Introduction

Litter decomposition is a fundamental global biogeochemical process. On the one hand, it is an important component of the global carbon (C) cycle by which carbon fixed during photosynthesis is returned to the atmosphere. On the other hand, perhaps more importantly, it plays a vital role in the recycling of nutrients to soil and plant communities. During the process of litter decomposition, dead plant materials are broken down into plant and microbe available nutrients, inorganic forms of C, and stable organic matter [1]. Hence, changes in the rates of decomposition can have profound effects on ecosystem attributes, such as productivity, plant species composition, and food-chain dynamics [2, 3]. Climate and litter chemistry are thought to be the primary drivers of litter decomposition and nutrient release [4]. Across multiple types of ecosystems, temperature, indices of water availability, and measures of litter quality such as nitrogen (N) availability, lignin content, or the lignin:N ratio, have important implications for the rates of mass and nutrient loss [5-7].

Climate has a direct effect on litter decomposition due to the effects of temperature and moisture. Generally, the litter decomposition rate increases with increasing temperature [8]. Temperature is generally considered the primary limiting factor, especially in high latitude and altitude ecosystems, and there is evidence that even a small increase in temperature could enhance decomposition activity and
the release of CO₂ from dead organic matter in these soils [9, 10]. Water availability affects the rates of mass loss and nutrient release primarily affecting the activity of the decomposer community [11]. In addition, precipitation can control the physical process of leaching, with greater rainfall accelerating the breakdown of surface litter and mass loss in the initial stage of the decay process [12]. In warm, moist conditions soluble litter substrates may be decomposed more rapidly than in cool, drought conditions, resulting in the higher accumulation of recalcitrant substances in warm rather than cool climates [13]. Therefore, climate factors such as mean temperature and precipitation or combined indices that incorporate both temperature and precipitation, such as actual evapotranspiration (AET), potential evapotranspiration (PET), and the climate decomposition index (CDI), are generally the best predictors of decomposition on regional and global scales [14, 15].

Although climate conditions usually have a direct influence on plant litter decomposition, they also have an indirect effect through the climatic impact on litter chemistry [10]. Recently, it has been shown that the magnitude of species-driven variation in litter decomposition rates is much larger than previously thought and even greater than the climate-driven variation [2]. Litter quality, especially the chemical characteristics of the organic constituents, is the prevailing endogenous control of the litter decomposition rate at the ecosystem level [16]. In general, decomposition rates increase with a decrease in the ratio of C to N (C:N ratio), which is therefore an important indicator of litter quality [17]. Other typical constituents limiting the rate of litter degradation are the initial tissue N, and P or lignin concentrations (including the lignin:N or N:P ratios) [18, 19]. For instance, litter with a high N concentration decays faster than litter with a low N concentration under equal lignin content [20]. Lignin is a complex aromatic heteropolymer in cell walls, and is one of the litter components that are most recalcitrant to decomposition [21, 22]. However, specific chemical characteristics are only proxies for the overall species-driven controls on litter decomposition rates. Therefore, recent studies have shown that species identity has a higher explanatory power for litter decomposition rates than litter chemistry parameters [1, 23]. Conifer species generally have slower decomposition rates due to lower quality litter (e.g., higher C:N ratios, higher tannins), while deciduous species have faster decomposition due to higher quality litter (e.g., lower C:N ratios, lower tannins) [24, 25].

Environmental gradient studies have been recognized as powerful tools for exploring and quantifying the influence of environmental conditions on ecosystem processes [3, 26]. In particular, elevation studies have the potential to provide information on the sensitivity of ecosystem processes to temperature, although the covariance of temperature with other elevation-dependent variables necessitates caution in interpretation [3]. Furthermore, ecological field experiments along environmental gradients can enable discrimination between direct environmental factors and other site-dependent factors, such as species’ traits and composition [27, 28]. Sergyemla Mountain is located in the southeast margin of the Tibetan Plateau, with strong climatic variation along elevation gradients, and clear vertical zonation of vegetation types. In the present study, three sites representing three vegetation types along the altitudinal gradient: mixed conifer and broadleaf forest (MCFB, 3,169 m a.s.l.), sclerophyllous evergreen broadleaf forest (SEBF, 3,453 m a.s.l.), and subalpine dark coniferous forest (SDCF, 3,957 m a.s.l.) were selected to compare the leaf litter decomposition rates and chemical fractions loss. Because soil temperature generally decreases with elevation, decomposition is expected to occur much slower and over a shorter season at higher altitudes than lower altitudes. Thus, we hypothesize that the rates of decomposition in the dark coniferous forest are the lowest, those in the sclerophyllous evergreen broadleaf forest are intermediate, and those in mixed conifer and broadleaf forests are the highest.

Materials and Methods

Study Site

Sergyemla Mountain (29°10′-30°15′N and 93°12′-95°35′E), which belongs to the joint zone between Nyainqentanglha Mountain and the Himalayas, is located in the Gongbu Nature Reserve of Nyingchi County, southeast Tibet. The mountain stretches from the northwest to the southeast, forming a large east-west profile, with the south-west-facing western section winding its way to the Nyenchu river valley approximately 2,900-3,000 m a.s.l. and the northeast-facing eastern section cutting along Lulang River drainage downward to a valley zone as low as 2,100 m a.s.l. Situated in the transition zone between humid and semi-humid climate regions of southeast Tibet, Sergyemla Mountain has warmer winters and cooler summers as a result of the India Ocean monsoon. The mean annual air temperature which obtained from 30-year records at a meteorological weather station located at 2,900 m is 8.5°C, with mean minimal temperature in January of -0.2°C and mean maximal temperature in July of 15.5°C. The average annual precipitation is approximately 654.1 mm, with 75% of the annual average precipitation occurring from June to September [29]. Annual sunshine duration lasts 1,150 hours and the highest monthly duration is 152 hours in December. The annual relative humidity is 79% [30].

As a vital part of the Nyenchu Forest area, Sergyemla Mountain is a region with high biodiversity. In this study, three typical forests were selected at altitudes of 3,169, 3,453, and 3,957 m along the east slope of Sergyemla Mountain to determine the leaf litter decomposition rates and chemical fraction losses. At the 3,169 m site, the forest is mixed conifer and broadleaf (MCFB), which is dominated by *Pinus armandi* Franch., *Quercus aquifolioides* Rehd. et Wils. and *Populus szechuanica* Schneid. var. tibetica Schneid. At the 3,453 m site, the forest is sclerophyllous evergreen broadleaf forest (SEBF), which is dominated by *Quercus aquifolioides* Rehd. et Wils. And at the 3,957 m site the forest is subalpine dark coniferous forest (SDCF), which is dominated by *Abies georgei* var. *smithii* (Table 1).
Table 1. Summary of site characteristics, with soil property data for the organic layer.

<table>
<thead>
<tr>
<th>Vegetation type</th>
<th>Lat (N)</th>
<th>Long (E)</th>
<th>Elevation (m a.s.l.)</th>
<th>Soil organic C (g·kg⁻¹)</th>
<th>Soil total N (g·kg⁻¹)</th>
<th>Soil C:N</th>
</tr>
</thead>
<tbody>
<tr>
<td>MCBF</td>
<td>29°48′55″</td>
<td>94°44′20″</td>
<td>3,169</td>
<td>74.38</td>
<td>1.38</td>
<td>53.90</td>
</tr>
<tr>
<td>SEBF</td>
<td>29°43′12″</td>
<td>94°43′51″</td>
<td>3,453</td>
<td>53.61</td>
<td>0.97</td>
<td>55.27</td>
</tr>
<tr>
<td>SDCF</td>
<td>29°38′57″</td>
<td>94°42′52″</td>
<td>3,957</td>
<td>54.16</td>
<td>1.34</td>
<td>40.42</td>
</tr>
</tbody>
</table>

MCBF – mixed conifer and broadleaf forest, SEBF – sclerophyllous evergreen broadleaf forest, SDCF – subalpine dark coniferous forest

**Experimental Design**

To analyze leaf litter decomposition rates and nutrient loss, three experimental sites were established at three elevation zones. The leaf litter material used in the decomposition experiment was collected from the same site where it had been formed. Fresh leaves were collected from each study site in September 2008, and were oven-dried at 40°C to a constant weight in the laboratory. A sample of 10 g dried litter were placed in nylon mesh bags (20×20 cm) of 1 mm mesh with the edges sealed. After the removal of newly fallen litter on the forest floor, 200 replicate litterbags were equitably placed in five plots at each altitudinal gradient site in October 2008. In total, 600 sample bags (40 replicates × 5 plots × 3 sites) were placed for this study. The plots (10 ×10 m) were fenced to exclude wild animals and all plots were located within a radius of approximately 2 km. In addition, the initial carbon and nutrients in litter samples of each study site were determined in September 2008. At each site, air and soil temperatures at 0 cm, 5 cm, and 10 cm depths were automatically monitored by a soil thermometer (DS1921G-F5#, Maxim Integrated Products, Dallas Semiconductor Inc., Sunnyvale, California) every 2 hrs.

Litter bags were collected every three months from December 2008 to December 2010. At each collection time, 10 litter bags were harvested at five selected plots. Bags were brushed to remove plants, arthropods, and sand. Collected litter bags were oven-dried at 65°C to a constant weight in the laboratory and the remaining weight of leaf-litter was determined for mass loss. The remaining leaf litter from five plots of each site was then mixed and ground to pass through a 0.5-mm screen, and used for the analyses of total carbon, total nitrogen, total phosphorus, carbohydrate, lignin, and cellulose contents.

**Litter Chemical Analysis**

Carbon and nutrients in litter samples were determined as described by Lu [31] and Zhang [32]. Total organic carbon was determined by using the dichromate oxidation-sulphateferrous titration method. Total N was determined by the micro-Kjeldahl method after digesting the subsamples in K₂Cr₂O₇-H₂SO₄. Samples of P were acid digested with an H₃PO₄ and H₂O₂ solution. The digested solution was then transferred to a 100-ml volumetric flask, rationed, and stored for measurement of the P content. Total P was determined by the phosphomolybdhenum-yellow colorimetry method. Carbohydrate was determined by the anthrone colorimetric method [33]. The lignin and cellulose concentrations were determined using acid-detergent fibre methods [34].

**Calculations and Statistical Analysis**

The constant potential mass loss over time for each site was calculated by the following exponential equation (Eq. 1) [35]:

\[ x_t = x_0 e^{-kt} \]

...where \( x_0 \) is the original mass of a litter sample, \( x_t \) is the amount of litter remaining after time \( t \), and \( k \) is the litter decomposition coefficient (month⁻¹). One-way ANOVA was used to test the differences in the percentage of litter mass and chemical fraction loss among three different vegetation types along the altitudinal gradient, and a least significant difference (LSD) test was used to distinguish significant differences at \( p = 0.05 \). Relationships between the litter mass loss and chemical fraction contents, air, and soil temperatures were tested using Pearson correlation analysis. All analyses were performed using the SPSS 11.5 statistical software package (SPSS Inc., USA).

**Results**

**Litter Decomposition Rates**

In general, the leaf litter mass loss followed the same pattern for the three different vegetation types along the elevation gradient (Fig. 1). The mass losses were rapid for the first six months of decomposition in the field, and then the mass were lost gradually from June 2009 to the end of the decomposition period. When the three different vegetation types were compared, the litter decomposition rate of the MCBF was the fastest, the litter decomposition rate of the SEBF was intermediate, and that of the SDCF was the slowest. The leaf litter decomposition coefficient (\( k \) values, month⁻¹) during the 24-month decomposition period of the MCBF site was 1.7 and 2.2 times greater than that of the SEBF and SDCF sites, respectively (Table 2).

**Chemical Fraction Contents**

During the 24-month decomposition experiment, the contents of C and other chemical fractions varied among
the different vegetation types, and the magnitude of variation was different among the chemical fractions (Fig. 2). The dynamics of C content during the experiment were different between the MCBF, SEBF, and SDCF. In the initial stages of the experiment, the litter C content of all three vegetation types slightly increased, but then the C content of the MCBF and SEBF continued to increase, while that of the SDCF decreased. The P and cellulose content generally showed an increasing trend during the 24-month decomposition period, but the carbohydrate content generally showed a decreasing trend. N, lignin content, and the lignin:N ratio increased during the initial stages and then decreased. In contrast, the C:N, C:P, and N:P ratios decreased during the initial stages and then slightly increased.

Litter Mass and Chemical Fraction Loss

Over the 24-month decomposition period, 62.3% of the litter mass at the MCBF site, 48.2% of the litter mass at the SEBF site, and 35.6% of the litter mass at the SDCF site were lost, respectively (Fig. 3). Statistical analyses revealed that the litter loss was significantly different among the three different vegetation types. The lost chemical fraction accounted for 60.4% of the C, 49.7% of the N, 32.2% of the P, 74.9% of the lignin, 63.6% of the cellulose, and 89.4% of the carbohydrate of the MCBF in 24-month net loss rates. Comparatively, the lost chemical fraction accounted for 48.0% of the C, 39.0% of the N, 37.9% of the P, 41.6% of the lignin, 49.8% of the cellulose, and 95.1% of the carbohydrate of the SEBF, and the lost chemical fraction accounted for 29.9% of the C, 21.2% of the N, 3.7% of the P, 52.4% of the lignin, 2.4% of the cellulose, and 96.8% of the carbohydrate of the SDCF. The results from one-way ANOVA demonstrated that the chemical fraction loss was also significantly different among the three different vegetation types along the elevation gradient.

Discussion

Litter Decomposition along the Altitudinal Gradient

The results of the present study supported our hypothesis that the rates of decomposition are highest in the MCBF (3,169 m a.s.l.), intermediate in the SEBF (3,453 m a.s.l.), and lowest in the SDCF (3,957 m a.s.l.). The average leaf litter decomposition coefficient during the 24-month decomposition period in the MCBF, SEBF, and SDCF sites were 0.04, 0.03, and 0.02 month⁻¹, respectively. After the 24-month decomposition period, the lost litter mass accounted for 62.3% of the MCBF, 48.2% of the SEBF, and 35.6% of the SDCF, respectively. This result is in agreement with previous reports that found that litter decomposition decreases as the elevation increases [36, 37]. For instance, the leaf litter decomposition rates were subtropical evergreen broadleaf forest (500 m a.s.l.) > temperate coniferous forest (1,150 m a.s.l.) > cold temperate dwarf forest (1,750 m a.s.l.) > and frigid zone alpine meadow (2,150 m a.s.l.) in the Wuyi Mountains in southeastern China [38]. Similarly, the litter decomposition rates of the wet forest (350 to 400 m a.s.l.) and the cloud forest (1,051 m a.s.l.) of the Luquillo Mountains in northeastern Puerto Rico were 1.12 and 0.70 year⁻¹, respectively [37]. These data are also similar to the latitudinal decomposition rule of tropics > subtropics > temperate zone > cold temperate zone > frigid zone [39].

Climate exerts an important influence on decomposition in hierarchical models of decomposition and climatic effects on decomposition at large scales are apparent from the relationship between actual evaportranspiration and decomposition rates [10, 38]. In alpine and subalpine areas, as altitude increases the climate shifts toward more stressful conditions for plant growth and organic matter decomposition: lower mean temperatures, higher precipitation, longer snow cover and, thus, a shorter growing season, lower atmospheric pressure, and higher solar radiation [27]. On the whole, the air temperature declines with increasing elevation on
Fig. 2. Variations in the C, N, P, lignin, cellulose, and carbohydrate contents and the C:N, C:P, lignin:N, and N:P ratios during the decomposition process of three subalpine forests on Sergyemla Mountain.
Sergyemla Mountain. The average air temperature during the 24-month decomposition period was 5.98°C at the MCBF site, which was 20.3% and 65.4% higher than that of the SDBF site (4.97°C) and the SDEF site (3.62°C), respectively. The soil temperatures at these three sites showed a similar pattern. The average soil temperature of the MCBF site, which were 6.99, 6.87, and 6.77°C at 0, 5, and 10 cm, respectively, were the highest, those of the SEBF site, which were 5.30, 5.59, and 5.63°C at 0, 5, and 10 cm, were intermediate, and those of the SDEF site which were 3.01, 3.63, and 3.82°C at 0, 5, and 10 cm, were the lowest among the three different altitudinal gradients.

The positive relationships between the litter mass loss and temperature along the elevation gradient were found in the present study. The percentages of litter mass loss increased significantly with increasing air temperature ($r = 0.39, p = 0.04$), 0 cm soil temperature ($r = 0.44, p = 0.02$), 5 cm soil temperature ($r = 0.43, p = 0.03$), and 10 cm soil temperature ($r = 0.43, p = 0.03$) in the subalpine forest ecosystem on Sergyemla Mountain (Fig. 4). Hence, the hypothesis was that decomposition occurred much slower and over a shorter season at higher altitudes than lower altitudes due to the generally decreasing soil temperature with elevation. This hypothesis turned out to be right because litter decomposition rates were significantly correlated with soil temperature. These correlations are consistent with studies that also found that litter decomposition decreases as air temperature falls along elevation gradients [3].

The data that litter mass loss increased with increasing temperature indicate that climatic constraints are the strongest regulators of decomposition. For instance, soil temperature can explain 95% of the variation in the decomposition rate along an elevation gradient in Peruvian forests [3]. Litter decomposition is an ecological process governed by decomposer organisms, such as soil fauna and microorganisms. Hence, the increase in soil temperature is likely to stimulate litter decomposition by creating conditions favorable for decomposer populations and activity [5, 40]. Soil moisture was not measured in the present study; however, decomposition in the lower elevation was limited by moisture and soil moisture, rather than temperature, which appears to be the primary factor that controls decomposition rates reported in previous studies [41, 42]. Hence, it would be necessary to determine soil moisture for future research because some alpine regions are currently showing a spatially consistent warming, but spatial variability in changing precipitation [43].

**Litter Decomposition and Chemical Fraction**

Climate is the most important regulator of litter decomposition, but when climate conditions are similar or maximize potential decomposition, litter quality controls are enhanced [4]. Litter quality, especially the chemical characteristics of the organic constituents, is of particular importance for the mass loss and dynamics of limiting nutrients during decomposition [16]. In general, in the initial stages of decomposition, soluble components disappear quickly, and non-lignified carbohydrates are also degraded [44, 45]. Nevertheless, almost no decomposition of lignified carbohydrates and lignin occurs, and results in the increase of concentrations of recalcitrant materials and some nutrients [16, 46]. In the later stages of decomposition, the contents of lignin, N, and P increase linearly with accumulated mass loss, but the absolute amounts of N and P start to decrease at some point after the onset of lignin disappearance [7, 40].

This study demonstrated that the concentrations of the chemical fractions of the litter varied among different vegetation types and among different stages of decomposition during the course of leaf litter decomposition in subalpine
forests in Sergyemla Mountain (Fig. 2). The C content of the litter slightly increased at the initial stage and then fluctuated at the later stage. Because C is the element that predominantly contributes to the weight loss of the leaf litter [16], the loss of C over the course of the experiment was similar to the loss of litter mass (Fig. 3). N and P have long been recognized as the most limiting nutrients that regulate plant growth and net primary productivity in terrestrial ecosystems, especially in alpine and arctic ecosystems, where the mineralization is slow due to low temperatures [47, 48]. The immobilization of N and P often occurs during litter decomposition, and the amount of these nutrients in fresh litters is frequently insufficient for decomposer organisms in temperate and tropical regions [21, 49]. The patterns of the changes of the N and P content found here were generally consistent with previous works [50-52], as the N and P contents in the later stages of decomposition were higher than the initial content values. Lignin has traditionally been combined with N to predict long-term decomposition patterns across biomes, because N concentrations have been found to be more important during early decomposition, while lignin is more important during later stages [5]. Lignin can also be associated with other polymers such as cellulose and hemicellulose, which constitute the complex cell wall that decomposes slowly [53]. In the present study, the lignin content increased during the initial decomposition stage and then decreased, whereas the cellulose content generally exhibited a slightly increasing trend in the three subalpine forests. Carbohydrate is a relatively labile decomposition component of litter [8, 46], thus the carbohydrate content declined rapidly in the early stage of litter decomposition.

The concentrations of N, P, lignin, and cellulose, as well as the ratios of C to N, C to P, N to P, and lignin to N, are common indicators of litter quality. Of the above indicators, the ratio of C:N and lignin:N can well reflect the decomposition rate in various ecosystems [54, 55]. N is well known to affect the decomposition rate of individual litter types [7, 13], and P may be also important factor, especially in P limiting ecosystems [46, 56], in early phase of decomposition, the decomposition rate largely depends on initial litter N concentrations, and in later stages the concentration of lignin or the ratio of lignin to N becomes the determining factor [57, 58]. The litter mass loss were significant positive correlation with N and P contents during a 24-month decomposition period at the MCBF site, but significantly negatively correlated with C:P, N:P and lignin:N (Table 3). This finding is grossly consistent with other studies of litter decomposition [17, 59, 60]. Meanwhile, the litter mass loss at the SEBF site was not significantly correlative with
chemical fraction contents of the litter, while the litter mass loss at the SDCF site only significantly positively correlated with the cellulose and P content (Table 3). The absence of a statistically significant relationship between litter mass loss and litter quality at high altitudes on Sergyemla Mountain may be because that it is climate, not litter quality, that control the litter decomposition process because of the low temperatures limiting in high elevations.

Table 3. The relationship coefficient ($r$ values) between litter mass loss and chemical fraction content during the 24-month litter decomposition.

<table>
<thead>
<tr>
<th>Index</th>
<th>Vegetable type</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>MCBF (3,169 m)</td>
</tr>
<tr>
<td>C</td>
<td>0.520</td>
</tr>
<tr>
<td>N</td>
<td>0.754**</td>
</tr>
<tr>
<td>P</td>
<td>0.955**</td>
</tr>
<tr>
<td>Lignin</td>
<td>-0.436</td>
</tr>
<tr>
<td>Cellulose</td>
<td>0.075</td>
</tr>
<tr>
<td>C:N</td>
<td>-0.572</td>
</tr>
<tr>
<td>C:P</td>
<td>-0.878**</td>
</tr>
<tr>
<td>N:P</td>
<td>-0.754*</td>
</tr>
<tr>
<td>Lignin:N</td>
<td>-0.814**</td>
</tr>
</tbody>
</table>

*Correlation is significant at the 0.05 level.
**Correlation is significant at the 0.01 level.

MCBF – mixed conifer and broadleaf forest,
SEBF – sclerophyllous evergreen broadleaf forest,
SDCF – subalpine dark coniferous forest

Conclusions

These observations of leaf litter decomposition in three subalpine forests along an elevation gradient on the east slope of Sergyemla Mountain demonstrated that the decomposition rate decreased as elevation increased. The concentrations of the chemical fractions of the litter varied among different vegetation types and among different stages of decomposition during the course of leaf-litter decomposition. The correlation analysis determined that there was a significant positive relationship between the litter mass loss and the average air and soil temperature along the elevation gradient. To a certain extent, the litter mass loss at the MCBF site (3,169 m a.s.l.) significantly correlated with litter quality, including the N and P contents and the C:P ratio, and so on. However, the litter mass losses at the SEBF (3,453 m a.s.l.) and the SDCF (3,957 m a.s.l.) sites were mainly not significantly correlated with litter quality. These results indicate that climate plays a critical role in leaf litter decomposition across environmental gradients, but litter quality also can partly explain the higher leaf litter decomposition in low elevation forests on the east slope of Sergyemla Mountain.

Acknowledgements

This study is funded by the National Natural Science Foundation of China (41001018) and the action-plan of the Chinese Academy of Sciences (CAS) for West Development (KZCX2-XB3-08). The authors are grateful to Professor Jie Lu (University of Tibet) for assistance in the field.

References

33. GAO J.F. Experimental technology of plant physiology. Xi’an, China: World Map and Book Press, 2000 [In Chinese].