Soil microbial community structure -PLFAs

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Abstract

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Protocol

Step 1.

Three grams of freeze-dried soil samples were used to extract the lipids by a single-phase $CHCl_3$: methanol: citrate buffer (15.2 mL at a 1:2:0.8 volume ratio).

Step 2.

The soil extracts were fractionated into neutral lipids, glycolipids, and polar lipids using a silica-bonded phase column (SPE-Si, Supelco, Poole, UK) with $CHCl_3$, acetone and methanol, respectively.

Step 3.

The recovered polar lipids were saponified and methylated to fatty acid methyl esters (FAME).

Step 4.

FAMES were quantified by a gas chromatograph (N6890, Agilent) and identified with an MIDI Sherlock Microbial Identification System (Version 4.5, MIDI, Inc., Newark, DE).

Step 5.

We divided all PLFAs into 6 microbial groups based on previously published PLFA biomarker data: Gram-positive (G+) bacteria (i14:0, i15:0, a15:0, i16:0, i17:0,a17:0), Gram-negative (G-) bacteria (16:1ω7c, 16:1ω9c, 17:1ω8c, 18:1ω5c, 18:1ω7c, cy17:0, cy19:0), actinomycetes (10Me16:0, 10Me17:0, 10Me18:0), saprophytic fungi (18:1ω9c and 18:2ω6c) and AMF (16:1ω5c).