

ESCAPE

EX-SITU CONSERVATION OF FINNISH NATIVE PLANT SPECIES

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Ympäristöministeriö
Miljöministeriet
Ministry of the Environment



Need to ESCAPE



Traditional conservation is no more enough

Traditionally nature conservation has focused mainly on protecting the remnants of areas of high biodiversity that have remained untouched by humans. Even though this approach remains crucial, conservationists now fear that it might not be enough in the face of global environmental change. Therefore, new approaches are being developed to complement the more traditional ways. ESCAPE is an example of this.

Environmental problems – need for conservation

- Habitat loss and fragmentation
- Invasive alien species
- Environmental pollution
- Climate change

Ex-situ conservation is an approach to nature conservation where species are protected **outside of their natural habitat**. In Finland this type of conservation is still relatively new. ESCAPE has been developing and testing methods for ex-situ conservation of native Finnish plant species.

What is *ex-situ*?

Zoos have hosted endangered animals for long and today they play an important role in conservation. If a species can no more persist in its original area, it may still survive in a zoo.

Botanic gardens have worked applying for decades the same approach to plants. Plants are not capable of moving as fast as many animals, and thus different approaches are needed for their *ex-situ* conservation.

Plants can be maintained outside their natural habitat as seeds (seed banks), living collections (gardens and greenhouses) or frozen tissue material (cryopreservation). From these gene banks they can be transferred back to an area where they have gone extinct or to a totally new area, which is deemed suitable for them in the future.

Developing *ex-situ* methods has been the main goals of ESCAPE. The project has centered around native Finnish plant species. Nevertheless, the lessons learned can be applied also more broadly to other species and habitats.

ESCAPE in numbers

- ESCAPE species, subspecies and variants in collections total of 176
- Red list taxa in garden collections 83
- Taxa in the seed bank collections 142
- Seeds in seed bank 1 413 014
- Plant tissues collected c. 3000
- Other plant parts collected c. 20 000
- 5-year project funded by EU Life+ 2011 Biodiversity Program
- Started in September 2012
- Total budget c. 2 000 000 €
- Targets set to fulfill the national action plan for plant *ex-situ* conservation
- Ministry of Environment as a co-financier
- 4 beneficiaries: University of Helsinki, University of Oulu, Metsähallitus, Finnish Environmental Institute
- Taxa in cryopreservation 35
- Taxa in the micropropagation scheme 25
- Populations reintroduced 9 (6 species)
- Habitats improved 19

Seed Banking



Genetic diversity of plants is preserved in seed banks. Seeds are collected from threatened species and from these seeds, new individuals can be grown for later reintroduction to the wild. The first plant stored in ESCAPE seed bank in 2013 was *Pulsatilla patens*. Mature seeds are collected from flowering plants and then carefully cleaned and dried. After these preparations seeds are stored in a freezer. Finnish plant seeds can be preserved in these conditions for as much as hundreds of years.

Seeds are regularly tested for germination to make sure they are viable. Sprouted seeds of endangered species are usually planted in greenhouses and new plant individuals are included in living collection of the botanic garden. The aim of 80 species stored in the seed bank was well met and exceeded. The number of collected seeds is also considerably higher than originally planned.

2 mm



Working in the seed bank



Pulsatilla patens – The first one

- Protected in Finland
- Grows on dry sand ridges and xeric heath forests
- Only c. 120 known locations left
- *P. patens* dispersed to Finland after the last Ice Age. It is threatened by the shortage of suitable habitats and collection for private gardens.

Clones and Freezing Coldness

Cryogenic preservation and **micropropagation** are especially useful for plant species that cannot be stored into seed bank, such as bryophytes or vascular plants that do not produce seeds. In cryogenic preservation plant tissues are preserved by cooling them in liquid nitrogen to $-196\text{ }^{\circ}\text{C}$. At this very low temperature, all enzymatic and chemical activities inside the cells are stopped, and, in theory, the tissues are preserved theoretically forever. After the tissues have been preserved in this freezing coldness, they can be recreated by micropropagation. Micropropagation means multiplying stock plant tissues into viable plants. First, a small portion of tissue is taken from a plant, and then new clone plants grow from this tissue. In general, grasses and herbaceous plants are easy to propagate, whereas woody plants are more challenging.

ESCAPE has been studying the best cryopreserving methods and succeeded in recreating quite a many preserved taxa. Micropropagation sometimes has proved challenging. Problems have occurred especially with algal and fungal contaminations. Nevertheless, the expected number of 30 taxa has been exceeded. The research is still ongoing, improving and developing, for this method is still very new in the field of conservation biology.

Developing methods for species – examples:

Dianthus superbis (serpentine variation) and *Lychnis alpina* var. *serpentinicola* - Method works well and is effective way to increase population size

Rubus humulifolius – Extinct in Finland, has been preserved in *ex-situ* and is currently in cryopreservation

Cypripedium calceolus (Orchidaceae) – Micropropagation not yet successful, testing continues with other orchids too



Cypripedium calceolus

There is no universal appropriate method for micropropagation of all plants, and thus the procedure must be developed for each taxa separately.



Populations suffering from a low number of individuals encounter a variety of challenges that reduce their chance of survival. Small population sizes are typical for rare species. Therefore, these species have a high risk to die out locally.

ESCAPE did **population strengthening** to secure the survival of selected rare plant species.

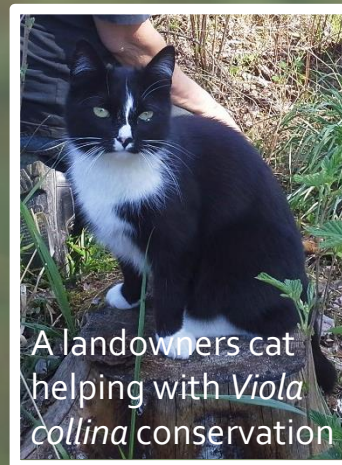
Population strengthening aims to increase the number of individuals in small natural populations. Additionally, new individuals were introduced to the plant populations in botanic gardens. This adds more genetic variation in the gardens' *ex-situ* collections and enables strengthening of natural populations also in the future if needed.

Altogether, **30 taxa** were cultivated in the botanic gardens and used to increase population sizes both in nature and in *ex-situ* collections. Some problems were encountered for example with overwintering: surviving the winter in greenhouse conditions proved challenging for certain species. Nevertheless, ESCAPE far exceeded the set targets for population strengthening, and gained valuable information to improve this conservation method.

Growing Stronger Together



Field inventory



A landowners cat helping with *Viola collina* conservation

Astragalus glycyphyllos

- critically endangered
- 3 populations
- grows in bright forest edges
- *Originally, only one individual remained in the target population. Luckily, this individual flowered and produced seeds from which new individuals were grown in the botanic garden. Now, these individuals have been successfully planted in natural populations.*

Viola collina

- endangered
- 5 populations
- grows in dry herb-rich forests, in open habitats
- threatened by forest management and overgrowth of vegetation
- *The right timing of seed collection proved to be challenging; the seeds of *V. collina* mature in cores that pop open when ready. The problem was tackled by bringing entire individuals to the greenhouse where keeping watch over the seeds was a lot easier, and the seeds were successfully collected.*



Assisted Migration

In the light of global climate change scenarios, also habitats are going to change and become inhabitable for a range of species. In the Northern Hemisphere, distribution areas of species are shifting northwards as the southernmost habitats become unsuitable. However, moving to new areas is challenging – if not even impossible – for some species. Threatened species with dispersal limitations are among the most vulnerable species. For them, methods of assisted migration may be applied. Assisted migration is an *ex-situ* based method, in which individuals of an endangered species are transferred to new areas that are predicted to be climatically suitable in the future.



Puccinellia phryganodes

- critically endangered
- listed on EU Habitat Directive
- two populations
- grows in seashore meadows
- threatened by the overgrowth of vegetation resulting from eutrophication and lack of grazing
- *P. phryganodes* was introduced to sites where cattle were grazing. Cattle keep the habitats open and suitable for the species, yet they can also cause harm by trampling the plants. Nevertheless, they provide valuable help against the overgrowth of vegetation threatening the species.

Artemisia campestris subsp. *bottnica*

- critically endangered
- listed on EU Habitat Directive
- one population
- grows in seashores of the Bay of Bothnia
- threatened by the overgrowth of vegetation resulting from eutrophication and crossbreeding
- *A. campestris* subsp. *bottnica* is threatened by the ongoing speciation process: it cross-breeds with the other subspecies. The hybrid is becoming ever more common, while future of the pure *A. campestris* subsp. *bottnica* is more and more uncertain. Therefore, the species is in a desperate need of management.



ESCAPE was the first actor doing assisted migration with Finnish plant species. Three species were selected to test and develop the method. Besides some challenges, the introductions have proved encouraging. One introduction is yet to be done by the end of the project.



Moving *Puccinellia phryganodes*

Back to the Roots

Plant populations can also be introduced back to the areas where it has died out recently. In **reintroductions**, new populations are planted to the species' original localities in the wild. These sites should still be suitable for the species, and no great changes should be foreseen in the near future. The main target is to establish a viable, breeding population to these original localities.

Altogether, ESCAPE has reintroduced six threatened plants and one moss species. So far, reintroductions have been mainly successful. The establishment success and changes in populations size will also be monitored after the project for the following 15 years. This will provide valuable information for the future *ex-situ* conservation.

Habitat Improvement

Reintroduction or assisted migration to an unsuitable place would be pointless. However, sometimes it is possible to significantly **improve the habitat** with slight changes. Most often this means removing competing vegetation so that the newly introduced plants will have a better chance to settle in their new home. Other methods include traditional grazing and controlled burning. During ESCAPE project 19 habitats have been improved. Most of them were improved only slightly by removing excessive vegetation by hand, others more strongly *e.g.* by removing spruce samplings with a chainsaw.



Armeria maritima subsp. *intermedia*

- critically endangered
- around 10 populations
- grows in sandy and dry seashore meadows
- threatened by the overgrowth of vegetation resulting from lack of grazing
- *Together with A. maritima subsp. intermedia, sheep were introduced to the sites to graze and keep the habitats open. However, the sheep turned out to be a bit too hard-working: they also ate the flowers of A. maritima subsp. intermedia. Thus, the plants were fenced off to keep them safe from grazers.*



Viola uliginosa

- endangered
- around 10 populations
- grows in repeatedly flooding habitats, *e.g.* on brook banks
- threatened by clearing of brooks, ditching, and constructions
- *V. uliginosa was not a classic case of reintroduction. The original locality had been built on, and thus the reintroduction was not possible to the same site. Luckily, a new, corresponding site was found close to the old one, and V. uliginosa was successfully reintroduced there.*



Bryophyte Specialities

Meesia longiseta

- endangered in Finland
- an EU directive species
- 101 populations
- grows in marshes and rich fens
- *M. longiseta* was included in the bryophyte micropropagation scheme. Moreover, the species was reintroduced successfully to a site where it had died out 10 years earlier. The introduced tussocks were growing strong.



Tortula cernua

- critically endangered in Finland
- 3 populations
- grows on calcareous substrates, e.g. walls, lime kilns, and waste pits
- Micropropagation methods were also developed successfully with *T. cernua*. Thus, in the future it is possible to propagate new bryophyte individuals even from a small amount of *T. cernua* tissue and spores.

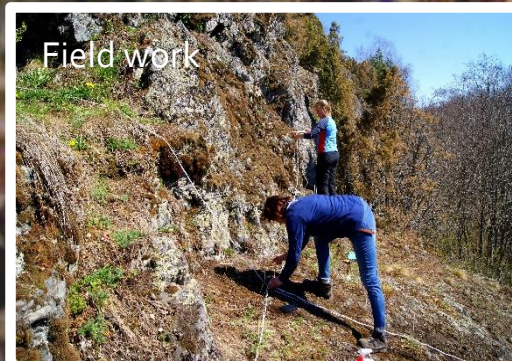
Mannia fragrans

- endangered in Finland
- 9 populations
- grows on calcareous or rich stones, and on soil or dry leas
- Population strengthening was done with *M. fragrans*. Close watch is kept on changes in patch number and size, shoot density, and reproduction.



Bryophytes have been conserved *ex-situ* in Finland for the first time during ESCAPE. There is very little experience also outside Finland. Three threatened bryophyte species representing different habitats and ecological requirements were selected as ESCAPE species. Candidates for bryophyte *ex-situ* were among the species in high threat categories also in the whole Europe and occurring in most threatened habitat types.

Bryophyte tissues have been shown to have high potential to survive deep-frozen even in nature. As bryophytes cannot be stored in seed bank, protocol for **tissue culture** and **cryopreservation** in liquid nitrogen tanks have been developed for many bryophyte species, including those studied in ESCAPE. Problems were encountered with alga and mold contaminations. However, **micropropagation** proved to be successful after starting from spores from sterilized spore capsules. ESCAPE has also tested the possibilities of bryophyte **reintroductions** and **population strengthening**.



In the Safety

of a Garden

Storing plants in **living outdoor collections** of botanic gardens is a complementary method for seed banking and cryogenic storage. Before ESCAPE only c. 11% of the threatened native plant species were *ex-situ* conserved in Finland. However, this proportion in living collections has increased by the end of the project up to 26%.

Outdoor collections have been assembled by gathering plants from their original growing sites and transferring them to gardens. Collections are also completed with seedlings from the seed bank sprouting tests. The possible crop can later be stored in the seed bank.

Plants in the collection are treated so that they could be reintroduced back to nature, and they are used in **population strengthening** and **assisted migration**. They also serve as a quick reservoir for *in-situ* conservation efforts. In addition, they play a big role in increasing public awareness of threatened plant species; in botanic gardens, people can see species that they unlikely meet elsewhere.



Managing outdoor collections in botanic gardens

1. All flowers are removed after flowering to prevent hybridization.
2. The flower beds are weeded and hoed to keep the beds clean and spacious.
3. Plants are regularly watered and their flowerbeds are fenced to prevent damage by rabbits.

The outdoor collections have been successful, and the aim of ESCAPE has been reached and surpassed for this part.



Public Awareness

Besides developing plant *ex-situ* conservation in Finland, ESCAPE aimed at increasing knowledge and interest on *ex-situ* issues in the society. New information was provided to conservation professionals through various presentations. ESCAPE has also targeted to get *ex-situ* conservation included in Finnish nature conservation legislation. A lot of attention was also paid to the general public. Numerous visitors of botanic gardens and other project partners visited touring exhibition on *ex-situ* conservation. Also, future generations were involved in ESCAPE organized workshop for school children. Kids were told about plants responses to changing environment and their conservation through games, songs, and other activities. Furthermore, ESCAPE gained significant visibility in public media.

Want to learn more about ESCAPE?

→ www.luomus.fi/en/ex-situ-conservation-finnish-native-plant-species

→ www.facebook.com/escape.luomus



Future Aspects

ESCAPE has succeeded in adding new species within *ex-situ* conservation. By the end of the project the proportion of threatened taxa within *ex-situ* conservation in Finland has increased to 53%. The project has also demonstrated various *ex-situ* methods and compiled a so-called tool box. We have learned a lot during ESCAPE, both how to and how not to do these actions. Today as well as in the future different conservation actions are surely needed. One possible *ex-situ* application could be for example green-roofs on which threatened plant and bryophyte species could be grown in the future.

Ex-situ conservation is still developing and suitable preserving methods for some species are still to be found. During this project, new information and experience was collected and with these, *ex-situ* conservation can be developed and improved. Based on ESCAPE actions guide books will be published for conservation. Guides will hopefully help to make *ex-situ* methods easier to approach and use. Research in this area is still needed so that in the future preserving of all kinds of plant species in Finland and all over Europe is possible. Success in *ex-situ* conservation gives hope for plant species in this ever-changing world.