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Ambient air — Biomonitoring with lichens — Assessing epiphytic lichen diversity

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National foreword

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Außenluft - Biomonitoring mit Flechten - Kartierung der Diversität epiphytischer Flechten

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Foreword

This document (EN 16413:2014) has been prepared by Technical Committee CEN/TC 264 “Air quality”, the secretariat of which is held by DIN.

This European Standard shall be given the status of a national standard, either by publication of an identical text or by endorsement, at the latest by August 2014, and conflicting national standards shall be withdrawn at the latest by August 2014.

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0 Introduction

0.1 Biomonitoring and air quality

The impact of air pollution is of growing importance worldwide. Local and regional assessment is necessary as a first step to collect fundamental information, which can be used to avoid, prevent and minimize harmful effects on human health and the environment as a whole. Biomonitoring may serve as a tool for such a purpose. As the effects on indicator organisms are a time-integrated result of complex influences combining both air quality and local climatic conditions, this holistic biological approach is considered particularly close to human and environmental health end points and thus is relevant to air quality management.

It is important to emphasize that biomonitoring data are completely different from those obtained through physico-chemical measurements (ambient concentrations and deposition) and computer modelling (emissions data). Biomonitoring provides evidence of the effects that airborne pollutants have on organisms. As such it reveals biologically relevant, field-based, time- and space-integrated indications of environmental health as a whole. Legislation states that there should be no harmful environmental effects from air pollution. This requirement can be met only by investigating the effects at the biological level. The application of biomonitoring in air quality and environmental management requires rigorous standards and a recognized regime so that it can be evaluated in the same way as physico-chemical measurements and modelling in pollution management.

Biomonitoring is the traditional way through which environmental changes have been detected historically. Various standard works on biomonitoring provide an overview of the state of the science at the time, e.g. [1], [2], [3]. The first investigations of passive biomonitoring are documented in the middle of the 19th century: by monitoring the development of epiphytic lichens it was discovered that the lichens were damaged during the polluted period in winter and recovered and showed strong growth in summer [4]. These observations identified lichens as important bioindicators. Later investigations also dealt with bioaccumulators. An active biomonitoring procedure with bush beans was first initiated in 1899 [5].

0.2 Biomonitoring and EU legislation

Biomonitoring methods in terrestrial environments respond to a variety of requirements and objectives of EU environmental policy primarily in the fields of air quality (Directive 2008/50/EC on ambient air, [6]), integrated pollution prevention and control (Directive 2008/1/EC [7], and Directive 2010/75/EU [8]) and conservation (Habitats Directive). The topics food chain ([9]) and animal feed ([10], [11], [12]) are alluded to as well.

For air quality in Europe, the legislator requires adequate monitoring of air quality, including pollution deposition as well as avoidance, prevention or reduction of harmful effects. Biomonitoring methods appertain to the scope of short and long-term air quality assessment.

Directive 2004/107/EC of 15 December 2004 relating to arsenic, cadmium, mercury, nickel and polycyclic aromatic hydrocarbons in ambient air ([13]) states that “the use of bio indicators may be considered where regional patterns of the impact on ecosystems are to be assessed”.

Concerning IPPC from industrial installations, the permit procedure includes two particular environmental conditions for setting adequate emission limit values. The asserted concepts of “effects” and “sensitivity of the local environment” open up a broad field for biomonitoring methods, in relation to the general impact on air quality and the deposition of operational-specific pollutants. The basic properties of biomonitoring methods can be used advantageously for various applications such as reference inventories prior to the start of a new installation, the mapping of the potential pollution reception areas and (long-term) monitoring of the impact caused by industrial activity. The environmental inspection of installations demands the examination of the full range of environmental effects. For the public authority, biomonitoring data contribute to the decision-making process, e.g. concerning the question of tolerance of impacts at the local scale.

The Habitats Directive (92/43/EEC on the conservation of natural habitats and of wild fauna and flora [14]) requires competent authorities to consider or review planning permission and other activities affecting a European designated site where the integrity of the site could be adversely affected. The Directive also

provides for the control of potentially damaging operations, whereby consent may only be granted once it has been shown through appropriate assessment that the proposed operation will not adversely affect the integrity of the site. The responsibility lies with the applicant to demonstrate that there is no adverse effect on such a conservation area. For this purpose, biomonitoring is well suited as a non-intrusive form of environmental assessment.

As an important element within its integrated environmental policy, in 2003 the European Commission adopted a European Environment and Health Strategy ([15]) with the overall aim of reducing diseases caused by environmental factors in Europe. In Chapter 5 of this document it is stated that the “community approach entails the collection and linking of data on environmental pollutants in all the different environmental compartments (including the cycle of pollutants) and in the whole ecosystem (bio-indicators) to health data (epidemiological, toxicological, morbidity)”. The European Environment and Health Action Plan 2004-2010 ([16]) which followed the adoption of this strategy focusses on human biomonitoring, but emphasizes the need to “develop integrated monitoring of the environment, including food, to allow the determination of relevant human exposure“.

0.3 Biomonitoring with lichens

Many lichens, due to their morphological, ecological and physiological peculiarities, are extremely sensitive to changes in their environment ([17], [18]) such as eutrophication ([19], [20]), climate ([21], [22]) and woodland management ([23], [24]).

Lichen diversity is an excellent indicator of pollution from phytotoxic gaseous substances ([18], [25]). Lichens respond relatively fast to a deterioration in air quality and can re-colonize urban and industrial environments as a consequence of changing conditions within a few years, as recorded in many parts of Europe (e.g. [26], [27]).

The method described here determines the actual state of lichen diversity before or after exposure to air pollution and/or other types of environmental stresses. Correlative studies between lichen diversity and epidemiological studies suggest that bioindicators can be useful tools for detecting the possible effects of air pollution on human health ([28]).

This European Standard proposes a standardized method to assess lichen diversity on tree bark and is largely based on the German VDI standard on lichen mapping ([29], [30]), the French national standard ([31]), the Italian guidelines ([32], [33]) and the publication by *Asta* et al. ([34]). The interpretation of geographic patterns and temporal trends of lichen diversity may be assisted by using ecological indicator values ([35], [36], [37], [38]), multivariate statistics, such as numerical analysis of matrices of species ([39], [40]), non-parametric models ([41], [42]) or other statistical tools.

1 Scope

This European Standard aims to provide a reliable, repeatable and objective method for assessing epiphytic lichen diversity. According to international literature on the topic (see e.g. [18] for an overall outline), it provides a framework for assessing the impact of anthropogenic intervention, particularly for estimating the effects of atmospheric pollution.

2 Terms and definitions

For the purposes of this document, the following terms and definitions apply.

2.1
biomonitoring
use of biological systems (organisms and organism communities) to monitor environmental change over space and/or time

Note 1 to entry: Biological systems can be further considerd as bioindicators.

2.2
bioindicator
organism or a part of it or an organism community (biocoenosis) which documents environmental impacts

Note 1 to entry: It encompasses bioaccumulators and response indicators.

2.3
bioaccumulator
organism which can indicate environmental conditions and their modification by accumulating substances present in the environment (air, water or soil) at the surface and/or internally

2.4
response indicator
effect indicator
organism which can indicate environmental conditions and their modification by either showing specific symptoms (molecular, biochemical, cellular, physiological, anatomical or morphological) or by its presence/absence in the ecosystem

2.5
lichen
ecologically obligate, self-supporting symbiotic association of a fungus (the mycobiont, generally an ascomycete) and one or more populations of green algae and/or cyanobacteria (the photobionts), which results in a stable vegetative structure (“thallus”) with a definite morphology

2.6
lichen community
biocoenosis
assemblage of populations of lichens, whose composition and aspect is determined by the properties of the environment and by their relationship with other epiphytes, animals, etc.

2.7
lichen diversity
species richness found on the bark of standard trees at a height ranging between 1 m and 1,5 m, above the base of the trunk at four different aspects (NSEW)

Note 1 to entry: See Annex B.

2.8
epiphyte
plant or plant-like organism growing on another plant, dependant on mechanical support but not deriving nutrients from the plant upon which it grows

2.9
study area
geographical area considered by the study

Note 1 to entry: It should be described in detail in terms of extent, land use classification and altitudinal range.

2.10
study domain
geographical extent in which the target population is studied

Note 1 to entry: It may coincide with the study area or it may be more restricted.

2.11
sampling point
geographic location identified by a pair of geographic coordinates (Lat, Long), being the reference point for the Sampling Unit, selected on the basis of a given sampling design

2.12
sampling unit
SU
either single tree or cluster of trees (tree-based sampling) or plot (geographical area of determined size, centred on a sampling point) where data are collected

2.13
probabilistic sampling
sampling conducted according to the statistical principles of sampling

Note 1 to entry: The essential principle of probabilistic sampling is that every individual particle or item in the population has an equal chance of being sampled.

2.14
stratified random sampling design
technique consisting of subdividing a heterogeneous population into sub-populations (strata), which are more homogeneous and mutually exclusive

Note 1 to entry: Within each stratum the samples are consequently independent and randomly selected.

2.15
target population
lichen communities living on the bark of standard trees at a height ranging between 1 m and 1,5 m, starting from the base of the trunk at each main aspect

Note 1 to entry: Standard trees should be defined in terms of species, bole circumference and inclination (see below) and should be located within the study area.

3 Principles

The procedure is widely applicable for the purposes of collecting lichen diversity data. The interpretation of the results, however, shall be adapted to the regional characteristics of the lichen flora and to the prevalent types of environmental stress. Different methods may be used to solve particular problems, or in particular areas.

For quality assurance purposes, investigations performed according to this European Standard require personnel or institutions to have the necessary level of expertise in the fields of lichenology and probabilistic sampling design.

4 Equipment

4.1 Field work preparation equipment

4.1.1 Maps.

The choice of the map scale depends on the study area dimension and on the intended use of the map. Different scale maps will be necessary, both small scale maps for the study areas (e.g. 1:250 000 scale map) and large scale maps (at least 1:25 000 scale maps but also 1:10 000 and 1:5 000 scale maps) that may be useful for the location of the SU in the field.

4.1.2 Geographic Information System (GIS) with land use strata based on the Corine Land Cover nomenclature.

Other important sources of information may be the analysis of aerial photographs, of town and country planning maps, or ecological maps. The topography may also be used as an additional stratum in those regions presenting significant variation in topographical relief.

4.1.3 Identification of SU on the 1:25 000 (or more detailed if necessary) scale map.

The limits of the SU, and of their possible replacements, will be drawn on the map in order to facilitate field work.

4.1.4 Algorithm for random sampling (scientific calculator or statistics software).

4.2 Field equipment

4.2.1 Chemical reagents for spot tests on lichen thalli.

To produce characteristic colour changes. In particular, the most commonly used reagents are calcium hypochlorite (C), potassium hydroxide (K) and para-phenylendiamine dilution (P).

4.2.2 Compass-clinometer, essential to find the correct positioning of the observation grids on the trunk of the selected trees and also to measure bole inclination.

4.2.3 Identification keys.

Keys may be useful to distinguish species in the field (see Bibliography).

4.2.4 Envelopes.

Specimens to be transported to the lab for identification should be placed in separate, labelled envelopes. The use of paper envelopes is recommended to avoid the growth of mould on the lichen samples.

4.2.5 Global Positioning System (GPS) receiver.

4.2.6 Knife.

This is important to remove lichen samples from the bark of selected trees.

4.2.7 Magnifying glass.

It is essential to have a lens that magnifies by at least × 10 but a × 20 lens is also recommended for crustose lichens.

4.2.8 Maps.

See 4.1.1.

4.2.9 Tape measure: at least a 20 m tape measure: useful for measuring tree circumference.

4.2.10 Observation grid, 10 cm × 50 cm grid (Figure 1), subdivided into five 10 cm × 10 cm quadrats, to be applied to the trunk of sample trees for example by means of rubber bands.

The grid shall be flexible enough to be easily placed on the bole but also sufficiently robust and resistant so as to prevent changes in shape and in dimensions with use.

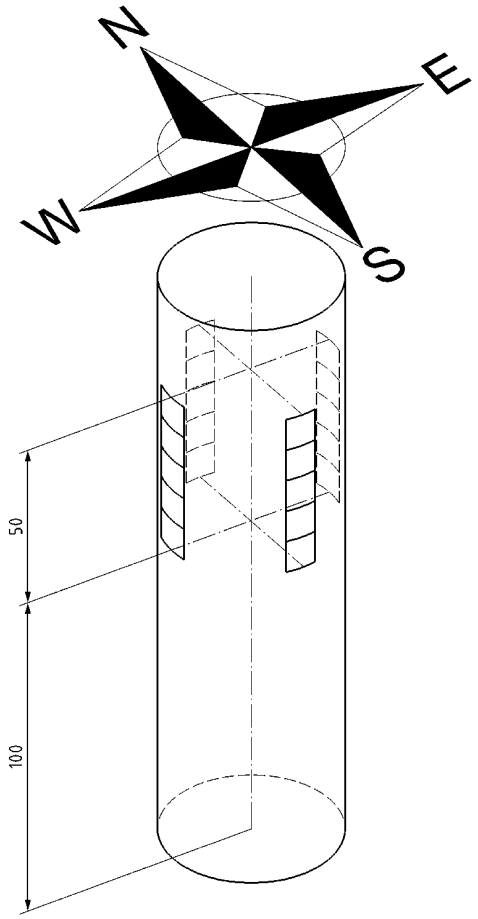


Figure 1 — Observation grids attached at four aspects along the trunk (see 5.4.8)

4.2.11 Survey sheets: Sampling Unit survey sheet (see Annex A).

4.3 Laboratory equipment

4.3.1 Chemical reagents traditionally used in lichenology.

Solutions of iodine (I), calcium hypochlorite (C), and potassium hydroxide (K), para-phenylendiamine (P). For solution preparation procedures refer to *Purvis* et al. ([43]). Blotting paper to highlight the spot test reaction on

lichen thalli. For the determination of several critical genera, such as *Lecanora*, *Pertusaria*, *Parmotrema*, *Cladonia*, *Lepraria*, some thin layer chromatography (TLC) analysis may be necessary and should be carried out according to the method suggested by *Culberson* ([44]) and *Culberson* et al. ([45]).

4.3.2 Identification keys (see Bibliography).

4.3.3 Online lichen checklists (see Bibliography).

They may be useful for nomenclature and ecological and distributional information of lichen species found in the field.

4.3.4 Optical microscope (required magnification: × 400 up to × 1 000), used for high-power magnification of lichen structures such as asci and spores.

An eyepiece micrometer will also be necessary to measure spore dimensions. Polarisator appliance is also recommended for the determination of several groups of species (e.g. *Lecanora* spp.).

4.3.5 Stereo microscope (minimum range × 10 to × 60), used for low-power magnification of lichen samples.

4.3.6 Usual small laboratory equipment (tweezers, scalpel or razor blades, microscope slides and cover slips, immersion oil, pipettes).

5 Sampling

5.1 General

Sampling is the act or process of selecting a part (the sample) of something (the target population), with the intent of reflecting its quality, style and nature. Since there are many possible designs, and the most effective one depends on the nature of the population being investigated, there is no unique sampling design that can be recommended for all studies. Rather, the probabilistic nature of the sampling design shall always be maintained. The following guidelines are provided in order to drive the main steps in defining the sampling design for individual studies. A synthetic flow chart showing the main steps to be followed is provided in Annex F.

5.2 Sampling objective

The sampling objective is to obtain an estimate of the parameter of the response variable (e.g. mean species richness or mean Lichen Diversity Value; LDV) over the study domain with a given precision. The precision level should be expressed in terms of confidence intervals for the defined probability level. It is required that the sampling objective is defined for each study.

EXAMPLE Obtain an estimate of the mean LDV for the study domain with a confidence interval ± 10 % of the mean value, at a probability (P) level of 95 %.

The computation of estimates and confidence intervals depends on the sampling design adopted. Therefore each study shall define precision and probability levels, taking into account the requirements of the study framework and considering the available resources.

5.3 Study type considered

Lichen diversity assessment and monitoring is a typical observational, mensurative study. Studies can be classified with respect to their temporal coverage:

a) the study is a baseline;

b) the study implies a series of subsequent measurements/repeated measurements in time over the study domain.

This European Standard includes guidelines focussed on Case a). When evaluation of change between subsequent measurements is of interest (Case b)), in addition to the following guidelines, it is important to consider the implications related to the statistical analysis for detecting changes (see [46]).

5.4 Sampling design

5.4.1 General

Sampling design is a randomized scheme to select a subset S (the sample) of elements from the population (P). It allows identification of the set of possible samples which can be selected by the scheme with the corresponding probabilities of selection.

5.4.2 Prior to sampling

The sampling scheme should be decided on the basis of the characteristics of the study area and the aim of the study. Some preliminary steps should be taken before selecting the most appropriate sampling scheme.

— The distribution of potentially suitable trees (see below) over the study area should be as well-known as possible, e.g. by using aerial imagery or analogous information source, or by means of a preliminary inspection of the study area.

— In the case of gradient studies quantitative information concerning the gradient should be considered and, if possible, a thematic layer map (e.g. prevailing point sources of pollution, or wind direction and intensity) should be produced.

— Any kind of restricted access, such as private estates and military areas, should be preliminarily checked in order to include or exclude those areas from the study area.

5.4.3 Standard tree species selection for a survey

Suitable tree species are those with sub-acidic, neutral or basic bark, excluding some conifers and other *taxa* with a high degree of bark exfoliation: a complete list of suitable species is provided in Annex C. Suitable *taxa* are arranged into groups, based on comparable characteristics of the bark (i.e. pH, texture, exfoliation, water retention).

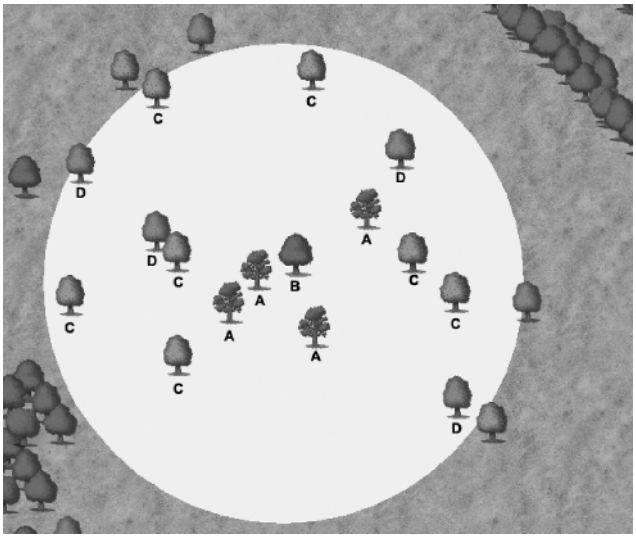
When choosing tree species to be sampled in a given study area, the following options (in preference order) should be considered (Figure 2):

a) a single tree species within the whole study area;

b) different tree species, within the same bark-type group;

c) different tree species within different bark-type groups (excluding unsuitable *taxa*, e.g. some conifers, *Platanus*, see Annex C).

The two latter options require direct assessment of lichen diversity differences, by means of appropriate statistical analysis. It is recommended to compare the results obtained on different tree species within the same study domain.



NOTE Four trees of species A, 1 tree of species B, 7 trees of species C and 4 trees of species D are included within the Sampling Unit:

— 5.4.3 a): only 4 trees of the species A (Group I) are selected; other possible trees are excluded because they belong to other species of the same bark group (species B), to other bark groups (species C) or to unsuitable species (species D);

— 5.4.3 b): 4 trees of species A (Group I) and 1 tree of species B (Group I) are selected; other possible trees are excluded because they belong to other groups (species C) or to unsuitable species (species D);

— 5.4.3 c): 4 trees of species A (Group I), 1 tree of species B (Group I) and 2 trees of species C (Group II) are selected; trees of species D are excluded because they belong to other groups or to unsuitable species.

Figure 2 — Standard tree selection at plot level, according to different strategies previously agreed for monitoring the study area

5.4.4 Standard tree parameter

A standard tree is defined as follows:

a) it is a suitable species (see 5.4.3);

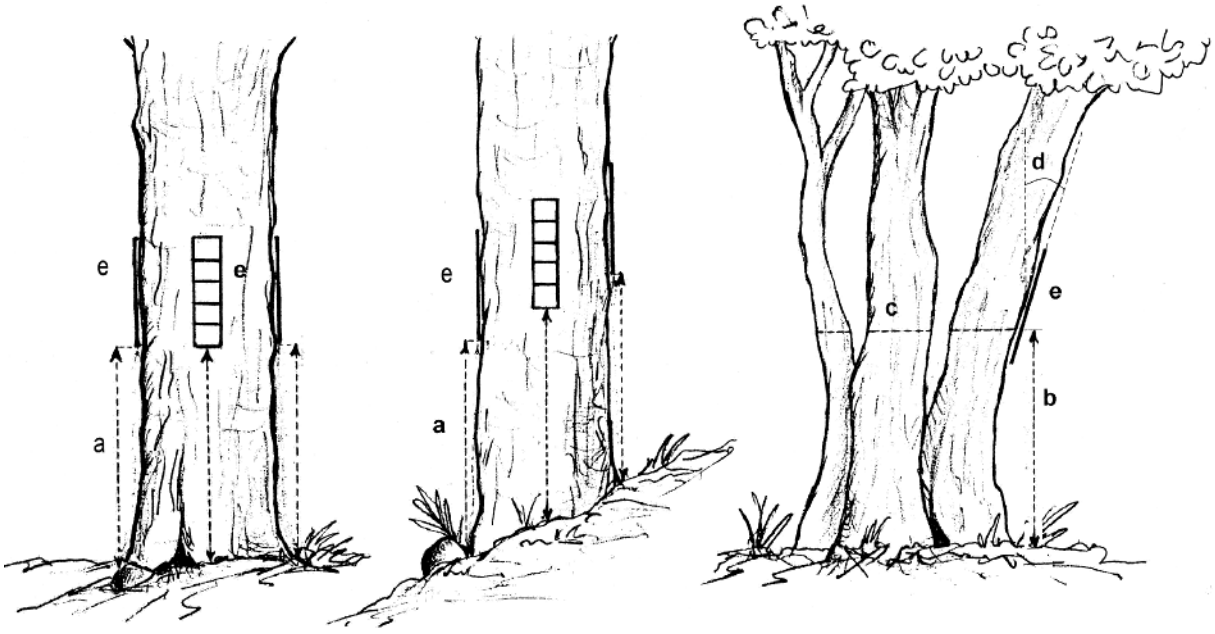
b) it has a trunk circumference (at 130 cm from the ground level; see Figure 3 for details on how to measure) between 50 cm and 250 cm;

c) each exposition (N, E, S, W) has an inclination (at the centre of each grid) < 20°;

d) the area of the trunk that is unsuitable for recording (damage, decortication, branching, knots and/or other epiphytes or climbing plants such as ivy, preventing growth of lichens) within each of the 4 grids when summed < 20 %.

Note that if the four grids cannot be adequately positioned on the trunk – meeting points c) and d) – some replacement procedures at tree level may be followed (see 5.4.8).

Stems that have clear separation below 100 cm above the ground should be considered separate trees.



Key

a height from the ground level to the lower edge of the grid (a = 1 m)

b height to measure the circumference of the trunk (b = 1,3 m)

c circumference of the trunk

d inclination at the center of the grid

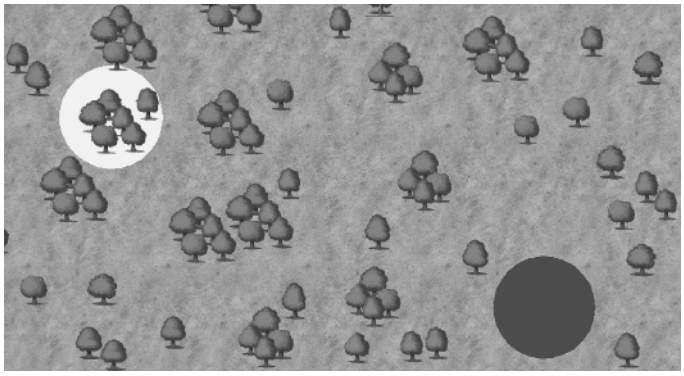
e observation grid

Figure 3 — Measurement height from the ground level for positioning the lower edge of the grid (1 m) and location for measuring the circumference and the inclination of the trunk (1,3 m)

5.4.5 Sampling scheme

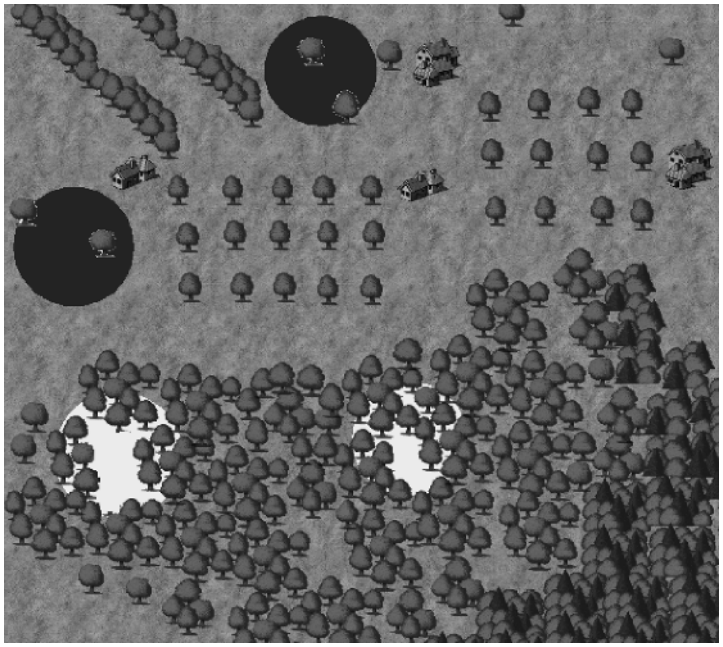
5.4.5.1 General

The sampling design varies according to the distribution of the standard trees within the study domain. Different designs are suggested as examples (see also Figure 4), mainly on the basis of ecological homogeneity or heterogeneity of the study domain. In each study, ecological variables to assess homogeneity should be explicitly provided (e.g. altitude, land use, resident population density, vegetation), according to the aim of the study.



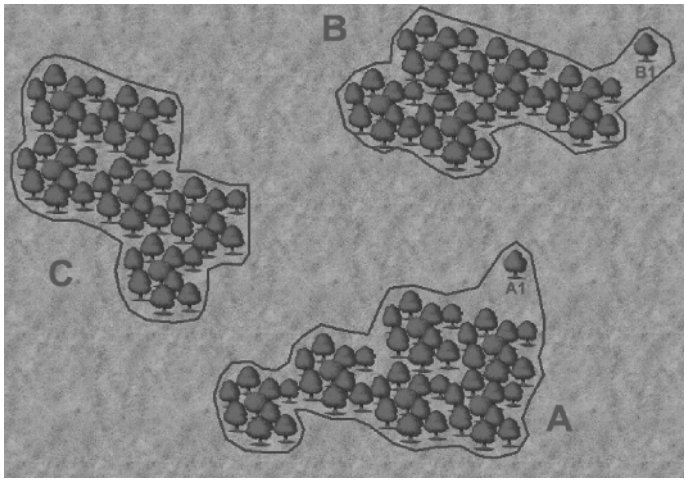
a) Case 5.4.5.2 a) for a homogeneous area; standard trees abundantly and homogeneously distributed over the study domain

Plot sampling: light plot has been randomly selected and defined (7 trees are included). Dark plot has been selected but a priori excluded because it does not contain any trees.



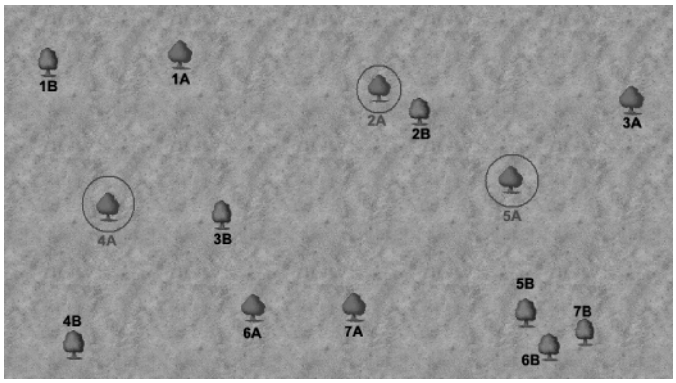
b) Case 5.4.5.3 a) for a heterogeneous area; stratified random design with two strata: agricultural area (dark circles); forested area (light circles)

Figure 4 ***(continued)***



c) Cases 5.4.5.2 b) and 5.4.5.3 b); standard trees abundantly scattered in clusters over the study domain

Cluster sampling. Trees A1 and B1 are included in clusters A and B, respectively, following the criterion of the threshold distance from the centroid of the cluster.



d) Cases 5.4.5.2 c) and 5.4.5.3 c); standard trees scattered infrequently over the study domain

Tree-based sampling: trees 2A, 4A and 5A have been randomly selected from among the population of isolated trees of species A. Trees 1B-7B belong to an unsuitable species and are excluded from the tree population.

Figure 4 — Examples of different sampling regimes (see 5.4.5)

5.4.5.2 Sampling schemes in ecologically homogeneous areas

a) Standard trees abundantly and homogeneously distributed over the study domain:

A simple random or systematic design is recommended. Plot sampling is recommended, with sample plots allocated according to a regular grid, with the starting point of the grid chosen at random.

b) Standard trees abundantly scattered in clusters over the study domain:

Tree-based cluster sampling or two stage sampling are recommended. Firstly define a criterion to identify clusters (e.g. defining a threshold maximum distance between adjacent trees to be included within the same cluster and/or defining a threshold minimum distance for two clusters to be considered as separate sampling units), then identify and list all the clusters and obtain a random sample of them. If the average

number of standard trees per cluster within the study domain is reasonable (≤10), perform measurements on all of them. Otherwise, proceed with a two stage sampling: obtain a random selection of trees within the cluster and perform measurements on all the trees selected. Sub-sampling of the SU introduces a further source of variability which may affect the quality of the data. It is important to take this into account when performing statistical analysis of the data.

c) Standard trees infrequently scattered over the study domain:

A simple tree-based random sampling is recommended. Obtain a list of the individual trees on the basis of the aerial photo and select the sample trees at random.

5.4.5.3 Sampling scheme in ecologically heterogeneous areas

a) Standard trees abundant and homogeneously distributed over the study domain:

A stratified random sampling design is recommended. Plot sampling is recommended. First, identify strata on the basis of the information available on the heterogeneous ecological variables (e.g. altitudinal maps, land use classification, forest type). Subsequently calculate the sampling density (see below); allocate sample plots on a random basis within the strata, and in proportion to the dimension of the strata.

b) Standard trees abundant, scattered in clusters over the study domain:

A stratified random sampling design is recommended. Tree-based cluster sampling or two stage sampling are recommended. Firstly, define a criterion to identify clusters (e.g. defining a threshold maximum distance between adjacent trees to be included within the same cluster and/or defining a threshold minimum distance for two clusters to be considered as separate SUs), based on the information available on the heterogeneous ecological variables (e.g. altitudinal maps, land use classification), identify and list all the clusters and obtain a random sample of them. If the average number of standard trees per cluster within the study domain is reasonable (≤ 10), perform measurements on all of them.

Otherwise, proceed with two stage sampling: obtain a random selection of trees within the cluster and perform measurements on all the trees selected. Sub-sampling of the sampling units introduces a further source of variability which may affect the quality of the data. It is important to take this into account when performing statistical analysis of the data.

c) Standard trees infrequently scattered over the study domain:

Use a tree-based stratified random design. Obtain a list of the individual trees per stratum on the basis of the aerial photo and select the sample trees at random.

5.4.6 Sampling unit

— **Plot sampling**: each plot represents a sampling unit. As selected plot size depends on tree density in the study domain, different sizes could be considered. In most cases, a circular sampling plot with a radius of 30 m is recommended. Each plot shall be allocated, centred on a sampling point. All the standard trees in the plot are measured.

— **Tree-based sampling**: each tree or cluster of trees represents a sampling unit.

5.4.7 Sampling density

The minimum number of plots to be selected should be calculated on the basis of available sample size formulae for different designs (e.g. [46]). Usually, these formulae require preliminary information on data variability in a given study area, so that a pilot study and/or the revision of data from previous campaigns (or surveys carried out in comparable areas) are recommended before embarking on a formal investigation. Further inputs in terms of expected precision are also required. Practical examples are reported in Annex D.

5.4.8 Surveying lichens

The observation grid (4.2.10, Figure 1) is attached for example by means of rubber bands along the trunk so that its lower edge is always 1 m above the base of the trunk (see Figures 1 and 3). The grid shall be placed at four different aspects of the trunk (NSEW).

It is possible to relocate each monitoring grid by a maximum shift of 20° (firstly in a clockwise, then in counter clockwise direction), to avoid parts of the trunk which are not suitable (see 5.4.4) for sampling (e.g. wounds, knots).

Even if a high lichen cover is present, a grid shall be shifted if the extent of the disturbances (namely damage, decortication, branches, knots and/or other epiphytes or climbing plants such as ivy, preventing growth of lichens) is higher than 20 % (however, lichens growing on bryophytes shall be included in the list of species).

At each aspect, examine all five quadrats of the grid for lichen species. Record the occurrence of each species within each quadrat, and calculate the frequency of each species in the grid (by summing up the quadrats in which the species are found, ranging from 0 to 5) and document them (see Annex A).

The record list shall include all species. However, a few small crustose lichens are particularly difficult to identify and/or are easily overlooked. Where the identification of certain thalli is troublesome both in the field and/or in the laboratory, it is advisable to include them in the calculation of diversity as "*Sp. nr. x*", having established that they are not damaged or poorly developed forms of species already occurring in the monitoring grid.

No lichens should be removed from within the observation grid as future surveys and/or Quality Control procedures may be planned. Where species identification requires laboratory investigation equivalent specimens should be removed from the trunk area outside of the quadrat. If this is not possible a small fragment should be removed from the specimen within the quadrat without causing harm to the thalli.

5.4.9 Identification in laboratory of critical specimens

Critical specimens should be identified at the laboratory (collected as described in 5.4.8), according to the methods described in the published literature (see Bibliography, Identification keys), using the equipment reported in 4.3.

There may be instances where it may be desirable to retain reference specimens for some species (excluding endangered species) for storage.

6 Lichen species frequencies

The basic results of the sampling of lichen diversity provide aggregated matrices of the species frequencies at several spatial levels of sampling:

— matrix of species × aspect at each tree;

— matrix of species × trees;

— matrix of species × sampling units.

Based on this information different ecological and distributional features may be evident (see Annex B).

7 Recommendations for Quality Assurance and Quality Control

Quality Assurance (QA) and Quality Control (QC) procedures can be implemented to control errors and document the overall quality of the survey/monitoring campaign (see [47], [48], [49], [50]). These procedures form an integral part of the study design and results (see e.g. [51], [52], [53], [54]). Studies failing to report

their QA procedures and Quality Control results (with particular reference to points a) and b)) shall be regarded as incomplete.

QA and QC procedures should be described in an ad hoc document, termed Quality Assurance Plan (QAP) of the study. The QAP should report the minimum acceptable set of QA/QC procedures. An example of information needed at the end of the survey is reported in Annex E. The QAP should cover the following areas of concern:

a) Project management: a description of the organization and management of the study and its requirements; this document should include:

    1) Identification of responsibilities: responsible person(s) from the funding agency and/or of the organization that will carry out the study should be clearly identified, as well as their role and responsibilities in the project.

    2) Description of the aim of the study, the problem being targeted and the final use of the information arising from the study.

    3) Sampling objective of the study, defined according to 5.2 and taking into account the data quality objectives (see below).

    4) Data Quality Objectives (DQO), i.e. qualitative and quantitative statements that clarify the study's technical and quality objectives: they may vary with the study of concern: they define the appropriate type of data, and specify tolerable levels of potential decision errors that will be used as the basis for establishing the quality and quantity of data needed to support decisions.

    5) Data Quality Indicators (DQI): the quantitative statistics and qualitative descriptors used to interpret the degree of acceptability or utility of data to the user. Two indicators can be considered: the Measurement Quality Objective, defined as the expected level of accuracy of the data; and the Data Quality Limits, defined as the minimum acceptable level of consistency between control data and surveyor data. Values for these two indicators should be established.

    6) Needs in terms of training and expertise of the personnel involved and of any certification and licence required.

    7) Permission to access private areas or areas with restricted access.

b) Standard Operating Procedures (SOP) should be adopted. SOPs should be consistent with this European Standard and shall provide details related to any adaptation needed for the particular conditions of the study area. If necessary, SOPs should be annexed to the QAP as a separate document. SOPs should be signed off by the responsible person (see item a) 1)) and explicitly accepted by the personnel involved. SOPs should include:

    1) Description of the sampling design adopted and its justification in relation to the study objectives.

    2) Description of the method to be used to identify sample location on the maps and in the field.

    3) Description of data collection methods in the field, including equipment and field forms.

    4) Description of the chain of custody of the data (recording in the field, storage in electronic format, transmission to central database – if any).

    5) Description of materials and consumables.

    6) Description of software and hardware to be used in the different phases of the study.

c) Data Quality Control (QC) activity: the set of activities aimed at ensuring compliance with DQO and DQIs should be described here. QC activity could include:

   1) Identification of requirements for surveying personnel; the certification of the survey team could consider the observer error in each sampling phase (see Annex F).

   2) Selection of plots and trees in the field.

   3) Tree species identification.

   4) Positioning of the sampling grid on the bole.

   5) Taxonomic identification of lichen species.

   6) Training procedures: description of what training procedures can be adopted to make personnel familiar with the SOPs.

   7) Calibration: description of what exercises will be carried out to promote calibration amongst the personnel involved.

   8) Field checks: each study could include field controls, carried out by an independent team, documenting the meeting of Data Quality Objectives (DQO). It is suggested that two phases be considered in the field checks:

      i) Observation of surveying personnel while working;

      ii) Independent measurements carried out in randomly selected plots.

   9) Description of criteria and procedures to be adopted to accept, reject or qualify study information.

Annex A
(informative)

Example of survey sheets

The following data should be recorded:

a) Sampling Unit survey sheet:

1) date of Sampling Unit (SU) survey;
2) SU code and sub-sample unit if expected (e.g. trees within a plot or a cluster of trees);
3) locality (region, municipality, etc.);
4) routes to reach the sampling unit;
5) description of the SU;
6) photo of the SU;
7) topographic map of the SU;
8) altitude (min. – max.);
9) land use;
10) selected tree species;
11) number of selected trees;
12) coordinates for each selected tree.

b) Tree survey sheet:

1) date of tree selection;
2) SU code;
3) tree code;
4) tree species;
5) tree circumference;
6) tree coordinates;
7) inclinations of the bole at the four aspects;
8) description of the location of the selected tree;
9) photo of the selected tree;
10) location of the selected tree on a topographic map;

11) optional: other variables potentially useful for interpreting the data (e.g. canopy coverage, height of the branches);

12) degrees and direction of grid(s) shifting (if necessary);

13) frequencies of lichen species on the examined trees (matrix of species for each tree).

SU code												
Date:					**Operators:**							
Tree code												
Tree species												
Tree circumference (cm)												
Grid aspect	**N**	**E**	**S**	**W**	**N**	**E**	**S**	**W**	**N**	**E**	**S**	**W**
Disturbance coverage (%)												
Shift of the grid (°)												
Lichen species												
1)*Xanthoria parietina*												
2)*Physcia adscendens*												
3)*Parmelina tiliacea*												
4)……..												
5)												
6)												
7)												
8)												
9)												
10)												
etc.												

Annex B
(informative)

Calculating lichen diversity metrics

B.1 General

Possible ways to interpret the data afterwards are reported as an example. The following examples are not exhaustive and other interpretative tools may be applied to basic data.

B.2 Lichen Diversity Value (LDV)

Amongst the possible results derivable from the basic species × SUs matrix, the Lichen Diversity Value (LDV – *Asta* et al. 2002, [34]) is one of the most used in applicative biomonitoring surveys. The first step in calculating the LDV of a sampling unit (j) is to sum the frequencies of all lichen species found on each tree (t) within the unit.

Since substantial differences in lichen growth may be expected on different sides of the trunks, the frequencies shall be summed separately for each aspect.

Thus, for each tree, there are four Sums of Frequencies (tree t: SF_{Nt}, SF_{Et}, SF_{St}, SF_{Wt}).

Next, for each tree, the Lichen Diversity Value is calculated:

$$LDV_t = SF_{Nt} + SF_{Et} + SF_{Wt} + SF_{St}$$

where

SF is the sum of frequencies of all lichen species found at one aspect of tree t;

N, E, S, W are respectively North, East, South and West.

The Lichen Diversity Value of a sampling unit *j* (LDV_j) is the arithmetic mean of the LDV_t of all trees within the sampling unit.

$$LDV_j = \left(SF_{1t} + SF_{2t} + SF_{3t} + SF_{4t} + \ldots\ldots + SF_{nt}\right) / n$$

where

SF is the sum of frequencies of all lichen species found at each tree (1t, 2t, etc.);

n is the number of trees sampled in unit j.

Further parameters may be derived from the data set of the species frequencies. They include e.g. morpho-functional guilds (e.g. macrolichens, nitrophilous vs. acidophilous species) that have been successfully used for detecting particular sources of atmospheric pollution (see VDI 3957 Part 13 [30]).

However, these alternative approaches require clear operational objectives, definitions that are not formally included nor described in this European Standard.

B.3 Diversity value of the indicators of eutrophication (e.g. LDVN *sensu* VDI 3957 Part 13)

It is calculated in the same way as LDV, but only considering those species (nitrophytes) associated with high eutrophication (e.g. nitrogen compounds and/or dust). As the national flora and the ecological requirements of the species themselves may vary considerably across Europe, proper lists of these species shall be provided at country or eco-regional level and they are not reported here.

Table B.1 — Example of calculation of LDV and LDVN

Tree 1	**Nitrophyte**	**N**	**E**	**S**	**W**	**Species abundance**
Species A	No	5	5	5	5	20
Species B	No	4	3	1	0	8
Species C	No	3	2	4	0	9
Species D	Yes	5	0	2	0	7
Species E	Yes	0	0	0	3	3
Sum of frequencies		17	10	12	8	LDV_t 47
Sum of frequencies of nitrophytes		5	0	2	3	$LDVN_t$ 10

Tree 2	**Nitrophyte**	**N**	**E**	**S**	**W**	**Species abundance**
Species A	No	0	0	0	0	0
Species B	No	0	0	0	3	3
Species C	No	2	1	5	5	13
Species D	Yes	5	5	0	0	10
Species E	Yes	5	5	5	5	20
Sum of frequencies		12	11	10	13	LDV_t 46
Sum of frequencies of nitrophytes		10	10	5	5	$LDVN_t$ 30

Annex C
(informative)

Suitable tree species

Table C.1 lists suitable tree species for sampling of lichen diversity, separated into tree groups with similar bark physico-chemical properties. Indicatively, species belonging to the same group can be used interchangeably. Trees not included in Table C.1 will need to be classified according to their bark properties.

Table C.1 — Suitable tree species

Group I	Group II	Group III	Group IV	Group V	To be tested	Excluded
Acer spp.	*Olea* spp.	*Abies alba*	*Alnus glutinosa*	*Fagus* spp.	*Robinia pseudoacacia*	*Araucaria* spp.
Ceratonia siliqua	*Prunus* spp.	*Larix decidua*	*Betula pendula*	*Carpinus* spp.	*Ailanthus altissima*	*Platanus* spp.
Fraxinus spp.	*Quercus* spp.	microthermic *Pinus* spp.			*Celtis* spp.	*Taxus baccata*
Juglans spp.	*Castanea sativa*	*Picea abies*			*Salix* spp.	*Cycas* spp.
Pyrus communis[a]				*Ostrya carpinifolia*	"Palms"	
Tilia spp.[a]				*Cupressus sempervirens*	mediterranean *Pinus* spp.	
Ulmus spp.	*Malus* spp.				*Alnus cordata*	
Populus spp.	*Ostrya carpinifolia*				*Ginkgo biloba*	
Ficus spp.	*Sorbus* spp.				*Magnolia* spp.	
					Citrus spp.	
					Crataegus spp.	
					Pseudotsuga menziesii	
					"other exotic cultivated plants"	
					any other species not explicitly reported in Table C.1	

[a] According to VDI standards indications ([29], [30]) *Tilia* spp. and *Pyrus communis* can be used in both groups because they hold a middle position with regard to their bark properties.

Annex D
(informative)

Sampling density calculations

Example: for simple random sampling (according to *Elzinga* et al. [46])

In the case of normally distributed populations, the sampling density to obtain estimates of mean with simple random sampling can be calculated as follows:

$$n = \frac{(Z_\alpha)^2 (s)^2}{(B)^2} \quad \text{(D.1)}$$

where

- n is the number of sampling units;
- s is the normalized standard deviation of the response variable (e.g. LDV, LDVN or number of species) in a given survey area, according to existing or preliminary pilot study;
- Z_α depends on P level (for α = 0,05; *P* = 95 %, Z = 1,96);
- B is the desired precision level of the response variable, in terms of half-width of confidence interval.

Subsequent corrections may be necessary (see *Elzinga* et al. [46]).

Annex E
(informative)

Information needed at the end of the survey

Each study should present the following basic data in a table:

— project management;

— Standard Operating Procedures adopted;

— name of the operator(s);

— project manager(s);

— temporal framework of the survey;

— geographical name of the study area;

— number and size of the sampling units;

— tree species surveyed;

— total number of trees surveyed;

— mean number of trees surveyed per sampling unit;

— list of considered response variables, if any;

— standard deviation of the response variable(s) in the sampling units;

— confidence limits of the response variable(s);

— precision of the estimates inter grids, intra-trees, inter-trees;

— Data Quality Control manager(s);

— Data Quality Control activity results.

Annex F
(informative)

Main phases of application of this European Standard

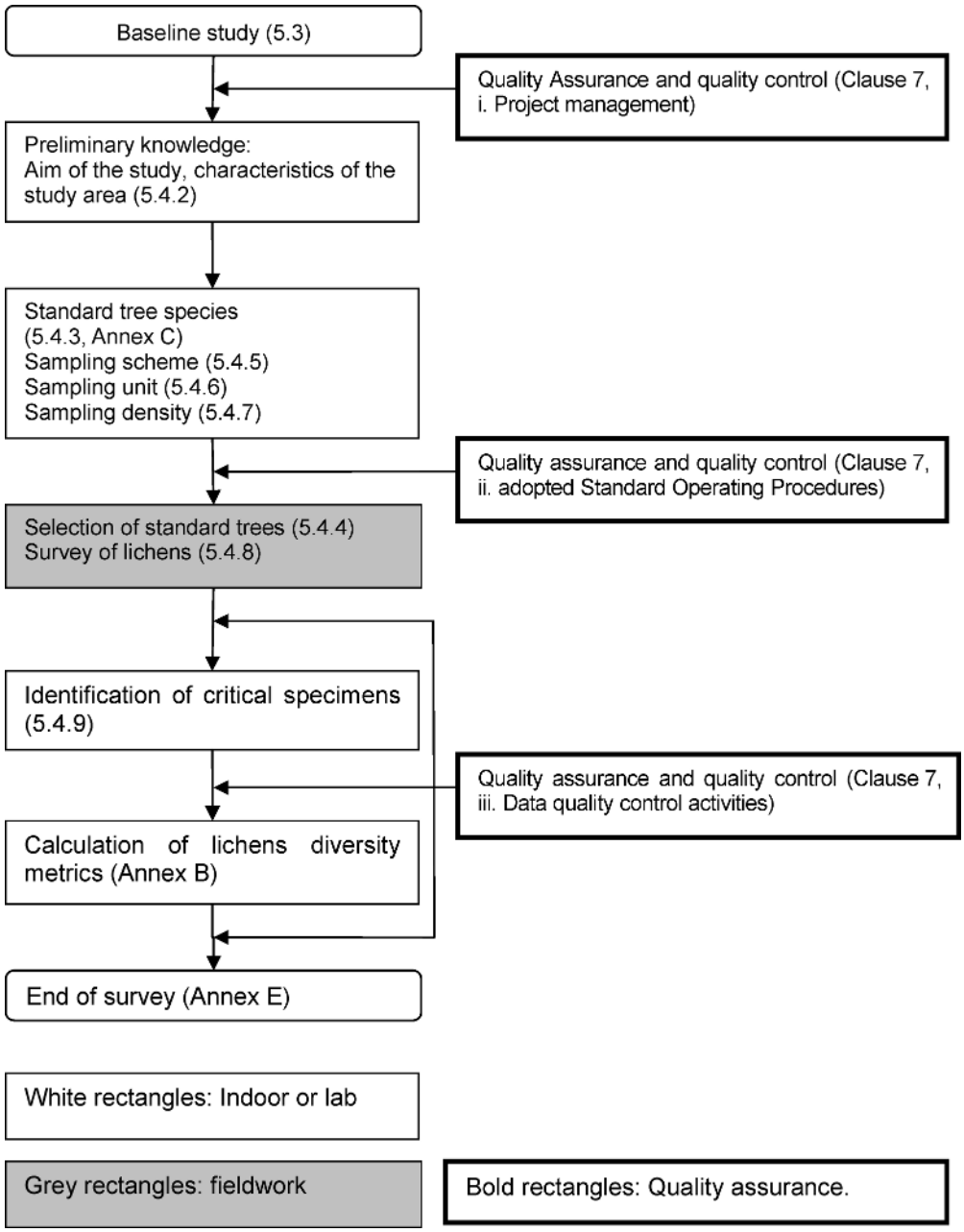


Figure F.1 — Main phases of application

Bibliography

[1] ARNDT U., NOBEL W., SCHWEIZER B. *Bioindikatoren: Möglichkeiten, Grenzen und neue Erkenntnisse*. Ulmer, Stuttgart, 1987

[2] GARREC J.-P., VAN HALUWYN C. *Biosurveillance végétale de la qualité de l'air: concepts, méthodes et applications. Editions Tec et Doc*. Lavoisier, Paris, 2002, 117 p.

[3] KLUMPP A., ANSEL W., KLUMPP G. *Urban Air Pollution, Bioindication and Environmental Awareness*. Cuvillier, Göttingen, 2004

[4] NYLANDER W. Les lichens du Jardin du Luxembourg. *Bull. Soc. Fr.* 1866, **13** pp. 364–372

[5] SORAUER, P. and RAMANN, E.: *Sogenannte unsichtbare Rauchbeschädigungen. Botanisches Centralblatt 80* (1899), pp. 50-60; 106-116; 156-168; 205-216; 251-262

[6] Directive 2008/50/EC of the European Parliament and of the Council of 21 May 2008 on ambient air quality and cleaner air for Europe

[7] Directive 2008/1/EC of the European Parliament and of the Council of 15 January 2008 concerning integrated pollution prevention and control

[8] Directive 2010/75/EU of the European Parliament and of the Council of 24 November 2010 on industrial emissions (integrated pollution prevention and control)

[9] Commission Regulation (EC) No 1881/2006 of 19 December 2006 setting maximum levels for certain contaminants in foodstuffs

[10] Directive 2002/32/EC of the European Parliament and of the Council of 7 May 2002 on undesirable substances in animal feed - Council statement

[11] Commission Directive 2005/87/EC of 5 December 2005 amending Annex I to Directive 2002/32/EC of the European Parliament and of the Council on undesirable substances in animal feed as regards lead, fluorine and cadmium

[12] Commission Directive 2006/13/EC of 3 February 2006 amending Annexes I and II to Directive 2002/32/EC of the European Parliament and of the Council on undesirable substances in animal feed as regards dioxins and dioxin-like PCBs

[13] Directive 2004/107/EC of the European Parliament and of the Council of 15 December 2004 relating to arsenic, cadmium, mercury, nickel and polycyclic aromatic hydrocarbons in ambient air

[14] Council Directive 92/43/EEC of 21 May 1992 on the conservation of natural habitats and of wild fauna and flora

[15] COM/2003/0338 final Communication from the Commission to the Council, the European Parliament and the European Economic and Social Committee - A European Environment and Health Strategy

[16] COM. 2004/0416 Vol. I final Communication from the Commission to the Council, the European Parliament, the European Economic and Social Committee - "The European Environment & Health Action Plan 2004-2010" {SEC(2004) 729}

[17] RICHARDSON D.H.S. *Pollution monitoring with lichens*. Richmond, Slough, UK, 1992

[18] NIMIS P.L., SCHEIDEGGER C., WOLSELEY P.A. *Monitoring with Lichens – Monitoring Lichens. NATO Science Series, IV*. Kluwer, Dordrecht, Vol. 7, 2002, 408 p.

[19] WOLSELEY P.A., JAMES P.W., THEOBALD M.R., SUTTON M.A. Detecting changes in epiphytic lichen communities at sites affected by atmospheric ammonia from agricultural sources. *Lichenologist*. 2006, **38** pp. 161–176

[20] DAVIES L., BATES J.W., BELL J.N.B., JAMES P.W., PURVIS O.W. Diversity and sensitivity of epiphytes to oxides of nitrogen in London. *Environ. Pollut.* 2007, **146** pp. 299–310

[21] INSAROV G.E., SCHROETER B. Lichen monitoring and climate change. In: *Monitoring with lichens – Monitoring lichens. NATO Science Series, IV*, (NIMIS P.L., SCHEIDEGGER C., WOLSELEY P., eds.). Kluwer, Dordrecht, Vol. 7, 2002, pp. 183–201.

[22] VAN HERK C.M., APTROOT A., VAN DOBBEN H.F. Long-term monitoring in the Netherlands suggests that lichens respond to global warming. *Lichenologist*. 2002, **34** (2) pp. 141–154

[23] MCCUNE B. Lichen communities as indicators of forest health. *Bryologist*. 2000, **103** pp. 353–356

[24] ROSE F., COPPINS S. Site assessment of epiphytic habitats using lichen indices. In: *Monitoring with lichens – Monitoring lichens. NATO Science Series, IV*, (NIMIS P.L., SCHEIDEGGER C., WOLSELEY P.A., eds.). Kluwer, Dordrecht, Vol. 7, 2002, pp. 343–8.

[25] VAN DOBBEN H.F., WOLTERBEEK H.T., WAMELING G.W.W., TER BRAAK J.F. Relationship between epiphytic lichens, trace elements and gaseous atmospheric pollutants. *Environ. Pollut.* 2001, **112** (2) pp. 163–169

[26] SEAWARD M.R.D., LETROUIT-GALINOU M.A. Lichen recolonization of the trees in the Jardin du Luxembourg, Paris. *Lichenologist*. 1991, **23** pp. 181–186

[27] SPARRIUS L.B. Response of epiphytic lichen communities to decreasing ammonia air concentrations in a moderately polluted area of The Netherlands. *Environ. Pollut.* 2007, **146** pp. 375–379

[28] CISLAGHI C., NIMIS P.L. Lichens, air pollution and lung cancer. *Nature*. 1997, **387** pp. 463–464

[29] VDI 3799 *Part 1 (1995) (withdrawn in 2004-10). Measurement of immission effects – Measurement and evaluation of phytotoxic effects of ambient air pollutants (immissions) with lichens – Mapping of lichens for assessment of the air quality. Berlin: Beuth-Verlag*

[30] VDI 3957 *Part 13 (2005). Biological measurement procedures for determining and evaluating the effects of ambient air pollutants by means of lichens (bioindication) – Mapping the diversity of epiphytic lichens as an indicator of air quality. Berlin: Beuth-Verlag*

[31] AFNOR (2008). Biosurveillance de l'environnement – Détermination d'un indice biologique de lichens épiphytes (IBLE). NF X43-903

[32] NIMIS P.L. (1999). Linee guida per la bioindicazione degli effetti dell'inquinamento tramite la biodiversitàdei licheni epifiti. In: Piccini C., Salvati S. (Eds.): Atti Workshop Biomonitoraggio Qualità dell'aria sul territorio Nazionale. ANPA, Ser. Atti, 2, 267-277

[33] ANPA. (2001). I.B.L. Indice di Biodiversità Lichenica. ANPA, Serie Manuali e Linee Guida 2/2001, Rome

[34] ASTA J., ERHARDT W., FERRETTI M., FORNASIER F., KIRSCHBAUM U., NIMIS P.L. et al. Mapping lichen diversity as an indicator of environmental quality. In: *Monitoring with lichens – Monitoring lichens. NATO Science Series, IV*, (NIMIS P.L., SCHEIDEGGER C., WOLSELEY P., eds.). Kluwer, Dordrecht, Vol. 7, 2002, pp. 273–9.

[35] HAWKSWORTH D.L., ROSE F. Qualitative scale for estimating sulphur dioxide air pollution in England and Wales using epiphytic lichens. *Nature*. 1970, **227** pp. 145–148

[36] WIRTH V. Ökologische Zeigerwerte von Flechten – erweiterte und aktualisierte Fassung. *Herzogia*. 2010, **23** pp. 229–248

[37] NIMIS P.L., MARTELLOS S. Testing the predictivity of ecological indicator values. A comparison of real and "virtual" relevés of lichen vegetation. *Plant Ecol.* 2001, **157** pp. 165–172

[38] NIMIS P.L., MARTELLOS S. (2002). ITALIC - the information system on Italian lichens. In: X. Llimona, H.T. Lumbsch, S. Ott (Eds.): Progress and problems in lichenology at the Turn of the Millennium. Bibliotheca lichenologica 82, 271-283

[39] GIORDANI P., BRUNIALTI G., ALLETEO D. Effects of atmospheric pollution on Lichen Biodiversity (LB) in a Mediterranean region (Liguria, NW-Italy). *Environ. Pollut.* 2002, **118** pp. 53–64

[40] GIORDANI P. Variables influencing the distribution of epiphytic lichens in heterogeneous areas: A case study for Liguria, NW Italy. *J. Veg. Sci.* 2006, **17** pp. 195–206

[41] MCCUNE B., MEFFORD M.J. *Nonparametric Multiplicative Habitat Modelling*. MjM Software Design, Gleneden Beach, Oregon, 2004

[42] GIORDANI P. Is the diversity of epiphytic lichens a reliable indicator of air pollution? A case study from Italy. *Environ. Pollut.* 2007, **146** pp. 317–323

[43] PURVIS O.W., COPPINS B.J., HAWKSWORTH D.L., JAMES P.W., MOORE D.M. *The lichen flora of Great Britain and Ireland*. Natural History Museum Publications, London, 1992

[44] CULBERSON C.F. Improved conditions and new data for the identification of lichen products by a standardized thin-layer chromatographic method. *J. Chromatogr. A*. 1972, **72** pp. 113–125

[45] CULBERSON C.F., CULBERSON W.L., JOHNSON A. A standardized TLC analysis of β-orcinol depsidones. *Bryologist*. 1981, **84** pp. 16–29

[46] ELZINGA C.L., SALZER D.W., WILLOUGHBY J.W., GIBBS J.P. *Monitoring plant and animal populations*. Blackwell Science, Inc, 2001, 360 p.

[47] FERRETTI M., ERHARDT W. Key issues in designing biomonitoring programmes. Monitoring scenarios, sampling strategies and quality assurance. In: *Monitoring with lichens – Monitoring lichens. NATO Science Series, IV*, (NIMIS P.L., SCHEIDEGGER C., WOLSELEY P.A., eds.). Kluwer, Dordrecht, Vol. 7, 2002, pp. 111–39.

[48] CLINE S.P., BURKMAN W.G. The role of quality assurance in ecological programs. In: *Air pollution and forest decline. IUFRO*, (BUCHER J.B., BUCHER-WALLIN J., eds.). Birmensdorf, 1989

[49] SHAMPINE W.J. Quality assurance and quality control in monitoring programs. *Environ. Monit. Assess.* 1993, **26** p. 143

[50] EPA (2002). Guidance for Quality Assurance Project Plans. EPA QA/G-5

[51] MCCUNE B., DEY J.P., PECK J.E., CASSELL D., HEIMAN K., WILL-WOLF S. et al. Repeatability of community data: species richness versus gradient scores in large-scale lichen studies. *Bryologist*. 1997, **100** pp. 40–46

[52] BRUNIALTI G., GIORDANI P., ISOCRONO D., LOPPI S. Evaluation of data quality in lichen biomonitoring studies: the Italian experience. *Environ. Monit. Assess.* 2002, **75** pp. 271–280

[53] BRUNIALTI G., GIORDANI P., FERRETTI M. Discriminating between the good and the bad: quality assurance is central in biomonitoring studies. In: *Environmental Monitoring*, (WIERSMA B., ed.). CRC Press LLC, 2004, pp. 443–64.

[54] GIORDANI P., BRUNIALTI G., BENESPERI R., RIZZI G., FRATI L., MODENESI P. Rapid Biodiversity Assessment in lichen biomonitoring surveys: implications for Quality Assurance. *J. Environ. Monit.* 2009, **11** pp. 730–735

Identification keys

[55] CLAUZADE G., ROUX C. *Likenoj de Okcidenta Europo*. Illustrita Determinlibro. Bulletin de la Societé Bot. du Centre-Ouest. Royan, 1985

[56] DIEDERICH P. (1989). *Les lichens epiphytiques et leurs champignons lichénicoles (Macrolichenes exceptes: lichens crustaces) du Luxembourg*. Ministre des Affaires Culturelles. Travaux scientifiques du Musée National D' Histoire Naturelle de Luxembourg XIV. Luxembourg

[57] DOBSON F.S. (2011). *Lichens: An illustrated guide to the British and Irish Species*. Sixth Edition, Richmond Publishing, Slough, 496 p.

[58] FOUCARD T. *Svensk Skorplavs Flora (crustose lichens)*. BTJ-Druck, Lund, 1990

[59] FRAHM J.-P., SCHUMM F., STAPPER N.J. *Epiphytische Flechten als Umweltgütezeiger – Eine Bestimmungshilfe. bod Verlag*. Norderstedt, 2010

[60] JAHNS H.M. *Farne, Moose, Flechten*. BLV-Verlag, München, 1987

[61] KIRSCHBAUM U., WIRTH V. *Flechten erkennen – Umwelt bewerten*. Hessisches Landesamt für Umwelt und Geologie, Wiesbaden, 2010

[62] KIRSCHBAUM U., WIRTH V. *Les lichens bio-indicateurs – Les reconnaître – Evaluer la qualité de l'air*. (VAN HALUWYN C., GAVÉRIAUX J.P., CUNY D., LEROND M., trans.). Ulmer-Verlag, Stuttgart, 1997

[63] KIRSCHBAUM U., WIRTH V. (2013). *Flechten einfach bestimmen: Ein zuverlässiger Führer zu den häufigsten Arten Mitteleuropas*. Quelle & Meyer, Wiebelsheim, 416 pp.

[64] KROG H., ØSTHAGEN H., TØNSBERG T. *Lavflora*. Universitätsforlaget, Oslo, 1980

[65] LORENTE V.C., SÁNCHEZ M.J.S. *Guia de liquenes epifitos*. Ministerio de Medio Ambiente. Secretaria General de Medio Ambiente, Madrid, 2001

[66] MCCUNE B. Lichen communities as indicators of forest health. *Bryologist*. 2000, **103** pp. 353–356

[67] MOBERG R., HOLMASEN I. *Flechten von Nord- und Mitteleuropa. G. Fischer-Verlag*. NewYork, Stuttgart, Jena, 1992

[68] NIMIS P.L. I Macrolicheni d`Italia. Chiavi analitiche per la determinazione. *Gortania*. 1987, **8** pp. 101–220

[69] OZENDA P., CLAUZADE G. *Les Lichens*. Masson, Paris, 1970

[70] POELT J. *Bestimmungsschlüssel europäischer Flechten. Cramer-Verlag*. Lehre, Vaduz, 1969

[71] POELT J., VEZDA A. *Bestimmungsschlüssel europäischer Flechten*. Ergänzungsheft I. *Bibl. Lichenol.* 1977, **9** pp. 1–258

[72] POELT J., VEZDA A. *Bestimmungsschlüssel europäischer Flechten*. Ergänzungsheft II. *Bibl. Lichenol.* 1981, **16** pp. 1–390

[73] PURVIS O.W., COPPINS B.J., HAWKSWORTH D.L., JAMES P.W., MOORE D.M. *The lichen flora of Great Britain and Ireland*. Natural History Museum Publications, London, 1992

[74] SCHREINER E., HAFELLNER J. *Sorediöse, corticole Krustenflechten im Ostalpenraum*. I. *Bibl. Lichenol.* 1992, **45** pp. 1–291

[75] SMITH C.M., APTROOT A., COPPINS B.J., FLETCHER A., GILBERT O.L., JAMES P.W. et al. *The Lichens of Great Britain and Ireland*. British Lichen Society, 2009

[76] TØNSBERG T. The sorediate and isidiate, corticolous, crustose lichens in Norway. *Sommerfeltia*. 1992, **14** pp. 1–331

[77] VAN HALUWYN C. Essai de clé de détermination des lichens épiphytes crustacés stériles du nord ouest de la France. *Bull. Ass. Fr. Lichenol.* 1988, **13** (1) pp. 5–14

[78] VAN HALUWYN C., LEROND M. *Guide des lichens*. Lechevalier, Paris, 1993

[79] WIRTH V., HAUCK M., SCHULTZ M. (2013). *Die Flechten Deutschlands. Band 1 und 2.* Ulmer-Verlag, Stuttgart

[80] WIRTH V., DÜLL R. *Farbatlas Flechten und Moose*. Ulmer-Verlag, Stuttgart, 2000

Online lichen checklists

[81] Online Atlas of Dutch Lichens – BLWG Verspreidingsatlas Korstmossen online: URL: http://www.verspreidingsatlas.nl/korstmossen. Distribution maps and photographs of lichens and lichenicolous fungi. Species list based on: APTROOT A., VAN HERK C.M., SPARRIUS L.B., SPIER J.L. Checklist van de Nederlandse Korstmossen en korstmosparasieten. *Buxbaumiella*. 2004, **69** pp. 17–55

[82] DIEDERICH P., ERTZ D., STAPPER N., SÉRUSIAUX E., VAN DEN BROECK D., VAN DEN BOOM P. et al. (2010). The lichens and lichenicolous fungi of Belgium, Luxembourg and northern France. URL: http://www.lichenology.info [24.05.2010]. Online checklist with distribution maps and images of lichens and lichenicolous fungi

[83] NIMIS P.L., MARTELLOS S. (2008). ITALIC – The Information System on Italian Lichens. Version 4.0. University of Trieste, Dept. of Biology, IN4.0/1. URL: http://dbiodbs.univ.trieste.it/

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