washington, DC 20036.
- Gornal, D.F., Bender D.M. and Hammock B.D. (1949). Quantitative kinetic assays for glutathione-S-transferase and general esterase in individual mosquitoes using an EIA reader. *Insect Biochemistry*, 19: 741-751.
- Hammok, N. S. and F. A. Al-mandeel (2020). Effect of Different Application Methods for Pendimethalin Herbicide on Growth and Productivity of Green Pea Plant (*Pisum sativum L.*). *Current Applied Science and Technology*, 20(3): 528-536, (September-December, 2020).
- Hassan, N.M., Alla, M.M.N. (2005). Oxidative stress in herbicide-treated broad bean and maize plants. *Acta Physiology of Plant*, 27: 429–438.
- Hernández, J.A. and Almansa M.S. (2002). Short-term effects of salt stress on antioxidant systems and leaf water relations of pea leaves. *Physiologia Plantarum*, 115: 251-257.
- Horbowicz, M., Sempruch, C., Kosson, R. et al. (2013). Effect of fluazifop-p-butyl treatment on pigments and polyamines level within tissues of non-target maize plants. *Pesticide Biochemistry and Physiology*, 107: 78-85.
- Jana, S. and Choudhuri M.A. (1981). Glycolate metabolism of three submerged aquatic angiosperms during aging. *Aquatic Botany*, 11: 67-77.

- Jat, R. S., Meena, H. N., Singh, A. L., Surya, J. N., and Misra, J. B. (2011). Weed management in groundnut (*Arachis hypogaea*) in India, A review. *Agricultural Reviews*, 32(3): 155–171.
- Kabir, R., S. Yeasmin, A.K.M Islam and Sarkar M .A.R. (2013). Effect of Phosphorus, Calcium and Boron on the Growth and Yield of Groundnut (*Arachis hypogea* L.). *International Journal of Bio-Science and Bio-Technology*, 5(3):51-59.
- Korav, S., A.K. Dhaka, R. Singh, N. Premaradhya and G.C. Reddy (2020). A study on crop weed competition in field crops. *Journal of Pharmacognosy and Phytochemistry*, 7(4): 3235-3240.
- Laemmali, U.K. (1970). Cleavage of structural proteins during the assembly of the head of bacteriophage T4. *Nature*, 227: 680-685.
- Lichtenthaler, H.K. (1987). Chlorophylls and carotenoids-pigments of photosynthetic biomembranes. In: Colowick, S.P., Kaplan, N.O. (ed.): Methods in Enzymology. Pp. 350-382. Academic Press, San Diego, New York, Berkeley, Boston, London, Sydney, Tokyo, Toronto.
- MacAdam, J.W., Nelson, C.J., Sharp, R.E. (1992). Peroxidase activity in the leaf elongation zone of tall fescue I. Spatial distribution of ionically bound peroxidase activity in genotypes differing in length of the elongation zone. *Plant Physiology*, 99: 872– 878.
- Macedo, R., Lombardi, A., Omachi, C., R€orig, L. (2008). Effects of the herbicide bentazon on growth and photosystem II maximum quantum yield of the marine diatom Skeletonema costatum. *Toxicology in Vitro*, 22: 716–722.
- Malencic, D., Miladinovi_c, J., Popovi_c, M. (2008). Effects of linuron and dimethenamid on antioxidant systems in weeds associated with soybean. *Open Life Sciences*, 3: 155–160.
- Malick, C.P. and M.B. Singh (1980). Plant enzymology and histo enzymology. Kalyani Publishers. New Delhi, 286.
- Molina, A., Bueno, P., Marín, M.C., Rodríguez-Rosales, M.P., Belver, A., Venema, K., Donaire, J.P. (2002). Involvement of endogenous salicylic acid content, lipoxygenase and antioxidant enzyme activities in the response of tomato cell suspension cultures to NaCl. *New Phytologist*, 156: 409–415.
- Murata, N., Takahashi, S., Nishiyama, Y., Allakhverdiev, S.I. (2007). Photoinhibition of photosystem II under environmental stress. *Biochimica et Biophysica Acta Bioenergetics*, 1767: 414–421.
- Olorunmaiye, P. M. (2009). Effect of integrated weed management on weed control and yield components of maize and cassava intercrop in a southern Guinea savanna ecology of Nigeria. *Australian Journal of Crop Science*, 3(3):129-136.
- Patel, V.Y., B.D. Patel, V.J. Patel, D.D. Chaudhari and N.J. Chaudhari (2020). Effect of herbicide combinations on growth, yield and nutrient uptake by weeds in irrigated wheat. *International Journal of Chemical Studies*, 8(5): 1279-1282.
- Pospísil, P., (2009). Production of reactive oxygen species by photosystem II. *Biochimica et Biophysica Acta Bioenergetics*, 1787: 1151–1160.
- Radwan, D.E.M., Fayez, K.A., Mahmoud, S.Y., Hamad, A., Lu, G. (2019). Physiological and metabolic changes of Cucurbita pepo leaves in response to zucchini yellow mosaic virus (ZYMV) infection and salicylic acid treatments. *Plant Physiology and Biochemistry*, 45: 480–489.
- Rutherford, A.W. and Krieger-Liszkay, A. (2001). Herbicide-induced oxidative stress in photosystem II. *Trends in Biochemical Sciences*, 26: 648–653.
- Saha, A., Debarati Bhaduri, Ashvin Pipariya, N.K.Jain and B.B. Basak (2015).Behaviour of pendimethalin and oxyflourfen in peanut field soil : effects on soil biological and

biochemical activities . *Chemistry and Ecology*. Vol. 31, No. 6, 550–566, http://dx.doi.org/10.1080/02757540.2015.1039526.

- Song, N.H., Le Yin, X., Chen, G.F., Yang, H. (2007). Biological responses of wheat (*Triticum aestivum*) plants to the herbicide chlorotoluron in soils. *Chemosphere*, 68: 1779–1787.
- Stroch, M., Lenk, S., Navr_atil, M., _Spunda, V., Buschmann, C. (2008). Epidermal UVshielding and photosystem II adjustment in wild type and chlorina f2 mutant of barley during exposure to increased PAR and UV radiation. *Environmental and Experimental Botony*, 64: 271–278.
- Taphee, G.B.and Jongur A.A.U. (2014). Productivity and Efficiency of Groundnut Farming in North Taraba State. *Journal of Agriculture and Sustainability*, 5(1): 45-56.
- Trevisan MTS, Scheffer JJC, Verpoorte R. (1997) Effect of elicitation on the peroxidase activity in some hop cell suspension cultures. *Plant Cell Tissue and Organ Culture*, 48:121–126.
- Van Breusegem, F., Slooten, L., Stassart, J.-M., Botterman, J., Moens, T., Van Montagu, M., Inz_e, D. (1999). Effects of overproduction of tobacco MnSOD in maize chloroplasts on foliar tolerance to cold and oxidative stress. *Journal of Experimental Botony*, 50: 71–78.
- Vanacker, H., Carver, T.L., Foyer, C.H. (1998). Pathogen-induced changes in the antioxidant status of the apoplast in barley leaves. *Plant Physiology*, 117: 1103– 1114.
- Yin, X.L., Jiang, L., Song, N.H., Yang, H. (2008). Toxic reactivity of wheat (Triticum aestivum) plants to herbicide isoproturon. J. Agric. Food Chem., 56: 4825–4831.
- Zhang, X. (1992). Research Methodology of Crop Physiology. *China Agriculture Press Beijing*, 2 (2): 208–211.
- Zhou, Z.S., Guo, K., Elbaz, A.A., Yang, Z.M. (2009). Salicylic acid alleviates mercury toxicity by preventing oxidative stress in roots of Medicago sativa. *Environmental Experimental Botony*, 65: 27–34.

ARABIC SUMMARY

التغيرات الفسيولوجية المصاحبة لبعض مبيدات الحشائش على أوراق نبات الفول السوداني، Arachis hypogaea L.

رحاب عيداروس محمد السيد سالم¹- أحمد السيد أحمد السبكي¹ أقسم وقاية النبات- كلية الزراعة- جامعة الزقازيق- مصر

تستخدم مبيدات الحشائش بكثافة لمكافحة الحشائش والمخاطر الممكن حدوثها نتيجة التأثيرات الجانبية على البيئة وصحة الانسان. تهدف هذه الدراسة لفهم السمية النباتية نتيجة استخدام تلك المبيدات على أوراق الفول السوداني، Arachis وصحة الانسان. تهدف هذه الدراسة لفهم السمية النباتية نتيجة استخدام تلك المبيدات على أوراق الفول السوداني، Arachis ايماز ابيك و كليثوديم). أستخدمت تركيزات مختلفة من مبيدات الحشائش تحت الدراسة (البنديميثالين، فلوزيفوب يي بيوتيل، ايماز ابيك و كليثوديم). تم تقدير صبغات البناء الضوئي، انزيمات النباتات الدفاعية و المواد الكيماوية غير الانزيمية في أوراق صنف جيزة 6 لنبات الفول السوداني. أوضحت النتائج المتحصل عليها وجود اختلافات معنوية في صبغات الكلوروفيل أ، ب والكاروتينويدات نتيجة معاملة أوراق الفول السوداني بتلك المبيدات سابقة الذكر. محتوى البروتين الكلي وتفريده كهربيا، أوضحت النتائج المتحصل عليها وجود اختلافات معنوية في صبغات الكلوروفيل أ، ب والكاروتينويدات نتيجة معاملة أوراق الفول السوداني بتلك المبيدات سابقة الذكر. محتوى البروتين الكلي وتفريده كهربيا، أوضحت النتائج المتحصل عليها وجود اختلافات معنوية في صبغات الكلوروفيل أ، ب والكاروتينويدات نتيجة معاملة أوراق الفول السوداني بتلك المبيدات سابقة الذكر. محتوى البروتين الكلي وتفريده كهربيا، أوضحت النتائج وجود اختلافات بين المعاملات وتأثير تلك مبيدات الحشائش على اختفاء بعض البروتينات الهامة للنبات وخاصة مع استخدام ضعف التركيز الموصى به. ما يخص الانزيمات النباتية الدفاعية (الاسكوربات بيروكسيديز، البيروكسيديزس، السوبر اوكسيد ديسميوتيز و الكاتاليزس)، وجد أن هناك زيادة معنوية في البروتينات الموصي به من تلك المبيدات. أيضاً، وجد زيادة معنوية في السكوربات بيروكسيديز، البيروكسيديزس، السوبر اوكسيد ديسميوتيز و الكاتاليزس)، وجد أن هناك زيادة معنوية في الالنزيمات عندام ضعف الموسي وحد زيادة معنوية في البوتينات الهامة للنبات وخاصة مع التركيز الموصى به من تلك المبيدات. أيضاً، وجد زيادة معنوية في محر اللبوية كان النباي وحل أول السوبر وكان الكيزيمات النباتية الدفاعية النوريات عندا منعف التركيز الموصى به من تلك المبيدات. أيضاً، وجد زيادة معنوية في محرويات الموصى به من تلك المبيدات. أيضاً، وجد زيادة معنوية في مرويات للللبراليمات عاملي الموص المول الموسي الموسي بالموسي بوليان ا

لذلك نوصي بتجنب استخدام ضعف المعدل الموصى به من مبيدات الحشائش (البنديميثالين، فلوزيفوب بي-بيوتيل، ايماز ابيك و كليثوديم) على نبات الفول السوداني وذلك لحدوث تداخل بينها وبين المكونات الحيوية المسئولة عن عمليات هامة داخل النبات، وكذا على كمية ونوعية المحصول.