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Abstract: Organic matter is the soil component most affected by wildfires, in terms of both abundance and composition. Fire-induced soil organic matter (SOM) alteration mostly depends on the degree of soil heating, heating duration and oxygen availability. In this study we investigated the direct impact of fire on the topsoil (0-2.5 cm) of two forests, which had experienced wildfires of different severity, moderate in Tuscany, Central Italy, and extreme in Victoria, south-east Australia. The purpose of this study was to assess the fire-induced changes on amount and composition of SOM in the bulk soil and different density fractions. In particular, changes in the abundance and composition of non-cellulosic neutral sugars and lignin phenols were analysed.
In contrast to the moderately severe fire in Italy, the extremely severe fire in Australia resulted in a very substantial loss of SOM, about half of the original stock in the mineral soil. The latter fire reduced by half both lignin and sugar contents in the soil, with neither SOM component being selectively oxidised. The moderate fire, conversely, had a greater effect on lignin than sugars. Both fires resulted in significant organic matter redistribution between the density fractions of soil, which exhibit different properties and turnover time of SOM. Although characterised by different impacts by virtue of their different severity, both fires affected significantly SOM composition in the topsoil, which confirms the main role fire can play in the C local and global cycle.

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Abundance and composition of neutral sugars and lignin in two forest soils affected by wildfires of different severity

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1. Introduction

Fire is a major ecological factor, affecting more land surface that any other natural disturbance (Lavorel et al., 2007; Scott et al., 2014). Soils suffer from many direct and indirect consequences of fire, which virtually alters all physical, mineralogical, chemical and biological properties of soil, either temporarily or permanently (Bento-Gonçalves et al., 2012; Certini, 2005; Neary et al., 1999). The organic component of soil is the one most affected by fire, in terms of both content and composition (González-Pérez et al., 2004; Certini et al., 2011). The assessment of the overall effect

of fire on soil organic matter (SOM) is a complex task because burnt soils are generally a patchwork of areas affected to different degrees by burning. For example, fire effects on SOM depend on the degree and duration of heating, the availability of oxygen and the type of combustion (smouldering or flaming), which may lead to different SOM transformations (González-Pérez et al., 2004; Rumpel et al., 2007; Shakesby and Doerr, 2006). Most often fire causes a substantial reduction of the organic matter (OM) stock in the litter layer (Bento-Gonçalves et al., 2012; Certini et al., 2011; Nave et al., 2011). On the other hand, there are contrasting results in the literature about quantitative changes in the mineral soil. For instance, the meta-analyses by Johnson and Curtis (2001) and Nave et al. (2011), have contrasting outcomes, concluding that in the short-term the A horizon does experience, respectively, a significant fire-induced increase and decline in C content. However, divergent results from different studies can arise from methodological differences, related to sampling strategy, including soil depth or time elapsed since fire. Furthermore, the degree of fuel consumption as well as local conditions, as for example vegetation/climate type, may affect the results differently in different studies. In terms of SOM quality, the reactions that take place during combustion, *e.g.* dehydration, dehydrogenation, volatilisation of nitrogenous compounds, decarboxylation, demethylation, cyclisation and polycondensation (Hernàndez et al., 1997; Knicker, 2007), can substantially change the composition of the parent material. Charcoal formation is the most abundant outcome of wildfires and essentially represents the temperature and oxygen-depletion dependent transformation of the organic compounds into highly recalcitrant aromatic structures (Alexis et al., 2010). Once incorporated into the soil, charcoal may resist decomposition for centuries or even millennia, thereby sequestering carbon (Almendros et al., 2003; Egli et al., 2012; Kuhlbusch, 1995; Schmidt and Noack, 2000).

Carbohydrates are believed to be among the most fire-affected components of SOM (Certini, 2005; González-Pérez et al., 2004; Knicker et al., 2006). On this basis, Martín et al. (2009) proposed the ratio of carbohydrates to total SOM as an index of fire impact on SOM quality. Such an index allows, in principle, also differentiating between low- and high- soil burn severity wildfires. Lignin,

which, after carbohydrates, is the second most abundant component of plant residues in terrestrial ecosystems, is rather resistant to fire and is totally oxidised only at 400-450 °C (DeBano, 1991; Kuo et al., 2008). Nevertheless, despite the high heat resistance of its backbone (Knicker et al., 2008; Sharma et al., 2004), lignin is affected by fire at much lower temperatures (200-250 °C) in terms of phenols distribution (Certini et al., 2011; Kuo et al., 2008; Nocentini et al. 2010; Rumpel et al., 2007). Therefore, the composition of SOM regarding lignin and phenols has a potential as indicator of fire occurrence and severity. Quantifying post-fire lignin phenols and sugars content in soil might be a useful tool to evaluate the wider impact of fire on soil quality.

In the mineral soil, the effects of fire are usually confined to the top few cm because of the low thermal conductivity of both minerals and pore spaces (Certini, 2005; Knicker, 2007; Rovira et al., 2012; Wan et al., 2001). Despite this, in several studies reporting on fire impacts, soil has been sampled to rather substantial depths, which is likely to have resulted in the dilution of the investigated fire effects (Rovira et al., 2012). The choice of an appropriate sampling depth is indeed of great importance when examining the direct impact of a specific fire event on soil.

In this study, we investigated the top 2.5 cm of mineral soil of two forests located in Italy and Australia, which have been affected by recent wildfires of moderate and extreme severity, respectively. The purpose of using two contrasting sites in terms of forest type and experiencing fires of different severity was to assess changes to SOM resulting directly from the fire and explore their implications. We focused particularly on the non-cellulosic neutral sugars and lignin phenols in soil, the SOM distribution among different soil density fractions and the composition of such fractions. The changes SOM experienced at the two sites were compared in relation to fire severity, hypothesising that we would be able to detect general anticipated differences in fire-severity related alterations in SOM between the two sites.

2. Materials and Methods

78 *2.1 Study sites and soil sampling*

79 The study sites were Orentano (hereafter called OR), 30 km east of Pisa, Tuscany, Central Italy, and
80 Mount Gordon (hereafter called MG), near Marysville, in the Victoria State, south-east Australia
81 (Fig. 1; Table 1).
82 Orentano has an elevation of 20 m asl, mean annual precipitation 893 mm and mean annual
83 temperature 14.3 °C. The vegetation cover is a mixed forest of Downy oak (*Quercus pubescentis*
84 Willd) and Maritime pine (*Pinus pinaster* Aiton*)* with a rich understory of common fern (*Pteridium*
85 *aquilinum* L.*)* and *Rubus spp*. Soils formed on sand and stony lacustrine deposits and are classified
86 as Haplic Acrisols Skeletic Siltic according to the World Reference Base for Soil Resources (IUSS
87 Working Group WRB, 2006). An area of 3.3 ha in July 2011was involved by a wildfire of
88 moderate to high severity, based on the visual scale of litter and vegetation consumption proposed
89 by Chafer et al. (2004). Most of the tree stems were still standing after the fire and were partly or
90 totally scorched. The soil was covered entirely by charcoal and ash, with no or very little uncharred
91 litter remaining. Soil sampling was carried out three days after the fire, on both the burnt area
92 (coordinates WGS84: 43°47'22.82"N, 10°39'52.30"E) and an adjacent (50 meters away) unburnt
93 area having the same characteristics of the burnt one prior to fire occurrence, used as control.
94 At Mount Gordon the sampling area is 530 m asl in elevation, has a mean annual precipitation of
95 670 mm and mean annual temperature of 13 °C. The site was chosen because it represented an end-
96 member in terms of fire severity. The sadly famous 'Black Saturday', which affected also MG, in
97 early February 2009, burned some 450,000 ha of eucalypt forest causing the loss of 173 lives
98 (Royal Commission 2009). Average fire-line intensity is estimated to have exceeded 70,000–80,000
99 kW m^{-1}, which is amongst the highest ever reported in Australia (Royal Commission 2009). Such
100 an extreme intensity was promoted by particularly extreme weather conditions, such as wind speeds
101 up to 100 km h^{-1} and air temperatures even exceeding 45 °C. Fuel loads were very high, since the
102 forest had not experienced a major fire since 1939 (fuel load, including the litter, amounted to 25–
103 40 t ha^{-1}), and fuel moisture was very low (3–4%) because of two weeks of drought (McCaw et al.,

2009). The sampling site (37°31'56.30"S, 145°43'17.14"E) is a *Eucalyptus spp.* mixed forest 3 km SW of Marysville on the road to Narbethong. Fire removed all ground fuel, green vegetation and woody stems <10 mm in diameter; accordingly, fire severity was classified as extreme, based on the classification of Chafer et al. (2004). A long unburnt site - last burned by wildfire in 1939 - was selected as control, approximately 3 km NW of Narbethong (37°32'54.10"S, 145°37'37.30"E). This site is 8.5 km away from the burnt site and as much as possible similar to the latter in terms of all environmental conditions, soil included, and time elapsed since the previous fire. Soils here are formed on sandy Devonian sediments and are classified as Umbric Cambisols according to the World Reference Base for Soil Resources (IUSS Working Group WRB, 2006). Sampling was performed in April 2009, two months after the fire and following some light rainfall, but before the more intense precipitation of winter had caused significant ash removal via erosion.

At both study sites, OR and MG, the sampling involved four parallel 20 m transects, laid out 5 m apart, at 5 m intervals. Twenty mineral soil samples were taken at each site down to 2.5 cm, after carefully removing the ash, charcoal, or any litter layer by a brush. At each burnt area, ten samples of charcoal particles were collected in randomly chosen 40 x 40 cm areas.

2.2 Physico-chemical properties

Soil solution pH was measured potentiometrically using deionised water to soil ratio of 5:1, while particle size analysis was performed according to the hydrometer method. Total C and N contents and stable carbon isotopic composition of the fine earth (the less than 2 mm soil fraction) and charcoal were measured by Carlo Erba NA1500 elemental analyser coupled to an isotope ratio mass spectrometer (Micromass-Optima). The measured pH values, all below 6, demonstrated that the two soils were carbonate-free, hence, that the measured C was entirely in organic forms. δ^{13} C isotope abundance was reported in per mil (‰) relative to the Pee Dee Belemnite standard (PDB).

2.3 Density fractionation

Density fractionation was carried out on two replicates of fine earth samples per site, obtained combining equal aliquots of ten soil samples for each composite sample. The procedure was based on the method by Golchin et al. (1994), modified according to Sohi et al. (2001) and Cerli et al. (2009). This density fractionation allows different fractions, related to the spatial arrangement and interactions of organic compounds with minerals, to be separated (Cerli et al., 2012). The procedure requires a sodium polytungstate (NaPT) solution adjusted at a specific density to isolate a free light fraction (f-LF). By addition of the same solution to the precipitated material followed by ultrasonic dispersion to break down the aggregates, the occluded light fraction (o-LF) is separated from the heavy fraction (HF), the latter fraction mainly comprising minerals. The aim of the fractionation scheme was to separate the o-LF with little or no interaction with mineral phase. We performed preliminary trials in order to determine the most suitable density cut off and sonication energy for the fractionation. We first used the most common density and sonication intensity, i.e. 1.6 and 1.8 g cm^{-3} and 100 and 300 J ml^{-1}, respectively (Cerli et al., 2009; Cerli et al., 2012; Golchin et al., 1997; Kiem and Kögel-Knabner, 2003), the ultrasonic energy being calibrated calorimetrically according to Schmidt et al. (1999). Based on the criterion of the highest SOM concentration of the o-LF, *i.e.* the higher release of the o-LF without mineral "contamination" (Cerli et al., 2009), we selected 1.8 g cm^{-3} and 300 J ml^{-1} for soils from both study areas (data not shown). Hereafter the soil samples were fractionated according to the following procedure: 125 ml of NaPT solution (density 1.8 g cm^{-3}) were added to 25 g of soil, gently shaken and allowed to stand for one hour. After centrifugation at 6800 *g* for 20 min, the suspended material (f-LF <1.8 g cm^{-3}) was separated by careful removal of the supernatant and filtrated on a glass fibre filter (cut-off 0.7 µm) for washing away the any residual NaPT. The remaining non-floating material was ultrasonically dispersed in NaPT solution with the same density (1.8 g cm^{-3}, soil to solution ratio 1:5) by applying 300 J ml^{-1} and allowed to stand for one hour. After centrifugation at 6800 g for 20 min, the o-LF <1.8 g cm^{-3} was recovered as described above. The remaining HF >1.8 g cm^{-3}, where OM is strongly bound to the mineral fraction, as the other fractions, was washed from the salt by repeated addition of deionised water

156 and following centrifugation. All fractions were washed with deionised water until the electrical
157 conductivity of the supernatant was <50 μS cm^{-1}. They were then oven dried at 50° C, weighed and
158 analysed for their C and N content.
159
160 *2.4 Lignin phenols determination*
161 There is not yet any method able to reliably quantify the total lignin content in soil. In fact, even the
162 cupric oxide (CuO) oxidation, which is a widely used method for this purpose, does not succeed in
163 completely depolymerising lignin, thus it cannot be considered as a strictly quantitative method
164 (Kögel, 1986). Nevertheless, CuO oxidation is able to release phenolic monomers and dimers from
165 the end groups of the lignin macromolecules and, as such, it is a reliable indicator of lignin amount
166 and composition in soil (Kögel, 1986; Guggenberger et al., 1994, 1995; Kiem and Kögel-Knabner,
167 2003; Otto and Simpson, 2006; Spielvogel et al., 2007; Thevenot et al., 2010). On this basis, we
168 submitted the bulk soil and density fractions to CuO oxidation according to the modified method
169 proposed by Kögel and Bochter (1985) and Kögel-Knabner (1995). Briefly, 50-200 mg of sample
170 (the higher the C concentration, the lower the quantity of sample), 250 mg CuO, 50 mg of glucose,
171 50 mg of $Fe(NH_4)_2(SO_4)_2 \cdot 6H_2O$ and 15 ml 2 M NaOH were digested in a Teflon pot at 172 °C
172 under N_2 atmosphere for 2 h. Afterwards, ethyl vanillin was added as an internal standard to assess
173 the recovery of lignin products. The solution was adjusted to pH 1.8 – 2.2 and left overnight for
174 humic acid precipitation. Thereafter, the lignin-derived phenols were purified by elution through a
175 C_{18} column (International Sorbent Technology) and extracted from the column by adding 5 x 0.5 ml
176 (2.5 ml in total) ethyl acetate. After evaporating ethyl acetate under N_2 flow, the dried residue was
177 dissolved in pyridine containing phenyl acetic acid as internal quantification standard, and then
178 derivatised by adding BSTFA [N, O-bis(trimethylsilyl)trifluoro-acetamide]. The silylated lignin
179 monomers were separated using a HP 6890 gas chromatograph (GC) equipped with a SGE BPX-5
180 column (65 m × 0.32 mm internal diameter, 0.25 μm film thickness) and a flame ionization detector
181 (FID). The GC oven temperature program was: 100 °C (2 min) to 172 °C at 8° C/min, to 184 °C at

4 °C/min, to 300 °C at 10° C/min and 310 °C for 5 min. Helium was used as carrier gas and samples were injected in split mode (1:10). CuO oxidation products are composed of vanillyl (V)-units (vanillin, acetovanillone, vanillic acid), syringyl (S)-units (syringaldehyde, acetosyringone, syringic acid) and cinnamyl (C)-units (ferulic and p-coumaric acids). The sum of V-, S- and C-type phenols (VSC) was used to estimate the total amount of lignin. Ac/Al_V and Ac/Al_S are the mass ratios of acid to aldehyde units for vanillyl and syringyl units, respectively. S/V is the mass ratio of syringyl to vanillyl units and C/V is the mass ratio of cinnamyl to vanillyl units. These mass ratios are generally used to assess the state of degradation of lignin, since the Ac/Al ratios increase while S/V and C/V ratios decrease with increasing decomposition. Fire was shown to immediately produce a degraded lignin signature, for pure organic matter, partly mimicking the effect of microbial degradation (Nocentini et al., 2010; Rumpel et al., 2007).

2.5 Non-cellulosic neutral sugars determination

Sugar monomers were determined according to Amelung et al. (1996) as modified by Rumpel and Dignac (2006). The analysis was performed on both bulk soil and density fractions. Briefly, 200-500 mg of soil, depending on C concentration, were hydrolysed with 4 M trifluoroacetic acid (TFA) at 105 °C for 4 h. After the samples had cooled down, 0.5 ml of myoinositol (concentration 2 mg l^{-1}) was added as internal standard. Thereafter, the hydrolysed samples were purified by filtration over glass fibre membrane (cut-off 1.2 µm) and dried using a rotary evaporator (58 °C). Ethylenediaminetetraacetic acid (EDTA) was added, according to Eder et al. (2010), to keep iron in solution in a non-reactive form to avoid possible co-precipitation of dissolved organic carbon. Derivatisation of the sample was carried out in screw top test tubes. Aldoses were reduced to their corresponding alditols after addition of 1 ml $NaBH_4$ dissolved in dimethyl sulfoxide (DMSO). Acetylation was carried out by adding 2 ml acetic anhydride and 2 ml glacial acetic acid and using methylimidazole (2 ml) as catalyst. The reaction was stopped after 10 min by adding 7 ml ice-cold deionized water, to decompose acetic anhydride, generating acetic acid. The derivatised sugar

monomers were extracted by liquid–liquid extraction with 1 ml dichloromethane using a vortex mixer. After the phase separation, the darker organic lower phase was removed with a Pasteur pipette and transferred into a GC vial. The analyses were performed by a HP 6890 gas chromatograph (GC) equipped with a SGE BPX-70 column (60 m × 0.32 mm internal diameter, 0.25 μm film thickness) and a FID. Split injection (1:10) was used. The temperature program started at 200 °C and rose to 250 °C at 8 °C min^{-1}, with 15 min isothermal at 250 °C. Carrier gas was helium.

TFA digests the monosaccharides originated from plant-derived hemicelluloses and microbial products, while it is not able to digest crystalline cellulose (Guggenberger et al., 1994). Hence, hereafter, we will use the term sugars to indicate the non-cellulosic neutral polysaccharides. In particular, the sugar monomers detected by this method are: rhamnose, fucose, ribose, arabinose, xylose, mannose, galactose, glucose. Fructose is transformed into the same alditol as glucose during the reduction step (Rumpel and Dignac, 2006); however, the fructose content in soil is so low that it can be neglected (Amelung et al., 1996). The concentration of individual sugar monomers was calculated based upon the internal standard myoinositol.

According to Oades (1984), the proportion of microbially-derived sugars in relation to plant sugars was estimated by calculating the mass ratios of hexoses/pentoses sugars: (galactose + mannose)/(arabinose + xylose) = (GM/AX). Low (<0.5) and high (>2) GM/AX ratios are peculiar of carbohydrates predominantly derived from plants and microorganisms, respectively (Oades, 1984).

2.6 Solid-state ^{13}C NMR spectroscopy

We analysed by solid-state ^{13}C cross polarisation with magic angle spinning (CP/MAS) nuclear magnetic resonance (NMR) spectroscopy both the bulk soil and the density fractions, except the heavy one (HF), which was too poor in C to provide reliable information (Mastrolonardo et al., 2013). The specimens to undergo NMR analysis were obtained combining equal aliquots of all the

independent samples collected from each site. Before ^{13}C NMR analysis, all samples were treated with 2% hydrofluoric acid, as described by Skjemstad et al. (1994), to remove paramagnetic minerals, which strongly reduce the signal-to-noise ratio of the spectra. NMR spectra of so-treated samples were recorded on a Bruker AMX300-WB spectrometer, working at 300.13 MHz for proton and at 75.47 MHz for carbon-13, and equipped with a 4 mm CP/MAS probehead. The spectra were recorded with a contact time of 2 ms under proton decoupling conditions with a spinning rate of 8 kHz. The ^{1}H 90° pulse length was 3.4 μs, the spin-lock field 72 kHz, and the recycle delay 4 s. From 4,000 to 40,000 scans were acquired depending on the sample. The chemical shifts were referenced to tetramethylsilane (TMS) using adamantane as external standard.

The contribution of seven main C forms to total signal was determined by integration of corresponding chemical shift regions: 0 to 45 ppm (alkyl C), 45 to 110 ppm (O-alkyl C, subdivided in Methoxyl/N-alkyl C, 45 – 60 ppm; O-alkyl C, 60 - 90 ppm; Di-O-alkyl C, 90 – 110 ppm), 110 to 165 ppm (aryl C, subdivided in aromatic C-H and C-C, 110 – 140 ppm; O substituted C, 140 – 165 ppm), 165 to 185 ppm (carboxyl C); no signals arising from aldehydes or ketones were observed in the 185 to 220 ppm region.

2.7 Statistics

Data from burnt and unburnt soils were compared by two-tailed paired t-test at 95% confidence level (SigmaPlot 12.0). Where data did not show a normal distribution (Shapiro-Wilk test), Spearman rank correlation was used.

3. Results and Discussion

3.1 Carbon and nitrogen in the bulk soil

At OR, the concentrations of carbon and nitrogen, and consequently the C–to–N ratios, did not change significantly between the burnt and unburnt soil samples (Table 2). Only a slight enrichment, statistically not significant, of carbon and nitrogen concentrations in the burnt soil samples was found. Perhaps, the fire-induced loss in OM experienced by the A horizon, if any, was counterbalanced by the input of organic residues, including charcoal, from aboveground biomass and litter. Johnson and Curtis (2001), based on a meta-analysis carried out on eight independent studies, found a significant increase in soil C in the A horizon of forests burnt less than 10 years earlier. They attributed such an increase, at least partly, to an accumulation in the mineral soil of unburnt and charred residues. The incorporation of fresh charcoal into the mineral soil at OR is supported by the lower $\delta^{13}C$ value of the burnt soil compared to the unburnt one (Table 2), since charcoal had lower $\delta^{13}C$ than unburnt soil (-28.5 ‰ in charcoal versus -27.4 ‰ in unburnt soil). At MG, the mineral soil affected by the extremely severely wildfire exhibited substantially lower C and N concentrations compared to the unburnt one: 88 and 164 g C kg^{-1}, and 4.3 and 5.8 g N kg^{-1}, respectively (Table 2). Such fire-induced losses are out of range when compared to data reported by Nave et al. (2011) in their meta-analysis. Actually, apart from the extreme fire severity, we had focused our attention to a very thin uppermost soil layer (2.5 cm), thus avoiding diluting possible differences due to fire as in other works, where the sampled soil layer was thicker. At MG, carbon and N were differently affected by fire, resulting in a significant decrease of the C–to–N ratio, which might be explained by an accumulation of recalcitrant organic N-forms in the charred material (Almendros et al., 2003; González-Pérez et al., 2004; Mastrolonardo et al., 2014; Santin et al., 2008). As at OR, also at MG fire modified significantly the $\delta^{13}C$, the burnt soil showing a $\delta^{13}C$ of -27.5 ‰ and the unburnt one of -27.7 ‰ (Table 2). In the case of MG, the addition of charred materials from the aboveground biomass evidently was not sufficient to counterbalance the large loss of OM experienced by the mineral soil. Nevertheless, it should be noted that the surface ash layer removed during sampling to expose the mineral soil was considerable (1.7 cm on average). It contained substantial amounts of total C (62 g kg^{-1}), mainly in the form of charcoal (Santin et al.,

2012). Although the ash layer does usually not last for a long time on hillslopes, it is conceivable that over time, at least some of the C retained in the ash layer would become incorporated into the mineral soil. Its loss from soil surface would be due mainly to wind and water erosion (Bodì et al., 2014; Rumpel et al., 2009), thus, the fate of ash layer and the C balance of soil will probably be largely dependent upon climatic condition and rainfall regime.

3.2 Carbon and nitrogen in density fractions

The C and N concentrations of soil density fractions are given in Table 2. Light fractions, f- and o-LF, are generally assumed to comprise mainly plant debris and ancillary animal residues, charcoal and microorganisms colonising organic residues (Golchin et al., 1994; Wagai et al., 2009). Their main differences generally are in the size and location within the soil matrix: the f-LF is assumed to feature larger almost undecomposed organic materials, while the o-LF should comprise finer and more altered organic particles than the f-LF (Cerli et al., 2012; Golchin et al., 1994).

The C–to–N ratio of density fractions of the unburnt soil at both OR and MG supports the expectation that SOM ranges between the less degraded light fractions, having a higher C/N, to the comparably more decomposed heavy fraction showing a lower C/N. These considerations are consistent with the higher $^{13}\delta$ C values for HF compared to light fractions, which support the hypothesis of a higher decomposition of the SOM contained in this fraction (Roscoe et al., 2001). In spite of low C and N concentrations, but due to its large relative mass (Table 2), HF stores one third of total SOC and almost one half of total soil N (Fig 2). The high relatively N content of HF could be partly due to inorganic N or preferential adsorption of N-rich organic compounds onto inorganic surfaces of soil minerals (Golchin et al., 1997).

Apart from obtaining functionally more homogeneous fractions with a narrower range of properties compared to bulk soil, density fractionation allows free SOM and SOM associated to minerals and physically protected into aggregates to be distinguished. As a consequence, comparing density fractions from burnt and unburnt soils, one could infer if and how the aggregates are able to protect

the OM against heating or if fire caused a partial collapse of the soil structure. As quite recently reviewed by Mataix-Solera et al. (2011), the response of soil aggregates to heating by fire can be highly variable. Fire can oxidise organic binding agents in aggregates thereby causing their breakdown. Alternatively, a fast vaporisation of the water included in aggregates can lead to their destruction in a similar way as slacking does (Albalasmeh et al., 2013). However, under certain conditions, *i.e.* for wettable soils with SOM as main binding agent subject to low severity fires, aggregate stability may improve as a consequence of enhanced soil water repellency (Mataix-Solera and Doerr, 2004).

At OR, fire apparently caused an increase of C and N stock and concentration of f-LF (Table 2; Fig. 2). This increase is probably due to the incorporation of some charred residues into the top centimetres of soil and the charring of OM in the mineral soil. The C stock in the o-LF was slightly lower in the burnt soil compared to the unburnt one, although the C and N concentrations were actually higher (Fig 2; Table 2). This leads us to hypothesise that fire could have caused a partial disruption of aggregates that, as a consequence, released some high density mineral particles with no or little interaction with OM, which may become part of the HF, and some OM free particles, which may become part of the f-LF (Fig. 3). The so released OM would also be more exposed to decomposition because easier accessible by microorganisms. The same phenomenon, a partial aggregates disruption, also occurred at MG, here resulting in an increasing in terms of relative weight of the HF (Table 2). However, at MG the most affected fraction was the f-LF, that experienced a depletion in the C and N content and contribution (Table 2; Fig. 2).

3.3 Soil polysaccharides content

At OR, neutral sugars in the burnt and unburnt soils amounted to 7.4 and 8.2 g kg^{-1}, respectively, which correspond to 76 and 97 g kg^{-1} of TOC (Table 3) and are in the range of data reported by other authors for forest soils (e.g. Guggenberger et al., 1994; Rumpel and Dignac, 2006). The apparent fire-induced decrease in neutral sugars was statistically significant if the latter were related

to TOC, but not in absolute terms. The galactose+mannose to arabinose+xylose ratio (hereafter called GM/AX) before fire occurrence was >2, indicating that sugars were synthetized mainly by microbial population (Guggenberger and Zech, 1994; Oades, 1984). Fire did not change that ratio, so indicating that neither plant- nor microbial-derived sugars were preferentially oxidised.

The TOC normalised sugar content slightly decreased due to fire in the f-LF, while in the other fractions, o-LF and HF, it increased (Table 3), suggesting that sugars associated to minerals were relatively preserved compared to the other OM compounds. In fact, it is commonly reported that sugars of microbial origins contribute to the formation and stabilisation of soil aggregates, hence benefiting from physical protection. Moreover, they seem to be stabilised by interaction with the mineral phase (Kiem and Kögel-Knabner, 2003; Martín et al., 2009; Rumpel et al., 2010). Rather surprisingly, the absolute content of sugars associated to HF increased after fire.

At MG, the absolute concentration of non-cellulosic neutral polysaccharides in the bulk soil was substantially lower in burnt than unburned samples (9 and 21 g kg^{-1} respectively) (Table 3). This apparent decrease, however, was not significant if sugar content was normalised to TOC, 105 vs. 126 g kg^{-1} of TOC for the burnt and unburnt soil, respectively. This suggests that the polysaccharides present in the mineral soil were not preferentially oxidised by fire, despite being part of the thermally labile SOM pool, and totally oxidised at relatively low temperatures (ca. 300 °C; De la Rosa et al., 2008). Such a hypothesis is not in accordance with our results at MG. However, it must be pointed out that the method we used was not able to also detect cellulose C, which may have a different behaviour towards fire compared to non-cellulosic sugars. Like at OR, the GM/AX ratio at MG indicates that carbohydrates were mainly originated from microorganisms and the ratio did not change after fire occurrence. As expected, sugars, most of which were stored in the f-LF, were apparently greatly lost because of the fire. Nevertheless, also the o-LF and HF were substantially affected by fire, both in absolute terms (normalised to mass proportion of density fractions) and relative to OC (Table 3).

3.4 Soil lignin content

At OR, the yield of phenolic CuO oxidation products in the burnt soil was significantly lower than in the unburnt one, both in absolute terms and relative to OC (Table 4). This suggests that lignin was somehow preferentially oxidised, despite its assumed moderate recalcitrance to heating (Knicker et al., 2005). Fire apparently left the VSC content of SOC associated to HF almost unaltered, while it affected the VSC content of the light fractions, particularly o-LF, both in absolute value and relative to OC. Therefore, occlusion into aggregates does not seem to guarantee lignin protection. Indeed, pyrolytic degradation of lignin polymers in aggregates could be favoured by inorganic catalysts, such as acidic clay minerals (Ohta and Venkatesan, 1992).

At MG, the absolute yield of phenolic CuO oxidation monomers was half in the burnt bulk soil compared to the unburnt one (Table 4), but such a difference disappeared if values were related to TOC. As in the case of sugars, lignin monomers did not appear to be preferentially oxidised by fire compared to other SOM constituents. In the unburnt soil, lignin absolute content was almost equally distributed among density fractions. Fire mainly affected the f-LF, causing large decrease in its VSC content. The o-LF showed the highest lignin contribution to TOC and the highest apparent lignin loss due to fire, while HF shows a relative accumulation of lignin compounds.

At both study sites, none of the indicators describing the composition and degradation of lignin, *i.e.* Acid-to-Aldehyde ratios of V and S-type units, S-to-V and C-to-V ratios, changed significantly in response to fire (Table 4). Hence, it seems that fire oxidised all lignin units without distinction and no selective consumption of specific residues occurred, which is in contrast with what has been reported by other authors, *i.e.* a higher thermal susceptibility of aldehydes in V and S phenols (Certini et al., 2011; Kuo et al., 2008; Nocentini et al., 2011; Ohta and Venkatesan, 1992; Rumpel et al., 2007).

Plotting the sugar vs. lignin content from burnt soil at OR and MG, we found a fairly good linear correlation, which was not present at all in the unburnt soils (Fig. 4). This correlation would depend on fire that, whatever the severity, would affect lignin and sugar leading to an overall decrease of

388 both of them. Although based on a relatively small sample size here, this intriguing correlation
389 deserves further investigation in future studies to ascertain its wider validity and the role fire
390 actually plays in determining the sugar/lignin ratio.
391
392 *3.5 NMR analysis*
393 The ^{13}C CPMAS NMR spectra of bulk soil from burnt and unburnt sites at OR and MG are
394 displayed in Fig. 5, while Table 5 shows the percentage distribution of the total signal among seven
395 selected regions.
396 At both OR and MG, the most evident difference between the spectra of burnt and unburnt soil was
397 the more intense signal of the former one in the Aromatic C region (110-160 ppm), which was
398 clearly due to some input of charred material, whose signal is in fact centred at ~130 ppm
399 (Skjemstad et al., 1996). At OR, this enrichment was counterbalanced by a decrease in Alkyl-C (0-
400 45 ppm region) and a less substantial decrease in O-alkyl C (60-90 ppm). The two sharp peaks at
401 around 72 and 104 ppm in the burnt soil, revealed the persistence of substantial amounts of
402 polysaccharides, possibly cellulose. Therefore unexpectedly, based on what has been reported by
403 several other authors (*e.g.*, Certini et al., 2011; Knicker et al., 2005; Knicker et al., 2006), O-Alkyl
404 C was not the most fire-affected region upon heating. A possible explanation might be related to the
405 high pH imposed by the ash layer overlying the mineral soil (Bodí et al. 2014) that could have
406 catalysed an alkaline hydrolysis of the polysaccharides forming the unburnt plant necromass,
407 leading to the accumulation of dissolved sugars in the topsoil. At MG, in spite of the extremely high
408 fire severity, the ^{13}C CPMAS NMR spectra of burnt and unburnt soil did not show major
409 differences, besides the evident enrichment of aromatic C in the burnt soil. Both soils were
410 characterised by dominant signals in the alkyl C region, generally assigned to lipids and other
411 aliphatic compounds, and in the O-alkyl C region, indicative for polysaccharides and amide C of
412 proteins (Knicker and Lüdemann, 1995). Actually, there were some differences in the relative
413 intensities of these signals from the unburnt to the burnt soils, quite small the one of alkyl C region

414 and more consistent the one of O-alkyl C, this latter attributable to the degradation of carbohydrate-
415 derived structures, mainly cellulose.
416 The NMR spectra of the light density fractions of burnt and unburnt soils from OR and MG are
417 shown in Fig. 6 and 7, respectively, while the signal distribution among the selected seven regions
418 of the spectra is in Table 5. The light fractions from the unburnt soils had similar patterns in the two
419 sites, although the o-LF revealed more advanced stage of alteration than f-LF, as chiefly indicated
420 by a lower O-alkyl C–to–alkyl C ratio (Baldock et al., 1992) and a much higher signal in the aryl C
421 region (Golchin et al., 1994). The higher relative intensity observed in the aromatic region at ~150,
422 130 and 115 ppm for o-LF with respect to f-LF suggests higher lignin content (Golchin et al., 1994;
423 Hatcher, 1987). The NMR analysis unravelled that in both soils the light fractions were to some
424 extent affected by fire (Figs. 6 and 7). In particular, at OR, the f-LF of burnt soil experienced an
425 increase of the peak at 130 ppm, and small signal losses in the alkyl (0-45 ppm), O-alkyl (60-90
426 ppm) and carboxyl C (160-185), overall indicating a slight carbonisation of the OM. In the o-LF,
427 the peak at 56 ppm (ascribable to lignin methoxyl carbon) and the signal at around 150 ppm (O-
428 substituted phenolic carbon) decreased much in the burnt soil compared to the unburnt one, which
429 indicated some lignin decomposition. At MG, the most evident change between the burnt and the
430 unburnt soil was the increase of the peak at 130 ppm for both the light fractions. Ancillary
431 differences are revealed by the decrease of the peaks at about 150 and 53, which indicate lignin
432 oxidation, and the intensification of the peak at 174 ppm, which implies an increase in carboxylic C.
433 This latter fact remains difficult to explain, since organic matter exposed to severe heating generally
434 loses carboxyl C (Knicker et al., 2005).
435

436 **4. Conclusions**

437 Our parallel investigation at Orentano and Mt. Gordon, two areas recently affected by wildfires of
438 markedly different severity, showed that in both cases, fire had a marked impact on SOM
439 composition of the uppermost 2.5 cm of mineral soil, which was partly independent of fire severity.

At Orentano, Italy, where fire severity in the mixed oak-pine forest was moderate, soil experienced a slight, statistically not significant, gain in carbon, mostly consisting in partly charred biomass from the organic layer and the standing vegetation. At Mt. Gordon, Australia, where the eucalypt forest had been burnt by an extremely severe fire, SOM experienced substantial loss not counterbalanced by the incorporation of charred materials, although it is likely that over time some of the OM still retained in the ash layer will be partly incorporated into the mineral soil.

Density fractionation enabled to examine SOM pools with different characteristics and turnover time in soil, and to assess the fire impact on each of them. In spite of contrasting fire severity, at the two study sites we found similar fire impacts on the SOM assumed occluded in aggregates. Apparently, because of fire, some organic particles were released from disrupted aggregates, the lighter of them becoming hence part of the free light fraction of SOM, while the OM bound to the freed mineral particles joined the heavy fraction. Such a SOM redistribution could imply substantial changes in the soil environment, in particular in the C cycle. It is reasonable, for example, to assume an enhancement of the overall decay rate of post-fire SOM because of easier access to microorganisms of the OM derived from disruption of aggregates. This outcome of fire should be taken into account, together with the significant immediate loss of SOM due to combustion and the increase in mean residence time of charred residues, when accounting for the fire impact on C cycle.

In terms of SOM composition, lignin in soil at OR was preferentially oxidised compared to sugars, particularly in the fraction occluded into aggregates, where presumably minerals offered different protection to different compounds. At MG, where the lignin and sugar content in soil were apparently halved in response to fire occurrence, none of the SOM components were preferentially oxidised, either in the bulk soil or the density fractions. Here, evidently, fire severity was so high that SOM was non-selectively combusted. Variables formerly proposed as reliable indicators of fire severity in soil, *i.e.* the sugar–to–total organic C ratio and phenols ratio in lignin, did not provide

encouraging results in this study. Further studies are needed to elucidate the complex fire impact on SOM composition and to eventually individuate chemical legacies useful to reconstruct fire history.

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Table 1

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Table 1. Location and soil pH, bulk density (mean and standard deviation; n=3) and particle size distribution of the study sites.

Site (Code)	Sampling area	Location WGS84	soil pH (H_2O, 5:1)	bulk density g cm^{-3}	Sand %	Silt %	Clay %
Orentano (OR) Italy	Burnt	43°47'22.82"N 10°39'52.30"E	5.12 (±0.37)	0.98 (±0.07)	11	71	17
	Unburnt	43°47'25.94"N 10°39'50.35"E	4.22 (±0.32)	1.11 (±0.17)	16	66	19
Mount Gordon (MG) Australia	Burnt	37°31'56.30"S 145°43'17.14"E	5.90 (±0.37)	0.81 (±0.06)[a]	29	52	18
	Unburnt	37°32'54.10"S 145°37'37.30"E	4.83 (±0.03)	0.64 (±0.14)[a]	33	44	22

[a] n=2 for mean and standard deviation values

Table 2

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Table 2. Elemental analysis, of charcoal, burnt and unburnt soil (mean values and standard deviation; n=20 for bulk soil samples; n=10 for charcoal samples; n=2 for density fractions).

Site (Code)	Sample	Fraction	% of bulk soil weight	C org g kg^{-1}	N g kg^{-1}	C/N	$\delta^{13}C$ ‰
Orentano (OR) Itlay	Burnt soil	bulk	-	100 (±34)	5.8 (±2.0)	17.2 (±1.7)	-27.8 (±0.3)*
		f-LF	15.0 (±0.6)	216 (±2)	12.0 (±0.2)	18.1 (±0.4)	-27.9 (±0.1)
		o-LF	6.5 (±1.1)	415 (±20)	21.0 (±0.6)	19.8 (±0.4)	-28.1 (±0.1)
		HF	78.5 (±1.7)	31 (±3)	2.5 (±0.2)	12.8 (±0.4)	-27.3 (±0.2)
	Unburnt soil	bulk	-	84 (±27)	5.2 (±2.1)	17.0 (±3.7)	-27.5 (±0.3)*
		f-LF	9.1 (±2.0)	192 (±14)	10.7 (±2.5)	18.3 (±2.9)	-27.7 (±0.1)
		o-LF	9.3 (±2.4)	304 (±130)	13.9 (±3.4)	21.3 (±4.1)	-28.0 (±0.1)
		HF	81.5 (±0.4)	24 (±1)	2.0 (±0.4)	12.1 (±2.0)	-27.1 (±0.8)
	Charcoal	-		338 (±62)	16.1 (±2.6)	20.7 (±3.6)	-28.6 (±0.5)
Mount Gordon (MG) Australia	Burnt soil	bulk	-	87 (±13)**	4.2 (±0.5)**	20.6 (±2.1)**	-27.5 (±0.2)**
		f-LF	9.0 (±4.0)	171 (±9)*	7.1 (±0.4)	24.2 (±0.2)	-27.7 (±0.1)
		o-LF	7.6 (±0.9)	324 (±33)	9.6 (±0.0)	33.7 (±3.7)	-27.7 (±0.1)
		HF	83.4 (±4.9)	40 (±3)	2.8 (±0.1)	14.3 (±0.6)	-27.3 (±0.0)
	Unburnt soil	bulk	-	163 (±55)**	5.8 (±0.1)**	27.5 (±3.4)**	-27.7 (±0.2)**
		f-LF	20.9 (±8.0)	252 (±11)*	8.3 (±0.1)	30.3 (±0.8)	-28.0 (±0.1)
		o-LF	11.1 (±1.4)	233 (±16)	6.6 (±0.4)	35.5 (±0.2)	-28.1 (±0.3)
		HF	68.0 (±9.4)	54 (±19)	3.2 (±0.5)	16.9 (±3.2)	-27.3 (±0.3)
	Charcoal	-	-	196 (±52)	6.0 (±1.4)	28.5 (±9.9)	-27.4 (±0.5)

* Significantly different means between burnt and unburnt soil ($P < 0.05$)

** Significantly different means between burnt and unburnt soil ($P < 0.01$)

Table 3

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Table 3. Non-cellulosic neutral sugars content and (galactose + mannose)/(arabinose + xylose) ratio (GM/AX) of burnt and unburnt soil (mean values and standard deviation; n=4 for bulk soil; n=1 for density fractions).

Site (Code)	Sample	Fraction	Σ Neutral Sugars g kg^{-1} bulk soil[a]	Σ Neutral Sugars g kg^{-1} OC	GM/AX
Orentano (OR) Itlay	Burnt soil	bulk	7.46 (±0.43)	76.6 (±6.59)*	2.79 (±0.90)
		f-LF	1.13	111.2	1.28
		o-LF	0.74	86.1	1.24
		HF	4.88	178.9	1.67
	Unburnt soil	bulk	8.27 (±2.30)	98.6 (±13.0)*	2.81 (±1.50)
		f-LF	1.11	119.7	1.55
		o-LF	0.70	54.6	149
		HF	2.50	133.1	2.55
Mount Gordon (MG) Australia	Burnt soil	bulk	9.18 (±1.90)*	105.2 (±26.2)	2.00 (±0.44)
		f-LF	0.61	88.0	1.42
		o-LF	0.45	52.2	1.30
		HF	3.40	99.1	2.10
	Unburnt soil	bulk	20.9 (±6.18)*	126.1 (±26.4)	1.60 (±0.18)
		f-LF	5.12	137.6	1.33
		o-LF	0.65	77.92	1.16
		HF	5.01	160.8	2.69

* Significantly different means between burnt and unburnt soil ($P < 0.05$)

[a] For density fractions, net amount of sugars was calculated by multiplying the sugar content of a given fraction by the mass proportion (% of bulk soil weight) of that fraction.

Table 4

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Table 4. Sum of vanillyl, syringyl and cinnamyl units (VSC), acid–to –aldehyde ratio of vanillyl and syringyl units, cinnamyl phenols– and syringyl phenols–to–vanillyl phenols, in the bulk soil and density fractions (mean values and standard deviation; n=4 for soil samples; n=2 for density fractions).

Site (Code)	Sample	Fraction	Σ VSC g kg^{-1} bulk soil[a]	Σ VSC g kg^{-1} OC	$(Ac/Al)_v$	$(Ac/Al)_s$	C/V	S/V
Orentano (OR) Itlay	Burnt soil	bulk	0.35 (±0.01)**	3.57 (±0.18)**	0.74 (±0.37)	0.78 (±0.13)	0.42 (±0.13)	0.38 (±0.22)
		f-LF	0.13(±0.08)	4.10 (±2.81)	1.57 (±2.19)	1.43 (±1.05)	0.56 (±0.12)	0.76 (±0.12)
		o-LF	0.08 (±0.02)	3.05 (±0.40)	0.57 (±0.77)	0.65 (±0.10)	0.59 (±0.39)	0.97 (±0.45)
		HF	0.25 (±0.15)	8.13 (±1.66)	1.09 (±0.13)	0.67 (±0.09)	0.45 (±0.10)	0.62 (±0.06)
	Unburnt soil	bulk	0.65 (±0.14)**	7.71 (±0.86)**	0.58 (±0.22)	0.76 (±0.32)	0.38 (±0.09)	0.50 (±0.11)
		f-LF	0.11 (±0.01)	6.48 (±1.83)	0.26 (±0.14)	1.09 (±0.78)	0.29 (±0.10)	0.82 (±0.08)
		o-LF	0.20 (±0.09)	8.26 (±5.16)	0.11 (±0.12)	0.83 (±0.52)	0.80 (±0.04)	0.81 (±0.13)
		HF	0.17 (±0.02)	8.98 (±1.40)	0.92 (±0.02)	0.60 (±0.03)	0.33 (±0.00)	0.57 (±0.01)
Mount Gordon (MG) Australia	Burnt soil	bulk	0.65 (±0.23)*	7.53 (±2.97)	0.96 (±0.14)	0.87 (±0.57)	0.24 (±0.10)	1.21 (±0.32)
		f-LF	0.17 (±0.09)	10.92 (±1.82)	0.41 (±0.29)	0.65 (±0.33)	0.56 (±0.05)	1.63 (±0.82)
		o-LF	0.21 (±0.14)	8.90 (±5.93)	1.16 (±0.42)	1.24 (±0.91)	0.59 (±0.43)	1.12 (±0.88)
		HF	0.54 (±0.25)	15.97 (±7.09)	0.80 (±0.28)	0.62 (±0.17)	0.45 (±0.13)	1.18 (±0.14)
	Unburnt soil	bulk	1.24 (±0.38)*	7.52 (±2.30)	1.05 (±1.21)	0.75 (±0.47)	0.32 (±0.29)	1.96 (±0.59)
		f-LF	0.38 (±0.10)	11.59 (±0.39)	1.41 (±1.43)	7.82 (±8.69)	0.29 (±0.09)	1.24 (±1.02)
		o-LF	0.29 (±0.14)	21.45 (±10.32)	0.23 (±0.27)	6.73 (±9.00)	0.80 (±0.05)	1.63 (±1.30)
		HF	0.46 (±0.29)	10.06 (±0.28)	0.80 (±0.13)	0.52 (±0.02)	0.33 (±0.00)	1.44 (±0.13)

* Significantly different means between burnt and unburnt soil ($P < 0.05$)

** Significantly different means between burnt and unburnt soil ($P < 0.01$)

[a] For density fractions, the net amount of VSC was calculated by multiplying the VSC content of a given fraction by the mass proportion (% of bulk soil weight) of that fraction

Table 5

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Table 5. Percentage distribution (%)[a] of the signal intensity between selected chemical shift regions of ^{13}C CPMAS NMR spectra of bulk soil at Orentano (OR) and Mount Gordon (MG). The contribution of aldehydes and ketones (chemical shift region 185-220) is not reported because negligible.

Site (Code)	Relative distribution (%) of chemical shift region (ppm)						
	Alkyl C	O-Alkyl C			Aryl C		Carboxyl C
	0 – 45	Methoxyl/N-alkyl 45-60	O-alkyl 60-90	Di-O-alkyl 90-110	C substituted (aromatic C-H and C-C) 110-140	O substituted 140-160	carboxyl C/amide/ester 160-185
Orentano (OR)							
Burnt							
bulk soil	30	7	20	6	22	7	8
f-LF <1.8 g cm^{-3}	31	8	24	7	22	4	4
o-LF <1.8 g cm^{-3}	34	7	23	6	16	7	6
Unburnt							
bulk soil	38	6	22	7	14	6	7
f-LF <1.8 g cm^{-3}	33	8	29	8	13	3	6
o-LF <1.8 g cm^{-3}	31	6	24	6	22	5	5
Mount Gordon (MG)							
Burnt							
bulk soil	27	8	24	7	18	8	8
f-LF <1.8 g cm^{-3}	30	8	26	8	16	5	7
o-LF <1.8 g cm^{-3}	31	6	23	8	24	4	4
Unburnt							
bulk soil	30	8	31	8	12	5	6
f-LF <1.8 g cm^{-3}	31	11	30	9	10	5	4
o-LF <1.8 g cm^{-3}	30	7	24	9	16	7	6

[a] Maximum error ± 10%.

Figure 1

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Figure 1 color

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Figure 2
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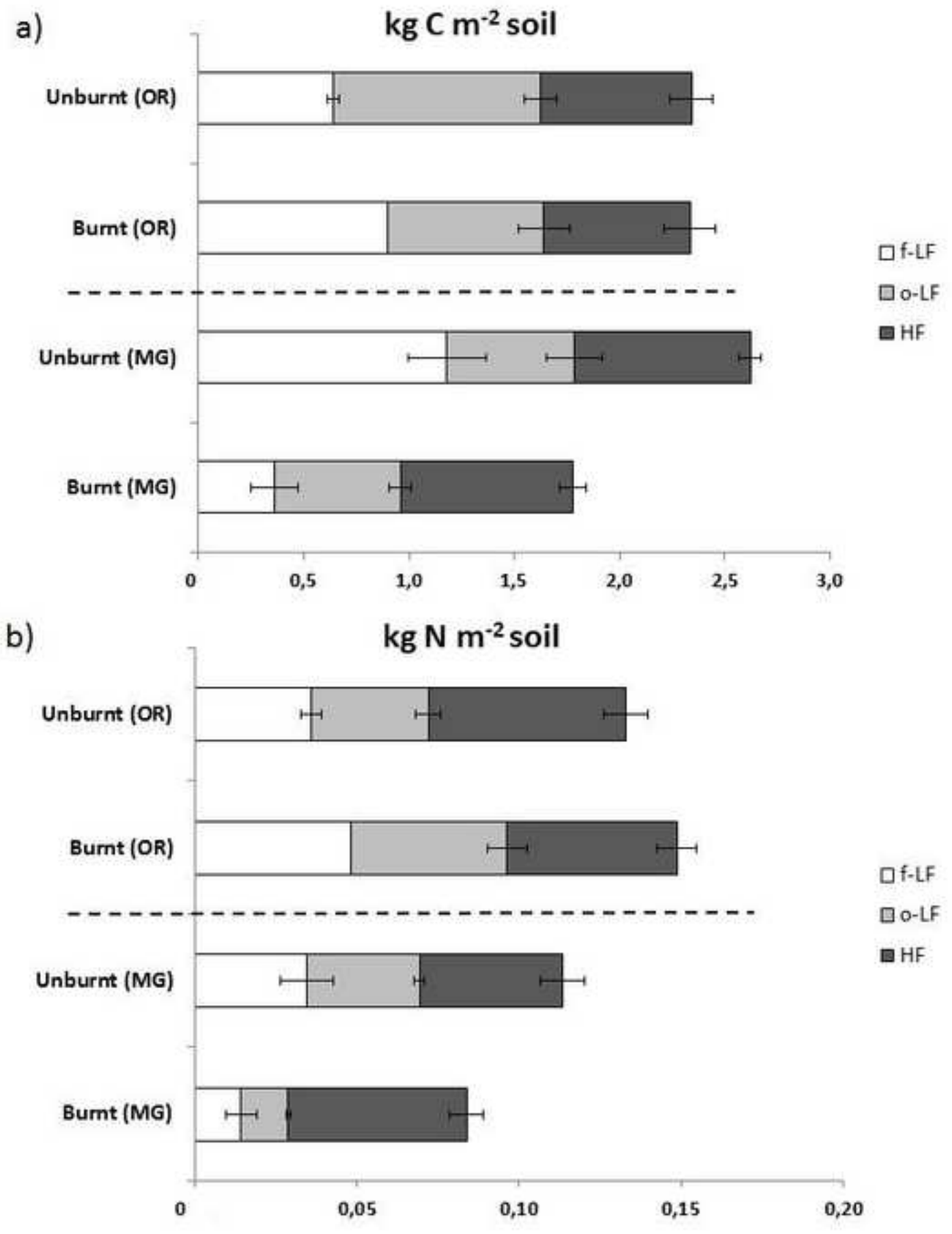

Figure 3

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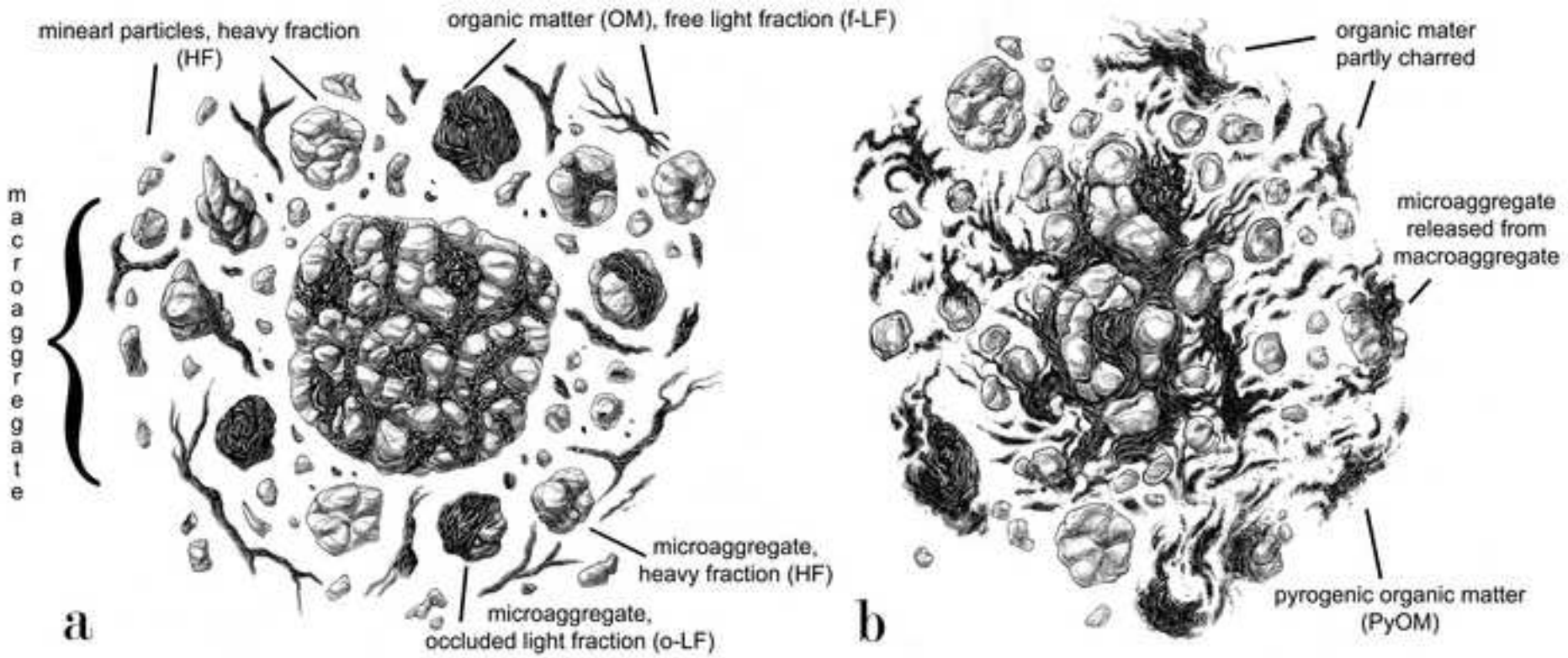

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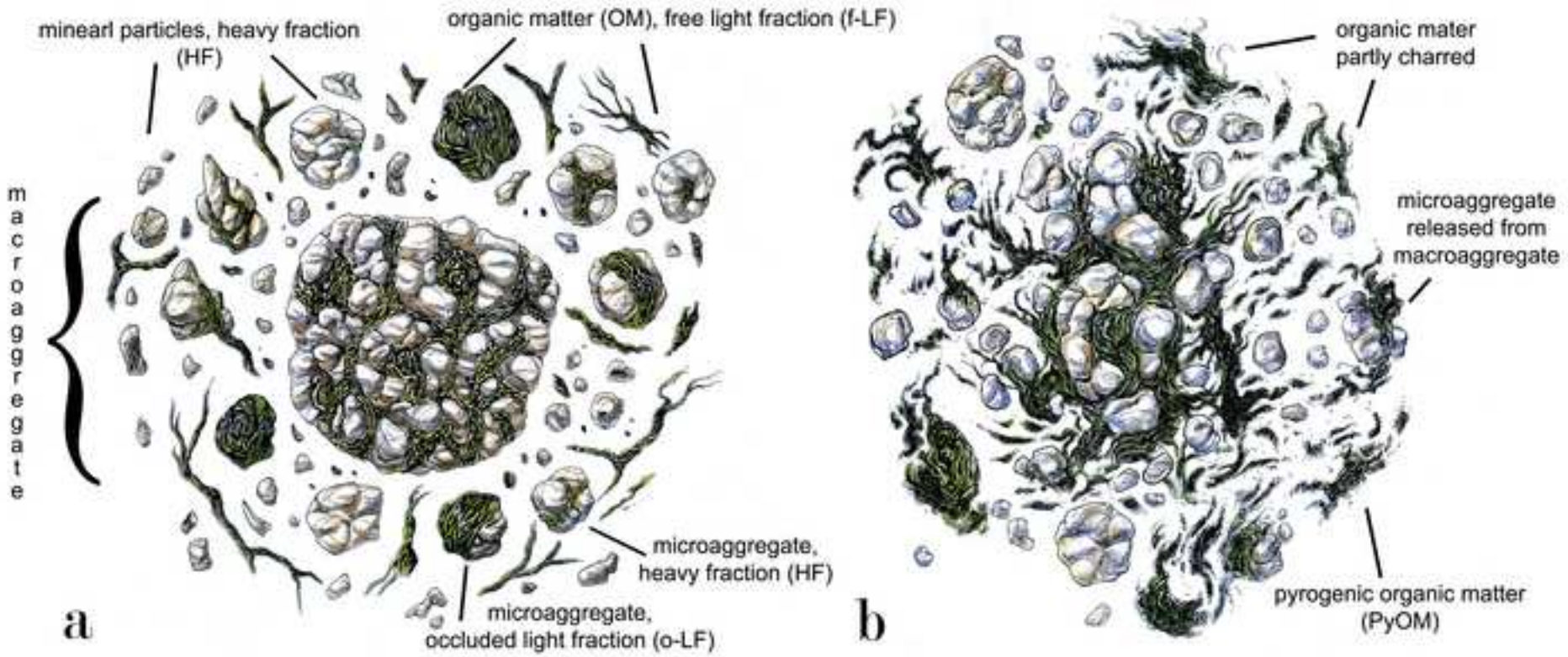

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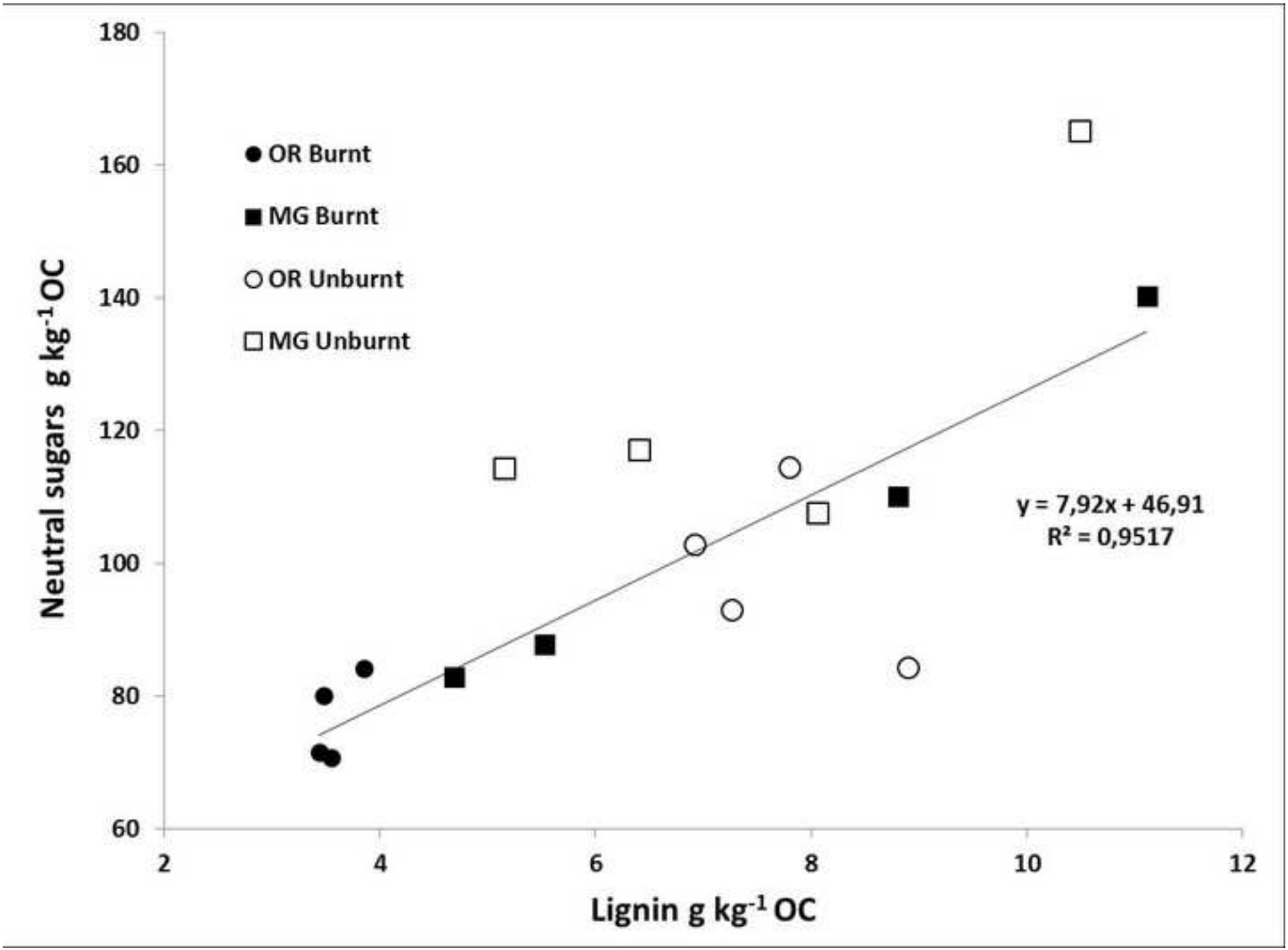

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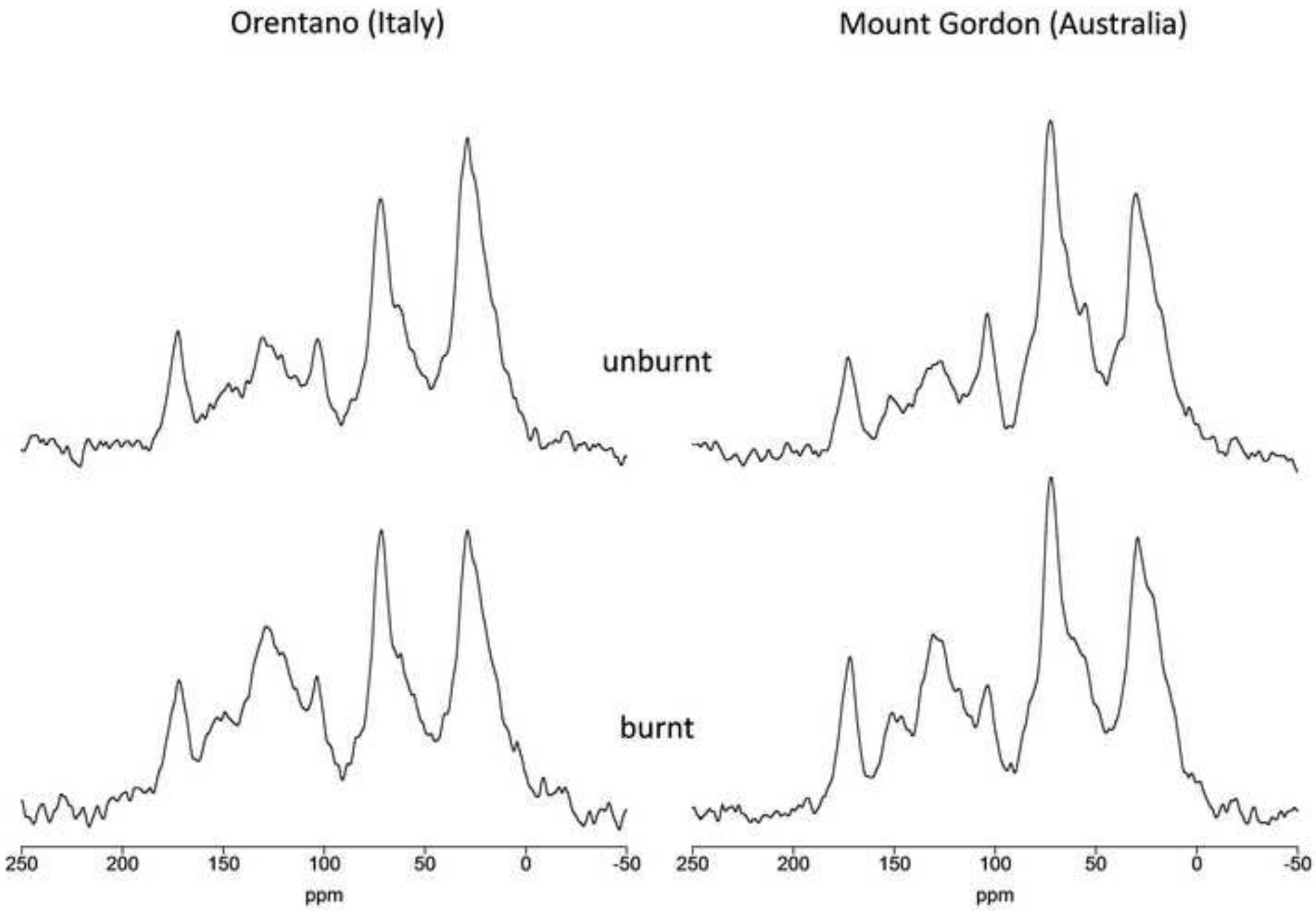

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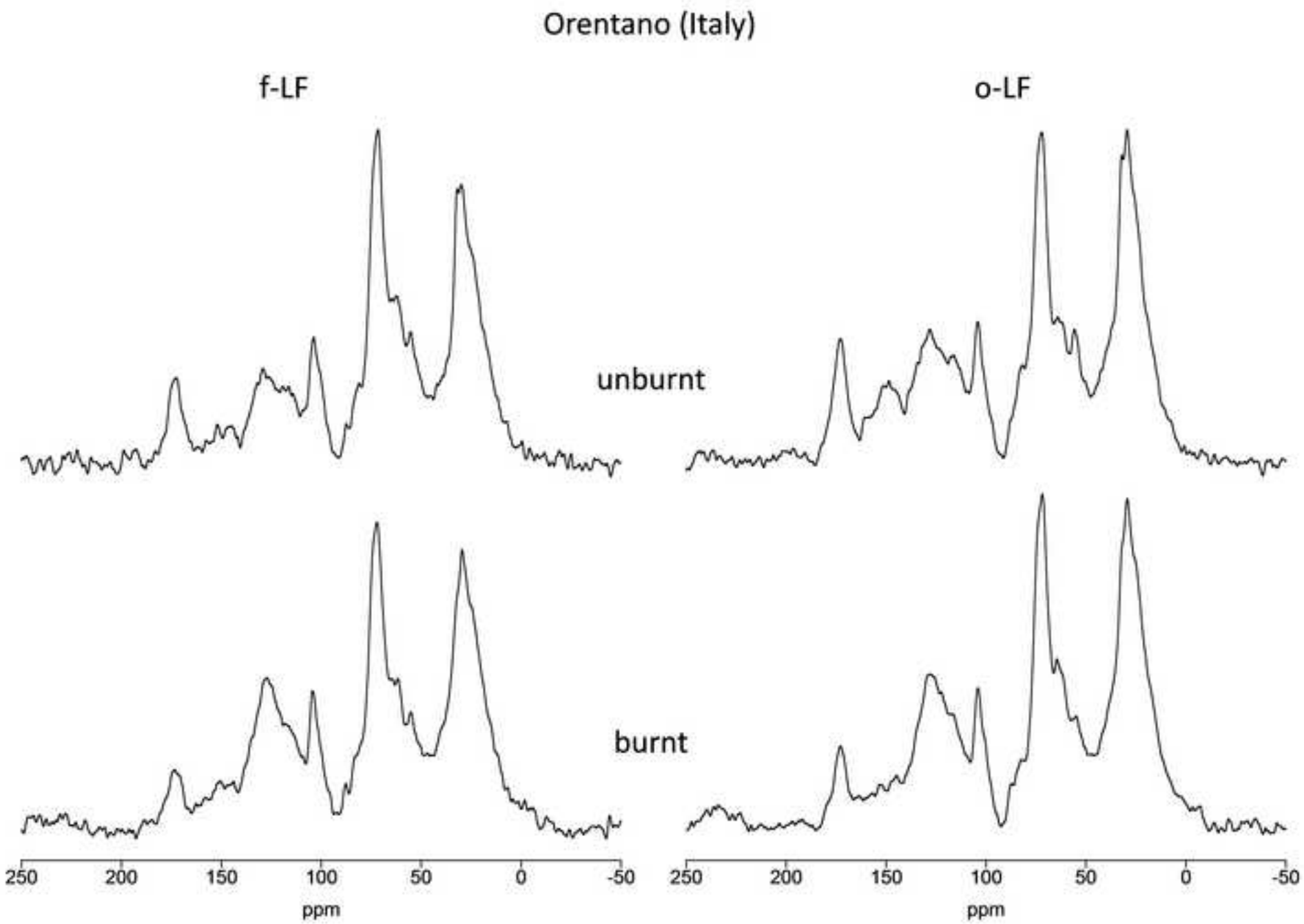

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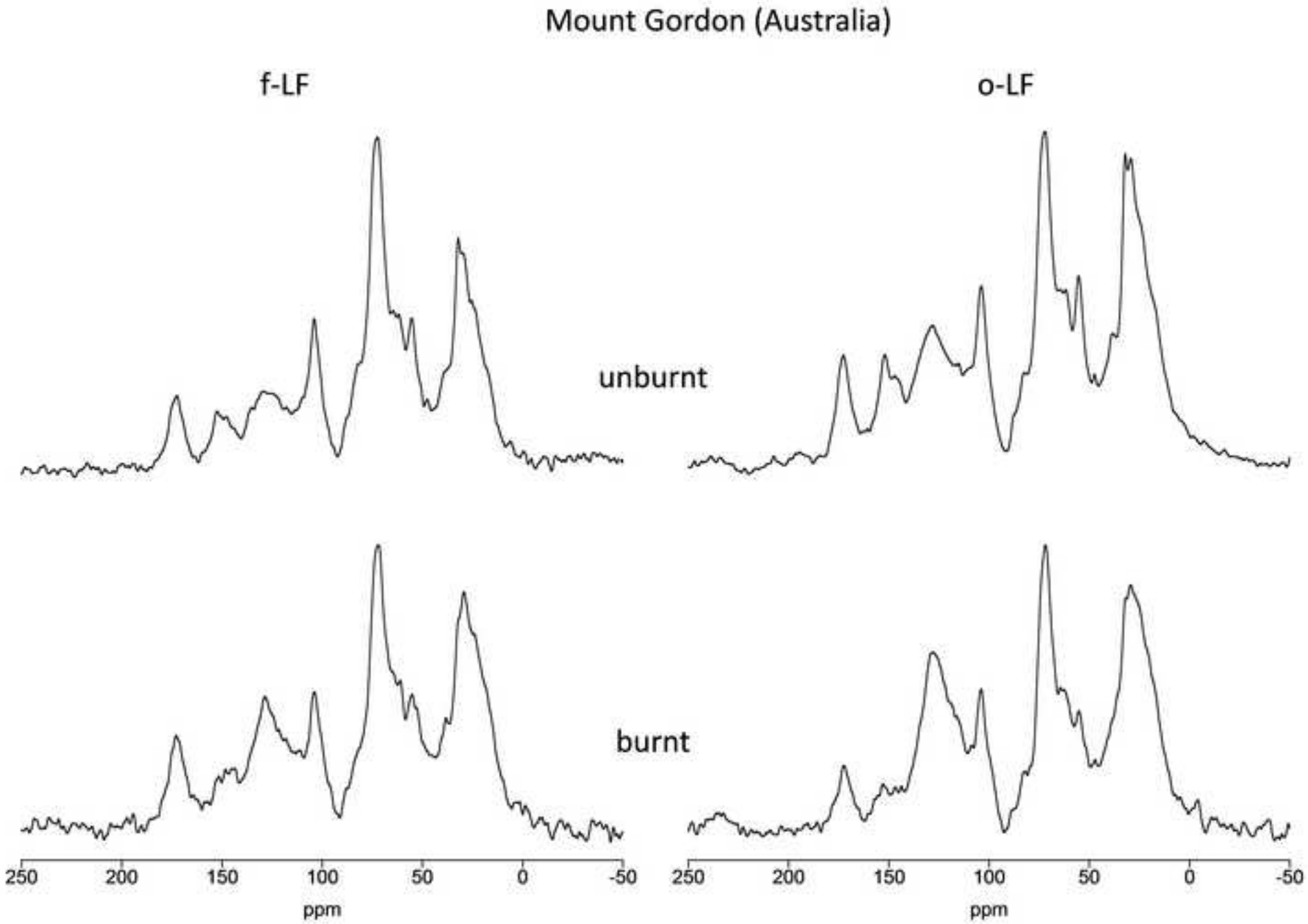

Figure captions:

Fig. 1. The study sites as they appeared during the sampling: a) Orentano, two days after the fire, and b) Mount Gordon, two months after the fire.

Fig. 2. Soil C and N content distribution among density fractions.

Fig. 3. a) Sketch of a soil macroaggregate and the density fractions of SOM: free light fraction (f-LF), occluded light fraction (o-LF) and the heavy fraction (HF); b) Conceptual model of fire impact on soil organic matter free and occluded in soil aggregates (picture by G. Borgogni).

Fig. 4. Relationship between the OC-normalised contents of lignin and neutral sugars, in burnt and unburnt bulk soils from Orentano and Mount Gordon. A regression curve is provided for the burnt bulk soils.

Fig. 5. ^{13}C CPMAS NMR spectra of the burnt and unburnt bulk uppermost mineral soil from Orentano and Mount Gordon.

Fig. 6. ^{13}C CPMAS NMR spectra of the light density fractions, f-LF and o-LF, of the burnt and the unburnt soil from Orentano.

Fig. 7. ^{13}C CPMAS NMR spectra of the light density fractions, f-LF and o-LF, of the burnt and the unburnt soil from Mount Gordon.