



## STUDY OF OXIDATIVE ENZYMES IN MANY GENOTYPES OF (*Zea mays* L.)

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### ABSTRACT

The aim of this study was to evaluate the activity of some oxidative enzymes in three genotypes of maize namely (Baghdad, 5018, Sara) under three sowing dates: (26July, 4August and 12August) and denote (D<sub>1</sub>, D<sub>2</sub> and D<sub>3</sub>). A field experiment was conducted at field of College of Agricultural Engineering Sciences, University of Baghdad/Al- Jadriya for the fall season 2021- 2022, factorial experiment was conducted using randomized blocks (RCBD) with three replicates. Significant differences were found among genotypes for the number of grains in the row, and the number of kernels in the ear. The results showed that superiority of the genotype (Baghdad) in the characteristic of Tasseling and Silking flowering 50%, plant yield with averages of 58.89d, 62.89d and 164.6g, respectively. While the genotype (Sara) was superior to the weight of 100 kernels, with an average weight of 27.34g. Genotype (5018) gave the highest value for Catalase (CAT) and Peroxidase (POD) enzyme, which was 2.16 u/mg and 0.51 u/mg, respectively, while genotype (Sara) gave the highest value for Superoxide dismutases (SODs), which was 50.15u/mg The (D<sub>1</sub>) showed an increase in most of the traits averages, especially (50% tasseling which gave 65.00d), (50% Silking which gave 68.00d), (weight of 100 grains which reached 26.91g) and (POD enzyme 0.56 u/mg). The (D<sub>3</sub>) gave the highest value for plant yield which was 162.1g, CAT enzyme 2.54u/mg and SOD enzyme 57.34u/mg.

**Keywords:** *Zea mays* L, Oxidative enzymes, Genotypes, Sowing date.

### دراسة انزيمات الاكسدة في عدة تراكيب وراثية من الذرة الصفراء (*Zea mays* L.)

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#### الخلاصة

هدفت الدراسة الى تقييم النشاط الانزيمي لثلاثة تراكيب وراثية من الذرة الصفراء : (بغداد، 5018، سارة) خلال ثلاثة مواعيد زراعة (26 تموز، 4 آب، 12 آب). اجريت تجربة في حقول كلية علوم الهندسة الزراعية جامعة بغداد/ الجادرية، للموسم الخريفي 2021-2022 وفق تصميم القطاعات العشوائية الكاملة RCBD (تجربة عاملية) بثلاثة مكررات. وجدت فروق معنوية بين التراكيب الوراثية لعدد الحبوب في الصف وعدد الحبوب في العنوص. اظهرت النتائج تفوق التركيب الوراثي (بغداد) في صفة 50% تزهير ذكري وانثوي وحاصل النبات بمتوسط 58.89 يوم و 62.89 يوم و 164.6 غم/نبات على التوالي. بينما تفوق التركيب الوراثي (سارة) في صفة وزن 100 حبة بمتوسط مقداره 27.34 غم، وأعطى التركيب الوراثي (5018) أعلى قيمة لإنزيم catalase و peroxidase بلغ مقدارها 2.16 وحدة/ملغم و 0.51 وحدة/ملغم على التوالي، بينما أعطى التركيب الوراثي (سارة) أعلى قيمة لإنزيم superoxidase بلغت 50.15 وحدة/ملغم.

أظهر الموعد الأول (D<sub>1</sub>) زيادة في معظم متوسطات الصفات وخاصة (50% تزهير ذكري 65.00 يوم) و (50% تزهير انثوي 68.00 يوم) و (وزن 100 حبة 26.91 غم) و (0.56 peroxidase وحدة/ملغم. بروتين).

أعطى الموعد الثالث (D<sub>3</sub>) أعلى قيمة لحاصل النبات بلغت 162 غم/نبات وإنزيم catalase 2.54 وحدة/ملغم، وإنزيم superoxidase 57.34 وحدة/ملغم.

الكلمات المفتاحية: الذرة الصفراء، انزيمات الاكسدة، تراكيب وراثية، مواعيد الزراعة.



## INTRODUCTION

The most important thing that the plant breeder needs is the productivity that is achieved by increasing the cultivated area and providing the appropriate environmental conditions, the most important of which is the planting date because of its direct effect of growth and development stages of the crop.

Cultivars has decreased causing the loss of original local cultivars. The response of crops to planting dates varies according to their types. Determining or choosing the appropriate date for planting is very important in creating a regulation of the growth stages of the crop and the formation of its organs in the appropriate conditions of temperature, humidity and light. Also, the detection of early genetic behavior in different environmental conditions is one of the breeding programs that aim to increase the production of a unit area and improve its characteristics (Nagham *et al.*, 2020).

NADPH oxides (NOXES) are one of the major sources of cellular Reactive Oxygen Species (ROS), and remain a subject of intense research interest due to their exclusive function in the production of reactive oxygen species under normal physiological conditions (Hayyan *et al.*, 2016). Catalase (CAT) was divided into three subgroups which are typical, atypical and catalase-peroxidase that has an important role in the oxidative enzyme system by breaking down hydrogen peroxide ( $H_2O_2$ ) into water and oxygen with high efficiency and effectiveness. High temperature significantly reduces the activity of Catalase enzyme (Scandalios, 2000). There is presence of an increase in the activity of Catalase enzyme under heat stress conditions (Singh, 2020). The Catalase family (CAT) consists of highly conserved enzymes that Catalyze the hydrolysis of  $H_2O_2$  to water and oxygen and thus play an important role in plant responses to biotic and abiotic stresses (Yuan *et al.*, 2017 ; Zhou *et al.*, 2018). Because of the heterogeneous regulation of gene expression Peroxidase is involved in many cellular processes during plant development stages and is stress responsive (Cosi & Dunand, 2009). High temperatures cause a decrease in the percentage of Peroxidase enzyme (Yang *et al.*, 2017). It has recently been shown that the accumulation of  $O_2^-$  leads to the activation of the enzyme SOD, which detoxifies  $O_2^-$  by converting it to  $H_2O_2$  and is thus one of the antioxidant enzymes that protects corn roots from oxidative damage. (Keyster *et al.*, 2012 ; Klein *et al.*, 2013). Higher temperatures reduce the activity of the enzyme superoxidase (Zhu *et al.*, 2010).

## MATERIALS AND METHODS

A field experiment was carried out in the fields College of Agricultural Engineering Sciences - University of Baghdad for the fall season (2020-2021), A total of three genotypes of maize namely (Baghdad – 5018 – Sara ), and denote ( $V_1$ ,  $V_2$  and  $V_3$ ) were used in this study by using RCBD with three replications, under three sowing dates ( $D_1$ ,  $D_2$  and  $D_3$ ). Average temperatures during the sowing season were got from the Iraqi Meteorological Authority.

### Estimation of enzyme activity

The samples were taken during flowering period of each experimental unit and for three sowing dates, 1g of fresh leaves were mashed for each sample with addition of 10.mL of protective potassium phosphate pH =7.8 and frozen at  $-18^\circ C$ . used device Spectrophotometer to estimate the enzymatic activity of each following enzymes.

### Estimation of activity catalase (CAT)

The enzyme activity was estimated according to the method Aebi (1974) using the following solutions:

**Solution A:** is prepared by dissolving 1.7420g of  $KH_2PO_4$  in little of distilled water and complete the volume to 200mL of distilled water.

**Solution B:** is prepared by dissolving 1.3608g of  $\text{KH}_2\text{PO}_4$  in little of distilled water and complete the volume to 200mL of distilled water.

**The First Solution**

Potassium phosphate buffer  $\text{KH}_2\text{PO}_4$  solution, which is prepared by adding a calculated volume of solution A to 50mL of solution B.

**The second solution:**

Hydrogen peroxide  $\text{H}_2\text{O}_2$  solution at a concentration of 30mm, we take 0.34mL of  $\text{H}_2\text{O}_2$  (30%) and complete the volume to 100mL using the buffer solution. Preparation method:- 1000  $\mu\text{L}$  of the sample were added with 1000 $\mu\text{L}$  of buffer solution and 1000 $\mu\text{L}$  of the prepared hydrogen peroxide were added and measured with a spectrophotometer at wavelength 240nm and the change was followed for five consecutive readings.

$$\text{Enzyme activity CAT (u/mg)} = \frac{\frac{\Delta \text{reading}}{\Delta t}}{0.1 \times 0.01 \times \text{protein concentration}}$$

0.1=mL sample volume

0.01 = 1 unit of enzyme

mg. mL = protein concentration

**Estimation of activity peroxidase (POD).**

The activity of the peroxidase was estimated according to the **nezih (1985)** method. A guaiacol substance was prepared. 1.36mL of guaiacol was placed in a beaker and the volume was completed to 250mL with distilled water.

Method of preparation: - Mix 1000 $\mu\text{L}$  of hydrogen peroxide with 1000 $\mu\text{L}$  of prepared guaiacol and 1000 $\mu\text{L}$  of the sample. The data was read by a spectrophotometer at wavelength 420nm and the change was followed every 30s for 3min.

$$\text{Enzyme activity CAT (u/mg)} = \frac{\frac{\Delta \text{reading}}{\Delta t}}{0.1 \times 0.01 \times \text{protein concentration}}$$

**Estimation of activity superoxidase dismutase (SOD)**

The activity of the enzyme (SOD) was estimated by **Beyer & Fridoich (1987)** using 6 solutions :

**Solution (1)**

Prepare (82.4 Mm) of  $\text{K}_2\text{HPO}_4$  and (165  $\mu\text{M}$ ) of EDTA-2Na by dissolving 3.5880 g of  $\text{K}_2\text{HPO}_4$  and 0.0154 g of EDTA in sum of distilled water and then fill the volume to 250.mL of distilled water.

**Preparation method** (82.4 Mm) of  $\text{K}_2\text{HPO}_4$ , (165  $\mu\text{M}$ ) of EDTA-2Na Dissolve 3.5880g of  $\text{K}_2\text{HPO}_4$  and 0.0154g of EDTA in sum of distilled water and then complete the volume to 250mL of distilled water.

The buffer potassium phosphate was prepared by gradually adding solution B to solution A until pH value = 7.8

**Solution (2)**

Prepare by dissolving 150mg of L- methionine in 5mL of distilled water.

**Solution (3)**

It was prepared by dissolving 0.1mL of Triton X-100 in 10mL of distilled water.

**Solution (4)**

It was prepared by dissolving NBT Nitro blue titrazolum 14.4mg in 10mL of distilled water and kept in an opaque vial.

**Solution (5)**

Mix the solutions (1,2,3,4) with a volume of 18.35mL of solution 1, 1.50 of solution 2, 0.75 of solution 3 and 1.00 of solution 4, so that the total volume becomes 21.60mL.

**Solution (6)**

It was prepared by dissolving 0.0018g of riboflavin in a little of distilled water and completing the volume to 100mL of distilled water.

**Method of preparation:** 1500µL of the prepared mixture were added to the test tubes and 500µL of distilled water were added to it, then 40µL of the sample were added to the tubes. The plank treatment was prepared in the same way with the difference that the tube contains distilled water instead of the sample, and a 40µL of riboflavin dye (solution 6), then the absorbance was read by a spectrophotometer at a wavelength of 560nm, then the tubes were exposed to illumination for 7-10min and the absorbance was read again at the same wavelength. The highest inhibition rate of the sample was estimated, as well as the enzyme activity was estimated according to the following equations.

$$\text{SOD(inhibition)} = \frac{(A2B-A1B)-(A2S-A1S)}{(A2B-A1B)} \times 100$$

A1B = blank absorbance before lighting

A2B = blank absorbance after lighting

A1S = absorbance of the sample before lighting

A2S = absorbance of the sample after lighting

$$\text{Enzyme activity SOD} = \frac{\text{sample inhibition}}{\text{highest inhibition}} \times \frac{\text{df}}{\text{Vs}} \text{ (u/mL) Measruing unit}$$

df = dilution factor

U = unit

Vs = sample size

**growth traits**

**Number of days from planting to 50% tasseling:**

The number of days from planting (the first irrigation) until the totasseling was recorded by 50% (Al-Sahoke, 1990)..

**Number of days from planting to 50% silking:**

The number of days from planting (the first irrigation) until the emergence of the silking was recorded by 50% (Al-Sahoke, 1990).

**Number kernels\ row :**

The number of grains in one row was calculated from the sum of all five random ear per plants from each unit experimental.

**Number kernels \ear:**

The number of grains in the ear was calculated by (number of rows per ear) × ( the number of grains per row).

**Weight of 100 grains (g):**

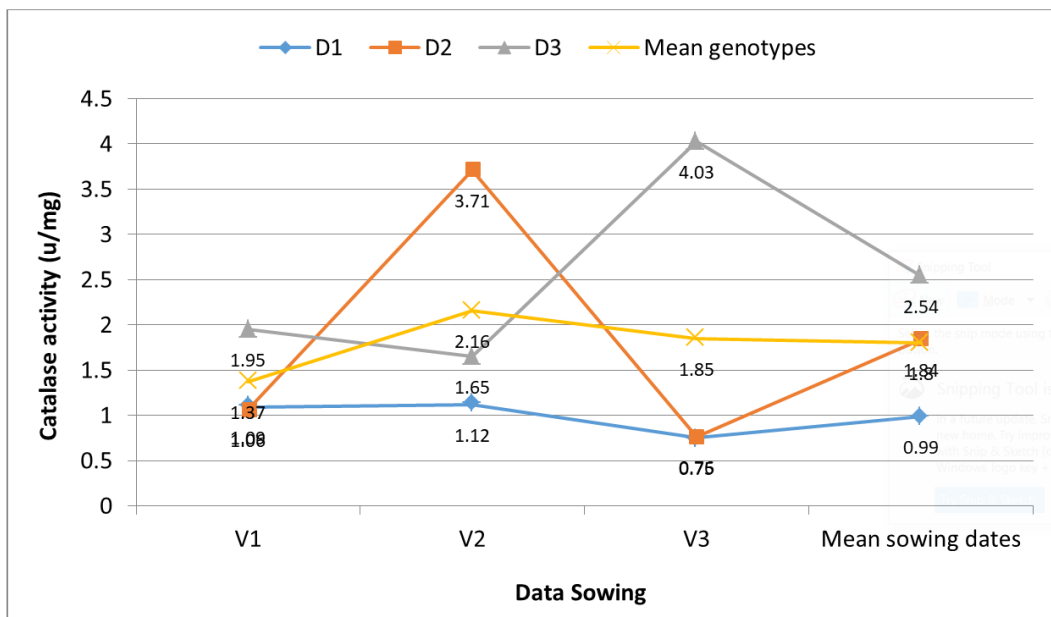
the yield of five grain plants was weighed (Al-Sahoke, 1990).

## RESULTS AND DISCUSSION

### Catalase (CAT)

Plants use complex antioxidant defense systems to overcome uncontrolled production of ROS and protect plants from oxidative damage when ROS production exceeds the activities of antioxidant enzymes such as catalase, causing damage to essential cellular components, (Hussain *et al.*, 2019). The results in (Figure, 1) indicated that there were significant differences between the genotypes, as the genotype (V<sub>2</sub>) gave the highest average of 2.16u/mg, followed by the genotype (V<sub>3</sub>), which gave an average of 1.85 u/mg, compared to the genotype (V<sub>1</sub>) which gave the lowest mean of 1.37 u/mg.

The results in (Figure, 1) indicated significant differences between the sowing date, where the sowing date D<sub>3</sub> out performed by giving it the highest average of 2.54 u/mg, followed by the sowing date D<sub>2</sub>, which gave an average of 1.84 u/mg compared to the sowing date D<sub>1</sub>, which gave the lowest mean of 0.99 u/mg, (Table, 1) indicates a rise in temperatures in pla date D<sub>1</sub>, D<sub>2</sub>. The activity of the catalase decreased and this is due to the inhibition of the biosynthesis of antioxidants (Yin *et al.*, 2008) and this was reached by Zhu *et al.*, (2010) that when the temperature increased, the activity of the catalase decreased and this is not confirmed by the results of Kumar *et al.*, (2012) when he concluded that the activity of the Catalase enzyme increases when temperatures rise. The results of the interaction in (Figure, 1) showed that sowing date D<sub>3</sub> gave the highest mean for genotype (V<sub>3</sub>) of 4.03 u/mg, while sowing date D<sub>1</sub> gave the lowest average for genotype (V<sub>3</sub>) 0.75 u/mg.



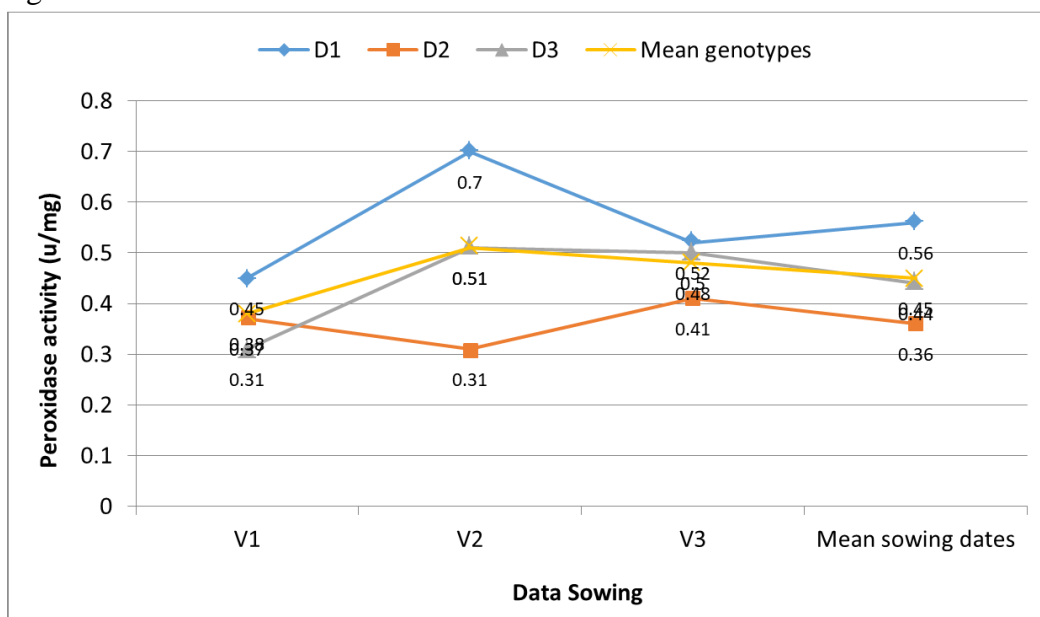
**Figure (1):** Effect of sowing dates on activity of catalase (CAT) (u/mg) for genotypes of maize in fall season 2020-2021.

**Table (1):** Effect Average maximum and minimum temperatures in fall season. 2020-2021.

Month	Jul	Aug			Sep			Oct		
AT Max	47,71	46,75	44,61	45,76	46,00	39,28	39,04	40,35	36,60	<b>31,44</b>
AT Min	<b>40,31</b>	<b>35,81</b>	<b>34,09</b>	<b>34,66</b>	<b>36,79</b>	<b>31,61</b>	<b>29,27</b>	<b>31,13</b>	<b>26,39</b>	<b>23,42</b>

### Peroxidase (POD)

The accumulation of ROS species and the levels of membrane damage in maize increase under heat stress conditions (Hussain *et al.*, 2019). The results of (Figure 3) indicated that there were significant differences between the genotypes, as the genotype (V<sub>2</sub>) gave the highest average Peroxidase enzyme 0.51u/mg, followed by the genotype (V<sub>3</sub>) which gave an average of 0.48u/mg, compared to the genotype (V<sub>1</sub>) which gave the lowest mean 0.38u/mg. This is because the differences between the genotypes in terms of enzyme activity are due to the specificity of the genotype according to Chakraborty & Pradhan (2011). While there were significant differences between the three sowing dates, where the date D<sub>1</sub> outperformed, giving the highest average 0.56u/mg, followed by the D<sub>3</sub> date, which gave an average 0.44u/mg compared to the D<sub>2</sub> date, which he gave the lowest average 0.36u/mg, because the activity of the Peroxidase enzyme increases at high temperatures as a defense mechanism against heat stress (Yin *et al.*, 2008), and when the enzyme activity is higher, H<sub>2</sub>O<sub>2</sub> is efficiently converted to water and oxygen and the integrity of the membrane is preserved under heat stress (Dođru, 2021). This is what Kumar *et al.*, (2012) concluded that the activity of the enzyme increases at higher temperatures. This is not confirmed by the results of (Zhu *et al.*, 2010) that by increasing the temperature, the activity of the peroxidase decreases. The results of the interaction in (Figure 3) showed sowing date D<sub>1</sub> showed the highest mean of genotype (V<sub>2</sub>) 0.70u/mg, while sowing date D<sub>2</sub> showed the lowest mean of genotype (V<sub>2</sub>) 0.31u/mg.



**Figure (3):** Effect of sowing dates on activity of peroxidase (POD) (u/mg) for genotypes of maize in fall season 2020-2021.

### Superoxidase (SOD)

Superoxidase is the first line of defense used by plants to inhibit  $O_2^-$  in  $H_2O_2$  and  $O_2$  (Hussain *et al.*, 2016). Results (Figure, 4) indicated that there were significant differences between genotypes, as genotype ( $V_3$ ) gave the highest average 50.15u/mg, followed by genotype ( $V_1$ ), which gave an average 45.14u/mg compared to genotype ( $V_2$ ). Which gave the lowest average 43.69u/mg, and the reason is due to the possibility of natural resistance to heat stress, which may be the reason for less generation of ROS under heat stress (Ranjeet *et al.*, 2014). While there were significant differences between the sowing dates, where the  $D_3$  gave the highest average of 57.34u/mg, followed by the  $D_1$  which gave an average of 43.74u/mg compared to the  $D_2$  which gave the lowest an average 37.91u/mg, because when plants are exposed to heat stress, the antioxidant systems become active and start searching for ROS, Where the antioxidant defense system plays a vital role in helping plants to withstand heat stress (Yadav *et al.*, 2017). (Table, 1) for temperature indicates that the enzyme activity increased by the third date at lower temperatures, and this is what Zhu *et al.*, (2010) found when high temperatures, the activity of the Superoxidase enzyme decreases, and this is not confirmed by the results of Kumar *et al.*, (2012) where they found that the activity of the enzyme increases at higher temperatures. The results of the interaction in (Figure 4) showed that  $D_3$  gave the highest mean for genotype ( $V_3$ ) of 64.24u/mg, while  $D_2$  gave the lowest mean for genotype ( $V_2$ ) of 22.97u/mg.

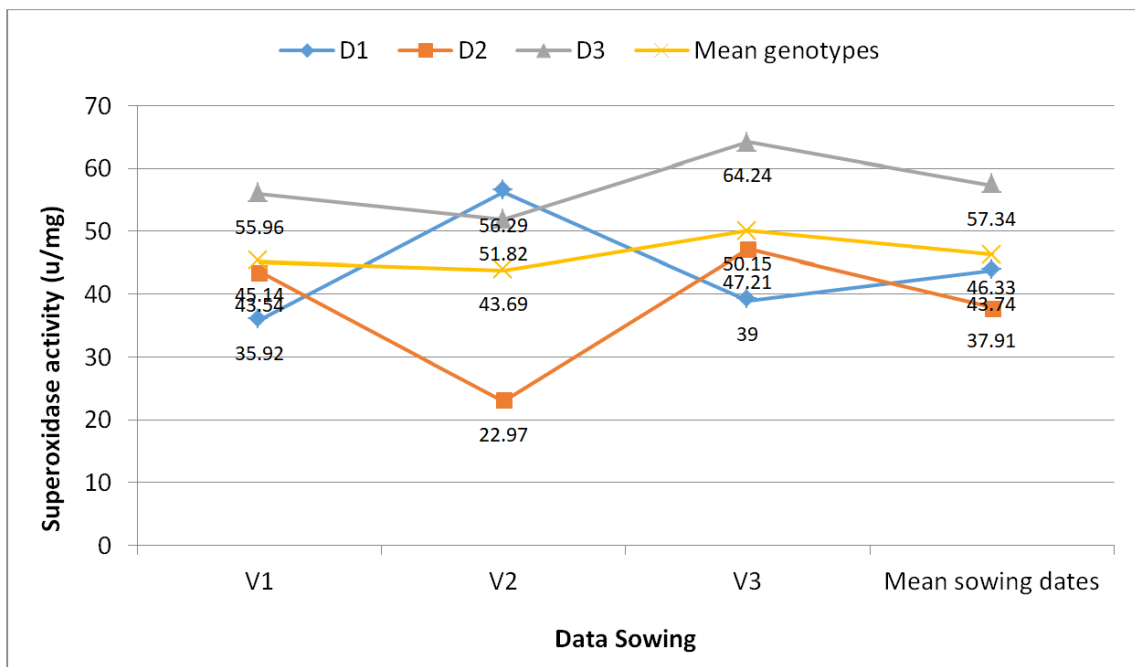
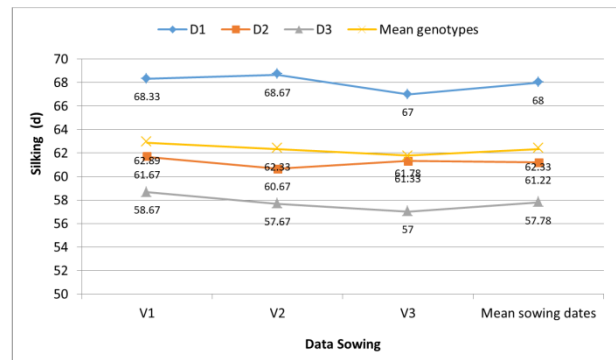
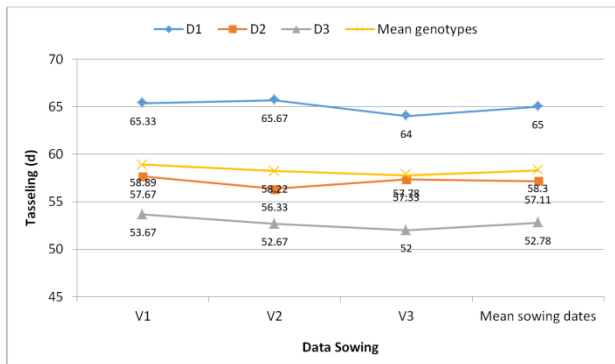


Figure (4): Effect of sowing dates on activity of superoxidase (SOD) (u/mg) for genotypes of maize in fall season 2020-2021.

### Tasseling and silking flowering 50%

According (Figure 5. a and b) it is clear that there are significant differences between the genotypes for the number of days of sowing trait up to 50% tasseling and silking straight. followed by genotype ( $V_2$ ), which gave an average number of days 58.22d and 62.33d, respectively, compared to genotype ( $V_1$ ), which took the most number of days 58.89d and 62.89d, respectively. This is attributed to the gene action and the genetic combination of each

genetic structure and the different influence of the environment on the performance of the genotypes, and this was confirmed by **Tollenaar *et al.*, (2006)** there is a significant effect of sowing dates, as the D<sub>3</sub> took the lowest average number of days from sowing to 50% silking, reach 52.78d and 57.78d, respectively. Followed by the sowing D<sub>2</sub>, it gave an average of 57.11d and 61.22d, respectively, while the sowing D<sub>1</sub> had the most number of days to tasseling and silking was 65.00d and 68.00 days, respectively. (**Table, 1**) that the accumulated temperatures were higher on sowing date D<sub>1</sub>, then began to decrease on sowing date D<sub>2</sub> and sowing D<sub>3</sub>, and this led to the flowering of the plants of the third date early due to the lack of accumulated heat units (**Parthasarath *et al.*, 2013**), where he found in his results that plants of the late date accompanied by a drop in temperature bloomed early. The results of the interaction in (**Figure 5. a and b**) showed that the date D<sub>3</sub> took the least number of days from planting up to 50% Tasseling and silking for all genotypes, and the genotype (V<sub>3</sub>) took the least number of days of 52.00d and 57.00d for tasseling and silking respectively. The maximum number of sowing date days for genotype (V<sub>2</sub>) was 65.67d and 68.67d for tasseling and silking, respectively, for the sowing date D<sub>1</sub>.



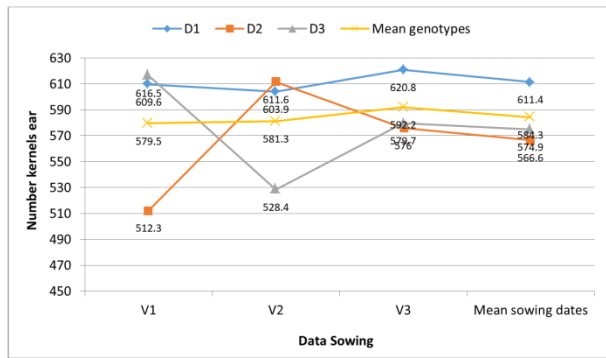
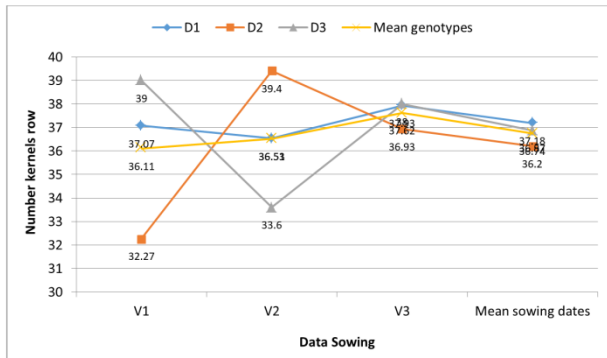
**(Figure 5.a):** Effect of sowing dates on 50% Tasseling for genotypes of maize in fall season 2020-2021.

**(Figure 5.b):** Effect of sowing dates on 50% Silking for genotypes of maize in fall season 2020-2021.

#### Number kernels row and number kernels ear

Indicates (**Figure, 6. a and b**) no significant differences between the genotypes of maize in number kernels row and number kernels ear. Also showed no significant differences between sowing dates for traits. While the results of the interaction showed significant differences between the genotypes and sowing dates for number kernels row. genotype (V<sub>2</sub>) the best in D<sub>2</sub> gave the highest average 39.40kernels row while genotype (V<sub>1</sub>) gave lowest average 32.27kernels row. As for interaction between genotype (V<sub>3</sub>) and D<sub>1</sub> showed the highest mean of number kernels ear, genotype 620.8kernels ear while genotype (V<sub>1</sub>) gave the lowest average 512.3kernels ear for D<sub>2</sub>.





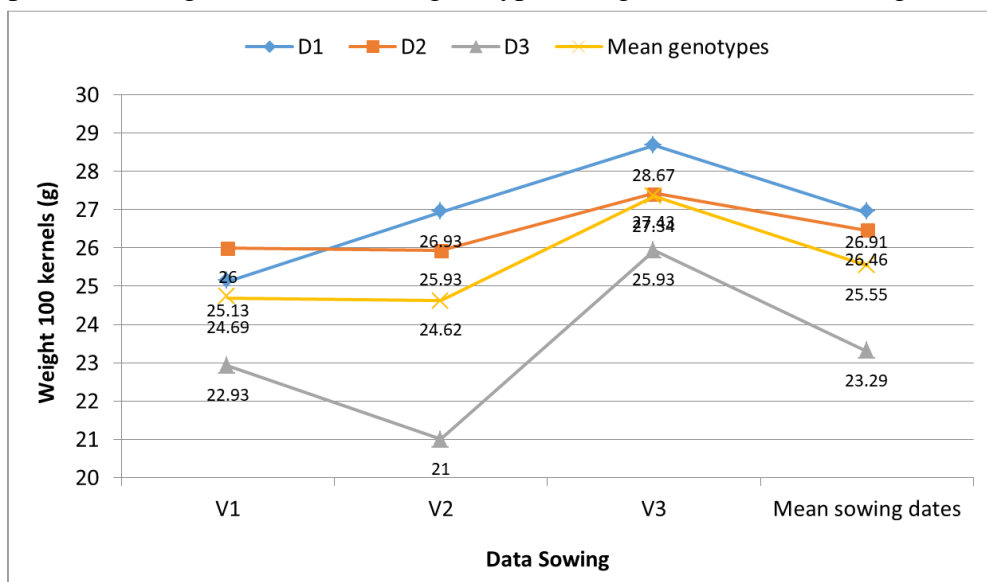
**Figure (6.a):** Effect of sowing dates on number kernels row for genotypes of maize in fall season 2020-2021

**Figure (6.b):** Effect of sowing dates on number kernels ear for genotypes of maize in fall season 2020-2021

**Weight 100 kernels g**

Evident from **(Figure, 7)** there are significant differences between the genotypes of trait weight 100 kernels g, genotype (V<sub>3</sub>) gave a higher average 27.34g, followed genotype (V<sub>1</sub>) gave an average 24.69g compared to genotype (V<sub>2</sub>), which gave the lowest average 24.62g. noticed there is significant effect of sowing dates in trait, D<sub>1</sub> was superior with the highest average 26.91g, followed by D<sub>2</sub> which gave an average 26.46g compared to the D<sub>3</sub>, which gave lowest average 23.29g, **(Table, 1)** shows that the delay in sowing date led to a decrease in length of grain filling period, which affected the weight 100 kernels and finally the grain yield, which may be due to the low accumulation of nutrients in seeds. **Qian et al., (2016)** and this is supported by **Rabbani & Safdry (2021)**.

The results of the interaction in **(Figure, 7)** Showed the D<sub>1</sub> gave the highest average for genotype (V<sub>3</sub>) 28.67g, while the D<sub>3</sub> for genotype (V<sub>2</sub>) gave the lowest average 21.00g.



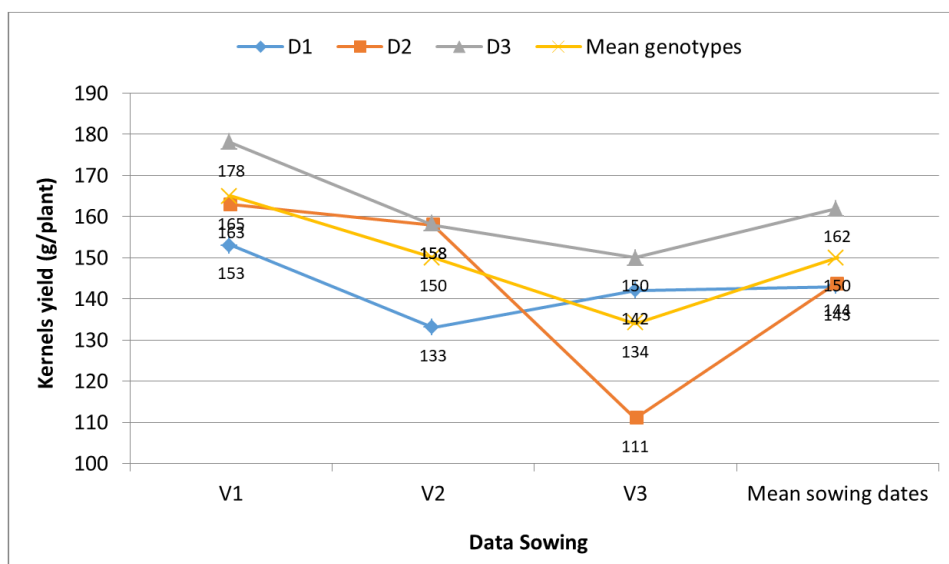
**Figure (7):** Effect of sowing dates on weight 100 kernels g for genotypes of maize in fall season 2020-2021.

### The kernels yield per plant (g per plant)

The results in (Figure 8) shows that the significant differences between the genotypes for trait kernels yield, genotype (V<sub>1</sub>) gave the highest average 165g/plant followed genotype (V<sub>2</sub>) gave an average 150g/plant. Compare genotype (V<sub>2</sub>) to the genotype (V<sub>3</sub>), which gave the lowest average 134g/plant, the reason is due to the morphological difference between genotypes in addition to difference between average traits of vegetative part and the traits of the yield and this is confirmed by Wahaib (2012). (Figure, 8) evident there are significant differences between the genotypes of trait plant yield. Genotype (V<sub>1</sub>) gave a higher average 165g/plant Followed by genotype (V<sub>2</sub>) gave average 150g/plant compared to the genotype (V<sub>3</sub>), which gave the lowest average 134g/plant. The reason is due to morphological difference between the genotypes in addition to difference between average traits of the vegetative part and the traits of yield (Wahaib,2012).

The D<sub>3</sub> recorded the highest average 162g/plants followed by the D<sub>2</sub>, which gave an average 144g/plants compared with D<sub>1</sub>, which gave the lowest average 143g/plants because the high kernels yield is related to the number of grains and the period of their filling. early or late sowing dates affect plants due to the environmental conditions to which the plant is exposed at the time of flowering, and thus affect the kernels yield (Tsimba *et al.*, 2013). (Table, 1) show the temperatures associated with the sowing dates. the, D<sub>1</sub> was exposed to high temperatures more than D<sub>2</sub> and D<sub>3</sub>.

The results of the interaction in (Figure, 8) between D<sub>3</sub> and genotype (V<sub>1</sub>) showed the highest average 178g/plant while D<sub>2</sub> with genotype (V<sub>3</sub>) gave the lowest average 111g/plant.



**Figure (8):** Effect of sowing dates on kernel yield g/plant for genotypes of maize in fall season 2020-2021.

### CONCLUSION

The best activity enzyme observed in catalase and superoxidase at the first sowing date (D<sub>1</sub>). Genotype (Baghdad) the best for 50% Tasseling, silking flowering and plant yield .g/plant. The best activity catalase and peroxidase observed in genotype (5018). Genotype (Sara) was the best for the weight 100 grain g.



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