Chapter 19
Abiotic and Biotic Stress Research in Plants: A Gizmatic Approach of Modern Omics Technologies

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Abstract Plants are the basic source of food for humans, birds, and animals because of which they occupy the primary position in the food chain. Being the immovable organisms, plants have to withstand adverse environmental and biological conditions as a result of which they are subjected to various types of abiotic and biotic stresses. These stresses affect the growth and reproduction of plants and also cause loss in the yield of crops. Plants have the natural ability to tolerate these stresses by modifying various molecular, cellular, or physiological mechanisms within them and by understanding these mechanisms; a stress-tolerant plant can be developed. Conventional approaches are inadequate to understand these complex mechanisms, so advanced molecular and system biology tools have been used to overcome these problems. One of the noticeable achievements that laid the foundation for development and improvement of stress tolerance in plants is the omics technologies such as genomics, transcriptomics, metabolomics, proteogenomics, etc. which use large scale data of biological origin such as genes, proteins, etc. along with heavy data mining or bioinformatics component. Recent developments in the field of omics technologies hold an immense potential to reshape and improve plants for stress tolerance by modifying their gene regulation or other regulatory networks. The modern omics technologies have been successfully applied for improvement of abiotic or biotic stress-tolerant plants. The present review gives a brief description of the various omics technologies implied for plant stresses (biotic and abiotic) and also discusses the future prospects related to the use of these technologies.

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1   Background

With growing global warming and abnormal climate change, plants are encountered with a number of abiotic and biotic stresses which ultimately affect their metabolism, growth, yield, and development (Pandey et al. 2017). These stresses may be natural or human-induced (Kumar and Verma 2018). The stress conditions result in hormonal imbalance, ion toxicity, susceptibility to disease, and nutrient mobilization which lead to affect the plant growth and development (Kumar and Verma 2018). Drought, salinity, heat, cold, chilling, heavy metal, freezing, nutrient, highlight intensity, and ozone are some of the abiotic stresses which affect growth and development of plants (Cavanagh et al. 2008). In addition to these abiotic stresses, plants also face the threat of various kind of biotic stresses which occur as a result of interaction between plant and other living organisms such as infection by pathogens (including bacteria, fungi, viruses, and nematodes) or attack by herbivore pests (Atkinson and Urwin 2012). Sometimes plants face multiple stresses at the same time where one stress condition enhances or reduces the effect of another stress condition. As a consequence of different abiotic stresses, the strength of occurrences of detrimental effects of biotic stresses on plants is increased and/or decreased (Pandey et al. 2017).

In response to various stresses, plants express different stress-coping mechanisms based on type and strength of the stress, for example, accumulation of proline during drought and salinity, formation of heat shock proteins, performance of photosynthesis, antioxidants, responses to pathogen, hormone signaling, osmolyte synthesis, etc. in order to detect changes of environmental conditions, prevent damage, and ensure their survival (Atkinson and Urwin 2012; Prasch and Sonnewald 2013). The response of plants to individual stress is found to be different from multiple stresses (Atkinson and Urwin 2012). The mechanism underlying the tolerance of plants to different kind of stresses is still unknown, and further studies are required in this context. "Omics" tools, including genomics, transcriptomics, proteomics, metabolomics, and phenomics, with the association of bioinformatics and computational approaches, enlightened us to understand the mechanism underlying the tolerance of plants and to produce stress-tolerant plants. Understanding the importance, this chapter provides an update on the recent findings related to various omics tools used in different kind of biotic and abiotic stresses and also lays down future prospect for various researches for improvement of stress tolerance in plants.

2   Stress and Plant Response

Mostly plants are exposed to multiple stresses at the same time which are unfavorable for their growth and development, for instance, during summer season plants face concurrently heat stress, drought stress, and high-light stress. These multiple stresses interact with one another so that the effect of one stress depends on another. Plants may consider the multiple stresses as entirely new stress and produce a tailored response to the new stress; however, sometimes the individual stresses would usually elicit opposing reactions. For instance, *Arabidopsis* confers resistance to osmotic, salt, and heat stress on expressing pigeon pea proline-rich protein; however, plants affected by these stresses are of stunted size (Priyanka et al. 2010).

The simultaneous occurrence of biotic and abiotic stresses is crucial where the effects of an abiotic stress weaken the plant defenses, and thus plants become more susceptible to pathogen. Sharma et al. (2007) observed that on exposing the high temperature stress in wheat, the plant becomes more prone to bacterial, viral, fungal, and nematode pathogens. However, sometimes it was also observed that abiotic stress interacts positively with pathogen stress. For example, on exposure to drought stress, plants may close stomata to reduce transpiration which has positive effect on resistance to pathogen (Beattie 2011). Similarly, viral infection can provide protection from drought stress (Xu et al. 2008). It is due to the fact that pathogenicity actively interferes with plant-water relations (Beattie 2011). The combination of stresses, i.e., abiotic-biotic or abiotic-abiotic, also neutralizes the effect of each other which leads to overall neutral or positive impact on plants. For example, in response to individual drought and ozone stresses in *Medicago truncatula* (alfalfa), the growth becomes affected; however, when both the stresses act in combination, the plant becomes tolerant to these stresses (Pandey et al. 2017). Hence, the mechanisms underlying plant responses to multiple simultaneous stresses are crucial. However, omics strategy makes us understand the exact mechanism behind such response which also helps researcher to develop broad-spectrum stress-tolerant crops.

The plants mainly respond to stresses by means of various crosstalks among the signal pathways (Takahashi et al. 2004). One of the earliest signals is reactive oxygen species (ROS) and reactive nitrogen species (RNS) which are involved in many biotic and abiotic stress responses and thus modify enzyme activity and gene regulation (Molassiotis and Fotopoulos 2011). Hormone such as abscisic acid (ABA) is also considered as important regulators of plant responses to abiotic stress, while defense against different biotic stresses is specified by antagonism between the salicylic acid (SA) and jasmonic acid (JA)/ethylene signaling pathways. However, ABA acts both synergistically and antagonistically to biotic stresses and thus generates a complex network of interacting pathways with crosstalk at different levels (Yasuda et al. 2008). Transcription factors (TFs) are also of prime importance for conferring multiple stress tolerance response as they control a wide range of downstream events (Xu et al. 2011a).

3 Omics Technologies to Understand the Stress Tolerance and Plant Response

The Sanskrit word "om" depicts fullness, and the term omics presumably has gotten from the Sanskrit "om." Along these lines, omics infers totality. As of now, the post-fix omics is employed to portray different advanced fields of science, for example, genomics, proteomics, metabolomics, and so forth, to depict studies involving the sum total of genes, proteins, and metabolites, respectively, within an organism or a cell. With the appearance of new advances and gained learning, the quantity of fields in omics and their applications in differing fields is quickly expanding in the post-genomics period. Modern researches use both molecular and systems biology for study of plant stress or the improvement of plant stress responses (Table 19.1). They provide new insights and open new horizons for understanding stresses and stress responses as well as their improvement and resistance to stresses (Duque et al. 2013). The applications of few of the omics approaches in studying plant stresses are discuss below in detail (Fig. 19.1).

3.1 Genomics

Genomics deals with the identification of gene sequences, intragenic sequences, gene structures, and annotations (Duque et al. 2013). The study of genes involves various processes such as DNA extraction, amplification using PCR techniques, DNA sequencing, as well as sequence assembly (Ramegowda et al. 2014). Functional genomics approaches help in identifying the functional genes involved in abiotic and biotic stress responses in plants and also unravel the interactions between genes involved in regulatory networks (Ramegowda et al. 2014). Functional genomics approaches involve sequencing- or hybridization-based methodologies. The sequencing-based approaches use expressed sequence tags (ESTs) sequencing and serial analysis of gene expression (SAGE) where ESTs have been used to explore the discovery of expressed gene and genome annotation and SAGE can measure the expression of gene. Array-based techniques are one of the widely used hybridization-based methodologies where target DNA is allowed to hybridize with cDNA or oligonucleotide probes attached to a surface to assess expression. This technique is widely used to disclose stress responses in different crops and industrially and agriculturally important plant species (Akpinar et al. 2013). Researchers have identified different stress-responsive genes in different variety of plants with functional genomics approaches (Arpat et al. 2004; Micheletto et al. 2007). Jeong et al. (2010) identified rice NAC-domain gene, *OsNAC10*, with functional genomics approach. They observed that when this gene is under the control of constitutive promoter *GOS2*, *RCc3* showed enhanced tolerance to drought condition.

In addition to functional genomics approaches, structural genomics with next-generation sequencing (NGS) platforms such as Roche 454 GS FLX Titanium or

Table 19.1 Different omics tools and technologies used

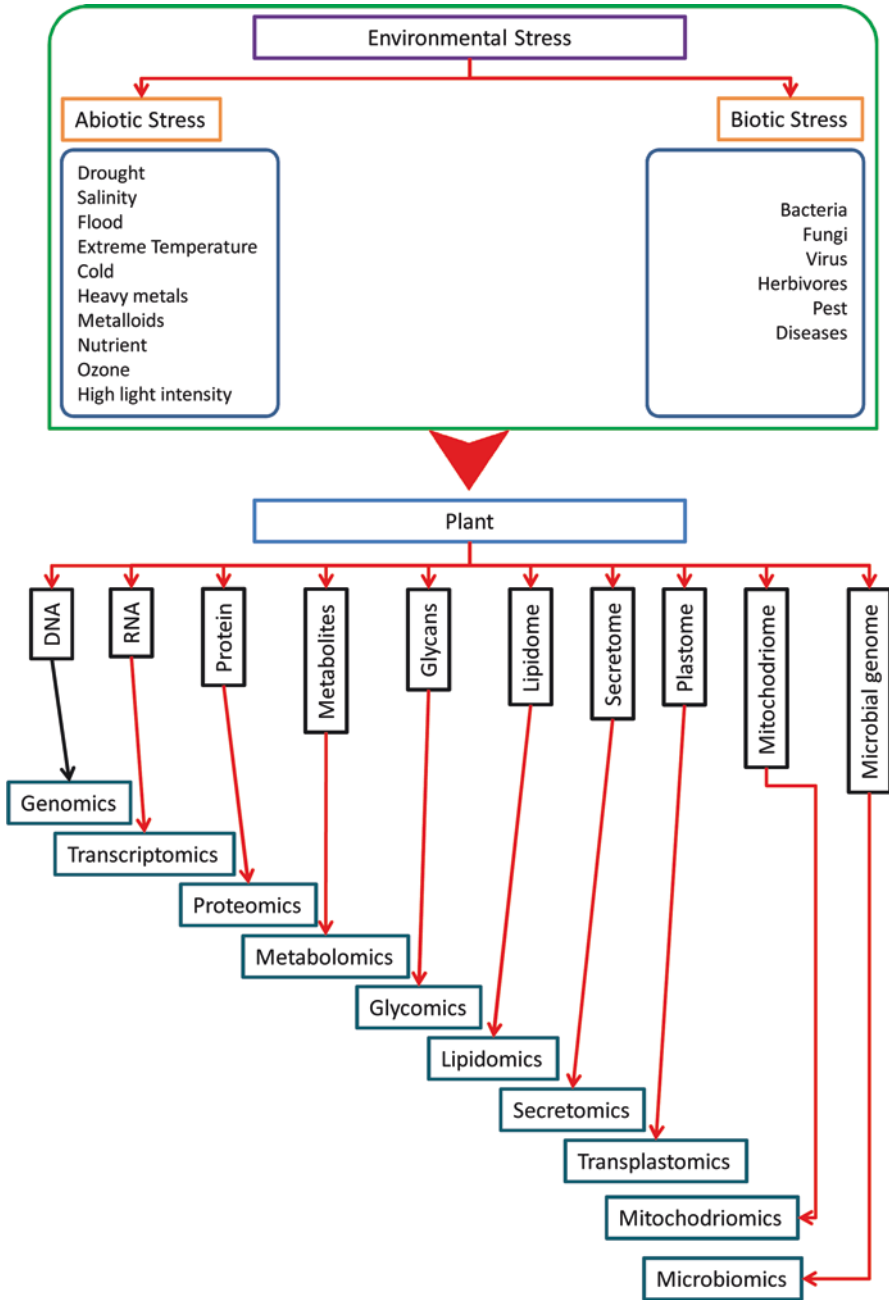| Omics tools	Technologies used
Genomics	Next-generation sequencing (NGS) – Roche Platforms (GS Jr, GS FLX+), Ilumina (MiniSeq, MiSeq, HiSeq, and NovaSeq), SOLID and Ion Torrent, PacBio (RSII and Sequel), OxNano (MiniON and GridION/Prome-thION); bead-based flow cytometric method; expressed sequence tag (EST) analysis
Proteomics	Protein extraction and purification; 1-D and 2-D PAGE; reversed-phase high-performance liquid chromatography (RPLC); MALDI-TOF-MS/MS; ESI-TOF-MS/MS; X-ray crystallography; nuclear magnetic resonance (NMR) spectroscopy; MS/MS-based isotope-coded affinity tags (ICAT) and isobaric tag for relative and absolute quantitation (iTRAQ); multidimensional protein identification technology (MudPIT)
Transcriptomics	RNA-Seq; expressed sequence tags (ESTs); cDNA-AFLP; RFLP-coupled domain-directed differential Display; microarray; next-generation sequencing (NGS) – Roche Platforms (GS Jr, GS FLX+), Ilumina (MiniSeq, MiSeq, HiSeq, and NovaSeq), SOLID and Ion Torrent, PacBio (RSII and Sequel), OxNano (MiniON and GridION/Prome-thION)
Metabolomics	Thin-layer chromatography (TLC); high-performance thin-layer chromatography (HPTLC); liquid chromatography-mass spectrometry (LC-MS); high-performance liquid chromatography (HPLC); capillary electrophoresis-mass spectrometry (CE-MS); gas chromatography-mass spectrometry (GC-MS); nuclear magnetic resonance (NMR); Fourier-transform infrared (FT-IR) spectroscopy
Plant glycomics	Chromatographic techniques – LC, HPLC; tandem mass spectrometry; MALDI-mass spectrometry – MALDI-TOF-MS/MS; ESI-mass spectrometry – LC-ESI-MS/MS; NMR spectroscopy – gCOSY, TOCOSY; Fourier-transform ion cyclotron mass spectrometry (FT-ICR-MS); Microarray – carbohydrate microarray, Neoglycolipid (NGL)-based oligosaccharide microarray, lectin microarray, glycogene microarray
Plant lipidomics	Liquid chromatography-mass spectrometry (LC-MS); ion trap mass spectrometer; triple quadrupole; MALDI-TOF MS; Fourier-transform mass spectrometer (FT-MS); direct-infusion ESI-based MS
Phenomics	Technologies used in genomics, transcriptomics, metabolomics
Metatranscriptomics	Technologies used in genomics and transcriptomics
Cytogenomics and mutagenomics	Technologies used in genomics; TILLING (Targeting Induced Local Lesions IN Genomes)
Plant miRNomics	Technologies used in genomics and transcriptomics
Plant secretomics	Technologies used in genomics, proteomics, and metabolomics
Signalomics	Technologies used in genomics, proteomics, and metabolomics
Thiolomics	Technologies used in genomics, proteomics, and metabolomics
Transplastomics and chloroplastomics	Technologies used in genomics, proteomics, and transcriptomics
Plant mitochodriomics	Technologies used in genomics, proteomics, and metabolomics
Micromorphomics	Technologies used in genomics, proteomics, and metabolomics
Microbiomics in plants	Technologies used in genomics
Cryobionomics	Technologies used in genomics, proteomics, and metabolomics

Fig. 19.1 Schematic diagram representing multipurpose application of omics approaches

Illumina Solexa Genome Analyzer and comparative genomics can also be used to get detailed information of genomic features of plants. The NGS-mediated shotgun sequences have been used to develop molecular markers to detect polymorphisms at the DNA sequence level which help plant breeders to create new breeds that can tolerate several biotic and abiotic stresses (Akpinar et al. 2013). Genomics in combination with other omics technologies such as transcriptomics and proteomics help plant breeders to create new stress-tolerant breeds (Agarwal et al. 2014) and to understand the regulation and expression of those genes (Yanik et al. 2013; Zadraznik et al. 2013).

3.2   Transcriptomics

Transcriptomics is the study of the complete set of RNA transcripts that are produced by the genome, under specific set of conditions by a cell using high-throughput methods. Comparison of transcriptomes permits the identification of genes that are differentially expressed in particular plant, or in response to different stresses. The transcriptomics technology has provided a valuable insight to diverse plant stress response and tolerance. It provides better understanding of abiotic stress candidate genes and plant-pathogen relationship. Various technologies such as hybridization-based approaches, sequence-based approaches, and RNA sequences are used for transcriptomic studies (Tan et al. 2009). Transcriptomics analysis of broccoli plants treated with N6-benzylaminopurine showed that genes encode for molecular chaperones and stress is upregulated (Liu et al. 2013). Transcriptomics analysis of oak plants showed upregulation of genes associated with plant defense when subjected to long-term mild drought (Spieb et al. 2012). On exposure to salt stress, transcriptomics analysis of plants assisted in the identification of important transcripts and related associations between various physiological processes (Maathius 2006). Exposure to both cold and salinity stresses resulted in downregulation of most of the photosynthetic genes and upregulation of genes encoding transcription factor (Evers et al. 2012). Vashisth et al. (2018) identified stress-responsive genes by transcriptome sequencing of *Artemisia annua* which help to tolerate different abiotic stresses including salt, cold, drought, and waterlogging.

3.3   Proteomics

Protein is regarded as the building block of all living organism. The term proteome implies to the overall proteins in a cell, tissue, or life form. Proteomics is the investigation of the whole protein complement of a cell, tissue, or living being underneath a particular, designate set of conditions. Proteomics encompass the identity, biochemical properties, and functional roles, and how quantities, modifications, and structures change during development and in response to internal and external stim-

uli these proteins. Various types of proteins respond to various stresses at pre- and post-transcriptional and translational levels. These stress inducible proteins could be comprehensively useful for developing stress tolerance in plant. Thus the proteomics study offers an avenue for the discovery of new proteins and pathways associated with crop physiological and stress responses. Few proteomic studies have been conducted in plants under various abiotic stresses including salt stress (Nam et al. 2012), drought stress (Castillejo et al. 2008), and heat stress (Rollins et al. 2013). Plant biologists also used proteomics approach to study the response of *Arabidopsis thaliana*, *Nicotiana tobaccum*, and *Agrostis stolonifera* in response to saline stress (Jiang et al. 2007; Razavizadeh et al. 2009; Xu et al. 2011b). Besides this, some economically important agricultural crop plants have also been investigated under saline stress with proteomics approach, e.g., canola (Bandehagh et al. 2011), sugar beet (Wakeel et al. 2011), potato (Aghaei et al. 2008), and cucumber (Du et al. 2010). Wang et al. (2014) reported 53 differentially expressed protein in *Kandelia candel* where expressions of some of the proteins are upregulated on subjecting to salt stress. Zhao et al. (2008) detected proteins involved in signal transduction while studying the guard cell proteome under drought stress in *Arabidopsis thaliana*. Passamani et al. (2017) used proteomics approach to study the response of salt stress in micro-propagated shoots of two sugarcane cultivars. It was observed that proteins which are involved in calcium-dependent protein kinase, photosystem I, phospholipase D, and glyceraldehyde-3-phosphate dehydrogenase were abundant in salt-tolerant ones. Proteomics also provide information of interactions between crop-pathogen, bacterial pathogens, and elicitors (Casasoli et al. 2008; Margaria and Palmano 2011).

3.4   Metabolomics

Metabolomics is a technology to determine and quantify metabolites involved in different life processes (Deshmukh et al. 2014). Since plants can synthesize a wide range of metabolites to adapt different stress condition, identification and quantification of these metabolites provide better understanding of stress biology in plants (Badjakov et al. 2012). The metabolic fingerprinting can be performed by different techniques such as nuclear magnetic resonance (NMR), MS, Fourier-transform ion cyclotron resonance mass spectrometry, or Fourier-transform infrared (FT-IR) spectroscopy. The identification and quantification of the metabolites can be done by NMR, GC-MS, liquid chromatography-mass spectrometry (LC-MS), capillary electrophoresis-mass spectrometry (CE-MS), and FT-IR spectroscopy (Bagati et al. 2018).

In response to salinity stress, researchers utilized metabolomic approach to know the metabolic effect of salinity in a variety of crop and related plant species including sea lavender (*Limonium latifolium*), rice (Hirai et al. 2004), tomato (Lenz et al. 2011), and grapevine (Cramer et al. 2011). Kaplan et al. (2004) found a set of metabolites in *Arabidopsis* in response to heat and cold stress. Morsy et al. (2007)

observed the accumulation of galactose and raffinose in chilling-tolerant genotype and declination of level of sugar in the chilling-sensitive genotype of rice. In response to drought stress, metabolic regulation in photosynthesis and accumulation of osmolytes occur. *Arabidopsis* can respond to drought stress on accumulating metabolites including amino acids such as proline, raffinose family oligosaccharides, γ-amino butyrate (GABA), and metabolites of tricarboxylic acid (TCA) cycle (Urano et al. 2009). The GC-MS profiling with microarray analysis was conducted by Gong et al. (2005) to study and compare the metabolic profile due to stress competence in the extremophile *Thellungiella halophila* with *Arabidopsis* which showed variation of metabolites between *Arabidopsis* and *Thellungiella* under salt and osmotic stresses. Nikiforova et al. (2005) studied the metabolite profiling of *Arabidopsis* due to sulfur stress. Similarly, Hernandez et al. (2007) identified a set of metabolites such as amino acid, polyols, and sugar which were increased due to phosphorus stress. Various studies on heavy metal-induced stress in plants were also done using NMR-based metabolites fingerprinting on *Silene cucubalus* cell cultures by Bailey et al. (2003).

3.5 Phenomics

Phenomics is the acquisition of multidimensional phenotypic data in an organism (Houle et al. 2010). It extends our knowledge of stress tolerance mechanisms in crop plants. The linking of phenomics to different omics-technologies such as genomics, transcriptomics, and metabolomics helps to analyze the performance of plants in different stress conditions and to fill the gap between response of gene and complex targeted traits for crop improvement (Singh et al. 2018). Plants are sensitive to high heat/cold temperature, and plants can tolerate these stresses on changing physiological and biochemical traits which can be studied by phenotyping (Singh et al. 2018). Phenomics can also be used to find salinity stress behavior in plants which occurs in two phases, i.e., osmotic phase and tissue-tolerant phase. It was observed that osmotic-sensitive plants are more prone to reduction of growth rate in comparison to osmotic-tolerant plants on exposure to NaCl. This reduction of growth rate can be calculated by daily imaging the plant or by infrared thermography (Sirault et al. 2009). James and Sirault (2012) also used infrared thermography to screen a large number of wheat genotypes in response to salinity tolerance. In tissue-tolerant phase, in response to NaCl stress, sensitive plants are not able to compartmentalize Na^+ away from cytosol resulting in early senescence in leaves which leads to alter the leaf color. These color changes are visible by light imaging techniques with a Scanalyzer 3D Discovery Platform (Rajendran et al. 2009). Similarly phenomics study with different phenotypic systems such as visible, infrared, and hyperspectral imaging is also able to identify plant's response to drought conditions (Iyer-Pascuzzi et al. 2010).

Plants are more prone to various pathogens resulting in loss of yield. Pathogens result in increasing chlorophyll fluorescence at early stage of infection. These

symptoms are generally monitored visually which is time-consuming; however, the high-throughput phenotyping detects the symptoms on monitoring chlorophyll fluorescence more easily even before its detection by human eyes (Yang et al. 2012). Researchers have also developed different high-throughput phenotyping systems such as hyperspectral imaging system, ground-based real-time remote-sensing system, remote pheromone trap monitoring system, etc. to detect plant pathogenic diseases at an early stage (Moshou et al. 2005; Fukatsu et al. 2012; Yang et al. 2012).

3.6   Meta-transcriptomics

Metatranscriptomics deals with the expression of the mRNA within microbial community to identify genes or genetic pathways (Jiang et al. 2016). With this approach, it is possible to identify induced systemic resistance offered by microbial strains in plants in response to various pathogens. For example, *Pseudomonas fluorescens* and *Bacillus* sp. induce resistance to tomato leaf curl virus and tobacco mosaic virus, respectively (Wang et al. 2009; Sangeetha et al. 2010). It also provides information of bacterial gene expression profiles in control/wild and treated plant species/mutant plant species with various agents to adapt defense response (Jones and Dangl 2006). Metatranscriptomics study also provided information of stress tolerance in *Salix* sp. grown in polluted land where Rubisco activase transcripts were downregulated and thiamine thiazole synthase and CP12 were uniformly upregulated (Brereton et al. 2016). The mRNA-based metatranscriptomics can reveal changes of rhizosphere microbial flora in different stress conditions. Hayden et al. (2018) used metatranscriptomics of wheat rhizospheric microflora in disease suppressive and non-suppressive soil to *Rhizoctonia solani* AG8. The results showed that in suppressive soil the polyketide cyclase, terpenoid biosynthesis backbone (dxs) and cold shock proteins (csp) genes get expressed. Whereas in non-suppressive soil, antibiotic genes, genes involved in detoxifying reactive oxygen species (ROS) and superoxide radicals genes get expressed.

3.7   Cytogenomics and Mutagenomics

The plant cytogenomics and mutagenomics are important breakthrough in history of genetics and plant breeding. With the aid of advanced technologies like DNA-based-specific fluorescence banding, GISH, and FISH-guided chromosome painting to the classical plant cytogenetics, the identification, localization, and mapping of chromosome-specific markers in plants are greatly facilitated, which are of high importance in plant breeding, molecular systematics, species identification, detection of hybrid nature, detection of alien chromosomes, chromosomal aberrations, and analysis of somaclonal variations and diversity (Chaudhary et al. 2011). On the

other hand, mutational approach offers a powerful tool to study the genetic and molecular mechanism protecting plants against diverse types of biotic and abiotic stresses. The induction of mutation in plants results in alteration of the susceptible alleles of resistant genes and often leads to improvement of yield, morphological traits and increased tolerance to biotic and abiotic stresses (Kharkwal and Shu 2009). For example the induction of mutation by γ-rays, X-rays, NaN_3, EMS, NMU, N-ethyl nitroso urea, colchicine, transposon, etc. in cereals (rice, wheat, maize, barley)/millets, grains, and vegetables results in the plants becoming more tolerant to both biotic and abiotic stresses (Sinjushin and Talukdar 2015). This induced mutation can be easily identified by genomic-based technologies on comparing with wild and normal type of plants (Sinjushin and Talukdar 2015).

3.8   Plant miRNomics

In plants, microRNAs or miRNAs are short (22 bp approx.), endogenously expressed and non-translated RNAs which are regulating the expression of messenger RNAs by targeting transcripts for cleavage or translational repression and sometimes considered as negative gene regulatory molecules. They are considered as conserved regions and play an important role in plant growth and development. Additionally miRNAs also occupy an imperative part in plant stress responses (Kruszka et al. 2012). The expression profiles of most of the miRNAs changed in response to stress, and this stress-responsive miRNAs control the attenuated plant growth and development under stress condition (Mendoza-Soto et al. 2012). Different types of miRNA are involved in various type of abiotic and biotic stress response. Among abiotic stress response, miR169 in *Arabidopsis* were modified to adapt to drought and cold (Li et al. 2008; Zhou et al. 2008). Similarly, in response to salt stress, the expressions of different salt-responsive miRNAs are altered, and some of the miRNAs respond to salt stress by regulating transcription factors (Ding et al. 2009). In response to biotic stress such as plant disease development due to viral infection, miRNAs are involved in silencing of virus-induced gene. Though the exact mechanism behind it is not clear, it is presumed that plants may utilize general RNA silencing machinery to destroy viral RNAs (Hohn and Vazquez 2011). Plants also resist bacterial infection by miRNA-guided regulations of multiple plant hormone pathways. In response to infection of *Pseudomonas syringae*, miR393a gets expressed in plants, and auxin receptors AFB1 are downregulated which result in repression of auxin perception and signaling and thus protecting plants against pathogenic bacteria (Navarro et al. 2006). There are some miRNAs which can respond to multiple biotic and abiotic stresses, for example, miR156 can respond to drought, salt, cold, heat, ABA, oxidative, hypoxia, and UV B (Kruszka et al. 2012; Sunkar et al. 2012).

3.9 Plant Glycomics

Glycomics describes studies designed to define the complete repository of sugars (glycans) and the interaction of these with other macromolecules that a cell or an organism produces under specified conditions of time, location, and environment. Since the sugars play key role in many biological processes such as signaling, stress responses, and immunity, glycomics study is beneficial to study stress responses of plants. Plant glycomic research has significant applications in biopharming of proteins and enzymes (Yadav et al. 2015). The genomics and proteomics don't provide information of biosynthesis of glycan; hence the study of glycomics is lagging behind; however advancement in technology aided the progress of glycomics (Yadav et al. 2015). The covalent linkage of glycan to protein and lipid is considered as the most structurally diverse modification which provides both structural and functional diversity. Attaching glycan to asparagine (N) residues of proteins in plant cell causes variations in the endoplasmic reticulum and the Golgi apparatus, and the *N*-glycan modifications in the Golgi apparatus result in complex *N*-glycans. This complex *N*-glycans protect the growth of root in response to salt/osmotic stress. They also regulate biosynthesis of cell wall and proliferation of cell (von Schaewen et al. 2008). Plant lectins with fungicidal and insecticidal activities are used to form transgenic plants which have the capacity to enhance resistance of plant to pests and phytopathogens (Melnykova et al. 2013). Introducing lectin-like kinases genes in plants genome results in the enhancement of plants defense against various environmental stresses (Melnykova et al. 2013).

3.10 Plant Lipidomics

Lipidomics has transpired as a unique field that allows the comprehensive analysis of cellular lipidome including chemical structures and the quantitative composition with respect to cell signaling, membrane architecture, transcriptional and translational modulation, and cell-cell and cell-protein interactions. In plants, lipids and lipid-based derivatives are essential cellular constituents that play a key role in storage of carbon energy, cell compartmentalization, protection against pathogens, and developmental processes.

Plants can alter membrane lipidome and membrane lipid remodeling to neutralize abiotic stresses. Different researchers utilized lipidomics approach to know the response of plants in connection with different biotic and abiotic stresses. Chen et al. (2013) observed that plant model such as *Arabidopsis* can alter membrane lipid composition in response to cold stress for adaptation and survival in that condition. Similarly lipidomics study of drought-tolerant and drought-sensitive thyme plants (*Thymus serpyllum* L. and *Thymus vulgaris* L.) showed that sensitive plants can decrease the level of galactolipids and phospholipids, while tolerant plants

showed increasing level of lipids involved in signaling (Moradi et al. 2017). Similarly, in response to laminar shear stress in *Taxus cuspidata*, phosphatidic acid (PA) content was found to be increased with enhanced activation of phospholipase D (PLD) and phospholipase C (PLC) in comparison to control cells which demonstrated that phospholipids and related phospholipases have imperative roles in mechanotransduction of *T. cuspidata* cells in response to shear stress (Han and Yuan 2009). Liu et al. (2017) performed lipidomics analysis to unravel the effect of nitrogen fertilization on lipid metabolism in *Camellia sinensis* L. and concluded that application of nitrogen fertilizer can balance the lipid metabolism and leads to formation of improved quality of tea with flavor/aroma origin compounds. Lipids also act as signaling molecules that can trigger genes against stress response, for example, ERECTA; proton antiporters TNHX1 and a proton pyrophosphatase TVP1 can protect *Arabidopsis* against salinity and drought stress (Han and Yuan 2009).

There are mainly two different techniques used in the identification and quantification of lipids, i.e., mass spectrometry (MS)-based techniques and non-MS-based techniques. Mass spectrometry (MS)-based techniques involve the use of ion trap mass spectrometer, triple quadrupole, MALDI-TOF MS, Fourier-transform mass spectrometer (FTMS), and direct-infusion ESI-based MS. Non-MS-based techniques involve nuclear magnetic resonance and LC-MRM platforms.

3.11 Plant Secretomics

Plant secretomics is an emerging field of proteomics which deals with secreted protein of plants that have significant importance in the formation of cell wall structure, cell-to-cell interaction, appropriate response to environmental stimuli, and defense against pathogens (Kamoun 2009). The secreted proteins in the apoplast or the extracellular space mediate major defense responses (Grant and Lamb 2006). Thus, plant secretome might play a key role in the early recognition and defense against pathogen attack and improves our insight of defense mechanism during plant-pathogen interactions. Researchers have identified different secreted proteins in response to fungal pathogens (Okushima et al. 2000; Chivasa et al. 2005; Oh et al. 2005). Plants can also produce different elicitor-responsive proteins in response to various pathogens. Thus whole protein extracts from plants augment our understanding of plant-pathogen interactions and defense signaling (Lee et al. 2006; Rampitsch et al. 2006). On attack of pathogens, plants produce salicylic acid which stimulates the expression or secretion of pathogenesis-related proteins which offer the plants to acquire resistance. In plant secretome analyses of *Capsicum annuum* infected with *Phytophthora capsici*, 75 secretory proteins were identified, most of which were stress-related proteins (Yeom et al. 2011). These stress-mediated pathogenesis-related proteins in apoplast are also effective in restraining the growth of pathogens in plants (Pechanova et al. 2010).

3.12   Signalomics

Plant growth regulators (PGRs) play important roles in the growth and development. They also act as signaling molecules in various metabolic processes in plants. For example, roots sense the onset of water stress and can communicate the need to close leaf stomata by altering abscisic acid (ABA) levels in the shoot. Day length and temperature regulate synthesis and transport of gibberellins, which promote stem elongation and stolon formation. Epinasty is characterized by downward curvature of leaves on the adaxial surface of the petiole which occurs in response to stress. This movement reduces foliar absorption of light, retards transpirational water loss, and reduces drought-induced wilting (Abeles et al. 1992). Recent studies in tomato and other crops suggest the possibility of alternative signals involved in root-based communication of drought stress. Holbrook et al. (2002) used two tomato mutants deficient in ABA synthesis to investigate the role of root-derived ABA. Additional experiments using split roots (partial drying) and grafting coupled with maintenance of turgor pressure (pressure chamber) indicated the presence of a root signal that was independent of the root genotype. Similar results were obtained by Fambrini et al. (1995), who used sunflower mutants deficient in ABA and partial root drying of tomato (Sobeih et al. 2004). These studies suggest the presence of an alternative root-based chemical signal capable of altering leaf ABA concentration and availability, facilitating stomatal responses.

3.13   Thiolomics

Thiol (sulfur) is an important constituent, for synthesizing of protein for growing plants is determined with the subsequent uptake and distribution of sulfate in response to demand and environmental factors. Cysteine (cys) is the first molecule in plant metabolism that contains both sulfur and nitrogen. The biosynthesis of cysteine is of utmost importance for the synthesis of a number of essential metabolites in plant. Ruiz and Blumwald (2002) reported that the biosynthesis of cys and glutathione (GSH) was greatly increased in *Brassica napus* when exposed to saline conditions. In gray poplar hybrid, the GSH contents increased in response to NaCl stress in leaves but not in roots (Herschbach et al. 2010). The roles of GSH in modulating plant growth and development have been explored in grass pea and in lentil genotypes under water stress (Talukdar 2013). The pivotal roles played by thiol-cascade in conferring tolerance to heavy metal stress in plants were demonstrated by Harada et al. (2001) who engineered tobacco plants expressing a rice CS gene in order to make it tolerant to toxic levels of cadmium. Similarly, a combined study of transcript, enzymatic, and metabolic profiling in the moss, *Physcomitrella patens*, revealed vital involvement of sulfate assimilatory genes under Cd stress (Rother et al. 2006). The central role played by the GSH and phytochelatins (PCs) in the

detoxification of the metalloid indicates a critical importance for S-metabolism in determining plant survival in As-contaminated soils (Ahsan et al. 2008).

3.14 Transplastomics and Chloroplastomics

Plastids are semi-autonomous, endosymbiotic organelles of prokaryotic origin. They contain circular double-stranded DNA and have retained their own nucleic acid and protein synthesis machinery (Wani et al. 2015). The plastid genome, termed as plastome, is present in several identical copies in each plastid. Transplastomics is the omics science related to study of plastid transformation. Plastid transformation represents an alternative to existing nuclear transgenic technologies for improving crop quality and productivity under adverse growth conditions. Transplastomic plants which are resistant to insects due to the presence of crystal proteins of *Bacillus thuringiensis* (Bt) are considered as safe biological insecticides which have immense role in agriculture (Romeis et al. 2006). Since 1994, several transgenic crops expressing Bt crystal proteins, e.g., Cry1Ab in maize and Cry1Ac in cotton, have been commercialized and grown worldwide on millions of hectares and have significantly decreased the use of insecticides (Kota et al. 1999). A novel and non-Bt-type insect resistance strategy has recently been demonstrated by expressing long double-stranded RNA (dsRNA) targeting in order to activate RNA interference that disrupts expression of the target gene in the insects (Zhang et al. 2015). Plastid transformation is also regarded as a potential tool to increase disease resistance to phytopathogenic bacteria and fungi due to high concentrations of the target protein accumulating in a single compartment and released only during hypersensitive reaction, e.g., msi-99 transgene, which encodes a magainin 2 analog antimicrobial peptide, that was first successfully introduced to the plastid genome and was proven to be efficient against different bacteria and fungi (DeGray et al. 2001). Similarly, the introduction of a single agglutinin gene of the *Pinellia ternata* herb resulted in high levels of protein expression in leaf chloroplasts and was effective against broad-spectrum resistance against various pests including aphids, flies, lepidopteran insects, and bacterial and viral pathogens (Jin et al. 2012). The expression of the chloroperoxidase-encoding gene of *Pseudomonas pyrrocinia* in transplastomic plants conferred a similar level of fungal resistance in vitro and in plants (Ruhlman et al. 2014). Hence, transplastomic plants conferred a broad spectrum of resistance not only against different pests and diseases but also against abiotic (salt, osmotic, and oxidative) stresses.

The chloroplast is the most remarkable organelle of plant cells which is the site of a myriad of different biochemical reactions. The knowledge of chloroplast transcriptomes largely leads to the emergence of a very interesting field, directly related to the improvement of crops by photosynthesis/Rubisco engineering. Besides lowering plant nitrogen demand by modulating Rubisco levels, efforts have been made for the improvement of some of its enzymatic traits such as CO_2 affinity, CO_2/O_2

specificity, etc. in tobacco (Zhu et al. 2010). Chloroplast transcriptome of *A. thaliana* has been explored during seed development, stratification, germination, and early seedling development (Demarsy et al. 2012). The thylakoid proteome of sugar beet was explored under iron deficiency conditions, while carbon metabolism-related proteins displayed greater abundance under this stress conditions (Andaluz et al. 2006). Furthermore, the proteome of rice leaves under cold stress was reported to contain 60 unique proteins, whose functions are related to protein synthesis and folding, cell wall synthesis, protein degradation, energy production, and signal transduction. These proteomes of leaves are the most affected organelles by cold stress and possibly can be regarded as mediators of plant responses to cold stress (Cui et al. 2005).

3.15   Plant Mitochodriomics

Mitochondria, the powerhouse of the cell, are an important organelle responsible for production of required amount of ATP for other cellular functions. The mitochondrial genome size of animals is about 16.5 kbp in length whereas between 200 and 2000 kbp in plants. With the discovery of mitochondria in 1840, various progresses have been made in understanding the role of mitochondria in the regulation of energy metabolism of the cell. But the advent of mitochondrial omics technology has given an edge to understand the various regulation mechanism of the cell. In many plants, proline (amino acid) accumulates in response to water stress which later degenerates (Kiyosue et al. 1996). Thus the mitochondria play an important role in recovery of water stress by degrading accumulated proline (Mani et al. 2002).

Plants which are subjected to abiotic stress face disruption of cellular homeostasis. Fratianni et al. (2001) when isolated mitochondria from potato cells acclimated to water stress were found to have an increased capacity of several transporters like dicarboxylate, nucleotide, and K channel which could possibly be related to oxidative stress and energy management. In plants, mitochondrial potassium channels exhibiting high conductivity were discovered and have been suggested to play a major role in energy dissipation and prevention of oxidative stress (Ruy et al. 2004). Durum wheat is a drought-tolerant cereal for which mitochondrial biology in relation to water stress has been extensively studied (Pastore et al. 2007). In addition, Pastore et al. (2007) suggested mitochondrial acclimation to water deficit by applying osmotic or salt stress to seedlings which resulted in the activation of UCP and the potassium channel. Pastore et al. (2003) suggested that the Mal/OAA shuttle was the main route for reoxidation of cytosolic NADH accumulating in stress conditions in durum wheat mitochondria. Most higher plant seeds are desiccation-tolerant, and researches have suggested that mitochondria have a unique property to withstand desiccation with the existence of protective mechanisms, e.g., maize and pea mitochondria isolated from imbibing seeds were found to oxidize succinate and external NADH oxidation at high rates, while TCA cycle activities developed later during germination (Logan et al. 2001; Benamar et al. 2003).

3.16   Micromorphomics

Micromorphomics is the micromorphology-based analysis which is applied to all living organisms which have a great role in stress physiology. In response to stress, plants can adapt, avoid, or may overcome these stresses by means of various physiological and biochemical mechanisms. Plant architecture thus plays an important role in stress physiology. The study of micromorphomics includes measurements from all levels of plant organization like molecules, organelles, cells, tissues, organs, whole plants, and populations which are often necessary to analyze in response to stress. In response to drought and salt stress, plants can roll the leaf extensively (Srivastava 2001), and in response to heavy metals content, necrosis along veins and leaf chlorosis occur in plant (Andre et al. 2006). "Stomatal clustering" is also recognized as a new micromorphological marker for environmental adaptation in terrestrial plants (Gan et al. 2010). Azmat et al. (2009) stated that increased stomatal density coupled with decreased stomatal size is considered as a good adaptive feature of plants in response to heavy metal toxicity and polluted environment. The larger number of small stomata ensures sufficient flow of carbon dioxide for photosynthesis, keeping transpiration to a minimum.

Roots are also considered a sensor organ which is influenced by different stress like drought, salinity, heat, etc. Production of ramified root system in response to drought was reported in many crop plants like rice, wheat, soybean, maize, and sunflower (Tahir et al. 2002; Jaleel et al. 2009). Increased root area is also observed in *Arabidopsis thaliana* when the plant is exposed to abiotic stress (Olmos et al. 2006). Elongation of root and decrease in root diameter in response to drought in many wild species were also reported. Small diameter root maximizes absorptive surfaces, thus increasing rates of water and nutrient uptake (Reader et al. 1993).

A flower is a modified stem which ensures maximal reproductive success. The reproductive parts in flowering plants are often highly sensitive to environmental stress than the vegetative phase. For example, in saffron fresh weight of flower stigma (stigma yield) initially increases with salinity, but in extreme saline condition, stigma yields rapidly decrease (Torbaghan et al. 2011). High temperature also affects the stigma position of flower affecting the pollen capturing capacity (Talukdar 2015). Similarly, exposure to high temperature resulted in male sterility in barley because of non-viable pollen and failure of anther dehiscence (Talukdar 2015).

3.17   Microbiomics in Plants

Plant microbiome shows a close symbiotic relationship between plants and their associated microorganisms. The structure of the plant microbiome is determined by different factors, including type of plant species, plant age, soil properties, nutrient status, and climatic conditions (Berg and Smalla 2009). As an example, the associated microbiome of medicinal plants *Matricaria chamomilla* and the African night-

shade, *Solanum distichum*, showed that despite their being grown in direct proximity to one another, their structural (16S rRNA genes) as well as functional (diazotrophic community, *nifH* gene encoding the nitrogenase reductase subunit) colonization profiles revealed a high degree of plant specificity (Koberl et al. 2011). Plant-associated microbes interaction helps the growth and development of the plant and also facilitates the plant to promote stress resistance. Pandey et al. (2016) have demonstrated that application of *Trichoderma harzianum* induces the rice genotypes to mitigate stress response by upregulating the genes for aquaporin, dehydrin, and malondialdehyde. Similarly, *Pseudomonas* sp. and *Acinetobacter* sp. augment production of IAA and ACC deaminase to grow barley and oats in salt-affected soil (Chang et al. 2014).

3.18 Cryobionomics

Cryopreservation is the long-term storage of biological samples in liquid nitrogen at ultralow temperatures (-196 °C) which ensures storage of nonorthodox seeds, the germplasm of vegetatively propagated species and biotechnologically important plant cell lines, etc. (Martinez-Montero and Harding 2015). However, during the process of cryopreservation, there is a possibility of cryo-injury and cryogenic/noncryogenic stress factors to cells/tissues; hence it is important to ensure of any destabilizing effects before they are reintroduced into natural environments which can be easily confirmed by cryobionomics approach (Martinez-Montero and Harding 2015). Generally cold acclimatization is necessary before the exposure to liquid nitrogen. It was observed that genes respond to adapt to the extreme stresses as imposed by cryopreservation which can be studied by transcriptomic approach (Volk 2010). In cryopreservation, it is necessary to dehydrate the tissue with sugars such as sucrose which also acts as osmotic cryoprotectant. It was observed from sucrose-mediated osmotic stress on the banana meristem results that most of the proteins were significantly up- or downregulated (Carpentier et al. 2007). Hence, in this case it is necessary to maintain the osmoprotective level of intracellular sucrose and augment the expression of some energy-conserving glycolysis genes and the conservation of the cell. Similarly it was observed that proteins from *Vanilla planifolia*-dissected apices responded to PVS3 cryoprotective treatments where some were up- or downregulated proteins (González-Arnao et al. 2011).

3.19 Bioinformatics

All the major omics technologies are highly dependent on this omics science, i.e., bioinformatics and computational tools. Various omics approaches provide high-throughput and large-scale properties which require proper handling, analyses,

visualization, and storage. Hence all omics research is tightly bound with strong bioinformatics and computational tools that perform the various analyses (El-Metwally et al. 2014). Computational tools and informatics analyses along with associated methods and algorithms in biology help to analyze biological data in an accurate, fast, and error-free manner (Orozco et al. 2013). Several bioinformatics tasks include genome sequence assembly, sequence alignment, gene prediction, DNA-protein and protein-protein interaction, interaction and regulatory networks analysis, etc. Special type of visualization is required from the huge amount of data analysis and processing by genome sequencer and mass spectrometers. Few of the examples of data visualization are integrated genome tool, PRIDE inspector and ConPath, 3omics, Peppy, etc. The large amount of data and results obtained from various bioinformatics analysis is required to be available and accessible to the scientific community. Several types of databases are available online for deposition and storing the biological data such as plant genomic information (Yu et al. 2013), plant transcriptomics information (Priya and Jain 2013), and plant proteomics information (Cheng et al. 2014).

3.20   *Multi-omics Approach*

Combination of different omics approaches such as metabolomics, transcriptomics, and proteomics can help to link metabolites with their producer genes, as well as allow the mapping of these metabolites/genes into metabolic pathways (Kumar et al. 2014), and the detailed output of plant metabolic pathways can be harnessed to the plant-specific metabolic pathway database. Similarly, phenomics which is the high-throughput analysis of plant physiology (Furbank and Tester 2011) can also be integrated with all other omics to identify the candidate genes involved in abiotic stresses. Identified genes by all the abovementioned omics technologies can be then functionally characterized using functional genomics approaches like gene over expression or silencing. The integrated approach of transcriptomics and metabolomics can reveal the connections between genes and metabolites (Urano et al. 2009). The combination of metabolomics with other omics techniques facilitates to correlate the metabolite levels and expression level of genes/proteins (Srivastava et al. 2013). Cramer et al. (2007) used integrated approach of metabolomics and transcriptomics to assess response of grapevine to water and salinity stress. Integrated approaches of transcriptomics and proteomics analysis also revealed that nearly five sulfate transporter genes in rice (Norton et al. 2008; Chakrabarty et al. 2009), three in *Brassica* (Srivastava et al. 2009), and at least one transporter in *Arabidopsis* (Sung et al. 2009) are upregulated in roots due to exposure to heavy metal like Arsenic.

4 Conclusions and Future Outlook

Omics technology plays an important role in biotic and abiotic stress responses in plants. It plays significant role in crop quality management under stress condition. With the use of genomics, transcriptomics, proteomics, metabolomics, etc. in plant breeding, plant breeder can improve the consistency and predictability by reducing the time and expense of producing better quality food crops that are resistant to various stresses. It also helps to understand the mechanism involved in plant-microbes and plant-pests' interactions and the various responses involved in these interactions. Thus, the use of various omics approaches has created a platform for further research by establishing networks of interaction between genes, proteins, and metabolites that are involved in stress response mechanism.

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