Chapter 1

1.1 All the abiotic stresses affecting plant trigger complex responses aiming to increase the stress tolerance. The responses include the emission of Volatile Organic Compounds (VOCs). The VOCs like ET, JA, NO, Na+ etc produced in response to the different stresses are also shown. A common mechanism links together the different stresses: all cause oxidative stress and hamper the production of reactive oxygen species. Excess light and heat, as well as exposure to oxidizing air pollutants, cause direct accumulation of ROS which crucially contributes to initiate the stress-related signal cascades

1.2 Generic pathway for plant response to stress. The extracellular stress signal is first perceived by the membrane receptors and then activate large and complex signaling cascade intracellularly including the generation of secondary signal molecules. The signal cascade results in the expression of multiple stress responsive genes, the products of which can provide the stress tolerance directly or indirectly

1.3 The effect of environmental stress on plant survival

1.4 Plants perceive PAMPs/MAMPs or effector proteins using extracellular or intracellular receptors and activate immune responses. The tomato receptor-like protein Ve1 and the rice receptor-like kinase Xa21 are examples of extracellular receptors that recognize *Verticillium* Ave1 and *Xanthomonas oryzae* pv. oryzae Xa21, respectively. Tomato I-2 and *Arabidopsis* RRS1-R are examples of intracellular NB–LRR-type receptors that perceive the *F. oxysporum* f. sp. lycopersici Avr2 effector and the *R. solanacearum* effector PopP2, respectively

1.5 Diagram of cold-responsive transcriptional network in *Arabidopsis*. Plants probably sense low temperatures through membrane rigidification and/or other cellular changes, which
might induce a calcium signature and activate protein kinases necessary for cold acclimation. Constitutively expressed ICE1 is activated by cold stress through sumoylation and phosphorylation. CBFs regulate the expression of COR genes that confer freezing tolerance.

1.6 Schematic diagram showing the molecular regulatory mechanism of heat shock proteins based on a hypothetical cellular model. Upon heat stress perceived by the plant cell, (a) monomeric heat shock factors (HSFs) are entering into the nucleus; (b) from the cytoplasm. In the nucleus, HSF monomers form active trimers; (c) that will bind; (d) to the specific genomic region (promoter or heat shock element, HSE) of the respective heat shock gene (HSG). Molecular dissection of the HSF binding region of HSE showing that it consists of one DNA binding domain and two domains for trimerization of HSFs. Successful transcription (e) translation and post-translational modification; (f) lead to produce functional HSP to protect the plant cell and responsible for heat stress tolerance.

1.7 A scheme showing the interaction interface and overlapping signaling pathways of abiotic and biotic stress at the cellular level.

1.8 Three-dimensional structure of GT-A (A) GT-B (B) proteins. In panel B, the two Rossmann domains are shown in red and green color. The sugar donor (magenta) and acceptor (blue) are shown in stick form.

1.9 Three-dimensional structure of CC domain of *Hordeum vulgare* (PDB-ID: 3QFL) (A) TIR domain of RPS4 protein of *Arabidopsis* (PDB-ID: 4C6R).

1.10 Three-dimensional structure of polygalacturonase inhibiting protein, a leucine rich protein of *Phaseolus vulgaris* (PDB-ID: 1OGQ).

1.11 Three-dimensional structure of aspartate protease from *Hordeum vulgare* (PDB-ID: 1QDM).

1.12 Three-dimensional structure of cysteine protease of *Hordeum vulgare* (PDB-ID: 2FO5).

1.13 The three-dimensional structures of representative members of.
the 13 classes [A-M] of serine proteases. A. Trypsin (Deg5 & Deg8) from *Arabidopsis* (PDB-ID: 4IC5 & 4IC6); B. Clp Endopeptidase from *Bacillus subtilis* (PDB-ID: 3KTG); C. C-terminal processing peptidases from *Scenedesmus obliquus* (PDB-ID: 1FC6); D. Lon proteases from *Bacillus subtilis* (PDB-ID: 3M6A); E. Lys-Pro-X carboxypeptidase from *Homo sapiens* (PDB-ID: 3N2Z); F. Nucleoporin autopeptidases from *Homo sapiens* (PDB-ID: 2Q5X); G. Prolyl oligopeptidases from *Trypanosoma brucei* (PDB-ID: 4BP8); H. Protease IV from *Escherichia coli* (PDB-ID: 3BEZ); I. Rhomboid from *Haemophilus influenzae* (PDB-ID: 2NR9); J. Serine carboxypeptidases from *Triticum aestivum* (PDB-ID: 1BCR); K. Signal peptidases I from *Escherichia coli* (PDB-ID: 1B12); L. Subtilase from *Cucumis melo* (PDB-ID: 3VTA)

1.14 The three-dimensional structure of cysteine protease inhibitor of *Solanum tuberosum* (PDB-ID: 3W9P).
1.15 The three dimensional structures of serine protease inhibitors [A-G]. A. Kunitz inhibitor from *Delonix regia* trypsin inhibitor (PDB-ID: 1R8N); B. Bowman-Birk inhibitor from *Medicago scutellata* (PDB-ID: 2ILN); C. Squash inhibitor from *Cucurbita pepo* (PDB-ID: 2BTC); D. Serpin from *Arabidopsis thaliana* (PDB-ID: 2ILN); E. Ragi seed Trypsin/α-Amylase Inhibitor/Lipid transfer protein from *Hordeum vulgare* (PDB-ID: 3GSH); F. Pin-I from *Fagopyrum esculentum* (PDB-ID: 3RDY); G. Pin-II from *Solanum lycopersicum* (PDB-ID: 1PJU)

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two templates employed for the modeling studies

3.4 Molecular model of *Fragaria* UGT prepared using pdb structure 2C1Z and 3HBF as templates. B: Ramachandran plot C: ERRAT plot D: Overall model quality plot generated using ProSA showing the Z-score of the model as black dot

3.5 a: Stereo image representing cartoon drawing of UGT from *Vitis vinifera* with conserved amino acids of the N-terminal domain shown in stick form. b: Stereo image representing cartoon drawing of UGT from *Vitis vinifera* with conserved amino acids of the C- terminal domain shown in stick form

3.6 Stereo view of the docked complex of UGT from *Fragaria ananassa* with kaempferol shown in stick form. The arrow shows the 3-OH group of kaempferol which take part in glycosylation event. UDP-glucose is also shown in stick form

3.7 Ribbon view of UGT from *Vitis vinifera* with docked acceptor and sugar donor in stick form is shown. Six conserved regions from N1 to N6 at the NTD and two regions C1 and C2 at the CTD marked with an arrow plays a crucial role in holding the acceptor in the binding pocket

3.8 Multiple sequence alignment of 30 F3GT protein sequences to show the eight conserved regions at the N- and C- terminal domain involved in the binding of flavonoid acceptor

3.9 Stereo view of docked complex of UGT from *Vitis vinifera* (2C1Z) with Kaempferol-3-O-glucoside and UDP shown in stick form

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3.13 Image showing docked complexes of three positive and two negative control ligands in the acceptor binding pocket of *Dianthus caryophyllus* (*Dianthus_1*).
Image showing docked complexes of three positive and three negative control ligands in the acceptor binding pocket of *Diospyros kaki* (*Diospyros*).

Image showing docked complexes of three positive and three negative control ligands in the acceptor binding pocket of *Petunia hybrida* (*Petunia_2*).

Image showing docked complexes of three positive control ligands in the acceptor binding pocket of *Scutellaria baicalensis*.

Chapter 4

4.1 Surface representation of UGT88E9 with bound quercetin (Yellow) and UPG (blue) shown in stick form. The NTD and CTD are shown in red and green color with the interdomain linker marked by arrows.

4.2 Conservation [Bit score (a) and Relative entropy (b)] of the PSPG motif of 89 UGTs from various plant sources.

4.3 The number of *CaUGTs* identified using various methods such as PSWM search using MEME-MAST, Blastp and HMM-profiles search shown with the help of a Venn diagram.

4.4 Genomic distribution of *CaUGTs*. Chromosomal distribution of *CaUGTs* in chickpea genome.

4.5 Phylogenetic analysis of *CaUGTs*. Dendrogram showing clustering of 96 *CaUGTs* along with two recent gene duplication events marked by arrows.

4.6 Functional annotation of *CaUGTs*. Dendrogram showing clustering of 96 *CaUGTs* with 38 well characterized UGT proteins from other plant species. The image shows distinct clustering of *CaUGTs* with the functionally related UGTs.

4.7 Multiple sequence alignment of members of group A1 cluster to show the eight conserved regions enclosed in boxes near the acceptor binding site.

4.8 Docked complexes of *CaUGTs* with their respective acceptor and sugar donor. A. The docked complex of *CaUGT* of group A1 with cyanidin (shown in stick form) interacting with H26 and H155. B. The docked complex of *CaUGT* of group B with cytokinin (shown in stick form) interacting with H21 and H404. C. The docked complex of *CaUGT* of group A2 in which 3-OH.
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4.13 Violin plot representing distribution of FPKM values of all the
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5.5 Multiple sequence alignment of cysteine protease inhibitors of chickpea

5.6 Multiple sequence alignment of Bowman-Birk inhibitor of chickpea and other plant species. The cysteines involved in the disulphide bond formation are marked with an arrow. P1 and P1’ residues are also shown

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5.12 Domain arrangement of chickpea APs and SPs

5.13 Image shows gene architecture and clustering of CaAPs. The genomic locations of aspartate proteases with IDs CaAP_S6, CaAP_S1, CaAP_S2, CaAPS3, CaAP_S7 were unknown and therefore assigned them on scaffolds

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5.15 Expression level for chickpea protease genes in various tissues by RNA-seq data analysis. Heatmap showing relative gene expression in various tissue samples. The color scale represents log transformed count per million (CPM), for proteases genes in
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5.16 Expression level for chickpea protease inhibitor genes in various tissues by RNA-seq data analysis. Heatmap shows relative gene expression in various tissue samples. The color scale represents log transformed count per million (CPM), for protease inhibitor genes in different tissues. The protease inhibitor genes with FPKM > 0 are included in the analysis. Dendrogram on the left side of the heatmap shows hierarchical clustering of genes using complete linkage approach

5.17 Homology models of 10 Kunitz inhibitors of chickpea are shown in ribbon representation. The lower panel shows the structural alignment of ten chickpea Kunitz inhibitors and Kunitz type dual inhibitor (TKI) of factor Xa (FXa) and trypsin of tamarind. The reactive loops are shown in the enclosed box.

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5.20 A. The docked complex of cysteine protease inhibitor (red) and bovine trypsin (green) showing inter and intra molecular hydrogen bonds at the interface. B. The docked complex of PI-I (red) and bovine trypsin (green) showing inter and intra molecular hydrogen bond at the interface. C. The docked complex of BBI (red) and papain (green & yellow) showing inter and intra molecular hydrogen bond at the interface

5.21 The root mean square deviation plot of C-alpha atoms of Bowman-Birk inhibitor, cysteine protease inhibitor, POP, and PI-I.

5.22 The image shows superposition of the initial structure (green) and the structure after 10 ns simulation (red).
5.23 The docked POP-ZPR complex after 10 ns simulation. The docked complex of POP and drug ZPR (yellow) showing intermolecular hydrogen bond between them. The β-propeller domain is shown in green color and α-β hydrolase domain is shown in red color. After 10 ns, the drug blocked the active site residues His696 and Ser563.

5.24 A. The docked cysteine protease inhibitor (red) and bovine trypsin (green) after 10 ns simulation. B. The docked complex of PI-I (red) and bovine trypsin (green) after 10 ns simulation. C. The docked complex of BBI (red) and papain (yellow & green) after 10 ns simulation.

Chapter 6

6.1 The schematic diagram shows the arrangement of domains present in the NBS-LRR proteins. The functional role of each domain is also shown.

6.2 The image shows eight groups of plant resistance genes based on the motif organization and membrane spanning regions.

6.3 The NBS domains of TNL proteins of chickpea (from P-Loop to GLPL) were shown that were used to construct the phylogeny.

6.4 The NBS domains of non-TNL proteins of chickpea (from P-Loop to GLPL) are shown, same are used to construct the phylogeny.

6.5 Circular representation of dendrogram reveals distinct clusters of non-TNL and TNL chickpea proteins. The two black circles show clades of two families of NBS encoding genes. The non-TNL family is divided into subfamilies CNL1 to CNL4. The TNL family is classified into subfamilies TNL1-TNL3. The diamonds represents the non-TNL proteins in which RPW8 domain fusion has occurred. The green circles depict pair of genes involved in segmental duplication events.

6.6 Distribution of non-TNL and TNL family members of NBS-LRR gene family on chromosome 1 to 8 and scaffolds of chickpea genome. Dashed and straight lines represent the non-TNL and TNL genes, respectively.

6.7 Exon-intron arrangement of non-TNL genes of chickpea.

6.8 Exon-intron arrangement of TNL genes of chickpea.
6.9 Heatmap shows relative gene expression of chickpea non-TNL genes in various tissues samples by RNA-seq data analysis. The color scale represents log (base 10) transformed FPKM values, calculated by comparing fragment kilo base transcript per million (FPKM) value for NBS-encoding genes in different tissues. The NBS genes with FPKM > 0 are included in the analysis. Dendrogram on the left side of the heatmap shows hierarchical clustering of genes using complete linkage approach.

6.10 Heatmap shows relative gene expression of chickpea TNL genes in various tissues samples by RNA-seq data analysis. The color scale represents log (base 10) transformed FPKM values, calculated by comparing fragment kilo base transcript per million (FPKM) value for NBS-encoding genes in different tissues. The NBS genes with FPKM > 0 are included in the analysis. Dendrogram on the left side of the heatmap shows hierarchical clustering of genes using complete linkage approach.

Chapter 7

7.1 Pie chart depicting the distribution of members of different classes of additional stress genes identified in chickpea.

7.2 Heatmap shows relative gene expression of chickpea chitinase and HSPs genes in various tissues samples by RNA-seq data analysis. The color scale represents log (base 10) transformed FPKM values, calculated by comparing fragment kilo base transcript per million (FPKM) value for identified genes in different tissues. The genes with FPKM > 0 are included in the analysis. Dendrogram on the left side of the heatmap shows hierarchical clustering of genes using complete linkage approach.

7.3 Heatmap shows relative gene expression of chickpea glucanase genes in various tissues samples by RNA-seq data analysis. The color scale represents log (base 10) transformed FPKM values, calculated by comparing fragment kilo base transcript per million (FPKM) value for identified genes in different tissues. The genes with FPKM > 0 are included in the analysis. Dendrogram on the left side of the heatmap shows hierarchical clustering of genes using complete linkage approach.

7.4 Heatmap shows relative gene expression of chickpea thaumatin genes in various tissues samples by RNA-seq data analysis. The color scale represents log (base 10) transformed FPKM values, calculated by comparing fragment kilo base transcript per million (FPKM) value for identified genes in different tissues. The genes with FPKM > 0 are included in the analysis. Dendrogram on the left side of the heatmap shows hierarchical clustering of genes using complete linkage approach.
genes in various tissues samples by RNA-seq data analysis. The color scale represents log (base 10) transformed FPKM values, calculated by comparing fragment kilo base transcript per million (FPKM) value for identified genes in different tissues. The genes with FPKM > 0 are included in the analysis. Dendrogram on the left side of the heatmap shows hierarchical clustering of genes using complete linkage approach.

7.5 Heatmap shows relative gene expression of chickpea LEA and LTP genes in various tissues samples by RNA-seq data analysis. The color scale represents log (base 10) transformed FPKM values, calculated by comparing fragment kilo base transcript per million (FPKM) value for identified genes in different tissues. The genes with FPKM > 0 are included in the analysis. Dendrogram on the left side of the heatmap shows hierarchical clustering of genes using complete linkage approach.

7.6 Heatmap shows relative gene expression of chickpea peroxidase genes in various tissues samples by RNA-seq data analysis. The color scale represents log (base 10) transformed FPKM values, calculated by comparing fragment kilo base transcript per million (FPKM) value for identified genes in different tissues. The genes with FPKM > 0 are included in the analysis. Dendrogram on the left side of the heatmap shows hierarchical clustering of genes using complete linkage approach.