<https://helda.helsinki.fi>

Effects of different forms of N deposition on leaf litter decomposition and extracellular enzyme activities in a temperate grassland

Dong, Lili

2019-07

Dong , L , Sun , T , Berg , B , Zhang , L & Zhang , Q 2019 , ' Effects of different forms of N deposition on leaf litter decomposition and extracellular enzyme activities in a temperate grassland ' , Soil Biology & Biochemistry , vol. 134 , pp. 78-80 . <https://doi.org/10.1016/j.soilbio.2019.03.016>

<http://hdl.handle.net/10138/327966>

<https://doi.org/10.1016/j.soilbio.2019.03.016>

cc_by_nc_nd

Downloaded from Helda, University of Helsinki institutional repository.

This is an electronic reprint of the original article.

This reprint may differ from the original in pagination and typographic detail.

Please cite the original version.

Accepted Manuscript

Effects of different forms of N deposition on leaf litter decomposition and extracellular enzyme activities in a temperate grassland

Lili Dong, Tao Sun, Björn Berg, Lili Zhang, Quanquan Zhang

PII: S0038-0717(19)30077-X
DOI: 10.1016/j.soilbio.2019.03.016
Reference: SBB 7444
To appear in: *Soil Biology and Biochemistry*
Received Date: 08 November 2018
Accepted Date: 14 March 2019

Please cite this article as: Lili Dong, Tao Sun, Björn Berg, Lili Zhang, Quanquan Zhang, Effects of different forms of N deposition on leaf litter decomposition and extracellular enzyme activities in a temperate grassland, *Soil Biology and Biochemistry* (2019), doi: 10.1016/j.soilbio.2019.03.016

This is a PDF file of an unedited manuscript that has been accepted for publication. As a service to our customers we are providing this early version of the manuscript. The manuscript will undergo copyediting, typesetting, and review of the resulting proof before it is published in its final form. Please note that during the production process errors may be discovered which could affect the content, and all legal disclaimers that apply to the journal pertain.

Effects of different forms of N deposition on leaf litter decomposition and extracellular enzyme activities in a temperate grassland

Lili Dong^a, Tao Sun^{b*}, Björn Berg^{c,d}, Lili Zhang^{e*}, Quanquan Zhang^f

^aErguna Forest-Steppe Ecotone Research Station, Institute of Applied Ecology, Chinese Academy of Sciences, Shenyang 110016, China;

^bKey Laboratory of Forest Ecology and Management, Institute of Applied Ecology, Chinese Academy of Sciences, Shenyang 110016, China;

^cDepartment of Forest Sciences, University of Helsinki, Helsinki 00014, Finland;

^dSection of Biology, University of Gävle, SE-801 76 Gävle, Sweden;

^eKey Laboratory of Forest Ecology and Management, Institute of Applied Ecology, Chinese Academy of Sciences, Shenyang 110016, China;

^fSchool of international education, Beijing University of Chemical Technology, Beijing 102202, China;

*Corresponding author: Tao Sun or Lili Zhang

Tel.: +86-024-83970896

Fax.: +86-024-83970200

E-mail address: suntao28329@163.com or llzhang@iae.ac.cn

20 **Abstract**

21 Despite the importance of decomposition for biogeochemical cycles, it is still not
22 clear how this process is affected by different forms of nitrogen (N). Equal amounts of
23 N with different ratios of inorganic N : organic N (0 : 0, 10 : 0, 7 : 3, 5 : 5, 3 : 7, and
24 0 : 10) were added to the soil in a steppe. We studied the response of litter
25 decomposition
26 to different forms of N enrichment. The treatment with 30% organic N resulted in the
27 fastest decomposition, which was higher than with inorganic N or organic N addition
28 alone. Our results highlight the need for studies of N deposition on carbon cycles that
29 consider different components in N deposition.

30 **Keywords:** *Litter decomposition; Inorganic nitrogen deposition; Organic nitrogen*
31 *deposition; Extracellular enzyme activity ; Grassland*

Litter decomposition, as an important first step for the formation of soil humus, is a key biogeochemical process that influences the recycling of nutrients, carbon (C) and energy within and among ecosystems. Despite the importance of litter decomposition for ecosystem biogeochemical cycles (Vitousek, 2004), how this fundamental process is affected by different forms of nitrogen (N) is still uncertain.

Numerous studies have focused on effects of N deposition on decomposition by examining only the addition of inorganic N (IN), but ignoring the organic nitrogen (ON) component, which may lead to an underestimation of N retention effects on ecosystems (Cornell, 2003). N deposition not only encompasses IN, but also ON (e.g. urea and amino acids; Neff et al., 2002), which may contribute to 30% of total atmospheric N deposition (Cornell 2011). N added as IN and ON may have different effects on litter decomposition, given that ON is more readily bioavailable (Seitzinger et al., 2002) and a preferential N source for decomposer microorganisms (Hobbie, 2005). However, the effects of different types of N deposition on litter decomposition have rarely been examined. One exception is a study by Hobbie et al. (2012) that reported that either inorganic or organic N accelerated the early stage of decomposition rate, but suppressed the later stage of decomposition for pin oak leaves in Northern American temperate forests.

In the present article, we determined the effects of different IN-to-ON ratios on leaf litter decomposition of *Leymus chinensis* (*L.chinensis*) in a temperate grassland. We hypothesized that all forms of fertilized-N would accelerate decomposition and that the rate of decomposition would increase with the proportion of ON.

This decomposition experiment was established in a temperate steppe in Inner Mongolia, China (50°10'46.1"N; 119°22'56.4"E). Detail descriptions can be found in Wang et al. (2018). Mean N deposition is 8.1 kg N ha⁻¹ among the growing seasons, (Li et al., 2015). *L.chinensis* is the dominant species in this region, and can contribute > 50% of the total aboveground biomass. In this study, NH₄NO₃ was chosen as the IN source, while urea and glycine were selected and mixed at equal proportions as the ON sources. Plots received one of the following treatments with different ratios of IN/ON (six replicate plots per treatment): N0 (control, no N addition), N1 (10:0), N2 (7:3), N3 (5:5), N4 (3:7), N5 (0:10). A total amount of 10 g N m⁻² yr⁻¹ was added in each plot once a year in May since 2014.

We constructed 15 cm × 15 cm nylon litter bags using 1-mm mesh. Previous studies have demonstrated that this mesh size is permeable to a variety of soil fauna (Berg and McClaugherty, 2014). Each bag was filled with approximately 8 g leaf litter from the non-fertilized sites. 12 litterbags were randomly deployed in each plot and fixed on the ground using 10-cm long metal pegs in late September 2015. Three replicate bags were retrieved from each plot in May and September in both 2016 and 2017. Harvested leaf litter was cleaned of soil, roots, and then subsampled for dry mass (65 °C) and enzyme activities (4 °C). According to the method of Saiya-Cork et al. (2002), we analyzed the activities of the following cellulolytic and ligninolytic enzymes: β-1,4-glucosidase (β-GC, EC 3.2.1.21), cellobiohydrolase (CBH, EC 3.2.1.91), phenol oxidase (POX, EC 1.10.3.2) and peroxidase (PER, EC 1.11.1.7). The effects of N addition on decomposition rates (*k*, Olson, 1963) were analyzed using ANOVA with

enzyme activity analyzed using repeated-measures ANOVA.

Litter decomposition rates (k) after two years of field-exposure were significantly different between N0 and all N treatments ($P < 0.01$ in all cases; Fig. 1), which were ca. 16-45% higher in N fertilization than in control treatments. Decomposition rate was highest in 30% organic N plots but was not significantly different from 50% or 70% organic N plots (Fig. 1). As hypothesized, all forms of N treatments (inorganic or organic N) stimulated the decomposition rates of *L. chinensis*. The positive effect of N additions on litter decomposition was probably due to the alleviation of N-limitations for microbial metabolism (Hobbie and Vitousek, 2000) and acceleration of microbial activities (Waldrop et al., 2004). However, our results showed that the magnitude of N effects varied among different IN-to-ON ratios, with both types (organic and inorganic) of N addition promoting decomposition rates more than just only either organic or inorganic N addition ($P < 0.01$ in all cases, Fig. 1, Table 1). Thus, the microbe-dominated decomposition process was more readily influenced by mixed types of N addition. This is probably because the mixed forms of N addition supply provide a wider use of N sources for decomposing microbes to grow on (Lochhead and Chase, 1943; Brown, 1980; Jones et al., 2004)

However, in contrast to our expectations, we did not observe the promotion of N addition effects on decomposition increase with the increasing proportion of organic N (Fig. 1). In fact, after two years of field exposure, decomposition rates in the 100% inorganic N treatments was not different from the 100% organic N treatments (Fig. 1). This is possible because of the energetic costs of assimilating different types of N

were different enough to result into differences in the rate of C use and thus mass loss rates.

Decomposition is dependent on a suite of microbe-mediated mechanisms (Cusack et al., 2013) that underlie extracellular enzymes' breakdown of the structural components of plant litter and the recover of organic N and phosphorus, thus creating a dynamic nutrient cycle system (Carreiro et al., 2000). N amendment significantly enhanced the hydrolytic enzymes' activity by 61%-100% for β -GC and by 84%-222% for CBH and peaked at N2 compared with the control in the first sampling ($P < 0.001$, Fig. 2a and 2b), leading to a larger and faster litter decomposition in the initial phase (Hobbie et al., 2012; Sun et al., 2016). The activity of oxidative enzymes, peaked in the last sampling at mass loss of 55% (Table 1), and varied among different forms of N addition (negative effect of IN vs. positive effect of mixed type vs. neutral effects of ON), and greatly increased at mixed type, especially under N2 ($P < 0.01$; Fig. 2c and 2d) indicating accelerated degradation of late lignin-dominated stage (Berg, 2014). Consequently, the "bell-shaped" response of decomposition rate to N addition could arise for reasons that N with different forms may not be equivalent in quantity or quality for the microbial community (Wang et al., 2011; Li et al., 2014), and the contrasting responses of lignin-degrading enzyme activity to IN versus ON-containing inputs helps explain the effects of N form on decomposition.

Collectively, our results highlight the need for studies of N effects on litter decomposition that consider the different N components in N deposition. Predicting the potential effects of natural N deposition on C and biogeochemical cycles requires

that organic N sources are also considered when simulating N deposition experiments.

Acknowledgments

We acknowledge Peng Zan, Weiping Gu, Pengyu Ma, and Luning Wen for litter sampling and processing in the field and laboratory. We also acknowledge chief editor Tim Clough and two anonymous reviewers for their helpful comments. The funding for this research was supported by the State Key Program of China (2016YFD0300904), the State Key Program of China (2016YFA0600800), Natural Science Foundation of China (31500361 and 31830015) and Key Research Program of Frontier Sciences of Chinese Academy of Sciences (QYZDB-SSW-DQC002).

References

- Berg, B., McClaugherty, C., 2014. Plant litter - Decomposition, Humus Formation, Carbon Sequestration. Springer Verlag, Heidelberg, Berlin. pp. 317.
- Berg, B., 2014. Decomposition patterns for foliar litter- A theory for influencing factors. *Soil Biology & Biochemistry* 78, 222-232.
- Berg, B., 1986. Nutrient release from litter and humus in coniferous forest soils- a mini review. *Scandinavian Journal of Forest Research* 1, 359-369.
- Brown, C.M., 1980. Ammonia assimilation and utilization in bacteria and fungi. In: Payne JW., Ed. *Microorganisms and nitrogen sources*. New York: John Wiley and Sons. 511-536.
- Carreiro, M.M., Sinsabaugh, R.L., Repert, D.A., Parkhurst, D.F., 2000. Microbial enzyme shifts explain litter decay responses to simulated nitrogen deposition.

- Ecology 81, 2359-2365.
- Cusack, D.F., 2013. Soil nitrogen levels are linked to decomposition enzyme activities along an urban-remote tropical forest gradient. Soil Biology & Biochemistry 57, 192–203.
- Cornell, S.E., Jickells, T.D., Cape, J.N., Rowland, A.P., Duce, R.A., 2003. Organic nitrogen deposition on land and coastal environments: a review of methods and data. Atmospheric Environment 37, 2173–2191.
- Cornell, S.E., 2011. Atmospheric nitrogen deposition: revisiting the question of the importance of the organic component. Environmental Pollution 159, 2214–2222.
- Fog, K., 1988. The effect of added nitrogen on the rate of decomposition of organic matter. Biological Reviews 63, 433-462.
- Hobbie, S.E., Vitousek, P.M., 2000. Nutrient limitation of decomposition in Hawaiian forests. Ecology 81, 1867-1877.
- Hobbie, S.E., 2005. Contrasting Effects of Substrate and Fertilizer Nitrogen on the early Stages of Litter Decomposition. Ecosystems 8, 644–656.
- Hobbie, S.E., Eddy, W.C., Buyarski, C.R., Adair, E.C., Ogdahl, M.L., Weisenhorn, P., 2012. Response of decomposing litter and its microbial community to multiple forms of nitrogen enrichment. Ecological Monographs 82, 389-405.
- Jones, D.L., Shannon, D., Murphy, D.V., Farrar, J., 2004. Role of dissolved organic nitrogen (DON) in soil N cycling in grassland soils. Soil Biology & Biochemistry 36, 749-756.
- Lochhead, A.G., Chase, F.E., 1943. Nutritional requirements of the predominant

- bacterial flora. *Soil Science* 55, 185-195.
- Li, S., Du, Y., Guo, P., Guo, L., Qu, K., He, J., 2014. Effects of different types of N deposition on the fungal decomposition activities of temperate forest soils. *Science of the Total Environment* 497–498, 91–96.
- Neff, J.C., Holland, E.A., Dentener, F.J., McDowell, W.H., Russell, K.M., 2002. The original composition and rates of organic nitrogen deposition: a missing piece of the nitrogen cycle? *Biogeochemistry* 57, 99-136.
- Olson, J.S., 1963. Energy storage and the balance of producers and decomposers in ecological systems. *Ecology* 44, 322-331.
- Seitzinger, S.P., Sanders, R.W., Styles, Renée., 2002. Bioavailability of DON from natural and anthropogenic sources to estuarine plankton. *Limnology and Oceanography* 47(2), 353-366.
- Saiya-Cork, K.R., Sinsabaugh, R.L., Zak, D.R., 2002. The effects of long term nitrogen deposition on extracellular enzyme activity in an *Acer saccharum* forest soil. *Soil Biology & Biochemistry* 34:1309 – 1315.
- Sun, T., Dong, L.L., Wang, Z.W., Lü, X.T., Mao, Z.J., 2016. Effects of long-term nitrogen deposition on fine root decomposition and its extracellular enzyme activities in temperate forests. *Soil Biology & Biochemistry* 93, 50-59.
- Vitousek, P.M., 2004. *Nutrient Cycling and Limitation: Hawai'i as a Model System*. Princeton University Press, New Jersey, USA.
- Waldrop, M.P., Zak, D.R., Sinsabaugh, R.L., 2004. Microbial community response to nitrogen deposition in northern forest ecosystems. *Soil Biology & Biochemistry*

- 185 36, 1443–1451.
- 186 Wang C., Han G., Jia Y., Feng X., Guo P., Tian X. 2011. Response of litter
187 decomposition and related soil enzyme activities to different forms of nitrogen
188 fertilization in a subtropical forest. *Ecological Research* 26, 505–513.
- 189 Wang, H.Y., Wang, Z.W., Ding, R., Hou, S.L., Yang, G.J., Lü, X.T., Han, X.G., 2018.
190 The impacts of nitrogen deposition on community N:P stoichiometry do not
191 depend on phosphorus availability in a temperate meadow steppe. *Environmental*
192 *Pollution* 242, 82-89.
- 193 Zhang, Y., Song, L., Liu, X.J., 2012. Atmospheric organic nitrogen deposition in
194 China. *Atmospheric Environment* 46, 195–204.

Fig. 1. Adjusted mean + SE decomposition rates (single exponential model). Different letters among columns indicate significant differences for Tukey's HSD post hoc comparisons among different forms of N treatment ($p < 0.05$). N0, Control; N1, IN-to-ON=10:0; N2, 7:3; N3, 5:5; N4, 3:7; N5, 0:10. The N2 treatment was consistent with the average contribution of ON to atmospheric N deposition in China (28%) and globally (30%) (Cornell et al., 2011; Zhang et al., 2012).

Fig. 2. Extracellular enzyme activity on litter substrates sampled during the two-year experiment under different forms of N treatment. An asterisk indicates that a particular treatment differed significantly from the control at a particular sampling time ($p < 0.05$). The hydrolytic enzymes' activity peaked in the second sampling while the oxidative enzymes at last sampling.

- Effects of different types of N addition on litter decomposition were examined
- Different ratios of inorganic N to organic N were used to fertilize grassland soil
- All forms of N addition increased leaf litter decomposition rates
- Mixed types of N promoted decomposition more than just only type of N addition

Table 1 Cumulative mass loss (in per cent of total initial dry mass) at each sampling in the different N treatments. Different superscript letters represent significant differences among different N treatments (a for the highest value at each sampling).

N treatment	Mass loss at each sampling (%)			
	May 2016	Sep. 2016	May 2017	Sep. 2017
N0	17.0 (1.5) ^c	31.5 (1.8) ^c	42.9 (1.3) ^c	48.4 (0.5) ^c
N1	22.6 (1.9) ^b	35.0 (1.1) ^b	46.8 (1.1) ^b	53.1(0.3) ^b
N2	28.9 (2.5) ^a	43.2 (2.2) ^a	53.6 (0.8) ^a	59.9 (0.7) ^a
N3	26.3 (1.8) ^a	42.9 (1.6) ^a	51.0 (0.7) ^a	57.3 (1.2) ^a
N4	25.0 (2.7) ^a	40.8 (1.3) ^{ab}	49.8 (1.0) ^b	56.7 (0.4) ^{ab}
N5	21.4 (1.7) ^b	36.9 (0.7) ^b	46.7 (0.9) ^b	56.1(0.6) ^{ab}