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# Operational Manual

*„Mihai Cristea” Plant Genetic Resources Bank,  
Suceava, Romania*



*Banca de Resurse Genetice Vegetale Suceava*



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## 1. Germplasm Acquisition and Accessioning

Genebanks can obtain the germplasm they want to conserve through a number of different ways. Conducting collecting missions is possibly the best way of acquiring germplasm material in the most reliable manner. Germplasm exchange with other genebanks is a second route to add genetic diversity to the collection. Obtaining and storing germplasm from researchers and plant breeders is another route to acquire genetic material. Such acquisitions should be guided by a formal mandate that the genebank concludes with its host organization or government and that provides the basis for a genebank acquisition policy. The actual accessioning of acquired germplasm samples, i.e. formally including it into the collection with its unique accession number, is a complex process during which the curator has to check a number of aspects such as the verification of the identity of the material, the health status, the availability of pertinent information, etc. It is further understood that also legal aspects form part of this activity, e.g. was the material collected/obtained in legal manner, are there any restrictions on its use, etc.

### Box 1.1. Germplasm Acquisition and Accessioning

**GA1** – Briefly describe any formal mandate that your genebank might have concluded with or received from your “mother organization” (e.g. institute, governmental body).

*This description should include details on:*

- a) *which species you conserve and make available;*
- b) *who decides on what your mandate is and, if different,*
- c) *from whom do you received the mandate;*
- d) *the main aspects of the mandate; and*
- e) *legal considerations on PGR as foreseen in national legislation.*

The Suceava Genebank (SVGB) is a public agricultural research institution operating under the authority of the “Gheorghe Ionescu-Sisesti” Academy of Agricultural and Forestry Sciences, headquartered in Bucharest. Established in 1990, the Genebank has been officially mandated to preserve, under both long- and medium-term storage conditions, seed-propagated plant species that are critical to Romanian agriculture.

The Genebank’s collection encompasses a remarkable diversity of plant species, covering all agronomic categories relevant to national agriculture, including cereals, grain legumes, vegetables, industrial crops, forages as well as medicinal, aromatic, and ornamental plants.

This genetic repository, maintained under controlled temperature and humidity conditions, comprises over 25,000 accessions. Of these, 19,037 represent distinct genotypes, belonging to 597 taxa across 454 species, 234 genera, and 44 botanical families.

At the species level, the collection includes key agricultural crops, with *Zea mays* L. subsp. *mays* (maize) represented by 5,754 varieties, *Phaseolus vulgaris* L. (common bean) by 2,731 accessions and *Triticum aestivum* L. (common wheat) by 2,550 genotypes.

A significant portion of the collection—approximately 55%—is composed of traditional landraces, often referred to as "peasants seeds". These varieties, emblematic of Romania's biocultural heritage, have largely disappeared from cultivation due to the shift towards intensive agriculture and the widespread adoption of modern hybrids and varieties.

The genetic material is accessible under the terms of the Standard Material Transfer Agreement (SMTA) of the International Treaty on Plant Genetic Resources for Food and Agriculture, including non-annex I materials.

The Suceava Genebank also serves as the National Coordinator for plant genetic resource-related activities and acts as Romania's National Focal Point for implementing the International Treaty on Plant Genetic Resources for Food and Agriculture.

The Genebank operates in accordance with LAW No. 45 of March 20, 2009, which regulates the organization and functioning of the "Gheorghe Ionescu-Sisesti" Academy of Agricultural and Forestry Sciences and the agricultural, forestry, and food industry research and development system. Furthermore, it adheres to Government Decision No. 112 of March 10, 2018, issued by the Ministry of Agriculture and Rural Development. Article 3 of this decision specifies, in accordance with CAEN codes, the crop categories under the Genebank's mandate and its associated responsibilities.

To ensure operational excellence, the Suceava Genebank has implemented a quality management system in compliance with EN ISO 9001:2015. Additionally, it follows the SR 13572:2016 standard for the implementation of an innovation management system.

**GA2** – Specific agreements. Does your genebank have any specific formal agreements with other Genebanks regarding the conservation of specified germplasm?

This should include:

- a) whether or not your genebank has any international agreements to conserve specified germplasm on behalf of other countries,
- b) a specific region, and/or
- c) the world, and
- d) which crops or genebanks fall under these agreements?

The Genebank has no specific international/regional agreements for any plant species.

**GA3** – In case your genebank has a germplasm acquisition policy, what does the policy entail?

Please specify which crops or which geographic area, if applicable.

The strategy in this field has evolved over time, reflecting the institution's development and the changing priorities of germplasm users. Currently, three primary approaches are employed to expand the collections: acquiring reproductive material from public-sector breeding organizations, conducting collection, exchange activities with interested parties worldwide, and repatriating plant genetic resources from other collections.

Priority is given to collecting landraces, obsolete cultivars, and crop wild relatives originating in Romania, as these are highly vulnerable to genetic erosion. In the case of crop wild relatives, their limited representation in the existing collections further underscores their importance. Following these, breeding materials with known and valuable traits are also targeted for acquisition.

**GA4** – How do you verify the identity of the germplasm material received (e.g. relying on the donor's information, comparing material with other accessions, involving (taxonomic) expertise, etc.)?

For samples obtained from other Genebanks or breeding and research organizations, we rely on the information provided by the respective donors.

Samples collected during our own missions are subject to taxonomic verification by curators before being introduced into permanent conservation. This verification is conducted during the initial multiplication cycle, either in the field or within the Genebank's greenhouses or solariums.

**GA5** – Describe if and how you conduct an assessment of the various quality aspects of the seeds, tissue culture or plant material received. *This description includes:*

- a) *quality aspects related to the correct identification of a given accession, but also*
- b) *health*
- c) *purity aspects of the sample/accession), and*
- d) *use of a quality control system (e.g. ISO).*

Newly acquired biological material undergoes a visual inspection by both the curator and the phytopathologist to ensure it meets the required purity and health standards before being added to the collections. This process is conducted in accordance with standard procedures and aligns with the Genebank's ISO-certified quality management system.

**GA6** – Describe whether and how the SMTA is being implemented:

- a) *extent of materials covered by SMTA (crops, numbers of accessions)*
- b) *ways of SMTA implementation and documentation of transfers of PGR*
- c) *other aspects (e.g. monitoring, supervision).*

The Standard Material Transfer Agreement (SMTA) has been in use since 2007, ensuring that all accessions are provided in compliance with its regulations. This includes its application to non-Annex I species.

From a technical standpoint, we utilize Easy-SMTA, an IT tool developed by the ITPGRFA, to streamline the process for users of the Multilateral System (MLS). By accessing the ITPGRFA platform (<http://mls.planttreaty.org/itt/>), we prefer generating a click-wrap SMTA rather than the traditional signed SMTA in English. This approach allows recipients to electronically sign the agreement directly on the platform, facilitating efficient interaction.

The process generates original documents for both the provider and the recipient, which are securely stored on the platform. These documents are subsequently used for reporting purposes to the ITPGRFA, ensuring compliance and transparency.

### **Box 1.2. Germplasm Collecting**

**GC1** – Describe here the details of the strategy that you follow in implementing germplasm collecting missions.

*This description should include:*

- a) *general aspects of planning and implementing a collecting mission,*
- b) *the criteria you use for priority setting;*
- c) *the actual strategy followed in sampling material from farmers' fields, from nature, etc.; and*
- d) *how your germplasm acquisition policy underpins the mission.*

Each collecting expedition is planned and conducted in alignment with the *Annual Research and Development Plan*, ongoing projects, and the budget allocated for this activity.

Priorities for collection are determined after analysing data from the BIOGEN database to address gaps in the collections, whether in terms of species, varieties, or geographical representation. Updated information on key agricultural crops, as well as traditional varieties still cultivated in the region, is gathered from local authorities or agricultural consultants. Collection missions are typically scheduled for March to April, prior to sowing when biological material has not yet been utilized, or in autumn, during or after the harvest. Sampling is carried out from farmers' fields or gardens, household or farm stores, and local markets. Alongside passport descriptors, the collection process also involves documenting local knowledge using a dedicated questionnaire for on-farm descriptors.

Weedy and crop wild relatives are primarily collected from natural reserves, with the consent of the custodians of these protected areas. This process is conducted in compliance with the provisions of the International Code of Conduct for Germplasm Collecting and Transfer (FAO, 1994).

As outlined in Government Decision 112/2018, Article 3, Paragraph (1), the Gene Bank's primary mandate includes "the exploration, inventory, collection, research, and conservation of phylogenetic resources...."

**GC2** – Provide any additional information on the germplasm collecting activities of your genebank, including the collaboration with others.

Collecting and exploration missions are also conducted with mixed, multinational teams, established through bi- or multi-lateral agreements that are approved by the relevant authorities in the participating countries.

## 2. Ensuring Security

### Box 2.1.1. Safety Duplication (of long-term conserved germplasm)

**SD1** – Please describe how your genebank implements the safety duplication of your germplasm material.

*This description should include the following aspects:*

- a) *the type of safety duplication (e.g. black-box; no specific arrangement; other);*
- b) *the location(s) where you store your safety-duplicates (country; genebank);*
- c) *whether or not you are using a formal agreement with the genebank(s) that store your duplicates?*
- d) *whether the safety-duplicates are stored under conditions comparable to your own? Please provide details;*
- e) *do you maintain safety-duplicates from other genebanks at your genebank? If so, do you know any details of that material?*

Unique accessions collected from Romania, along with all non-confidential material of local or foreign origin, are sent to the Svalbard Global Seed Vault for conservation under identical storage conditions. These materials are stored as "black boxes" based on a formal agreement. The first shipment to the Seed Vault was made in early 2020.

Currently, most of the collection is duplicated at Romanian institutions that have provided seed samples to the Suceava Gene Bank. However, apart from the Svalbard Global Seed Vault, there are no formal storage agreements with other Genebanks.

Additionally, no agreements are in place for the storage of duplicate samples on behalf of other Genebanks.

**SD2** – Do you have a safety duplication policy? If so, please provide essential details.

The Genebank continues the process of duplicating the base collection, by obtaining freshly regenerated material, preparing it and sending it to Svalbard Seed Vault.

The plan is that all AEGIS accessions to be duplicated in the world collection in Svalbard Seed Vault.

### Box 2.1.2. Structure

**SS1** – Please provide details on how your genebank building has been designed to resist natural disasters (e.g. earthquakes; flood; storm).

The Genebank is situated in Suceava, a town in the northeastern part of Romania, at an altitude of 325 meters, within a geologically stable region. The building has been

specifically designed to withstand earthquakes exceeding 7 on the Richter scale, ensuring its structural resilience.

Romania's temperate climate minimizes the risk of extreme environmental events such as monsoons, typhoons, or hurricanes. Additionally, the Genebank is located at a safe distance from the nearest running water, the Suceava River, which is approximately 3.4 kilometers away in a straight line. The building is positioned 87 meters higher in elevation than the river, providing effective protection against potential flooding.

**SS2** – Please describe the security arrangements that you have in place to protect your genebank against burglars, fire and others.

*Please include details on the following arrangements, as applicable:*

- a) *fences;*
- b) *security doors;*
- c) *alarm system;*
- d) *fire detectors;*
- e) *standby generator;*
- f) *others (please specify).*

The institute's perimeter is enclosed by fencing that separates it from the public areas. A video surveillance system monitors the building and access points, extending its coverage to the genebank and greenhouse areas. Smoke detectors are installed indoors, and each room is equipped with one or two fire extinguishers to facilitate fire response. The fire protection system is further reinforced by both interior and exterior hydrants. Additionally, the compressors for the cold rooms are supported by an electrical backup system powered by a standby generator.

**SS3** – Please provide information on any other structural security aspects that you might have in place.

None.

### **Box 2.1.3. Security Equipment**

**SE1** – Provide details on the kind of emergency (back-up) equipment or arrangements that you have in place to ensure permanent electricity and cooling. *Aspects to consider are:*

- a) *“back-up” compressors for your cold rooms;*
- b) *generator;*
- c) *regular maintenance and trial runs;*
- d) *other.*

The Genebank maintains a spare compressor for each type used in the cold rooms to ensure uninterrupted operation. The main power supply is supported by an automatic standby generator, which activates immediately in the event of a power outage or when the main power is turned off. A modern system for monitoring preservation conditions is in place, which sends alert notifications via email and phone in case of any irregularities.

Daily, the compressors and cold rooms are inspected by the Genebank's specialized engineer, and all parameter changes are recorded and stored by a computer program. Regular maintenance is performed on the compressors currently in operation, while periodic tests are conducted on those designated for cold chambers that are not yet in use. Additionally, the operating status of the generator is checked and documented weekly to ensure its reliability.

**SE2** – Describe how you monitor temperature and relative humidity in your cold stores and drying room.

Since seed samples are stored in airtight containers, the relative humidity of the cold rooms is not controlled. However, the temperature is strictly monitored, with values recorded by a computerized system.

A monitoring system has been implemented to track the parameters of the cold rooms. This system collects data every 15 seconds, including internal conditions (e.g., temperature) and the operational status of key components such as the compressor (on/off), fan (on/off), and defrost cycle (on/off). For freezing installations, the system also monitors the door status. All collected data is stored in a database for purposes such as auditing, graphical analysis, and sending alert emails when any critical parameter exceeds the acceptable range.

In the drying chambers, relative humidity and temperature are monitored using dehumidifier sensors, which are cross-checked with independent devices. These independent devices also record the evolution of relative humidity and temperature values over time.

Further details regarding the thermo-hydric conditions can be found in Box 3.1.3.A: Seed Storage Conditions.

#### **Box 2.1.4. Institutional and Personnel Security**

**IPS1** – Provide details on the “institutional security”, in particular with respect to the provision of financial means to operate the genebank *Aspects to consider are:*

- a) *timely transfer of funds from the “mother” organization to the genebank;*
- b) *do you have direct access to the “mother” organization that provides the budget?;*
- c) *internal “security” of accessing these funds;*
- d) *long-term security and stability of funding (compensation of inflation rates, avoiding variation in years)*
- e) *any other observations that are relevant in this context.*

The Gene Bank operates as a public institution, primarily funded through the state budget to support its core activities and staff salaries. Additional funding can be secured through extra-budgetary sources, such as research projects. The annual budget is developed based on proposals submitted by the Gene Bank and subsequently approved by the “Gheorghe Ionescu Sisesti” Academy of Agricultural and Forestry Sciences in Bucharest. There are no concerns regarding the security of funding for the institution.

**IPS2** – Describe how you secure adequate staffing of your genebank.

The institution operates under the provisions of Government Decision No. 112/2018 and has an organizational structure comprising 25 positions. Currently, 23 of these positions are filled by individuals employed under permanent individual employment contracts.

#### **Box 2.1.5. Contingency Plans**

**CP1** – Describe the kind of emergency or contingency plan that your genebank has in place to cope with disaster situations.

The Genebank has an accredited contingency plan, approved by an authorized state institution. This plan incorporates administrative and organizational measures designed to mitigate a wide range of risks. It includes provisions for information and property security, an evacuation strategy for the institute's staff and assets, and a framework to ensure the resilience and continuity of the institute's activities.

**CP2** – Provide information on the kind of training, security drills and other activities that your genebank gives to its staff to deal with emergency situations, if any.

The Genebank has implemented a system of periodic training and information sessions for staff, focusing on security risks, emergencies, and health risks, in compliance with occupational safety and fire prevention regulations. An annual Program of Measures, which includes both technical and organizational components, is developed to address these areas.

Employees undergo emergency training at least every six months, while members of the intervention team receive training monthly. Additionally, competent institutions conduct periodic inspections of the facilities, the organization of activities, and compliance with relevant regulations to ensure adherence to safety and security standards.

### **3. Germplasm Maintenance**

#### **3.1. Maintenance of Viability**

##### **A. Seed Collections**

##### **Box 3.1.1.A. Initial seed viability**

**IV1** – Describe the procedures or practices that you have in place to ensure the highest possible initial viability of your seed, in particular during regeneration and post-harvest (e.g. cultivation practices, pollination aspects, use of specific equipment as shelters, storage of harvested seeds, cleaning, etc.).

During regeneration or multiplication activities, all procedures are carefully followed to meet the biological requirements of each species. This includes measures such as isolating cross-pollinated genotypes, maintaining appropriate distances between plants, addressing agrotechnical needs, and applying other methods to ensure an initial germination rate of over 85%.

All regenerated or multiplied crops are manually harvested at the physiological maturity stage. Post-harvest procedures vary depending on the biology of the species. For example, vegetable fruits such as tomatoes, peppers, eggplants, cucumbers, and cucurbits are stored in a post-maturation room on shelves at room temperature for 2 to 10 days before seed extraction.

As a general practice, after threshing, all samples are placed in the first drying room. From there, individual samples are cleaned and transferred to the second drying room to complete the drying process. Before packaging and storing the seeds in freezers or refrigerators, sub-samples are taken to test their germination capacity.

**IV2** – Describe procedures how you deal with a) dormancy and b) hard seeds.

Various methods are employed to dormancy breaking and facilitate germination of hard seeds, following the procedures outlined by the International Seed Testing Association (ISTA). These methods include techniques such as scratching, pre-cooling or heating, and wetting seeds with solutions of gibberellin or potassium nitrate. These approaches are designed to enhance germination and ensure accurate testing results, as per ISTA's standardized guidelines.

**IV3** – Please provide any other information on procedures that you follow to ensure highest possible initial viability.

None.

#### **Box 3.1.2.A. Seed Viability Monitoring**

**VM1** – Describe the routine seed viability monitoring system that you use.

*The monitoring system should include the following aspects:*

- a) *frequency of testing;*
- b) *sampling method applied;*
- c) *any thresholds that you use;*
- d) *whether you apply different procedures for crops/species with erratic initial viability or irregular viability lifespan;*

Seed lots intended for both the active and base collections originate from the same source. Viability testing is conducted every five years on seeds from the active collection, which are currently stored at +4°C in glass jars with lids. If significant declines in germination rates are observed, seeds from the same samples stored in the base collection—maintained at -20°C in aluminum foil bags—are also tested.

The Viability Assessment Laboratory selects samples for monitoring by applying specific filters based on the latest germination test results recorded in the BIOGEN database. A corresponding Excel list is then provided to the Conservation Laboratory. An appropriate quantity of seeds, determined by species, size, and availability, is delivered to the designated researcher for testing.

Viability tests are conducted under controlled conditions in a growth chamber (Binder KBW / KBWF 240), following both Gene Bank and ISTA standards. For most cultivated

species, the initial germination percentage must exceed 85%, while for wild species, it is approximately 60%, as per international guidelines.

Further details on germination testing procedures are outlined in the Genebank's ISO Quality Management System.

**VM2** – Please describe the information “system” that you might have in place that allows you to make more species -or even accession- specific decisions regarding when the next monitoring should take place.

Following the regeneration or multiplication phases and subsequent seed processing, viability data, along with the number of regeneration cycles, are recorded in the BIOGEN database. If the seeds fail to meet the minimum germination standards required, the material remains under the supervision of the collection curator to undergo another regeneration or multiplication cycle.

For monitoring seed samples included in the collections, files indicating the accessions to be verified can be generated from the BIOGEN database upon request, based on the reference year.

**VM3** – Please provide information on non-specific thresholds that you might use for viability of seeds (i.e. percentage of germination) and for the amount of seeds left of an accession to initiate regeneration. In case you differentiate between self- and outbreeding species, please answer for each category separately.

Regardless of the species' pollination type, whether the material is collected locally or obtained through international exchanges, regeneration and multiplication are prioritized. This process proceeds irrespective of the germination capacity or seed quantity, which often fail to meet the standards required for permanent conservation.

#### **Box 3.1.3.A. Seed Storage Conditions (for the different types of collections, i.e. short/medium- or long-term storage)**

**SC1** – Please provide details on temperature and relative humidity conditions of your storage and drying rooms. In case they vary from room to room, please provide details for each.

The drying of biological material is conducted until a moisture content of 5–6% is achieved. This process utilizes dehumidifiers that lower the relative humidity in the drying chambers to levels between 10–15%, while maintaining an ambient temperature below 20°C. The moisture content of the seeds is periodically monitored using an infrared thermobalance.

Both drying chambers operate under identical temperature and humidity conditions.

The seed storage chambers for the active collection are maintained at a thermostatically controlled temperature of +4°C, while those for the base collection are set at -20°C. Relative humidity (RH) is not regulated in either storage environment.

**SC2** – Provide details on the type of containers and the packaging procedures (and the corresponding equipment, if any) that you use.

Previously, seed samples in the active collection were stored in glass jars with threaded caps containing rubber sealing rings. However, in 2024, a transition began to change the packaging type for these samples. The seeds are now being transferred from glass jars to aluminum foil bags.

For the base collection, seed samples are packaged in aluminium foil bags, designated for specific purposes such as germination, regeneration, or as residual samples, to facilitate easier handling. Each package is labeled with double tags and a QR code: one label is attached to the exterior of the bag, while the other is placed inside with the seeds.

The packaging process for the tri-laminated aluminium foil bags is carried out using a vacuum and sealing device.

**SC3** – What is the range of seed moisture contents (smc) of your stored seeds of different species; what measures do you apply to keep and/or monitor the (low) moisture level? Do you treat different species differently?

The seeds are arranged in trays in a thin layer, accompanied by a label containing their identity data, which remains with the biological material throughout the process. The seeds are dried to a moisture content of 5-6%, regardless of the species. Moisture content is determined using a small sample of the material, which is ground and weighed before and after infrared drying, with the moisture level expressed as a percentage.

Once the desired moisture level is achieved, packaging is carried out promptly. Long-term maintenance of this moisture level relies solely on the airtight sealing of the containers, as no monitoring system is in place to track changes over time.

**SC4** – Provide data on the total storage capacity (number of containers, number of accessions) and an estimated percentage to which extent this capacity has been filled.

The Suceava Genebank is equipped with facilities for both the active and base collections. For the active collection, maintained at +4°C, there are four conservation rooms, each covering an area of 23 square meters. These rooms are fitted with a total of 15 racks, each containing 12 shelves, capable of storing between 30,000 jars of 700 ml or 45,000 jars of 400 ml.

For the base collection, stored at -20°C, the facility includes three thermally insulated cells, each with an area of 15 square meters. Each cell is equipped with three racks (two fixed and one movable in the center) and approximately 120 drawers per cell. Depending on the size of the seeds and the number of aluminum foil bags, each drawer can hold between 200 and 1,000 packages.

As of January 2025, the storage capacity utilization of the Suceava Genebank is approximately 60% for the active collection and 25% for the base collection. The transition to new packaging in the active collection is being implemented to increase storage capacity and improve the sealing of the packages. This change helps

preserve the initial moisture content and maintain the viability of the seed samples over time.

**SC5** – Please include any other aspects regarding storage conditions at your genebank that you regard as important (e.g. anticipated lifespan of freezing and drying equipment and related prudent financial management).

There is no additional data relevant to seed conservation.

## **B. *In vitro* Culture Collections**

### **Box 3.1.1.B. Initial viability**

**IV1** – Describe the procedures or practices that you have in place to ensure the highest possible initial viability of your plant material, in particular during culture of donor plants (e.g. cultivation practices [field, greenhouse], phytosanitary pretreatments, like use of pesticides).

The *in vitro* culture laboratory, established in 1991, was initially designed for the cultivation of *Solanum tuberosum* (potato) as its primary species. In 2020, the research plan was expanded to include *Allium sativum* (garlic).

The potato and garlic collections maintained in the Bank's field serve as the primary source of biological material for initiating *in vitro* cultures, with additional material sourced from collection missions.

All activities related to the maintenance of field collections are conducted in strict compliance with technical and technological requirements, including soil preparation, maintaining appropriate spacing between and within rows, and ensuring optimal planting times. During the vegetation period, specific measures are implemented to promote the growth of vigorous, healthy plants and high-quality planting material. These measures include weed removal, soil loosening, hilling (for potatoes), and the application of treatments to control diseases and pests.

The selection of genotypes for inclusion in the *in vitro* collection is based on field observations conducted in the genebank and the morphological characteristics of the vegetative organs.

**IV2** – Describe procedures of explant isolation (organ source in the plant, manipulations) and sterilization (chemical and handling) of the explants.

To minimize the risk of contamination during the initiation phase of *in vitro* culture, potato tubers are transferred from the storage room to the laboratory. They are thoroughly washed, inspected macroscopically, placed on shelves, and allowed to sprout.

The preparation and inoculation of potato meristems are carried out once the shoots begin to grow, ensuring the process is completed before the tubers show signs of wilting or dehydration. This procedure is performed under aseptic conditions using a binocular magnifier within a laminar flow hood that provides sterile air.

For disinfection, the apices of potato shoots grown in the laboratory are treated with 70% alcohol for one minute. Similarly, garlic bulbils are disinfected by soaking them in 96% alcohol for 90 seconds.

In both cases, the biological material is rinsed three times with sterile distilled water following disinfection to ensure the removal of any residual alcohol.

**IV3** – Please provide any other information on procedures that you follow to ensure highest possible initial viability.

The selection of biological material, which serves as the foundation for all inoculations, is conducted based on an assessment of its health and vigour.

### **Box 3.1.2.B. Viability Monitoring**

**VM1** – Describe the routine *in vitro* viability monitoring system that you use.

*The monitoring system should include the following aspects:*

- a) *regular control of contamination events,*
- b) *control of hyper-hydricity,*
- c) *control of health state (if different from a above),*
- d) *etc.*

Infections during both the initiation and later stages of *in vitro* culture are relatively rare. Observations are conducted daily during the initiation phase, while for the multiplication and conservation stages, monitoring occurs on a weekly or monthly basis, respectively.

Over the years, cases of hyper-hydricity in potatoes have been extremely limited.

Research on the micromultiplication and preservation of garlic varieties is ongoing. Instances of hyper-hydricity have been observed accidentally in a small number of inoculums, which exhibited abnormal morphological development.

**VM2** – Describe the information “system” (i.e. an “expert system”) that you might have in place that allows you to make more species - or even accession-specific decisions regarding when the next monitoring should take place.

There is no formal monitoring system in place for the plantlets. The protocol relies solely on periodic examinations of the plantlets and the expertise accumulated over time.

Special attention is given to experimental variants where modifications are required, such as changes in the composition of the culture medium, the type of explant, or the type of culture flask used. These adjustments are carefully observed and evaluated to optimize the outcomes of the *in vitro* culture process.

**VM3** – Please provide information on non-specific thresholds that you might use for vigour of *in vitro* cultures (i.e. multiplication rates, loss by weak growth) and for the amount of culture vessels (tubes, jars) left of an accession to initiate additional multiplication measures.

There is significant variability in the vigour of *in vitro* plantlets across different local varieties, similar to the variability observed in plants grown in the experimental field.

Key morphological traits, such as leaf size, shoot diameter, shoot length, shoot number, and the ability to regenerate microtubers, provide valuable insights into the performance of a variety on a specific culture medium. Additionally, the presence or absence of necrotic leaves or microtubers showing signs of senescence can influence the multiplication rate and regeneration capacity during subcultures.

Typically, each genotype grown on a specific culture medium is initiated with four vials, each containing 20 inocula. If two vials need to be removed due to issues, they are promptly replaced to maintain the collection and prevent the loss of the variety. This approach ensures the preservation of genetic diversity and the continuity of the *in vitro* collection.

### **Box 3.1.3.B. Storage Conditions (for the different types of collections i.e.**

#### **short/medium- or long-term storage)**

**SC1** – Please provide details on light, temperature and relative humidity conditions of your culture and storage rooms, as applicable. In case they vary from room to room, please provide details for each.

In the growth room, culture vessels are maintained at a temperature of 20–22°C, with a photoperiod of 16 hours under white LED light, providing an intensity of approximately 1800–2000 lux.

In the *in vitro* conservation chamber, the temperature is maintained at approximately 6–10°C (most often between 6–7°C), occasionally reaching 10°C during hot summer days. The photoperiod is set to 10 hours of light per 24 hours, with a light intensity of 1000–1200 lux generated by cool white LED tubes.

Humidity levels are not controlled.

**SC2** – Provide details on the type of cultivation vessels (tubes, jars, plastic vessels etc.) and the transfer procedures (including the corresponding equipment, if any) that you use.

For the inoculation phase of individual meristems, small glass vials with dimensions of 2 cm in diameter and 7.5 cm in height are utilized.

During the micromultiplication and preservation phases, larger glass jars sourced from the food industry, with diameters of 5–6 cm and capacities ranging from 170 to 220 ml, are employed. In recent years, Magenta bottles with dimensions of 77×77×97 mm have also been introduced into the laboratory's workflow.

The *in vitro* culture laboratory is fully equipped with all the necessary facilities to support its activities. These facilities are organized to ensure a seamless technological flow, starting from washing and sterilization, progressing to the sterile room, and finally to growth or preservation under slow-growth conditions.

The laboratory is equipped with a range of specialized tools and devices, including a laboratory glassware washing machine, oven, autoclave, water distillation and bidistillation units, culture vessels, a laminar flow hood for sterile air, a binocular magnifier with a camera, tools for handling explants, and air-conditioned rooms for

growth and conservation. These resources enable the laboratory to maintain high standards of efficiency and precision in its operations.

**SC3** – Please include any other aspects regarding *in vitro* culture and storage conditions at your genebank that you regard as important.

The availability of the conservation chamber and the reliable operation of thermostatic systems maintained at 6 – 10°C are particularly beneficial for extending the interval between subcultures. Additionally, they help prevent the onset of senescence, even when using culture media without growth inhibitors.

## D. Field Genebank Collections

### Box 3.1.1.D. Initial viability

**IV1** – Describe the procedures or practices that you have in place to ensure the highest possible quality of your planting material, in particular during the growing from donor plants (e.g. cultivation practices in the field or greenhouse], phytosanitary pre-treatments, etc.).

At the Suceava Genebank, field genebank conservation is employed for collections of potato (*Solanum tuberosum*), garlic (*Allium sativum*), and certain onion varieties, including *Allium cepa* var. *aggregatum* G. Don (potato onion) and *Allium cepa* - Proliferum Group.

Plant material samples, such as tubers, bulbils, and bulbs, regardless of their origin, undergo a macroscopic inspection to identify and remove any material affected by diseases or pests. No pre-treatments are applied to the material before planting.

**IV2** – Describe any particular procedures you use (e.g. which organ of the donor plant you use to reproduce the planting material).

Potato is propagated through tubers, while garlic and certain onion varieties, such as *Allium cepa* var. *aggregatum* and the *Allium cepa* Proliferum Group, reproduce vegetatively through bulbils and bulbs.

**IV3** – Please provide any other information on procedures that you follow to ensure highest possible initial quality.

No other information.

### Box 3.1.2.D. Viability Monitoring

**VM1** - Describe the routine field genebank monitoring system that you use.

*The monitoring system could include the following aspects: regular control of disease or pest contamination, other types of damages to the plants, etc.*

Each year, the area selected for the regeneration and multiplication of accessions is carefully chosen to accommodate the number of genotypes and to meet crop rotation requirements. This approach helps prevent or minimize the incidence of diseases and pest attacks specific to the crops.

The collection is planted in the field following all technological cultivation norms, including soil preparation, spacing between rows and plants within the same row, and adhering to the optimal planting schedule.

The samples to be planted are assigned an identity label, in accordance with the lists recorded in the field register. Prior to planting, all samples are individually inspected to remove any biological material showing signs of damage caused by diseases or pests.

During the vegetation period, starting from the plant emergence phase, various morpho-physiological characteristics are documented based on the specific descriptors for the crop. Additionally, observations are made regarding resistance to diseases and pest attacks.

**VM2** – Describe the information “system” that you might have in place that allows you to make more species- or even accession-specific decisions regarding when the next monitoring should take place.

There is no system specifically adapted to any particular species or variety.

**VM3** – Please provide information on non-specific thresholds that you might use for the quality of the individual plants (e.g. loss by weak growth) and for the amount of plants of an accession left in the field before additional initiating multiplication measures.

Not applicable.

#### **Box 3.1.3.D. Maintenance Conditions**

**SC1** – Please provide details on your cultural practices (e.g. cultivation practices; pruning; irrigation; protection against animals, etc.; pest and disease management; etc. applied to your field genebank material.

All field activities, including weed removal, soil loosening, hilling for potatoes, and treatments for diseases and pests, are performed at optimal times. These practices ensure the development of vigorous, healthy plants and the production of high-quality planting material.

**SC2** – In the case of annual or sub-perennial species that cannot over-winter in the field genebank, what measures do you take?

Samples are harvested once the material reaches physiological maturity and weather conditions permit the collection of clean, soil-free storage organs. The harvested samples are stored in the vegetative material conservation chamber under controlled temperature (5–7°C) and humidity (85–90% relative humidity).

Garlic and onions are planted in the experimental field in autumn, typically at the beginning of October.

**SC3** – Please include any other aspects regarding field genebank maintenance conditions at your genebank that you regard as important.

No other information.

## 3.2. Maintaining Genetic Integrity

### A. Seed Collections

#### Box 3.2.1.A. Seed Containers and Sample Size

**SCSS1** – Do you document the initial number of seeds of individual accessions (either as received from collecting missions or through exchange)?

For accessions obtained through collecting missions, the initial stock is documented only if the accession meets all the necessary conditions to proceed directly to conservation, without requiring prior multiplication in the experimental field.

For material received from breeding collections, documentation focuses on recording the number of seeds provided.

**SCSS2** – Please describe what kind of containers (and equipment) you use, the procedure you follow with respect to sub-sampling, seed numbers per container, etc.

Until 2024, seeds from accessions in the active collection were stored in glass jars with screw caps, while those in the base collection were packaged in aluminum foil bags. However, a transition to aluminum foil bags for the active collection began and is still ongoing. This shift not only optimizes the use of available storage space but also ensures that seeds are stored under conditions that minimize deterioration, thereby supporting the long-term conservation of genetic resources.

The number of seeds in each accession is determined either by direct counting, using a seed counter device, or by calculating based on the mass of 1,000 grains. The measured quantity is recorded for each package and documented in the database to ensure accurate inventory management.

For the active collection, each accession is placed into 1 to 3 separate sub-samples, while for the base collection, seeds are packaged in 3 to 6 aluminum foil envelopes, depending on the available quantity of seeds.

Seeds from the same accession, but obtained from different regeneration or multiplication cycles, are stored in separate containers.

**SCSS3** – What is the number of seeds that you use as the minimum threshold per accession? Are these seed numbers of a given accession based on genetic parameters (such as reproduction biology; heterogeneous samples)? Please provide URL of your protocols if these are available online.

The minimum number of conserved seeds is determined by the biological characteristics and reproductive system of the accession. For local populations, wild species, and allogamous plants with small to medium-sized seeds, the required quantity ranges from 2,000 to 15,000 seeds.

In contrast, modern cultivars and autogamous species are typically represented by fewer seeds, ranging from 1,000 to 5,000, depending on seed size. Breeding materials,

which are confidential, are stored exclusively in the base collection with a minimum requirement of 300 seeds.

For genetic stocks provided by breeders and old landraces no longer under cultivation, there are no specific limits set on the number of seeds to be conserved.

**SCSS4** – Please provide details on other aspects that are important in this context.

None.

#### **Box 3.2.2.A. Pollination Control**

**PC1** – Please describe the regeneration procedures that you follow for self- and outbreeding species.

*Please include in your description the following aspects:*

- a) *any control measures to minimize or avoid cross-pollination between accessions;*
- b) *the use of pollination cages for insect-pollinated species;*
- c) *the use of specific pollinators for insect-pollinated species;*
- d) *strategies to ensure that males and females participate equally in the reproduction;*
- e) *strategies to avoid any genetic drift (minimum number of plants, minimum number of plants at flowering stage before pollinators introduction, similar quantity of seeds harvested from each plant, etc.*

For regeneration and multiplication activities, we utilize both our in-house facilities and collaborations with selected breeding institutions, particularly for allogamous species.

In the case of entomophilous species, insect cages with *Bombus terrestris* are employed to ensure effective pollination.

For wind-pollinated species, pollination is facilitated through the manual application of vibrations. In cucurbit and sunflower genotypes, artificial pollination and isolation are accomplished using paper or textile bags.

Maize, as the most numerically significant and historically important crop in our collection, receives special attention and is included annually in the regeneration and multiplication plan. Pollination in maize is controlled through various methods, such as spatial isolation using hemp curtains or individual isolation of female flowers, followed by manual pollination with a pollen mixture. To prevent genetic drift, a minimum of 50 plants per accession are maintained.

**PC2** – Provide any other relevant information on procedures that you apply to control pollination of your germplasm.

None.

#### **Box 3.2.3.A. Regeneration Environment and Procedures**

**RE1** – Describe the regeneration environment and conditions that you apply. If applicable, you might want to distinguish between different types of germplasm (e.g. wild relatives, landraces, modern varieties, breeding material, genetic stocks, etc.). *Consider the following aspects:*

- a) *in how far are the environmental conditions of the current regeneration of individual germplasm accessions comparable to the environmental conditions that existed at the original collecting or breeding site?*
- b) *do you use controlled environments?*
- c) *do you collaborate with other Genebanks in Europe?*
- d) *others.*

Most species in the collections of the Gene Bank can be regenerated or multiplied under the pedo-climatic conditions of the area where the institution is located.

Generally, only a small number of wild species are included in regeneration or multiplication programs. Instead, most varieties of medicinal, aromatic, and fodder plants are recollected from their areas of origin.

Genetic stocks and most breeding lines are regenerated by the donor breeding institutions, as these collections are often protected by intellectual property rights (IPR).

Currently, there are no collaborations with other gene banks in Europe in this field. However, at the national level, a regeneration and multiplication network has been established for local vegetable varieties, with funding provided by the Ministry of Agriculture and Rural Development.

**RE2** – Please include any other relevant points on regeneration environment.

None.

#### **Box 3.2.4.A. Seed Processing Procedures**

**SPP1** – Describe the protocol(s) that you use for threshing and seed cleaning.

Accessions harvested from the experimental field or greenhouses, whether as seeds or fruits, are collected once they reach physiological maturity. These are packaged in labeled paper or raffia bags and sent to the seed processing facilities. The processing respects the physiological and technological requirements of each species, such as natural drying to achieve a seed moisture content of 12–14% and post-maturation of the fruit, among other needs.

The Genebank is equipped with various types of selectors to ensure optimal cleaning of plant residues. However, before the seeds are handed over to the conservation laboratory, they also undergo manual cleaning to ensure the highest quality.

**SPP2** – Describe the protocol(s) that you use for seed drying, including whether you use different drying procedures for different types of species.

The drying process for all seed accessions is conducted in two dedicated drying chambers, one measuring 30 m<sup>2</sup> and the other 20 m<sup>2</sup>. These chambers are equipped with professional-grade dehumidifiers designed to maintain optimal conditions, specifically a relative humidity of 10–15% and temperatures below 20°C.

**SPP3** – Please describe how you keep the time between harvesting and the actual (long-term) storage of seeds as short as possible.

Following harvesting or collection, all seed accessions undergo immediate processing, which includes threshing and transferring the seeds to the first drying room, the largest one measuring 30 m<sup>2</sup>. From this room, individual accessions are gradually processed through either mechanical or manual cleaning. This stage is typically completed by the end of the same year in which the regeneration, multiplication, or collection of the biological material occurred.

After cleaning, the seeds are moved to the secondary drying chamber, where they remain for a period of 1 to 3 months. During this time, the seed moisture content is reduced to the target level of 5–6%.

Once drying is complete, germination tests are conducted in the Genebank's laboratory to evaluate seed viability. Based on the results of these tests and the available seed quantity, the accessions are packaged and distributed across one to three of the Genebank's collections: active, base, and duplicate (Svalbard Global Seed Vault).

**SPP4** – Please describe how and where you store (in a temporary manner) newly harvested seeds.

*Please provide details on the temperature and relative humidity of the storage room/space; what type of containers do you use, if any.*

One of the drying rooms is designated for the temporary storage of materials received by the Genebank, regardless of species or source. These materials may originate from field genebanks, collecting missions, or donor institutions. The second drying room is specifically used to complete the drying process, where samples are organized according to the size and chemical composition of the seeds.

Both drying rooms operate under controlled conditions, maintaining a relative humidity of 10–15% and temperatures below 20°C. In the temporary storage drying chamber, materials are packed in various types of containers, including paper bags, textile materials, and aluminum foil trays.

In the second drying chamber, samples are arranged on shelves in aluminum trays, where the drying process is carried out in a thin, evenly distributed layer.

**SPP5** – Describe the criteria you use to decide on seed quantity per accession for the long-term storage.

Details are given at SCSS3.

#### **Box 3.2.5.A. Genetically Modified Material**

**GMM1** – In case you treat GMO material differently from “normal germplasm”, please provide here the details for each of the deviating procedures (and equipment).

Suceava Genebank does not own, or process organisms known to be genetically modified.

**GMM2** – Describe the policy and procedures (if any) in your genebank, related to ensuring that distributed samples are not containing GMOs.

Suceava Genebank does not own, or process organisms known to be genetically modified.

## **B. *In vitro* Culture Collections**

### **Box 3.2.1.B. *In vitro* Culture Vessels and Sample Size**

**SCSS1** – Indicate if you document the initial number of explants of individual accessions when culture is initiated (from one or from more clonal donor plants).

Currently, 4 – 5 potato tubers, selected from a mix of clonal, healthy plants grown in the field collection, are used as the genetic source for meristem sampling. Starting with 2025 the procedure is to be changed by introducing the two clonal donor plants.

For garlic, cloves are selected from multiple bulbs based on their vigor and health.

Approximately 10 – 15 meristems per accession are extracted under a binocular microscope, and the details are recorded in an Excel file specifically dedicated to laboratory work.

**SCSS2** – Please describe in general terms the type of culture vessels (as far as not already done in section SC2 in Box 3.1.3.B), media and phytohormones you use, as well as the procedures you follow with respect to cutting technique, callus exclusion, etc.

The type of culture vials used is described in section 3.1.3.B.

The culture media are prepared based on the Murashige and Skoog (MS) formulation from 1962, which is widely recognized for plant tissue culture.

The primary growth regulators incorporated into the media include kinetin, benzyl adenine, and  $\alpha$ -naphthyl acetic acid, with optional additions of Daminozide or Cycocel.

The prepared culture medium is dispensed into glass jars (20 ml per 170 ml vial), which are then covered with aluminum foil and sterilized in an autoclave at 121°C for 20 minutes.

For each genotype, 20 nodal segments are distributed across four jars. Any callus that forms, regardless of its stage, is removed. After inoculating the medium with the plant material, all jars are sealed with double layers of polyethylene foil, secured with two rubber bands.

**SCSS3** – Please indicate whether or not you use a minimum number of *in vitro* plantlets per accession.

During the multiplication and conservation phases, 40 plantlets are typically used, evenly distributed across two culture media, with a minimum of 20 specimens in each medium.

**SCSS4** – Please provide details on other aspects that are important in this context.

None.

#### **Box 3.2.2.B. *In vitro* Culture Procedures**

**SPP1** – Describe the numbers of sub-clones you may cultivate per accession (assuming that this is not crop-specific).

Currently, each sample in the *in vitro* potato collection consists of the progeny of three to four meristems that exhibited the most favourable development during the regeneration phase. For each culture medium (micromultiplication, microtuberization, and conservation), 20 microcuttings are sub-cultured, resulting in the production of an equal number of plantlets (sub-clones).

**SPP2** – Describe the sub-culture duration (if not crop-specific).

Subculturing is performed every 2 – 3 months for *in vitro* plantlets grown on micromultiplication media in the growth chamber, maintained at 22°C. For plantlets transferred to the conservation cell, kept at 6 – 10°C, subculturing occurs every 6 – 8 months.

Plantlets maintained under slow-growth conditions on conservation media, supplemented with growth inhibitors and subjected to restrictive environmental conditions in the conservation cell, can remain viable for 2 – 3 years between subcultures.

**SPP3** – Describe the criteria you use to decide on *in vitro* plant quality (if not crop specific).

The overall development of the plantlets, including the presence of green leaves, the formation of microtubers, and proper rooting, indicates healthy growth. Conversely, signs such as necrosis, etiolated leaves, and devitalized shoots are indicative of senescence. These deteriorating tissues should be promptly removed and eliminated through subculturing to preserve the biological material and ensure its viability.

#### **Box 3.2.3.B. Genetically Modified Material**

**GMM1** – In case you treat GMO material differently from “normal germplasm”, please provide here the details for each of the deviating procedures (and equipment).

NOT APPLICABLE

### **D. Field Genebank Collections**

#### **Box 3.2.1.D. Accession Sample Size**

**SCSS1** – Indicate if you document the initial number of plants of individual accessions (either as received from collecting missions or through exchange).

The number of vegetative organs for each field genebank accession, whether received from external sources or obtained from the previous year's harvest, is recorded in the list of crops maintained as live plants in the field genebank. This parameter serves as

a reference for comparing and analyzing the accession's development in subsequent stages, including emergence, growth, and harvest.

**SCSS2** – Please describe what kind of procedures you follow, if any, with respect to sub-sampling and subsequent place/container/etc. of maintenance.

Sub-sampling procedures are not applied to field collections of potato, garlic, and onion. This means that no specific techniques are used to divide or reduce the collected material into smaller representative portions for analysis or preservation. Instead, the entire collected material is likely handled as a whole for these crops.

**SCSS3** – What is the number of plants that you use as the minimum threshold per accession? Are these plant numbers of a given accession based on genetic parameters (such as reproduction biology; heterogeneous samples)?

An accession maintained in the field typically comprises 10 to 15 plants for potato and onion crops. For garlic, however, a sample includes 20 to 60 plants to also accommodate distribution requirements.

**SCSS4** – Please provide details on other aspects that are important in this context.

None.

#### **Box 3.2.2.D. Multiplication**

**PC1** – Please describe the multiplication procedures that you follow for your field genebank material (both annual and perennial species)

*Please include in your description the following aspects if they would apply to your field genebank management procedures):*

- a) *any control measures to minimize or avoid cross-pollination between accessions (if applicable/relevant);*
- b) *the use of pollination cages for insect-pollinated species;*
- c) *the use of specific pollinators for insect-pollinated species;*
- d) *strategies to ensure that males and females participate equally in the reproduction);*
- e) *strategies to avoid any genetic drift (minimum number of plants, minimum number of plants at flowering stage before pollinators introduction, similar quantity of seeds harvested from each plant, etc.).*

NOT APPLICABLE

**PC2** – Provide any other relevant information on procedures that you apply to control pollination of your germplasm in case of harvesting planting material from your field genebank material.

NOT APPLICABLE

#### **Box 3.2.3.D. Planting Material Processing Procedures**

**SPP1** – Describe the protocol(s) that you use for threshing and seed cleaning, if used as an intermediate step for the management/multiplication of your field genebank accessions.

NOT APPLICABLE

**SPP2** – Please describe how and where you store (in a temporary manner) newly harvested planting material.

*Please provide details on the temperature and relative humidity of the storage room/space; what type of containers you use, if any, etc.*

During the winter, the potato collection is stored in a climate-controlled cell where air conditioning maintains temperatures between 4 – 5°C, and the relative humidity ranges from 75% to 90%. The tuber samples are packed in paper bags, labeled according to the field list, and placed on metal racks for organized storage.

After harvesting, onion and garlic bulbs are stored in a post-ripening room at approximately 20°C with a relative humidity of up to 70%. This ensures proper conditions for post-harvest handling and preservation of the bulbs.

**SPP3** – Describe the criteria you use to decide on the number of plants per accession intended for the long-term conservation.

For potato, garlic, and certain onion collections, the criteria used to determine the number of plants per accession intended for long-term conservation typically include propagation capacity, health status, availability of space and resources and distribution needs (specific to garlic).

### 3.3. Ensuring Availability

#### A. Seed Collections

##### Box 3.3.1.A. Ensuring Availability of Germplasm – Policy Aspects

**AGP1** – Describe the germplasm distribution policy that you follow at your genebank.

*You might want to consider in your response the following aspects:*

- a) *crop/species specificity;*
- b) *whether or not sufficient seed stock is available; who the requestor is;*
- c) *what the purpose of the germplasm request is;*
- d) *any restrictive conditions and/or*
- e) *the total amount of accessions sent per request for distribution of germplasm;*
- f) *use of a formal agreement to distribute the germplasm.*

All accessions in the active collection that are not protected by Intellectual Property Rights (IPR), have sufficient stock, and demonstrate adequate germination capacity are distributed for breeding, research, or educational purposes. This distribution is carried out under the Standard Material Transfer Agreement (SMTA) in accordance with the International Treaty on Plant Genetic Resources for Food and Agriculture.

To support the on-farm or garden conservation of Romanian crop landraces, the Genebank provides up to five accessions from its collections to individuals interested in preserving agricultural traditions and heirloom varieties. For autogamous, annual species, 15 – 50 seeds are offered per accession, following the Suceava Genebank Transfer Protocol.

**AGP2** – Do you have as part of your service-rendering policy aspects such as a “maximum time” between receiving a germplasm request and distribution of the germplasm?

For researchers and breeders, the response time is kept as short as possible, not exceeding 10 days, and is determined by the time required to complete the formalities for signing the Standard Material Transfer Agreement (SMTA).

For individuals, sample shipments begin 10 – 14 days after the end of the designated registration periods. Requests are processed in chronological order to ensure that the biological material is delivered on time to all geographical regions of the country.

Further details can be found at [www.svgenebank.ro](http://www.svgenebank.ro) (in Romanian).

**AGP3** – Describe how you treat “related information” about the requested accessions that you make available to the requestor, i.e. provide details on the typical information you send out with the germplasm

Material provided for breeding, education or research purposes is accompanied by passport descriptors and, when available, characterization and evaluation data.

For samples sent to individuals, the accompanying label includes the following details: the accession number, the location of origin, and the scientific and local names of the material.

### **Box 3.3.2.A. Ensuring Availability of Germplasm – Seed/Germplasm Stock Aspects**

**AGSS1** – Please provide details on the minimum/maximum amount of seed, plant, in vitro samples that you distribute (where relevant, differentiated by species groups, i.e. self-pollinating, cross-pollinating and/or whether an accession is homo- or heterogeneous).

For research and breeding purposes, there is no fixed limit on the number of seeds requested, as long as the requests are reasonable. The quantity of seeds provided is determined based on the specific request and the availability of stock.

For the distribution of local varieties to small growers within the country, the number of seeds provided ranges from 15 to 50, depending on the seed size. For most varieties, 25 seeds are typically offered.

**AGSS2** – Describe how you store the seeds/other germplasm of a given accession with respect to the use of single or multiple bags or containers per accession.

The Genebank distributes material exclusively from its active collection and does not maintain sub-samples of conserved accessions specifically for distribution purposes.

The varieties provided to individuals are sourced from stocks produced through annual multiplication, which are kept separate from the stored collections. This ensures that the conserved collections remain intact while meeting the needs of distribution.

**AGSS3** – Describe how you manage the availability of adequate seed/other germplasm stock per accession, including the use of an absolute lower minimum of seeds per accession as the threshold to decide to regenerate.

By querying the BIOGEN database, which provides information on the total seed stock and germination capacity, priority lists are created to guide the multiplication and regeneration of accessions. This process ensures that accessions with declining germination rates or insufficient stock are prioritized for regeneration, aligning with standard genebank management practices.

**AGSS4** – Provide here information on any other aspects that are relevant to manage seed/other germplasm stocks.

None.

### **Box 3.3.3.A. Ensuring Availability of Germplasm – Health Aspects**

**AGHA1** – Describe how you store seed/other germplasm with respect to germplasm health considerations, including whether you have a “policy” of storing only “disease free” (as far as you can see or determine) accessions, at least for the quarantine pests and diseases.

All seed accessions regenerated or multiplied by the Genebank undergo strict health monitoring to ensure they are free from diseases or pests. This monitoring is conducted during their growth and development in the field genebank, as well as during the preparation phases for storage or distribution.

After seed extraction, all samples are thoroughly inspected and issued an internal health certificate by the Genebank’s phytosanitary specialist. Only seed samples that receive this certificate, confirming they are “disease-free,” are eligible for storage or distribution. This process ensures the health and quality of the biological material provided by the Genebank.

**AGHA2** – Describe how you follow plant quarantine rules and regulations when exporting germplasm abroad (especially to countries at another continent).

The Suceava Genebank adheres to strict norms for preventing the spread of quarantine diseases. To ensure compliance, it collaborates with the Plant Protection Agency, which inspects the material and issues phytosanitary certificates. These certificates confirm that the plant material is free from pests and diseases, meeting the required health standards.

This collaboration ensures that all distributed or stored material is safe and aligns with national and international phytosanitary regulations.

**AGHA3** – Describe if and how you distribute germplasm accompanied by a phytosanitary certificate or a “plant passport”.

All necessary steps are taken to obtain phytosanitary certificates issued by the Plant Protection Agency before dispatching seeds. This process ensures compliance with European regulations and the legislation of other countries that require genetic material to meet specific phytosanitary standards.

Phytosanitary certificates verify that the seeds have been inspected and are free from pests and diseases, as required by international agreements such as the International Plant Protection Convention (IPPC).

These certificates are issued by or under the authority of the National Plant Protection Organization (NPPO), following inspections and related activities.

**AGHA4** – Provide any other relevant information on procedures that you follow with respect to germplasm health aspects.

NOT APPLICABLE

#### **Box 3.3.4.A. Germplasm Supply**

**GS1** – Describe the policy of your genebank with respect to the sample size that you use for distribution purposes, including whether you differentiate between germplasm from self - or outbreeding species, heterogeneous accessions, and possibly other aspects.

Answer is given at AGSS1.

**GS2** – As GS1 above, but in case your germplasm samples do not possess the minimum viability, would you increase the number of seeds?

If the germination rate of seeds is lower than the recommended standard, the Genebank compensates by increasing the number of seeds provided to the beneficiary.

Additionally, the beneficiary is informed about the lower germination rate to ensure transparency and proper planning for their intended use.

**GS3** – Please provide information on any other aspects related to seed supply.

None.

#### **B. *In vitro* Culture Collections**

##### **Box 3.3.1.B. Ensuring Availability of Germplasm – Policy Aspects**

**AGP1** – Describe the germplasm distribution policy that you follow at your genebank. *You might want to consider in your response the following aspects: is the user informed about the option to get provided with in vitro cultures and whether they are available all the time of the year; are in vitro samples an option or the only way to get material; who the requestor is; what the purpose of the germplasm request is; any restrictive conditions and/or the total amount of accessions sent per request for distribution of germplasm; use of a formal agreement to distribute germplasm.*

The biological material from the *in vitro* potato collection is available year-round in the form of microtubers. This material is primarily intended for institutions with laboratories

capable of maintaining or properly utilizing the offered genotypes. The main recipients are breeding and research institutions.

As with other genotypes distributed by the Genebank, the Standard Material Transfer Agreement (SMTA) is used to regulate the transfer of these materials.

**AGP2** – Indicate if you have as part of your service-rendering policy aspects such as a “regular or a maximum time” between receiving a germplasm request and distribution of the germplasm.

For researchers and breeders, the response time is kept as short as possible, not exceeding 10 days. This timeframe is determined by the completion of the formalities required for signing the Standard Material Transfer Agreement (SMTA).

**AGP3** – Describe how you treat “related information” about the requested accessions that you make available to the requestor, i.e. provide details on the typical information you send out with the germplasm.

The material is accompanied by passport descriptors

#### **Box 3.3.2.B. Ensuring Availability of Germplasm – Germplasm Stock Aspects**

**AGSS1** – Please provide details on the maximum amount of *in vitro* samples that you distribute.

Microtubers can be transferred from the culture medium into sterile vials under controlled conditions. Depending on their size and the developmental stage of the microshoots, 10–15 microtubers are typically prepared and offered for distribution.

**AGSS2** – Describe how you store the samples of a given accession with respect to the use of vessels for culture and vessels for distributions (glasses or plastic bags).

There are no pre-prepared samples available for distribution. However, from the collection of potato genotypes grown on microtuberization medium, 1 – 2 vials containing microtubers can be extracted and packed for shipment as needed.

**AGSS3** – Describe how you manage the availability of adequate plants per accession, including the use of an absolute lowest minimum of plants per accession as the threshold to decide to regenerate.

At a minimum of 10 plantlets, distributed across two vials, the initial stock of 20 specimens should be replenished to maintain the collection's viability and ensure sufficient material for future distribution or research purposes.

**AGSS4** – Provide here information on any other aspects that are relevant to manage stocks (e.g. transfer of material through greenhouse transfer phases in case a user cannot handle *in vitro* cultures).

Microtubers can be utilized, with appropriate care, even by entities that lack *in vitro* culture laboratories.

#### **Box 3.3.3.B. Ensuring Availability of Germplasm – Health Aspects**

**AGHA1** – Describe how you store germplasm with respect to germplasm health considerations, including whether you have a “policy” of storing only “disease-free” (as far as you can see or determine) accessions, at least for the quarantine pests and diseases.

At present, there are no facilities available to conduct health control checks or issue certificates regarding the health status of the material. However, the use of meristem culture procedures significantly improves the quality of the plant material by eliminating viruses and other pathogens

While no formal certification is provided, the macroscopic appearance of the material is considered “disease-free,” reflecting the effectiveness of the meristem culture process in producing clean plant material.

**AGHA2** – Describe how you follow plant quarantine rules and regulations when exporting germplasm abroad (especially to countries at another continent).

The Suceava Genebank adheres to strict norms for preventing the spread of quarantine diseases. To ensure compliance, it collaborates with the Plant Protection Agency, which inspects the plant material and issues phytosanitary certificates.

**AGHA3** – Describe if and how you distribute germplasm accompanied by a phytosanitary certificate or a “plant passport”.

See AGHA2.

**AGHA4** – Provide any other relevant information on procedures that you follow with respect to germplasm health aspects

None.

#### **Box 3.3.4.B. Germplasm Supply**

**GS1** – Describe the policy of your genebank with respect to the sample size that you use for distribution purposes.

The answer is given at question AGSS1.

**GS2** – Please provide details of your routine methodology of containers etc. that you use to distribute *in vitro* cultures.

Glass vials are used for storing and transporting microtubers, ensuring sterility through secure closures. These vials have a sufficient capacity to hold 10–15 microtubers, making them suitable for safe handling and distribution.

**GS3** – Please provide information on any other aspects related to *in vitro* plant supply.

None.

#### **D. Field Genebank Collections**

##### **Box 3.3.1.D. Ensuring Availability of Germplasm – Policy Aspects**

**AGP1** – Describe the germplasm distribution policy that you follow at your genebank. *You might want to consider in your response the following aspects: crop/species specificity; whether or not sufficient seed stock is available; who the requestor is; what the purpose of the germplasm request is; any restrictive conditions and/or the total amount of accessions sent per request for distribution of germplasm; use of a formal agreement to distribute the germplasm.*

Only samples of local garlic varieties included in the Genebank's distribution program for the autumn campaign are considered for this category of genetic resources. These samples are specifically selected and made available as part of the Genebank's efforts to preserve and distribute local garlic biodiversity.

**AGP2** – Indicate if you have as part of your service-rendering policy aspects such as a “maximum time” between receiving a germplasm request and distribution of the germplasm.

The distribution protocol for garlic local varieties ensures compliance with the optimal planting period for the requestors. By considering the planting period, the Genebank ensures that the distributed material is suitable for successful cultivation and aligns with the agricultural calendar of the recipients.

**AGP3** – Describe how you treat “related information” about the requested accessions that you make available to the requestor, i.e. provide details on the typical information you send out with the germplasm.

Answer is given at AGP3, Box 3.3.1A.

#### **Box 3.3.2.D. Ensuring Availability of Germplasm – Seed/Germplasm Stock Aspect**

**AGSS1** - Please provide details on the minimum/maximum amount of plants or organs (cuttings, bulbs, tubers, etc.) per plant that you distribute per accession (where relevant, differentiated by species groups, i.e. annual or perennial; woody or herbaceous; other) and/or whether an accession is clonally or sexually propagated).

Samples prepared for garlic distribution typically consist of approximately 10 bulbils per variety.

**AGSS2** – Describe how you manage the availability of adequate organs per accession, including the use of an absolute lower minimum of plants per accession as the threshold to decide to multiply.

The entire garlic field collection is cultivated annually. The most representative genotypes are multiplied specifically for distribution purposes.

**AGSS3** – Provide here information on any other aspects that are relevant to manage plant material stocks.

None.

#### **Box 3.3.3.D. Ensuring Availability of Germplasm – Health Aspects**

**AGHA1** – Describe how you maintain field genebank (and any intermediate storage step) accessions with respect to health considerations, including whether you have a “policy” on accepting/planting only “disease-free” planting material (as far as you can see or determine) accessions, at least for the quarantine pests and diseases.

All distributed biological material, including garlic bulbils, undergoes a macroscopic inspection to ensure quality and health. During this process, any bulbils showing signs of disease or pest attacks are removed. Only bulbils that are, to the best of the Genebank's knowledge, free from disease are approved for shipment to users.

**AGHA2** – Describe how you follow plant quarantine rules and regulations when exporting germplasm abroad (especially to countries at another continent).

NOT APPLICABLE

**AGHA3** – Describe if and how you distribute germplasm accompanied by a phytosanitary certificate or a “plant passport”.

NOT APPLICABLE.

**AGHA4** – Provide any other relevant information on procedures that you follow with respect to germplasm health aspects.

None.

#### **Box 3.3.4.D. Germplasm Supply**

**GS1** – Describe the policy of your genebank with respect to the sample size that you use for distribution purposes, including whether you differentiate between germplasm from annual or perennial species, clonally or sexually propagated accessions, and possibly other aspects.

The answer is given at AGSS1, Box 3.3.2.D.

**GS2** – Please provide information on any other aspects related to seed supply.

None.

### **4. Providing Information**

#### **Box 4.1. Genebank Documentation System**

**GD1** – Please provide details on the technical aspects of the genebank information management system(s) that you use.

- a) On which software is the system based (i.e. Oracle, Fox Pro, MS Access, MS excel, MS Word, other?).
- b) In case you use a manual information management system, please provide details.
- c) In case your “internal” database(s) is/are different from the publicly available database(s), please provide details on both,
- d) Describe which activities of the genebank are covered by the system.

The current BIOGEN system relies on FoxPro, while online databases use MS Access.

Genebank has initiated a new project to modernize and streamline the work of its researchers by replacing the current BIOGEN software with a new application called GeneDataBank. This new system is built entirely on Open-Source technologies, utilizing a relational database (PostgreSQL) and a Java-based back end. Access to the application will be provided via a web browser, making it user-friendly and widely accessible.

The GeneDataBank application incorporates several modern features to enhance functionality and efficiency, including:

- QR code generation for containers in conservation, improving tracking and management.
- Mapping capabilities to indicate the geographic origin of samples.
- Graph generation to visualize germination history and trends.
- Advanced filtering options for easier data selection.
- Data export functionality in Excel format for further analysis or reporting.

The current manual information management system is being progressively developed alongside the database (<https://github.com/genedatabank/manual/ro/GeneDataBank-ro.md>).

The "internal" database(s) store all accession-related dates, such as those for passports, C&E, and management, while the online database is limited to non-confidential passport information. The BIOGEN database system supports activities including collection, multiplication/regeneration, characterization/evaluation, viability testing/monitoring, germplasm conservation, and distribution.

**GD2** – Provide details on which types of data you handle in your documentation system, e.g. passport data, characterization & evaluation data, cultivar data, material distribution, etc.

The BIOGEN software currently used by Genebank is a comprehensive system that manages a wide range of data related to genetic resources.

It includes:

- Passport data: Information about the origin and identity of genetic material.
- Local knowledge data: Details about traditional uses and cultural significance of genetic material.
- Herbarium data: Records of preserved plant specimens.
- Management data: Information on the handling and maintenance of genetic resources.
- Viability data: Data on the germination and survival rates of stored material.
- Characterization and evaluation data: Descriptions of genetic traits and performance under various conditions.
- Taxonomy data: Classification and naming of plant species.
- Storage data: Details about the conditions and methods used for preserving genetic material.
- Distribution data: Records of the transfer and sharing of genetic resources.

**GD3** – In case your internal database(s) is/are different from the publicly available database(s), please provide details on both.

The answer is given at GD1 c.

**GD4** – Describe in which form you send accession specific data (e.g. as hard copy, electronically – if the latter, please specify (in plain text) which file format, i.e. Excel, Access, others is used).

If requested, the available data from the Genebank can be provided either as a hard copy or electronically in Excel file format. This ensures flexibility and accessibility for researchers or other interested parties who require the data for their work.

**GD5** – Provide information on how technical support for development and maintenance of the documentation system is arranged.

Technical support for the Genebank's systems and applications is provided by the IT compartment of the Genebank. This ensures that users and researchers have access to assistance for any technical issues or questions related to the Genebank's software, databases, or data distribution processes.

**GD6** – Describe your genebank policy with respect to backing-up of the database contents, including with which frequency

Database backups at the Genebank are performed weekly in electronic format on different stations and annually on external memory devices. These backups are managed by the IT compartment of the Genebank, ensuring the safety and integrity of the data.

**GD7** – Provide any other information on your information management system that is not covered in one of the above questions.

None.

#### **Box 4.2. Information Exchange**

**IE1** – Please describe how you make your passport data available to users (i.e. as hard copy; via the internet; other?).

Currently, the passport data from the BIOGEN database and the national inventory is made publicly available on the SVGB website, but only with minimum descriptors. For more comprehensive access, all passport descriptors of the national inventory database are accessible via the European Search Catalogue for Plant Genetic Resources (EURISCO) website.

**IE2** – Please indicate if your data is available as machine-to-machine web-services. In case it is, describe:

- a) what types of data (passport data, characterization & evaluation data etc.) and
- b) which web-service interfaces are available (i.e. GBIF IPT, BioCase, TapirLink).

In the old BIOGEN database, data was not accessible via machine-to-machine web services. However, the new application, GeneDataBank, operates on a server-client

architecture, enabling clients to access comprehensive data, including Passport, Deposit, Viability, Duplicate, and more. The web service facilitating communication between clients (web browsers) and the server (GeneDataBank) utilizes the classical Internet Protocol (IP).

**IE3** – Please indicate if your data is published to EURISCO. Describe which data is published to EURISCO and at which intervals.

Passport data from the BIOGEN database and the national inventory is currently published on EURISCO and updated annually. This process will continue once the transition to the new GeneDataBank is completed.

Additionally, characterization and evaluation (C&E) data for AEGIS accessions are also published on EURISCO, ensuring broader accessibility and alignment with European plant genetic resource documentation standards

**IE4** – Please provide any other information on information exchange that is important for others to know.

None.

**IE5** – Describe the kind of information you distribute together with the germplasm to persons that request germplasm.

Please consider the following data types: Passport, Characterization; Evaluation, and/or Germplasm management data (e.g. viability percentage; protocols followed for routine operations; etc.

The answer is given at AGP3, Box 3.3.1.A.

