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## Physiological and Biochemical Parameters as An Index for Herbicides Damage in Wheat Plants

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### ABSTRACT

In order to evaluate physiological and biochemical changes of wheat plants (*Triticum aestivum* L.) under the stress of four herbicides namely, (pinoxaden, tribenuron-methy, pyroxsulam and clodinafop-propargyl) at recommended dose on three wheat cultivars (Sakha 95, Giza 171 and Shandweel 1). In a totally randomized method, an experiment was performed in El-Sharkia Governorate, Egypt. During 2020 season. The analysis of variance showed that the content of the main pigments of leaf (chlorophyll a, chlorophyll b and carotenoid), activity of catalase, peroxidase, oxidase enzymes and ascorbic acids. Data showed that the herbicide pinoxaden was the highest toxicity while tribenuron-methy was the lowest one on the three cultivars of wheat in chlorophyll content. Chlorophyll a, the reduction percentages were 17.2, 16.8 and 13.6 % in Giza 171, Shandweel 1 and Sakha 95, respectively compared with control. The reduction percentages in chlorophyll b were 14.8, 14.2 and 11.0 % in Sakha 95, Giza 171 and Shandweel 1, respectively compared with control. The opposite picture in carotenoid the reductions were 18.8, 15.3, and 9.7 % in Giza 171, Sakha 95 and Shandweel 1 compared with control. On the other hand, the four herbicides led to a significant decrease in the activity of wheat antioxidant enzymes compared with control. Results showed that the oxidase and peroxidase enzymes were high sensitivity to all treatments compared with control, but ascorbic acid and catalase enzymes appear low sensitivity on all cultivars of wheat. These results showed that all used herbicides reduce plant tolerance from damage and enzymes activities, therefore it can be used as an index in physiological research.

### INTRODUCTION

Wheat (*Triticum aestivum* L.) belongs to the family "Poaceae" and genus "Triticum". It is one of the most important world's leading cereal crops. In Egypt, wheat is the main crop used as a source of food Mohamed and Amin (2014). 85% of the total area under wheat cultivation, and the next species is *Triticum durum*, which occupies 14% of the wheat area (Kumer, 2010). (Rao, 2011). Many factors are responsible for decreased wheat products such as weed (Singh *et al.*, 2018). Narrow (grassy) and broad-leaved weeds dominated in wheat fields represent a very serious problem which may increase rapidly and cause losses in wheat yield ranged from 20-50 % depending on many factors particularly weed species (Dangwal *et al.*, 2010 and Dangwal *et al.*, 2011). *Phalaris minor* and

*Chenopodium album* weeds caused 44 to 66% reductions in wheat grain yield in weed-infested plots Hesammi (2011). Herbicide selectivity depends on certain variables, such as ingredients, characteristics, application of plants, absorption and metabolism. (Hess, 2010). A lot of herbicides are registered for use in wheat in Egypt. Herbicides are used to decrease weed infestation allowing the wheat to grow and another role of herbicides can provide effective and economic control (Kieloch *et al.*, 2006; Pesticide Manual, 2013 and Saha *et al.*, 2018). A wide range of weeds, at a low rate of application and low mammalian toxicity, is used for control. Pinoxaden, in the phenyl pyrazalin chemical class, is a herbicide that inhibits acetyl coenzyme A carboxylase (ACC). A shifted herbicide that reaches the plant via leaves and is transported basipetally and acropetally is financially accessible as pivotal (4.5 percent EC). It is an ACC inhibitor. The herbicide is post-emergent utilized against the yearly gramineous weeds on winter wheat planting from the earliest starting point of the pack framing (Gronwald, 1991 and Mykhalska *et al.*, 2014). Tribenuron-methyl the objective site for this herbicide is the enzyme acetyl-CoA carboxylase which is found in the stroma of plastid, this herbicide used to control broad-leaved weeds in wheat fields (Adamczewski *et al.*, 2014, Kieloch *et al.*, 2014). This herbicide has a place with the sulfonylurea herbicide gathering, which denies acetolactate synthesis (ALS) authorization (Cui *et al.*, 2012, Han *et al.*, 2012, Adamczewski *et al.*, 2014). Pyroxsulam is a triazolopyrimidine sulfonamide, an acetolactate synthase inhibitor, that gives post-emergence control of grass and broadleaf weeds in wheat fields, financially open as pyroxsulam (4.5%OD). Therefore, the damage to crops in the next alteration as wheat is one of the important consequences of using herbicides has the attention of farmers in recent years. Leaves are an integral component of plants because they perform a variety of important roles, such as food production (photosynthesis), food storage, transport of water, gas, etc (respiration). It is known that higher plants contain chlorophyll a and chlorophyll b in the approximate ratio of 3:1 (Rehab Salem, 2016). Chlorophyll is structurally derived from dicarboxylic acid methyl phytylesters, consisting of a porphyrin head with four rings centrally connected to magnesium atoms and a phytol tail (C<sub>20</sub>H<sub>39</sub>OH) with a long aliphatic alcohol chain. Chlorophyll (a) (C<sub>55</sub>H<sub>70</sub>O<sub>6</sub>N<sub>4</sub>Mg) and chlorophyll (b) (C<sub>55</sub>H<sub>70</sub>O<sub>6</sub>N<sub>4</sub>Mg) play a major role in solar energy absorption for the purpose of photochemical photo shooting reactions. (Murray *et al.*, 2015). Plants have a defense system content of antioxidant enzymes such as peroxidase, catalase, ascorbic acid and oxidase enzymes (Gill and Tuteja, 2010). To know this defense system and physiological response of plants to herbicides may facilitate the investigation of phytotoxicity. Although recommended for wheat, there are filed reports of phytotoxicity in the crop after treatment of herbicides which drive the investigation on the capacity of these products to cause oxidative stress in wheat. Thus, this study aimed to evaluate the effect of herbicides on the antioxidant activity of wheat plants. Therefore, the aim of this study was to evaluate the effect of some herbicides on chlorophyll pigments and antioxidant enzymes in wheat plants.

## MATERIALS AND METHODS

### Wheat Cultivars:

Grains of three Egyptian wheat cultivars were kindly obtained from Agriculture Research Center, Cairo, Egypt. These were Shandweel 1, Giza 171 and Sakha 95. The three cultivars were sown in the middle of November (15<sup>th</sup>) in winter season of 2020 in sandy clay soil texture and the physical and chemical analysis of soil were shown in Table (1).

**Table1:** Physical and chemical analysis of the experimental soil tested

Character		Value
Physical analysis	Sand %	72.90
	Silt %	17.30
	Clay %	9.80
	Texture	Sandy-Clay
Chemical analysis	P <sup>H</sup> (1:2.5)	8.20
	CO <sub>3</sub>	----
	HCO <sub>3</sub>	0.10mg kg <sup>-1</sup> soil
	SO <sub>4</sub> <sup>-2</sup>	4.61mg kg <sup>-1</sup> soil
	Cl <sup>-</sup>	3.00mg kg <sup>-1</sup> soil
	Ca <sup>+2</sup>	2.00mg kg <sup>-1</sup> soil
	Mg <sup>+2</sup>	2.00mg kg <sup>-1</sup> soil
	Na <sup>+</sup>	4.35mg kg <sup>-1</sup> soil
	K <sup>+</sup>	0.26mg kg <sup>-1</sup> soil
EC (dS m <sup>-1</sup> ) (1:5)		1.20

**Herbicides Used:**

1. Pinoxaden, phenylpyrazoline: 8-(2,6-diethyl-4-methylphenyl)-7-oxo-1,2,4,5-tetrahydropyrazolo [1,2-d] [1,4,5] oxadiazepin-9-yl] 2,2-dimethylpropanoate; known commercially as Axial®, 4.5% OD sprayed at the rate of 550ml/feddan. Its mode of action is based on inhibition of the Acetyl-coenzyme A carboxylase.

2. Clodinafop-propargyl, Aryloxyphenoxy-propionate: Propynyl (*R*)-2-[4-[(5-chloro-3-fluoro-2-pyridinyl)oxy]phenoxy]propanoate; known commercially Topik®, 15% WP sprayed at the rate of 140 gm/feddan, Syngenta Co., Clodinafop-propargyl formulations interact with and inhibit the enzyme, acetyl co-enzyme A carboxylase (ACCase), which is essential for the production of lipids (fatty acids) needed for plant growth.

3. Pyroxsulam, triazolopyrimidine sulfonamide: N-(5,7-dimethoxy [1,4] triazolo[1,5-a] pyrimidin-2-yl)-2-methoxy-4-(trifluoromethyl) pyridine-3- sulfonamide; known commercially as Pallas®, 4.5% OD sprayed at a rate of 160 cm<sup>3</sup> /feddan. Inhibition of acetolactate synthase -inhibiting.

4. Tribenuron-methyl, Sulfonylurea: (1-Methyl 2[[{N-(4-methoxy-6-methyl-1,3,5 triazin 2-yl) methyl amino} carbonyl] amino] sulfonyl] benzoate) known commercially as Granstar® 75% DF sprayed after 25 days from sowing at the rate of 0.80 gm/feddan, Inhibition of acetolactate synthase –inhibiting. All tested selective herbicides were applied at 25 days after sowing (DAS) with 200 L water/feddan. It is widely used for selective weed control and the most commonly used herbicide in the world on wheat plants.

**The Experiment:**

The experiment was conducted in a randomized, complete block design with three replicates, with an experimental block size of 160 m<sup>2</sup>, with an individual plot size of 16 m<sup>2</sup> (5.34m length x 3width). The soil of experimental sites was sandy clay having pH 8.20, organic matter 8.7 (g/kg) soil with available N 4.35(mg/kg soil), available, and available K was 210 (mg/kg soil). The doses of herbicides per hectare was calculated according to the surface of feddan (4200m<sup>2</sup>), where pinoxaden (4.5%EC) at 550 cm<sup>3</sup>/ fed., tribenuron-mehtyl (75% DF) at 8gm. /fed. and pyroxsulam (4.5% OD) at 160 cm<sup>3</sup>/ fed., also to the plot area (16m<sup>2</sup>) and then the herbicides were solubilized in a suitable amount of water (200 L/fad.) to spraying (Hala Kandil, 2011; El-Kholy *et al.*, 2013 and and Abouziena *et al.*, 2008). The experimental farm is located at 30° 42' 0 N, 31° 48' 0 E. Each plot received herbicide solution

as recommended doses by U.S.A Environmental Protection Agency (Pinoxaden, 2005), Tribenuron-mehtyl, 2009 and Pyroxulam, 2008).

#### **Sample Preparation:**

Wheat plants were sprayed at the age of the fourth leaf (Tottman ,1987), then after 48h from spraying twelve randomly, plants were chosen from each plot which was pulled up then put in a bag in an icebox with dry ice until reach to the laboratory (Frederique *et al.*, 2006 and Kawa *et al.*, 2012).

#### **Measuring Phytotoxicity of the Test Herbicides on Wheat Cultivars:**

Samples of leaves were taken as control before each application and immediately transferred to the laboratory after 96 hours of treatment to determine chlorophyll (a), chlorophyll (b) and carotenoids. Photosynthetic pigments were extracted from fresh leaves using 85% acetone according to Fideel (1962). The optical density was measured spectrophotometrically using spectronic 20D colorim (Wettstain, 1957).

$$\text{Cholorphyll (a) (mg /L)} = (9.784 \times E662) - (0.99 \times E644).$$

$$\text{Cholorphyll (b) (mg /L)} = (21.426 \times E644) - (4.64 \times E662).$$

$$\text{Carotenoids (mg/L)} = (4.695 \times E440) - [0.268 \times (\text{Chlorophyll a} + \text{b})].$$

Where E is: reading of colorimeter.

#### **Enzymes Assay:**

The fresh leaf samples (0.5 g) of *T. aestivum* cultivars were prepared and homogenized in extraction buffers as described by (Hameed *et al.*, 2015; El-Banna and Abdelaal, 2018 and Rasoul *et al.*, 2020). Enzyme activities were expressed on a weight basis. Catalase activity, {(UM H<sub>2</sub>O<sub>2</sub> decomposed) min<sup>-1</sup>g<sup>-1</sup>(FM)} (E.C. 1.11.1.6), was determined according to protocol (Aebi, 1984 and Hadwan, 2018). Peroxidase activity {(UM guaiacol oxidized) min<sup>-1</sup> g<sup>-1</sup> (FM)} (E.C. 1.11.1.7), was determined based on the technique of (Hammerschmidt, *et al.* 1982). According to the methods of (Khan, 1975 and Sikora *et al.*, 2019) was used to measure the polyphenol oxidase enzyme absorbance at 475 nm {(OD) per min per mg of total protein} (EC 1.10.3.1). Ascorbate peroxidase {(UM ascorbate oxidized) min<sup>-1</sup> g<sup>-1</sup> (FM)} (EC.1.11.1.11), the extraction and assay of Ascorbate peroxidase activity were performed as described by Guo *et al.*, (2005).

#### **Statistical Analysis:**

Data were subjected to ANOVA to find out the difference among treatments. All the data of this experiment were subjected to proper statistical analysis of variance according to Snedecor and Cochran (1980) and the differences among treatments were compared using LSD at 0.05level.

## **RESULTS AND DISCUSSION**

#### **Effect of Some Herbicides on Chlorophyll Content in Wheat Cultivars:**

The results presented in tables (2 & 3) indicated that the changes in the content of the main pigments decreased the content of chlorophyll a, chlorophyll b and carotenoids in wheat plants. The results showed that wheat plants had a low level of tolerance to spraying herbicides.

According to the interaction effect between herbicide treatments and cultivars, it is interesting to mention that decreased photosynthetic pigments. These results indicated that Shandweel 1 was the highest sensitivity of wheat cultivars while Sakha 95 was the most negatively affected by all used herbicides, while the lowest values for photosynthetic pigments achieved by Tribenuron-mehtyl. The results showed that pinoxaden herbicide was the highest toxicity in reduced the content of chlorophyll. Chlorophyll a, the reduction percentages values were 22.5, 21.5 and 16.6% in Shandweel 1, Giza 171 and Sakha 95, respectively. In chlorophyll b reduction percentages were 16.3, 22.4 and 24.6 % in

Shandweel 1, Giza 171 and Sakha 95, respectively, the corresponding picture in carotenoid content reduction values were 23.8, 22.5 and 12.1% in Giza 171, Sakha 95 and Shandweel 1 respectively compared with control. On the other hand, wheat cultivars showed that in chlorophyll a Shandweel 1 was the most sensitive cultivar, the mean value was (1.402) and the lowest one was Sakha 95, the mean value was (1.243).

**Table 2:** Reduction percentages of four herbicides on chlorophyll pigments (chlorophyll a, chlorophyll b and carotenoid) of wheat cultivars leaves.

Chlorophyll pigments	Herbicides	Shandweel 1**	Reduction %	Giza 171 **	Reduction %	Saka 95**	Reduction %
Chlorophyll a	Tribenuron-mehtyl	1.422	12.2	1.32	13.4	1.247	10.5
	Pyroxsulam,	1.395	13.9	1.285	15.7	1.213	13.0
	Clodinafop-propargyl,	1.318	18.6	1.253	17.8	1.195	14.3
	Pinoxaden,	1.254	22.5	1.190	21.9	1.163	16.6
	Control	1.619		1.525		1.394	
Chlorophyll b	Tribenuron-mehtyl	1.84	4.1	1.722	7.1	1.63	4.4
	Pyroxsulam,	1.756	8.5	1.621	12.6	1.502	11.9
	Clodinafop-propargyl,	1.630	15.1	1.586	14.5	1.429	16.2
	Pinoxaden,	1.607	16.3	1.439	22.4	1.285	24.6
	Control	1.919		1.854		1.705	
Caroteniod	Tribenuron-mehtyl	2.508	7.8	2.364	8.9	2.185	7.9
	Pyroxsulam,	2.495	8.3	0.072	20.2	2.065	12.9
	Clodinafop-propargyl,	2.428	10.7	2.02	22.2	1.950	17.8
	Pinoxaden	2.391	12.1	1.978	23.8	1.839	22.5
	Control	2.72		2.595		2.372	

\*\* Amounts mg /kg of chlorophyll a, chlorophyll b and carotenoid

**Table 3:** Effect of cultivars (A), herbicides (B) and their interactions (A×B) on chlorophyll a, b, and carotenoids contents (mg/g as fresh weight) of wheat (*Triticum aestivum* L.) plant

Wheat cultivars	Herbicide treatments					Means (A)
	Control	Tribenuron-mehtyl	Pyroxsulam	Clodinafop-propargyl,	Pinoxaden	
<b>Chlorophyll a</b>						
Shandaweel	1.619a	1.423c	1.395d	1.316e	1.254g	1.402A
Giza-171	1.525b	1.320e	1.286f	1.253gh	1.190j	1.315B
Saka- 95	1.395d	1.248h	1.213i	1.196j	1.164k	1.243C
Means (B)	1.513 A	1.330 B	1.298 C	1.254 D	1.203 E	
LSD at 5 %	For (A)= 0.003		For (B)= 0.004		For (A×B)= 0.006	
<b>Chlorophyll b</b>						
Shandaweel	1.920a	1.840c	1.756d	1.631g	1.608i	1.751A
Giza-171	1.855b	1.723e	1.621h	1.586j	1.440l	1.645B
Saka- 95	1.705f	1.630g	1.503k	1.429m	1.285n	1.510C
Means (B)	1.826A	1.731B	1.627C	1.549D	1.444E	
LSD at 5 %	For (A)=0.004		For (B)=0.004		For (A×B)=0.007	
<b>Carotenoids</b>						
Shandaweel	2.720a	2.509c	2.496d	2.428e	2.392f	2.509A
Giza-171	2.595b	2.364g	2.144i	2.010k	1.979 l	2.218B
Saka- 95	2.372g	2.186h	2.066j	1.951m	1.839n	2.083C
Means (B)	2.562A	2.353B	2.235C	2.130D	2.070E	
LSD at 5 %	For (A)=0.010		For (B)=0.006		For (A×B)=0.013	

Also .in chlorophyll b Shandweel 1 was the highest sensitivity, the mean value was (1.751), while Sakha 95 was the lowest one the mean value was (1.510). The corresponding picture in carotenoid Sakha 95 was the lowest one, and the highest one Shandweel 1 compared with control. As mentioned, that these results due to the herbicide pinoxadan are intensively used in the wheat crop which inhibits photosynthesis by binding to the quinine-binding (QB) niche on the D1 protein of the photosystem 2 complex, localized in the chloroplast thylakoid membranes contents of chlorophyll a and chlorophyll b and carotenoids. These herbicides acted as inhibitors or stimulants. Such disturbance may be due to interference with the following criteria: A-Plastide formation; small dense protoplasmic inclusion in the cell plant and may act as a special center of chemical activity. These plastids, when exposed to light, become pigmented and chlorine.

Transformation of plastids into chloroplasts (in chlorophyll-containing plastids, with or without other pigments embedded individually or considered in the cytoplasm of a plant cell. Thus, pesticides may interfere positively or negatively with the formation of plastids or chloroplasts. This may elucidate an increase or decrease in chlorophyll levels in pesticide-treated plants. Several investigators have shown that the relative susceptibility of the receiving plant and, in turn, the pigment content (chlorophyll a and b) and carotenoid in herbicides – treated plants could be explained in different modes of action as follows: inhibition of pigment synthesis due to accumulation of carotenoid precursor phytofluene and photogene, and loss of chlorophyll, carotene. The chloroplast ribosomes and the grana structure of this accumulation resulted from the blockage of dehydrogenation reactions in carotenoid biosynthesis in herbicide-treated leaves. These results are consistent with Wu et al. (2003) in the study of foliar sprays with buprofezine, imidacloprid and jinganny, and the findings have shown that the chlorophyll content is significantly reduced and therefore photosynthetic. Fungicides carbendazim, captan, thiram and mancozeb have been found to have decreased chlorophyll content consistent with fungicide dose and application days. (Aminjani and Mahdiyeh, 2013; Hanci and Cebeci, 2014) suggested that chlorophyll and carotenoid content often decrease and reduce in plants exposed to pesticides. Also, Giza 178 c.v. achieved the highest photosynthesis pigments content followed by Giza 182 c.v. which recorded the lowest value (Kattab, 2019; Rasoul et. al., 2020). The applications of herbicides were able to reduce chlorophyll and amino acids and total soluble carbohydrate (Kumer, 2012; EL- Sobki , 2019). Tribenuron-methyl and flumetsulam + florasulam treatments did not cause any visible phytotoxicity, while pyroxsulam, diclofop-methyl and tralkoxydim treatments recorded a low index of phytotoxicity on wheat plants (Hoda *et al.*, 2018).

#### **Effect of Herbicides on the Activity of Antioxidant Enzymes in Wheat Cultivars:**

It appears that the cells under stress increase the production of antioxidant enzymes that scavenge free radicals. Once free radicals are formed, the cells start some physiological defense mechanisms to prevent damage (Anerjee *et al.*, 2001). The results of the variance analysis showed that the effects of three herbicides (pinoxaden, clodinafop propargyl and pyroxsulem) on the activity of antioxidant enzymes were observed in Tables (4 &5).

It was observed that the recommended dose of the herbicide treatments reduced all the activity of antioxidant enzymes compared with control. The application pinoxaden,, clodinafop propargyl and pyroxsulem reduced the catalase enzyme activity. The reduction percentages were 5.8, 15.5, 22.7% in Shandweel 1 cultivar; 4.2 ,17.1,21.5 % in Giza 171 and 7.7 ,12.4 ,18.4% in Sakha 95, respectively.

Data in Table (5) presented that peroxidase enzyme activity was significantly affected by the three herbicides. The activity of this enzyme was completely inhibited at pinoxaden herbicide. The application of pinoxaden, clodinafop propargyl and pyroxsulem reduced the activity of enzyme peroxidase by 30.1, 33.2, 37.9% in Shandweel 1 cultivar; 30.0, 36.0 41.5 % in Giza 171 and 35.3, 36.8 and 38.9 % in Sakha95, respectively. The

results showed that polyphenol oxidase enzyme activity was significantly affected by the three herbicides. The activity of this enzyme was completely inhibited at pinoxaden herbicide. The application of pinoxaden, clodinafop propargyl and pyroxsulem reduced the activity of the enzyme polyphenol enzyme by 18.4,31.2,43.1% in Shandweel 1 cultivar; 14.5, 31.0, 38.0 % in Giza 171 and 10.4,19.7 and 26.2 % in Sakha 95, respectively.

**Table 4:** Reduction percentages of some herbicides on the activity of antioxidant enzymes in wheat cultivars.

Antioxidant's enzymes	Herbicides	Shandweel 1	Reduction %	Giza 171	Reduction %	Saka 95	Reduction %
Ascorbate peroxidase enzyme	Pyroxsulem	84.205	4.	79.495	3.9	75.105	5.1
	clodinafop propargyl	81.375	7.5	76.225	7.8	71.705	9.4
	Pinoxaden	80.51	8.4	71.955	13.0	68.605	13.3
	Control	87.93		82.645		79.17	
Polyphenol oxidase enzyme	pyroxsulem	14.195	18.4	13.085	14.5	10.665	10.4
	clodinafop propargyl	11.96	31.2	10.56	31.0	9.555	19.7
	Pinoxaden	9.905	43.1	9.485	38.0	8.79	26.2
	Control	17.395		15.3		11.905	
Peroxidase activity enzyme	Pyroxsulem	5.17	30.1	4.87	30.0	4.15	35.3
	clodinafop propargyl	4.945	33.2	4.455	36.0	4.05	36.8
	Pinoxaden	4.595	37.9	4.07	41.5	3.935	38.6
	Control	7.4		6.96		6.41	
Catalase activity enzyme	Pyroxsulem	11.76	5.8	11.405	4.2	10.0	7.7
	clodinafop propargyl	10.545	15.5	9.875	17.1	9.5	12.4
	Pinoxaden	9.645	22.7	9.34	21.5	8.79	18.9
	Control	12.485		11.905		10.845	

Therefore, on the other hand, data cleared that the same picture Ascorbate peroxidase enzyme activity was significantly affected by the three herbicides. The activity of this enzyme was completely inhibited at pinoxaden herbicide. The application of pinoxaden,, clodinafop propargyl and pyroxsulem reduced the activity of enzyme Ascorbate peroxidase enzyme by 4.2, 7.5, 8.1% in Shandweel 1 cultivar; 3.9, 7.8, 13% in Giza 171 and 5.1, 9.4 and 13.3 % in Sakha95, respectively. Also, results observed that the Shandweel 1 cultivar was the highest sensitivity of the three cultivars for all herbicide treatments therefore, this study can advise not to use this cultivar in agriculture because of high sensitivity.

In the test, the effect of pinoxaden is based on inhibition of Acetyl-coenzyme A carboxylase, clodinafop-propargyl, inhibition of the enzyme, acetyl co-enzyme A carboxylase (ACCase), which is essential for the production of lipids (fatty acids) needed for plant growth, pyroxsulam inhibition of acetolactate synthase, inhibition of On wheat cultivars plants. The used herbicides reduced catalase activity, increased H<sub>2</sub>O<sub>2</sub> levels and increased lipid oxidation which is in agreement with (Nohatt, *et al.*, 2016; Nilgum *et al.*, 2019). Oxidative stress caused by increased concentration of oxygen species may cause cell death due to inhibition of the membrane lipid peroxidation enzyme (Ma *et al.*, 2013). Pyroxsulam acts by blocking the transport of electrons in photosynthesis, such as H<sub>2</sub>O<sub>2</sub>. (Gill and Tuteja, 2010). Researches have shown these herbicides as inhibiting the enzyme activity (Abedi and Pakniyat, 2010). The study is useful for farmers, agriculturalists and



researchers in this field to identify antioxidant enzyme changes in order to avoid negative herbicide effects on the growth and yield of wheat plants.

**Table 5:** Effect of cultivars (A), pesticides (B) and their interactions (A×B) on ascorbic acid, oxidase, super-oxidase and catalase enzymes (mg/g as fresh weight) of wheat (*Triticum aestivum* L.) plant

Wheat cultivars	Herbicide treatments				means
	Control	Pyroxsulem	clodinafop propargyl	Pinoxaden	
<b>Ascorbate peroxidase enzyme</b>					
Shandaweel	87.93a	84.21a-c	81.38c-e	80.51c-f	83.95A
Giza-171	82.70b-d	79.50c-g	76.23f-h	76.96e-g	79.22B
Saka- 95	79.17d-g	75.11gh	71.71hi	68.61i	74.53C
Means (B)	83.27A	79.60B	79.60C	76.44D	
LSD at 5 %	For (A)= 3.59 For (B)= 2.35 For (A×B)= 4.86				
<b>Polyphenol oxidase enzyme</b>					
Shandaweel	17.40a	14.20d	11.96f	9.91i	13.81A
Giza-171	15.30b	13.08e	10.56h	9.49j	12.68B
Saka- 95	11.91f	10.67gh	9.56ij	8.79k	10.37C
Means (B)	14.87A	12.65B	10.69C	9.39D	
LSD at 5 %	For (A)= 0.27 For (B)= 0.19 For (A×B)= 0.38				
<b>peroxidase activity enzyme</b>					
Shandaweel	7.40a	5.18g	4.95h	4.60j	5.78A
Giza-171	6.96b	4.87i	4.46k	4.07m	5.30B
Saka- 95	6.41d	4.15l	4.05m	3.94n	4.91C
Means (B)	6.92A	4.73B	4.48C	4.20D	
LSD at 5 %	For (A)= 0.007 For (B)= 0.040 For (A×B)= 0.063				
<b>Catalase activity enzyme</b>					
Shandaweel	12.49a	11.76bc	10.55e	9.65fg	11.29A
Giza-171	11.91bd	11.68c	9.88f	9.34h	10.76B
Sakha- 95	10.85d	10.86d	9.50gh	8.79i	9.95C
Means (B)	11.75A	11.43B	9.97C	9.26D	
LSD at 5 %	For (A)= 0.17 For (B)= 0.15 For (A×B)= 0.28				

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## ARABIC SUMMARY

### التغيرات الفسيولوجية والبيوكيميائية في نباتات القمح كمؤشر لأضرار مبيدات الحشائش

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إجريت تجارب حقلية لتقييم التغيرات الفسيولوجية والبيوكيميائية لنباتات القمح *Triticum aestivum* L. تحت تأثير أربع مبيدات حشائش (بينوكسادين، تريبينورون ميثيل، بيروكسولام وكلوديناغوب- بروبارجيل) بالجرعة الموصى بها على ثلاثة أصناف من نباتات القمح (سحا 95، الجيزة 171 وساندويل 1) خلال موسم النمو 2020 في محافظة الشرقية، مصر. أظهر التحليل الاحصائي أن محتوى أوراق نباتات القمح المختبرة من الأصباغ (الكلوروفيل (أ)، الكلوروفيل (ب) والكاروتين) وإنزيمات الكاتلايز، البيروكسيديز، الأوكسيديز وحمض الأسكوربيك. أظهرت النتائج أن مبيد الحشائش بينوكسادين سجل أعلى المبيدات المختبرة سمية بينما أعطى مبيد تريبينورون ميثيل أقل المعاملات تأثيراً على أصناف القمح الثلاثة المختبرة في محتوى الكلوروفيل. حيث أوضحت النتائج أن الكلوروفيل (أ) سجل نسب خفض قدره 17.2 , 16.8 و 13.6% في أصناف جيزة 171, شندويل 1 وسحا 95 على التوالي، مقارنة بمعاملة الكنترول. بينما سجل كلوروفيل (ب) نسب خفض قدره 14.8 , 14.2 و 11.0% في أصناف سحا 95 , جيزه 171 و شندويل 1 على التوالي، مقارنة بمعاملة الكنترول. في حين سجلت قيم الكاروتين قيم قدرها 18.8 و 15.3 و 9.7% في أصناف جيزة 171، سحا 95 و شندويل 1 على التوالي، مقارنة بمعاملة الكنترول. من ناحية أخرى، سجلت مبيدات الحشائش الأربعة انخفاض معنوي في نشاط إنزيمات القمح المضادة للأكسدة مقارنة بالكنترول. كما أوضحت النتائج أيضاً أن إنزيمات الأوكسيديز والبيروكسيديز أظهرت حساسية عالية لجميع مبيدات الحشائش المختبرة، مقارنة بمعاملة الكنترول، بينما سجلت إنزيمات حمض الأسكوربيك والكاتلايز حساسية منخفضة في جميع أصناف القمح المختبرة. أظهرت النتائج أن جميع مبيدات الحشائش المختبرة تقلل من تحمل النباتات من الضرر، وتؤثر على نشاط الإنزيمات وبالتالي يمكن إستخدامها كمؤشر لحدوث السمية النباتية.