

**Safeguarding of potato onion (*Allium cepa* L.
Aggregatum group) and garlic (*Allium sativum* L.)
crop diversity in North Europe – Baltic region
(SafeAlliDiv)**

Interim Activity Report (June 2017–June 2018)

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Interim Activity Report

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Photograph: Czech landrace of potato onion (*Allium cepa* L. *Aggregatum* group). ©H. Stavěliková, Crop Research Institute, Olomouc, Czech Republic.

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INTRODUCTION

The aim of the SafeAlliDiv project, funded by the ECPGR Grant Scheme, is to promote active on-farm conservation of European *Allium* accessions. The genetic analyses undertaken within the project will complement the existing molecular marker analysis of Nordic potato onions that was recently carried out in Finland and Sweden, with analysis of additional material from partner countries, in order to reach a better understanding of existing genetic diversity in collections.

The proposed workplan given in the SafeAlliDiv Activity proposal¹ is as follows:

1. Meeting in 2017 aimed at investigating the situation in *Allium* crop cultivation and maintenance in participating countries
2. Meeting in 2018 aimed at discussing/presenting the action plan for each country for safeguarding of focused *Allium* crops
3. To perform molecular analysis for potato onion (*Allium cepa* L. *Aggregatum* group) to determine the genetic diversity and relationships among European accessions
4. To prepare high level scientific publication on obtained molecular analyses results
5. To prepare popular publications in farmers magazines in involved countries in order to increase awareness of wider public on possibilities for cultivation and use of focused Alliums.

This Interim Activity Report provides information on actions implemented to date (items 1 to 3).

FIRST MEETING AND OUTCOMES

The first SafeAlliDiv project meeting was held 11-12 July 2017 in Tallinn, Estonia. In order to clarify the existing situation in potato onion conservation and diversity, before this meeting, project partners received and completed a questionnaire, providing the following information on three *Allium* species (garlic (*Allium sativum* L.), potato onion (*Allium cepa* L. *Aggregatum* group) and onion (*A. cepa* L.): number of accessions, type of collection (field or seed), type of conservation (*ex situ*, *in situ*), source of new accessions (collecting missions, seed companies, breeders, etc.), number of described accessions, number of accessions with chemical analyses and DNA analyses, safety duplication, cooperation with other genebanks in accession exchange, current projects related to particular species, other GR activities. Information on commercial production for each species was also included in the questionnaire: number of national varieties, harvest area (ha), production (t), import (t) and home-garden scale.

Participants from Croatia, Czech Republic, Estonia, Finland, Latvia, Lithuania, Norway and Sweden presented the situation in *Allium* crop cultivation and conservation in their countries, with particular emphasis on potato onion/shallot (*Allium cepa* L., *Aggregatum* group) and garlic (*Allium sativum* L.). The group agreed on criteria to select and acquire accessions, to be sent to the Latvian State Forest Research Institute for molecular analysis. It was agreed to perform also morphological characterization for the same accessions.

The group visited potato onion fields in the Lake Peipus area, where traditional cultivation is based on heterogeneous seed-propagated material, in contrast to the customary vegetative propagation in other

¹ The Activity proposal, including the list of partners, is available from the [SafeAlliDiv webpage](#).

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countries via bulbs. It was agreed that the different propagation methods influence the genetic and morphological heterogeneity of accessions as well as their phytosanitary status.

The tasks agreed at the first meeting and their respective achievements are listed below:

Workplan for the first year and achievements

| | Tasks agreed at the first meeting | Achievements |
|----|--|--|
| 1. | To contact Vera Shumilina from the N.I. Vavilov Institute of Plant Genetic Resources (VIR), St. Petersburg, Russian Federation, to enquire about the possibilities of obtaining shallot accessions from the Baltic region for inclusion in the molecular analyses. | Vera Shumilina promised to send 7 accessions for DNA analysis. The process took longer than expected, since it was necessary to send an official request. The first request was sent by Helena Stavělíková on 11 December 2017 to the director of VIR, Prof. Dzuybenko. Due to changes in the administration in VIR, Dainis Rungis sent a repeated request to Vera Shumilina on 12 April 2018, who replied that the new director of VIR, Elena Konstantinovna Khlestkina was revising the accession request forms. To date, no further information has been received from Vera Shumilina regarding the <i>Allium</i> accessions. |
| 2. | To distribute the lists of Croatian, Finnish and Norwegian potato onion accessions maintained in the Czech collection to the respective country members for identification of duplicates. | On 17 July 2017 Helena Stavělíková sent the lists to Smiljana Goreta Ban (Croatia), Terhi Suojala-Ahlfors (Finland) and Ingunn M. Vågen (Norway). |
| 3. | To send a summary of the results from the questionnaire to project partners. | Helena Stavělíková sent the questionnaire summary to project partners on 12 September 2017 (see Table 1). |
| 4. | To send a modified questionnaire to all members of the <i>Allium</i> WG. | Helena Stavělíková sent the modified questionnaire, without the section about commercial production, to all members of the ECPGR <i>Allium</i> WG (AWG). Completed questionnaires were received from 23 of 36 countries represented in the AWG. The results are summarized in Table 2. |

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| | Tasks agreed at the first meeting | Achievements |
|----|---|--|
| 5. | To initiate an electronic discussion between Terhi Suojala-Ahlfors (Finland), Dainis Rungis (Latvia) and Matti Leino (Sweden) about tools for DNA analysis. | The discussion took place and 15 microsatellite (SSR) markers were selected and tested (which were also utilised to genotype Swedish <i>Allium</i> accessions). The DNA markers tested were: AMS04, AMS06, AMS07, AMS08, AMS10, AMS12, AMS13, AMS14, AMS16, AMS22, AMS23, AMS25, AMS26, AMS29 and AMS30. After selection of the most informative and high quality markers, the <i>Allium</i> collection was genotyped with 11 markers: AMS06, AMS08, AMS10, AMS12, AMS13, AMS14, AMS16, AMS23, AMS30, AMS22 and AMS25. |
| 6. | To send the Descriptors for <i>Allium</i> (<i>Allium</i> spp.) ² to all project partners. | All project partners received the Descriptors for <i>Allium</i> (<i>Allium</i> spp.) electronically (18 July 2017) as well as a hard copy (during August 2017) from the ECPGR Secretariat. |
| 7. | To select and send the material for DNA analysis in good quantity and quality to Dainis Rungis, the Latvian State Forest Research Institute. | On 23 November 2017, all project partners received instructions about sample preparation and sending for DNA analysis. A total of 264 accessions were received for DNA analysis. DNA extraction was done on bulked tissue samples from 2 individuals for vegetatively propagated accessions (262 accessions), and from 9 separate individuals for the two generatively propagated Estonian accessions (Jõgeva 3, Kolkja KA). A total of 280 DNA samples were analysed (Table 3). |

Table 1. Questionnaire to project partner countries' summary table

| Genetic resources | Number of accessions | | | | | | | | | |
|---|----------------------|----------------|---------|---------|--------|-----------|-------------|--------|--------|------------|
| | Croatia | Czech Republic | Estonia | Finland | Latvia | Lithuania | Netherlands | Norway | Sweden | Total |
| Garlic (<i>Allium sativum</i> L.) | 52 | 628 | 1 | 5 | 67 | 68 | | | 3 | 824 |
| Potato onion (<i>Allium cepa</i> L. <i>Aggregatum</i> group) | 25 | 132 | 7 | 25 | 41 | 12 | | 20 | 29 | 291 |
| Onion (<i>A. cepa</i> L.) | 9 | 28 | 0 | 0 | 1 | 51 | 226 | 3 | 33 | 351 |

² IPGRI, ECP/GR, AVRDC. 2001. Descriptors for *Allium* (*Allium* spp.). International Plant Genetic Resources Institute, Rome, Italy; European Cooperative Programme for Crop Genetic Resources Networks (ECP/GR), Asian Vegetable Research and Development Center, Taiwan

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| Genetic resources | Number of accessions | | | | | | | | | | |
|--|----------------------|----|---|----|---|---|----|-----|----|--|-------------|
| | | | | | | | | | | | |
| Leek (<i>A. ampeloprasum</i> L.) | | 9 | 0 | 0 | 0 | 4 | 99 | 1 | 0 | | 113 |
| Chives (<i>A. schoenoprasum</i> L.) | | 4 | 0 | 15 | 0 | 6 | 24 | 7 | 71 | | 127 |
| Bunching onion (<i>A. fistulosum</i> L.) | | 16 | 0 | 0 | 0 | 4 | 42 | 7 | 2 | | 71 |
| Ramsons (<i>A. ursinum</i>) | | | | | | | | 1 | | | 1 |
| Alpine leek (<i>A. victorialis</i>) | | | | | | | | 4 | | | 4 |
| Other species | | | | | | | 37 | | | | 37 |
| Private collection with high number of <i>Allium</i> species | | | | | | | | 260 | | | 260 |
| Total | | | | | | | | | | | 2079 |

Table 2. Summary of replies to questionnaire on total number of *Allium* accessions from 23 responding AWG countries

| Genetic resources | Total number of accessions |
|---|----------------------------|
| Garlic (<i>Allium sativum</i> L.) | 3299 |
| Potato onion (<i>Allium cepa</i> L. <i>Aggregatum</i> group) | 1090 |
| Onion (<i>A. cepa</i> L.) | 5342 |
| Leek (<i>A. ampeloprasum</i> L.) | 1363 |
| Chives (<i>A. schoenoprasum</i> L.) | 272 |
| Bunching onion (<i>A. fistulosum</i> L.) | 459 |
| Ramsons (<i>A. ursinum</i>) | 1 |
| Alpine leek (<i>A. victorialis</i>) | 4 |
| Ball head onion, round-headed leek, round-headed garlic (<i>Allium sphaerocephalon</i> L.) | 1 |
| Mouse garlic (<i>Allium angulosum</i> L.) | 4 |
| <i>A. altaicum</i> Pall. | 11 |
| Siberian chives <i>A. nutans</i> L. | 25 |
| Garlic chives <i>A. tuberosum</i> Rotter. et Spreng | 20 |
| Tree onion <i>A. proliferum</i> Schrad. | 18 |
| Private collection with high number of <i>Allium</i> species | 260 |
| Other species | 3012 |
| Total | 15181 |

This information is very important for the continuing work of the *Allium* WG. It is necessary to know the current size and status of collections.

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Table 3. Number of samples for DNA analysis per country

| Country | Name of representative | Institute | Number of accessions for DNA analysis | Number of samples DNA extracted |
|----------------|------------------------|--|---------------------------------------|---------------------------------|
| Croatia | Smiljana Goreta Ban | Institute of Agriculture and Tourism | 25 | 25 |
| Czech Republic | Helena Stavělíková a | Crop Research Institute | 129 | 129 |
| Estonia | Küllli Annamaa | Estonian Crop Research Institute (ECRI) | 4 | 20 (*) |
| Finland | Terhi Suojala-Ahlfors | Natural Resources Institute Finland (Luke), Horticulture | 24 | 24 |
| Latvia | Līga Lepse | Institute of Horticulture | 38 | 38 |
| Lithuania | Danguolė Juškevičienė | Institute of Horticulture, LRCAF | 12 | 12 |
| Norway | Ingunn Molund Vågen | NIBIO – Norwegian Institute of Bioeconomy Research | 23 | 23 |
| Sweden | Matti Leino | Nordiska museet, Swedish Museum of Agriculture | 9 | 9 |
| Total | | | 264 | 280 |

(*) 2 vegetatively propagated accessions, plus 9 individuals from each of 2 generatively propagated accessions (Jõgeva 3, Kolkja KA).

The development of a guidebook on potato onion cultivation practices continues.

SECOND MEETING AND OUTCOMES

The second SafeAlliDiv project meeting was held 17-18 April 2018 in Olomouc, Czech Republic. The tasks from the first meeting and their fulfilment were discussed. Participants from Croatia, Czech Republic, Estonia, Finland, Latvia, Lithuania, Norway and Sweden presented information on the potato onion/shallot (*Allium cepa* L., Aggregatum group) accessions sent for DNA analysis.

Dainis Rungis, from the Latvian State Forest Research Institute, presented preliminary results of the molecular analysis. The DNA analysis results are very extensive. A total of 264 *Allium* accessions

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were received from the project partners. DNA was extracted from 2 bulbs of each accession (bulked prior to extraction, except for the 2 Estonian generatively propagated accessions (Jõgeva 3, Kolkja KA). The accessions were genotyped as diploids, but noting where three or more alleles were found in one sample, which could indicate triploid germplasm or the presence of genetic diversity within the accession.

The genotyping results indicated that 30 groups of accessions with identical multilocus genotypes (143 individuals) were identified, and 132 unique genotypes. Feedback has been received from the project partners regarding safety duplications and other factors to take into consideration during analysis. Genotype data from previous projects (carried out in 2012 and 2016 with funding from the Finnish national plant genetic resources programme) has been received from Finland and Sweden. The final analysis of genotype data should be completed by September 2018 and the first draft of a publication will be prepared and sent to project partners. In addition, the data will potentially be calibrated with the Finnish and Swedish genotype data, enabling integration of the datasets.

The group visited the Genebank of Vegetable and Specialty crops in Olomouc, where they could observe a collection of garlic and discuss the practical questions of maintenance of vegetatively propagated Alliums.

Further tasks to be carried out were agreed at the second meeting. Their respective status and achievements are listed below (schedule updated at time of writing the report):

Workplan for the period May 2018-January 2019

| | Task agreed at second meeting, April 2018 | Achievement/status by June 2018 |
|---|--|---|
| 1 | Mid May, 2018 – Dainis Rungis sends complete data on DNA analyses | 11 May 2018 – All partners of project received complete data on DNA analyses. |
| 2 | June 4, 2018 – feedback on safety duplications and other information sent to Dainis Rungis | 04 June 2018 – feedback sent by Helena Stavělíková (Czech Republic), Terhi Suojala-Ahlfors (Finland), Līga Lepse (Latvia) and Ingunn Vågen (Norway) |
| 3 | June-August 2018 – evaluation of the morphological traits for accessions in the field in the 2018 season | |
| 4 | October 15, 2018 – D. Rungis completes a summary based on received feedback and other information | |
| 5 | October 31, 2018 – first draft developed and sent to all project participants for contributions and comments | |
| 6 | December 15, 2018 – the second draft with contribution of each project partner is completed | |
| 7 | January 31, 2019 – the final version is circulated for final corrections/additions | |