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Effects of sulfuric, nitric, and mixed acid rain on litter decomposition, soil microbial biomass, and enzyme activities in subtropical forests of China

Yanna Lv[a], Congyan Wang[c], Yanyan Jia[b], Wenwen Wang[b], Xu Ma[b], Jingjing Du[b], Gaozhong Pu[b], Xingjun Tian[b,*]

[a] School of Pharmacy and Biological Sciences, Weifang Medical University, Weifang 261053, China
[b] School of Life Sciences, Nanjing University, Nanjing 210093, China
[c] School of the Environment and Safety Engineering, Jiangsu University, Zhenjiang 212013, China

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ABSTRACT

Acid rain pollution is changing gradually from sulfuric acid rain (SAR) to mixed acid rain (MAR) and then to nitric acid rain (NAR) with the rapidly growing number of motor vehicles. The influences of changed acid rain types on ecosystem functions, particularly on litter decomposition, remain unclear. Two dominant litter types from a coniferous forest and a broad-leaved forest were incubated in microcosms with original forest soils and treated by five types of acid rain with different SO_4^{2-} to NO_3^- ratios (1:0, 5:1, 1:1, 1:5, and 0:1). During a six-month incubation period, litter mass losses, soil microbial biomass, and enzyme activities were investigated. Results showed that various acid treatments inhibited litter decomposition, soil microbial biomass, and most enzyme activities, and the inhibitory effects of NAR were more significant than those of SAR and MAR. The resistance to external acid of microbial communities in broad-leaved forest was higher than that in coniferous forest. NAR and MAR treatments slowed down soil carbon (C), nitrogen (N), and phosphorus (P) mineralization by attenuating the correlations between litter mass losses and the enzymes involved in C, N, and P cycling. Results reveal that the ratio of SO_4^{2-} to NO_3^- in acid rain is an important factor which profoundly influences litter decomposition process. In the future, a decreasing ratio of SO_4^{2-} to NO_3^- in acid rain will be observed in subtropical forests. Thus, soil C would accumulate as a consequence of future acid precipitation, and this may seriously affect the balance of ecosystem C, N flux.

1. Introduction

Acid rain has now become a global environmental problem because of population increase (Mouli et al., 2005) and rapid economic development (Li, 2003; Wang et al., 2012). The influences of acid rain on the natural ecosystems are an increasing environmental concern. Acid rain has many effects on ecosystems, such as the acidification of soil (Liao et al., 2007), the reduction in functioning of microbial communities (Kuperman and Edwards, 1997), the decrease of enzyme activities (Jagels et al., 2002; Singh et al., 2004), and the alteration of forest species composition (Schaberg et al., 2001). In China, the rapid economic growth, with its attendant energy demand and consumption, has caused very serious acid rain pollution throughout the country (Tu et al., 2005). Acid precipitation has occurred in about 40% of the entire territory, especially in fast developing industrial regions, such as the areas around the Yangtze River (Ling et al., 2010).

The major sources of acid rain are sulfur dioxide (SO_2) and nitrogen oxides (NO_x), which react with the water molecules in the atmosphere to produce acids (Zhang et al., 2007). Since the late 1990s, given the policies on controlling and mitigating SO_2 emissions and energy structure changes, sulfate ion (SO_4^{2-}) in precipitation has decreased significantly. On the other hand, NO_x emissions have increased significantly because of the rapidly growing number of motor vehicles, and then the relative contribution of nitrate ion (NO_3^-) to acidification has increased significantly along with them. Consequently, the ratio of SO_4^{2-} to NO_3^- in precipitation has decreased from 6 in 2003 to 5 now in Nanjing, China (Tu et al., 2005; Wang et al., 2010). Thus, acid rain pollution is gradually changing from sulfuric acid rain (SAR) to mixed acid rain (MAR), and then to nitric acid rain (NAR). This change of acid rain types further complicates the ongoing challenge of ecosystem stability and increases risks to the ecosystem (Xu and Ji, 2001).

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* Corresponding author at: School of Life Sciences, Nanjing University, Hankou Road 22, Nanjing 210093, China. Tel.: +86 25 83686787; fax: +86 25 83686787.
E-mail addresses: lynlyna@163.com (Y. Lv), tianxj@nju.edu.cn (X. Tian).

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Most studies about the effects of acid rain on forest ecosystems focus on the effects of a single acid rain type, especially SAR (e.g., Dangles et al., 2004; Wang et al., 2010). However, very little is known about the effects of different acid rain types on forest ecosystems, especially on subtropical forests in China.

Coniferous forest and broad-leaved forest are typical stages in the progression of subtropical forests. They play important roles in global carbon (C) dynamics, partly because of their high productivity and nutrient turnover (Hughes et al., 1999; He et al., 2007). Meanwhile, leaf litter decomposition and its accompanied release of nutrients and formation of soil organic matter are fundamental processes in humus formation, nutrient cycling, and ecosystem C flux (Hoorens et al., 2003; Wang et al., 2010, 2012). Thus, studying and identifying the effects of different acid rain types on litter decomposition process in the subtropical soils of coniferous and broad-leaved forest is important for understanding their functions in nutrient cycling.

Soil extracellular enzymes and microbial biomass play a pivotal role in litter decomposition and nutrient cycling in forest ecosystems (Dick et al., 2000; Enowashu et al., 2009). Measurement of soil extracellular enzyme activities and microbial biomass has been recommended as the most appropriate indicator of microbial decomposition, soil fertility, and ecological stability (Ajwa et al., 1999; Caldwell, 2005). Thus, changes in soil enzyme activities and microbial biomass during acid rain treatment can explain the ecological effects of acid rain on litter decomposition process in forest ecosystems.

Microcosm experiments under controlled laboratory conditions are useful for investigating the factors that influence litter decomposition (Naeem et al., 2000; Manning et al., 2008). Thus, this study was carried out through a microcosm experiment, in order (1) to compare the difference in litter decomposition between coniferous forest and broad-leaved forest in response to different types of simulated acid rain, (2) to measure the effects of different acid rain types on soil microbial biomass and enzyme activities, and (3) to identify the major degradative enzyme contributors involved in litter decomposition in subtropical forest after exposure to different types of acid rain. In this work, we hypothesize that (1) acid rain treatments would depress litter decomposition process of the two forest types, and the inhibitory effects of SAR, MAR, and NAR with the same acidity would be equivalent, and (2) the negative effects of acid rain on the litter decomposition of broad-leaved forest would be more significant than on those of coniferous forest because the initial soil acidity of coniferous forest is higher than that of broad-leaved forest.

2. Materials and methods

2.1. Site description

The study was conducted in two forest types, coniferous forest (dominated by *Pinus massoniana*) and broad-leaved forest (dominated by *Quercus variabilis*), of Zijin Mountain (32°5′ N, 118°48′ E), Nanjing, China. Zijin Mountain has an area of 24 km^2, an altitude of 447.1 m, and a subtropical humid climate. The annual mean temperature is 15.4 °C, with a monthly mean temperature reaching a maximum of 28.2 °C in July and a minimum of 1.9 °C in January. The rainy season is from June to July, and the average annual precipitation is 1106.5 mm. The litter coverage rate reaches 90%. The soil is classified as slightly acidic Humic Cambisol with a pH of about 5.0 (FAO, 1987). Considerable amount of nutrients and organic matters are accumulated in the humus layer. The bedrock materials are sandstones and shales. The annual average pH value of rainfall is approximately 4.98, with an acid rain frequency of approximately 55.8% (Wang et al., 2007).

Table 1
Initial litter characteristics of *Pinus massoniana* needles and *Quercus acutissima* leaves. Data with different superscript letters in a row are significantly different.

Composition	Needles	Leaves
Total C (%)	51.05^a	48.84^b
Total nitrogen (N) (%)	0.78^a	0.66^b
Total hydrogen (H) (%)	6.78^a	6.56^b
Klason lignin (%)	41.18^a	30.88^b
Total polyphenol (mg gallic acid equiv. g^{-1})	192.97^a	206.37^a
C:N	65.18^b	73.63^a
Klason lignin:N	52.59^a	46.54^b

Table 2
Initial soil properties (0–5 cm depth) of coniferous forest and broad-leaved forest. Data with different superscript letters in a transverse row indicate a significant difference.

Properties	Coniferous forest soil	Broad-leaved forest soil
pH	5.17^b	5.26^a
Moisture (%)	33.87^a	33.31^a
Total C (%)	7.07^a	8.50^a
Total N (%)	0.39^b	0.48^a
Total H (%)	1.19^a	1.43^a
C:N	18.26^a	17.71^a

Table 3
Major ions in two basic solutions of control and simulated acid rain treatments (μmol L^{-1}).

pH	Cl^-	F^-	Ca^{2+}	NH_4^+	Mg^{2+}	K^+	Na^+
5.6	21.04	0.19	5.34	5.01	1.38	1.32	0.30
5.0	23.85	0.22	6.67	5.33	1.42	1.36	0.42

2.2. Experiment design

Three discrete sites (1.5 m × 1.5 m), separated from each other by approximately 10 m, were respectively chosen in coniferous forest and broad-leaved forest. In October 2011, *P. massoniana* needles and *Q. variabilis* leaves were collected from the forest floor of the aforementioned sites (The initial litter quality is shown in Table 1). All litter samples were taken back to the laboratory and oven-dried at 60 °C for 24 h to achieve a constant weight for further study. Meanwhile, three soil samples from each forest type were collected from the top layers (0–5 cm) of each site (The initial soil properties are shown in Table 2). All soil samples were kept in sealed bags and immediately (after ∼2 h) taken back to the laboratory for further study. Soil samples were passed through a 2-mm sieve to remove leaves, plant roots, gravel, and stones. Three soil samples from each forest type were then homogenized and kept in a refrigerator at 4 °C for 24 h in preparation for further incubation.

To study litter decomposition process, 0.5 g of the oven-dried litter was mixed with 40 g soil in a 125-mL polypropylene specimen chamber. The needles were incubated in coniferous forest soil, and the leaves in broad-leaved forest soil. This experiment employed a full-factorial experimental design with two litter types (needles and leaves) × six treatments expressed by control, SAR, MAR1, MAR2, MAR3, and NAR. Five stock solutions of acid rain were prepared by mixing 0.5 mol L^{-1} H_2SO_4 and 0.5 mol L^{-1} HNO_3 at ratios of 1:0, 5:1, 1:1, 1:5, and 0:1. Basic solutions of control and acid rain treatments were then prepared according to Wang et al. (2010) (Table 3). For control treatment, the pH value of basic solution was finally buffered to 5.6 by adding the stock solutions of H_2SO_4 and HNO_3 with a ratio of 5:1. For the five acid rain treatments, the pH values of basic solution were finally buffered to 5.0 by adding the aforementioned five stock solutions, respectively. In this study, pH 5.6 and the ratio of 5:1 for H_2SO_4 and HNO_3 were the natural values of unpolluted rainfall, and pH 5.0 was approximately the annual average pH value of rainfall in the study sites. As such,