



Moisture-driven divergence in mineral-associated soil carbon persistence

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Mineral stabilization of soil organic matter is an important regulator of the global carbon (C) cycle. However, the vulnerability of mineral-stabilized organic matter (OM) to climate change is currently unknown. We examined soil profiles from 34 sites across the conterminous USA to investigate how the abundance and persistence of mineral-associated organic C varied with climate at the continental scale. Using a novel combination of radiocarbon and molecular composition measurements, we show that the relationship between the abundance and persistence of mineral-associated organic matter (MAOM) appears to be driven by moisture availability. In wetter climates where precipitation exceeds evapotranspiration, excess moisture leads to deeper and more prolonged periods of wetness, creating conditions which favor greater root abundance and also allow for greater diffusion and interaction of inputs with MAOM. In these humid soils, mineral-associated soil organic C concentration and persistence are strongly linked, whereas this relationship is absent in drier climates. In arid soils, root abundance is lower, and interaction of inputs with mineral surfaces is limited by shallower and briefer periods of moisture, resulting in a disconnect between concentration and persistence. Data suggest a tipping point in the cycling of mineral-associated C at a climate threshold where precipitation equals evaporation. As climate patterns shift, our findings emphasize that divergence in the mechanisms of OM persistence associated with historical climate legacies need to be considered in process-based models.

soil carbon | radiocarbon | mineral stabilization | terrestrial carbon cycle | soil carbon persistence

Globally, mineral-associated organic matter (MAOM) represents the majority of soil organic carbon (C) [~60%; (1)], and recent syntheses support the idea of a higher degree of stability for MAOM than particulate organic matter (OM) (1–3). Mineral association is generally assumed to confer stability to soil organic matter (SOM) by physically or chemically decreasing microbial decomposer access. However, this stability is not absolute. MAOM can be destabilized through the introduction of “fresh” substrate that provides the energy necessary for plants and microbes to mine nutrients concentrated in the MAOM pool, such as nitrogen (4–6). Typically, as the input of fresh substrate decreases exponentially with depth, the persistence [i.e., mean system “age,” (7)] of the MAOM increases concomitantly (8, 9). This commonly observed pattern reflects a strong interdependence of moisture availability, substrate inputs, decomposition, and the persistence of MAOM.

A wide array of factors influences the cycling of SOM, ranging from climate to physicochemical properties (8, 10, 11). However, the simplest of variables, *depth*, consistently explains half or more of the variation associated with bulk SOM and MAOM abundance and persistence (as approximated by radiocarbon measurements) (1, 12, 13). However, depth itself is not a direct or mechanistic driver of MAOM accrual or turnover. The relationship between depth and SOM abundance and persistence integrates variation in a myriad of mechanistic drivers including substrate inputs, oxygen availability, mineral surface reactivity, pH, and other soil properties that affect rates of SOM decomposition. Key to understanding the relationship of depth to SOM is linking SOM profiles to vertical transport of water and solutes—as soil morphological development, net primary production (NPP) and SOM production are directly dependent on moisture availability [i.e., hydroclimatic regime; (14–16)].

At the continental scale, soils and hydroclimatic regimes can be bifurcated into two categories based on moisture availability (17, 18). In water-limited systems (referred to here as *arid*), evaporation is limited by moisture availability, and the ratio of mean annual precipitation (MAP) to potential evapotranspiration (PET) is less than one (MAP/PET < 1). In energy-limited systems (referred to here as *humid*), evaporation is limited by available evaporative energy (MAP/PET > 1). In the conterminous USA, this threshold lies near the 98th meridian and approximately 635 mm annual precipitation [Fig. 1; (19)].

Significance

Mineral-associated organic matter (MAOM) comprises the largest reservoir of C in soils globally, and the vulnerability of this pool to climate change is currently unknown. This work demonstrates that moisture availability drives differences in the soil factors regulating MAOM concentration and persistence, with a strong bifurcation in MAOM characteristics and cycling between arid and humid soils. In wetter climates where conditions favor greater root abundance and interaction of inputs with mineral surfaces, mineral-associated soil organic C concentration, and persistence are strongly linked, whereas this relationship is absent in drier climates. Shifting climate patterns are likely to result in nonlinear responses in the stability of MAOM.

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This geographic and hydroclimatic boundary is associated with sharply contrasting soil morphology (17), pH values (20), and MAOM abundance (21). However, how this hydroclimatic threshold influences the stability and turnover of MAOM is currently unknown, as is the relative importance of climate versus physicochemical properties in determining MAOM persistence at the continental scale. We examined surface and subsurface soils (surface, mid-depth, and basal horizons, *SI Appendix, Fig. S1*) from 34 sites (20 arid, 14 humid) across the National Ecological Observatory Network (NEON; Fig. 1) to investigate differences in MAOM abundance and persistence associated with shifts in moisture availability, C inputs, and soil physicochemical characteristics. Here, we provide mechanistic insight into the drivers of MAOM persistence across climate thresholds by synthesizing mineral-associated fraction radiocarbon abundance [$\Delta^{14}\text{C}$ (‰)]

and %C measurements with a novel and diverse suite of complementary soil physicochemical and climate variables.

Results and Discussion

MAOM Persistence Regulated by Substrate Limitation in Humid Soils and Transport Limitation in Arid Soils. We report a strong contrast in the relationship between MAOM abundance and persistence between arid and humid hydroclimatic regimes (Fig. 2). In humid soils, decreasing MAOM abundance was coupled with increasing persistence (as inferred from radiocarbon measurements of MAOM fractions; $r^2 = 0.63$, $P < 0.0001$), and this relationship strongly covaried with depth (*SI Appendix, Figs. S2 and S3*). In contrast, this relationship was wholly absent in arid soils (Fig. 2). Additionally, we noted strong correlations among concentrations

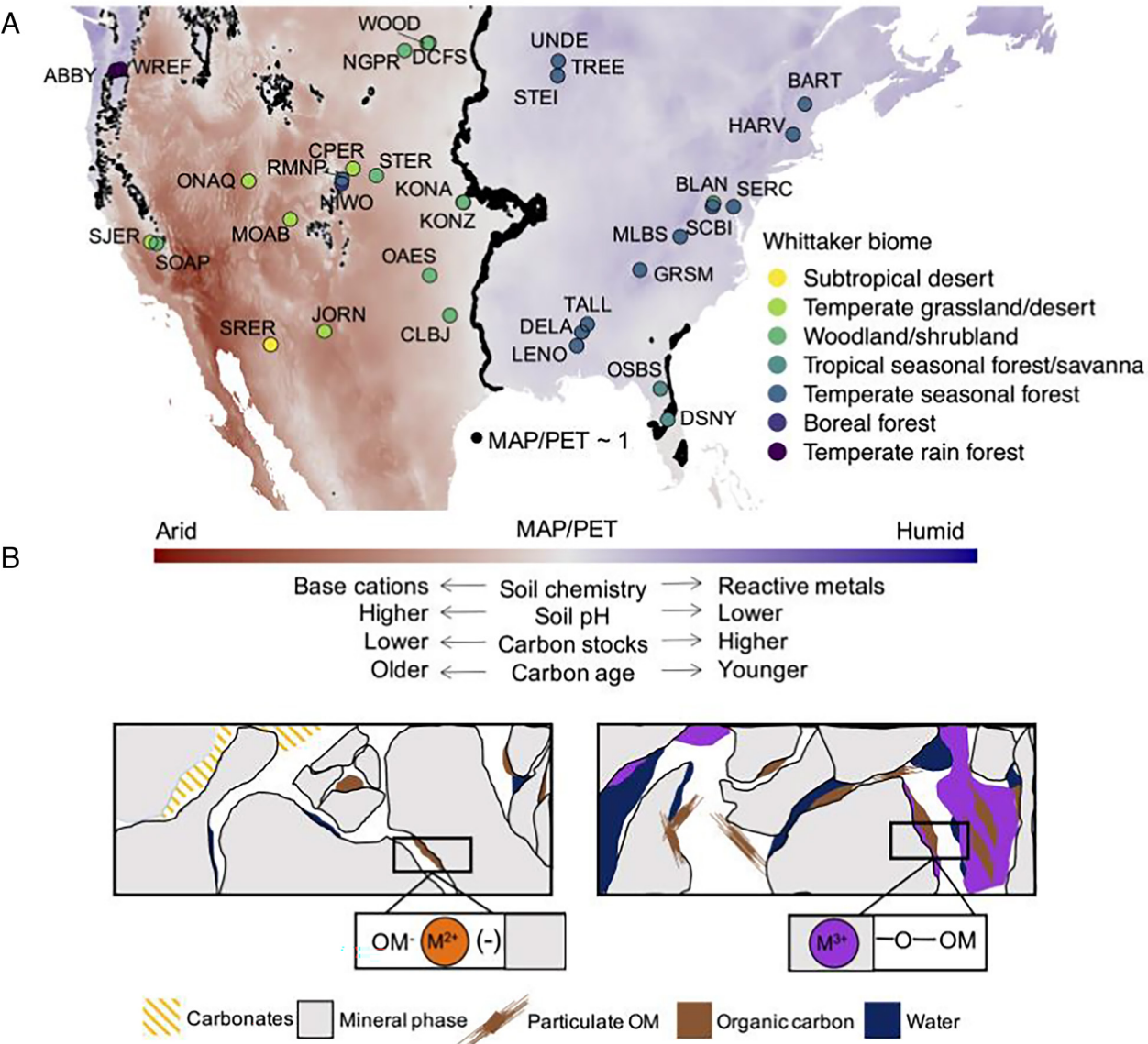


Fig. 1. Moisture availability as an integrative predictor of MAOM formation and persistence at the continental scale. (A) Moisture availability, here shown by the ratio of MAP to PET, follows a strong longitudinal gradient across the conterminous US, driving spatial patterns of dominant vegetation, soil properties, and SOC cycling. Map shows $\ln$ -transformed MAP/PET. MAP and PET data (at 0.25-degree resolution) represent yearly total for 2018, derived from TerraClimate (22). Black points indicate where MAP and PET are roughly equal (MAP/PET ~ 1), concurrent with the historical division between soil climate regimes as defined by Marbut (17). Map uses Cartesian coordinates. (B) Conceptual summary of divergence in expected dominant mechanisms of MAOM formation linked to differences in relative abundance of soil water, base cations (shown in orange) versus reactive iron and aluminum (shown in magenta), inorganic carbon, and particulate OM (12, 13, 23, 24). Call-out boxes represent simplified surface interaction mechanisms, ranging from predominantly electrostatic (e.g., cation bridging) in arid systems to inner-sphere (e.g., covalent bonds) in humid systems.

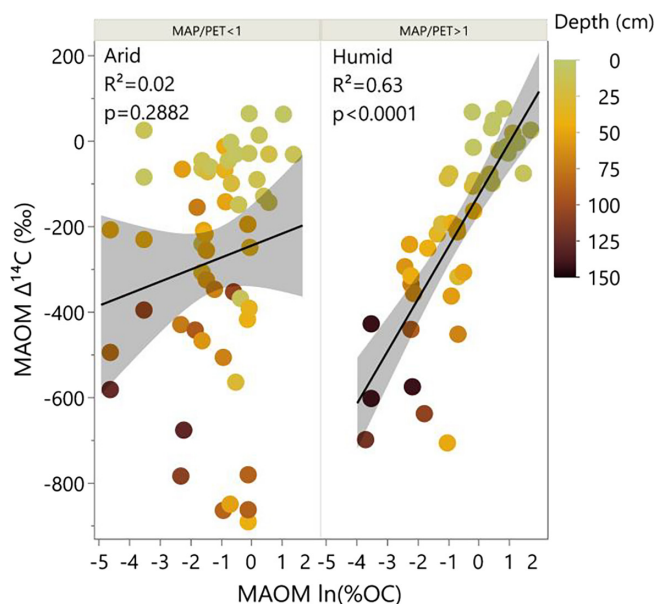


Fig. 2. MAOM %C and $\Delta^{14}\text{C}$ are strongly coupled by transport in humid soils. This relationship is wholly absent in arid soils. Shaded areas reflect the 95% CI for the line of fit.

of MAOM C, water-soluble C, and root abundance in humid soils, whereas these relationships were weaker or wholly absent in arid soils (Fig. 3 and *SI Appendix*, Fig. S4). We examined the composition of the MAOM pool and its relation to soil physicochemical properties to further investigate how moisture availability modifies the cycling of MAOM. To do this, we combined a suite of physicochemical measurements associated with known mechanisms of SOM stabilization [exchangeable cations, Fe and Al from reactive mineral phases, and particle-specific surface area (PSSA)] with traditional and novel characterization techniques assessing the composition and decomposition status of the MAOM and water-soluble pools [cupric oxide oxidation, ultraviolet–visible (UV–VIS) spectroscopy, Fourier transform ion cyclotron resonance mass spectrometry (FT-ICR-MS), and excitation emission mass

spectrometry (EEMS)]. We found that humid and arid systems differed significantly in the persistence of MAOM, the relationships between MAOM and soil physicochemical properties (Fig. 4), and the molecular composition of the MAOM and soluble OM fraction (*SI Appendix*, Table S2). Collectively, these differences support a moisture-driven divergence in the mechanisms regulating the cycling of MAOM.

OM is deposited and transported through the soil matrix by multiple processes including vertical and lateral transport of readily soluble organics and in situ root deposition. Soluble OM may be derived from percolation through organic horizons or root deposits, but roots are generally considered the largest primary contributor to soil C globally, depositing soluble OM in the form of root exudates and particulate OM through root sloughing and death (26). These root-derived organic inputs contribute to MAOM formation through direct interaction with minerals and through decomposition and conversion to microbial necromass which may then be stabilized on mineral surfaces (27). Root abundance and architecture vary widely among terrestrial ecosystems and are often a direct reflection of moisture availability. Rooting depths may be more variable in arid soils where plants utilize a variety of strategies for moisture acquisition, but rooting depth and total root biomass are more often greater in more humid forested ecosystems (28, 29). This variability in root characteristics suggests that their contribution to MAOM may also vary with climate. Consistent with these general trends, root mass in humid soils was significantly higher than in arid systems (*SI Appendix*, Fig. S5). We also observed a strong bifurcation in the relationship of roots to MAOM concentration between humid and arid soils where root abundance and MAOM C concentration were strongly linked in humid but not arid soils (Fig. 3). This pattern may result from moisture limitations on root decomposition or exudate production in arid soils, as connections between root mass and the water-soluble C pool in arid soils are much weaker than in humid soils (*SI Appendix*, Fig. S4). Conceptually and quantitatively, moisture availability, root abundance, and water-soluble C abundance are intrinsically linked. Greater moisture availability allows for greater root production, which can result in increased production and mobilization of root exudates (30, 31). The abundance of

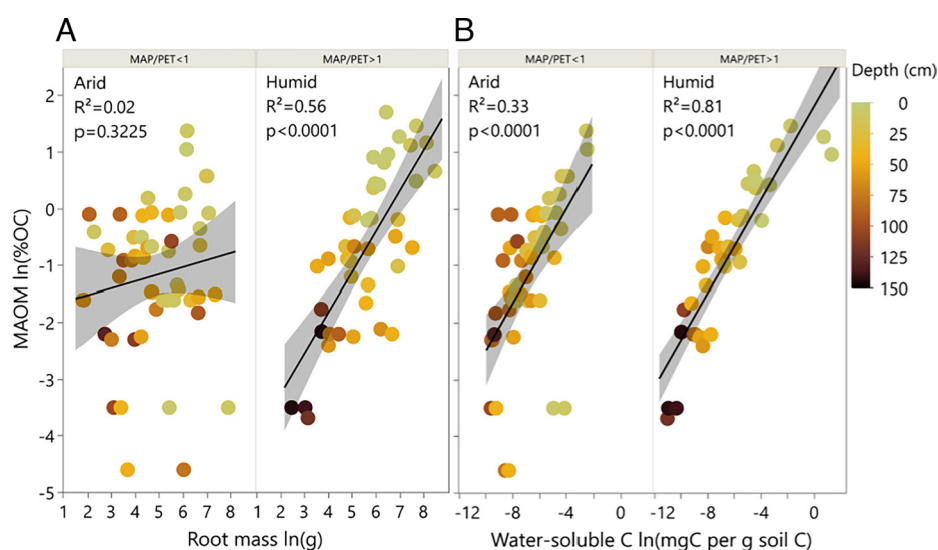


Fig. 3. The concentration of MAOM is strongly associated with both root abundance and water-soluble C abundance. (A) The relationship of root mass with MAOM %C. Root masses were derived from the NEON data product, Root biomass and chemistry (25). Root masses were significantly linked to MAOM C concentrations in humid but not arid sites. (B) The relationship of C solubility with MAOM %C. Specifically, the x axis unit is dissolvable C normalized to the bulk soil C value (mg soluble C per g of bulk soil C). Therefore, there is not simply more soluble C because there is more C in general. Rather it shows that C solubility differs with depth and among the two hydroclimatic regimes (arid and humid). Though linkages between C solubility and MAOM %C are stronger in humid soils, there is still highly significant relationship between C solubility and MAOM %C in arid soils.

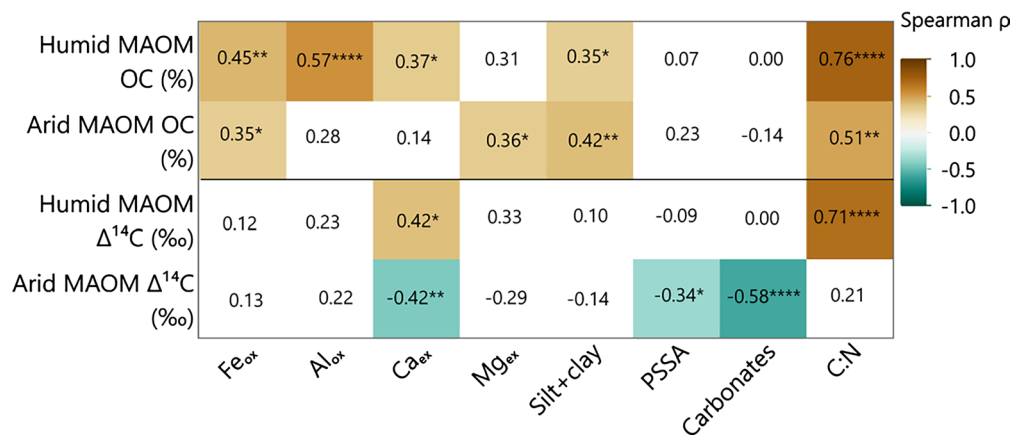


Fig. 4. Humid and arid soils differ in the soil properties associated with MAOM C concentration and persistence. Note that there are no soil properties associated with increasing persistence in humid soils, whereas multiple properties are associated with increasing MAOM persistence in arid soils. Potential explanatory variables are on the horizontal axis, while response variables, MAOM concentration (%OC) and persistence ($\Delta^{14}\text{C}$) are on the vertical axis. Darker shades and higher absolute values in individual squares indicate stronger correlations between explanatory and response variables. White squares indicate insignificant relationships ($P > 0.05$). Specifically, the graph expresses the Spearman correlation coefficients (ρ) between potential explanatory variables and the %OC and $\Delta^{14}\text{C}$ of MAOM. P values are given within each cell. Significance is indicated as follows: * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$, **** $P < 0.0001$. Fe_{ox} = bulk soil oxalate-extractable Fe; Al_{ox} = bulk soil oxalate-extractable Al; Ca_{ex} = bulk soil exchangeable Ca; Mg_{ex} = bulk soil exchangeable Mg; silt+clay = silt and clay percent by weight; PSSA = particle-specific surface area; Carbonates = bulk soil estimated carbonate content based on % inorganic C by weight; C:N = molar ratio of C to N.

water-soluble C itself was most strongly connected to MAOM concentration (Fig. 3), especially in humid soils where greater moisture may allow for increased interaction of inputs with mineral surfaces. These relationships suggest an important mechanistic role for moisture availability and root-derived soluble C in the regulation of MAOM abundance.

The pool of water-soluble OM in soils is generally considered hydrologically active (moving through the profile) and/or bio-available (32), and interacts directly with mineral surfaces and MAOM. In this study, water-soluble OM molecular composition and decomposition status differed strongly between humid and arid soils (SI Appendix, Table S2). Water-soluble OM in humid soils is derived from more recently fixed photosynthates and contains more microbially derived compounds than that of arid soils. In contrast, the water-soluble fraction of OM in arid soils is more biodegraded and contains more aromatic and high molecular weight compounds. Consistent with the idea that there is significant interaction and exchange between the soluble OM and MAOM pools (e.g., ref. 33), trends in MAOM molecular composition mirrored those of the water-soluble OM fraction. Arid MAOM ranked higher in decomposition/degradation status than humid MAOM across a spectrum of metrics (SI Appendix, Table S2). The MAOM in the surface horizons of arid soils was also less abundant and more persistent than in humid systems (A horizon %C, $P < 0.001$ & $\Delta^{14}\text{C}$, $P < 0.05$, respectively, SI Appendix, Table S1). This suggests that these fundamental differences in decomposition status, abundance, and persistence of MAOM between humid and dry systems can be explained within a conceptual framework that links MAOM cycling to moisture-dependent production and transport of fresh substrates at depth and within the soil matrix.

We propose that in humid soils, the availability of excess moisture leads to significantly higher NPP (humid versus arid soils, $P < 0.001$; SI Appendix, Table S1), resulting in increased rates of substrate inputs and transport of these inputs vertically and laterally. This finding is consistent with a higher contribution of fresh inputs to water-soluble OM in humid soils [based on a higher Freshness Index; (34, 35)], as well as higher MAOM C:N, and lower MAOM decomposition status (SI Appendix, Tables S1 and S2). This elevated level of fresh inputs in humid systems may allow for a greater exchange of C and nutrients from mineral surfaces through

a priming mechanism, whereby more labile and/or lower molecular weight organic acids displace existing MAOM (4, 36). However, higher levels of inputs may also sustain a larger pool of MAOM, possibly through the “microbial carbon pump” mechanism in which labile inputs drive greater production of microbial biomass (37), reflected in the higher concentrations of microbial leachates found in the soluble OM pool in humid soils [Fluorescence Index; (38, 39); SI Appendix, Table S2]. This balance between priming and biomass production may be directly dependent on the composition, function, and carbon use efficiency of the microbial community (37, 40, 41). In fact, increasing evidence of the importance of considering microbial diversity in soil C cycling is leading to the development of both conceptual (42) and mathematical (43) models that explicitly include microbial traits. These OM transformation processes create strong links between MAOM abundance and persistence directly driven by the vertical and lateral transport of labile inputs, with transport being a product of moisture availability. In humid systems, a continuous supply of fresh inputs, transport through the soil matrix, and interaction with mineral surfaces, allows for a steady and uniform turnover of the MAOM pool, reflected in the strong linkage between abundance and persistence (Fig. 2 and SI Appendix, Fig. S6) and lower biodegradation status (SI Appendix, Table S2) of this MAOM in comparison with arid soils.

In arid soils, lower NPP and shallower depths of moisture penetration limit the interaction of soluble OM with MAOM and microbial communities. As such, decomposition is more limited by depth of water penetration (water may not reach very deep in the profile), time (soils may rapidly dry following a wetting event), and spatial separation between substrate and microbe [lack of moisture prevents mobilization of substrates; (44)]. This creates a system in which partially degraded OM and less energetically favorable substrates (high molecular weight and aromaticity) persist within the soil matrix for long periods of time. These contrasts are supported by differences in the aromaticity and molecular weight of water-soluble OM (SI Appendix, Table S2). In these arid soils, microbial communities may have to make rapid use of smaller and more ephemeral inputs, and in fact have been shown to preferentially decompose plant C in drier soils versus SOM in wetter soils (45). The energetically unfavorable nature of the soluble OM pool in these soils may additionally

promote longer persistence of MAOM due to the lack of low molecular weight organic acids associated with C exchange at mineral surfaces (4) or the nutrients needed to prime the microbial C pump (5, 37). This results in an only partially degraded, but stable, pool of MAOM in arid soils—lower in abundance but seemingly higher in persistence than OM in humid soils—and a disconnect between MAOM abundance and persistence (Fig. 2 and *SI Appendix*, Fig. S6).

Moisture Availability Drives Divergence in Soil Physicochemical Properties Regulating MAOM Abundance and Persistence.

The abundance and persistence of MAOM is a reflection of the balance between biotic (e.g., decomposition) and abiotic (e.g., mineral sorption) processes. Substrate quality may influence the carbon use efficiency of decomposer communities (46), or the propensity of the OM to bind to mineral surfaces (33). Mineral surface reactivity and the availability of reactive surface area may influence the overall size of the MAOM pool. Meanwhile, soil pH often determines the relative importance of different metals and mineral phases for the retention of MAOM (8, 12, 33, 47, 48). We assessed the explanatory power of soil physicochemical properties linked to substrate quality and mineral reactivity as predictors of MAOM abundance (%C) and persistence [$\Delta^{14}\text{C}$ (‰)] (Fig. 4).

Based on the existing literature and our measures of MAOM degradation status, C:N is generally thought to be representative of one aspect of substrate quality, with higher C:N substrates tending to be fresher and less degraded (49–51). After depth, the C:N of the MAOM had the highest explanatory power for abundance in humid soils (Fig. 4). This is consistent with a conceptual framework in which MAOM accretion is linked to supply and transport of fresh substrate in humid soils more than sorptive capacity of the mineral matrix. In arid soils, higher C:N was also associated with increasing MAOM abundance but to a lesser degree. Additionally, in humid soils, lower persistence was strongly associated with increasing C:N. This again suggests a dominant role of fresh substrate inputs in driving lower persistence and a MAOM pool with a lesser degree of degradation. These trends are consistent with recent work illustrating the strong dependence of soil respiration on net inputs and rainfall at the global scale (52).

Historically, texture has been one of the favored variables associated with the regulation of the SOC cycle in modeling efforts (e.g., ref. 53). However, recent empirical work has suggested a divergence in the relative importance of organo-mineral stabilization mechanisms between arid and humid soil systems. Though soil texture remains an important soil property in the context of soil C cycling, specific bonding mechanisms, associated with an abundance of reactive Fe and Al phases in humid soils and base cations in arid soils, are now thought to play a dominant role in determining soil C abundance and persistence (12, 21, 23). This divergence in stabilization mechanisms is directly related to differences in moisture availability and shows a threshold response at the point where MAP equals PET. This threshold in moisture availability also corresponds to a shift in soil pH, from acid in humid soils, to alkaline in arid soils (Fig. 1A). Increased weathering rates due to increased moisture availability and transport in humid soils creates an abundance of secondary Fe and Al phases of varying crystallinity that can form strong inner-sphere bonds with organics. In arid soils, historic moisture limitations on mineral weathering result in soils rich in divalent base cations (Ca, Mg) that create cation bridges between mineral surfaces and organics (Fig. 1B). Recent work has illustrated the importance of this bifurcation in stabilization mechanisms for C stocks in bulk soils (12) and the abundance of C associated with Fe and Al phases (21). We explored the explanatory power of these physicochemical

properties for the abundance and persistence of the MAOM pool, specifically.

Consistent with the emerging paradigm of Fe/Al versus Ca/Mg stabilization, arid and humid soils showed divergence in the relative importance of soil physicochemical properties associated with variation in MAOM abundance (Fig. 4). Extractable Fe and Al pools (oxalate-extractable Fe and Al) were strongly associated with increasing MAOM abundance in humid soils, but less so in arid soils. In accordance with historical and contemporary understanding of soil texture's influence on the SOM cycle (e.g., refs. 12 and 54), increasing silt+clay was associated with increasing MAOM abundance in arid soils. However, surprisingly, divalent base cations (exchangeable Ca and Mg), specifically associated with increasing SOM abundance in circum-neutral to alkaline soils (12, 23, 55), were associated with increasing MAOM abundance in both systems. These relationships among soil properties and C abundance were generally consistent with previous work. However, none of the physicochemical variables examined were found to promote higher persistence of MAOM in humid soils. This lends additional evidence to a conceptual model in which turnover of MAOM in humid soils is driven more by the rate and mobility of inputs rather than limits on decomposer access (52).

In arid soils, the concentration of inorganic C (as a proxy for calcium carbonate content) was by far the best predictor of MAOM persistence. In that the depth and concentration of accumulated carbonates is partially a function of moisture availability over time (56, 57), inorganic C accumulation may effectively be an integrator of long-term climate conditions in these drier systems. Alternatively, because carbonates can dissolve and re-precipitate on mineral surfaces, it may be possible that OM co-precipitates with carbonates or becomes entrained in carbonate concretions. Such a process would have the potential to stabilize organics against biodegradation over extremely long time periods.

Exchangeable Ca was a significant explanatory variable for MAOM persistence in both systems, but with an equal and opposite influence. Higher Ca concentrations in humid soils were associated with decreasing persistence, possibly due to the positive influence of base cations on productivity and pH buffering in wetter forested systems (58). As with abundance, increasing persistence in arid soils was associated with increasing Ca, likely due to the same cation bridging mechanisms associated with increasing MAOM abundance.

These findings illustrate fundamental moisture-dependent differences in the drivers of MAOM stabilization and destabilization between humid and arid soils (Fig. 5), the implication being that moisture, though not as well examined as temperature, has a strong nonlinear influence on the stability of not just bulk SOM (21, 59, 60), but also on MAOM, which is often assumed to be less sensitive to climatic change (3, 61). The patterns observed between MAOM abundance and persistence in humid and arid soils are reflective of climate conditions over many thousands of years. It is currently unknown how quickly these patterns could change in the future, but this work indicates that any sustained shifts in hydroclimatic regime are likely to drive associated changes in the persistence of MAOM.

Implications for MAOM Stocks under Future Climate. This work illustrates a strong dependence of MAOM abundance and persistence on climate, specifically available moisture. These results suggest that MAOM is likely vulnerable to climate change, and as climate patterns shift, nonlinear responses in the stability of MAOM are possible. For example, laboratory incubations of these soils indicate that heterotrophic respiration rates show

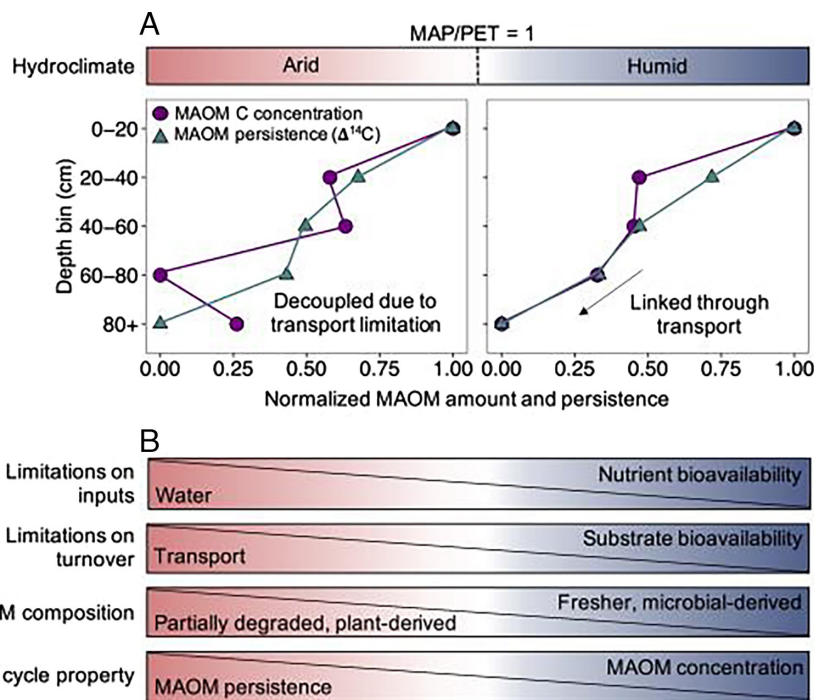


Fig. 5. In humid soils, MAOM abundance and turnover are linked through transport of new inputs to depth, whereas this linkage is absent in arid soils. These relationships are driven by differences in moisture availability versus substrate availability. (A) Points show mean organic C concentration (% log transformed) or mean radiocarbon abundance ($\Delta^{14}\text{C}$) for 20-cm depth bins. For both variables, the x-axis was rescaled (minimum = 0 and maximum = 1) within each moisture regime to emphasize depth trends. (B) Arid and Humid soils differ in the molecular composition of the soluble (dissolved OM) and MAOM fractions, as well as proposed limitations on MAOM concentration and persistence.

higher moisture sensitivity in arid subsystems, but a higher temperature sensitivity in humid subsystems (24). This implies that changes in precipitation frequency and intensity could alter mineralization rates of MAOM stocks, especially in arid subsystems where moisture limitations on decomposer access limit MAOM turnover. Conversely, under future scenarios changes in productivity (from elevated CO_2) or temperature may have a larger impact on the balance of SOM dynamics in the humid subsystems where turnover and transport of MAOM are more tightly linked. We emphasize that while our findings highlight the importance of historical climate legacies in generating patterns of MAOM abundance and persistence, additional manipulative field experiments are needed to improve projections of their rate of change under future climate scenarios. Notably, models that are used to project potential carbon cycle—climate change feedbacks do not explicitly represent divergent moisture sensitivities between hydroclimatic regimes. Both models and projections of uncertainty in SOM response to climate change should consider the moisture-driven divergence in MAOM abundance and persistence.

Materials and Methods

Soil Sampling. Soils were sampled to 2 meters or the depth of refusal from 34 sites across the NEON between 2015 and 2018; see Nave et al. (62) for a detailed description of core collection and compositing. Sites were distributed across the climatic gradient of the conterminous USA and therefore varied widely in dominant vegetation and soil characteristics (63). Five cores from each site were composited by genetic horizon, and uppermost A horizons, uppermost B horizons and lowest B or B/C horizons were selected for further characterization (*SI Appendix*, Fig. S1). Soils were sieved to 2 mm and air-dried.

Climate Data. Site-level climate data and detailed descriptions of climate metrics are published in Possinger et al. (24). Briefly, MAP and PET for each site were generated with the ClimateNA v.5.10 software package, available at

<https://tinyurl.com/ClimateNA>, based on methodology described by Wang et al. (64). Site-level netprimary productivity (NPP) ($\text{kg C m}^{-2} \text{y}^{-1}$) was determined using MODIS data (65, 66).

Soil Inorganic Chemical and Physical Characterization. Soil texture and mass SSA were determined by laser diffraction (67) following removal of OM with H_2O_2 at 50 °C overnight (68). Ammonium oxalate extractions were conducted following standard United States Department of Agriculture Natural Resources Conservation Service (USDA NRCS) protocols (69), and abundance of measured metals (Al_{ox} , Fe_{ox}) is expressed on a mass percent of bulk soil basis. Exchangeable cations (Ca_{ex} , Mg_{ex}) were extracted using ammonium acetate at pH 7.1 at the Oregon State University Central Analytical Laboratory (69).

SOM Characterization. Soils were density separated into free particulate, occluded particulate, and MAOM fractions using a sodium polytungstate solution adjusted to a density of 1.65 g cm^{-3} and sonication for the disruption of aggregates at $750 \text{ J g soil}^{-1}$ (70, 71).

Percent C and N were measured by EA-IRMS (Isoprime Ltd., Cheadle, UK). Samples containing carbonates were measured both prior to and following acid fumigation (72). Samples were prepared for radiocarbon measurement according to Vogel et al. (73) and measured for radiocarbon abundance by accelerator mass spectrometry (AMS) at the Center for AMS at Lawrence Livermore National Laboratory and the WM Keck Carbon Cycle AMS facility at the University of California, Irvine. We examined the molecular composition of the MAOM through CuO oxidation (74–76), and the molecular composition of the soluble OM fraction through EEMS (Flouromax-3 fluorometer; Jobin Yvon Horiba) and FT-ICR-MS (12 T Bruker Solarix). See Heckman et al. (13) and *SI Appendix*, Table S2 for added detail. Note that for Fig. 3, no dissolved organic C extractions were performed for 6 of the 14 A horizons for the humid sites and an additional three A horizons for the humid sites are from slightly different A horizons than were used for the broader dataset (the A horizon directly below the surface A horizon).

NEON Root Mass Data. This manuscript utilizes root mass data from the NEON data portal (25) for three soil profiles collected in the single, tower-adjacent “megapits” at each site. This root mass dataset is thus not derived from the same

locations as the soil profiles presented in this manuscript, but at most NEON sites these "megapit" root observations were taken within 100 m of the soil plots where the soil profiles were sampled for this analysis, under very similar vegetation cover, soil conditions, and topography. For profile level root abundances, the root mass per square meter to the base of the profile was used. For horizon/layer level data, an adjustment was necessary to match the depth increments of the NEON data to those sampled as part of our project. NEON root data was sampled by 10 cm depth increments, while our soils were sampled by genetic horizon. To merge the datasets, top and bottom depths of genetic horizons were rounded to 10 cm increments. All megapit root masses that fell within the upper and lower rounded bounds of the individual genetic horizons were summed to create root masses for each genetic horizon for each pit at each site. Data were averaged across the three pits to create one root mass per genetic horizon per site. No root data for the site Konza Prairie Agroecosystem (KONA) was available.

Statistical Analysis. Analyses were carried out in JMP® Pro 14.0.0 (SAS Institute). Simple linear regression was used to examine the relationships among MAOM %OC, MAOM $\Delta^{14}\text{C}$ soil depth (cm), root mass (g), and DOC (mg per g soil C). Spearman correlation matrices were constructed for the relationship between MAOM %OC and $\Delta^{14}\text{C}$ soil physicochemical properties and climate variables (Fig. 3). Specifically, the JMP Add-In, Correlate Y by X, was used to create both parametric and nonparametric correlations and *P* values. Due to the non-normal distribution of many variables, nonparametric Spearman correlations were selected as the best representation of relationships between explanatory and response variables. Because many potential explanatory variables were tested for each response variable, *P* values shown in Fig. 3 were adjusted for multiple comparisons following Benjamini and Hochberg (77), which reduces false positives and minimizes false negatives.

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Data, Materials, and Software Availability. All study data are included in the article and/or *SI Appendix*. Some data is additionally available online in a publicly accessible database (<https://doi.org/10.5281/zenodo.7552468>) (78).

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