

1 **Effect of plateau pikas disturbance and patchiness on ecosystem carbon emission of**
2 **alpine meadow on the northeastern part of Qinghai-Tibetan Plateau**

3 Yu Qin¹, Shuhua Yi^{2, 1*}, Yongjian Ding^{1, 3}, Wei Zhang^{1, 3}, Yan Qin^{1, 3}, Jianjun Chen^{4,5}, Zhiwei
4 Wang^{1,6}

5 1. State Key Laboratory of Cryospheric Sciences, Northwest Institute of Eco-Environment
6 and Resources, Chinese Academy of Sciences, 320 Donggang West Road, Lanzhou 730000,
7 China

8 2. School of Geographic Sciences, Nantong University, 999 Tongjing Road, Nantong, Jiangsu,
9 226007, China

10 3. University of the Chinese Academy of Sciences, No.19A Yuquan Road, Beijing 100049,
11 China

12 4. College of Geomatics and Geoinformation, Guilin University of Technology, 12 Jiangan Ro
13 ad, Guilin, 541004, China

14 5. Guangxi Key Laboratory of Spatial Information and Geomatics, 12 Jiangan Road, Guilin, 5
15 41004, China

16 6. Guizhou Institute of Prataculture, Guizhou Academy of Agricultural Sciences, Guiyang,
17 550006, People's Republic of China

18 * E-mail: yis@lzb.ac.cn

19 Tel: +86-931-4967356

20 **Abstract**

21 Plateau pikas (*Ochotona curzoniae*) disturbance and patchiness intensify the spatial
22 heterogeneous distribution of vegetation productivity and soil physicochemical properties,
23 which may alter ecosystem carbon emission process. Nevertheless, previous researches have
24 mostly focused on the homogeneous vegetation patches rather than heterogeneous land
25 surface. Thus, this study aims to improve our understanding of the difference in ecosystem
26 respiration (Re) over heterogeneous land surface in an alpine meadow grassland. Six different
27 land surface: large bald patch, medium bald patch, small bald patch, intact grassland, above
28 pika tunnel and pika pile were selected to analyze the response of Re to pikas disturbance and
29 patchiness, and the key controlling factors. The results showed that (1) Re under intact
30 grassland were 0.22-1.07 times higher than pika pile and bald patches; (2) soil moisture (SM)
31 of intact grassland was 2-11% higher than those of pika pile and bald patches despite pikas
32 disturbance increased water infiltration rate, while soil temperature (ST) under intact
33 grassland was 1-3°C less than pika pile and bald patches; (3) Soil organic carbon (SOC) and
34 total nitrogen (TN) under intact grassland were approximate 50 % and 60 % less than above
35 pika tunnel, whereas 10-30 % and 22-110 % higher than pika pile and bald patches; and (4)
36 Re was significantly correlated with SM, TN and vegetation biomass ($P < 0.05$). Our results
37 suggested that pikas disturbance and patchiness altered ecosystem carbon emission pattern,
38 which was mainly attributed to the reduction of soil water and supply of substrates. Given that
39 the wide distribution of pikas and large area of bald patches, the varied Re under
40 heterogeneous land surfaces should not be neglected for estimation of ecosystem carbon
41 emission at plot or region scale.

42 **Keywords:** pikas disturbance; patchiness; ecosystem respiration; alpine meadow; the
43 Qinghai-Tibetan Plateau

44 **Introduction**

45 Ecosystem respiration (Re) is the key process to determine the carbon budget in the terrestrial
46 ecosystem. Thus, even a small imbalances between CO₂ uptake via photosynthesis and CO₂
47 release by ecosystem respiration can lead to significant interannual variation in atmospheric
48 CO₂ (Schimel et al., 2001; Cox et al., 2000; Grogan and Jonasson, 2005; Oberbauer et al.,
49 2007; Warren and Taranto, 2011). Dependent on autotrophic (plant) and heterotrophic
50 (microbe) activity, ecosystem respiration is mainly controlled by abiotic factors (primarily
51 temperature and water availability) (Chimner and Welker, 2005; Flanagan and Johnson, 2005;
52 Nakano et al., 2008; Buttlar et al., 2018), and supply of carbohydrate fixed by leaves,
53 vegetation litter and soil organic matter (Janssens et al., 2001; Reichstein et al., 2002).
54 Therefore, any external disturbance altering environmental conditions and affecting
55 vegetation growth would exert profound influence on ecosystem carbon emission.

56 One of the basic function of terrestrial ecosystem is to regulate carbon balance between
57 the atmosphere and ecosystem (Canadell et al., 2007; Le Quéré et al., 2014; Ahlström et al.,
58 2015). However, this balance would be broken by widespread land degradation (Post and
59 Kwon, 2000; Dregne, 2002), which accompanied with the reduction of photosynthetic fixed
60 carbon dioxide from atmosphere and carbon sequestration by soils (Defries et al., 1999;
61 Upadhyay et al., 2005). It was estimated that land degradation had resulted in 19-29 Pg C loss
62 worldwide (Lal, 2001). Over the past decades, grasslands have experienced patchiness
63 throughout the world and this process is still ongoing (Baldi et al., 2006; Wang et al., 2009;
64 Roch and Jaeger, 2014). Patchiness generally refers to a landscape that consists of remnant
65 areas of native vegetation surrounded by a more heterogeneous and patchy situation (Kouki
66 and Löfman, 1998). Other than climate change (Yi et al., 2014), vegetation self-organization
67 (Rietkerk et al., 2004; Venegas et al., 2005; McKey et al., 2010) or anthropogenic
68 disturbances (Kouki and Löfman, 1998; Yi et al., 2016), rodents burrowing activities were
69 also considered as the origin of the patchiness (Wei et al., 2006; Davidson and Lightfoot,
70 2008). This patchiness intensified spatial heterogeneity of land surface and led to the
71 changing of the structure and function of the original ecosystem (Herkert et al., 2003;
72 Bestelmeyer et al., 2006; Lindenmayer and Fischer, 2013). For instance, there is abundant
73 evidence that patchiness not only intensified the spatial heterogeneous distribution of

74 ecosystem organic carbon (C) and vegetation productivity (Yan et al., 2016; Qin et al., 2018)
75 but also altered the pattern of coupled water and heat cycling between the land surface and the
76 atmosphere (Saunders et al., 1991; You et al., 2017; Ma et al., 2018). Consequently, this may
77 alter ecosystem carbon emission process (Juszczak et al., 2013).

78 Plateau pikas (*Ochotona curzoniae*, hereafter pikas) are small mammals endemic to the
79 alpine grasslands on the Qinghai-Tibetan Plateau (QTP) (Smith and Foggin, 1999; Lai and
80 Smith, 2003). Living in underground, they excavated deep layer soil to surface through
81 foraging and digging activities (Lai and Smith, 2003) and led to substantial bald piles on the
82 ground. The bald pile was considered to gradually become bald patches under soil erosion,
83 gravity, freeze-thaw and other factors (Chen et al., 2017; Ma et al., 2018). As a consequence,
84 natural vegetation patches and adjacent bald patches with different sizes, and pikas piles
85 represent the most common landscape pattern in the alpine meadow grassland on the QTP.
86 Previous studies have demonstrated that pikas disturbance and patchiness weaken the function
87 of alpine meadow as a carbon sink (Liu et al., 13; Peng et al., 2015; Qin et al., 2018) and
88 accelerated ecosystem carbon emission rate (Qin et al., 2015a). Nevertheless, most of these
89 studies have mainly focused on ecosystem carbon emission rate under the homogeneous land
90 surface rather than heterogeneous land surfaces. Thus, the specific aims of this study were to
91 (1) investigate the spatial heterogeneity of Re among different surface types (plateau pika pile,
92 different sizes of bald patches and vegetation) of alpine grassland; (2) illuminate the potential
93 regulating mechanism of pikas disturbance and patchiness to ecosystem respiration (Re) in an
94 alpine meadow grassland in the northeastern part of Qinghai-Tibetan Plateau (QTP).

95 **Materials and methods**

96 **Site description**

97 This study was conducted at the permanent plots at Suli Alpine Meadow Ecosystem
98 Observation and Experiment Station (98°18'33.2", 38°25'13.5", 3887 m a.s.l.), Northwest
99 Institute of Eco-Environment and Resources, Chinese Academy of Science. The study area is
100 characterized by a continental arid desert climate, with low mean annual air temperature, little
101 rainfall, and high evaporation (Wu et al., 2015). The mean annual air temperature was
102 approximately -4°C and the annual precipitation ranged from 200 to 400mm, respectively
103 (Chang et al., 2016). The permafrost type at our site is transition and the active layer depth is

104 2.78 ± 1.03 m (Chen et al., 2012). The dominant plant species in the study area were *Kobresia*
105 *capillifolia*, *Carex moorcroftii* (Qin et al., 2014). Soils was classified as “felty” with a pH of
106 8.56, 30.96 % silt and fine, 57.52 % fine sand and 10.68 % coarse sand, and soil bulk density
107 is 1.41 g cm⁻³ within a 0-40 cm depth of the soil layer (Qin et al., 2015b). The grassland in
108 this area suffered from degradation due to permafrost degradation and external disturbance
109 from grazing livestock and small mammals, i.e. plateau pikas (Yi et al., 2011, Qin et al.,
110 2015a). As a result, a mosaic pattern of vegetation patches, bald patches with different sizes
111 and pika piles was common.

112 **Field observation**

113 At early June 2016, three 100 m × 100 m plots were established as replicates. Each 100 × 100
114 m plot was in a distance of less than 50 m, which has the similar plant and terrain. In each
115 plot, six representative land surfaces were selected: (1) large bald patch with size larger than
116 9.0 m² (LP), (2) medium bald patch with size of 1.0-9.0 m² (MP), (3) small bald patch with
117 size of less than 1.0 m² (SP), (4) intact grassland patch (IG), (5) above pika tunnel (PT), (6)
118 old pika pile (PP) (Figure 1) (Yi et al., 2016; Qin et al., 2018). **There were no other mammals,**
119 **e.g. marmot and zokor in our study plots. All of the piles in each plot were created by plateau**
120 **pikas.** They were distinguished easily in aerial photographs. Large bald patches had less
121 vegetation cover and the smallest side was larger than 3 m. Medium patches also covered by
122 less vegetation cover and the largest side was in a range of 1 to 3 m and small bald patches
123 were characterized by less vegetation cover and the largest side was less than 1 m. Intact
124 grassland was characterized by high vegetation cover and no large and medium bare land was
125 found. Pika tunnel and pika pile usually co-existed. Pika tunnel is approximately 6 cm in
126 diameter and pika pile is in the front of pika tunnel, 60 cm in diameter and less vegetation
127 cover. We calculated the threshold area of large, medium and small patches by aerial
128 photograph. Each aerial photograph has 12 million pixels. At a height of 20 m, the resolution
129 of each pixel is ~1 cm and each photograph covers ~26 m × 35 m of ground. Pixels in each
130 aerial image were first classified into two groups, i.e. vegetated or bare patches (Yi, 2017).
131 Then patches with different sizes were created using OpenCv Library. And finally, fractions
132 of vegetation and bare patches (large, medium and small patches) were calculated. **For each**
133 **surface type in each plot, six 1 m × 1 m quadrats were set up, of which three was used for soil**

134 saturated hydraulic conductivity measurement and three for soil compactness measurement,
135 soil and vegetation sampling. We also set up another three 1 m × 1 m quadrats and three 2 m
136 × 2 m quadrats in each surface type in a 100 m × 100 m plot for measuring soil temperature,
137 soil moisture and ecosystem respiration.

138 (Insert Figure 1 here)

139 A meteorological tower was established in our observation station since 2008. Air
140 temperature (°C) at 2.0m was measured by HMP45C (Vaisala, Helsinki, Finland), and
141 precipitation was measured using an all-weather precipitation gauge (Geonor T-200B,
142 Norway) (Wu et al., 2015). Soil temperature and moisture at 10 cm were measured by using
143 an auto-measurement system (Decagon Inc., USA) from early June to the late August. The
144 system consisted of an EM50 logger and five 5TM sensors. The data were logged
145 automatically every 30 minutes. Soil saturated hydraulic conductivity and compactness were
146 measured one time in each month from June to August. Soil saturated hydraulic conductivity
147 was measured by Dual Head infiltrometer (Decagon Inc., USA). The measurement process
148 included soak time 15 minutes, hold time 20 minutes at low pressure head (5 cm) and high
149 pressure head (15 cm) with 2 cycles. Each measurement takes 95 minutes altogether. Soil
150 compactness was measured with TJSD-750 (Hangzhou Top Instrument co., LTD, Hangzhou,
151 China) from the soil surface to 10 cm depth. Ecosystem respiration rates were measured using
152 the LICOR-8150 Automated Soil CO₂ Flux System, which was an accessory for the
153 LI-8100A could connect 16 individual chambers at one time and were sampled and controlled
154 by the LI-8100A Analyzer Control Unit. The air temperature inside of the chamber was
155 measured using the internal thermistor of the chamber. The ecosystem CO₂ fluxes were
156 calculated by the equation as follow.

$$157 \quad Fc = \frac{10VP_0 \left(1 - \frac{W_0}{1000}\right) \partial C'}{RS(T_0 + 273.15) \partial t}$$

158 where Fc is the soil CO₂ efflux rate ($\mu\text{mol m}^{-2} \text{s}^{-1}$), V is volume (cm^3), P_0 is the initial pressure
159 (kPa), W_0 is the initial water vapor mole fraction (mmol mol^{-1}), R is the ideal gas constant, S
160 is soil surface area (cm^2), T_0 is initial air temperature (°C), and $\partial C'/\partial t$ is the initial rate of
161 change in water-corrected CO₂ mole fraction ($\mu\text{mol}^{-1} \text{s}^{-1}$).

162 Six LICOR-8100-104 long-term opaque chambers (20cm in diameter LICOR, Inc.,
163 Lincoln, NE, USA) were used to measure alternately between three replicates for six land
164 surface types. Therefore, 3 days at least were required to complete one rotation measurements
165 of ecosystem respiration. To measure ecosystem respiration, eighteen polyvinyl chloride
166 collars with a 20 cm inner diameter and a 12 cm height were inserted into the soil with 3-4 cm
167 exposed to the air (Qin et al., 2013). All of the collars were installed at least 24 h before the
168 first measurement to reduce disturbance-induced ecosystem CO₂ effluxes. Ecosystem
169 respiration rates were measured every 7-10 days from June 16 to August 20 in 2016
170 depending on weather conditions. A round-the-clock measurement protocol was carried out
171 and ecosystem respiration rates were measured every 30 minutes. Each measurement takes 1
172 minute and 45 seconds, including pre-purge 10 seconds, dead band 15 seconds, observation
173 length 1 minute and post-purge 20 seconds.

174 **Soil and vegetation sampling**

175 Soil samples were collected during the periods of late July to early August 2016. In each
176 surface type of each plot, five soil cores were collected using a stainless-steel auger (5 cm in
177 diameter) at depths of 0-10, 10-20, 20-30 and 30-40 cm, and bulked as one composite sample
178 for each depth in each quadrat. Another five soil cores were sampled by cylindrical cutting
179 ring (7 cm in diameter and 5.2 cm in depth) to determine soil bulk density from each land
180 surface type. Pika tunnel was approximate 6 cm in diameter and 40 cm in depth. Therefore,
181 soil samples were available to collect at depth of 40cm. Totally, 512 soil samples were
182 collected. Soil samples were firstly air-dried, then removed gravel and stone with manual
183 sieving and finally weighed. The remaining soil samples with diameter less than 2 mm were
184 ground to pass through a 0.25 mm sieve for analysis of soil organic carbon (SOC) and soil
185 total nitrogen (TN) concentration. SOC was measured by dichromate oxidation using
186 Walkley-Black acid digestion (Nelson and Sommers, 1982). TN was determined by digestion
187 and then tested using a flow injection analysis system (FIAstar 5000, Foss Inc., Sweden).
188 Aboveground and belowground biomasses were determined within a 1 m × 1 m quadrat on 4
189 August 2016 during peak biomass and species diversity. There were a total of 108
190 aboveground and belowground vegetation samples (3 plots × 6 land surface types × 3
191 replicates) from the study area. Aboveground biomass was determined by clipping all

192 above-ground living plants at ground level, drying (oven-dried at 65°C for 48 h) and weighing.
193 Belowground biomass was sampled by collecting five soil columns, and each soil column was
194 5 cm in diameter and 40 cm in depth. Soil cores were washed with a gentle spray of water
195 over a fine mesh screen until soil separated from the roots, and then drying (oven-dried at
196 65°C for 48 h) and weighing.

197 **Statistical analysis**

198 The soil organic C and total N densities in different land surface were calculated using the
199 equation (2) and (3):

$$200 \quad \text{SOC} = \sum_{i=1}^n \rho * (1 - \sigma_{\text{gravel}}) * C_{\text{SOC}} * D_i \quad (2)$$

$$201 \quad \text{TN} = \sum_{i=1}^n \rho * (1 - \sigma_{\text{gravel}}) * C_{\text{TN}} * D_i \quad (3)$$

202 where SOC is soil organic C density (kg m⁻²), TN is soil total N density (kg m⁻²), ρ is the soil
203 bulk density (g cm⁻³), σ_{gravel} is the relative volume of gravel (% w/w), C_{SOC} is soil organic C
204 content (g kg⁻¹), C_{TN} is soil total N content (g kg⁻¹) and D_i is soil thickness (cm) at layer i ,
205 respectively; $i=1, 2, 3$ and 4 .

206 The data were presented as mean \pm standard deviation. Statistical analyses were performed
207 using the SPSS 17.0 statistical software package (SPSS Inc., Chicago, IL, USA). Two-way
208 analysis of variance (ANOVA) and a multi-comparison of a least significant difference (LSD)
209 test were used to determine differences at the $p=0.05$ level. The relationships of ecosystem
210 respiration with biotic and abiotic factors were analyzed by Pearson correlation analysis using
211 R.

212 **Results**

213 **Ecosystem respiration**

214 Pika disturbance had significant effect on ecosystem respiration in June and July (Table 1,
215 $P<0.05$), while the significant effect of patchiness on ecosystem respiration was found in July
216 and August (Table 1, $P<0.05$). During the growing season, ecosystem respiration maximized
217 in August and minimized in June (Figure 2). In June, ecosystem respiration under intact
218 grassland, above pika tunnel, small patch and pika pile had no significant difference and the
219 lowest ecosystem respiration was found under large and medium patches (Figure 2). Average

220 ecosystem respiration under intact grassland was $4.01 \mu\text{mol m}^{-2} \text{s}^{-1}$ in July, which was
221 24.35 % to 137.39 % higher than other surface types (Figure 2). In August, average ecosystem
222 respiration were $4.07 \mu\text{mol m}^{-2} \text{s}^{-1}$ and $4.85 \mu\text{mol m}^{-2} \text{s}^{-1}$ for intact grassland and above pika
223 tunnel, $2.59\text{-}3.81 \mu\text{mol m}^{-2} \text{s}^{-1}$ for bald patches and $1.18 \mu\text{mol m}^{-2} \text{s}^{-1}$ for pika pile (Figure 2).

224 (Insert Table 1, Figure 2 here)

225 **Microclimate and soil hydrothermal characteristics**

226 Mean temperature and total rainfall during the growing seasons from 1 May to 30 September
227 in 2016 were $6.18 \text{ }^{\circ}\text{C}$ and 343.4 mm , respectively (Figure 3). Soil temperature and moisture
228 were significantly different among various land surface types (Table 1, $P < 0.05$). The monthly
229 average soil temperature was in a range of $8.20\text{-}13.72 \text{ }^{\circ}\text{C}$ during June to August, which was
230 approximate $1\text{-}3 \text{ }^{\circ}\text{C}$ higher under pika pile and bald patches than the intact grassland (Figure
231 4a, $P < 0.05$). The monthly mean soil moisture from June to August was approximate 30 % for
232 intact grassland and above pika tunnel, 25 % for small patch and pika pile, and 20 % for
233 larger and medium patch (Figure 4b). Soil saturated hydraulic conductivity had no significant
234 difference among different land surfaces (Table 2, $P > 0.05$). However, soil saturated hydraulic
235 conductivity under intact grassland was approximate 40 % higher than medium and large
236 patches and 17 % lower than pika pile (Figure 5).

237 (Insert Table 2, Figure 3 to 5 here)

238 **Soil and vegetation properties**

239 Both pikas disturbance and patchiness significantly affected soil compactness, SOC density,
240 TN density and vegetation biomass (Table 2, $P < 0.05$). Soil compactness was over 0.30 Pa in
241 intact grassland and above pika tunnel, approximate 0.20 Pa for bald patches and less than
242 0.10 Pa for pika pile (Figure 6), respectively. Mean SOC and TN density under intact
243 grassland were 52.45 % and 59.14 % less than above pika tunnel, whereas they were
244 9.69-30.12 % and 22.47-109.62 % higher than pika pile and bald patches (Figure 7).
245 Aboveground and belowground biomass under intact grassland were approximate 30 %
246 higher than above pika tunnel, 90 % higher than pika pile, 123-252 % and 134-289 % higher
247 than bald patches (Figure 8a, b).

248 (Insert Figure 6 to 8 here)

249 **Factors regulate ecosystem respiration**

250 We analyzed the relationships of ecosystem respiration with biotic and abiotic factors for six
251 land surface types (Figure 9). Correlation analysis showed that ecosystem respiration had no
252 significant correlation with soil temperature ($P>0.05$, Figure 9). However, ecosystem
253 respiration was significantly and positively related to soil moisture ($P<0.01$), soil total
254 nitrogen ($P<0.05$), aboveground ($P<0.05$) and belowground biomass ($P<0.05$) (Figure 9).

255 (Insert Figure 9 here)

256 Discussion

257 Effect of pikas disturbance on ecosystem respiration

258 Pikas burrowing activities increased oxygen content in deep soil, which contributed to the
259 decomposition of soil organic matter (Martin, 2003). The deposition of urine and feces by
260 small herbivorous mammals could also promote ecosystem nutrition circulation (Clark et al.,
261 2005). It was suggested that excreta deposited by pikas and frequently haunted in or near their
262 burrows supplied organic C available to microbial decomposition with an increase in
263 ecosystem CO₂ emission (Cao et al., 2004). Indeed, SOC and TN densities reached up to
264 14.54 and 0.98 kg m⁻² in above pika tunnel, which was 2.45 and 2.10 times higher than that of
265 intact grassland (Figure 7), respectively. The consistent results reported that the contents of
266 available soil nutrients around the pikas burrow were higher than those in control sites on an
267 alpine meadow (Zhang et al., 2016). We also found that SOC and TN densities under pika pile
268 decreased 13.35 % and 42.93 % than intact grassland. This was because pika burrowing
269 activity transferred of deeper, nutrient-poor soil to the soil surface, improved soil aeration
270 increased rate of organic carbon mineralization and soil erosion took away soil nutrition (Wei
271 et al., 2006; Qin et al., 2015a; Chen et al., 2017). However, except July, no significant
272 difference of Re was found between intact grassland and above pika tunnel, while Re under
273 pika pile was 42.08 % less than intact grassland (Figure 2). The similar result was also found
274 in an alpine meadow on the QTP (Peng et al., 2015), which indicated that ecosystem
275 respiration decreased with increasing of pika holes because of grassland biomass regulated
276 soil C and N with increasing number of pika holes. These results confirmed that pikas
277 disturbance did not increase ecosystem carbon emission directly, but facilitated CO₂ emission
278 into the atmosphere through pika holes (Qin et al., 2015a). The difference of ecosystem
279 respiration between intact grassland and pika piles was mainly related to changes in

280 vegetation biomass and soil moisture. For example, both aboveground and belowground
281 biomass decreased 244.62 % and 279.89 % under pika piles compared with the intact
282 grassland (Figure 8). The reduction of vegetation biomass production decreased aboveground
283 plant respiration and root respiration by decreasing carbon allocation (e.g., root exudates and
284 litter, and available SOC) (Raich and Potter, 1995; Högberg et al., 2002; Yang et al., 2018).
285 Consistent with previous studies which demonstrated that pikas burrowing activity increased
286 water infiltration rate (Hogan, 2010; Wilson and Smith, 2015), our results also showed that
287 soil saturated hydraulic conductivity in pika pile was significantly higher than bald and
288 vegetation patches (Figure 5). Nevertheless, the increased water infiltration was unable to
289 increase soil moisture under pika piles. For example, soil moisture under pika piles was
290 approximate 5 % lower than intact grassland (Figure 4). Our result was discrepant with
291 previous studies which reported old pika mound had the highest soil moisture during the
292 summer (Ma et al., 2018) and moderate pika burrowing activities increased surface soil
293 moisture (Li and Zhang, 2006). This difference may be contributed to the high pika density in
294 alpine meadow (Guo et al, 2017). Moreover, pika piles were loose (Figure 6) with less
295 vegetation cover (Figure 8), which was not beneficial for soil moisture storage.

296 **Effect of patchiness on ecosystem respiration**

297 Our results clearly showed that patchiness resulted in significant reduction of ecosystem
298 carbon emission. Compared with the intact grassland, ecosystem respiration decreased
299 approximate 17-48 % for bald patches (Figure 2). Two possible mechanisms could account
300 for the effects of patchiness on ecosystem respiration. On one hand, the reduction of SOC and
301 TN decreased microbial respiration by decreasing substrate supply to microbes in the
302 rhizosphere (Nobili et al., 2001; Scott-Denton et al., 2010). Our results indicated that
303 patchiness caused evident loss of SOC and TN (Figure 7) due to reduction in C input from
304 vegetation and increasing in C output from soil erosion (Qin et al., 2018). Previous study have
305 shown that the spatial heterogeneity of soil respiration was attributed to uneven soil organic
306 carbon and total nitrogen content (Xu and Qi, 2010). Soil organic carbon was considered as
307 the basic substrate of CO₂ emission by microbial decomposition (Sikora and Mccoy, 1990)
308 and soil total N enhanced ecosystem CO₂ emission by providing a source of protein for
309 microbial growth (Tewary et al., 1982). On the other hand, low moisture availability would

310 limit microbial respiration by restricting access to C substrates, reducing the diffusion of C
311 substrates and extracellular enzymes, and limiting microbial mobility (Yuste et al., 2003;
312 Wang et al., 2014). Our results showed that soil moisture under large and medium patches
313 decreased 10 % than intact grassland (Figure 4). Previous studies had reported that the soil
314 compaction of bald patches decreased the rate of water infiltration (Wuest et al., 2006; Wilson
315 and Smith, 2015), which was similar with our results showed that bald patches had less
316 saturated soil hydraulic conductivity (Figure 5). Low vegetation cover under bald patches was
317 not beneficial for water retention and utilization, where most of soil water was mainly lost as
318 a way of evaporation (Yi et al., 2014). We have measured evaporation of the intact grassland,
319 isolate grassland, large patches, medium patches and small patches since the early June 2016.
320 Three years results indicated that evaporation under bald patches were higher than the intact
321 grassland (data were not shown here).

322 **Factors affected ecosystem respiration**

323 Most previous studies showed that soil temperature explained most of the temporal variation
324 of ecosystem respiration on the alpine grassland on the QTP (Lin et al, 2011; Qin et al., 2015c;
325 Zhang et al., 2017). Our results indicated that soil temperature under pika piles and bald
326 patches was approximate 1 to 3 °C higher than intact grassland (Figure 4), which mainly
327 resulted from the heterogeneity of surface albedo, surface soil water retention, heat
328 conduction properties and radiation (Beringer et al., 2005; Pielke, 2005; Yi et al., 2013; You et
329 al., 2017). It was suggested that pikas disturbance create a better soil temperature buffer for
330 them to avoid the extreme cold in winter (Ma et al., 2018), whereas high soil temperature
331 under bald patch was a disadvantage for the recovery of vegetation because patch surface had
332 the smallest soil moisture content (Figure 4) and the largest daily range of soil temperature
333 (Ma et al., 2018). **It was well known that rising of soil temperature under natural condition
334 enhanced ecosystem respiration by stimulating decomposition of soil organic matter (Conant
335 et al., 2008), increasing plant biomass (Yi et al., 2014) and activity of microbial enzymes
336 (Bond-Lamberty and Thomson, 2010). However, obvious relationship between Re and soil
337 temperature was not found in the present study (Figure 9), which suggested that other factors
338 involved in controlling Re induced by pikas disturbance and patchiness.** Our results showed
339 that Re were positively correlated with soil moisture, soil total nitrogen, aboveground and

340 belowground biomass (Figure 9). Pikas disturbance and patchiness led to the drying and
341 loosening of soil (Figure 4 and 6). It was considered that loose, dry surface sediments and
342 strong winds were the primary factors responsible for soil erosion (Dong et al., 2010b) and
343 wind erosion was especially common in arid and semi-arid regions (Zhang and Dong, 2014).
344 This resulted in the reduction of soil organic carbon, total nitrogen and vegetation biomass
345 (Figure 7 and 8). The alteration of these biotic and abiotic factors induced by pikas
346 disturbance and patchiness led to the decline of ecosystem respiration. Nevertheless, the
347 decline of ecosystem respiration did not completely offset the sequestration of C fixed by
348 photosynthesis because of the lower vegetation cover under bald patches and pika piles.
349 Given the large area covered by bald patches in alpine grasslands, patchiness was more
350 susceptible to erosion and exert greater influence on ecosystem respiration than pikas
351 disturbance. Recent study has also reported that bald patches of various sizes on the
352 grasslands played a much more important role than pikas direct disturbance in reducing
353 vegetation cover, aboveground biomass, soil carbon and nitrogen (Yi et al., 2016).

354 **Effect of pikas disturbance on patchiness**

355 Natural vegetation patches, bald patches with different sizes and pikas piles coexisted on the
356 alpine meadow (Figure 1), which supported that alpine grassland had also experienced
357 fragmentation (Qin et al., 2018). Several proposed mechanisms may be accounted for the
358 formation and development of patchiness in alpine grassland. As one of dominant form of
359 land utilization, alpine grasslands are widely used for grazing. Previous studies suggested that
360 overgrazing destroyed the original vegetation and led to decrease in the coverage and
361 looseness of soil (Dong et al., 2013), which was prone to form bald patch due to soil erosion
362 (Fécan et al., 1998; Zhang and Dong, 2014). Other than livestock, alpine grassland is also
363 habitats for many small mammals such as plateau pika, zokor (*Eospalax fontanierii*), marmot
364 (*Marmota himalayana*) and fox (*Vulpes ferrilata*). Pikas were considered to create a patchy
365 matrix by changing soil properties (Chen et al., 2017), digging tunnels and burying activities
366 (Dong et al., 2013). On one hand, pikas bury vegetation by fresh excavated soil, then small
367 bare soil patches are formed and further large soil patches are then formed by linking small
368 bare soil patches by wind and/or water (Wei et al., 2007; Ma et al., 2018). On the other hand,
369 pikas dig tunnel underground. Although pikas make burrows are the primary homes to a wide

370 variety of small birds and lizards (Smith and Foggin, 1999), the collapse of pika tunnels
371 results in the emergence of bald soil patches (Zhou et al., 2003; Cao et al., 2010). Moreover,
372 alpine grassland is underlain by extensive permafrost (Chen and Wu, 2007). The repeated
373 freeze and thaw cause the crack of the sod around the barren area (Yang et al. 2003) and
374 create precondition for forming bald patch. However, to date, there are no direct evidences to
375 demonstrate the potential mechanism for forming and developing of patchiness for alpine
376 grassland on the QTP. It is, therefore, critical to perform long-term repeated monitoring
377 studies to determine whether bald patches are developed from pika piles or burrow tunnels
378 and what the major factors affecting bald patch expansion are (Yi et al., 2016).

379 **Conclusions**

380 In this study, we investigated soil physicochemical properties, vegetation biomass and
381 ecosystem respiration (Re) under six land surfaces originating from pikas disturbance and
382 patchiness. We also analyzed the dominant factors regulated the Re. Our results showed that
383 pikas disturbance and patchiness decreased soil moisture but increased soil temperature,
384 which may be conducive to pikas survive in cold season but disadvantage for vegetation
385 growth. Patchiness caused evident decreasing in SOC and TN density, while both SOC and
386 TN density showed different response under pika piles and burrows. Both pikas disturbance
387 and patchiness decreased ecosystem carbon emission, and ecosystem respiration sharply
388 correlated with soil moisture, TN and vegetation biomass. Our results indicated that pikas
389 disturbance and patchiness led to the changing of ecosystem respiration process owing to the
390 drying of soil and the reduction of substrate supply. However, the decline of ecosystem
391 respiration may not able to offset the sequestration of C fixed by photosynthesis.

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660 **Table 1.** Two-way ANOVA results of the effect of patches fragmentation and pikas
 661 disturbance on soil temperature, soil moisture and ecosystem respiration.

		Soil temperature			Soil moisture			Ecosystem respiration		
		Jun	Jul	Aug	Jun	Jul	Aug	Jun	Jul	Aug
Patchiness	<i>F</i>	10.44	20.63	3.51	218.23	205.44	62.56	7.03	18.98	2.71
	<i>P</i>	<0.001	<0.001	0.03	<0.001	<0.001	<0.001	0.002	<0.001	0.12
Pikas disturbance	<i>F</i>	16.85	20.14	3.68	4.80	12.97	3.21	0.4	4.93	11.58
	<i>P</i>	<0.001	<0.001	0.03	0.012	<0.001	0.05	0.68	0.023	0.009

662 **Table 2.** Two-way ANOVA results of the effect of patches fragmentation and pikas
 663 disturbance on soil compactness, aboveground biomass, belowground biomass, soil hydraulic
 664 conductivity, SOC and TN density.

		Soil compactness	Aboveground biomass	Belowground biomass	Saturated hydraulic conductivity	SOC density	TN density
Patchiness	<i>F</i>	28.10	12.15	7.24	0.75	4.49	10.78
	<i>P</i>	<0.001	0.002	0.023	0.54	0.04	0.003
Pikas disturbance	<i>F</i>	55.86	8.77	11.98	0.42	372.10	69.49
	<i>P</i>	<0.001	0.017	0.002	0.67	<0.001	<0.001

666 **Figure legends**

667 **Figure 1.** An aerial photo of field observation of ecosystem respiration at six surface types: (1)
668 Large bald patch (LP), (2) Medium bald patch (MP), (3) Small bald patch (SP), (4) Intact
669 grassland patch (IG), (5) above pika tunnel (PT) and (6) old Pika pile (PP).

670 **Figure 2.** Ecosystem respiration of different surface types: (1) large bald patch (LP), (2)
671 medium bald patch (MP), (3) small bald patch (SP), (4) intact grassland patch (IG), (5) above
672 pika tunnel (PT) and (6) old pika pile (PP). The upper solid lines, the bottom solid lines, the
673 bold solid horizontal line and the empty dot mean the maximum value, minimum value,
674 median and abnormal value. Letters on the error bars indicate significant differences among
675 different surface types at $P < 0.05$.

676 **Figure 3.** Daily average air temperature and precipitation of the study site in 2016.

677 **Figure 4.** Monthly average soil temperature and soil moisture under different surface types:
678 (1) large bald patch (LP), (2) medium bald patch (MP), (3) small bald patch (SP), (4) intact
679 grassland patch (IG), (5) above pika tunnel (PT) and (6) old pika pile (PP).

680 **Figure 5.** Soil saturated hydraulic conductivity (SHC) under different surface types: (1) large
681 bald patch (LP), (2) medium bald patch (MP), (3) small bald patch (SP), (4) intact grassland
682 patch (IG), (5) above pika tunnel (PT) and (6) old pika pile (PP).

683 **Figure 6.** Soil compactness under different surface types: (1) large bald patch (LP), (2)
684 medium bald patch (MP), (3) small bald patch (SP), (4) intact grassland patch (IG), (5) above
685 pika tunnel (PT) and (6) old pika pile (PP).

686 **Figure 7.** Soil organic carbon (SOC) (a) and total nitrogen (TN) (b) density of different
687 surface types: (1) large bald patch (LP), (2) medium bald patch (MP), (3) small bald patch
688 (SP), (4) intact grassland patch (IG), (5) above pika tunnel (PT) and (6) old pika pile (PP).

689 **Figure 8.** Aboveground biomass (AGB) (a) and belowground biomass (BGB) (b) under
690 different surface types: (1) large bald patch (LP), (2) medium bald patch (MP), (3) small bald
691 patch (SP), (4) intact grassland patch (IG), (5) above pika tunnel (PT) and (6) old pika pile
692 (PP).

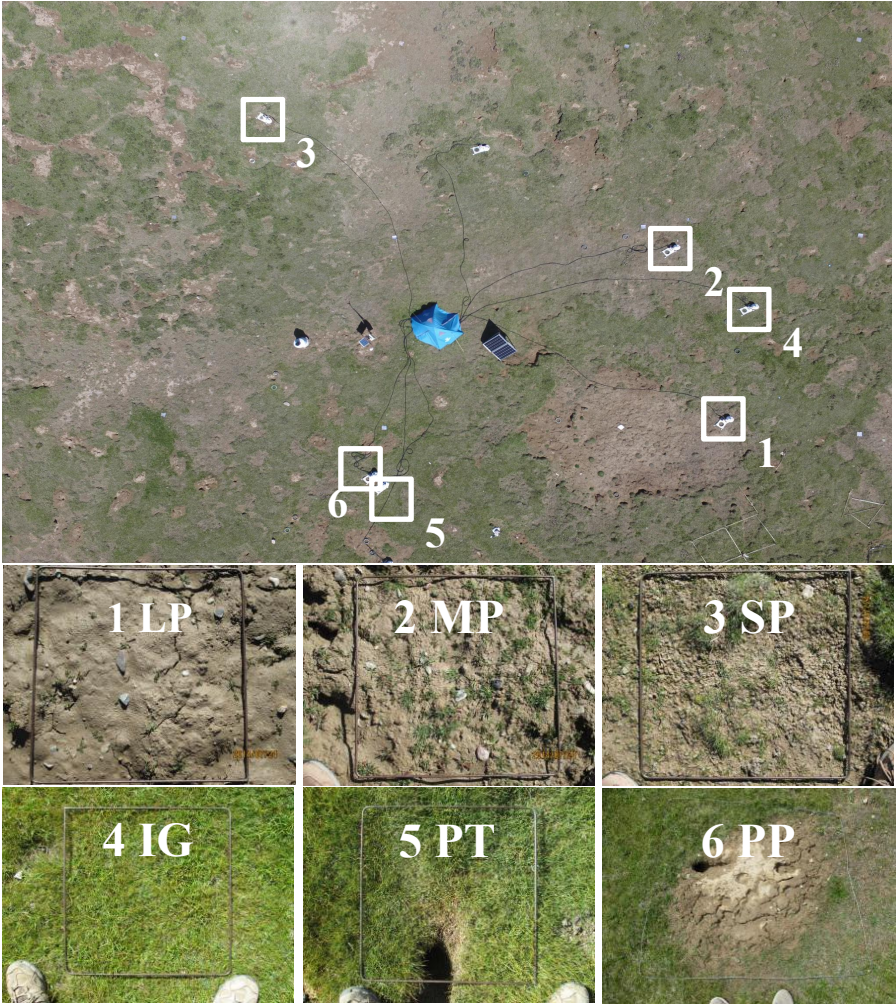
693 **Figure 9.** The correlation coefficient charts between ecosystem respiration (R_e) and biotic
694 and abiotic factors for all six land surfaces. The diagonal line in the figure shows the
695 distributions of the variables themselves. The red line means the frequency distribution of

696 **variables**. The lower triangle (the left bottom of the diagonal) in the figure shows scatter plots
697 of the two properties. The upper triangle (the upper right of the diagonal) in the figure
698 indicates the correlation values of the two parameters; the asterisk indicates the degree of
699 significance (** indicates significant differences at $P < 0.001$, **** indicates significant**
700 **differences at $P < 0.01$, * indicates significant differences at $P < 0.05$**). The bold bigger
701 numbers mean the higher correlation.

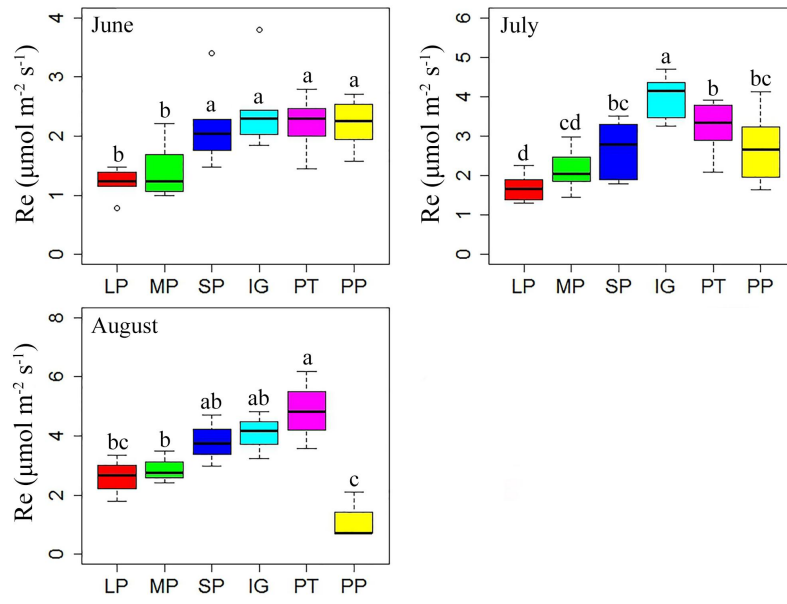
702 **Figure 1.**

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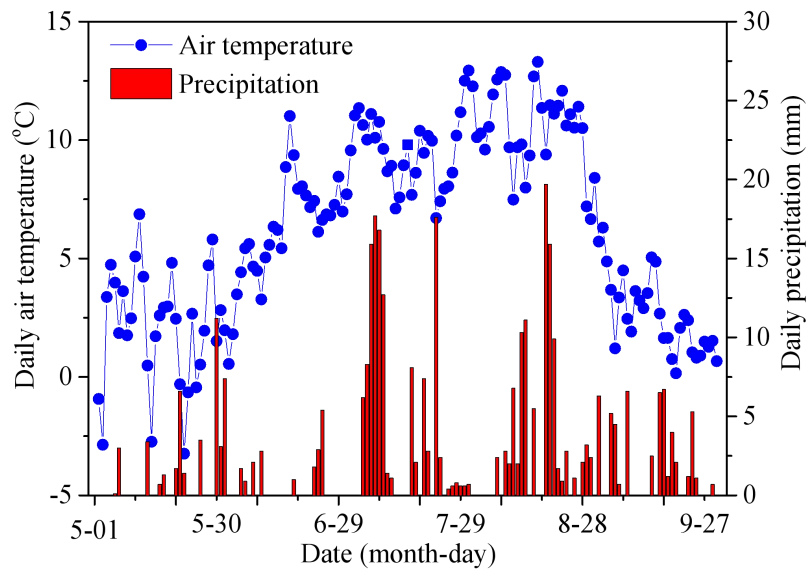
705 **Figure 2.**



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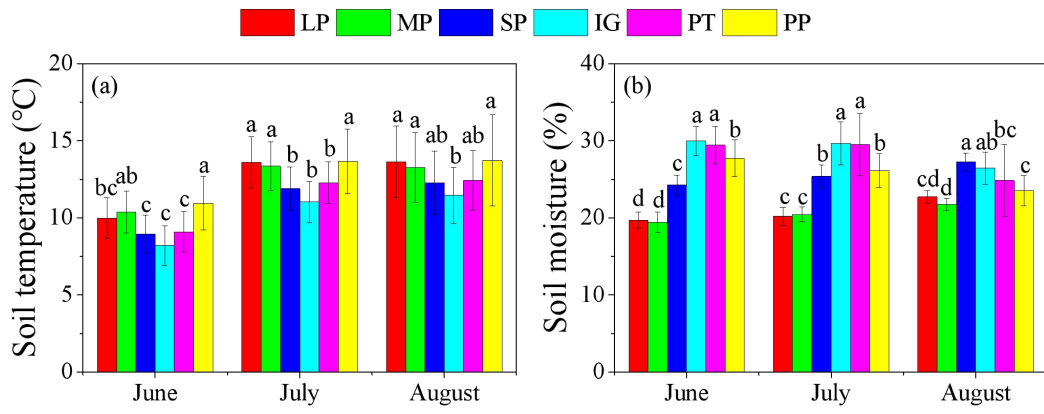
708 **Figure 3.**



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711 **Figure 4.**

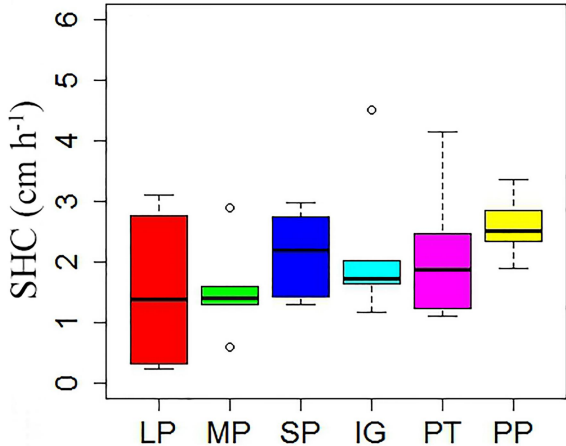


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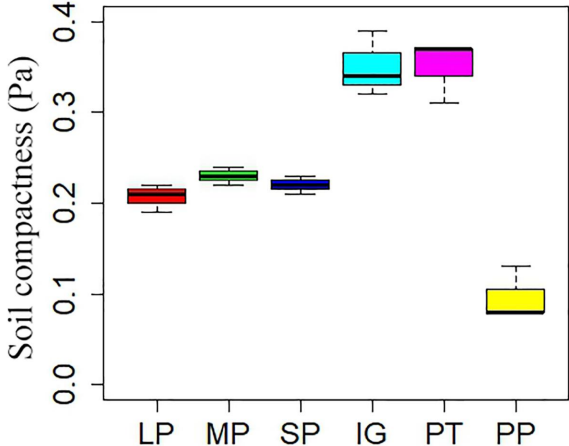
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715 **Figure 5.**



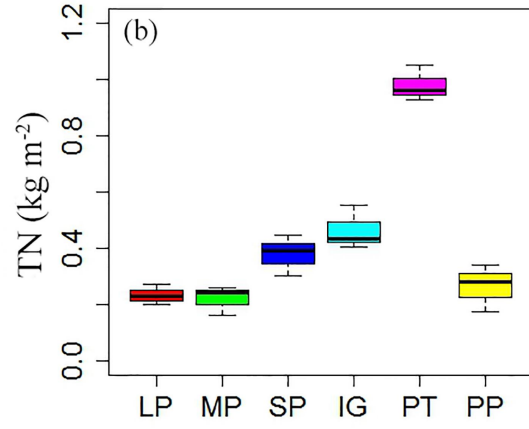
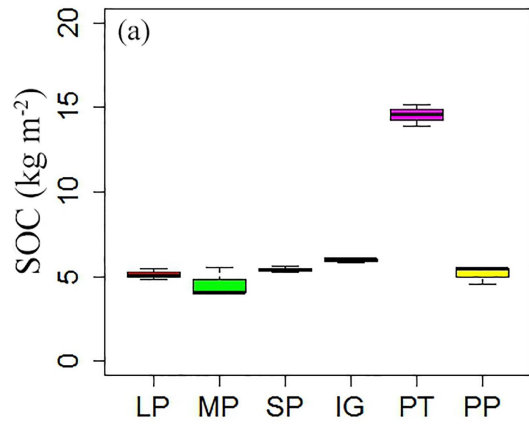
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717 **Figure 6.**



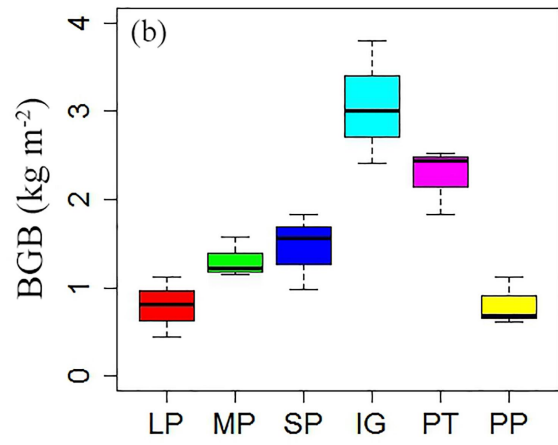
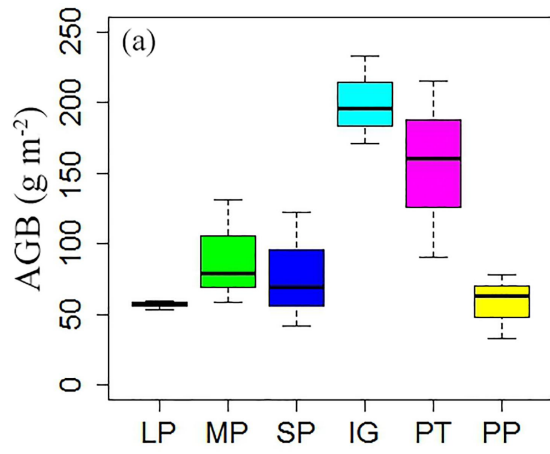
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719 **Figure 7.**



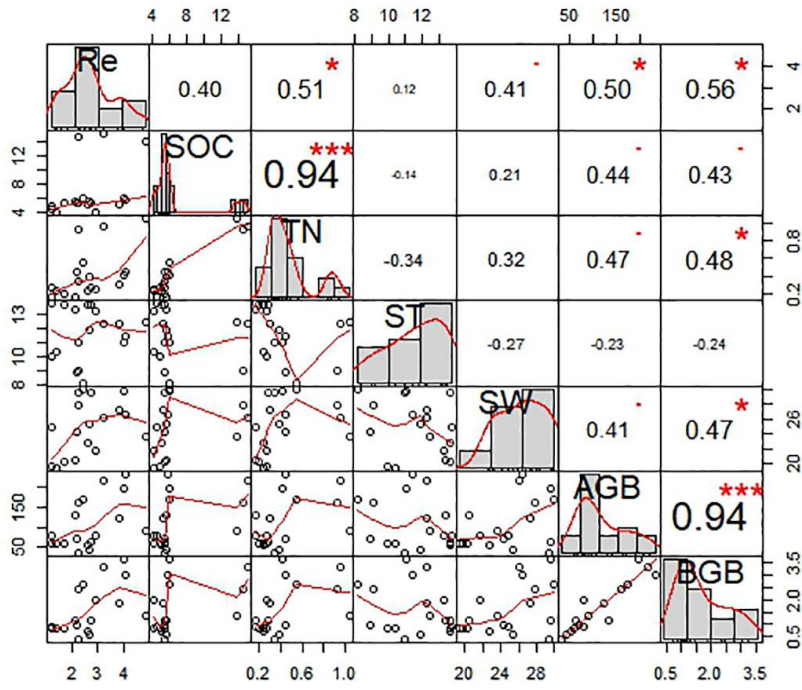
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721 **Figure 8.**



722

723 **Figure 9.**



724