

Comparative transcriptomics identifies distinct molecular profiles of drought and bacterial stress responses in plants



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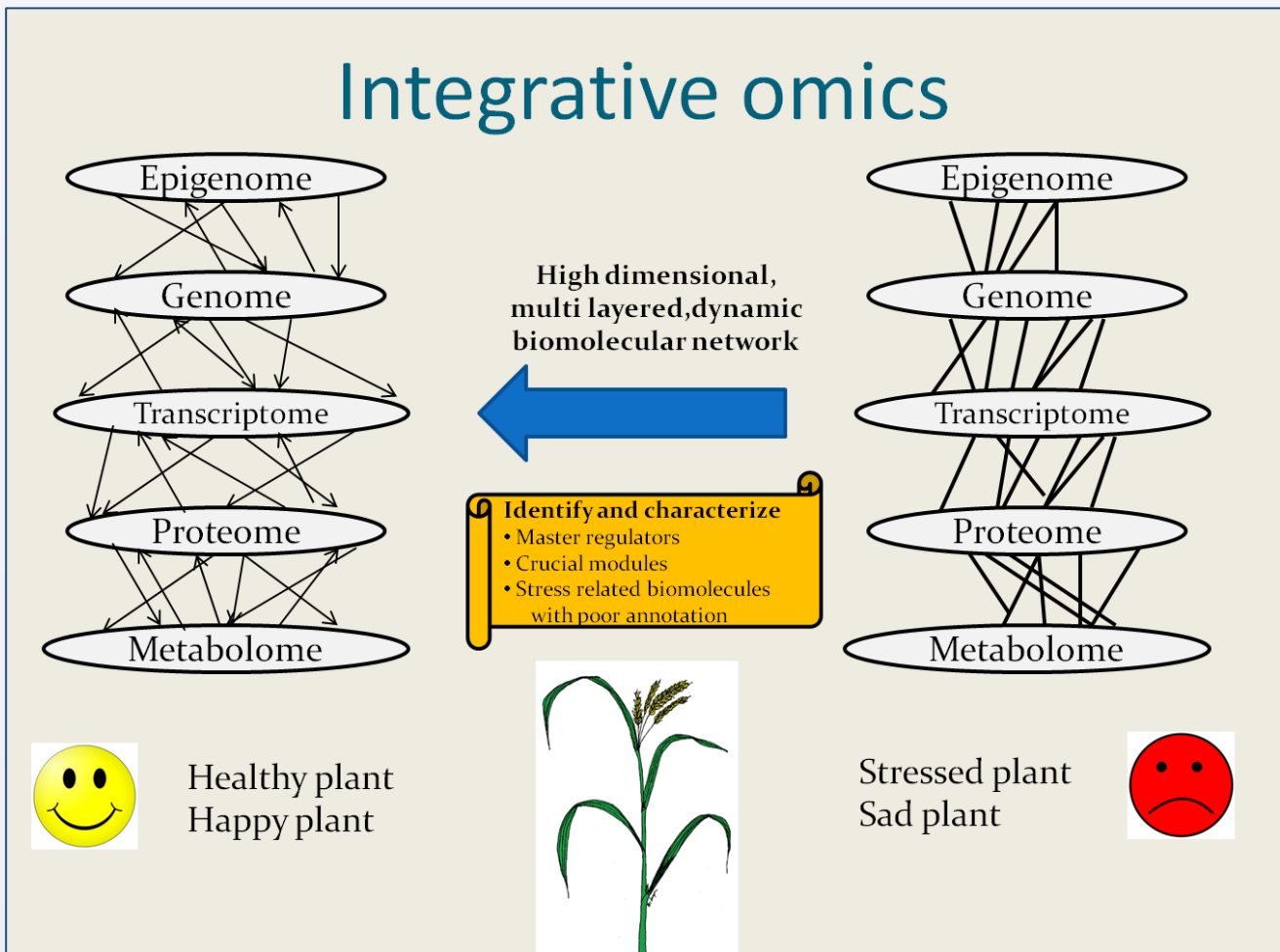
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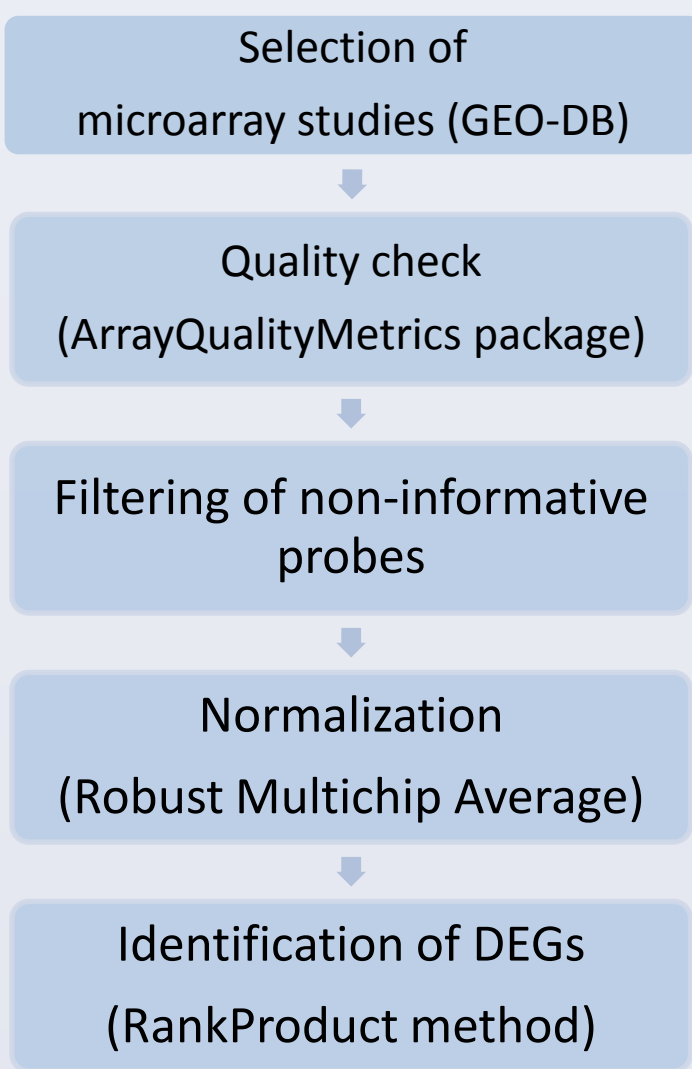
INTRODUCTION



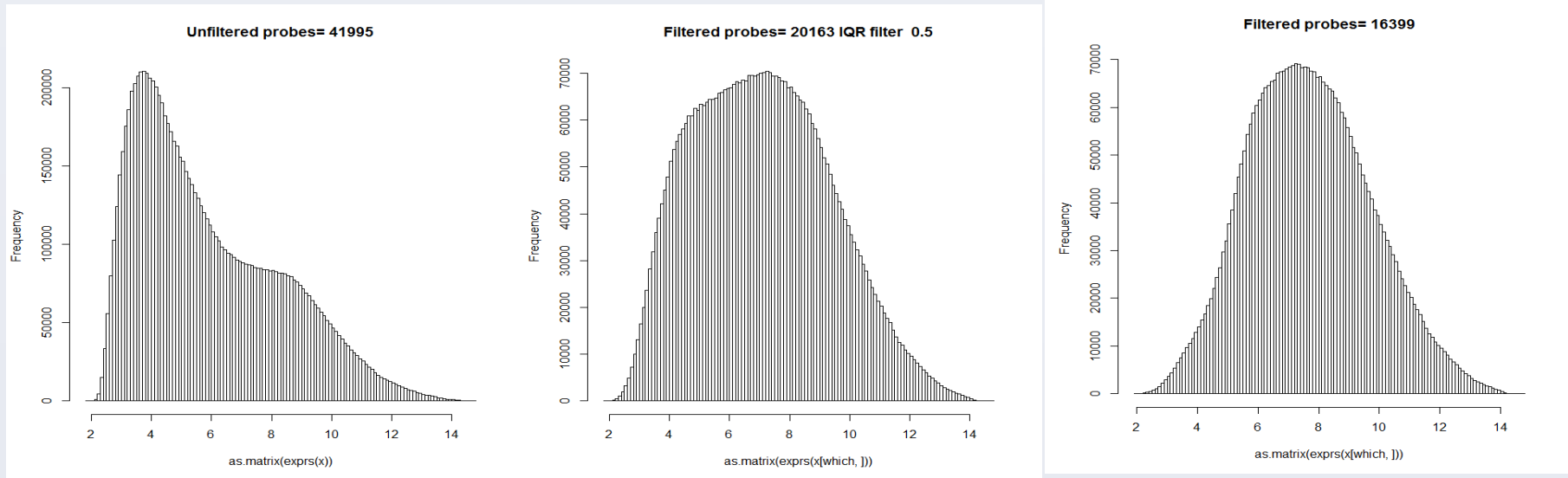
More often than not plants are exposed simultaneously to multiple stresses resulting in enormous changes in the molecular landscape within the cell.

Here, we performed meta-analysis of drought (abiotic), bacterial (biotic) stress microarray studies in rice (monocot) and arabidopsis (dicot) and compared the identified differentially expressed genes (DEGs) which elucidated the common and specific molecular profiles for different stresses across plant species.

METHODS



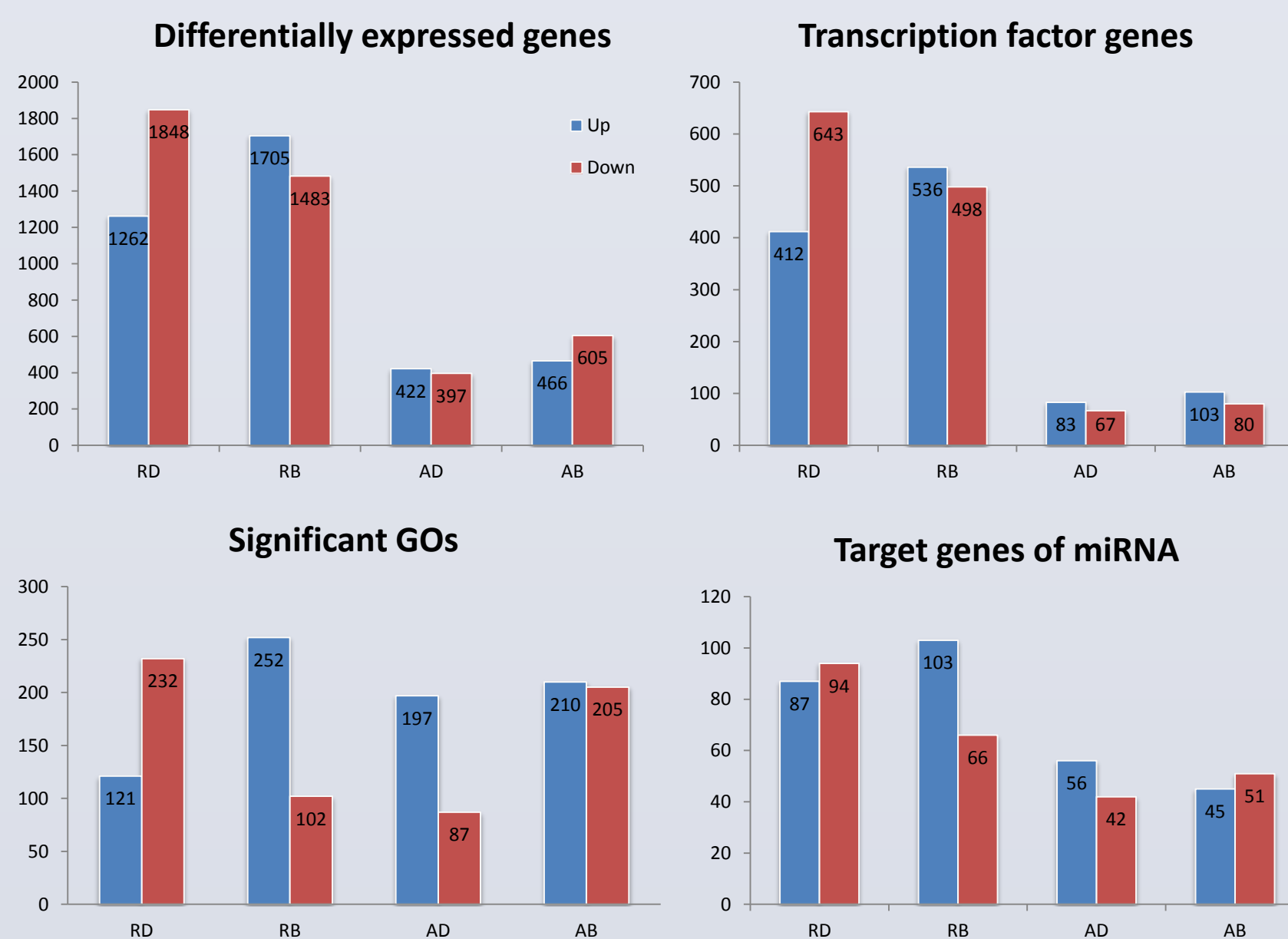
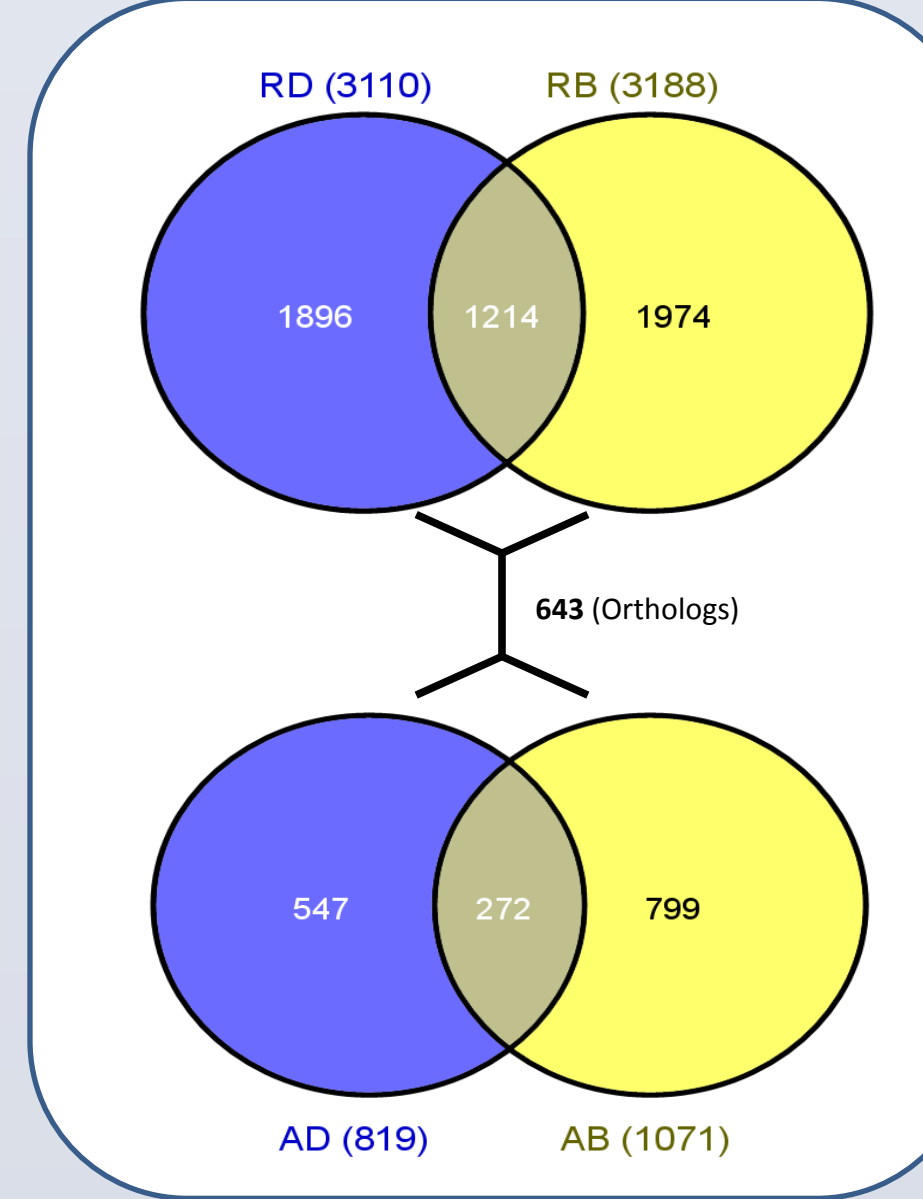
Distribution of expression values of microarray probes after filtering



Stress condition	No. of Series	No. of Samples	No. of controls	No. of treatments	Total DEGs
Rice Drought (RD)	5	78	35	43	3110
Rice Bacteria (RB)	7	227	61	166	3188
Arabidopsis Drought (AD)	7	114	61	53	819
Arabidopsis Bacteria (AB)	5	106	40	66	1071
Total		525	197	328	

RESULTS

Overlap of DEGs in different stresses



Gene Ontology (GO) enrichment analysis

(terms most significantly enriched in rice shown in bold, arabidopsis shown in italics and both shown in plain text)

Drought Stress

Response to: water, heat, high light intensity, osmotic stress
Regulation of: transcription DNA-dependent, protein modification process, hormone levels, signal transduction, cell communication
B.P.: positive regulation of macromolecule biosynthetic process
M.E.: protein serine/threonine phosphatase activity, phosphofructokinase activity

COMMONLY UPREGULATED

Regulation of: gene expression, RNA metabolic process, macromolecule biosynthetic process
Metabolism: toxin catabolic process, transaminase activity, glucose, hydrolase activity, hydrolyzing O-glycosyl compounds
Response to: oxidative stress, abscisic acid, salicylic acid, ethylene, jasmonic acid, chitin, carbohydrate, wounding, other organism
cell part, catalytic activity, oxidoreductase activity, localization, metabolic process: cellular nitrogen compound, electron Transport, small molecule. Response to organic substance, fungus, endogenous, chemical and hormonal stimuli

Bacterial stress

Response to: Bacterium, hypoxia
Regulation of: cell size, cellular component size, defense response, innate immune response
B.P.: protein ubiquitination, cell wall thickening, glycolysis, translation, host programmed cell death by symbiont
M.E.: protein serine/threonine kinase activity
C.C.: endoplasmic reticulum, ribosome and its parts, mitochondria and its parts

COMMONLY DOWNREGULATED

Regulation of: negative regulation of catalytic Activity
B.P.: L-serine metabolic process
C.C.: nucleosome
M.E.: heme binding, sexual reproduction, protein-DNA complex assembly
two-component response regulator activity

Response to: cytokinin, auxin, gibberellin, metal ion, light stimulus
B.P.: photosynthesis, circadian rhythm cofactor biosynthetic process
M.E.: tetrapyrrole binding, substrate-specific transmembrane transporter activity
C.C.: apoplast, chloroplast and its parts

Regulation of: innate immune response, Homeostatic process,
Response to: hydrogen peroxide
B.P.: phosphoinositide metabolic process
M.E.: steroid metabolic process
C.C.: NAD(P)H dehydrogenase complex (plastoquinone)

KEGG Orthology (KO) enrichment analysis

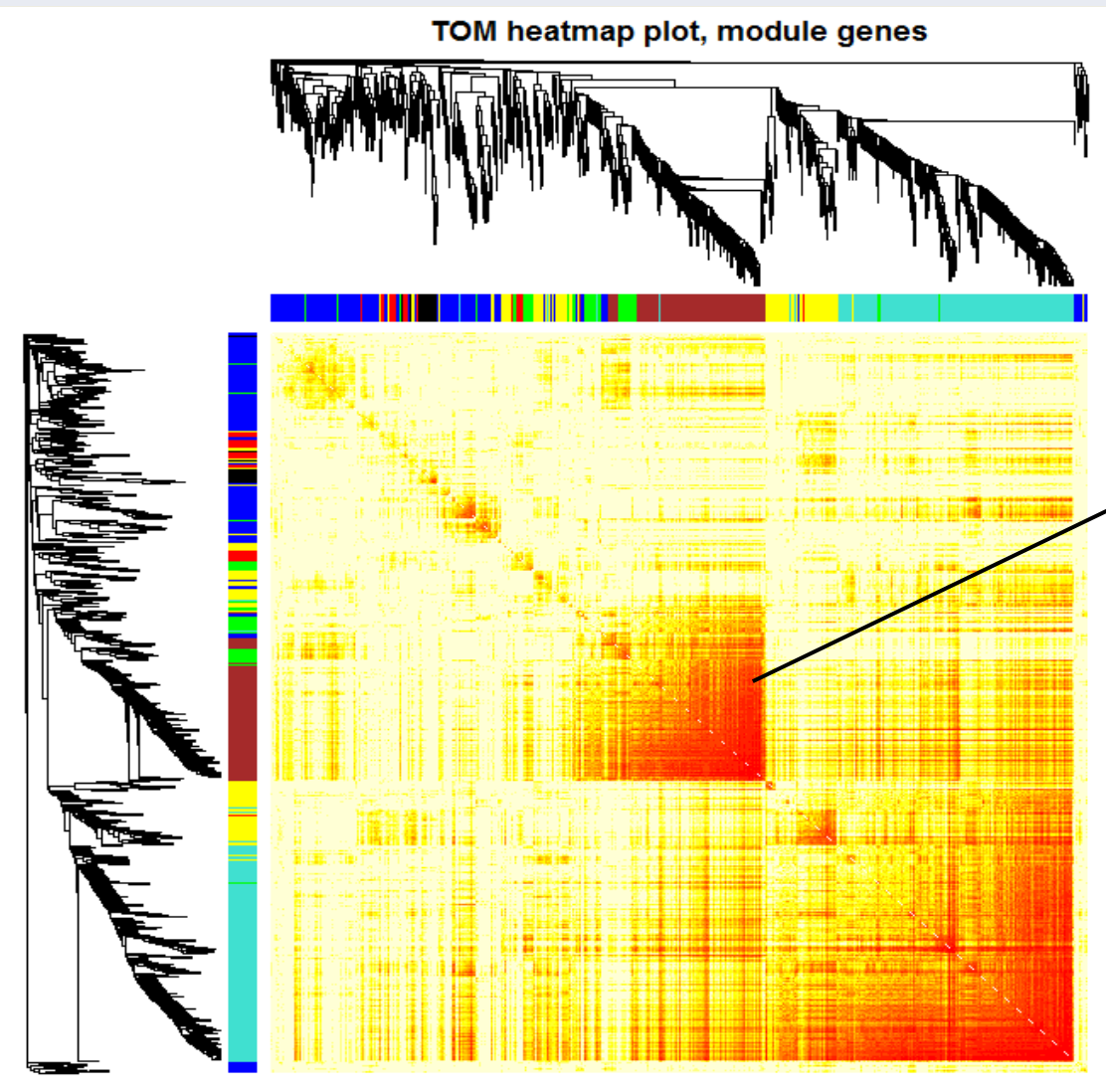
(* - hypergeometric P-value <0.05; ** - hypergeometric P-value <0.001)

KO ID	Definition	Pathway	ADD	ABD	ABU	ADU
K00799	Glutathione S-transferase	Glutathione metabolism; Chemical carcinogenesis			**	**
K05359	Arogenate/prephenate dehydratase	Phenylalanine, tyrosine and tryptophan biosynthesis			*	*
K14492	Two-component response regulator ARR-A family	Plant hormone signal transduction	*	**		
K14488	SAUR family protein	Plant hormone signal transduction	*	*		
K13993	HSP20 family protein	Protein processing in endoplasmic reticulum		*		*
K01177	Beta-amylase	Starch and sucrose metabolism		*		**
K08081	Tropine dehydrogenase	Tropine, piperidine and pyridine alkaloid biosynthesis	*	*		
KO ID	Definition	Pathway	RDD	RBD	RBU	RDU
K02639	Ferredoxin	Photosynthesis	**			
K00815	Tyrosine aminotransferase	Ubiquinone and other terpenoid-quinone biosynthesis		**	**	
K03283	Heat shock 70kDa protein	Spliceosome; MAPK signaling pathway			*	*
K01183	Chitinase	Amino sugar and nucleotide sugar metabolism				*
K10999	Cellulose synthase A	Starch and sucrose metabolism	**			

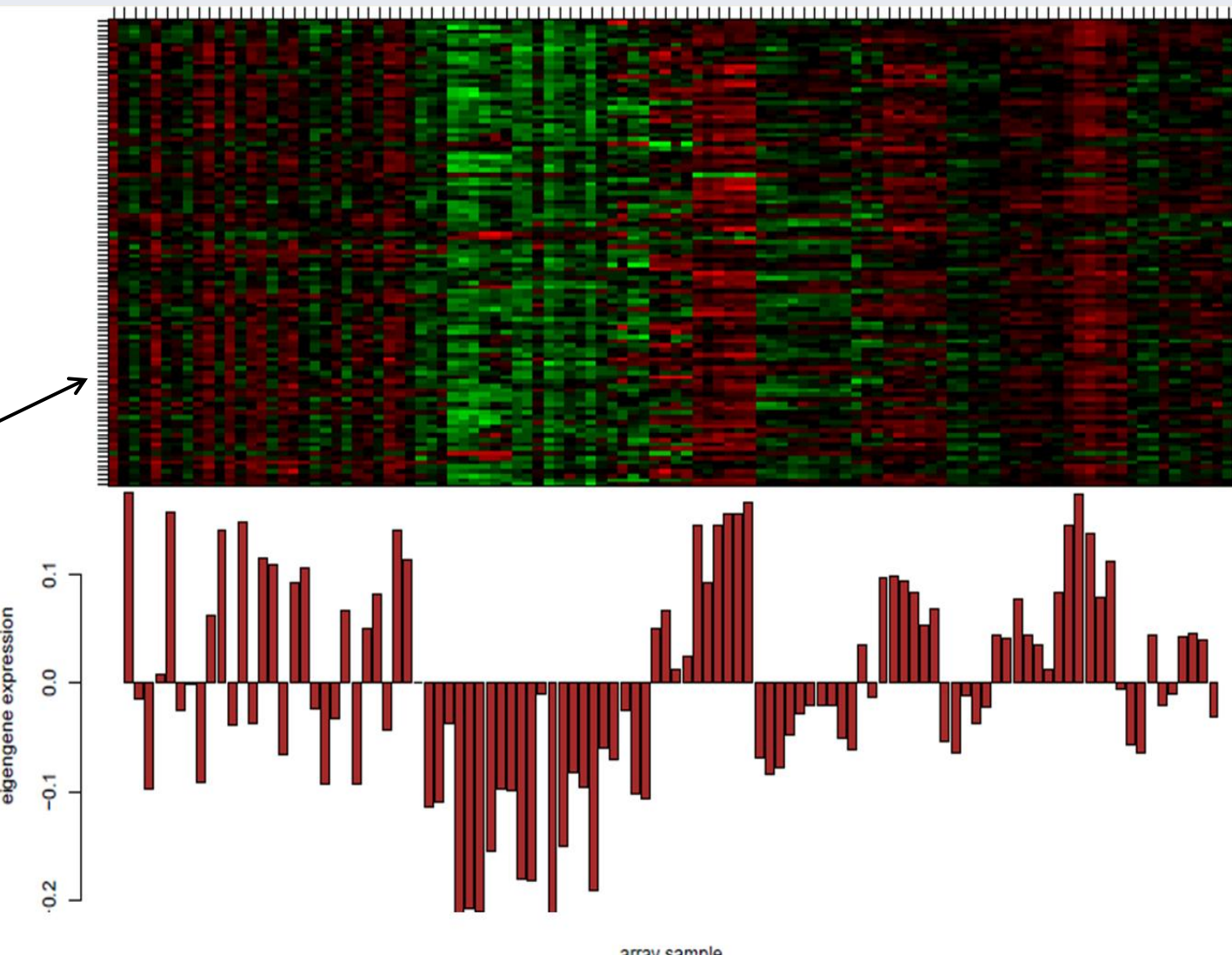
Weighted Gene Co-expression Network Analysis (WGCNA)

WGCNA package for R was used to identify gene co-expression modules from the gene expression values using Pearson's correlation matrix, soft thresholding, topological overlap matrix and hierarchical cluster tree

Heatmap of arabidopsis co-expressed DEGs divided into eight modules under bacterial stress



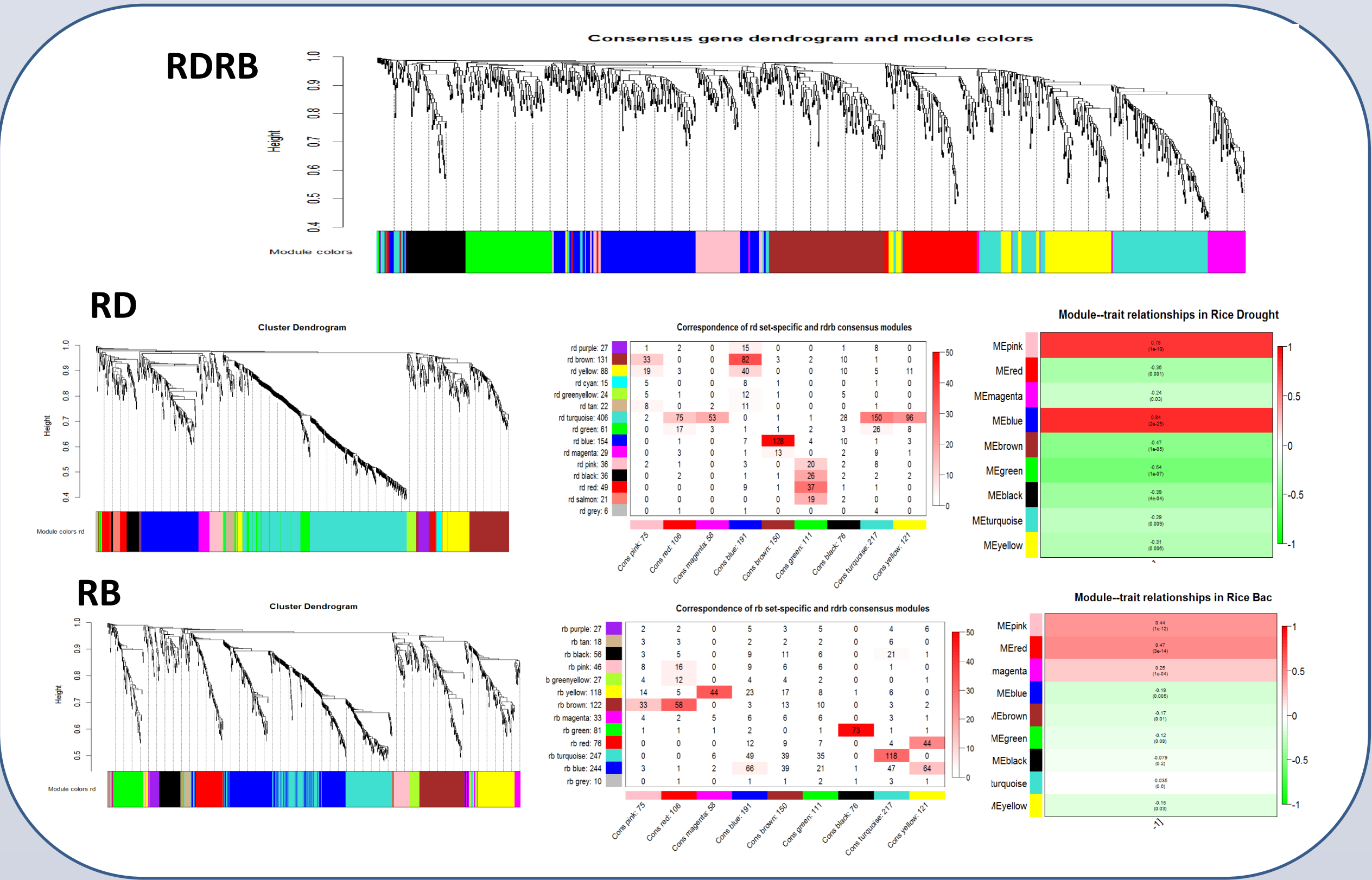
Eigengene expression graph of brown module containing 263 genes (~81% upregulated genes) across 106 microarrays



Top five genes in brown module with highest number of co-expressed genes within the module (as a function of no. of connections, k)

Locus	Annotation	kTotal	kWithin	kOut	kDiff
AT1G01560	ATMPK11; MAP kinase/ kinase	77.68	64.70	12.98	51.71
AT5G13320	PBS3 (AVRPPHB SUSCEPTIBLE 3)	74.43	62.51	11.92	50.59
AT3G52430	PAD4 (PHYTOALEXIN DEFICIENT 4); lipase/ protein binding / triacylglycerol lipase	65.75	57.14	8.60	48.53
AT1G07000	ATEXO70B2 (exocyst subunit EXO70 family protein B2); protein binding	86.13	67.00	19.12	47.87
AT1G73805	Calmodulin binding	81.85	64.85	17.00	47.85

Consensus WGCNA of 1214 DEGs common to RD and RB elucidates gene modules co-expressed across stresses



CONCLUSION

Our comparative analysis and comprehensive characterization of DEGs identified key features and regulatory modules that are generic and specific to drought and bacterial stress responses and also revealed conservation of many features in diverse plants species.