

1 **Impact of elevated precipitation, nitrogen deposition and warming on soil**
2 **respiration in a temperate desert**

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13

14 **Abstract**

15 Soil respiration (R_s) is the most important source of carbon dioxide emissions from soil to
16 atmosphere. However, it is unclear what the interactive response of R_s would be to environmental
17 changes such as elevated precipitation, nitrogen (N) deposition and warming, especially in unique
18 temperate desert ecosystems. To investigate this an *in situ* field experiment was conducted in the
19 Gurbantunggut Desert, northwest China, from September 2014 to October 2016. The results showed
20 that precipitation and N deposition significantly increased R_s , but warming decreased R_s , except in
21 extreme precipitation events, which was mainly through its impact on the variation of soil moisture
22 at 5 cm depth. In addition, the interactive response of R_s to combinations of the factors was much
23 less than that of any single-factor, and the main interaction being a positive effect, except interaction

24 from increased precipitation and high N deposition ($60 \text{ kg N ha}^{-1} \text{ yr}^{-1}$). Although R_s was found to be
25 a unimodal change pattern with the variation of soil moisture, soil temperature and soil $\text{NH}_4^+\text{-N}$
26 content, and it was significantly positively correlated to soil dissolved organic carbon (DOC) and pH,
27 but from a structural equation model found that soil temperature was the most important controlling
28 factor. Those results indicated that R_s was mainly interactively controlled by the soil multi-
29 environmental factors and soil nutrients, and was very sensitive to elevated precipitation, N
30 deposition and warming. But the interactions of multiple factors largely reduced between-year
31 variation of R_s more than any single-factor, suggesting that the carbon cycle in temperate deserts
32 could be profoundly influenced by positive carbon-climate feedbacks.

33 **Key words:** precipitation; nitrogen deposition; warming; soil respiration; temperate desert

34

35 **Highlights**

36 1. Impacts of rainfall, N addition and warming on R_s were studied in a temperate desert.

37 2. Rainfall and N deposition significantly increased R_s , but warming reduced it.

38 3. The interactive response of R_s was much lower than any single-factor.

39 4. Soil temperature was the most important controlling factor for R_s .

40

41 **1. Introduction**

42 Global climate warming, changes in precipitation patterns and increased atmospheric nitrogen
43 (N) deposition have all occurred since the industrial revolution, especially in temperate regions
44 (IPCC, 2013), which will be expected to significantly change soil respiration (R_s) that is the most
45 important source of carbon dioxide (CO_2) from soil to atmosphere (Wu et al., 2011): the annual CO_2

46 flux from R_s was ten-fold that of fossil fuel emissions (Eswaran et al., 1993; Batjes, 1996;
47 Gougoulias et al., 2014). Therefore, even a small change in R_s will profoundly affect greenhouse
48 gas balance and climate (Heimann and Reichstein, 2008). Although a number of experiments of the
49 effects of warming, precipitation, and N deposition on R_s have been conducted in alpine grassland,
50 tundra regions, peatlands and temperate forest (Lafleur and Humphreys, 2008; Strong et al., 2017;
51 Yang et al., 2017; Zhao et al., 2017), studies in temperate desert ecosystems are scarce, especially
52 the impact on R_s of the interactions of these changes. A field study of multi-factor interactive effects
53 on R_s was therefore conducted in a temperate desert ecosystem to help in understanding the response
54 of R_s to climate change and N deposition in future and highlight the main driving factors.

55 R_s includes autotrophic respiration (R_A), which is mainly from plant roots and mycorrhizal
56 activities; and heterotrophic respiration (R_H), which is mainly from the activities of microorganisms
57 (Hanson et al., 2000). Soil moisture is an critical limiting factor for plant roots and microbial
58 activities in desert ecosystems (Huang et al., 2015a): R_s was significantly increased by 47-70% in a
59 degraded steppe in Inner Mongolia, China, by increasing precipitation (Chen et al., 2013), with the
60 effect especially strong in summer (Zhang et al., 2017). In addition, in arid ecosystems, increasing
61 precipitation significantly stimulated plant growth, enhanced soil microbial activity and abundance
62 (Huang et al., 2015a), and changed soil nutrient and substrate concentration, such as dissolved
63 organic carbon (DOC), inorganic nitrogen content, moisture and temperature (Huang et al., 2015b).

64 Warming significantly increased soil temperature, another important controlling factor for
65 plants growth and microbial activity (Sheiket et al., 2011; Huang et al., 2015a). R_s rates were
66 significantly increased in a forest soil and Tibetan Plateau grassland by warming (Chen et al., 2017a),
67 reducing R_s with decreasing soil moisture in the growing season, but increasing R_s in the non-

68 growing season (Fang et al., 2017; Li et al., 2017); no significant impact was observed from
69 warming (Liu et al., 2016a). Therefore, how R_s is affected by warming induced variations in the soil
70 environment is still unclear. In addition, low and short-term N deposition enhanced R_s , while higher
71 and long-term N deposition inhibited R_s due to changes in plant growth and microbial activity (Zhu
72 et al., 2017), but no impacts have also been reported (Luo et al., 2017; Zhang et al., 2017). A meta-
73 analysis showed that the effects of N enrichment on soil CO_2 fluxes depended on temperature and
74 soil properties (Zhong et al., 2016); desert soils may be even more sensitive to its variation.

75 A nation-wide analysis showed that warming, elevated N deposition and precipitation
76 significantly increased R_s in China (Feng et al., 2017). Some studies have shown that the warming
77 effect on R_s mainly depended on the variation of soil moisture in a dry forest soil (Li et al., 2017).
78 Luo et al. (2008), using a modeling analysis, found that interactive effects became increasingly
79 weaker with increasing intensity of the factors, but a recent meta-analysis showed that interactive
80 effects were much greater than single factors (Zhou et al., 2016a). Thus how multi-factor
81 interactions impact R_s is still unclear. Therefore, an *in situ* experiment was carried out in the
82 Gurbantunggut Desert to (1) investigate the single-factor and interactive responses of R_s to warming,
83 precipitation and N deposition, and (2) identify the main controlling factors on R_s .

84

85 **2. Materials and methods**

86 *2.1. Study site*

87 A field experiment was carried out at the southern edge of the Gurbantunggut Desert (44°26'
88 N, 87°54'E and 436.8 m a.s.l.), northwest China, from September 2014 to October 2016. This is the
89 largest fixed/semi-fixed temperate desert in China. The mean annual temperature and precipitation

90 are 7.1°C and 215.6 mm, respectively (Cui et al., 2017), and annual potential evaporation exceeds
91 2000 mm. From late November to mid-March of the following year, a 20–35 cm depth of snow
92 cover the whole desert (equivalent to 38–64 mm rainfall; Huang et al., 2015c). The growing season
93 is from April to October. This desert soil is of extremely low fertility and high alkaline (Cui et al.,
94 2017). Soil organic carbon, total N content, soil NO₃⁻-N, NH₄⁺-N contents and C:N ratio are 2.21
95 ±0.71 g kg⁻¹, 0.08 ± 0.003 g kg⁻¹, 4.49 ± 0.71 mg kg⁻¹, 1.38 ± 0.74 mg kg⁻¹ and 21.39 ± 1.84,
96 respectively (Table 1; Cui et al., 2017). Plant species are dominated by *Haloxylon ammodendron*
97 and *Haloxylon persicum*, and the vegetation was extremely sparse, with only 30% coverage, with
98 some spring ephemeral plants (May–June), some annuals, and perennials herbaceous plants (July–
99 August; Liu et al., 2016). Spring ephemerals account for > 60% of the community cover and 85%
100 of the biomass. Summer ephemerals, annuals and perennials usually account for only a small
101 proportion of the community biomass before June, but dominate the community after the die-back
102 of the spring annuals (Huang et al., 2015c).

103

104 2.2. Experimental treatments

105 A striking N deposition rate (35.2 kg N ha⁻¹ yr⁻¹) has occurred in the Gurbantunggut Desert due
106 to the rapid development of agriculture and industry with main form of ammonium nitrate
107 (NH₄NO₃), and wet (19.6 kg N ha⁻¹ yr⁻¹) and dry (15.6 kg N ha⁻¹ yr⁻¹) deposition are almost half
108 (Song et al., 2015). In addition, according to the forecast of Galloway et al. (2008) that atmospheric
109 N deposition will double from the early 1990s to 2050, and the predictions of Liu et al. (2010) that
110 precipitation in this region would be increased by 30% in next 30 years. In September 2014 to
111 August 2016, an *in situ* complete block interactive experiment was therefore conducted to study the

112 impact of N deposition and increased precipitation on R_s (Experiment 1). The three levels of N
113 deposition ($0 \text{ kg N ha}^{-1} \text{ yr}^{-1}$ (control, N0), $30 \text{ kg N ha}^{-1} \text{ yr}^{-1}$ (low, N1) and $60 \text{ kg N ha}^{-1} \text{ yr}^{-1}$ (high,
114 N2)) and two levels of precipitation ('natural' precipitation (W0) and an increase of 30% (an extra
115 60 mm precipitation annually (W1)) were applied (Cui et al., 2017). Therefore there were six
116 treatments (W0N0, W0N1, W0N2, W1N0, W1N1 and W1N2) with four replicates of each treatment;
117 each replicate plot was $10 \text{ m} \times 10 \text{ m}$ with a 5-m wide buffer zone. The additional precipitation and
118 N deposition (NH_4NO_3) were added twelve times in April, July and September, equivalent to 5 mm
119 precipitation and 2.5 or 5 kg N ha^{-1} per application over a week. The NH_4NO_3 was diluted in 50 L
120 water (equal to 0.5 mm precipitation), and evenly applied following the simulated precipitation. The
121 same amount of water was applied to the control plots (W0N0).

122 Rapidly warming ($0.6 \text{ }^\circ\text{C}$ per decade), increasing precipitation ($3\text{-}5 \text{ mm yr}^{-1}$ since 1979) and
123 receiving high N deposition (3 kg N ha^{-1} since 1980) are affecting the Gurbantunggut Desert (Liu
124 et al., 2013; Li et al., 2015), which would be expected to affect rate of R_s . Therefore, another
125 interactive experiment was established at the same time, simulating the three most likely climate
126 scenarios in the future: (1) warming only (W0N0T1); (2) increased precipitation and N deposition
127 without warming (W1N1T0); (3) the interaction of increasing precipitation, N deposition and
128 warming (W1N1T1); all compared with the current climate (W0N0T0). Therefore, there were four
129 treatments (W0N0T1, W1N1T0, W1N1T1, W0N0T0) with four replicates (plots) of each treatment.
130 Open-top chambers (OTCs) were used to simulate warming. The OTCs were designed with 5 mm
131 transparent tempered glass and stainless steel angle iron to the ITEX standard (Marion et al., 1997).
132 They were 2 m high and 4 m in diameter, with each OTC area being 12 m^2 . However, the design
133 was improved such that the top and bottom OTC areas were the same so that precipitation and

134 snowfall were the same as that to the surrounding environment; this also avoids overheating inside
135 the OTCs. The timings of applications of water and N were the same as in Experiment 1.

136

137 2.3. *Measurements*

138 R_s in all plots were measured twice or thrice a week (continuous measurements over 3 days
139 were made following simulated precipitation and N deposition) using gas chromatography and static
140 chambers (50 cm×50 cm×10 cm) at locations where grow only spring ephemeral plants without any
141 annuals and perennials in order to minimize the between-treatment spatial heterogeneity due to
142 sparse annuals, and perennials (Liu et al., 2012). Gas samples were collected between 10:00-12:00
143 (GMT + 8) throughout the experimental period, which was detected in this period were close to the
144 diurnal averages (Fig.3b and 3d, Fig. 1S). Gas samples were collected from the headspace of each
145 chamber 0, 10, 20 and 30 min after closing the chamber per time. The gas samples analyzed within
146 three days using a gas chromatograph (GC; Agilent 7890A, Agilent Technologies, Santa Clara, CA,
147 USA) equipped with a flame ionization detector for quantitative R_s (Liu et al., 2012). R_s rates were
148 calculated from four concentrations of the gas sample based on a first order differential linear or
149 non-linear equation and were temperature- and pressure- corrected (Liu et al., 2012; Zhang et al.,
150 2014). Soil samples were taken monthly from around the static chambers to a depth of 10 cm using
151 an auger (3.5 cm in diameter). Fine roots and small stones were separated out using a 2 mm sieve.
152 Dissolved organic carbon (DOC) was extracted with deionized water (soil: water ratio = 1:10) by
153 shaking on an orbital shaker at 10000 rpm for 5 min and analyzed using a TOC analyzer (multi N/C
154 3100, Jena, Germany; Jones and Willett, 2006). Brookes' (1985), Chloroform fumigation extraction
155 was used to measure microbial biomass carbon (MBC) and microbial biomass nitrogen (MBN). Soil

156 organic carbon (SOC) were measured using the potassium dichromate method (Jiang et al., 2014),
157 and soil NO₃⁻-N and NH₄⁺-N analyzed as per Yue et al. (2016). Soil pH was measured on a 1:5 soil:
158 deionized water suspension using a pH Meter (Seven Easy, Mettler-Toledo, Switzerland). Caipos
159 Soil and Environment Monitoring Systems (Caipos GmbH, Austria) were used to monitor soil
160 moisture/temperature at 5 and 20 cm depth every hour.

161

162 *2.4. Effects of each treatment on R_s*

163 The each treatment effect was analyzed using the following formula to better evaluating the
164 effect of precipitation, warming and N deposition on R_s (Yue et al., 2016).

$$165 \quad \text{The treatment effect} = (\text{TR}_s - \text{CR}_s) / \text{CR}_s \times 100\%,$$

166 Where the treatment effect is W0N1, W0N2, W1N0, W1N1, W1N2, W1N1T1 or W0N0T1
167 effect on R_s (a positive value shows that the treatment has increased R_s and a negative value shows
168 decrease of R_s), corresponding TR_s represents R_s from the W0N1, W0N2, W1N0, W1N1, W1N2,
169 W1N1T1 or W0N0T1 plots (mg C m⁻² h⁻¹) and CR_s indicates the R_s from the control plots (W0N0,
170 mg C m⁻² h⁻¹).

171

172 *2.5. Statistical analyses*

173 Treatments effect on soil organic carbon (SOC), NO₃⁻-N, NH₄⁺-N content, pH, DOC, MBC
174 and MBN were examined in each treatment by least significant difference LSD (p<0.05). The single-
175 factor and interaction effects of precipitation, warming and N deposition on R_s were detected by
176 multi-way analysis of variance (ANOVA), and the accumulated effect of precipitation, warming and
177 N deposition on R_s were tested by repeated measures ANOVA. In addition, the relationships of R_s

178 and DOC, MBC, MBN, soil temperature, soil moisture, NH_4^+ -N content, soil NO_3^- -N, and pH were
179 described using a linear or non-linear regression model. The factors of key controls on R_s were used
180 to analyze by structural equation models (SEMs). SPSS software (version 20.0) was used to conduct
181 all statistical analyses, and statistical significant differences were set with $P < 0.05$. All Figures were
182 created using the Sigmaplot software package (version 10.0), but SEMs analyses were carried out
183 using AMOS 22.0 (Amos Development Corporation, Chicago, IL, USA).

184

185 **3. Results**

186 *3.1. Treatments effects on soil environmental and properties*

187 Soil temperatures at 5 depth were mostly increased between 11:00 and 22:00 every day by
188 warming; the average annual soil temperatures at 5 and 20 cm depth were significantly increased
189 by 4.41 and 3.67 °C, respectively (Fig. 1a). Soil moisture at 5 cm depth was decreased by warming
190 by only 0.61v/v% (Fig. 1b), and a very small decrease of 0.01v/v% in soil moisture at 20 cm depth
191 was observed (Fig.1b). Soil moisture at 5 and 20 cm depth were largely increased by the increased
192 precipitation (Fig.1b). N deposition and warming significant increased soil NH_4^+ -N and NO_3^- -N
193 contents (Fig. 1c), but no significant change was found from increased precipitation. Soil MBC and
194 MBN were greatly increased by N deposition, but significant negative effects on soil MBC and
195 MBN were observed by warming and the interaction of precipitation and N deposition (Fig. 1d). No
196 significant change in SOC and DOC was observed in any treatment (Fig. 1c and 1d).

197

198 *3.2. Precipitation, warming and N deposition effects on R_s*

199 In our study, a weak R_s emission rate (-2.46 to 50. 26 $\text{mg C m}^{-2} \text{ h}^{-1}$) was observed at control

200 plots with an average emission rate of $12.18 \text{ mg C m}^{-2} \text{ h}^{-1}$ from September 2014 to October 2016
201 (Fig. 2c). Annual cumulative rate of R_s was $1090.11 \pm 450.78 \text{ kg C ha}^{-1}$, with non-growing season
202 account for 20.7% of the annual emission (Table 1). R_s was significantly enhanced by increasing 5-
203 mm precipitation and N deposition from 12.18 to 16.23 and 14.97 $\text{mg C m}^{-2} \text{ h}^{-1}$ (average),
204 respectively ($P < 0.001$; Fig. 2c and 2d; Table 2), with annual R_s increased by 33.1% and 19.2-22.8%,
205 respectively (Table 1). And the low N deposition effect on R_s was much higher than that high N
206 deposition (Fig. 2c and 2d). However, R_s was reduced mostly by warming, although not significant
207 ($P = 0.084$; Table 2). And high temperatures and low humidity at times of peak sunshine during the
208 diurnal variation significantly inhibited its emission rate (Fig 3a and 3b, Fig.2S), but it was also
209 significantly increased by warming following extremely rainfall events that increased soil moisture
210 (Fig. 3c and 3d). The diurnal trend in R_s was consistent with that of soil temperature at 5 cm depth
211 (Fig. 3). In addition, the interactive responses of R_s to increasing precipitation, warming and N
212 deposition were much lower than that from any single-factor (Table 1), and with the interaction of
213 60 kg ha^{-1} N and extra precipitation decreasing R_s by 4.25% (Table 1). Overall, annual R_s rates were
214 significantly impacted by precipitation, N deposition, and their interaction (Table 2), but no
215 significant net change was caused by warming (Table 2), although R_s rates were decreased by 9.99%
216 (Table 1).

217

218 *3.3. Temporal variation and its control*

219 The results of repeated measures ANOVA showed that significantly accumulated effects on R_s
220 were found by N deposition and interaction between N deposition and precipitation or warming
221 rather than alone increasing precipitation and warming (Table 2). A large between-year variation in

222 R_s was observed with a coefficient of variation (CV) up to 41.4% (a much higher R_s rate was
223 observed in 2016 than 2015), but variation was reduced by increasing precipitation, N deposition
224 and warming and their interaction, except with an increase in N deposition of 30 kg ha⁻¹ (Table 1).
225 The results of regression analysis showed that R_s was significantly increased by increases in pH and
226 DOC (Fig. 4e and 4f), but no significant relationships were found with MBC, MBN or NO₃⁻-N
227 content (Fig. 4a, 4b and 4c). In addition, different response characteristics of R_s in the impacts of
228 increased soil moisture, soil temperature and NH₄⁺-N content were found. Soil moisture was the
229 most important controlling factor when it was <4.2 % and soil temperature was >26.5⁰C (Fig. 4g
230 and 4h, Fig. 2S). Secondly, soil temperature was the most important limiting factor when soil
231 moisture was >15.9 % and soil temperature <2.7 °C (Fig. 4g and 4h, 2S). Thirdly, there was no
232 significant impact on R_s when soil NH₄⁺-N content was <6.3 mg N / kg. A significant increase in R_s
233 occurred when soil NH₄⁺-N content was between 6.3 and 12.6 mg N / kg, but R_s was inhibited when
234 soil NH₄⁺-N content was between 12.6 and 31.6 mg N / kg (Fig. 4d).

235

236 **4. Discussion**

237 *4.1. Single-factors impacts of precipitation, N deposition and warming on R_s*

238 Annual R_s was 1090 kg C ha⁻¹ in this temperate desert, with the non-growing season accounting
239 for 20.7 % of the annual flux (Table 1). This is consistent with previous study in here (Zhou et al.,
240 2014; Huang et al., 2015a) because SOC content was very low (Fig. 1c), and vegetation was very
241 sparse in this desert (Liu et al., 2016b). Increasing precipitation significantly increased R_s (Fig. 2c).
242 It is also consistent with the results of a meta-analysis and previous study in here (Huang et al.,
243 2015a ; Liu et al., 2016c). This is because that the growth of desert plants and microbial activity are

244 significantly activated by increasing precipitation (Huang et al., 2015a), and microbial biomass,
245 mass-specific respiration, microbial biomass carbon (MBC) and nitrogen (MBN), and microbial
246 PLFAs were consistently significantly enhanced by increased precipitation (Zhang et al., 2013;
247 Huang et al., 2015a). However, R_s in our study was much higher in moderate soil moisture
248 conditions than with too little or too much soil moisture (Fig. 4g). This suggests that R_s is mainly
249 R_H rather than R_A in this desert, namely from soil microorganism, because (1) too little or too much
250 soil moisture could significantly inhibit microbial activity due to variation of soil temperature and
251 soil properties (Ma et al., 2013), while moderate soil moisture could significantly enhance microbial
252 activity (Skopp et al., 1990), and (2) the biomass of fine roots was no significantly enhanced at our
253 sites by increased precipitation (Cui et al., 2017). This is consistent with results from a desert steppe
254 in northern China where the contribution of R_H (78.1%) was significantly higher than that of R_A
255 (21.9%) under increasing precipitation (Liu et al., 2016a).

256 N deposition also significantly increased R_s , especially in low N deposition (Fig. 2d). This is
257 consistent with results from an alpine meadow and in the Loess Plateau (Fang et al., 2017; Zong et
258 al., 2017), and with a meta-analysis showing that N deposition increased R_s by 8.8% (Zhou et al.,
259 2016a). This is because N deposition, on the one hand, could increase fine root biomass, although
260 this was not significant in our study (Cui et al., 2017); on the other hand, increases microbial activity
261 and abundance by low N deposition (Huang et al., 2015b). But this was inconsistent with a young
262 *Cunninghamialanceolata* forest (Wang et al., 2017), and beneath shrubs of *H. ammodendron*, soil
263 high N content, has the opposite effect in our study site (Chen et al., 2013; Huang et al., 2015b).
264 What's more, the results of nonlinear regression analysis showed that higher R_s rates occurred at
265 moderate soil $\text{NH}_4^+\text{-N}$ contents (between 6.3 and 12.6 mg N / kg), while lower R_s occurred in much

266 lower (<6.3 mg N / kg) or much higher (>12.6 mg N / kg) soil NH₄⁺-N contents (Fig. 4d), but this
267 effect of N deposition on R_s is not consistent with other ecosystems (Burton et al., 2004; Chen et al.,
268 2013; Liu et al., 2015; Chen et al., 2017b). This is because the desert soil is extremely limited than
269 other ecosystem (Adams, 2003), so low N deposition enhanced plants growth and microbial activity,
270 but high N inhibited microbial activity and community composition, reduced R_s (Zhou et al., 2014;
271 Huang et al., 2015b). Overall, soil NH₄⁺-N content was an important controlling factor for R_s
272 because microbial activity, abundance and species diversity were regulated by soil NH₄⁺-N content
273 in this desert, and R_s was very sensitive to variation of N deposition.

274 Warming decreased R_s (Fig. 2f), although not significantly (P=0.084; Table 2), which was
275 consistent with results from a semi-arid alfalfa-pasture of the Loess Plateau (Fang et al., 2017). In
276 addition, a significant decrease in R_s was observed on an extreme drought or hot sunny day when
277 soil moisture was reduced, and sharply reduced R_s when soil temperature reached 37 °C (Fig. 3a
278 and 3b, 1S). This is because: (i) microbial activity is significantly inhibited by extreme temperatures
279 and low soil moisture, may reduce population size by 50-80% (Sheik et al., 2011); (ii) fine root
280 growth is inhibited in high temperature and low soil moisture. Others have noted this phenomenon
281 as occurring at about 16:00 each day (Ma et al., 2013), but in our study the effect was advanced to
282 14:00 by warming, which may reduce carbon emission from soil to atmosphere. However, this is
283 not consistent with results from a tundra ecosystem, subtropical forest or alpine regions where R_s
284 was significantly increased by warming due to the limitation of soil temperature in these ecosystems,
285 and no significant change in soil moisture (Noh et al., 2016; Wu et al., 2016; Zhou et al., 2016b). In
286 addition, a significant increase in R_s was found following enhanced precipitation with warming (Fig.
287 3c and 3d), which indicates that soil moisture was the most important controlling factor for R_s under

288 a warming climate. This is consistent with other studies (Chen et al., 2017a; Zhao et al., 2017).
289 However, statistical analysis showed that no overall significant impact on R_s was found during the
290 experimental period by warming, and it was reduced by 9.99%. This is because our gas samples
291 were taken at 10:00 – 12:00 each day, when average soil temperatures were increased by about one
292 degree. Thus mean annual R_s was not sensitive to temperature changes this small in contrast to the
293 very significant effects of short-term diurnal changes in soil temperature, observed between 12:00
294 and 17:00 (Fig. 3a and 3c). However, gas samples during 10:00-12:00 in this study could catch the
295 mainly warming effects on R_s , except some extremely precipitation and drought events in summer
296 (Fig.1S), which will require further systematic evaluation. Those results indicated that R_s depends
297 mainly on variations of soil moisture and temperature in the context of warming, and climate change
298 is likely to have a very significant effect on temperate deserts.

299

300 *4.2. The interactive effects of precipitation, N deposition and warming on R_s*

301 Interactive responses of R_s were much lower than those of any single-factor, but still increased
302 R_s overall, except interaction between precipitation and high N deposition (Table 1). This is
303 consistent with results in dry ecosystems (Morillas et al., 2015; Martins et al., 2016), but not with
304 the results of a meta-analysis that precipitation and N deposition interactive experiments were a
305 greater extent positive effect on R_s (Zhou et al., 2016a). This can be explained in our study that soil
306 MBC or MBN were much less in interactive treatments than that of single-factor (Fig. 1d), which
307 showed that a number of microorganisms were much less in interactive treatments than that of
308 single-factor due to much stronger N effect. As we found that R_s was reduced with increasing N
309 deposition and precipitation by as much as 4.25% in W1N2 plots (Table 1), which showed that the

310 inhibiting effect of soil NH_4^+ -N content was much stronger when there was sufficient soil moisture
311 (Fig. 2e). This is consistent with the results in a *Populuseuphratica* community in a desert ecosystem
312 (He et al., 2015). This was because (i) microbial activity was inhibited by high or low soil moisture
313 and high soil NH_4^+ -N or NO_3^- -N content (Burton et al., 2004); and (ii) high N tent to reduce
314 extracellular enzyme activity and the fungal population (Maris et al., 2015). In addition, the
315 interactive effect of the three factors on R_s in this desert was much lower than interaction of two
316 factors of precipitation and N deposition (Table 1), and is consistent with the results of modeled
317 interactive effects, which showed that three-factor interactions were rare while two-factor
318 interactions were more common (Luo et al., 2008). Fortunately, the interactive effect of three factors
319 or two factors (precipitation, N deposition and warming) in this desert could largely reduce between-
320 year variation on R_s (Table 2), which may because (i) the limits of soil moisture, soil temperature
321 and soil N content were relieved for key biological processes by increasing precipitation, N
322 deposition and warming (Huang et al., 2015a; Liu et al., 2016b); (ii) various factors antagonistic to
323 each other (Zhou et al., 2016a). However, the variation in the growing season on R_s can be increased
324 by warming, elevated precipitation and N deposition because of their dominant effects on plant
325 growth and microbial activity (Huang et al., 2015b), but it was the exact opposite in the non-growing
326 season due to reduce the limit of temperature (Zeng et al., 2016). Those results showed that R_s would
327 be reduced under interactive effect of increasing rainfall, temperature and N deposition in the future,
328 and took place a positive carbon-climate feedbacks.

329

330 4.3. Temporal variation in treatments on R_s and controlling factors

331 Significantly accumulated effects on R_s were found by elevated N deposition rather than alone

332 increasing precipitation and warming (Table 2). A previous study in here has showed that R_s was
333 decreased to N addition with experimental duration (Zhou et al., 2014), which was inconsistent with
334 our results (Fig. 2d) because in our study relatively lower rate of N addition than that Zhou et al.
335 (2014), and the composition of microbial community and soil propertie were altered gradually by
336 long-term and high N deposition (Fig. 1c and d; Huang et al., 2015b; Zong et al., 2017). In addition,
337 significantly accumulated effects in the interaction between N deposition and precipitation or
338 warming on R_s were also found (Table 2), and R_s was decreased by 4.25% by interaction between
339 increasing precipitation and high N deposition (Table 1), which indicated that the response of R_s to
340 N deposition largely dependent on soil moisture in desert soil. This may be attributed to the
341 antagonistic interaction between elevated N deposition and precipitation on R_h (Zhou et al., 2016a).
342 Those results indicated that N deposition produced strong accumulated effects on R_s in this desert,
343 and was enhanced largely with increasing soil moisture, which would reduce carbon emission from
344 soil to atmosphere.

345 Regression analysis shows that R_s exhibited a unimodal change pattern with variations of soil
346 NH_4^+-N (Fig. 4d), mositure (Fig. 4g), and temperature (Fig. 4h), and R_s was signicantly postively
347 correlated to soil dissloved organic carbon (DOC) and pH (Fig. 4e and 4f). However, structural
348 equation modeling indicated that soil temperature was the most important controlling factor than soil
349 NH_4^+-N and soil moisture (Fig. 5), unsupported our hypothesis, but it is consistent with most
350 research results (Wu et al., 2016; Zhou et al., 2016b; Chen et al., 2017a). In addition, large inter-
351 annual variation was observed (CV = 41.4%) during our experiment (Table 1), while the variation
352 of annual precipitation and air temperature were only 4.41% and 7.78%, respectively (Table 1), but
353 close to the CV of spring root biomass of ephemeral plants (47.14%) with 24 times of aboveground

354 biomass of spring ephemeral plants in 2016 than that in 2015 (Cui et al., 2017), which indicated that
355 the increase of R_s in 2016 was mainly from the root respiration of ephemeral plants. This is
356 consistent with previous study that ephemeral plants mediated inter-annual variation of carbon
357 fluxes in this desert (Huang et al., 2015c; Liu et al., 2016). It is different from other ecosystems
358 where inter-annual variations of R_s were mainly dependent on variations in annual precipitation and
359 air temperature (Gerard et al., 1999; Asensio et al., 2007; Chen et al., 2012). Overall, our results
360 indicate that annual variation in R_s in this temperate desert is mainly controlled by soil temperature,
361 but between-year variation in R_s is mainly controlled by ephemeral plants.

362

363 **5. Conclusion**

364 Climate change and elevated N deposition play important roles in controlling R_s in temperate
365 deserts. We found that increasing precipitation and N deposition significantly increased R_s in the
366 Gurbantunggut Desert, but warming reduced R_s , mostly because of the variation of soil moisture.
367 In addition, we found that the interactive responses of R_s was much lower to precipitation, N
368 deposition and warming than that any single factors. What's more, R_s are mainly mediated by soil
369 moisture, soil temperature and soil $\text{NH}_4^+\text{-N}$ content, but soil temperature are the most important
370 with between-year variation in R_s mainly controlled by ephemeral plants. Those results showed that
371 R_s is very sensitive to increasing precipitation, N deposition and warming, and their interactive
372 effects could reduce soil carbon emissions and so reduce the impacts of climate change.

373

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379

380 **Inferences**

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544 **Fig. 1** Comparative effects of warming by Open Topped Chambers, increased precipitation, and N
545 deposition on soil temperature (a), soil moisture (b) at 5 and 20 cm depth; soil organic carbon (SOC),
546 NH₄⁺-N content, NO₃⁻-N content and pH (c), dissolved organic carbon (DOC), microbial biomass
547 carbon (MBC) and microbial biomass nitrogen (MBN, d). The data are mean ± SE, n = 4 in c and
548 d, different letter indicate significant effect at P < 0.05.

549

550 **Fig. 2** Variation in rainfall (mm, a), and air temperature (°C, a) from September 2014 to October
551 2016 at the Gurbantunggut Desert, and the soil moisture and temperature to increasing precipitation
552 and warming (b), and the response of R_s (mean, n = 4) to precipitation (c), N deposition (d-e) and
553 warming (f). W0 and W1 indicate under ambient precipitation (without water addition) and 60 mm
554 yr⁻¹ precipitation addition; N0, N1, and N2 indicate 0, 30 and 60 kg N ha⁻¹ yr⁻¹ nitrogen addition;
555 while W1N0, W1N1, and W1N2 indicate 0, 30 and 60 kg N ha⁻¹ yr⁻¹ nitrogen addition under 60
556 mm yr⁻¹ precipitation addition; W1N1T1, W0N0T1 and W0N0T0 indicate the interaction between
557 increasing precipitation (60 mm yr⁻¹), N deposition (30 kg N ha⁻¹ yr⁻¹) and warming by OTCs,
558 warming alone (without increasing precipitation and N deposition) and control plots, respectively.
559 Black arrows indicate simulated precipitation (5 mm per time) and N deposition (0.25 or 0.5 g N m⁻²

560 ² per time). Each point represents the mean of four replications (chambers). Standard deviations for
561 R_s are not showed for figure clarity.

562

563 **Fig. 3** Post-rainfall diurnal variation in R_s (mean \pm SE, n = 4, b) with variation in soil temperature
564 and soil moisture (a), and a sunny day variation in R_s (mean \pm SE, n = 4, d) with variation in soil
565 temperature (T5, T20, c) and soil moisture at 5 (W5) or 20 (W20) cm depth caused by warming in
566 open topped chambers (OTCs). Positive values indicate increment by warming, and negative values
567 indicate decline. A red straight line indicates the average value of R_s inside the OTCs in (b) and (d),
568 and a green straight line represents the average value of R_s out of OTCs in (b) and (d). Red *, **
569 and *** indicate significant effect at $P < 0.05$, $P < 0.01$, and $P < 0.001$, respectively.

570

571 **Fig. 4** The relationship of soil respiration with microbial biomass carbon (MBC, a); microbial
572 biomass carbon (MBN, b); soil $\text{NO}_3\text{-N}$ (c); $\text{NH}_4^+\text{-N}$ content (x, d); soil dissolved organic carbon
573 (DOC, e); pH (f); soil moisture (g) and soil temperature (h).

574

575 **Fig. 5** Structure equation modeling (SEM) test the multivariate (soil moisture, soil temperature, soil
576 $\text{NH}_4^+\text{-N}$ content, DOC and pH) effects on R_s (n=34). Single-headed arrows show that the effect of
577 different key controls on R_s were analyzed. The green arrows indicated positive effects, and red
578 arrows showed negative effects. And the width of the arrows indicate the strength of the relationship.
579 The numbers are standardized path coefficients, which can show the importance of the variables in
580 the model. Goodness-of-fit statistics for the model are shown below the model. * indicate significant
581 effect at $P < 0.05$.

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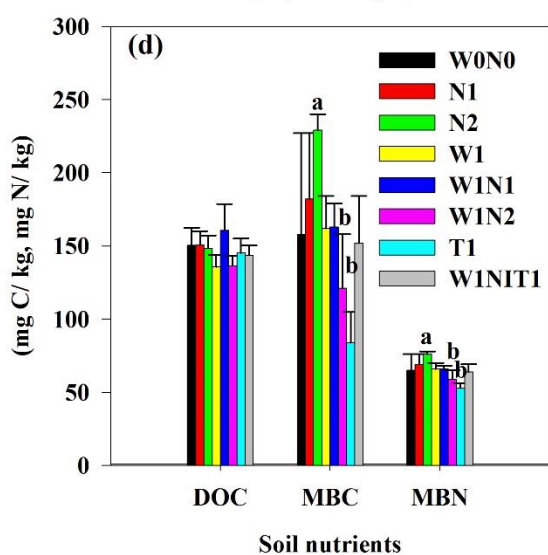
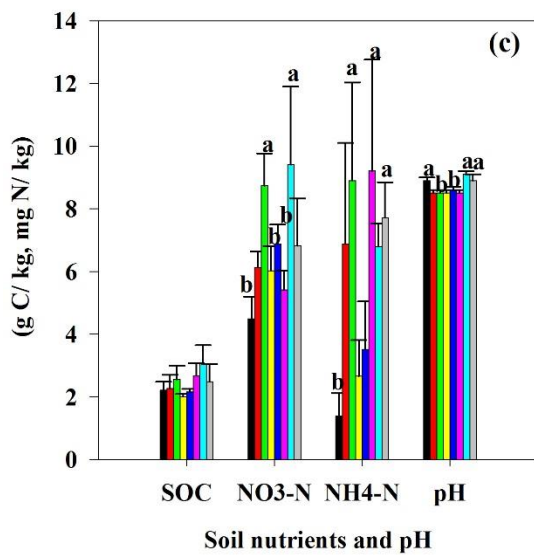
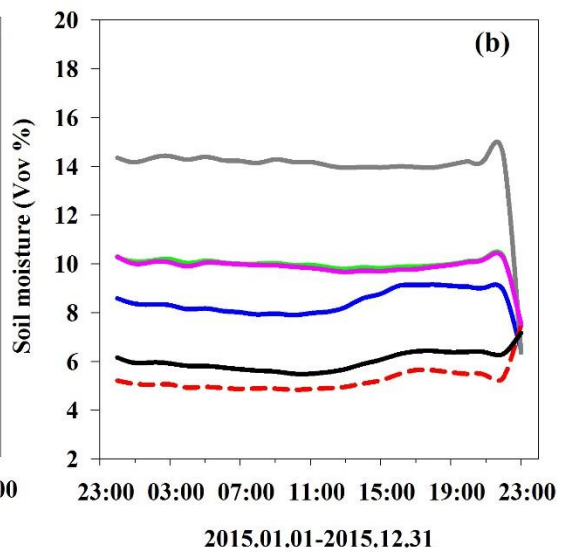
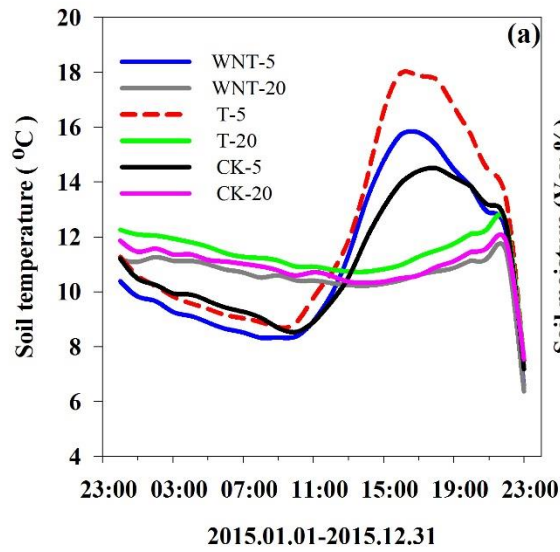
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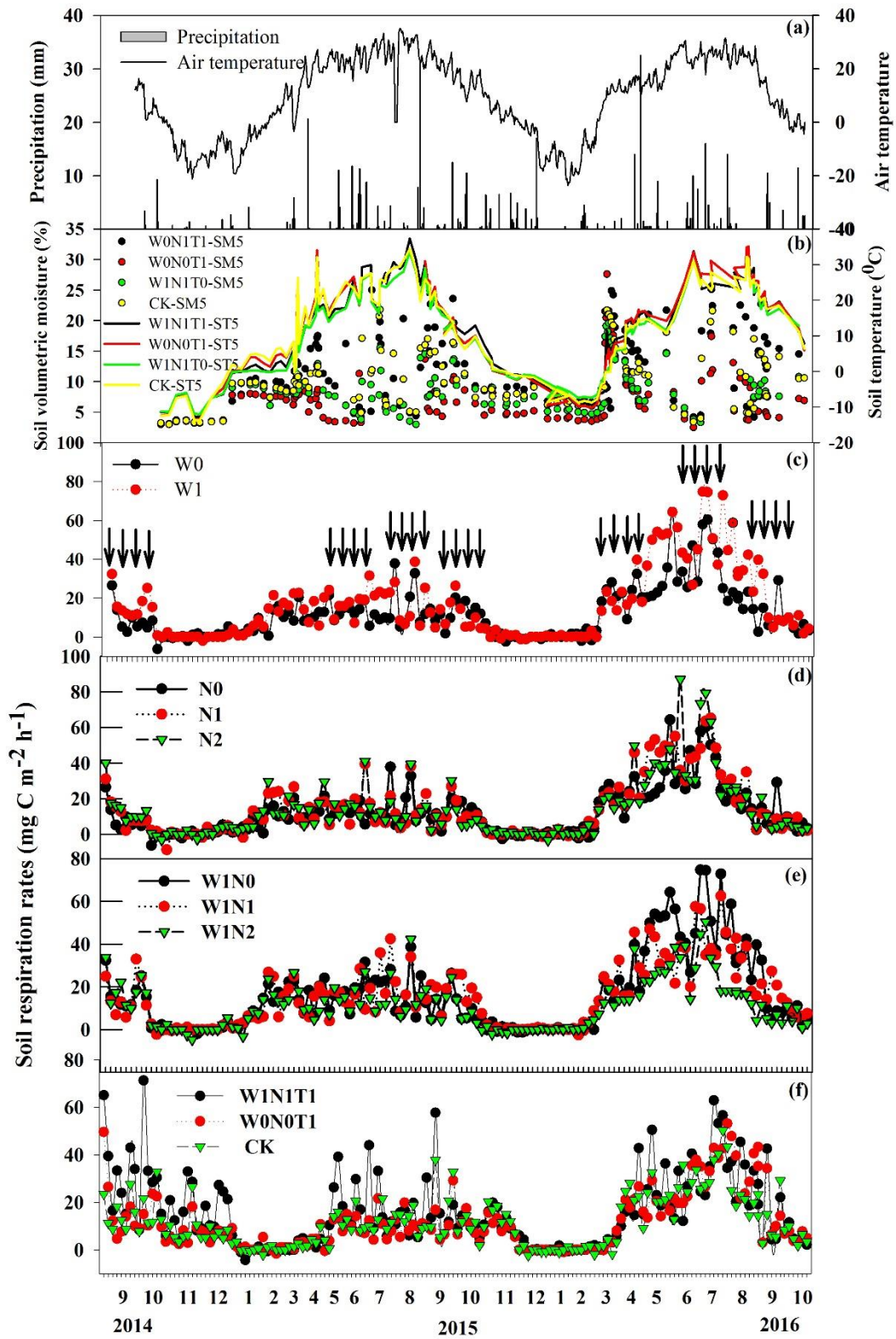
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605 **Fig. 1**



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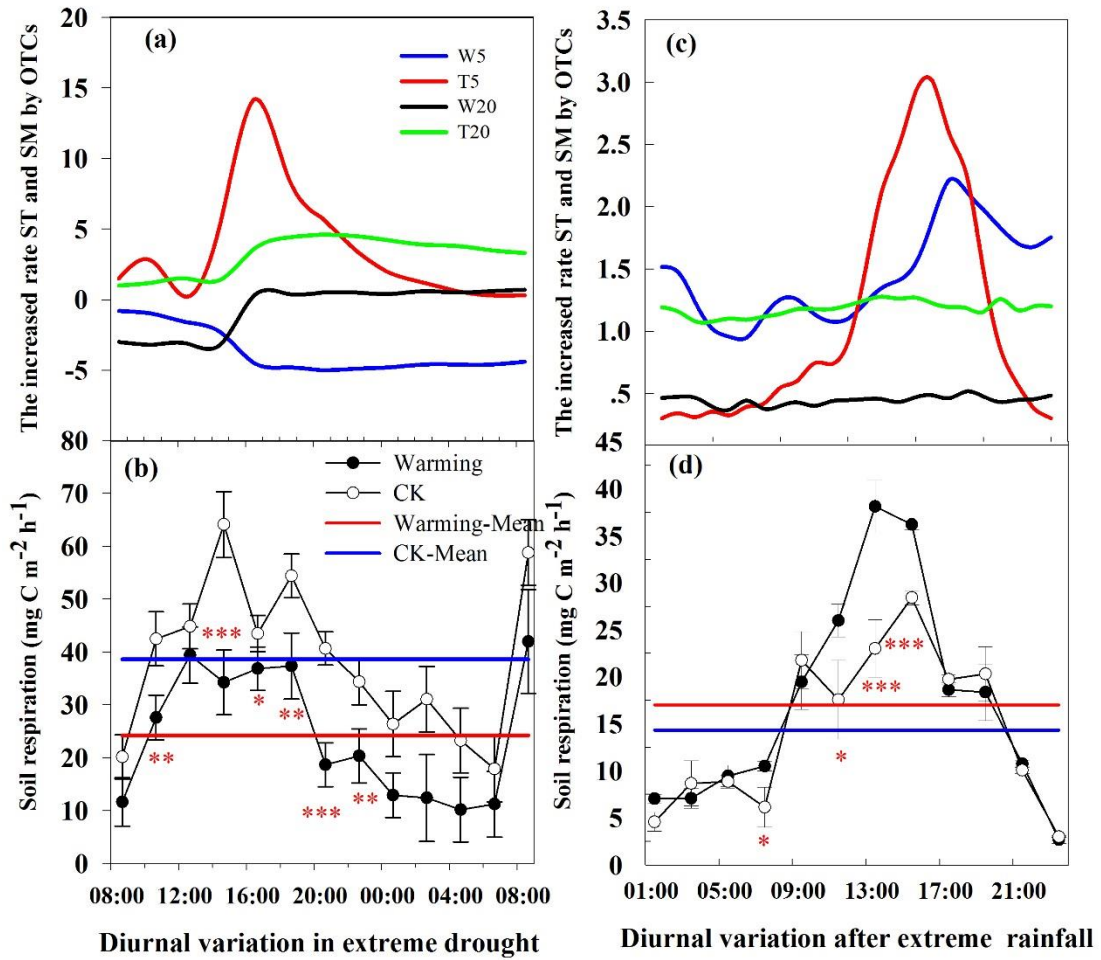
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622 **Fig. 3**

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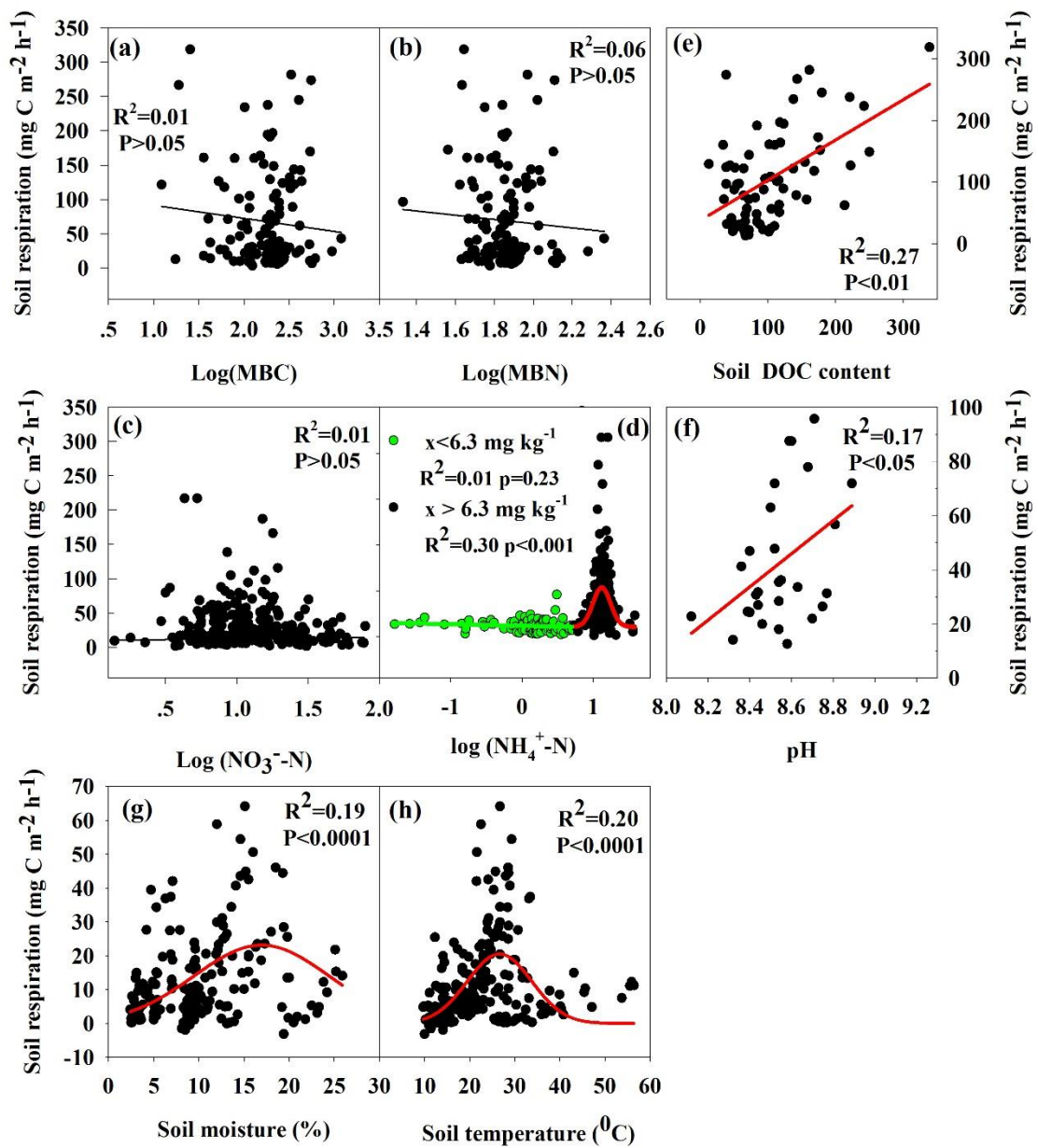
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638 **Fig. 4**



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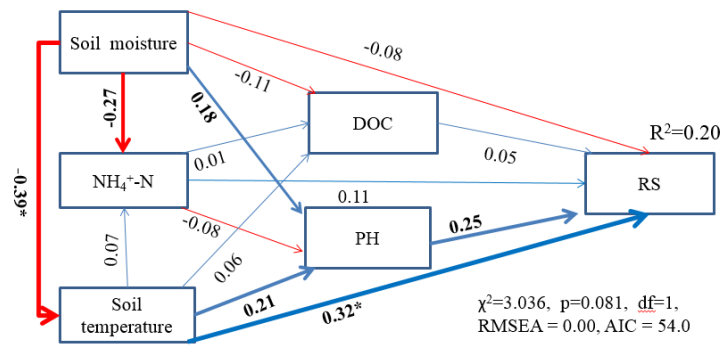
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649 **Fig. 5**



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671 **Table 1** The annual, growing season (GS), non-growing season (NGS), and between-year fluxes

672 and variation of soil respiration (R_s) in September 2014 to September 2016 (mean \pm SE), including
 673 the contribution of GS and NGS, and the treatment effect. The positive values stand for increase R_s ,
 674 and the negative value stand for reduced R_s .

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Treatments	Prec. mm	temp. °C	R_s rate (kg C ha ⁻¹)							
			W0N0	W0N1	W0N2	W1N0	W1N1	W1N2	W1N1T1	T1
Annual	175.75	4.63	1090.11	1338.26	1299.41	1450.78	1304.77	1043.77	1196.84	981.19
	± 7.75	± 0.36	± 450.78	± 599.12	± 537	± 543.70	± 383.29	± 233.23	± 334.31	± 371.34
CV (%)	4.41	7.78	41.35	44.77	41.33	37.48	29.37	22.34	27.93	37.85
Treatments effect (%)	---	---	---	22.76	19.20	33.09	19.69	-4.25	9.79	-9.99
Growing season										
2014.9-2015.8	120.5	14.67	508.30	561.95	570.38	650.66	669.93	562.04	561.10	425.35
2015.9-2016.8	114.5	21.18	1220.27	1546.62	1506.88	1641.62	1274.93	1052.22	1183.86	1089.51
CV (%)	3.61	25.68	46.18	52.03	50.96	48.30	32.79	33.21	50.47	62.00
Non-Growing season										
2014.9-2015.8	47.5	-6.13	131.03	177.09	192.03	256.43	251.55	248.50	301.43	184.49
2015.9-2016.8	69	-11.2	320.62	390.84	329.54	352.85	413.12	224.79	347.29	262.02
CV (%)	26.10	41.37	59.37	53.23	37.29	22.38	34.38	7.09	10.00	24.82
NGS Contribution	---	---	20.65	22.07	21.57	22.98	25.89	24.13	27.10	22.80

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690 **Table 2** Tests of significance of year (Y), warming (T), precipitation (W) and nitrogen addition

691 (N) on soil respiration (R_s) by multivariate ANOVA (F and P values). The accumulated effect of
 692 precipitation, N deposition and warming on R_s in 2015 and 2016 (F and P values) as assessed by
 693 repeated measures ANOVA. *, ** and *** indicate significant effects at $P < 0.05$, 0.01, and 0.001,
 694 respectively.

Three-way ANOVA	n	F	P
Y	2	26.171	<0.001***
N	424	7.709	<0.001***
W	565	17.124	<0.001***
W×N	424	9.392	<0.001***
W×Y	424	6.899	<0.001***
N×Y	424	5.561	0.004**
Y×W×N	424	5.963	0.003**
T	424	2.320	0.084
T×Y	424	0.536	0.464
Repeated measures ANOVA			
Y	2	30.487	<0.000***
N	383	12.887	<0.000***
W	281	2.934	0.087
T	142	0.965	0.326
W×N	281	12.755	<0.000***
T×W×N	281	39.927	<0.000***

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