DEPENDENCE OF CO₂ FLUX ON THE KEY ABIOTIC AND BIOTIC PARAMETERS IN SEMI-NATURAL GRASSLANDS EITHER TRADITIONALLY GRAZED OR EXCLUDED FROM GRAZING


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(Received 31st Mar 2016; accepted 26th Jan 2017)

Abstract. Traditional farming has been ceased in large areas regarding several Central-European countries during the last decades. Nevertheless, only minimal knowledge is available about the impacts of these land use changes on the fluxes of grasslands. Within the framework of this research the dependence of stand level CO₂ flux (NEE) on temperature, light (PPFD), leaf area (LAI), evapotranspiration and soil moisture has been examined in a dry sandy pasture grazed by grey cattle for decades. The examinations have been completed both in a grazed area and in a 6 hectare patch excluded from grazing. Measurements have been taken for 3 years during the vegetation periods in the area of the Kiskunság National Park by a mobile Plexiglas chamber as well as infrared gas analysers. Among the examined abiotic and biotic parameters the dependence of NEE on PPFD and LAI has been significant in both areas. Exponential and linear correlation turned out to be the strongest in both grassland types between CO₂ fixation and light dependence and CO₂ fixation and leaf area, respectively. The strongest correlation has been observed between the PPFD and NEE in both types of grassland. The dependence of CO₂ flux on light and leaf area has been stronger in the extensively grazed area compared to the other examined grass excluded from grazing. Based on this fact it can be assumed that the conversion of land use in traditionally grazed grasslands has negative influence on the relation among NEE and the key abiotic and biotic parameters. It can be explained, beside the accumulation of litter, by the altered vegetation dynamic processes. Extreme dry periods effect, in a negative way, the dependence of CO₂ flux on temperature, soil moisture and evapotranspiration. It calls the attention to the impacts of weather extremities on the mechanisms in semi-arid sandy grasslands.

Keywords: canopy chamber; grazing exclusion, in situ experiment; net ecosystem CO₂ exchange; semi-arid grassland

Abbreviations: EC – eddy covariance; ET – evapotranspiration; LAI – leaf area index; NEE – net ecosystem CO₂ exchange; PPFD – photosynthetically active photon flux density; SWC – soil water content; T_air – air temperature

Introduction

Grasslands constitute one of the largest ecosystems of the temperate zone covering approximately 20% of Europe (Soussana et al., 2007). Similarly in the Carpathian Basin extended areas are covered by semi-natural grass communities (Czóbel et al., 2010), including dry grasslands. Our knowledge on the mechanisms of grasslands, their carbon balance and greenhouse gas fluxes has been significantly enlarged on both European
level (Soussana et al., 2007) and that of the Pannonian biogeographical region (e.g. Nagy et al., 2007; Czóbel et al., 2010; Horváth et al., 2010). One part of the studies published so far applied either large scale methods (eddy covariance, NDVI) or modelling (Barcza, 2011). Due to their nature these processes do not provide adequate information on smaller scale changes.

The chamber method has been used extensively for measuring gas exchange between soil surfaces and the atmosphere (Delle Vedove et al., 2007), and has the advantages of low cost and ease of use. Besides measuring soil respiration, it can also be applied for determining fluxes of CO₂, CH₄ and N₂O. Transparent chambers are used above short canopy, for measurements of the net flux including the effect of photosynthesis and soil exhalation (Farkas et al., 2011).

Different aspects of global climate change are likely to affect semi-natural and managed terrestrial ecosystems (Frank, 2007). Weather anomalies can have negative effects on the functioning of grasses by decreasing their production and carbon uptake, especially the summer drought and heat (e.g. Ciais et al., 2005). Furthermore, it is also known that drought stress significantly decreases mycorrhizal colonization (e.g. Nilsen et al., 1998; Lutgen et al., 2003).

Several publications discuss the dependence of NEE on light, temperature and LAI; however, the majority of these studies examine only one key ecological variable. Moreover, surprisingly low amount of publications focus on the examination of connections between the CO₂ flux of grasslands managed in an altered way and the main biotic and abiotic factors.

The correlation between carbon fixation and the main ecological variables has been examined in a northern temperate grassland for three growing seasons by using the eddy covariance technique. Change in soil moisture was the most important ecological factor controlling C gain in the studied grassland ecosystem. Soil moisture content was positively correlated with leaf area index (LAI). Gross photosynthesis was strongly correlated with changes in LAI (Flanagan et al., 2002). High correlation between the actual LAI and net primer production has been observed in many cases (e.g. Gilmanov, 2005) even when including interannual differences (Wylie et al., 2003). In grasslands much of variation in NEE is constrained by amount of precipitation (Flanagan et al., 2002), however responses of the ecosystem carbon exchange to climatic factors may change according to the phenological stage (Yuste et al., 2004).

The Eastern and Central European countries, changed their agricultural land use when they switched from a state to a market economy. Since 1989 livestock numbers have generally decreased by more than 50% up to 70%. Some of the grasslands were even completely abandoned from agricultural use (Isselstein et al., 2005). Nevertheless, only minimal knowledge is available about the impacts of these land use changes on the mechanisms and fluxes of grasslands.

Several publications discussed the consequences of ceasing grazing activities; however these studies mainly examined the changes occurring in the species composition, structure, production of vegetation and in the C and N content of the soil following the abandonment of intensively grazed grasslands (e.g. Osem et al., 2004; Su et al., 2005). In case of CO₂ flux examinations typically leaf-level measurements have been made. These latter studies showed the higher net photosynthesis and transpiration of species excluded from grazing (Chen et al., 2005).

A recent publication discussed the effects of grazing exclusion on net ecosystem CO₂ exchange and net primary production in Bugac dry sandy grassland (Czóbel et al.,
2012). EC measurements implemented in the past decade justify that – apart from the extremely arid years – this area is to be considered as a net sink for carbon (Nagy et al., 2007). As a result of the grazing exclusion the CO$_2$ uptake potential of the non-grazed stand increased by 13% compared to the grazed stand. It was more significant in the extreme dry year, however, in wet year slightly lower average carbon sequestration was detected at the non-grazed stand, than that of the grazed area. Significant carbon sequestration potential was only detected during wet periods in both stands. Grazing exclusion had no significant effect on the NEE flux as inferred from chamber measurements results (Czóbel et al., 2012).

The grasslands ecosystems of the Carpathian Basin are particularly vulnerable to current and forecasted changes in land use, especially due to global climate changes. However, the possible structural and functional responses of this ecosystem to different management practices is poorly known (Czóbel et al., 2010, 2013).

The aim of our study was to investigate the dependence of CO$_2$ flux on selected key abiotic and biotic parameters in semi-natural grasslands either traditionally grazed or excluded from grazing. The three-year-long study was carried out within the frame of the GreenGrass project (Soussana et al., 2007).

**Materials and Methods**

*Site description and grazing exclusion*

This site is situated at Bugacpuszta (46°41’N, 19°36’E) in the Hungarian Great Plain. The grassland is part of the Kiskunság National Park and has been under extensive management for the last 20 years (Nagy et al., 2007). The soil is a Chernozem sandy soil. In the first 30 cm layer the share of soil fractions are: sand : silt : clay 0–10 cm: 79% : 8% : 13%; 10–20 cm: 85% : 6% : 9%; 30–50 cm: 93% : 3% : 4% (Horváth et al., 2010). The vegetation is semi-arid sandy grassland dominated by *Festuca ovina* ssp. *pseudovina*, *Carex stenophylla* and *Salvia pratensis* (Czóbel et al., 2010).

Of the large (10.000 ha) territory of the dry sandy grassland subject to extensive grazing with Hungarian Grey cattle for decades and displaying only minor (< 3 m) terrain differences, an area of some 6 ha, taking the shape of a semicircle and separated with electric fence, was selected to represent grazing exclusion. During the grazing season (from 1 May to 31 October), the average grazing rate varied between 0.53 and 0.75 LSU/ha in the control area – adjacent to the fenced off area – subject to extensive grazing with Hungarian Grey cattle (Czóbel et al., 2012).

*Instrumentation*

Net ecosystem gas exchange rates of CO$_2$ (NEE) were measured episodically on the grazed and non-grazed part of the grassland during the vegetative period using chamber technique and a portable infrared gas-analyser (*CIRAS-2*, PP Systems, Hitchin, UK) operated in open system mode. The photosynthetic system was connected to a water clean, portable, non-destructive, self-developed chamber (d=60 cm, made from plexiglass) taking air samples from the connecting (inner and outer) tubes (Czóbel et al., 2004). Carbon dioxide exchange rate has been calculated from the differences or changes in CO$_2$ concentrations (for more details, see Czóbel et al., 2004, 2005). Stand level chamber measurements were carried out in clear and sunny days between 10:00 and 16:00 in order to avoid the unsteady meteorological paramereters affecting the NEE.
values. On a peculiar NEE measurement day the carbon flux of 3 grazed and non-grazed, randomly selected plots were measured alternately for an average of 60 min per plot. The mean and standard deviation of the data collected were calculated for each treatment type (grazed and grazing excluded). Gas exchange rates have been calculated from the rate of CO$_2$ / water vapour concentration change.

Photosynthetically active photon flux density (PPFD), air temperature ($T_{air}$) and evapotranspiration (ET) data were measured by CIRAS-2 (PP Systems, Hitchin, UK) sensors parallel with the carbon flux data. A TDR reflectometer (ML2, Delta-T Devices Co., Cambridge, UK) was used to detect the volumetric soil water content (SWC) at a soil depth of $-0.05$ m. An infrared normalized difference vegetation index camera (ADC, Dycam Inc., Chatsworth, USA) and the Soil Adjusted Vegetation Index were used to estimate the green leaf area index (LAI).

Mean annual air temperature and precipitation were provided by the nearest meteorological station. For interpretation of the effect of interannual variation, years of measurements were classified into groups, dry year (2003), normal year (2002), and wet year (2004).

The annual precipitation for 2004 (677 mm) was significantly above the 1951–1999 mean (512 mm) for Bugac, whereas 2002 (416 mm) was closest to average, and 2000 (308 mm) was significantly below average.

**Statistical analysis**

The Kolmogorov–Smirnov test was performed to test for normal distribution of the data. For normally distributed data the t-test was applied to identify significant differences between datasets, for non-normally distributed data the nonparametric Mann–Whitney test was performed instead. Statistical analyses were calculated using SigmaPlot2000 (SPSS Inc., Chicago, USA). Regressions and correlations were fitted and computed using SigmaPlot2000. Polynomial nonlinear regression was used and analyses of variance were calculated using SigmaPlot2000 in order to evaluate the influence of abiotic and biotic parameters on NEE. In each case the regression showing the strongest correlation has been indicated.

**Results and Discussion**

**Dependence of NEE on air temperature**

Temperature dependence of CO$_2$ fixation is positive in the area excluded from grazing while negative in grazed grassland. In both cases the correlation is low and not significant. The relationship between NEE and air temperature were different between the drought stressed and unstressed period. By examining the temperature dependence of annual data it can be concluded that in case of wet year linear and close correlation could be observed in both stands. It seems, that the severity of drought observed in dry year has masked temperature dependent pattern of NEE visible in less severe drought years. This finding is supported by the observation by Ciais et al. (2005).

**Dependence of NEE on light**

As regards of CO$_2$ fixation and PPFD the connection is significant ($P < 0.01$) in both types of grassland (Fig. 1, 2).
Figure 1. Dependence of NEE on light in a grazing excluded dry grassland /Bugac, Hungary/; symbols represent normal year, dry year, and wet year ($R^2 = 0.449$, $P < 0.01$)

Figure 2. Dependence of NEE on light in an extensively grazed dry grassland /Bugac, Hungary/; symbols represent normal year, dry year, and wet year ($R^2 = 0.674$, $P < 0.01$)

This significance is slightly stronger in the grazed land; nevertheless, it is exponential in both grassland types. Another similarity is that the values of dry and normal years fit the regression curve the best. The values of the wet year have been measured at a lower radiation level and can be found farther from the curve. It can be explained by the larger leaf area as well as the favourable abiotic conditions (Tair, SWC).
**Dependence of NEE on leaf area**

The relation between the NEE and LAI values proved to be strong (e.g. Flanagan et al., 2002; Wylie et al., 2003; Gilmanov et al., 2005). The dependence of CO₂ fixation on leaf area shows positive and significant correlation ($P < 0.05$) in both cases (Fig. 3, 4).

![Figure 3. Dependence of NEE on leaf area in a grazing excluded dry grassland /Bugac, Hungary/; symbols represent normal year, dry year, and wet year ($R^2 = 0.381, P < 0.05$)](image)

![Figure 4. Dependence of NEE on leaf area in an extensively grazed dry grassland /Bugac, Hungary/; symbols represent normal year, dry year, and wet year ($R^2 = 0.392, P < 0.05$)](image)
The extent of linear relation is slightly stronger in the grazed grassland which indicates the adaptation of the vegetation to the pressure of extensive grazing. Further accordance can be found in the two types: the correlation between NEE and LAI is the strongest in the wet year as well as if not including data from drought stressed or autumn regrowth periods when vegetation was less active then during intensive growth (Wang et al., 2004). This latter conclusion matches the observations of other researchers based on the comparison of NDVI and NEE data (e.g. Wang et al., 2004; Nagy et al., 2007).

**Dependence of NEE on evapotranspiration**

The dependence of CO$_2$ fixation on evapotranspiration shows different results in the two stands; however, ET dependence is not significant in either group. Stronger and positive correlation is measured in grassland excluded from grazing while in case of the grazed area the correlation is low and negative from the aspect of carbon fixation. The extent of regression is stronger in the not grazed grassland ($R^2 = 0.204$), however in contrast to the results of Chen et al. (2005) based on leaf-level measurements the stand level ET values fluctuated in similar range as data measured in the grazed grassland. The somewhat surprising negative correlation of the grazed stand is the result of a few outliers. It is justified by the fact that when examining the years separately in the wet year strong positive correlation can be seen between NEE and ET in the grazed grassland. As for the non-grazed stand the relation between these two parameters is the strongest also in the wet year. Carbon release is observed in some part of the middle-range ET values which can be explained by the drought stress as well as by the reduced physiological activity that indicates the initiation of resting period. The positive NEE value of the wet year is justified on the one hand by the smaller leaf area, lower temperature and radiation, on the other hand by the increased soil respiration due to the intensive growing period and higher soil moisture.

**Dependence of NEE on soil moisture**

Regarding CO$_2$ uptake and soil moisture content low level, negative and linear correlation is observed in both the area excluded from grazing and in grazed land. This finding is in contrast of the results of Flanagan et al. (2002) based on eddy covariance measurements. The negative relation is not significant in either stands, although it is stronger in case of the area excluded from grazing ($R^2 = 0.127$). No significant differences are seen between the SWC content of the two grassland types that is the most surprising in case of 2003, but indicates that the extreme drought has decreased soil moisture of both areas in approximately the same extent. The high precipitation and soil moisture in 2004 allowed a much higher NEE and an extended period of net carbon gain relative to 2002 and 2003. By excluding the lowest SWC values exponential and also negative curves could be regressed in both stands. It shows that not only low SWC values can be related to small amount of carbon fixation, but also high soil moisture can result small or even positive NEE provided that other ecological factors are unfavourable. It seems that only certain soil moisture ranges (very low and usually the very high) can considerably reduce synphysiological activities while in the intermediate range the SWC determines carbon uptake in smaller extent than expected.
Conclusions

From the three years long chamber CO$_2$ flux experiment, conducted at special semi-arid Pannonian climate, the following main conclusions can be drawn:

Among the examined abiotic and biotic parameters the dependence of NEE on PPFD and LAI has been significant both in the extensively grazed and grazing excluded area. Exponential and linear correlation turned out to be the strongest in both grassland types between CO$_2$ fixation and light dependence and CO$_2$ fixation and leaf area, respectively. The strongest correlation has been observed between the photosynthetically active radiation and stand level CO$_2$ flux ($P<0.01$) in both types of grassland. The dependence of CO$_2$ flux on light and leaf area has been stronger in the extensively grazed area compared to the other examined grass excluded from grazing. Based on this fact it can be assumed that the conversion of land use in traditionally grazed grasslands has negative influence on the relation among NEE and the key abiotic and biotic parameters. It can be explained, beside the accumulation of litter, by the altered vegetation dynamic processes. Extreme dry periods effect, in a negative way, the dependence of CO$_2$ flux on temperature, soil moisture and evapotranspiration. It is justified by the fact that in case of sufficient precipitation strong correlation can be observed between NEE and the mentioned other parameters. It calls the attention to the impacts of weather extremities on the mechanisms in semi-natural sandy grasslands.

Acknowledgements. The research was supported by the GREENGRASS (EVK2-CT2001-00105) project (www.bgc-jena.mpg.de/public/carboeur/projects/green.htm). The publication was supported by Research Centre of Excellence project (1476-4/2016) of Faculty of Agricultural & Environmental Sciences of Szent István University.

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