The effect of a dry spring on seasonal carbon allocation and vegetation dynamics in a poplar bioenergy plantation

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Abstract

In this study the seasonal variation in carbon, water and energy fluxes as well as in net primary productivity (NPP) of different tree components is presented for a 2-year-old poplar (Populus spp.) plantation. A thorough ecophysiological study was performed at ecosystem scale, at tree and at leaf level, in this high-density bioenergy plantation. Seasonal variation in NPP and fluxes was analysed in relation to meteorological parameters at the field site. The growing season length in terms of carbon uptake was controlled by leaf area development until the maximum leaf area index (LAImax) was reached. Afterwards, a shift to belowground carbon allocation was observed. A dry period in spring caused a reduced leaf area production as well as a decrease in net ecosystem exchange and gross primary production (GPP) due to stomatal closure. Water use efficiency and fine root growth increased in response to limiting soil water availability in the root zone. When soil water availability was not limiting, GPP was controlled by a decrease in solar radiation and air temperature. The results of this study indicate that the productivity of recently established bioenergy plantations with fast-growing trees is very sensitive to drought. The interaction between soil water availability and factors controlling ecosystem GPP is crucial in assessing the CO2 mitigation potential under future climate conditions.

Keywords: growing season length, intra-annual variability, net ecosystem exchange, net primary productivity, Populus spp., short-rotation coppice, soil water availability, water use efficiency

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Introduction

Forest ecosystems play a crucial role in the global carbon cycle (Kauppi et al., 1992; Birdsey et al., 1993; Grace, 2004), in particular by mitigating the increasing anthropogenic CO2 emissions (Kauppi & Tomppo, 1993; Bonan, 2008). In response to an increasing global energy demand this mitigation potential is applied by increasing the use of biomass from forestry for bioenergy (Burger, 2009; Aguilar et al., 2011). In this context, Europe has committed in the development of short-rotation coppice cultures (SRC) of fast-growing tree species, as poplar (Populus spp.) and willow (Salix spp.) (Kauter et al., 2003; Aylott et al., 2008; AEBIOM, 2011). It is therefore impor-

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ecophysiological characteristics controlling tree growth is required to optimize the wood production of high-density SRC under projected climatic conditions (Calfapietra et al., 2010). Increased drought due to climate change is predicted to reduce energy production and carbon mitigation for perennial energy crops, such as Miscanthus × giganteus and Phalaris arundinacea (Hastings et al., 2009; Zhou et al., 2011). On the other hand, drought tolerance improved under elevated atmospheric CO₂ concentrations in a number of lignocellulosic bioenergy crops (Oliver et al., 2009). Hence, the study of carbon allocation patterns to different tissue components throughout the growing season in relation to changing environmental conditions remains relevant (Kozlowski, 1992; Barbaroux et al., 2003; Campioli et al., 2011).

With the long-term goal of assessing whether SRC are efficient in mitigating climate change, an experimental SRC with poplar (POPFULL: http://webh01.ua.ac.be/popfull/) is being intensively studied in Flanders (Belgium). The objective of this study is to analyze the seasonal variations in net ecosystem exchange (NEE), photosynthesis and carbon allocation between organs in relation to environmental parameters in this type of high-density plantations. An atypical dry spring to summer period during the year of investigation (2011) provided the opportunity to examine the effect of drought on ecosystem dynamics. Given the increasing risk of summer droughts, ecosystem water use is gaining interest in climate change research. Divergent results have been reported in studies examining the controlling factors of ecosystem water use efficiency (WUE) (Krishnan et al., 2006; Reichstein et al., 2007a; Yang et al., 2010). In this context, environmental control of the plantation WUE was studied throughout the second growing season after planting. Because poplar is known for its vulnerability to drought (Rood et al., 2000; Marron et al., 2003; Larchevèque et al., 2011), we hypothesized that plantation productivity would decrease during the dry period (Souch & Stephens, 1998). An increased WUE (Monclus et al., 2006; Chamaillard et al., 2011) and root–shoot ratio were expected in response to dry conditions (Yin et al., 2005; Guo et al., 2010). Therefore, an in-depth data collection and analysis were performed using an integration of different hierarchical scales: leaf level, tree level and ecosystem scale.

Materials and methods

Study site

This study was carried out within an operationally managed bioenergy plantation of poplars, established in April 2010 in Lochristi, East-Flanders, Belgium (51°06′44″N, 3°51′02″E; 6.25 m above sea level). The total area of the site is 18.4 ha of former agricultural land consisting of cropland (65% of the surface area, recently grown with a monoculture of maize) and extensively grazed pasture (35% of the surface area). The long-term average annual temperature at the site is 9.5 °C and the average annual precipitation is 726 mm, equally distributed over the year. A detailed soil analysis prior to planting characterized the soil type as a sandy texture, with clay-enriched deeper soil layers. In the upper soil layer, carbon (C) and nitrogen (N) concentrations were significantly lower in former cropland as compared with former pasture, and decreased exponentially with depth in both former land-use types. For more information on the site and the soil characteristics, see Broeckx et al. (2012a).

Large replicated monoclonal blocks were established on a total of 14.5 ha, with 12 poplar genotypes representing different species and interspecific hybrids of Populus deltoides Marsh., P. maximowiczii Henry, P. nigra L. and P. trichocarpa Hook (Table 1). After soil preparation, 25 cm long dormant and unrooted hardwood cuttings were planted on 7–10 April 2010 at a density of 8000 ha⁻¹. The cuttings were planted in a double-row design with alternating distances of 0.75 m and 1.50 m between the rows and 1.1 m between the individuals within each row. Neither irrigation nor fertilization was applied. More details on the plantation establishment and on the layout can be found in Broeckx et al. (2012a). All measurements reported in this study were performed between January and December 2011, i.e. during the second growth year of the plantation. Depending on the study scale and on the measurement technique, the measurements were performed on a different number of genotypes, ranging from two genotypes for below-ground measurements to 12 genotypes for leaf area index (LAI) and ecosystem-scale measurements (Table 1). Although the number of genotypes varied, the choice of genotypes was nested hierarchically and always represented phenotypically and genetically contrasting genotypes.

Environmental parameters

Meteorological parameters were recorded half-hourly at the study site. Air temperature (Ta), relative humidity and precipitation were recorded on an extendable eddy covariance (EC) mast at 5.4 m above the ground surface using Vaisala probes (model HMP45C; Vaisala, Helsinki, Finland). The incoming short-wave radiation (SWR, 0.3–3 μm) was collected using a pyranometer (model CNR1; Kipp & Zonen, Delft, the Netherlands). Precipitation (P) was recorded using a tipping bucket rain gauge (model 3665R; Spectrum Technologies Inc., Plainfield, IL, USA), whereas the water table was recorded with a pressure transducer (model PDCR1830; Campbell Scientific, Logan, UT, USA) installed in a pipe inserted into the ground to a depth of 1.85 m. More information on the logging and the gap filling of the environmental factors can be found in Zona et al. (2012, 2013).

Soil water content (SWC, m³ m⁻³) was measured half-hourly using moisture probes (TDR model CS616; Campbell Scientific) placed at depths of 20 cm and 40 cm at one location close to the EC mast. As a complement, soil water potential (Ψₛ) was measured half-hourly from June to November 2011 using
Table 1  Overview of the 12 poplar genotypes planted in a short-rotation coppice culture. Parentage, genotype and measurements performed on each genotype have been indicated. Regression coefficients a and b as well as the coefficient of determination $R^2$ of the genotype-specific allometric relationship are also shown (DM = $a \cdot D^b$, where DM (dry mass) and D represent the aboveground woody biomass and the stem diameter at 22 cm respectively)

<table>
<thead>
<tr>
<th>Parentage</th>
<th>Genotype</th>
<th>a</th>
<th>b</th>
<th>$R^2$</th>
<th>Measurement</th>
</tr>
</thead>
<tbody>
<tr>
<td>T × M</td>
<td>Bakán †</td>
<td>0.1617</td>
<td>2.3966</td>
<td>0.9975</td>
<td>EC – LAI – bud phenology – stem diameter – $\Psi_S$ – leaf gas exchange</td>
</tr>
<tr>
<td>T × M</td>
<td>Skado †</td>
<td>0.6882</td>
<td>2.0298</td>
<td>0.9698</td>
<td>EC – LAI – bud phenology – stem diameter – $\Psi_S$ – leaf gas exchange – $R_{soil}$ – fine roots</td>
</tr>
<tr>
<td>D × N</td>
<td>Muur †</td>
<td>0.1237</td>
<td>2.4221</td>
<td>0.9944</td>
<td>EC – LAI – bud phenology</td>
</tr>
<tr>
<td>D × N</td>
<td>Oudenberg †</td>
<td>0.1799</td>
<td>2.3327</td>
<td>0.9985</td>
<td>EC – LAI – bud phenology – stem diameter – $\Psi_S$ – leaf gas exchange</td>
</tr>
<tr>
<td>D × N</td>
<td>Vesten †</td>
<td>0.0723</td>
<td>2.5741</td>
<td>0.9884</td>
<td>EC – LAI – bud phenology</td>
</tr>
<tr>
<td>D × N</td>
<td>Ellert †</td>
<td>0.1349</td>
<td>2.3850</td>
<td>0.9662</td>
<td>LAI – bud phenology</td>
</tr>
<tr>
<td>D × N</td>
<td>Hees †</td>
<td>0.2176</td>
<td>2.2785</td>
<td>0.9962</td>
<td>EC – LAI – bud phenology</td>
</tr>
<tr>
<td>D × N</td>
<td>Koster †</td>
<td>0.1517</td>
<td>2.3435</td>
<td>0.9988</td>
<td>EC – LAI – bud phenology – leaf gas exchange – fine roots</td>
</tr>
<tr>
<td>D × N</td>
<td>Robusta †</td>
<td>0.2004</td>
<td>2.2817</td>
<td>0.9969</td>
<td>LAI – bud phenology</td>
</tr>
<tr>
<td>D × (T × D)</td>
<td>Grimminge †</td>
<td>0.1270</td>
<td>2.4541</td>
<td>0.9976</td>
<td>EC – LAI – bud phenology – stem diameter – $\Psi_S$ – leaf gas exchange – $R_{soil}$</td>
</tr>
<tr>
<td>N</td>
<td>Brandaris †</td>
<td>0.1498</td>
<td>2.3478</td>
<td>0.9907</td>
<td>LAI – bud phenology</td>
</tr>
<tr>
<td>N</td>
<td>Wolterson †</td>
<td>0.1389</td>
<td>2.3804</td>
<td>0.9958</td>
<td>LAI – bud phenology – leaf gas exchange</td>
</tr>
</tbody>
</table>

D, Populus deltoides; M, Populus maxima; N, Populus nigra; T, Populus trichocarpa; EC genotypes highly represented in the footprint of the eddy covariance tower; LAI, leaf area index; $\Psi_S$, soil water potential; $R_{soil}$, soil respiration.

*Produced by INBO (Geraardsbergen, Belgium).
†Produced by Vermeerderingstuinen Nederland (Zeewolde, the Netherlands).

Calibrated equitensimeter probes (type EQ-2; Delta-T Devices Ltd, Cambridge, UK) installed at depths of 20 and 40 cm at four locations around the EC mast in four different genotypes (Table 1). We opted for characterizing soil water availability along the growing season through the time course of $\Psi_S$ values, averaged among the four locations. Therefore, $\Psi_S$ values (only available from June to November 2011) were extrapolated to the whole year based on the relationship observed between SWC and $\Psi_S$ measurements at each measuring depth. A feed-forward Neural Network (Matlab R2012a; Mathworks, Natick, MA, USA) was used to interpolate missing values. The correlation between predicted and measured values ranged between 0.83 and 0.98 for the different soil depths.

### Eddy covariance measurements

Ecosystem-scale CO$_2$, water vapour and energy fluxes were continuously (at 10 Hz) measured with standard EC techniques. The system included a sonic anemometer (Model CSAT3; Campbell Scientific) for the measurement of the three-dimensional wind components, wind speed, wind direction and the energy fluxes. The sonic anemometer and the inlets of the sampling lines were situated at 5.8 m above the surface during the first period (1 January 2011 to 31 August 2011); afterwards they were raised to 6.6 m (after 31 August 2011). The location of the mast was chosen according to the prevalent wind direction, to maximize the footprint of the EC mast, as described in Zona et al. (2012). A LI-7000 fast-response gas analyser (LI-COR; Lincoln, NE, USA) was used to measure CO$_2$ and latent heat (LE) fluxes. Latent heat flux was converted into evapotranspiration (ET) using $T_{\text{air}}$ and latent heat of vaporization ($\lambda$; Rebmann et al., 2012):

\[
ET = \frac{LE}{\lambda} \tag{1}
\]

Fluxes of CO$_2$, LE and sensible heat (H) were calculated using the EdiRe software (R. Clement, University of Edinburgh, UK) and averaged over 30 min. The Bowen ratio was defined as the ratio between H and LE (Bowen, 1926; Teuling et al., 2010). The ratio of ET over P was called the evaporative index (Williams et al., 2012). Half-hourly NEE data were partitioned into gross primary production (GPP) and ecosystem respiration ($R_{eco}$). Accordingly, the net ecosystem productivity (NEP) was defined as (Gower et al., 2001; Krishnan et al., 2006):

\[
NEP = GPP - R_{eco} \tag{2}
\]

A filter was used to set negative GPP values to zero. The ecosystem WUE was defined as the GPP/ET ratio (Krishnan et al., 2006; Granier et al., 2007; Reichstein et al., 2007a). For more details on the EC system and postprocessing of the data, see Zona et al. (2013).

### Soil CO$_2$ efflux

The CO$_2$ efflux from the soil, i.e. soil respiration ($R_{soil}$), was measured on an hourly basis by an automated soil CO$_2$-flux system (LI-8100; LI-COR). Sixteen chambers operating as closed systems were connected to an infrared gas analyser through a multiplexer (LI-8150; LI-COR). The chambers were located in the neighbourhood of the EC mast – within the footprint – and spatially distributed to cover both former land-use types in two genotypes (Table 1; Verlinden et al., 2013). The system was installed at the end of March 2011 and continuously logged soil CO$_2$ efflux for each chamber successively every hour until the
end of the year. Soil CO₂ efflux was extrapolated for January to March 2011 by a feed-forward Neural Network analysis (Matlab R2012a; Mathworks) based on the close relationship with soil temperature. The correlation between predicted and measured values ranged between 0.84 and 0.93 for the different measurement chambers. Soil temperature was monitored all year at 0–20 cm depth using a thermocouple (TCAV-L, Campbell Scientific) at one location close to the EC mast.

Leaf gas exchange and photosynthetic parameters

Leaf gas exchange measurements were performed with a portable open-path gas exchange measurement system (LI-6400; LI-COR) equipped with a leaf chamber fluorometer (LI-6400-40; LI-COR). Measurements were completed in monthly measurement campaigns from May to September 2011 on four replicate trees of six genotypes located on former cropland. All measurements were done in the upper canopy on the most recently matured leaf of the current-year shoot. Leaves were first acclimated for 10 min in the chamber at the atmospheric CO₂ concentration (400 ppm) and under a saturating photosynthetic photon flux density (PPFD) of 1500 μmol s⁻¹ m⁻². Afterwards light-saturated assimilation rate at atmospheric CO₂ concentration (Aₑ) and stomatal conductance (gₛₑ) was recorded before establishing the response of the net assimilation rate to varying intercellular CO₂ concentrations (A–Cᵢ curve). Each curve consisted of 10 steps of external CO₂ concentrations set in succession to 400, 300, 250, 150, 100, 50, 500, 750, 1000 and 1250 ppm. Leaves were allowed to equilibrate at least 3 min at each step before data were logged. All measurements were done at a constant block temperature (25 °C) and at a controlled vapour pressure deficit (VPD) of ca. 1 kPa (1.07 kPa ± 0.03, mean ± SE). Day respiration (Rₑₑₑ) was estimated from the A–Cᵢ curves by fitting the biochemical photosynthesis model of Farquhar (Farquhar et al., 1980).

Radial stem growth

Stem diameter at 22 cm above soil level was regularly measured for four genotypes between March and September 2011 with a digital caliper (0.01 mm precision, CD-15DC; Mitutoyo, Kawasaki, Kanagawa, Japan). Measurements were performed on five trees per genotype, chosen to represent the diameter distribution of each genotype using the quantiles of the total approach (Cermák & Michálek, 1991; Cermák & Kučera, 1994). As the effect of former land use on stem diameter diminished during the second growing season (Broeckx et al., 2012a), only stem growth of trees located on former cropland was monitored. Data are presented as the relative radial growth for 2011, i.e. stem diameter expressed relative to the end-of-season stem diameter.

Leaf area index and bud phenology

Canopy development was monitored by measuring LAI on a monthly basis from April to November 2011. Measurements were performed in four replicated measurement plots per former land-use type for each genotype. An LAI-2200 Plant Canopy Analyzer (LI-COR) with a 45° view cap was used, by comparison of above- and below-canopy readings. In each plot two diagonal transects were defined between the rows; measurements were then taken along each transect with the sensor parallel to the row and perpendicular to the row (cfr. Broeckx et al., 2012b). A weighted average LAI was calculated according to the ground surface area occupied by each genotype in the plantation at each monthly measurement.

For the assessment of the timing of bud break and bud set, a phenological score was assigned weekly to each of the 12 genotypes. A sigmoidal curve was then fitted through the evolution of weekly scores (Matlab R2012a; Mathworks) and used to calculate the day of the year at which the score – corresponding to the following descriptions – was reached: the timing of spring bud flush was defined according to the score describing the apical bud as ‘bud sprouting, with a tip of the small leaves emerging out of the bud scales, which couldn’t be observed individually’ (based on UPOV, 1981); the end of leaf production, associated with autumnal bud set and the end of stem growth height, was defined as the time when ‘an apical bud was present but not fully closed, bud scales were predominantly green and no more rolled-up leaves were present’ (Rohde et al., 2010). These phenological observations were used to define the length of the growing season as the time between bud flush and bud set, corresponding to the time between leaf appearance and maximum LAI (LAIₘₐₓ). In comparison, a second definition of the length of the growing season is used further in the discussion, i.e. the period with a net CO₂ uptake from the atmosphere as indicated by NEE<0 for a period of more than 1 week (carbon uptake period, CUP).

Fine root production

Root biomass was quantified every 2 weeks from February to November 2011. At each campaign 20 soil samples per genotype of the upper 15 cm soil layer were collected using a hand-driven corer (Eijkelkamp Agrisearch equipment, Netherlands; Oliveira et al., 2000). In the laboratory roots were then hand-picked, washed, sorted in poplar and weed roots and finally oven dried at 70 °C to estimate root biomass. In addition, poplar roots were classified into four diameter classes: <1 mm (class L1), 1–2 mm (class L2), 2–5 mm (class L3) and >5 mm (class L4). Dead roots were only observed in the <1 mm class (L1), and were separated from living roots based on the dark colour and the lack of cohesion of the periderm (according to Janssens et al., 1999). Belowground biomass production was constrained to poplar fine root biomass production. Fine roots were defined as roots with diameter <2 mm (classes L₁+L₂). More details on root collection and data processing can be found in Berhongaray et al. (2012).

Net primary productivity (NPP)

Biomass measurements of each tree component were used to calculate NPP in terms of g C m⁻² day⁻¹. For each tree component, monthly average NPP/GPP ratios were calculated. Standing biomass was estimated using allometric relationships

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established between aboveground woody dry mass (DM) and stem diameter (D):

\[ DM = a \cdot D^b \]  

(3)

where a and b are regression coefficients specific to each genotype (cfr. Laureysens et al., 2004). Ten trees per genotype, representing the population diameter range, were harvested at the end of the second growing season to establish genotype-specific allometric relationships (Table 1). Specific leaf area (SLA, the ratio between fresh leaf area and leaf DM), measured in September 2011 from six leaves of different size and tree height per genotype, was used to estimate leaf biomass from LAI measurements (Jonckheere et al., 2004). Biomass increment of stems and leaves was calculated as the difference between two consecutive biomass estimations and the time between the data collection. Fine root productivity between sampling dates was estimated using the decision matrix of Fairley & Alexander (1985). This method calculates production, mortality and decomposition of fine roots between consecutive sampling dates using fine root biomass and necromass. Carbon concentrations were determined from ground plant material by dry combustion (Carlo Erba Instruments, Rodano, Milan, Italy). Carbon concentrations were considered constant during the growing season, carbon concentration of DM being a conservative parameter (Vande Walle et al. 2005).

**Data integration and analysis**

Weekly averages were calculated for all continuously monitored parameters (NEE, GPP, \( R_{ec} \), \( R_{soil} \), \( \Psi_c \), \( T_{air} \), P, SWR, VPD and heat fluxes). In addition, midday fluxes of NEE, GPP, \( R_{ec} \) and VPD (constrained between 10 h and 16 h local time for the entire year) were used, as photosynthetic carbon uptake was restricted to daytime and as leaf and tree level measurements were performed within this time range. All continuous and discontinuous measurements were plotted over time to analyse seasonal trends and to identify possible patterns of interaction. Short-term regressions between continuous and derived parameters (GPP, SWR VPD and ET) were established for periods with a different water status: both a period with limiting water availability at 20 cm depth (15 May to 20 June 2011) and a period with low water availability at 40 cm depth (15 June to 20 July 2011) were compared to a period with ample soil water availability (August 2011). All plots and regressions were made in SigmaPlot Version 12.0 (Systat Software, San Jose, CA).

**Results**

**Seasonal variability**

Based on bud phenology measurements, the growing season started on 31 March 2011 (±11 days) – averaged over the different genotypes in the plantation – corresponding to leaf expansion and the onset of stem diameter increment (Fig. 1a). At the onset of the growing season, high NPP/GPP ratios for leaf and stem biomass were found with maxima of 24% in March and 21% in April respectively. In terms of ecosystem CUP the growing season started on 31 March 2011, in accordance with the first definition. A short period of net carbon uptake (by definition excluded from the CUP) occurred earlier, starting on 13 March, and net carbon uptake temporarily decreased to zero around 31 March. A rather symmetrical seasonal NEP pattern was observed, slightly shifted towards the left, i.e. towards the beginning of the growing season (Fig. 2a).

Stem diameter increment, reflecting stem biomass production, stopped in September 2011 (Fig. 1a). No more leaves were produced from 23 September (±7 days) onwards – averaged over the plantation – once LAI had reached a maximum. The average plantations \( LAI_{max} \) was rather low (\( LAI_{max} = 2.63 \); Fig. 1a). An increase in fine root production was concomitantly observed in September (Fig. 3). Overall, the above- to belowground productivity ratio decreased during the growing season (Fig. 3). The ecosystem stopped being a carbon sink on 1 October (Fig. 2a). Over the whole year, the poplar SRC was a moderate carbon sink of 95.7 g C m\(^{-2}\) (±15.3) (Zona et al., 2013). Leaf, stem and fine root dry biomass production corresponded to 2 Mg ha\(^{-1}\) yr\(^{-1}\), 5.8 Mg ha\(^{-1}\) yr\(^{-1}\) and 0.5 Mg ha\(^{-1}\) yr\(^{-1}\) respectively. It is worth noting that the photosynthetically active season, as defined by CUP, ended shortly after bud set (Fig. 1a).

On a yearly basis, \( R_{soil} \) contributed 54% to \( R_{ec} \) (Fig. 2b). A decreasing trend in the contribution of \( R_{soil} \) to \( R_{ec} \) was, however, observed towards the end of the year. A significant and positive linear correlation was observed between SWR and \( T_{air} \) which in turn positively affected VPD. Consequently, a closely related seasonal time course was observed between SWR and VPD (Fig. 4a). VPD and GPP showed a reversed seasonal pattern during the growing season, reflecting a reduced carbon uptake at high VPD values. Overall, the ecosystem WUE increased during the growing season (Fig. 5a).

**Dry spring conditions**

Spring 2011 was particularly dry at our site. The monthly precipitation at the site in March, April and May represented only 26%, 16% and 25% of the long-term average monthly precipitation, respectively, as measured by the Royal Meteorological Institute (Ukkel, 50° 47’ 55”N, 4° 21’ 29”E). The monthly precipitation amounts measured in Ukkel were statistically characterized as exceptional (i.e. occurring once every 30 years) for March 2011, normal for April 2011 and very atypical
(i.e. occurring once every 10 years) for May 2011 (Royal Meteorological Institute, http://www.meteo.be/meteo/view/en/65239-Home.html). Low precipitation was mirrored by a progressive decrease in water table of almost 1 m, i.e. from 45 cm below the soil surface at the beginning of March to a minimum of 142 cm below the soil surface at the beginning of July 2011 (Fig. 4b). However, the effects on soil water potential ($\Psi_S$) were visible only from May onwards continuing until July (Fig. 4c). Values of $\Psi_S$ reached a minimum of −1.8 MPa at 20 cm depth in the beginning of June and a minimum of −1.2 MPa at 40 cm depth in mid-July (Fig. 4c). These values were close to and even exceeded the wilting point (i.e. at $\Psi_S$ of −1.6 MPa; Granier et al., 2007; Migliavacca et al., 2009). Given this naturally occurring drought, we acknowledge that we studied the interacting effect of soil water availability and seasonality, in absence of an irrigated control plot.

A decrease in leaf NPP and consequently a decreased leaf growth rate were observed in May and June 2011 (Figs 1b and 3), when soil water availability was low in the near-surface soil layer. The reduced leaf area development was accompanied by a decrease in midday GPP and NEE (Fig. 1b). NEE and GPP indeed showed a similar seasonal time course as the evolution of LAI during the growing season. This seasonal trend was less pronounced in midday $R_{eco}$ (Fig. 1b). At the leaf level, net assimilation rate ($A_{sat}$) was reduced in response to low water availability in the near-surface soil layer while day respiration ($R_{day}$) was not affected (Fig. 6a). A temporary return towards carbon neutrality occurred in August when the ecosystem was recovering from drought (Fig. 2a and d). Simultaneously with the decreasing leaf production, fine root productivity reached a temporary maximum in June (Fig. 4), indicating a stimulation of fine root production in response to limiting water availability in the near-surface soil layer. In contrast, stem biomass growth was only slightly affected by the dry conditions (Fig. 1a), as supported by the seasonal pattern in stem NPP (Fig. 3).
A decrease in GPP was observed towards the beginning of June (Fig. 2d), in response to decreasing $\Psi_S$ at 20 cm depth (Fig. 4c). GPP was not responsive to VPD when water availability was limiting at 20 cm depth (Fig. 7a). A modest increase in the Bowen ratio with decreasing $\Psi_S$ at 20 cm depth was observed (Fig. 5a). This was caused by an increasing $H$ and decreasing LE (Fig. 2c). A rapid saturation of ET with VPD occurred at low water availability in the upper 20 cm soil layer (Fig. 7b). A local maximum in WUE was reached shortly before the minimum $\Psi_S$ at 20 cm depth (Fig. 5a). At the leaf level, a simultaneous reduction in $g_{\text{sat}}$ and $A_{\text{sat}}$ was observed towards the beginning of July (Fig. 6a and b). A similar, but less pronounced, spring reduction was found in leaf nitrogen (N) concentration (Fig. 6b). 

An increase in GPP was observed in response to the June rainfall event (Figs 2d and 4b) that caused an increase in $\Psi_S$ at 20 cm depth (Fig. 4c), although the water table did not change at this time (Fig. 4b). Simultaneously, $\Psi_S$ at 40 cm depth was decreasing as a delayed effect of the spring dry period by depletion of deeper soil water (Fig. 4c). A peak in ET and evaporative index corresponded to the decreasing soil water availability at 40 cm depth (Fig. 5b). A second decrease

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in GPP was observed during the intense rainfall event in the beginning of July (Figs 2d and 4b). This sudden rainfall caused an increase in water table and a recovery of $\Psi$ back to 0 MPa (Fig. 4c). A strong decrease in SWR and $T_{\text{air}}$ was observed consistent with reduced GPP (Fig. 4a and d). A trend of increasing GPP with $T_{\text{air}}$ and SWR was observed when soil water availability was low at 40 cm depth (Fig. 7c and d).

**Discussion**

**Growing season patterns**

High NPP/GPP ratios for leaf and stem diameter in the beginning of the growing season are consistent with previous findings for beech (Campioli et al., 2011). This reflects the growth strategy of deciduous trees in temperate regions, maximally exploiting optimal spring growth conditions (Campioli et al., 2011). An early net carbon uptake event of <1 week from 13 March onwards was caused by increasing GPP. This early ecosystem carbon uptake reflected the photosynthetic carbon uptake by the understory vegetation at one hand, and the genotypic variation in timing of bud flush on the other hand. The ecosystem carbon uptake started when environmental conditions favoured photosynthesis and strongly increased when the earliest genotypes started to flush leaves. A temporary decrease to zero in net carbon uptake around 31 March was explained by reduced SWR and hence reduced GPP, in addition to increasing respiration at the very beginning of the growing season, offsetting GPP. High photosynthetic rates in the beginning of the growing season resulted in high NEP, when SWR was high and $R_{\text{soil}}$ was still low because of the low soil temperature (Verlinden et al., 2013). This observation was previously found for temperate deciduous forests (Falge et al., 2002).

The timing of LAI$_{\text{max}}$ near the end of the growing season reflects the indeterminate growth habitus of poplar (Howe et al., 2000). A similar seasonal LAI development was observed in the perennial energy crop switchgrass (Albaugh et al., 2012). A typical temperate annual cycle was observed with a fast increase in NEP in spring associated with photosynthetic C uptake (Krishnan et al., 2006). A very close relationship was observed when comparing both definitions of the duration of the growing season (i.e. the time between bud burst and bud set vs. the CUP), although they address processes at different scales. The increasing fine root production in September could indicate a shift in carbon allocation from aboveground biomass to belowground biomass towards the end of the growing season (Scarscia-Mugnozza, 1991; Dickmann & Pregitzer, 1992), as evidenced by a decreasing above- to belowground productivity ratio. Root growth generally continues longer than shoot growth even after leaf abscission (Lyr & Hoffmann, 1967; Cannell & Willet, 1976). The fact that root growth is favoured over shoot growth at the end of the growing season has been previously reported not only for mature forests (Burke & Raynal, 1994) but also for young poplar plantations (Heilmann et al., 1994).

The low LAI$_{\text{max}}$ suggested a rather open stand where a fraction of the net radiation reached the soil surface (cfr. Granier et al., 2000). The coinciding seasonal course of SWR and soil heat flux (data not shown) suggested that at least part of the solar energy was indeed absorbed by the soil in our plantation. Total ecosystem ET in open stands is generally not proportional to tree LAI and hence to tree transpiration (Granier et al., 2000), which was mirrored in the different seasonal time courses of ET and LAI. Canopy openness had therefore promoted an increase in transpiration of the understory weed vegetation with incoming radiation (cfr. Granier et al., 2000), especially in the beginning of the growing season when LAI was low and incoming SWR was rather high.

The observed contribution of $R_{\text{soil}}$ to $R_{\text{eco}}$ (54%) was in the lower end of the range reported for European forest ecosystems (Janssens et al., 2001), although this plantation was recently established and accompanied by severe soil disturbance due to ploughing. As roots, litter and soil microbial activity are the main contributors to soil respiration, the young age of the plantation
and the still developing root system likely explained this low proportional contribution. A positive correlation between $R_{\text{soil}}$ and root biomass was observed in the plantation (Verlinden et al. 2013) with low root biomass values compared with other studies (Vande Walle et al., 2007; Abou Jaoudé et al., 2011). Several factors may explain the decreasing trend of this relative contribution, including (i) the limited leaf fall in the soil chambers causing a higher contribution of leaf decomposition in the plantation as compared with the chambers; (ii) a higher ecosystem respiration at the end of the year due to an increase in wood volume and an increase in perennial weed biomass as compared with the beginning of the season and (iii) a low soil respiration at the end of the year due to a high local water table and a low soil temperature possibly underestimating the contribution of $R_{\text{soil}}$ to $R_{\text{eco}}$. The first factor could be considered an artefact of the limited chamber area of the $R_{\text{soil}}$ measurements. The effect of water table was previously described (Verlinden et al. 2013), with water saturated soils causing surface sealing and anaerobic conditions, and hence reduced diffusion of gases and of microbial

Fig. 4  Time course in 2011 of meteorological parameters. (a) Vapour pressure deficit (VPD) and short-wave radiation (SWR). (b) Precipitation (P; hatched bars) and water table (black dots) in cm below the surface. (c) Soil water potential ($\psi_s$) at 20 and 40 cm soil depths. (d) Air ($T_{\text{air}}$) and soil temperature ($T_{\text{soil}}$) in the upper 20 cm soil layer. Vertical dotted and dashed lines represent the minimum soil water potential at 20 cm depth (before June rainfall events) and restoration of soil water potential at 20 cm depth (before July heavy rainfall event) respectively.
activity. Earlier studies also reported an exponential response of soil respiration to soil temperature and a decrease in soil respiration at low and high SWC (Vincent et al., 2006; Lee et al., 2010), affecting seasonality of $R_{\text{soil}}$.

Impact of the dry spring

The minimum values of $\Psi_S$ reached in the plantation suggested that the trees were potentially suffering from drought, affecting several plant- and ecosystem-scale processes. Over the growing season (April–September), cumulative ET (284 mm) was more than compensated by cumulative rainfall (360 mm). However, seasonal variations were observed, with an average evaporative index of 1.62 during the first half of the growing season (April–June) indicating drought, as compared to 0.52 during the second half of the growing season (July–September). Seasonal variation in the evaporative index has already been related to lower ET values in drier periods in a switchgrass bioenergy plantation (Wagle & Kakani, 2012).

Effects on net primary productivity

The observed decrease in leaf NPP and hence in leaf growth rate in response to low water availability were reported in previous studies: a decreased LAI was found in container-grown hybrid poplar under drought conditions (Souch & Stephens, 1998), in willow SRC stands in response to soil water deficits (Lindroth et al., 1994) as well as in other perennial energy crops such as several Miscanthus species, reed canary grass and sugarcane (Smit & Singels, 2006; Zub & Brancourt-Hulmel, 2010; Zhou et al., 2011). A decreased maximum individual leaf area in response to drought has also been reported in hybrid poplar (Monclus et al., 2006, 2009), confirming that leaf production and expansion are very sensitive to drought in fast-growing species such as poplar and willow. The lower correspondence in sea-

Fig. 5 Time course in 2011 of (a) water use efficiency (WUE) and Bowen ratio [ratio between sensible (H) and latent (LE) heat flux] and of (b) evapotranspiration (ET) and evaporative index [ratio between ET and precipitation (P)]. Vertical dotted and dashed lines represent the minimum soil water potential at 20 cm depth (before June rainfall events) and restoration of soil water potential (before July heavy rainfall event) respectively.
sonal time course of \( R_{\text{eco}} \) compared to GPP with the evolution of LAI was consistent with previous observations of a stronger response of GPP to increasing drought in European forests (Granier et al., 2007). Similar observations of a larger effect of soil water availability on gross ecosystem productivity than on \( R_{\text{eco}} \) were already reported for a hybrid poplar plantation in the southern boreal forest region of Canada (Cai et al., 2011).

Observations of increased carbon allocation to the roots in response to water stress were previously reported for poplar seedlings (Zhang et al., 2004; Yin et al., 2005; Guo et al., 2010), as well as for other tree species (Tomlinson & Anderson, 1998; Leuschner et al., 2001). However, during the first year of their experiment on water use in hybrid poplar, Souch & Stephens (1998) found no effect of drought on biomass partitioning between roots, stems or branches. A reduced allocation to fine roots in the upper soil layers in response to low soil moisture has been reported before in poplar (Dickmann et al., 1996) and in balsam fir (Olesinski et al., 2011). Older trees have fewer possibilities and needs for expansion of their root systems, but in newly planted trees root growth tends to explore the soil volume and to increase the ability to compete with neighbouring plants. Consequently, root growth uses reserves during periods of low supplies of photosynthates (Persson, 2002). In poplar, active root growth in response to a declining water table might be an adaptive trait related to its natural riparian habitat (Rood et al., 2003).

Effects on ecosystem and leaf level gas exchange

Increasing \( H \) when water availability was limiting at 20 cm depth suggested that the higher energy load from incoming radiation increased leaf temperature and hence sensible heat, due to reduced transpiration. Reduced photosynthetic rates, induced by a decrease in \( g_{\text{sat}} \) with increased VPD were previously observed in \( P. \) tremuloides stands (Hogg et al., 2000). The similar reduction in stomatal conductance and \( A_{\text{sat}} \) towards the beginning of July implied a strong stomatal control upon photosynthesis (Regier et al., 2009; Larchevêque et al., 2011). The high \( T_{\text{air}} \) during this period and the limited GPP even at low VPD emphasized that water availability was the limiting factor. These results were supported by a rapidly saturating ET with increasing VPD when soil water availability was low in the upper 20 cm. Decreasing photosynthetic rates when soil water potential was below the wilting point in the upper 20 cm soil layer (−1.8 to −1.6 MPa) suggested that water and nutrient uptake occurred in this near-surface soil layer where a significant part of fine roots occurred (cfr. Al Afas et al., 2008; G. Berhongaray, unpublished results). Sufficient water availability at 40 cm depth (0 to −0.3 MPa) indicated that deeper roots could not support the necessary water uptake for the whole system at
this time, in contrast to previous studies (Lyr & Hoffmann, 1967; Rood et al., 2003). The increasing ecosystem WUE with decreasing soil water availability and decreasing $g_s$ was consistent with earlier studies (Baldochci, 1997; Williams et al., 1998; Krishnan et al., 2006).

Reduced leaf N concentration due to water stress was previously reported for poplar, in combination with a reduced net assimilation rate and $g_s$ (Yin et al. 2009). Reduced leaf N concentration may have been one cause of reduced NPP considering the positive correlation between N content and fresh stem biomass as previously reported by Monclus et al. (2009). However, the minimum N concentration in the leaves of more than 2% suggested that N was not the limiting factor of $A_{sat}$.

The reduced leaf N concentration during the spring dry period could have been caused by a lower N uptake due to either (i) a reduced transpiration caused by stomatal closure; (ii) a reduced soil N availability (Larsen et al., 2011) and/or (iii) a reduced uptake capacity of the roots (Kreuzwieser & Gessler, 2010) in response to low water availability. The hypothesis of a limitation in N availability was supported by the progressive decrease in N$_2$O emission and the occurrence of N$_2$O uptake at the end of the growing season 2011 (Zona et al., 2012). A similar limitation in N uptake during drought showed from an increased effect of N input on the yield of Miscanthus spp. under water stress (Zub & Brancourt-Hulmel, 2010).
The constant Bowen ratio during the June rainfall event reflected high transpiration rates and less sensible heat, indicating a reduced stomatal control despite low \( A_{\text{sat}} \) and \( g_{e,\text{sat}} \). This was supported by a sharp increase in ET, associated with a peak in the evaporative index. A short-term increase in \( g_{e,\text{sat}} \) and hence tree transpiration in response to rainfall as also observed by Granier et al. (2007) could explain this high carbon uptake – that could not be observed due to the discontinuous leaf gas exchange measurements – in combination with evaporation of water intercepted by the canopy. The maximum GPP reached by the end of June was associated with an increase in water intercepted by the canopy. The maximum GPP, previously observed in a boreal aspen stand (Hogg et al., 2007), was consistent with reduced GPP suggested temperature as the limiting factor of photosynthetic carbon uptake at that time. These results were confirmed by the stronger control of GPP by air temperature when water was non-limiting in the root zone. Both stomatal control due to water limitation in the superficial root zone and increased VPD reduced the effect of temperature on GPP. This observation was supported by a faster saturation of GPP with increasing SWR when water was more limiting. A stronger control of solar radiation on the carbon assimilation rate as compared with VPD was previously observed in a boreal aspen stand (Hogg et al., 2000), with sufficient water availability. Similarly, Reichstein et al. (2007b) reported a significant correlation between GPP and water availability in more water-limited (Southern European) sites, whereas in rarely water-limited (Northern European) sites a significant correlation between GPP and \( T_{\text{air}} \) was found.

In this study we analysed the seasonal dynamics in \( \text{CO}_2 \), water and energy fluxes during the second growth year of a poplar bioenergy plantation. The results showed that the plantation was already a modest carbon sink at this time, with the CUP closely corresponding to leaf area development up to \( \text{LAI}_{\max} \). The unusually dry period that occurred in spring allowed us to evaluate the impact of low water availability on ecosystem productivity. As we hypothesized, a reduced leaf area development was found in response to low water availability in the near-surface soil layer. High SWR and VPD induced strong stomatal control on carbon uptake when soil water availability was limiting in the upper soil layer. An increase in ecosystem WUE was observed in association with stomatal closure. Decreasing SWR and \( T_{\text{air}} \) were the limiting factors of photosynthetic carbon uptake when water was not limiting in the root zone. These results indicate that an increasing frequency and intensity of summer heat and drought could affect the productivity of recently established bioenergy plantations with fast-growing trees, when the developing root system is not able to provide the necessary water from deeper soil layers.

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**References**


