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Impact Factor: 4.1 · DOI: 10.1016/j.scitotenv.2015.10.022

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# Evident elevation of atmospheric monoterpenes due to degradation-induced species changes in a semi-arid grassland

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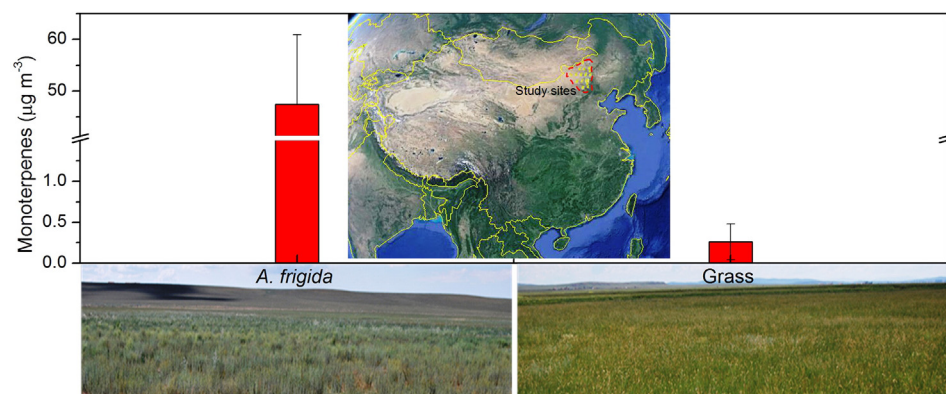
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## HIGHLIGHTS

- Ambient BVOCs concentrations were measured in a degraded semi-arid grassland.
- Degraded grassland may become a significant BVOCs source to the atmosphere.
- Climate change-induced vegetation change alters ambient BVOCs concentration.
- *A. frigida* and sedge emit high monoterpenes and isoprene, respectively, in grassland.

## GRAPHICAL ABSTRACT



## ARTICLE INFO

### Article history:

Received 14 June 2015

Received in revised form 20 August 2015

Accepted 6 October 2015

Available online xxxx

Editor: D. Barcelo

### Keywords:

Monoterpene

Isoprene

Biogenic volatile organic compounds

*Artemisia frigida*

Sedge

Steppe

## ABSTRACT

Biogenic volatile organic compounds (BVOCs) emitted from plants have substantial effects on atmospheric chemistry/physics and feedbacks on ecosystem function. The on-going climate change and anthropogenic disturbance have been confirmed to cause the evident degradation of grassland with shift of plant community, and hence BVOCs emissions were suspected to be altered due to the different BVOCs emission potentials of different species. In this study, we investigated BVOCs concentration above ground surface during growing season in a degraded semi-arid grassland ( $41^{\circ}2' \text{N}$ – $45^{\circ}6' \text{N}$ ,  $113^{\circ}5' \text{E}$ – $117^{\circ}8' \text{E}$ ) in Inner Mongolia. The observed monoterpenes' concentrations varied from  $0.10$  to  $215.78 \mu\text{g m}^{-3}$  ( $34.88 \pm 9.73 \mu\text{g m}^{-3}$  in average) across 41 sites. Compared to non-degraded grassland, concentrations of monoterpenes were about 180 times higher at the sites dominated by subshrub — *Artemisia frigida*, a preponderant species under drought stress and over-grazing. The biomass of *A. frigida* explained 51.39% of the variation of monoterpenes' concentrations.  $\alpha$ -pinene,  $\beta$ -pinene and  $\gamma$ -terpinene dominated in the 10 determined monoterpenes, accounting for  $37.72 \pm 2.98\%$ ,  $14.65 \pm 2.55\%$  and  $10.50 \pm 2.37\%$  of the total monoterpenes concentration, respectively. Low isoprene concentrations ( $\leq 3.25 \mu\text{g m}^{-3}$ ) were found and sedge biomass contributed about 51.76% to their spatial variation.  $\alpha$ -pinene and isoprene emissions at noon were as high as  $515.53 \pm 88.34 \mu\text{g m}^{-2} \text{h}^{-1}$  and

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$7606.19 \pm 1073.94 \mu\text{g m}^{-2} \text{h}^{-1}$  in *A. frigida*- and sedge-dominated areas where their biomass were  $236.90 \text{ g m}^{-2}$  and  $72.37 \text{ g m}^{-2}$ , respectively. Our results suggested that the expansion of *A. frigida* and sedge caused by over-grazing and climatic stresses may increase local ambient BVOCs concentration in grassland.

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## 1. Introduction

Some plants emit substantial biogenic volatile organic compounds (BVOCs), which may significantly impact air quality and biosphere structure/function (Fuentes et al., 2001; Guenther et al., 1995; Lerdau, 2007; Peñuelas, 2008; Peñuelas and Llusà, 2003; Peñuelas and Staudt, 2010; Shallcross and Monks, 2000). It is well known that BVOCs are important for plants in defense against abiotic and biotic stresses (Frost et al., 2008; Peñuelas and Llusà, 2003). Extensive studies suggest that plants with strong BVOCs emission may expand, because they gain more protection by releasing BVOCs in the face of abiotic stressors, like warming, drought, elevated  $\text{CO}_2$  and UV, compared to low and non-BVOCs emitting plants (Lerdau, 2007; Loreto and Schnitzler, 2010; Peñuelas, 2008; Peñuelas and Llusà, 2003; Peñuelas and Staudt, 2010). Thus, the ambient BVOCs concentration may increase, but still lack of evidence, in some climate-sensitive ecosystems where the high BVOCs emission plants may expand due to climate change. Valolahti et al. (2015) recently investigated how long-term warming (13 years of treatment) affects the BVOCs emission in subarctic ecosystems. Other than the direct effect of warming, their results show that climate warming-induced vegetation change is the major factor indirectly increasing the BVOCs emission (Valolahti et al., 2015).

Grassland, as one of the major biome types, covers about one-quarter of earth's land surface. Generally, the dominant herbaceous species in non-degraded grassland emit low BVOCs (Guenther et al., 2006). However, grassland is undergoing significant changes due to climate change and anthropogenic disturbance (Guenther et al., 1995; Lerdau and Slobodkin, 2002). Currently degradation is the most severe ecological problem in the Eurasian grassland – one of the largest contiguous terrestrial biomes (Werger and van Staaldin, 2012). Shrub encroachment has been observed in many native grasslands globally over the past two centuries (Idso, 1992; Liu, 2003; Morgan et al., 2007). The newly dominant species, which adapt to stressed conditions, may substantially impact the community-level BVOCs emission and hence the ambient BVOCs concentration. A study in a semi-arid grassland of Inner Mongolia discovered that subshrub – *A. frigida*, as an overgrazing indicator, is an important monoterpene emitter, whose emission factor based on dry biomass was about 20 times higher than grass (Wang et al., 2012). *A. frigida* is a common subshrub in North American and Eurasian grasslands and its abundance tends to increase globally under elevated  $\text{CO}_2$  concentration and anthropogenic disturbance (Liu, 2003; Morgan et al., 2007; Werger and van Staaldin, 2012). In the Colorado shortgrass steppe in USA, 5-year double  $\text{CO}_2$  enrichment resulted in a 20-fold increase in *A. frigida* coverage (Morgan et al., 2007). Currently more than 60% of grassland in China has been degraded by over-grazing and climate change. Remote sensing data and field investigation (Liu, 2003) have shown that between 1960 and 2000, *A. frigida* coverage increased by 10 times in the Xilinguole region (20% of total area in Inner Mongolia). Therefore, ambient BVOCs concentration in grassland may increase globally.

In this study, we investigated ambient BVOCs concentrations during the growing season in a temperate semi-arid grassland in Inner Mongolia that represents the Eurasian grassland ecosystem. Our aim was to give (1) a first overview on the ambient BVOCs concentrations (isoprene and monoterpene) near ground surface in degraded and non-degraded grasslands, and (2) to assess the effect of species changes and environmental conditions on ambient BVOCs concentration in arid and semi-arid grassland.

## 2. Materials and methods

### 2.1. Site description

The sampling sites were located in a typical semiarid grassland in Xilinguole League of Inner Mongolia ( $41^\circ 35' - 46^\circ 46' \text{N}$ ,  $111^\circ 09' - 119^\circ 58' \text{E}$ ). Xilinguole League covers about  $2 \times 10^5 \text{ km}^2$  including two county-level cities, one county and nine banners. Historically, *Stipa grandis* and *Leymus chinensis* have been the dominant species in this region. However, over-grazing and drought have significantly increased the abundance of *A. frigida*, and *A. frigida* is becoming the dominant species (Liu, 2003). In this study, we focused on the degraded grassland dominated by *A. frigida*. In total, 31 degraded sites (2–4 sites in each county and banner excluding cities) were included, 24 sites dominated by *A. frigida*, 3 sites by *Artemisia sieversiana* and 4 sites by *Artemisia ordosica*. In order to compare with non-degraded grassland, another 10 sites covered by *S. grandis*, *L. chinensis* and/or sedge (mainly *Carex korshinskyi* Kom) were selected. All sites were far away from road and city, and the covering area of dominant species except sedge at each site was larger than 1 ha.

### 2.2. Ambient BVOCs sampling and analysis

Stainless-steel electro-polished canisters (2 L) provided by University of California, Irvine (UCI) were used to sample ambient air at a height of 0.2 m above plant canopy. All canisters were cleaned to  $-50 \text{ mTorr}$  by a vacuum cleaner (Entech 3100A) in lab. Gas samples were collected between 9:00 am and 3:00 pm from 22 July to 8 August 2009. All gas samples were transported to State Key Laboratory of Organic Geochemistry at Guangzhou Institute of Geochemistry, Chinese Academy of Sciences for BVOCs analysis. At each site, we harvested all aboveground plants in a  $0.5 \times 0.5 \text{ m}$  quadrat to measure biomass.

Concentrations of BVOCs were determined by a Model 7100 preconcentrator (Entech Instruments Inc., California, USA) coupled with an Agilent 5973 N gas chromatography-mass selective detector (GC-MSD, Agilent Technologies, USA). Detailed cryogenic concentration steps are described elsewhere (Wang and Wu, 2008; Yi et al., 2007; Zhang et al., 2013). Briefly, 250 mL air inside the canister was first concentrated through a liquid-nitrogen cryogenic trap at  $-160^\circ \text{C}$ . The trapped BVOCs were then transferred by pure helium to a secondary trap at  $-40^\circ \text{C}$  with Tenax-TA as adsorbent. Most of the  $\text{H}_2\text{O}$  and  $\text{CO}_2$  were removed during these two steps. The secondary trap was then heated to get target BVOCs transferred by helium to a third cryo-focus trap at  $-70^\circ \text{C}$ . After the focusing step, the trap was rapidly heated and the BVOCs were transferred to the GC-MSD system. The mixture was first separated by an HP-1 capillary column (HP-1,  $60 \text{ m} \times 0.32 \text{ mm} \times 1.0 \mu\text{m}$ , Agilent Technologies, USA) with helium as carrier gas and determined by MSD. The GC oven temperature was programmed to be initially at  $-50^\circ \text{C}$ , held for 3 min; ramped at  $15^\circ \text{C min}^{-1}$  up to  $10^\circ \text{C}$ ; then raised to  $250^\circ \text{C}$  at  $5^\circ \text{C min}^{-1}$  for 10 min. Triplicate samples were analyzed for each canister. Standard gas provided by Rowland/Blake group in UCI was used to identify and quantify isoprene and monoterpenes. The MSD was used in selected ion monitoring (SIM) mode and the ionization method was electron impacting (EI).

### 2.3. Diurnal variations of BVOCs emission and ambient concentration

Because ambient concentrations of monoterpenes significantly correlated to the biomass of *A. frigida* and isoprene to sedge, we sampled

separately in areas dominated by *A. frigida* and sedge (*C. korshinskyi* Kom) to determined diurnal variations of monoterpene and isoprene emission in summer. Gas emission was determined by static chamber technique (Wang et al., 2012) every 1–3 h at Duolun Restoration Ecology Research Station (42°50' N, 116°18' E) in Inner Mongolia. Our gas chamber contained an upper chamber and pedestal (Supplementary Fig. 1). The upper chamber was built from Teflon film (140  $\mu\text{m}$  thickness) enclosed around stainless-steel-framework (35 cm height, 40 cm diameter). The stainless-steel pedestal (5 cm height) had a gutter around the outside of the upper rim that was filled with water to create a seal with the upper chamber during sampling. Two gas samples were taken at 3–4 min intervals from the chambers by syringe, and the temperature in the chamber was recorded at the start and end of the sampling. The emission rate was calculated from the linear change of concentrations in the chamber headspace as a function of time, base area, chamber volume, and the molar volume of monoterpene or isoprene at the average air temperature in chamber.

Ambient air samples were collected by glass syringe every 20–60 min at a height of 0.2 m above *A. frigida* canopy. Because sedge patches were small (<10  $\text{m}^2$  each) at this site, we used the concentration of the first gas sample at zero minute from our chamber as a rough ambient concentration, which might be overestimated but without wind effect. All gas samples were analyzed within two hours by gas chromatography (GC-4400, East & West Analytical Instruments, Inc., China) with photoionization detector (PID at 10.6 eV). This GC-PID system can directly analyze some BVOCs without pre-concentration and allowed us to do in-situ measurements with high-frequency sampling. In this study, we reliably detected isoprene and  $\alpha$ -pinene. The analysis methods in detail were described by Wang et al. (2012).

#### 2.4. Statistical analyses

One-way ANOVA was used to compare the difference of BVOCs concentration and biomass among sites with different dominant species. The contribution of biotic and abiotic factors to the variability of BVOCs concentration was determined by stepwise multiple linear regression. Standard errors were calculated for means and error bars in figures.  $p$  values under 0.05 and 0.05–0.10 were considered statistically significant and marginally significant, respectively. All the analyses were conducted by SAS software (SAS Institute Inc., Cary, NC, USA).

### 3. Results

#### 3.1. Ambient BVOCs concentration

Across 41 sites in Xilinguole League, all gas samples and biomass were collected within two weeks in the middle of growing season. Air temperature varied between 16.1 and 36.2  $^{\circ}\text{C}$  with mean value of 27.2  $^{\circ}\text{C}$ , and mean PAR was 1125  $\mu\text{mol s}^{-1} \text{m}^{-2}$  ranging from 212 to 1948  $\mu\text{mol s}^{-1} \text{m}^{-2}$  when the samples were collected. Ambient concentration of total monoterpenes and isoprene showed a large variability, especially among sites with different dominant species (Fig. 1). Monoterpenes concentration ranged from 0.10 to 215.78  $\mu\text{g m}^{-3}$  ( $34.88 \pm 9.73 \mu\text{g m}^{-3}$  in average) throughout all sites and was about 180 times higher in *A. frigida*-dominated areas ( $47.41 \pm 13.52 \mu\text{g m}^{-3}$ ) than in areas dominated by healthy grass species (Fig. 1a). In areas with mixed *A. frigida* and *A. sieversiana* populations and mean biomass of  $1712.12 \pm 278.53 \text{ g m}^{-2}$ , monoterpenes concentration was as high as  $88.16 \pm 39.59 \mu\text{g m}^{-3}$  (Fig. 1 a,c). Regression analysis, including air temperature, PAR, biomass of different species, showed that the variability of monoterpenes concentration was mainly explained by *A. frigida* biomass (51.39%) and PAR (4.67%). If we only considered sites (total 8) under conditions of *A. frigida* biomass  $\geq 1000 \text{ g m}^{-2}$  and PAR  $\geq 1000 \mu\text{mol s}^{-1} \text{m}^{-2}$ , monoterpenes concentration reached up to  $137.22 \pm 10.77 \mu\text{g m}^{-3}$ . We mainly determined 10 monoterpene compounds including  $\alpha$ -pinene,  $\beta$ -pinene camphene, (+)-2-carene,

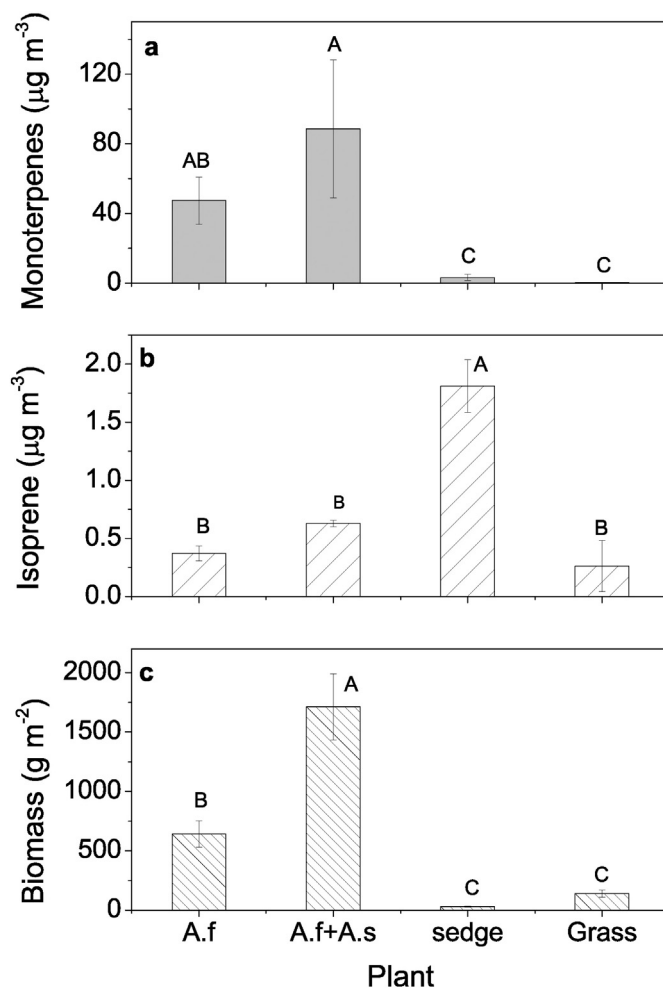


Fig. 1. Average concentrations of total monoterpenes (a) and isoprene (b), and biomass (c) in the sites dominated by *A. frigida*, *A. frigida* + *A. sieversiana*, sedge and grass separately. Different letters indicated significant difference among sites.

$\beta$ -myrcene,  $\alpha$ -phellandrene, ocimene,  $\delta$ -limonene,  $\gamma$ -terpinene and terpinolene.  $\alpha$ -pinene,  $\beta$ -pinene and  $\gamma$ -terpinene dominated the ambient concentration of total monoterpenes and accounted for  $37.72 \pm 2.98\%$ ,  $14.65 \pm 2.55\%$  and  $10.50 \pm 2.37\%$ , respectively (Fig. 2). When total monoterpenes concentration was lower than  $2 \mu\text{g m}^{-3}$ , terpinolene's contribution increased to  $22.38 \pm 2.84\%$  (Fig. 2). Compared to monoterpenes, isoprene concentration was lower ( $\leq 3.25 \mu\text{g m}^{-3}$ ). Mean value of isoprene concentration in sedge-dominated areas (generally  $< 900 \text{ m}^2$ ) was  $1.81 \pm 0.23 \mu\text{g m}^{-3}$ , much higher than in other areas (Fig. 1b). Considering all sites, sedge biomass contributed about 51.76% to the variation of isoprene concentration.

#### 3.2. Diurnal variations of BVOCs emission and their ambient concentrations

Ambient concentrations of both  $\alpha$ -pinene and isoprene showed similar diurnal variation, which spiked around noon and varied with PAR (Figs. 3a, 4). The highest ambient concentrations of  $\alpha$ -pinene and isoprene at noon were as high as  $281.94 \mu\text{g m}^{-3}$  and  $120.11 \mu\text{g m}^{-3}$  when PAR were  $1806 \mu\text{mol s}^{-1} \text{m}^{-2}$  and  $1506 \mu\text{mol s}^{-1} \text{m}^{-2}$ , respectively. PAR explained 80.22% and 80.75% of the diurnal variation of  $\alpha$ -pinene and isoprene concentrations, respectively.  $\alpha$ -pinene and isoprene emission rates also had the same diurnal variation as their ambient concentration and were controlled by PAR (Figs. 3, 4). At noon  $\alpha$ -pinene and isoprene emission rates reached up to  $515.53 \pm$



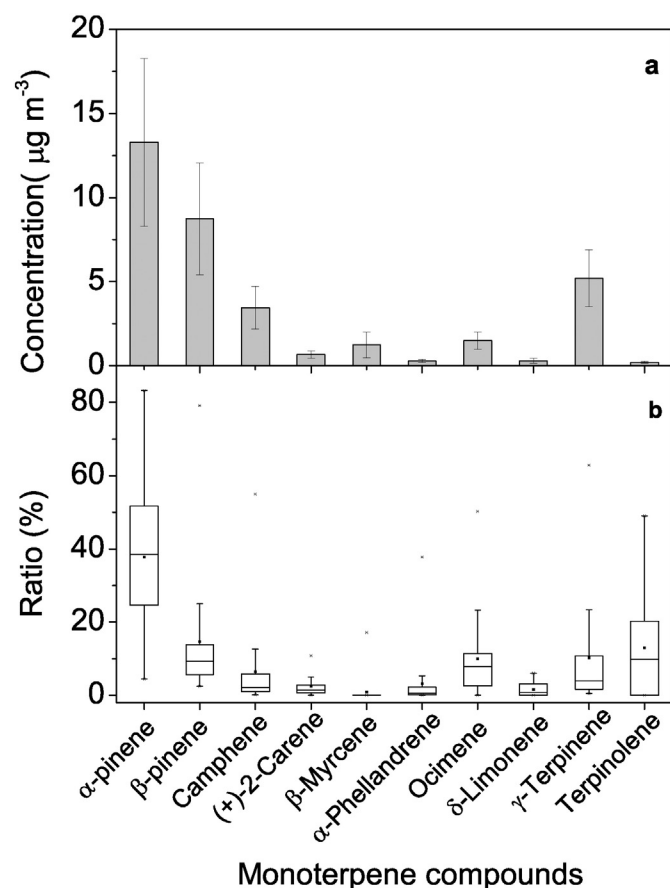


Fig. 2. Concentration of each monoterpene species (a) and the contribution of each monoterpene to the total monoterpene concentration (b) across 41 sites in Xilinguole League.

$88.34 \mu\text{g m}^{-2} \text{h}^{-1}$  and  $7606.19 \pm 1073.9 \mu\text{g m}^{-2} \text{h}^{-1}$  in *A. frigida* and sedge dominant areas where their biomass were  $236.90 \text{ g m}^{-2}$  and  $72.37 \text{ g m}^{-2}$ , respectively.

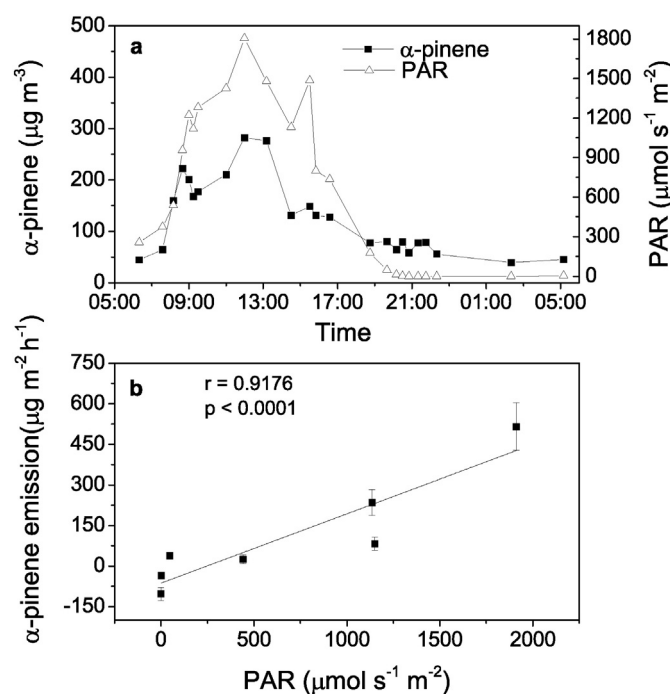


Fig. 3. Diurnal variations in ambient  $\alpha$ -pinene concentration (a) and the relationship between  $\alpha$ -pinene emission and PAR (b).

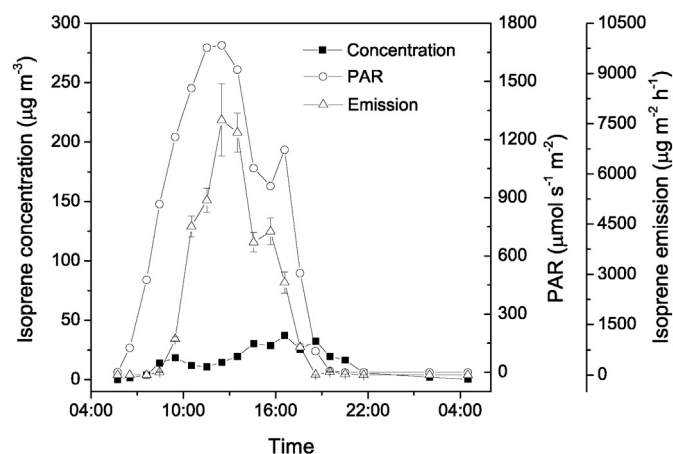


Fig. 4. Diurnal variations in isoprene emission, ambient isoprene concentration and PAR.

#### 4. Discussions

##### 4.1. High ambient monoterpenes concentration in *A. frigida*-dominated grassland

As expected, degradation-induced species changes significantly increased ambient BVOCs concentration in the semi-arid grassland. Compared to non-degraded grass areas, concentrations of monoterpenes were about 180 times higher at the sites dominated by subshrub – *A. frigida*, a preponderant species under drought stress and over-grazing in Inner Mongolia. In *A. frigida*-dominated areas with biomass  $\geq 1000 \text{ g m}^{-2}$ , the concentration of monoterpenes ranged from  $94.5$  to  $186.1 \mu\text{g m}^{-3}$  in hot and sunny days (Temperature  $\geq 25^\circ\text{C}$  and  $\text{PAR} \geq 1000 \mu\text{mol s}^{-1} \text{m}^{-2}$ ). Such high concentrations were at least an order of magnitude higher than reported maximum values in most other studies in forest ecosystems (Clement et al., 1990; Hakola et al., 2012; Hakola et al., 2003; Harrison et al., 2001; Noe et al., 2012), although the biomass in grassland is much lower than tree biomass in forest. The possible reason for such high concentration is that *A. frigida* contains high amounts of essential oil (1.5% w/w dry basis), in which monoterpenes and oxygenated monoterpenes accounted for 10.4 and 75.6%, respectively (Lopes-Lutz et al., 2008). Based on the diurnal pattern of monoterpene emission, the emission from *A. frigida* is light-dependent (Fig. 3b), similar to some species such as oak and beech (Dindorf et al., 2006; Staudt and Seufert, 1995) but different from most coniferous trees (Hakola et al., 2000; Juuti et al., 1990; Noe et al., 2012; Tarvainen et al., 2005). Thus, strong sun light ( $1125 \mu\text{mol s}^{-1} \text{m}^{-2}$  in average) during our measurements in the grassland might be another reason for the observed high concentrations. Therefore, the increase of *A. frigida* resulting from over-grazing and climate change in grassland may raise ambient BVOCs concentration by more than 100 times, and also possibly results in increasingly detected odor if environmental stresses continue to increase (Peñuelas, 2008; Peñuelas and Staudt, 2010). In the grassland ecosystems where NO<sub>x</sub> levels are relatively low, the high ambient BVOCs concentration may significantly affect the atmospheric chemistry via reacting with the OH radicals, which will increase the lifetime of CH<sub>4</sub>, an important greenhouse gas (Di Carlo et al., 2004).

##### 4.2. Grassland might be a potential isoprene source in future

Isoprene concentrations we observed in Inner Mongolia were similar to reported values ranging from  $0.1$  to  $7.5 \mu\text{g m}^{-3}$  in boreal forest (Lappalainen et al., 2009; Noe et al., 2012). Based on the regression analysis, sedge was the main contributor, although only small sedge patches existed in our study areas. Bai et al. (2006) also found that isoprene emission from grassland in Inner Mongolia mainly came from sedge

(Bai et al., 2006). Different from ambient samples collected by canisters, we found much higher isoprene concentration roughly at zero minute in the chamber. This might be caused by high isoprene emission that can increase isoprene concentration from 0 to  $100 \mu\text{g m}^{-3}$  within 20 s based on the emission rate. Recently, growing evidence showed that isoprene emission from sedge in high-latitude wetlands was unexpectedly high (Ekberg et al., 2009; Faubert et al., 2010; Holst et al., 2010; Patrick et al., 2011; Tiiva et al., 2007a, 2007b) and that nitrogen deposition significantly and quickly increased sedge coverage in grassland (Bassin et al., 2007; Bowman et al., 2008) because sedge grows well in acidic soil. Moreover, our diurnal observation showed that sedge (*C. korshinskyi* Kom) in semi-arid grassland emitted isoprene at a high rate ( $>9 \text{ mg m}^{-2} \text{ h}^{-1}$ ) compared to boreal wetlands, possibly due to drought and high temperature ( $>25^\circ\text{C}$ ). This indicates that drought and warming might significantly increase isoprene emission from sedge-dominated, high-latitude meadows in the future. Therefore, grassland may be also a potential isoprene source under climate change.

Supplementary data to this article can be found online at <http://dx.doi.org/10.1016/j.scitotenv.2015.10.022>.

## Acknowledgments

We thank J. Meng for her help in the field measurement. We also thank K. M. Yang and A. B. Huang in East & West Analytical Instruments for the technical support in GC analysis. This study was funded by the National Natural Science Foundation of China (20907071/41025012), State Key Laboratory of Organic Geochemistry (OGL-200801) and Youth Innovation Promotion Association of Chinese Academy of Sciences. H. Wang and X. Wang contributed equally to this work.

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