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ANTHROPOGENIC DISTURBANCES AFFECT POPULATION SIZE AND BIOMASS ALLOCATION OF TWO ALPINE SPECIES FROM THE HEADWATER AREA OF THE URUMQI RIVER, CHINA

RUIMING ZHAO, HUA ZHANG AND LIZHE AN*

MOE Key Laboratory of Cell Activities and Stress Adaptations, School of Life Sciences, Lanzhou University, Lanzhou 730000, China
**Corresponding author's email: zhaorm1001@163.com; Tel: +8613919831830; Fax: +86 0931 4967199*

Abstract

The survival of alpine plants are seriously threatened by increasing anthropogenic activity. *Saussurea involucrata* and *Rhodiola quadrifida* are particularly affected because of their high medicinal value. To assess the impact of anthropogenic disturbance on the two species, their population size and biomass allocation were examined at three levels of disturbance at low and high altitudes. Anthropogenic disturbance was the most serious threat to the populations and changed the population density, biomass, and biomass allocation of both species significantly ($p<0.05$). The changes differed with the species and the altitude, and were also affected by the interaction between these two factors. Population density and biomass of the two species decreased with an increase in the level of anthropogenic disturbance. These results imply that the decrease in population size and in biomass allocation to reproductive organs due to anthropogenic disturbances may make the plant populations even smaller and scarce. Meanwhile, change of making their survival dependent on the extent of anthropogenic disturbance: unless such disturbance is checked and the species are protected, they will probably disappear from the headwater area of the Urumqi River. This influence of anthropogenic disturbances may be potential threats to population ability of survival and reproduction.

Key words: Conservation; Biomass allocation; Population density; *Rhodiola quadrifida*; *Saussurea involucrate*.

Introduction

The subnival alpine habitat is one of the most unsuitable habitats for plant growth and development. The high altitudes, low temperatures, a short growing season, unstable substrate, intense radiation, and high winds all make the terrain extremely inhospitable (Körner, 2003; Cavieres *et al*., 2005). Vegetation in the alpine belt mainly comprises cryophytic and xerophytic perennials growing in clusters as pioneer communities (Xu *et al*., 2014). Although unitary in their species composition, such communities of alpine plants play an important role in these ecosystems (Kikvidze *et al*., 2015). In some alpine ecosystems, cushion plants are considered to be 'nurse plants' or 'ecosystem engineers' because they are often long-lived and dominant pioneer species, improve the microclimate, and promote the growth of other plants, arthropods, and microorganisms (Butterfield *et al*., 2013; He *et al*., 2014). Additionally, other alpine plants such as *Saussurea involucrata*, *Saussurea gossypiphora*, *Rheum nobile*, and *Rhodiola quadrifida* improve the local micro-environment and contribute to the biodiversity of alpine ecosystems (Chen *et al*., 2013a; Sistla *et al*., 2013; Liczner & Lortie, 2014).

Over the past few decades, many parts of the world have witnessed large-scale anthropogenic damage to the environment that controls the structure and function of ecosystems and ecosystem services (Rockström *et al*., 2009). This damage is particularly obvious in the alpine ecosystem above the treeline in china because the system is highly sensitive to anthropogenic disturbance and climate change (Chen *et al*., 2013b). For example, over-collection of plant (*Cordyceps sinensis*, *Rhodiola rosea*, *Saussurea involucrata*, etc.) and over-grazing have caused vegetation and soil degradation, which resulted in serious social and ecological problems (Zhu & Lou, 2010; Anonymous, 2015; Zhang *et al*., 2016; Xu *et al*., 2015). Intensive and unrestrained exploitation of wild Chinese herbs had damaged natural resources. There are 2000 wild Chinese herbs which are at risk of extinction (China Statistics Bureau, 2010). Therefore, severe soil erosion and ecological deterioration had seriously threatened the habitats of many wild Chinese herbs, especially in fragile ecological environments, such as high altitude areas (Meng *et al.*, 2012). Alpine vegetation is threated by the loss of biodiversity and fragmentation of habitats caused by anthropogenic disturbance (Hagen *et al*., 2014; Wang *et al*., 2016). With the widening of the distribution range of other species along the altitudinal gradient because of the increasingly warmer climates accompanied by major changes in the structure of the alpine plant community (Roux & McGeoch, 2008; Walther, 2010). The habitat available to alpine plants is expected to decrease gradually (Nagy & Grabherr, 2009). Many studies have demonstrated the upward movement of alpine flora, probably as a result of gradually increasing temperatures (Parolo & Rossi, 2008; Felde *et al*., 2012; Spasojevic *et al*., 2013). The most likely result of these changes is the vastly increased risk of extinction of alpine plants and a substantial reduction in community diversity (Kreyling, 2010; Sorte & Jetz, 2010).

Both anthropogenic disturbance (mainly grazing and unsustainable collection of economically useful plants) and climate change (in the form of warming) can change the distribution of plant species as they struggle to remain within the range of their climatic tolerances (Colwell *et al*., 2008; Chen *et al*., 2009; Feeley & Silman, 2010). Although plants can survive despite anthropogenic disturbance and climate change, once the distribution and the composition of plant communities begin to change, the migrations may cause larger changes in the extent of suitable habitats, which, in turn, affect the size of plant populations and increase the risk of extinction (Feeley & Silman, 2010). To minimize such risks, it is essential to evaluate the response of alpine plants (especially of constructive species and endemic species) to anthropogenic disturbance and climate change. In fact, most plant species in the alpine subnival belt are characterized by a narrow distribution, small populations, and a marked endemism (Xu *et al*., 2014)—characteristics which make them particularly sensitive to anthropogenic disturbance and climate change, thereby increasing the risk of their extinction. Several studies have shown the influence of anthropogenic disturbance and climate change on plants in alpine zones (Chapin & Körner, 1995; Körner, 2003; Puhe & Ulrich, 2012; Belgacem & Louhaichi, 2013; Chen *et al*., 2013b; Broennimann *et al*., 2014; Hughes *et al*., 2014). However, these studies focused mainly on alpine plants and ecosystems of the mountainous regions in the Arctic and the Antarctic, in Europe, North America, and Africa, and in the Qinghai-Tibetan plateau. Alpine plants in the headwater area of the Urumqi River and other high-altitude regions of the Tianshan mountains have been largely ignored.

The headwater area of the Urumqi River is part of the middle sections of the Tianshan mountains. Earlier investigations focused mainly on glacial geomorphology in studying the impact of anthropogenic disturbance and climate change (Li *et al*., 2011; Sun *et al*., 2013; Wang *et al*., 2014), but little attention was given to alpine plants (from the alpine subnival belt) in this region, especially the typically alpine subnival plants that survived, such as *S. involucrata*, *Chorispora bungeana*, *Cancrinia tianschanica*, *Thylacospermum caespitosum*, *Sibbaldia tetrandra*, and *R. quadrifida* (An *et al*., 1999; Liu *et al*., 2012). Plants from the alpine subnival belt in the eastern parts of the Tianshan mountains are affected by anthropogenic disturbance in the form of heavy grazing pressure (the region houses some significant summer saeters in Xinjiang) (Zhang *et al*., 2002). At the same time, the structure and distribution of alpine subnival plants are changing as a result of overexploitation. More valuable plant species are the worst affected (Chik *et al*., 2015). In addition, the habitat of alpine subnival plants is shrinking as the snowline continues to move upwards as a result of anthropogenic disturbance and a warmer climate (Liu *et al*., 2012, 2015; Yang *et al*., 2015).

It was against this background that we decided to test the hypothesis that population density, biomass, and biomass allocation of alpine subnival plants from the headwater area of the Urumqi River have been affected by anthropogenic disturbance, which may threatens the survival of those plants and limits their distribution. To test the hypothesis, we investigated the main ways in which anthropogenic disturbance affects population density, biomass, and biomass allocation of two alpine subnival plant species, namely *S. involucrata* and *R. quadrifida*, and also the ways in which such impacts vary depending on the severity of the disturbance, the species, the altitude, and the combined effect of these three factors.

Material and Methods

Description of the site: The study site was in the headwater area of the Urumqi River in eastern parts of the Tianshan mountains. The site is part of the Xinjiang Uygur Autonomous Region of China. The average elevation varies from approximately 4100–4300 m. The Tianger peak (43°06′39.34″ N, 86°47′49.78″ E) is the highest peak, approximately 4486 m above the sea level. The current snowline is at approximately 4000–4100 m. At the study site, the annual average temperature is below 5°C during the day and –4°C at night. The temperature fluctuates from nearly 4°C to –10°C during the period favourable for the growth of plants. The average daily temperature is below 0°C and the average annual precipitation is 337.5 mm in the region above 3600 m. The period with stable snow cover is from October to April, and the average depth of snow is 10–15 cm in the vicinity of the Daxigou weather station (Liu *et al*., 2015). In summer, precipitation is mainly in the form of snowfall (which generally lasts for 1–2 days) and accounts for the majority of total annual precipitation. Vegetation is dominated by *S. involucrata*, *T. caespitosum*, *S. tetrandra*, *C. bungeana*, *C. tianschanica*, and *R. quadrifida* (An *et al*., 1999; Sheng *et al*., 2011).

Plant material: *Saussurea involucrata* (family Compositae) grows along mountain slopes, in valleys, on meadows, and in rock fissures at elevations of 2400–4100 m. The species was named ‘Tianshan snow lotus’ (the snow lotus that grows on sky-high mountains) because of the very high altitudes at which it grows and is distributed mainly in the Xinjiang province of China, although it is also found in Kazakhstan, Kyrgyzstan, and Mongolia (Chen *et al*., 1999; Flora of China Editorial Committee, 1999). *Saussurea involucrata* is an important medicinal herb in traditional medicine in China, Mongolia, and Kazakhstan, where it is used in the treatment of rheumatoid arthritis and regulation of the menstrual cycle. The species also has anti-inflammatory and analgesic properties; is an anti-oxidant; counters hypoxia, fatigue, and aging; and is used in treating hormone-related gynaecological disorders (Chik *et al*., 2015). *Rhodiola quadrifida* (family Crassulaceae) grows in alpine regions and on stony slopes and rocks in China (Xinjiang and the Qinghai-Tibet Plateau), in East Siberia, Mongolia (Hentii, Hangai, Hovsgol, Hovd, and Mongol Altai), and Russia (Altai, Sayan) (Skopińska-Różewska *et al*., 2008). *Rhodiola quadrifida* is named ‘ere-gombo’ in Mongolia and ‘tsan’ in Tibet and was used in the traditional systems of medicine in these countries for the treatment of fatigue, high blood pressure, dysentery, inflammation, and depression and also as a tonic (Flora of China Editorial Committee, 1999; Wiedenfeld *et al*., 2007). *S. involucrata* and *R. quadrifida* are important component of alpine vegetation and commonly plays a crucial role in alpine ecosystem (Haider *et al*., 2014). These species can increase the abundance and diversity of plant in the alpine zone (Sarwar & Qaiser, 2012; Chik *et al*., 2015), improve soil nutrient content (Dunyan *et al*., 1998), and supply food for mammalia (Travina *et al*., 2000). Indeed, many of these species have not yet been adequately studied, so the true ecological role of these species is not yet known.

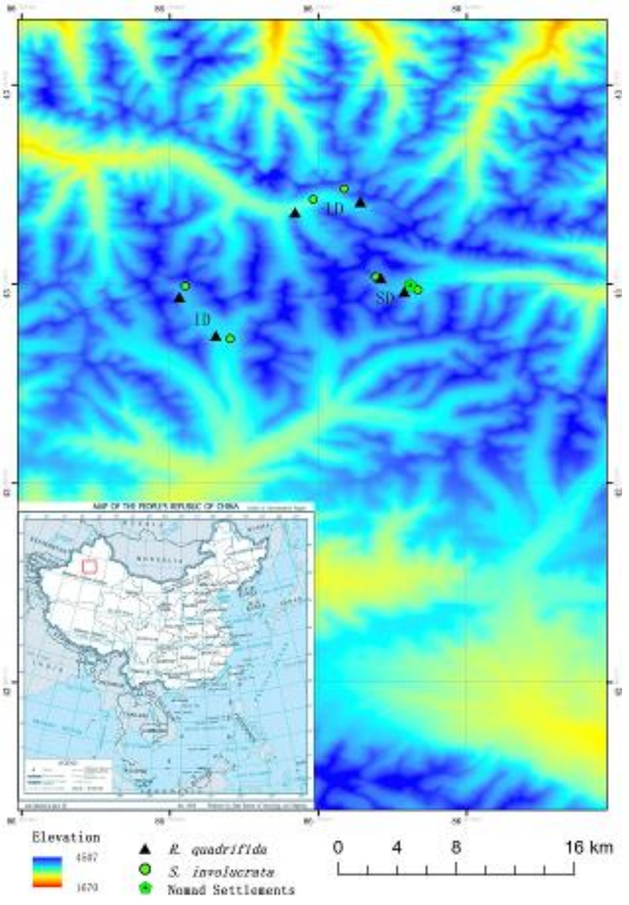

Fig. 1. Map showing the study region of the headwater area of Urumqi River and the 11 study sites at the three levels of disturbance in low and high altitudes to *S. involucrata* and *R. quadrifida*. LD, light disturbance; ID, intermediate disturbance; SD, severe disturbance.

Experimental design and data collection: The experiment was carried out in 2015. After a survey in July, populations of *S. involucrata* and *R. quadrifida* were assessed at three levels of anthropogenic disturbance (light, intermediate, and high) and two elevations, high (3800–4000 m) and low (3400–3600 m). Thus, the six populations were as follows: light disturbance (LD) at low and high altitudes, intermediate disturbance (ID) at low and high altitudes, and severe disturbance (SD) at low and high altitudes (Fig. 1). Different levels were defined by the degree of disturbance and accessibility by human (Table 1). For each population, five quadrats of 10 × 10 m were established at random. In each plot (quadrat), the numbers of reproducing and non-reproducing plants were marked and recorded. At the end of their growing period, three reproducing plants and three non-reproducing plants were collected from each quadrat to estimate the population's standing reproductive allocation and biomass allocation. Plant biomass was divided into reproductive biomass (all the reproductive structures) and non-reproductive biomass and also into above-ground biomass and below-ground biomass, dried at 70°C for 48 h, and weighed (Ploschuk *et al*., 2005; Damhoureyeh, 2017). To limit the loss of plants, only six plants were collected from every quadrat. The population biomass and biomass allocation were calculated as follows:

Reproductive biomass $(g\ m^{-2}) = N_1 \times W_1 / 100$
Above-ground biomass $(g\ m^{-2}) = (N_1 \times W_1 + N_1 \times W_2 + N_2 \times W_3) / 100$
Below-ground biomass $(g\ m^{-2}) = (N_1 \times W_3 + N_2 \times W_5) / 100$
Total biomass $(g\ m^{-2}) = (N_1 \times W_1 + N_1 \times W_2 + N_2 \times W_3 + N_1 \times W_4 + N_2 \times W_5) / 100$
Proportion of reproducing plants (%) $= N_1 / (N1 + N2) \times 100$
Proportion of non-reproducing plants (%) $= N_2 / (N1 + N2) \times 100$
Proportion, by weight, of reproductive organs (%) = Reproductive biomass / total biomass
Proportion, by weight, of stems and leaves (%) = (Above-ground biomass – reproductive biomass) / total biomass
Proportion, by weight, of roots (%) = Below-ground biomass / total biomass

N_1: number of reproducing plants in a quadrat; N_2: number of non-reproducing plants in a quadrat; W_1: average weight of plant reproductive organs in a quadrat; W_2: average weight of above-ground vegetative organs in a quadrat (reproducing plants); W_3: average weight of above-ground vegetative organs in a quadrat (non-reproducing plants); W_4: average weight of below-ground vegetative organs in a quadrat (reproducing plants); W_5: average weight of below-ground vegetative organs in a quadrat (non-reproducing plants).

Statistical analysis: All experimental results were presented as the mean ± standard error of five replicates. Statistical analyses were performed using SPSS ver. 16.0 (SPSS, Chicago, Illinois, USA) using multi-factor analyses of variance (ANOVA). A multi-factor ANOVA was used to evaluate the effects of anthropogenic disturbance, species, and altitude, both singly and in combination, on population density, biomass, and reproductive allocation. In all cases, Duncan's test was used at a significance level of $p<0.05$.

Results

Anthropogenic disturbance: All the plant populations that were part of the study were growing on alpine screes (Table 1). The degree of disturbance was mainly a function of accessibility: the severe-disturbance (SD) populations were the most accessible; the intermediate-disturbance (ID) populations less so; and the light-disturbance (LD) populations were the most inaccessible (Table 1). The main forms of anthropogenic disturbance in the SD and ID populations were collection and grazing, whereas in the LD populations, collection was the only form of disturbance (Table 1). The flowers of *S. involucrata* and the roots of *R. quadrifida* are collected, mainly by local nomads and by tourists, for the economic and medicinal value of those plant organs.

Changes in population density: The population density of reproducing plants of *S. involucrata* and *R. quadrifida* was significantly affected by the level of anthropogenic disturbance ($p<0.001$), by the species ($p<0.001$), by the altitude ($0.01<p<0.05$), and by the interaction between the level of disturbance and (a) the species ($p<0.001$) and (b) the altitude ($0.01<p<0.05$). However, the interaction between species and altitude and the three-way interaction between the level of disturbance, species, and altitude was not significant ($p>0.05$; Table 2). Similarly, the population density of non-reproducing plants and of all the plants had significant positive correlation with the level of disturbance ($p<0.001$), species ($p<0.001$), and altitude ($p<0.001$); to the interaction between the level of disturbance and (a) species ($P_{Non\text{-}reproduction\ plants\ density}$ < 0.001; 0.01 <$P_{Total\ plants\ density}$ <0.05) and (b) altitude (0.001 <*p*< 0.01); and to the interaction between species and altitude (*p*<0.001). However, the three-way interaction involving the level disturbance, species, and altitude ($P_{Non\text{-}reproduction\ plants\ density}$ <0.05) was significantly correlated only to the density of *non-reproducing* plants: the three-way interaction was not significant in the case of total plant density (*p*>0.05; Table 2). As shown in Table 2, the population density of *S. involucrata* and *R. quadrifida* was affected by the level of anthropogenic disturbance, species, altitude, and the interaction between these three factors. As the level of disturbance increased, population density of all the three types – reproducing plants, non-reproducing plants, and total plants – decreased significantly (*p*<0.05) in the case of *S. involucrata* at both altitudes (Fig. 2a, 2c, and 2e). At the low altitude, the actual values (density, or the number of plants/m^2) were as follows. Reproducing plants: 0 at SD, 0.006 at ID, and 0.064 at LD; non-reproducing plants: 0.038 at SD, 0.102 at ID, and 0.252 at LD; total plants: 0.038 at SD, 0.108 at ID, and 0.316 at LD. At the high altitude too, all the three kinds of population densities of *S. involucrata* at SD were significantly reduced (*P* < 0.05), compared to those at LD (Fig. 2a, 2c, and 2e). In *R. quadrifida*, the results were similar to those in *S. involucrata* at both altitudes (Fig. 2b, 2d, and 2f).

Table 1. The anthropogenic disturbance difference of three levels at accessibility, livestock number, collection trace, number of collection people and number of collection frequency.

Levels of anthropogenic disturbance	Accessibility	Livestock number (km^{-2})	Collection trace	Number of collection people (a^{-1})	Number of collection frequency (a^{-1})
Severe disturbance (SD)	Easy	950	Many	> 50	>12
Intermediate disturbance (ID)	Difficult	240	General	10–15	3–5
Light disturbance (LD)	Extremely difficult	0	Rare	< 2	1

Table 2. Results of multivariate generalized linear model (GLM) analysis for the effects of human disturbance, species, and altitude on population density, biomass and reproductive allocation of *Saussurea involucrate* and *Rhodiola quadrifida*.

Parameter	Disturbance	Species	Altitude	Disturbance × Species	Disturbance × Altitude	Species × Altitude	Disturbance × Species × Altitude
Density of reproductive plants	166.68***	189.97***	12.15**	18.40***	3.61*	0.90	3.14
Density of non-reproductive plants	53.99***	277.24***	24.79***	17.70***	5.94**	17.18***	5.01*
Total plant density	131.928***	55.12***	30.37***	4.01*	7.68**	14.02***	1.601
Reproductive biomass	212.82***	81.16***	0.43	37.36***	6.15**	60.70***	14.71***
Above-ground biomass	230.56***	31.31***	12.58***	12.76***	30.30***	37.98***	2.14
Below-ground biomass	107.40***	430.80***	17.38***	81.84***	12.67***	15.01***	7.46**
Total biomass	135.48***	299.17***	18.14***	55.99***	16.06***	19.71***	6.96**
Proportion of reproductive plants	32.65***	1396.00***	16.94***	4.85*	5.43**	0.78	3.28*
Proportion of non-reproductive plants	32.65***	1396.00***	16.94***	4.85*	5.43**	0.78	3.28*
Proportion of reproductive organs weight	73.55***	288.15***	63.46***	58.71***	1.29	80.75***	3.41*
Proportion of stems and leaves weight	7.98***	56310.00***	57.83***	1.08	1.39	12.73***	17.80***
Proportion of root weight	150.33***	30560.00***	56.81***	134.49***	5.82**	259.49***	30.07***

F-ratios from multi factor variance analysis are given. Degrees of freedom (d.f.) are same for all parameters.
(Disturbance, 2; species, 1; altitude, 1; disturbance × species, 2; disturbance × altitude, 2; species × altitude, 2; disturbance × species × altitude, 2).
$^{***}p<0.001$, $^{**}0.001<p<0.01$, $^{*}0.01<p<0.05$.

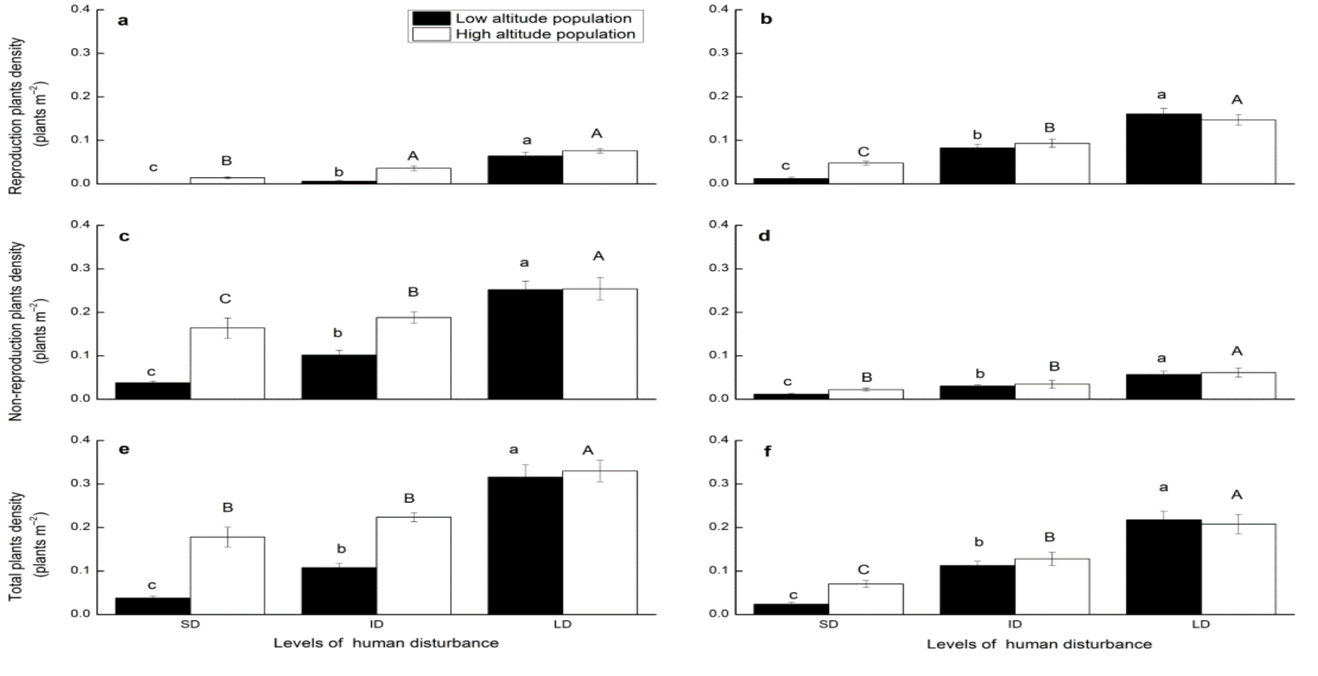

Fig. 2. Changes in population densities of reproducing plants, non-reproducing plants, and total plants in *S. involucrata* (a, c and e) and *R. quadrifida* (b, d and f) at three levels of anthropogenic disturbance. Data are mean values ± standard error (*N* = 5). Different letters between samples indicate significant differences using Duncan's test (*p*<0.05). LD, light disturbance; ID, intermediate disturbance; SD, severe disturbance.

Changes in biomass: As with the populations of the two species, their above-ground biomass, below-ground biomass, and total biomass were significantly affected by the level of anthropogenic disturbance (p<0.001), by the species (p<0.001), by the altitude (p<0.001); by the interaction between the disturbance and (a) species (p<0.001) and (b) altitude (p<0.001); and by the interaction between species and altitude (0.01 <p<0.05). However, the three-way interaction between the level of disturbance, species, and altitude showed no significant influence (p>0.05; Table 2). The biomass of reproducing population was affected only slightly. Biomass of the reproducing populations had significant positive correlation with the level of disturbance (p<0.001), to the species (p<0.001), to the interaction between the level of disturbance and (a) species (p<0.001) and (b) altitude (0.001 <p<0.01), to the interaction between species and altitude (p<0.001), and to the three-way interaction between the level of disturbance, species, and altitude (p<0.001). Altitude, by itself, however, had no significant influence on biomass (p>0.05; Table 2). At lower altitudes, values of the three kinds of biomass (above-ground, below-ground, and total) were markedly greater in the case of *S. involucrata* (p<0.05) and decreased as the level of anthropogenic disturbance increased (Fig. 3). The highest values were reached in *S. involucrata* at LD. At lower altitudes, the actual values of biomass, in grams per square metre, were as follows: reproductive biomass, 0.00 at SD and 0.191 at LD; above-ground biomass, 0.078 at SD and 1.109 at LD; below-ground biomass, 0.061 at SD and 0.612 at LD; and total biomass, 0.139 at SD and 1.721 at LD. At high altitudes, all the four values were significantly lower (p<0.05) in *S. involucrata* at SD than those at LD (Fig. 3). The same pattern was seen in *R. quadrifida* (Fig. 4).

Changes in allocation of biomass: The proportions of both reproducing plants and non-reproducing plants in the total populations were significantly affected by the degree of anthropogenic disturbance (p<0.001), the species (p<0.001), by the altitude (p<0.001); by the interaction between the degree of disturbance and species (p<0.05), (0.001 <p<0.01); and by the three-way interaction between the degree of disturbance, species, and altitude (0.01<p<0.05). However, there was little interaction between the degree of disturbance and altitude (p>0.05; Table 2). Moreover, the proportions (by weight) of reproductive organs, stems and leaves, and roots were significantly affected by the degree of anthropogenic disturbance (p<0.001), by the species (p<0.001), and by the altitude (p<0.001). However, the interactions between the degree of disturbance and (a) species and (b) altitude, between species and altitude, and between the degree of disturbance, species, and altitude were slightly different. The proportion of reproductive organs (by weight) had significant positive correlation with the interaction between the degree of disturbance and species (p<0.001), between species and altitude (p<0.001), and between the degree of disturbance, species, and altitude (0.01 <p<0.05). The proportion of stems and leaves (by weight) was affected neither by the interaction between the degree of disturbance and species (p<0.05) nor by that between the degree of disturbance and altitude (p<0.05; Table 2). The proportion of roots (by weight) was significantly affected by the interaction between the degree of disturbance, species, and altitude. At both low and high altitudes, the proportion (by weight) of reproductive organs in the populations of *S. involucrata* increased markedly (p<0.05) as the degree of anthropogenic disturbance decreased, whereas the proportion of non-reproductive parts showed an opposite pattern (Fig. 5a and 5b). In *S. involucrata*, the highest proportions of reproductive organs were reached at LD (from 0.00% at SD to 19.90% at LD) and those of the non-reproductive organs at SD. However, these proportions in *R. quadrifida*—whether at high altitudes or at low altitudes—were not significantly affected by the degree of anthropogenic disturbance except at SD at low altitudes (Fig. 5a and 5b). Thus at both high and low altitudes, the allocation of biomass to reproductive organs in *S. involucrata* increased markedly (p<0.05)—and the allocation to the roots decreased markedly—as the degree of anthropogenic disturbance decreased (Fig. 6a and 6b). However, the proportions (by weight) of the reproductive organs, roots, and stems and leaves were not significantly affected (p>0.05) by the differences in the degree of anthropogenic disturbance (Fig. 6a and 6b). At low altitudes, the proportions of reproductive organs in *S. involucrata* were 0.00% at SD, 3.57% at ID, and 10.90% at LD. At high altitudes, the corresponding values were 5.32%, 12.35%, and 20.85%, respectively. Unlike *S. involucrata*, *R. quadrifida* allocated most of its biomass to roots (Fig. 6c and 6d). At low altitudes, the proportions of reproductive organs in *R. quadrifida* were 0.55% at SD, 0.65% at ID, and 1.92% at LD. At high altitudes, the corresponding values were 0.61%, 0.43%, and 0.64%. The allocation of biomass—especially to reproductive organs in *S. involucrata* and to roots in *R. quadrifida*—was deeply influenced by the degree of anthropogenic disturbance. This pattern of allocation is bound to lower both reproductive capacity and dispersal ability of the two species.

Discussion

Anthropogenic disturbance and climate change have proved detrimental or even catastrophic in many parts of the world in the recent past (Rockstrom *et al*., 2009). Alpine plants in alpine subnival habitats are confronted by extreme environmental conditions (Cavieres *et al*., 2005), and the survival, growth, and development of such species are threatened by anthropogenic disturbance. The present study indicated that collection by local nomads and tourists, although but illegal, was the most serious form of anthropogenic disturbance to *S. involucrata* and *R. quadrifida*, and their populations were also affected by grazing. Wild population of *S. involucrata* are dwindling rapidly because of overexploitation (Kang *et al*., 2010), and the area over which the species is distributed has shrunk from more than 20 million hectares in the 1960s and the 1970s to a few hundred hectares at present (Chik *et al*., 2015). This rare species could become extinct in less than a decade. In 2015, a news item that police had to be brought in to investigate the incident of a traveller illegally collecting *S. involucrata* in Urumqi in Tianshan caused serious concern throughout China (Anonymous, 2015). Indeed, anthropogenic interference in the form of such illegal and unsustainable collection will profoundly change the population density, biomass, and reproductive success of *S. involucrata* and *R. quadrifida*, threatening the survival and dispersion of both the species.

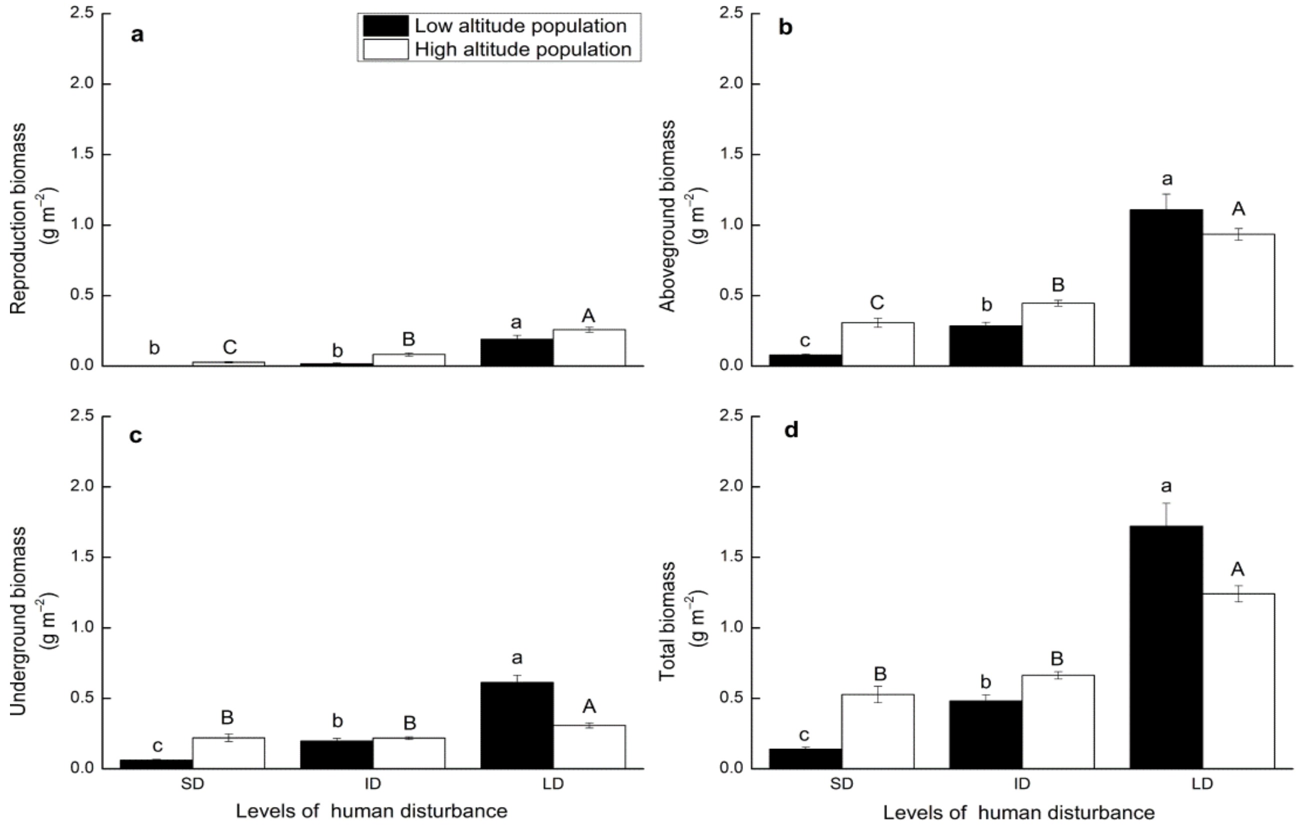

Fig. 3. changes in reproductive biomass, above-ground biomass, below-ground biomass, and total biomass of *S. involucrata* at different levels of anthropogenic disturbance. Data are mean values ± standard error (*N* = 5). Different letters between samples indicate significant differences using Duncan's test (p<0.05). LD, light disturbance; ID, intermediate disturbance; SD, severe disturbance.

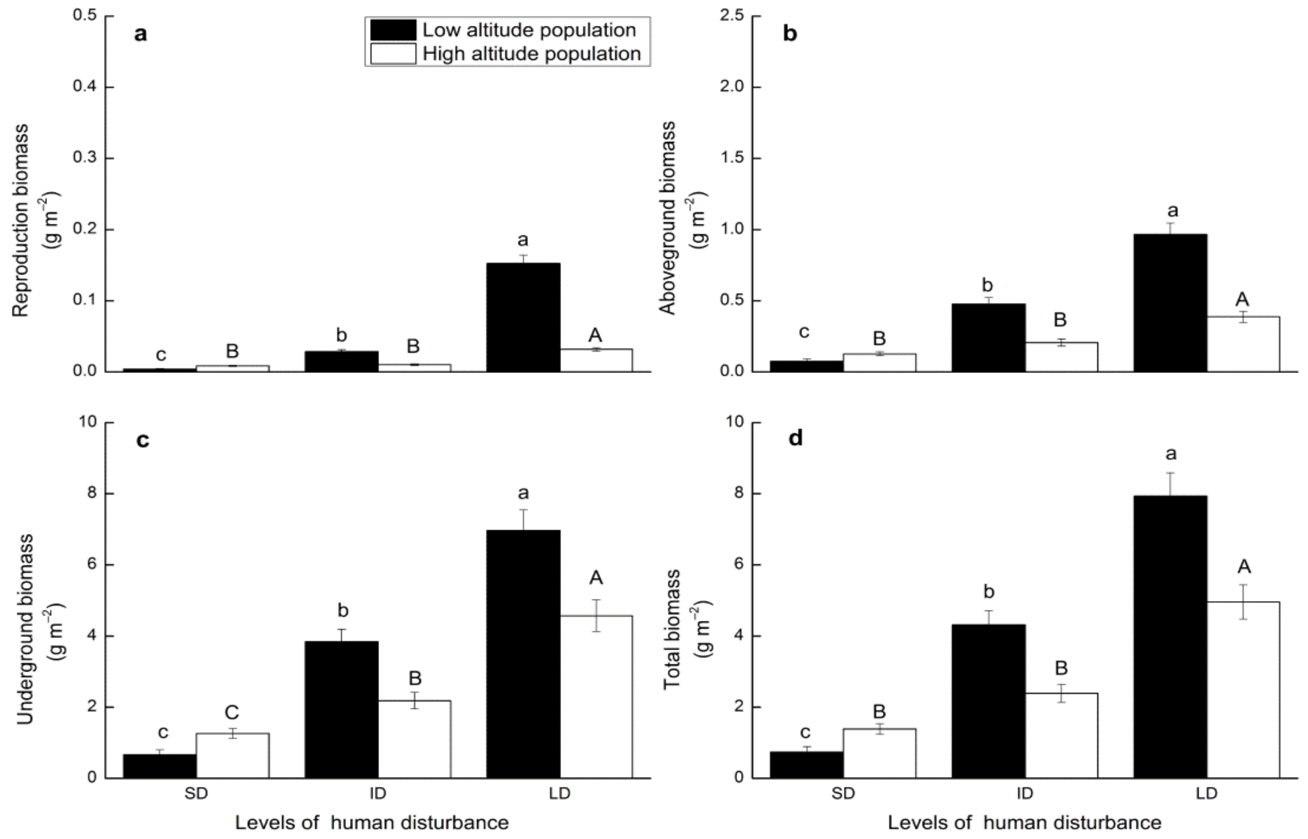

Fig. 4. Changes in reproductive biomass, above-ground biomass, below-ground biomass, and total biomass of *R. quadrifida* at different levels of anthropogenic disturbance. Data are mean values ± standard error (*N* = 5). Different letters between samples indicate significant differences using Duncan's test (p<0.05). LD, light disturbance; ID, intermediate disturbance; SD, severe disturbance.

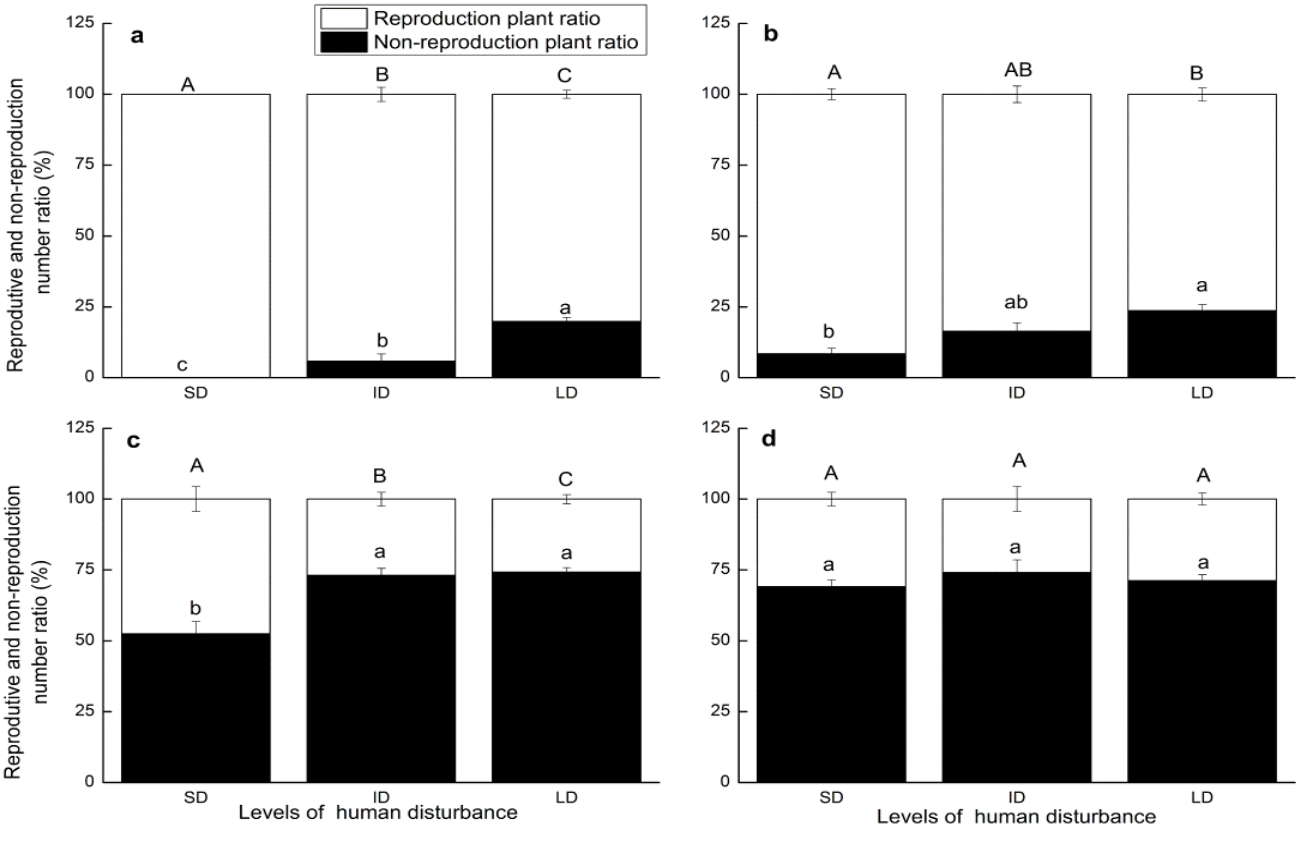

Fig. 5. Changes in the proportions of reproductive and non-reproductive plant numbers in *S. involucrata* (a and b) and *R. quadrifida* (c and d) at different levels of anthropogenic disturbance. Data are mean values ± standard error (N = 5). Different letters between samples indicate significant differences using Duncan's test (p<0.05). LD, light disturbance; ID, intermediate disturbance; SD, severe disturbance.

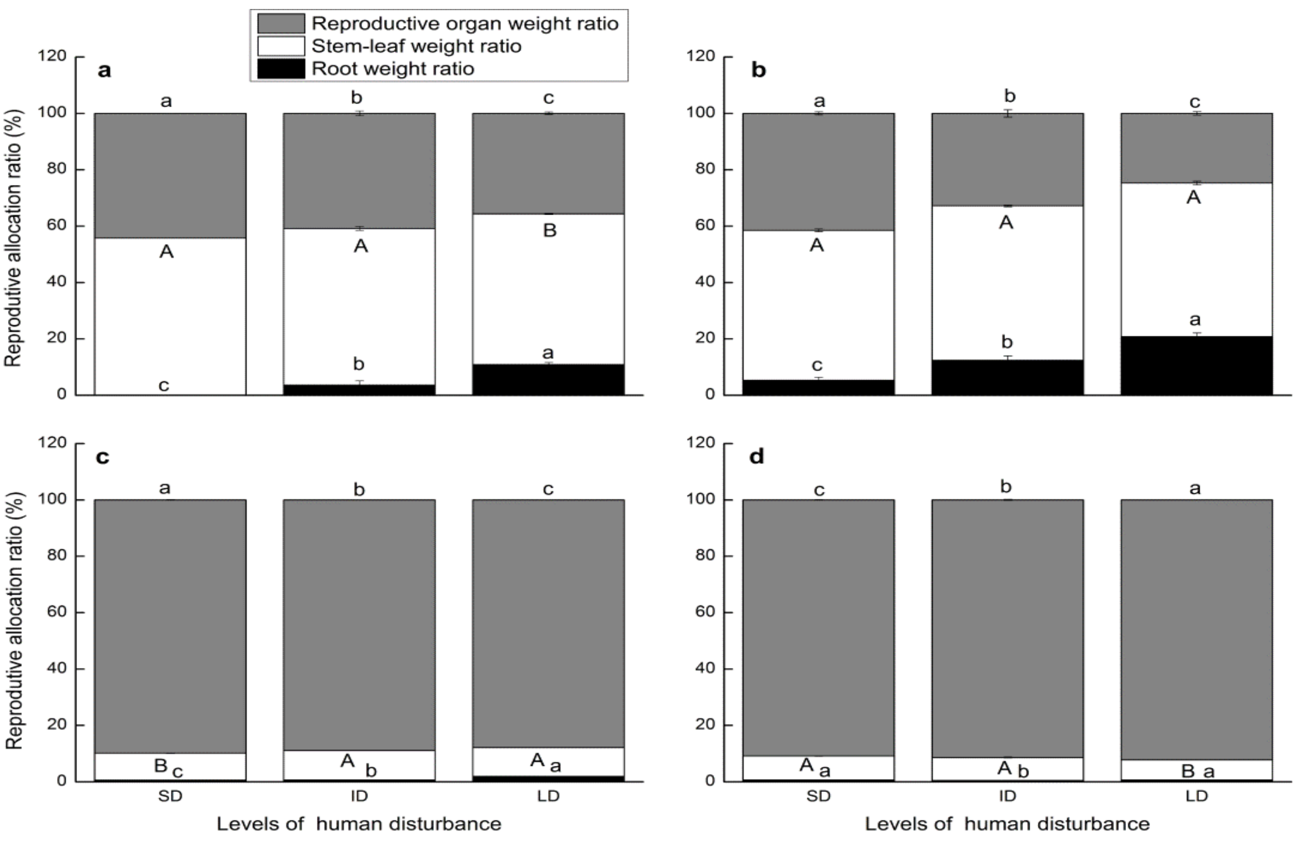

Fig. 6. Changes in the proportions (by weight) of reproductive organs, stems and leaves, and roots in *S. involucrata* (a and b) and *R. quadrifida* (c and d)at different levels of anthropogenic disturbance. Data are mean values ± standard error (N = 5). Different letters between samples indicate significant differences using Duncan's test (p<0.05). LD, light disturbance; ID, intermediate disturbance; SD, severe disturbance.

The data was taken from the results of field investigation and interview investigation with local herdsmen. Accessibility is the ease with which the sample plots can be reached from residential areas and roads. Collection trace was the trace left by collection plant. Number of collection people is the collection people at one year (that mainly on July, August and September). Number of collection frequency is the collection frequency by one people at one year (that mainly on July, August and September).

Normally, decreasing population size (or the number of individuals) is a direct consequence of overexploitation. The results of the present study point out that the density of *S. involucrata* and *R. quadrifida* in all the three forms—reproductive, non-reproductive, and total—decreased significantly with increased intensity of anthropogenic disturbance in low altitudes as well as in high altitudes (Fig. 1). Such a decrease may increase the risk that the two species will soon be extinct in the wild. Small populations are more vulnerable to the Allee effect (Groom, 1998): as population density decreases, smaller populations decline at ever-accelerating rates because individuals find it increasingly difficult to reproduce. Effectiveness of the breeding system, group defences against predators (human beings, who over-exploit *S. involucrata* and *R. quadrifida*, are the main predators in this case), and reproductive efficiencies may all decline at low population densities (Levitan *et al*., 1992). At the same time, harsh habitat conditions exercise stronger effects on individuals because they have fewer neighbours to protect them from the environment (Schiel & Choat, 1980). Demographic stochasticity can also increase the risk of extinction for small populations (Lande *et al*., 2003, Jeppsson & Forslund, 2012). Because death, birth, and and reproduction are discrete events, small populations may decline owing to chance events, although the average rates of survival and reproduction contribute to population growth (Wootton & Pfister, 2013). In the present study, the density of reproductive plants and the proportion (by weight) of reproductive organs in *S. involucrata* and *R. quadrifida* decreased significantly as the degree of anthropogenic disturbance increased at both altitudes (Figs. 1 and 4). In fact, anthropogenic disturbance can lower reproductive success in a population. In addition, a decrease in population or in population density will result in loss of plant genetic diversity and lower the rate of population growth (Wootton & Pfister, 2013). With fewer genetically different individuals and closely interrelated offspring after several generations, the potential for inbreeding depression in small populations is very high. Greater homozygosity is likely to show up many deleterious recessive mutations, and loss of heterozygotic advantages is closely associated with high offspring relatedness (Charlesworth & Charlesworth, 1987). In small populations, genetic drift may cause beneficial alleles to be lost and increase the chances of deleterious alleles being expressed, thereby slowing down the rate of population growth (Lande, 1994). Therefore, given the present situation, the numbers of *S. involucrata* and *R. quadrifida* are likely to decrease dramatically if anthropogenic disturbance continues, eventually leading to the two species being extinct.

Both *S. involucrata* and *R. quadrifida* showed similar patterns of change in biomass at each of the three levels of anthropogenic disturbance both at higher and lower altitudes. Reproductive biomass, above-ground biomass, below-ground biomass, and total biomass decreased significantly with an increase in the level of anthropogenic disturbance (Figa. 2 and 3). Such reductions in biomass will have adverse effects on reproduction and diffusion of the populations: lower biomass, especially vegetative biomass (the proportion by weight of stems and leaves), means lower above-ground net primary productivity of a population or a community (Zeng *et al*., 2015), which, in turn, may reduce the ability of *S. involucrata* and *R. quadrifida* populations to reproduce and to spread. Coupled with the depletion due to overexploitation, the populations of the two species will be too small to sustain their ecological and economic functions—once lost, these functions will be practically impossible to restore given the sensitivity of such depleted populations to anthropogenic disturbance and climatic extremes (Chen *et al*., 2013b; Hagen *et al*., 2014; Sarmiento *et al*., 2015). Traditionally, allocation has been the measure of reproductive allocation for individual plants and for populations (Cheplick, 2005). Any factor that influences plant size also affects reproductive allocation, and individuals in a population can vary enormously in size owing to a variety of factors (Wang *et al*., 2016). Our results showed that allocation of biomass to reproductive organs (which is reflected in the proportion, by weight, of the reproductive organs) in both *S. involucrata* and *R. quadrifida* were deeply influenced by anthropogenic disturbance and decreased significantly as the levels of anthropogenic disturbance increased (Fig. 4). Such inadequate allocation will, in turn, lower the ability of the population to reproduce and to disperse. Both growth and reproduction are fundamental to sustaining plant populations, but greater allocation to one often comes at a cost to the other (Reekie & Avila-Sakar, 2005). Long-lived species will allocate more resources for survival than to reproduction, as reproductive output and survival of juveniles are more variable than the survival of adults (Gaillard *et al*., 2000; Gaillard & Yoccoz, 2003). The balance between reproduction and survival will depend on how changes in the environment and anthropogenic disturbance affect the two functions (Bårdsen *et al*., 2008, 2010). In other words, some investment in reproduction is essential if a population is to reproduce and spread successfully. Thus, the balance between production of biomass and its allocation (especially to reproduction) in the populations of both *S. involucrata* and *R. quadrifida* would be destroyed if human interference continues, which may ultimately result in their extinction.

It is thus clear that anthropogenic disturbance was one of the most important factors that changed the density of, and biomass and biomass allocation in, the populations of *S. involucrata* and *R. quadrifida* significantly, furthermore these population parameters were also affected by the altitude, and the interaction between the species and the altitude (Table 2). Increasing anthropogenic disturbance in the form of exploitation of natural resources, infrastructure development, and tourism in alpine areas threatens not only individual species but entire ecosystems (Hagen *et al*., 2014). In the alpine subnival belt, the size and structure of the populations of *S. involucrata* and *R. quadrifida* are unlikely to be restored once they are depleted beyond a point by anthropogenic disturbance. In these habitats, natural recovery is usually slow (Jorgenson *et al*., 2010; Rydgren *et al*., 2013) because the plants grow and develop under conditions marked by short growing seasons, low summer and winter temperatures, and strong winds (Hagen *et al*., 2014). More critically, the population density and biomass of the two species have decreased with increases in the level of anthropogenic disturbance, making the plant resources even more precious and scarce. Decreased resources of *S. involucrata* and *R. quadrifida* may lead to even more intense anthropogenic disturbance. Populations that are easily accessible will be consistently the worst affected. Yet the level of anthropogenic disturbance will keep rising, although at present the less accessible sites show only light or intermediate disturbance. The increasingly warmer climate will shrink the available habitat further, and other species will expand their range (Felde *et al.*, 2012; Spasojevic *et al*., 2013), thereby increasing the threat to the survival and development of *S. involucrata* and *R. quadrifida*. Unless anthropogenic disturbance is checked and *S. involucrata* and *R. quadrifida* are protected, these species will probably disappear from the headwater area of the Urumqi River.

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