

CO₂ fluxes of transitional bioenergy crops: effect of land conversion during the first year of cultivation

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Abstract

The present study examined the effect of land conversion on carbon (C) fluxes using the eddy covariance technique at seven sites in southwestern Michigan (USA). Four sites had been managed as grasslands under the Conservation Reserve Program of the USDA. Three fields had previously been cultivated in a corn/soybean rotation with corn until 2008. The effects of land use change were studied during 2009 when six of the sites were converted to soybean cultivation, with the seventh site kept as a grassland. In winter, the corn fields were C neutral while the CRP lands were C sources, with average emissions of $15 \text{ g C m}^{-2} \text{ month}^{-1}$. In April 2009, while the corn fields continued to be a C source to the atmosphere, the CRPs switched to C sinks. In May, herbicide (Glyphosate) was applied to the vegetation before the planting of soybean. After tilling the killed-grass and planting soybean in mid June, all sites continued to be C sources until the end of June. In July, fields previously planted with corn became C sinks, accumulating $15\text{--}50 \text{ g C m}^{-2} \text{ month}^{-1}$. In contrast, converted CRP sites continued to be net sources of C despite strong growth of soybean. The conversion of CRP to soybean induced net C emissions with net ecosystem exchange (NEE) ranging from $155.7 (\pm 25)$ to $128.1 (\pm 27) \text{ g C m}^{-2} \text{ yr}^{-1}$. The annual NEE at the reference site was $-81.6 (\pm 26.5) \text{ g C m}^{-2} \text{ yr}^{-1}$ while at the sites converted from corn/soybean rotation was remarkably different with two sites being sinks of $-91 (\pm 26)$ and $-56.0 (\pm 20.7) \text{ g C m}^{-2} \text{ yr}^{-1}$ whereas one site was a source of $31.0 (\pm 10.2) \text{ g C m}^{-2} \text{ yr}^{-1}$. This study shows how large C imbalances can be invoked in the first year by conversion of grasslands to biofuel crops.

Keywords: bioenergy crops, carbon fluxes, eddy covariance, land use change

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Introduction

The demand for alternatives to petroleum has increased biofuel production from traditional food crops such as corn, sugarcane and soybean (Fargione *et al.*, 2008). The US Energy Independence and Security Act (EISA) of 2007 mandates that 36 billion gallons of renewable fuels are to be produced by 2022, of which $6.1 \times 10^{10} \text{ L}$ are expected to come from cellulosic feedstocks (Kim *et al.*,

2009). However, to evaluate policies regarding biofuel production, we need to know not only how much harvestable carbon (C) the system can produce, but also its eventual impact on soil organic C stocks, which bears on its global-warming potential (Robertson *et al.*, 2000; Lal *et al.*, 2004; Robertson & Swinton, 2005). Land use change (LUC) such as conversion to bioenergy systems is responsible for significant changes in the global C budget through alteration of the rates of accumulation and turnover of soil C and vegetation biomass as well as soil erosion (Richter *et al.*, 1999; Lal, 2002; Zhang *et al.*, 2010). Both direct and indirect LUC effects have been identified. Direct LUC emissions are associated with

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the clearing of land and its preparation to grow crops for biofuel production and include changes in soil C and aboveground C production (Kim *et al.*, 2009). Indirect LUC refers to the potential effects which may be caused by cultivating biomass for bioenergy, biofuels or biomaterials on lands which were previously used for other cultivation (e.g., feed, food or fiber production), or would have been in the future in a business-as-usual (baseline) scenario without the biomass use for bioenergy, biofuels or biomaterials (Uwe *et al.*, 2009).

Several studies (Whendee & Ryan, 2001; West *et al.*, 2004) have pointed out that the conversion of natural habitats to cropland would release CO₂ because of burning and microbial decomposition of organic C stored in plant biomass and soils. Additionally, the rapid release of CO₂ from fire used for clearing land or from the decomposition of leaves and fine roots is followed by a prolonged period of enhanced CO₂ emission because of stimulated decomposition of coarse roots, branches and soil organic matters. The amount of CO₂ released upon land conversion incurs a 'C debt' that may or may not be 'paid off' by sequestration in future years (Fargione *et al.*, 2008). By excluding the CO₂ emissions due to land conversion, early studies provided an incomplete picture because they counted the C benefits of using land for biofuels but not the C costs (i.e., the C sequestration sacrificed when diverting land from its existing uses). In practice, conversion of land to biofuel production can potentially contribute to CO₂ stabilization only if the conversion increases C sequestration (Searchinger *et al.*, 2008).

To quantify the effect of land conversion for the C budget of bioenergy production, we examined the net exchange of CO₂ using the eddy covariance (EC) technique in seven bioenergy production systems in southwestern Michigan. Four of the seven sites had been managed as grasslands during the past 20 years, while the other three were conventional agricultural fields. The effects of LUC were studied in six large fields including three grasslands of the Conservation Reserve Program (CRP) and three agricultural sites converted in 2009 to soybean. A seventh CRP grassland remained as grassland and served as a reference site in this study. Our objectives were to: (i) examine the temporal dynamics of net ecosystem exchange (NEE) of C during the initial stage of the land conversion and (ii) quantify the effect of major management activities on C fluxes during the conversion year. We hypothesized that the long-term unmanaged CRP (<http://www.fsa.usda.gov/FSA>) grasslands would turn into major C sources after conversion, and that specific management activities would play a profound role in determining the magnitude and direction of NEE.

Materials and methods

Site description

The experiment was conducted under the aegis of the Great Lakes Bioenergy Research Center (GLBRC, <http://glbrc.org/>), and the study sites were located at the W.K. Kellogg Biological Station (KBS) (42°24' N, 85°24' W, 288 m a.s.l.) of Michigan State University, located in southern Michigan, USA. The region lies on the north-eastern edge of the US Corn Belt. The climate is temperate and humid, with a mean annual air temperature at KBS of 9.7°C and an annual precipitation of 920 mm, evenly distributed throughout the year with about half falling as snow. The soil textural class of all sites is sandy clay loam with a pH range from 5.8 to 6.4. Soil C and N contents were significantly higher in the grassland sites compared with the agricultural sites (Table 1).

Aerial photos show that all seven sites were in either row crops or pasture for at least the past 72 years. Three of the sites (AG-S1, AG-S2, AG-S3) had been cultivated conventionally as a corn/soybean rotation for the past 10 years and were in row crops for at least 30 years before that. The other four sites (CRP-S1, CRP-S2, CRP-S3 and CRP-Ref) were monocultural grasslands, dominated by smooth brome (*Bromus inermis* Leyss), and had been maintained as CRP grasslands (<http://www.fsa.usda.gov/FSA>). The CRP is a land retirement program designed to mitigate agriculture's adverse affects on the environment and currently includes about 10 million ha across the United States (USDA, 2008). When originally established its purpose was to conserve and improve soil, water and wildlife resources by establishing perennial plant cover on highly erodible and other environmentally sensitive land through 10–15 year leases (Feather *et al.*, 1999). The US Department of Agriculture administers the CRP.

To prepare the sites for soybean cultivation in 2009, the nonselective herbicide glyphosate [N-(phosphonomethyl) glycine; Syngenta Agro, Syngenta Corporation, Wilmington, DE, USA] was applied at 2.85 kg ha⁻¹ on DOY 125 (sites CRP-S1, CRP-S2, CRP-S3), DOY 126 (AG-S1) and DOY 149 (AG-S2, AG-S3) using a pull-type sprayer (Demco Dethmers MFG Company, Boyden, IA, USA). Soybean cultivar 92M91, genetically engineered to resist Glyphosate, was planted on DOY 160 at the agricultural sites and on DOY 161–162 at the CRP sites (except the CRP-Ref site). Fertilizer (140 kg P ha⁻¹) was applied at site AG-S3 only, based on previous soil test results.

EC and microclimate measurements

The turbulent exchange of CO₂ between canopy and atmosphere was measured throughout 2009 using EC

Table 1 Soil physical and chemical properties of study sites

Site	Area (ha)	Previous cultivation	2009 cultivation	Textural class	Sand (g kg ⁻¹ soil)	Silt (g kg ⁻¹ soil)	Clay (g kg ⁻¹ soil)	Soil pH	CEC [meq (100 g) ⁻¹]	Bulk density (g cm ⁻³)	Nitrogen (g kg ⁻¹ soil)	Carbon (g kg ⁻¹ soil)
CRP-S3	17.9	Grassland	Soybean	Sandy clay loam	700.1 ^a	32.5 ^b	267.4 ^a	5.9 ^a	6.00 ^{ab}	1.42 ^c	2.03 ^{cd}	23.76 ^b
CRP-S2	13.1	Grassland	Soybean	Sandy clay loam	684.9 ^a	47.5 ^b	267.6 ^a	6.2 ^a	5.46 ^b	1.34 ^c	2.27 ^{bc}	26.40 ^{ab}
CRP-S1	19.5	Grassland	Soybean	Sandy clay loam	670.0 ^a	60.1 ^{ab}	269.9 ^a	6.1 ^a	6.02 ^{ab}	1.41 ^c	2.79 ^a	30.94 ^a
AG-S1	11.2	Corn	Soybean	Sandy clay loam	642.5 ^a	52.6 ^{ab}	304.9 ^a	6.4 ^a	8.08 ^{ab}	1.55 ^b	1.32 ^e	14.21 ^c
AG-S2	14.1	Corn	Soybean	Sandy clay loam	624.9 ^a	42.6 ^b	332.5 ^a	6.4 ^a	7.07 ^{ab}	1.73 ^a	1.34 ^e	13.71 ^c
AG-S3	23.0	Corn	Soybean	Sandy clay	535.2 ^a	102.4 ^a	362.4 ^a	5.8 ^a	8.60 ^a	1.61 ^b	1.62 ^{de}	16.38 ^c
CRP-Ref	9.1	Grassland	Grassland	Sandy clay loam	600.1 ^a	52.5 ^b	347.4 ^a	6.2 ^a	6.50 ^{ab}	1.41 ^c	2.69 ^{ab}	30.60 ^a

Means followed by same letters are not significantly different by *t*-test ($P < 0.05$). CEC, cation exchange capacity.

technique (Lee *et al.*, 2004). A 3 m EC tower was located in the center of each field, with a homogeneous fetch of at least 200 m in all directions. Our EC systems consisted of a LI-7500 open-path infrared gas analyzer (Li-Cor Biosciences, Lincoln, NE, USA), a CSAT3 three-dimensional sonic anemometer [Campbell Scientific Inc. (CSI), Campbell Scientific, Inc., Logan, UT, USA] and a CR5000 datalogger (CSI). The LI-7500 was calibrated at least every 4 months in the laboratory using zero-grade nitrogen for zeroing CO₂ and H₂O sensors, and a dew-point generator (LI-610, Li-Cor Biosciences) and CO₂ standards (Scott-Marrin, Inc., Riverside, CA, USA, 489.03 ppm) for setting H₂O and CO₂ spans and calibration, respectively. The 30-min mean flux of CO₂ was computed as the covariance of vertical wind speed and the concentration of CO₂. Quality measures before the mean calculation included: (1) the removal of spikes >6 standard deviations from the 30-min mean through block averaging, (2) humidity and pressure correction of the sonic temperature (Schotanus *et al.*, 1983) and (3) correction of the coordinate system by the planar fit method. The algorithm uses the formulation of Leuning (2004) in the planar fit coordinate system (Wilczak *et al.*, 2001), which was defined from the entire year's mean wind data in all sites investigated using the EC PROCESOR software package (<http://www4.ncsu.edu/~anoorme/>). The 30-min mean fluxes were further corrected for fluctuations in air density using the Webb–Pearman–Leuning expression (Webb *et al.*, 1980), including the term for the warming of the infrared gas analyzer above air temperature (Grelle & Burba, 2007). The flux data were almost continuous during the study period with <10% of data missing. Upon screening the data for inadequate turbulent mixing and out-of-range fluxes (e.g., NEE outside of $\pm 50 \mu\text{mol m}^{-2} \text{s}^{-1}$), the amount of usable data was 76–86% for the total time period.

The following micrometeorological variables were measured above canopy: air temperature T_a (°C) and relative humidity (RH; %) using HMP45C (CSI), photosynthetically active radiation (PAR; $\mu\text{mol quanta m}^{-2} \text{s}^{-1}$) using LI-190SB (Li-Cor Biosciences), incoming and reflected solar radiation (W m^{-2}) using the CNR1 (Kipp & Zonen, Delft, the Netherlands) and precipitation P (mm) using tipping bucket rain gauges TE525WS-L (Texas electronics, Dallas, TX, USA). The 30-min means of soil temperature T_s (°C) were monitored at three different depths (2, 5 and 10 cm) using the CS107 probes (CSI). Volumetric soil water content was averaged for the top 30 cm using a vertically inserted CS616 Time Domain Reflectometer probe (CSI). Soil heat flux (W m^{-2}) was measured using the HFT3 flux plates (CSI). Snow depth on the ground was monitored using a SR50 sonic ranging sensor (CSI).

C pools

Above- and belowground net primary productivity

Aboveground net primary productivity (ANPP) was determined by harvesting the vegetation in ten 1 m² subplots randomly distributed in each field. Biomass samples were collected before complete leaf drop when there were only a few leaves and petioles on the dried stems. Plants from each subplot were clipped at ground level and collected together with soybean surface litter. Both the litter and live plant components were dried in a forced-air oven at 65 °C for at least 4 days until a constant dry mass was reached. ANPP was calculated as

$$\text{ANPP} = (G + S + L) + W + \text{Li}, \quad (1)$$

where *G* is the grain mass, *S* the stem mass, *L* the soybean leaf mass, *W* the weed mass and *Li* is the litter mass from the 10 subplots (i.e., averaged by site). Dried plant material was threshed using a small grain thresher to separate seeds from soybean shells. Belowground net primary productivity (BNPP) was estimated by the auger method (Bohm, 1979) at plant physiological maturity in autumn 2009. BNPP measurements were taken in the same ten subplots where the aboveground biomass was harvested. In each subplot, two soil cores were taken to a depth of 20 cm using a root auger of 7.5 cm diameter (Eijkelkamp Agrisearch Equipment BV, Giesbeek, the Netherlands). The first core was positioned at the center of the plant to include most larger roots and second core at the midway point between two plant rows where there were mainly fine roots. Following the recommendations of Anderson & Ingram (1993), roots were separated from the soil by soaking in water and then gently washing over a series of sieves with mesh sizes of 2.0 and 0.5 mm. Cores that could not be sorted immediately were frozen (0 °C) until they got processed. Roots from each subplot were oven dried at 65 °C and weighed. Root biomass was extrapolated from the cored area to the area of each subplot to estimate the root mass of one plant, and multiplied by the plant density to calculate BNPP, which was then added to the ANPP measured in the subplots to yield overall NPP.

Soil chemical and physical properties. Soil sampling was performed in May 2009 before soybean planting and in November after harvesting. Samples were collected in 10 georeferenced positions at each site. At each position, samples were collected from 0 to 25 cm depth with the root auger described above. The samples were taken between the plant rows and were composited from two 7.5 cm diameter cores within 0.2 m of each other. A total

of 70 samples were collected. All samples were sieved (4 mm) to remove plant residues and stones and then oven dried at 65 °C for at least a week. The samples were ground using a shatterbox (Shatterbox Model 8530, SPEX CertiPrep, Metuchen, NJ, USA) to pass through a 250 µm sieve. Total C was measured using an automatic CHN analyzer (Costech Analytical Technologies Inc., Valencia, CA, USA). Bulk density and soil texture were measured by the core method (Blake & Hartge, 1986) and Bouyoucos hydrometer method (Gee & Bauder, 1986), respectively. Other chemical properties including pH and cation exchange capacity were analyzed by standard methods at Michigan State University's Soil Testing Laboratory.

Statistical analysis

All productivity parameter, soil properties and winter NEE were tested using a one-way analysis of variance (ANOVA) and separation of means was subjected to Student's *t*-test using statistical software JMP, Version 7 (SAS Institute Inc., Cary, NC, 1989–2007). Statistical significance was accepted if *P*-value is <0.05.

Results

Microclimate

During 2009, the annual average air temperature (*T_a*) was 8.5 °C and the precipitation was 920 mm which was distributed throughout the year (Fig. 1a and b). However, there were significant intersite differences in soil volumetric water content (VWC) (Fig. 1b), particularly from January through March before conversion. The mean (± SE) VWC from January to March was 30.7% (± 3.4) at the CRP reference site, 23.2% (± 2.3), 18.0% (± 3.0) and 23.3% (± 2.7) at the grassland sites to be converted, and 9.7 (± 4.3), 8.5% (± 5.0) and 13.3% (± 6.4) at the agricultural sites to be converted. Soil temperature (*T_s*) at 5 cm depth and *T_a* showed a strong correlation (*P* < 0.0001, *R*² = 0.87) throughout the year. Mean annual soil temperature *T_s* did not vary significantly among the seven sites and was 9.6 °C at the reference site, 10.1 °C at the CRP grasslands and 11.0 °C at the agricultural sites (Fig. 1a and b). Photosynthetic photon flux density and net radiation showed a typical seasonal pattern for this latitude (Fig. 1c). During the second half of December 2008 all sites were covered by snow, but due to a rain event the snow was melted at the beginning of January 2009, clearing the snowpack at all sites for the first half of the month. During the second half of January and throughout February and March snow cover existed at all the sites (Fig. 1d).

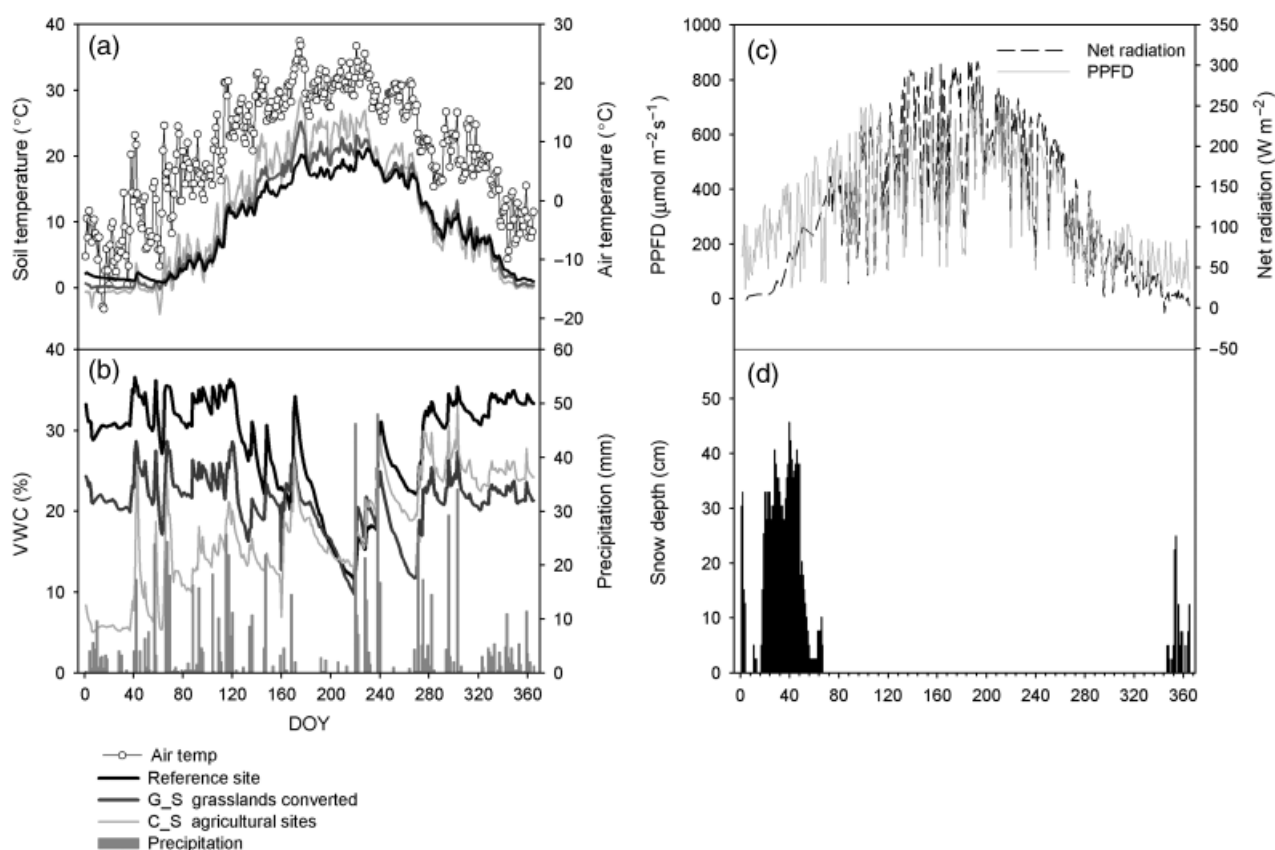


Fig. 1 Annual time series of: (a) mean daily air temperature above the canopy and soil temperature measured at 5 cm depth; (b) mean daily soil volumetric water content and precipitation events; (c) annual course of daily mean photosynthetic photon flux density (PPFD) and net radiation; and (d) snow depth (radiation and snow were measured at the reference grassland site). DOY = day of year.

Ecosystem C pools and fluxes

During the conversion year, the mean (\pm SE) annual NPP at the three sites converted from grassland to soybean was significantly higher (Fig. 2) compared with the three agricultural sites converted to soybean, reflecting in part the growth of *B. inermis* early in the season in the grasslands while the agricultural sites were relatively barren (Table 2). The mean soybean yield across all six sites was $2.22 (\pm 0.35) \text{ Mg ha}^{-1}$, which was comparable to the average soybean productivity of 2.16 Mg ha^{-1} for southwest Michigan (http://www.nass.usda.gov/QuickStats/PullData_US.jsp) (Table 2).

NEE of CO₂ is denoted as positive in the case of net emission to the atmosphere, and negative for net uptake. Seasonal changes in NEE reflected the seasonal transition from winter into the growing season, until the herbicide application (Figs 3 and 4). The mean daily NEE in winter (from DOY 1 to 90) was $0.97 (\pm 0.09)$, $1.10 (\pm 0.11)$ and $1.04 (\pm 0.11) \text{ g C m}^{-2} \text{ day}^{-1}$ at sites CRP-S1, CRP-S2 and CRP-S3, respectively (Fig. 3a–c). In contrast, winter NEE was lower at the agricultural sites, where the corn had been harvested the previous

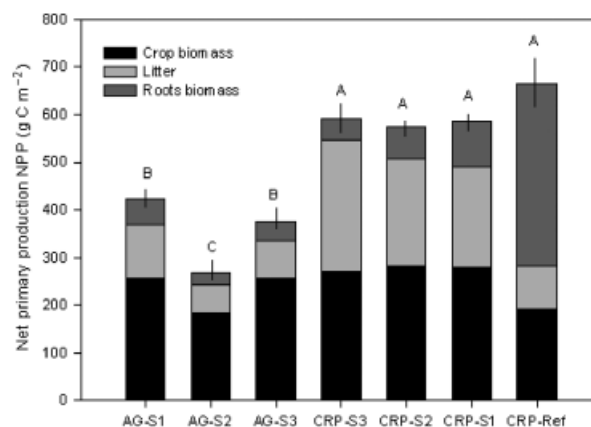


Fig. 2 Net Primary Production of the sites: conventional agriculture to no-till soybean land use change (AG-S1, S2, S3); Conservation Reserve Program (CRP) land to no-till soybean land use change (CRP-S1, S2, S3); and reference site with continued CRP land use (CRP-REF). Difference significant at $p < 0.05$.

autumn, and varied from $0.70 (\pm 0.07)$ at site AG-S1, $0.57 (\pm 0.06)$ at site AG-S2, to $0.44 (\pm 0.05) \text{ g C m}^{-2} \text{ day}^{-1}$ at site AG-S3. In April, the CRP grasslands switched

Table 2 Total yield, grain carbon, above- and belowground net primary productivity (NPP), total NPP, and annual net ecosystem exchange (NEE) at the sites investigated

Site	Yield (T ha ⁻¹)	Grain (C g C m ⁻²)	ANPP (g C m ⁻²)	BNPP (g C m ⁻²)	NPP (g C m ⁻²)	Annual NEE (g C m ⁻²)
CRP Ref	nd	nd	281.5 (± 126.0)	399.5 (± 101.2)	681.0 (± 161)	-81 (± 26.5)
CRP-S3	2.36 (± 0.13)	125.1 (± 7.22)	546.8 (± 21.0)	43.5 (± 11.2)	590.3 (± 23.8)	155.7 (± 25.0)
CRP-S2	2.18 (± 0.26)	116.1 (± 14.26)	506.2 (± 16.0)	67.8 (± 20.6)	574.0 (± 26.0)	139.6 (± 27.8)
CRP-S1	2.29 (± 0.18)	122.0 (± 9.78)	489.4 (± 21.4)	97.5 (± 22.3)	586.9 (± 31)	128.1 (± 27.0)
AG-S1	2.40 (± 0.19)	127.3 (± 10.10)	369.4 (± 19.0)	52.4 (± 4.3)	421.8 (± 19.4)	-56.0 (± 20.7)
AG-S2	1.54 (± 0.18)	81.9 (± 9.59)	243.1 (± 22.1)	24.5 (± 4.8)	267.6 (± 22.6)	31.0 (± 10.2)
AG-S3	2.56 (± 0.24)	135.7 (± 13.20)	336.4 (± 24.1)	39.9 (± 7.4)	376.3 (± 25.2)	-90.8 (± 25.7)
Mean CRP-S	2.27 (± 0.09)	121.0 (± 4.57)	514.1 (± 29.5)	69.6 (± 27.0)	583.7 (± 46.9)	141.1 (± 13.8)
Mean AG-S	2.16 (± 0.54)	114.9 (± 28.9)	316.3 (± 65.5)	38.9 (± 13.9)	355.2 (± 39.1)	-38.6 (62.7)

nd, not detected.

from a C source to a sink as *B. inermis* entered its growth phase.

After the application of the herbicide at the three grassland sites on DOY 125, there was a sharp increase in NEE approximately within 50 h, reflecting the cessation of photosynthetic assimilation and the eventual decomposition of the killed grass biomass. Mean (± SE) daytime NEE during the 15 days before herbicide application was -2.26 (± 0.81), -1.27 (± 0.62) and -1.38 (± 0.60) g C m⁻² day⁻¹, respectively, for CRP-S3, CRP-S2 and CRP-S1 (Fig. 5a). Subsequent to the herbicide application, emission of CO₂ ranged from 1.65 (± 0.22) g C m⁻² day⁻¹ at site G_S1 to 2.3 (± 0.30) g C m⁻² day⁻¹ at site G_S3 from DOY 125 through the end of June (DOY 181). Meanwhile, the CRP reference site continued to show net CO₂ uptake well into the summer (Fig. 5b). Herbicide application also induced a decrease in the latent heat flux (LE in W m⁻²), with mean (± SE) values over the 15 days before the application of 75.9 (± 15.9), 54.1 (± 20.2) and 61.4 (± 19.8) W m⁻² for sites G_S1, G_S2 and G_S3, respectively. After the herbicide application until the end of June, LE varied from 52.9 (± 7.8) W m⁻² at G_S1, 39.4 (± 5.5) W m⁻² at G_S2 to 38.6 (± 5.2) W m⁻² at G-S2, while during the same period of time, LE was 120 (± 9.8) W m⁻² at the reference site (Fig. 5c and d).

The monthly cumulative values of NEE after the herbicide application varied from 46.1 (± 7.9), 19.9 (± 8.0) and 19.0 (± 9.2) g C m⁻² month⁻¹ in May to 64.6 (± 8.5), 63.4 (± 9.0) and 84.7 (± 8.9) g C m⁻² month⁻¹ in June, respectively, at sites CRP-S3, CRP-S2 and CRP-S1. During the soybean growth period (June–October), the daily NEE was -0.05 (± 0.24), -0.26 (± 0.25), -0.4 (± 0.25) g C m⁻² day⁻¹, respectively, for CRP-S3, CRP-S2 and CRP-S1G, and -1.57 (± 0.19), -0.74 (± 0.24), and -1.5 (± 0.26) g C m⁻² day⁻¹ from AG-S1, AG-S2 and AG-S3, respectively. The NEE over the soybean growing season was not significantly

correlated with soil C content ($P = 0.32$; $R^2 = 0.19$) for all sites.

The cumulative annual NEE showed net CO₂ emission for the three converted CRP grassland sites, whereas the CRP reference site showed net uptake (Fig. 3). Two of the sites previously cultivated as corn/soybean rotation were a net C sink (Fig. 4), ranging from -90.8 (± 25.7) to -56.0 (± 21) g C m⁻² yr⁻¹ although one site (AG-S2) approached C neutrality. Site AG-S2 was characterized by a lower soil C content and lower biomass production compared with the others, with an annual NEE of 31 (± 10) g C m⁻² yr⁻¹.

The relationship of NEE to PAR during the soybean growing season was clearly different between the grassland and agricultural sites (Fig. 6). The Pearson correlation coefficient indicated that daily PAR explained 85% (range 84–92%) of variation in daily NEE in the grassland sites (Fig. 6a–c) from July through September and 79.9% (range 55–95%) at the agricultural sites (Fig. 6d–f). A weak response of NEE to PAR was observed at site AG-S2 where only 57% of the variability could be explained by the PAR (Fig. 6e). During the same time the NEE vs. PAR response at the reference site showed a significant but lower correlation compared with the other sites ($P < 0.001$; $R^2 = 0.66$) (data not shown). The cumulative NEE for 2009 at the CRP site indicated net CO₂ emission, ranging from 155.7 (± 25.0) at G_S1, 139.6 (± 27.8) at site G_S2 to 128.1 (± 27.0) g C m⁻² yr⁻¹ at site G-S3.

Discussion

During the winter the mean NEE was 1.09 (± 0.11), 1.03 (± 0.11) and 0.93 (± 0.33) g CO₂ m⁻² day⁻¹ at three CRP grasslands. These values corresponded well to the wintertime CO₂ evolution reported by Ham & Knapp (1998) and Volk & Niklaus (2002). However, winter NEE observed at the reference site was 2.1

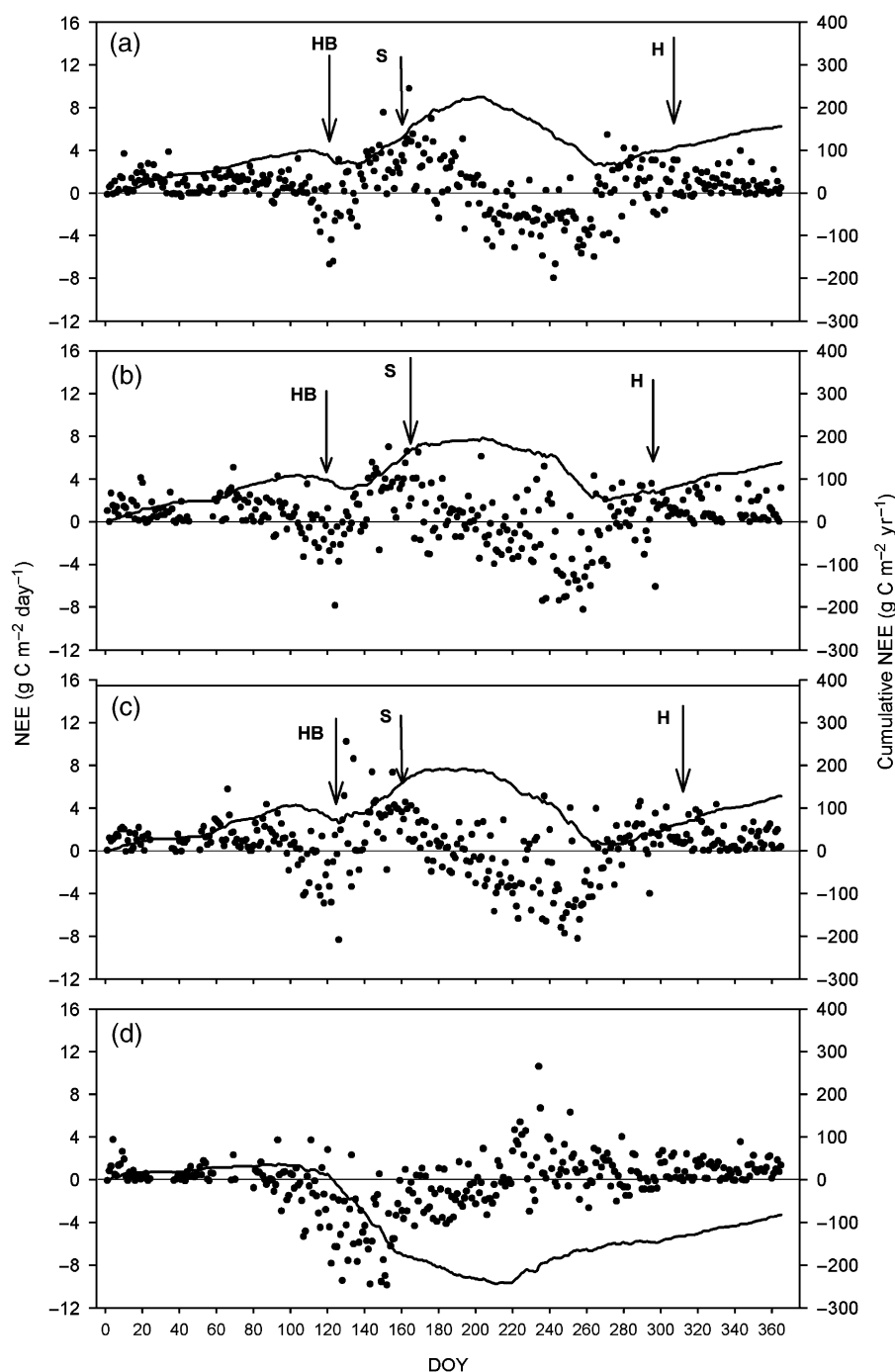


Fig. 3 Annual patterns of daily NEE (dots) in $\text{g C m}^{-2} \text{ day}^{-1}$ and cumulative NEE (lines) in g C m^{-2} across the CRP converted grassland sites (a-c, representing sites CRP-S3, CRP-S2 and CRP-S1, respectively) and the CRP reference site (d). Arrows show the timing of herbicide application (HB), sowing of soybean (S), and soybean harvest (H). DOY = day of year.

(± 0.3) $\text{g CO}_2 \text{ m}^{-2} \text{ day}^{-1}$, and hence it was 43% lower than the average of the other grassland sites of this study. At the reference site, the biomass was harvested in 2008 but it had been left undisturbed throughout the year at sites G_S 1, G_S 2 and G_S 3, which could explain the much larger emission of CO₂ in these sites

compared to the reference site. The average daily NEE in the winter showed a positive correlation with soil C content of the first 10 cm (adjusted $R^2 = 0.77$, $P = 0.021$; Fig. 7) at sites G_S 1, G_S 2 and G_S 3. This is consistent with the results obtained by Howard (2007) which showed a significant correlation between daily CO₂ flux

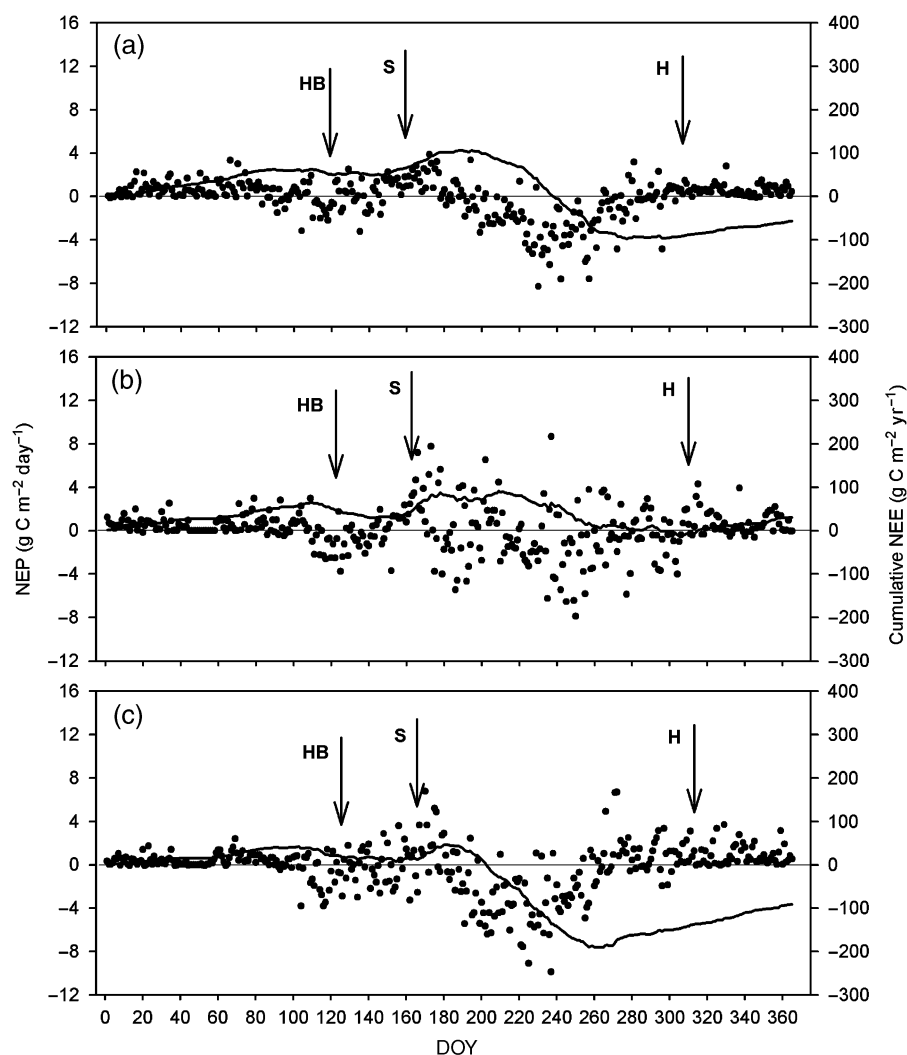


Fig. 4 Annual changes of daily NEE (dots) in $\text{g C m}^{-2} \text{ day}^{-1}$ and cumulative NEE (lines) in g C m^{-2} at the agricultural sites (a-c, representing sites AG-S1, AG-S2, and AG-S3, respectively). DOY = day of year.

and the amount of live biomass present at stand level in a grass pasture where an average efflux from December through March of $3.09 (\pm 0.14) \text{ g CO}_2 \text{ m}^{-2} \text{ day}^{-1}$ was reported. At global scale, Raich & Potter (1995) found soil CO_2 efflux rates of about $4\text{--}6 \text{ g CO}_2 \text{ m}^{-2} \text{ day}^{-1}$ at an air temperature near 0°C . Gilmanov *et al.* (2004) conducted a literature review on wintertime CO_2 efflux from various nonforest ecosystems and reported values for prairies ranging from $0.77 \text{ g CO}_2 \text{ m}^{-2} \text{ day}^{-1}$ in a northern mixed prairie in southwestern Canada to $4.4 \text{ g CO}_2 \text{ m}^{-2} \text{ day}^{-1}$ in a grassland in Switzerland. The average NEE in the winter months at our agricultural fields was 44% lower than that of the grasslands and varied from $2.5 (\pm 0.28)$ at site AG_S1 to $1.61 (\pm 0.19) \text{ g CO}_2 \text{ m}^{-2} \text{ day}^{-1}$ at site AG_S3. The lower emission observed at the corn fields appeared to be related to the lower soil C content and lower ground litter compared with the

grassland sites (Fig. 7). One-way ANOVA indicated that there was a significant difference between daily NEE of grassland and two agricultural fields during winter months ($F = 9.23$, $P < 0.05$; Fig. 9).

Glyphosate is concentrated nonselective foliar systemic herbicide for controlling a broad spectrum of annual and perennial plants including grasses, sedges, broad-leaved weeds and woody plants. It is formulated as a liquid and acts by blocking the shikimate pathway through inhibition of 5-enolpyruvyl-shikimate-3-phosphate synthase (EPSPS) (Duke *et al.*, 2003). Glyphosate is particularly effective because most plants metabolically degrade it very slowly or not at all, and it translocates well to metabolically active tissues such as meristems (Cerqueira & Duke, 2006). In our study, its effect on NEE at the CRP grassland fields was evident within 2 days of application (Fig. 5), although the plants did not show

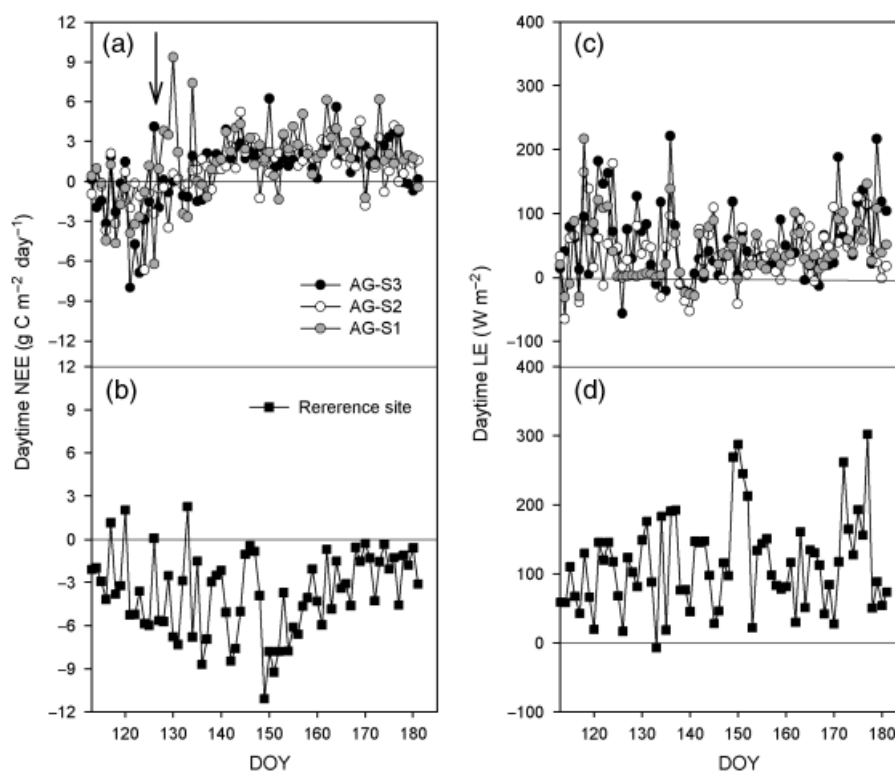


Fig. 5 Effect of herbicide application (day 125) on daily NEE (a) and LE (c) at the converted grassland sites compare to the CRP-grassland reference site (b, d) measured from 15 days before to 55 days after the application. DOY = day of year. Arrow show the day of the herbicide application.

evident morphological symptoms (yellowing leaves) for about 15 days. In a similar study in a corn/soybean rotation, Baker & Griffis (2005) found the effect of Glyphosate application on daytime NEE 5 days after the application when the ecosystem switched from a sink ($-4 \text{ g C m}^{-2} \text{ day}^{-1}$) to a source ($2 \text{ g C m}^{-2} \text{ day}^{-1}$). Additionally, they reported that the nighttime NEE was not substantially affected afterward. This suggests that herbicides directly affected the photosynthetic activity of the plants, by suppressing leaf metabolism but not the decomposition of organic materials.

The NEE showed a greater response after herbicide application in the fields converted from grassland compared to those converted from agriculture (Figs 3 and 4). The large amount of dead *B. inermis* biomass present on the ground in the converted CRP sites, together with their higher soil organic C content, likely explain the higher emission of CO₂ compared with the agricultural sites. This effect is particularly evident in June when the mean CO₂ emission from the grasslands was $92 (\pm 11.2) \text{ g C m}^{-2} \text{ month}^{-1}$ while in the agricultural sites it was $49.0 (\pm 1.7) \text{ g C m}^{-2} \text{ month}^{-1}$.

In general, changes in soil C invoked by LUC can be reversed when the ecosystem returns to its former state, although this may take decades (Gelfand *et al.*, in re-

view). This paper reports the C flux responses in the first year alone and shows that the converted CRP grasslands were C sources that year. Follow-up studies will show how long the enhanced C emission lasts. The loss of soil organic C by conversion of natural vegetation to cultivated annual crops is well known (Post & Kwon, 2000). A meta-analysis by Guo & Gifford (2002) reported a decline of soil C stock of approximately 42% after conversion from a forest to an annual crop, and 59% after conversion from a pasture to an annual crop. The same study showed that soil C stocks increased 8% after conversion from forest to pasture and 19% after conversion from crop to pasture. The CRP grasslands converted to soybeans in our study had about twice the C in the soil than the agricultural sites. One-way ANOVA indicated that there was no difference between soil C in the first 10 cm in May vs. November across all the sites (Fig. 8).

To quantify the effect of LUC from either unmanaged vegetation or food crops to biofuel production systems, it is important to consider not only the nature of the biofuel crops (e.g., annual vs. perennial, grain vs. cellulosic), but also how the conversion is managed. The sites investigated in this study were all managed as no-tillage systems. The application of conventional soil

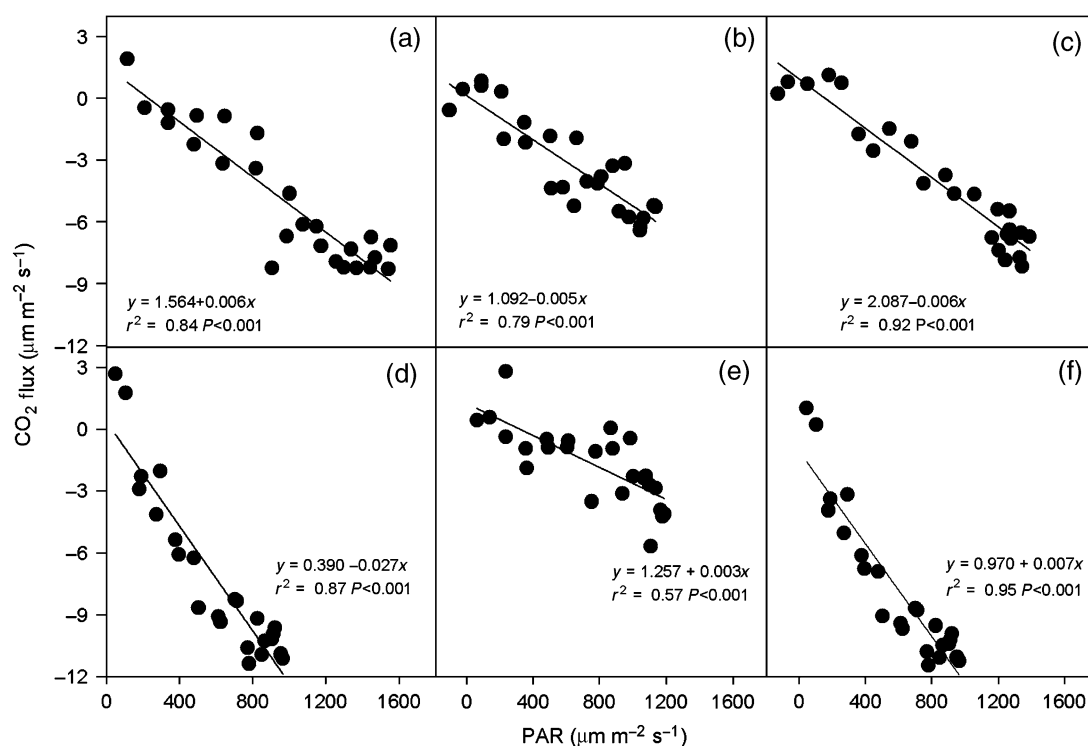


Fig. 6 Response of net ecosystem exchange of CO_2 (NEE) to photosynthetically active radiation (PAR) measured daily between 07.30 am and 07.30 pm from July to September. Panels a–c show the converted CRP-grassland sites while d–f show the agricultural sites. All of the regression fits were significant at $p < 0.001$.

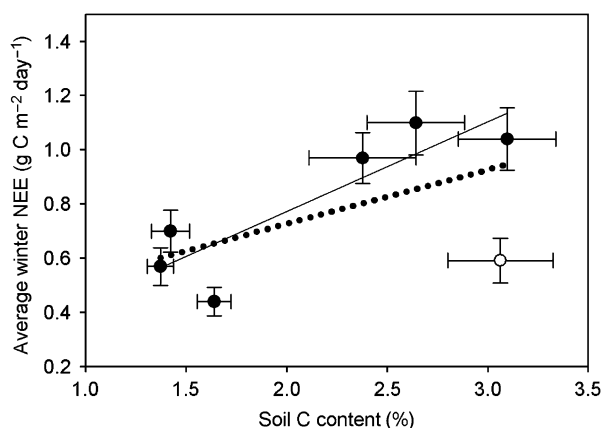


Fig. 7 Relationship between soil organic carbon (C) in the upper 10 cm and mean winter time NEE including the CRP-grassland reference site (dotted line not significant at $p = 0.05$) and solid line shows regression without the reference site: $y = 0.021 + 0.032x$; $R_2 = 0.77$, $p < 0.05$.

tillage could induce a larger emission of CO_2 with a consequently larger impact on global warming potential (Bhardwaj *et al.*, 2010).

Our EC flux data clearly show the C imbalances invoked by the conversion of the CRP grasslands to

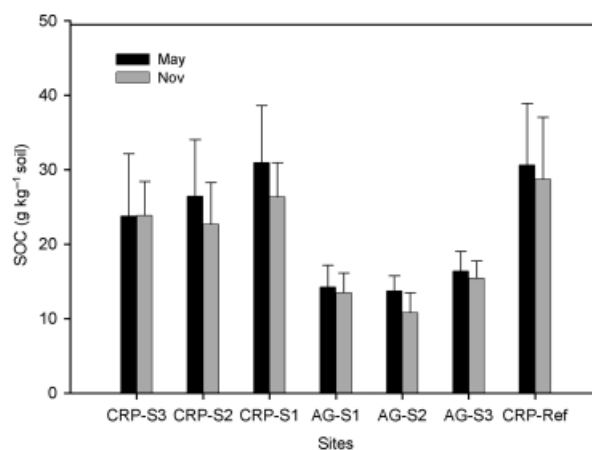


Fig. 8 Soil organic carbon (SOC) measured in the upper 10 cm in May and November. None of these sites showed a significant change in SOC between the two sampling dates ($p < 0.05$).

soybean biofuel production (Fig. 3). Compared with the reference site, the herbicide application onto the perennial grassland midway into its growing season abruptly halted primary production and resulted in net CO_2 emission (i.e., positive NEE) for several weeks

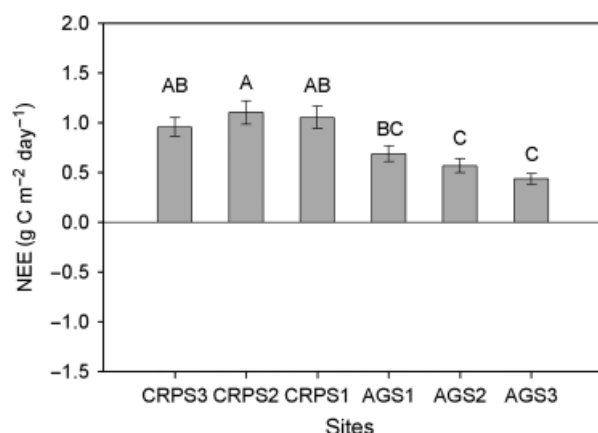


Fig. 9 One way ANOVA of the NEE fluxes across the sites in winter months (From DOY 1 to DOY 90). Difference significant at $p < 0.05$.

until the soybean crop began to grow. By the end of the year that C imbalance was reflected in a large positive value for the cumulative NEE, reflecting a 'C debt' that has to be considered in evaluating the climate implications of biofuel crop production (Gelfand *et al.*, in review). The effect of the herbicide was more modest in the agricultural sites, which had much less live biomass due to previous cultivation of annual crops, and those sites ended the year with negative or near-neutral values of cumulative NEE (Fig. 4).

Conclusions

Using direct measurements of cluster EC towers, this study shows how large C imbalances can be invoked in the first year by conversion of grasslands or row crops to biofuel production, and that longer-term gains in soil C under CRP management can quickly be negated under a new crop production regime. The CRP management over the last 20 years at the grassland sites had resulted in accumulation of soil C to approximately twice the concentrations of a traditional corn/soybean rotation. The conversion of CRP grasslands back to an annual cropping system invoked a large emission of CO₂ during the first year of cultivation, mainly due to the dead aboveground biomass present after the herbicide application and decomposition of roots. Meanwhile, the undisturbed reference CRP grassland continued to be a C sink. While this enhanced CO₂ emission is presumably a transient phenomenon that may last less than a couple of years, it must be factored into the greenhouse gas accounting as a 'C debt' that may or may not be repaid in subsequent years depending on the C balance of the biofuel production system (Fargione *et al.*, 2008). More research is needed to understand the longer-term dy-

namics of C balance following conversion to a bioenergy crop production system.

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