AEGIS Project 11_038

Brassica selection criteria for the identification of the Most Appropriate Accessions: relate to the *Brassica oleracea* of Iberian collection

Introduction

Unique *Brassica oleracea* crop types are cultivated on the Iberian peninsula; these include *B. oleracea* vars *acephala*, *costata* and *capitata*. Farming practices for these crops still involve the use of landraces, and various collecting missions have resulted in ex-situ collections held at BPGV and MGB. However the genetic diversity and relationship among accessions is not known, as the name of an accession is not necessarily indicative of its identity, and seed swaps among farmers mean that geographic and genetic distance are not necessarily related. This made it difficult to select accessions to be put forward as European accessions as duplication and genetic structure are not known.

The objectives of the project were:

i) apply the selection criteria previously proposed for *B. rapa* to the Iberian *B. oleracea* collection, maintained in their respective countries of origin, at BPGV and MBG;

ii) apply molecular markers (ITS and SSR) to understand the genetic variability of the Iberian landraces, to improve the crop types classification and to eliminate duplicates;

iii) propose MAAs from the Iberian Collection.

Methods

The criteria previously applied to *B. rapa* by members of the Brassica WG could not readily be applied to the Iberian landraces as they relied on the name of an accession being indicative of genetic uniqueness. Therefore, further investigations such as molecular marker assays and other data such as seed viability and stock levels would be required to distinguish among the landrace accessions in order to ensure that the best representation of genetic diversity is entered into the European collection.

Germination tests were carried out on a total of 248 accessions from BPGV and seed stock levels were checked.

In order to cope with the physical and financial limits of the project, the investigation was limited to 100 accessions. These were selected from *B. oleracea costata*, *acephala* and *capitata* landraces held at BPGV and MGB. The 100 accessions were selected from a limited geographic region; Entre Douro e Minho (Portugal) and Galicia (Spain) – westwards of around 8^{o} 00’ 00” W longitude. Within this region both collecting site and elevation were used as selection criteria; both can be regarded as markers for ecogeographical differences. This meant that the 100 accessions selected would be as diverse as possible. Additionally, the accessions put forward for selection by MBG had already been characterised for morphological and agronomic traits.

ITS and SSR markers were used to understand genetic diversity and relationships among accessions

Results/Conclusions

Both *B. oleracea capitata* and *acephala* accessions from Spain and Portugal were found in mixed clusters as a result of the ITS investigation. *B. oleracea costata* accessions clustered together and appeared to be genetically distinct.

Cluster analysis of the SSR results showed that all three crop types did not cluster independently, and no relationship between accession name or geographical origin was noted. Two accessions were regarded as probable duplicates as they were genetically similar to other accessions; these were removed from further consideration as MAAs. Both sets of markers gave useful information on the biology and origin of the different crop types. Some accessions did not yield enough high quality DNA for the genotyping and sequencing analysis.

A total of 42 accessions (18 held at BPGV and 24 at MBG) were selected as MAAs to be included in the European collection. This was based on:

- selecting accessions representing the genetic variability over all three crop types as revealed by the ITS and SSR work
- the germination and seed stock levels were over set thresholds as detailed in the full project report
- MBG used pre-existing characterisation data to inform the selection process
- Only accessions for which both SSR and ITS data were available – some accessions only had had data for one marker type

Molecular marker data have enabled more detailed selections of accessions to be made rather than those based purely on passport data, and have allowed the identification of duplicate accessions. This type of work is therefore an option when considering groups of potential duplicates for the European Collection, particularly for landraces where accession names are not informative.