PhD School on Agriculture, Environment and Bioenergy

(http://sites.unimi.it/dottorato_aab/)

(XXXVI cycle, 2020-23)

Project draft

1.Field of interest

Patologia Agraria AGR/12, Genetica Agraria, AGR/07

2.Project title

Mechanisms of plant response to drought in maize

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4.Relevance of the topic and state of the art:

The maize species has always been considered as a model for genetic and cellular analyses and, since the advent of genomic technologies, it has become even more attractive. With a fully sequenced genome and the availability of different molecular tools, maize is nowadays considered one of the most accessible higher plant systems. Numerous gene models and molecular markers are available, along with seed stocks carrying different genetic variants (http://www.maizegdb.org). Maize is also a species of agronomic and cultural importance. In Europe its cultivation is widespread in Southern countries, including Italy, where environmental and social conditions are favorable. Many local accessions had been cultivated in small areas, isolated from other populations until the late 1950s, when they were almost completely replaced by the introduction of much more productive dent hybrids (1,2). However, the cultivation of some local varieties persists in specific locations. Although less productive, they show increased stability, accomplished through generations of selection for valuable functional traits. These genotypes may contain useful and untapped allelic variations and represent an important resource for the identification of novel alleles and haplotypes conferring resistance to environmental biotic and abiotic stress conditions (3). More recently, attention has also been given to the study of association between these varieties and specific microbial communities (4).

5.Layout of the project (draft)

This project aims at the identification and functional characterization of key genes controlling morphological traits and involved in plant response to drought conditions. It will also contribute to the characterization of the agrobiodiversity in maize for what concerns the capability to recruit endophytes, putatively involved in response against fungal infections, and tolerate drought stress conditions.

5.1 Materials & Methods

1. Gene functional studies. A group of mutants in genes controlling plant morphology and architecture and putatively involved in drought stress response is available in the laboratory of the proponents (5,6). For most of them the involved gene has been isolated. A detailed phenotypic characterization will be achieved by using various approaches, including physiological and cellular analysis. For one mutant, whose chromosomal position has been determined, a candidate gene approach will be undertaken to unravel the responsible gene. Response to drought will also be investigated during early stages of development in mutant and wild type sibling plants and gene expression profiles will be determined in referent inbred lines grown under different conditions. For the most interesting genes, allelic variants will be searched in a collection of maize accessions.

2. Effect of isolated endophytes on maize seedling growth. Specific bacterial strains isolated from local maize varieties will be inoculated to plants of the maize reference line B73 and other maize lines and the effect of the treatment will be determined at early stages of plant development. Plants will be grown under normal-watered as well as drought conditions. Treated and non-treated plants will be compared for plant growth and physiological parameters. Likewise, treated and non-treated plants will be experimentally inoculated with the plant pathogenic and toxin-producing fungus *Fusarium verticillioides* to compare the symptoms caused on the plant and mycotoxin content in the kernels, to identify candidate biocontrol agents against this pathogen.

5.2. Schedule and major steps (3 years):

First year

Activities
Mutant phenotypic and molecular analysis
Expected results
Description of the gene function; detection of a novel gene involved in plant development

Second year

Activities
1. Analysis of mutant response to drought. Analysis of gene expression in plant grown under limited water supply
2. Optimization of endophytes inoculation during seedling germination and growth
Expected results:
1. Detection of drought responsive genes
2. Protocol for bacterial inoculation in maize

Third year

Activities
1. Sequence analysis of key genes in local maize varieties
2. Analysis of the effect of isolated endophytes on seedling growth in normal and water scarcity condition
Expected results
Detection of variants in genes involved in drought stress response
Detection of bacterial strains beneficial to plant growth and response to drought and pathogen infection.

6. Available funds
COAT, GEMMA

6. Literature
1. Brandenburg J.T., Mary-Huard T., Rigaill G., Hearne S.J., Corti H., et al. (2017) Independent introductions and admixtures have contributed to adaptation of European maize and its American counterparts. *PLOS Genetics* 13(3): e1006666

2. Brandolini A., Brandolini A. (2009) Maize introduction, evolution and diffusion in Italy. *Maydica* 54: 233– 242.

3. Newton A.C., Akar T., Baresel J.P., Bebeli P.J., Bettencourt E., Bladenopoulos K.V., Czembor J.H., Fasoula D.A., Katsiotis A., Koutis1 K., Koutsika-Sotiriou M., Kovacs G., Larsson H., Pinheiro de Carvalho M.A.A., Rubiales D., Russell J., Dos Santos T.M.M., Vaz Patto M.C. (2010) Cereal landraces for sustainable agriculture. A review. *Agronomy for Sustainable Development* 30: 237–269.

4. Kroll S., Alger M.T., Kemen E. (2017) Genomic dissection of host-microbe and microbe-microbe interactions for advanced plant breeding. *Current Opinion in Plant biology* 36, 71-78

5. La Rocca N., Manzotti P., Cavaiuolo M., Barbante A., Dalla Vecchia F., Gabotti D., Gendrot G., Horner D., Krstajic J., Persico M., Rascio N., Rogowsky P., Scarafoni A., Consonni G. (2015) The maize *fused leaves1* (*fdl1*) gene controls organ separation in the embryo and seedling shoot and promotes coleoptile opening. *Journal of Experimental Botany* 66:5753-5767.

6. Castorina G, Persico M, Zilio M, Sangiorgio S, Carabelli L, Consonni G (2018) The maize *lilliputian1* (*lil1*) gene, encoding a brassinosteroid cytochrome P450 C-6 oxidase, is involved in plant growth and drought response. *Annals of Botany*, 227–238,