## **Executive summary of UGC MRP**

## (F. 41-515/2012 (SR) dt. 25.07.2012) sanctioned to Dr. NP Singh

Different environmental stresses like drought, salinity, high and low temperature conditions affect the plant productivity. The temperature stress is one of the major abiotic factors that affect crop yield. In India high temperature has a majot impact on the wheat productivity. Plants also synthesizes a wide set of proteins to acclimatize under both biotic and abiotic stresses and develop a defensive mechanism with the activation of many genes in response to different stresses. Plants usually experiences collective effect of temperature and light that result in negative impact on the photosynthesis. However, high temperature exposure to plants disturbs the overall plant's life cycle and ultimately affects metabolic processes. In recent years, transcriptomics and proteomics has been used to identify stress-responsive genes and proteins induced under different temperatures, salinity, cold and water deficit conditions in several vegetative tissues in rice and wheat and general stress related proteins. Since the storage proteins of wheat determines the quality and end use properties of the grain. Therefore, keeping in view the importance of the wheat as one of the important cereal crop in human diet and lack of information about the variation in protein quality due to high temperature stress conditions, the present research was taken up to evaluate the impact of high temperature stress on seed protein quality of Wheat.

The seed protein content was varied from 7.8% to 13.3% in lines 'HD 2687' and 'WH 1021' respectively. The seed protein fractionation studies showed that the glutenins fraction was the major protein fraction ranged from 42.9% to 45.9% followed by gliadins in the range of 28.0% to 32.3%. However, the proportion of albumins and globulins collectively ranged from 23.9% to 27.1%. The electrophoretic patterns of the wheat seed protein resolved two zones of banding patterns. These were HMW i.e. high molecular mass and LMW i.e. low molecular mass gluten proteins. The low molecular weight proteins in the range of mol. wt. 15 kDa to 80 kDa constituted gliadins which were divided into four major classes i.e.  $\alpha$ ,  $\beta$ ,  $\gamma$  and  $\omega$  gliadins on the basis of their molecular weights. The polypeptide patterns of these eleven lines showed three different patterns in the HMW region,  $\omega$  gliadins region and LMW gliadin region. The selected wheat lines were further studied for the polypeptide variations in the LMW - gliadin fractions. The gliadin fractions of all the seven lines resolved

polypeptides in the range of mol. wt. 25 kDa to 64 kDa. Considerable variations were observed in the region of mol. wt. 25 kDa to 64 kDa

The variation in the polypeptide patterns in four seed protein fractions was observed in heat tolerant and heat susceptible wheat genotypes. Whereas, only two polypeptides were observed in region of 14.8 kDa to 16.0 kDa. When comparing the protein profile of heat tolerant lines PBW 502, WL 711, and PBW 343; heat stress susceptible lines NW1014 and WH147, differences were observed, the comparison of polypeptide band of WL 711 under heat stress induced conditions demonstrates that the gradual decrease in intensity of the polypeptides with increased stress intensity. In case of PBW 343 under different heat stress induced conditions the intensity of polypeptide of mol. wt. 86.5 kDa was increased from temperature stress 20°C to 32°C. The glutenin fraction polypeptide pattern was divided into three different regions of mol. wt. 72.0 kDa to 88.0 kDa, 29.0 kDa to 47.0 kDa and 14.8 kDa to 16.0 kDa. Under different heat stress induced conditions the intensity of the polypeptide of mol. wt. 15.2 kDa was increased from 20°C to 32°C in the heat stress tolerant line PBW 343. Whereas, the polypeptide of mol. wt. 46.6 kDa was observed under 24°C, 28°C and 32°C temperature stress conditions. The polypeptide of 14.5 kDa was common among all the treated, control even heat tolerant and susceptible plants, this polypeptide upon 2mercaptoethanol treatment cleaved into 13.5 kDa and 10.4 kDa polypeptides. The polypeptide of the mol. wt. 16.0 kDa was cleaved into 14.4 kDa and 10.4 kDa polypeptide and was observed in PBW 502 control, PBW 343 control and treated plants protein samples. Interestingly, this polypeptide of mol. wt. 33.0 kDa in heat susceptible wheat variety NW 1014 was cleaved into 30.5 kDa and 29.3 kDa polypeptide subunits in control samples but in treated plant protein sample it was cleaved off into 32.0 kDa, 30.5 kDa and 29.3 kDa polypeptide subunits. The most variation was observed among heat tolerant and susceptible wheat lines in higher molecular weight polypeptides. The polypeptide of 70.0 kDa was observed only in WL 711 control and treated samples and that polypeptide cleaved into 71.0 kDa and 55.0 kDa polypeptide subunits. The polypeptides which possessed interlinked disulfide linkage in PBW 343 control and treated samples but not present in other lines were 18.9 kDa, 24.8 kDa, 26.9 kDa, 32.0 kDa, 39.8 kDa, 44.3 kDa, and 55.5 kDa.

In heat tolerant line PBW 502, a total of 133 spots were detected in both control and stressed plants, among these spots 90 protein spots were matched, 6 spots were

found only in heat stressed grains, and 37 spots were found only in control grains. The spots with 1.5 fold or above were considered as up abundant protein and spots with 0.5 fold or lower were considered as low abundant protein under heat stress. A total of 28 spots were shows low abundance under heat stressed conditions. When CBB-R250 stained SDS PAGE of PBW 502 protein samples under control and heat stressed conditions were analyzed they shows several proteins shared between heat stress treatments and control were differentially regulated. The scatter plot of shared proteins spots was generated after pair wise comparison of control and 32°C heat stressed protein samples. When control and heat stressed samples were evaluated, about 47% protein spots were not regulated, about 22% protein spots were up regulated and about 31% protein spots were down regulated. In genotype PBW 343 a total of 148 spots were detected in both control and stressed plants, among these spots 112 protein spots were matched, 36 spots were found only in control grains. A total of 13 spots showed low abundance under heat stressed conditions. The scatter plot of shared proteins spots was generated after pair wise comparison of control and 32°C heat stress protein samples showed that about 46% protein spots were not regulated, about 42% protein spots were up regulated and about 12% protein spots were down regulated. In heat susceptible line NW1014, a total of 154 spots were detected in both control and stressed plants, among these spots 135 protein spots were matched, 19 spots were found only in heat stressed grains. The total of 63 spots were shows low abundance under heat stressed conditions. The scatter plot of shared proteins spots was generated after pair wise comparison of control and 32°C heat stress protein samples. When control and heat stressed samples were evaluated, about 41% protein spots were not regulated, about 12% protein spots were up regulated and about 47% protein spots were down regulated. Whereas, the line PBW 343 consists of 9 polypeptide subunits in each control and heat stressed conditions, and WL 711 consists of 5 and 4 polypeptide subunits in control and heat stressed conditions respectively, and heat susceptible line NW 1014 consists of 4 and 3 polypeptide subunits under control and heat stressed conditions respectively.