

**Metagenomic insights
into the effect of frass fertilizer
application on rhizosphere
microbial community:
The case study of Italian turnip**

Speaker:
Dr. Giuseppe Di Cuia

*G. Di Cuia¹, M. D'Imperio¹, G. Mulè¹,
M. Gonnella¹, F. Gai², A. Parente¹, M.
Ferrara¹*

*¹ Institute of Sciences of Food Production,
CNR, via G. Amendola, 70126 Bari, Italy.*

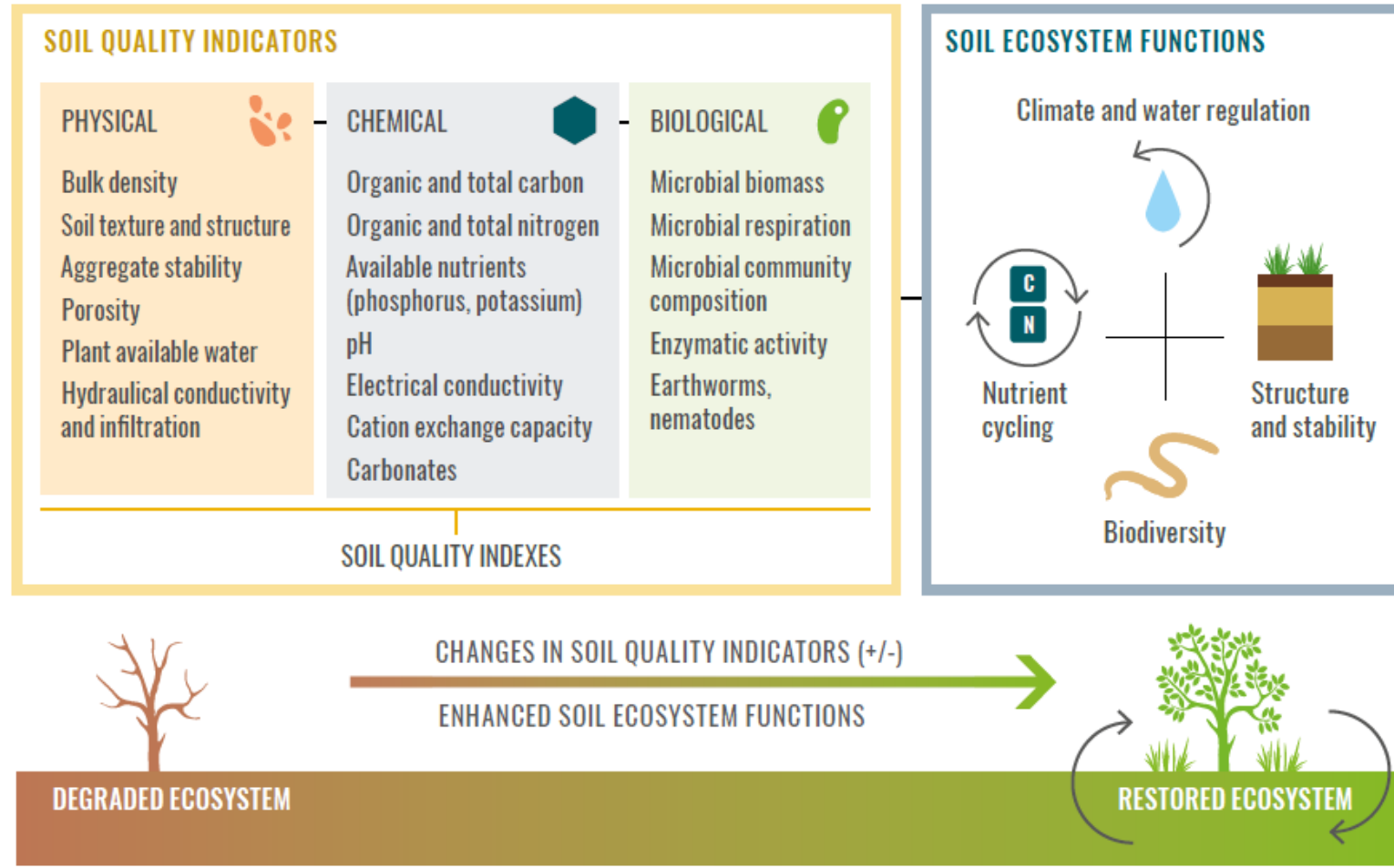
*² Institute of Sciences of Food Production,
CNR, Largo Paolo Braccini, 10095
Grugliasco, Italy.*



EAAP
European Federation
of Animal Science

Metagenomic insights into the effect of frass fertilizer application on rhizosphere microbial community: the case study of Italian turnip

The intensification of current agricultural practices and production systems is a primary cause imputable for the deterioration of soil quality, erosion, organic matter depletion and decline of soil biodiversity.



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Insect farming products

Larvae



Animal feed:

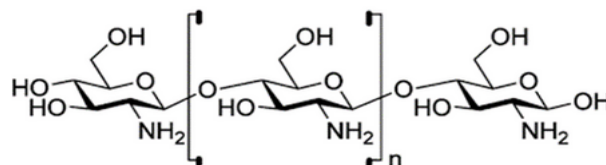
- *Fat: 15 – 35%*
- *Proteins: ~ 40%*
- *Fibers: 7%*
- *Ashes: 14- 28%*
- *Calcium: 5 - 8%*

Exuviae and adults



Industry:

- *Plant resistance inducers*
- *Biopolimers*
- *Food supplements*



Frass



Agriculture:

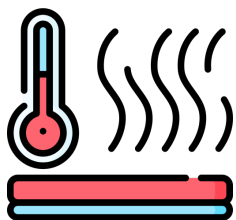
- *Soil amendment*
- *Cultivation substrate*

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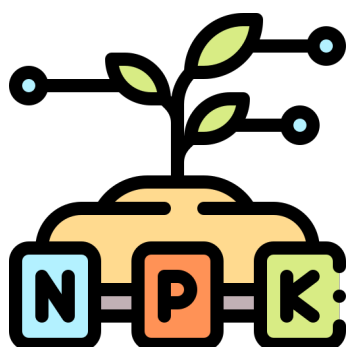


According to the **European Regulation 1925/2021**, amending certain Annexes to Regulation (EU) No 142/2011 as regards the requirements for placing on the market of insect products and the adaptation of a containment method, insect frass is intended:

"a mixture of excrements derived from farmed insects, the feeding substrate, parts of farmed insects, dead eggs and with a content of dead farmed insects of not more than 5% in volume and not more than 3% in weight."



70°C 1 h
(UE) 2021/1925



Regulation 1925/2021 setting EU standards for insect frass (i.e. heat treatment of 70 °C for at least 60 minutes, setting of microbiological standards) entered into force on 28 November 2021.

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Species: Italian turnip, local variety (cv. “Novantina”) in Apulian region (southern Italy)

Growing environment: open field

Main goal: evaluation of insect frass (*Tenebrio molitor* L.) as organic fertilizers in the sustainable agricultural process in the framework of the local agro-biodiversity enhancement, as partial or total substitutes of chemical fertilizers on the bio-morphology, quality and yield of local vegetable crops.



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Open field application

Figure 1. Distribution of fertilizers along the rows.

Quantity:

Rate of Nitrogen (N) fertilizer as reference: **130 kg/ha** according to
 Apulia Region Official Bulletin n. 38 31/03/2022

Treatments		
100% chemical	100% frass	100% organic
100% poultry manure	75% chem 25% frass	50% chem 50% frass
75% chem 25% poultry	50% chem 50% poultry	Control

Figure 2. Burying of fertilizers before transplanting.

Figure 3. Transplanting

Figure 4. Growing and harvesting

Fig. 1



Fig. 2



Fig. 3



Fig. 4



Experimental Design: randomized block with 3 replicates

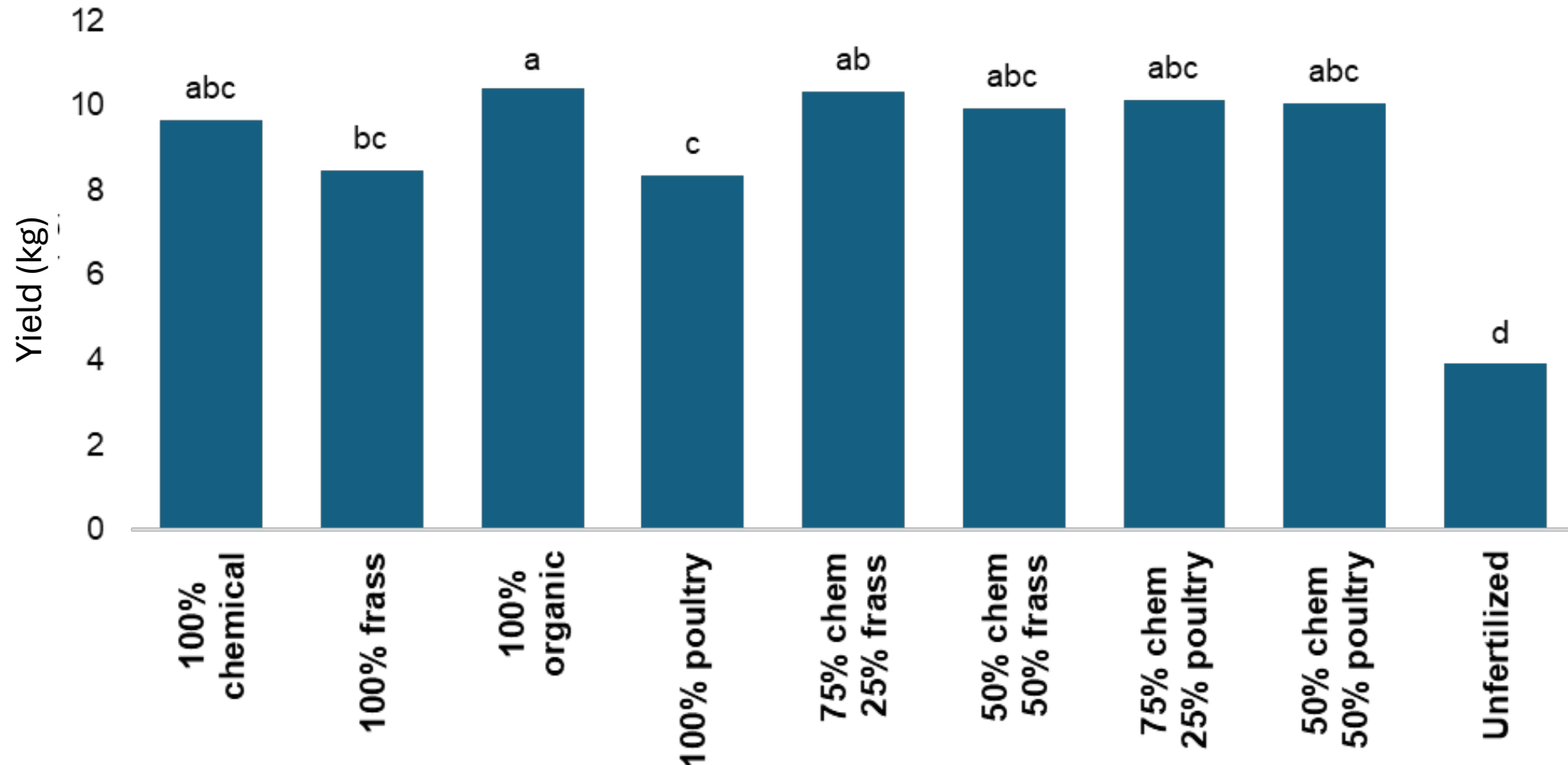
Measurements and analysis: crop performance, yield, nutritional quality (data not shown), and soil metagenomics

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Results - Open Field

Crop performance and production

Total yield (kg of fresh product)



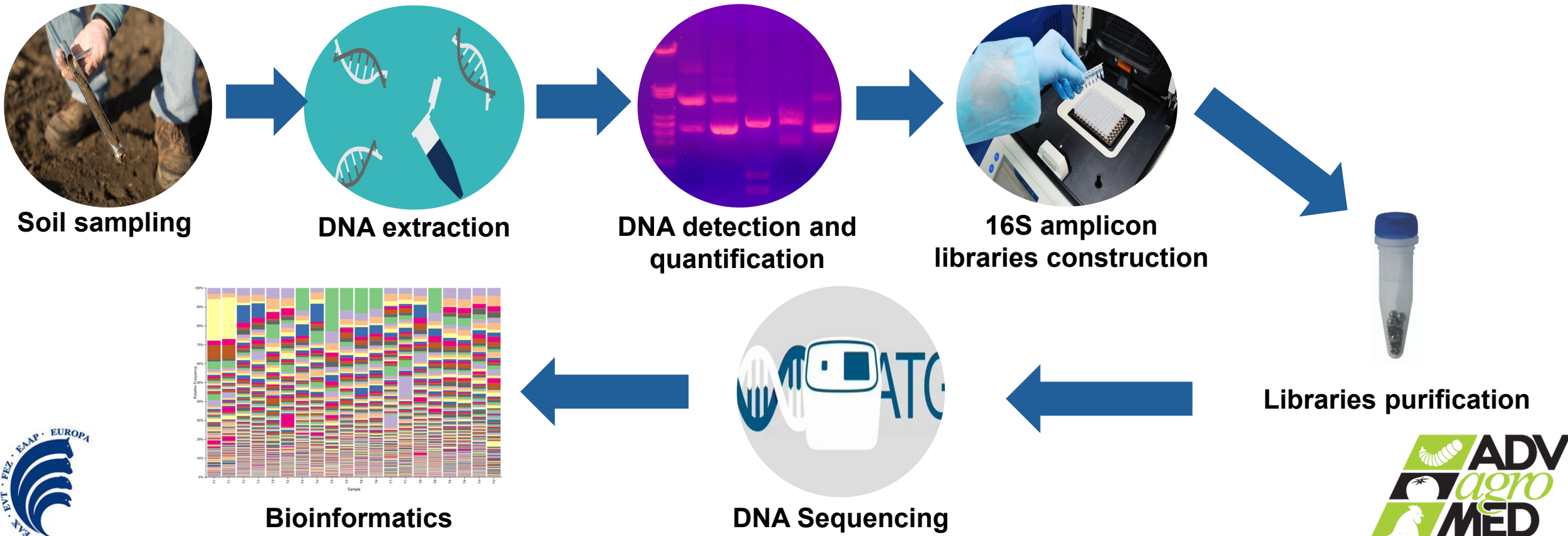
Data are expressed as mean of the treatments. The different letters indicate values significantly different, according to LSD test ($\alpha \leq 0.05$).

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Metagenomics

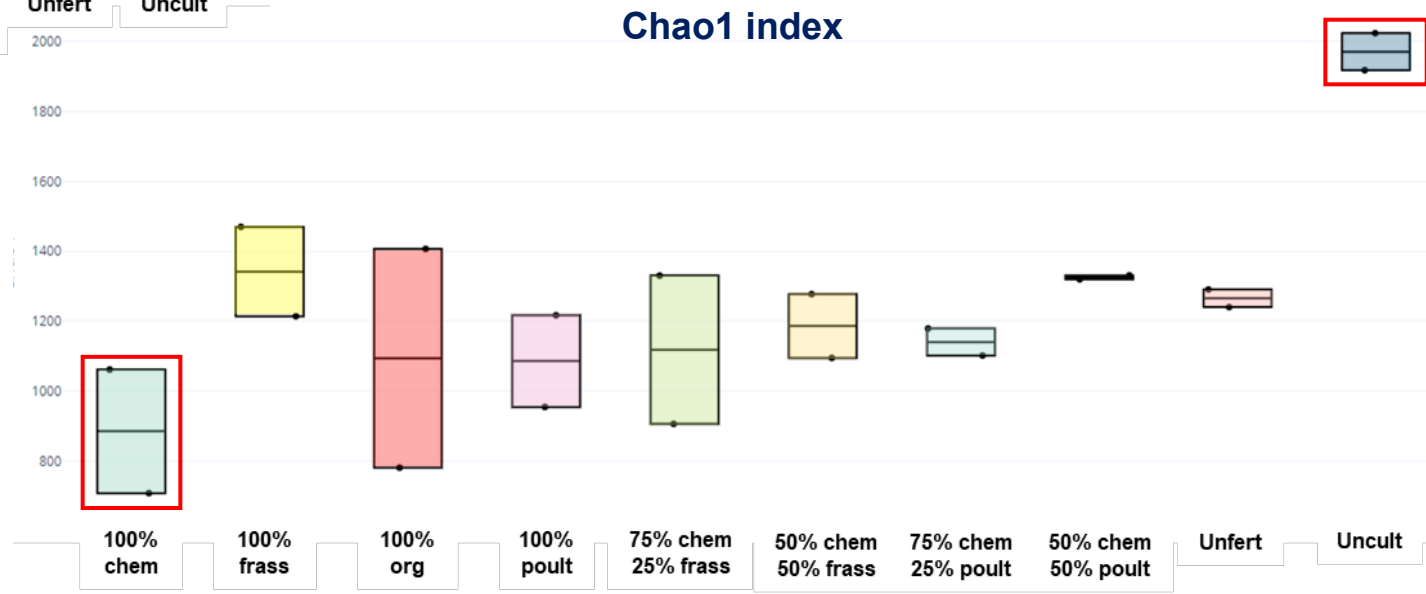
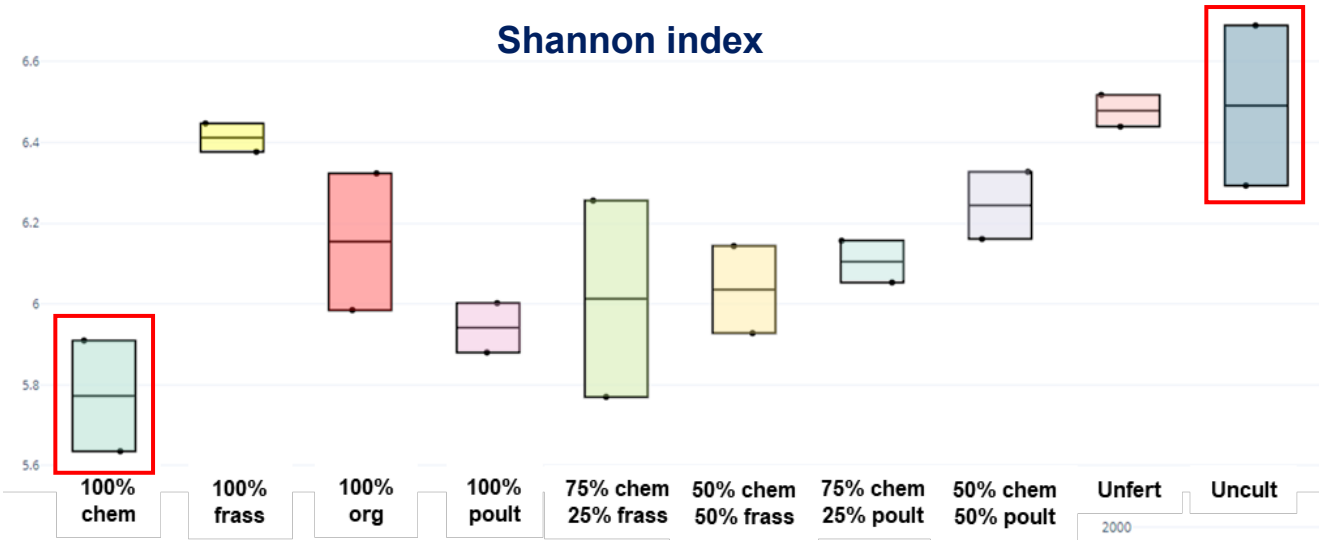
A modern approach that combines different techniques to study the genetic material of microorganisms recovered directly from samples (soil, water, clinic, human gut, etc.)

Workflow

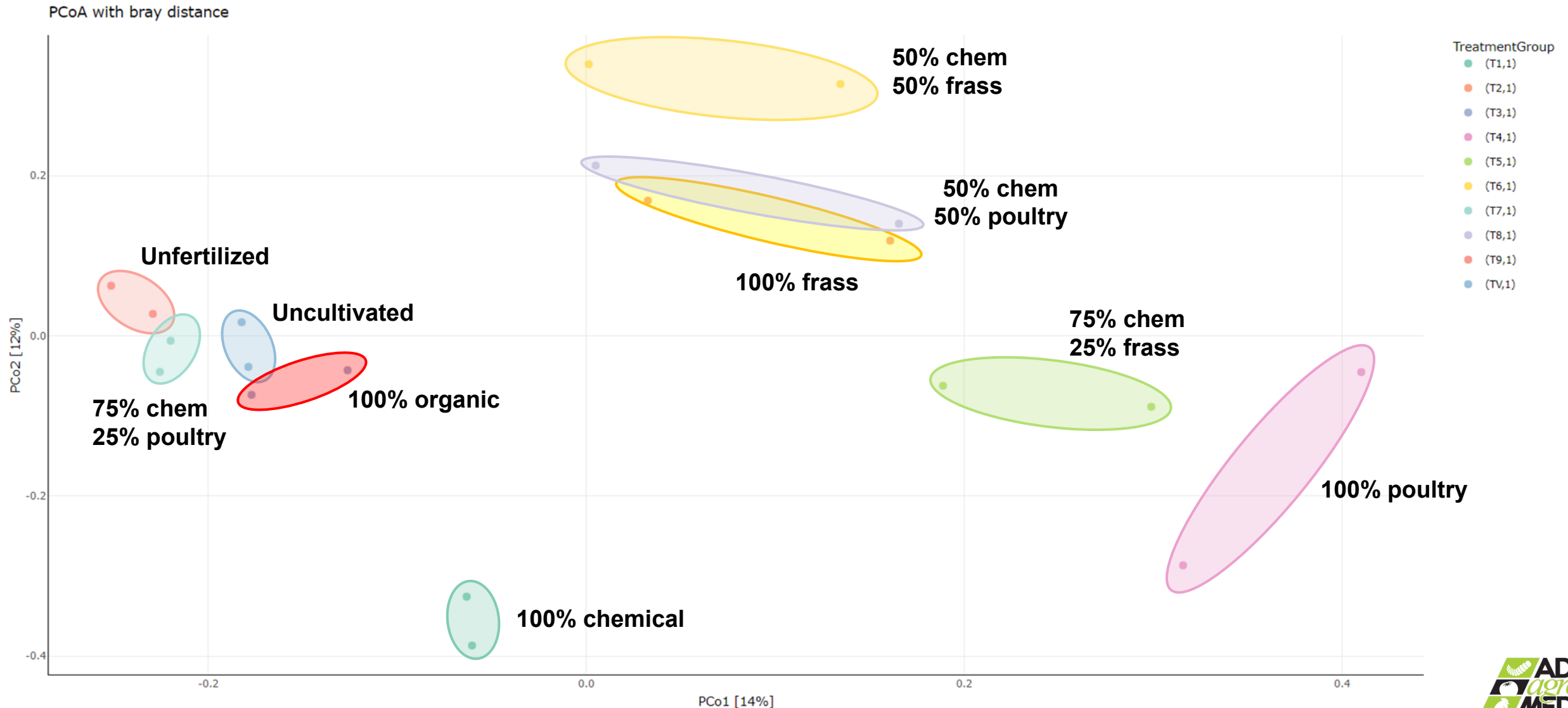


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Alfa diversity



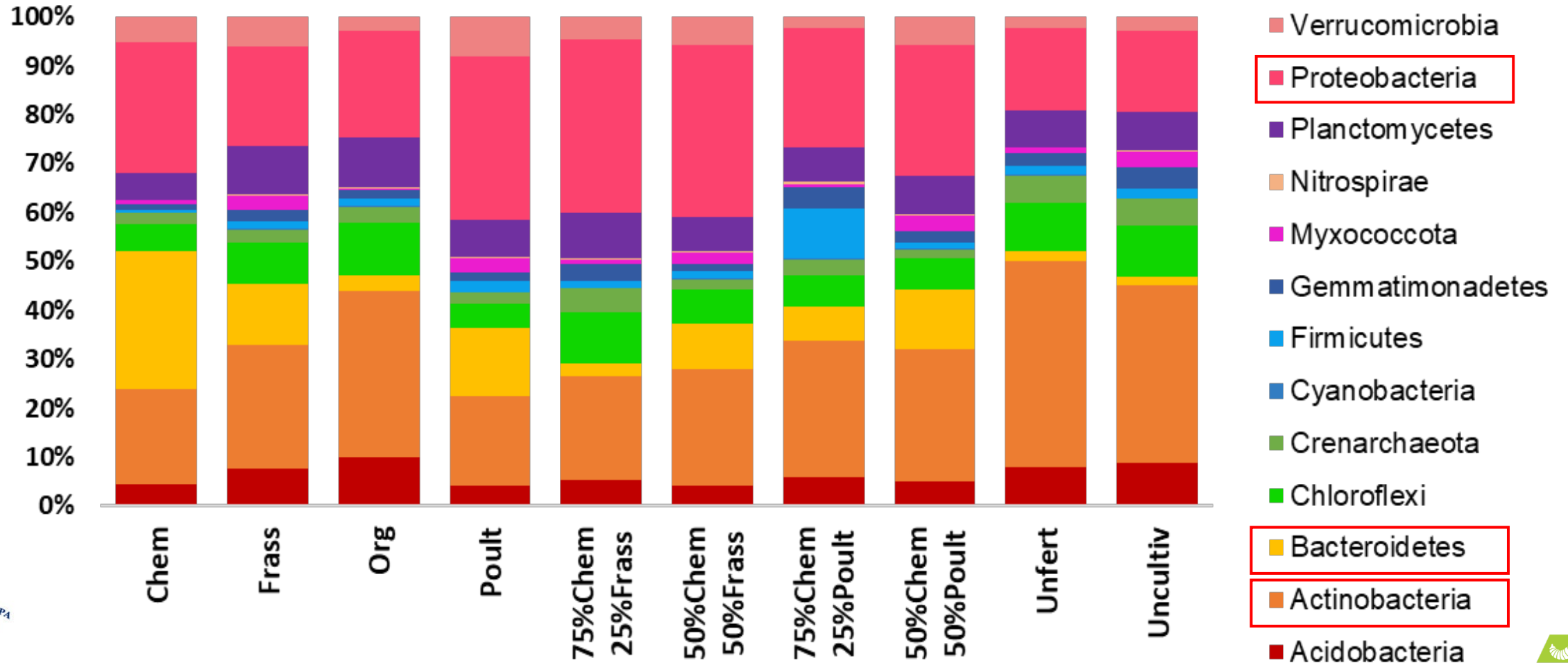
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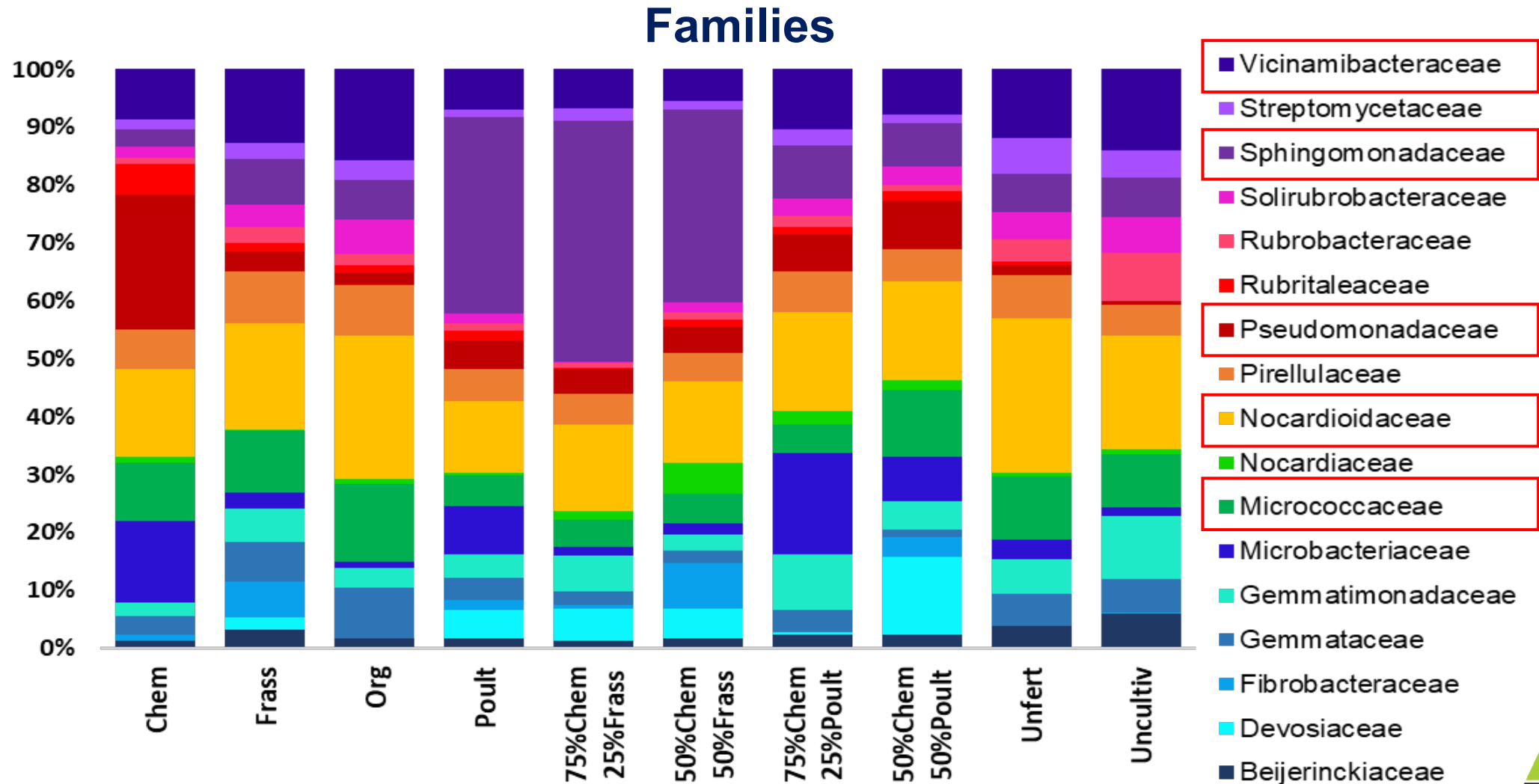
Results

Phyla



Data are expressed as abundance (%) of the 13 most represented bacterial phyla. Statistical analysis was performed using one-way ANOVA and Tukey – Kramer post-hoc test, corrected with Benjamini–Hochberg (FDR) test; ($p \geq 0.05$).

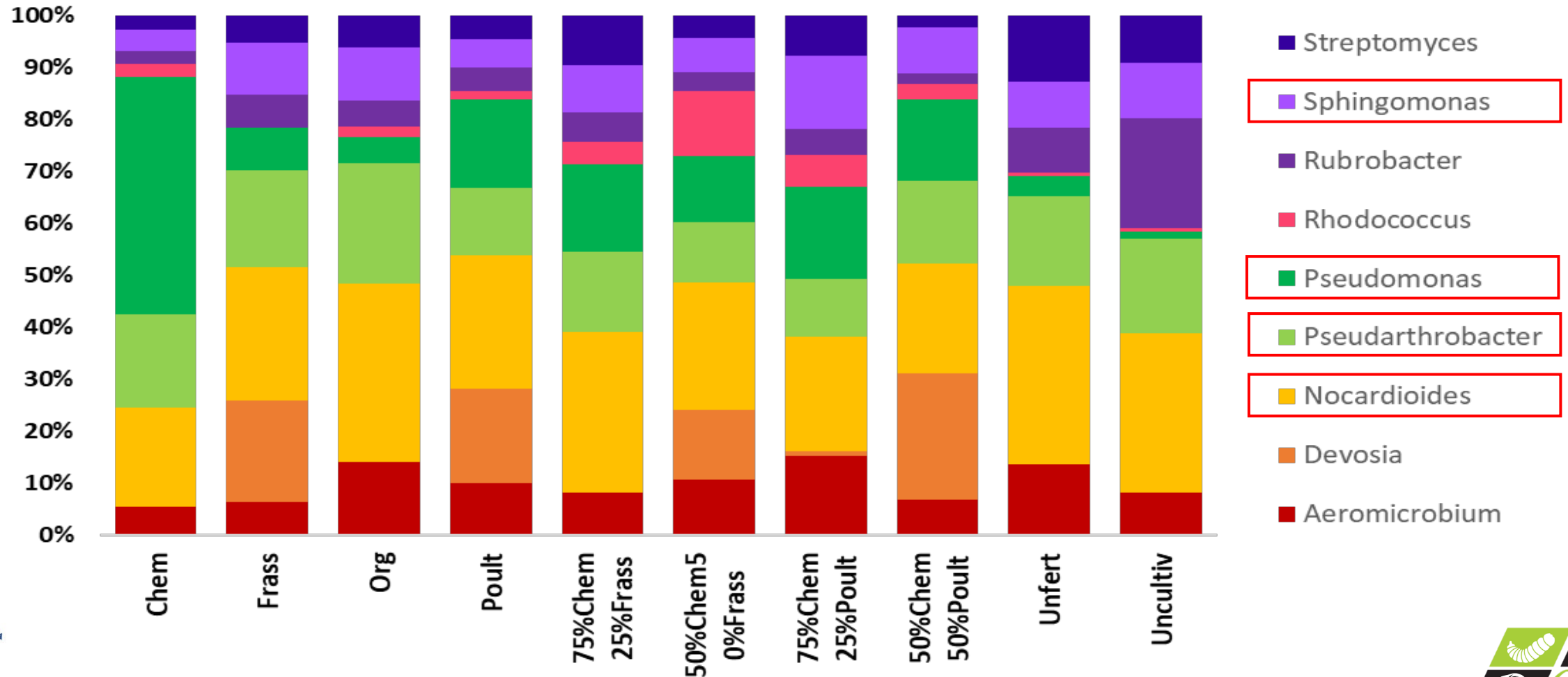
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Data are expressed as abundance (%) of the 17 most represented bacterial families. Statistical analysis was performed using one-way ANOVA and Tukey – Kramer post-hoc test, corrected with Benjamini–Hochberg (FDR) test; ($p \geq 0.05$).

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Genera



Data are expressed as abundance (%) of the 9 most represented bacterial genera. Statistical analysis was performed using one-way ANOVA and Tukey – Kramer post-hoc test, corrected with Benjamini–Hochberg (FDR) test; ($p \geq 0.05$).

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Conclusions

The insect frass fertilization:

- Can have a key role into circular economy and organic fertilization of agroecosystems;
- Can be a substitute of chemical fertilizers in plant fertilization;
- Can contrast soil biodiversity loss due to chemical fertilizers applications, increasing agroecosystems resilience;
- Can increase soil microbial community if applied yearly, although further studies should be conducted (ongoing activities of cumulative effects of fertilization on soil microbial communities).



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ACKNOWLEDGEMENTS

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Contacts:

Dr. Giuseppe Di Cuia

giuseppe.dicuia@ispa.cnr.it

Dr. Angelo Parente (WP4 Leader)

angelo.parente@ispa.cnr.it

Dr. Massimo Ferrara

massimo.ferrara@ispa.cnr.it



UNIVERSITÀ
DEGLI STUDI
DI TORINO

Department of Agricultural, Forest and
Food Sciences

Prof. Laura Gasco (COORDINATOR)

Prof. Ilaria Biasato



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