(Format No. 13)

SUMMARY OF DOCTORAL THESIS

Name: Gamila Mohamed Idris Elhadi

Title: Genome Wide Association Studies for Wheat Kernel Hardness and Related Traits in Response to Heat and Combined Heat-Drought Stresses
(高温および高温・乾燥複合ストレスに対するコムギ種子硬度
および関連形質のゲノムワイド関連解析)

Kernel hardness of wheat is one of the most important characteristics for milling and baking quality. It is defined as the force needed to crush the kernels. Wheat grain has three major components; those are starch, protein and lipid. Interactions between these three components determine the quality composition of the wheat grain and its suitability. Wheat endosperm texture ranges from very soft to hard. Soft wheat kernels are easy to be fractured, which results in production of large number of intact starch granules, whereas flour that produced by hard wheat, having broken granules and higher levels of starch damage. Hard wheat is more suitable for bread while it is good to use flour of soft wheat for cookies, cakes and pastries due to less protein and starch damage. The flour of bread wheat is used to make bread, chapatti, biscuits, and pastry products.

Kernel hardness is genetically controlled by two puroindoline genes, *Pina* and *Pinb*, which are 60% identical, which are located at the distal end of the short arm of chromosome 5D. Kernel hardness has several related traits, among them kernel weight and shape traits. Kernel weight is considered to be an important approach for further improving yield potential. Also it considered as the most heritable trait among yield components. In addition, milling yield could be increased by optimizing kernel weight and size. Kernel weight is closely associated with kernel size traits, such as kernel length, kernel width, and kernel diameter. Therefore, improving kernel weight and size is a prime breeding target for wheat yield potential and end use quality.

Beside puroindoline genes which was considered major determinants of wheat hardness, many studies have demonstrated the complex nature of this trait and suggest that hardness is affected by several factors. These factors include abiotic stresses such as heat and drought. Several studies have demonstrated that stress from high temperatures and drought can accelerate kernel filling. These stresses compress the timing of key events during wheat kernel development, such as increasing the production of storage proteins and starch synthesis in the endosperm under stress, which together affect kernel hardness beside kernel weight and shape related traits, leading to yield losses and decreased quality. Therefore, identifying varieties that have both high yield and high quality is critical for food security in the context of global climate change. These varieties will also be crucial for breeding programs.

Several studies described kernel hardness and shape related traits under normal conditions, but extensive studies under stress conditions have not conducted. In addition, the genetic factors that affected the change in hardness remained unclear. Hence, better understanding of the change or stability in hardness under stress environment is essential. Thus, to improve wheat genotypes that maintain high yield and quality even under stress condition, knowledge of genotypic and environment interaction is necessary. Therefore, the current study aimed at investigating the effect of heat and combined heat-drought upon hardness and shape related traits and to explore the genetic loci for kernel hardness and shape related traits. I evaluated hardness, weight and shape-related traits and applied genome-wide association analysis to a panel of wheat multiple synthetic derivative (MSD) lines harboring genomic fragments from Aegilops tauschii, grown under optimum conditions in Japan and under heat and combined heat-drought conditions in Sudan. My results revealed promising markers and alleles that will contribute to enhance and maintain high yield and quality under stressed condition and they could be used in wheat breeding after validation.

Chapter one outlines the objectives of this study, providing the hardness and shape related traits overview, importance, and measurements, along with providing the literature review for the relative studies. Also, it elucidates the impact of abiotic stress upon yield and quality beside its impact on hardness and shape related traits.

In chapter two, we studied the effect of heat and combined-heat drought upon kernel weight and shape related traits along with performing genome wide association to a panel of 160 MSD lines. We aimed to explore the genetic loci for weight and shape related traits acts under stress conditions that can be useful for enhancing yield under stress conditions. We identified tolerant line MSD187 that has a good performance under optimum as well as stress condition. We identified stable marker under all condition on chromosome 5D which associated with a candidate gene encoding a RING-type *E3 ubiquitin*-protein ligase and originated from Aegilops tauschii. This marker contributes to increase the kernel size under stress condition and hence yield. The tolerant line MSD187 harbor the positive allele for this marker, which contributes to increase kernel size.

In chapter three, we investigated the impact of heat and combined heat-drought upon kernel hardness. We aimed to identify the genetic loci that contributes for hardness under optimum condition in Japan, and heat and combined heat-drought conditions in Sudan, and to investigate the association between hardness stabilization and stress tolerance. We observed that less reduction of kernel weight is associated with either low change or stable kernel hardness. Also, we found a significant association with hardness under stress on chromosome 4D, along with dissecting several candidate genes associated with the change of hardness under stress.

Chapter four, outlines the general discussion for the study along with providing and elucidating the general understanding for the two studies.

The current work, aimed at investigating the effect of stress upon both yield and quality through studying the effect of heat and combined heat drought stress upon kernel weight and shape related traits beside hardness. The MSD tolerant lines that identified in this study, which performed good under heat and heat drought stress conditions. These lines, along with the stable markers, favorable alleles and candidate genes elucidated here, represent a good resource with which to enhance wheat grain yield under stress and optimum conditions after validation. Among the tolerant lines, (MSD187) which have good performance under all conditions. In addition, the (MSD187) with tolerance potential to heat and heat drought conditions had more stable hardness than the sensitive line (MSD259). Moreover, the significant peak on chromosome 4D, that observed for hardness under stress conditions. This indicates that occurrence of MTAs contributes to the hardness changes under stress conditions. In addition, the MTAs and candidate genes that obtained for hardness could play an important role in understanding the factors that control the changes of the hardness under a stress conditions and their relationships to the stress tolerance. This will help breeders to develop stress-tolerant cultivars that maintain high yield and stable hardness, capable of resisting the adverse effects of global warming, thereby improving food security.