Soil as a Biological System

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I have summarized the in issues discussed in the mini review that I have published in 2020. Starting from the seven grand questions described by Selman Waksman in 1927 I have discussed the main known aspects of soil as a biological system, the main research approaches and one knowledge gap for each question.

Keywords: molecular technique; stable isotope probes; soil DNA; soil microbiota; soil proteins

1. Introduction

This contribution summarizes what reported by Nannipieri (2020) about what is known and what is unknown on soil as a biological system. The review by Nannipieri (2020) has given an historical perspective starting from the seven grand questions proposed by Selman Waksman in 1927 [1]:

- 1. What organisms are active under field conditions and in what ways?
- 2. What associative and antagonistic influences exist among soil microflora and fauna?
- 3. What relationships exist between soil organic matter (SOM) transformations and soil fertility?
- 4. What is the meaning and significance of energy balance in soil, in particular with reference to C and N?
- 5. How do cultivated plants influence soil transformations?
- 6. How can one modify soil populations and to what ends?
- 7. What interrelationships exist between physicochemical conditions in soil and microbial activities?

After 50 years Arthur D McLaren [2] briefly discussed some advances on the topics treated by the questions. Nowadays many issues about these questions are still poorly known despite more than 90 years of research and advances in soil biology and biochemistry. It is not possible to discuss here all these knowledge gaps due the vastness of the matter involving activity, abundance and composition of soil organisms, interactions among them and with different plants, effects of different soil properties, environmental conditions, management practices and polluting agents, etc,. I shall summarize what is known nowadays on soil as a biological system and then I shall mention one knowledge gap per each question as an example of future research.

2. The main known properties of soil as a biological system

The main known properties of soil as a biological system can be summarized in the following ways:

- The microbial diversity is huge as shown by the use of molecular techniques and also microbial biomass is huge but soil organisms only inhabit less than 1 % of the available space likely because most of the microenvironments are hostile for living organisms [3]. However, a well-defined core set of taxa is present in soil sampled from different part of the World [1][4]. There are hot spots (for example, soil surrounding roots and that surrounding a plant residue) where microbial activity and biomass are higher than in other parts of soil;
- Microbiota, such as bacteria and fungi play the most important role in soil processes, but organisms such as protozoa, nematodes, microarthropods, macroarthropods, Enchytraeidae and Earthworms, collectively named soil fauna, are also important. Viruses are also present but they have been less studied than microbiota and fauna [3];
- Processes, such as organic C and organic N mineralization, are carried out by different microbial species and a decrease in microbial diversity generally does not affect the rate of these processes. Redundancy does not occur for processes, such as nitrification, which are carried out by a limited number of species [3];
- Surface-reactive particles of soil can adsorb important biological molecules, such as DNA and protein, and protect them
 from degradation. The adsorbed extracellular DNA can be taken up by competent bacterial cells and the relative
 genome incorporated in the DNA of the cell. In this way, the adsorbed extracellular DNA acts as a reservoir of genetic
 information. Enzymes adsorbed by surface-reactive cells can remain active even under conditions unfavorable for
 microbial activity (3);

 Interactions between microorganisms, microorganisms and plants, between microorganisms and fauna can be positive, negative and neutral. These interactions are important because they can regulate several processes; for example, in the case of bacteria they can regulate symbiosis, virulence. Competence for transformation, conjugation, antibiotic production, motility, sporulation and biofilm formation.

The rhizosphere soil, that is the soil around the roots, is a hotspot particularly studied because microorganisms inhabiting this environment can have beneficial but also negative effects (pathogens) on plants. Usually soil properties have a more marked effects on microbial diversity of the rhizosphere soil than plant, despite plants can recruit microbial species from soil due to rhizodeposition, which includes root exudates, mucilage, root debris and whole detached cells [5].

3. The main research approaches used to study soil as a biological system

The main research approaches used to study soil as a biological system include:

- Use of simplified systems, as those with two soil components. For example, this type of studies has allowed
 understanding the mechanisms responsible for the interactions of organic molecules (such as DNA, proteins,
 pesticides, etc) and pure clay minerals. Of course caution is required to extrapolate results of these studies to the *in*situ conditions;
- Use of molecular techniques to determine microbial diversity and the presence of functional genes in soil. Generally
 these studies are based on the assumption that the detected microbial species carry out in soil the same processes
 detected *in vitro* and the presence of the functional gene means that the relative processes occur in the studied soil.
 However, these studies should also involve the use of approaches, such as the metatranscriptomic and proteomic,
 determining the expression of genes;
- The use of labelled compounds (taking N as an example, the use of ¹⁵N enriched compounds) with the holistic approach, where the system is portioned into pools (for example, organic N, plant N, exchangeable and fixed ammonium-M, nitrate-N, microbial biomass N, leached N, gaseous N) and fluxes (for example, ammonia volatilization, nitrification, denitrification, N mineralization, N immobilization, etc), has allowed to quantify the distribution of a nutrient (in my example N) in the soil-plant system;

The stable isotope probing (SIP) links directly the function to the identification of species using the substrate and combines the holistic approach with the use of molecular techniques. I have discussed the advantages as well as the drawback of this important technique [3].

4. Conclusion

Finally I have discussed the knowledge gaps for each questions, which can be summarized in the following way:

- The *in situ* determination of active microorganisms is still not possible;
- · Interactions between microorganisms and soil fauna are largely unknown;
- It is needed to have methods determining important organic pools, such as easily degradable and less degradable organic pools, for a better quantification of nutrients present in soil as organic forms;
- The carbon use efficiency (CUE, the ratio between the formed microbial biomass divided for the consumed substrate) as affected by microbial diversity is unknown and to fill this knowledge gap is important because CUE can give insights into the organic C stored in soil. This can be beneficial for counteracting the increase in the carbon dioxide concentration:
- It is important to analyze root exudates in the presence of soil;
- The manipulation of soil microbiome is important to have beneficial soil services, such as the stimulation of plant growth and the biological control of pathogen, However, microbial inoculations often fail for the insufficient number of active microorganism and for the use of ineffective carriers;
- It is still challenging to understand how microorganisms perform their activities at the microenvironment scale.

I have concluded my mini review by suggesting the combination of technology-driven research with hypothesis-driven research in the future studies and underlining the need of imaginative research for simulating the soil microenvironment at micro-scale.

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