



# Omics Technologies in Unraveling Plant Stress Responses; Using Sorghum as a Model Crop, How Far Have We Gone?

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### Abstract

Owe to sedentary and sessile nature of plants, they require efficient innate mechanisms that will allow them to adapt to various sudden environmental changes that pose harmful conditions. Moreover, as animals; more importantly human survival on earth depends largely on plants survival, there is therefore an immense pressure for production of agricultural crops with higher yields and adaptive strategies to environmental stresses. There is therefore need to focus attention to more effective and efficient means that will provide applicable information to improve plants adaptation mechanisms towards their survival in this era of uncertain climatic change. The whole genome sequencing of cereal crops such as maize, rice, wheat, barley, sorghum genomes together with the use of omics approaches such as genomics, transcriptomics, proteomics, metabolomics and bioinformatics have created a platform for further research in establishing networks of interaction between genes, proteins and metabolites that are involved in stress response mechanisms. This review thus highlights these omics approaches employed in understanding potential roles of these signaling molecules and metabolic pathways using Sorghum; a naturally stress tolerant crop as a model, towards optimum development and/or improvement of other economic crops that are often susceptible to diverse environmental stresses.

### Keywords

Abiotic and biotic stresses; Transgenic plants; Quantitative trait loci (QTL); Next generation sequencing (NGS); Single nuclear polymorphism (SNP)

### Introduction

Plant science is one of the oldest areas of scientific research but is still an extremely important and rapidly developing area of study [1]. Plant life plays an essential role in many of today's challenging issues. For example, growing robust crops with increased yield not only affects the agriculture industry, but also improves the well-being of humans. It also plays a role in environmental sustainability and contamination. According to Grierson [2], the growing and

increasingly prosperous human population needs abundant safe nutritious food, shelter, clothes, fibre and renewable energy, and needs to address the problems generated by climate change, while preserving habitats. These global challenges can only be met in the context of a strong fundamental understanding of plant biology and ecology and translation of this knowledge into field-based solutions.

During evolution, plants have developed complex strategies that regulate biochemical and physiological acclimation in order to respond to biotic stress (viral, bacterial, fungal, and oomycete infections [3] and abiotic stress (salinity, drought, water logged, heavy metals and high/low temperature [4,5] evolving from the environment. Biotic and abiotic stresses severely reduce agricultural productivity worldwide [6,7]. Thus, understanding the adaptation strategies employed by plants in order to tolerate and/or resist these biotic and abiotic stresses are essential for improving plant productivity under these challenging conditions; more importantly in economic crops such as maize, rice and sorghum that are often exposed to these stresses. In furtherance of these, extensive study of how a naturally stress tolerant crop like sorghum has evolved mechanisms in coping with these environmental challenges will provide a data based application platform which can be employed in the development or improvement of other stress susceptible economic crops such as maize and rice towards the production of hybrid varieties.

In recent years, the Omics technologies have considerably contributed to studying biotic and abiotic stresses responses in plants [8]. Omics tools have played significant role in crop quality improvement and protection which has brought about increase in agricultural food production by enhancing the quality, taste and nutritional composition of food crops. Through the use of genomics, transcriptomics, proteomics, and metabolomics, the consistency and predictability in plant breeding have been improved, reducing the time and expense of producing better quality food crops that are resistant to stress but still exhibit a high nutritional value [9]. For example, Omics technologies have been able to create information on possible plants-microbes and plants-pests interactions to transfer genes which can influence crop response to climatic conditions, tools now used in genetic engineering for biotechnological applications [10]. Linking genes to traits provides more scientific certainty leading to improved cultivars and understanding the mechanisms of insect and weed resistance [9]. Omics enables a systems biology approach toward understanding the complex interactions between genes, proteins, and metabolites within the resulting phenotype (Figure 1). This integrated approach relies heavily on chemical analytical methods, bioinformatics, and computational analysis and many other disciplines of biology, leading to crop protection and improvements [9,11].

In recent years, genomics knowledge based on Next Generation Sequencing (NGS), gene editing systems, gene silencing, and over-expression methods have provided a large amount of genetic information to help reveal the mechanisms of biotic and abiotic stress responses in plants [12,13]. At the transcriptome level, technological innovations have made it possible to overview the changes that occur at the transcriptomic level under different environmental stress conditions. Microarrays and RNA sequencing techniques are employed to elucidate the differential expression of genes involved in

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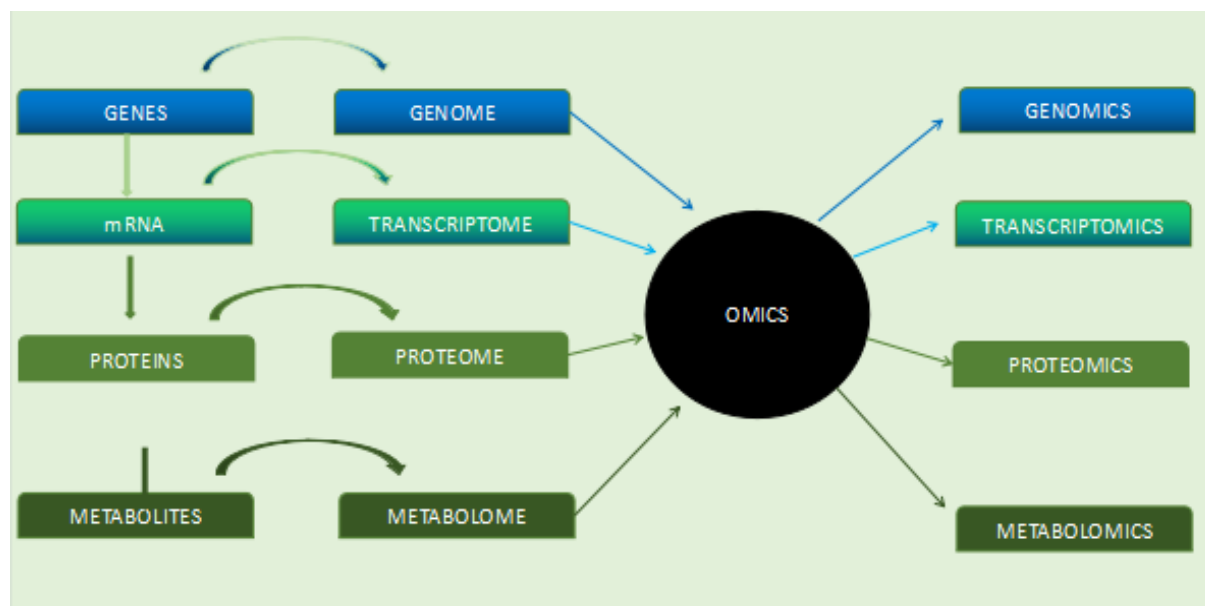


Figure 1: A schematic diagram outlining the flow of information and the application of Omics Technologies.

biotic and abiotic stress responses in a variety of plant species [14,15]. Proteomics and metabolomics are two emerging “-omic” techniques in the post-genomic era [16]. Proteomics has been able to successfully identify and characterize many proteins in cereal and leguminous crops responsible for stress responses and their regulations [17]. Metabolomics focuses on the global profile of the low molecular weight (<1000 Da) metabolites which are the end products of metabolisms in bio-fluids, tissues and even whole organism that are secreted in response to abiotic stresses like drought, salinity, heavy metal toxicity, etc [18]. Bioinformatics has been crucial in every aspect of omics-based research to manage various types of genome-scale data sets effectively and extract valuable knowledge. Integration of these omics approaches will therefore enhance understanding of the molecular mechanisms involved in plants development and more importantly their response towards tolerating environmental stress and thus facilitate knowledge exchange between model crops [11].

*Sorghum bicolor* L. Moench, a  $C_4$  plant, ranks the fifth economically important cereal crop worldwide [19]. The availability of the full genome sequence [20] makes sorghum a  $C_4$  plant in addition to the  $C_3$  plants *Arabidopsis* and rice to study gene products involved in adaptation to drought stress and high tolerance to heavy metal toxicity [21, 22]. Sorghum is sometimes called a “camel” among crops owe to its very high water-use efficiency and its  $C_4$  photosynthesis; it is therefore a good plant resource to explore and identify genes and proteins involved in stress resistance and/or tolerance [23]. Sorghum is widely grown as a staple cereal crop, particularly in Africa and parts of Asia, and also for various other uses in other parts of the world [23]. It is highly cultivated in high temperature and low rainfall areas and in soil with salinity problem [24]. It is the principal source of food for the poor people, in India and Africa. In Australia and the US Southern Plains, grain sorghum represents a drought-tolerant alternative to maize production for livestock feeding, while in Europe, China and North America, interest is also growing rapidly in the use of sweet and/or grain sorghum forms as a potentially drought-tolerant and nutrient- efficient alternative to maize for bio-energy production [25]. Aside from sorghum been a  $C_4$  plant other

attributes that contributes to its hardiness are its well organized deep root system at early seedlings [26], its non-senescence abilities [27] and epicuticular waxy layer [28]. These characteristics make sorghum tolerate stress from drought compared to other cereal crops.

According to IPCC (2007), by the end of the 21<sup>st</sup> century, global surface temperatures are predicted to increase in the range of + 1.4 to + 5.8 °C, and this is expected to increased incidence of drought stress frequency worldwide [29]. However, knowing that sorghum has the ability to withstand long drought condition including other abiotic and biotic stresses, understanding it’s morpho-physiological and biochemical mechanisms will provide better insight towards development of robust crops that can tolerate and adapt to these adverse environmental conditions, alleviating the predicted and/or expected climate change in the future. Thus sorghum is proposed as a model plant for improvement of other cereals crops such as maize, rice that are often susceptible to these abiotic and biotic stresses.

### Brief Description of Omics Technologies

In molecular biology, the term “-ome” refers to the study of the global or total changes in an organism, thus DNA/gene refers to genome, proteins refers to proteome and metabolites refers to metabolome. Omics (genomics, transcriptomics, proteomics, metabolomics, etc.) has come to refer generally to the study of large, comprehensive biological data sets focused on investigating changes in these -omes. Hence plant omics is described as a holistic study of any or all -omes, specifically in plants [1]. The suffix “omics” has been attached to many fields of study, conferring buzzword status and attention (Figure 2 and 3); and it is quickly becoming a vast field and impossible to cover properly in just one article. Thus the following four omics technologies; genomics, transcriptomics, proteomics, metabolomics will be the main focus of this review.

### Genomics

This is the science that deals with the discovery and noting of all the sequences in the entire genome of a particular organism. Genome

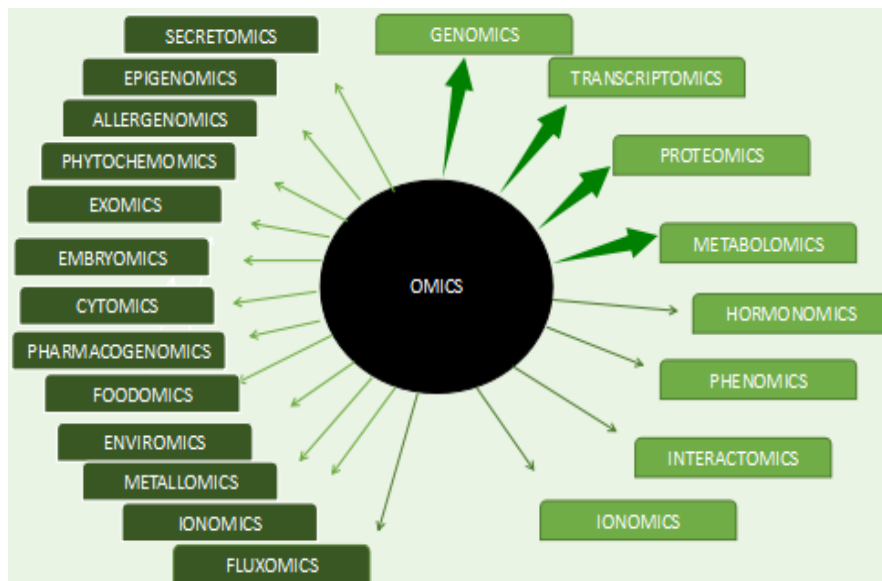


Figure 2: A Schematic illustration of different types of Omics technologies; big arrow indicates the four Omics on focus.

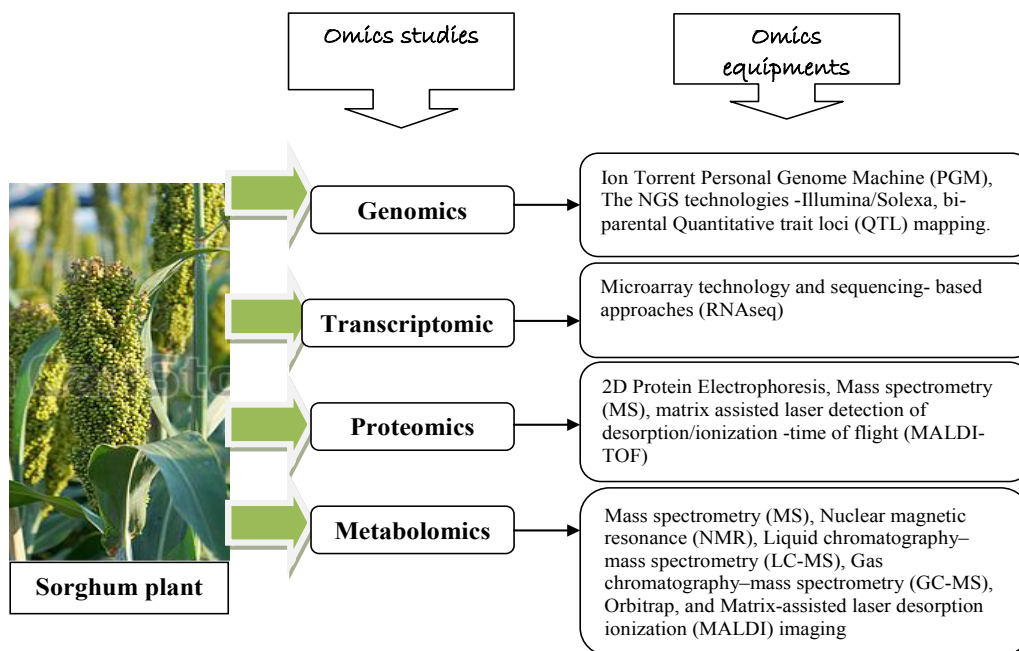


Figure 3: Omics Technologies and the various applications used for their study. Below figure illustrates the afore-discussed Omics Technologies and various applications used in studying them, using sorghum as a model plant.

is the total genetic make-up of an organism. Genomics is a good entry point for the omics field, particularly as the omics nomenclature started with the coining of the word “genomics” [9]. A plant’s domestication and breeding history are recorded in its genome [30]. The completion of the sequence of the first plant genome, *Arabidopsis thaliana* (i.e., thale cress), ushered in the post genomic era in plant research. The Human Genome Project revealed that there are about 20500 human genes, whereas The *Arabidopsis* Genome Initiative [31], reported *A. thaliana* to have 25498 genes. These genomes were

completed along with the rice genome in the early 2000s as reported by the International Rice Genome Sequencing Project. Databases of genetic sequences have been compiled based on submissions from researchers to foster collaborations. GenBank is an annotated collection of publicly available DNA sequences that is frequently updated to provide the latest DNA sequence information [32].

The recent advances in application of DNA marker technologies for detection of Single Nucleotide Polymorphism (SNP) have been used for detection of desirable traits. The Next Generation

Sequence (NGS) technologies using Illumina/Solexa, Ion Torrent Personal Genome Machine (PGM) and Pacific Biosciences (PacBio) sequencing methods have also revolutionized genomic and genetic research through application of a precise and fast phenotyping which is often more expensive and durable than getting single nucleotides polymorphism (SNP) data for thousands of markers [33,34]. Application of bi-parental Quantitative trait loci (QTL) mapping or association mapping techniques to understand the inheritance and genetic variance of complex quantitative traits is also currently in use [35]. QTL mapping is fast and accurate and can possibly discover rare allele and give good estimates of the allelic effects in a given genetic background.

Genomics therefore provides knowledge-based approaches for crop plant biotechnology by enabling precise and controllable methods for molecular breeding and marker-assisted selection, thus accelerating the development of new crop varieties.

### Transcriptomics

This involves the study of complete set of mRNA (also known as the transcriptome), where the non-coding regions have been edited and it only carries information that will be translated by ribosomes to protein. According to Jeanette and Van [9], transcriptomics is described as expression profiling, as it is a study of the expression levels of mRNAs in a given cell population. Unlike the genome, which is roughly fixed for a given cell line, with the exception of mutations, the transcriptome is dynamic as it is essentially a reflection of the genes that are actively expressed at any given time under various conditions [9]. Transcriptomics determines how the pattern of gene expression changes due to internal and external factors such as biotic and abiotic stress.

Transcriptomics is a powerful tool for understanding biological systems, and transcriptomic techniques such as sequencing-based approaches (RNAseq) / next-generation sequencing (NGS) [36] and Microarrays methods [37] provide capability for furthering the understanding of the functional elements of the genome. The Massive Parallel Signature Sequencing (MPSS) which is the prototype of next-generation sequencing (NGS) involves four rounds of restriction enzyme digestion and ligation reactions to determine the nucleotide sequence of complementary DNA (cDNA) ends generating a 17–20 base pairs (bp) sequence as the fingerprint of a corresponding RNA [38]. An analysis of changes at the transcript level can be used to identify new signaling proteins and metabolic processes which are important for plant development and also for stress tolerance [37].

Gene expression studies identify functional gene products that give rise to the phenotype, an information that can be used to create hybrid plant. The task of any breeder is to find reliable traits that can be used to select genotypes that best fit a target environment [39]. By adding a specific gene or genes to a plant, or knocking down a gene with RNAi, the desirable phenotype can be produced more quickly than through traditional plant breeding [33]. These techniques are also applied in biotechnology sector for biopharmaceuticals and industrial compounds.

### Proteomics

It is the study and characterization of the complete set of proteins present at a given time in the cell [40]. As proteins are present everywhere in plants and are responsible for many cell functions; through proteomics it can be determined whether the expression of mRNA results in protein synthesis, which thus illuminates the gene

function. The word “proteome” is derived from PROTEINS expressed by a GENOME. The hundreds of thousands of distinct proteins play key functional roles in the growth, development, yield, flavor, and nutritional value of plants and food products [41]. Through protein expression profiling, proteins can be identified at specific time as a result of response to a stimulus such as disease and insect infestation or temperature and drought, consequently indicating the cellular function of expressed proteins. The use of comparative proteomics has therefore been used to determine and understand the molecular mechanisms involved in susceptibility or resistance which has been applied to enhance plants resistance traits [42, 43]. Plant proteomics has further been extended to translational proteomics that study proteome from expression to functional, structural, and finally the manifestation of desired traits. Through this, proteomics is been applied for food safety authenticity for human health and increased economic values [43].

The proteome has dynamic capabilities unlike the genome that is static in nature. The study of proteins introduces post-translational modifications (PTMs) and provides the knowledge that is important for understanding the biological functions [44]. The PTMs that play important roles during the growth and development of a plant and/or in response to various stress conditions cannot be understood from the genome sequence projects and/or transcript abundance alone. Thus proteomics knowledge provides functional genomics with completeness toward understanding the process [45]. Technologies used in proteomics includes, 2D Protein Electrophoresis [46], Mass spectrometry (MS), matrix assisted laser detection of desorption/ionization -time of flight (MALDI-TOF) which separates based on charges and molecular sizes respectively of which MALDI-TOF has been described to be more accurate in determining proteins molecular weights [47].

### Metabolomics

It is the study of chemical processes providing a linkage between genotypes and phenotypes [48]. Proteomics identifies the gene products produced, whereas metabolomic studies determine whether the expressed proteins are metabolically active and identify the roles and biochemical processes of the resulting various metabolites. The metabolome is dynamic and subject to environmental and internal conditions. Metabolic profiling therefore provides an instantaneous picture of what is occurring in the cell at a particular plant stage or under influence of external environmental factors [49].

Metabolic profiling through mass spectrometry (MS) and nuclear magnetic resonance (NMR) analyses have been used to ascertain plant metabolic responses and to investigate regulations and changes that are related to environmental conditions of light, temperature, humidity, soil type, salinity, fertilizers, pests and pesticides, and genetic perturbations [48]. Other applications used are liquid chromatography–mass spectrometry (LC-MS) for studying secondary metabolites [50,51], gas chromatography–mass spectrometry (GC-MS) for volatile and primary metabolites such as sugars and amino acids [51 - 53], Orbitrap for accurate mass measurements due to its fast scan times with improved deconvolution, run times and high mass accuracy [50, 51], and Matrix-assisted laser desorption ionization (MALDI) imaging for visualization of the spatial distribution of proteins and metabolites [54].

### Applications of Omics Technologies

Generally, all plants must respond to external stimuli which could result from environmental hazard and other living factors (abiotic



and biotic stresses) [55]. They must adjust their omics profile in order to cope with changes in the environment. Metabolic pathways leading to the synthesis of anti-stress metabolites are enhanced [56], while some biochemical pathways (like photosynthesis, carbon fixations among others) may be inhibited in order to create avoidance, tolerance or resistance to the stresses [57]. The plants organ or tissue type, developmental stage and genotype are other factors that also influences plant response to stress [58]. At specific developmental stages, plants are either more or less sensitive to particular stress or possibly multiple stresses [56].

Sorghum is a plant that possesses the characteristics feature to withstand long drought stress [59]. The adaptation of sorghum to drier environments led to its domestication in north-eastern Africa [60]. Been a tropical plant, sorghum does not tolerate frost. Thus at low temperature (below 20°C) several early developmental processes including emergence, seedling vigor and general metabolism are affected [61].

Early-stage chilling stress is a common type of abiotic stress in young sorghum [62]. Use of genomics approaches has recently been helpful in the discovery of Sorghum key traits that modulate its survival under low environmental temperature conditions. Anami [63] identified 350 Quantitative Traits Locus (QTLs) related to abiotic and biotic stress tolerance in sorghum. Bekele [23] reported that emergence and root establishment are very vital for development and survival of sorghum seedlings under cold stress. It is shown that sorghum possesses highly interactive epistatic quantitative trait loci (QTL) hotspots and an unknown QTL on chromosome Sb06 in its genetic make-up that conferred the significant tolerance effect on prolonged cold stress, and the traits were found to regulate different physiological mechanisms contributing to maintenance of growth and development despite the cold temperatures. Kebede [64] and Harris [65] also extensively studied chromosomes Sb02, Sb03 and Sb05 and reported that several QTL on these chromosomes are involved in the stay-green characteristics and responsible for sorghum maintenance of its greenish colour when exposed to drought stress for 60 days. Marker assisted techniques (MAS) like single marker analysis, simple interval mapping (SIM), Composite interval mapping (CIM) and Association mapping have also been successfully used to identify specific loci responsible for low temperature tolerance in sorghum genome in a population and these genes have been isolated and introgressed into elite lines, and as a result, remarkably showed resistance to early chilling stress [66 - 68]. Recently, Upadhyaya [69] evaluated 242 accessions of ICRISAT sorghum mini core collection for seed germination and seedling vigor at 12°C to measure its cold tolerance using 162,177 single nucleotide polymorphism marker for Genome-wide Association analysis (GWAS). Results of this analysis showed only one marker locus (Locus7-2) associated with low-temperature germination which was supported by four lines of previous experiments. Of more recent work of Maulana [70] showed chilling stress tolerance of sorghum germplasm, by using association analysis between markers and seedling traits to identify chromosomes that harbors QTLs associated with cold tolerance in sorghum. Other studies have also reported QTL for early growth parameters under cold conditions [23,68,71,72].

In the work of Chen [73], using Genome-wide association study (GWAS) for traits responsive to heat stress at the vegetative stage to dissect the genetic basis of heat tolerance in sorghum in an association panel. It was found that nine SNPs and five SNPs associated with leaf firing and leaf blotching respectively, which could serve as candidate DNA markers for selecting heat tolerance. Moreover,

candidate genes near these SNPs are shown also to be directly linked to biological pathways involved in heat stress responses. Thus these plants must have adapted to the diverse agro-climatic regions by developing abundant trait diversity [74]. In order to develop quality inbred lines, these diverse traits in sorghum must therefore be identified and dissected. For efficient location/identification of these traits, remarkable improvements have being made on the use of association mapping for the detection of Quantitative loci (QTL) on chromosomes. Bouchet [75] developed a Nested Association Mapping (NAM), a great resource for trait mapping in sorghum and it has demonstrated a high value for dissection adaptive traits. Using NAM, 10 diverse global lines crossed with an elite reference line RT×430 to discover several QTLs having genetic variation of 65% of flowering time variance and 75% of plant height variance in sorghum, and 57,411 recombination events were captured. To ascertain this development, though the efficiency of NAM population for trait dissection was compared with GWAS using joint linkage mapping for flowering time and plant height, result showed that the detection of QTLs was remarkably three times greater for NAM than GWAS and the flowering time and plant height were more consistent with the NAM population [75].

Several other improvement in mapping of Quantitative trait loci like Multi-Parent Advanced Generation Inter-cross (MAGIC) to produce immortal MAGIC inbred lines among others, have been developed to dissect the genetic architecture of abundant Quantitative trait loci (QTL) in population of sorghum [76, 77] and these panels have been used for GWAS of several traits [77- 79]. MAGIC; a wide genomic tool and powerful next generation mapping resources has demonstrated to have a high utility for gene mapping. By intercrossing 19 diverse founder lines, through a series of paired crosses with a genetic male sterile source, followed by 10 generations of random mating, showed no evidence of population stratification, consequently the desirability of the population for genome-wide association studies (GWAS). The genotyping-by-sequencing of a subset of 200 MAGIC lines identified 79,728 SNPs spanning high gene rich regions and 19 founders were found to form three clusters, each with considerable genetic diversity [77].

Transcriptomics analysis of sorghum bicolor indicates its survival under combined heat and drought stresses. Johnson [37], used microarray consisting of 28585 gene probes to identify gene expression changes when sorghum is exposed to drought and heat stresses. Results show changes in gene expression of approximately 4% and 18% of genes on the chip following drought and heat stresses respectively and in response to combined stress, and approximately 20% of probes were differentially expressed. Whilst many of these transcript changes were in common with those changed in response to heat or drought alone, the levels of 2043 specific transcripts (representing 7% of all gene probes) were found to only be changed following the combined stress treatment. Ontological analysis of these 'unique' transcripts identified a potential role for specific transcription factors including MYB78 and ATAF1, chaperones, unique heat shock proteins (HSPs) and metabolic pathways such as polyamine biosynthesis in the combined stresses responses. It is therefore proposed that functional characterization of the genes and pathways thus identified could lead to new targets for the enhancement of plant stress tolerance, which will be particularly important in the face of climate change and the increasing prevalence of these abiotic stresses.

Furthermore, transcriptome analysis required accurate annotation of each transcriptional unit and its expression profile is

very essential. Thus in RIKEN Biomass Engineering programme, *Sorghum bicolor* BTx623 primary genome database was established in order to understand stress tolerance mechanism and its applications to biomass engineering [80]. Normalized full-length cDNAs library from five growth stages of whole sorghum plant parts and three growth stages of panicles was constructed and over 38,981 high quality expressed sequence tags (ESTs) clones were obtained. Applying three tissues specific RNA-Seq analyses (panicles, seeds and stems); results obtained from the data supports the ESTs defined genes existence. The database which is publicly available is a valuable resource in comparing various transcriptome data [80].

RNA sequencing is another omics tool used for transcriptome analysis of plants to discover their defense mechanisms during adverse environmental conditions. Dugas [81], in the course to elucidate genes and genes network that contributes to sorghum's tolerance to long-term drought utilized next generation sequencing (NGS) technology to examine the transcriptome of three independent sorghum plants by inducing the water limiting with osmotic stress and exogenous Abscisic acid (ABA). RNA-Seq sequencing was performed by Illumina's Real Time Analysis (RTA) and aligned to the Sorghum genome download [81]. Result of the RNA-Seq of Sorghum bicolor in response to osmotic stress and Abscisic acid (ABA) in this experiment can be accessed [81]. Numerous genes were highly expressed and validated. 34,496 gene models were generated from RNA-SEQ cDNA of which only 34,144 of the genes were discovered to be annotated genes. Further analysis on transcriptional activities found that only 28,335 unique genes out of which 25,568 and 2,649 are high confidence and low confidence protein coding respectively and 118 non-coding pri-microRNAs.

Furthermore, different transcription factors (TF) family such as AP2-EREBP and DRE2COREZMRAB17 are over represented in the differentially expressed genes from shoots when treated with ABA and set of 6 TF: Bzip-Sb04g034190, C3H-Sb09g006050, Sb03g003110, HSF-Sb09g033750, Sb10g021800 and MYB-related-Sb04g034190, were up-regulated in all tissues/treatment combinations [81]. These transcription factors accumulation are discovered to be involved in plant defense by up-regulating the expression of genes responsible for combating oxidative stress generated from limited water availability [82]

Early season cold stress (12-15 °C) limits the stand establishment and seedling growth of sorghum in low temperate regions. Chopra [83], performed a new strategy of transcriptome profiling between known cold sensitive (BT×623) and tolerant sorghum (HKZ) lines using RNA sequencing technology by integrating analysis of differential gene expression and identification of bi-allelic single nucleotide polymorphism (SNP) 41,603 Single nucleotide polymorphism (SNP) were identified and 114 SNP sites were validated using endpoint genotyping technology. GMAP (a genomic mapping and alignment program for mRNA and EST) was used to align the raw sequence to the sorghum reference genome derived from Phytozome v12.1.6 that hosts 93 assembled and annotated genomes, from 82 Viridiplantae species [83]. When comparing the 1910 differentially expression patterns between contrasting sorghum genotypes under cold and control temperatures, about 625 and 344 transcripts showed significant changes for BT×623 and HKZ respectively at 14 to 28°C. This work also showed significant increase in accumulation of transcription factors like Dehydration-responsive element-binding factors, C-repeat binding factors and Ethylene responsive transcription factors are up-regulated during the cold

stress and these factors are known to enhance the expression of stress responsive genes [82].

Ndimba [84], proteomics analyses on whole sorghum organs (leaves sheath and roots) under salinity stress using 2DE-PAGE and MALDI-TOF mass spectrophotometer reported increased concentrations of hyper-osmotic stress responsive protein alpha-galactosidase polypeptides in all organs and high expression of Heat shock protein (HSP70); a stress responsive protein in the root of stressed plants. The alpha galactosidases which are known to play an important role in the cell wall metabolism and in plant growth and development thus indicate that the plant uses these proteins to continue its metabolism and enhance its growth and developments under the salinity stressed condition.

Furthermore, various methods have been used recently to determine early stage chilling response in sorghum and other tropical crop plants such as maize [71,85,86], and it was found that sorghum contains considerable genetic variability traits for cold stress sensitivity, non-senescence character and minimum basal temperatures.

Heavy metals are among the most widespread soil contaminants, and they could cause damage to plants by inducing alterations in major physiological and metabolic processes [87,88]). The effects of certain toxic metals like Cadmium (Cd), Zinc (Zn), Lead (Pb) on plants growth and development have been a concern for the plant scientists [89], however, it has been demonstrated that sorghum plants were able to accumulate large quantities of heavy metal in the shoots [90, 91], as well as highly tolerant to metal pollution [21,22].

A more recent proteomics research by Roy [92] on sorghum's response to cadmium stress was carried out using two-dimensional gel electrophoresis (2DE). A total of 33 differentially expressed proteins were found after the seedlings were exposed to various concentration of cadmium and analyzed using MALDI TOF/TOF MS. Result indicated that 15 proteins were up-regulated while 18 were down-regulated. This proteomic results showed that the high number of down-regulated proteins could have had an inhibitory effect on carbon fixation, ATP production, and regulation of other proteins syntheses such as proteins involved in carbohydrate metabolism, transcriptional regulation, translation and stress responses. This is because that result indicates the rapid decrease in ATP synthase, Ribulose-bisphosphate carboxylase (Rubisco) and Haloacid dehalogenase involved in ATP production in response to stress. Thus, sorghum seedlings under the presence of heavy cadmium stress would rather up-regulate proteins involved in ATP utilization than production.

Furthermore, 3 proteins cytochrome P450, flavonoid 3', 5'-hydroxylase and glutathione-S-transferase (GST) proteins were markedly expressed at a higher level in transgene compared to the control experiment in the sorghum seedlings [93]. These are stress related proteins which are involved in scavenging free radicals generated in the cell. Therefore, expressions of these anti-oxidative enzymes could serve as a signal transduction for gene expression which will result in protein synthesis and metabolism effluxes control [93]. Another protein that was up-regulated in the presence of stress is G2/mitotic-specific cyclin-2. This protein is involved in cellular metabolism, suggesting that this protein may play essential role in cell growth during stress. Physiological analysis on the leaves showed an increase in weight of leaves meaning the leaves were able to accumulate large quantities of heavy metals.

Other very distinct omics studies on sorghum's molecular responses or adaptation to environmental stresses have also been reported. Ourania [94] determined the physiological responses of young sorghum seedling grown under drought stress induced by polyethylene glycol (PEG) 6000 at two levels of stress (2.5% and 5% PEG), for 7 days using Gas-chromatography combined with Mass-spectrometry (GC-MS). Comparative metabolite profiling of the test experiment with the control experiment revealed that drought stressed sorghum substantially increased its metabolite content, yielding 143 quantifiable compounds with highly reproducible patterns in both leaves and roots, a result which is higher than that of the unstressed sorghum plant. Result revealed such compounds belongs to different classes, including amino acids, organic acids, sugars, polyols, phosphates, polyamines, nitrogenous compounds, etc. majority of the sugars were stated to be increased in both roots and leaves of stressed sorghum with the most profound increase found in the level of D-glucose and fructose for leaves and D-lyxose and D-(+) trehalose for roots, of which an accumulation of D-(+) trehalose substantiates earlier findings pointing to its association with drought stress in bacteria, fungi and plants which tolerate extreme drought conditions [95]. Ourania [94] study does not report proline and glycinebetaine accumulation in the stressed sorghum plant at both concentrations. But given the fact that proline, glycinebetaine and sugars functions as osmolytes in most plants under osmotic stress [96-98]), thus proline and glycinebetaine's absence in the sorghum exposed to drought is certainly intriguing and should be further studied.

Other metabolites of great importance were observed in concentrations of galacturonic acid and isopropyl b-D-1-thiogalactopyranoside, both being significantly increased under water deficit stress [99]. More importantly, 2-keto-L-gulonic acid was produced exclusively in response to stress while, acetol was found only in control plants. These findings are consistent with the well-established role of sugars in osmotic adjustment and adaptation to various abiotic stresses [99]. Furthermore, certain amino acids such as D-alanine, L-alanine, L-lysine, L-ornithine, L-serine and L-threonine presented significantly higher concentrations upon stress induced at 2.5 % than 5 % PEG. The high concentrations recorded for these amino acids in the stressed sorghum plant is in-line with previous existing findings that stress induces protein degradation in plants leading to high yield of amino acids which plays a vital role as osmo-regulatory elements [95]. Polyamines are generally viewed as protective agents that help to scavenge free radicals under various stress conditions [100,101].

Biotic stress mostly common to sorghum is *Striga* parasite. The *Striga* problem is particularly acute in Africa, where the native giant witchweed; *S. hermonthica*, is widespread and most damaging to many cereal crops including millet, rice and maize [102]. Genomic studies have discovered high polymorphism of multiple disease resistance genes (NBS-LRR; nucleotide binding site plus leucine-rich repeat genes) in the genome of sorghum. Sorghum NBS-encoding genes were also significantly enriched in the regions of the genome containing fungal pathogen disease resistance QTL; with the diversity of the NBS-encoding genes influenced by the type of co-locating biotic stress resistance QTL [103]. Quantitative diseases responses (QDR) hotspot in sorghum's genome containing 15 QTL for 6 traits have being discovered from 4 studies [104-106].

More recently, Gobena [107], reported the identification of a Mutant allele at the *LGS1* (*LOW GERMINATION STIMULANT 1*)

locus regulating *Striga* resistance in sorghum thereby reducing *Striga* germination stimulant activity at the early stage and the protective chemistry resulting from mutation. To validate this discovery, *Striga*-resistant sorghum varieties were mated with Chinese landrace "Shanqui Red" which result into high germination stimulant activity generating a Genetic mapping population of 600 Recombinant Inbred Lines (RIL) [107].

Another fungal pathogen is *Colletotrichum sublineola*. These pathogens cause anthracnose in sorghum during these conditions [108]. Symptoms of its infections are leaf blight and stem rot leading to retarded growth and reduction in yield. In order to identify anthracnose resistance loci in sorghum, Felderhoff [108] crossed 'BK7' with susceptible inbred 'Early Hegari-Sart' to generate biparental mapping population of F<sub>3,4</sub> and F<sub>4,5</sub> sorghum lines using a total of 5186 and 2759 informative Single nucleotide polymorphism (SNP). This experiment identified anthracnose resistant loci on chromosomes 7 and 9 in the two populations. Independent genetic analysis conducted on the lines derived from a cross between sweet sorghum 'Mer81-4' and 'BK7' which narrowed the resistance locus on chromosome 9 was used to validate this Quantitative trait loci (QTL) and the resistance allele derived from 'BK7' population was also genotyped. Result indicated they contain multiple classes of defense-related genes based on sequence similarities and gene ontologies. Furthermore, Burrell [109] sequence the anthracnose resistance genomic DNA from SC748-5 (a grain sorghum that has been identified to have strong resistance to the disease; [110], and also mapped the major QTL, using both composite interval mapping and inclusive composite interval mapping (ICIM), result shows a consistent QTL on chromosome 5. Patil [111] also carried out a QTL analysis on two anthracnose-resistance lines of resistant cultivars SC155-14E and SC414-12E. Result shows a series of QTL associated with anthracnose-resistance, three of the QTL were reported to control stable resistance across all environments, while two QTL were environment specific and a major anthracnose-resistance QTL on chromosome 9 of the SC155-14E genome was reported to be observed in all environments and controlled from 19 to 62% of the phenotypic variance.

Other studies not limited to the aforementioned above have shown how omics technologies have been involved in interpreting sorghum plants adjusting its omics profiles and physiology in order to adapt/respond to abiotic and biotic stresses. QTLs conferring resistance to various foliar diseases have also been identified and these include anthracnose, rust, zonate leaf spot, drechslera leaf blight and target leaf spot. A total of 12 quantitative trait loci (QTLs) for 5 foliar diseases on 3 sorghum linkage groups (SBI-03, SBI-04 and SBI-06) were detected, accounting for 6.9 - 44.9% phenotypic variance [112], Insect resistance: a total of 29 QTL were detected by multiple QTL mapping (MQM); 4 each for leaf glossiness and seedling vigor, 7 for oviposition, 6 for deadhearts, 2 for adaxial trichome density and 6 for abaxial trichome density [112]. Interestingly, QTL identified in this study correspond to QTL/genes for insect resistance at the syntenic maize genomic regions, suggesting the conservation of insect resistance loci exist between these crops [113].

Diana [114], used next generation sequencing (NGS) technology to examine the functional transcriptome profile of sorghum, and the transcriptional activity yields 28,335 unique genes from sorghum root and shoot tissues subjected to polyethylene glycol (PEG) - induced osmotic stress or exogenous abscisic acid (ABA). These genes expressed were revealed to play a vital role among various metabolic pathways including abscisic acid and 13-lipoxygenase, salicylic acid,



jasmonic acid, and plant defense pathways. Furthermore, Diana [114], examined transcriptome analyses data of sorghum with published transcriptome analyses for rice [115,116], maize, [117,118] and *Arabidopsis* [119,120] and discovered more than 50 differentially expressed, drought-responsive gene orthologs for which no function had been previously ascribed, using decision tree. From the analyses, only two sorghum genes, Sb01g045990 and Sb03g005990, and their orthologs were found to be differentially expressed in all four species.

### Merits and Demerits of these Applications towards Abiotic and Biotic Plant Stress Responses

Several researches have been conducted on plant responses to stress through the study of their omics profiles. These researches have generated results which have created new dimensions in various sectors of life involving quality crop utilizations which extends to improving the lives of humans at large. Plants have two major divisions, the shoot (leaves, stem and fruits), and the roots. Series of biochemical reactions like carbon fixation, glycolysis, nitrogen fixation, photosynthesis, starch formation, protein synthesis and degradation, fatty acid synthesis among others takes place in either of the divisions at regulated intervals. Plants omics studies through extensive researches on the plant morpho-physiological and biochemical characteristics using high throughput technologies have been able to decipher where, when and how these molecular reactions takes place when exposed to environmental biotic and abiotic stresses. These large data gotten from plants omics studies have been well organized and saved and for further studies and consultations in bioinformatics tools. Then the question comes, what are the benefits of these extended researches on plants?

Plants omics researches have contributed in the plant science field and all walks of life in many ways (Figure 4) but not limited to:

- Development of new biological tools used for system analysis.
- Discovery of stress responsive genes for transgenic practices.
- Breeding of pre-harvest sprouting/submergence stress cultivars to increase yield.
- Improvement/protection of crops through intercropping practice in farming.
- Prolonged access to quality foods in the market.
- Development of novel biological tools used for system analysis.

Plant Omics research has come a long way since the first two-colour microarrays were hybridized [121]. In early experiments, only a few treatments were conducted and significance was often assessed based on whether an arbitrary threshold of a twofold change was exceeded [122]. Many tools have been developed to visualize microbe and animal omics data, especially microarray data, on biological pathways focusing mostly on microarray data [123, 124]. However, when the first 'full genome' *Arabidopsis* arrays became available in 2002 [125], so also the rice genome in the early 2000s [126,127], there was still a clear lack of user-friendly tools for plants. Due to these facts, user-friendly tools that can compile the large omics data of plants across different species, integrate and interpret them for proper visualization in a holistic view were developed, and this led to the advent of Mapman software [128]. Mapman, a tool which was initially developed for use with *Arabidopsis* [122], and it works by

relying on its ontology to classify genes and metabolites, and provide a modular system to visualize the results in the context of pathways and processes. It is a software tool that supports the visualization of profiling data sets in the context of existing knowledge [122] and its use have been extended to the analyses of other plant species. MapMan is composed of a set of Scavenger modules, the Image Annotator module and the Page-Man module. Scavengers organize genes, enzymes, proteins or metabolites into functional categories ('BINs' and 'sub-BINs') to generate hierarchical and essentially non-redundant gene ontologies ('mapping files', a file that stores the association of genes, metabolites or proteins to MapMan BINs). The Image Annotator module visualizes data on diagrams ('maps') of biological pathways or processes relying on mapping files. The Page-Man visualizes data by compressing the response of whole pathways (all of the genes in a sub-BIN or BIN) down to single-coloured rectangles. Mapman have being used to compare transcriptional responses between crops which are phylogenetically different. Usadel [122] used Mapman to analyse the transcriptional response of the crop plant maize to diurnal changes and an extension of the night and also visually and systematically compare responses in the maize and *Arabidopsis* and this has yielded promising results. Indeed, MapMan ontologies are already available for rice [129], potato [130], grapevine [131], *Chlamydomonas* [132], tobacco [133]. It has also being used to visualize gene expression differences for sorghum seed of different genotypes [134] and for dedicated transcript arrays for tomato [135].

Plant MetGenMAP is another tool like Mapman. This system is noted for its efficient and rapid retrieval of significantly altered biochemical pathways and biological processes under given conditions and transcriptional events and/or metabolic changes in a pathway can be easily visualized. Je-Gun [136] demonstrated the functions and application of the system using data sets from *Arabidopsis* (*Arabidopsis thaliana*) and tomato (*Solanum lycopersicum*), respectively. Further tools were released that support the functional analysis of Omics data for plant pathways, such as KaPPA-view [137], Cyc-related web sites [138], Vanted [139] and Reactome [140].

These strategies should therefore be applied to a very wide range of crops and wild species which are susceptible to stresses and possibly generating molecular solutions to its effect on plants productivity. With these achievements, researchers and software programmers are working on developing more robust and user-friendly tools for omics data analysis and interpretation as the full understanding of how plants survive in very harsh environmental conditions could create an insight in understanding humans' life processes in respect to their environments.

### Discovery of Stress Responsive Genes for Transgenic Practices

Linking genes to traits provides more scientific certainty leading to improved cultivars and understanding the mechanisms of insect and weed resistance. This success has led to discovery of plant resistance to herbicides for better pest management [9]. The extensive study of *Arabidopsis*, sorghum, rice, maize and other leguminous plants omics profile in response to stress has brought about improvement of crops that are highly susceptible to environmental stresses. Today's agriculture faces a daunting task of ensuring food security to the increasing human population on this planet [141]. A great proportion (more than 60%) of this population depends on some energy giving food as their staple food, of which rice (*Oryza sativa* L.) is one of the mostly consumed foods worldwide [142]. Rice contributes up to 20%



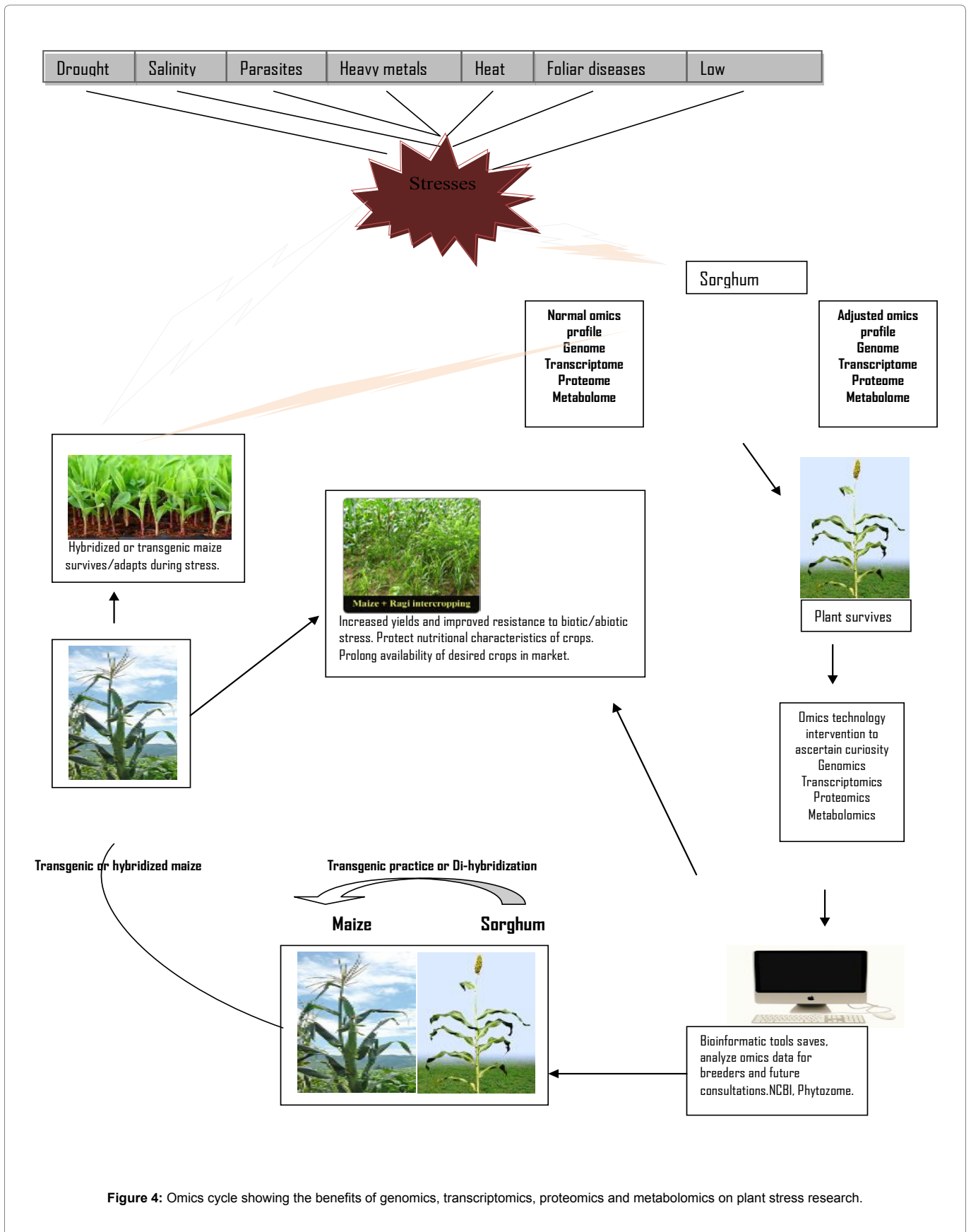


Figure 4: Omics cycle showing the benefits of genomics, transcriptomics, proteomics and metabolomics on plant stress research.

of the calories consumed by human nutrition worldwide. Therefore, rice production must increase during the coming time in order to keep pace with increasing world population. Both abiotic and biotic stresses frequently prevent the attainment of optimum growth and yield of rice. These stresses include high salinity, drought, heat, and cold which have negative effect on the yield and vegetative production of rice, and cause a key risk to worldwide food safety [143,144].

Amongst the various environmental stress factors listed above, salinity is the main hazardous factor limiting crop productivity, especially rice at its young stage [145]. Due to this fact, omics based approach have been used to decipher how rice can be improved against salinity stress. Researchers have used Next Generation Sequencing (NGS) to discover Quantitative Trait Loci (QTL) for salinity stress tolerance in plants [146]. Plants react to salinity by limiting the intake of toxic ions like sodium ( $\text{Na}^+$ ) and regulate their osmotic potential by producing compatible solutes (sugars, glycinebetaine, proline) [98] and partitioning toxic ions into the tonoplasts to maintain low  $\text{Na}^+$  levels in the cytoplasm [147]. Salinity tolerant transgenic rice plants were generated by getting ideas from the above observation [148]. Xu [149] produced transgenic rice by introduction and over-expression of late embryogenesis abundant (LEA) protein from barley. Their study demonstrated that the transgenic rice possessed a better growth rate under 200 mM of salinity and better recovery upon removal of stress. Similarly, genetically engineered rice has also been developed with the capacity to produce glycinebetaine by a gene (*codA*) encodes choline oxidase and it has been found to have better salt (150 mM NaCl) tolerance than the wild type.

Saijo [150], reported better tolerance of transgenic rice plants that over-expresses OsCDPK7 (a calcium-dependent protein kinase) gene to treatment with 200 mM NaCl, as the wild type were found to have their youngest leaf drooped 3 days after treatment with 200 mM NaCl. Several reports have shown a host of other genes related to antioxidants, transcription factors, signaling, ion homeostasis and transporters found to have key role in salinity and drought tolerance [147,151-158]. Heat stress in plants leads to the generation of reactive oxygen species which leads to oxidative stress and this alters various molecular and biochemical activities in plants [159]. In plants generally, heat stress is responsible for the up-regulation of several heat inducible genes, commonly referred as "heat shock genes" (HSGs) which encode Heat shock proteins (HSPs) and these active products are very much necessary for plant's survival under fatal high temperature [160]. Several studies have indicated some successful transgenic cases where over expression of a certain genes is improved by fusion with another gene in order to enhance its thermo-tolerant abilities. Katiyar-Agarwal [161], reported successful improvement of rice heat tolerance level after the incorporation of over-expressed HSP genes (*Hsp101*) of *Arabidopsis* in transgenic rice to enhance thermo-tolerance. Moreover, over-expressing rice chloroplast *sHSP* (*Oshsp26*) gene conferred better tolerance to heat stress and other associated oxidative stress in *E. coli* [162]. Murakami [163] also reported that over-expression of *sHSP17.7* confers heat and ultra-violet tolerance to rice plants. Lee [164] shows that successful alteration in the expression level of heat-shock proteins (HSPs) as a result of changes in the transcription factor (*AtHSF1*) responsible for HSPs in *Arabidopsis* plants was able to produce transgenic high temperature stress tolerant *Arabidopsis*. Malik [165], also reported the increase in thermo-tolerant in transgenic carrot cell lines and other plants by constitutive expression of carrot *Hsp17.7* gene driven *CaMV35S* promoter. Tang [166] demonstrated tolerance in transgenic potato plants through over-expression of both superoxide

dismutase and ascorbate peroxidase in chloroplasts against oxidative stress and high temperature.

A few studies have also been conducted drought susceptible leguminous crop pea through protein profiling for understanding abiotic stress response. 2D-GE, MALDI-TOF/TOF, and LC-ESI-QTOF was employed to dissect the signaling pathway leading to inhibition of biological nitrogen fixation under drought stress [167]. The study successfully identified 18 nodule proteins regulated by both pea and rhizobium genomes under drought stress and unraveling the SNF regulation machinery in nodules. Jaiswal [168] also investigated the dehydration-responsive microsomal proteome with 2D-GE and MALDI-TOF/TOF and they identified 184 proteins that showed significant differential expression.

Other omics studies on plant responses to stress have been able to decipher the regulatory roles and mechanisms used by specific transcription factors in recognizing cognate binding sites on a particular gene in response to environment factors. Qin [169] reported the mechanisms used by C-repeat-binding factor/dehydration responsive element binding protein 1 (CBF/DREB1) regulon and Dehydrating responsive element binding protein 2 (DREB2) to activate and bind on DRE/CRT cis-acting element (core motif: G/ACCGAC) located in the promoters of CBFs/DREB1s genes leading to its expression in response to cold and drought stress stimuli. It was alongside reported that DREB2A have dual functions in modulating the expression of drought, drought and heat, and heat downstream genes under both heat and water deficit stress [170]. A comparative metabolic study of DREB1A- and DREB2A-overexpressing plants has revealed that the metabolic configuration of DREB1A over-expressors resembles that of plants subjected to freezing stress; likewise, DREB2A over-expressors share similarities with drought-exposed plants [171] and their over-expression in transgenic plants have greatly enhanced tolerance to cold, drought and high temperature respectively [170,172].

In plant metabolomics studies, hormone abscisic acid (ABA) regulates the adaptive response of plants to environmental stresses such as drought, salinity, and chilling via diverse physiological and developmental processes [173]. ABA biosynthetic pathway has been deeply studied, and many of the key enzymes involved in ABA synthesis have been used in transgenic plants to improve abiotic stress tolerance [174]. Ji [174] reported transgenic plants over-expressing the genes involved in ABA synthesis showed increased tolerance to drought and salinity stress. Similarly, many studies have illustrated the potential of manipulating *CBF/DREB* genes to confer improved drought tolerance [175]. Other successful transgenic approaches aiming to improve photosynthesis under chilling stress conditions in tomatoes through changes in the lipid biochemistry of the membranes have also being reported [176].

Detecting markers in crop genomes will help identify candidate genes for toughness to climate changes/pathogens which can be introgressed into crop germplasm. Thus, this knowledge will provide plant breeders with selection targets for fast-tracking the development of high performing varieties with more durable pathogen and climatic resistance than the traditional plant breeding.

### Breeding of Pre-harvest sprouting/submergence stress cultivars to increase yield

Pre-harvest sprouting (PHS) causes substantial losses in grain yield and quality, and therefore, it is one of the major factors

negatively affecting the quality of crops in the areas with high levels of precipitation during grain maturation. Cereal crops with low levels of seed dormancy are susceptible to PHS when wet and moist conditions occur prior to harvest [177]. A defined level of seed dormancy is under genetic and environmental control, and it is an essential component of seed quality [178].

In order to alleviate the effects of PHS in crop varieties, genotype together with a number of other factors such as stage of maturity, environmental conditions during grain ripening, crop morphology, biotic and abiotic stress must be studied extensively in resistant plants [179] and a positive result could possibly be used in breeding of PHS resistance cultivars. Since PHS is associated with seed dormancy as stated by Hu [177], it is important to gain deeper knowledge on the molecular mechanisms leading to changes in omics profile involved in dormancy regulation in PHS resistance plants. Proteomics offers the opportunity to examine simultaneous changes and to characterize patterns of protein accumulation occurring during seed dormancy maintenance or release in plants tissues and organs [180].

Recent proteomics and transcriptomics studies have indicated that antioxidant defense mechanisms, redox regulation of seed mRNAs and protein thiols, integrated with hormonal signaling (like Abscisic acid, Cytokinins, Ethylene, dormin and auxins) play a key role in controlling seed dormancy in wheat [181-183], barley [184,185], and rice [157,186]. When seed dormancy is controlled in these crops and possibly other crops, it will significantly contribute to the development of breeding of Pre-harvest sprouting tolerant crops and protect crop spoilage before harvesting.

A major earlier success for crop breeding using genomic markers was the marker-assisted introgression of the Submergence 1A (Sub1A) gene for submergence-tolerance into high-yielding commercial rice varieties [187]. Submergence stress in rice can cause annual losses in excess of US \$1 billion [188]. According to Bailey-Serres [189], increased tolerance to submergence in cultivars with the Sub1A gene, an ethylene response factor, is achieved by limiting shoot elongation during the inundation period. Ismail [188] reported success of alleviation of this stress due to the Sub1A cultivars now grown by over 4 million farmers in Asia.

### Improvement/Protection of crops through intercropping practice in farming

In order to prevent loss of valuable crops due to various stresses that arises from climatic change, weed and infections from parasites, farmers introduce intercropping technique on their farmland. This intercropping technique enables stress responsive plants to combat the stress generated by the stressors and protects incompetent plants from injury caused by the stressors.

In Africa, crop yield losses associated with *Striga* related activities is about 40% [190] and in particular, recent intense droughts, water logging, and extreme temperatures have severely affected maize growth and yield [191]. These has greatly affected adequate food supply as there is rapid increasing global population which is expected to reach 9000 million by 2050 [190]. According to International Crops Research Institute for the Semi-Arid Tropics (ICRISAT) [192], 60% of farm households in Nkayi, Zimbabwe, will be exposed to greater vulnerability by 2050 due to climatic change. Farmers in order to prevent these from causing adverse effects in the future introduce intercropping technique. Data of omics researches on plants survival during and after stress provides insight for farmers in choosing the

right plant to intercrop and combat the disturbing stresses (abiotic and biotic stress). Several studies have reported successful intercrop practice using sorghum, maize and other model plants to cope with the increasing climate change risks and parasitic infections.

According to Ejeta and Butler [193], intercropping Sorghum with trap crops such as Soy beans (*Glycine max* L.), groundnut (*Arachis hypogea* L.), Bambara nut (*Vigna subterranea* (L.) Walp), Sunflower (*Helianthus annus* L.) and cowpea (*Vigna unguiculata* (L.) Walp) may help to reduce the number of *Striga hermonthica* seed in the soil. Carson [194] showed that intercropping of sorghum (*Sorghum bicolor* (L.) Moench) and groundnut significantly reduced *Striga hermonthica* (Del) Benth emergence. This was associated with a decrease in soil temperature in the intercropped plots. In Gambia, Lagoke [195] reported that alternating sorghum or millet (*Pennisetum americanum* (L.) K. Schum) with groundnut resulted highly reduced growth of *S. hermonthica* in soil. Dereje [196] reported a high yield from intercropping sorghum with groundnut in the control of *Striga hermonthica* infection at Assosa Zone, Benshangul Gumuz Region, Western Ethiopia, East Africa. This research indicated that intercropping with groundnut drastically reduced the emergence of the parasite than intercropping with soybean and other legumes.

Recent report from International Crops Research Institute for the Semi-Arid Tropics [192] in Kenya also indicated that farmers who replaced maize with sorghum in intercrop practice with groundnut at 2:4 ratio mitigates the effects of drought and ensure food supply and provides the farmers enough grains till July 2017. Those that intercropped with soybean and other leguminous and cereal crops also had high yield but not as significant as the one of groundnut, whereas some farmers who intercropped maize and groundnut had massive crop failure.

According to Weston [197], Sorghum shoots and roots also produce allelopathic compounds like sorgonole, phenolic compounds, dhurrin and cyanide which can suppress the growth and establishment of competing weed species. Crop rotation systems that integrate sorghum or intercropping or addition of green sorghum manure and extracts from its shoot and root is therefore widely used to control weed infestation. The application of sorghum root and shoot extracts for parasitic weed (*striga*) control has been tested in several crops [198]. Thus this can be used for other crop that *striga* parasite have been an obstacle for quality productivity.

### Prolonged Access to Quality Foods in the Market

Through the use of genomics, proteomics, transcriptomics, and metabolomics, the consistency and predictability in plant breeding have been improved, reducing the time and expense of producing better quality food crops that are resistant to stress but still exhibit their nutritional value. Omics has provided insights to the molecular mechanisms of stress resistance in crops thereby increasing their chances of survival in the environment and enhance quality productivity. Cultivating hybrid or transgenic crops would make the plants survive and possibly have quality growth and development outside their season [199]. This in turn would increase their abundant availability for consumers' consumptions. This further contributes to the therapy of food related diseases like kwashiorkor, Osteomalacia and so on which has become a burden to underdeveloped and developing countries. For instance, growing transgenic or hybridized maize from sorghum during dry seasons may make maize available for consumption at both dry and rainy seasons.

Salinity has been a major environmental hazard of rice cultivation at young stage in the world today [145]. A number of single nucleotide polymorphism (SNP) and simple sequence repeat (SSR) markers analysis have been successfully used to discover salt tolerant cultivars of rice [200], and similarly, Xu [149] produced transgenic rice by introducing and over-expressing late embryogenesis abundant (LEA) protein from barley.

The Quantitative trait loci (QTL/genes) for insect resistance detected at the syntenic maize and sorghum's genomic regions by Satish [113] can be used to improve resistance of stress susceptible crops thereby generating abundant hybrids or transgenic crops with intact nutritional qualities. Sui [176] reported successful transgenic approaches aiming to improve photosynthesis under chilling stress conditions in tomatoes through changes in the lipid biochemistry of the membranes. These have not only enhanced crops adaptation to stresses but indirectly increase their availability in the market for better human and animal nutrition.

## Demerits

Though few demerits exist as regards omics application towards understanding and unraveling different plant abiotic and biotic stresses response because omics research in plant science has expanded with improving technologies with time, but there still some reservations.

Proteomics studies on plants have been helpful in the discovery of the function of wide range of proteins expressed as a result of stress generated from the environment or infections from parasites. Despite the wide range of achievements recorded in plant proteome studies in response to stress, there is a disadvantage of proteomics research on plants responses to stress. Abhilash [201] reported the inability of proteomics research to comprehensively identify the isoforms of each protein. No research has included proteins isoforms, though Ndimba [84] reported the presence of alpha-galactosidase with different isoelectric point indicating different isoforms could be expressed but did not identified them by any omics application. Roy [92] also reported the increased expression of cytochrome P450 enzyme in sorghum subjected to cadmium stress and the enzyme possesses diverse isoforms in nature but none of the isoforms was identified in the experiment.

Another demerit is the heterosis generated from plant hybrids. Availability of plants omics profile for stress responsive plants has encouraged breeders to indulge in breeding practice where hybrids will inherit genes from both parents and also develop its own distinct genes, a considerable number of genes have been shown to have an allele-specific type of expression that could certainly result in heterosis [202-204] performed a study to examine the morphology, physiology and leaf proteome in the two inbred lines (drought-resistance and sensitive parental inbred line) and their reciprocal F1 generation under drought condition in order to verify whether hybrids can really be better adapted to drought conditions than their inbred parents. Though the hybrids inherited and developed some distinct characteristics from their parents like larger plant body, increased synthesis of drought resistance proteins and metabolites and changes in physiological characters in order to adapt to the limited water-supply condition but the larger body and stomata opening inherited from both parents developed both positive and negative heterosis in that they enhance photosynthesis and carbon dioxide (CO<sub>2</sub>) accumulation respectively but also associated with increased transpiration rate which is very detrimental to the growth and development of the hybrids under limited water conditions.

The fact that voluminous data are always generated from omics researches and with plants having complex-dynamic-biological-network systems, often times results into generalization, simplification and lots of assumptions. Thus, omics approaches in understanding plants abiotic and biotic stresses must be tied to the appropriate and correct modeling of these data sets.

## Development of a more Robust Tolerant Hybrid Crops

Improvement of crops in agricultural sector has been a major challenge in the last decades for molecular biologists and biotechnologists due to the rising adverse effect of biotic and abiotic factors (most especially drought and low temperature) with increasing population [205]. These circumstances have paved ways for robust researches on ways to combat these challenges through in-depth molecular omics applications that have led to detection of desirable traits on chromosomes loci expressed in response to the climatic change and other biotic factors. Thus hybrid crops which can withstand these changing climatic conditions are rapidly evolving.

With the recent advances in genomic technology and statistical analysis methods, there is growing interest in using association mapping strategies to identify genes underlying quantitative or complex traits of particular agricultural or evolutionary importance. Using genomic sequence and single nuclear polymorphism (SNP) information, association mapping based on linkage disequilibrium analysis of a naturally varying population can resolve complex traits down to the sequence and genome-wide levels [206]. Though, association mapping based on linkage disequilibrium is less effective at estimating the effects of alleles in general and rare alleles specifically, which may nevertheless be valuable for breeding [207]. In-depth detection of economically and agronomically important traits on loci in stress resistance crops is important. Therefore, improvement of these mapping techniques is needed. Joint Linkage Association Mapping (JLAM) or Nested Association Mapping (NAM) has been put forward to enhance association mapping techniques [23]. This technique has been used to detect Quantitative trait loci underlying drought tolerance in maize, sorghum and a number of other crops [208,209]). More improvements are needed in these techniques so as to discover more important tolerant traits that initially have no ascribed functions and their success can be expected to widely inform public and private breeding programs in the future.

Proteomics researches have gone a long way in the post-genomic era. Many proteins as we know have diverse isoforms, for example Cytochrome P450. These important proteins are synthesized in stressed plants and biotechnologists seize these opportunities to develop hybrid crops which will significantly increase their synthesis under stressed conditions. More in-depth analyses on each protein's isoforms would be of great achievement in developing hybrid crops in that these discoveries would create assurance and ascertain quality of choosing the right parents for the hybrids so as to combat stress and equally increase yield.

Few researches are performed on hybrids in green laboratories/ fields in order to assure their efficiencies and few publications discuss current commercial breeding programs using genomics; however Chen [210] in his research reported limitations in the F1 hybrids of two inbred lines of maize despite it has developed its response to drought stress. Cooper [211] also reported proprietary genomic approaches to facilitate breeding of drought-tolerant maize hybrids



in the United State (US). These are wonderful researches as they will create insights on the void to fill in developing a more robust hybrid crop which will be of economical importance. Extensive research should be done in these areas on other crops and test-run in green laboratories or fields in order to ascertain their qualities and shortcomings before distributing their seeds to farmers for planting.

In order to achieve a more economical and quality hybrids, researchers use the method of predicting the genotypic values (GV) of untested hybrid combinations in maize based on parental information. This has been made possible through integration of high-throughput omics technologies with other prediction tools. Recently, Matthias [212] measured endo-phenotypes of parent lines in maize to forecast the genotypic values (GV) of T0, T1 and T2 hybrid progeny by using prediction equations trained with "omics" information (genomic, transcriptomic and metabolic data) from other parent lines and phenotypic information on their hybrid offspring. This extensive study also revealed the physiological epistasis that is transmitted from parent to their hybrid offsprings and also proposed that the use of Whole-genome Prediction (WGP) is efficient in predicting accurate hybrid biomass over traditional pedigree-based best linear unbiased predictor (BLUP). Previously, Feher [213], in order to understand the mechanism of heterosis in hybrids prior to selecting optimum parental lines, used parental metabolite profiling to predict quality hybrid biomass in maize by incorporating the standard concept of additivity/dominance, which he named Combined Relative Level (CRL). This multivariate diallel approach not only predicted maize hybrid biomass but also the metabolite profiles of the hybrids during early stage of seedling development. Many other studies have been carried out in the prediction of hybrid in maize [214-216], more of these researches are needed on other crops.

These omics integration together with statistical and computational software generated higher success rates in the prediction in developing a more robust hybrid. A success in these small-scale experiments implies a valid strategy for large-scale experiments, where biomass of all possible hybrids would be predicted. Therefore, intimate collaborations among plant molecular biologists, physiologists, breeders, farmers and politicians are required. Previous work regarding gene identification and functional characterization has been performed by molecular biologists; photosynthetic damage, stomatal aperture movement and osmotic adjustments in stress responses are topics for specialists in plant physiology; and tolerant resource collection and evaluation are performed by breeders and farmers are enlightened to plant quality/improved seeds. If possible, farmers should be given the seeds at no cost and full implementations should be closely monitored by government. Through a strong dialog and cooperatives, it will be possible to deliver better agricultural products that utilize less input, have lower environmental costs and provide higher levels of social well-being.

There is need for development/upgrade of existing software tools for analyzing omics studies in plants. Advancement in Mapman, Kappaview, PlantMetgem and other software used in integrating, interpreting and visualizing plant omics results will give more molecular results and predict in-vitro changes that had taken place at the molecular level in plant cell. Results are saved in bioinformatic database for breeders' consultations and optimum knowledge of this would pave way for selection of quality crops for development of hybrids.

## Conclusion

As various omics technologies with high technical potentials for studying large sets of biological molecules are rapidly emerging

with advance high-throughput methods, it is thus very essential to understand the molecular systems that underlie various plants functions. Integration of knowledge from omics-based research with various emerging analytical applications together with improved bioinformatics methods will help in gaining biological insights towards identifying significant information that can be used to understand and improve plants qualities.

Though understanding specific plant functions that appear in a particular plant species is very important in discovering useful genes that can improve the plant functions, with no limitations to sorghum, various plants species with various individual characteristics of interest can be exploited using omics to identify and confer the beneficial characteristics of interest from one plant to another, thereby creating various tolerant or resistant hybrid crops necessary for survival and increase of quality of life for both human and animal.

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