



ADOC: Allelic Diversity of Orthologous Candidate genes for drought tolerance in crops : example of the ASR gene family



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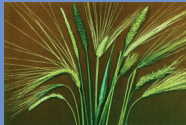


ABSTRACT

GCP (Generation Challenge Program) is funding a project called ADOC with the objective of producing a dataset of allelic diversity at orthologous candidate genes for drought tolerance in 7 crops (barley, rice, sorghum, common bean, chickpea, cassava and potato). The ASR (ABA, Stress and Ripening) gene family is one of the target genes chosen in this program. We gathered all sequences available in the databases on this gene family. No sequences was found for chickpea. We used these sequences to design consensus primers in order to amplify all genes of this family in all crops. These primers amplified some bands. These were picked, reamplified and sequenced. In a second step, specific primers will be designed and their whole sequence polymorphism will be assessed in a DNA bank of 283 reference accessions for each crop already characterized with neutral markers. This will allow us to analyze the (ortho)allelic diversity, to study the linkage disequilibrium in the selected crops along selected genes. This reference germplasm could allow testing association between genes polymorphism and trait variability.

PROJECT

- Research of **available sequences in databases** for 5 gene families (ASR, invertase, Sucrose synthase, DREB and ERECTA) in these 7 crops:



barley



rice



sorghum



common bean



chickpea



cassava



potato

- Design **consensus primers** in order to sequence more genes of these families in all crops and complete these data
- Choice one or more genes in each family (based on physiological data) and identify **orthologous genes** by phylogenetic analysis
 - Design **specific primers** for PCR amplification of overlapping sequences of these genes
 - **sequencing in 283 genotypes** in each crops
 - Analysis of **allelic diversity** and **linkage disequilibrium**
 - initiate a **public dataset**

PARTNERS :

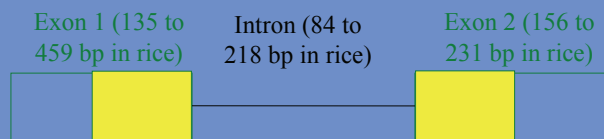


THE EXAMPLE OF THE ASR GENE FAMILY

1) Sequences available :

Family	species	both gDNA and cDNA	only cDNA
POACEAE	<i>Oryza sativa</i>	5	5
	<i>Sorghum Bicolor</i>	3	5
	<i>Hordeum vulgare</i>	0	3
	<i>Zea mays</i>	4	6
BRASSICACEAE	<i>Arabidopsis thaliana</i>	0	0
FABACEAE	<i>Phaseolus vulgare</i>	0	1
	<i>Cicer arietinum</i>	0	0
	<i>Glycine max</i>	0	2
	<i>Medicago truncatula</i>	1	1
SOLANACEAE	<i>Lotus japonica</i>	1	1
	<i>Solanum tuberosum</i>	3	4
EUPHORBIACEAE	<i>Lycopersicon esculentum</i>	3	4
	<i>Manihot esculenta</i>	0	2
SALICACEAE	<i>Hevea brasiliensis</i>	0	1
	<i>Populus trichocarpa</i>	2	2

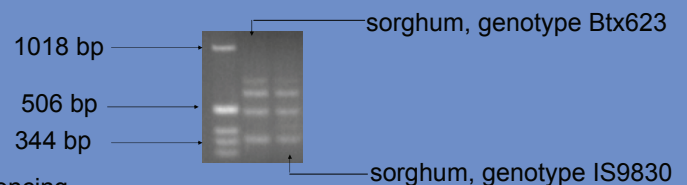
2) ASR gene structure :



Highly conserved domain : ABA/WDS (Pf02496)

Some **consensus primers pairs (degenerated in 3')** are designed for each crop cluster : forward primer in exon 1 and reverse primer in exon 2.

Example of a sorghum amplification with POACEAE consensus primers :



3) Picking these bands, reamplification with the same primers and sequencing

4) SNP identification (example of rice)



synonymous SNP, position 132

INDEL, position 216 to 219

non synonymous SNP, position 410



Acknowledgment : Generation Challenge Program (GCP, SP1)

