Soil incubations

Subsamples of dried and sieved soil (30 g) reserved for incubations were placed in 70 mL polystyrene cups. Deionised water was added to each cup to reach 42% gravimetric moisture content. Each cup was placed into a 500 mL canning jar with a lid modified to hold a 1.5 cm, butyl rubber stopper for sampling. Soils were incubated in the dark at 25°C in a controlled environment incubator. Headspace samples (40 mL) were collected on 16 occasions over the course of 135 days (eight times in the first two weeks, and less frequently thereafter). Prior to headspace sampling jars were opened to allow equilibration with ambient air outdoors and then closed. Jars were then immediately placed in the incubator for periods ranging from 24 h during the early days of incubation to 90 h at the final sampling date, to allow approximately 10 000 µmol mol^{-1} CO_2 to accumulate. CO_2 production rate per hour was calculated based on the length of time after closing. Four jars without soil were used as blanks to account for time zero CO_2 concentrations and $\delta^{13}C$ values. Headspace samples were analysed for CO_2 concentration with a PICARRO G2201i isotopic CO_2/CH_4 analyser (Picarro Inc., Santa Clara, California, USA).

Measured CO_2 production rates over time were fitted to a two pool exponential decay model in which two pools of C, one labile and one intermediate, decay exponentially (Cheng & Dijkstra 2007, Wedin & Pastor 1993):

$$R_t = R_L * e^{-kL*t} + R_I * e^{-kI*t}$$

where R_t is the respiration rate at time t (µg C g $soil^{-1}$ day^{-1}), R_L is the initial rate of decomposition of the labile pool (at the start of the incubation; µg C g $soil^{-1}$ day^{-1}), k_L is the decay rate of C in the labile pool (day^{-1}), R_I is the initial rate of decomposition rate of C in the intermediate pool (µg C g $soil^{-1}$ day^{-1}), and k_I is the decay rate of C in the intermediate pool (day^{-1}). The size of the labile pool was defined as R_L/k_L and the size of the intermediate pool as R_I/k_I (60, 68). The size of the resistant pool was calculated as the difference between the total measured organic C and the sum of the estimated labile and intermediate pools. This same procedure of curve fitting and pool size estimation was also applied to the portion of CO_2 that was released from the originally present soil C (i.e not the added plant-derived C), which was determined via isotopic partitioning (see below).

To calculate the contribution of plant- and soil-derived C to respired CO_2 at each sampling point, isotopic partitioning was used as follows:

$$\text{Soil fraction}_{CO2} = (\delta^{13}C_{CO2} - \delta^{13}C_{UP\text{-}CO2})/(\delta^{13}C_P - \delta^{13}C_{UP\text{-}CO2})$$

where $\delta^{13}C_{CO2}$ is the C isotopic composition of CO_2 measured in each incubation jar after subtracting the mean blank value, $\delta^{13}C_{UP\text{-}CO2}$ is the mean $\delta^{13}C$ of CO_2 from unplanted controls, and $\delta^{13}C_P$ is the $\delta^{13}C$ of the plant shoots in each pot. The plant C fraction was defined as 1 minus the soil C fraction. These fractions were then applied to the measured CO_2 amounts in each jar to calculate plant- and soil-derived amounts. Based on these, we calculated total CO_2 released from plant- and soil-derived C during the full length of the incubation. To calculate the soil-derived C, we fitted a two pool decay model to the curve of soil-derived CO_2 (as described above) and used the obtained equation to calculate the cumulative amount of CO_2. The behaviour of the plant-

derived CO_2 showed more complex dynamics, so estimation of total plant-derived C release was done by interpolation of pairs of consecutive rates to obtain average rates for each period of time, which were then applied to the duration of each period and then added together to obtain the cumulative plant-derived CO_2.

Soil fractionation analysis

A 10 g soil subsample was shaken in deionised water and sieved (70 µm). The filtrate (containing material smaller than 70 µm) was further centrifuged at 3000 rpm to separate MAOM from the liquid containing dissolved organic matter. In the material that did not pass through the sieve (i.e. greater than 70 µm), the denser AggC fraction was separated from the lighter POM fraction via addition of sodium iodide (density 1.8 g cm^{-3}) and centrifugation at 3000 rpm. Carbon and nitrogen (N) content of the fractions were measured via combustion analysis.

In vitro fungal assessment

Under sterile conditions, 1/2 potato dextrose agar (PDA) plates were each inoculated with a single plug of fungal inoculum from a full strength PDA plate with four replicates per isolate. The perimeters of the colonies were delineated every two-to-three days, as well as those of the agar plugs at the start of the assay. Once growth reached the edge of the plate, the mycelium was harvested by removing the fungal colony from the plate into a funnel lined with nylon mesh fabric and pouring boiling water to drain the agar. The recovered mycelium was dried at 60°C and weighed.

Soil PLFA analysis

Soil was extracted with methanol-chloroform-phosphate buffer (2:1:0.8 in volume), and then fractionated into lipid types with a silica gel column. This was followed by mild alkaline methanolysis to produce phospholipid fatty acid (PLFA) methyl esters (PLFAMEs), which were then dried. The PLFAMEs were analysed for composition and quantification of concentration, relative to an internal standard, via gas chromatography. Gas chromatography was performed on an Agilent 7890A GC (Agilent Technologies, Wilmington, Delaware, USA) and PLFAME profiles were identified using the MIDI PLFAD1 calibration mix and the software SHERLOCK version 6.2 (MIDI Inc., Newark, Delaware, USA). The abundances of individual PLFAs were calculated as µg PLFA g-1 dry soil. The PLFAMEs used as markers followed Joergensen et al. (2022).