

Use of phospholipid fatty acid (PLFA) analysis to describe microbial communities in soil

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It is far more complex to count and identify microorganisms than it is nematodes, earthworms and other soil organisms. We need reliable ways to measure microbial populations that: i) are representative of who is there, ii) provide useful information on type (species, trophic groups) and their metabolic status, iii) are not subject to interference from soil particles.

A recent revolution in microbial ecology has led to development of methods that analyze components extracted directly from cellular tissues: phospholipids and nucleic acids (DNA, RNA) and provide much more detailed information than previous methods (e.g., counts, biomass).

1. PLFAs are associated with membranes of microorganisms (see Fig. 1).

- great diversity of fatty acids (differing in chain length, saturation, substitution, etc.) which differ among bacterial and fungal species (Fig. 2)
- originally used to identify individual bacterial species, so there is large database relating peaks to species and groups.

FIG. 1

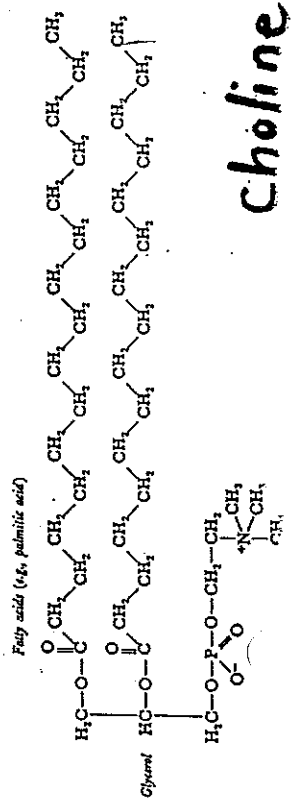
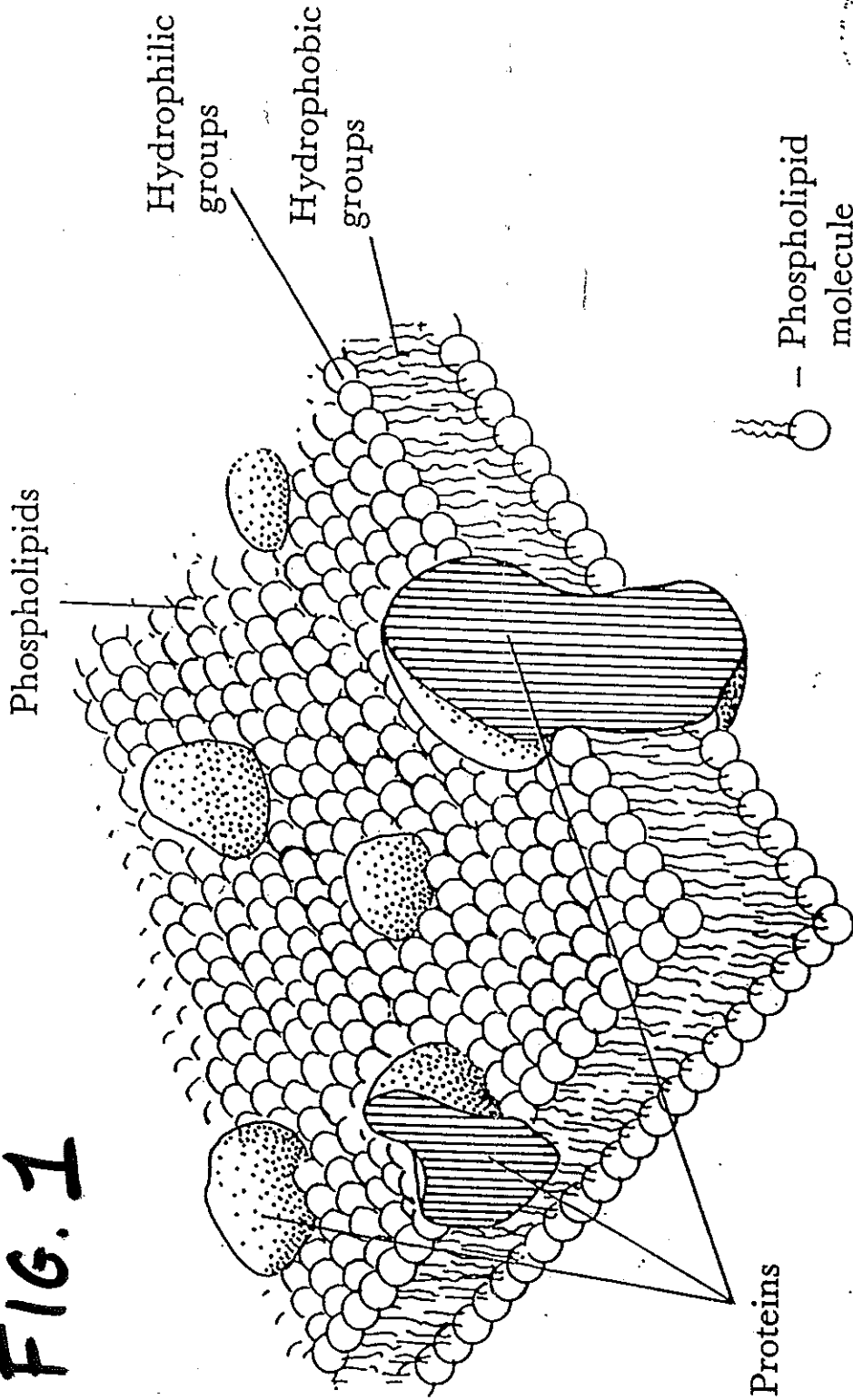


FIG. 2
EXAMPLE:

2. Can use PLFAs to measure:

- Total biomass
- Specific groups; e.g. bacteria vrs fungi, sulfate reducers, methane oxidizers.
- All peaks together can be used as unique "fingerprint" of a particular soil (Fig. 3).

3. Approach involves direct extraction of PLFAs from soil which are then cleaned up, separated, derivatized, and analyzed by gas chromatography (Fig. 4). Method requires a high level of technical expertise and equipment.

4. Results: In two farming systems studies, the importance of various environmental and management conditions on microbial communities was analyzed in replicated field trial comparisons of:

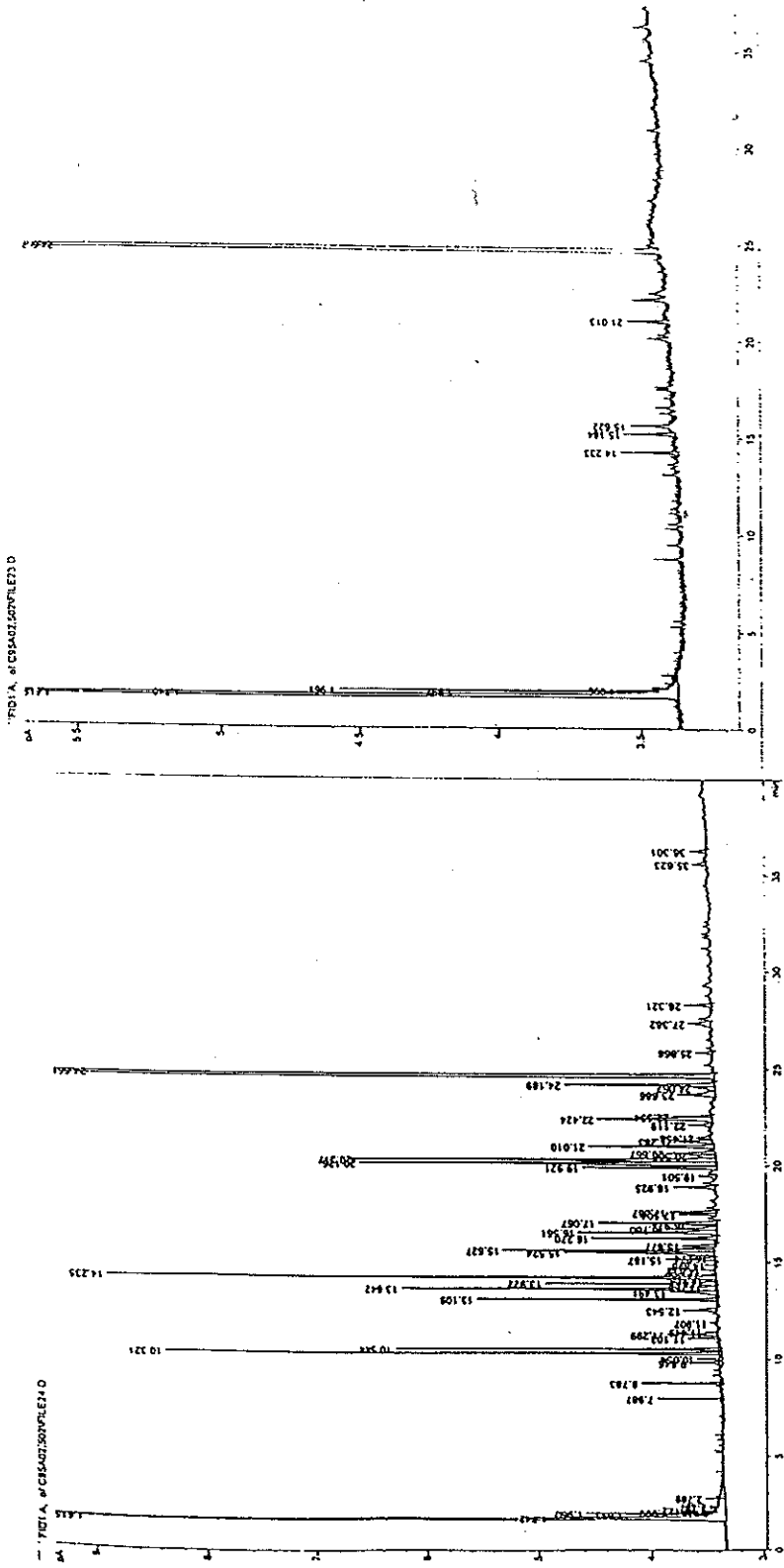
- *3 agronomic management systems for tomatoes (SAFS)*
- *8 straw management systems for rice (straw decomposition project)*

Findings:

a. Soils from the two field sites could be clearly distinguished from each other.

b. The relative importance of different factors in determining the character of a microbial community were:

crop or soil type > season > certain management practices (e.g., fertilization or organic matter incorporation) > management system (e.g., organic vs. conventional) > field variability.



SUB-SURFACE SOIL

SURFACE SOIL

Fig. 3. Example Gas Chromatographs of PLFAs Extracted from Soils.
(Each peak corresponds to a PLFA)

2

4

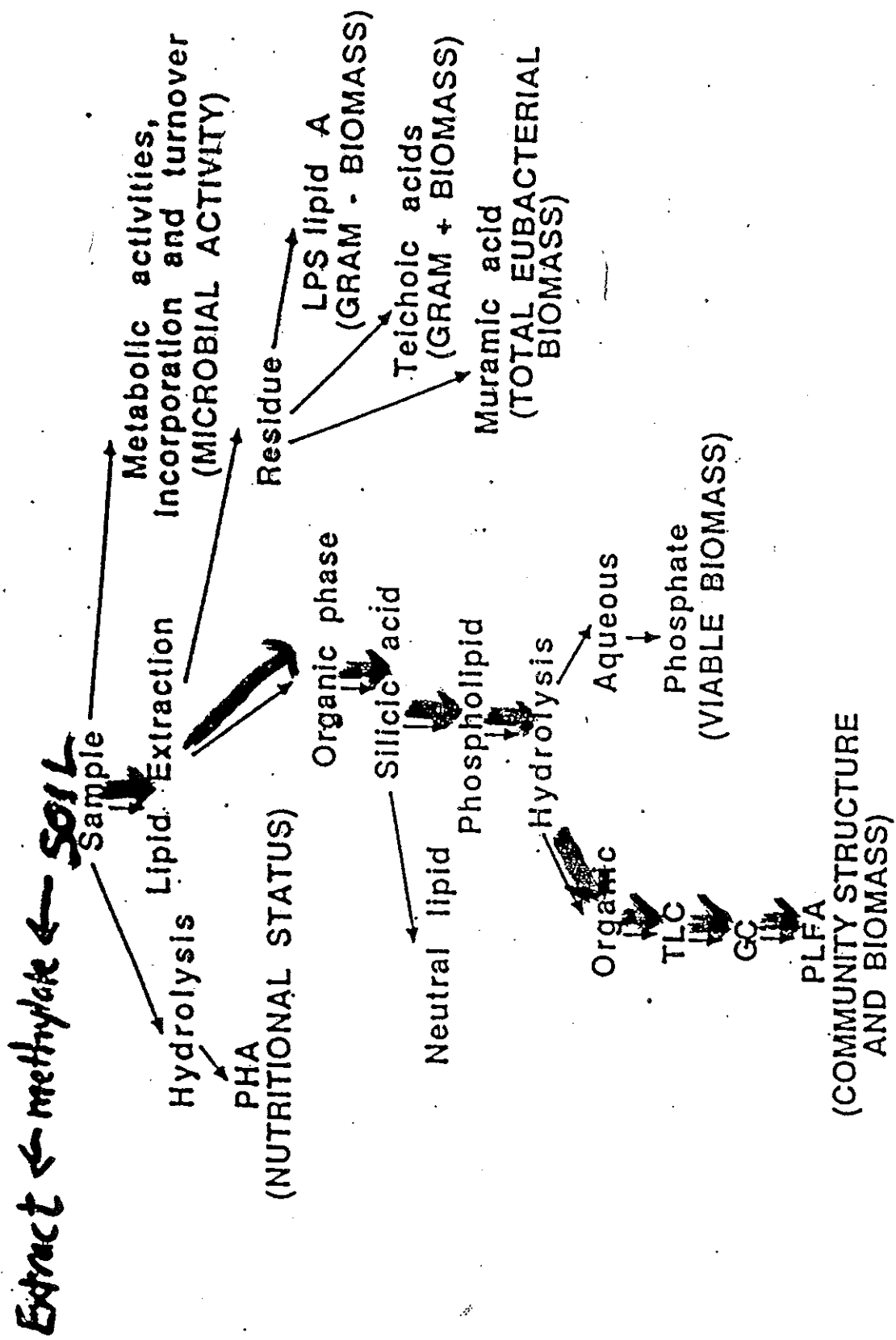


Figure 4. Flow diagram for the biochemical analysis of natural microbial communities using lipids (after White 1983).

c. With regard to specific groups of organisms, some of the differences included (preliminary results):

- greater relative importance of fungi in organic than conventional**
- greater relative importance of actinomycetes in low input than organic and conventional**
- greater relative importance of gram positive bacteria in conventional than other systems.**
- fungi were not very abundant in any system.**

5. Future directions: characterizing more soil types and crops, trying to relate unknown PLFAs that consistently increase or decrease under various conditions with specific groups of organisms.