



Ciliated protists as indicators of soil health: Three case studies from Italy



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**KEEP SOIL ALIVE
PROTECT SOIL
BIODIVERSITY**

**GLOBAL SYMPOSIUM
ON SOIL BIODIVERSITY**

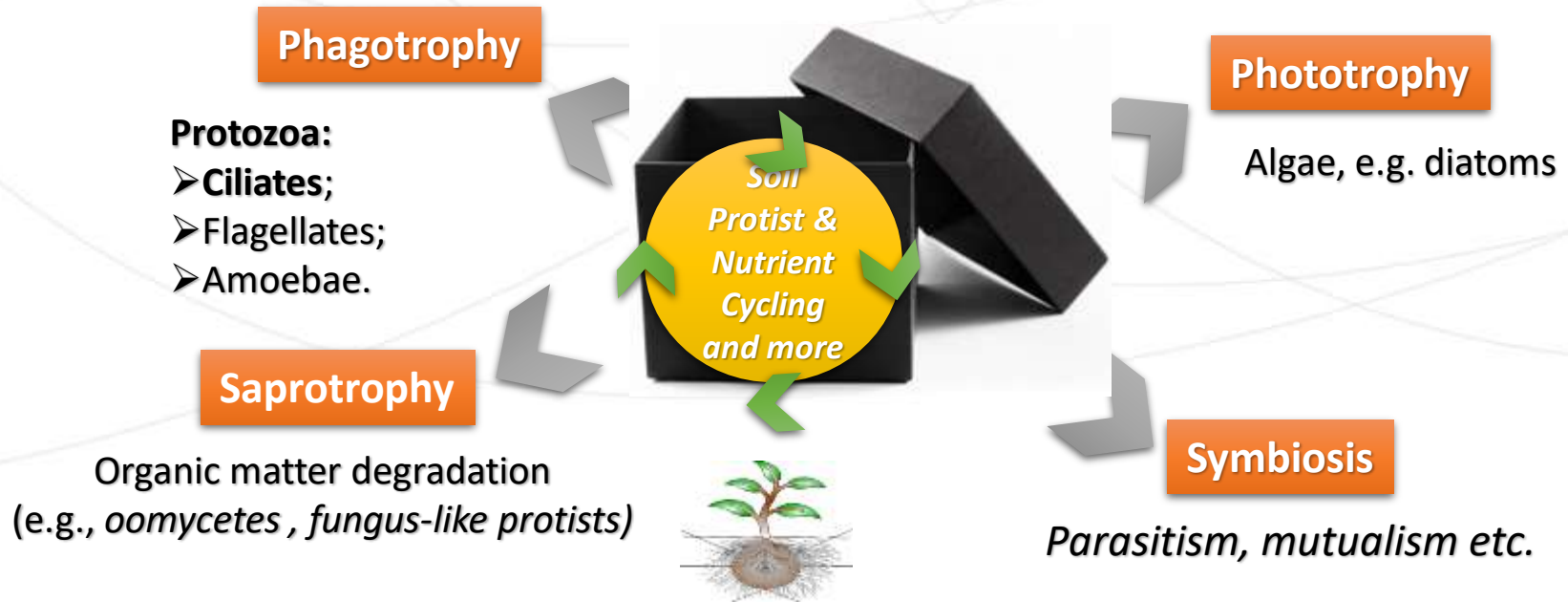
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➤ *Protists: key multi-channel feeders in the soil food web*

✓ One of the most diverse and abundant group of soil eukaryotes an essential component of the *rhizosphere microbiome*.

The Functional versatility of soil protists!



All functional groups of soil protists provide critical roles in nutrient cycling.



Aims: to provide an overview of the *outcomes and challenges encountered using ciliated protists as indicators of Soil Health* in the framework of several projects conducted in Italy since 2009.

1st The BioPrint Pilot Project



2nd Ciliates in organic vineyards



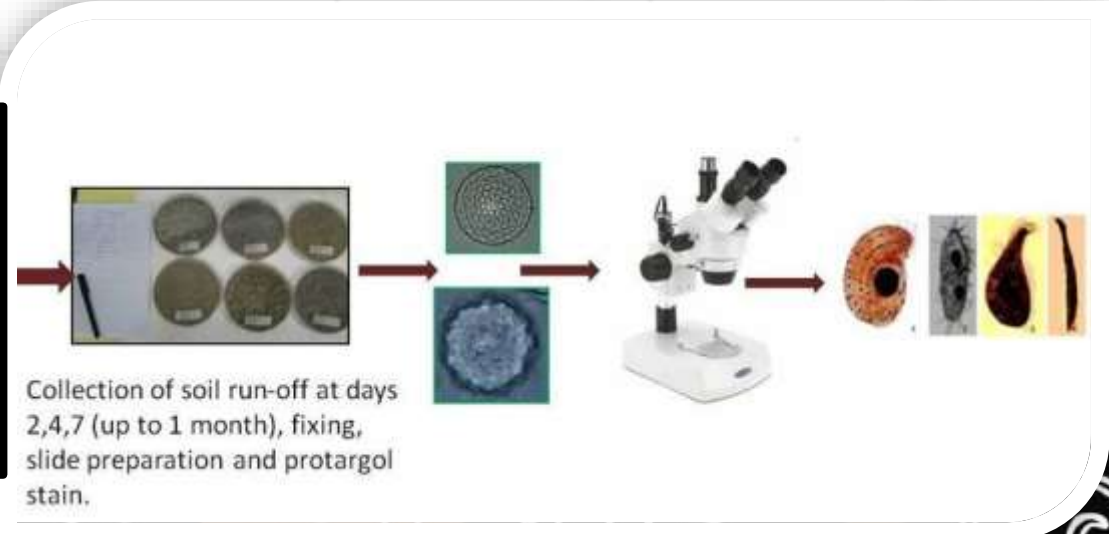
3rd Ciliates in industrial sites – Soil Mapping Lombardia



A “Classical” Methodological Approach:

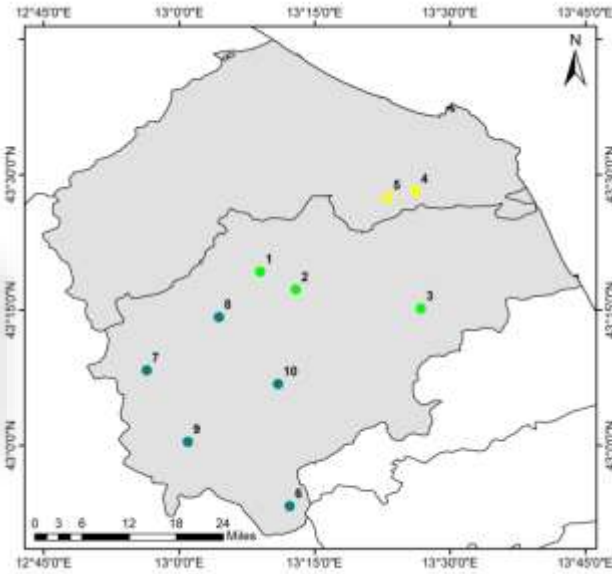
Qualitative-quantitative analysis of the ciliate diversity and community structures at the investigated sites.

- 1) Non-Flooded Petridish” methods (Foissner in Protocols in Protozoology, 1992)
- 2) Live observations;
- 3) Fixing and Slide preparation;
- 4) Protargol staining;
- 6) Identification (genus/species level) & direct counting on slide;
- 7) Data analysis (diversity indices & multivariate statistics)



Case studies across Italy. 1st The BioPrint Pilot Project

Main aims: to evaluate the capacity of ciliates to discriminate between different land uses (forests and agroecosystems) and farming management practices (organic vs conventional) with different level of soil disturbance.



Agroecosystems

- Light green dot- **ORG**anic (n:3; minimum tillage)
- Yellow dot- **CON**ventional (n:2; Sod seeding – No-tillage and chemical weed control)

Forests

- Dark green dot- **FOR** (n:5; undisturbed soil)
(BF:beech forest; OF: oak forest; OF: chestnut forest)



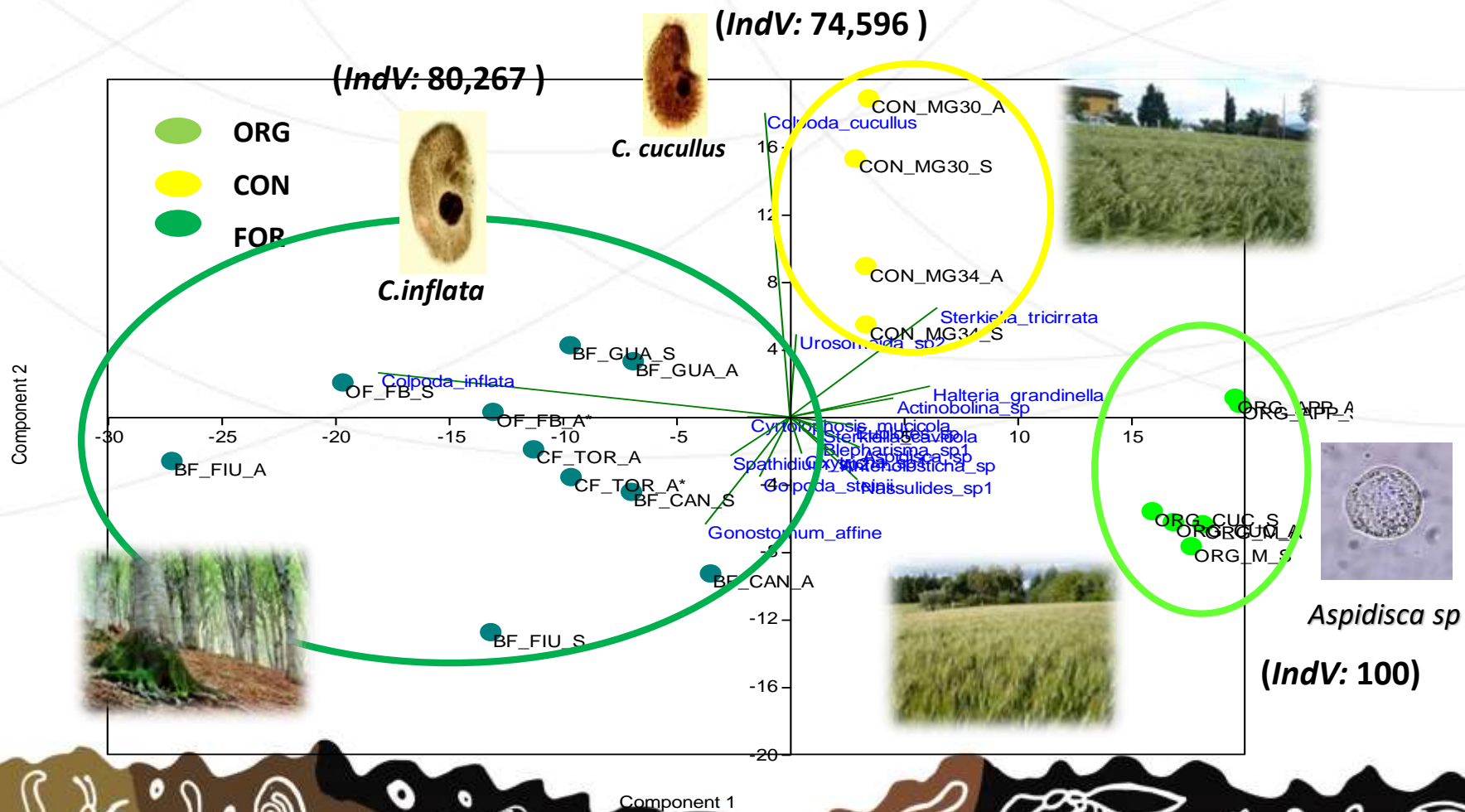
Time Schedule and Sampling

- Soil samples were collected in spring (S) and autumn (A) (2011)
- 10 soil samples (0–10 cm depth in an area of 100 m²) were randomly collected with a Edelman auger, mixed together to obtain a composite sample.



Different land use types host different ciliated protist communities and are characterized by different sets of Indicator Species (ISA, Dufrene & Legendre, 1997)

PCA for spatial taxonomic patterns of soil ciliates for square-root transformed species-abundance data for the 3 site types

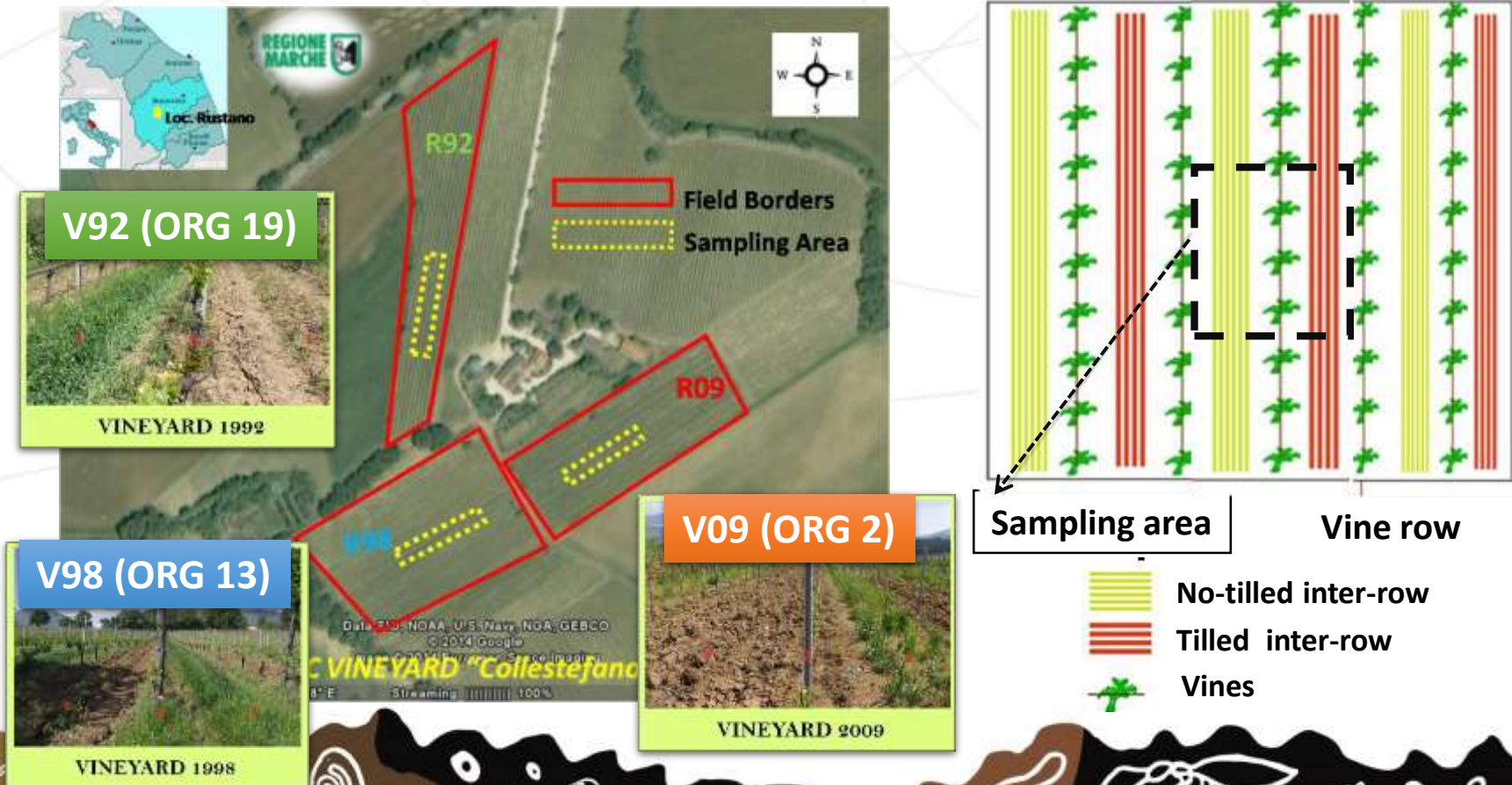


Axes 1 and 2 respectively accounted for 51.5 % and 15.9 % of the total variation present. One-way ANOSIM revealed significant global differences among the 3 sites (Global R= 0.85; p=0.0001) and between each pair of groups (**p<0.01).

Case studies across Italy. 2nd Ciliates in organic vineyards

Aim: to assess the long term effect of organic floor management on Soil Health by mean of ciliates communities analysis in 3 vineyards which were organically managed for **19 years (V92)**, **13 years (V98)** and **2 years (V09)** respectively

3 sampling in May, June and July 2011

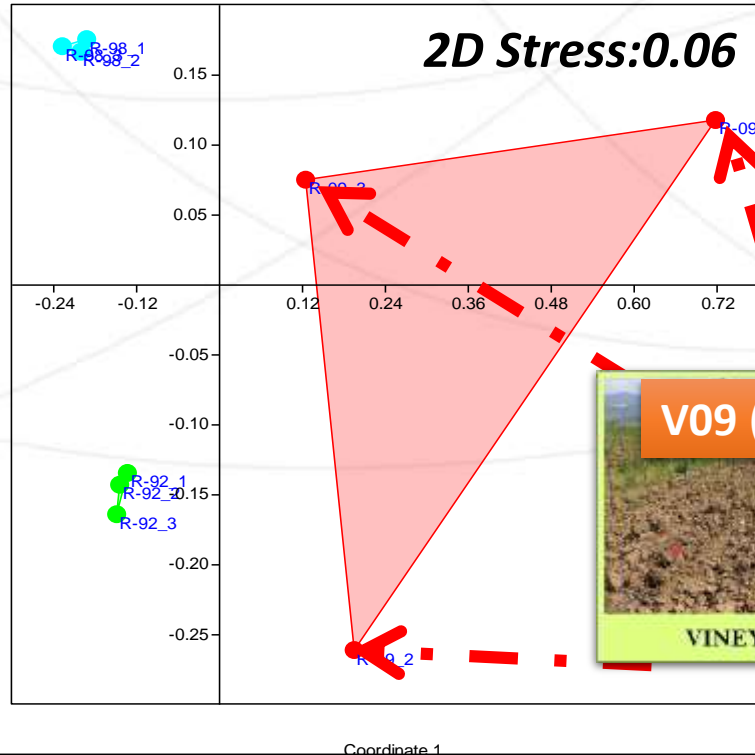
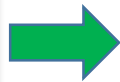


Vineyard floor management: alternate management of the tractor-rows on either side of the vine row with tillage and no-tillage annual cover crop species.

The “oldest” vineyards host more stable ciliate communities than the “youngest” vineyard

Non-metric Multidimensional scaling (nMDS)

for spatial taxonomic patterns of soil ciliates from log transformed species-abundance data on Euclidean Distances, for the 3 vineyard, **V92**, **V98** & **V09** - Convex Hull



•Overall, the ciliate communities show less fluctuations (> stable) in the “older” vineyards compared with the “younger” **V09** vineyard (**V92**>**V98**>**V09**).

•This effect may be due to the greater soil resilience, possible achieved during the long term organic management of the vineyards **V92** and **V98**

Case studies across Italy. 3rd Ciliates in industrial sites

Aims: i) to evaluate the potential of soil ciliate communities to discriminate between different levels of soil contamination/disturbances in four industrial areas (**Incinerator**; **SIN, Site of National Interest**; **Viscolube, Plant of regeneration of exhausted oils**; **Cement factory**) of Lombardia Region; ii) to assess relationships among ciliate communities and abiotic (environmental & contaminant parameters).

Four areas for a total of 30 sites were sampled.

Contaminated Site of National Interest



- As; Tl; Cu; Hg; Zn; Pb.....
- POP (PCB, PCDD, PCDF...)

Italcementi, Cement Factory



- Cu
- POP

Viscolube



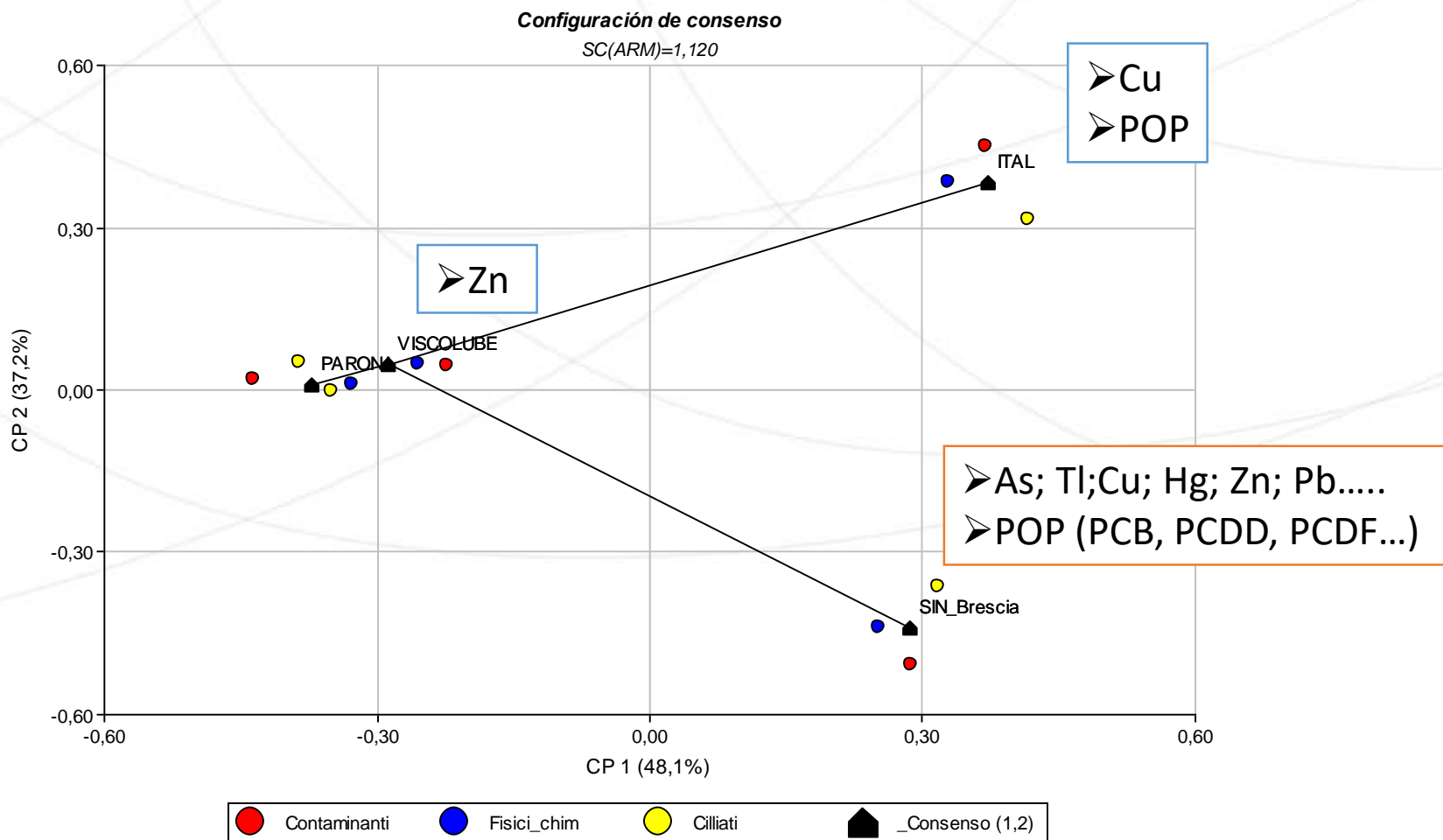
- Zn

Incinerator, Parona



What is the contribution of Chem-Physical and contaminant variables in shaping the community structures of ciliates at the 4 sites?

A Statistical Shape Analysis: Generalized Procrustes Analysis (GPA)



Total Consensus Matrix (groups): 0.917;

Group contribution values: Ciliates: 0.956; Chemical-Physical factors: 0.948;

Contaminants: 0.838.

To what extent and how do ciliate communities contribute to soil bioindication?

The 1st C. Study ORG vs CON vs FOR

- *discriminating between natural (FORest) and agroecosystems, and different management systems (ORGanic vs CONventional).*
- *providing “land-use” sets of Indicator Species*

The 2nd C. Study Ciliates in the vineyards

- *acting as proxy of soil resilience in agroecosystems (and thus, as indicators of sustainable land management).*

The 3rd C. Study Ciliates in industrial sites

- *discriminating between different levels of soil contamination in polluted sites.*
- *Showing significant relationships with abiotic (environmental and contaminant) factors.*

Altogether, these outcomes add new knowledge toward a more informed use of ciliates as bioindicators of soil health and broaden our understanding of how land use intensity, agricultural management and contamination levels can shape ciliate communities.

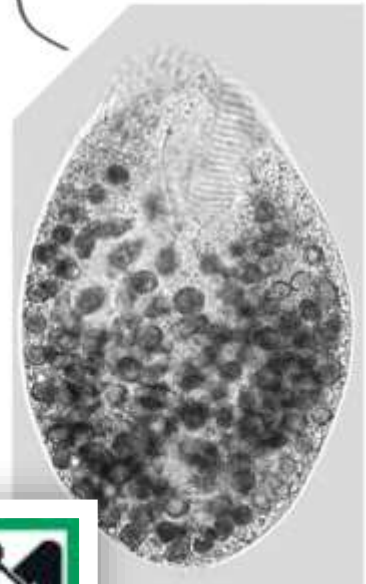
.... a contribution to alpha-taxonomy!

Soil ciliates: Novel species from Italy



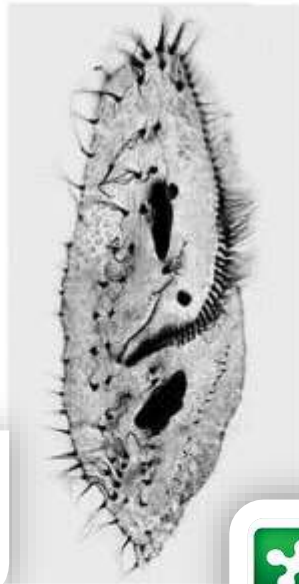
Kumar et al., 2014

Paraparentocirrus sibillinensis



Bharti et al., 2015

Gonostomum paronense



Pseudouroleptus plestiensis

Bharti et al., 2014



.....and many others!

.....biodiversity inventories are essential to obtain the **baseline knowledge_ which is the prerequisite to monitor ecosystem integrity_** and thus to detect and evaluate impacts of natural and/or anthropogenic disturbances.....(Cotterill et al., 2013)

Acknowledgments



.....for trusting ciliates!

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*and....to all farmers for having
facilitated and supported the
research on their land.*



Thank you!

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