



## Effect of Drought Stress on Germination , Chlorophyll and Carbohydrates contents in *Triticum aestivum*

Aseel Mohammed Imran

Babylone university , college of farmacy , department of pharmacognocy.

### Abstract:-

This study was conducted to determine the effect of water stress on germination , chlorophyll a, b, total chlorophyll and carbohydrates content in *Triticum aestivum* , five concentrations of PEG were used (100 , 200 , 300 , 400 , and 500 mg/l) in addition to control ( distilled water ). Results of analysis of variance showed a significant decrease in germination in contrast with control and decrease in chl a , b , total chl and carbohydrates in low levels of drought stress , but in high levels of drought stress (400 and 500 mg/l ) its found a significant increase in chl a , b , total chl and carbohydrates contents in contrast to control.

### Introduction:-

Water availability is one of the most limiting environmental factors affecting crop productivity and is a well known fact that crop growth is frequently subjected to water stress during the course of its life time. Stress imposed during these periods drastically affectes crop growth , ultimately leading to a massive loss in yield and quality ( Govindarajan et al., 1996) .

There is some evidence that roots are the primary sensors of water deficit in the soil , causing the observed physiological and biochemical perturbations in the stems and the decline in growth to be generally interconnected with charges in plant nutrition, carbon dioxide balance and water relations. Plant nutrient elements and available water are absorbed by plant roots in independent processes but they are closely related to one another (Levitt, 1980) .

Abiotic stresses are widespread problems around the world , germination characteristics negatively affected by abiotic stresses in more crops (Ansari et al., 2012) . Seed germination is the most sensitive stage to abiotic stress (Patade et al., 2011) osmo priming can contribute to improve seedling emergence in different plant species by improvement of ATP activity, RNA , and acid phosphatase synthesis (Fu et al., 1988) , also by improve of amylases, lipases and protease synthesis (Ashraf and Foolad, 2005). Bahrami et al,(2012) found that germination percent decreased with increasing drought level in *Sesamum indicum*. Ansari and Sharif-Zadeh (2012) found a decrease in germination percent in *Secale montanum* when increase the drought stress.

Chlorophyll accumulation plays vital role in the crop productivity, as they are only pigments responsible for CO<sub>2</sub> assimilation. Its destruction as often observed under water stress in deleterious to the crop productivity. Pirzad et al., 2011 found that maximum amount of chlorophyll a , chlorophyll b and total chlorophyll obtained from irrigation level 85 and 70 % in contrast with control (100%) and minimum amount of total chlorophyll obtained in level 55% of field capacity. Wter stress decreased the chlorophyll content in rice leaves (Zhu and Huang , 1994). The accumulation of osmolyt compounds in the cells, as aresult of water stress is often associated with a possible mechanism to tolerate the harmful effect of water shortage. Pirzad et al,(2011) , Jones



and Turner (1980) found that contents of sugars did not change in fully expanded leaves. Riduan et al, (2005) found a different tolerance to drought stress in total leaf sugar content in different species of peanut (in peanut CV. Jerapah, Gajah, Macan and Simpai decreased under drought stress while that of Singa and Kelinci are the same as that under optimal condition.

Screening of different crop plants to abiotic stress is used to find out most resistant variety ( Zafar-UI-Hye et al., 2007) . While screening the wheat cultivars for most drought sensitive and most drought tolerant genotype is was considered that the success of these approaches under green house and lab conditions depends on their same behavior under field condition also (Raza et al., 2012).

Recently, poly ethylene glycol (PEG) has been used to control water potential in seed germination studies to determine plant drought tolerance at germination and seedling stages (Dodd and Donovan, 1999). In this method water potential can be controlled precisely and a large number of treatments can be performed quickly, PEG with 6000 or higher molecular weight can not enter the pores of plant cells and PEG is not toxic to plant cells (Verslues et al., 2006).

The objective of this study was to determine the germination of *Triticum aestivum* and the amount of chlorophyll a, chlorophyll b ,and total chlorophyll and carbohydrate content under drought stress.

#### **Materials and methods :**

fifteen healthy and sterilised seeds in sodium hypochloride, for 5 min (Martin,1990) of *T. aestivum* and were kept in sterilised petri dishes on filter paper and moistened with 10ml extracts. Each treatment had 3 replicates each with 15 seeds. Control consisted of distilled water. The Petri-dishes were maintained under laboratory conditions at 25 C<sup>0</sup> temperature with diffused light during day. The petridishes were covered to prevent the loss of moisture by evaporation. PEG 6000 was prepared by dissolving the required amount of PEG (100 , 200 , 300 , 400 , 500 mg/l) in distilled water (25 °c) and complete the volume to 100 ml , by distilled water.

#### **Statistical analysis:-**

The data obtained were subjected to three way analysis of variance within randomized complet block design (RCBD). And the mean values were separated at p<0.05 applying least significant difference test (LSD).

#### **Physiological parameters:-**

#### **Germination percentage:-**

After ten days, germinated seeds were measured according to the low: (Belcher and Miller 1974).



$$\% \text{ germination} = \frac{n}{N} * 100$$

n = number of germinated seeds.

N = total number of seeds.

### Chlorophyll extraction and determination :-

0.05 g of complete leaves were homogenized with 2 ml acetone 80% concentration in mortar and pestle , then filtrated and added 2 ml acetone 80% to the mortar and again this treatment after that the filterate was taken for spectrophotometer at 645 and 663 nm wavelengths. To estimate total chlorophyll and chlorophyll a , b by spectrophotometry , the following equations were used ( Turner, 1981)).

$$\text{Chlorophyll a (g/l)} = (0.0127 \times \text{OD663}) + (0.00269 \times \text{OD645}) \quad (2)$$

$$\text{Chlorophyll b (g/l)} = (0.0229 \times \text{OD645}) + (0.00468 \times \text{OD663}) \quad (3)$$

$$\text{Total chlorophyll (g/l)} = (0.0202 \times \text{OD645}) + (0.0082 \times \text{OD663}) \quad (4)$$

After that converted units to mg /l

### Carbohydrates extraction and determination :-

1 mg dry weight of leaves from each treatment , add 1 ml phenole 5% and 5 ml H<sub>2</sub>SO<sub>4</sub> (80%) were added , remain solution for 10 min , after that the solution was taken to spectrophotometer at 490 nm ( blank treated by mixed 1 ml phynol (5%) and 5ml H<sub>2</sub>SO<sub>4</sub> (80%) . and then made calibration curve was made by using glucose and the following equation was used ( Duboies et al., 1956).

$$Y = 289.63 * + 3.8435$$

### Results:-

#### Germination percentage:-

With an increased level of drought stress , the mean of germination percentage reduced in control 90% while in level 100 , 200 , 300 , 400 , and 500 mg/l germination percentage became 71, 50, 50, 49, and 29% in sequence. However there was a significant difference between the control and all levels of stress .(fig 1). And there was a significant differences between concentration 10 mg/ml and other concentrations and there was no significant differences among concentrations 200 , 300 , 400 mg/ml and between 400 and 500 mg/l .

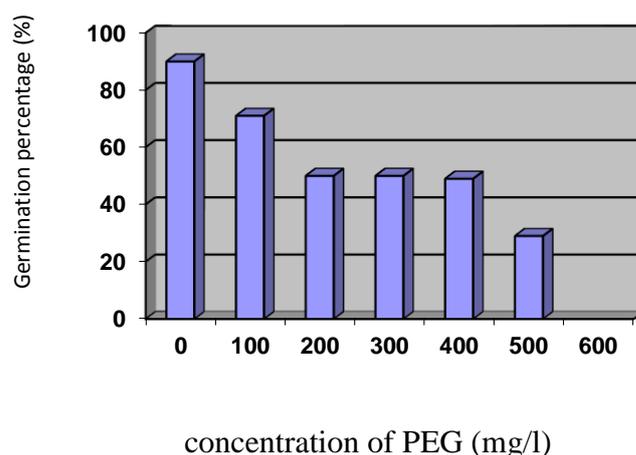


### Chlorophyll content:-

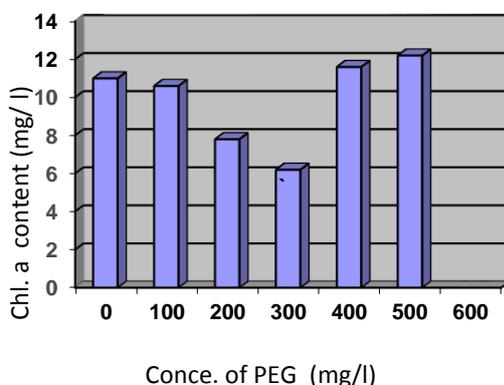
Chlorophyll a, b and total chlorophyll showed a significant decrease in concentrations 100 , 200 , 300 % (except chl a in conc 100mg/l) they were became 10.6 , 7.8, 6.2 mg/l when contrast to control (11 mg/l) , chl b became 11.1 , 7.9 , and 8.9 mg/l In contrast with control (11.5 mg/l) , while concentration 400 and 500 mg/l showed a significant increase in chl a , b and total chl amount ( chl a is 11.6 , 12.2 mg/l ) , ( chl b 12.2 , 13.2 mg/l ) ( total chl 14.3 , 15.3 mg/l ) in contrast with control (fig 2 , 3 , 4 ).

### Carbohydrates content:-

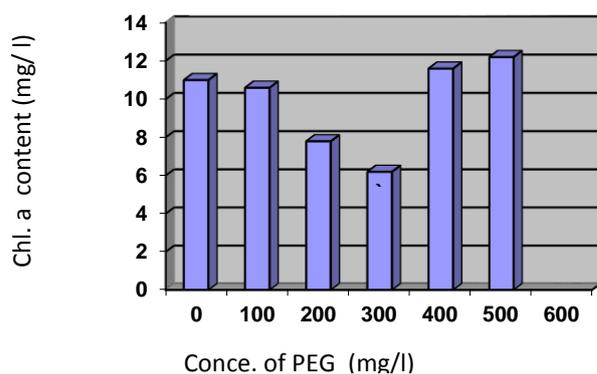
Fig 5 showed a significant increase in carbohydrate amount under 50 mg/ml PEG ( 45 Mg/ml ) in contrast with control (59 Mg/ml ) while in other concentrations (100 , 200 , 300 , 400 mg/l ) carbohydrates content decreased (47, 31 , 35, 37 Mg/ml ) in contrast with control , and there were a significant differences among all levels of drought stress.



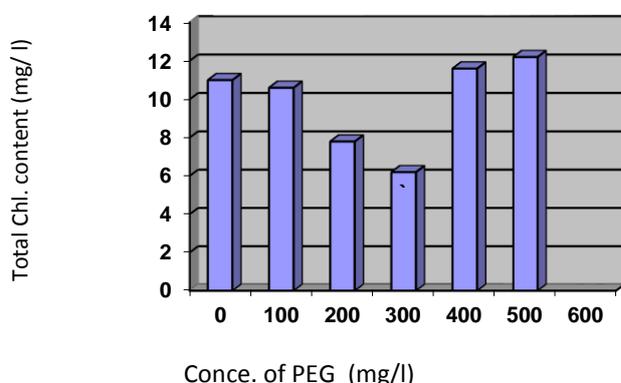
Figur- 1 effect of drought stress on germination percentage



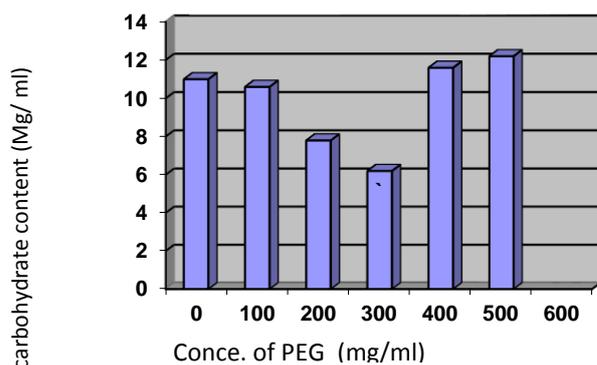
Figur -2- effect of drought stress on chlorophyll a content(mg/ l)



**Figur -3- effect of drought stress on chlorophyll b content(mg/ l)**



**Figur -4- effect of drought stress on total chlorophyll content(mg/ l)**



**Figur -5- effect of drought stress on carbohydrate content(mg/ l)**



## **Discussion:-**

Water stress may result in delayed , reduced , or complete prevention of germination ( Hegarty , 1977). This current study also reveals the save pattern of germination with PEG treatment . Our results are in agreement with the results from other species , such as *Xanthoceras sorbifolia* (Guo et al., 2012), *Secale montanum*

( Ansari and Sharif-Zadeh , 1997) , and *Sesamum indicum* ( Bahramiet al., 2012) Thus reduction in germination percentage may be attributed to lower infusibility of water through the seed coat and initial water uptake of the seed under stress condition

( Turk et al., 2004) and decreased external water potential (H adas , 1979).

In low water stress , chlorophyll a , b , and total chlorophyll contents decreased while by increasing stress their values increased to the heighth amount. Beeflink et al. (1985) reported increase chlorophyll content in onion under drought stress . Kirnak et al, (2001) , Chen et al, (1991) have associated the increased electrolyte leakage to reductions in chlorophyll concentrations ( due to leaf senescence) while Perzad et al,( 2011) found that chlorophyll a , b and total chlorophyll increase in *Matricaria chamomillia* when increasing water stress to 85 and 70 % of field capacity , and then decrease in 55% of field capacity . Drought stress cause an increasing in K<sup>+</sup> concentration ( Majid et al., 2007). Veberic et al ,(2005) found a high transpiration rates were observed in K-fertilized in apple leaves , and this treatment increase photosynthesis .

The decrease in carbohydrates content in low levels of drought stress then increase in high levels similar to that in chlorophyll content, partitioning of carbohydrates , the most common form of fixed carbon translocated from photosynthesizing tissues (sucrose) to heterotrophic plant parts , largely determines how plants grow and respond to various environmental conditions (Koch, 1996) . Our results agree with results of reseaches like Shellenbaum et al,(1999) who found that in tobacco plants which exposed to drought stress pools of sucrose were greater than those of unstressed plants. Jone and Turner (1980) found that content of sugar did not change in fully expanded leaves of sunflower, Riduan et al, (2005) found that total leaf sugar of peanut Singa and Kelinci were the same as that where drought stress and optimal conditions.

## **Conclusion:-**

Distinction of significant differences in germination in *T. aestivum* under all levels of drought stress , and decrease in chlorophyll and carbohydrates contents under low levels of drought stress and then a significant increase in chlorophyll a , b , and total chlorophyll , and carbohydrates contents were found.

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