

Ex-situ Conservation of Woody Plants within the Framework of the Millennium Seed Bank Partnership in Georgia

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ABSTRACT

Georgian flora comprises of the vascular plants grown in wild conditions on the territory of country, comprising 185 families, 1,048 genera and 4,275 species. High endemism level of Georgian flora reflects its richness - 21% of the local flora is endemic.

Nearly 10% of Georgian flora comprises of around 400 tree and shrub species. About 40 per cent of them are included in the Red List of Caucasus Endemics.

Within the framework of the Millennium Seed Bank partnership, 144 populations of 131 species of trees and shrubs common in Georgia were collected at the place of natural distribution and deposited in the National Seed Bank of Georgia (NSBG) under the Global Tree Seed Bank Project (Georgia) with the funding from the Garfield Weston Foundation (2015-2019).

The created genetic bank includes rare, endemic, relict, endangered and economically important species, they have food, medicine, fuel, construction, paint and other properties.

This collection also comprises relict species of Colchic forests important for the study of the history of the Caucasus flora from the following genera: *Rhododendron*, *Betula*, *Ruscus*, *Ilex*, *Daphne*, *Hedera*, *Laurocerasus*, amongst others;

The National Seed Bank is established for long-term storage of the wild plant species of Georgia.

Seed duplicates from different populations of all target species of the above mentioned project are kept in the Millennium Seed Bank of the Royal Botanic Gardens, Kew, UK.

In addition to the seed bank, a collection of seedlings of some of these species has been created in the collection plot of the Plant Conservation Department.

Key words: ex-situ conservation, tree and shrub species, seed bank, seedlings collection.

1. Introduction

The Caucasus, and Georgia in particular, has been attributed to the world's biodiversity hotspots.

Georgia has one of the highest concentration of plant species known from any temperate part of the world. The number of species in the Caucasus amounts to 6,350 and the number of endemic species to 1,500 (Myers et al 2000).

Georgian flora comprises of the vascular plants grown in wild conditions on the territory of country, comprising 185 families, 1,048 genera and 4,275 species. High endemism level of Georgian flora reflects its richness - 21% of the local flora is endemic and consists of about 900 species. Among them about 600 species are endemic to the Caucasus and about 300 are endemic to Georgia (Davliandize *et al.* 2018).

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Nearly 10% of Georgian flora comprises of around 400 tree and shrub species (Kikodze, Faruk 2016) About 40 percent of them are included in the Red List of Caucasus.

Ex-situ conservation via establishing seed banks and living collections is one of the most efficient and comparatively low-cost method for the protection of plant biodiversity.



Figure 1: The Republic of Georgia on the world map.

As of partnership with the Millennium Seed Bank of the Kew Botanical Garden: The global project – The Millennium Seed Bank Partnership (MSB) was launched by the Seed Conservation Department of the Royal Botanic Gardens, Kew in 2000. Scientists of the Department of Plant Conservation joined this global initiative in 2005 with the project “Ex-situ conservation of the wild flora of Georgia in the Caucasus Regional Seed Bank (Georgia) and the Millennium Seed Bank (UK) 2005-2010 (N4714 P4/6, 22 August 2005). Georgia was the first former Soviet country to join the MSB-1 and MSB-2 (2010-2020) projects. Priority for the project was creation of the seed bank of endemic, rare, threatened, vulnerable and economically important plants.

Staff members of the Department of Plant Conservation of the National Botanical Garden of Georgia together with colleagues from the Institute of Botany of the Iliia State University continue their work (2020-2030).



Figure 2, 3, 4: Field collection process of the target species for the Global Tree Seed Bank Project. Top left: *Salix alba*; bottom left: *Prunus laurocerasus*; right: *Sorbus migarica*

2. Materials and Methods

The collection of seeds of the target plants is always carried out in wild populations. The data gathering is accomplished for each accession in a special data form, which includes the geographic, botanical (associated species), habitat, collectors and other important information in a comprehensive manner. The acquired data will later be inputted in the BRAHMS (Botanical Research and Herbarium Management System) electronic database which is managed under the MSB and MSB DW (Data Warehouse).

The National Seed Bank (NSB) is designed for the long-term storage of seeds. All seed bank procedures are in accordance with international standards and guidelines (Baskin 2001; Smith 2003).

The procedures for seed preparation are following: dry room is used for seed drying upon the arrival to the seed bank. Afterwards the seeds are cleaned, sorted, infested, damaged or empty seeds are removed, weighted and counted. The quality and maturity of seeds is carefully assessed by the specialist group using the binocular stereoscope (Motic SMZ-161, Hong Kong). If the collection batch is not too small, some seeds are cut-tested and stained with tetrazolium chloride (TZ test) for the viability testing. Eventually, the “adjusted seed number” will be determined based on the percentage of potentially non-viable versus the viable seeds. It is crucial that the relative humidity of the seeds is reduced to 15-20% rH. This is a safe level for storing seeds at the negative temperature. Aluminium

foil bags are used for packing dry seeds, which are later sealed using special apparatus (Hulme Martin Heat Sealers Ltd, UK). The packages are stored at -20°C at the local facility.

The target tree and shrub species were mostly orthodox and the studies were consistent with existing methodology.

Among the target species were species of the genus *Salix* (willow). Seeds of *Salix*, although orthodox, are known to be short-lived. If processed appropriately, however, they can remain viable for years stored at low temperature and moisture conditions (Lopez-Fernandez et al., 2018). MSB Seed Collections Manager, Janet Terry, introduced it to our colleagues at the training in the new protocol for processing short-lived *Salix* seeds.

Germination capacity of seeds is tested on Petri dishes on 1% plain agar medium inside the incubator (LMS Ltd, UK) under controlled illumination and temperature regime. Results and conditions of germination tests along with the data of seed processing (cleaning, weighing, cut testing) and relative humidity measurements are transferred to the electronic database (BRAHMS), which automatically calculates percentages of germination and viability.

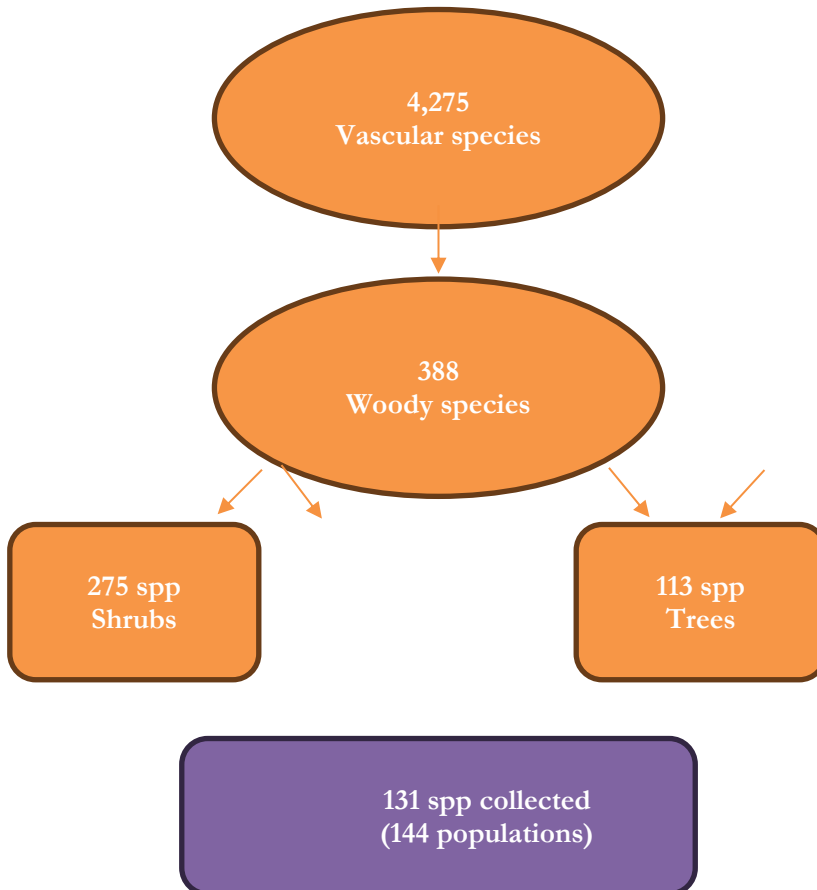


Figure 5: The number of taxa and populations collected in the scope of Global Tree Seed Bank Project in Georgia in reference to the vascular flora of Georgia.

3. Discussion

Considering the existing global challenges for nature conservation today ex-situ conservation of plants via the assembly of seed bank and living collections is one of the most effective and financially feasible means.

The aim of the following work was creation of seed bank of nationally distributed rare woody endemic and relict species, also those near extinction or economically important. As a target 131 plants extant on Georgian territory were selected, that are included in the Red List of Georgia (Solomon J., Shulkina T., Schatz E. G. (2014) Red List of the endemic plants of the Caucasus).

The seed bank is created with the consideration of populations in their distribution areas of the plants. The collection process was done with international standards (maximum 20% collected from wild) with acquired data (dedicated field data form) from the natural distribution areas of target species. The lab processing of the collected seeds is conducted similarly to the long term storage of collections at the Millennium Seed Bank (Kew) (Baskin, C. C. & Baskin, J.M. (2001).

The majority of woody species in Georgia are orthodox and complies with the -20°C storage law for the long term storage. However, there are a few genera with are not following this method: *Salix*, *Populus*, *Hedera*, *Juniperus* and some others. For the optimal storage of those plant groups additional studies need to be carried out. The presented work was done for the genera *Salix* and the new protocol for 12 species was worked out, the methodology was elaborated and the post-storage germination rate was therefore high.

4. Conclusion and Results

Among the 388 species of trees and shrubs distributed in Georgia 275 are shrubs and 113 are trees.

During 2015-2019, seeds and herbarium samples of 144 populations of 131 species were collected from 388 species of trees and shrubs common in Georgia within the scope of the Global Tree Seed Bank Project.

Out of 131 species collected, 19 species are included in the red list of Georgia (Red List of Georgia 2014). These species are: *Acer ibericum* M.Bieb. (*Acer monspessulanum* subsp. *ibericum* (M.Bieb.) Yalt.); *Arbutus andrachne* L.; *Betula medwedienii* Regel; *Betula megrelica* Regel (Endemic of Georgia); *Betula raddeana* Trautv. (Endemic of Caucasus); *Buxus colchica* Pojark. (*Buxus sempervirens* L.); *Corylus colchica* Albov (Endemic of Caucasus); *Juniperus foetidissima* Willd.; *Ostrya carpinifolia* Scop.; *Pterocarya fraxinifolia* (Poir.) Spach; *Pyrus demetrii* Kuth.; *Pyrus ketzkebovelii* Kuth. (Endemic of Georgia); *Pyrus sachokiana* Kuth. (Endemic of Georgia); *Salix kikodseae* Goerz (Endemic of Caucasus); *Salvia garedjii* Troitsky (Endemic of Caucasus); *Staphylea colchica* Steven; *Taxus baccata* L.; *Ulmus minor* Mill.; *Zelkova carpinifolia* (Pall.) K.Koch.

The seed bank of the selected species was created for the first time under the project's scope.

This collection also comprises relict species of Colchic forests important for the study of the history of the Caucasus flora from the following genera: *Rhododendron*, *Betula*, *Ruscus*, *Ilex*, *Daphne*, *Hedera*, *Laurocerasus* and others.

The assembled gene bank comprises of rare, endemic, relict, near extinction and economically important woody species with conservation status distributed in Georgia. Those plants additionally have nutritional, medicinal, fuel, construction, dye and other qualities. The assembled seed bank is a foundation for the conservation and sustainable development.

Minimum number of 500 seeds from each species (131 taxa) are deposited at the NSB and the corresponding labelled herbarium specimen kept at the National Herbarium of Georgia (TBI). The duplicates of the seed collection and herbarium voucher are sent to the Millennium Seed Bank and Herbarium of the Royal Botanic Gardens, Kew (K).

Using the new protocols (Lopez-Fernandez, M., et al. 2018) 12 species of *Salix* seeds were collected and placed in the seed bank, duplicates were sent to MSB. The seed germination rate was as high as 75% to 100%.

The information gathered in the seed bank is managed using the BRAHMS database.

During the implementation of the project, was created ex-situ seedlings collection of several endemic species with conservation status.

The documented living collections for later use in *in-situ* reintroduction works, scientific research and public awareness.



Figures 6, 7: Living collections acquired from the wild seed collections under the scope of project. Left: *Ilex colchica*. Right: *Lonicera stevensiana*

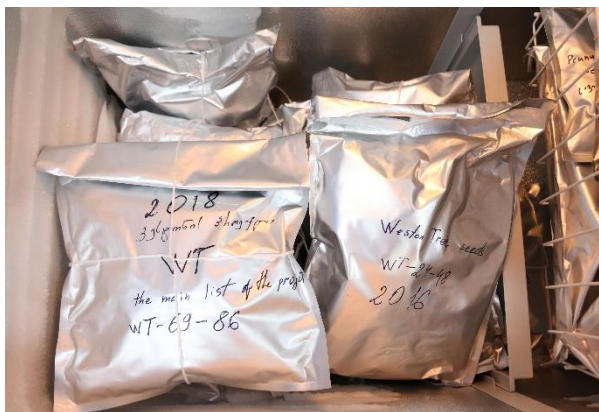


Figure 8: Seed accessions from the Global Tree Seed Bank Project stored at the National Seed Bank of Georgia

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References

- Barblishvili T. (2020) New protocol for processing short-lived *Salix* seeds trialled at National Seed Bank of Georgia. Samara, ISSN:1475-8245
- Baskin, C. C. & Baskin, J.M. (2001) Seeds, ecology, biogeography and evolution of dormancy and germination. Academic Press, California.
- Davlianidze M. and et al. (2018) Nomenclatural checklist of flora of Georgia. Ilia State University Institute of Botany, Tbilisi.
- Kikodze D. and Faruk A. (2016) Collecting rare, endangered and protected woody plants in Georgia. Samara, ISSN:1475-8245
- Kozlowski G. and Gratzfeld J. (2013) *Zelkova* – an ancient tree. Global status and conservation action. Natural History Museum Fribourg, Switzerland.
- Lopez-Fernandez, M. et al. (2018). Deterioration of willow seeds during storage. Scientific Reports. 8 (1).
- Miller J.S., McCue K., Consiglio T., Stone J., Eristavi M., Sikharulidze Sh., Mikatadze-Pantsulaia Ts., Khutsishvili M. (2006) Endemic Medicinal Plants of Georgia (Caucasus). 45p.
- Mikatadze-Pantsulaia Ts., Kolbaia S. and Gogoladze A. (2019) Safeguarding wild plant genetic resources of Georgia within the Millenium Seed Bank Partnership. European Journal of Sustainable Development. ISSN:2239-5938
- Mikatadze-Pantsulaia Ts., Barblishvili T., Trivedi C., Faruk A. (2017) Caucasus Regional Seed Bank. Nova Science Publishers Inc. New York, USA.
- Myers N., Mittermeier R. A., Mittermeier C. G., da Fonseca G. A., Kent J. (2000) Biodiversity hotspots for conservation priorities. Nature. doi: 10.1038/35002501.
- Red List of Georgia (2014). Resolution 190 of the Government of Georgia, February 20, 2014.
- Solomon J., Shulkina T., Schatz E. G. (2014) Red List of the endemic plants of the Caucasus: Armenia, Azerbaijan, Georgia, Iran, Russia, and Turkey. Monographs in Systematic Botany from Missouri Botanical.
- Smith R. D., Dickie J. B., Linington S. H., Pritchard H. W., & Probert R. J. (2003). Seed Conservation: Turning Science into Practice. London. The Royal Botanic Gardens, Kew.
- Trivedi C. (2012) Seed scientists and horticulturalists working together to conserve threatened Georgian species. Samara, ISSN:1475-8245