Dear Dr. Subke,

thank you very much for your comprehensive response to our reply on the referee and short comments in the Biogeosciences Discussions forum.

Please find attached the revised manuscript incl. supplements as well as the marked-up versions to track the changes which were made by us.

As already discussed in our reply, in the revised manuscript the covarying moisture gradient became a central part of the discussion and served as explanatory factor for most of our observations. Thus, we avoid an overinterpretation of the data with respect to the role of the fungal community in particulate OM decomposition. We completely rephrased the conclusions section and highlighted the importance of the moisture gradient for studies dealing with the OM dynamics along salinity gradients. Also the abstract was modified with regard to that point.

Moreover, we integrated the many helpful suggestions by the referee and short comments. Therefore, please refer to our previously uploaded reply (which is again attached to this document) and the changes which were made in the revised manuscript.

Best regards,

Norbert Bischoff, on behalf of all co-authors
Response to comments of Referee #1

Dear Referee #1 (R#1),

Thank you for taking your time to go through our manuscript and give critical comments and advices. We agree with you that we partially overinterpreted the results (deriving from PLFA measurements the functionality and resilience of the microbial community) and that we should discuss some data more carefully. After discussing your comments we decided to add a fourth hypothesis to the existing three: (i) soil OC stocks decrease with increasing salinity, (ii) the proportion and stability of particulate OM is larger in salt-affected than in non-salt-affected soils, (iii) sodicity reduces the proportion and stability of mineral-associated OM, and (iv) fungi : bacteria ratios, as derived from PLFA measurements, decrease along the salinity gradient. By that, we connect our objectives with the hypotheses, as you suggested. Moreover, we are not going to relate the PLFA data directly to the proportion and stability of particulate OM, as this in fact would result in an overinterpretation. Setting up a fourth hypothesis allows for a separate discussion of the microbial community data. We are going to discuss the possibility that a functionally diverse fungal community contributed to the progressive decomposition of particulate OM. In addition, we will clearly state that a similar water stress along the salinity gradient could be responsible that we have not found a different alteration of OM along the transect. In the introduction we will remove the explanation on Solonetz soils, but focus on salinity and sodicity, as these were the main issues of our study. By that we follow many of your helpful advices. We are going to await the decision of the editor and, if positive, work on the manuscript revision.

General comments

R#1: Considering the increasing extent of salt-affected soils, this MS deals with an important and timely issue. Understanding the carbon dynamics in salt-affected soils is an interesting topic within the scope of Biogeosciences.

The biggest problem of the presented study is that the salinity gradient was confounded by a large difference in soil moisture between the saline sites and the non-saline site. This is discussed by the authors (p. 12 l 37-p.13 l 2), but the importance is under stated. Soil moisture has an enormous impact not only on plant productivity, but on decomposition processes, which are inhibited strongly by lack of water (Manzoni et al., 2012). The possibility cannot be excluded that the alteration of OM was found to be similar at saline and non-saline sites, because decomposition was inhibited by lack of water at the non-saline site, especially if the non-saline site is drier than the saline site throughout the course of the year. It is therefore not possible to conclude that microbial activity was resistant to salinity in the studied soils, since it could have been inhibited by low water availability at all sites, caused by different mechanisms. As a result, a major revision of the discussion is needed. As a suggestion, it could make sense to use water potential as a parameter, to allow for an easier comparison between sites and distinguish between the effects of salinity and moisture.

Authors (A): We agree that the salinity gradient was possibly confounded by a large difference in soil moisture. Hence, the interpretation of the data is currently not straightforward. After evaluating the data for the first time, we were aware of the problem and intended to calculate the water potential of the soils, as you have suggested. Thereby, we faced the problem, that we could not measure the matric potential directly by use of soil water retention curves, since we had no undisturbed soil cores of the studied soils. Thus, we had to use Pedo-Transfer-Functions (PTF’s) which estimate the matric
potential via soil parameters, such as soil texture, bulk density, organic carbon content, and actual soil water content. Such a PTF was proposed in Vereecken et al. (1989) with the Van Genuchten model. Another possibility is to calculate the model parameters for the Van Genuchten model via the software "RETC". Both, the use of the PTF’s in Vereecken et al. (1989) and the use of "RETC" have not yielded plausible results for our soils. This might be due to the fact, that the PTF’s were empirically developed for temperate soils without influence of salinity. As a consequence, we cannot calculate the matric potential for the soils in our study and, thus, neither the water potential. However, this is not limiting the significance of our study, since (i) it is a natural phenomenon that salinity co-varies with soil moisture in the study area, thus, our transect represents the occurring natural conditions, (ii) the soil moisture measurement given in Table 2 represents just a "snapshot" at the moment of soil sampling and not a mean value during a longer period of time. To draw conclusions about the possible effect of the matric potential or water potential, respectively, on processes like soil OM decomposition, we would need to measure these parameters for a longer period of time. Nevertheless, we agree with you that the effect of soil moisture is a critical aspect in the manuscript and should be discussed more extensively. In the revised manuscript we intend to revise the discussion thoroughly, particularly with respect to the effect of matric potential vs. osmotic potential and the overall water potential on soil OM decomposition. In particular we are going to state that it is possible that we have not found differences between the soils with respect to soil OM decomposition, because of a similar water stress/water potential in all soils.

R#1: Another serious issue is that the dataset is very limited, to the extent that statistical hypothesis testing was not possible. Effectively, the number of independent samples along the salinity gradient is only 3.

A: The number of independent samples along the salinity gradient was 3 or 4 (for the Non-sodic Solonchaks), respectively. Thus, statistical hypothesis testing was not possible, as noted on p. 8 l. 34-36 of the manuscript. However, this does not mean that the dataset is limited. We decided to conduct an in-depth analysis by measuring many soil parameters per soil profile and relate them to each other in order to reveal processes which take place within the soil. This was done in many previous studies (Fierer et al., 2003; Kemmitt et al., 2008; Shen and Bartha, 1996). By that, we actually obtained a very large and detailed data set. For example, only by use of isotopic data ($^{13}$C, $^{14}$C) and neutral sugar measurements in combination with PLFA we could reveal that POM was not distinctly altered in the studied soils, maybe due to a functionally diverse and resilient microbial community, which is capable of decomposing POM at a similar rate in salt-affected and non-salt-affected soils. As you have mentioned in the previous comment, in the revised manuscript we will add to this explanation, that it is possible that a lower soil moisture in the non-salt-affected soils has led to similar POM decomposition in the salt-affected and non-salt-affected soils.

R#1: The manuscript is generally well written, if a bit lengthy in some areas (Results) and underdeveloped in others (discussion). However, there are some sentences that contain clumsy English structures.

A: In the revised manuscript we are going to shorten the results section (e.g. the part about soil mineralogy and by generally not repeating the numbers from the tables too extensively). On the other hand, we are going to work on the discussion section including more detail and discussing also controversial positions, such as the fact that soil moisture could have a crucial impact on soil OM
decomposition along the transect. Sentences that contain clumsy English structures are going to be revised.

Specific comments

R#1: p2 l17-19: While you measured the microbial community composition, I do not understand how you derive from the results that the functioning and capability to decompose of the community was virtually unaffected by salt. This seems like an overinterpretation of the data.

A: As mentioned above, we agree that this could be an overinterpretation of the data. We are going to soften this conclusion in the revised manuscript.

R#1: p3 l 6-7: This is a bit confusing, since Na+ is also a water-soluble salt. Another issue: Here you refer to Solonchaks and Solonetzes, but later in the MS you switch to sodic and non-sodic Solonchak. Naming should be consistent.

A: Na+, as such, is not a water-soluble salt but a monovalent cation. To make the sentence clearer, we may change it in the revised manuscript to: "Solonchaks contain high loads of water-soluble salts in general, while Solonetzes are particularly characterized by Na+ as the dominant cation on the exchange sites, irrespective of the quantity of salts." Here we distinguish between Solonchak and Solonetz to explain the difference between non-sodic and sodic. But, we agree with you, that we could shorten the explanation regarding "Solonetz" in the revised manuscript, as this particular soil type was not part of our transect.

R#1: p 3 l 136: which previous studies?

A: Thank you for this attentive note. Previous studies are for example Mavi et al. (2012), Setia et al. (2013, 2014). We are going to add this to the revised manuscript.

R#1: p 4 l 4: What is the expectation for the third objective?

A: After your comment about the "overinterpretation" of the PLFA data (microbial community composition / functioning), we decided to attenuate the conclusion on the results of the third objective. Moreover, we came to the conclusion that it is not straightforward to relate the PLFA data to the results of soil OC stocks and quantities and properties of functionally different OM fractions. Thus, we will restate our third objective to "(iii) analyse changes of the microbial community composition". This objective will be kept quite general, as to our knowledge there are no studies which have determined microbial community compositions in Solonchaks or Solonetzes so far (which we stated on p. 3 l. 34-36). We are going to include this in the revised manuscript.

R#1: p 4 l. 13-16: As a suggestion, the focus of the MS would become clearer, if the hypotheses would follow your stated objectives above.

A: In the revised manuscript we are going to integrate your suggestion. We will set up three objectives and add a fourth hypothesis regarding the microbial community composition.

R#1: p. 8. l. 26: Was plant biomass the only response variable that was tested?
A: Yes, plant biomass was the only response variable that was tested, because this was the only parameter for which we had a sufficient number of samples/replicates. This was because it is a parameter which is easy to measure without the need of lots of time and money.

R#1: p.8 l.37: By “involved the consideration of several response variables”, do you mean multivariate statistics? It is an unclear sentence.

A: In the revised manuscript we will change the sentence to: “Data of PLFA and neutral sugars were analyzed by PCA in order to consider multiple response variables. Confidence regions (68%) for the group centroids of the independent factor variables were added to the biplots.”

R#1: p.9 l. 27-36: This section is never clearly brought up in the discussion and I am not sure if these results contribute important information.

A: We determined the soil mineralogical composition principally because of two reasons: (i) to characterize the mineralogical composition of water-soluble salt minerals in the salt-affected soils, and (ii) to determine the clay mineralogical composition particularly with respect to expandable clay minerals, such as smectite, as these affect the physical properties of sodic soils crucially (see p. 6 l.4-5). The mineralogical characterization of the water-soluble salt minerals is primarily descriptive, but informative and important as we study salt-affected soils. The clay mineralogical composition turned out to be similar between the soils and therefore cannot explain differences between the soils later on in the discussion. Thus, we may move this section to the Supplements in the revised manuscript.

R#1: p. 10 l. 21: What could be the reason for decreasing d13C ratios? Leaching? This is missing a discussion. Could also be linked to the 14C increase with depth.

A: In our opinion, decreasing d13C ratios cannot be caused by leaching as the net-movement of water in the salt-affected soils is upwards. Decreasing d13C ratios, and as such increasing 14C activities, with depth could be related to a faster soil OM turnover. In the Solonchaks of our study this could occur due to the water stress in the topsoil (osmotic stress and matric stress) while the subsoil is generally wetter due to the proximity to the groundwater and a lower salt content. Hence, the conditions for microbes to process soil OM could be better in the subsoil than in the topsoil. This would explain the observed pattern in the Solonchak, but not the increase of 14C activity in the Kastanozem. Since this is very speculative, we decided to leave it out from the discussion. But we may add it to the revised manuscript with the advice that this asks for further investigation in future studies.

R#1: p. 12 l.9: I don’t see any differences in community composition between soil types. Consider changing the wording of “less pronounced”.

A: Indeed, there are differences in the microbial community composition between the soil types. Please consider the confidence regions in Fig. 6a with a larger variability on PC2 for the salt-affected soils. This corresponds to a larger variability of fungal PLFA in the salt-affected soils. Though the differences are small, they are existent and should be mentioned. However, we are going to change the wording to “small” instead of “less pronounced” in the revised manuscript.

R#1: p. 13 l.16-18: Again, since the Kastanozem was so dry, I would be careful to talk about a lack of inhibition by salinity. Were the OC stocks actually large compared with what would be expected in a steppe soil? Bring this statement into context with data from other studies.
As mentioned in a previous response to one of your comments, in the revised manuscript we are going to include a more intensive discussion on the fact that the very dry conditions in the Kastanozem could have led to a similar water stress in the Kastanozems and Solonchaks, with the respective consequences on soil OM input and soil OM decomposition. So far this was only little discussed in the manuscript (p. 12 l. 37-39, p. 13 l. 1-5). As already mentioned in the manuscript, the OC stocks of Solonchaks were large when compared to data from other studies, while the Kastanozems of the transect revealed smaller OC stocks than previously observed in other studies (see chapter “Discussion: Soil OC stocks along the salinity gradient”).

Technical comments:

R#1: p. 4 l. 22: Upslope of the lake?
A: “to about 5m above the lake”

R#1: p.9 l. 18: lowest EC1:5. Also in other places in the MS “small” should be replaced by “low”, and “large” by “high”.
A: Thank you for this correction. We are going to correct this in the revised manuscript.

R#1: p.10 l.23: Consider changing the order of Figure 3 and 4, so that it matches the first appearances in the text
A: In the revised MS, we are going to change the order of Figure 3 and 4.

R#1: p. 10 l.36: Did you mean Fig. 3?
A: Correct. We are going to change this in the revised MS.

R#1: p.15 l. 19: This led us to the conclusion.
A: Thanks for the correction. We are going to integrate this in the revised MS.

References:


Response to comments of Referee #2

Dear Referee #2 (R#2),

Thank you for taking your time to go through our manuscript and give us helpful advices and corrections.

R#2: This study aimed to understand the role of salinity in shaping soil organic matter. The study is somewhat confounded because the salinity gradient covaries with a moisture gradient. The saltiest soil is closest to the water table and had the highest moisture content while the low salinity soil was far from the water table and had much lower soil moisture. Consequently, it is not possible to separate out the effects of moisture and salinity on the soil carbon and microbial community. Despite this limitation the manuscript presents a robust dataset that is, on the whole, well contextualized. The presentation of the data is quite dense and the manuscript is made less comprehensible by the excessive use of abbreviations. The authors should work to simplify the results where data is sometimes redundantly presented in the text, tables and figures. There also seems to be an excess of supplemental data that is simply an alternate presentation of the data shown in the tables. Generally, I think the authors could do more to explain why their findings do not match those reported by others. The moisture gradient seems to be the most obvious reason to me yet this is not well discussed in the manuscript.

A: It is true that the salinity gradient covaries with a moisture gradient, which is a broadly occurring situation in the study area. Salt-affected soils are those close to the groundwater table and are thus generally moister than the non-salt-affected soils which occur at a larger distance to the groundwater table. Therefore, this should not be seen as a limitation of the study. It is just the natural association of the soils in the semi-arid steppe. We agree with you that we should discuss this in more detail in the revised manuscript. In particular we are going to include the fact that it is difficult to separate the effect of salinity and moisture on soil OM dynamics and the microbial community. We will discuss that the missing effect of salinity on the soil OM dynamics along the transect could also be explained by the covarying moisture gradient.

According to your comment we are going to delete some abbreviations, e.g. SPT. If data is redundantly presented, we will delete redundant data, e.g. the d13C ratios in Table 3 which are similarly presented in Figure 4. In our opinion there is no excess of supplemental data and it is not an alternate presentation of the data shown in the tables. Only Figure S5 and S6 are partially redundant to Figure 5, but we consider these figures as informative as they highlight precisely how the two sugars glucose and arabinose differ between the soil types with respect to plant samples and organic matter fractions. This is not possible to show in a PCA in such detail.

In the revised manuscript we are going to explain in more detail why we think that our results differ from those observed in other studies. As was already discussed, this could be due to the covarying moisture gradient, particularly the pronounced aridity in the Kastanozem, which may have led to a smaller OM input and consequently smaller OC stocks. Moreover, soil OM decomposition could have been inhibited and thus the soil OM transformation appears similar to those of the salt-affected soils.

R#2: Page 3 - 31. Suggest start new sentence, i.e. change “OM, while particulate OM” to “OM. In contrast, particulate OM”

A: Thank you for this advice. We are going to change it accordingly in the revised manuscript.
If the salt interfered with the internal standard peak how can you be sure it did not interfere with any of the other peaks?

We are very sure that the salt did not interfere with any of the other PLFA peaks, as the peaks appeared clear and with a characteristic shape. Moreover, the shape and the appearance of the peaks were similar in the salt-affected and non-salt-affected soils. This was not the case for the internal standard peak, which clearly differed in the topsoils of the salt-affected soils by a clear overlap with another unspecific peak.

Figure 3 is not referred to in the results section.

By accident, on p.10 l. 36 we referred to “Table 3” instead of “Figure 3”. We are going to change this in the revised manuscript.

Note to self salty soils have more clay and more moisture – these are factors that stabilize C

To argue that a higher clay content may contribute to a larger C stabilization in the salt-affected soils is a good point. We are going to integrate this in the revised manuscript. To discuss soil moisture as a factor that stabilizes C along the transect is, in our opinion, difficult, because the “available” soil moisture (i.e. water potential) is possibly similar along the transect, either due to a low osmotic potential (Solonchaks) or a low matric potential (Kastanozems). Thus, we would not like to integrate this into the discussion.

Can you write out SPT this is not used frequently enough to warrant abbreviation?

Yes, we are going to write it out in the revised manuscript.

Can you just refer to the loss as mobilized C, I think that would make it less confusing. I had to reread the methods to understand this part of the results.

We think it would not be correct to refer to the loss simply as mobilized C, since the mass loss in the salt-affected soils is largely due to the dissolution of C-free salts. Hence, in these soils we observe a large mass loss but only a minor loss of mobilizable C. In the Kastanozem, however, less total soil mass was lost during the density fractionation but this was associated with a larger portion of mobilized OC. Since we think that this differentiation is important, we would like to keep it in our revised manuscript.

This is a sentence for the discussion.

This is what we explained in the previous comment. We agree with you, that this sentence fits better into the discussion. Hence, we are going to move it in the revised manuscript.

Page 10 - 37 What does B.P. stand for ?Before Present?

Yes, B.P. means “Before Present”.

Figure 4 is also not referred to in the results- only the tables. Perhaps the data should not be redundantly presented in both locations?
A: Indeed, figure 4 is referred to in the results (p. 10 l. 23). However, we agree that the data on δ13C is somewhat redundant. In the revised manuscript we are going to delete δ13C values from Table 3 and refer only to Figure 4.

R#2: Figure 5 – Is there a need to show the grey dots in each panel?

A: The PCA on neutral sugars was applied on the entire data set, i.e. neutral sugar data of all three soil types and all three fractions was analyzed in one PCA. This resulted in the biplot shown in Figure 5. To highlight differences between the soils we split the biplot into three panels and indicated the fractions of each soil by different colors. The biplot shows all considered data (i.e. the entire data set); this includes the grey dots which do not belong to the particular soil type of a panel.

R#2: Page 11 – 21-33 Have you considered doing a PerMANOVA to determine if these differences in sugar composition are significant?

A: PerMANOVA is a robust tool to test multivariate data on statistical significance. However, a minimum sample size is required to obtain reliable results. As mentioned in the statistics section of the Material & Methods part, we only have 3–4 field replicates (i.e. 3–4 soil profiles per soil type). One might argue that we consider more than 3–4 samples per soil type in the PCA, but this is because the soil profiles were sampled in horizons and data of each horizon is also considered in the PCA. As these horizon samples are nested within the soil profile they cannot be treated as independent samples. If so, they would be referred to as “pseudo-replicates” and application of statistical hypothesis testing on such data would result in underestimation of p-values. Therefore we decided to refrain from a PerMANOVA and analyze the data descriptively.

R#2: Page 11 – 35 this sentence is confusing ” The relative contribution of PLFA observed within the PLFA profiles“

A: In the revised manuscript we are going to change the sentence to: “The relative proportion of PLFA on the entire data set was as follows:”

R#2: Page 12 – 5 As with sugar composition you should be able to test statistically if the sites and soil profiles are statistically distinct in terms of microbial community structure.

A: Please refer to our previous comment regarding statistical hypothesis testing on neutral sugar data.

R#2: Pag 12- 20 Given the high CV for these soil types I’m not sure that soil type is such a great predictor of carbon stock.

A: Soil type is, for sure, not the best predictor of C stocks but it is, as a single variable, possibly more precise than solely predictors such as temperature, moisture, parent material, or clay content. Anyway, this was not the reason why we discuss C stocks of soil types in that part of the manuscript. In this section we aim to compare our measured data on C stocks to data of previous studies investigating similar soils. This is particularly important as our data is different from what was found in other studies. We therefore would like to keep this part in the revised manuscript.

R#2: Page 13 – 14 How are you sure the soils are not affected by erosion?
This is explained on p.4 l.30. Sample locations were plane with <0.5° slope inclination. Thus, the probability of erosion is reduced to a minimum.

**R#2:** Page 13-15 – Could reduced decomposition due to salt stress and anaerobic conditions from the high moisture content be contributing to the higher organic matter content in the Non-sodic and sodic Solonchaks?

**A:** We would expect that a reduced decomposition due to salt stress would result in an accumulation of particulate OM. This was not the case in the studied soils: salt-affected and non-salt-affected soils contained similar proportions of particulate OM. Moreover, the analysis of C isotopes and neutral sugars indicated a comparable degree of OM alteration between the soils, as already discussed in the manuscript, while we would expect a smaller OM alteration if decomposition would be reduced in the salt-affected soils.

Higher OC stocks in the salt-affected soils were particularly found in the topsoils. Anaerobic conditions in the topsoil are very unlikely as the gleyic properties of the soils show that anaerobic conditions can be primarily expected in the subsoil, but not in the topsoil. For example, in the four soil profiles next to the lake, Fe and Mn mottling reaches on average 84 ± 16 cm soil depth, thus indicating the maximum average ground water level during flooding. In the subsoil differences between OC stocks were smaller. Thus, we do not consider anaerobic conditions as a factor explaining the high OC stocks in the Solonchaks.

**R#2:** Page 13 – 30 Could you remind us what your second hypothesis was?

**A:** In the revised manuscript we are going to repeat the second hypothesis at the beginning of that paragraph.

**R#2:** Page 13 – it seems that the water availability to plants and microbes might be similar in the dry salt free Kastanozem and the wetter but salty Solonchaks (i.e. similar osmotic pressure). This could explain why above ground biomass was similar and explain the similarities in soil C.

**A:** This is a good point and was already noted in the manuscript (p. 12 l. 37 – p. 13 l. 5). However, we think we should discuss this issue more intensively in the revised manuscript. This was already noted by Referee #1 and we are going to revise the discussion in the revised manuscript with particular focus on that point.

**R#2:** Page 15-19 this lets us assume?

**A:** We are going to change that in the revised MS to “This led us to the conclusion...”
Response to short comment by M.W.I Schmidt and his two master students, respectively

Dear M.W.I Schmidt,
Dear master students,

Thank you for taking your time to discuss our manuscript and give advices for improvements.

Short comment #1 (SC #1): *A note upfront from the submitting person: This review was prepared by two master students in geography or earth system science at the University of Zurich. The review was part of an exercise during a second semester master level seminar on “the biogeochemistry of plant-soil systems in a changing world”, which I organize. We would like to highlight that the depth of scientific knowledge and technical understanding of these reviewers represents that of master students. We enjoyed discussing the manuscript in the seminar, and hope that our comments will be helpful for the authors.*

Rising temperature and anthropogenic influences are the main reason why salt affected soils become more frequent. This study aims to investigate the organic matter dynamics of three different soil types (Kastanozem, non-sodic Solonchak, sodic Solonchak), along a salinity gradient in the South-Western Siberian Kulunda steppe. Soil samples and the aboveground plants and underground biomass have been characterized by a variety of methods. The results of this study were different from similar studies in the literature, and the authors had to reject their initial hypothesis. Surprisingly, organic carbon stocks in the salt-affected were not smaller than in the non-salt-affected soils. Also the abundance and stability of the particulate organic matter was not influenced by salinity. The proportion and stability of mineral-bound organic matter was not reduced under high sodicity levels. Thus, salt-affected soils contribute significantly to the organic carbon storage in the examined region. Also most of the organic carbon was present in stable mineral-organic associations which implies a long-term sequestration. We liked the readability of the paper. The abstract, the introduction, the discussion and the conclusion are interesting to read. It is a very relevant topic that is important under future climate. However, we had problems to understand the experimental setup. Could the sampling and experimental setup be summarized in a figure or table?

Authors (A): Thank you for this evaluation of our manuscript. We are going to explain the experimental setup more clearly in the revised manuscript, particularly the part on p. 4 l. 31-39 will become changed. However, please note that we included already a figure explaining the experimental setup in the existing manuscript (Figure 1).

SC #1: Also, for the belowground plant samples we did not understand how they were taken.

A: To characterize the isotopic composition (d13C) and neutral sugars of plant samples, we retrieved whole plants of the dominant plant species (see Table 1) from the soil. Subsequently, we split the plant into two parts: roots and shoots.

SC #1: Were they taken in the profile? Or in about 5 meter distance in every depth, or just once?

A: With respect to plant samples, we took three replicates in about 5m distance to the profile. This is explained on p. 4 l. 38 – p. 5. l.2 and also shown in Figure 1.

SC #1: As we are only in our second master semester the method section was too long for us. We understand that this section is important for replication. Would it be possible to shorten this section
and/or move the details (set up, used instruments, packages, etc.) in the appendix? For non-experts it would help for faster understanding.

**A:** We agree that the method section is very long. But this is owed to the many methods we used to collect our data. Methods like density fractionation, neutral sugars analysis or PLFA have to be explained in such detail. Also other methods, as the determination of OC, TN, and d13C, are non-trivial and deserve a paragraph of explanation. However, we decided to move the part about soil mineralogical composition into the supplement of the revised MS as it does not contribute substantial data which is discussed later on.

**SC #1:** We also found many references to figures and tables in the supplement. We are wondering why they are referred to so often, sometimes more often than figures in the the normal text. Could it be, that some figures from the supplement should be Moved back to the main text?

**A:** The supplemental data (figures and tables) give additional information which contribute to the understanding of the manuscript but are not necessary for a deep discussion of the data. Hence, we would like to keep it as is and not move part of it into the main text of the MS.

**SC #1:** On page 6 in line 3 you the text says “Sample quantity allowed only for two treatments for qualitative analysis” Why are just two treatments for qualitative analysis allowed. Where there not good enough or to less soil samples?

**A:** In XRD analysis there are usually four treatments used to distinguish the clay mineralogical composition of a soil sample: (i) Mg$^{2+}$-saturation, (ii) Mg$^{2+}$ + ethylene glycol saturation, (iii) K$^+$-saturation and (iv) K$^+$-saturation + heating to 550K. We had not enough sample mass to conduct all four treatments, thus we had to decide for two of the treatments. As we were interested in the quantity of expandable (swelling) clay minerals such as smectite, we decided to use the “standard” treatment (Mg$^{2+}$-saturation) and the Mg$^{2+}$ + ethylene glycol saturation, as the combination of both yields the necessary results.

**SC #1:** Also on page 11 & 12 in line 20 respectively 13 there was written “data not shown” but for us it was not clear why there are not shown and why you have to state that. If the data are important could you put the data in the supplement?

**A:** On p. 11 l. 20 we state the relative proportion of each neutral sugar on the entire data set. This is to give an overview to the data and not necessary to repeat in a table. Otherwise it would be a redundant presentation of data. On p. 12 l. 13 we write about fungi: bacteria ratios. We agree with you that it would be informative to the reader if we add the data to the supplements.

**SC #1:** Table 1: The last column shows “a” but we do not understand why.

**A:** As is noted in the heading of the table, these letters indicate whether there are significant differences between the samples or not.

**SC #1:** For table 2 & 3 a line between each soil type would help to read the table. It would also be nice to clarify in the tables itself what the values in parenthesis mean (standard error).

**A:** We are going to add a line between soil types for better readability. We already clarified in the heading of the table the meaning of the value in parenthesis (standard error).
SC #1: The figure 1 was for us quite unclear. We could not make sense of the position in the plant sample dots. Does the position represent on which side they were taken? Why there are green dots in the Sodic Solonchaks could be stated in the text. However, for us it was not clear. As we wrote above, the experimental set up was mixed with the rest of the text. Not all profiles have the same depth, but this different depth is not represented in the figure.

A: Yes, the dots represent the approximate position where the samples were taken. This is also indicated by the arrows which highlight the distance of the sampling locations to each other.

As stated on p. 4 l. 32-36, four soil profiles were analyzed on the foot slope of the transect because of the larger site heterogeneity there. However, laboratory analyses afterwards revealed that one of the four soils was not sodic and had to be grouped together with the non-sodic Solonchaks. This exactly is shown in Figure 1. We also explained the meaning of the colors in Figure 1. The different depth of the groundwater table, which resulted in different depths of the soil profiles, is clearly shown in Figure 1.

SC #1: Also in the figure 3 it was for us not that clear why the depth is not the same as in the profiles.

A: $^{14}$C analyses are very costly and to measure all samples of a profile was therefore not possible for us. We therefore decided to measure all samples until the topmost C horizon of a profile, because the topmost horizons are those with the highest OC contents. Moreover, in the topmost horizons we observed the largest differences between the soils with respect to their OC stocks. Only in the Non-sodic Solonchak we had not enough LF material in the Cz horizon to analyze the $^{14}$C activity. We agree with you that we should mention this in the figure caption and the Material & Methods section. This is going to be included in the revised MS.

SC #1: In the conclusion we would also appreciate an outlook for future studies. What would be important to look at?

A: An important issue would be to determine the water potential of all soils as the sum of matric potential + osmotic potential. Determination of the matric potential can be done by collecting undisturbed samples and measuring a soil retention curve. A time-series of soil moisture measurements could then be related to the soil water retention curve to obtain the matric potential at the particular soil moisture over the year. The osmotic potential can be determined via
measurements of the electrical conductivity of the soil solution. By that we could verify whether the water stress, as indicated by a low water potential, is similar between the soils.

Another promising approach would be to relate our results to measurements of enzyme activities. By that we would be able to directly determine whether the microbial activity is inhibited by salt stress or not. In combination with incubation studies of the bulk soil we could compare soil OM decomposition rates between the salt-affected and non-salt-affected soils. In the incubation studies we could adapt the soil moisture to the values observed in the field to simulate field conditions.

In the revised manuscript we are going to give a brief overview on that future research prospects.

SC #1: Some minor comments: - Strange starting sentence of the introduction "... soils...important...." I why do they get more important. They will get more frequent and just to study them will get more important. Maybe “twice as” could be a nicer starting.

A: We agree with you and starting the sentence in the abstract with “Salt-affected soils will become more frequent in the next decades...” is a more precise statement. We are going to change that in the revised MS.

SC #1: Page 3/ line 42 It is a german sentence; “Todate, these soils cover already an area. . .” do you need “already”?

A: “Already” indicates that the soils cover a considerable area worldwide.

SC #1: Page 6/ line 26 Ulnits are at two lines

A: This manuscript is not yet text-edited. If published in Biogeosciences, text-editing will be done.

SC #1: Page 6/ line 33 It is written Sect. 2.5, but chapters are not numbered

A: The manuscript was written with a template offered by Copernicus Publications. This template does not include numbering of sections. But, if the manuscript gets published in Biogeosciences, numbering of sections will become necessary and thus we included the section number already.

SC #1: Page 9/ line 30 . . .very broad, peak broadening is related. . . I you might make two sentences?

A: We agree with you and will correct this in the revised MS.

SC #1: Page 15/ line 19 This let’s. . . I informal english

A: In the revised MS we are going to change that to “This led us to the conclusion...”
Organic matter dynamics along a salinity gradient in Siberian steppe soils

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Abstract

Salt-affected soils will become increasingly important in the next decades as arid and semi-arid ecosystems are predicted to expand as a result of climate change. Nevertheless, little is known about organic matter (OM) dynamics in these soils, though OM is crucial for soil fertility and represents an important carbon sink. We aimed at investigating OM dynamics along a salinity and sodicity gradient in soils of the south-western Siberian Kulunda steppe (Kastanozem, Non-sodic Solonchak, Sodic Solonchak) by assessing the organic carbon (OC) stocks, the quantity and quality of particulate and mineral-associated OM in terms of non-cellulosic neutral sugar contents and carbon isotopes (δ¹³C, δ¹⁴C activity), and the microbial community composition based on phospholipid fatty acid (PLFA) patterns. Above-ground biomass was measured as a proxy for plant growth and soil OC inputs. Our hypotheses were that (i) soil OC stocks decrease along the salinity gradient, (ii) the proportion and stability of particulate OM is larger in salt-affected Solonchaks as compared to non-saline Kastanozems, (iii) sodicity reduces the proportion and stability of mineral-associated OM, and (iv) the fungi : bacteria ratio is negatively correlated with salinity. Against our first hypothesis, OC stocks increased along the salinity gradient with most pronounced differences between topsoils. In contrast to our second hypothesis, the proportion of particulate OM was unaffected by salinity, thereby accounting for only <10% in all three soil types, while mineral-associated OM contributed to >90%. Isotopic data (δ¹³C, δ¹⁴C activity) and neutral sugars in the OM fractions indicated a comparable degree of OM transformation along the salinity gradient, and that particulate OM was not more persistent under saline conditions. This we attribute to a resilient microbial community composition and function, which was nearly unaffected by salt occurrence, and capable of decomposing OM at a similar rate in salt-affected and non-saline soils. Also our third hypothesis was rejected, as saline-sodic soils contained more than twice as much mineral-bound OC than non-saline Kastanozems, what we ascribe to the flocculation of OM and mineral components under higher ionic strength conditions. Contrary to the fourth hypothesis, the fungi : bacteria ratio in the topsoils remained fairly constant along the salinity gradient. A possible explanation why our hypotheses were not affirmed is that soil moisture covaried with salinity along the transect, i.e. the Solonchaks were generally wetter than the Kastanozems. This might cause comparable water stress conditions for plants and microorganisms, either due to a low osmotic or a low matric potential, resulting in (i) similar plant growth and, hence, soil OC inputs along the transect, (ii) a comparable persistence of particulate OM, and (iii) unaffected fungi : bacteria ratios. We conclude that salt-affected soils contribute significantly to the OC storage in the semi-arid soils of the Kulunda steppe while most of the OC is associated to minerals and therefore effectively sequestered in the long-term.
Introduction

Salt-affected soils occur predominantly in arid and semi-arid environments where rainfall is insufficient to leach salts from the soil (Mavi et al., 2012). They form either anthropogenically as a result of agricultural mismanagement or naturally due to the accumulation of salts from mineral weathering, dust deposition, precipitation or capillary rise of shallow groundwater tables (Essington, 2004). According to FAO (2001), salt-affected soils include Solonchaks (high salinity) and Solonetzes (high sodicity). While Solonchaks contain high elevated loads of water-soluble salts, while Solonetzes are primarily distinguished by Na⁺ as a dominant cation present on the exchange sites and usually a pH > 8.5, irrespective of the quantity of salts. This difference in the amount and composition of salts within both soil types leads to contrasting physico-chemical properties. Solonchaks have a compact soil aggregation, whereas soil particles tend to disperse in Solonetzes. In Solonchaks, as a result of high sodicity, causing a poor soil structure and the translocation of clay (lessification) and organic matter (OM) as well as clogging of pores which results in reduced water infiltration, increased surface run-off, and the risk of erosion (Qadir and Schubert, 2002; Sumner, 1993). Salt-affected soils, i.e., both Solonchaks and Solonetzes, respectively, are harsh environments for plants as high salt contents reduce the osmotic potential and subsequently limit plant water uptake (Läuchli and Grattan, 2007). Nutrient uptake is impeded due to ion competition and the high pH, while the poor soil structure (particularly in Solonetz caused by high sodicity) has adverse effects on soil water balance and plant development (Qadir and Schubert, 2002). As a result, plant residue inputs into the soil are reduced and, thus, lead to small soil OM contents (Wong et al., 2010). However, OM is a key component of soils, being a reservoir for nutrients and determining a soil’s agricultural productivity, while, at the same time, it is an important carbon (C) repository and plays a pivotal role in the course of climate change (Lal, 2004). Particularly by improving soil structure and increasing the selectivity of exchange sites for Ca²⁺–soil OM can ameliorate sodic soils (Nelson and Oades, 1998; Sumner, 1993). Independent from soil genesis, salt-affected soils are further classified according to their electrical conductivity (EC, in dS m⁻¹) and sodium adsorption ratio (SAR) of the saturated paste extract into saline (EC >4 and SAR <13), sodic (EC <4 and SAR >13), and saline-sodic (EC >4 and SAR >13; U.S. Salinity Laboratory Staff, 1954). Both parameters control the soil structure due to their impact on the dispersion of clay and OM significantly. Numerous studies showed that the desorption of OM from clay particles increases with SAR, while a rise in EC or the proportion of divalent cations counterbalances the dispersing effect of Na⁺ by inducing flocculation (Mavi et al., 2012; Nelson and Oades, 1998; Setia et al., 2014). High soil pH is likewise supposed to increase losses of organic C (OC) through solubilization of OM (Pathak and Rao, 1998). Peinemann et al. (2005) concluded that in salt-affected soils, mineral-associated OM can be rapidly lost through dispersion and subsequent leaching as dissolved OM, while particulate OM represents a relatively stable fraction as its decomposition is reduced due to an inhibited microbial activity. In line with this, previous work revealed in incubation and field studies that the microbial decomposition of soil OM is reduced along salinity gradient at elevated salinity (Rath and Rousk, 2015; Rietz and Haynes, 2003). However, little is known about the composition of soil microbial communities functioning in salt-affected soils, and particularly for Solonchaks and Solonetzes, there are no studies available that characterized microbial community compositions. Baumann and Marschner (2011) and Pankhurst et al. (2001) observed decreased fungi : bacteria ratios at enhanced salinity, while Barin et al. (2015) found the opposite, indicating that more research is required to come to firm conclusions. Though, based on conclusions results from sorption-desorption experiments, previous studies noted the sensitivity of mineral-organic associations in salt-affected soils (Mavi et al., 2012; Setia et al., 2013, 2014), to
date, no study quantified the amount and properties of mineral-associated and particulate OM in these soils. This is surprising, as the occurrence of salt-affected soils is predicted to increase as a result of climate change due to enhanced aridity (Amini et al., 2016). To date, currently, these soils cover already a global area of 831 Mio. ha worldwide (Martínez-Beltran and Manzur, 2005), of which Solonchaks and Solonetzes constitute about 260 Mio. ha and 135 Mio. ha, respectively, and 135 Mio. ha, respectively (IUSS Working Group WRB, 2014). Thus, our objectives were to (i) elucidate the effect of salinity and sodicity on (ii) soil OC stocks, (iii) determine the quantities and properties of functionally different soil OM fractions (particulate vs. mineral-associated OM), and (iii) relate our results to changes of the microbial community composition. We approached this by comparing soil OC stocks, the amount and properties of density-separated OM fractions (contents of hydrolysable non-cellulosic neutral sugars; δ13C and δ14C activity), and the PLFA-based microbial community composition of three soil types representing an increasing impact of salinity and sodicity along a transect of increasing salinity and sodicity in the south-western Siberian Kulunda steppe. Non-cellulosic sugars were chosen as an OM quality parameter, as they enter the soil in large amounts with litter, root residues and plant rhizodeposits as well as by products of microbial and faunal metabolism and represent a major energy source for heterotrophic soil microbial communities (Cheshire, 1979; Gunina and Kuzyakov, 2015). Additionally, soil aggregate stability was determined to assess the effect of salinity, particularly Na⁺ sodicity on the structural stability of the soils. We hypothesized that (i) soil OC stocks decrease along the salinity gradient with increasing salinity, because high salinity decreases plant growth and subsequently lowers soil OC inputs, (ii) the proportion and stability of particulate OM is larger in salt-affected soils as compared to non-salt-affected soils since microbial decomposition and transformation of OM is reduced under high salinity levels, and (iii) sodicity reduces the proportion and stability of mineral-associated OM, and (iv) the fungi : bacteria ratio is negatively correlated with salinity.

Material & Methods

Study site and soil sampling

The studied transect is located in the south-western Siberian Kulunda steppe which is part of the Altaysky Kray (Russian Federation). The area belongs to the dry steppe type with a mean annual temperature of 2.6 °C and a mean annual precipitation of 285 mm (climate data from “WorldClim” data base; Hijmans et al., 2005). The studied transect (52°3’36.51”N, 79°36’0.71”E) ranged from a lake over a terraced hillslope to about 5 m upslope above the lake (Figure 1). The groundwater table varied from ca. 140 cm next to the lake to >300 cm at the highest point of the transect. Three different soil types developed along the transect primarily as function of the groundwater table. At shallow groundwater depth close to the lake, Sodic Solonchaks dominated, while Mollic Solonchaks (non-sodic) prevailed backslope with slightly higher groundwater at about 170–180 cm. Upslope the groundwater table reached >300 cm and capillary rise did not reach the soil surface, thus, Haplic Kastanozems and Calic Kastanozems occurred which were generally grouped as Kastanozems.—A detailed soil typology classification according to IUSS Working Group WRB (2014) of the analyzed profiles is given in Table S1. We sampled the soils at plane areas along the terraced slope to avoid the influence of erosion on the soil profiles. Three plots, each with a soil profile down to the groundwater table and locations for plant analyses, were established per soil type; only in the Kastanozems the groundwater was too deep to be reached. Four plots were analyzed on the footslope next to the lake, where site heterogeneity was larger, but one of the four soils was
not classified as Sodic Solonchak but as Haplic Solonchak. This soil profile was grouped together with the Mollic Solonchaks since these soils corresponded to a lower level of sodicity and they were referred to as Non-sodic Solonchaks. Therefore, Kastanozems and Sodic Solonchaks were represented by three soil profiles, while Non-sodic Solonchaks were characterized by four soil profiles. Composite soil samples were taken according to generic horizons in the profiles. Plant samples (shoots and roots) were taken within the plots 5 m distant from around each profile for determination of OC, total nitrogen (TN), and δ¹³C, and non-cellulosic neutral sugars. The above-ground biomass was determined in triplicate around each profile by cutting off plants in a 40 cm x 40 cm square and subsequent drying (70°C) and weighing of plant material. The major plant species are listed in Table 1.

10 Sample preparation and basic soil analyses

Samples from generic horizons of the profiles were air-dried and sieved to <2 mm. Visible plant materials were removed and big clods were gently broken to pass the sieve. An aliquot of the fine earth fraction was dried at 105°C to determine the residual soil water content. Soil bulk density was determined gravimetrically in triplicate for generic horizons by use of a soil sample ring. Soil pH was measured in a 1 : 2.5 (w : v) soil-to-water suspension after equilibration for one day. Carbonate content was analyzed by the Scheibler volumetric method (Schlichting et al., 1995). The texture of the soils was determined according to the standard sieve-pipette method (DIN ISO 11277, 2002) and the content of oxalate- and dithionite-extractable Fe was analyzed as described in McKeague and Day (1966). Soil aggregate stability was measured based on a method modified from Hartge and Horn (1989) and explained in detail in Bischoff et al. (2016). It was calculated as the difference between the mean weight diameter (MWD) of aggregates of a dry- and a wet-sieving method, expressed as ΔMWD, with a large ΔMWD corresponding to low aggregate stability and a small ΔMWD relating to high aggregate stability.

The soil mineralogical composition was analyzed to characterize the soils with respect to their composition of water-soluble salts and the amount of expandable clay minerals. Clay mineralogy significantly affects the physical properties of sodic soils (Essington, 2004). The quantity of expandable clay minerals was similar in all three soil types and cannot explain differences in the OM dynamics between the soils. All data on soil mineralogical composition are provided in the Supplements (S1).

Soil salinity parameters

The content and composition of water-soluble salts was determined by shaking the soil in a 1 : 5 (w : v) soil-to-water suspension at 15 rpm during 1 h and leaving the sample for one day to reach equilibrium. After measuring the EC the extract was centrifuged at 3,000 g for 15 min and filtered through 0.45-µm syringe filters (Cellulose acetate). An aliquot of the extract was measured for Na⁺, K⁺, Ca²⁺, and Mg²⁺ with an inductively coupled plasma optical emission spectrometer (Varian 725-ES; Agilent Technologies, Santa Clara, USA) while another aliquot was analyzed for Cl⁻, NO₃⁻, and SO₄²⁻ with an ion chromatograph (ICS-90; Dionex Corp., Sunnyvale, USA). The concentrations of Na⁺, Ca²⁺, and Mg²⁺ (mmol l⁻¹) in the extract were used to calculate the SAR according to Eq. (1).

\[
SAR = \frac{Na^+}{(Ca^{2+} + Mg^{2+})^{0.5}}
\]
Soil mineralogical composition

X-ray diffractograms of ball-milled <2-mm fractions were recorded with an X’Pert PRO MPD Θ–Θ diffractometer (PANalytical, Almelo, Netherlands) equipped with a Cu anode producing Kα radiation. The powder samples were scanned from 2° to 85° 2Θ with a step size of 0.02° 2Θ and 3 s per step. A subset of samples was evaluated at the micro-scale using a Quanta 600 FEG environmental scanning electron microscope (ESEM; FEI Company, Hillsboro, USA) with an acceleration voltage of 20 keV. As the analysis was carried out in low vacuum mode (0.6 mbar), sputtering of the samples with gold or carbon was not necessary. The microscope was equipped with an Apollo XL EDX detector (Ametek Inc., Berwyn, USA). Clay mineralogical analyses were carried out for one representative soil profile of each soil type. Clay fractions (<2 µm) were obtained by pre-treating the soil with acetic acid (removal of carbonates), H2O2 (removal of OM), and dithionite-citrate (removal of iron oxides), subsequent separation by sedimentation (Stoke’s law) and final Mg2+ saturation to cause flocculation and thus easier handling of samples. X-ray diffraction patterns were recorded using the same system and settings as for the powder analyses of bulk soil but with Co-Kα radiation generated at 40 kV and 40 mA. Oriented mounts were prepared on porous ceramic tiles to avoid segregation of fine particles during sedimentation (Dohrmann et al., 2009) and scanned from 2° to 35° 2Θ with a step size of 0.02° 2Θ and 4 s per step. Sample quantity allowed only for two treatments for qualitative analysis: (i) Mg2+, (ii) Mg2+ + ethylene glycol. The ethylene glycol treatment was used as it detects expandable clay minerals like smectite, which strongly affect the physical properties of sodic soils.

Determination of organic carbon, δ13C, and total nitrogen

Ball-milled <2-mm fractions were measured for OC and TN as well as for δ13C via dry combustion in an Elementar vario MICRO cube C/N Analyzer (Elementar Analysensysteme GmbH, Hanau, Germany) coupled to an IsoPrime IRMS (IsoPrime Ltd, Cheadle Hulme, UK) after removing inorganic C by fumigation with HCl and subsequent neutralization over NaOH pellets (modified from Walthert et al., 2010). The measured δ13C values were corrected by calculating response factors from standard compounds (CaCO3, cellulose, caffein) and expressed in the delta notation related to the Vienna PeeDee-Belemnite-Standard (0‰). The complete removal of inorganic C from all samples was confirmed by δ13C values which are in the typical range of soil OM (-22.5‰ to -28.1‰).

Density fractionation and 14C analysis

Density fractionation (modified after Golchin et al., 1994) separated the soil into a light fraction (LF), containing mostly particulate OM, and a heavy fraction (HF), consisting of mineral-associated OM as well as mineral components free of OM. As particulate OM contents are mostly very low in the subsoil, we fractionated the soil only until the first C horizon of each profile. In brief, 25g soil was weighted in duplicate into beakers and 125ml sodium polytungstate (SPT; ρ = 1.6 g cm⁻³) was added, gently stirred with a glass rod and ultra sonification was applied with an energy input of 60 J ml⁻¹ during 8 min to break down aggregates. After centrifugation at 3,000 g for 20 min the LF was separated from the HF by decanting the floating LF on polyethersulfone filters and repeating the procedure if the separation between both fractions was insufficient. LF remaining on the filter was washed with deionized water to remove residual sodium polytungstate until the washing solution had an EC <60 µS cm⁻¹. The HF remaining in the beaker was washed with deionized water until the EC of the washing solution was <100 µS cm⁻¹, but at maximum four times in the salt-affected soils, as no residual sodium...
polytungstate was detected afterwards by ESEM–EDX analysis, which was carried out with a Quanta 200 FEG environmental scanning electron microscope (FEI Company, Hillsboro, USA) coupled to an XL–30 EDX detector (Ametek Inc, Berwyn, USA). The washing solutions of both LF and HF, respectively, were collected, filtered through 0.45-µm syringe filters (PVDF), and measured for non-purgeable OC with a LiquiTOC (Elementar Analysensysteme GmbH, Hanau, Germany) to account for the loss of OC during washing of the samples (mobilized OC, MobC; Gentsch et al., 2015). The LF and HF were freeze-dried, weighted, homogenized in a mortar, and subsequently measured for OC and TN as well as δ¹³C as described in Sect. 2.4, after removal of inorganic C. The mobilized OC was added to the OC content of the LF or HF, respectively.

Three representative soil profiles were selected, one per soil type, for analysis of ¹⁴C activities of OM fractions at the Max Planck Institute for Biogeochemistry Jena (Germany). As the low quantity of LF material in the subsoil did not allow for an accurate ¹⁴C measurement at deeper depth, we only analyzed ¹⁴C activities until the topmost C horizon of the respective soil profile. Inorganic C was removed by 2M HCl until pH remained <3.5 and samples were subsequently neutralized with 2M NaOH to pH 6. After freeze-drying ¹⁴C analysis was performed with a 3MV Tandetron™ AMS ¹⁴C system (Steinhof et al., 2011) and ¹³C isotope activities were converted to percent modern carbon (pMC) according to Steinhof (2013), while pMC was defined according to Stuiver and Polach (1977), see Eq. (2):

\[ pMC = \frac{A_{\text{act}}}{A_{\text{abs}}} \times 100\% \]  

(2)

where \( A_{\text{act}} \) is the normalized sample activity and \( A_{\text{abs}} \) corresponds to the activity of the absolute international standard; both activities were background-corrected and δ¹³C-normalized. OxCal 4.2 software (University of Oxford) was used to calculate conventional ¹³C ages by selecting the IntCal13 calibration curve (Reimer et al., 2013), if pMC was <100%, and the calibration curve from Hua et al. (2013), if pMC was >100%.

Biomarker analyses

Non-cellulosic neutral sugars

Non-cellulosic neutral sugars were analyzed in the LF and HF from generic horizons of each soil profile. In the LF neutral sugars were only analyzed in some of the topmost horizons, as its content was too low in most samples to provide sufficient material. Additionally, neutral sugars were determined in plant material (shoots and roots). Neutral sugars were analyzed slightly modified according to Rumpel and Dignac (2006), including the EDTA purification step from Eder et al. (2010). In brief, 600mg of HF and 50mg of LF or plant material was hydrolyzed in 4M trifluoroacetic acid (TFA) at 105°C during 4 h after 1.5ml myo-inositol was added as an internal standard. After cooling to room temperature the extract was filtered through glassfiber filters (Whatman™ GF6) and TFA was removed in a rotary evaporator. The samples were redissolved in ultrapure water and the pH was adjusted to 4–5 by adding NH₃. Ferric Fe was complexed by adding 4ml EDTA and incubating the samples in the dark during 10min. From now on darkened glassware was used to prevent photolysis of Fe(III) ligand complexes. After freeze-drying and adding two drops of NH₃ the reduction of aldoses to their corresponding alditols (derivatization) was performed at 40°C during 1.5 h with NaBH₄ dissolved in dimethyl sulfoxide. Acetylation was carried out by adding 2ml acetic anhydride and 0.2ml glacial acetic acid, thereby using methylimidazole as a catalyst. Ice-cold deionised water was added after 10 min to stop the reaction. Sugar monomers were extracted by liquid-liquid extraction with dichloromethane and subsequently measured by gas chromatography on a 7890A GC system (Agilent Technologies, Santa Clara, USA) equipped
with a SGE forte GC capillary column (0.25mm diameter and 0.25µm film thickness; SGE Analytical Science, Melbourne, Australia) and a flame ionization detector, using He as a carrier gas. External standards were used to detect eight different sugars: arabinose, xylose and ribose (pentoses), galactose, glucose and mannose (hexoses), and fucose and rhamnose (desosysugars).

5 Phospholipid fatty acids

Directly after sampling, sieving to <2 mm and removing visible plant materials, 1.0–1.5g field-moist soil was weighted into cryovials and 3ml RNAlater® was added to prevent sample degradation (Schnecker et al., 2012). An aliquot was dried at 105°C to determine the soil water content. The cryovials were kept cool until they were frozen to −20°C within 72 h. For PLFA analysis we used a modified method from Gunina et al. (2014). Briefly, samples were transferred from cryovials into test tubes and washed with ultrapure water to remove residual RNAlater®. Lipids were extracted twice with a chloroform-methanol-citrate buffer (1:2:0.8 v/v/v) and separated into glycolipids, neutral lipids, and phospholipids by solid phase extraction with activated Silica gel (Sigma Aldrich, pore size 60Å, 70–230 mesh). Phospholipids were derivatized into fatty acid methyl esters (FAME) with 0.5M NaOH dissolved in methanol and with BF₃ as catalyst. FAME were analyzed with a 7890A GC system (Agilent Technologies, Santa Clara, USA) equipped with a 60m Zebon ZB-5MSi capillary GC column (0.25mm diameter and 0.25µm film thickness; Phenomenex, Torrance, USA) and a flame ionization detector, using He as a carrier gas. As an internal standard we used nonadecanoic acid (FA 19:0) and 17 fatty acids were used as external standards. Peak identification of the internal standard turned out as problematic in the salt-affected topsoils. Therefore we could not reliably quantify individual PLFA but only their relative proportion in the sample. As a result the sum of all PLFA was not used as a proxy of the microbial biomass contents but PLFA were used to characterize the composition of functional microbial groups. We applied a principal components analysis (PCA) on the relative distribution of all 17 PLFA to identify clusters of correlated PLFA, which presumably derive from identical microbial functional groups. The assignment of individual PLFA to certain microbial groups based on the PCA was in agreement with the literature (Frostegård et al., 2011; Olsson, 1999; Ruess and Chamberlain, 2010; Zelles, 1999). Thus, the following PLFA were used to distinguish functional microbial groups: 18:2ω6,9 and 18:1ω9c as marker for saprophytic saprotrophic fungi (SAPapFungi), 16:1ω5c to identify arbuscular mycorrhizal fungi (AMF), i15:0, a15:0, i16:0, i17:0 and a17:0 were related to gram-positive bacteria (Gram+), 10Me16:0 characterized actinomycetes (Actino), 16:1ω7c and 18:1ω7c identified gram-negative bacteria (Gram–), and 14:0, 15:0, 17:0 and 18:0 related to unspecific bacteria (UnspBact). The PLFA Cy19:0 and 20:4ω6c were not used as markers for microbial groups as they hardly reached the detection limit and were sometimes difficult to distinguish from other unspecific peaks in the gas chromatogram.

Calculation of organic carbon stocks

Organic C stocks (Mg ha⁻¹) were calculated according to Poeplau & Don (2013) for all horizons and the entire soil profile as well as until 1m depth using Eq. (3):

\[ \text{OC stock} = \sum_{i=1}^{n} \frac{FSM}{V_i} \times C_i \times D_i \]  

(3)

where \( n \) is the number of horizons, \( FSM \) is the fine-earth soil mass (g), \( V \) is the volume (cm³), \( C \) is the OC content (% of soil mass) and \( D \) is the length of the horizon (cm).
Statistical analyses

Data analysis was performed in R, version 3.2.5 (R Core Team, 2016). From replicated measurements we calculated arithmetic means and standard errors. To test for the effect of soil type on above-ground plant biomass a linear mixed effects model was fitted (package lme4; Bates et al., 2012). We accounted for the nested structure of sampling, i.e. the soil type was used as fixed effect while the soil profiles (of each soil type) were included as random effects. Residuals and random effect estimates of the fitted model were visually assessed by Q-Q-normal plots but no deviations from normality were observed. The difference of the response variable between the soil types was tested based on the linear mixed effects model fit, including corrections for multiple comparisons (analogous to the Tukey test), with Satterthwaite degrees of freedom, on the basis of the R packages lsmeans (Lenth and Herve, 2015), lmerTest (Kuznetsova et al., 2015), and multcomp (Hothorn et al., 2008).

Soil sample related parameters were analyzed descriptively, as their sample size was only 3–4 per soil type, which was insufficient for statistical hypothesis testing. Data of PLFA and neutral sugars were analyzed by PCA in order to consider multiple response variables. Confidence regions (68%) for the group centroids of the independent factor variables were added to the biplots. Analysis of data from PLFA and neutral sugars involved the consideration of several response variables which was done by PCA, thereby adding confidence regions (68%) for the group centroids of the analyzed factor variables. Figure 1 was drawn in Inkscape, while the other graphs were generated using ggplot2 (Wickham, 2009).

Results

Basic soil and site properties

The soil moisture during sampling (% of dry weight) was very small-low in the Kastanozems (3.6–4.5%) and larger-higher in the salt-affected soils with shallow groundwater table (Non-sodic Solonchaks: 14.9–20.5%, Sodic Solonchaks: 16.4–30.6%; Table 2). Thus, soil moisture covaried with salinity along the transect. The pH in the Kastanozems increased from about 7 in the topsoil to 9 in the subsoil, while the Solonchaks revealed a nearly constant pH throughout the soil profile between 8.5 and 9. While Kastanozems had no carbonates in the topsoil, the carbonate content peaked in the Ck horizon with 51 ± 12 mg g⁻¹ (Table 2). The salt-affected soils exhibited larger-higher carbonate contents, between 53 ± 16 mg g⁻¹ and 152 ± 34 mg g⁻¹ in the Non-sodic Solonchaks and 115 ± 49 mg g⁻¹ and 264 ± 22 mg g⁻¹ in the Sodic Solonchaks. The aggregate stability was larger-higher in Kastanozems and Sodic Solonchaks (AMWD: 0.41 ± 0.06 mm and 0.33 ± 0.03 mm, respectively) than in Non-sodic Solonchaks (1.02 ± 0.29 mm; Table 2). The Kastanozems consisted mostly of sandy loam, while the Solonchaks were more loamy with larger-higher clay and silt contents. Oxalate- and dithionite-extractable Fe was consistently low in all three soil types (<0.4 mg g⁻¹FeO, <5 mg g⁻¹FeD, Table 2).

Soil salinity parameters

The EC1:5 was small-low (<250 µS cm⁻¹) in the Kastanozems with a slight increase from top- to subsoil, while the largest-highest EC1:5 in the Solonchaks was found in the topsoil (Table 2). In the Non-sodic Solonchaks the EC1:5 decreased from 3416 ± 1053 µS cm⁻¹ in the topsoil to 796 ± 333 µS cm⁻¹ in the subsoil, while the Sodic Solonchaks had the largest-highest EC1:5 with 5350 ± 1476 µS cm⁻¹ in the topsoil and the smallest-lowest EC1:5 with 1093 ± 702 µS cm⁻¹ in the subsoil. The SAR1:5 revealed a similar pattern, with small-low SAR1:5 (<2) in the
Kastanozems and largest-higher values in the Solonchaks (Table S2). In the Non-sodic Solonchaks the SAR dropped from 9.6 ± 2.2 in the topsoil to 3.9 ± 1.0 in the subsoil, while Sodic Solonchaks had the largest highest SAR with 36.0 ± 10.4 in the topsoil and 8.0 ± 4.6 in the subsoil. The composition of water-soluble anions and cations was different in the two salt-affected soils (Figure S1). While the Non-sodic Solonchaks had an almost balanced concentration of SO₄²⁻ and Cl⁻ on the one hand, and Na⁺, Ca²⁺ and Mg²⁺ on the other hand, the Sodic Solonchaks were dominated by SO₄²⁻ and Na⁺, with smaller quantities of Cl⁻.

### Soil mineralogical composition

The three soil types had a quite homogenous mineralogical composition, dominated by quartz and feldspar as well as calcite and dolomite in the carbonate-rich horizons, whereas almost all samples showed small quantities of amphibole and muscovite (Figure S2). In the Solonchaks also gypsum was present. Calcite and dolomite XRD peaks were very broad, peak broadening is related to very fine crystallite sizes. The clay fraction showed small amounts of illite, kaolinite, and chlorite, while smectites were partially present in the subsoil, and in the Sodic Solonchaks also in the topsoil. In the smectite-rich horizons, the quantities of smectite and illite exceeded those of chlorite and kaolinite significantly (Figure S3). In the Solonchaks, the quantities of water-soluble salts were small when related to the bulk soil. Mass balance calculations (data not shown) and analyses by ESEM-EDX (Figure S4) revealed that water-soluble salts mostly consisted of thanadite (α-Na₂SO₄) and halite (NaCl), but also bischofite (MgCl₂·6H₂O) could be present.

### Soil organic carbon stocks

Soil OC stocks increased with salinity and sodicity from Kastanozems over Non-sodic Solonchaks to Sodic Solonchaks (Figure 2). Differences were most pronounced in the topsoils, while subsoil OC stocks were similar between the soil types. Down to a depth of 100 cm Kastanozems had 70.9 ± 2.8 Mg OC ha⁻¹, Non-sodic Solonchaks 94.2 ± 6.9 Mg OC ha⁻¹ and Sodic Solonchaks 129.5 ± 25.6 Mg OC ha⁻¹. Thus, OC stocks in Non-sodic Solonchaks were 32.8 ± 9.7% larger than in Kastanozems and OC stocks of Sodic Solonchaks exceeded those of Kastanozems even by 82.6 ± 36.1%. The C : N ratios were comparable along the salinity gradient and ranged from about 10 in the topsoil to 5–8 in the subsoil (Table S2).

### Soil organic matter fractions

#### Organic carbon contents and isotopic composition

All three soil types were dominated by HF-OC with >90% of bulk OC, while LF-OC accounted for <10% (Table S3). The proportion of HF-OC revealed no clear depth gradient within the soil profiles. The OC content of the HF increased in A horizons with salinity and sodicity from Kastanozems (7.7 ± 0.3 mg g⁻¹) to Non-sodic Solonchaks (18.3 ± 2.7 mg g⁻¹) to Sodic Solonchaks (19.3 ± 5.0 mg g⁻¹), while OC contents were similar in the subsoil (Table S3). OC contents in the LF were smaller lower in the Kastanozems (120–219 mg OC g⁻¹) than in Non-sodic Solonchaks (197–279 mg OC g⁻¹) and Sodic Solonchaks (247–265 mg OC g⁻¹).

Table S3). Kastanozems and Non-sodic Solonchaks had the largest-higher LF-OC contents in the subsoil but LF-OC contents were equal over depth in the Sodic Solonchaks. HF material was enriched in δ¹³C as compared to LF material (Figure 3). In the LF δ¹³C ratios ranged from −27.5% to −26.4% (Kastanozems), −27.0% to −28.1% (Non-sodic Solonchaks) and −24.3% to −26.0% (Sodic Solonchaks). Remarkably, the δ¹³C ratios in the LF decreased from top- to subsoil in the Solonchaks, while the Kastanozems revealed a typical increase of δ¹³C ratios from top- to subsoil. The δ¹³C ratios of the LF were similar to the root
signals of the plants, while no relation to the shoot signals was apparent (Figure 4). Ratios of $\delta^{13}$C in the HF were comparable between the three soil types and ranged from −23.8% to −23.0% in the Kastanozems, from −23.3% to −22.8% in the Non-sodic Solonchaks and from −23.4% to −22.5% in the Sodic Solonchaks (Table 3). As residual sodium polytungstate (SPT) had to be removed during density fractionation for subsequent determination of OC parameters, all samples were washed with deionized water (see Sect. 2.6.5). This resulted in a loss of HF material. About 8–29 mg HF g$^{-1}$ soil was lost in Kastanozems, while the loss was larger in salt-affected soils due to the high solubility of salts and accounted for 61–86 mg HF g$^{-1}$ soil in Non-sodic Solonchaks and 46–76 mg HF g$^{-1}$ soil in Sodic Solonchaks, with larger losses in samples with high EC (Table 3). Despite larger HF losses were observed in Solonchaks, the percentage of MobC related to bulk OC was small in these soils (maximally 9.4 ± 1.6%), while Kastanozems had larger proportions of MobC (15.6 ± 0.5% to 45.7 ± 12.0%). This indicates that the water-soluble salts in the salt-affected soils were mostly not associated with OC. The quantities of MobC from the LF were larger in salt-affected soils and accounted for up to 258 mg OC g$^{-1}$ LF, but maximally 3.4% of bulk OC in all three soil types (Table 3). The proportion of MobC increased with depth in both LF and HF, respectively. The $^{14}$C activities in the LF were similar in the Kastanozem and the Sodic Solonchak and amounted mostly >100 pMC (Table 3, Figure 4), corresponding to recent C with $^{14}$C ages of maximally 60 years B.P. In the Non-sodic Solonchak the $^{14}$C activity was >100 pMC in the topmost horizon (Az1) but lower in the underlying horizons, i.e. 91.67 pMC (ca. 730 years B.P.) in the Az2 horizon and 93.86 pMC (ca. 580 years B.P.) in the Bkz horizon, respectively. This unusually high age of LF material indicated a possible contamination with HF material. The $^{14}$C activities in the HF were smaller than in the LF, corresponding to higher $^{14}$C ages, and no trend related to the three soil types was apparent. Remarkably, $^{14}$C activities increased from ca. 30 cm depth to 50–60 cm depth after a typical decrease from the topsoil. The $^{14}$C activities in the HF corresponded to $^{14}$C ages of 150–950 years B.P. in the topsoil horizons and 1200–2900 years B.P. in the underlying horizons, while the highest $^{14}$C age occurred in the comparatively deep Cz horizon (ca. 90cm) of the Non-sodic Solonchak with 4600 years B.P.

**Non-cellulosic neutral sugars**

The neutral sugar content of the LF from the topmost horizons was similar in the Kastanozems and the Non-sodic Solonchaks with 47 ± 5 mg g$^{-1}$ and 46 mg g$^{-1}$, respectively, while Sodic Solonchaks contained more neutral sugars (105 ± 27 mg g$^{-1}$; Table 3). Related to the OC content, sugar contents were comparable between all soil types and ranged from 328–410 mg g$^{-1}$ OC. The HF contained less sugars than the LF, thereby sugar contents decreased from top- to subsoil according to the decrease of OC contents (Table 3). In topsoils sugar contents of the HF increased from Kastanozems (1.0 ± 0.2 mg g$^{-1}$) over Non-sodic Solonchaks (3.1 ± 0.6 mg g$^{-1}$) to Sodic Solonchaks (5.7 ± 0.8 mg g$^{-1}$), while sugar contents were similar in the subsoil. Based on the OC content, sugar contents were similar in the Kastanozems and Non-sodic Solonchaks and ranged between 136–172 mg g$^{-1}$ OC, with no clear depth gradient. Sodic Solonchaks contained more sugar per g OC than the other two soil types, with 322 ± 61 mg g$^{-1}$ OC in the topsoil and smaller sugar contents in the subsoil (165 mg sugar g$^{-1}$ OC). The averaged proportion of each sugar in the total sugars was as following: xylose (27 ± 8%), glucose (20 ± 2%), arabinose (19 ± 2%), galactose (18 ± 3%), mannose (7 ± 3%), rhamnose (5 ± 1%), fucose (3 ± 1%), and ribose (1 ± 1%; data not shown).

The PCA of neutral sugars from plants, LF and HF material revealed two significant components (eigenvalue > 1), the first component (PC1) with 54.9% explained variance and the second component (PC2) related to 18.7%
explained variance (Figure S5). The composition of neutral sugars was different between plants, LF material and HF material, while differences between the three soil types were smaller. Plants of all soil types were enriched in xylose and those of salt-affected soils also in arabinose, while HF material of all soils was augmented with mannose, galactose, fucose, ribose, and rhamnose. Differences between soil types were apparent with respect to arabinose and glucose. In the Kastanozems OM in the LF and HF became enriched in arabinose during decomposition of plant material, while the opposite was observed in the salt-affected soils (see also Figure S5). The relative proportion of glucose remained similar in the Kastanozems but increased in the salt-affected soils in the course of decomposition (see also Figure S6). However, on the whole, neutral sugars in LF but also HF material were similarly altered in all three soil types with respect to their initial composition in the plant tissue, as indicated by a comparable shift of the three fractions in all soil types along the first axis in the biplot, suggesting a comparable degree of soil OM alteration between the soil types.

### Phospholipid fatty acids

The fungi : bacteria ratio was similar in the topsoils of the three soil types and amounted in A horizons 0.24 ± 0.01 in Kastanozems, 0.27 ± 0.04 in Non-sodic Solonchaks, and 0.17 ± 0.05 in Sodic Solonchaks (Table 4). In the subsoil the salt-affected soils had slightly higher fungi : bacteria ratios than the non-salt-affected Kastanozems. The relative contribution of PLFA observed within the PLFA profiles was as follows: PLFA from unspecific bacteria (36.7 ± 2.2%), Gram+ (25.6 ± 0.7%), Gram– (11.9 ± 1.3%), SAP-saprotrophic fungi (11.3 ± 0.9%), AMF (8.4 ± 1.8%) and from actinomycetes (6.1 ± 0.6%). Thus, bacterial PLFA constituted 80.4 ± 1.1% while fungal PLFA represented 19.6 ± 1.1% of the analyzed fatty acids. The PCA of the PLFA-based microbial groups extracted two significant components (eigenvalue >1) and showed a clear differentiation between bacterial and fungal PLFA (Figure 6), the former stretching along the first component (PC1) and the latter correlating with the second component (PC2). Accordingly, bacterial PLFA explained 57.8% of the variability of total PLFA, while fungal PLFA corresponded to 22.0% of the total variability. PLFA of Gram+, Gram– and actinomycetes were positively correlated with each other, but had a negative correlation to the group of unspecific PLFA. Among the fungal PLFA, those of AMF correlated negatively to those of SAP-saprotrophic fungi. Differences in the microbial community composition existed between soil horizons and were largely explained by the variability of bacterial PLFA, with a larger abundance of Gram+, Gram– and actinomycetes in topsoil horizons and a larger abundance of unspecific PLFA in the subsoil (Figure 6). Changes of the microbial community composition between the three soil types were less pronounced overall and mostly due to a larger variability of fungal PLFA in the Solonchaks as compared to the Kastanozems. Higher relative abundance of AMF in the salt-affected soils than in the non-salt-affected Kastanozems, whereas the composition of bacterial PLFA was similar between all soils. However, the fungi : bacteria ratio was rather constant between the three soil types and amounted to 0.22 ± 0.03 in Kastanozems, 0.28 ± 0.05 in Non-sodic Solonchaks, and 0.27 ± 0.03 in Sodic Solonchaks, with slightly larger fungi : bacteria ratios in the subsoils of the Solonchaks (data not shown).
Discussion

Soil OC stocks along the salinity gradient

Salt-affected soils, such as Solonchaks, are normally characterized by poor plant growth resulting in small soil OC inputs and subsequently low soil OC stocks (Wong et al., 2010). Muñoz-Rojas et al. (2012), for example, reported soil OC stocks in Solonchaks of southern Spain in 0–75 cm depth of 53.6 Mg ha\(^{-1}\) (coefficient of variation (CV): 60%) under shrub and/or herbaceous vegetation. Batjes (1996) calculated in the framework of a global meta-analysis average soil OC stocks of Solonchaks of 42 Mg ha\(^{-1}\) (CV: 67%) in 0–100 cm depth, while he noted that particularly Mollic Solonchaks had much larger soil OC stocks of 101 Mg ha\(^{-1}\) (CV: 44%). Kastanozems, on the other hand, contained on average 96 Mg ha\(^{-1}\) (CV: 50%) in the first meter, at which Haplic Kastanozems had soil OC stocks above that average of 138 Mg ha\(^{-1}\) (CV: 44%; Batjes, 1996). Based on data from Bischoff et al. (2016), we calculated soil OC stocks in Kastanozems of the dry steppe type of the Kulunda steppe down to 60 cm, which accounted for 110 ± 6 Mg ha\(^{-1}\). All of the previously published data confirm that salt-affected soils like Solonchaks have normally smaller OC stocks than the non-salt-affected Kastanozems. Contrary, in our study, salt-affected soils had larger OC stocks as compared to the nearby Kastanozems. With average OC stocks of 70.9 ± 2.8 Mg OC ha\(^{-1}\) in 0–100 cm depth of the Kastanozems, the values were clearly below those observed by Batjes (1996) and calculated from Bischoff et al. (2016). On the other hand, average OC stocks of 94.2 ± 6.9 Mg OC ha\(^{-1}\) and 129.5 ± 25.6 Mg OC ha\(^{-1}\) in 0–100 cm of the Non-sodic Solonchaks and Sodic Solonchaks, respectively, were clearly above the values reported by Batjes (1996) and Muñoz-Rojas et al. (2012). Larger OC stocks in salt-affected soils than in Kastanozems are also in contrast to earlier work which found a negative effect of salinity on soil OC stocks (reviewed by Wong et al., 2010). Possible reasons for the observed differences are climatic variations between the studies (strong aridity in the Spanish Solonchaks from Muñoz-Rojas et al., 2012) or alterations in soil texture (finer textured Kastanozems in the study from Bischoff et al., 2016) which may eventually change the soil water balance and thus plant growth and soil OC inputs. However, it appears that the covarying moisture gradient along the salinity transect is a better explanation for the observed differences. Possible reasons for the observed differences are climatic variations between the studies (strong aridity in the Spanish Solonchaks from Muñoz-Rojas et al., 2012) or alterations in soil texture (finer textured Kastanozems in the study from Bischoff et al., 2016) which may eventually change the soil water balance and thus plant growth and soil OC inputs. Moreover, during sampling we observed very dry conditions in the Kastanozems (only 4.0 ± 0.3% soil water related to dry soil mass), while the Solonchaks were generally wetter due to their shallow groundwater table (15–30% soil water, Table 2) such that in these soil water stress was mostly related to a small osmotic potential. Overall, the water stress in the three soil types could have been similar, either as a result of matric osmotic stress or osmotic-matric stress, leading to comparable moisture conditions for plant growth. Accordingly, and in contrast to previous work, along our transect plant growth (as measured by above-ground biomass) was not reduced under high salinity along the transect (Table 4), which is in contrast to previous work (Läuchli and Grattan, 2007; Wong et al., 2010). As this was discussed as a prerequisite of expected reduced OC stocks at elevated salinity (Wong et al., 2010), it can be one way we consider it as the most likely reason why our study revealed different results. We did not find a negative relation between OC quantity and salinity. Since the δ\(^{13}\)C ratios suggested that soil OM was mostly root-derived in the studied soils (Figure 4), one might argue that above-ground biomass is a poor proxy for soil OC input. However, under the assumption that root residue inputs are correlated with the above-ground biomass (evidence is given by...
Tityanova et al. (1999) who observed significant correlations (p < 0.01, R > 0.5) between the above-ground and below-ground biomass of typical plants in Siberian grasslands. We might note that both, above-ground and below-ground soil OC inputs, were comparable between all three soil types. Possible reasons for the observed differences are climatic variations between the studies (e.g., aridity in the Spanish Solonchaks from Muñoz-Roa et al., 2012) or alterations in soil texture (e.g., fine-textured Kastanozems in the study from Bischoff et al., 2016) which may eventually change the soil water balance and thus plant growth and soil OC inputs.

Moreover, Wong et al. (2010) argued that small OC stocks in salt-affected soils can also be the result of erosion-induced OC losses, as particularly sodic soils are prone to erosion. Since we paid particular attention to the fact that all soils were not affected by erosion, we can rule out erosion as a factor that modified OC stocks in our study.

**Partitioning and composition of soil OM in different functionally different OM fractions**

Considering processes of soil OC stabilization, semi-arid soils should have large proportions of particulate OC, as the formation of stable mineral-organic associations is attenuated due to low water availability and a high soil pH (Kleber et al., 2015). However, in the semi-arid soils of the studied transect particulate OC contributed <10% of bulk OC, while mineral-bound OC accounted for >90% (Table 3). This contrasts observations from steppes (mostly Chernozems) of European Russia (Breulmann et al., 2014; Kalinina et al., 2011, Canada (Plante et al., 2010), or China (Steffens et al., 2010), where particulate OC represented >20% of bulk OC. Nevertheless, our results are in line with Bischoff et al. (2016) who reported that maximally 10% of OC was present as particulate OM in Chernozems and Kastanozems of the Kulunda steppe. Thus, we support previous observations from this region and conclude that mineral-bound OM is the dominant OM fraction in both, salt- and non-salt-affected soils of the studied region.

In our second hypothesis we expect that the proportion and stability of particulate OM is larger in the salt-affected than in the non-salt-affected soils. Against our second hypothesis, Sodic and Non-sodic Solonchaks salt-affected soils contained similar proportions of particulate OC like non-salt-affected soils (the non-salt-affected Kastanozems, with 4–8% particulate OC in all three soil types (Table 3)). Comparable 14C activities in the LF of the three soil types (small 14C activities in the Non-sodic Solonchak were probably due to a contamination with HF material) indicated a similar turnover of particulate OM, thus contradicting our hypothesis of increased stabilization of particulate OM under high salinity levels. Based on OC determinations in particle-size separates and analyses of lignin components along a salinity gradient in the Argentinian Pampa, Peinemann et al. (2005) concluded that based on OC determinations in particle-size separates and analyses of lignin components along a salinity gradient in the Argentinian Pampa, that particulate OM is a relatively stable fraction in salt-affected soils due to a reduced microbial transformation of the plant-derived residue inputs.
This is not corroborated by our results. The isotopic C composition (δ13C activity, δ15C) and the composition of neutral sugars suggest a comparable alteration of OM (i.e. degree of OM decomposition) between the three soil types (Figures 4, 5). As for the first hypothesis, a possible explanation for the observed differences is that soil moisture covaried with salinity along the transect. Given that the water stress is similar in all three soil types, either due to a low osmotic or matric potential, OM decomposition can be likewise reduced in both the salt-affected and non-salt-affected soils, respectively. This results in a similar proportion and stability of particulate OM as well as a comparable alteration of soil OM along the transect, as indicated by the similar composition of C isotopes and neutral sugars in the studied soils. Hence, soil moisture can be considered a master variable in the OM dynamics of salt-affected soils, as it controls OM input and decomposition and, thus, can interfere with the effect of salinity on the quantity and quality of soil OM.

Furthermore, with respect to mineral-associated OM, Peinemann et al. (2005) concluded that mineral-bound OM is relatively susceptible to losses in salt-affected soils due to weak chemical bonding and subsequently weak OM stabilization. Our third hypothesis was built upon this conclusion but in contrast to that. Against our third hypothesis, the OC content of the HF of the salt-affected soils was more than twice as large as of the non-salt-affected Kastanozems (Table 3). Moreover, during washing of the density separates (sodium polytungstate wet removal) relatively less OC was mobilized from the HF of the salt-affected soils (3–10% MobC) than from the HF of the Kastanozems (16–46% MobC, Table 3), suggesting a lower chemical stabilization of mineral-bound OM in the non-salt-affected soils. We explain the large contents of mineral-associated OC under high salinity levels by consideration of basic chemical principles. According to Sumner (1993), dispersion of clay minerals is only possible below their critical flocculation concentration (CFC). This concept relates the dispersive effect of Na+ on the soil structure to the corresponding salt concentration of the soil solution (Rengasamy et al. 1984; Sumner et al. 1998). The authors classified soils into flocculated, potentially dispersive and dispersive depending on the EC and SAR of the soil water extract. Sumner et al. (1998) classified soils with large proportions of non-swelling-expandable illitic clays, while Rengasamy et al. (1984) considered soils with swelling-expandable 2:1 clays, similar to the smectite-rich soils of the studied transect. According to their classification, all of the salt-affected soils of our study fall into the category flocculated; even A horizons of the Sodic Solonchaks with an average SAR of 36 ± 10 remain flocculated, presumably due to the high electrolyte concentration as indicated by a high EC of 5350 ± 1476 µS cm⁻¹ (Table 2). This is underpinned by the aggregate stability of the Sodic Solonchaks (Table 2) and the lack of clay leissation or OM translocation, which are processes for which require the dispersion of clays and OM as a prerequisite. Similarly, in laboratory experiments, Setia et al. (2013, 2014) confirmed that the dispersive effect of Na+ on OM and mineral components is only evident at low electrolyte concentrations, particularly at low concentrations of divalent cations like Ca²⁺. These studies suggest, that the content of water-soluble salts in the soils of the studied transect is large enough to provoke flocculation of OM and mineral components and the formation of stable mineral-organic associations. Moreover, Nelson and Oades (1998) showed that the solubility of Na⁺-coated OM is larger than that of OM coated with Ca²⁺. Thus, particularly in the Non-sodic Solonchaks where Ca²⁺ is a dominant cation in the soil solution (Figure S1), the solubility of OM can be reduced. Furthermore, the Solonchaks had higher clay and silt contents than the Kastanozems (Table 2). This may also account for the higher HF-OC contents in the Solonchaks, as OM has an increased affinity to sorb on minerals in the clay- and silt-sized fraction (Kleber et al., 2015).
Interestingly, during the sodium polytungstate removal in the density fractionation procedure we found larger losses of HF material in the salt-affected soils as compared to the non-salt-affected Kastanozems, which we ascribe to the leaching of water-soluble salts (Table 3). However, the loss of MobC was much lower in the salt-affected soils. This indicates that the water-soluble salts were mostly not associated with OC, presumably because these salt minerals have a fast turnover (frequent formation and dissolution as function of the actual soil water content) and a small number of reactive surfaces.

Summing up these findings, in salt-affected soils particulate OM can be more labile than previously assumed, as evidenced by its small quantity in the Sodic and Non-sodic Solonchaks together with its low 14C ages. Salinity did not alter the proportion and stability of particulate OM, possibly due to the covarying moisture gradient. This suggests that soil moisture is a master variable which has to be considered when analyzing the effect of salinity on soil OM dynamics. Mineral-bound OM, on the other hand, is stabilized in the studied salt-affected soils as the large high electrolyte concentration in the soil solution promotes the flocculation of OM and minerals components. On the other hand, particulate OM is not as stable in salt-affected soils as previously assumed, as the degree of decomposition of this OM fraction was similar between salt-affected and non-salt-affected soils.

Microbial community composition along the salinity gradient

Microbial communities are sensitive to environmental changes and react to differences in the osmotic and matric potential (Rath and Rousk, 2015; Schimel et al., 2007). Particularly fungi but also Gram+ are thought to be more resistant against drought than Gram− due to their ability to produce osmolytes (Schimel et al., 2007). However, previous work on differences of the microbial community composition along salinity gradients could not support the view that fungi are superior to bacteria under water stress. Decreasing fungi : bacteria ratios caused by high salinity levels, as several studies observed even a negative relationship between fungal abundance and salinity (Baumann and Marschner, 2011; Chowdhury et al., 2011; Pankhurst et al., 2001). This suggests that in salt-affected soils not only drought dictates the abundance of certain microbial groups but that also toxic effects of certain ions or impeded nutrient uptake as a result of ion competition may exist. In our study, the fungi : bacteria ratio was not related to the salinity gradient and was similar between the topsoils of the three soil types (Table 4). Hence, our fourth hypothesis has to be rejected. As with hypothesis 1 and 2, a possible explanation is the covarying moisture gradient along the salinity transect which could have led to comparable water potentials (either due to low matric or osmotic potential) along the salinity gradient. Chowdhury et al. (2011) analyzed the effect of an alternating matric and osmotic potential on the PLFA-based microbial community composition. They detected a decreasing fungi : bacteria ratio with decreasing osmotic potential, while the opposite effect was evident with declining matric potential. Thus, with respect to our transect, both effects (decreasing matric vs. osmotic potential) could have cancelled each other out which resulted in similar fungi : bacteria ratios in the topsoils along the salinity gradient. Differences were only evident in the subsols, where salt-affected soils showed higher fungi : bacteria ratios than the non-salt-affected Kastanozems (Table 4). In the Sodic Solonchak fungi : bacteria ratios even increased from top- to subsoil (less pronounced also in the Non-sodic Solonchak), which is contrary to what was found in previous studies of temperate soils (Ekelund et al., 2001; Fierer et al., 2003; Taylor et al., 2002). This could indicate a larger C availability in the subsoil of the salt-affected soils (Fierer et al., 2003), which is also suggested by the δ13C ratios of the LF, which decrease from top- to subsoil in the Solonchaks (Figure 3).
With respect to the PLFA-based microbial community composition, PCA revealed a higher abundance of AMF in the salt-affected soils than in the Kastanozems (Figure 6). Evelin et al. (2009) reviewed the role of AMF in alleviating salt stress for plants. They concluded that AMF increased nutrient uptake, photosynthetic rate, water-use efficiency, and improved osmoregulation in the host plant. Thus, salt stress in plants caused by high salinity levels, such as a hampered nutrient uptake due to ion competition or exposure to osmotic stress, can be alleviated by symbiosis with AMF. This could explain the higher relative abundance of AMF in the Solonchaks of the studied transect. Moreover, the fungal PLFA composition revealed a larger variability in the salt-affected soils, indicating that these soils have a more variable fungal community composition. Both results can have consequences, if we consider that fungi are thought to be the primary decomposers of particulate OM (evidence is given by Bossuyt et al., 2001; Fray et al., 2000; Six et al., 2006): a fungal community whose abundance and diversity is unaffected by salinity is capable of decomposing particulate OM at the same rate in salt-affected and non-salt-affected soils. Thus, the decomposition of particulate OM proceeds also in the salt-affected soils, explaining the comparatively low contents of particulate OM and the observation that the proportion of particulate OM was unrelated to salinity.

Conclusions

This study aimed at investigating OM dynamics along a salinity gradient in soils of the south-western Siberian Kulunda steppe. Based on previous research, three hypotheses were tested: (i) soil OC stocks decrease along the salinity gradient, because high salinity decreases plant growth and subsequently lowers soil OC inputs, (ii) the proportion and stability of particulate OM is larger in salt-affected soils as compared to non-salt-affected soils as microbial decomposition and transformation of OM is reduced under high salinity levels, and (iii) sodicity reduces the proportion and stability of mineral-associated OM as the presence of Na+ and a high pH causes dispersion of OM and mineral components. Based on our results, all three hypotheses were rejected. The findings of this study suggest that soil moisture is a master variable shaping the soil OM dynamics along a salinity gradient of semi-arid steppe soils. The covarying moisture gradient along the salinity gradient serves as an explanatory factor for (i) the increasing soil OC stocks with increasing salinity, (ii) the constant proportion and stability of particulate OM along the transect, and (iii) a similar fungi : bacteria ratio in the topsoils along the studied gradient. As new emerging hypothesis, we suppose that the higher soil moisture in the salt-affected soils compensates the negative effects of high salinity on plant growth and the microbial community. By measuring the water potential, as the sum of matric and osmotic potential, one could test whether water stress occurs in both salt-affected and non-salt-affected soils, respectively. Since the covariation of salinity and moisture is a natural phenomenon in groundwater-affected Solonchaks of semi-arid steppes, this aspect deserves more attention in future studies.

Our data also showed that high salinity can cancel out the effect of sodicity on the dispersion of OM and mineral components. This we ascribe to the high ionic strength of the soil solution fostering the flocculation of soil constituents and increasing the formation and stability of mineral-organic associations. Given similar OC inputs into the soils along the transect this can be the reason for the larger OC stocks in the salt-affected soils. Against our first hypothesis, soil OC stocks increased along the salinity gradient with the most pronounced differences in the topsoil. Contrary to previous studies, plant growth (as determined by above-ground biomass) was not reduced under high salinity levels, suggesting that the soil OC input was similar between salt-affected and non-
salt-affected soils. In contrast to our second hypothesis, the abundance and stability of particulate OM was not related to salinity levels. Remarkably, most of soil OC (>90%) existed in mineral-organic associations (HF >1.6 g cm\(^{-3}\)) and only a small proportion (<10%) was present in particulate OM (LF <1.6 g cm\(^{-3}\)). The composition of C isotopes (\(\delta^{13}C, \delta^{14}C\) activity) and neutral sugars in the density separates suggested a similar degree of OM alteration in salt-affected and non-salt-affected soils. This let’s assume that the microbial activity was not reduced under high salinity levels. We ascribe this to a functionally diverse and resilient microbial community, as indicated by a fungi : bacteria ratio unaffected by salinity and an even larger fungal PLFA variability under saline conditions, which is capable of decomposing particulate OM at a similar rate in salt-affected and non-salt-affected soils. Contrary to our third hypothesis, the proportion and stability of mineral-bound OM was not reduced under high sodicity levels. High ionic strength of the soil solution fosters the flocculation of soil constituents and, hence, increases the stability of mineral-organic associations. This, in conclusion, can be the reason for the larger OC stocks in the salt-affected soils: at similar soil OC inputs along the transect and a similar rate of particulate OM decomposition, mineral-associated OM accumulated in the salt-affected soils due to the high ionic strength of the soil solution. In summary, salt-affected soils contribute significantly to the OC storage in the semi-arid soils of the Kulunda steppe. Most of the OC was present in stable mineral-organic associations and, thus, effectively sequestered in the long-term.

Acknowledgements

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References


Table 1: Vegetation (dominant species) and above-ground biomass on each soil type. Given are arithmetic means and the standard error of the mean in parentheses. Significant differences (p < 0.05) were not present and are denoted as same lowercase letters.

<table>
<thead>
<tr>
<th>Soil type</th>
<th>Vegetation / dominant species (from most to least dominant)</th>
<th>Above-ground biomass g m⁻²</th>
</tr>
</thead>
<tbody>
<tr>
<td>Kastanozem</td>
<td>Festuca valesiaca – Thymus maschallianus – Koeleria glauca</td>
<td>164.8 (37.7) a</td>
</tr>
<tr>
<td>Non-sodic Solonchak</td>
<td>Leymus poboanus – Artemisia nitrosa – Atriplex verrucifera</td>
<td>133.7 (17.6) a</td>
</tr>
<tr>
<td>Sodic Solonchak</td>
<td>Atriplex verrucifera – Leymus poboanus – Hordeum brevisubulatum</td>
<td>139.5 (21.7) a</td>
</tr>
</tbody>
</table>
Table 2: Basic soil parameters as function of soil type and horizon. Given are arithmetic means and the standard error of the mean in parentheses. Abbreviations: n = sample size, BD = bulk density, EC = electrical conductivity, SAR = sodium adsorption ratio, Aggstab = aggregate stability, MWD = mean weight diameter, FeO = oxalate-extractable Fe, FeD = dithionite-extractable Fe, n.d. = not determined.

<table>
<thead>
<tr>
<th>Soil type</th>
<th>Horizon n</th>
<th>Depth cm</th>
<th>BD g cm⁻³</th>
<th>Moisture % of dry weight</th>
<th>pH H₂O:1:2.5</th>
<th>EC₁:₅ μS cm⁻¹</th>
<th>SAR₁:₅</th>
<th>CaCO₃ mg g⁻¹</th>
<th>Aggstab Δ MWD (mm)</th>
<th>Clay mg g⁻¹</th>
<th>Silt mg g⁻¹</th>
<th>Sand mg g⁻¹</th>
<th>FeO mg g⁻¹</th>
<th>FeD mg g⁻¹</th>
<th>FeO : FeD</th>
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<td>1.47 (0.07)</td>
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<td>7.1 (0.1)</td>
<td>27 (3)</td>
<td>0.4 (0.1)</td>
<td>0 (0)</td>
<td>0.41 (0.06)</td>
<td>127 (7)</td>
<td>230 (20)</td>
<td>643 (24)</td>
<td>0.21 (0.20)</td>
<td>4.9 (0.1)</td>
<td>0.04 (0.04)</td>
</tr>
<tr>
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<td>8.0 (0.2)</td>
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<td>0.4 (0.0)</td>
<td>0 (0)</td>
<td>n.d.</td>
<td>n.d.</td>
<td>n.d.</td>
<td>n.d.</td>
<td>n.d.</td>
<td>n.d.</td>
<td>n.d.</td>
</tr>
<tr>
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<td>1.60 (0.07)</td>
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<td>8.8 (0.1)</td>
<td>152 (35)</td>
<td>0.9 (0.5)</td>
<td>51 (12)</td>
<td>n.d.</td>
<td>95 (13)</td>
<td>121 (22)</td>
<td>784 (35)</td>
<td>0.04 (0.04)</td>
<td>3.0 (0.2)</td>
<td>0.01 (0.01)</td>
</tr>
<tr>
<td></td>
<td>C</td>
<td>2 175.0 (15.0)</td>
<td>1.70 (0.05)</td>
<td>4.3 (0.4)</td>
<td>9.0 (0.1)</td>
<td>236 (101)</td>
<td>1.7 (0.3)</td>
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<td>91 (5)</td>
<td>125 (22)</td>
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<td>0.07 (0.07)</td>
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<tr>
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<td>9.6 (2.2)</td>
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<td>1.02 (0.29)</td>
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<td>330 (17)</td>
<td>497 (26)</td>
<td>0.31 (0.04)</td>
<td>2.8 (0.7)</td>
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</tr>
<tr>
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<td>8.8 (0.1)</td>
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<td>7.0 (0.3)</td>
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<td>n.d.</td>
<td>n.d.</td>
<td>n.d.</td>
<td>n.d.</td>
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<td>n.d.</td>
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<td>8.8 (0.1)</td>
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<td>5.3 (0.9)</td>
<td>152 (34)</td>
<td>n.d.</td>
<td>203 (32)</td>
<td>320 (56)</td>
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<td>796 (333)</td>
<td>3.9 (1.0)</td>
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<td>157 (34)</td>
<td>250 (81)</td>
<td>593 (114)</td>
<td>0.24 (0.08)</td>
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</tr>
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<tr>
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<td>307 (45)</td>
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<td>911 (639)</td>
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<td>0.10 (0.05)</td>
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Table 3: Parameters of OM fractions as function of soil type and horizon. Given are arithmetic means and the standard error of the mean in parentheses. Where n differs for a certain parameter from those indicated in the third column, it is indicated by a separate n in brackets. For LF material neutral sugars were only determined in A horizons, since the sample quantity was too low in the underlying horizons. Abbreviations: OC = organic carbon, MobC = mobilized organic carbon, n.d. = not determined.

<table>
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<tr>
<th>Soil type</th>
<th>Horizon</th>
<th>n</th>
<th>mg fraction</th>
<th>mg fraction lost</th>
<th>mg OC</th>
<th>C : N</th>
<th>mg MobC</th>
<th>% MobC of total OC</th>
<th>% OC of total OC</th>
<th>mg sugar</th>
<th>mg sugar</th>
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<tbody>
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<td></td>
<td></td>
<td></td>
<td>g² soil</td>
<td>g² soil</td>
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<td>g² fraction</td>
<td>g² fraction</td>
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<td>g² fraction</td>
<td>g² fraction</td>
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<tr>
<td>Kastanozem</td>
<td>Ah</td>
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<td>5.3 (0.6)</td>
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<td>34.8 (5.3)</td>
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<td>46.1 - 193.4 - 138.4</td>
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<tr>
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</tr>
<tr>
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<td>7.7 (0.3)</td>
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</tr>
<tr>
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<td>1</td>
<td>997.2 -</td>
<td>66.6 -</td>
<td>1.6 -</td>
<td>7.9 -</td>
<td>0.2 -</td>
<td>n.d.</td>
<td>n.d.</td>
<td>0.3 -</td>
<td>164.8 -</td>
</tr>
</tbody>
</table>
Table 4: Fungi : Bacteria ratio as function of soil type and horizon. Given are arithmetic means and the standard error of the mean in parentheses.

<table>
<thead>
<tr>
<th>Soil type</th>
<th>Horizon</th>
<th>n</th>
<th>Fungi : Bacteria ratio</th>
</tr>
</thead>
<tbody>
<tr>
<td>Kastanozem</td>
<td>Ah</td>
<td>3</td>
<td>0.23 (0.01)</td>
</tr>
<tr>
<td></td>
<td>AC</td>
<td>3</td>
<td>0.20 (0.00)</td>
</tr>
<tr>
<td></td>
<td>Ck</td>
<td>3</td>
<td>0.20 (0.06)</td>
</tr>
<tr>
<td></td>
<td>C</td>
<td>1</td>
<td>0.14 -</td>
</tr>
<tr>
<td>Non-sodic Solonchak</td>
<td>Az</td>
<td>4</td>
<td>0.27 (0.04)</td>
</tr>
<tr>
<td></td>
<td>B</td>
<td>4</td>
<td>0.28 (0.06)</td>
</tr>
<tr>
<td></td>
<td>C</td>
<td>4</td>
<td>0.32 (0.09)</td>
</tr>
<tr>
<td></td>
<td>Cl</td>
<td>1</td>
<td>0.16 -</td>
</tr>
<tr>
<td>Sodic Solonchak</td>
<td>Az</td>
<td>3</td>
<td>0.17 (0.05)</td>
</tr>
<tr>
<td></td>
<td>AC</td>
<td>3</td>
<td>0.16 (0.03)</td>
</tr>
<tr>
<td></td>
<td>C</td>
<td>2</td>
<td>0.36 (0.11)</td>
</tr>
<tr>
<td></td>
<td>Cl</td>
<td>2</td>
<td>0.46 (0.11)</td>
</tr>
</tbody>
</table>
Figures

Figure 1: Schematic representation of study sites and the experimental design. Same colors of the soil profiles and plant samples mark the same soils. A detailed soil type classification of the grouped soils is given in Table S1.
Figure 2: Soil OC stocks (Mg ha⁻¹) for three soil types, (a) as function of horizon and (b) for a depth of 100 cm and the entire soil profile (light and dark grey). Mean depths of the profiles were 157 ± 20 cm (KS), 175 ± 9 cm (nSC) and 141 ± 5 cm (sSC). Given are arithmetic means ± SE, while dots show individual measurements (in plot b) for the entire soil profile. Abbreviations: KS = Kastanozem, nSC = Non-sodic Solonchak, sSC = Sodic Solonchak.
Figure 3: δ¹³C ratios of plant components (upper three panels) and of OM present in the light fraction (LF) and the heavy fraction (HF) as function of soil depth (lower three panels) for three soil types. Grey dots in the upper three panels show individual measurements, while the black dots show arithmetic means ± standard error of the mean. In the lower three panels, the three and four replicates per soil type are shown.
Figure 43: $^{14}$C activity (pMC) for three soil types and two OM fractions as function of soil depth. Rectangles on the left of each panel indicate diagnostic horizons. Due to low quantity of LF material in the subsoil, $^{14}$C activities were only analyzed until the topmost C horizon. Abbreviations: LF = light fraction, HF = heavy fraction.
Figure 4: δ¹³C ratios of plant components (upper three panels) and of OM present in the light fraction (LF) and the heavy fraction (HF) as function of soil depth (lower three panels) for three soil types. Grey dots in the upper three panels show individual measurements, while the black dots show arithmetic means ± standard error of the mean. In the lower three panels, the three and four replicates per soil type are shown.
Figure 55: Biplots derived from a principal components analysis of non-cellulosic neutral sugars from plants, the light fraction (LF) and the heavy fraction (HF), plotted for each soil type separately. The grey dots belong to those samples not considered for the particular soil type. Abbreviations: Man = mannose, Ara = arabinose, Rha = rhamnose, Rib = ribose, Glu = glucose, Fuc = fucose, Xyl = xylose, Gal = galactose.
Figure 6: Biplots derived from a principal components analysis of functional microbial groups as identified from PLFA analysis. Colors and 68% confidence regions are grouped by a) soil type and b) horizon. Abbreviations: KS = Kastanozem, nSC = Non-sodic Solonchak, sSC = Sodic Solonchak, Gram+ = gram-positive bacteria, Gram- = gram-negative bacteria, Actino = actinomycetes, SapFungi = saprotrophic fungi, UnspcBact = unspecific bacteria, AMF = arbuscular mycorrhizal fungi.
Supplement of

Organic matter dynamics along a salinity gradient in Siberian steppe soils

N. Bischoff et al.

Correspondence to: N. Bischoff (bischoff@ifbk.uni-hannover.de)
### Tables

**Table S 1:** Assignment of soil types according to IUSS Working Group WRB (2014) to groups and soil types, respectively, used in the present study.

<table>
<thead>
<tr>
<th>Group / Soil type</th>
<th>Plot</th>
<th>Soil type according to IUSS Working Group WRB (2014)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Kastanozem</td>
<td>I</td>
<td>Calcic Kastanozem (Loamic)</td>
</tr>
<tr>
<td></td>
<td>II</td>
<td>Haplic Kastanozem (Arenic, Loamic)</td>
</tr>
<tr>
<td></td>
<td>III</td>
<td>Haplic Kastanozem (Arenic, Loamic)</td>
</tr>
<tr>
<td>Non-sodic Solonchak</td>
<td>IV</td>
<td>Mollic Solonchak (Loamic)</td>
</tr>
<tr>
<td></td>
<td>V</td>
<td>Mollic Solonchak (Loamic)</td>
</tr>
<tr>
<td></td>
<td>VI</td>
<td>Mollic Solonchak (Alcalic, Loamic)</td>
</tr>
<tr>
<td></td>
<td>VII</td>
<td>Haplic Solonchak (Alcalic, Loamic)</td>
</tr>
<tr>
<td>Sodic Solonchak</td>
<td>VIII</td>
<td>Gleyic Sodic Solonchak (Alcalic, Loamic)</td>
</tr>
<tr>
<td></td>
<td>IX</td>
<td>Sodic Solonchak (Alcalic, Loamic, Humic)</td>
</tr>
<tr>
<td></td>
<td>X</td>
<td>Sodic Solonchak (Alcalic, Loamic, Humic)</td>
</tr>
</tbody>
</table>
Table S 2: Organic carbon (OC), total nitrogen (TN), and C : N ratio of OM as function of soil type and horizon. Given are arithmetic means and the standard error of the mean in parentheses. Abbreviation: n = sample size.

<table>
<thead>
<tr>
<th>Soil type</th>
<th>Horizon</th>
<th>n</th>
<th>OC  ( \text{mg g}^{-1} )</th>
<th>TN  ( \text{mg g}^{-1} )</th>
<th>C : N</th>
</tr>
</thead>
<tbody>
<tr>
<td>Kastanozem</td>
<td>Ah</td>
<td>3</td>
<td>9.28 (0.34)</td>
<td>0.96 (0.03)</td>
<td>9.8 (0.1)</td>
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<tr>
<td></td>
<td>AC</td>
<td>3</td>
<td>5.33 (0.27)</td>
<td>0.63 (0.03)</td>
<td>8.2 (0.3)</td>
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<tr>
<td></td>
<td>Ck</td>
<td>3</td>
<td>2.05 (0.28)</td>
<td>0.30 (0.06)</td>
<td>8.2 (0.7)</td>
</tr>
<tr>
<td></td>
<td>C</td>
<td>2</td>
<td>1.27 (0.28)</td>
<td>0.25 (0.05)</td>
<td>5.7 (1.0)</td>
</tr>
<tr>
<td>Non-sodic Solonchak</td>
<td>Az</td>
<td>4</td>
<td>17.38 (2.90)</td>
<td>1.78 (0.31)</td>
<td>9.8 (0.1)</td>
</tr>
<tr>
<td></td>
<td>B</td>
<td>4</td>
<td>4.04 (0.62)</td>
<td>0.52 (0.07)</td>
<td>7.7 (0.2)</td>
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<tr>
<td></td>
<td>C</td>
<td>4</td>
<td>1.74 (0.13)</td>
<td>0.29 (0.01)</td>
<td>6.1 (0.4)</td>
</tr>
<tr>
<td></td>
<td>Cl</td>
<td>4</td>
<td>1.10 (0.18)</td>
<td>0.24 (0.02)</td>
<td>4.8 (0.6)</td>
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<tr>
<td>Sodic Solonchak</td>
<td>Az</td>
<td>3</td>
<td>24.53 (2.34)</td>
<td>2.53 (0.24)</td>
<td>9.7 (0.2)</td>
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<td></td>
<td>ACz</td>
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<td>11.07 (2.77)</td>
<td>1.10 (0.29)</td>
<td>10.0 (0.1)</td>
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<tr>
<td></td>
<td>C</td>
<td>2</td>
<td>3.46 (1.04)</td>
<td>0.40 (0.10)</td>
<td>8.8 (0.4)</td>
</tr>
<tr>
<td></td>
<td>Cl</td>
<td>3</td>
<td>1.34 (0.36)</td>
<td>0.24 (0.03)</td>
<td>6.0 (1.0)</td>
</tr>
</tbody>
</table>
Figure S1: Concentration of water-soluble anions and cations (µmol g⁻¹ soil) for three soil types as a function of soil depth. For better visualization measured data are omitted and only the local polynomial regression fits (LOESS) are shown.
Figure S2: X-ray powder diffractograms of bulk soil from three profiles and respective horizons. Intensities are square root transformed for better visualization of trace mineral phases.
Figure S3: X-ray diffractograms of clay fractions from two treatments of three soil types and four different horizons. Abbreviations: S = smectite, V = vermiculite, C = chlorite, I = illite, K = kaolinite, and Q = quartz.
Figure S-4: Scanning electron micrographs (a, c, d) and EDX spectra (b) of the fine earth fraction of Solonchak. The two common salt minerals halite (a, b) and thenardite (c, d) are shown. In a), halite is represented by the white region embedded in gypsum crystals. In c), thenardite is the finely textured coating on the larger mineral. d) is the enlargement of c).
Figure S 25: Percentage contribution of arabinose to the total non-cellulosic neutral sugars of three soil types, separated for plants, light fraction (LF) and heavy fraction (HF).
Figure S 3c: Percentage contribution of glucose to the total non-cellulosic neutral sugars of three soil types, separated for plants, light fraction (LF) and heavy fraction (HF).
Supplement S1 – Soil mineralogical composition

Material & Methods

X-ray diffractograms of ball-milled <2-mm fractions were recorded with an X’Pert PRO MPD Θ-Θ diffractometer (PANalytical, Almelo, Netherlands) equipped with a Cu anode producing Kα radiation. The powder samples were scanned from 2° to 85° 2Θ with a step size of 0.02° 2Θ and 3 s per step. A subset of samples was evaluated at the micro-scale using a Quanta 600 FEG environmental scanning electron microscope (ESEM; FEI Company, Hillsboro, USA) with an acceleration voltage of 20 keV. As the analysis was carried out in low-vacuum mode (0.6 mbar), sputtering of the samples with gold or carbon was not necessary. The microscope was equipped with an Apollo XL EDX detector (Ametek Inc., Berwyn, USA).

Clay mineralogical analyses were carried out for one representative soil profile of each soil type. Clay fractions (<2 µm) were obtained by pre-treating the soil with acetic acid (removal of carbonates), H2O2 (removal of OM), and dithionite-citrate (removal of iron oxides), subsequent separation by sedimentation (Stoke’s law) and final Mg2+ saturation to cause flocculation and thus easier handling of samples. X-ray diffraction patterns were recorded using the same system and settings as for the powder analyses of bulk soil but with Co-Kα radiation generated at 40 kV and 40 mA. Oriented mounts were prepared on porous ceramic tiles to avoid segregation of fine particles during sedimentation (Dohrmann et al., 2009) and scanned from 2° to 35° 2Θ with a step size of 0.02° 2Θ and 4 s per step. Sample quantity allowed only for two treatments for qualitative analysis: (i) Mg2+, (ii) Mg2+ + ethylene glycol. The ethylene glycol treatment was used as it detects expandable clay minerals like smectites, which strongly affect the physical properties of sodic soils.

Results

The three soil types had a quite homogenous mineralogical composition, dominated by quartz and feldspars as well as calcite and dolomite in the carbonate-rich horizons, whereas almost all samples showed small quantities of amphibole and muscovite (Figure S4). In the Solonchaks also gypsum was present. Calcite and dolomite XRD peaks were very broad. This peak broadening is related to very fine crystallite sizes. The clay fraction showed small amounts of illite, kaolinite, and chlorite, while smectites were partially present in the subsoil, and in the Sodic Solonchak also in the topsoil. In the smectite-rich horizons, the quantities of smectite and illite exceeded those of chlorite and kaolinite significantly (Figure S5). In the Solonchaks, the quantities of water-soluble salts were small when related to the bulk soil. Mass balance calculations (data not shown) and analyses by ESEM-EDX (Figure S6) revealed that water-soluble salts mostly consisted of thenardite (α-Na2SO4) and halite (NaCl), but also bischofite (MgCl2•6H2O) could be present.
Figure S 42: X-ray powder diffractograms of bulk soil from three profiles and respective horizons. Intensities are square root transformed for better visualization of trace mineral phases.
Figure S5: X-ray diffractograms of clay fractions from two treatments of three soil types and four different horizons. Abbreviations: S = smectite, V = vermiculite, C = chlorite, I = illite, K = kaolinite, and Q = quartz.
Figure S6: Scanning electron micrographs (a, c, d) and EDX spectra (b) of the fine earth fraction of Solonchaks. The two common salt minerals halite (a, b) and thenardite (c, d) are shown. In a), halite is represented by the white region embedded in gypsum crystals. In c), thenardite is the finely textured coating on the larger mineral, d) is the enlargement of c).
References
