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ISTITUTO AGRARIO
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DIPARTIMENTO
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Towards an international standard for lichen monitoring – theory and practice



Presentations given at the harmonization field course

*7 - 11 June 2010
Trentino - Northern Italy*

Supported by:



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IASMA Research and Innovation Centre - Fondazione E. Mach / Società Lichenologica Italiana / Dipartimento di Scienze della vita - Università degli Studi di Trieste / TerraData environmetrics

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Edited by:

Fabiana Cristofolini, Giorgio Brunialti, Paolo Giordani, Marco Ferretti

Supported by:

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**Towards an international standard for lichen monitoring –
theory and practice**

Editor

Fondazione Edmund Mach - Istituto Agrario di San Michele all'Adige
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Picture on the cover

Villa Welsperg e Pale di S.Martino- Val Canali-Trentino

By Carlo Albino Turra

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INDEX

<i>Introductions</i>	5
<i>Course programme</i>	7
CEN/TC 264/WG 31 "Biomonitoring methods with ... lichens": state of the art <i>Paolo Giordani</i>	9
Towards an international standards for lichen monitoring: results from the first Italian intercalibration test (2009) <i>Giorgio Brunialti</i>	25
Sampling design in theory and in practice: does and don'ts <i>Marco Ferretti</i>	39
GIS overview – concepts and definitions <i>Francesco Geri</i>	63
Old growth structures and lichen conservation in subalpine coniferous forests: a lesson from the Paneveggio Forest (NE Italy) <i>Juri Nascimbene</i>	69
Towards an international standards for lichen monitoring: results 2010 <i>Giorgio Brunialti, Paolo Giordani, Fabiana Cristofolini, Elena Gottardini, Marco Ferretti</i>	81
Final remarks <i>Paolo Giordani</i>	93
<i>Participants</i>	95

INTRODUCTIONS

Biomonitoring provides information on biological effects caused by anthropogenic disturbances on sensitive organisms. Lichens are very sensitive components of terrestrial ecosystems and among the most used organisms in environmental assessment. They are widely used as both bioaccumulators of trace elements (due to their resistance to heavy metals) and as bioindicators (due to their sensitivity to several anthropogenic disturbances). Despite this wide range of application, an international harmonized monitoring method was missing, and only recently, an attempt for an internationally harmonized biomonitoring method with lichens has been developed at European (CEN) level. However, questions related to its theoretical and practical application are yet to be clarified, with the only exception of a national initiative carried out in Tuscany (central Italy) in 2009 (see Giorgio Brunialti, this volume). To achieve this goal, from June, 7 to June, 11, 2010, we organised an international initiative entitled:

Towards an International standard for lichen monitoring – theory and practice A harmonization field course /

The course has been organised in Trentino (Northern Italy) by the Fondazione E. Mach (S. Michele all'Adige, Trento), in close co-operation with the Italian Lichenological Society, Terradata environmetrics (spin-off of the University of Siena) and the Department of Life Science, University of Trieste

The meeting was organized as a course - to provide basic concepts for application - and as an exercise - to compare experimental design options and field performances of different crews. During the theoretical session, the results of a previous Italian National training course (carried out in Tuscany in 2009) were presented and participants were introduced to the basic principles of sampling design and GIS software. The fieldwork (supported by the Natural Park Paneveggio-Pale di San Martino and carried out in Dolomitic Valley Val Canali) included ring-tests aimed to assess the performance of teams from different Countries in the application of the norm. In particular, a control team (made-up by three fully experienced experts familiar with the norm) certified the accuracy and the precision of the teams in localising the plot, assessing standard trees within the plot, positioning the sampling grid on the tree and sampling lichen diversity, before and after an intensive training on the basic steps of the CEN methodology.

Experts from several European Countries, including Spain, Portugal, UK, Slovenia, Estonia and Italy attended the course and contributed to draft the main conclusions during a final workshop, where provisional results and critical points were discussed. In particular, a working group ensuring continuous training and harmonization was felt necessary in order to support the development, application and maintenance of quality assured methodology in the field of lichen biomonitoring.

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Programme

Monday 7 June 2010

- 13.30 – 14.00 Registration
14.00 – 17.30 SESSION 1. OPENING OF THE MEETING AND INTRODUCTORY STATEMENTS
14.00 – 14.30 Opening Addresses and programme – *Fabiana Cristofolini*
14.30 – 16.00 CEN/TC 264/WG 31 "Biomonitoring methods with ... lichens": state of the art - *Paolo Giordani*
16.00 – 17.30 Results from the first Italian intercalibration test (2009) – *Giorgio Brunialti*

Tuesday 8 June 2010

- 9.00-12.30 SESSION 2. SAMPLING DESIGN
9.00 – 10.30 Sampling design in theory and in practice: does and don'ts – *Marco Ferretti*
10.30-11.00 *Coffee break*
11.00 – 12.30 Sampling design in theory and in practice: the use of GIS for implementing sampling design - *Francesco Geri*
12.30 – 14.00 *Lunch break*
14.00- 17.00 SESSION 3. INDOOR EXERCISE
Participants will carry out a computer exercise on sampling design- *Francesco Geri*
17.00 – 19.00 *Transfer from S.Michele to Exercise site Val Canali - Parco Paneveggio- Pale S. Martino*

Wednesday 9 June 2010

- 8.30 - 9.30 Lichen diversity in the study area – *Juri Nascimbene*
9.30 – 18.30 SESSION 4. FIELD EXERCISES
9.30 – 11.00 Lichen diversity sampling
11.00- 11.30 *Coffee break*
11.30 – 12.30 Lichen diversity sampling –
12.30 – 13.30 *Lunch break*
13.30- 15.00 Training
15.00 – 18.30 Lichen diversity sampling –

Thursday 10 June 2010

- 8.30-15.00 SESSION 5. FIELD EXERCISES
8.30 – 10.00 Plot location and tree selection
10.30 -12.30 Lichen diversity sampling
12.30 – 13.30 *Lunch break*
13.30 -15.00 Lichen diversity sampling
15.00 -17.00 *Transfer from exercise site -Val Canali- to Trento*

Friday 11 June 2010

- 9.00-12.00 FINAL WORKSHOP
9.00 -9.45 Presentation of results - *Giorgio Brunialti*
9.45 – 10.30 Discussion on critical issues emerged during the exercises - *Paolo Giordani*
10.30 – 11.00 *Coffee break*
11.00 -11.30 Suggestions for the final draft of the international standard- *Paolo Giordani*
11.30 – 12.00 Final remarks and conclusion of the meeting- *Fabiana Cristofolini*



CEN/TC 264/WG 31
"Biomonitoring methods with ... lichens": state of the art

Paolo Giordani
Italian Lichenological Society
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Società Lichenologica Italiana
S.I.L.



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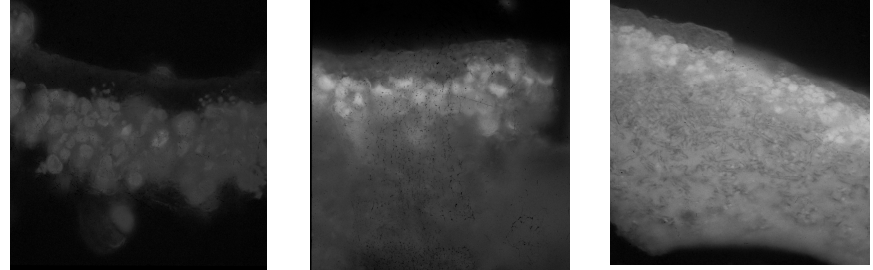
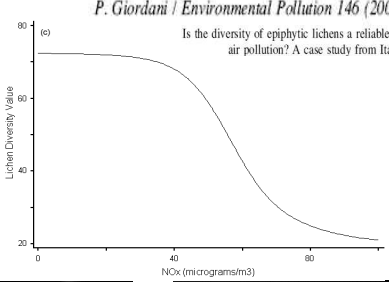
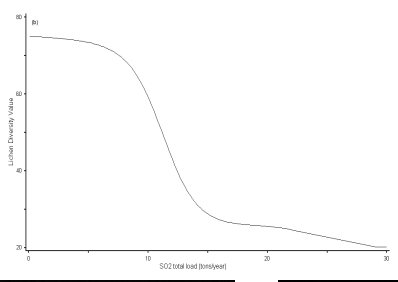


TerraData
G.P. VITTORELLI & C.

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Biological basis of lichen biomonitoring

P. Giordani / Environmental Pollution 146 (2007) 317–323
Is the diversity of epiphytic lichens a reliable indicator of air pollution? A case study from Italy



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The way to the Norm...

- Scientific literature since 1964
- German Norm VDI 1995 (emend. 2005)
- Italian ANPA guidelines 1999-2001
- “European” protocol Asta et al. 2002
- **French norm AFNOR 2006**



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A technical norm

- Wide consensus within all the possible stakeholders
- Approved by a recognized organism at national and/or international level (UNI, CEN, ISO)
- Best practice, best available state of the art (meeting point between scientific knowledge and applicability)
- Public process

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The aims of a norm

- To guarantee the quality of a measuring (monitoring) process
- To provide information about the sampling and non-sampling errors
- To provide the repeatability of the measurements
- To limit the subjectivity
- To allow the control process of each step of the measurements

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The CEN steps towards the final norm

ENQ + FV / PQ-UQ + FV	
Maximum timeframe expressed in months	Milestones/Description of events
0	Receipt of TC resolution (e-mail date)
+ 6 months	Step 1 Circulation of first working draft
+ 6 months (see note 1 for tolerances)	Step 2 Dispatch of Enquiry draft to CMC
+ 2,5 months	Step 3 Submission to Enquiry (the elapsed time of 2,5 months between step 2 and step 3 include the timeframe for the CMC processing and the translation procedure)
+ 5 months	Step 4 Closure of Enquiry
+ 8 months (see note 1 for tolerances)	Step 5 Dispatch of Formal Vote draft to CMC
+ 3,5 months	Step 6 Submission to Formal Vote (the elapsed time of 3,5 months between step 5 and step 6 include the timeframe for the CMC processing and the translation procedure)
+ 2 months	Step 7 Closure of Formal Vote
+ 1 month	Step 8 Ratification
+ 2 months	Step 9 Definitive text available (=DAV) (the elapsed time of 3 months between step 7 and step 9 include the timeframe for the CMC processing and the translation procedure)
36 months	

Source: CEN

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A norm for lichen monitoring



CEN/TC264/WG31 meetings

Biomonitoring methods with mosses and lichens CEN/TC 264/WG 31

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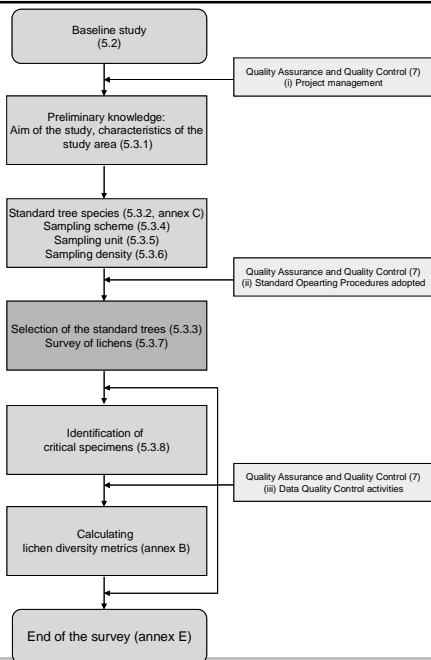
Paris (january 2008)
 Bad Durkheim (june 2008)
 Trieste (january 2009)
 Lille (june 2009)
 Bad Durkheim (january 2009)
 Trento (may 2010)

**Preliminary work item 00264107 -
 Lichens - Version May 2010**

Air Quality – Biomonitoring with lichens – Assessing epiphytic lichen diversity

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Flow chart



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The draft: scope

- to provide a **reliable, repeatable** and **objective** method for assessing epiphytic lichen diversity.
- it provides the **framework for assessing the impact of anthropogenic impact**, particularly for estimating the **effects of atmospheric pollution**.

Terms and definitions

Target population

in this document it is defined as: '**Lichen communities living on the bark of standard trees at an above ground level height ranging between 1 and 1.5 m. Standard trees should be defined in terms of species, bole circumference and inclination**) should be located within the study area'.

Principles of the norm

- The procedure **is widely applicable** and it is addressed to those subjects who are interested in collecting lichen diversity data.
- The **interpretation of the results has to 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.
- **Investigations performed according to this guideline require personnel or institutions having the necessary expertise, in the fields of lichenology and probabilistic sampling designs.**
- Quality assurance standards have to be achieved.

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Equipment

Material needed for each step of the work is described, **when necessary with high level of details**, otherwise general recommendations are provided:

- Field work preparation equipment
- Field equipment
- Laboratory determination equipment

Stereo microscope (**minimum range $\times 10$ to $\times 60$**). Used for lower-power magnification on lichen samples.

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

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Sampling

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 must be always maintained.**

Sampling objective

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

E.g.: "Obtain an estimate of the mean LDV for the study domain with a confidence interval $\pm 10\%$ of the mean value, at a probability (P) level of 95%"

Baseline studies vs. monitoring s. str.

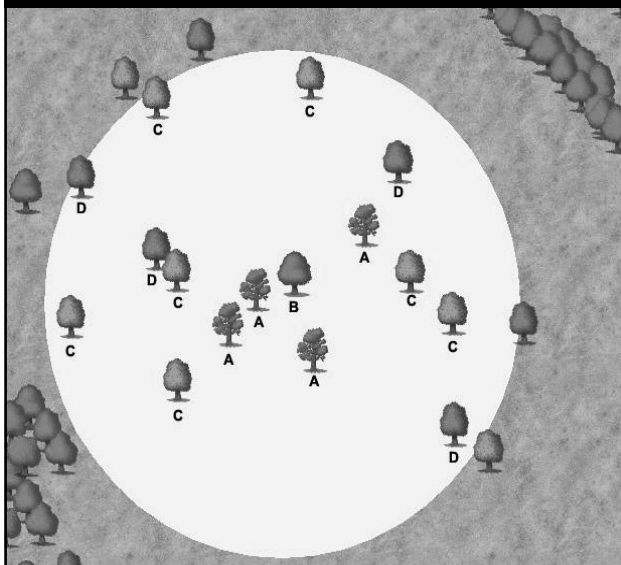
- This Norm includes guidelines focused on **baseline studies.**
- When evaluation of change between subsequent measurement is of interests in addition to the following guidelines, **it is important to consider the implications related to the statistical analysis for detecting changes**

Prior to sample

The sampling scheme should be decided on the basis of the characteristics of the study area and the aim of the study:

1. **the distribution of potentially suitable trees over the study area** should be as much known as possible, e.g. by means of a preliminary inspection throughout the study area.
2. In case of gradient studies (e.g. prevailing punctual source of pollution, or wind direction and intensity,...), **quantitative information concerning the gradient should be considered.**
3. **Any kind of restricted access**, such as private estates, military areas, etc. **should be preliminarily checked**, in order to include or exclude those areas from the study domain and to define the target population accordingly.

Standard tree species selection



When choosing tree species (in preference order):

- i. A single tree species within the whole study area (e.g. A).
- ii. Different tree species, within the same bark-type group (e.g. A and B).
- iii. Different tree species within different bark-type groups (e.g. A, B and C), excluding unsuitable taxa, e.g. some Conifers, *Platanus* - D).

Suitable tree species

<i>Group I</i>	<i>Group II</i>	<i>Group III</i>	<i>Group IV</i>	<i>Group V</i>	<i>To be tested</i>	<i>Excluded</i>
<i>Acer</i> spp.	<i>Olea</i> spp.	<i>Abies alba</i>	<i>Alnus glutinosa</i>	<i>Fagus</i> spp.	<i>Robinia pseudoacacia</i>	<i>Araucaria</i> spp.
<i>Ceratonia siliqua</i>	<i>Prunus</i> spp.	<i>Larix decidua</i>	<i>Betula pendula</i>	<i>Carpinus</i> spp.	<i>Ailanthus altissima</i>	<i>Platanus</i> sp.
<i>Fraxinus</i> spp.	<i>Quercus</i> spp.	Microthermic <i>Pinus</i> spp.			<i>Celtis</i> spp.	<i>Taxus baccata</i>
<i>Juglans</i> spp.	<i>Castanea sativa</i>	<i>Picea abies</i>			<i>Salix</i> spp.	<i>Cycas</i> spp.
	<i>Pirus communis</i> *				<i>Ostrya carpinifolia</i>	"Palms"
	<i>Tilia</i> spp.*				<i>Cupressus sempervirens</i>	Mediterranean <i>Pinus</i> spp.
<i>Ulmus</i> spp.	<i>Malus</i> spp.				<i>Alnus cordata</i>	
<i>Populus</i> spp.	<i>Ostrya carpinifolia</i>				<i>Ginkgo biloba</i>	
<i>Ficus</i> spp.	<i>Sorbus</i> spp.				<i>Magnolia</i> spp.	
					<i>Citrus</i> spp.	
					<i>Crataegus</i> spp.	
					<i>Pseudotsuga menziesii</i>	
					"other exotic cultivated plants"	
					Any other species not explicitly reported in this table	

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Standard tree individual parameters

A standard tree individual is defined as follows (some news!):

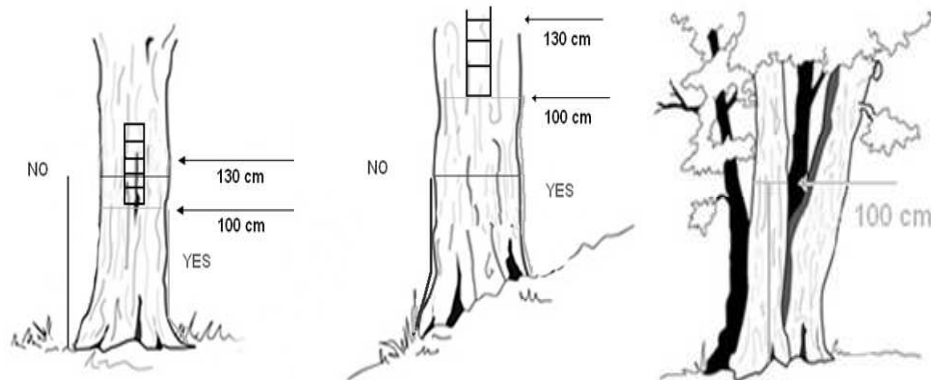
- i. It belongs to a **suitable species**.
- ii. It has a **trunk circumference** (at 130 cm from the ground level) **>50 cm and < 250 cm**.
- iii. Each exposition (N, E, S, W) has an **inclination** (at the same height) **< 20°**.
- iv. The coverage of the **trunk without disturbances** (damages, decortication, branches, knots and/or other epiphytes) **within each of the 4 grids is complexively < 20%**.

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Measurements of height from the ground level

for positioning the lower edge of the grid (100 cm) and for measuring the circumference and the inclination of the trunk (130 cm):

- correct (green, right) and incorrect (red, left) measures **along the trunk**;
- measurements **for tree laying on a slope**;
- definition of individual trees**, in the case of furcate stems: the two trunks have to be considered separate trees, being divided below 100 cm.



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Sampling scheme: heterogeneity vs. homogeneity

Different **designs** are suggested on the basis of **ecological homogeneity or heterogeneity of the study domain**.

In each study ecological **variables considered to assess homogeneity should be explicitly provided** (e.g. altitude, land use, resident population density, vegetation, etc.),

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Sampling unit

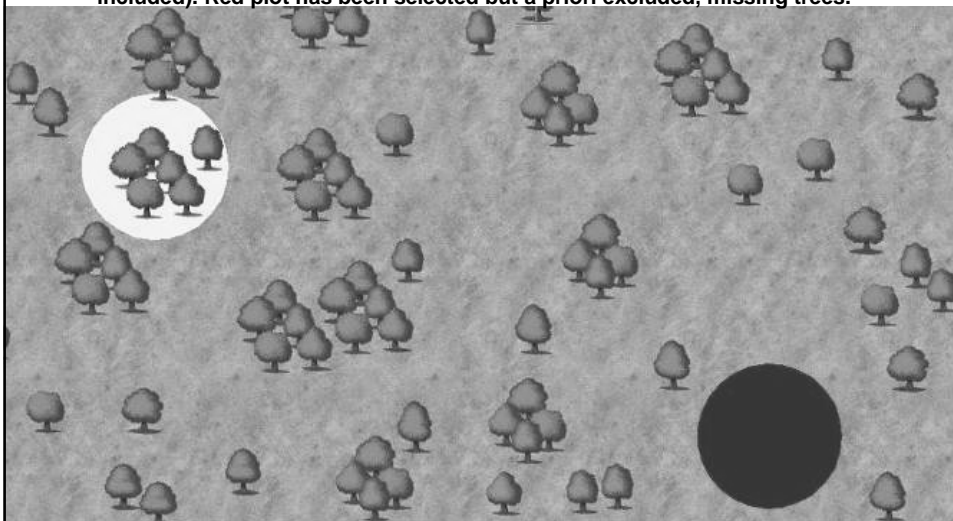
- **Plot sampling:** each plot represents a sampling unit.
 - A circular sampling plot with a radius of at least 30 m is recommended.
 - All the standard trees in the plot are measured.
 - Different dimensions could be considered, depending on the tree density in the study domain.
- **Tree-based sampling:** each tree or cluster of trees represent a sampling unit.

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Homogeneous area

Standard trees abundant and homogeneously distributed over the study domain

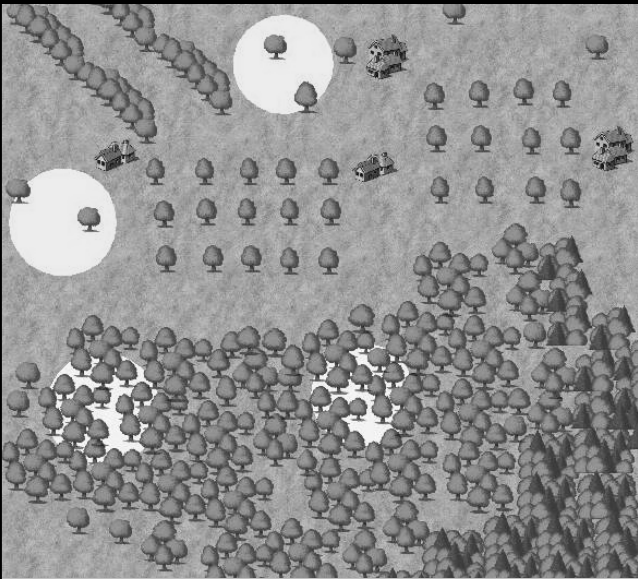
Plot sampling: light blue plot has been randomly selected and installed (7 trees are included). Red plot has been selected but a priori excluded, missing trees.



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Heterogeneous area

Standard trees abundant and homogeneously distributed over the study domain



The diagram shows a landscape with a mix of agricultural fields and forested areas. Two yellow circles highlight agricultural areas, and two light blue circles highlight forested areas. Small icons of tractors and trees are scattered across the landscape. The text 'Stratified random sampling' is written to the right of the diagram.

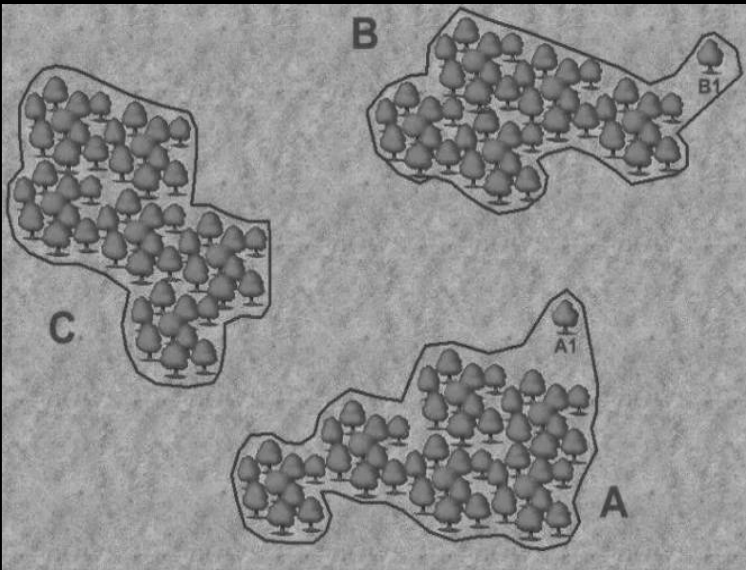
Stratified random sampling

with two strata:
agricultural area (yellow circles);
forested area (light blue circles).

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Homogeneous or heterogeneous area

Standard trees abundant, scattered over the study domain



The diagram shows a landscape with three distinct clusters of trees, labeled A, B, and C. Each cluster is enclosed in a black outline. Within cluster A, a single tree is labeled 'A1'. Within cluster B, a single tree is labeled 'B1'. The text 'Cluster sampling.' is written to the right of the diagram. Below the diagram, the text explains that trees A1 and B1 are included into clusters A and B, respectively, based on a threshold distance from the centroid of the cluster.

Cluster sampling.

Trees A1 and B1 are included into cluster A and B, respectively, following the criterion of the threshold distance from the centroid of the cluster

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Homogeneous or heterogeneous area

Standard trees infrequent, scattered over the study domain.

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

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Sampling density

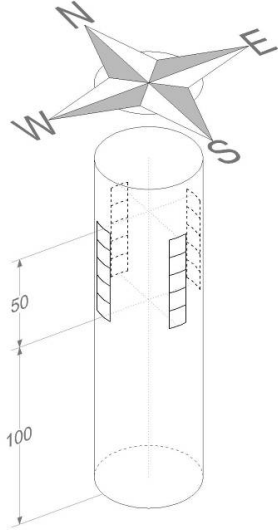
N vs C.L. by Tol. Prob. with Dist. to Limit=5,235 S=22,450
 C.I. One Mean

C.L.	N (Tol. Prob. 0.70)	N (Tol. Prob. 0.75)	N (Tol. Prob. 0.80)	N (Tol. Prob. 0.85)	N (Tol. Prob. 0.90)	N (Tol. Prob. 0.95)
0.7	25	28	32	35	38	42
0.8	35	40	45	50	55	60
0.9	55	65	75	85	95	105
1.0	85	100	115	130	145	160

The minimum number of plots to be selected should be calculated on the basis of available sample size equations for different designs, using previous data on the same (or comparable) study domain.

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Surveying lichens



- A flexible grid of five 10 × 10 cm squares each is attached along the trunk so that the lower edge of each grid is 1 m above the highest point of the ground.
- The grid must be placed four times to correspond the centre with the 4 aspects (NSEW) of the tree.
- All lichen species present within each grid are recorded and the frequency of occurrence of each species in the 5 squares of each quadrat segment noted.

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Shifting the grids

- It is possible to relocate each monitoring grid by a maximum shift of 20° (firstly in a clockwise, then in counterclockwise direction), to avoid parts of the trunk which are not suitable for sampling (e.g. wounds, knots, etc).
- a grid must be shifted if the overall sum of disturbances (namely damaged or decorticated parts; knots; cover of other epiphytes) is > 20%.

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The results

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

- matrix of species × exposition at each tree
- matrix of species × trees
- matrix of species × SUs

Calculating lichen diversity metrics

A set of possible ways to interpret the data are reported as an example. The following examples are not exhaustive and other interpretative tools may be applied to basic data. Among the others

Lichen Diversity Value (LDV)

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

where

SF: Sum of frequencies of all lichen species found at one aspect of tree t

N, E, S, W: north, east, south, west

n: number of trees sampled in unit j

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.

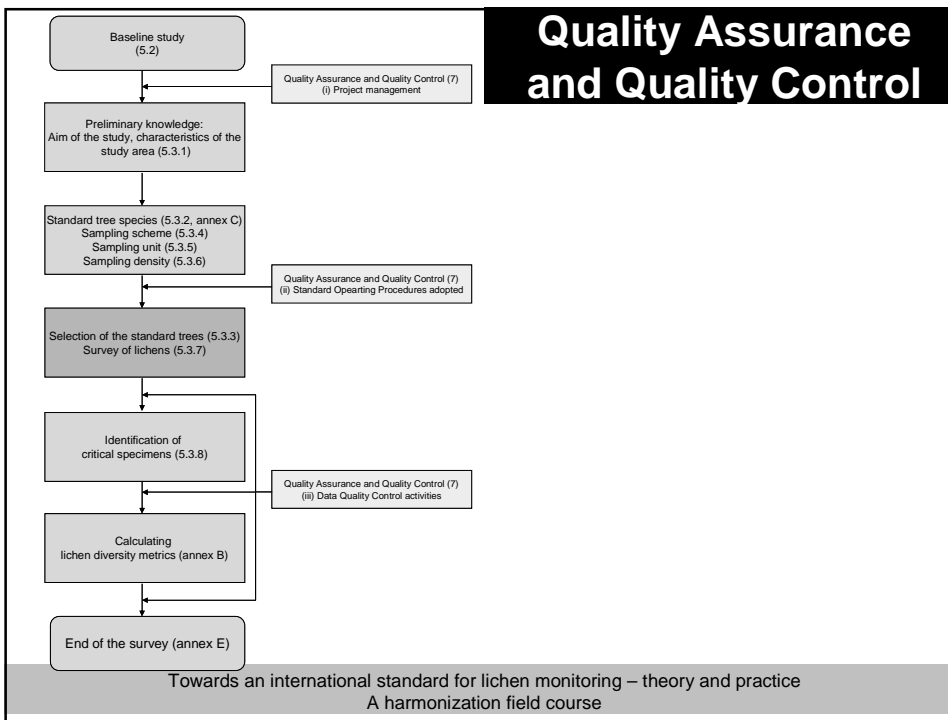
$$LDV_j = (SF_{1t} + SF_{2t} + SF_{3t} + SF_{4t} + \dots + SF_{nt})/n$$

Diversity value of the indicators of eutrophication (e.g., NIW sensu VDI, 2005).

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	LDVt 46
Sum of Frequencies of nitrophytes		10	10	5	5	NIWt 30

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Quality Assurance and Quality Control



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Giorgio Brunialti

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Background



- Lichen monitoring requires appropriate skill in different fields. Sampling design, sampling density and taxonomic knowledge have been reported as key problematic issues.
- Data comparability in space and time remains a central problem.
- A proposal for an European guideline is being prepared by the CEN.



Origin of the test



- Till now, only the performance of teams in species identification has been assessed.
- Very few reports exist dealing with the whole field procedure and none dealing with the entire survey design and implementation.
- To fill this gap, we decided to organize a comparative test.
- **Main objective:** to compare the results obtained by different well experienced operators facing the same problem, under the same field condition and provided with the same Standard Operating Procedures (SOPs).



Test concept



- The test was organized over a relatively small area
- to evaluate different sources of variability and error.
- Participants received a standard information package with digital maps, expected lichen flora and the set of SOPs.
- They were asked to perform **five exercises**, characterized by the progressive reduction of the operational freedom to the teams.
- In order to avoid a bias, participants were unaware of some exercises.



Participants

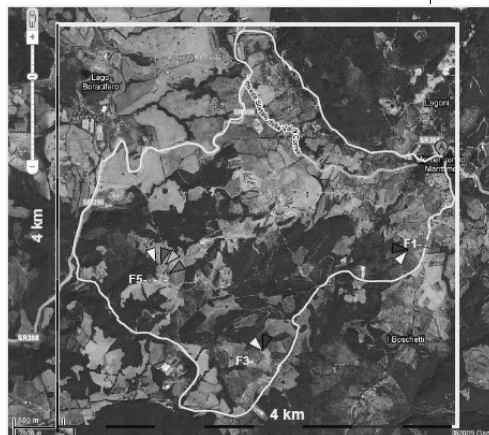
TerraData environmetrics	(Brunialti, Frati)
Fondazione E. Mach-IASMA	(Cristofolini, Cristofori, Gottardini)
Università di Siena	(Munzi, Paoli, Pisani)
Università di Genova	(Giordani)
Università di Firenze	(Benesperi)
ARPAT	(Critelli, Di Capua, Innocenti)
Università del Molise	(Genovesi, Ravera)

- Six teams
- All the participants were at their first application of the SOPs



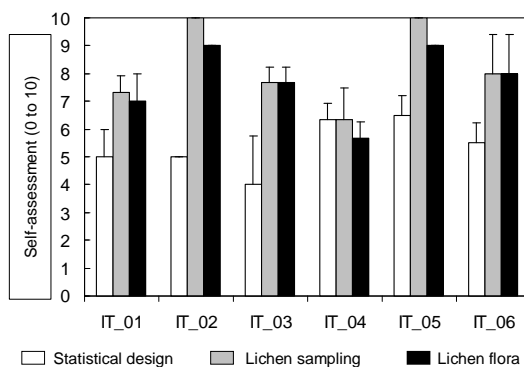
Study area

- Tuscany, Central Italy.
- 22 – 26 June 2009
- 7 km²
- Altitude: 46-764 m
- Deciduous woods:
Quercus cerris forests and
Castanea sativa plantations
- Cultivated areas with
scattered *Quercus pubescens* trees



Operator's experience

Each team member self-assessed on the following aspects:
 familiarity with statistical sampling and its implementation in the field, familiarity with lichenological sampling and with the lichen flora of the area.



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Result

Size of the sampling grid

The size of the grids of each team were measured before and after the test

Team code	Length (cm)	% variation	Width (cm)	% variation	Sampled area (cm ²)	% variation
IT_01	50.9±0.25	1.8	10.4±0.25	3.8	527.9	5.6
IT_02	50.3±0.10	0.7	9.3±0.29	-7.5	465.8	-6.9
IT_03	50.1±0.25	0.3	9.4±0.26	-6.5	468.7	-6.3
IT_04	50.0	0.0	10.00	0.0	500.0	0.0
IT_05	50.4±0.06	0.7	9.8±0.33	-2.5	490.9	-1.8
IT_06	50.4±0.25	0.8	9.1±0.23	-9.1	457.8	-8.4
Total, %		2%		13%		14%



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Conclusion

Size of the grid

- We need a standard grid
- The sampling protocol must specify the precision of this tool to control measurement error and to ensure data comparability



Exercise 1: in theory

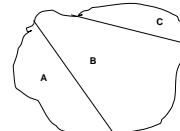
Identify the sampling scheme and sample size

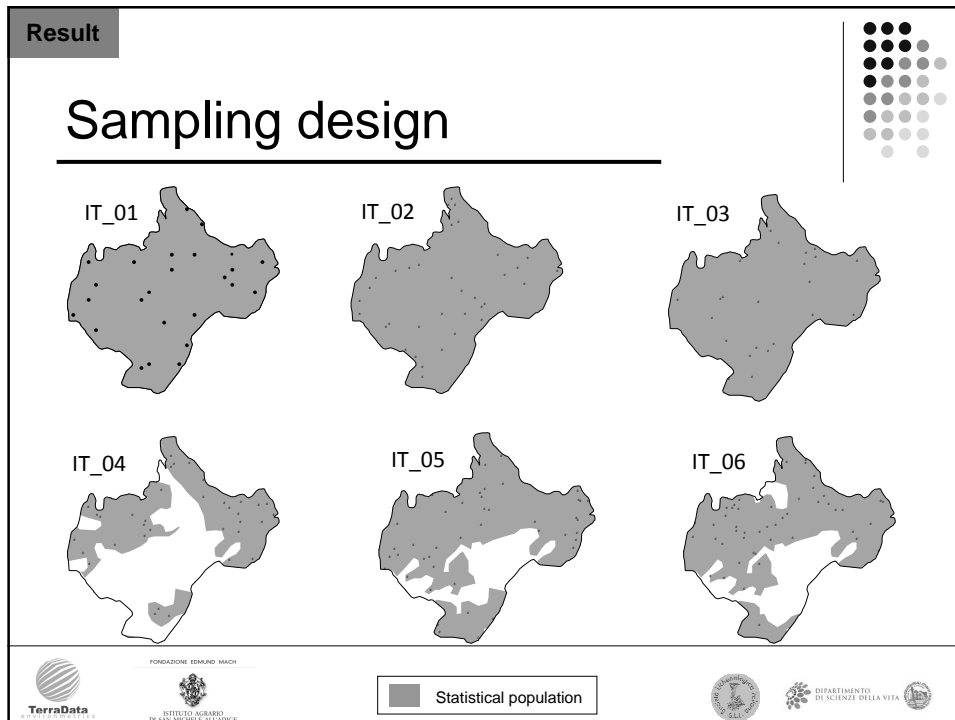
“Given the common study area, identify the statistical population and to select a sample on the basis of the SOPs”

Expected result: whether and how the participants had come to unequivocal conclusions on the population statistics on the subject of their survey and sample type.

Exercise 1 – Given study area, different landuse categories (A, B, C)

- Identify the population
- Select a sample





Result

Sampling design

Team	Declared precision	Actual precision	Declared Scheme	Decl. Strata	Actual scheme	Plots, n
IT_01	11% (p:95%)	15% (p:95%)	StRS	4	StRS	29
IT_02	10% (p:95%)	10% (p:95%)	StRS	2	Unclear	35
IT_03	10% (p:95%)	?	StRS	8	StRS	23
IT_04	10% (p:95%)	10% (p:95%)	StRS	2	StRS	35
IT_05	10% (p:95%)	10% (p:95%)	SRS	-	SRS	43
IT_06	10% (p:95%)	10% (p:95%)	StRS	2	StRS	48

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Conclusion

Sampling scheme and sample size



- The exercise met the goals
- Each team was able to select without difficulty a target population, reaching the stated accuracy
- Interestingly, there is a predominance of use of StRS, but with significant differences in the stratification that affects sampling density.

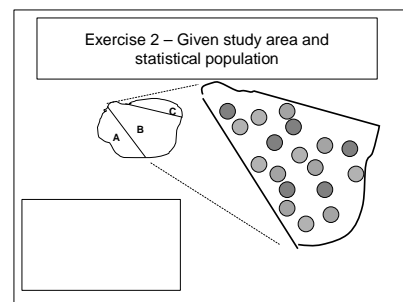


Exercise 2: in theory + field



“Given a defined and small statistical population, the team had to identify and to select a sample according to a simple random sampling (SRS)

with $n = 6$ circular plots (30 m radius) and to sample lichen diversity”.



Expected result: evaluate the sampling error. The data of 5 teams were then integrated to evaluate the effect of the sample size (a total of 30 plots).



Result

Species richness

Team	N trees	Tree species (n)	NSp
IT_01	8	Qc (2), Qi (4), Qp (2)	10.8±1.7
IT_02	13	Qc (4), Qp (9)	8.8±2.6
IT_03	15	Qc (3), Qi (2), Qp (10)	9.0±2.5
IT_04	16	Qc (2), Qi (3), Qp (11)	6.4±5.0
IT_05	10	Cs (3), Qc (3), Qp (4)	14.8±6.3

Qc: Q. cerris Qi: Q. ilex Qp: Q. pubescens Cs: Castanea sativa

Kruskal-Wallis test (df: 4, n=66) showed a significant difference among the five teams ($H = 14.9$; $p < 0.01$).



Conclusion

Sampling error

- The variability between teams is mainly related to environmental heterogeneity of the study area

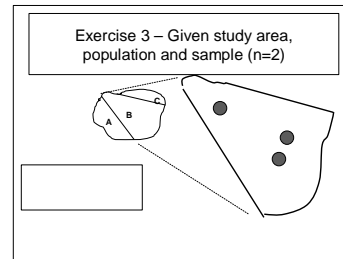
- It is therefore associated with sampling error



Exercise 3 (blind): common plots

Five teams sampled two common plots (blind test)

“Given defined population and sample, the team have to do the measurements according to SOPs”



Expected result: evaluate the measurement error (all inclusive: selection of the trees, placement of the grid, identification of the species).



Result

Common plots

Team code	N trees	N _{Sp}
IT_01	4	13.0±3.4
IT_02	9	13.2±2.6
IT_03	8	10.0±1.8
IT_04	4	10.5±1.3
IT_05	4	13.0±2.9

Friedman test (df: 4, n = 4) showed no significant differences among the 5 teams ($\chi^2 = 7.43, p > 0.05$).



Conclusion

Common plots

- This exercise confirmed that the marked differences found in exercise 2 are mainly due to environmental variability instead of measurement error.
- Indeed, when the teams sample the same plots, we find a considerable difference in the identification of suitable trees.
- Lack of harmonization among teams on the characteristics of standard trees.
- In this case, this source of variability doesn't affect species richness.



Result

Exercise 4 (blind): re-measuring

A control team re-measured 5 plots previously measured by the other teams

“given a plot sampled by a team, a control team sampled the plot independently”



Result

Independent control

The control team found no standard trees at four out of five plots.

Only one plot was re-measured, obtaining the same results.

Team code	Parameter	Value Team	Value control	Likely reason for disagreement
IT_01	Trees, n	1	0	Tree out of the plot
	SpN	9	-	
IT_02	Trees, n	3	0	no standard tree
	SpN	10	-	
IT_03	Trees, n	4	2	no standard tree
	SpN	9	9	
IT_04	Trees, n	2	0	no standard tree
	SpN	12	-	
IT_05	Trees, n	1	0	no standard tree
	SpN	10	-	



Conclusion

Independent control

- The results are mainly influenced by the disagreement in the definition of standard tree.
- This differences may be resolved by harmonization in the field.

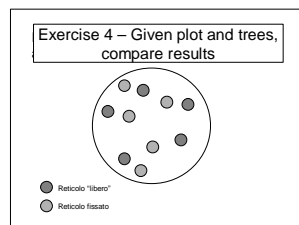


Exercise 5: Lichen identification

Lichen species identification in a common plot

In a given plot, each team sampled the same subsample of 10 trees out of the total.

- 5 trees: the position of the sampling grid was fixed *a priori*,
- 5 trees: each team placed independently the grid.



Expected result: to evaluate the measurement error of lichen diversity count and its dependence on the positioning of the grid.



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Result

Lichen species identification

p<0.001 (Friedman test: $\chi^2 = 16$; d.f.= 2; n= 10)	Tree code	Number of species			
		IT_01	IT_05	IT_06	CV%
Trees without a fixed position of the grid p<0.05 (Friedman test: $\chi^2 = 7.9$; d.f.= 2; n= 5)	1	15	21	16	18.5
	2	19	29	29	22.5
	3	9	10	14	24.1
	4	12	14	17	17.6
	5	23	25	24	4.2
Trees with the position of the sampling grid fixed a priori p<0.05 (Friedman test: $\chi^2 = 8.3$; d.f.= 2; n= 5)	6	14	25	24	29.0
	7	18	28	21	23.0
	8	13	18	19	19.3
	9	16	26	26	25.5
	10	17	25	23	19.2



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Conclusion

Lichen identification

- These results show that the final step of the monitoring program, lichen identification, greatly affects data comparability.
- These differences among teams may be resolved with training courses to improve knowledge on lichen species.



From Tuscany to Trentino...

from Italy to Europe

To answer the unanswered questions:

- Influence of training on the field.
- Harmonization on the field for the definition of standard trees.
- Identification of the plot: influence on the number of selected trees.



*Towards an international standard for lichen monitoring
- theory and practice*

San Michele all'Adige, 7-11 2010

Sampling design in theory and in practice: dos and don'ts

Marco Ferretti

TerraData environmetrics, Siena

ferretti@terradata.it

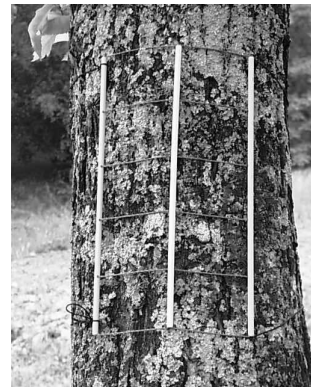


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Talk outline

- Getting started
- Origin and control of sampling errors
- Most common sampling schemes (incl. those in the CEN draft)
- Dos and don'ts



(Giordani e Brunialti, 2008)

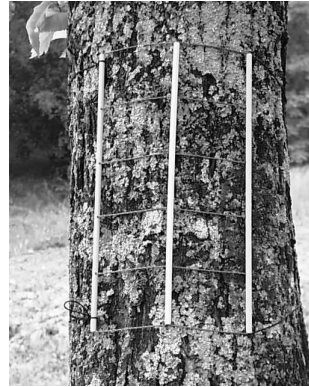


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(Giordani e Brunialti, 2008)



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1. Errors occur (always)

“To make mistakes is not desirable. To believe that no mistakes are made is naïve. To repress mistakes is not credible. To not know mistakes takes away the opportunity to improve upon them. To loose track of mistakes can lead to wrong estimates and conclusions.”

(Kaufmann and Schwyzer, Swiss NFI, 2001)

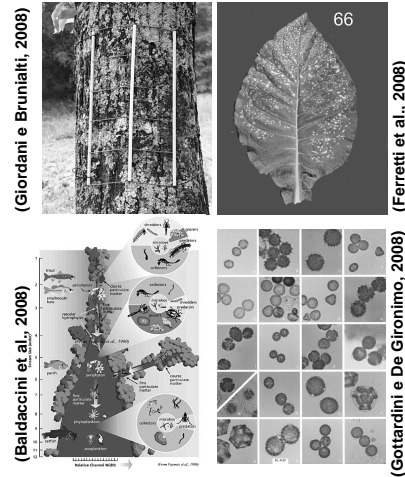


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2. Errors originate from different sources

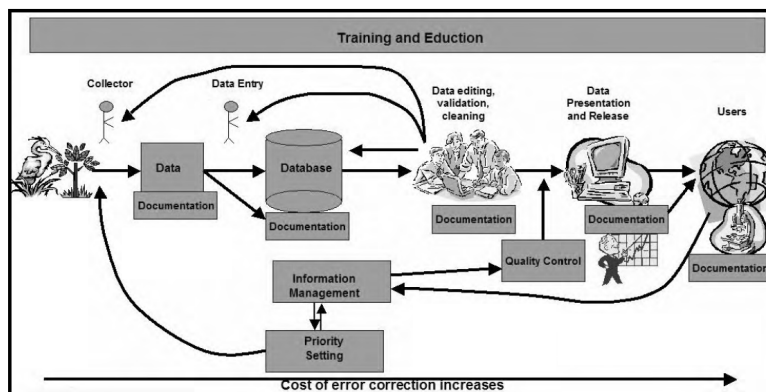
- Sampling error
- Measurement errors
- Model errors
- Non-statistical errors



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3. Errors are ubiquitous



Information Management Chain showing that the cost of error correction increases as one moves along the chain. Education, Training and Documentation are integral to all steps (from Chapman 2005a).



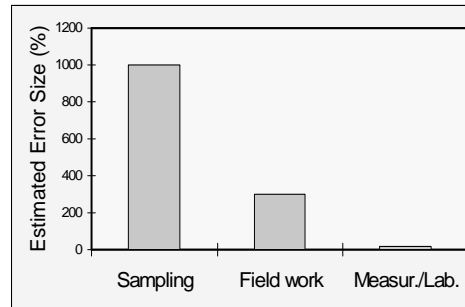
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4. The bad news: sampling error is the most critical

"..sampling is the first and least corrigible practical step of the whole...process. Errors made in this step definitely destroy all the efforts taken to assure quality during all the following steps..."

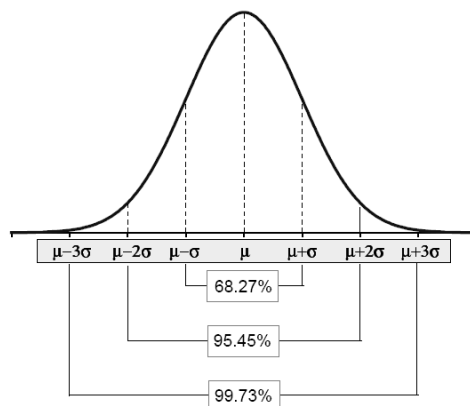
(Wagner G., The Science of Total Environment, 1995: 63-71)



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5. The good news: sampling error can be controlled (if...)

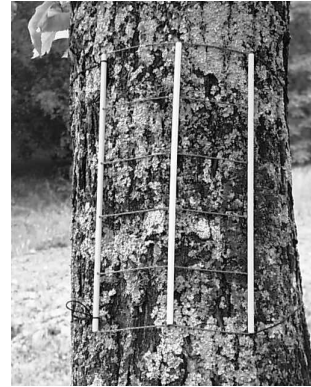


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(Giordani e Brunialti, 2008)




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Environmental surveys

Environmental population are usually constituted by a community of N individuals (animals or plants) spread over a study area



Survey variable  Quantitative characteristics
 X of the units (X_1, X_2, \dots, X_N)

(after Fattorini, 2008)



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Aim: Estimation of some population indexes

- $T = X_1 + X_2 + \dots + X_N$ Total
- $\bar{X} = T / N$ Mean

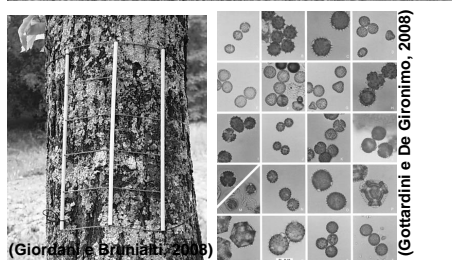
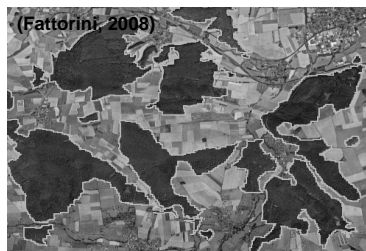
(after Fattorini, 2008)



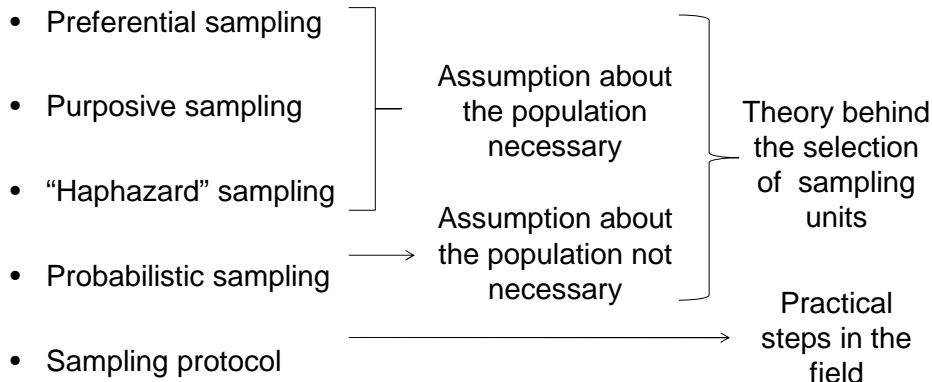
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Examples



Some terms often used



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Sample statistics and population parameters

- Sample statistics (totals, mean, variance,...) originated from measurements (observations) carried out on a subset (the sample) of the whole populations.
- Sample statistics vary – within the same population – when the sample varies.
- Sample statistics are necessary to estimate population parameters (totals, mean, variance, ...).
- Population parameters (totals, mean, variance,...) are indexes that characterizes the population.
- They are (assumed to be) fixed, mostly unknown, and they change only if the population changes.



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Statistical inference

The process of estimating population parameters (total, mean, proportion, variance,...) starting from sample statistics.

The estimation of the probability that an (unknown) population parameter is between known deviation from the known sample statistics.

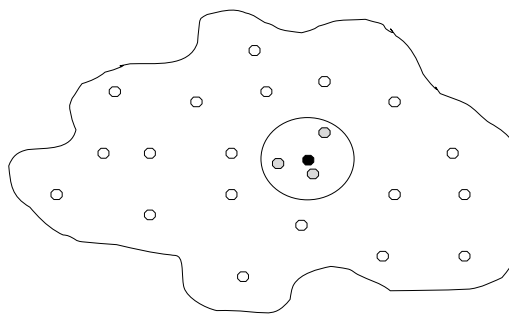


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Vascular plants in a forest - Floating plot sampling

A point is randomly thrown onto the area and the sampled units are those included in a circular or square plot of a pre-fixed size a centered at the random point.



(after Fattorini, 2008)

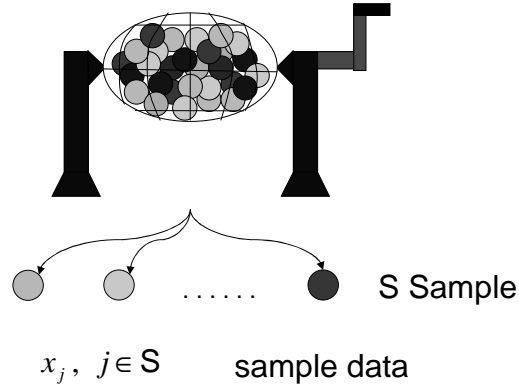


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Probabilistic sampling (after Fattorini, 2008)

Randomized scheme to select a subset S of units from the population

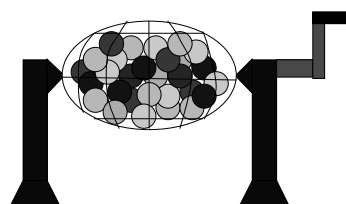


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Genesis of sampling errors (after Fattorini, 2008)

Sampling design: the set of the possible samples which can be selected by the scheme with the corresponding probabilities of selection



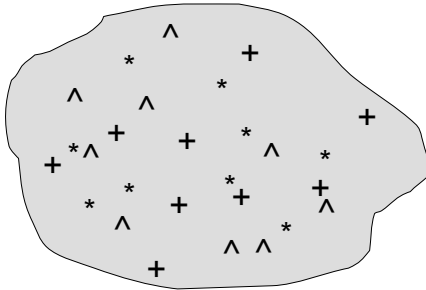
Sample	Probabilities
S_1	P_1
S_2	P_2
\vdots	\vdots
\vdots	\vdots
\vdots	\vdots
S_K	P_K



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Example



* Sample 1
+ Sample 2
.....
^ Sample n

Consequence:

Different sample statistics

What about population parameters?



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Genesis: estimation criterion (after Fattorini, 2008)

A function of the sample information to evaluate the interest parameter

Horvitz-Thompson (HT) estimator

$$\hat{T} = \sum_{j \in S} \frac{x_j}{\pi_j}$$

$\pi_j =$ Inclusion probabilities of unit j



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Genesis: sampling strategy (after Fattorini, 2008)

Sampling design + Estimation criteria

Sample	Probabilities	Estimate
S_1	P_1	\hat{T}_1
S_2	P_2	\hat{T}_2
⋮	⋮	⋮
S_K	P_K	\hat{T}_K



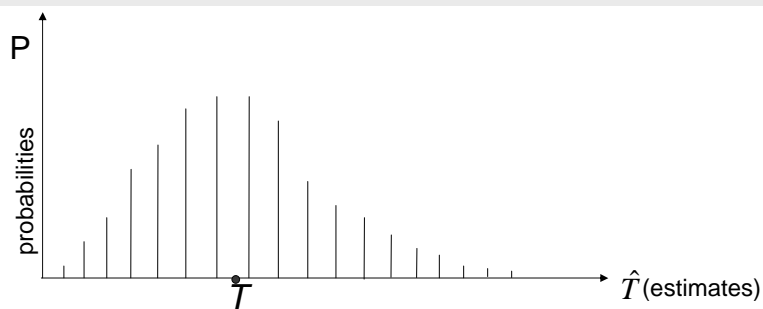
Probability distribution of the estimator



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Sampling error (after Fattorini, 2008)



$$\hat{T} - T \rightarrow \text{Sampling error}$$

Estimators are chosen on the basis of their probability distributions



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Example: tree population

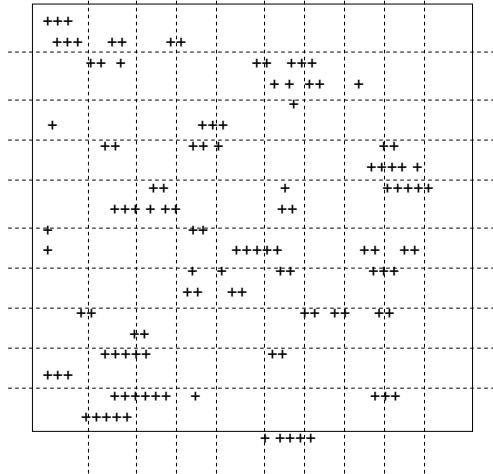
Area A: 1 km²

Objective: estimation of the total number of trees in A

Population: N not overlapping spatial cells, 100*100 m each, in A (N=100)

True values (example)

- Total (T): 400
- Mean per cell (μ): 4.0
- Standard deviation (σ): 5.005



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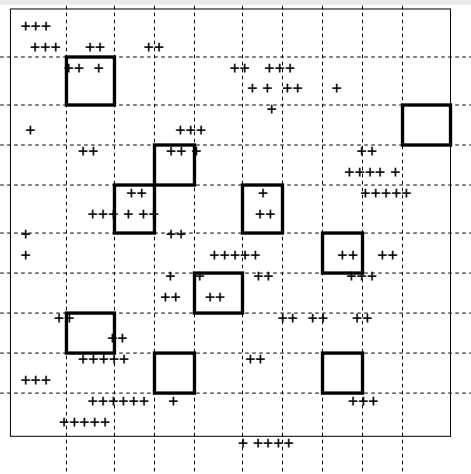
Example: sampling tree population

Simple random sampling (n=10)

- Sample mean (\bar{x}): 5.0
- Standard deviation(s): 6.146

Sample statistic=estimator (es.)

- Mean (\bar{x}): 5.0
- Total (τ): 500



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Estimation criteria and sampling errors

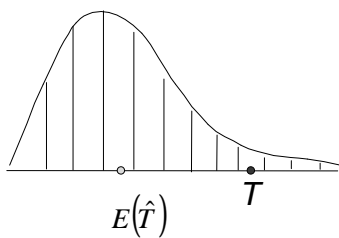
- **Accuracy** (unbiasedness). Closeness to the true value. An estimator is unbiased if the mean of all its possible values is equal to the population parameter.
- **Consistency**. When the sample size increases, the estimation gets closer to the true value of the population parameter.
- **Efficiency**. Given two unbiased estimators, the most effective one is the one with the lowest sampling variance.
- **Precision**. The closeness of repeated measures of the same quantity.



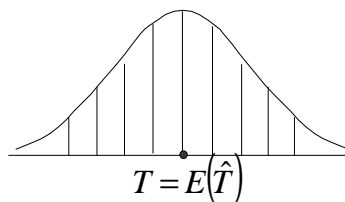
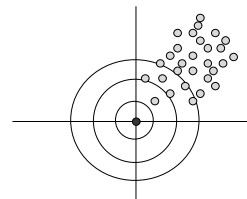
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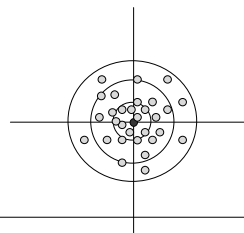
Bias (after Fattorini, 2008)



Biased



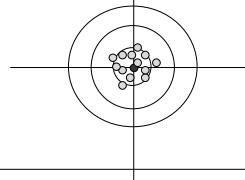
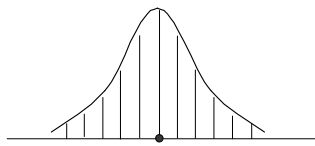
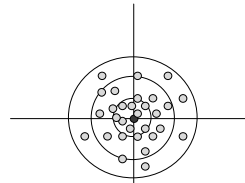
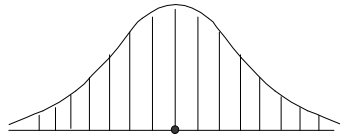
Unbiased



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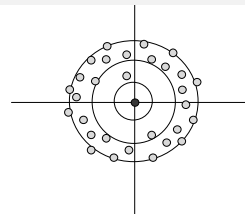
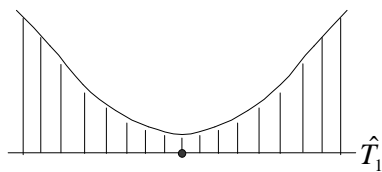
Accuracy (after Fattorini, 2008)



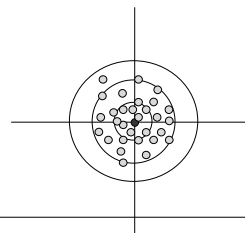
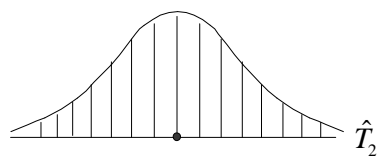
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Normality (after Fattorini, 2008)



$$V(\hat{T}_1) = V(\hat{T}_2)$$

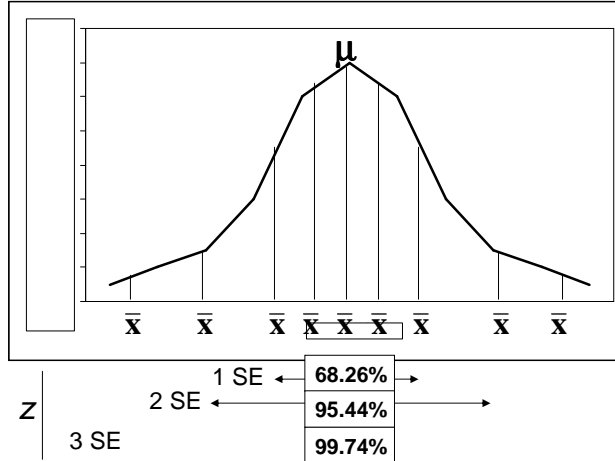


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Standard error

$$SE(\bar{x}) = \frac{s}{\sqrt{n}}$$

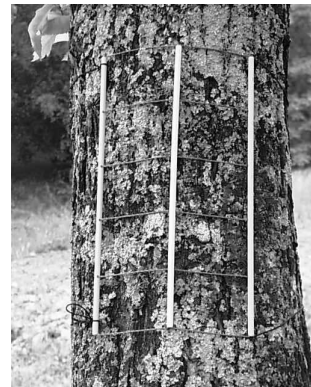


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(incl. those in the CEN draft)
- Dos and do'nts



(Giordani e Bruniatti, 2008)



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Clear objectives

Typical objective:
"Status and changes"

Important:

- Significance level (α);
- Size of the difference we wish to detect (δ);
- data variability;
- hypothesis;
- Sample size

$$\bar{X} - (t_{\alpha(2),v})SE < \mu < \bar{X} + (t_{\alpha(2),v})SE$$

$$t_{\beta(1),v} = \frac{\delta}{\sqrt{\frac{s^2}{n}}} - t_{\alpha,v}$$

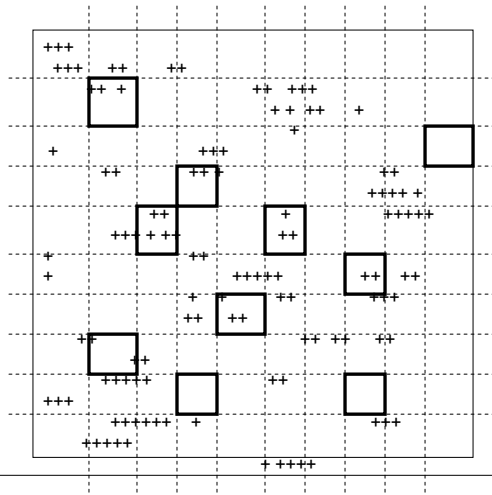
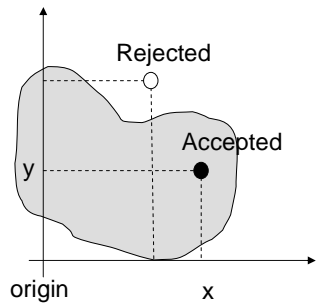
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Most common Sampling schemes

- Simple random sampling
- Systematic sampling
- Stratified random sampling
- Restricted random sampling
- Cluster sampling
- Two stage sampling

www.terradata.it

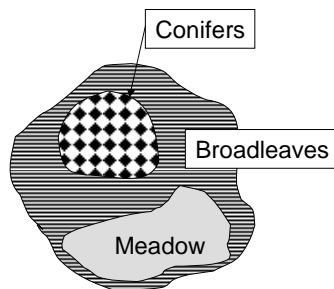
Simple random sampling



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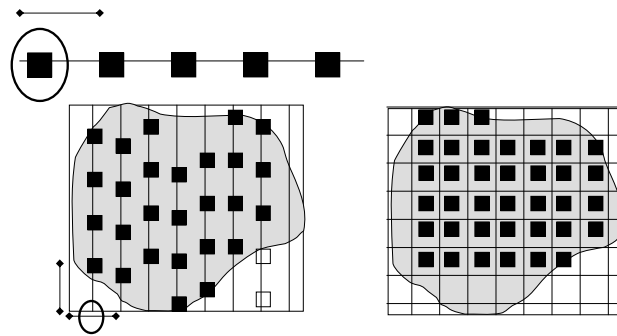
Stratified random sampling



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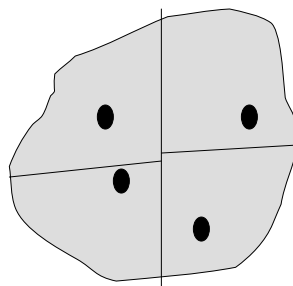
Sistematic sampling



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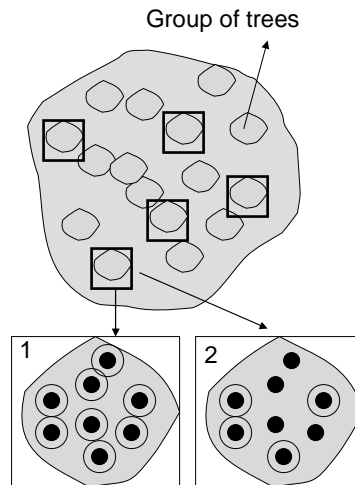
Restricted random sampling



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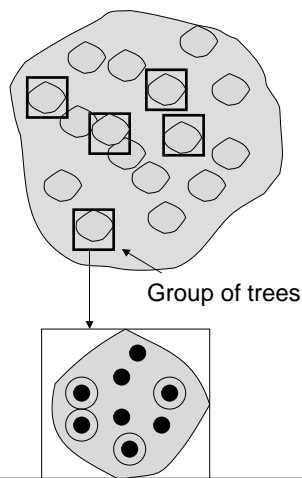
Cluster sampling



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Two stage sampling



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Sampling in the draft Norm

2.15 Target population: The totality of the possible sampling units, the totality of the situation to which conclusions can be applied. It is the target of the inferential process. In this document it is defined as: 'Lichen communities living on the bark of standard trees at an above ground level height ranging between 1 and 1.5 m. Standard trees should be defined in terms of species, bole circumference and inclination – see below) should be located within the study area'.

1 Scope

This document aims to provide a reliable, repeatable and objective method for assessing epiphytic lichen diversity. According to international literature on the topic (see e.g. Nuno et al., 2002 for an overall outline), the method can be used for assessing the impact of several anthropogenic disturbances, mainly for estimating the effects of atmospheric pollution.

2 Terms and definitions

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

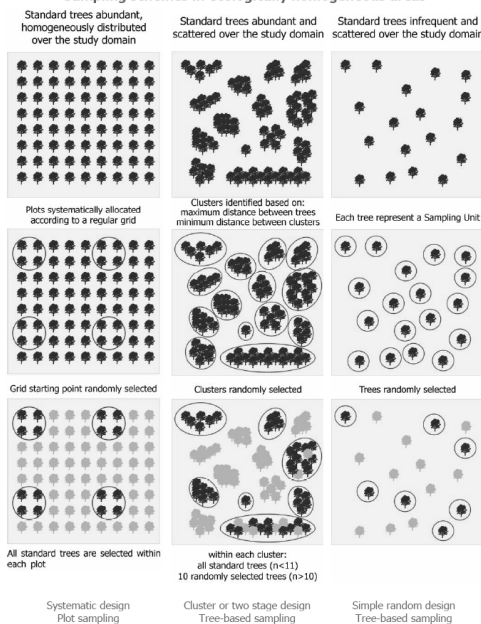
- 2.1 **Lichen:** ecologically obligate, stable, self-supporting symbiotic association of a fungus (the mycobiont, generally an ascomycete) and one or more populations of green algae and/or cyanobacteria (the photobiont), which results in a stable vegetative structure ("thallus") having a definite morphology.
- 2.2 **Lichen diversity:** in this document, an operational definition is considered in terms of Lichen Diversity Value, calculated as the sum of the frequencies of each lichen species within the observational grid (see § 7.1).
- 2.3 **Stratified sampling:** technique consisting in subdividing a heterogeneous population into sub-populations or strata, which are more homogeneous, mutually exclusive and collectively exhaustive. An independent sample is consequently taken within each of the strata.
- 2.4 **Epiphyte:** a plant or plant-like organism growing on another plant, dependent on mechanical support but not deriving nutrients from the plant upon which it grows (LIAS, 2008).
- 2.5 **Phorophyte:** a certain plant or plant-like organism (usually a tree) providing structural (e.g. mechanical) support for an epiphyte (LIAS, 2008).
- 2.6 **Sampling point:** in this context geographic location identified by a pair of geographic coordinates (Lat., Long.), being the centre of a sampling unit (SU), selected on the basis of a stratified sampling design.
- 2.7 **Sampling unit (SU):** in this context, geographical circular or squared area of determined size, centred on a sampling point which has been selected on the basis of a given sampling design.
- 2.8 **Stratum (plural: strata):** homogeneous statistical sub-population, determined from a stratification criterion (quantitative or qualitative variable) within a heterogeneous population.
- 2.9 **Taxa (plural: taxa):** a group of any rank, which can be placed in a particular level in a taxonomic hierarchy, according to the evolutionary relationships with other taxa.
- 2.10 **Lichen community:** a lichen community or biocoenosis is an assemblage of populations of lichens and fungi, whose composition and aspect is determined by the properties of the environment and by the relations with other epiphytes, animals, etc. [...]
- 2.11 **Reference population:** a set of entities concerning which statistical inferences are to be drawn, often based on a sample taken from the population.
- 2.12 **Probabilistic sampling:** part of statistical practice concerned with the selection of individual observations intended to yield some knowledge about a population of concern, especially for the purposes of statistical inference.
- 2.13 **Study area:** the geographical area considered by the study. It must be defined in details in terms of extent, land use categories, and elevation.
- 2.14 **Study domain:** The geographical extent in which the target population is located. It may coincide with the study area, or it may be more restricted, when some areas are excluded from sampling (e.g. particular land use classes, restricted access, etc.).
- 2.15 **Target population:** The totality of the possible sampling units, the totality of the situation to which conclusions can be applied. It is the target of the inferential process. In this document it is defined as: 'Lichen communities living on the bark of standard trees at an above ground level height



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Sampling schemes in ecologically homogeneous areas



a. Standard trees abundant and homogeneously distributed over the study domain

A 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 abundant, scattered over the study domain

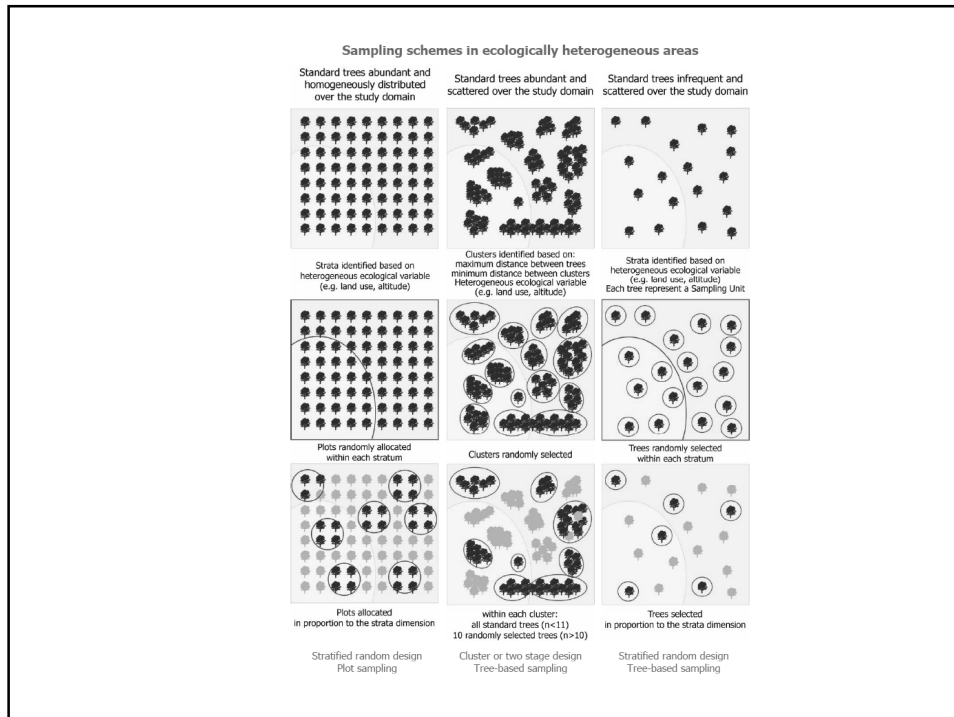
Tree-based cluster sampling or two stage sampling is recommended. Firstly define a criterion to identify clusters (e.g. defining a threshold maximum distance between adjacent trees to be included within the same cluster and/or defining a threshold minimum distance for two clusters to be considered as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

c. Standard trees infrequent, 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.



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5.3.2.2 Sampling scheme in ecologically heterogeneous areas

a. Standard trees abundant and homogeneously distributed over the study domain

A stratified random design is recommended. Plot sampling is recommended. First, identify strata on the basis of the information available on the heterogeneous ecological variable (e.g. altitudinal maps, land use classification, etc.). 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 over the study domain

Tree-based cluster sampling or two stage sampling is 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), eventually based on the information available on the heterogeneous ecological variable (e.g. altitudinal maps, land use classification, etc.), then identify and list all the clusters and obtain a random sample of them. If the number of standard trees per cluster 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.

c. Standard trees infrequent, scattered over the study domain.

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

Sampling density (examples)

- Status, simple random sampling

- n= number of sample units
- Z_{α} = standard (P:0.05 > Z: 1.96)
- S= standard deviation
- B= half-width confidence interval.

$$n = \frac{(Z_{\alpha})^2 (s)^2}{(B)^2}$$

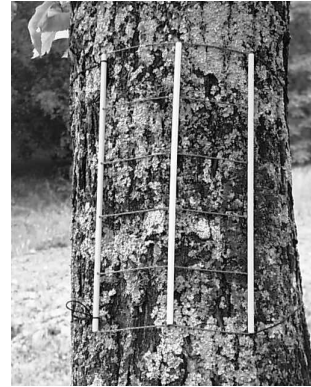
- Change, simple random sampling

- s= standard deviation;
- Z_{α} = Z values for Type I error
- Z_{β} = Z value for Type II error
- MDC= Minimum Detectable Change

$$n = \frac{2(s)^2 (Z_{\alpha} + Z_{\beta})}{(MDC)^2}$$

Talk outline

- Getting started
- Origin and control of sampling errors
- Most common sampling schemes (incl. Those in the CEN draft)
- Dos and don'ts



(Giordani e Brunialti, 2008)



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Dos

- Discuss intensively with your counterparts about objectives, expected precision, and user needs
- Identify correctly your target population
- Perform a pilot study to get basic knowledge about possible problems, constraints, expected data variability
- Balance your sampling effort in relation to required precision level and available resources
- Always adopt a probabilistic sampling design in order to make your data defensible. However remember...
- We are biologist, not statisticians. Seek proper advise from qualified statisticians BEFORE starting your investigations.



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Don'ts

- Avoid preferential sampling: your data will be highly questionable and you will be never able to estimate your errors
- Do not confound a standard protocol with a sampling design. By a protocol you ensure consistency of behaviour in the field, not a sound sampling.
- Do not over estimate your statistical package – if there are flaws in your data, they will remain even after the most intensive and sophisticated processing.



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“The results of inadequate monitoring can be both misleading and dangerous not only because of their inability to detect ecologically significant changes, but also because they create the illusion that something useful has been done”.

(Peterman, 1990)



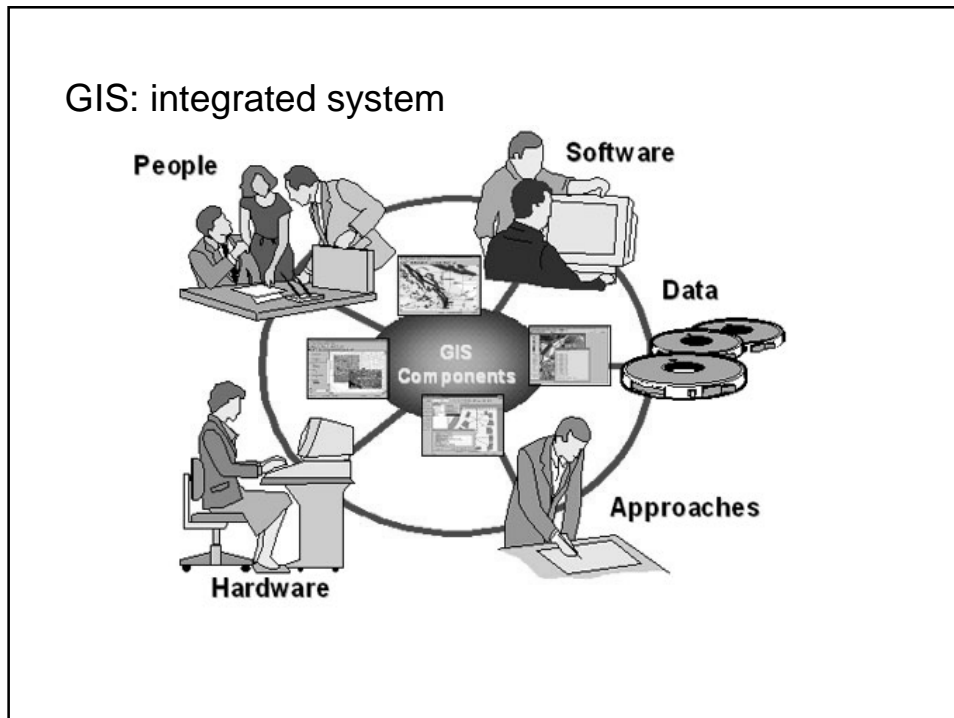
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	<p style="text-align: center;">GIS overview</p> <p style="text-align: center;">Concepts and definitions</p> <p style="text-align: center;">Francesco Geri Dipartimento di Scienze Ambientali "G.Sarfatti" Università di Siena francesco.geri@unisi.it</p>

Definition of GIS

- GIS is an acronym for GEOGRAPHIC INFORMATION SYSTEM.
- GIS are system designed to input, store, retrieve, analyze and output geographic data and information.
- GIS are complex system composed of a set of components that allow it to perform its many interrelated tasks.

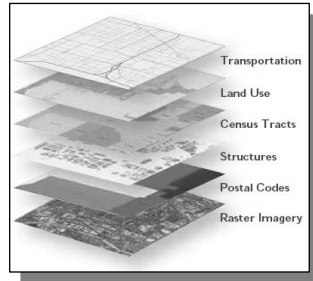


Software GIS

GIS system contains algorithms and modules specifically designed to:

- Organize geographic data within appropriate reference system
- Selectively query data and aggregate them for multiple goals
- Count and measure both individual objects and collections of object
- Classify objects based on user specified properties
- Overlay related thematic map data
- Capacity to combine these individual techniques into order of sequences of operation (model builder)

GIS: main features



A screenshot of a GIS software interface showing a data table. The table has columns for Name, Area, Perimeter, and other attributes. The data rows represent various geographic features like 'Città', 'Frazione', and 'Località'.

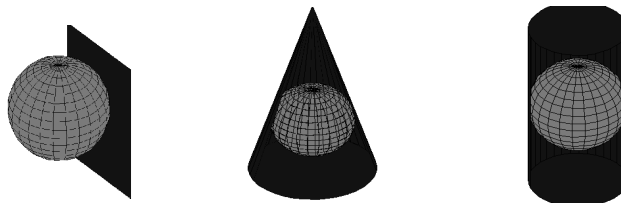
Name	Area	Perimeter	...
Città	100000	100000	...
Frazione	50000	50000	...
Località	25000	25000	...

- Overlay data
- Database Management System

Spatial Reference System

Projection System

Map projections are attempts to portray the surface of the earth or a portion of the earth on a flat surface. Map projections are necessary for creating maps. All map projections distort the surface in different ways. Depending on the purpose of the map, some distortions are acceptable and others not. Generally speaking there are 3 fundamental kind of map projections: perspective, conical and cylindrical.



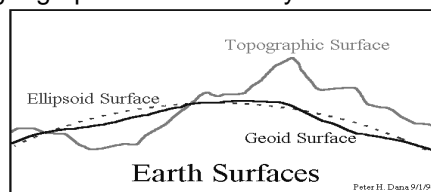
Spatial Reference System

Datum

A datum is mathematical surface on which a mapping and coordinate system is based.

Datum is a set of parameters that describe the position and the type of ellipsoid used: dimension, orientation, origin point etc.

Referencing geodetic coordinates to the wrong datum can result in position errors of hundreds of meters. Different nations and agencies use different datums as the basis for coordinate systems used to identify positions in geographic information systems.



Spatial Reference System

Coordinate systems

The coordinates are used to uniquely identify the location of a point on the Earth's surface.

There are many different coordinate systems, based on a variety of geodetic datums, units, projections, and reference systems in use today.

Simplifying, there are 2 main groups of coordinates:

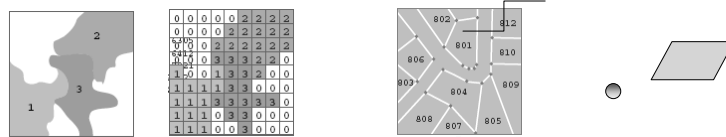
- 1) Geographic (degree)
- 2) Projected (meters)

Data representation

GIS data represents real objects (such as roads, land use, elevation) with digital data.

Real objects can be divided into two abstractions: discrete objects (a house) and continuous fields (such as rainfall amount, or elevation).

Traditionally, there are two broad methods used to store data in a GIS for both abstractions: **raster** and **vector**.

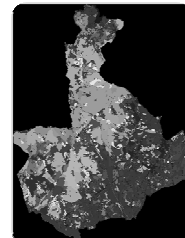
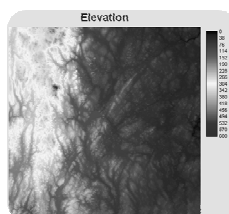
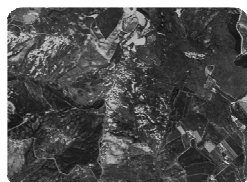


Data representation

Raster data type consists in a grid of cells with each cell storing a single value.

Raster data can be images (raster images) with each pixel (or cell) containing a color value.

Additional values recorded for each cell may be a discrete value, such as land use, a continuous value, such as temperature, or a null value if no data is available.



Data representation

In a vector data model, geospatial data is represented as geometrical shapes. The basic units in vector models are points, line and polygons.

Each of these geometries is linked to a row in a database that describes their attributes.

The vector format have 3 main components: geometrical (the shape of features), attribution (the information stored in table) and topology (relationship between features).

Shape	Name	Area	Perim	Comment
Polygon	Urban Area Type	6252276	47024	TRAPICANO TERRE
Polygon	Urban Area Type	21019495	69167	ALCANTO
Polygon	Urban Area Type	14300481	68872	LOAVELLE
Polygon	Urban Area Type	7843842	67791	TRAVAROLA
Polygon	Urban Area Type	11088481	67684	TRUCCHIOLO
Polygon	Urban Area Type	10087186	67607	MONTEFONTE D'ORZUELA
Polygon	Urban Area Type	11488168	67582	SAITANO
Polygon	Urban Area Type	6284882	67373	TRAGANDEA
Polygon	Urban Area Type	11491548	66814	TRAPICANO
Polygon	Urban Area Type	5844182	43111	TORRELA DI VETRA
Polygon	Urban Area Type	4482738	43045	VALLE D'AVAROTTO
Polygon	Urban Area Type	10522019	67176	TRIVITTO D'ALBA
Polygon	Urban Area Type	66411571	51209	SAN GIOVANNI D'AVESSE
Polygon	Urban Area Type	13247715	66371	TRINZANO
Polygon	Urban Area Type	12424582	66082	TRIVITTO D'ALBA
Polygon	Urban Area Type	6221264	36707	SAN GIACOMO D'ORZUELA
Polygon	Urban Area Type	30091481	47191	TRIVITTO D'ALBA
Polygon	Urban Area Type	26761201	47114	TRIVITTO D'ALBA
Polygon	Urban Area Type	44762621	65786	CATOLINE D'ORZUELA
Polygon	Urban Area Type	16969162	65511	SAITANO
Polygon	Urban Area Type	11846776	63122	TRUCCHIOLO
Polygon	Urban Area Type	52117821	24822	TRIVITTO
Polygon	Urban Area Type	59110389	43079	TRIVITTO D'ALBA
Polygon	Urban Area Type	10719189	61391	SAN GIOVANNI D'AVESSE
Polygon	Urban Area Type	89942389	63849	TRIVITTO D'ALBA

GIS SOFTWARE



ESRI Arcview / Arcgis



ERDAS Imagine / ER mapper



CLARK LABS Idrisi



Quantum GIS



GRASS



gvSIG

Old growth structures and lichen conservation in subalpine coniferous forests: a lesson from the Paneveggio Forest (NE Italy)

Juri Nascimbene

Dept. of Life Science, University of Trieste

With the collaboration of P.L. Nimis (Trieste), Lorenzo Marini (Padova), Renzo Motta (Torino), Marco Carrer (Padova)

The research on the lichen biota of the Parco di Paneveggio Natural Park: a long history

Ferdinand Arnold 1897: during his field trip in South Tyrol he visited several localities included in the Park providing a large amount of floristic information on the lichen biota of this area: **more than 400 species**

Nascimbene 1995-1997:

Floristic survey and description of the main lichen communities

The first checklist

Thor & Nascimbene 2005: a new floristic survey

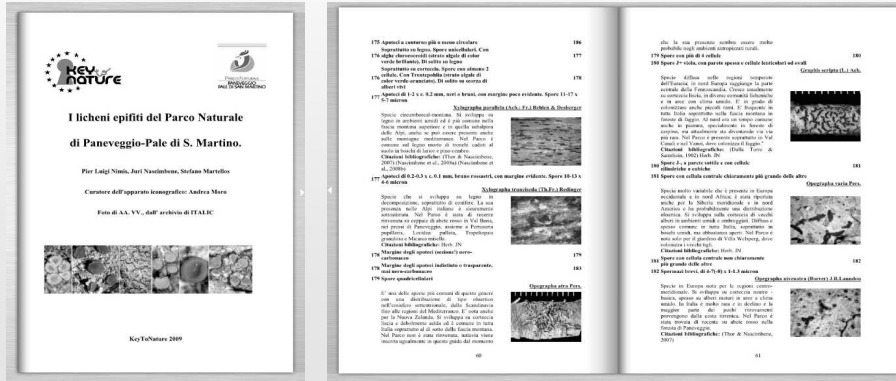
Nascimbene & Nimis 2005-2008:

Lichens in old growth forests

Effect of forest management

Updated checklist

Interactive identification tools for epiphytic lichens



The updated checklist includes c. 650 species (242 epiphytic lichens).
 However, a realistic estimate of the lichen biota hosted in the Park could
 be c. 900 species...let's go and find the lacking lichens!!

▪ Which are the main drivers undergoing this large lichen diversity?

Differences in **climatic conditions** between different parts of the Park:
from suboceanic (e.g. Val Vanali) to continental (e.g. Paneveggio)

Several **rock types**: from calcareous, to arenaceous, to metamorphic and porphyric
rocks

Presence of different **forest types**: from beech, to spruce, to larch-stone pine-dominated
forests

Presence of **old growth forests** (large, old trees, deadwood)

Unpolluted watercourses hosting several and rare aquatic lichens

...and...a long tradition of lichen research of course!

What are we doing on epiphytic
lichens in old growth forests?

General context...

In the European Alps, forests are known to be important for biodiversity conservation. Several forest types are included among the habitats worthy of conservation in Europe (EU 92/43 Directive).

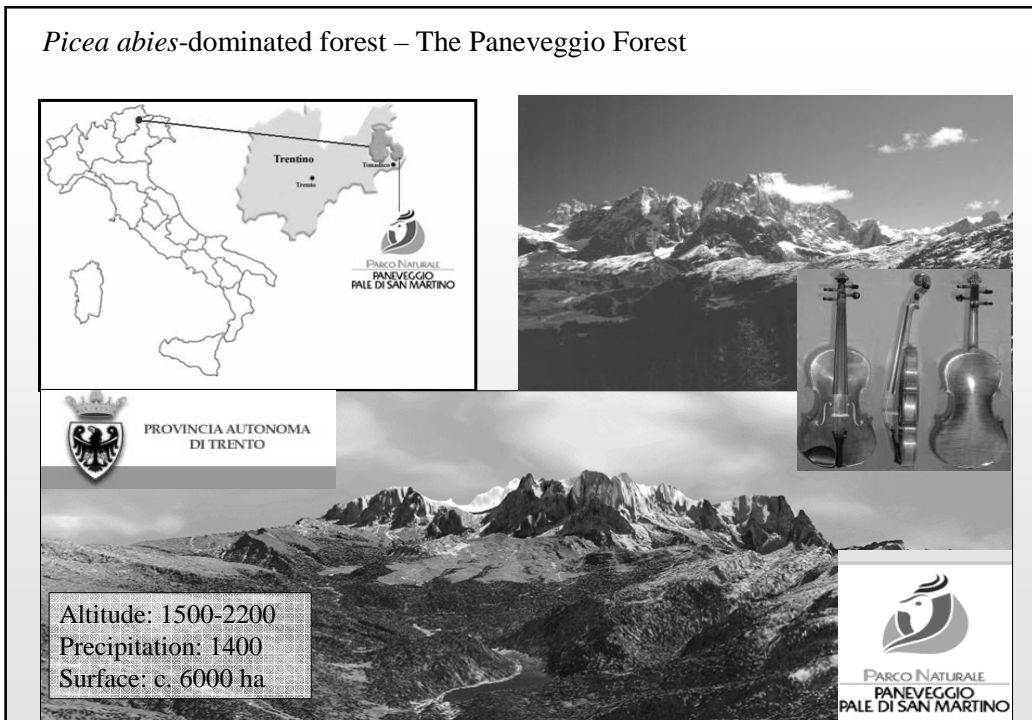
In the Italian Alps, despite the noticeable reduction of human activities since the beginning of the 20th century, spruce forests are still intensively managed for timber production.

Management practices are mainly based on the shelterwood system and on selective cutting (with several variants)

In the Italian Alps, the role of lichens in shaping forests biodiversity is still largely unknown and several forest substrates (e.g. dead wood) are overlooked

Testing the relations between tree parameters, the availability of CWD and the richness and composition of lichen communities could be a first step to gather information for forest managers interested in biodiversity conservation and monitoring.

Picea abies-dominated forest – The Paneveggio Forest



-forest management is systematically planned since **1847**

-Ecological research has a long tradition (e.g. 40 years studying the ecology of *Picea abies*)

-In **1990** a 123 ha Forest Reserve was established for long term-monitoring (in 5 1 ha plots) and ecological research which is mainly focused on the dynamics of the establishment and evolution of *Picea abies* (University of Firenze and Torino)



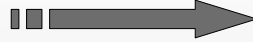
What was stimulating in studying lichens in this forest?

In the long-term monitoring plots, a lot of information was available to be related to lichen occurrence

-Age

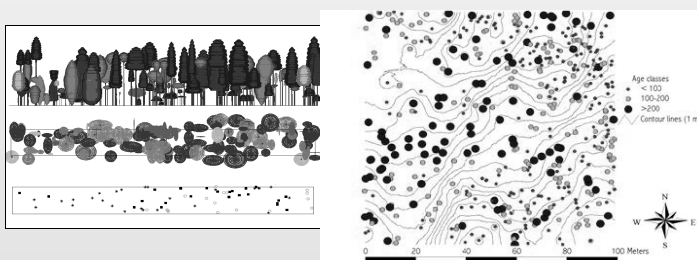
-DBH

-H of the first branches



-Surface of the crown

-CWD (logs, stumps, snags; decay stage)



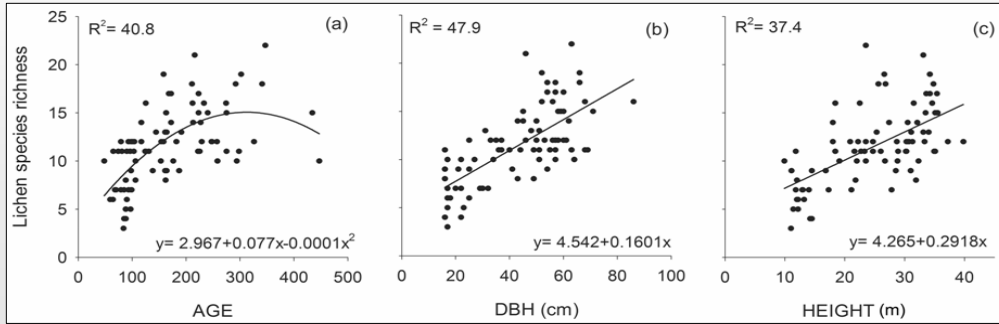
Influence of tree age, tree size and crown structure on lichen communities in mature Alpine spruce forests

For epiphytic lichen surveying, trees were stratified into 3 age classes representing the main tree generations potentially corresponding to different stages of the management process:

- (1) <100 years old, immature trees usually not suitable for felling,
- (2) 100-200 years old, mature trees suitable for felling, and
- (3) >200 years old, over-mature trees normally rare or absent in managed stands.

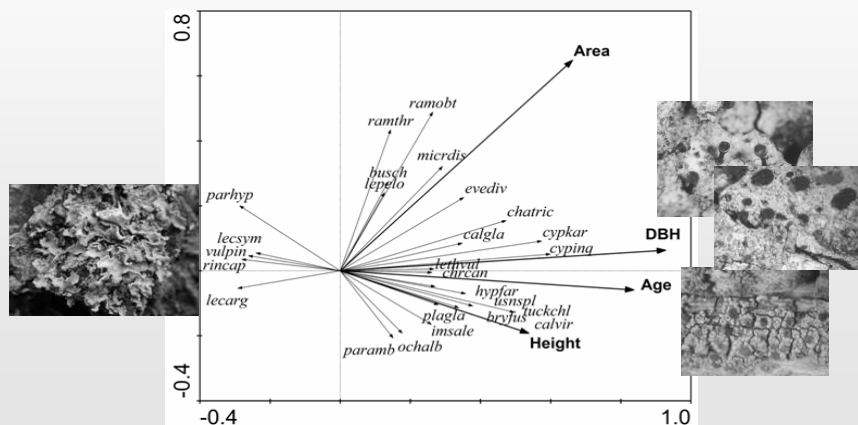


The longer time for lichen colonization and higher size of trees could account for the higher species richness on over-mature trees.



Species composition is influenced by tree age, tree size, and crown structure which could account for microclimatic differences between young and over-mature trees.

Several nationally rare species are associated to over-mature trees, including most of the calicioid lichens. This result suggests that the rarity of some epiphytic species in Italy could be related to the virtual absence of over-mature trees in managed forests

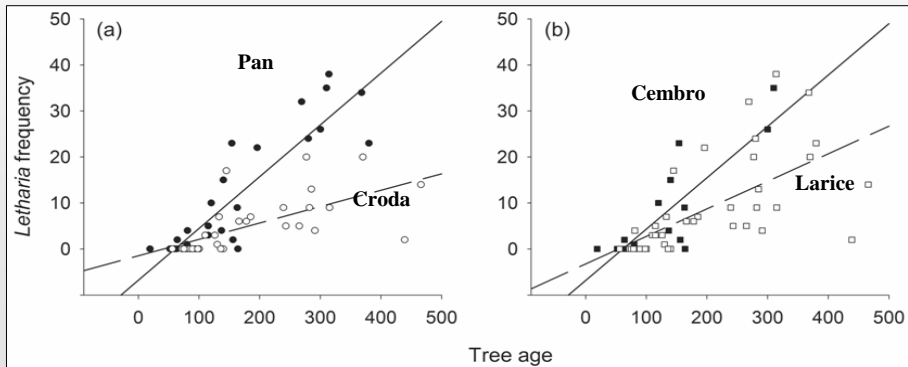




We also tested the influence of tree age on the habitat-restricted lichen *Letharia vulpina*. Results confirm that age is the most relevant tree-level variable in explaining its frequency.

Letharia frequency is positively related to tree age in both localities and on the two tree species on which it grows

On these basis this easily identifiable lichen could be used to rapidly identify trees/sites which potentially host nationally rare species which are often difficult to detect due to their small size.



And what about the role of dead wood?

We studied lichen diversity on stumps in relation to wood decay

Increasing wood decay



...please, look at some methods used for the lichen survey

10 cm x 10 cm



The main genera were *Cladonia* (17 species), *Buellia* (4 species), *Lecanora* (4 species), *Micarea* (4 species), and *Cyphelium* (3 species).
 7 calicioid lichens and 6 nationally rare species

Even if the total number of species do not differ among decay stages, we found some modifications in the behaviour of species composing lichen communities in the different decay stages.

Species	Decay stage			P*
	2	3	4	
Total	9.313	7.333	6.771	0.36
Crustose	4.708 ^a	2.083 ^b	2.167 ^b	0.04
Foliose	1.8125	1.041	0.562	0.10
Fruticose	2.792	4.208	4.042	0.10
Epiphytic	4.375 ^a	2.375	1.042	0.04
Epixylic	3.917	2.917	2.500	0.33
Terricolous	1.021 ^a	2.042	3.229	0.01

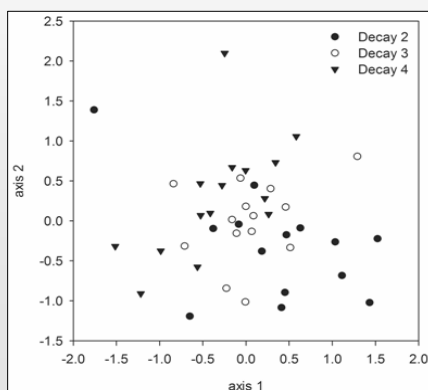
Each decay stage have some associated species (es. *Lecidea turgidula* classe 2, *Cladonia coniocraea*, classe 3; *Imadophila ericetorum* classe).

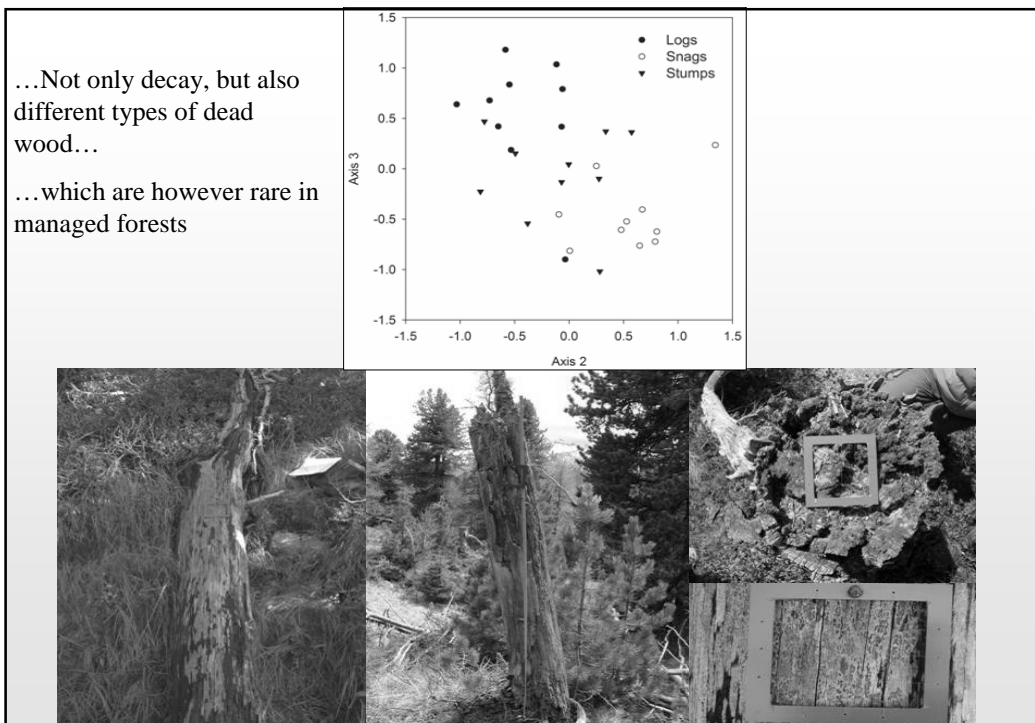
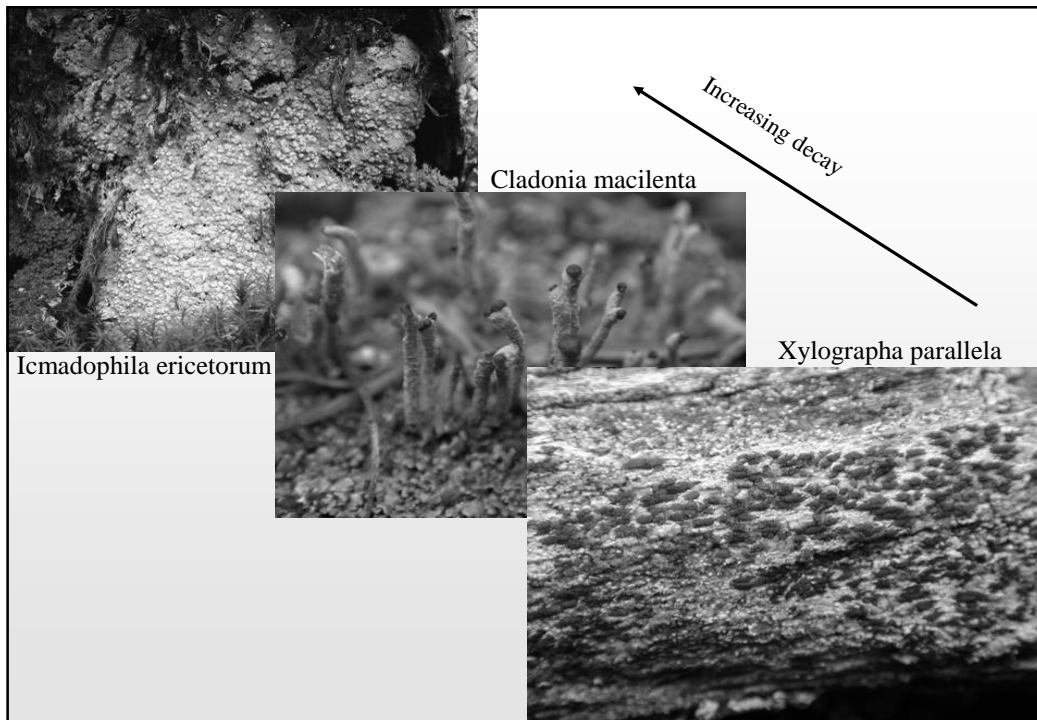
Species composition varies among decay stages but communities broadly overlap

Ecological explanation

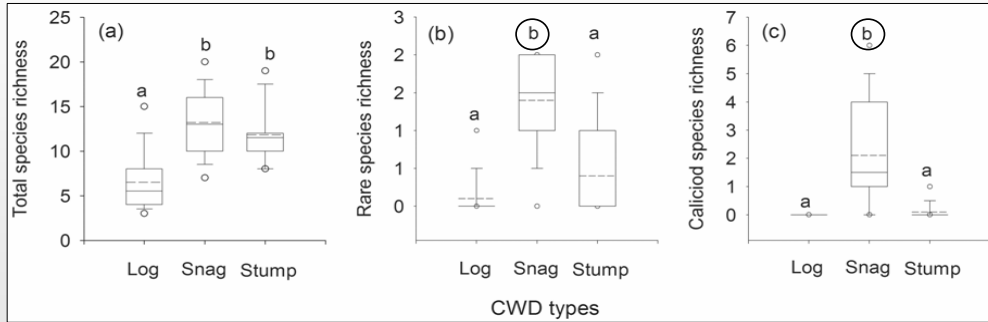
Wood decay is a continuous process and different decay stages can be found at the same time on one stump

During this process lichen communities are subjected to functional (growth forms) and ecological (substrate) modifications as a response to changing stump conditions.





Snags and stumps are probably the main substrates for lignicolous lichens, but snags were found to host a higher number of nationally rare species and calicioid lichens



What is important for conservation of epiphytic lichens?

Age and size of trees

What should be done?

Please, leave trees older than 200 years which are now virtually absent in production forests

For example with a longer rotation period

What is important for conservation of lignicolous lichens?

Different wood decay stages

Different types of dead wood

What should be done?

Please, leave snags and logs which are usually lacking in production forests where only stumps can be found

Maintain also different decay stages for example using a progressive thinning within stands

Towards an international standard for lichen monitoring. 7 - 11 June 2010 Trento

Towards an international standard for lichen monitoring: results 2010

Giorgio Brunialti, Paolo Giordani, Fabiana Cristofolini, Elena Gottardini, Marco Ferretti

Scientific Organization

Supported by



Main results

- Indoor exercise

- Outdoor exercises
 - Plot A, before training: standard trees and lichen assessment
 - Plot C, identification of the plot center
 - Plot B, standard trees and lichen assessment
 - Plot A, after training: lichen assessment

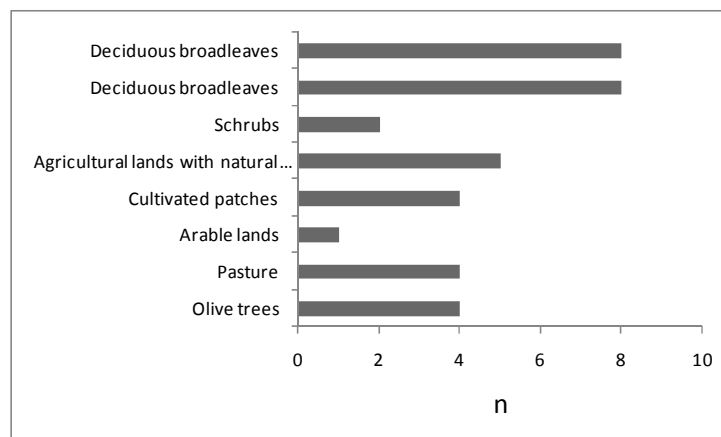
Indoor exercise: monitoring design



- Study domain:
 - Area: a remote area in Tuscany, Italy
 - Land Use Categories (LUC) included:
 - 2.2.3 Olive trees
 - 2.3.1 Pasture
 - 2.1.1.1 Arable lands
 - 2.4.2 Cultivated patches
 - 2.4.3 Agricultural lands with natural vegetation
 - 3.2.2 Schrubs
 - 3.1.1.2 Deciduous broadleaves
 - 3.1.1.4 Deciduous broadleaves



LUC considered by the teams



LUC considered by the teams



Code	Land Use Categories								Total LUC considered
	2.2.3	2.3.1	2.1.1.1	2.4.2	2.4.3	3.2.2	3.1.1.2	3.1.1.4	
1	0	1	0	1		0	1	1	4
2	1	0	0	0		1	0	1	4
38	1	0	0	0		0	0	1	3
47	1	0	0	0		1	0	1	4
56	1	1	1	1		1	1	1	8
9	0	1	0	0		0	0	1	3
10	0	0	0	1		1	1	1	5
11	0	1	0	1		1	0	1	5
Total	4	4	1	4		5	2	8	8



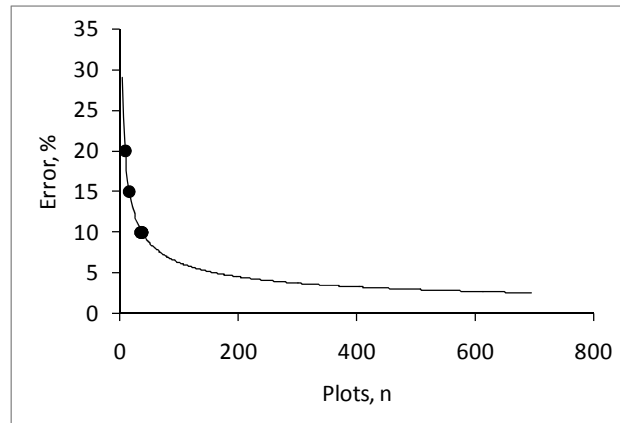
Suggested sampling schemes



Code	Total LUC considered	Sampling scheme	n strata	Note	Precision level (% of mean LDV)	P	n
1	4	StRS	4	-			
2	4	StRS	4	-	10	95	35
38	3	StRS	3	-	20	95	9
47	4	SRS	0	-	15	95	15
56	8	SiRS	0	-	15	95	15
9	3	StRS	3	-	10	95	36
10	5	StRS	4	LUT 2.4.2 and 2.4.3 merged into a single strata	10	95	35
11	5	StRS	3	LUT 2.3.1.1, 2.4.2, 2.4.3 merged into a single strata	10	95	35



Suggested sampling densities in relation to expected error (%)



Outdoor exercises

- Dimension of the individual quadrats of the sampling grids used by each team
- Plot A, before training: standard trees and lichen assessment
- Plot B, standard trees and lichen assessment
- Plot C, identification of the plot center
- Plot A, after training: lichen assessment



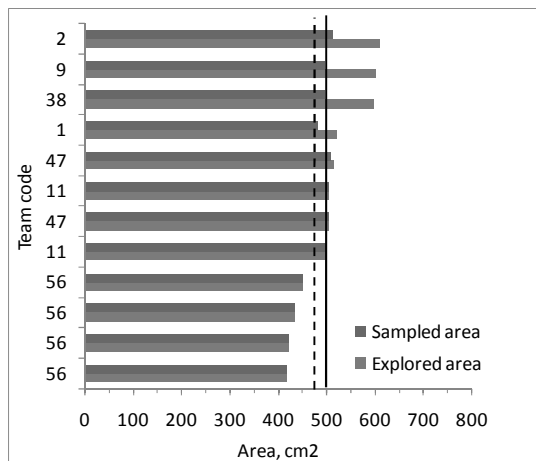
Sampling grid dimension



	Length			Width		
	min	mean	max	min	mean	max
Total	49.5	52.5	60.2	8.4	9.6	10.5
1	9.7	10.0	10.3	8.4	9.6	10.3
2	9.7	10.0	10.2	8.4	9.6	10.4
3	9.7	9.9	10.0	8.4	9.6	10.2
4	9.7	10.0	10.1	8.4	9.6	10.1
5	9.7	9.9	10.1	8.4	9.6	10.2



Sampling grid Explored area vs. sampled area

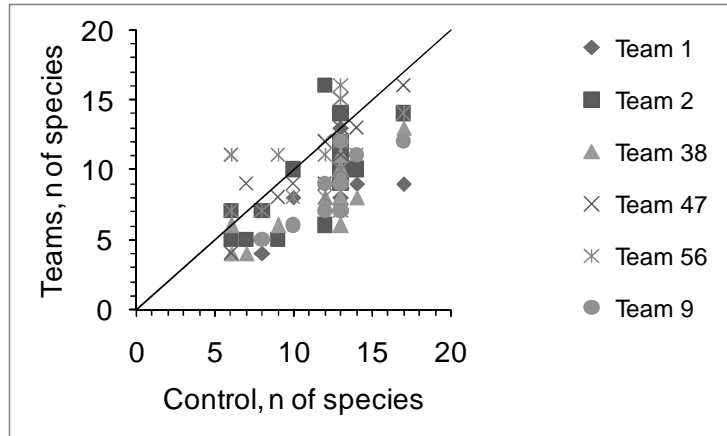


— standard area

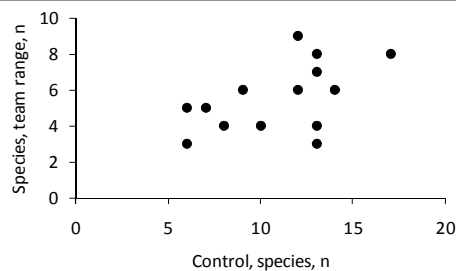
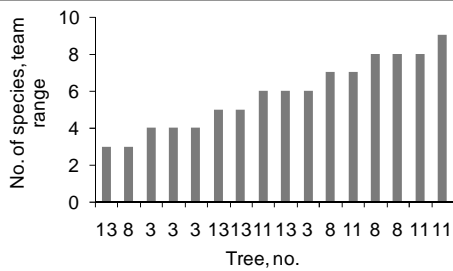
- - - mean area



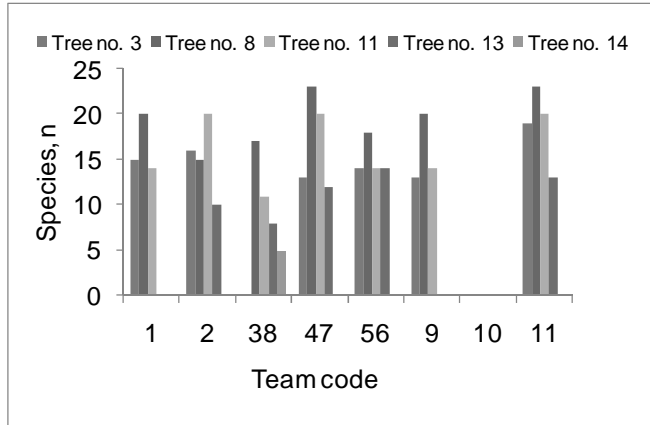
Outdoor – Plot A Species richness, individual grids



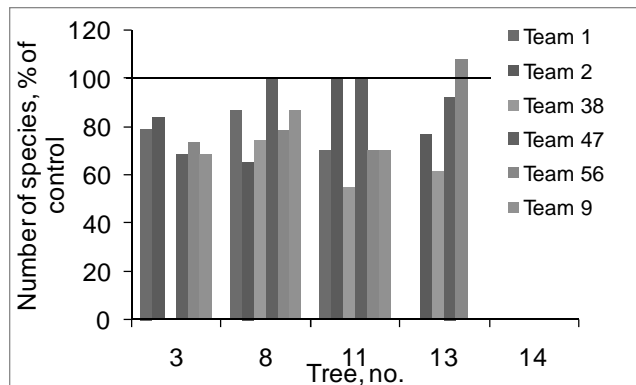
Outdoor – Plot A Range of values, individual grids



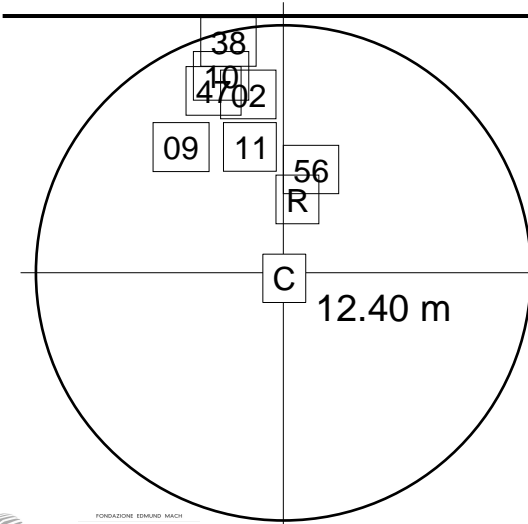
Outdoor – Plot A Species richness, tree level



Outdoor – Plot A Species richness, tree level



Outdoor - Plot C identification of the plot center



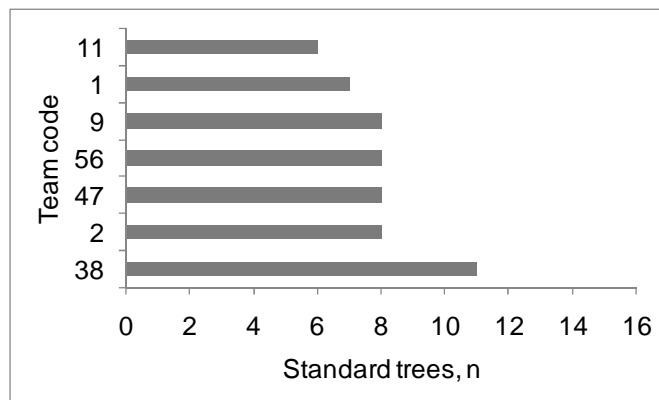
Numbers = team codes

R = repetition of
identification of plot
center, same GPS, same
operator

Mean distance from "true"
plot center = 8.33 m

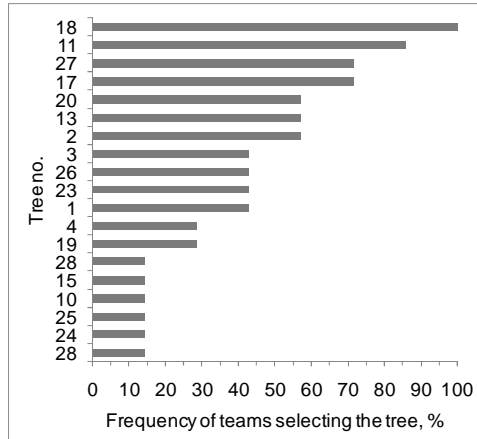


Outdoor - Plot B Standard trees selected by each team

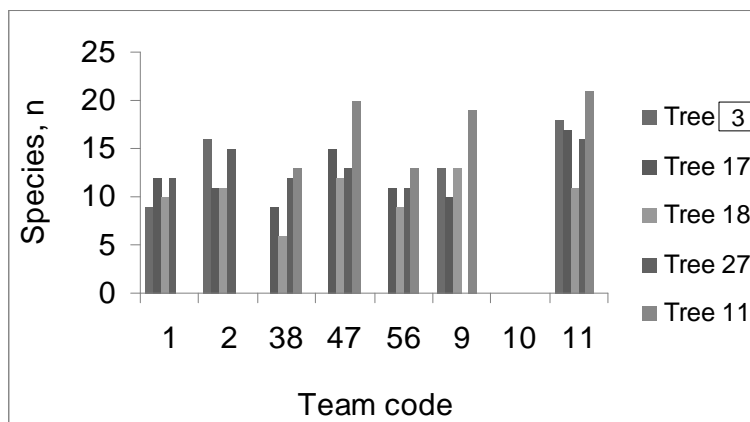


Outdoor - Plot B Frequency of selection

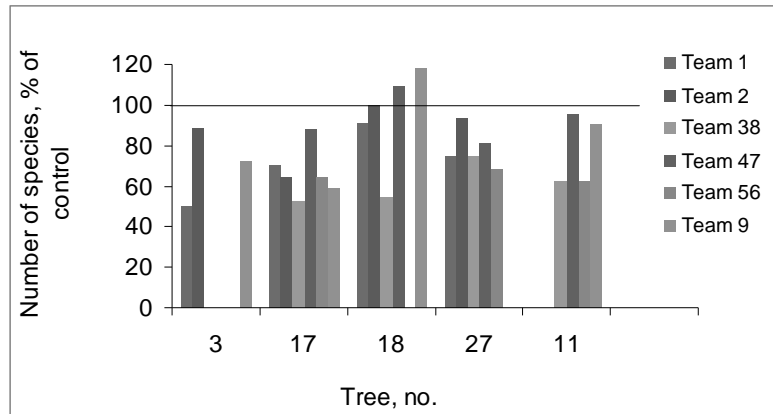
Total trees selected n=19



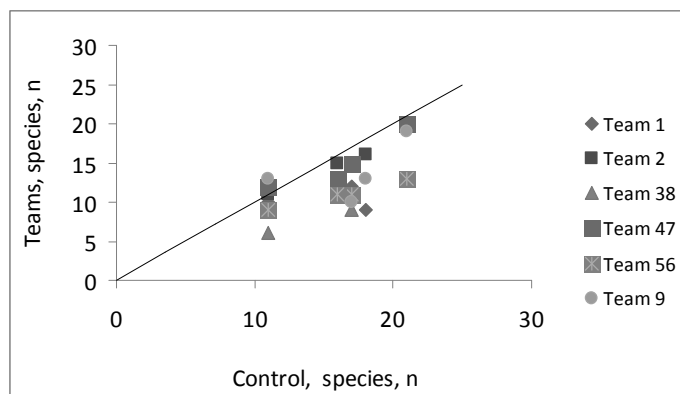
Outdoor – Plot B Species richness, tree level



Outdoor – Plot B Species richness, tree level



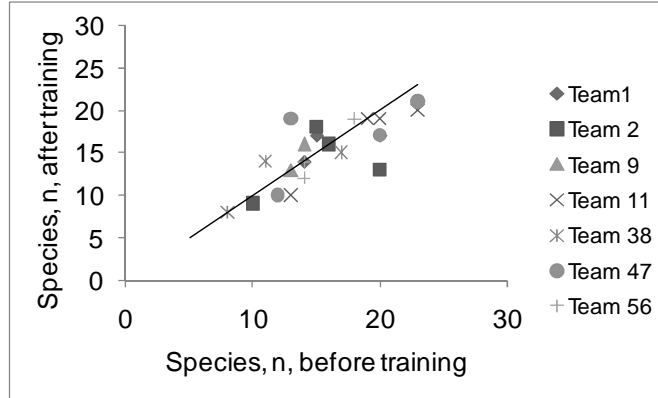
Outdoor – Plot B Species richness, tree level



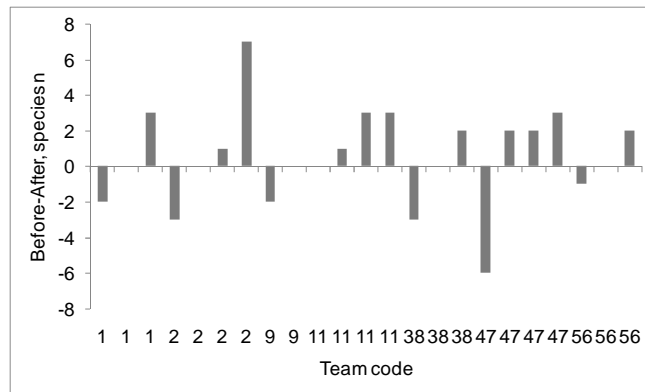
Outdoor – Plot A, after training Before-After, tree-level



Lichen assessment



Outdoor – Plot A, after training Before-After, tree-level



Conclusion

Indoor exercises

Issues for consideration

- Low familiarity with probabilistic sampling
 - Poor definition within the Norm?
 - Need for training?
 - Solvable problem
- Low familiarity with GIS systems
 - Need for training
 - Solvable problem



Conclusion

Outdoor exercises

Issues for consideration

- Differences in size of the sampling grids
 - Important?
 - Different areas of trunk explored
 - Easy to be solved – set tolerance limits!
- Differences in the selection of standard trees
 - Poor definition of standard characteristics by the Norm?
 - Poor application of the Norm by the teams?
- Differences in species frequency
 - Expected (familiarity with local flora)



FINAL REMARKS

Paolo Giordani

Discussion on critical issues emerged during the exercises; suggestions for the final draft of the international standard.

Topic	Proposal
Definition of "ground level"	Avoiding roots: starting from where the trunk shape becomes homogeneous.
Determining the level for measurements of the circumference	1.30 m above ground level is calculated with a rigid tape located starting from the ground level perpendicularly to the longitudinal axes of the trunk.
Where to put the grid	1 m above ground level at each aspect.
Grid material and characteristics	Specify tolerance limits for sampling grid dimension.
Epiphytes coverage	Rewording the paragraph, making more clear that e.g. small epathic e.g. Frullania species have not to be included in the calculation of 20% disturbance coverage?
Obstacles around the trees	Do not change: do not insert specific limitations for this.

Towards an international standard for lichen monitoring – theory and practice

Participants at the harmonization field course

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<i>Brunialti</i>	<i>Giorgio</i>	Siena	Italy
<i>Caporale</i>	<i>Stefania</i>	Santa Maria Imbaro	Italy
<i>Cristofolini</i>	<i>Fabiana</i>	San Michele a/A	Italy
<i>Ferretti</i>	<i>Marco</i>	Siena	Italy
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