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**Soil organic matter in the Peruvian Andes**

*Unravelling factors controlling soil organic carbon distribution and the underlying organic matter stabilization mechanisms*

Yang, S.

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## Chapter 6

### Synthesis

The large soil organic carbon (SOC) stocks in the alpine grasslands of the Peruvian Andes are crucial to ecosystem services and global carbon (C) dynamics. However, few studies have focused on how soil formation and environmental factors affect SOC stocks and the underlying mechanisms controlling the SOC stability in the Peruvian Andes. To address these knowledge gaps, the objectives of this thesis were: (1) to quantify the SOC stocks and to find key factors controlling the SOC spatial patterns, (2) to have insights into the underlying mechanisms of soil organic matter (SOM) stabilization, (3) to investigate SOM stabilization controlled by aggregates as affected by precipitation and lithology, and (4) to investigate potential relationships between SOM molecular composition and SOM stabilization in the studied alpine grasslands of the Peruvian Andes. Major conclusions on SOC stocks and SOM stabilization are synthesized in this chapter. Implications and possible future research are presented in the final part of this chapter.

### 6.1. Main conclusions

#### 6.1.1. SOC stocks and significant predictors

In the wet site, the SOC stocks estimated using 69 samples were  $215 \pm 21 \text{ Mg ha}^{-1}$  (Chapter 2), which is higher than the global average level reported by Lal (2004). In the wet site, the SOC stocks were  $405 \pm 42$  for the limestone soils (LSs) and  $226 \pm 6 \text{ Mg ha}^{-1}$  for the acid igneous rock soils (ASs). In the dry site, the SOC stocks were  $153 \pm 27$  for the LSs and  $172 \pm 13 \text{ Mg ha}^{-1}$  for the ASs. The LSs and the ASs in both wet and dry sites had higher SOC stocks when compared to corresponding soil groups (Batjes, 2014). When compared to other alpine grasslands of the Andes, SOC stocks are higher in Páramo soils of the Ecuadorian and Venezuelan Andes with higher precipitation rates (Sarmiento and Bottner, 2002; Tonneijck et al., 2010). In contrast, SOC stocks are lower in Southern Peru and Bolivia with drier climate conditions (Muñoz-García and Faz-Cano, 2012; Rolando et al., 2017a; Zimmermann et al., 2009). When compared to soils from

other alpine grasslands (Garcia-Pausas et al., 2007; Yang et al., 2008), our soils also have higher SOC stocks.

These findings suggest that precipitation drives SOC accumulation in Andean soils. This is further supported by our observation that SOC stocks were higher in the wet site when compared to the dry site for both LSs and ASs (Chapter 4). This trend coincides with the general observation that SOC stocks are high in cool and humid environments (Wiesmeier et al., 2019). The observed SOC stock pattern indicates that precipitation is crucial to the distribution of SOC stocks in the Andean region. In general, precipitation controls SOC sequestration through primary productivity, OM input and microbial decomposition (Wiesmeier et al., 2019). In addition, precipitation can also influence OM stabilization by controlling soil mineralogy through weathering processes (Doetterl et al., 2015b; Homann et al., 2007). Lithology is a key factor controlling the SOC stocks, as lithology became the significant predictor when soil depth and moisture were controlled as conditional variables (Chapter 2). The effects of lithology on SOC stocks can be explained by differences in soil mineralogy, which affect OM stabilization related to soil aggregates and mineral surfaces (Kleber et al., 2015; Lützow et al., 2006).

Compared to precipitation and lithology, other soil formation and environmental factors were less important to predict the SOC stock patterns (Chapter 2). It is surprising that land use was not a significant predictor for SOC stocks. This may be closely related to the special land use strategy in which a rotation in the order of cultivation, land set-aside and grazing were repeated every 3-5 years. This type of land use pattern keeps SOC stocks in a dynamic balance (Chapter 2). Grazing intensity only had significant effects on the SOC stock of the top 10cm, which suggests that its impacts on the SOC input and decomposition processes might be more severe for the topsoil than for the subsoil (Piñeiro et al., 2010). SOC stocks of the top 10cm were stimulated by increasing altitudes (Chapter 2), which can be explained by declined microbial decomposition due to lower temperatures (Du et al., 2014). The limited importance of topographical factors (slope position and slope angle) might be attributed to the homogeneous solar radiation and soil wetness for different aspects as the results of the tropical latitude (9 °S) of the study area.

### 6.1.2. OM stabilization mechanisms

OM occluded in aggregates had a limited contribution to SOM stabilization in our studied soils, as indicated by (1) limited stimulation of SOC mineralization after the incubation with crushed aggregates compared to intact aggregates, (2) unclear correlations between aggregate stability and SOC contents and stability, and (3) inconsistent patterns between aggregate-size distribution and SOC stability (Chapter 4). For the ASs, the limited contribution of aggregates in SOM stabilization can be explained by the lack of large-sized aggregates. For the LSs, it might be a consequence of the strong adsorption of OM on mineral surfaces. As OM occlusion in aggregates only had a minor contribution to OM stabilization, OM adsorption on mineral surfaces is proposed to be the dominant stabilization mechanism (Chapter 4).

In the wet site, the stabilization mechanisms of SOM were different between LSs and ASs, although their SOC stability was not significantly different. In the wet-ASs, SOM was stabilized by complexation with and/or adsorption on Fe and Al (oxides) rather than clay contents, allophanes or Al toxicity (Chapter 3). In the wet-LSs, OM stabilization was promoted by Ca-induced cation bridges in addition to interactions between OM and Fe and Al (oxides). Similar to our results, SOM has also been reported to be stabilized by Fe and Al (oxides) in alpine grasslands of the Andes (Buytaert et al., 2006b; Podwojewski et al., 2002; Tonneijck et al., 2010). The wet-LSs had significantly higher SOC stocks than the wet-ASs, which can be explained by OM stabilization being exclusively controlled by  $\text{Ca}^{2+}$  bridges in the wet-LSs (Chapter 3). Our results suggest a shift in OM stabilization from adsorption on Fe and Al (oxides) to the presence of  $\text{Ca}^{2+}$  bridges, with increasing pH values driven by lithology (Chapter 3). Similar shifts in OM stabilization mechanisms with changes in lithology and pH have been widely reported in other soils (e.g. Heckman et al., 2009; Kaiser et al., 2011; Masiello et al., 2004).

In the dry site, the LSs had lower SOC stocks and lower SOC stability compared to the ASs (Chapter 4). The lower SOC stability in the dry-LSs might be explained by the weak adsorption of SOM on mineral surfaces due to low pH values (Heckman et al., 2009). The lower pH suggests the weak interaction between OM and mineral surfaces through polyvalent cation bridges because  $\text{Ca}^{2+}$  is replaced by  $\text{H}^+$  with lower pH values (Jenny, 1941). Furthermore, the positive correlations between SOC mineralization rates and C/N ratios exclusively found

in the dry-LSs suggest that the decomposition of OM in the dry-LSs was more dependent on OM quality in comparison to other soils.

### *6.1.3. Effects of precipitation and lithology*

When precipitation levels decreased, SOC stocks and stability decreased in the LSs but did not clearly change in the ASs. The different responses between LSs and ASs might be attributed to differences in soil mineralogy (calcareous vs. acidic bedrock) related to weathering processes and OM input that is driven by precipitation (Chapter 4). By contrast, aggregate-size distribution was largely controlled by lithology rather than by precipitation. This suggests that soil aggregation is mainly regulated by soil mineralogy in our studied area (Chapter 4).

We observed lower SOC stability and lower soil pH values with lower precipitation amounts in the LSs. Although the lower pH values in the dry-LSs compared to the wet-LSs are not common from a pedogenetic point of view, they coincide with the higher belowground OM input in the dry-LSs. This is supported by: (1) the larger contributions of  $\alpha$ ,  $\omega$ -dioic acids,  $\omega$ -hydroxyl alkanolic acids and long-chain fatty acids, and (2) the depletion of these compounds after the incubation (Chapter 4). The higher belowground OM input in the dry-LSs caused more intensive acidification and lower pH values when compared to the wet-LSs. This further weakens the capacity of mineral surfaces to stabilize OM in the dry-LSs. A potential explanation for the higher OM input is that plants need more developed root systems to adapt to the lower precipitation in the dry-LSs. In contrast, the ASs had no clear difference in pH values between wet and dry sites. This might be explained by an absence of the effects of OM input on soil acidification in soils formed on the already acidic bedrocks.

Recent studies indicated that climate factors, including precipitation, control OM stabilization by regulating soil mineralogy through weathering processes (Chaplot et al., 2010; Doetterl et al., 2018, 2015b). Our results support this view and suggest that interactions between precipitation and lithology are crucial for the stabilization of OM. Furthermore, OM input controlled by precipitation might play an important role in regulation weathering process in the LSs rather than in the ASs.

### *6.1.4. Molecular composition of SOM*

Although the method of TMAH-pyrolysis-GC/MS does not yield quantitative data, we can still compare our results with other studies using similar methods. We observed large fractions of lipid-derived compounds, which are consistent with the results of Nierop et al. (2007) in alpine grassland soils in the Ecuadorian Andes. In contrast, soils outside alpine grasslands of the Andes had smaller fractions of lipid-derived compounds (Barré et al., 2018; Brock et al., 2019; Nierop and Verstraten, 2003). In addition, we found smaller fractions of pyrolysis products derived from lignin and polysaccharides (<10%) in most of our soil samples (Chapter 5). This is consistent with the study of Nierop et al. (2007) conducted in alpine grasslands of the Ecuadorian Andes. After a 76-day incubation, unsaturated FAs were depleted, whereas short-chain saturated FAs (e.g. C16:0 and C18:0) accumulated. In addition, the depletion of unsaturated FAs was strongly correlated to SOC mineralization rates. This suggests that depletion of unsaturated FAs is likely attributed to the microbial mineralization.

The results indicated that double bonds (saturated vs. unsaturated) and carbon chain length are two major factors controlling FA stability against decomposition. Unsaturated FAs were more vulnerable than saturated FAs, which is unlikely explained by their stabilization by occlusion in aggregates. Instead, the vulnerability might be explained by their chemical recalcitrance and/or association with mineral surfaces (Chapter 5). Long-chain FAs had lower stability than short-chain FAs. A possible explanation for this is that short-chain FAs received more protection by occlusion in aggregates compared to long-chain FAs. Only a few other studies have examined the effects of double bonds and carbon chain length on FA stabilization in soils (e.g. Dent et al., 2004; Moucawi et al., 1981). Microbial transformation of FAs was observed during the 76-day incubation. However, the microbial transformation had no significant effect on the controls of double bonds and carbon chain length on the stability of FAs.

Although chemical recalcitrance is recently considered less important in SOM stabilization than previously believed (Lehmann and Kleber, 2015; Schmidt et al., 2011), the chemical composition of SOM can potentially affect their stabilization by interacting with the soil matrix (Angst et al., 2018b, 2017). For this, our results provided evidence to support that the chemical properties of FAs influence their interactions with the soil matrix and further control their stabilization. Finally, our results also indicate that soil types and soil horizons also influence the effects of double bonds and carbon chain length on FA stabilization. This suggests that

more studies are required before generalizing our findings to other soils (Chapter 5).

## **6.2. Implications**

### *6.2.1. Implications for SOC sequestration in the Andes*

Land use change is well-known to affect SOC stocks. A change from cropland to grassland generally causes SOC accumulation, whereas the opposite results in SOC depletion (Poeplau et al., 2011). A possible explanation is that aggregates, which promote SOM stabilization, are destroyed by agricultural activities such as cultivation and tillage (Guo and Gifford, 2002). However, our results showed no significant differences in SOC stocks between different land use types. Similar results have been reported in the Peruvian Andes by Rolando et al. (2017b) and Zimmermann et al. (2009). A possible explanation is that the aggregation had a limited role in SOM stabilization in our study (Chapter 4). Thus, SOC stocks were not significantly affected by aggregate destruction caused by agricultural activities. A second explanation is that the local land use pattern, in which a rotation in the order of cultivation, land set-aside and grazing was applied every 3-5 years, might keep the SOC stocks at a high level and in a dynamical balance. This applied land use pattern might give a potential solution for how to sustain high SOC stocks in alpine grassland of the Peruvian Andes with intensive agriculture productions.

SOC stocks and SOC stability in the LSs were more sensitive to precipitation changes when compared to the ASs. We previously assumed that the SOC stocks in the ASs were more vulnerable to the potential change in precipitation compared to the LSs because the LSs had better soil structure. However, this assumption is not supported by our data because SOM stabilization was not clearly controlled by soil aggregation. Our findings suggest that the LSs may lose more SOC stocks than the ASs when the climate becomes drier. This suggests that lithology should be considered when evaluating potential feedbacks of SOC storage and stability to global change, as well as when making management plans to sustain ecosystem services in the Peruvian Andes.

### *6.2.2. Implications for SOC stocks estimation*



The results of Chapter 2 highlighted the importance of using the entire soil profile to estimate SOC stocks instead of using a constant soil depth. In general, SOC stocks are estimated using constant soil depths (e.g. Doetterl et al., 2015; Du et al., 2014). However, this increases the risk of inaccurate estimations of SOC stocks and potential effects of environmental factors on SOC stocks, because (1) deep soils may also contain large SOC stocks, and (2) SOC stability may be different between topsoil and subsoil (Batjes, 2014; Wiesmeier et al., 2012). Our results showed that SOC stocks were underestimated and the effects of soil formation and environmental factors on SOC stocks were overestimated when using a constant soil depth (Chapter 2). Thus, we confirm the necessity to include the entire soil profile to avoid bias in the estimation of SOC stocks.

### *6.2.3. Implications for SOM stabilization mechanisms*

The results of this thesis indicated that SOM stabilization in the Andean grasslands was controlled both by lithology and precipitation in relation to SOM input. The lower SOC stocks and stability in the dry-LSs, when compared to the wet-LSs, were explained by the weaker interaction between OM and mineral surfaces via  $\text{Ca}^{2+}$  bridges as controlled by lower pH values (Chapter 4). The lower pH values were likely further controlled by intensive soil acidification caused by larger belowground OM input. Doetterl et al., (2015b, 2018) reported that SOC stability is controlled by soil mineralogy, which is further governed by climate factors. In addition to the mineralogical control, our results suggest that OM input from the vegetation might also play an important role. Thus, it is important to consider the effects of biotic factors like vegetation and OM input for future research on mineral-controlled SOM stabilization due to the interactions between biotic factors and weathering processes. However, a very limited number of studies have reported OM input in the alpine Andes (e.g. Oliveras et al., 2014).

When compared with other studies using the TMAH-pyrolysis-GC/MS, our results suggest that it is likely that soils of the Andean highland, including soils in our study, have higher aliphatic compounds and lower contributions of lignin and polysaccharides (Barré et al., 2018; Brock et al., 2019; Nierop et al., 2007; Nierop and Verstraten, 2003). Thus, an open question remains whether the large SOC stocks in the Andean highland are associated with the chemical composition of SOM. It is important to understand the underlying mechanisms related to SOM input, decomposition and stabilization that controlled by biotic and abiotic factors.

Recently, chemical recalcitrance is considered less-important in SOM stabilization than previously thought. Instead, increasing evidence indicates that SOM stabilization is controlled by the interactions between OM molecules and the soil matrix that reduce OM decomposition rates (Dungait et al., 2012; Lehmann and Kleber, 2015; Schmidt et al., 2011). However, to the author's point of view, this does not necessarily mean that the molecular composition of SOM is not important for SOM persistence. Molecular composition not only determines the chemical recalcitrance of SOM but also affects the interactions between SOM and the soil matrix. Based on the SOM adsorption models, molecular composition largely controls the affinity of SOM to the association sites of mineral surfaces (Kleber et al., 2007). Recent studies also indicated that SOM composition had influences on SOM stabilization by interacting with the soil matrix (Angst et al., 2018b, 2017). Furthermore, the results of Chapter 5 showed direct evidence for this and gave better insights into the effects of SOM molecular composition on SOM persistence. Thus, SOM composition may still be important because of the potential effects of molecular composition on SOM stabilization by interacting with the soil matrix.

### **6.3. Future research**

The research involved in this thesis has improved the understanding of mechanisms and processes affecting the storage, distribution, stabilization and molecular composition of SOM in the studied areas of the Peruvian Andes. However, more studies are needed to gain further understanding of SOC sequestration and stabilization in the Peruvian Andes and in general.

With regard to SOC sequestration, very few studies have focused on C input of the Andean grasslands (e.g. Oliveras et al., 2014). As C input is an important factor for the SOC sequestration (Wiesmeier et al., 2019), studies focusing on the quality and quantity of input C may help understand SOC sequestration better in the Andean alpine grasslands. Furthermore, future studies could focus on the application of the knowledge highlighted in this thesis to improve the soil management and to sustain the ecosystem services in the alpine grasslands of the Peruvian Andes.

For SOM stabilization controlled by occlusion in soil aggregates, we found a small contribution of aggregates in SOM stabilization, even in LSs with good soil structure. The small contribution of aggregates needs further investigation. Future studies can also gain insights into the microbial activity, SOM decomposition and SOM stabilization in soil aggregates.

For the chemical composition of SOM, more open questions can be studied. One question is why the soils in alpine grasslands of the Andes have larger abundances of lipids compared to soils from other regions (e.g. Barré et al., 2018; Brock et al., 2019; Nierop and Verstraten, 2003). The underlying mechanisms of the selective preservation of lipids can be an interesting research focus. In addition, the results of Chapter 5 cannot explain whether the vulnerability of unsaturated FAs compared to saturated FAs is attributed to the differences in chemical recalcitrance or in the interactions between FAs and mineral surfaces. Further studies can apply a combination of adsorption and decomposition experiments to investigate the affinity of saturated and unsaturated FAs to the mineral surfaces and their stability against decomposition. Finally, as we found that soil types and horizons have influences on the controls of FA properties on their stabilization, more studies are needed before generalizing our findings to other soils.