##### **PRT-CGN-PG-109 PROTOCOL MULTIPLICATION AGRICULTURAL CRUCIFERS**

This protocol applies to all parties involved in the multiplication of CGN material.

**Introduction**

Multiplications have to fulfil minimum quality requirements to ensure maintenance of genetic identity and integrity (avoiding seed mixing among accessions and minimizing loss of diversity through genetic drift) and high seed quality (absence of diseases and sufficient germination ability).

Contamination with Genetically Modified Organisms (GMO) should be prevented.

*Any deviation from this protocol should be reported to CGN, after which it will be recorded by CGN in the Multiplication logbook (FOR-CGN-PG-002).*

**Multiplication**

Maintaining genetic integrity

* isolation
* Agricultural crucifers are largely cross pollinators. The material is therefore propagated per accession in multi-crop isolation fields, spaced at least 40 m apart. The isolation crop is rye or Triticale.
* population size
* Multiplication is done on about 80 plants, with a minimum of 25 to a maximum of 150 plants.
* In planting out, the required number of plants are chosen non-selectively from the population. However, plants that lag considerably behind in growth may be left out because their poor performance could mean that these plants eventually do not contribute to seed multiplication of the accession.
* The number of plants that have been used in multiplication is recorded for every accession. These data are copied into the Multiplication logbook.
* sowing
* The number of seeds to be sown is determined by CGN.
* If material germinates poorly or very slowly, these findings are recorded and copied into the Multiplication logbook.
* Vernalisation
* The CGN specifies the accessions to be vernalised and the minimum length of the vernalization period. These data are copied into the Multiplication logbook.
* Cultivation
* No selection is made. If an accession is more heterogeneous than what is expected based on the passport data, or if it is a mixture of different species, the CGN is notified. The CGN determines whether and how selection may be made. These data are copied into the Multiplication logbook.
* As soon as the silicles start to turn colour, the field is spanned with a net to prevent birds from eating them.
* pollination
* There is pollination by on-site insects.
* harvest
* All inflorescences with the mature seed are harvested and bagged by field number.
* All seed produced will be returned to the CGN.

Maintaining identity

* Characteristics
* During sowing, cultivation and harvesting, accessions should be clearly marked with a label indicating the field number. All field numbers assigned prior to seed sowing should remain unchanged up to and including harvesting and seed cleaning.

Maintaining seed quality:

* Monitoring
* Control against diseases and pests takes place. Diseases or pest problems are recorded and copied in the Multiplication logbook. In case of detection of diseases, which threaten proper seed multiplication, the CGN is alerted.
* Seed treatment after harvest
* Bags with harvested plants need to be pre-dried.
* The method of seed cleaning is being decided in consultation with CGN.

**Concluding actions**

* Recorded deviations from this protocol should be sent to CGN along with the seed. These records are copied into the Multiplication logbook.
* Harvested seed is sent to CGN as soon as possible, in any case no later than six months after harvest. The seed bags are identified by their field number and their CGN number.