Chapter 1

Translational Genomics for Crop Breeding: Abiotic Stress Tolerance, Yield, and Quality, An Introduction

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Abstract

In the context of global climate change and population explosion, feeding the world's population and addressing the issues of malnutrition, especially in developing countries, are daunting tasks before the global scientific community. The yield gains achieved through conventional breeding are not very promising, as several abiotic stresses such as drought, salinity, cold, flooding, submergence, and mineral toxicity have been leading to significant yield losses and reducing the quality of produce. In recent years, advances in genomics research and next generation sequencing (NGS) technologies have largely facilitated understanding and identifying gene networks that are involved in controlling genetic variation for agronomically valuable traits in elite breeding populations. The availability of genome-sequence information, transcriptomic resources, molecular markers, and genetic maps for major crops such as rice, maize, and sorghum have enabled adoption of genomics-assisted breeding (GAB) approaches, including marker-assisted backcrossing (MABC) and marker-assisted recurrent selection (MARS). Nevertheless, during the last decade significant genomic resources were also developed in less-studied crops and efforts are underway in deploying these genomic tools in breeding. Furthermore, the new bioinformatics approaches and decision-support tools developed are able to enhance the precision in selection and complement the success of GAB approaches.

This volume essentially focuses on the research on abiotic stress tolerance and the quality enhancement of agricultural produce. Further, this introductory chapter summarizes the key success stories and lessons learned in the field of genomics tools for crop improvement. In addition, this chapter also emphasizes the essence of deploying genome-wide association mapping and nested association mapping (NAM), as well as genomic selection (GS) approaches for crop improvements, in the context of the availability of a plethora of low-cost and high-throughput sequencing technologies.

Introduction

Despite continuous efforts to improve agricultural crops, changes in climate in the past two decades have had a tremendous influence on crop production and productivity. Further, climate changes will have a significant impact on the food security of humankind, especially in developing countries (Lake et al. 2012). It is expected that global temperatures will increase about 3°C by 2100 (Schneider et al. 2007), a change that would drastically curtail global crop production. Additionally, other critical abiotic stresses such as drought, salinity, cold, flooding, submergence, mineral toxicity, and so forth hamper the growth, development, yield, and seed quality of crops. In fact, these abiotic stresses represent the main cause of crop failure worldwide, curtailing average yields of all major crops by more than 50%. Furthermore, the quantitative inheritance and low heritability of resistance to these stresses, coupled with a strong Genotype x Environment x Management interaction of the yield response of crops to abiotic stresses, greatly limit a more accurate dissection and manipulation of such response. Since the world population is increasing at an alarming rate, minimizing these losses is also a major concern for all nations, particularly those with a strong increase in food demand. Besides increasing the production potential, the nutritional quality of the produce needs to be improved to avoid the malnutrition that billions are already facing, particularly in developing countries (Müller and Krawinkel 2005; Bouis et al. 2010).

In the context of rapidly growing demand for the staples of our sustenance, conventional breeding programs are struggling to achieve the yield gain required to adequately meet the burgeoning demand for food and plant-derived products (Tester and Langridge 2010). Accordingly, genomics-assisted breeding (GAB, Varshney et al. 2005) approaches are increasingly being adopted to improve the accuracy and effectiveness of selection while allowing for the dissection of the traits controlling the adaptive response of crops to unfavorable conditions. While availability of molecular markers and genetic maps are the prerequisites for GAB, several orphan crops, neglected at the global level but important for food security at the regional level, until recently lacked the genomic resources and platforms to implement GAB. However, in recent years significant progress has been achieved in the development of genomic resources in a number of orphan crops that have thus become genomic resource-rich crops (Varshney et al. 2009, 2010). As a result, GAB activities including trait mapping and marker-assisted selection (MAS) methods, such as marker-assisted backcrossing (MABC) and marker-assisted recurrent selection (MARS), are increasingly being adopted in breeding programs for major crops and have begun to be deployed in less-studied crops as well (Kulwal et al. 2012; Varshney et al. 2012).

Volume I of this book series presents reviews of genomics applications in crop breeding for biotic stress resistance, while this volume (Volume II) focuses on research endeavors in abiotic stress tolerance and the enhancement of the quality of agricultural produce. Four chapters in Volume II deal with ongoing research on tolerance to submergence, salinity, drought, and phosphorus (P) deficiency in rice. Another chapter discusses work on cloning and molecular breeding work for aluminum toxicity in sorghum. Research on freezing tolerance, an important trait in the Western world, in wheat and barley is summarized in another chapter. One chapter is focused on molecular breeding efforts for stay-green tolerance, an important drought tolerance trait in sorghum. Four chapters in the volume are devoted to quality improvement traits in rice, maize, peanut, and tomato. In the last chapter, authors discuss advances in sugarcane genomics and its applications for enhancing yields and ongoing efforts in genomic selection (GS). Some highlights of these chapters have been summarized in this introductory chapter.

Enhancing Tolerance to Abiotic Stresses in Rice

Submergence stress affects more than 15 Mha in lowland rice-growing areas of South and Southeast Asia. Chapter 2 provides insights into the ongoing efforts at the International Rice Research Institute (IRRI), Manila, Philippines, to improve submergence tolerance in rice. Following the identification of *Sub1* (*Submergence1*) locus and three ethylene responsive factors (ERFs) in rice, Septiningsih and colleagues report the development of eight *Sub1* varieties by the IRRI, six of which are already widely grown in several countries.

More than 444 Mha of global rice-growing area is affected by soil salinization (FAO, 2010). Soil salinization is a major problem in coastal areas of the regions where rice-based farming predominates. Reportedly rice yields are reduced by up to 50% when grown under moderate (6 dS/m) salinity levels (Ren et al. 2005). The losses due to soil salinization can be overcome by soil reclamation or by improving salinity tolerance in the crops. Efforts toward understanding the genetic basis of the trait for crop improvement has revealed that several genes are independently involved in salinity tolerance at different stages of crop cycles. In Chapter 3, Platten and colleagues provide an overview of genomics applications in enhancing salt tolerance in rice.

Drought is the major limiting factor to crop production, and cereals especially experience various kinds of drought stresses, depending on the timing and intensity of the water stress relative to the reproductive stage of the crop. In the case of rice, in Asia about 34 Mha of rain-fed lowland rice and 8 Mha of upland rice (Huke and Huke 1997) are frequently subjected to drought stress. Progress in developing high-yielding, drought-tolerant rice cultivars by conventional breeding has been slow, largely because of difficulties in precisely defining the target environment, complex interactions of drought tolerance with environments and management practices, and lack of appropriate screening methodology.

However, during the past decade the availability of large-scale genomic resources and genome sequences have enabled the adoption of various GAB approaches in rice (see Collard et al. 2008). These efforts are summarized in Chapter 4 by Kumar and colleagues.

Sixteen essential elements are required for rice during the crop cycle. The major nutrients such as nitrogen (N), phosphorus (P), and potassium (K) are largely supplied as chemical fertilizers. The excess application of P, owing to its insoluble nature, leads to deficiencies of copper (Cu), iron (Fe), manganese (Mn), and zinc (Zn). Additionally, erosion of P-enriched soils enhances eutrophication in fresh water (Wolf 1996). Most of the rain-fed rice grown in Asia and Africa is cultivated on problematic soils, especially when P becomes unavailable to the crop as it adheres to soil particles. Hence, in this context, development of crops with enhanced efficiency of P utilization and production of higher biomass is essential. Chapter 5 by Heuer and colleagues essentially discusses the issues related to P deficiency in rice production. Further, the authors also highlight the need for the adoption of molecular breeding approaches and summarize the molecular breeding efforts for enhancing P utilization efficiency in rice.

Enhancing Tolerance to Abiotic Stresses in Wheat and Barley

Freezing/cold tolerance in crop plants is most important in the context of global climate change. Freezing tolerance is important in temperate cereals such as wheat (*Triticum* spp.), barley (*Hordeum vulgare*), and rye (*Secale cereale*). Long exposures of winter wheat and barley varieties to non-freezing cold temperatures (Dhillon et al. 2010) will accelerate flowering time (vernalization) and improve freezing tolerance (cold acclimation). In the case of wheat, Kobayashi et al. (2005) reported that *Vrn-Fr1* controls both frost tolerance and vernalization. Chapter 6, by Gabor and colleagues, reports on the developmental plasticity of these *Triticeae* crops

upon onset of the cold stress. This chapter also summarizes the accumulated knowledge of the past 20 years in the area of genetics and genomics of the mechanism of freezing tolerance and the genomic tools available for enhancing freezing tolerance in the *Triticeae* crops.

Enhancing Tolerance to Abiotic Stresses in Sorghum

Aluminum (Al) is the third most abundant element on earth after oxygen and silicon (Ma et al. 2001). It is a light metal that makes up 7% of the earth's crust. Half the arable soils across the globe and especially those in Africa, Asia, and South America are affected by aluminum toxicity (http://www.news.cornell.edu/ stories/Aug07/SoilsKochian.kr.html). Chapter 7 by Magalhaes and colleagues deals with the existing diversity with respect to aluminum tolerance in sorghum and maize germplasm accessions as well as the molecular, physiological, and genetic basis of Al tolerance in both crops. The authors provide insights into the structure and functional analysis of membrane transporters such as Al-activated malate transporter (ALMT1) and multidrug and toxic compound efflux (MATE) involved in Al tolerance.

Furthermore, 20 to 30% of production losses in sorghum are due to lodging. Stay-green trait, however, has been an indirect selection criterion used by breeders for enhancing lodging resistance. Stay green is associated with increased grain yield and grain size in the sorghum crop under terminal drought, a common occurrence in arid and semiarid regions across the globe (Jordan et al. 2012). Among several genotypes identified with the stay-green trait, BTX645 has been a useful resource in developing commercial hybrids (Harris et al. 2007). Four major quantitative trait loci (QTLs) and several minor QTLs can enhance stay-green traits and several efforts are underway to introgress these QTLs into various genetic backgrounds. However, Vadez and colleagues in Chapter 8 report that this undertaking has been quite challenging owing to limited polymorphism among the parental lines for this trait. The physiological, genetic, and molecular breeding aspects for the stay-green trait are discussed at length in this chapter.

Improving Quality and Yield Through Molecular Breeding in Rice, Maize, Peanut, and Sugarcane

Besides increasing production and productivity, agricultural produce also should fulfill the requirement of consumer acceptance in terms of quality, in order to fetch a good market price. Hence, grain quality improvement also forms the major concern for cereal breeders. In Chapter 9, Hori and Yano describe rice grain quality traits in terms of physical and cooking qualities that are of interest to the consumer. In fact, grain quality is influenced by climate changes, such as high temperatures at the grain ripening stage, and grain components such as amylose, amylopectin, and proteins are greatly affected by such changes. This chapter summarizes the efforts to understand the genetics of grain quality traits and the multiple genes/QTL contributing to grain quality. The chapter also emphasizes the need for developing novel quality evaluation instruments/approaches, such as TILLING (Target Induced Local Lesion IN Genome) that can enhance the qualities related to the cooking and eating of *japonica* rice.

About 190 million children under the age of five years are suffering from malnutrition, especially in the underdeveloped and developing countries in Asia and sub-Saharan Africa (WHO, 2009). Malnutrition can be overcome by supplementing dietary requirements with micronutrients or through promotion of dietary diversification. However, these strategies have been only partially adopted and appear not to have improved the nutrient deficiencies in South African children since 1994 (Labadarios et al. 2005). Nevertheless "biofortification" – breeding staple crops with increased nutritional value – has emerged as a potential long-term strategy to improve nutritional security. Babu and

colleagues in Chapter 10 provide a comprehensive overview on biofortification in maize and highlight two specific cases of genetic improvement in maize that resulted in high nutritional value, particularly with respect to essential amino acid content in the endosperm. Besides emphasizing the molecular marker-assisted QPM (quality protein maize) breeding, the chapter also throws light on its impact in the developing world. Furthermore, the essence of provitamin A, Fe, and Zn, and low-phytate content, and the possibilities for genetically engineered, high-lysine maize are elegantly discussed in the chapter.

Peanut is the most important food legume and oilseed crop cultivated in arid and semi-arid regions of the world. About 45-51% of the dry weight of peanut seeds is oil (Chamberlin et al. 2011). Among fatty acids, oleic and linoleic acids are major fatty acids that determine the oil quality; hence the ratio of the two, the O/L ratio, is critical. In Chapter 11, molecular breeding efforts aimed at improving the oil quality in peanut, undertaken at the Kazusa DNA Research Institute and the Chiba Prefectural Agriculture and Forest Center, both located in Japan, is discussed by Kolikonda and colleagues. This chapter also provides the cost comparisons (costs involved) of conventional and molecular breeding programs.

The cultivated tomato, the most popular vegetable crop in the world, is an important model system for genetics and genomics studies. Marker-assisted selection has been employed extensively in tomato breeding for improving many simple traits. Kinkade and Foolad in Chapter 12 look for QTL analysis approaches and focus on the use of new "omics" technology and its potential use for improving fruit quality in tomato breeding. Progress on reverse genetics approaches, such as TILLING and the bioinformatic workflows to handle high-throughput identification of mutations in candidate genes are discussed.

In sugarcane, up until two decades ago most of the breeding efforts for improvement were purely traditional. Chapter 13, by Gouy and colleagues, highlights recent advances in genomics and its applications for enhancing sugar yields. The chapter also highlights the ongoing efforts on genomic selection (GS) for enhancing yield gains in sugarcane.

Summary and Outlook

This volume presents a number of comprehensive and informative articles written by eminent scientists in the area of crop genomics and molecular breeding. It is important to mention here that the traits and crops discussed in this volume provide just some examples on how genomics can help facilitate the enhancement of tolerance to abiotic stresses and quality in crops.

Volume I of this series offers comprehensive reviews of biotic stress tolerance in a range of crops. As compared to the selected examples of GAB for biotic stress tolerance, it is clear that although rice has made significant progress in GAB for abiotic stress tolerance, most success stories of GAB are related to biotic stresses. This may be attributed to the partially qualitative inheritance and higher heritability of disease resistance as compared to abiotic stress tolerance. Similar to submergence tolerance, if the QTLs contribute higher phenotypic variance, GAB approaches such as MAS and MABC can be deployed in breeding programs. However, in the case of tolerance to abiotic stresses and yield, where several and small-effect QTLs are involved, simple molecular breeding approaches such as MABC and MAS are not as effective. In those cases, MARS (Bernardo and Charcosset 2006) and GS (Heffner et al. 2009, 2010, 2011; Heslot et al. 2012; Nakaya and Isobe 2012) are expected to be the most promising approaches. In this context, and in addition to biparental linkage mapping, mapping approaches such as genome-wide association studies (GWAS; Huang et al. 2010; Zhao et al. 2011; Li et al. 2012; Pasam et al. 2012) and nested association mapping (NAM; Hung et al. 2011; Kump et al. 2011; Cook et al. 2012) can be

implemented due to the availability of low-cost, high-throughput sequencing technologies. While GAB approaches are routinely deployed in the private sector and in developed countries, availability of breeder-friendly decision support tools is required for enhanced adoption of GAB in developing countries. In this context, some tools like integrated system for marker-assisted breeding (ISMAB) (https://www.integrated breeding.net/ib-tools/breeding-decision/marker-assisted-back-crossing-tool), OptiMAS (https://www.integratedbreeding.net/node/1407), GS modules (Pérez-Rodríguez et al. 2012; de Los Campos et al. 2013) and platforms like Integrated Breeding Platform (IBP) (https://www.integratedbreeding.net/) are being developed.

We hope that these two volumes will allow graduate students and young scientists to better appreciate the potential of GAB and will encourage them to pursue careers in this exciting area of crop improvement. In addition, GAB practitioners as well as policy makers should be able to use these volumes for developing the road map for the improvement of target crops in their respective geographical areas.

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