



**POSTERS**



UNDERSTANDING THE GENOME  
FUNCTIONING TO ADAPT SUNFLOWER  
TO CLIMATE CHANGE

## Place of genomic selection in sunflower breeding

Genomic Selection (GS) is a method based on the whole genome genotyping of an individual. The genotyped markers are used to predict the individual genetic performance. GS uses two types of datasets: a training population whose individuals are phenotyped and genotyped and an untested population that is only genotyped and that have to be predicted. The training population is used to establish a link between the phenotype and the combination of the markers. Thanks to GS, it is possible to perform selection cycles without phenotyping the individuals.

The GS is cheaper, faster and theoretically more accurate than classical selection. However, if its effectiveness in animal selection is certain, some barriers remain when applied to plants. Indeed, it is not always well adapted to the structure of crops genome and in particular to the Genotype-Environment interactions.

In the current climatic context, one purpose of crop breeding is to find varieties with high and stable yield in different water conditions. Sunflower, *Helianthus annuus*, is a cost-saving crop: it needs few inputs and is well-adapted to water stress situations. As such, this species is a model plant to identify the loci and molecular markers associated with water stress tolerance.

At the moment, the average sunflower yield is about 20-25 q/ha while the potential yield due to genetic progress is 60 q/ha [1]. Yet, the yield gain per year kept reducing since the 1990's [2]. Hence, the performance of sunflower must be improved to keep the crop competitive and adapted to the future climate situation. Yield and water tolerance are quantitative polygenic traits. For such complex traits, if the technical locks are overcome, Genomic Selection can be a solution for crop breeding.

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# Identification of metabolic pathways and candidate genes associated to leaf senescence process in sunflower by integration of transcriptomic, metabolomic and phenotypic data

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Leaf senescence is a complex mechanism controlled by multiple variables, either from a genetic and environmental origin that strongly impacts on crop yield. Senescence process is the last stage in leaf development, characterized by a decrease in photosynthetic activity, nutrients recycling and ultimately cell death.

This study aims to evaluate the post-anthesis leaf senescence process in sunflower. To this purpose, the inbred lines R453 and B481-6 previously characterized as early and delayed senescence genotypes were assayed under field condition. Measurements of green leaf area, quantum yield, chlorophyll content, radiation interception, photosynthesis and grain weight, showed differences in the triggering and progress of the senescence process, showing yield increase in delayed senescence genotype. The integration of gene expression and metabolomics profiles clearly differentiated the molecular and physiological states of the contrasting lines, observing an early activation of the metabolic process associated with protein, lipids, cell wall degradation and nutrient recycling in the early senescence genotype. Furthermore, NAC MYB and WRKY transcription factors families, associated to leaf senescence in model species, also showed high expression levels in the early senescence genotype.

Our results provide a comprehensive overview of the sunflower post-anthesis leaf senescence at physiological, transcriptome, and metabolome levels which may help to understand the molecular mechanisms associated with this process. Additionally, this knowledge could contribute to the design of future breeding interventions to impact the crop yield in a climate-change scenario.

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UNDERSTANDING THE GENOME  
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## Identification of differentially expressed genes associated with mitochondrial stability, DNA repair and recombination in fertile and sterile anthers of sunflower

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Identification of new *Helianthus* cytoplasmic male sterility (CMS) sources and the associated restorers has been a continuous objective for commercial sunflower production in the world. This type of pollen infertility is associated with the expression of novel open reading frames originated from aberrant rearrangement of mitochondrial genome. It is known various CMS sources described until now, showed the same organization at the *atpA* locus as in PET1 CMS, although these male sterile germplasms have different origin, being produced by inter- or intraspecific crosses, mutagenesis or spontaneously aroused. Previously, our investigation provided the first evidence that sunflower plant with gibberellin induced male sterility is also associated with a similar transcript to mitochondrial *orfH522* characteristic for PET1 CMS. However, little is known about the physiological and molecular events leading to occurring of this sterile type of rearrangement. It is suggested that PET1, like other CMS systems is related to recombination events determined by nucleo-cytoplasmic incompatibility, such has been shown in various interspecific hybridization. However, by some researchers was hypothesized that *orfH522* could be originated from a substoichiometric DNA molecule that have been amplified during the first cross, rather than being the result of a new recombination event in the sterile line due to the nucleo-cytoplasmic incompatibility. Our data are in agreement with this statement, taking into account that the phenotype of plants with male sterility gibberellin induced are not the result of nucleo-cytoplasmic incompatibility.

Two genetic models, CMS-Rf system (Drofa F1 and their parental lines) and near-isonuclear lines (fertile SW501 line and its CMS-PET1 analogue) treated with GA3 at developing inflorescence buds period (male sterile phenotype) and sprayed with distilled water (control plants) were analyzed. The florets at different stages of microsporogenesis were subjected to microscopically and molecular analyses.

Thus, comparative investigations of fertile and sterile phenotype revealed 80-90% of meiocytes disrupted at pachytene, that are arrested during all following meiotic divisions in CMS anthers and various abnormalities, chromosome aberrations (delayed chromosomes, chromosome bridges, chromosome laggards, micronuclei, stickiness chromosomes etc.) in the most microsporogenesis stages of plants with GA3 induced sterility. In the same time GA3 treated plants showed high quantity of the mitochondrial *orfH522* transcript, which could be a causal relationship to the suboptimal cellular energetic status, and so to the abnormal pollen development. Based on these findings, we conclude that meiosis cell division, mtDNA replication and integrity that are necessary for mtDNA maintenance in plants with induced male sterility are not reliable. In this connection, quantitative real-time PCR was employed to analyze some genes involved in DNA repair, base-excision repair (GE514261, BU027250, GE512999, GE502158); mismatch repair, mitochondrial genome maintenance, DNA recombination, DNA metabolic process (GE487746, GE501301, GE506904, DY925268, GE513407, GE502378, BU026262); mitochondrial morphogenesis and organization (GE501868, CD852591, BQ915164) in plants with GA3 induced male sterility compared to those with CMS and fertile ones during microsporogenesis. The obtained data revealed various genes expression profiles related to sterility type, genetic background and stage of pollen development. The most of studied transcripts are gibberellin responsive.



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# Water-stressed sunflower transcriptome analysis revealed important molecular markers involved in drought stress response and tolerance

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Water stress is one of the main factors limiting worldwide agricultural productivity and efficiency. Although sunflower (*Helianthus annuus* L.) is a deep-rooted crop, drought reduces yield, quality of oil and seeds produced. A holistic approach that integrates physiological, biochemical and molecular genetic tools could provide opportunities for breeding novel genotypes with stable behavior under water-deficit conditions.

We focused on responses of many genotypes in normal and stress conditions, using genomics (RNA-seq, quantitative RT-PCR), physiological (growth, water statute, stomatal conduction evaluations, transpiration rate) and biochemical analyzes (LC-MS). Through the evaluation of stem growth rates, leaf water contents and phenotypical observations, we precisely identified genotypes that had different responses to water stress.

Abscisic acid being involved in stress processes, we determined its production in different genotypes with full range of sensitivity, with or without water restriction: high ABA content in leaves under normal conditions is a biochemical marker of drought sensitivity.

Several studies showed that decrease in leaf conductance is closely related to higher ABA production, indicating it could act as a water-stress signal to regulate stomatal conductance. ABA leaf profiles in field confirmed those obtained under controlled conditions.

Variability in responses could be observed transcriptomically with the identification of hydrous stress core genes and sensitivity clusters. Transcriptome analysis confirmed genetic and genotypic variability: applied water deficit triggers transcriptome level changes.

By analyzing genes involved in ABA biosynthesis and signaling, we noted that a set of genes composed of HaNCED1, HaABA1, HaABA2 had a similar expression level regardless of the status of water stress / genotype. In contrast, two genes encoding 9-cis-epoxycarotenoid dioxygenases, HaNCED3 and HaNCED5 showed distinct expression profiles. In sunflower, HaNCED3 expression seems to be correlated with an ABA increase in response to stress, but not with tolerance to drought, and expression of HaNCED5 was affected by water deficit.

We hypothesized the existence of an additive character for HaANN5, HaH1-S and HaCPT4 genes: the more they were expressed, the more tolerant was the genotype. In contrast, HaCLPC1 was overexpressed in sensitive genotypes regardless of the condition.

Bioinformatics analyses of RNA-Seq data pointed out ABI1 homolog as a molecular marker of drought tolerance: under stress it was overexpressed for sensitive genotypes, while one of the most repressed genes was PYL4 homolog. In the genotype least sensitive to drought, low ABI1 and high PYL4 expression lead to a greater reactivity to ABA, more efficient stomatal closure (confirmed by measuring conductance) and better tolerance.

Our transcriptome data provided more details on molecular mechanisms that could explain the differences of ABA accumulation profiles observed in our sunflower lines. We propose that sunflower water stress tolerance is correlated with a transcriptome fine-tuning leading to an efficient activation of ABA-dependent genes and not by ABA overproduction.

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# Differential impact of climate change on *Sclerotinia sclerotiorum* and *Verticillium dahliae*, two fungal pathogens of sunflower

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Facing challenges of climate change, the agricultural sector must adapt itself to feed a growing population (9 billion people in 2050). However, if the nature of crops and agricultural practices could change, bio-pathogenic aggressors could also.

Many fungal diseases affect sunflower crops but we chose two of them to compare their reactions towards climate change. The first one is *Sclerotinia sclerotiorum* which is very common in France even if only 1% of the sunflower-sown fields are infected by this fungus in the South West. Then, *Verticillium dahliae* has been an emergent fungus in France since the last 3 years: it has been observed in 41% of the fields (Terres Inovia survey, 2016).

Furthermore, their life cycles and patterns of infection are very different. On the one hand, *S. sclerotiorum* infects the aerial parts of sunflower plant by disseminating its spores thanks to the wind. It forms a mycelium network and typical sclerotes. On the other hand, *V. dahliae* spreads from the roots to the top of the plant by using the xylem tissue. As it reduces water transfer, the plant wilts before normal maturity.

Therefore, with climate change, these diseases due to their ecological preferences will respond differently as we will illustrate. In fact, the former is aerial whereas the latter is telluric. Thus, if increasing temperatures will change their life cycle similarly (earliness, longer period of infection...), their ability of spreading may also be modified. For example, the precipitation could have a bigger impact on an aerial fungus than on a telluric one.

Lastly, we know that agricultural practices are going to change to be more environmentally friendly and adapted to abiotic constraints resulting from climate change. These modifications will also affect the development of *S. sclerotiorum* and *V. dahliae* in a direction unpredictable today.

Considering all these aspects, we are going to state hypothesis on the following process: how climate change could modify the interactions between these two fungi life cycles, the sunflower development and finally the agricultural practices changes.

## Keywords:

sunflower / *Sclerotinia sclerotiorum* / *Verticillium dahliae* / climate change / interactions

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# Development of mutagenized sunflower populations: discovery and characterization of mutants for Sclerotinia Head Rot resistance candidate genes

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Sclerotinia Head Rot (SHR), caused by the necrotrophic fungus *Sclerotinia sclerotiorum*, is one of the most damaging diseases in sunflower. Resistance to this pathogen is quantitatively inherited with predominantly additive gene action, which has hampered the identification of reliable sources of resistance and the subsequent breeding of elite germplasm. The sunflower germplasm bank of INTA preserves a large proportion of genetic diversity in the form of pre-breeding lines, open-pollinated populations, composites and wild sunflower populations. These genetic resources have been used in QTL-mapping and transcriptomic analysis, leading to the identification of more than 1000 candidate genes. However, a validation step is still required to warrant effective incorporation into breeding programs. Reverse genetics tools such as mutagenized populations are a useful resource to validate candidate genes and produce stable genetic stocks of agronomically important mutants. This study aimed to validate the involvement of a set of candidate genes in SHR resistance and evaluate their contribution to disease response through the analysis of mutants.

Three mutagenized sunflower populations were generated, using ethyl methanesulfonate (EMS), ionizing radiation or sodium azide. Two high-throughput genotyping techniques were optimized to identify variants in each of these populations: (1) CEL1 cleavage of heteroduplex followed by capillary electrophoresis; (2) target enrichment by microfluidics-based multiplex PCR sequencing combining Fluidigm and Illumina technologies.

DNA fragments for 10 candidate genes were screened for mutations in the EMS-treated M2 population (1146 plants, 7989552 bp) using the CEL1 method. Two point variants were found in two different candidate genes. Preliminary analysis of the radiation-treated M2 population (188 plants, 3262552 bp), allowed identification of 28 point mutations, encompassing 15 of the 48 screened genes.

The response to SHR of the two sunflower mutants detected in the EMS-treated population was evaluated under field conditions using assisted inoculation. The mutant of the candidate gene encoding a mitochondrial calcium uptake protein expressed in stamen and pollen showed increased resistance to SHR relative to the control ( $p < 0.05$ ).

In sum, the results presented here offer new prospects for efficient breeding by generating novel allele variants for SHR resistance and pyramiding validated candidate genes.

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## *Cadophora malorum*, a new threat for sunflower production in Russia and Ukraine

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Russia devotes more than six million hectares to sunflower (*Helianthus annuus* L.) crop every year being by far, together with Ukraine (5.2 millions hectares), the most important producer countries worldwide. From 2014 an increase of wilt diseases is being observed in these two countries. In some cases *Verticillium dahliae* was identified as the causing agent of this symptomatology, but in others symptoms are similar to those of *Verticillium* wilt in the beginning but later a sudden death is observed. This phenomenon has been observed to have a relatively high impact in sunflower production in Orenburg and Saratov oblasts in Russia, and in Kiev, Cherkasy and Mykolaiv oblasts in Ukraine. Disease incidence was estimated between 15-40% depending on the field, and yield from infected plants was near to zero. Symptomatic plants were collected for observation and pathogen isolation. Fungal colonies were consistently isolated from all those samples in PDA medium. Colonies were flat, with felty to fleecy rings, the color varying from white-beige to brown-olivaceous. This morphology is completely different from that of *V. dahliae* colonies. On the basis of morphology, the fungus was identified as *Cadophora* sp. Species identification was conducted by amplification of the  $\beta$ -tubulin locus using primers BTCadF/BTCadR. Sequences of 750 bp of the isolates were deposited in GenBank and BLAST analyses showed 92% identity with *C. malorum* (accession KU532784.1) in the NCBI (National Center for Biotechnology Information) database. Pathogenicity of the isolates was confirmed in a greenhouse experiment by root immersion in a solution of conidia with 3-week-old plants of HA458 (public) and DP1 (Dupont Pioneer) inbred lines. Six weeks after inoculation, symptoms developed in 100% of the plants in both sunflower genotypes. Wilting and dwarfism were observed in the inoculated plants, and at least half of the nodes of those plants had necrotic leaves. Water-inoculated plants did not show any symptom. Re-isolations were made from two inoculated plants per each original isolate, and all the recovered isolates were morphologically identical to the original ones, fulfilling thus Koch postulates. To the best of our knowledge, this is the first time the *C. malorum* species is described as a pathogen of sunflower. Resistance screenings to determine tolerant hybrids are in progress in order to provide farmers with a tool to minimize economic losses. Our finding will eventually affect the goals of sunflower breeding programs for Eastern Europe.

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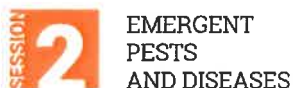
## An holistic approach to the diversity of *Verticillium dahliae* affecting sunflower in Europe

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Verticillium wilt and leaf mottle of sunflower, caused by the fungus *Verticillium dahliae* (Vd) has become a major constraint to sunflower oil production in temperate European countries. In spite that genetics, molecular traits and pathogenic abilities of fungal strains affecting crops like tomato, lettuce, spinach, mint, olive tree and cotton among others are widely known, information about Vd from sunflower is very scarce. Understanding and characterizing the diversity of populations of Vd in those countries where sunflowers are frequent and severely affected by Vd are essential for efficient breeding for resistance. The objective of our research was to analyze genetic, molecular and pathogenic traits of 38 Vd isolates affecting sunflower in European countries (Bulgaria, France, Italy, Romania, Spain, Ukraine and Turkey). Vegetative compatibility groups (VCG) were determined on the basis of formation stable heterokaryons with nit mutant tester strains. Molecular characterization was carried out by genotyping markers used in Vd affecting crops other than sunflower. Pathogenic characterization was done first by investigating the host range with six crop species, and second by inoculation in seven sunflower genotypes known to have different responses to Verticillium wilt. Almost all isolates from France, Italy, Spain and Ukraine were assigned to VCG2B. On the other hand, some isolates from Bulgaria, Turkey, Romania and Ukraine, were assigned only to VCG6. Other group of isolates from the last countries could not be assigned to any VCG. All the isolates were identified as race 2 according to molecular markers defined for Vd in tomato and lettuce. Markers of defoliating and non-defoliating pathotypes distinguished two well-differentiated clusters. One cluster grouped the 18 isolates from Eastern Europe (Cluster E), sharing about 47% similarity. All isolates from countries of the West of Europe shared 40% similarity and were grouped in a second cluster (cluster W). Interestingly, all the isolates in cluster W were VCG2B, while isolates in cluster E belonged to an unknown VCG or to VCG6. Two big groups of pathogenic variants were identified in the pathogenic characterization with seven sunflower genotypes and it is remarkable that these groups are in agreement with the molecular characterization. Thus, one cluster included the isolates from West Europe and two isolates from Ukraine, and the other grouped isolates from East Europe. According to this data, three pathogenic races were differentiated and called race V1, race V2-EE (East Europe) and race V2-WE (West Europe). Three differentials are proposed for race identification: HA 458 (universal susceptible), HA 89 (resistant to V2-EE and susceptible to V2-WE) and INRA2603 (susceptible to V2-EE and resistant to V2-WE).

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## *VdOCH1*, encoded $\alpha$ -1,6-mannosyltransferase, manipulated the microsclerotia formation and virulence of *Verticillium dahlia*

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Sunflower yellow wilt is a widespread and destructive disease caused by the soil-borne pathogen *Verticillium dahliae* (*V. dahliae*). To better understand the pathogenesis mechanism of *V. dahliae* on sunflower, T-DNA insertion library was generated via *Agrobacterium tumefaciens* mediated transformation system (ATMT). 800 putative transformants were obtained and 42 were confirmed as positive transformants based on both PCR identification with hygromycin B primer pair and GFP signal under fluorescence microscopy. Transformants varied in colony morphology, growth rate, conidia production and pathogenicity on sunflower compared to the wild type strain. A mutant, named as VdGn3-L2, was chosen for further analysis based on its morphology deprivation on the microsclerotia formation. The flanking sequence of T-DNA insertion site of VdGn3-L2 was identified via hiTAIL-PCR, and the interrupted gene encoded an initiation-specific  $\alpha$ -1,6-mannosyltransferase, named as VdOCH1. In yeast fungi,  $\alpha$ -1,6-mannosyltransferase adds the initial  $\alpha$ -1,6-mannose of a long  $\alpha$ -1,6-mannose chain to the N-linked oligosaccharides (Man8GlcNAc2) and further triggers the mannose out chain elongation (up to 200 mannose residues) and mannosylation. In filamentous fungi, this process initiated by OCH1 is reported only in *Aspergillus fumigatus*, *Neurospora crassa* and *Fusarium oxysporum* f. sp. *cubense*, which adds the initial  $\alpha$ -1,6-mannose of a short chain of  $\alpha$ -1,6-mannoses and galactoses to the N-linked oligosaccharides of cell wall proteins. However, the function of OCH1 in the other filamentous fungi is still elusive. In our study, the deletion mutant  $\Delta$ VdOCH1 were impaired in certain biology characteristics such as fungal growth, conidia production and microsclerotia formation. Beside above characteristics,  $\Delta$ VdOCH1 mutants were also more sensitive to the cell wall perturbing reagents, such as SDS and Congo red, impaired on the penetration ability through cellophane membrane and low down pathogenicity on sunflower dramatically. The above impaired phenotypes can be restored to the wild type level by complementation the deletion mutant with the full-length VdOCH1 gene. In conclusion, VdOCH1, encoded  $\alpha$ -1,6-mannosyltransferase, manipulating the biological characteristics, microsclerotia formation and pathogenicity ability of *V. dahliae* on sunflower.

# Molecular characterization of the major resistance gene Or7 controlling resistance to *O. cumana* in sunflower

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*Orobanche cumana* is a root-parasitic plant of the Orobanchaceae family. This weed specifically attacks sunflower crops and can cause important yield loss in Europe and Asia. Resistant cultivars play an important role for the control of sunflower broomrape in fields. Some of them carry the Or7 locus, a major resistance gene which confers resistance to *O. cumana* race F, the main race in South of Spain. We used a high-throughput genotyping tool to map the locus to the chromosome 7 by GWAs on 55 lines and by a segregating population of 355 RILs. In silico analyses and genetic data identified a genomic region of 850kb containing the Or7 gene. We then restricted the genomic region of the Or7 gene by genotyping a large segregating population of 14000 F2 plants, identifying 270 F2 recombinant plants between the two markers surrounding the 850kb genomic region. After selfing each of the 270 F2 plants, all F3 families were evaluated for their resistance to *O. cumana* in field and F4 progenies of these families were produced and phenotyped in the same field, and in controlled conditions to confirm the F3 families phenotype. Finally the Or7 locus mapped in a window of around 55kb, containing 2 predicted genes coding for proteins known to play important roles in plant resistance against microorganisms. Finally, we obtained and screened two BAC libraries from the susceptible and resistant lines. The genomic sequences of both parental lines were obtained showing polymorphisms with large structural variation. We performed a molecular diversity analysis of the region, in a panel of 170 wild relatives, wild and cultivated sunflower accessions, which showed only one haplotype on Or7+ lines on a gene very close to the 55kb window. Some wild accessions of the *Helianthus* genus shared this haplotype, and the genotyping of these accessions is underway.

## Phenotyping tools for the resistance of sunflower to *O. cumana*

*O. cumana* (the sunflower broomrape) is a parasitic plant that specifically infects sunflower and is one of the main constraints of sunflower crop in Europe. High throughput phenotyping of sunflowers for their resistance to broomrape is one of the main challenges to breed varieties with new resistances. However, it fixes on sunflower roots before emerging overground as a flower shoot. In addition, *O. cumana* populations differ by their virulence and aggressiveness from their geographical locations. Taking together, these biological and genetical features make high throughput phenotyping difficult in the field. Therefore, developing new phenotyping protocols in controlled conditions is a good way to better characterize the physiological effect of the infection on sunflower development and to screen for resistant sunflowers.

We used the Toulouse Plant Microbe Phenotyping facility to characterize a set of sunflower lines showing a scale of resistance from susceptibility to total resistance to different broomrape races. By measuring the height of sunflowers, we calculated the effect of the infection on sunflower growth of the different lines according to the broomrape races and we are working on the analysis of the acquired images.

We also highlight new starting projects that aim to (i) identify resistant sunflowers at the early stages of the interaction by measuring its effect on sunflower physiology and (ii) to develop an automatic mini-rhizotron phenotyping tool to access to sunflower roots.

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# Agronomic and physiological evaluation of some new introduced hybrids of sunflower (*Helianthus annuus L.*) under drought conditions in Morocco

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Drought was always present in Morocco's history. Its importance as a structural element of the country's climate increased during the last decades, with a net reduction in the precipitation and an increasing temperature trend, as a result of climate change. The present study was carried out in Meknes area (Morocco) in order to characterize and evaluate new sunflower hybrids for their agronomic performance and tolerance to water stress often occurring at bud and flowering stage. Seven introduced hybrids were conducted in Ain Jemaa under rainfield conditions during 2016-2017. Agronomic parameters, namely plant height, head diameter, number of seeds/head, 1000-seed weight, seeds weight/head, and seed yield/ha, as well as physiological traits, like as chlorophyll level, stomatal conductance and relative water content, were measured. Seed yield was shown to be significantly correlated with number of seeds per head, head diameter and plant height. The highest seed yield, 2.84 t/ha, was observed for the variety 'Laila', while the lowest yield is recorded for the variety 'Merida'. In 'Laila' hybrid, during flowering stage, there was a significant and strong positive correlation between stomatal conductance and seed yield. As flowering process was happening under drought conditions, it could be suggested that that hybrid was able to maintain open stomata and, thus, to yield better than the other studied hybrids.

#### Keywords:

sunflower / hybrid / drought / seed yield / stomatal conductance.

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# Analysis of factors controlling oil content and fatty acid composition: climate change effects and adaptation strategies

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Sunflower is a major oilseed crop which produced up to 15.8 M tons in 2014 at world level. Its oil is widely used because of its nutritional and industrial characteristics.

The two characteristics that define seed quality are oil concentration (OC) and fatty acid composition. Concerning fatty acid composition, the agri-food industry is looking for high levels of oleic acid concentration (OAC) for its high stability and its nutritional properties. In a context of climatic change, OAC and OC can be impacted by environmental changes. Thus, the purpose of this study was to identify the different effects of those changes on OAC and OC.

OC is dependent on environmental factors such as temperature, CO<sub>2</sub> concentration, water stress and radiation. OAC is mostly governed by night temperature for a given hybrid. These factors are strongly influenced by climate change. One example of those factors is temperature which is increasing with global warming: high temperatures at the end of the flowering period decrease the activity of the FAD enzyme conducting to higher OAC values in the seeds. Therefore OAC is often affected by sowing time and latitude. Agronomic factors should be taken in account for OC (e.g sowing density, N fertilization, irrigation, genetic). The moment a sunflower plant is impacted by a factor should also be considered since the physiological condition (growth, flowering, maturation) of the plant makes it more or less vulnerable to the stress.

To help minimizing these impacts by targeting adequate adaptation strategies, we explored different options, like sunflower variety breeding or crop management system. Statistical models can also be of great help in predicting sunflower OC and OAC in a specific context, especially because of the difficulty of understanding the complex interactions between both agronomic and environmental factors (i.e Sunflo model).

## Keywords:

sunflower / oil concentration / climate change / environmental factors / adaptation strategies

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# Interest and difficulties of conservation agriculture for adaptation of sunflower to climate change

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For fifteen years, sunflower yields remain stable in spite of genetic improvement, mostly because nowadays sunflower crops endure droughts more frequently due to climate change.

Climate change is responsible for rising temperatures, increasing the frequency of violent weather-related events and making available water resources scarcer.

Conservation agriculture (CA), which has been practiced in France since the 2000s, proposes solutions to reduce the negative impacts of climate change. Based on no-till farming, permanent soil cover and long-term rotation, it allows reducing soil erosion and improving soil water retention and efficiency.

However, growing sunflower in conservation agriculture is challenging because of technical impediments. The rates of crop emergence and plant population establishment are lower than those in conventional agriculture, due to a frequent poor quality of soil-seed contact. We observe lower yields and a worse quality of sunflower in CA.

To adapt sunflower crop in CA in the context of climate change, genetics and technical levers can be used. New varietal characteristics are needed to improve sunflower performance in CA, such as cold tolerance, to realize early sowing.

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## How is sunflower vulnerable to drought-induced embolism? Implication for breeding in an increasingly arid world

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Drought-induced xylem embolism is a key process closely related to plant mortality and productivity decline during drought events. However, this process has been little investigated in crop species to date, despite the observed decline of crop productivity under severe drought conditions. We assessed the intraspecific variability of embolism resistance in four sunflower (*Helianthus annuus* L.) accessions grown in well-watered conditions. Vulnerability to embolism (P50) was determined by the in situ flow centrifuge method (cavitron), and possible trade-offs between xylem safety, efficiency, anatomy and growth were assessed.

Sunflower is moderately vulnerable to embolism (P50 =  $-2.99 \pm 0.15$  MPa) relative to other herbaceous and woody species, which have P50 values ranging from  $-0.5$  to  $-7.5$  MPa (Ahmad et al. 2017, Lens et al. 2016) and from  $-0.5$  to  $-18.8$  MPa (Delzon et al. 2010, Choat et al. 2012, Larter et al. 2017), respectively. We found significant intraspecific differences in vulnerability to xylem embolism in sunflower, with the accessions at the two extremes of the scale differing in P50 by about 0.55 MPa. Vulnerability to embolism was also positively related to growth and negatively related to xylem-specific hydraulic conductivity, highlighting a trade-off between embolism resistance and growth, but not between xylem safety and efficiency. Future studies should investigate (i) the variability of embolism resistance across a wider range of accessions, (ii) the extent of the negative correlation between hydraulic conductivity (hydraulic efficiency) and growth traits in crops, and (iii) the possible existence of a trade-off between embolism resistance and yield potential to facilitate the identification of breeding strategies to improve yields in an increasingly arid world.

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## The use of crop wild relatives in sunflower breeding for drought response

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One of the major limitations for agricultural productivity around the world, particularly in warm, arid and semi-arid regions, is water stress. Drought is the most restraining factor in sunflower production worldwide, severely reducing yield, oil volume, oil quality, and other important yield traits. Sunflower, being a crop with medium water requirements ( $K_y < 1$ ), has the ability to tolerate a short period of drought. The future of sunflower is probably related to its potential adaptation to climate change. The challenge for the sunflower breeding community is to breed sunflower adaptable to these marginal environments and at the same time to increase seed yield. To develop drought-tolerant lines and hybrids in sunflower breeding, breeders need to be aware of the relationship between drought resistance traits and yield and apply effective screening methods for these traits. Genotypes should have such advantages as enhanced leaf area, earliness, and earlier stomatal closure.

The narrow genetic base of cultivated sunflower has been broadened by the infusion of genes from crop wild relatives (CWR). When choosing CWR which can increase the adaptive capacity for tolerance to drought and high temperatures, it is important to opt for species which inhabit desert and semi-desert areas. *Helianthus deserticola* inhabits the desert and semi-desert areas. The name of this species implies what kind of conditions it can withstand in the nature, as it grows on stabilized sandy soil and desert soil, which makes it perfect candidate for introduction of traits connected with drought resistance into cultivated sunflower through interspecific hybridization. Traits connected with this strategy include fast growth and flowering rate, lower plant height in the maturity, increased photosynthesis activity and stoma conductivity. *H. deserticola* shows both increased photosynthesis activity and stoma conductivity so it is excellent candidate for the adaptation to desert environments.

In this research, the plants were first selected from the interspecific population DES-1474-1, DES-1474-2 originating from annual wild species *H. deserticola* and then subjected to inbreeding and used with the intention to make cms female and Rf male inbred lines resistant to drought. Several agronomically important traits of new cms and Rf inbred lines and their hybrids were evaluated and could be used for the production of new drought resistant sunflower hybrids.

# Phenotyping of segregating generations derived from sunflower interspecific crosses (*H. annuus*) and (*H. argophyllus*)

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Limited water availability suggests that in future water insufficiency will be norm which leads to the major losses in yield and yield components. Management under these conditions as compare to unlimited water availability may result in reducing yield losses. Study was started to improve crop production by introgression of drought tolerant traits from wild sunflower to cultivated type of sunflower resultant material of filial generations F1, F2, and F4 is analyzed on the basis of some phenotypic and physiological indicators. When compare to parents UCA-16 and Argo-5-IV these segregating populations have mean value in F1 and when compared by leaf parameters like leaf rolling, hairiness, leaf colour and waxes produced by cuticular layers behaved differently. In selected material of successive generations on cell membrane injury, achene yield and single head type will be further grown by keeping ultimate objective of drought tolerant inbred line.

**Keywords:**

introgression / production / leaf color / tolerant inbred line / leaf rolling.

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# Sunflower breeding for the improvement of resistance to drought and extreme temperatures at NARDI Fundulea, Romania

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Climate changes characterized by higher temperatures, extreme climatic hazards and low water for agriculture determine the extension of the areas affected by drought. Agriculture is most affected by the climate variability, the extreme meteorological phenomenon, diminishing the yields with 35-50% each year.

The Romanian National Strategy (2013-2020) deals with adaptation to the climate changes. The National Agenda for the climate changes is based on specialized tools regarding the climate risks at the national and regional levels.

Sunflower is considered to be moderately resistant to drought, but in hot conditions, the plants suffer reduction in fertility, yield performance and quality of products. In literature there are mentioned some adoptive mechanisms of plants to drought: escape, avoidance and tolerance, as well as their genetic variability.

In our research work we have used different sources from our sunflower germplasm collection, some of them coming from the interspecific hybrids between wild *H. argopyllus* and cultivated sunflower. Some of our best elite lines have been introduced in a process of improvement of resistance to drought, using recurrent selection. We have used several parameters or characteristics in selection of the tolerant plants: deeper rooting depth and more efficient root uptake of water, area of leaves and number of leaves, plant ability to recover after wilting under heat stress. Each generation of selection was planted in drought natural conditions (missing water in soil and high air temperature), in the area named as pole of drought in Romania. The generations obtained in the glass house have been tested for resistance, using methods for laboratory conditions (PEG 6000). All generations of selection were tested for resistance to low temperatures in germination and emergence time. There have been selected the more tolerant ones.

In this process of selection, we obtained inbred lines (CMS and pollen fertility restorer lines) having very good tolerance to drought as well as resistance to low temperatures. By crossing these lines there have been obtained hybrids having good tolerance to drought, some of them with good resistance to low temperatures in emergence time. The hybrids have been tested in comparative trials in 4 locations, situated in areas with drought conditions, as well as in Fundulea institute field, having normal meteorological conditions. The seed yield released by hybrids has shown a high level in conditions of drought. In Constanta area it has been experimented the three planting times of the hybrids, the best results regarding the level of the seed yield, being in case of half of March planting time.

We selected 6 hybrids, the most tolerant and productive, these being now tested in the network of Institute for Plant Variety Testing and Registration.

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## Aspects and perspectives of sunflower research in Tunisia

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In Tunisia, the improvement of oilseed crop production is of a great importance. In addition to drought, the lack of improved local varieties is considered the major constraint to the development of oilseed crops, especially, sunflower (*Helianthus annuus* L.).

Different studies from 1965 till today have been carried out in the National Institute of Agronomic Research of Tunisia (INRAT) and the National Institute of Agronomy of Tunisia (INAT). The main aim of those studies was to develop productive and high-oil-content varieties of sunflower in Tunisia with improving resistance to diseases (*Plasmopara helianthi* Novot, *Puccinia Helianthii*, *Sclerotinia* .sp), pests (*Homoesoma nebulella*), weeds (*Cumana* .sp), cold and drought. In the last Century, studies were interested in the imported sunflower varieties from Russia, Hungary, Germany, Canada and Czech Republic. However, the recent studies were focused on the diversity of local sunflower populations cultivated by farmers in the north-west of Tunisia. In this abstract, we report the main results published on sunflower in Tunisia.

In 1965, the laboratory of genetic of INRAT introduced different varieties (Admiral hybrid, Cernanka 66, Peredovik, Airelle, Pionier Sibiri, Vniimk, Smena 15636, Giant 1476 B...), aiming to fix the crop management of sunflower in Tunisia. Experimental trials showed that low temperature is less dangerous than drought and early sowing of sunflower (first decade of March) was successful, contrary to maize and sorghum.

From 1972-1980, studies were interested mainly in identification of sunflower diseases, and in screening the most tolerant varieties. Downy mildew (*plasmopara helianthi* Novot) is observed for the first time in an out-season cultivation (sowing in October 77) so, description of symptoms, parasite and the means of control were preconized. *Macrophomina phaseoli* is regularly observed and isolated from all fields, it was noted that the disease may cause a reduction of yields evaluated to more than 25%. Results showed also that early varieties are the most sensible except one (Airelle) that presented a good level of tolerance. Vascular diseases of sunflower in Tunisia were attributed to *Fusarium oxysporum* sp. *Helianthi novot. sp* and the wilt of sunflower to *tracheomyces*.

During 90's - 2000, research on imported varieties continued and was interested in determining the effect of sowing date (from December 1999 to January 2000) on qualitative aspects of oil. The results demonstrated a decrease in yield as well as an increase in the number of sterile seeds with a decrease in the 1000 seed weight from 61.87 to 44.19 g. The total oil yield was not affected by the sowing dates, only its composition was affected. The rate of oleic acid increased with the delaying of the sowing dates. The content increased from 19.2 to 25.2%; however the rate of linoleic acid decreased from 49.4 to 59.4%. Known as adapted to soil and climate tunisian conditions, the two studied varieties (Albena, Blizar) were registered in the official catalogue of Tunisia in 2005.

At the beginning of this century, sunflower was well expanded reaching 13.000 ha, but still used mainly for confectionery purposes, to orientate the use of sunflower to oil industry, research was interested mainly in determining the most adapted oleic Pioneer varieties. In 2006 - 2009, results showed that early sowing (January - beginning of Mars) before conventional sowing date (end of Mars - Avril) may reduce the effect of drought occurring in the flowering stage and the effect of birds on seeds.

Among results two oleic Pioneer varieties (PR63D82, PR64A71), found well adapted to Tunisian bioclimatic conditions were registered in the official catalogue of Tunisia in 2010.

During 2011-2017, research concerned mainly local populations cultivated in the north-west of Tunisia, on which different aspects of diversity were conducted. Added to



■■■ Agro-morphological and phenological characterization, genetic diversity was carried out using SSR and SNP markers. Results showed variation within populations, and noted that selection of genotypes with a short growth cycle related to the lowest growth degree day and leaf area, with a high productive potential in drought conditions is a very interesting aspect. However, Genetic analysis highlighted some uniqueness of the tunisian material compared to control references and a low genetic dispersion between populations. Among results, No relation was found between genetic and geographical distances within tunisian populations which suggest the seed exchanges between farmers or agricultural institutes. The genetic analysis allowed us to define a core collection with 8 populations from Slougia, Dougga, Oued Zarga, Ksar Mezouar, Ain Chelou, Sejane, Tounga, and Oued Beja capturing 90% of molecular allelic content of cultivated populations in the north-west of Tunisia.

Based on the previous agro-morphological, phenological, genetic studies and some histologic aspects of tap-root, different populations (C8.3.1.2; C7.3.2.2; C1.3.1.1; C5.2.3.2; C31.1.1.2) were pre-selected and tested In vitro, simulating water stress by PEG6000. In the field, sunflower populations were submitted to different water regimes. Oil content and fatty acid composition were analyzed in normal and water limited conditions. Compared to a reference variety (PR64A71), C8.3.1.2 and C31.1.1.2 were selected and proposed in 2016 for inscription in the Tunisian national official catalogue. Named ATIA, this variety of sunflower has shown precocity compared to (PR64A71) with an average cycle of 130 days and a growth degree day (Emergence-flowering) of 554 to 689 (GDD). Seeds of ATIA have black color, and characterized by 1000 seed weight evaluated to 104 g, 25 to 30% crude protein content and 32% oil amount with 50.23% of oleic acid content.

Despite the researchers effort to develop sunflower cultivation in Tunisia, this crop still need supports from the Tunisian government to be well expanded , without forgetting the main role of private seed society.

# Sunflower hybrid idiomorph and fulfillment of the yield potential in the eastern forest-steppe of Ukraine

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Stable fulfillment of the yield potential of sunflower in production depends on the adaptability of F1 hybrids to changing environmental conditions. The problem attracts a special attention in connection with increasing frequency and level of stressful abiotic environmental factors, which has been noted recently.

Thus, in 2004–2017 in the Eastern Forest-Steppe of Ukraine, the hydrothermal coefficient (HTC) during the sunflower vegetation period varied considerably from 0.65 to 1.32. Significant lack of warmth (2004, 2008, 2009, 2014, 2015, 2017) and moisture (2009, 2010) at the initial developmental stages of, high temperatures (2010, 2012, 2016) and drought (2005, 2010, 2012, 2014) during anthesis and at the beginning of seed formation, severe drought during the seed filling period (2009, 2011, 2017) conspicuously influenced the fulfillment of the genetic potential of hybrids.

The yields of F1 hybrids in experiments ranged from 2.26 t/ha (2010) to 4.0 t/ha (2007). In 2004, 2005, 2010, 2012, 2016 and 2017, which were unfavorable for sunflower, the average yield was 2.26 – 2.91 t/ha. The maximum yield was recorded in 2006 and 2007, which were comfortable in terms of the hydrothermal mode both for the 1st and for the 2nd halves of the vegetation period (3.99–4.00 t/ha). While the average yields of hybrids significantly varied over the years of research (from 2.3 to 3.8 t/ha), significant differences in their ecological plasticity (from 0.02 to 1.83 units) were noted.

Basic breeding traits with the highest level of interdependence with yield are defined; a model of dependence of yield on these traits – growth in the leaf surface area and the coefficient of calathidium parastichy- was developed. A possibility of obtaining high-yielding F1 hybrids with combination of the growth rate in the leaf surface area during the vegetation period in the range of 0.6–0.7 and the coefficient of calathidium parastichy  $\leq 0.618$  as well as with 0.5–0.6 and 0.8, respectively, was demonstrated.

We determined optimal values of the idiomorph of F1 oil sunflower hybrids for the Eastern Forest-Steppe of Ukraine with a yield of  $\approx 4$  t/ha, which corresponds to an early-ripening or a mid-early hybrid with the index of reproductive capacity of 6,000–8,500/m<sup>2</sup>, 1000–seed weight of 60–85 g, the coefficient of calathidium parastichy of 0.6–0.7, intensive initial growth, providing the leaf surface area in the “3 pairs of leaves” phase of 1,100–1,700 cm<sup>2</sup>/plant within 17.5–19 days, the ratio of the length of the “emergence-anthesis” period to the length of the “anthesis - physiological maturity” period of 0.55–0.57, the plant height of 165–168 cm and the oil content in seeds of  $\geq 50\%$ .

These results offer an opportunity for targeted selection of promising hybrid combinations that can give high yields in the conditions of the Eastern Forest-Steppe of Ukraine.

## Evaluation of the temperature effect on economic features of sunflower hybrids of different ripeness groups

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One of the main factors that limit sunflower production is high air temperature. Sunflower hybrids belonging to different ripeness groups (short-season, early-ripening and mid-early groups) were tested in the northern part of the moderately arid zone of Ukraine during the period of 1998-2015. Results of combined analysis of variances for seed yield, oil content in seeds and oil yield showed significant effects of hybrids` ripeness group, environments and hybrids` ripeness group  $\times$  environments interaction.

The differences between hybrids belonging to different ripeness groups were determined in terms of the variability in these features, in years with various heat availability (high, medium, low) during the vegetation period. That suggested using the vegetation period length as a mechanism to avoid negative effects of heat stress. The influence of a temperature regime of a year on the variability of these features was assessed in year groups characterized by different temperature conditions, with the intergroup intervals of 2°C and 5°C for estimating the yield variability and the variability of oil content in seeds, respectively. In the northern part of the moderately arid zone of Ukraine, the 2°C increment in the average maximum daily temperature in June (from 26-27°C to 28-29°C) meant a various reduction in the yields of hybrids of different ripeness groups: by 0.17 t/ha in early-ripening hybrids; by 0.36 t/ha in short-season hybrids; by 0.07 t/ha in mid-early hybrids. The 2°C increment in the average maximum daily temperature in July (from 29-30°C to 31-32°C) reduced the yields of mid-early hybrids by 0.60 t/ha.

The variability directions of the oil content in seeds of hybrids of the three ripening groups were different, when the average maximum temperature in the 1st and 3rd ten-day periods of July increased above 27°C. Under such conditions, mid-early hybrids maintained the oil amount in seeds, but early-ripening and short-season hybrids reduced it. The compensation effect for losses in oil output due to a decrease in its content under the influence of high temperatures in July and August was observed and attributed to increased yields of hybrids.

The study of the interphase vegetative period lengths of 15 industrial hybrids, regardless of the ripeness group, revealed shortening in the lengths of the "sprouts-calathidium formation" and "calathidium formation – anthesis" periods, depending on the temperature during the corresponding period. A negative correlation between the length of the "anthesis - physiological maturity" period and temperature was only proved for several hybrids.

Over the period of 2001 to 2015, 47 sunflower hybrids, which belong to short-season, early-ripening and mid-early groups, with components of increased heat resistance and potential yields of to 4.8 t/ha, were registered in the State Register of Plant Varieties Suitable for Cultivation in Ukraine. The created hybrids were implemented in the agricultural production of Ukraine.

## Sunflower breeding for production stability Example of NS hybrids

Sunflower breeding process involves the selection of hybrids which have the tendency to maximize yield potential and its stability in different growing regions. Perspective genotypes are usually evaluated in different environments before selecting desirable ones that show stability across environments. In identifying such genotypes, G×E interactions are of major concern for the breeders, because such interactions confound the selection of superior cultivars by altering their relative productiveness in different environments. The most widely used way to biometrically assess stability of genotypes is AMMI (Additive Main and Multiplicative Interaction) analysis. AMMI analysis efficiently examines the variation resulting from the influence of the main (additive) effects (genotype and environment), as well as the variation resulting from the interaction effect (non-additive effects). Estimates based on the AMMI model are more precise in comparison with other analysis because a more detailed analysis of the G×E interaction provide agronomically important observations.

The adaptability and stability of 13 commercial hybrids, five older and seven modern ones, developed at the Institute of Field and Vegetable Crops, Novi Sad, was evaluated during six years across 19 localities by AMMI analysis. Three out of six years were extremely hot, two were extremely dry and one was rainy. Analysis showed that in total average during six years the best producing sunflower hybrids were four modern hybrids with average yields over 3.90 t/ha. High average yields of tested hybrids showed good adaptability to different production areas. AMMI analysis revealed that expression of hybrid yield potential was mainly influenced by locality and not the year. In terms of stability during six years period, AMMI analysis showed that hybrids with the lowest scores of interaction were two modern ones. Calculated AMMI stability value (ASV) also revealed that all tested NS sunflower hybrids were exceptional in terms of yield stability as ASV values in all hybrids were lower than 2, which is considered highly stable.

The obtained data revealed that genetic improvement through modern breeding has a potential to increase production stability of sunflower hybrids. The obtained data provide strong evidence that direct selection for seed yield leads not only to a yield gain, but also to a selection of genotypes with stable performance in different, often extreme, environments creating the preconditions for delivery of successful commercial hybrids.

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# Year × Variety × Fertilizer Regime effects on sunflower seed quality and oil yield in the humid tropics

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The productivity of sunflower depends upon the genetic makeup of the varieties, nutrient availability in the soil at appropriate quantities and time and prevailing agro-climatic conditions during the growing period. To this effect, two field experiments were conducted at the Institute of Food Security, Environmental Resources and Agricultural Research (IFSERAR), Nigeria during the late cropping seasons of 2014 and 2015. The experiment was aimed at evaluating the agronomic performance of four sunflower varieties (SAMSUN 1, SAMSUN 2, SAMSUN 3 and SAMSUN 4) as affected by year and fertilizer regime (control, split and single application of 60 kg N and 56 kg P<sub>2</sub>O<sub>5</sub> at 3 weeks after sowing, WAS). The field trials were laid out in a randomized complete block design using a 3 × 4 factorial arrangement in three replicates. Data were collected on seed yield, protein and oil content and oil yield. Year significantly ( $P < 0.05$ ) affected seed yield, protein and oil content, and oil yield. Year 2014 (610.2 mm) was wetter than 2015 (370.0 mm). Consequently, year significantly ( $P < 0.05$ ) affected all the traits measured with 2014 recording higher values than 2015 for all the traits, except protein content. The varieties were significantly ( $P < 0.05$ ) different from each other and SAMSUN 4 contained the highest protein and oil content. Fertilizer application significantly ( $P < 0.05$ ) increased seed yield, seed quality and oil yield relative to the control treatment. Single application of fertilizers recorded significantly ( $P < 0.05$ ) higher oil content and oil yield than split application and was on par with split for seed yield. Oil yield ranged between 212.91 and 362.78 kg/ha with the single application resulting in significantly ( $P < 0.05$ ) higher oil yield than the control. All the interaction effects were significant for protein and oil content, except Year × Variety for oil content. Therefore, single application of N and P fertilizers at 21 WAS is recommended for enhanced productivity of sunflower in the humid tropics.

**Keywords:**

oil yield / seed quality / seed yield / sunflower

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# Is predicting oleic acid percentage in sunflower oil from air temperature accurate enough?

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According to the literature (1, 2, 3), the percentage of oleic acid (OA%) in sunflower oil is closely related to minimum night temperature (MNT) during a short period during grain filling (between 100 and 300°C.days after flowering; base temperature 6°C). The response of OA% to MNT follows a sigmoid function and is subjected to genetic variability among traditional and high oleic hybrids. Merrien et al. (4) simplified the model by using minimum temperatures (Tmin as a proxy of MNT) and the period between 10 and 30 days after flowering. Both models were calibrated with linoleic and high-oleic cultivars, using data from fields in natural or semi-controlled conditions (Echarte et al., 2010). The models concluded to an increase of OA% with MNT or Tmin on a 11-17°C range in France (4) and 14-21°C in Argentina (3).

In our study, we tested the possibility to predict OA% from Tmin - provided by weather stations - using the post-registration network of Terres Inovia and a range of high-oleic sunflower hybrids. The other variable used was flowering date (FD, observed or simulated with SUNFLO). 228 experiments conducted from 2005 to 2015 in France were selected and the previous models were tested on 27 hybrids. Observed OA% came from trials either freely pollinated (oleic trials) or protected from cross-pollination by bags (oleic + linoleic trials). Tmin ranged from 12°C to 19°C on the 10-30 days after flowering. Values of OA% less than 75 % were discarded as they may have resulted from cross pollination with linoleic varieties. Mean OA% values (84 to 91%) and stability across environments differed among the 27 hybrids. Using Tmin from weather stations (distance 0 - 34 km) resulted in poor relations with OA% except for some hybrids where OA% and Tmin were positively related. Surprisingly, OA% decreased with oil concentration (OC%) when OC% was less than 44 % suggesting common limiting factors. Multiple linear regressions were attempted for explaining OA% at variety level (e.g  $r^2 = 0.43$  for NK Sinfoni with 3 variables: Tmin, OC% and FD).

Several assumptions were examined to explain these poor relations compared to the literature: however, we found no clear effects of the distance to the weather station, of the pollination conditions (freely or under bag) or the growing season. Some authors reported the positive influences of intercepted radiation (5) and water stress (6) on OA%. We conclude that models based solely on temperature are probably not accurate enough for predicting oil quality at field level for high-oleic hybrids.

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# Projections of changes in productivity of sunflower in the Republic of Moldova according to CMIP5 ensemble of 21 GCMs for RCP2.6, RCP4.5 and RCP8.5 scenarios

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In the present study, relationships between observed mean temperature (T) and precipitation (P) during the growing season and average annual rainfed sunflower (*Helianthus annuus* L.) yield based on statistical data at the Republic of Moldova's agricultural enterprises of various categories were explored and used to estimate potential impacts of future climate change on anticipated average yields by 2016–2035, 2046–2065 and 2081–2100 time periods on national and district level, based on the projected T and P changes from the CMIP5 ensemble of 21 GCMs, covering the end of 20th (base period) and 21st (future scenario) centuries introduced by three Representative Concentration Pathways (RCPs): RCP8.5, RCP4.5, and RCP2.6. The analysis of the data shows that the influence of climatic conditions during the growing season on sunflower yield in the 1981–2010 years was statistically significant in the majority of districts of the Republic of Moldova's Agro Ecological Zones (AEZs). The combined effect of P and T during growing season defined from 18 to 60% of the variability of average annual yield of sunflower during the 1981–2010 base period in Northern AEZ, from 38 to 75% of the variability of yield in Central AEZ, and from 38 to 77% of the variability of yield in Southern AEZ. In comparison with the 1981–2010 base period, by 2035 the rainfed sunflower yield may decrease in dependence of the assessed emission scenario from 20 to 33% (RCP2.6) and/or from 23 to 37% (RCP8.5). The maximum values of yield decrease may be reached by 2100, due to changes in values of main climate indicators P and T the yield of sunflower may decrease from 23 to 37% (RCP2.6) and/or from 44 to 76% (RCP 4.5). Without adaptation measures (if maintaining the current cultivation technologies and used varieties), due to changes in climatic conditions in the most districts of the Republic of Moldova, according to the RCP8.5 high emission scenario by the end of the XXI century, the cultivation of sunflower will be impossible or economically not cost effective. Adaptation measures should be implemented in short-term to offset negative impacts as shifting sowing dates, changing cultivars (earliness), revising soil management (conservation agriculture), fertilization, and plant protection practices, introducing irrigation. Crop management still offers a range of opportunities to cope with drought-prone conditions for sunflower cultivation. Without irrigation, the search and use of cultivars with lower base temperatures and shorter thermal times for emergence will become of great importance. Supplemental irrigation is an effective way to maintain or increase sunflower yield (and oil concentration) in dry conditions. More water in winter could be stored (dams, lakes, etc.) for securing summer irrigation where possible.

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# Predicting sunflower grain yield using remote sensing data and models

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Predicting grain yield (GY) and quality (oil, protein) a few weeks before harvest is of strategical interest for the cooperatives which collect, store and market grains. Several approaches have been used for that on various crops: (a) monitoring a network of representative fields using sensors and crop indicators; (b) running agroclimatic or process-based models at field level then upscaling the predictions at landscape level; (c) combining observations from remote sensing (satellite) and models (statistical or dynamic) for improving GY prediction. Climate variability and water deficit related to global warming both increase the uncertainty on model predictions. Correcting the predictions by assimilating vegetation and soil data into crop simulations is a way to reduce this uncertainty. Among field crops, sunflower has been scarcely used in remote-sensing studies in spite of the plasticity of green area index (GAI) and its key role in yield and oil achievement (Claverie et al., 2012; Merrien et al., 2012). In this study, we will combine remote sensing and two types of models (statistical and dynamic) to predict sunflower yield for a range of cultivars and crop practices.

In 2014 and 2015, 187 sunflower fields were monitored in Midi-Pyrénées (SW France) at different periods of the growing season and GY was provided by the farmers. GAI was estimated by the inversion of radiative transfer model (Baret et al., 2007) from satellite images (Landsat 8 and Deimos in 2014; Landsat 8 and Spot 5 in 2015) at 6 (2014) and 11 (2015) dates throughout the growing season. From these estimations, GAI<sub>max</sub> and GAD (Green Area Duration) on 10 August were calculated and used in several predictive linear models of GY. In parallel, the crop model SUNFLO (Casadebaig et al., 2011) was run on farmer's fields using soil conditions, daily weather, and management data (incl. variety). Both inversion and assimilation techniques based on GAI from remote sensing were used to correct the poor representation of environmental conditions (initial N and water conditions, soil type) and reduce the error on GY prediction. As statistical models used robust relations between GY and GAD and/or GAI<sub>max</sub>, a better performance was achieved with these models. However, assimilating GAI and correcting initial conditions improved the predictions of GY by SUNFLO but GY was still overestimated even after correcting it by limiting factors (weeds, fungal diseases, low plant population). Predicting oil will be the next issue to address with this approach.

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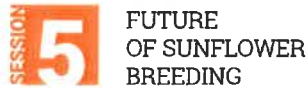
# Heliaphen, an outdoor high-throughput phenotyping platform designed to integrate genetics and crop modeling

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Heliaphen is an outdoor high-throughput phenotyping platform allowing automated management of growth conditions and monitoring of plants during the whole plant cycle. A robot moving between plants growing in 15L pots monitors plant water balance and phenotypes plant or leaf morphology, from which we can compute more complex traits such as the response of leaf expansion (LE) or plant transpiration (TR) to water deficit.

Here, we illustrate the platform capacities on two practical cases: a genetic association study for yield-related traits and a simulation study, where we use measured traits as inputs for a crop simulation model. For the genetic study, classical measurements of thousand-kernel weight (TKW) were done under water stress and control condition managed automatically on a sunflower bi-parental population. The association study on TKW in interaction with hydric stress highlighted five genetic markers with one near to a gene differentially expressed in drought conditions from a previous experiment. For the simulation study, we used the SUNFLO crop growth model to assess the impact of two traits measured in the platform (LE and TR) on crop yield in a large population of environments. We conducted simulations in 42 contrasted locations across Europe and 21 years of climate data. We identified ideotypes (i.e. genotypes with specific traits values) that are more adapted to specific growing conditions, defined by the pattern of abiotic stress occurring in these type of environments.

This study exemplifies how phenotyping platforms can help with the identification of the genetic architecture of complex response traits and the estimation of eco-physiological model parameters in order to define ideotypes adapted to different environmental conditions.



## Place of participatory breeding to promote a sustainable sunflower sector

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Sunflower is the second most cultivated oilseed crop in France by its area. Although it is a drought-adapted crop, work is to be done to adapt this important plant to climate change. Thanks to breeding programs, it is possible to start this challenge.

Thus, it is now essential to promote a sustainable agriculture that, at the same time, preserves the environment and remains productive for farmers and transparent to consumers. Finding a viable agriculture is sometimes slowed by broad-scope varieties which are not well adapted to local pedo-climatic conditions. Indeed, for years, the "classic" centralized selection has been associated to the development of high specialized and standardized productive systems, questioned today by the evolution of societal expectations and the need for adaptation to the environment and climate change. Today, genetic selection must maintain a crop biodiversity (of species and varietal) adapted to answer the necessity of resilience of production systems and others varied challenges. Participatory breeding development involves close farmer-researcher collaboration. Farmers need high value-added crops, so they are looking for genetic adaptations to local context and low inputs or organic systems. Moreover, local varieties can overcome diversity losses, biological vulnerability and a good capacity to be transformed. Participatory breeding can achieve these objectives by connecting breeders, farmers, researchers and transformers into a network.

Participative or decentralized selection can be described and classified in three models according to the place they give to farmers in the dynamics of selection. The consultative selection includes the farmers only at the end of the process, for real-condition tests. The col-laborative selection, integrates the farmers at various levels of the decision-making and ex-perimental processes. Finally, the collegial selection, the most well-known shape, is the "small-scale participatory breeding", that involves completely the farmers in all the processes of selection ; from the phase of determination of the objectives to the final choice of the variety. Even if there are not a lot of studies on participatory breeding upon sunflower, there is a concrete example in France, through the program launched in 2003 "Aquitaine cultive la biodiversité", that helps us highlighting the pros and cons of such a selection. Around thirty open-pollinated varieties of sunflowers were collected from farmers and networks of preservation of the genetic resources. With the aim to obtain technical references as a supplement to the observations in real conditions of cropping in the farms, ten varieties of sunflowers were sowed on a experimental platform in 2013.

Finally, the participative selection should enable, thanks to a continuous recultivation, to protect the germinal power of hundreds of thousand collected varieties, jointly in the fields of the farmers and on platforms of selection. So, it guarantees a dynamic management of the cultivated biodiversity. However, the diversity of stakeholders involved at the different scales of the sector, the regulatory aspect remains a major hindrance that it seems important to emphasize.

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