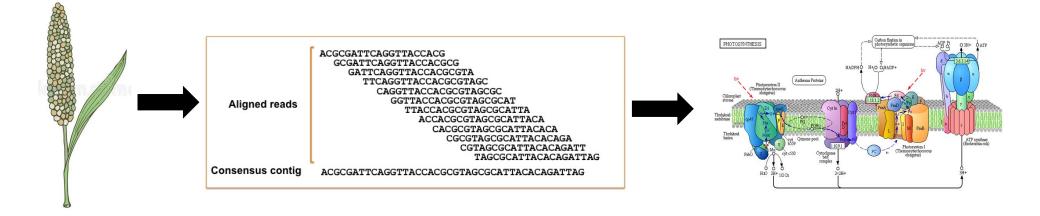


Transcriptomic analysis reveals the differentially expressed genes and pathways involved in drought tolerance in pearl millet [*Pennisetum glaucum* (L.) R. Br]



Ambika Dudhate

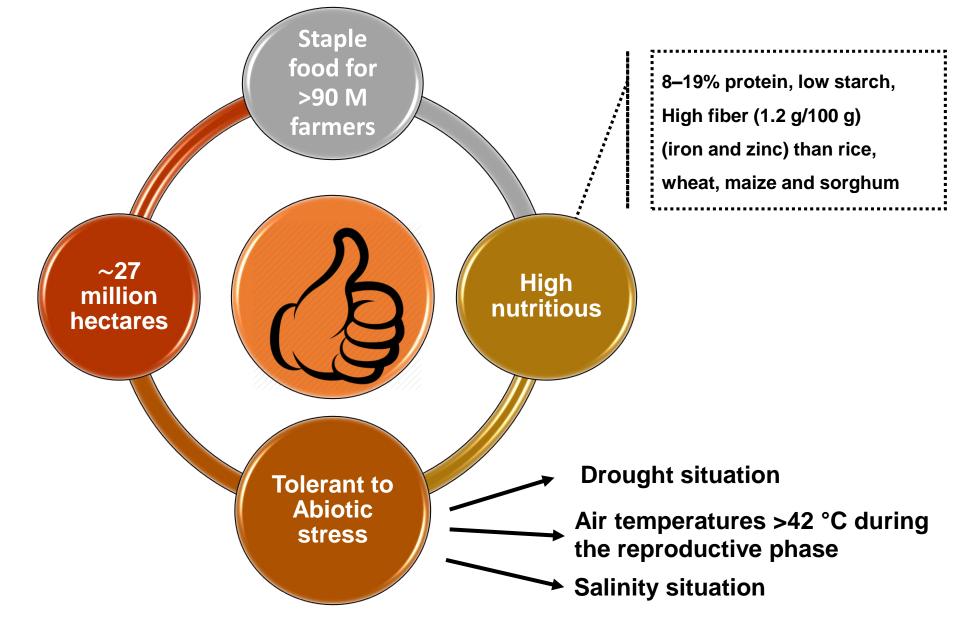
PhD-III year The Laboratory of environmental stress tolerance mechanism, The University of Tokyo, Japan

Introduction to Pearl Millet

- \circ C₄ crop.
- o Genus Pennisetum.
- o Other names- Bajara, Burush millet etc.
- \circ Diploid 2n=14.
- Arid areas of sub-Saharan Africa and India
- Novel source for investigation of molecular mechanism of drought.

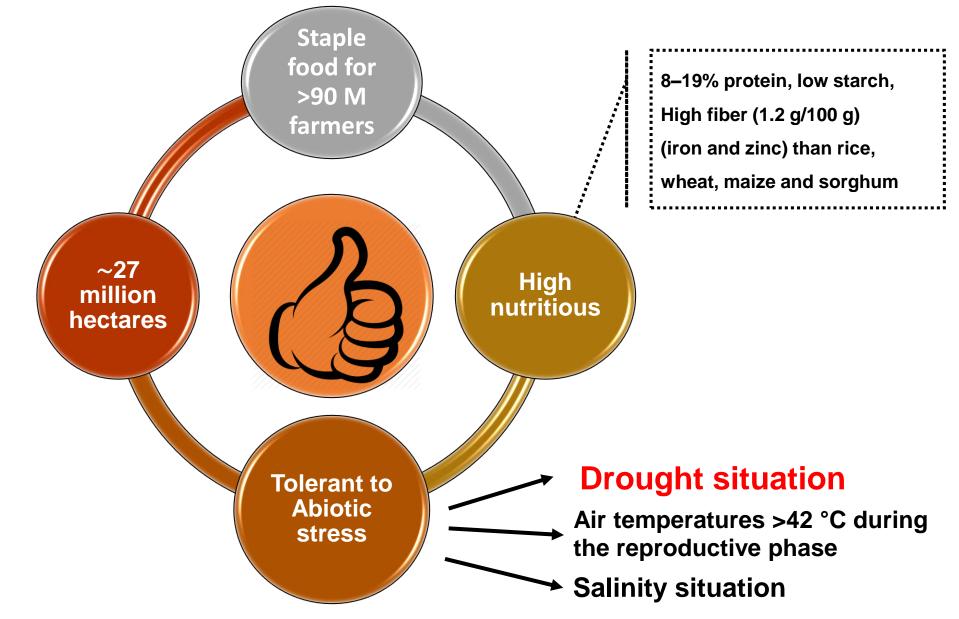


Why Pearl millet as study crop?



3

Why Pearl millet as study crop?





Release of Pearl millet genome in 2017.

Pearl millet genome sequence provides a resource to improve agronomic traits in arid environments

Rajeev K Varshney 🖾, Chengcheng Shi [...] Xun Xu 🖾

Nature Biotechnology 35, 969-976 (2017)

doi:10.1038/nbt.3943

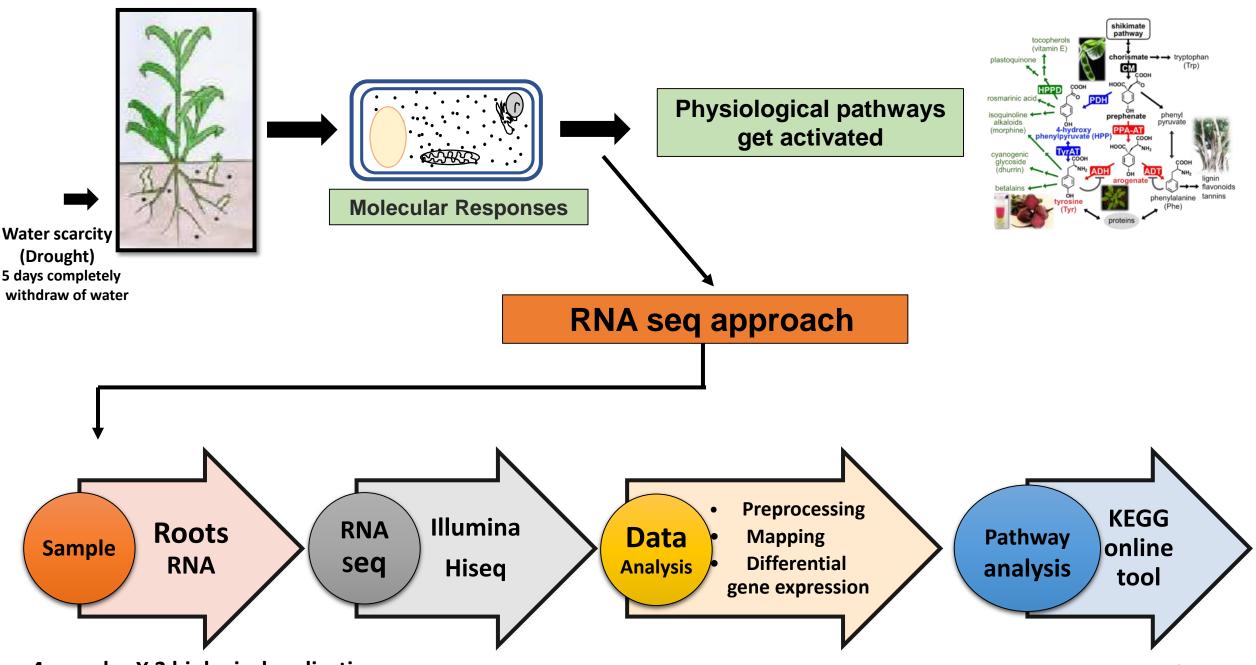
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Agricultural genetics

Next-generation sequencing

Received: 18 April 2017 Accepted: 17 July 2017 Published: 18 September 2017 Erratum: 05 April 2018 Corrected online 28 February 2018



4 samples X 3 biological replications

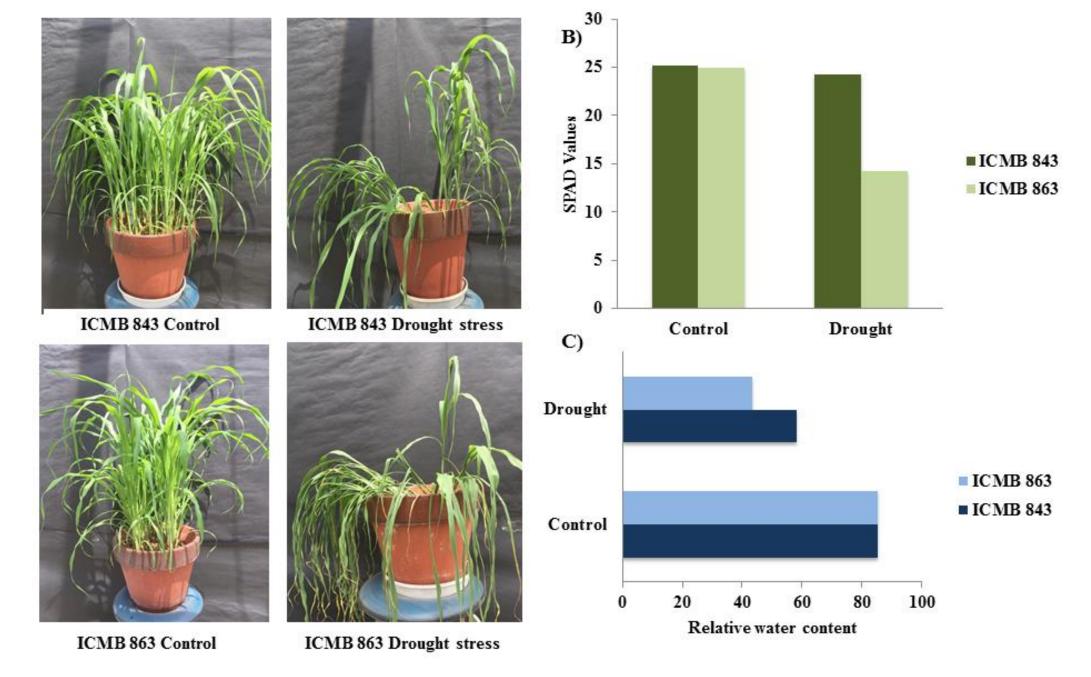


Fig 1. Physiological analysis of pearl millet for drought.

Foxtail millet (Setaria Italica) a better reference genome than Rice (Oryza Sativa)

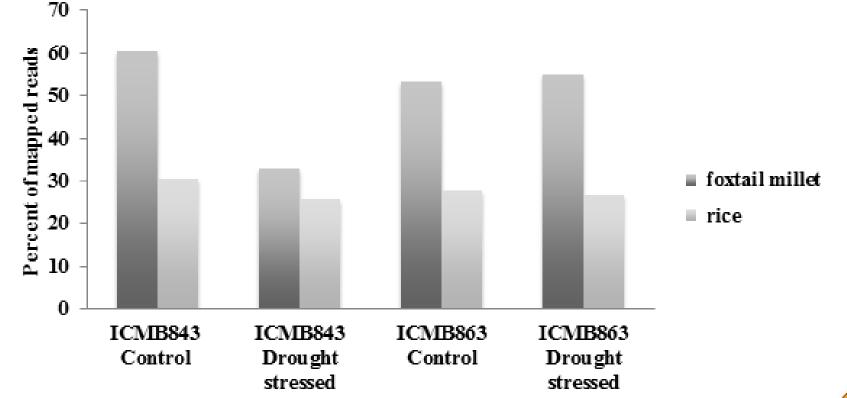


Fig 2. Mapping of pearl millet reads to the foxtail millet genome and the rice genome.



Lack of strong annotations to newly released pearl millet genome.....

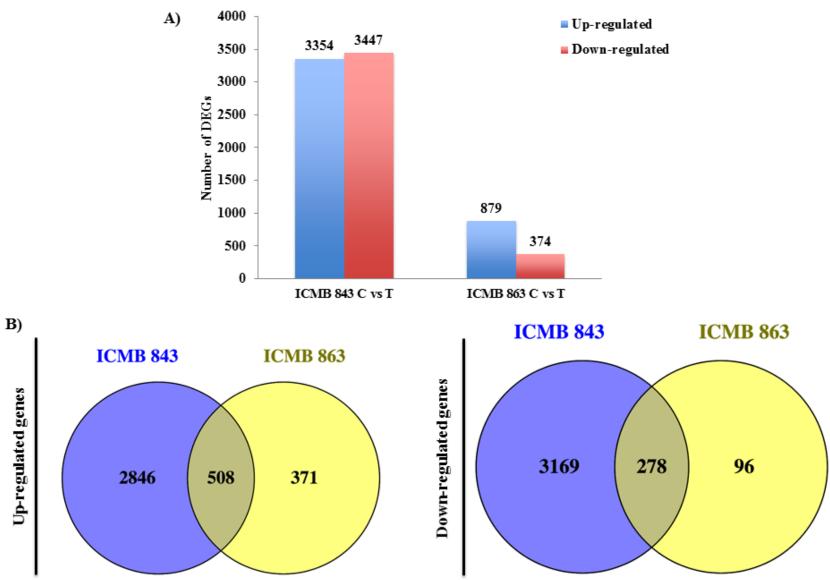


Fig 3. DEG analysis in comparisons study. A) Number of DEGs in all combinations with fold change >2 or <-2 and FDR-corrected pvalue <0.05) Blue and red bars indicate up- and down- regulated respectively. C: control and T and Treated: Drought- treated B) Venn diagram showing an up-regulated (right side) and down regulated (left side) genes in lines ICMB 843 and ICMB 863 under drought treated situation.

Fig4: Pathways Enriched with highest number DEGs were

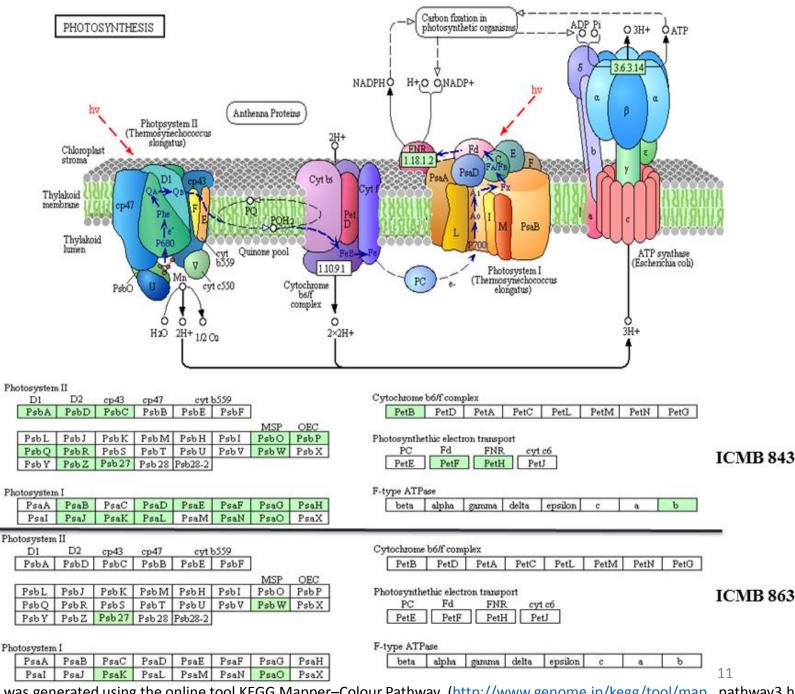
- Photosynthesis
- MAPK signaling pathway
- Plant hormone signal transduction

d					Alanine aspartate and glutamate metabolism Alpha Linolenic acid metabolism Amino sugar and nucleotide sugar metabolism AMPK signaling pathway Apelin signaling pathway Apoptosis Arginine and proline metabolism Autophagy other Beta Alanine metabolism Biosynthesis of amino acids Carbon fixation in photosynthetic organisms Carotenoid biosynthesis Cell cycle Cysteine and methionine metabolism Endocytosis Fatty acid degradation Fatty acid metabolism Galactose metabolism Glycerolipid metabolism Glycerolipid metabolism Glycerolipid metabolism Glycerolipid metabolism Glycerolipid metabolism Glycerolipid metabolism Glycerolipid metabolism MoxY signaling pathway Fructose and Gluconeogenesis Glycolysis and Gluconeogenesis Glycolysis and Gluconeogenesis Glycolysis and Gluconeogenesis Photosphate metabolism Nositol phosphate metabolism Nositol phosphate metabolism NAPK signaling pathway plant Methane metabolism Oxidative phosphorylation Pentose and glucuronate interconversions Pentose phospholipid metabolism Photosynthesis Photophyrin and chlorophyll metabolism Protein processing in endoplasmic reticulum Purine metabolism Pyrimidine metabolism Pyruvate metabolism Pyruvate metabolism Pyruvate metabolism Pyruvate metabolism Pyruvate metabolism Pyruvate metabolism Pyruvate metabolism Pyruvite metabolism Pyruvite metabolism Pyrimidine metabolism Typtophan metabolism Ubiquitin mediated proteolysis Valine leucine and isoleucine degradation
	I.843UP.	.843Down.	1.863UP.	I.863.Down.	10

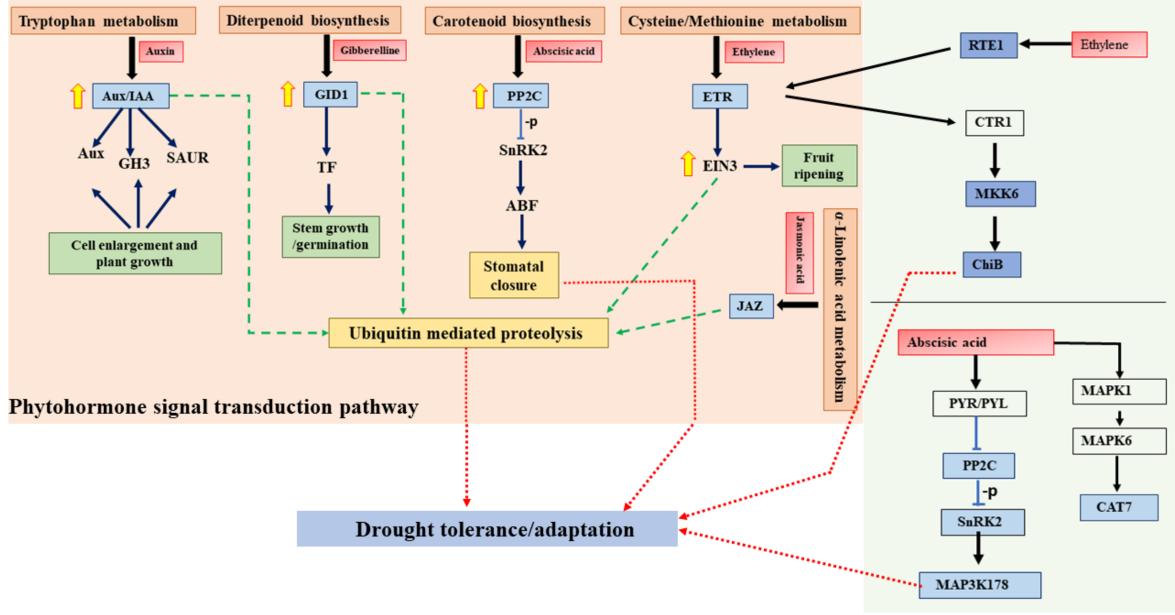
2

25

Fig 5. DEGs involved in photosynthesis. Genes up-regulated by drought stress are shown (25 genes in ICMB 843 and 4 in ICMB 863) in green White boxes. boxes indicate the non-droughtresponsive genes.

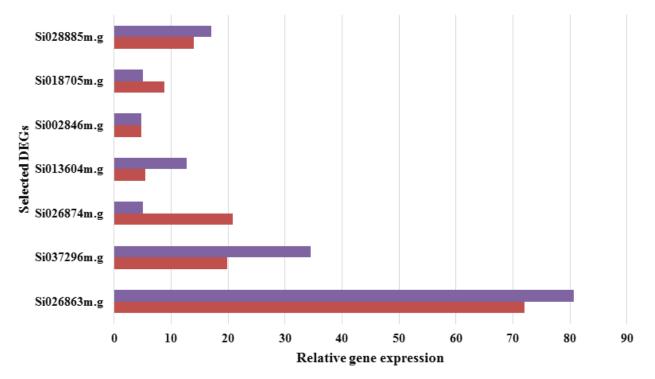


This image was generated using the online tool KEGG Mapper–Colour Pathway (<u>http://www.genome.jp/kegg/tool/map</u> pathway3.html).



MAPK signaling pathway

Fig 6. DEGs involved in phytohormone and MAPK signaling pathway



RT PCR (Fold change) RNA seq (RPKM) Fig 7. Validation of RNA-Seq result with RT PCR.

Name of the gene (Identifier)	Annotations	Fold (RT PCR)
Si028885m.g	Mixed linked glucan synthase	1.9
Si018705m.g	Chlorophyll centre-I related protein	5.11
Si002846m.g	Dehydration responsive elements binding protein 1F	4.45
Si013604m.g	Uncharacterized protein	12.8
Si026874m.g	Uncharacterized protein	5.1
Si037296m.g	Glutathione s-transferase (GSTU6)	34.5
Si026863m.g	Abiotic stress responsive factor	80.7

Conclusion remark

- Two pearl millet inbred lines, ICMB 843 and ICMB 863 were used
- ICMB 843 is relatively more tolerant to drought than ICMB 863 at seedling stage.
- Photosynthesis, plant hormone signal transduction and MAPK signaling pathways were induced by drought in pearl millet.
- Identified DEGs and metabolic pathways are targets for future studies in order to understand the molecular mechanism of drought tolerance in pearl millet.



RESEARCH ARTICLE

Transcriptomic analysis reveals the differentially expressed genes and pathways involved in drought tolerance in pearl millet [*Pennisetum glaucum* (L.) R. Br]

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Abstract

Pearl millet is a cereal crop known for its high tolerance to drought, heat and salinity stresses as well as for its nutritional quality. The molecular mechanism of drought tolerance in pearl millet is unknown. Here we attempted to unravel the molecular basis of drought tolerance in two pearl millet inbred lines, ICMB 843 and ICMB 863 using RNA sequencing. Under greenhouse condition, ICMB 843 was found to be more tolerant to drought than ICMB 863. We sequenced the root transcriptome from both lines under control and drought conditions using an Illumina Hi-Seq platform, generating 139.1 million reads. Mapping of sequenced reads against the foxtail millet genome, which has been relatively well-annotated, led to the identification of several differentially expressed genes under drought stress. Total of 6799 and 1253



OPEN ACCESS

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Laboratory of Environmental Stress Tolerance Mechanisms

Welcome to our website

Food comes first, then morals. Bertolt Brecht 1898-1956

Desertification. Salinization. Heavy-metal pollution. Environmental problems are threatening farming industry the worldwide. Since plants can not move, they developed "stress tolerance mechanisms" to cope with various environmental changes.

What is happening at DNA, cellular or whole plant level when plants are suffuring from the stresses? Why are some plants able to survive such severe conditions

Saline-alkali lands in Northeast China

The background image is the picture taken in alkalinesalt affected land in Northeast China, in Oct. 2009. Salt can be observed as white crusts on the ground. Most plants, including crops, cannot survive in such area. Both natural (climate, wind, water, etc.) and human-related (increased population, overgrazing, and policies) factors are responsible for the salinization or alkalinization of Northeast China.

Thank you for attention



