

WORKSHOP PROCEEDINGS

VALORISING THE DIVERSITY OF THE FIG TREE, AN ANCIENT FRUIT CROP FOR SUSTAINABLE MEDITERRANEAN AGRICULTURE: THE PRIMA PROJECT "FIGGEN"



05TH. JULY 2023

Auditorium of INTAEX (Avenida Adolfo
Suárez s/n) Badajoz (SPAIN)






PROGRAMME

Part I


Chairmen M. López-Corrales




09.00 - 09.15 Carmen González Ramos. Director of CICYTEX: Opening-Welcome




09.15 - 09.30 Prof. T. Giordani (University of Pisa): Presentation of the project "FIGGEN"




09.30 - 09.45 Dr G. Usai (University of Pisa): The haplotype phased genome of fig (Ficus carica L.): a crucial resource for fig breeding




09.45 - 10.00 Prof. M. López-Corrales (CICYTEX). Spanish germplasm bank and the different production systems developed in fig tree by CICYTEX




10.00 - 10.15 Prof. A. Kuden (Çukurova University). Phenotyping analysis of potted fig plants exposed to drought and salt stress and adult fig genotypes



10.15 - 10.30 Prof. G. Baraket (Faculty of Sciences of Tunis - UTM). The usefulness of the transdisciplinary approach for a sustainable commercial production of the fig tree in the Mediterranean region in relation to climate change.



10.30 - 10.45 Dr. Haffar Sahar (Faculty of Sciences of Tunis - UTM). Usefulness of molecular markers in the characterization of two spontaneous and cultivated compartments of fig trees in Tunisia.



10.45 - 11.00 Monserrat Pons i Boscana (Responsible for the fig germplasm bank of the Balearic Islands, Spain). Varieties autochthonous to the Balearic Islands: Field of Son Mut Nou.



11.00 - 11.30 Coffee-break and poster vision.







PROGRAMME

Part II


Chairmen M. López-Corrales




11.30 - 11.45 Dr. Paolo Belloni (Responsible for the fig germplasm bank "I giardini di Pomona" - Italy). The Fig Germplasm Collection at Pomona Gardens




11.45 - 12.00 Prof. Dr. Oğuzhan Çalışkan (Mustafa Kemal University). Caprifig (*Ficus carica* var. *caprificus*) genetic resources in the eastern Mediterranean region of Türkiye.




12.00 - 12.15 Dr. Olfa Saddoud Debbabi (National Bank of Genes - Tunisia). Conservation of the genetic resources of the fig tree in Tunisia




12.15 - 12.30 Francisco Balas Torres (Head of production and R&D FIKI Europe). FIGGEN relevance for companies: an experience from Extremadura.



12.30 - 12.45 Dr. Ghassan Zahid (Çukurova University): Comparative metabolome and transcriptome analysis of anthocyanin biosynthesis in fig (*Ficus carica* L.)



12.45 - 13.00 Dr. Fateh Aljane (Institute of Arid Regions of Medenine - Tunisia): Genetic Resources of the Fig Tree in Arid Zones (Tunisia): considerable potential, diversity and impacts of Climate Change



13.15 - 13.30 Dr. Arzu Ayar (Fig Research Institute - Turkey): Studies on the genetic diversity of female figs in the fig gene bank of Türkiye



13.30 - 14.00 Discussion and conclusions





PROGRAMME

POSTERS AND MORE INFORMATION

The workshop will be organised as a hybrid event (both on-site and on-line). The participation is free, but the registration is mandatory. The link to the webinar and the possible admission to participate on site, will be e-mailed all registered participants before the event.

REGISTRATION: [CLICK HERE TO REGISTER](#)

For more info:: margarita.lopez@juntaex.es

POSTERS

- 1 – Dr. Ana Isabel Galván. Mycotoxins and toxigenic fungi in dried figs from different farming systems.
- 2 – Dr. Ana Isabel Galván. Effect of temperature on growth and mycotoxin production of *Aspergillus section flavi* on dried figs in Extremadura.
- 3 – Agricultural Engineer Antonio Jesús Galán. Freezing as potential alternative to the application of phosphides for the control of insects during the storage of dried figs.
- 4 - Dr. Arzu Ayar (Fig Research Institute - Turkey): Some breba fig (*Ficus carica* L.) genotypes in Türkiye
- 5 - Dr. Muhammed Gündeşli (Gaziantep University): Fig selection studies in the Eastern Mediterranean Region

The FIGGEN project - Valorising the diversity of the fig tree, an ancient fruit crop for sustainable Mediterranean agriculture

FIGGEN is a three-year project promoted by PRIMA (Partnership for Research and Innovation in the Mediterranean Area) programme supported by the European Union. The ambition is to make the fig tree one of the most suitable and profitable crop in the Mediterranean area in a climate change context.

Among tree crops, the fig tree shows a good adaptation to dry, calcareous and saline environments, typical of different regions in the Mediterranean basin and the Middle East, where it has been cultivated for millennia. This crop has great potential for expansion thanks to valuable nutritional and nutraceutical characteristics and is particularly suitable for the application of sustainable agriculture based on biodiversity, such as mixed farming systems like agro-forestry.

Despite its importance, the fig tree has undergone low genetic improvement and most cultivation in the Mediterranean area is based on local cultivars that are currently highly threatened by genetic erosion due to various pests and diseases, abiotic stresses, intensive urbanization, monovarietal crops, migration from rural to urban areas.

FIGGEN wants to contribute breeding efforts to address crop tolerance to multiple abiotic stresses, improving productivity, efficiency and sustainability of agricultural farming systems.

Breeding depends on the collection; conservation and sharing of appropriate crop genetic resources among plant breeders and farmers. In this sense the project will create a participatory context involving main actors of the value chain following a transdisciplinary approach where socio-economic knowledge and recent scientific advances in assessing biodiversity will be combined with traditional knowledge of local private and public stakeholders.

FIGGEN aims to enhance the biodiversity of the fig tree and to select genotypes better adapted to environmental conditions coming from climate changes that can promote fig breeding and more sustainable fig production of the future.

www.figgen.eu





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ORAL PRESENTATIONS

Proceedings Workshop - Valorising the diversity of the fig tree, an ancient fruit crop for sustainable Mediterranean agriculture: the PRIMA project “FIGGEN” 6



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Prof. T. Giordani (Department of Agriculture, Food and Environment - University of Pisa - Italy)

Presentation of the project “FIGGEN”

FIGGEN - "Valorising the diversity of the fig tree, an ancient fruit crop for sustainable Mediterranean agriculture" is a project of PRIMA, the Partnership for Research and Innovation in the Mediterranean Area and is promoted by EU.

It started in 2020 and will end in March 2024.

The consortium is composed by 5 units: the Department of Agriculture, Food and Environment of University of Pisa from Italy that is coordinator, the Center For Scientific and Technological Research of Extremadura (CICYTEX) and the Institute for Mediterranean and Subtropical Horticulture "La Mayora" of the Spanish National Research Council (CSIC), from Spain, the Faculty of Science of the University of Tunis El Manar from Tunisia and the Department of Horticultura of Cukurova University from Turkey.

The objectives are:

- a) Exploring, valorising and evaluating fig cultivars from available Spanish, Tunisian and Turkish fig collections:
 - analysing genetic characteristics using a genotyping by sequencing approach
 - analysing morphological and physiological traits of fig cultivars to select plants with characteristics wanted by stakeholders and most suitable to be cultivated in drought and salinity conditions (more adapted to climate change)
- b) Identifying genetic traits linked to yield, fruit quality and drought and salinity adaptation that can be exploited in future breeding programs
- c) Disseminating project's products and results to stakeholders.

To achieve these objectives the project is structured in 5 WorkPackages. Wp0 Project coordination and management, WP1 Participatory assessment of the potential of fig cultivars in which periodic Living Labs with stakeholders are organised where researchers and participants exchange knowledge for the evaluation and selection of fig cultivars, following a transdisciplinary approach. WP2 Fig tree valorisation, characterization and selection, include genotyping and phenotyping analyses on 270 fig cultivars from Spanish, Turkish and Tunisian germplasm banks. A first product of these analyses is the identification of 20 cultivars tolerant to drought and salinity conditions and with traits expected by stakeholders. In WP3 Genome wide association studies (GWAS), data from these analyses will be combined to identify genetic characteristics associated with traits related to the fig production and quality. These specific genetic traits, after the end of the project, can be used for marker assisted selection and future breeding programs to develop new cultivars more adapted to climate change.

Finally WP4 is related to Dissemination, Exploitation, Communication and Outreach activities and has the objective to maximize the impacts of the project.





The team of Agricultural and Food Economics of our Department carried out a desk analysis of the market of the fig, focusing in the Mediterranean region to understand the importance of fig cultivation and production, critical issues, and development opportunities. In addition this team prepared guidelines with indications on Living Labs procedure, organization and reporting.

In spring 2021 and 2023 first two Living Labs were held in Spain, Tunisia, and Turkey. Among participants there were representatives of companies, farmers, plant growers, policy and administration, social associations, universities, research centres, consumer associations.

Following the selection of drought and salinity tolerant cultivars, a catalog with a description of 20 selected cultivars will be shown to the public during the third LL, scheduled for next autumn. The plants that best satisfy the expectations of the participants will be made accessible to them for validation in field trials (in real production facilities and contexts).

As concerns genotyping analysis, the team of Agricultural Genetics of our department produced a high-quality reference genome sequence of the fig tree combining the last published genome sequence with the latest methodologies of chromosome conformation capture. This allowed a deep characterization of the genome, with the identification and annotation of about 34 thousand protein coding genes. This reference sequence is a prerequisite for genotyping analyses on 270 fig cultivars that will be carried out following a Genotyping By Sequencing approach and for GWAS that are scheduled in the last part of the project.

As concerns phenotyping analyses, these have been performed both on adult plants, considering a number of plant and fruit traits, and on propagated plants in pots. These plants were grown in drought and salinity condition with the aim to select tolerant cultivars that could be more adapted to climate changes.

With regards to communication and dissemination activities, all partners contributed to publication of press releases, to the creation of a project logo, social media accounts and a project website. We participated to national and international conferences where our project has been presented and project results have been reported. Finally, up to now, we published 8 scientific papers in open access peer reviewed journals.





Usai G., Vangelisti A., Castellacci M., Simoni S., Mascagni F., Natali L., Cavallini A., Giordani T. (Department of Agriculture, Food and Environment, University of Pisa - Italy)

The haplotype phased genome of fig (*Ficus carica* L.): a crucial resource for fig breeding

The availability of genome sequences is crucial for the application of modern breeding techniques in crops. Understanding the genome's haplotypes is especially important for studying allele-specific expression and regulation patterns, particularly in fruit trees that maintain heterozygosity through clonal propagation. While the fig tree (*Ficus carica* L.) shows great potential for commercial expansion due to its nutritional properties and adaptability to challenging environmental conditions, high-quality genomic resources have only recently become available.

In this study, we present a haplotype-phased assembly of the fig tree genome using single-molecule, real-time sequencing and chromosome conformation capture. By integrating approximately 55x coverage of HiC reads with the existing genomic assembly, we generated two pseudo-haplotypes consisting of 538 sequences each, representing about 98% of the estimated 356 Mb fig genome. Furthermore, we assigned 400 out of 538 sequences (approximately 96% of both pseudo-haplotypes) to the corresponding 13 chromosomes, resulting in the new version 2.0 of the fig genome.

Using approaches including RNA-seq data, protein alignment, and *de novo* prediction, we predicted 33,954 and 33,379 protein-coding genes per pseudo-haplotype, respectively, with functional annotation for approximately 82% of the protein-coding genes. Synteny analysis identified 20,441 allelic gene pairs (about 65% of the total genes) that form the genetic map of the fig tree.

By analyzing structural variations within the pseudo-haplotypes, we discovered a total of 832,619 SNPs, 996,026 INDELs ranging from 1 to 50 bases, and 308 large structural variations longer than 50 bases, resulting in a total of 1,828,953 variations. Additionally, we calculated the intra-genomic diversity, estimated to be around 5.2 variations per kilobase.

The structural genomic variations were integrated with the positions of the 20,441 allelic gene pairs, considering different regions such as promoter regions, 5'-UTR, CDS, intronic regions, and 3'-UTR. Currently, we have results available for CDS, intronic, and 3'-UTR regions. The p-distance values for CDS regions range from a minimum of 0 (indicating identical sequences) to a maximum of 0.032. For intronic regions, the maximum value observed was 0.722, while for 3'-UTR regions, the maximum value was 0.464. In particular, intronic regions have shown the highest variability so far.





The fig genome sequence is being utilized to assess the genetic variability of fig varieties in Spanish, Tunisian, and Turkish collections. This analysis is conducted through genotyping by sequencing as part of the PRIMA project FIGGEN (Partnership for Research and Innovation in the Mediterranean Area). The objective is to perform genome-wide association studies (GWAS) to identify genes or molecular markers associated with fruit quality traits and adaptation to challenging environmental conditions influenced by climate change, ultimately leading to the genetic improvement of the fig species.





Margarita López-Corrales (Centro de Investigaciones Científicas y Tecnológicas de Extremadura - Spain)

Spanish germplasm bank and the different production systems developed in fig tree by CICYTEX

In Spain, the fig germplasm bank is located at the Centro de Investigaciones Científicas y Tecnológicas de Extremadura (CICYTEX) (Guadajira, Badajoz, Spain). At present, this bank has about 300 different varieties, 90% of which are varieties prospected in the Iberian Peninsula and the Canary and Balearic Islands. The planting frame is 5 x 5 m and 3 trees/variety with localized irrigation. Morphological characterization has been carried out using the UPOV TG165/1 descriptor and molecular characterization using nine microsatellite-type molecular markers. There is a high varietal diversity, with 31 varieties from different autonomous communities forming part of the nuclear collection. Balearic Islands are an ancient origin of domestication of fig even before the last glaciations, more genetically related with Eastern Mediterranean than the Iberian Peninsula.

Extremadura is the main region in terms of surface area and fig tree production in Spain. Traditionally, the fig tree has been grown under rainfed conditions and mainly for dry consumption with wide planting frames (10x10m or 12x 12m). Average yields range from 1 to 3 t/ha depending on soil type. In recent decades, CICYTEX has been researching this crop to position it as an alternative fruit crop due to the high demand for its high nutritional and functional quality fruits. First, it has identified the most interesting varieties for fresh consumption such as 'San Antonio', 'Dalmatie', 'Albacor', 'Negra Cabezuela' and 'De Rey'. For dry consumption, 'Calabacita' and 'Cuello Dama Blanco' stand out, the latter having a double aptitude. Then they studied the agronomic and fruit quality performance of the varieties both in intensive and super-intensive under localized irrigation conditions. Also, different training systems are being studied, both in vase and espalier with the use of meshes suspended and located on the ground.

In the case of varieties for dry consumption, intensive planting frames range between 5x5m and 6x6m. Trees are trained in high goblet and light annual pruning is required to eliminate dead branches and renew branches. average yields range between 7 and 7 t/ha in the eighth year of planting. The figs are harvested from the ground, placing the drip lines in the center of the lanes to avoid contact between the figs and the wet areas. Super-intensive dry fig plantations are only suitable for the Calabacita variety, at a 2 x 5 m frame (1000 plants/ha). The orientation of the tree line must be north-south to optimize illumination. In addition, a system of nets suspended about 50 cm above the ground has been implemented to facilitate fig harvesting and improve the hygienic and sanitary quality of the figs. To reduce the costs of this harvesting system, super-intensive plantations of fig trees on trellises at a 3 x





2.5m frame (1333 trees/ha) are being studied. Along the line of fig trees, an anti-weed net is placed, where the ripe figs fall and are collected with a fruit vacuum cleaner.

In the varieties for fresh consumption, in intensive systems the trees are trained in low vase. In this way, operators can harvest the fruit from the ground, with yields ranging between 17 and 30t/ha depending on the variety. To optimize harvesting and improve fruit illumination, the espalier training system has been studied with a planting frame of 4x 4m. In general, higher yields were obtained in the fifth year of planting and figs with higher soluble solids content.





Prof. Ayzin B. Küden (Çukurova University, Faculty of Agriculture, Department of Horticulture, Adana - Turkey)

Phenotyping analysis of potted fig plants exposed to drought and salt stress and adult fig genotypes

One of the aims of project FIGGEN is to characterize at phenotypic level 270 fig genotypes from germplasm banks or local crops located in Spain, Turkey, and Tunisia. In addition, another goal is to select 20 genotypes tolerant to drought and salt conditions and with traits of plant and fruit expected by stakeholders.

For these aims, propagated 18-months-old plants were used in three drought experiments set in Spain (CICYTEX), Tunisia (UTM), and Turkey (CU-TAGEM). Number of genotypes was 52 in Spain, 116 in Turkey and 110 in Tunisia. From these, the chlorophyll content, relative water content (RWC) and growth-related traits were collected.

Phenotypic analysis on propagated plant (water stress experiment)

In each country four propagated plants per genotype were irrigated normally according to water consumption (control plants), while other four plants were irrigated with 70% of water consumption (stressed plants). The experiments lasted about 50 days. During the experiment relative water content (RWC), leaf temperature, chlorophyll content (SPAD values) have been taken at three time points: at the start, at midpoint 1 (Mid-I, 15 days from the beginning of the trial, T15) and midpoint 2 (Mid-II, 30 days from the start of the trial, T30).

Under water stress conditions, one of the leaf responses is the stomatal closure that may increase leaf temperature due to the reduction of transpiration. So, leaf temperature is an indirect parameter related to water status, linked to transpiration process: plants are water stressed if temperature increases due to transpiration reduction. Growth parameters such as plant height and stem diameter, the total number of leaves were measured at the beginning and at the end of the experiment. Phenotypic data were processed following these steps:

Temperature. To obtain an indirect estimation of water status linked to transpiration, for each plant (replicate) temperature of fully expanded leaves have been managed as follow:

Leaf temperature of each replicate was collected at T15 and T30. Then the difference between leaf temperature minus air temperature has been calculated. For each replicate of control and stressed plants, the mean of these last values between T15 and T30 has been calculated following this equation: $[(T_{\text{leaf T15}} - T_{\text{air T15}}) + (T_{\text{leaf T30}} - T_{\text{air T30}})] / 2$.

SPAD. The SPAD value of fully expanded leaf from each replicate of control and stressed plants was collected at T15 and T30. Then mean, between SPAD values of T15 and T30 was calculated for each replicate.

RWC. Leaf samples (about 3 cm²) from a fully expanded leaf from each replicate of control and stressed plants were collected at T15. Plant water status was estimated by measuring relative water content: $RWC = (FW - DW) / (TW - DW)$, where FW, TW and DW denote fresh, turgid (water saturated) and dry weight, respectively.





Leaf number. Firstly, for each replicate we calculated the variation of the number of leaves between the end (day 50) and the beginning (day 0) of the experiment in control ($Lc_{50} - Lc_0$, ΔLc) and stressed plants ($Ls_{50} - Ls_0$, ΔLs).

The same calculations were also done for the **plant height** and **stem diameter**.

After this first data processing, for each trait, values from single replicate (four replicate for control and four replicates for treated plants) have been collected. After that, the median of the four replicates of control and the median of the four replicates of stressed plants were calculated.

Finally, to evaluate the variation between control and stressed plants, the difference between the two median values was calculated, obtaining a value for each genotype.

The final values for each of the six traits were sorted in descending order in the case of temperature, in ascending order for the remaining traits, thus obtaining six rankings where the tolerant genotypes were included in the first positions.

Phenotypic analysis on propagated plant (salt stress experiment)

The salinity test was carried out on 30 months old potted plants in June, July and August in 2022 in three experimental countries, Spain, Tunisia and Turkey.

Two different salinity levels were investigated in the experiment, which was carried out in four replications. The applications discussed in the study are given below.

-Application-1: Control plants (plants not exposed to salinity stress)

-Application-2: Plants subjected to salinity stress (irrigated with 100 millimolar NaCl).

The same parameters measured in drought experiment were recorded.

Through comparisons among genotypes rankings related to drought and salinity experiments. two genotypes from Spain, fifteen genotypes from Turkey and eleven genotypes from Tunisia were determined to be tolerant/resistant to both drought and salinity stresses.

Phenotypic analyses on adult plants

In summer 2021 also phenotypic analyses on mother plants of a part of 270 genotypes have been carried out in Spain (CICYTEX), Tunisia (UTM), Turkey (CU). Fifty-two genotypes from Spain, 105 female and 11 male (total 116) genotypes in Turkey have been studied.

Due to Covid pandemic restrictions, 60 out of 110 genotypes have been analysed in Tunisia.

For each genotype analysed, 26 phenotypic traits were evaluated.

The phenotypic analyses carried out in the three countries focusing mainly on parameters such as **weight, firmness, ostiole width, juiciness, harvesting date, maturation index, and resistance to crack**.

These parameters are among those suggested by stakeholders during the first Living lab organized in Spain, Tunisia and Turkey.

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Prof. Ghada Baraket (Department of Biology, Faculty of Science - University of Tunis El Manar - Tunisia)

The usefulness of the transdisciplinary approach for a sustainable commercial production of the fig tree in the Mediterranean region in relation to climate change

FIGGEN wants to contribute breeding efforts to address crop tolerance to multiple abiotic stresses, improving productivity, efficiency and sustainability of agricultural farming systems.

Breeding depends on the collection, conservation, characterization and sharing of appropriate crop genetic resources among plant breeders and farmers. In this sense the project will create a participatory context involving main actors of the value chain following a transdisciplinary approach where socio-economic knowledge and recent scientific advances in assessing biodiversity will be combined with traditional knowledge of local private and public stakeholders.

This participatory context is achieved through the organization of three periodic Living lab (LL) where scientific knowledge from experts in agricultural and food economics, tree physiology, agronomy, genetics, bioinformatics and ex situ conservation of germplasm are integrated with the knowledge from local stakeholders including breeders, nurseries, growers, consumers and marketing companies ensuring that FIGGEN outputs will be exploited and will have significant impacts in Mediterranean countries.

We adopted the Delphi method during our two Living Labs. It's a method for structuring a group communication process effectively to deal with complex problem. It's relies on a panel of experts to obtain the most reliable consensus of their opinions. The multistep process was adopted to perform this method and is implemented through different sequential rating rounds.

The first LL aiming to identify the most important functional trait that a selected cultivar should have. The process of phenotyping of first year bring to the development of a Draft catalog of selected fig cultivars tolerant to drought. This catalog was shown to stakeholders in the second LL. Based on the characteristics of the fruits and cultivars, the most important cultivars for Tunisian stakeholders were 'Zidi Kessra' and the caprifig 'Jrani'. For the Spanish stakeholders the most important cultivars were 'Clon 300', 'Canadria' and 'Panachée'. The Turkish stakeholders chose and gave the highest values to the cultivars 1008 YEŞİLGÜZ, 1005 ŞEKER İNCİRİ, 1029 SARILOP, 1045 MORGÜZ and 709 KIZIL MOR.

A final set of cultivars will be selected after phenotyping analyses on salt treated plants and a definitive catalog with drought and salinity tolerant genotypes will be developed. A third LL will be organized with stakeholders to evaluate selected cultivars and to gather further information on their potential and on the problems and opportunities for introducing them in





the value chain. The genotypes chosen will be made available of the users for evaluation in field trials.



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Sahar Haffar, Sana Ben Mustafa, Aymen Aounallah, Amel Hannachi Salhi and Ghada Baraket (Department of Biology, Faculty of Sciences of Tunis, University of Tunis El Manar, University Campus El Manar, Tunis - Tunisia)

Usefulness of molecular markers in the characterization of two spontaneous and cultivated compartments of fig trees in Tunisia

The genetic diversity of populations is a major factor influencing the evolution of species and their adaptation to the environment. Thus, the greater the diversity, the more adaptation to the environment is important. The measure of the genetic diversity within and between populations is crucial to guide methods of conservation of plants and their sustainable use for genetic improvement. Assessing the genetic diversity profile and genetic structure of populations of a species helps to determine its current status and risks, and can therefore serve as a basis for the development of acceptable scientific management policies and appropriate conservation initiatives.

It is therefore essential to conserve genetic diversity in order to strengthen the resilience of populations in the face of changing environments and to maintain a large genetic pool for future genetic improvements. Additionally, established molecular databases provide a large volume of information on a wide range of markers that have the potential to help identifying patterns of genetic diversity among plant populations and uncover the complexity of demographic and adaptive processes acting on natural populations. Therefore, the widespread availability of different molecular markers and the increase in computing power have favored the development of sophisticated methods and techniques that are beginning to meet these expectations.

In this context, CDDP (Conserved DNA derivative polymorphism) markers were designed starting from sequences of gene families present in multiple copies in the plant genome and targeted since they are mainly linked to biotic and abiotic stresses. The SCoT (Start Codon Targeted) tagging system based on the short conserved regions of plant genes flanking the ATG translation start codon was used in this study for a more complete understanding of the diversity available in the wild and cultivated Tunisian fig tree, for a better exploitation of the conserved germplasm and the definition of breeding strategies of the species *Ficus carica* L.

12 SCoT and 12 CDDP primers were used on 62 Tunisian fig trees genotypes. These primers revealed 317 SCoT-CDDP markers (92,96 %) that were found to be polymorphic. The diversity parameters showed the efficiency of the SCoT-CDDP primers and the high level of polymorphism detected in the two compartments of fig trees studied. A high number of effective migrants was detected ($N_m = 6,052$) reflecting the significant gene flow recorded between cultivated and wild fig trees. Indeed, the value of (G_{st}) recorded is 0,076 and AMOVA revealed significant differences in the distribution of perceived genetic diversity rather within groups (89%) than between groups (11%) of fig trees studied. The combined dendrogram displays three main groups. The topology of the tree obtained from the combined data of the





two markers analyzed suggests the presence of a diversity that operates according to the sex (common fig or caprifig) and the type (wild or cultivated) of the fig tree and independently of the geographical origins and the horticultural classification of the fig trees considered. The results suggest that CDDP and SCoT markers can be used as reliable and informative markers to detect polymorphism levels and draw genetic links in the species *Ficus carica* L.





Montserrat Pons i Boscana (Responsible for the fig germplasm bank of the Balearic Islands - Spain).

Varieties autochthonous to the Balearic Islands: Field of Son Mut Nou

Conservation of genetic resources.

In the trial farm of Son Mut Nou, Lluçmajor, Mallorca, Balearic Islands, the main objective is to collect and keep together a large collection of figs of traditional varieties, in order to carry out an easy and rigorous research, characterization and valorisation.

Further, in Son Mut Nou, the majority of the more important fig tree varieties from all the collections in the world have been added. Nowadays, the main field is distributed in 12 parts, with 2836 fig trees, of 1237 different varieties. From them, 242 are traditional from Balearic Islands, 175 from Spain, 506 from Europe, 120 from America, 104 from Asia, 35 from Africa, 7 from Oceania, 30 caprifig trees, and 18 remarkable trees.

| TYPE | Place of origin | N. of varieties | Synonyms | Total |
|---------------------|------------------------|------------------------|-----------------|--------------|
| Fig | Balearic Island | 242 | 119 | 361 |
| Fig | Spain | 175 | 6 | 181 |
| Fig | Europe | 506 | 261 | 767 |
| Fig | America | 120 | 40 | 160 |
| Fig | Asia | 104 | 5 | 109 |
| Fig | Africa | 35 | 1 | 36 |
| Fig | Oceania | 7 | 1 | 8 |
| Caprifig | | 30 | | 30 |
| Remarkable fig tree | | 18 | | 18 |
| TOTAL | | 1237 | 433 | 1658 |

It has been carried out a characterization both at molecular and morphological level, and have been determined the principal agronomical features of every single variety, especially of those ones which are in genetic erosion risk.





Traditional variegated varieties of fig in the Balearic Islands

Among the hundreds varieties of fig tree in the world, there are only one or two dozen with a very special and beautiful characteristic: they are variegated. The fruit of these cultivars alternates normal color stripes and lighter or white stripes. It is also always manifested in young shoots, and occasionally in leaves.

This phenomenon is being studied to determine and understand the causes that provoke it. It is suspected that may be induced by a disease virus-like depigmentation resulting in the absence of chloroplasts in the stripes.

In the Balearic Islands variegated varieties are known by the adjective "rimadas", and there are 5 traditional cultivars, two black-skinned figs (Martineca rimada and Bordissot negra rimada) and three green-skinned (Bordissot blanca rimada, Paratjal rimada and Coll de dama rimada).





Dr. Paolo Belloni (Responsible for the fig germplasm bank "I giardini di Pomona" - Italy)

The Fig Germplasm Collection at Pomona Garden

I'm Paolo Belloni,

30 years ago I founded "I Giardini di Pomona" - the national association for the enhancement of biodiversity. We have always dealt with the recovery and conservation of traditional fruit varieties

Pomona was the Latin goddess who protected gardens and orchards.

"I Giardini di Pomona" is a private structure founded through tourist hospitality, guided tours and the sale of products processed on the farm.

"I Giardini di Pomona" covers an area of about 10 ha.

Pomona APS (Social Promotion Association) has three priority purposes:

- To pass on to future generations this enormous heritage of cultivated plants selected over the millennia by hundreds of generations of farmers.
- the careful use of water by the restoration of the whole system of cisterns present in the various camps.
- to renaturalize soils that are heading towards desertification.

Over 1000 varieties of ancient fruit are preserved here. The varieties which at list date to 1950 include, among the others, 600 cultivar of figs and about 150 of pomegranates by the Cossio collection.

«I Giardini di Pomona» collaborates with the network of Apulian permaculturists. Our food forests integrate the conservation of biodiversity with the principles of a saving use of water.

The Pomona APS has enhanced the school food education projects of the Region Lombardia.

Continuous attention is devoted to environmental education through guided tours, Erasmus projects and workshops for the little ones.

The Pomona APS is well known for the pomological exhibitions on ancient fruit, created to show the richness of the Italian botanical heritage and the risks associated with its disappearance.

For 20 years (from 1993 to 2013) the Pomona APS presents Italian fruit varieties from all over the peninsula at the international exhibition organized by the "Croqueurs de pommes" in France which is held every 5 years.

But, why FIGS?

Because they are delicious fruit, healthy, highly energetic, easy to dry and preserve and with great versatility in cooking.

The good adaptability to climatic changes and good resistance to heat (+45°/50°) and cold: from -14° to -20° depending on the cultivar.





the adaptability to cultivation in dry farming and the resistance to saltiness and brackish air. This makes cultivable many areas immediately facing the sea, not usable with other crops of fruit trees;

the ability, in biferous cultivars, to produce two different fruits at different times on the same tree. Unique characteristic among fruit trees;

it is a tree both easy to cultivate and to reproduce, even by those with no agronomic skills;

It shows an extraordinary variability of the fruit in terms of skin and pulp color, shape and size of the figs and allows you to enjoy the very extensive and diversified range of flavors of each variety.

We intend to enhance all parts of the plant: buds, leaves, latex, roots;

For example, we produce an alcohol-glyceric extract from buds and one-year-old rootlets to create a food supplement for the natural protection of the stomach and intestines.

We are interested in participating in all the projects for the valorization of the *Ficus carica* species and have created two lines of products dedicated to health and beauty.



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Oguzhan Caliskan¹, Safder Bayazit¹, Muruvvet Ilgin², Nesrin Karatas³, Derya Kilic¹

Caprifig (*Ficus carica* var. *caprificus*) genetic resources in the eastern Mediterranean region of Turkey

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Fig (*Ficus carica* L.) culture has been widely practiced in Mediterranean countries since ancient times. Turkey is one of the important fig germplasm centers. The coastal areas of the Aegean and Mediterranean regions of Turkey, which have mainly Mediterranean climate, are very suitable for fig culture. Several earlier studies have reported the morphological and genetic diversity of edible fig genotypes and endorsed the use of reproducible morphological parameters to characterize genotypes. However, studies on the caprifig germplasm and its potential use in caprification and breeding have been limited. We have started several projects since 2014 to determine the morphological, pollinator, and genetic diversity of six standard caprifig cultivars (from the Aegean region) and 90 caprifig genotypes selected from the Eastern Mediterranean region of Turkey. Our results showed that profichi fruits and their pollen morphology can be used to identify caprifigs, select caprifigs for caprification, and determine the paternal individuals for edible fig-breeding programs. Currently, our studies are continuing to develop new fig individuals for different breeding purposes using selected caprifig genotypes.

The genus *Ficus* includes more than 800 species, with most found in the tropics or subtropics and only a handful with fruits considered edible. All of the edible cultivated figs belong to the species *Ficus carica* L. The profichi crop of caprifigs (*Ficus carica* var. *caprificus*) in this species is very important for edible figs. This crop has to use in caprification processing for fruit set and yield in the main crop of edible figs (caduceus group) deciduous. Turkey is the major fig producer and exporter in the world and there are two main cultivars, 'Bursa Siyahı' and 'Sarılöp'. Since these cultivars are in the caducous group, caprification is applied. However, caprifigs can affect the fruit size, fruit skin and flesh color, total soluble solids, harvesting period, phytochemical profiles, and some aroma compounds in edible figs (Condit, 1947; Gaaliche et al., 2011). Therefore, it is necessary to determine the quality and productive caprifigs.

Several earlier studies have reported the morphological diversity of edible fig genotypes and endorsed the use of reproducible morphological parameters in the characterization of the genotypes (Caliskan et al., 2023). Unfortunately, to date, there has been little research into the genetic resources available for caprifig germplasm (Khadivi-Khub and Anjam, 2014).





Total of 90 caprifig genotypes were selected by [Caliskan et al. \(2017\)](#) and six caprifig cultivars ('Ak İlek', 'Armut İlek', 'Elma İlek', 'Hamza', 'Küçük Konkur', and 'Taşlık') were used to compare the genotypes. A total of 45 morpho-pomological characters and 13 pollen traits were investigated ([Caliskan et al., 2021](#)). In addition, 15 SSR primers were used for the genetic characterization of the caprifig germplasm ([Caliskan et al., 2018](#)).

The data showed that fruit size, fruit number per shoot, the time of *Blastophaga* wasps exit from caprifig fruits, ripening period, coinciding with the female flowers in edible figs, amount of gall flower and male flowers, amount of pollen production, pollen viability and germination percentage, the inclusion of the mammoni and mamme crops, and free from disease and pests were very critical characteristics in the identifying caprifig resources. In addition, the fruit skin color of caprifigs was not as rich as that of edible figs and 90% of the genotypes were green in color. There were a small number of black, purple, and brown skin-colored genotypes. The internal pulp color of caprifigs is most often white, whereas a color distribution ranging from dark purple to light purple was observed in the fruit pulp color. In addition, Mersin06 and Osmaniye02 were determined as persistent genotypes for the first time in Turkey fig genetic resources. Parthenocarpic edible fig breeding studies were started with these genotypes

The polar length, equatorial diameter, pollen shape, number of porates, porate width, exine thickness, and abnormal pollen ratio were important variables in discriminating the caprifigs germplasm. Therefore, these traits can also be used to identify caprifigs.

Genetic distance values and cluster analyses revealed high genetic similarities, except for the standard cultivars, among the caprifig groups (Adana, Mersin, Hatay, Osmaniye, and Kahramanmaraş). In addition, factorial correspondence analysis separated the caprifig groups, suggesting that caprifig populations from Turkey were unmixed, probably because of low gene flow, likely because germplasm has not yet been moved among geographical areas and because many caprifig populations have grown from propagation by seed. In our population structure analysis, the caprifig accessions could be grouped according to the regions from where they were sampled. Our molecular data revealed great genetic diversity within this caprifig germplasm. Besides, presented here support the efficiency of microsatellite markers for both the description of genetic diversity and management of caprifig germplasm.

These results revealed the great genetic variation available in caprifig germplasm resources from the eastern Mediterranean region of Turkey. The present study provides essential information to design a caprifig germplasm collection without duplication of plant material, to sustainably manage fig breeding programs, and to establish strategies for conserving caprifig genetic resources.

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Conservation of the genetic resources of the fig tree in Tunisia

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Plant genetic resources are very important for food security. Moreover, they are part of the history and identity of Tunisia, which occupies a strategic geographical position in the Mediterranean and North Africa. Aware of this importance, Tunisia has adopted the international undertaking (1983), the convention of biological diversity (1992), the international treaty (2001) and the Nagoya Protocol (2010).

National Genebank of Tunisia has been established for the conservation of plant, animal and micro-organisms genetic resources. Fruit trees genetic resources are very diversified and represented by several species in Tunisia.

Fig (*Ficus carica* L.) is very old cultivated a fruit tree species. It is diversified and it is represented by more than 100 cultivars. Several studies have been interested last decades to identification and characterization of this germplasm. Several collections have been established in order to conserve this species *ex situ*. Recently, in 2014, National Genebank of Tunisia (NGBT) has settled a national fruit tree field genebank in Takelsa in North Est of the country, according to international genebanks standards.

Accessions have been collected during collecting missions in several regions of Tunisia. Each accession has a passport data with all the information requested for genebank. It has been characterized using morphological traits according to International biodiversity descriptors for fig. Molecular markers have been used in order to identify cultivars, to resolve problems of homonymy and synonymy and to study genetic diversity of accessions collected. SSR markers have been usually the most suitable markers for this purpose.

Endangered accessions have been propagated by grafting to be *ex situ* conserved. Some accessions requested from farmers, NGOs and researchers have been shared.

Moreover, NGBT is in contact with national stakeholders to establish a strategy for conservation of this species. It is involved in actions of implementation of labels for fig cultivars such as PDO (Product of Denomination of Origin) and PGI (Product of Geographical Indications). It has organized several actions for capacity building and for strengthening awareness to conserve fruit trees genetic resources. For a better conservation of fig germplasm of Tunisia it is crucial to settle a national data base clustering all the data from several institutions. A national fig collection needs to be enriched with fig cultivars grouping the major part of Tunisian fig diversity.





Francisco Balas Torres (Head of production and R&D FIKI Europe - Spain)

FIGGEN relevance for companies: an experience from Extremadura

The cultivation of the fig tree is of great socioeconomic importance in Extremadura. FIKI EUROPA SL is a company dedicated to the cultivation and marketing of figs in different formats with extensive experience in public-private cooperation frameworks and a significant investment in R&D. We consider that there are three challenge areas in fig production and marketing: the challenges of the agri-food chain, those of climate change, and regulatory challenges.

Among the challenges of the agri-food chain, it is worth highlighting the short shelf life of figs, since they are highly perishable fruits and push the whole logistics chain (producer, transport, wholesaler, retailer, consumer) to be very fast and very efficient in order to avoid losses. It is also needed to adjust ripening dates to the best sales dates, as well as the shape and colour of the fruits, which may vary from one market to another. In general terms, figs are required to be sweet and tasty and are increasingly required to have nutritional benefits such as vitamins, minerals and fiber.

The challenges derived from the environment and more specifically from the climate change scenario are especially complicated. Our production area is located in the province of Badajoz, within the Extremadura region, where we have a continental Mediterranean climate with a slight Atlantic influence that is characterized by very hot and very dry summers that coincide with the fig harvest season. When spring or fall is drier than normal, drought problems often manifest. In last decades, a general rise in temperatures has been observed, as well as a decrease in rainfall. This makes us fear more and more frequent episodes of drought at harvest time with the consequent effects such as anomalies in the reproductive cycles of plants, severe drought, frost, lack of water for irrigation, soil degradation and the appearance of new pests. and diseases.

Regarding regulatory challenges, but also socioeconomic, all producers in Europe must be prepared for the new EU Green Deal regulatory framework, which will affect all areas of activity of agricultural producers and which are summarized in the search for sustainability, energy efficiency and zero waste in order to mitigate the effects of climate change in the coming decades. In addition, a high level of food safety, traceability and certifications will be required. In some areas of Europe we face a very significant demographic challenge, with the population leaving the rural areas in search of new opportunities, generating a lack of manpower. Finally, the cultivation of the fig tree suffers from a general shortage of registered inputs to manage orchards.

The challenges that emanates from these three areas gives us an idea of how the perfect fig should look like in order to be optimally produced and marketed. Some of these characters are: extended shelf life, high yield, parthenocarpic, nice colour, fitting shape, no cavity, tight





ostiole, drying ability, juiciness, aroma, sweetness, salinity tolerance, drought resistance, pest resistance, disease resistance, low input requirements, suited to new cultivation systems, adaptation to new harvesting methodologies. Therefore, new and more and better adapted varieties are needed. To achieve these objectives, breeding is the most appropriate tool, since it is the engine of innovation throughout all production and marketing processes. The FIGGEN project has addressed some of these challenges by identifying genotypes that meet these criteria, thus being an aid to producers and breeders for the future. There is still a lot of work to be done, but initiatives like FIGGEN have a positive impact on the sector.



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Ghassan Zahid, Ayzin Küden and Yıldız Aka Kaçar (Çukurova University - Turkey)

Comparative Metabolome and Transcriptome Analysis of Anthocyanin Biosynthesis in Fig (*Ficus carica* L.).

Ficus carica L., also known as the common fig, is a species of flowering plant in the Moraceae family. Fig trees are well adapted to the Mediterranean climate and have a long history of cultivation in countries such as Türkiye. In addition to being an important food source, figs and fig trees also have cultural and economic significance in Türkiye. Additionally, they contain a significant amount of phenolic compounds (PCs), sugars, and antioxidants, all of which are important for preventing heart and metabolic diseases, microbial infections, obesity, diabetes, and cancer in humans. These PCs are also renowned for their antioxidant capacity, particularly anthocyanins, a type of pigment that is found in a wide range of plant-based foods, including fruits, vegetables, and grains. They are responsible for the red, blue, and purple colors of many foods, and are part of the class of compounds known as flavonoids. Anthocyanins have been the subject of numerous studies due to their potential health benefits.

Turkey is the world's leading producer of figs and recognizes this fruit as a key part of its cuisine. However, despite their diversity and rich medicinal value, local figs are overlooked and poorly examined for the biochemical fingerprints associated with their nutritional quality, and little information is available on their molecular structure beyond fig anthocyanin biosynthesis and coloration. Therefore, this study was first designed to analyze the peels and pulps of eight Turkish figs for their total phenolic composition, sugars, and antioxidant capacity. Thirty-one phenolic compounds were detected using High-Performance Liquid Chromatography coupled with Mass Spectrometry (HPLC-MS/MS) method. Among the peels and pulps, the highest mean value of colored and colorless phenolic compounds was found in the '01-1N-58' (dark violet-purple) genotype. Therefore, based on the results, the peels of the '01-1N-58' genotype and the peels of the 'Sarilop' cultivar were selected for transcriptomic analysis. For this, RNA-Seq analysis was performed on the peels of the dark-purple '01-1N-58' genotype and greenish-yellowish 'Sarilop' cultivar using Illumina NovaSeq 6000™. Based on these genes, gene ontology (GO) analysis was performed. A total of 21102 differentially expressed genes were observed, and 3542 up-regulated and 3164 down-regulated genes in the libraries of two fig samples were identified. Additionally, qRT-PCR analyses were also conducted to validate the results of the expression of genes in RNA-Seq. qRT-PCR analysis was also conducted to validate the results of the expression of genes in RNA-seq. Most significantly, our study indicated that the rich bioactive compounds in peels and high variability among eight fig cultivars and genotypes might allow better use of various fig fruit resources when consumed with dark-colored peels for maximum health benefits.





Dr Fateh Aljane (Drylands and Oases Cropping Laboratory, Institute of Arid Regions (IRA) - Médenine - University of Gabès - Tunisia)

Fig Genetic Resources in Tunisian Arid Regions: A Considerable Potentiality, Diversity and Impacts of Climate Changes

In Tunisia, Fig (*Ficus carica* L.) is a traditional fruit trees and is well adapted to arid and semi-arid climates. They have an important ecological and socio-economic role and can valorize marginal soils. A high number of local varieties characterizes fig and the denominations are usually based on the color, size and time of fruit ripening or geographical origin resulting in denominations confusion (synonymies and homonymies).

In Tunisia, female fig is characterized by a frequent exchange of varieties among different regions to compare to others fruit trees but a less effort has been made for the propagation and cultivation of male (caprifig) varieties. The selection of fig varieties is often based on the appreciation of farmers, the production of the Breba crop/ main crop, the productivity, the pomological characteristics of fresh and dried figs. Concerning the pollinator, the main selection criteria are the abundance production of caprifigs (profichis), the maturity date, the abundance of *Blastophaga psenes* and pollen viability. Fig varieties identification is generally based on phenotypic and morphological characters (fruit shape, fruit color, fruit dimensions, fruit weight, ostiole diameter), agronomic parameters (Plasticity and adaptation of the variety, resistance to biotic and abiotic factors) and biochemical composition (acidity, sugars, polyphenols, *etc.*). Nevertheless, this approaches influenced by environmental conditions and a limited number of characters. However, the molecular (RAPD, SSR, SNP markers) characterization of fig varieties was developed. These molecular information of fig genetic resources will be used to guide the ex situ conservation measures, selection and to determinate characters of interest such as yield, quality, disease resistance and abiotic stress tolerance.

The management of fig genetic resources raises many challenges and constraints, namely: the selection, conservation (the ex situ an in situ conservation are the challenges and is being carried out in cooperation with Non- Governmental Organisations (NGOs) and farmers), propagation, field management, phytosanitary protection and the exchange of plant material within the regions.

The risk of transfer of diseases and pests (Fig Mosaic Disease) and the emergents pests (*Lonchaea aristella*, *Hypocryphalus scabricollis*) perhaps the major challenges of the exchange of plant material between agrosystems.

Climate changes have an effect of on fig trees, namely a very difficult climate conditions, drought on figs plantation (trees that are dried out) and a highly endangered fig germplasm. Furthermore, climate changes undergone by the arid regions, very vulnerable affect agrosystems and their genetic resources and risk causing irreparable damage to all their components: Date palm, fruit trees, vegetable and other crops. The depletion of the fruit trees





genetic resources in arid areas, especially fig has a direct effect on the sustainability of arid agrosystems.

For the adaptation of arid regions to the effects of climate changes, we must respect agrosystems in terms of plant and animal biodiversity (Resistant species, diversification of income, *etc.*), monitoring of the species and varieties introduced, restoration of plant biodiversity and supervision of economic and social activities in arid areas.



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Dr. Arzu Ayar (Fig Research Institute, İncirliova/Aydın - Turkey)

Studies on the genetic diversity of female figs in the fig gene bank of Turkey

According to FAO (2022) data, Fig production is carried out in an area of 281.522 hectares in the World. 19% of the world's fig production areas belong to Turkey. World fig production is 1.264.943 tons. Turkey ranks first between world with 320 thousand tons of fig production. Türkiye ranks first in dried fig exports with 69%. Export income is \$293.372.

In this study, first the presence of figs in Turkey (establishment of fig field gene bank, number of genotypes and collection areas) was emphasized. Some information about fig descriptors (Tree, fruit and leaf growth) and finally other applications in the fig field gene bank has been given.

The fig tree (*Ficus carica* L.) gene source is very important for Turkey. Conservation, enrichment and evaluation of female genotypes collection in the fig field gene bank is one of the most important duties of the Fig Research Institute Directorate.

The female fig field gene bank in Turkey began to be established in the 1970s. The collection is enriched by studies such as selection and collection carried out in the fig growing regions of Turkey. Nowadays, the number of varieties and types is 292. 98% of female figs originate from Turkey. 11% of this is registered variety. The most collecting region of the genotypes is the Aegean Region with 36 %. On the other hand, Marmara and Mediterranean regions are in the 2nd place with 22 %.

Female genotypes have been evaluated according to 112 criteria (IPGRI and CHEAM, 2003). These are; Tree growth habit, leaf and fruit growth criteria and phenological observations.

Tree growth habit was evaluated in 5 groups as erect, semi erect, open, spreading and weeping.

From fruit quality criteria; fruit inner colour, fruit shape and symmetry, flesh thickness, colour of liquid drop around ostiole, amount and size of fruitlets, ostiole opening, colour and adhesion of scales around the ostiole opening, lenticels quantity and shape etc. criteria differed between genotypes.

All female fig genotypes in the fig field gene bank were photographed and recorded. A digital archive of the examined IPGRI data of all genotypes was created. These records are an important step in transmitting the genetic resources to future generations and to be resources /literatures for different breeding studies.

Taking multiple data can better reveal the similarity-difference status of a genetic population. Multiple data collection studies of the genotypes continue in the female fig field gene bank. These studies within the scope of the "Fig (*Ficus carica* L.) Genetic Resources Conservation and Characterization Project" (continuous project) are supported by the General Directorate of Agricultural Policy.





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Mycotoxins and toxigenic fungi in dried figs from different farming systems

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The common fig tree (*Ficus carica* L.) is a typical species of the Middle East and Mediterranean Region since it is well-adapted to high temperatures and low water regimes. Its cultivation has been traditionally subsidiary to other fruit-bearing or herbaceous plants so not too much effort has been done to improve harvest of fig fruits which is mainly based on traditional collecting of ripened fruits (≈ 30 % moisture content) from the soil once they drop from trees. This makes dried figs fruits highly vulnerable to contamination with filamentous fungi. Some of them, if the environmental conditions are propitious, may produce mycotoxins, mainly aflatoxins and ochratoxin A. So there is a need to evaluate the effect of different farming practices and drying on toxigenic fungi development and further mycotoxin production to ensure quality and safety of dried figs. The aim of this study was to analyse mycotoxins and toxigenic fungi of different samples of dried figs of the variety 'Calabacita' collected from two fields from Extremadura (Spain) with different farming systems (rainfed and irrigated practices) before (30% m.c.) and after their drying ($<24\%$ m.c) in a greenhouse. Two samplings at two different times were carried out. Samples were extracted with immunoaffinity column and analysed by direct injection into the high-performance liquid chromatography with fluorescence detection (HPLC-FLD). In addition, the presence of potentially toxigenic moulds was also analysed by real-time PCR by using specific primers involved in mycotoxin biosynthesis. Results showed that 3 samples were contaminated with ochratoxin A and 1 with aflatoxins, concretely aflatoxins B₁, B₂ and G₂. All contaminated samples belonged to dried fig ($<24\%$ m.c.) cultivated under a rainfed system. In addition, a high percentage of potentially ochratoxigenic and aflatoxigenic moulds was detected. Therefore, it is necessary to design strategies to control toxigenic moulds in dried figs by improving farming system practices and accelerating drying.





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Effect of temperature on growth and mycotoxin production of *Aspergillus* section *Flavi* on dried figs in Extremadura

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Aspergillus section *Flavi* is one of the most important fungal hazards in dried fig. Under specific conditions, they can produce mainly aflatoxins (AFs) together with other mycotoxins such as cyclopiazonic acid (CPA) and kojic acid (KA). Conditions during the traditional sun drying and industrial process of dried figs are favourable for mould development and mycotoxin contamination.

The aim of this work was to evaluate the impact of temperature on native *Aspergillus* section *Flavi* from Extremadura on growth and mycotoxin production.

For this aim, the moulds were isolated from dried figs that showed fluorescence during their processing. They were typing by clustering analysis applying ISSR-PCR and subsequently identified by sequencing of the ITS and β -tubulin regions. Six strains were selected to study the impact of different temperatures (22, 32 and 37 °C) at 0.92 a_w on growth in semisolid YES by measuring the turbidity in a Bioscreen C. The Time to Detection (TTD) of growth initiation was determined under the different variable assayed. Finally, at the end of growth, the metabolites aflatoxin B₁ (AFB₁), aflatoxin B₂ (AFB₂), aflatoxin G₁ (AFG₁), aflatoxin G₂ (AFG₂), KA and CPA were analysed by LC-MS.

The isolates obtained belong to *A. flavus*, *A. parasiticus* and *A. tamarii*. Two strains from each species was selected for the assays.

Regarding to the capacity of growth under different temperatures, the TTD was variable between species and highly affected by temperature. At 32°C was recorded the lowest TTD values for the three species studied. In terms of mycotoxin production, *A. flavus* had the highest production of AFB₁ and AFB₂ at the lowest temperature, 22°C. However, it had the highest production of KA and CPA at 37°C and 32°C, respectively. On the other hand, *A. parasiticus* obtained the highest production of AFB₁, AFB₂, AFG₁, AFG₂ at 37°C, whereas KA production was optimal at 32°C. In addition, *A. tamarii* only produced KA and the production was higher at 22°C. In conclusion, the





impact of temperature on mycotoxin producing of *Aspergillus* section *Flavi* is species dependent. These findings may help us to improve the production process of dried figs to avoid mycotoxin hazards associated to *Aspergillus* section *Flavi* species.





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Freezing as a potential alternative to phosphine application for insect control during storage of dried figs

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The consumption of dried figs has increased significantly in recent years. A society with an increasingly health-conscious lifestyle, combined with a product that is rich in nutrients and beneficial to health, has meant that dried figs have become increasingly important. Dehydration of the fruit takes place naturally on the tree until they fall to the ground, where they finish drying in the sun. During this period, figs can be attacked by various types of insects, which can affect their hygienic and sanitary quality.

In order to prevent these insects from appearing during the storage and marketing of dried figs, the industry uses aluminium and/or magnesium phosphide (i.e. "phosphine"). This treatment is increasingly being abandoned due to legal restrictions and the negative impact of the use of chemicals.

Our aim was to evaluate the effectiveness of different freezing treatments as an alternative to phosphine. The treatments were 48 hours at temperatures of -5°C and -18°C. The treated fruits had an average infestation of 44.6%, naturally caused by insects of the species *Cadra abstersella*, *Plodia interpunctella* (Lepidoptera) and *Carpophilus hemipterus* (Coleoptera).

The results showed that the most effective temperature was -18°C, which caused 100% mortality of the above carpophagous insects. Treatment at -5°C caused only 55% mortality in Lepidoptera and 90% in Coleoptera.

The results suggest that freezing (-18°C for 48 h) is an effective alternative for the control of storage pests in dried figs, reducing post-harvest treatment times and, above all, avoiding the use of chemical products.





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Some breba fig (*Ficus carica* L. *domestica*) genotypes in Turkey

Although Turkey ranks first in the world's table fig exports, the lack of sufficient production of early table varieties limits exports. Breba fig orchards is not common in Turkey. Usually, the breba trees is in the form of a few trees on the garden edges. One of the Breba cultivars, 1071 Karabakunya is common around the Karaburun region of Izmir, and the 1013 Beyaz Orak is common around the Manisa region and are quite productive. In recent years, due to the humid weather and cracking of the fig fruits in the Mediterranean Region, early fruiting breba varieties are preferred in fig cultivation (Adana, Antalya).

The harvest of fig varieties that ripen the breba fruit is usually started at the end of June. These fig varieties are also of great importance in terms of extending the marketing period of table figs and filling the gap in the early period.

Fruit quality characteristics of 22 female fig genotypes in fig field gene bank ripening breba fruit under Turkey conditions were investigated.

With this research, the genotypes in the Female Fig Field Gene Bank were scanned, and the fig varieties ripening breba fruit were observed and evaluated according to the IPGRI criteria.

In the research, the average fruit weight (g), fruit width (mm), fruit length (mm), ostiole opening (mm), flesh thickness (mm) and water-soluble dry matter amount (TSS)(%) etc. was evaluated according to the Weighted-Rankit method. Class intervals was organized according to the average fruit quality measurement from breba fruits of all genotypes are taken as a basis.

Genotypes originating in Turkey, 1019 Karabakunya, Siyah Çiçek, Fethiye PRT, 250 Yediveren and 1071 Karabakunya breba fruit came forward the fore in table fruit quality criteria. Breba fruit of Banana (foreign origin) genotype was ranked third.

Among the prominent genotypes in the research, except 250 Yediveren, the other genotypes are in dark colour. Fruit inner colour is amber in Fethiye PRT and 1071 Karabakunya, and pink in the other genotypes. The peel of the fruit is generally easily peeled off in all genotypes.

Flesh thickness is an important feature in terms of road resistance. The flesh thickness of the genotypes varies between 3.12 (1071 Karabakunya)- 4.34 (Siyah Çiçek) mm.

In order to increase the yield of breba cultivar/ genotypes in the world and in Turkey, pruning, fertilization and so on, studies are carried out involving applications. Also, studies on pruning continue in our institute.

