



## CHLOROPHYLL-BASED SCREENING FOR SALINITY TOLERANCE IN WHEAT GENOTYPES

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### ABSTRACT

Salinity is one of the major environmental factors that reduce wheat productivity. Salinity causes osmotic imbalance in the plant which in turn results in the deterioration of many vital processes, including reduction in the total chlorophyll content. In this experiment the salt tolerance potential of selected wheat genotypes was assessed at different level of NaCl concentration (0mM, 50mM, 100mM, and 200mM). Two wheat genotypes ZAS 42 and ZAS 08 were least affected by the higher NaCl concentration and are reported as salt tolerant.

**Keywords:** wheat, salinity, chlorophyll, genotypes.

### INTRODUCTION

Wheat productivity is affected by biotic and abiotic stresses. Salt stress is one of the most prevalent abiotic stresses that impacts plant growth (Pessarakli, 1991). It is estimated that over 800 million hectares (Mha) of the land is salt affected, which makes over six percent of the total land area in the world (FAO, 2005). Pakistan has about 8.133 million hectares (Mha) of land affected by either salinity or sodicity (Akhtar *et al.*, 2009; Azhar and Tariq, 2003).

Wheat is a moderately salt-tolerant crop and its yield is substantially reduced as the soil salinity level rises to 100 mM NaCl (Shahzad *et al.*, 2012; Munns *et al.*, 2006). Wheat is cultivated both on rain fed and irrigated areas in Pakistan. Irrigated areas contribute 95% towards total national wheat production while rain-fed areas contribute only 5% (Agricultural Statistics of Pakistan, 2011). Irrigated water generally contains sodium, calcium and magnesium salts. After irrigation, the water is used by the crop or evaporates but the salt, however, is left behind in the soil and accumulates. The salt ions when taken up by the plants create an osmotic imbalance which in turn results in the deterioration of many vital processes. One of the major detrimental impacts of salinity is reduction in total chlorophyll content. The loss of chlorophyll makes plants unable to sustain vitality and eventually death occurs. The amount of chlorophyll present in stressed plants can efficiently be used as a parameter to compare the effect of salinity on different wheat genotypes. The present research work was designed to evaluate the salt tolerance potential of wheat genotypes on the basis of reduction occur to the total chlorophyll content.

### MATERIALS AND METHODS

#### Plant material and experimental conditions

Five wheat genotypes (ZAS 42, ZAS 67, ZAS 70, ZAS 08, and ZAS 34) were used in this study. All material was grown in plastic pots in the glass house of Institute of Biotechnology and Genetic Engineering, NWFP Agricultural University Peshawar. Each pot was filled

with 5.25 kg of a mixture of silt and farmyard manure (1:1). Ten days seedlings were transplanted into 15 ml glass tubes filled with different concentrations of NaCl (0, 50, 100 and 200 mM). The seedlings were stressed for a period of 10 days before harvested for chlorophyll analysis. The experiment was conducted in three replications and the experimental lay out was CRD. Genotypes were ranked in their salt tolerance potential by determining the chlorophyll content.

#### Chlorophyll extraction and quantification

Total chlorophyll content was determined by the method of Arnon (1949). ~100 mg leaf tissue was quickly frozen in liquid nitrogen and homogenized in 3ml of 80% acetone. The homogenate was centrifuged at 15, 000 rpm for 10 min at 4°C and the supernatant was collected. Chlorophyll content was determined through spectrophotometer by measuring absorbance at 645 and 663 nm of the supernatant and calculated by using this formula:

$$\text{Chlorophyll Content} = 20.2 (A_{645} - A_{730}) + 8.02 (A_{663} - A_{730}) = \mu\text{g chl/ml}$$

### RESULTS AND DISCUSSIONS

The experiment was conducted in three replications and the design used was CRD. The total chlorophyll content determined per replication of each genotype is given in the Table-1.

The average chlorophyll content of each genotype in microgram per ml was calculated and the percent decrease in the total chlorophyll content of each genotype was determined.

Wheat genotypes ZAS 08, ZAS 34, and ZAS 42 were least affected at 50 mM NaCl concentration while ZAS 70 showed great loss in its total chlorophyll content. ZAS 67, showed a moderate loss to the total chlorophyll at 50 mM NaCl concentration. The reduction occurred in the total chlorophyll content of wheat genotypes at 100 mM NaCl is shown in the Table-1. ZAS 70 exhibited the greatest depletion in its chlorophyll content. The percent



decrease in the total chlorophyll content in ZAS 34 ranged from 22% to 35%.

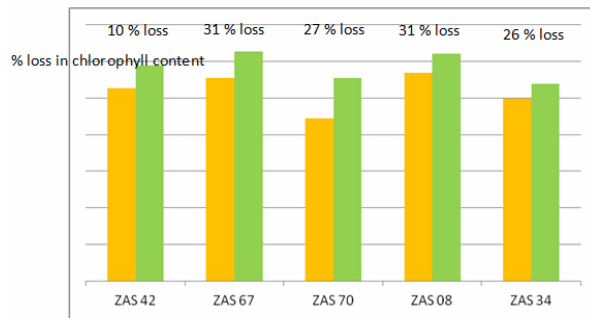
At 200 mM NaCl concentration ZAS 08 were least affected. And total loss of chlorophyll content ranged from 42 to 51% approximately. ZAS 70 and ZAS 67 were the highly affected genotypes at the 200mM NaCl stress and loss in chlorophyll content ranged from 60% to 69%.

ZAS 34 was considered as moderately tolerant wheat genotypes. ZAS 70 was the highly susceptible genotypes to salt, and expressed the greater loss at all stress levels. ZAS 42 and ZAS 08 were determined as the highly tolerant wheat genotypes which exhibited great tolerance at the highest salt concentration.

**Table-1.** Wheat genotypes grown on varying concentration of NaCl.

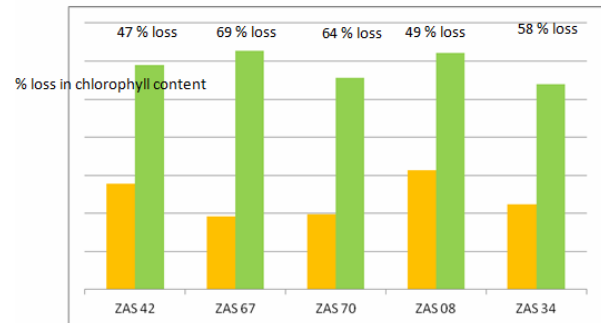
| S/No | Genotypes | 50mM NaCl |       |       |         | 100mM NaCl |       |       |         | 200mM NaCl |       |       |         |
|------|-----------|-----------|-------|-------|---------|------------|-------|-------|---------|------------|-------|-------|---------|
|      |           | R1        | R2    | R3    | Control | R1         | R2    | R3    | control | R1         | R2    | R3    | Control |
| 1    | ZAS 42    | 77.87     | 81.49 | 77.76 | 88.44   | 72.55      | 72.88 | 69.67 | 87.13   | 41.82      | 39.66 | 43.65 | 88.91   |
| 2    | ZAS 67    | 82.72     | 81.93 | 84.77 | 94.17   | 78.23      | 87.72 | 87.22 | 93.22   | 27.59      | 27.93 | 31.23 | 93.12   |
| 3    | ZAS 70    | 67.32     | 65.73 | 67.45 | 83.29   | 45.79      | 44.53 | 41.06 | 83.70   | 34.35      | 27.17 | 27.11 | 82.97   |
| 4    | ZAS 08    | 63.66     | 64.33 | 61.88 | 82.11   | 43.42      | 43.36 | 49.89 | 82.90   | 31.89      | 28.59 | 36.04 | 81.60   |
| 5    | ZAS 34    | 73.06     | 74.98 | 76.39 | 80.76   | 55.19      | 56.24 | 55.38 | 78.44   | 33.55      | 32.42 | 35.00 | 80.19   |

Wheat genotypes grown in 50mM NaCl



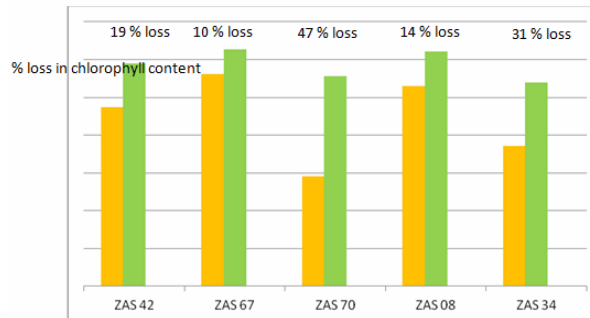
. Green bar represent control  
. Yellow bars represent stress

Wheat genotypes grown in 200mM NaCl



. Green bars represent control  
. Yellow bars represent stress

Wheat genotypes grown in 100mM NaCl



. Green bar represent control  
. Yellow bars represent stress

## CONCLUSIONS

Salinity is a threat to agricultural productivity. Wheat which is the staple food of people in Pakistan is severely affected by salinity. Searching for salt tolerant genotype is the need of the day. Plant breeders have developed several high yielding varieties that grow on normal soil, but to overcome the threatening problem of increasing salinity, salt tolerant germplasm still to be produced. The development of salt tolerant wheat varieties seems to be a very desirable solution to tackle the salinity problem and ensure food security. Two wheat genotypes ZAS 42 and ZAS 08 were identified, in this study, as salt tolerant and are recommended to be used in future breeding programs.

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