Alpine soil nutrient and microbial biomass properties at different altitudes in winter

P Yin¹,², XJ. Liu¹,², JH Liao², X Hu¹,²,*

¹Bamboo Diseases and Pests Control and Resources Development Key Laboratory of Sichuan Province, Leshan Sichuan, 614004, China
²School of Life Sciences, Leshan Normal University, Leshan Sichuan, 614004, China

*Corresponding author: 250095515@qq.com

Abstract. Soil ecology processes play an important role in mineral element transformation and plant distribution in winter. To further understand the ecological processes in alpine soil in winter, soil samples were collected at four altitudes (890 m, 1,800 m, 2,400 m, and 3,020 m) at the Mount Emei, Sichuan Province, China. Additionally, the soil temperature, water content, microbial biomass, and soil nutrients contents were determined at these altitudes. A regular trend of changes in the soil temperature with the increase in the altitudinal gradient was established. Except for TP and TK, the contents of soil OM, TN, NH₄⁺-N, NO₃⁻-N, MBC, and MBN were the highest at the altitude of 2,400 m, characterized by unfrozen soil, a lower temperature, and a higher humidity. Our findings indicate that the organic matter content and mineral nutrition level of the high-altitude soils at Mount Emei are high and provide a pool of nutrients in amounts that are sufficient for the growth of plants in the non-growing season and the spring of the next year. The research results obtained not only fill in the knowledge gap of the soil ecological processes in winter in Mount Emei, but also provide a theoretical basis for research on soil nutrient cycling and vegetation distribution patterns in the Mount Emei area.

1. Introduction

The study of soil ecology for the growing season is a "traditional project" of forest ecology, while soil ecological process is relatively inadequate in the long cold season. An increasing number of studies have shown that vigorous vitality exists in soils in winter and even under ice and snow [1-3]. For example, Sommerfeld et al. (1993) found that the soil under snow in winter continued to discharge CO₂ and N₂O from respiration. Moreover, they found that the release flux of N₂O in winter was even greater than that in the growth season [4]. In addition, Taylor & Jones (1990) reported that litter decomposition in winter usually accounted for more than 40%-60% of the yearly rate [5].

Research over the recent 20 years has shown that in the cold season, soil microbes are neither frozen nor dormant but maintain considerable microbial activity. Even when the soil is frozen, microbial activity continues [6-8]. The activity of microbes inevitably affects the decomposition process of litter in winter, which has a substantial effect on the dynamic changes of the available carbon sources in the soil and ultimately influences the processes of soil nutrition and material circulation [9-10].
Soil ecology issues have become a topic of intensive research interest. However, most studies are focused on investigations on the growing season and farmland ecosystems. Few examinations have been performed on soil ecology in the mountains in winter, mainly in countries in North America and Northern Europe (i.e., the United States, Canada, Sweden, Finland, and Norway), and to a lesser degree in New Zealand, Australia, and Japan. On the other hand, work in this field has begun in the world’s "third pole" area of the Qinghai Tibet Plateau and Himalaya [2–3, 11–20].

However, investigations in these domains have not been conducted in the region of Mount Emei, which is a typical alpine landform. Mount Emei (elevation of 3,099 m) is a complex terrain containing a wide variety of plant species (more than 3,000). The vertical band spectrum of Mount Emei has formed a range of complex plant microenvironments, which has led to the availability of considerable differences in their soil properties. In this study, the soil nutrient characteristics and microbial biomass content of the soil at different altitudes in the Mount Emei area were investigated. The aim was to provide a theoretical basis for research on soil nutrient cycling and vegetation distribution pattern in the global alpine ecosystem, and to enrich the knowledge of the ecological processes in the mountains to fill in the existing scientific gap in this aspect, especially for the region of Mount Emei.

2. Study area and study method

2.1. Study area
Mount Emei is located on the southwest edge of the Sichuan Province in China. The average annual rainfall in Mount Emei is approximately 1,480.5 mm and the annual relative humidity about 80%.

Four plots (20 m × 20 m) were selected in the four different vegetation belts (890 m; 1,800 m; 2,400 m; and 3,020 m) of Mount Emei in December 2017. Then, three sampling points were randomly set at more than 5 m apart in each of the soil plots. Next, soil samples were collected by a soil sampler, and their contents of soil organic matter (OM), ammonium nitrogen (NH₄⁺-N), nitrate nitrogen (NO₃⁻-N), total nitrogen (TN), total phosphorus (TP), total potassium (TK), microbial biomass carbon (MBC), and microbial biomass nitrogen (MBN) were determined.

2.2. Determination method
Soil temperature and soil water content at a soil depth of 5 cm were measured by a portable soil temperature and humidity detector. NH₄⁺-N and NO₃⁻-N contents were analyzed by indophenol blue colorimetry and ultraviolet spectrophotometry, respectively. In addition, MBC and MBN contents were determined by the chloroform-fumigation direct-extraction (CFE) technique. The OM, TN, TP, and TK contents were analyzed by the acid-dichromate oxidation method, the semi-micro Kjeldahl method, ammonium molybdate spectrophotometry and flame photometry, respectively [21-22].

2.3. Data processing and analysis
Microsoft Excel 2010 was used for data processing, and one-way ANOVA was conducted by SPSS 13.0. Values of $P < 0.05$ were considered statistically significant.

3. Results And Analysis

3.1. Soil temperature and soil water content
In general, soil temperature decreased gradually with elevation. The soil temperature of the lowest elevation (890 m) was the highest, reaching 6.83 °C (at a depth of 5 cm below the ground level), whereas the soil temperature at the highest elevation (3,020 m) was the lowest (-1 °C). Except at the highest altitude, soil freezing was not observed at any other elevation. The soil water content showed an irregular trend with elevation increase. The soil water content at an intermediate altitude (2,400 m and 1,800 m) was high, reaching 29.35% and 20.3%, respectively. The lowest water content of the soil was 11.5%, which was detected in the highest altitude (Figure 1).
3.2. Soil nutrient pools and microbial biomass contents

As can be seen from Fig. 2, soil OM, TN, NH$_4^+$-N, NO$_3^-$-N, MBC, and MBN contents have the same change trend; their highest values were measured at the altitude of 2,400 m. However, except for the NH$_4^+$-N content, the differences in soil OM, TN, NO$_3^-$-N, MBC, and MBN contents did not reach statistically significant levels at the various altitudes studied (Table 2). The results of the ANOVA analysis revealed that the impact of elevation on soil NH$_4^+$-N content had reached highly significant levels ($P < 0.01$) (Table 2). The amount of soil NH$_4^+$-N was the highest (10.87 g/kg) at the altitude of 2,400 m but the lowest (5.38 g/kg) at the altitude of 3,020 m.

Soil TP content increased slowly with the elevation and reached a maximum value of 2.10 g/kg at the altitude of 3,020 m. No significant difference in TP content at the different altitudes was found by the statistical analysis. Soil TK exhibited a trend of irregular change and was the highest in the plot at the elevation of 1,800 m, reaching 37.42 g/kg, which was significantly higher than those of the other elevations (Table 1).
Fig. 2 Soil nutrient and microbial biomass contents at different elevation levels.

Table 1. Summary of statistically significant effects of different elevation on soil nutrient and microbial biomass.

| category          | variable | P     | Response                      |
|-------------------|----------|-------|-------------------------------|
| Microbial pools   | MBC      | 0.189 | ns                            |
|                   | MNB      | 0.319 | ns                            |
|                   | OM       | 0.265 | ns                            |
|                   | TN       | 0.330 | ns                            |
|                   | TP       | 0.323 | ns                            |
| Soil nutrient pools | TK   | 0.000 | 1800 m > 890 m > 2400 m, 3020 m |
|                   | NO$_3$-N | 0.052 | ns                            |
|                   | NH$_4$+-N | 0.004 | 2400 m > 1800 m, 890 m > 3020 m |

Note: “ns” is not significant.

3.3. Correlation analysis of the differences in soil microbial biomass, inorganic nitrogen, and soil basal values

As can be seen from Table 2, soil water content was positively highly correlated with MBN and NO$_3$-N content, and correlated with NH$_4$+-N content. A significant positive correlation was also observed between TN and NH$_4$+-N contents. No significant correlation was observed between the other soil parameters.

Table 2. Correlation analysis of the differences in the values of the contents of soil microbial biomass and inorganic nitrogen, and the soil basal properties at different elevation levels.

|          | MBC    | MBN    | NO$_3$-N | NH$_4$+-N |
|----------|--------|--------|----------|-----------|
| Soil water content | 0.7719 | 0.9632** | 0.9296** | 0.8146*   |
| Soil temperature  | 0.0736 | -0.0136 | 0.0001   | -0.0243   |
| OM content       | 0.785  | 0.2997  | 0.473    | 0.3666    |
| TN content       | 0.4439 | 0.395   | 0.5646   | 0.844*    |
| TP content       | -0.0257 | -0.354  | -0.2237  | -0.3141   |
| TK content       | -0.2314 | -0.0004 | -0.0604  | -0.1069   |

Note: ** extremely significant level ($p<0.01$), * significant level ($p<0.05$).
4. Discussion

The elevation gradient and its closely related soil water temperature condition significantly affect the transformation of soil mineral elements and soil microbial dynamics, influencing the distribution and growth of plants [23].

We found that the soil temperature in winter in Mount Emei decreased with altitude, which is consistent with the results of previous studies. Nevertheless, the soil moisture content showed an inconsistent trend. In this study, the soil water content increased with elevation at 2,400 m below sea level, and suddenly decreased at the altitude of 3,020 m. Our results are not consistent with those obtained in this area in the summer. For example, in a previous examination, the soil water content was found to rise with the elevation of altitude in summer [24]. Because the soil was frozen and covered by 2–20-cm layers of ice at the altitude of 3,020 m in winter, the amount of the available soil water could not be detected by the detector.

In this study, except for TP and TK, the soil nutrients and microbial biomass contents were the highest at the altitude of 2,400 m, characterized by unfrozen soil, lower temperature, and higher humidity. The contents of soil TN, NH$_4^+$-N, and NO$_3^-$-N, were the highest at the at 2,400-m altitude in winter. Perhaps because the amount and variety of litter are more in the mixed forest of deciduous broad-leaved conifers, which makes the highest content of the organic matter and the highest water content. In addition, the soil is not frozen at the altitude, which is the most beneficial to the growth and reproduction of microorganisms, and can contribute to rapid soil mineralization, thus becoming one of the main sources of soil nitrogen. On the other hand, it may be that the consumption of organic matter and nitrogen source is less because of the lower temperature (close to 0 °C) at 2,400 m altitude, which is also the main cause of the highest soil nitrogen contents in winter.

Zhang et al. discovered differences in the amounts and activities of soil microbes at various elevations in Mount Gongga, which increased with the decrease of altitude in the upper half of the mountain. On the contrary, in the lower half of the mountain, the high altitude and low temperature were not conducive to the growth and reproduction of soil microbes. In Mount Emei, we established the highest MBC and MBN contents at the altitude of 2,400 m in winter. This might have been caused by the abundant water content, high soil fertility, plentiful organic matter, and the sufficient amounts of nutrients at the 2,400-m altitude, which is conducive to the multiplication of microorganisms. Previous studies support our conclusion that soil organic matter content is significantly correlated with soil microbial biomass [25-27].

5. Conclusion

The soil at Mount Emei has a high level of OM and mineral nutrition pools in winter, especially at the elevation of 2,400 m. Our findings indicate that high-altitude soils provide a larger potential nutrient pool that is sufficient to maintain plant growth in the non-growing season and the coming spring season.

Acknowledgements

This work was supported by the project of the Science and Technology Department in Sichuan province (2018NFP0107), the scientific research fund of Sichuan Provincial Education Department (16ZA0306, 18ZA0245), the Natural Science Foundation of China (31500346), the scientific research fund of bamboo diseases and pests control and resources development key laboratory of Sichuan Province (17ZZ004, 17ZZ005), the scientific research project of Leshan Normal University (XJR17006).

References

[1] Huang X, Wen W Q, Zhang J, et al. Soil faunal diversity under typical alpine vegetations in West Sichuan[J]. Chinese Journal of Applied Ecology, 2010, 21 (1):181-190.

[2] Hu X, Yin P, Zong H, et al. Response of soil microbial dynamics to changes in snow regime in alpine area [J]. Journal of Ecology and Rural Environment, 2014, 30 (4):470-474.
Hu X, Yin P, Wu Y, et al. Effect of snow depth and snow duration on soil N dynamics and microbial activity in the alpine areas of the eastern Tibetan Plateau [J]. Russian Journal of Ecology, 2014, 45 (4): 263-268.

Sommerfeld R.A., Mosier A.R and Musselman R.C. CO2, CH4 and N2O flux through a Wyoming snowpack and implications for global budgets [J]. Nature, 1993, 361: 140-142.

Taylor B.R. and Jones H.G. Litter decomposition under snow cover in a balsam fir forest [J]. Can. J. Bot. 1990, 68: 112-120.

Mikan C J, Schimel J P, Doyle A P. Temperature controls of microbial respiration in arctic tundra soils above and below freezing [J]. Soil Biology & Biochemistry, 2002, 34 (11): 1785-1795.

Schimel J P, Mikan C. Changing microbial substrate use in arctic tundra soils through a freeze-thaw cycle [J]. Soil Biology & Biochemistry, 2005, 37 (8): 1411-1418.

Hu X, Wu N, Yin P, et al. Effects of snow pack and litter input on soil microbial count and biomass in the Eastern Tibetan Plateau [J]. Journal of Ecological Science, 2013, 8 (3): 97-102.

Wang S J, Yuan H H, Wang J S, et al. Composition structure of soil fauna community under the typical vegetations in the Wuyi Mountains, China [J]. Acta Ecologica Sinica, 2010, 30 (19): 5174-5184.

Laossi K R, Barot B, Calvalho D, et al. Effects of plant diversity on plant biomass production and soil maerofauna in Amazonian pastures [J]. Pedobiologia, 2008, 51: 397-407.

Brooks P.D., McKnight, D. and Elder K. Carbon limitation of soil respiration under winter snowpacks: potential feedbacks between growing season and winter carbon fluxes [J]. Global Change Biol., 2005, 11: 231-238.

Buckeridge K.M. and Grogan P. Deepened snow alters soil microbial nutrient limitations in arctic birch hummock tundra [J]. Appl Soil Ecol, 2008, 39: 210-222.

Decker K.L., Wang D. and Waite C. Snow removal and ambient air temperature effects on forest soil temperatures in northern Vermont [J]. Soil Science Society of American Journal, 2003, 67: 1234-1242.

Edwards A.C., Scalenghe R. and Freppaz M. Changes in the seasonal snow cover of alpine regions and its effect on soil processes: A review [J]. Quatern Int. 2007, 162-163: 172-181.

Elliott A.C. and Henry H.A.L. Freeze-thaw cycle amplitude and freezing rate effects on extractable nitrogen in a temperate old field soil [J]. Biol Fert Soils, 2009, 45: 469-476.

Hiltbrunner E., Schwikowski M. and Körner C. Inorganic nitrogen storage in alpine snow pack in the Central Alps (Switzerland) [J]. Atmos Environ., 2005, 39: 2249-2259.

Larsen K.S., Jonasson S. and Michelsen A. Repeated freeze-thaw cycles and their effects on biological processes in two arctic ecosystem types [J]. Appl. Soil Ecol., 2002, 21: 187-195.

Michele F., Berwyn L.W. and Anthony C.E. et al. Labile nitrogen, carbon, and phosphorus pools and nitrogen mineralization and immobilization rates at low temperatures in seasonally snow-covered soils [J]. Biol Fertil Soils., 2007, 43: 519-529.

Mikan C.J., Schimel J.P. and Allen P.D. Temperature controls of microbial respiration in arctic tundra soils above and below freezing [J]. Soil Biol. Biochem. 2002, 34: 1785-1795.

Liu, L., Wu, Y. and Wu, N. Effects of freezing and freeze-thaw cycles on soil microbial biomass and nutrient dynamics under different snow gradients in an alpine meadow (Tibetan Plateau) [J]. Pol J Ecol., 2010, 58: 717-728.

R.K. Lu, Soil and Agricultural Chemistry Analysis. Beijing, China Agricultural Science and technology press, 1999.

J. Li, M. G. Li, J. Yang, C. F. Wang, Y. Ai, and R. L. Xu, The community structure of soil Sarcodina in Baiyun Mountain, Guanzhau, China. European Journal of Soil Biology, 2010, 46(1): 1-5.

WANG B, CHEN Y M, ZHOU Z Y. Study of Soil Nitrogen Mineralization at Different Altitudes on Western Slopes of Helan Mountains, China. Journal of Desert Research,
2007, 27 (3): 483-490.

[24] HU X, YI N, CAI S, ZENG S, et al. The soil nitrogen cycle of different elevation in Mt. Emei. Journal of Jinan University (Natural Science & Medicine Edition), 2015, 36 (5):378-382.

[25] Sui Y Y, Jiao X G, Gao C S, et al. The relationship among organic matter content and soil microbial biomass and soil enzyme activities [J]. Chinese Journal of Soil Science, 2009, 40 (5): 1036-1039.

[26] Sayer E J, Powers J S, Tanner E V J. Increased litter fall in tropical forests boosts the transfer of soil CO2 to the atmosphere[J]. Plosone, 2007, 2 (12): 1-6.

[27] Brant J B, Sulzman E W, Myrold D D. Microbial community utilization of added carbon substrates in response to long term carbon input manipulation [J]. Soil Biology & Biochemistry, 2006, 38: 2219-2232.