

Soil Microbial Molecular Biomass

An indicator of the abundance of microorganisms

GenoSol Plateform – INRA of Dijon



WHY THE ABUNDANCE OF MICROORGANISMS?

The amount of **microbial biomass** is a determining factor in the biological quality of soils due to its role in the **regulation, transformation and storage of nutrients**. This amount of microbial biomass is shown to be a **sensitive, robust and early indicator of soil threats** (changes in agricultural practices, contaminations, change in organic status, etc...; Ranjard et al., 2006; Chaussod et al., 1996). The measurement of **Microbial Molecular Biomass** is a technique developed to estimate the abundance of soil microorganisms **just like the fumigation/extraction method** (Marstorp et al., 2000; Blagodatskaya et al., 2003; Widmer et al., 2006; Bouzaiane et al., 2007). Its **ease of implementation** allows it to be applied at moderate flow rate. For example, this Molecular Biomass has been measured on the 2.200 soils of the Network for Measurement of Soil Quality (RMQS) and a national mapping has therefore been established, as well as the first interpretation frame of reference associated with this measurement (Dequiedt et al., 2011). These efforts confirmed the robustness of the indicator in detecting modifications linked to changes in soil uses.

On 1 hectare (carbon eq.)

2.500 kg of bacteria
3.500 kg of microbial fungi
the equivalent of 6 to 10 cows

HOW ARE ANALYSES CONDUCTED AND INTERPRETED?

The measurement of Microbial Molecular Biomass **relates to the efficiency of DNA extraction from the soil**. The technique is based on quantification of extracted DNA directly from soil sample. The extraction and quantification protocols were developed and standardised by the GenoSol platform (Terrat et al., 2012; Plassart et al., 2012) and take approximately **1 day per sample** with the possibility to work on up to thirty samples at the same time:

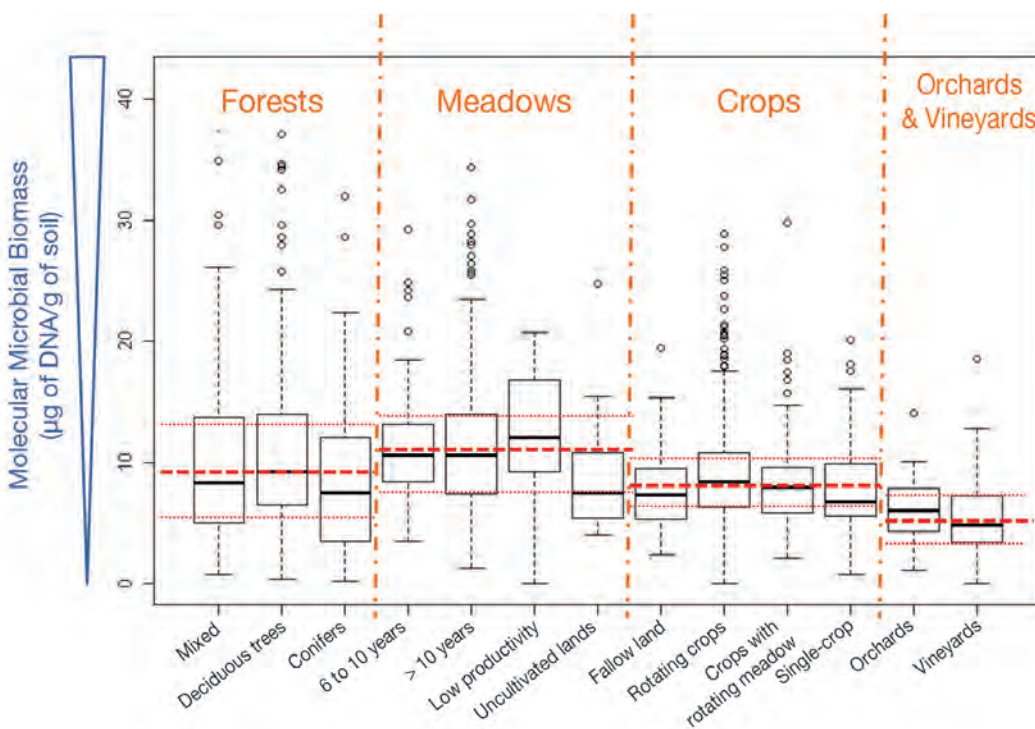
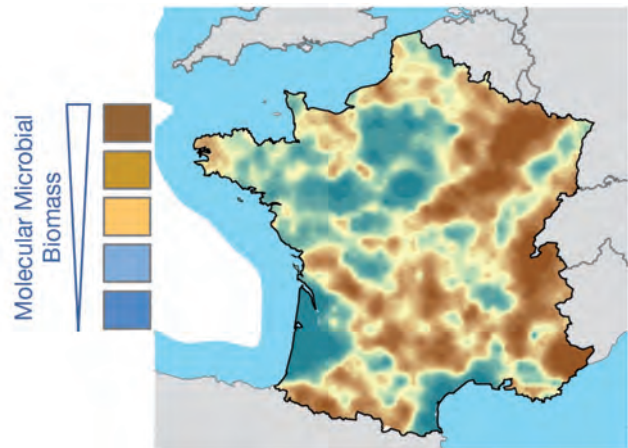
- **DNA extraction** from 1 g of dry soil (**standardised protocol which corresponds to the optimisation of normalised protocol ISO-TC 90-SC4-WG 4N119**),
- Deposit and **migration of DNA on agarose gel**,
- **Colouration** of DNA in the gel with ETHidium Bromide (BET),



Agarose gel after migration, colouration and revelation of DNA under ultraviolet light

- **Revelation** of DNA dyed in BET which fluoresces under ultraviolet light,
- **Quantification of DNA** by comparing it to a dyeset.

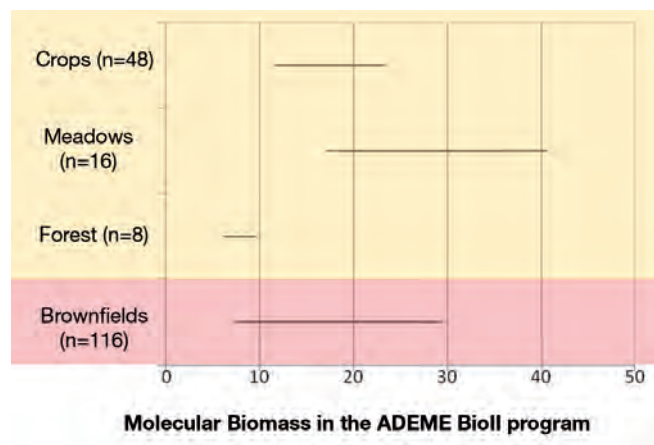
The **interpretation** of results from the indicator is conducted by **comparing values with a local referent** (test situation) or by **positioning within the national frame of reference** (MicroSol database[®]) using adapted mathematical tools (comparison of averages, mapping, decision trees, etc...).



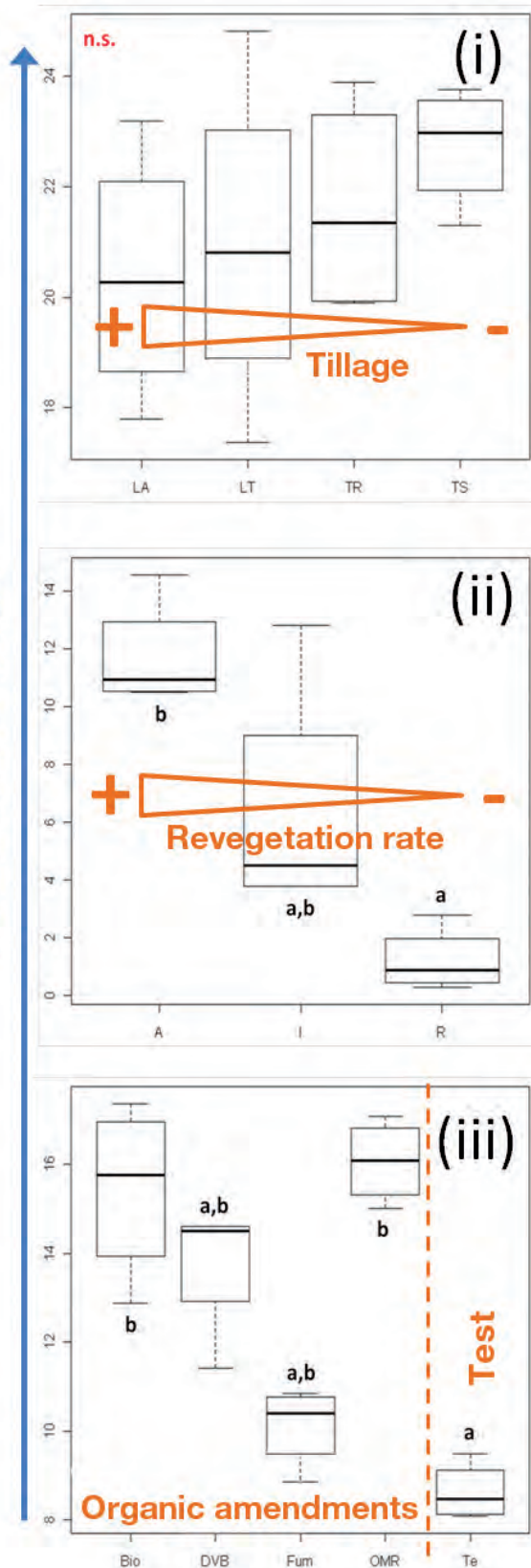
Interpretation frame of reference (MicroSol database[®]): spatial distribution of Molecular Biomass and variation range depending on the use of soils at the national level.

APPLICATION TO FIELD ISSUES

Studies at the national scale (RMQS; Dequiedt et al., 2011) showed **the predominance of local parameters** (occupation of soils, types of soils, practices, etc...) in comparison with global parameters (climate, location, geo-morphology, etc...) on the **variation range of microbial Molecular Biomass**.



Microbial Molecular Biomass



Results achieved on sites of the ADEME Bioll program: i) Thil site, ii) Saint-Etienne site and iii) Qualiagro site, show sensitivity of the indicator to:

- **tillage intensity**: the biological response reveals a tendency (not significant) to the **decrease of Microbial Molecular Biomass in the most tillage-intensive situations** (ploughing) while superficial tillage tends to increase this value and reduce heterogeneity between replicates (i),
- **recovery rate** of contaminated soil: biomass values significantly increase with **the percentage of revegetation** of the area under study by plants (ii),
- **soil organic status**: values are significantly **higher in amended situations** in regard with the test situation. The sensitivity of the indicator also allows for the discrimination of different amended situations depending on **the biochemical quality of organic inputs** and on their efficiency to stimulate microbial life (iii).



INTERESTS AND LIMITS OF THE MOLECULAR BIOMASS INDICATOR

Interest

- **Strong sensitivity to changes in practices**, crop routings (tillage, organic amendments, etc...).
- **Standardised (normalised) acquisition and interpretation techniques, tested and easy to implement at a low cost (about 50 €).**
- **Little specific material** to be used (centrifuge, pipettes, etc...); only the **soil crusher** is specific (MPBio FastPrep).
- **A compulsory step**, DNA extraction is required before all other molecular characterisations.
- **Availability of a frame of reference** (MicroSol database[®]) **to interpret results** (RMQS, network, INRA and ITA experimental sites, etc...).

Limits:

- **High sensitivity to the soil organic status and type which can mask other effects** (for instance, HAP or metal contamination).



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CONTACT

Plateforme GenoSol – INRA de Dijon - 17 rue de Sully - BP 86510 - 21065 Dijon Cedex France
http://www.dijon.inra.fr/plateforme_genosol

RANJARD Lionel (Dir. Scientifique) - Tel : +33 (0) 3 80 69 30 88 - lionel.ranjard@dijon.inra.fr

DEQUIEDT Samuel (Dir. Technique) - Tel : +33 (0) 3 80 69 33 83 - samuel.dequiedt@dijon.inra.fr