



# Procedure 008 :

## Collection and sampling of terrestrial moss (*Hylocomium splendens*)

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## 1. Aim of procedure

This procedure describes all the various steps of collection and sampling of terrestrial moss (*Hylocomium splendens*; etasjemose) for the Environmental Specimen Bank for environmental pollutants. This procedure describes every step of the sampling and handling of samples to ensure that the procedure can be used in a clear way. The procedure should cover all aspects of reproducibility, quality and registration of data for the sampling material.

The procedure is based on the ICP Vegetation (2015) manual and the illustrations therein are representative for the sampling described here.

## 2. Sampling in field

### 2.1. Collection and handling

Collection and handling of the moss should be carried out manually, in order to avoid contamination of potential environmental pollutants and protect the samples from any kind of impact that will affect their usages as research and reference materials.

The sampling should be done as described here, which is in accordance with the ICP Vegetation Manual (2015):

1. The sampling location must be at least 3 meters away from the nearest tree crown: preferably in openings within the forest (diameter > 10m) or planted areas (diameter > 5m), without significant impact of overhanging trees and things coming from the trees, preferably on the ground or on the surface of rotting tree stumps. Sampling under shrubs and trees should be avoided also on grasslands or peatlands. This also applies to slope areas with running water.
2. Sampling sites should be located so that they represent the non-urban part of the region. The sampling location should be situated at least 300 m away from main roads, villages and industries, and at least 100 m away from smaller roads and houses.
3. In order to compare data from this survey with data from previous surveys, it is important to collect moss from the same site (or nearby, for example less than 2 km away but with similar biotopic conditions) that was used in previous moss studies.
4. For each sampling location, a pooled sample consisting of 10 sub-samples is collected. If possible, these sub-samples should be collected within a 50x50m area. Cleaned and burned (6h, 450 °C) glass jars should be used. To protect the collected moss from the rubber cover in the lid on the glass, cover the lid with a layer of aluminium foil before closing the lid.
5. 3 L of moss (tightly packed together) should be collected. Since some of the POP compounds are volatile and / or subject to photochemical degradation, the samples must be kept cold and dark at all times.
6. Smoking is prohibited during collection and further handling of the samples. Collection is done with bare hands that are washed well with large amounts of *Sphagnum* (peat moss) prior to collection.
7. Large contaminants on the moss samples such as particles, soil and animals should carefully be removed. Pieces of moss with sand or ants should be avoided. The sample should be carefully cleaned for any kind of dead material, and only the green and brown-green shoots from the last three years of growth are included in the sample. Brown parts should not be included, even though the green parts only represent the last two to three years of growth.

## 2.2. Registration of field data

For each sampling location, a collection sheet with information regarding location, time and person responsible for collection should be filled out. Any deviations from procedures should also be written here.

The coordinates for the position of the collection site should be written preferably as WGS84 (World Geodetic System 1984) in decimal degrees (Statens kartverk, 2009), although as an alternative, UTM/EUREF89 (Universal Transverse Mercator) with the zone 33N (epsg projection 32633) can be used.

## 2.3. Storage before shipping

The samples should be frozen (or kept cool) as soon as possible after the collection until the samples are being sent to the Environmental Specimen Bank.

# 3. Transport of sample

## 3.1. Packing

The collected moss samples should be kept frozen or cool during the whole transport to the Environmental Specimen Bank.

The samples should be wrapped in aluminium foil and put in PE-bags (alternatively; MAGIC VAC® bags). The way of packing should ensure that the samples are transported cold and safe for any kind of damages of the material, and that the mussels are not getting in contact with any areas or compounds that may contaminate the samples. The parcels should be marked clearly with sender's name and address, sent to Environmental Specimen Bank and addressed to a contact person there.

## 3.2. Transport routines

The moss samples should be transported as quickly as possible to the Environmental Specimen Bank, e.g. as an "express over-night"-parcel. A contact person at ESB should be informed on forehand regarding time of delivery to ensure that the parcel is received in a proper way. If post or shipping companies are used, the parcel must be sent in a traceable manner.

# 4. Equipment and cleaning procedures

All equipment that is in contact with the samples should be cleaned by solvents or burned in the oven at 450°C for 8h. Nitril-gloves should be worn when handling the samples to avoid contamination.

# 5. Registration of data, marking and freezing samples

Data from the sampling sheet are transferred to the database. In the data base, information about where the samples are stored will be included. This information should include which rack and section of it, which shelf and box where the sample is stored. All glasses should be labelled with (freezing safe labels) unique sampling numbers. After transfer of sample material to the glasses, they should be closed and frozen at -25 °C in the freezer of the Environmental Specimen Bank.



## 6. Referenses

ICP Vegetation (2015)

## 7. Appendix

Appendix 1. Sampling sheet

Site no.	Site name	Position (in degrees)		YEAR:	Other samples collected			Species*	Person sampling
		Latitude (N)	Longitude (E)	Dato	POP-samples	Humus	Vegetation		
3	Aremark								
24	Kjelsås								
29	Nordmoen								
43	Kise								
61	Narbuvooll								
82	Heidal								
127	Åmot								
139	Birkeland								
148	Risør								
190	Lund								
213	Fitjar								
245	Hovlandsdal/ Fjaler								
271	Valderøy/ Godøy								
275	Molde								
315	Verrabotn								
330	Joma								
372	Festvåg								
393	Andenes								
401	Øverbygd/ Målselv								
432	Lakselv								

\*BP = Birch (*Betula pubescens*), VM = Blueberry (*Vaccinium myrtillus*), DF = Wavy hair grass (*Deschampsia flexuosa*), CV = Heather/shrub (*Calluna vulgaris*), Emp= Crowberry (*Empetrum*)