REPUBLIC OF TURKEY MINISTRY OF AGRICULTURE AND FORESTRY
GENERAL DIRECTORATE OF AGRICULTURAL RESEARCH AND POLICIES

Drought tolerance improvement in plants
An integrated view from breeding to genomics

Enes YAKIŞIR
August 10, 2020

Outline

1. Introduction
2. The increasing importance of drought tolerance in variable climates
3. Addressing the complexity of plant response to drought
4. Molecular dissection of drought tolerance
5. Modern breeding approaches for developing superior germplasm for drought tolerance
6. Future outlook

Introduction

Drought is the most devastating abiotic stress affecting crop productivity, which is caused by insufficient rainfall and/or altered precipitation patterns.

The seriousness of drought stress depends on its timing, duration and intensity.

Introduction

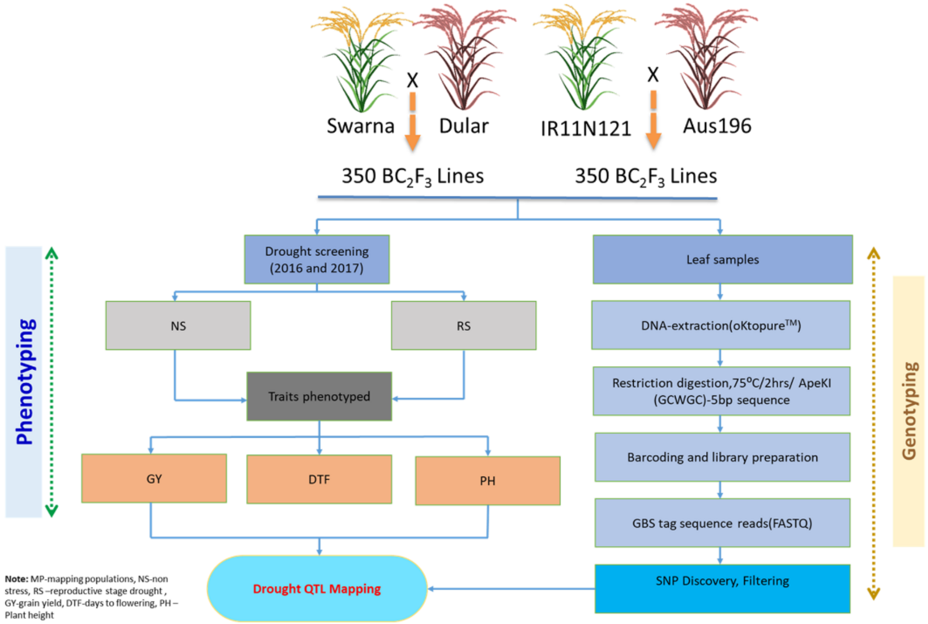
The impact of drought on crop production has been evidenced as early as the beginning of the seventeenth century, known as ‘‘Sahel drought’’, caused due to human intervention effects of deforestation, overgrazing and industrialization.

Increase in greenhouse emissions has resulted in altered precipitation, increase in arid land, desertification and finally reduction in crop productivity.

Introduction

Tolerance to drought is a complex quantitative trait controlled by several small effect genes or QTLs and is often confounded by differences in plants phenology.

To address the complexity of plant responses to drought, it is vital to understand the physiological and genetic basis of this response.



Swarna X Dular

IR11N121 X Aus196

350 BC_2F_3 Lines

350 BC_2F_3 Lines

Phenotyping

Drought screening (2016 and 2017)

NS

RS

Traits phenotyped

GY

DTF

PH

Genotyping

Leaf samples

DNA-extraction(oKtopure^{TM})

Restriction digestion,75^{0}C/2hrs/ ApeKI (GCWGC)-5bp sequence

Barcoding and library preparation

GBS tag sequence reads(FASTQ)

SNP Discovery, Filtering

Drought QTL Mapping

Note: MP-mapping populations, NS-non stress, RS –reproductive stage drought , GY-grain yield, DTF-days to flowering, PH – Plant height

Recent advances in crop physiology, systematic plant phenotyping and genomics have led to new insights in drought tolerance, thus providing crop breeders with greater knowledge of the gene networks and providing new tools for plant improvement to increase crop yield.

The increasing importance of drought tolerance in variable climates

The global water shortage caused by an increasing world population and worldwide climate change is considered as one of the major challenges facing agriculture today.

This shortage of water may threaten sustainable crop farming, since agricultural activities account for 75 % of global water consumption and irrigation consumes over 90 % of water used in many developing countries.

The increasing importance of drought tolerance in variable climates

It is also anticipated that by 2030, developing countries will be most severely affected by climate change because:

(a) climate change will have the greatest impact on the tropics and subtropics,

(b) most of the predicted population growth to 2030 will occur in developing countries and

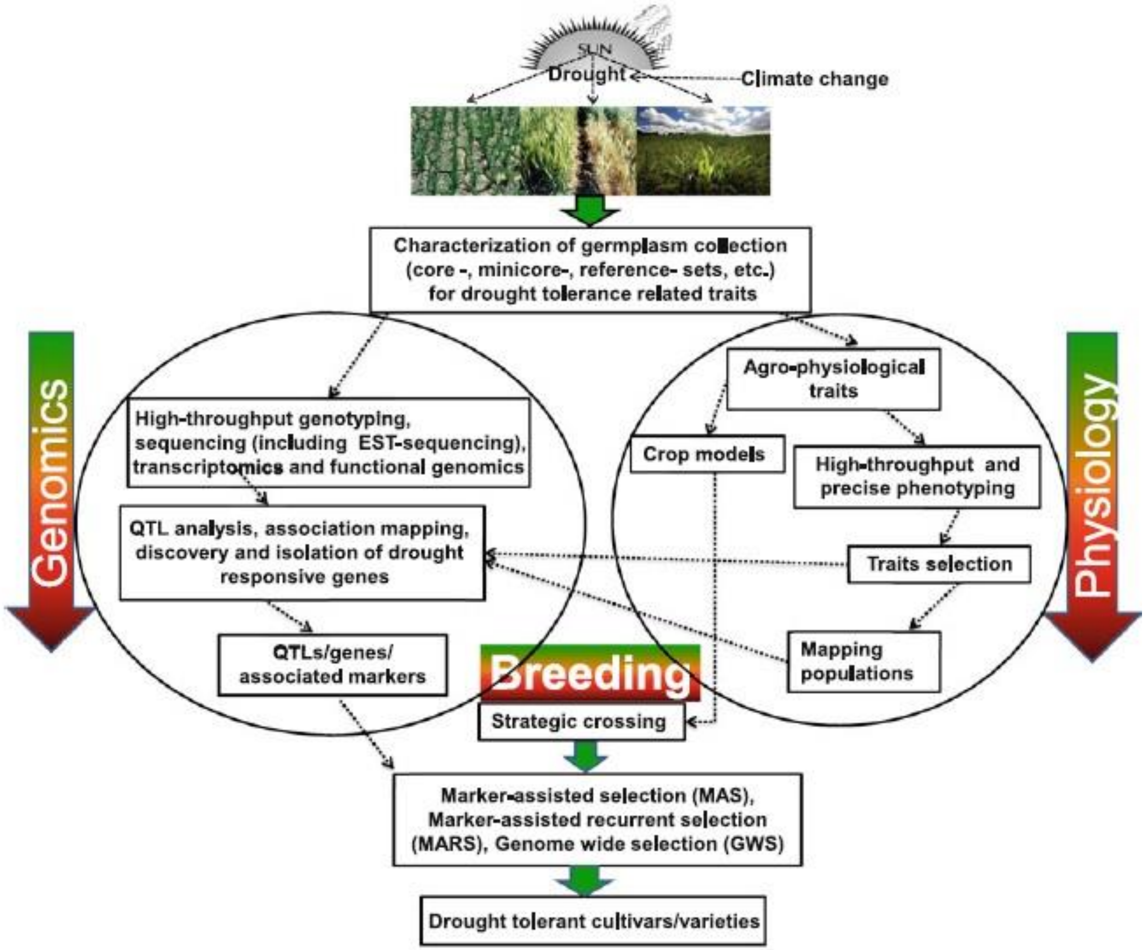
(c) more than half of the workforce in developing countries is involved in agriculture.

The increasing importance of drought tolerance in variable climates

This raises serious concerns and places huge responsibilities on the shoulders of scientists for developing ‘‘*drought-suited varieties*’’ through molecular breeding and genetically modified approaches.

However, it is clear that the demand to produce sufficient major food crops (wheat, rice and maize) for the growing population has always been increasing.

The increasing importance of drought tolerance in variable climates



Addressing the complexity of plant response to drought

Among the various abiotic stresses that curtail crop productivity, drought is the most recalcitrant to breeding, because plants use various mechanisms to cope with drought stress.

The physiological dissection of complex traits like drought is a first step to understand the genetic control of tolerance and will ultimately enhance the efficiency of molecular breeding strategies.

Developing and integrating a gene-to-phenotype concept in crop improvement requires particular attention to phenotyping and ecophysiological modelling, as well as the identification of stable candidate genomic regions through novel concepts of '*genetical genomics*'.

Addressing the complexity of plant response to drought

Criteria for using physiological traits in breeding programmes

The use of physiological traits in a breeding program, either by direct selection or through a surrogate such as molecular markers, depends on their relative genetic correlation with yield, extent of genetic variation, heritability and genotype x environment interactions.

For instance, in drought environments, osmotic adjustment, accumulation and remobilization of stem reserves, superior photosynthesis, heat- and desiccation-tolerant enzymes, etc. are important physiological traits.

The information on important physiological traits can be collected on potential parental lines involving screening of entire crossing block, or a set of commonly used parents, thus producing a catalogue of useful physiological traits.

However, if enough resources are available, screening for physiological traits could be applied to segregating generations in yield trials, or any intermediate stage, depending on when genetic gains from selection are optimal.

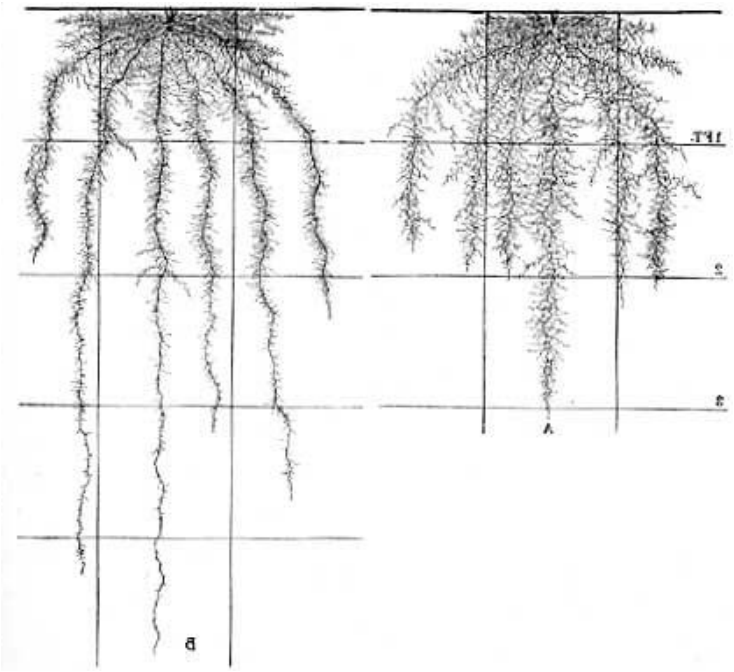
Addressing the complexity of plant response to drought

Conceptual framework for drought adaptation

The conceptual framework for yield drought adaptation by Passioura has three important drivers:

(1) water uptake (WU)
(2) water-use efficiency (WUE)
(3) harvest index (HI)

Addressing the complexity of plant response to drought



Crucial to target specific physiological mechanisms and to identify those traits most relevant to the patterns of drought stress found in the target environment.

For example, in crops grown with residual soil moisture that experience terminal drought, such as chickpea (*Cicer arietinum*), genotypes with deeper, more profuse roots have an advantage through better water extraction deeper in the soil profile.

In other crops also, deeper/profuse roots were found to increase plant access to water from deeper soil layers and support greater crop growth under drought conditions.

Molecular dissection of drought tolerance

In several genetic studies, drought tolerance has been found to be a complex quantitative trait controlled by a large number of minor genes/QTLs.

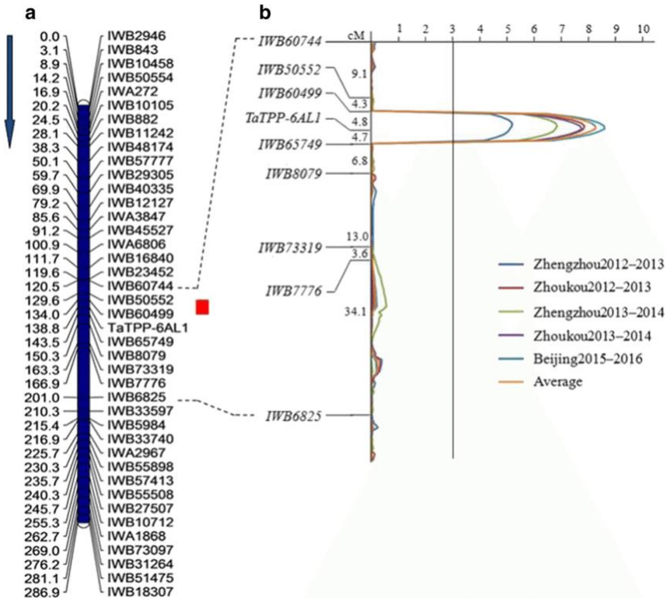
Recent advances in genome mapping and functional genomics technologies have provided powerful new tools for molecular dissection of drought tolerance.

The molecular markers and/or candidate genes identified provide a better understanding of the molecular basis of drought tolerance and, once validated, can be used in molecular breeding.

Molecular dissection of drought tolerance

Traditional QTL mapping involves:

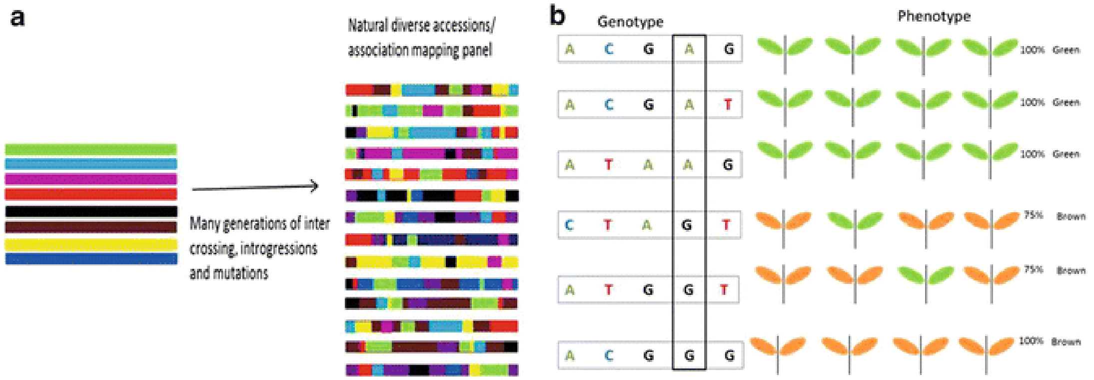
(1) development of mapping populations segregating for drought tolerancerelated traits
(2) identification of polymorphic markers,
(3) genotyping of the mapping populations with polymorphic markers,
(4) construction of genetic maps,
(5) precise phenotyping for drought tolerance-related traits, and
(6) QTL mapping using both genotypic and phenotypic data.



Molecular dissection of drought tolerance

The association mapping (AM) approach involves:

(1) selection of a diverse association panel/group of individuals from a natural population/ germplasm collection,
(2) precise recording of phenotypic data on the panel,
(3) candidate gene sequencing or highdensity marker genotyping of the panel,
(4) study of population structure (the level of genetic differentiation among groups within the selected population) and kinship (coefficient of relatedness between pairs of each individual within the population) and
(5) association analysis based on information gained through population structure, kinship, and correlation of phenotypic and genotypic/haplotypic data.



Molecular dissection of drought tolerance

QTLs for drought tolerance have been identified for several major and important crop species like rice, maize, wheat, barley, sorghum, pearl millet, soybean and chickpea.

These QTLs were identified for a variety of important traits including:

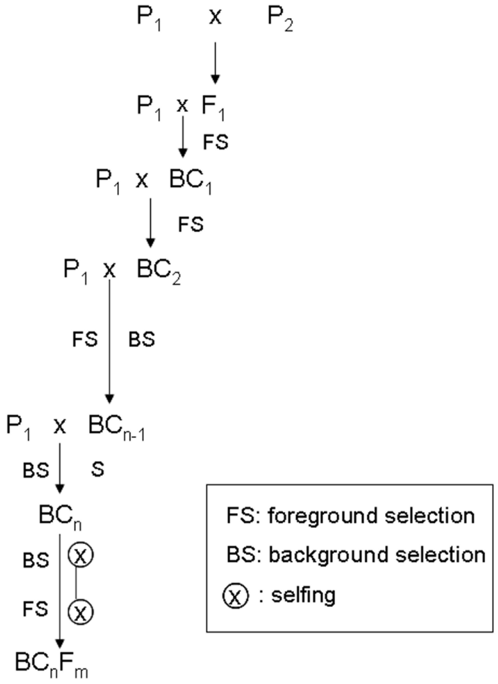
(1) yield and yield contributing traits under water-deficit conditions (in the case of wheat, maize, rice, soybean and pearl millet),
(2) physiological responses including water-soluble carbohydrates, carbon isotope ratio, osmotic potential, chlorophyll content, flag leaf rolling index, grain carbon isotope discrimination, relative water content, leaf osmotic potential, osmotic adjustment, chlorophyll and chlorophyll fluorescence parameters to drought stress (in the case of wheat, maize and rice),
(3) flowering time including anthesis to silking interval (in maize),
(4) root traits (rice, maize, wheat, soybean and chickpea),
(5) stay green (sorghum) and (6) nitrogen fixation (soybean).

Modern breeding approaches for developing superior germplasm for drought tolerance

Marker-assisted backcrossing (MABC)

When the QTLs identified for drought tolerance traits contribute higher phenotypic variation, they are considered major QTLs.

These QTLs, after validation in desired germplasm, can be used for introgressing drought tolerance from the donor genotypes (generally used for identification of the QTL for the trait) into elite, less drought-tolerant cultivars or breeding lines (recipient parents) without transfer of undesirable or deleterious genes from the donors (linkage drag).



FS: foreground selection
BS: background selection
ⓧ : selfing

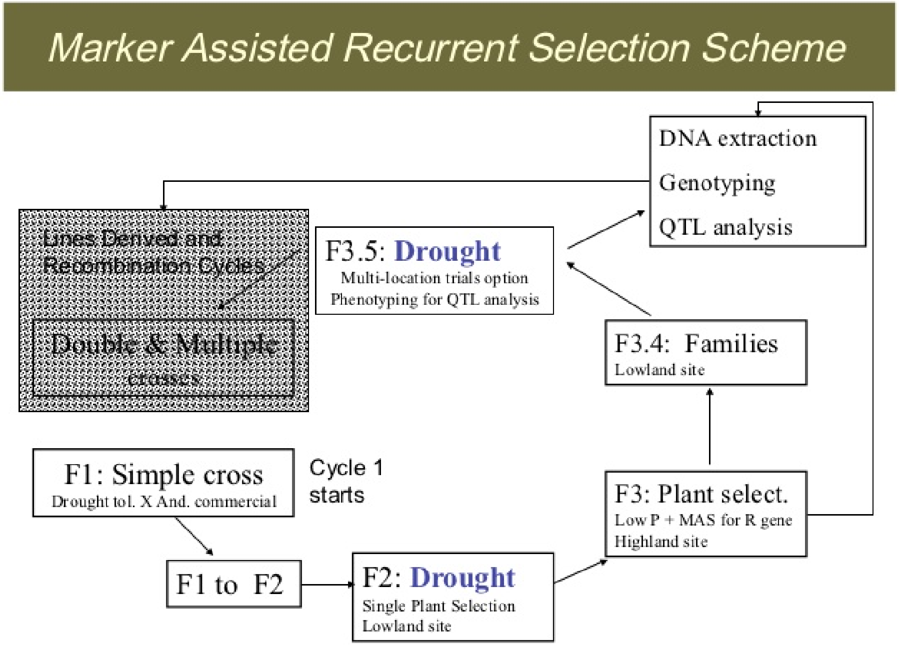
The process is commonly referred to as marker-assisted backcrossing (MABC).

Superior lines or cultivars are developed that contain only the major gene/ QTL from the donor parent, while retaining the whole genome of the recurrent parent.

Modern breeding approaches for developing superior germplasm for drought tolerance

Marker-assisted recurrent selection (MARS)

To overcome the limitations of MABC, particularly when multiple QTLs control the expression of a complex trait, the MARS approach, which involves intermating selected individuals in each selection cycle, has been recommended.



It generally involves the use of an F2 base population, and can be used in self-pollinated crops like wheat, barley and chickpea for developing pure lines with superior per se performance.

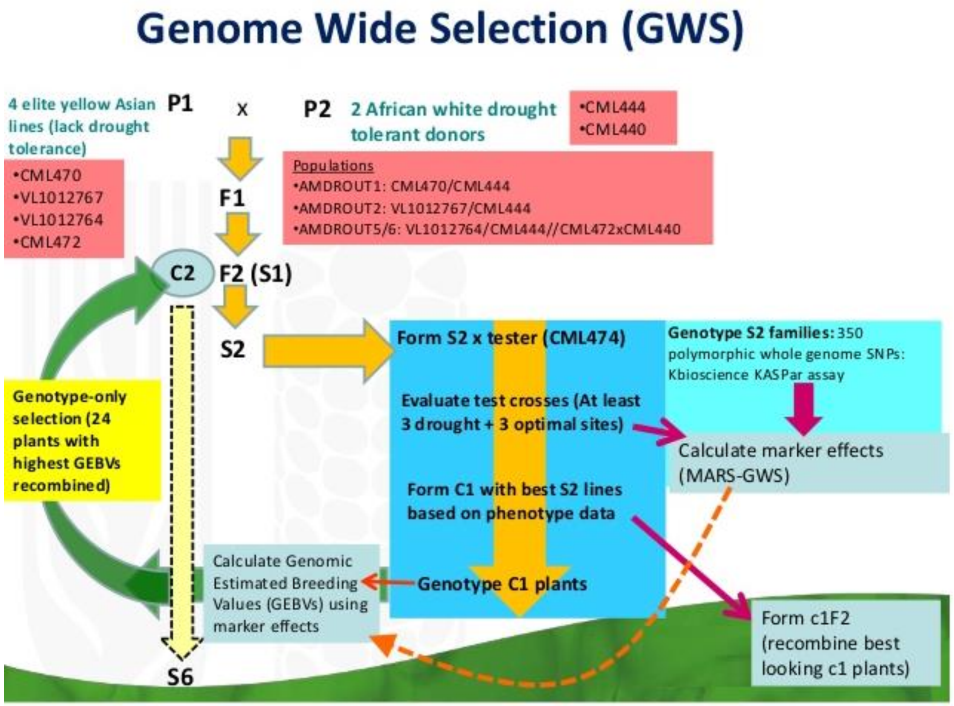
The successful use of MARS has been reported in sweet corn, sunflower and soybean.

Modern breeding approaches for developing superior germplasm for drought tolerance

Genome-wide selection (GWS)

Genome-wide selection (GWS) or genomic selection (GS) is another important approach to develop superior germplasm lines with overall excellent performance in a target environment.

Genome-wide marker genotyping is used for GWS rather than selected markers showing significant associations (as in case of MARS) with the traits of interest.



In summary, individuals in a phenotyped population (generally referred to as the 'training population') are genotyped using genome-wide markers and breeding values of alternative alleles of all the markers are fitted as random effects in a linear model.

Future outlook

It is evident that precise phenotyping is essential to screen larger core collection/mapping population for identifying the most appropriate QTL and candidate genes for use in plant breeding.

To meet the real-world challenge of increased crop production, the information available from functional genomics and systems biology needs to be integrated at the crop level; thereby, crop physiology will have a fundamental role in achieving this goal.

A new generation of crop models combined with systems biology studies should enable us to significantly narrow the gap between genes and complex phenotypes by predicting the field performance of crop genotypes.

Future outlook

Similarly, recent advances in genomics make it possible to not only conduct large-scale and high-throughput marker genotyping, but also sequence or re-sequence the genomes of germplasm collections, thus facilitating the identification of QTLs and candidate genes associated with drought tolerance.

While commonly used MABC has not been very effective in developing superior lines for drought tolerance, modern breeding approaches such as MARS and GWS are powerful tools for pyramiding multiple QTLs for drought tolerance or introgressing multiple complex traits such as heat tolerance in addition to drought tolerance.

Future outlook

In summary, it is essential to integrate crop physiology, genomics and breeding approaches to dissect complex drought tolerance traits, understand the molecular basis of drought tolerance and develop the next-generation crops for our changing climate.

Although work is ongoing in some major crops, it is anticipated that integrated physiology, genomics and breeding approaches will be initiated/ accelerated in the so-called orphan crops that are important for food security in many developing countries.

Thank you

enes.yakisir@tarimorman.gov.tr

REPUBLIC OF TURKEY MINISTRY OF AGRICULTURE AND FORESTRY

GENERAL DIRECTORATE OF AGRICULTURAL RESEARCH AND POLICIES