MOLECULAR GENETIC STUDIES ON SOME DROUGHT AND SALINITY TOLERANCE GENES IN BREAD WHEAT

By

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B.Sc. Agric. Sci. (Biotechnology), Fac. Agri., Cairo Univ., 2001 M.Sc. Agric. Sci. (Agric. Biochemistry), Fac. Agri., Cairo Univ., 2008

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ABSTRACT

The aims of this study were identifying of some wheat varieties and double haploid response to drought and salinity stress to determine and isolation of tolerant genes and select a gene for cloning it in tobacco plant. The behavior of seven wheat (tritium aestivum) genotypes under salinity and drought stress was studied. The genotypes used were double haploids (DH1, DH2, DH3 and DH4) and local verities (Sakha93, Sids1 and Gemmiza9) and relative water content (RWC) was estimated for the varieties and DH under drought stress 30% polyethylene glycol 6000, after 72 hours of treatment, The genotypes DH4 had the higher RWC (91.93%) while Sids1 had the lowest RWC of 68.23%. under water stress 30% PEG-6000. While, at salinity level 2.5% NaCl, DH3 displayed the highest relative water content (92.71%) and Gimmeza9 had the lowest RWC 75.88%.

Using primers and polymerase chain reaction (PCR) technique, the genes WZY2, WUB3, ZFP22, TaOF1b, STRP and Di19a were detected in all genotypes and gave fragments with MW. 194,234,182,204,228 and 747bp respectively, with variance expression between different genotypes. Three genes were selected for study the expression of whole sequence genes. DH4 the highest RWC genotype under drought stress and DH3 was had the highest RWC genotype under salinity stress at different times 0, 3, 6,12,24,48 and 72h, in leaves and roots, The wzy2 gene appeared 474bp which varied in expression between leaves and roots, while STRP with 879bp appeared in DH4 in root only at 48h, however in DH3 under salinity stress no gene expression was appeared. On the other hand, Di19a with 747bp appeared in DH3 at 24 and 48h under salinity stress. The three genes were isolated and the sequences of the genes were compared with genbank, wzy2 gene was selected and cloned by carried on pBi121 vector, propagated in E. coli and transformed in tobacco leaf disc mediated Agrobacterium by using tissue culture technique the callus, leaves and roots were obtained, the whole plant was transferred to adaptation. The transformed wzy2 gene was detected using PCR reaction technique by using primers compared with control.

Key words: wheat, *tritium aestivum*, drought and salinity stress, relative water content, cloning, agrobacterium, transformation.

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LIST OF ABBREVIATIONS

6BA 6-Benzylaminopurine ABA Absisic acid hZIP basic region/leucine zipper protein Cauliflower Mosaic Virus promoter CaMV35s International Wheat and Maize Improvement Center **CIMMYT** Dimethyl sulfoxid **DMSO** DH **Doubled Haploid DHNS** Dehydrin (DHN) is a multi-family of proteins present in plants Guanidine thiocyanate **GTC** β-glucuronidase **GUS** HR Hypersensitive response IAA Indole-3- acetic acid LEA Lata embryogenesis abuandant MS Murashige and Skoog media NOS Nopaline synthase terminator which induce termination process rate **NPTII** Neomycin phosphotransferase PCD Programmed cell death **PEG** Polyethylene glycol **ROS** Reactive Oxygen species **RWC** Relative water content SA Salicylic acid SOS Salt overlay sensitive genes **STRP** Salt Tolerance Related Protein **SUMO** Small Ub-related modifier TFS Transcription factors Uh Ubiquitin proteins **UPS** The ubiquitin 26S proteasome system **USDA** United States Department of Agriculture **ZFP** Zinc finger protein DH Double Haploid

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