



## Biophysical interactions in a short rotation willow intercropping system in southern Ontario, Canada

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### ABSTRACT

Recently, there has been interest in shifting to carbon-neutral sources of energy, including bioenergy from short rotation woody crops. This study looked at the growth and yields of short rotation willow in an agroforestry intercropping system compared to a conventional single variety plot system used as a control. Three willow clones (*Salix dasyclados* SV1, *Salix miyabeana* SX67 and *Salix purpurea* 9882-41) were established in each field setup, where in the agroforestry field willow plots were located between 15 m wide rows of 20-year-old mixed tree species. Differences in photosynthetic photon flux density (PPFD), soil temperature, soil moisture and soil/foliar nitrogen between the two field setups were investigated from 2006 to 2007. Willow yields were significantly higher in the agroforestry fields during both years of the study, with 0.8 and 0.5 odt ha<sup>-1</sup> for the agroforestry and control fields, respectively, in 2006, and 3.0 and 1.1 odt ha<sup>-1</sup> respectively, in 2007. There were opposite trends in clonal yields between the two field setups in 2006, but in 2007, clonal yields were in the same order across fields with averages of 2.8, 2.2 and 1.2 odt ha<sup>-1</sup> for SV1, SX67 and 9882-41, respectively. Daily average photosynthetic photon flux density was 210 μmol m<sup>-2</sup> s<sup>-1</sup> (16%) lower in the agroforestry system, and PPFD was correlated with soil temperatures that were on average 0.4 °C and 2.7 °C lower in the agroforestry field in 2006 and 2007, respectively ( $r = 0.82$  and  $0.93$ ). Soil temperatures were negatively correlated with soil moisture levels that were on average 1.4% and 1.9% higher in the agroforestry field in 2006 and 2007, respectively ( $r = -0.54$  and  $-0.41$ ), and soil moisture content was positively correlated with willow yields ( $r = 0.49$  and  $0.72$ ). There was less soil available nitrogen in the agroforestry field, but no difference in foliar nitrogen between fields. An experiment excluding root competition in the top 1 m of soil between intercropped trees and willows in the agroforestry field found no significant competition for soil moisture or nitrogen in the first two years of growth. Results of this study suggest that moderate shading in an intercropping setup can result in a buffering effect on microclimate conditions, where there is less variation in soil moisture content and soil temperature across a range of weather conditions.

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### 1. Introduction

The Ontario greenhouse industry, worth about US\$ 2.3 billion per year, is seeking alternative energy sources to oil and natural gas, since about 25% of the operating costs for greenhouses are for heating (Picchi et al., 2006). A feasibility report by Picchi et al. (2006) indicated a large potential for short rotation willow (in the form of wood pellets or wood chips) to be used by greenhouse centres in Niagara and Essex counties in southern Ontario. A maximal radius of 75–100 km from either of these centres is estimated to be the “economic growing zone”, or a cost-effective

distance to transport woody feedstock. Within the economic growing zone, there are about 170,000 ha of suitable land to grow short rotation willow. Much of this potential land is old tobacco land (Canada Land Inventory [Agriculture] Class 3 or 4 land with sandy soils) which is marginal by most standards and is subject to considerable erosion (Picchi et al., 2006).

Recently, there has been interest in using agroforestry systems for short rotation willow production, mainly because there are large amounts of agriculturally exhausted land in industrialized countries, and agroforestry systems have been very successful with production on marginal land in tropical and subtropical climates (Gruenewald et al., 2007). Tree-intercropping is a specific type of agroforestry practice which incorporates alternating rows of trees and other crops (e.g. short rotation willow). Agroforestry intercropping allows for structural and functional diversification of

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agricultural systems through, for example, different age structures and harvest rotations lengths (e.g. 3-year harvests of willow and 20–40 year harvests of higher value hardwood), and through incorporation of nut/fruit-bearing or syrup-producing tree species as an intercrop. In addition, the presence of older trees in the landscape can enhance soil productivity over the longer term by slowing erosion, stabilizing soil structure, and balancing nutrient inputs and outputs (Schroth, 1995; Gordon and Newman, 1997; Williams et al., 1997; Nair and Graetz, 2004; Thevathasan and Gordon, 2004; Thevathasan et al., 2004; Reynolds et al., 2007).

Results from experiments conducted at the University of Guelph Agroforestry Research Station over the last 20 years suggest that in an agroforestry intercropping system where tree rows are spaced 15 m apart (with crops grown in the alleyway between tree rows), a 2 m wide buffer is needed between trees and crop plants to avoid direct competition for light, moisture and nutrients. In this system, the central 11 m of the alleyways is used to grow crop plants, and this zone is characterized by favourable growing conditions where the following processes are enhanced: nutrient cycling, nitrogen mineralization, soil organic carbon addition, earthworm activity, and carbon assimilation (Thevathasan and Gordon, 2004; Reynolds et al., 2007). Trials with agricultural crops have found that yields of C<sub>3</sub> plants such as winter wheat and winter barley in intercropping systems have been generally the same as those in monocrop systems (per unit area of crop), even producing higher yields during times of drought (Thevathasan and Gordon, 2004).

For this study, three willow clones were grown in both an agroforestry intercropping system where willow plots were intercropped with 20-year-old mixed tree species, and in a conventional monocrop system consisting of single-variety plots which was used as a control. In the agroforestry system, willow plots were located in the central 11 m of 15 m wide intercropping alleyways. The goal of this study was to compare willow growth and yields between field setups (and clonal varieties within each setup), and to attempt to explain growth and yield differences through two of the primary drivers of growth: light levels (photosynthetic photon flux density) and soil moisture content. Correlation analyses of data from the 2006 and 2007 seasons looked at relationships between photosynthetic photon flux density (PPFD), soil temperature and soil moisture content to see whether differences in PPFD may be primarily responsible for differences in soil moisture content between a monocrop (control) and an agroforestry intercropping setup. Photosynthetic light saturation curves were also generated for the three willow clones to understand how lower light levels might affect rates of photosynthesis of willows in an agroforestry setup. In 2007, a root barrier experiment was set up in the agroforestry field to determine whether a 2 m buffer between willows and intercropped trees was enough to avoid significant root competition for soil moisture and nitrogen. It was hypothesized that there would be no significant root competition between willows and intercropped trees in the agroforestry setup, and that moderately lower light levels in the agroforestry setup would contribute to lower soil temperatures and higher soil moisture levels, which may improve willow growth and yields.

## 2. Methods

### 2.1. Site description

The field sites were located at the University of Guelph's Agroforestry Research Station in Guelph, Ontario (latitude 43°32'28" N, longitude 80°12'32" W) on Class 3–4 land according to the Canada Land Inventory (Agriculture) designation. The soils were classified as Gray Brown Luvisols (Canadian soil classification

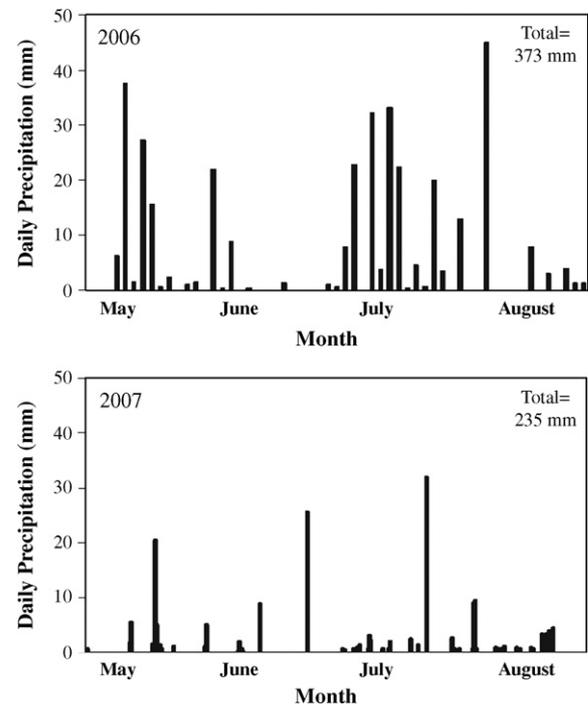


Fig. 1. Daily average precipitation in 2006 and 2007.

system) or Haplic Luvisols (WRB/FAO classification system) with a fine sandy loam texture (Haynes, 1998). The study was conducted from 2006 to 2007, where the 2006 season was much cooler and wetter than 2007, with a mean daily temperature of 12.5 °C and mean daily precipitation of 3.03 mm between May and August (Guelph Turfgrass Institute Weather Station). In 2007, the mean daily temperature was 17.5 °C and mean daily precipitation was 1.92 mm over the same time period (Fig. 1).

Two separate fields (agroforestry and control/monocrop) were used for the study. For comparison of soil parameters which may have influenced productivity between the two fields, an analysis of soil texture, inorganic and organic carbon fractions was done at 0–30 cm depth (Table 1). Soil carbon samples ( $n = 144$ ) were analyzed using a Leco<sup>®</sup> Carbon Determinator CR-12 (Model No. 781-700, Leco Instruments Ltd.) following the dry combustion technique (Carter and Gregorich, 2008). Soil pH (water basis) was measured in 2007 ( $n = 36$ ) using standard methods (Carter and Gregorich, 2008), and electrical conductivity was also measured in 2007 ( $n = 36$ ) using an HH2 WET Sensor (Delta-T Devices Ltd.).

### 2.2. Field design

The field design was two-factor factorial where factor one was tree-intercropping (present or absent), and factor two was willow clone (three clones). Plots (11 m × 50 m) of each of the three willow clones (*Salix dasyclados* SV1, *Salix miyabeana* SX67 and *Salix purpurea* 9882-41, obtained from the State University of New York's College of Environmental Science and Forestry) were arranged randomly within each field with four replications, for a total of 12 plots in each field. Willow plots consisted of five double rows, with alternating inter-row distances of 0.75 m and 1.5 m. Spacing between cuttings in a row was 0.55 m, resulting in a planting density of approximately 20 000 stems ha<sup>-1</sup>.

In the agroforestry field, trees intercropped with willow plots consisted of 20-year-old mixed tree species (predominantly *Juglans nigra* L. with some *Quercus rubra* L., *Fraxinus Americana* L. and *Robinia pseudoacacia* L.) which were planted in 370 m long rows with 5 m spacing between trees in a row, and 15 m width

**Table 1**  
Summary of soil characteristics (0–30 cm depth) from the two field sites.

Field	Texture: sand, silt, clay (%)	Carbon: total, inorganic, organic (%)	pH	Electrical conductivity (mS m <sup>-1</sup> )
Agroforestry	51.1, 34.7, 14.2	3.15, 1.84, 1.31	7.4	95.8
Control	53.4, 33.0, 13.6	3.29, 1.86, 1.43	7.5	89.9

None of the measured soil parameters were significantly different between the two fields (*t*-test, *p* > 0.05).

between tree rows (the alleyway). Tree rows were oriented northwest to southeast. Willow plots were located within the alleyways between the large tree rows with a 2 m buffer on either side, placing them in the central 11 m of the 15 m wide alleyways. The control field had no established tree rows, and willow plots were separated by at least a 2 m buffer.

### 2.3. Field establishment and management

On 3 May 2006, 18–25 cm long willow cuttings were planted flush to the ground using a Step Planter (Salix Maskiner AB, Sweden). Both fields were fertilized at the beginning of the second growing season (May 2007) with 75, 25 and 50 kg ha<sup>-1</sup> of N, P and K, respectively. Fertilization rates followed general recommendations by Adegbi et al. (2001) based on rates of nitrogen removal by willow crops at similar planting densities, with higher P and K inputs based on observations of growth of other crops at the field sites.

Pre-emergent weed control during 2006 was achieved using Dual<sup>®</sup> II Magnum<sup>®</sup> (Syngenta Canada) applied at a rate of 0.92 kg ai ha<sup>-1</sup> (S-metalachlor). For the 2007 season, Goal 2XL<sup>®</sup> (Dow AgroSciences) was used for pre-emergent control at a rate of 1.12 kg ai ha<sup>-1</sup> (oxyfluorfen). As weeds emerged later in the season each year, weed control was done by hand and using a tractor-drawn disker and rototiller.

Willows were harvested at 2–4 cm height after the first growing season using a sickle bar mower (Gaspardo, Italy).

## 2.4. Measurements and experiments

### 2.4.1. Willow growth and yields

To estimate willow yields at the end of 2006, a 3 m<sup>2</sup> area (12 individuals) at each of two random locations within each plot was harvested (cut 2–4 cm above the soil surface) (*n* = 576). In 2007, a 1 m<sup>2</sup> area (4 individuals) at each of three random locations in each plot was harvested (*n* = 288). Dry weight for each individual was measured by weighing the stems after drying in an oven at 65 °C for one week. Yields in odt ha<sup>-1</sup> (oven-dried tonnes per hectare) were calculated using all individuals sampled within a quadrat as a single average estimate per unit area, then converted to yields per hectare.

Percent survival was measured by counting the number of surviving plants (from establishment in May 2006) along three rows in each plot in September of 2006 and 2007. Percent survival was not factored into yield calculations, since destructive harvesting at the end of each season was randomized and included areas with willow mortality.

Willow height was tracked throughout each growing season on the same individuals harvested for yield estimates at the end of each year. Number of stems per individual was also recorded for harvested willows.

Leaf area index (LAI) was measured in August of both years. In 2006, destructive measurements were taken on five randomly sampled individuals within each plot (*n* = 120) using a LI-3100 Leaf Area Meter (LI-COR Biosciences, USA). In 2007, LAI was measured directly using an AccuPAR LP-80 meter (Decagon Devices Inc., USA) in 9 systematic locations per plot in each field (*n* = 216).

### 2.4.2. Photosynthetic photon flux density

Photosynthetic photon flux density measurements were done above the willow canopy under sunny, clear conditions on 1 August 2006, 30 May 2007, and 30 July 2007 at 9 a.m., 12 p.m., 3 p.m. and 6 p.m. at nine pre-determined points within each plot using a handheld Quantum Meter (Apogee Instruments Inc., USA). PPFD measurements in 2007 were averaged across the two sampling dates.

### 2.4.3. Photosynthetic light saturation curves

Photosynthetic light saturation curves were generated for the three willow clones using automatic settings on a LI-COR 6400 Portable Photosynthetic System (LI-COR Biosciences, USA). Measurements were taken on three individuals of each clone in each field, using the most fully expanded leaves the furthest up the stem (*n* = 18). Measurements were taken on 24 July 2007 between 9 a.m. and 11 a.m. During sampling, the average air temperature was 26.7 °C, and relative humidity was 53%. Lines of best fit for the curves were calculated using GraphPad Prism: Scientific graphing, curve fitting and statistical tool (Version 5, GraphPad Software, San Diego, CA, 2007). Data points were fit to the asymptotic model  $y = A + B \times \exp(C \times x)$  (Zheng et al., 2006).

### 2.4.4. Soil temperature

Soil temperature was measured at 15 cm depth continuously in 30 min intervals at twelve locations in each field from June to August of both years using HOBO<sup>®</sup> H8 Outdoor 4-Channel External Logger sensors (Onset Computer Corporation, USA). In 2006, measurements were taken in transects across willow plots; in the agroforestry field this looked at the effect of shading gradients in the agroforestry alleyways (centre vs. edge comparison). In 2007, sensors were located at each of three randomized locations within each plot where soil moisture/nitrogen and willow growth measurements were taken.

### 2.4.5. Soil moisture

Soil samples for moisture content were collected by hand using an auger at 0–30 cm depth from May to September in both years. In 2006, samples were taken in three systematic locations per plot (diagonally across each plot) on a weekly basis; this pattern in the agroforestry field allowed for a test of the effect of intercropped tree rows on soil moisture. In 2007, soil samples were taken bi-weekly at each of three random locations per plot where soil temperature and willow growth measurements were taken. A 10 g subsample was analyzed for gravimetric moisture content using the oven-dry method with a 48-h drying time at 105 °C (Reynolds, 1970).

### 2.4.6. Soil and foliar nitrogen

Soil available nitrogen (nitrate and ammonium) was measured on the same samples taken for soil moisture analyses in both years of the study. Samples were frozen until analysis, where 20 g subsamples underwent KCl extraction (Keeney and Nelson, 1982), and the extracts were analyzed using a Technicon<sup>®</sup> Autoanalyzer<sup>®</sup> II (Mandel Scientific Company Ltd., Canada). Only 2007 soil samples were analyzed for nitrogen.

To understand whether soil nitrogen differences were actually affecting willow growth, leaf samples for foliar nitrogen were collected on two sampling dates in July and August 2007. Two to three leaves from one stem of each individual in the middle six rows of each plot were removed by hand. After drying leaves at 65 °C for 48 h, they were ground through a 200 µm mesh in a Wiley Mill (General Electric Co., USA), percent nitrogen was determined for a 1–2 g subsample for each plot using dry combustion in a Leco FP-428 (Leco Instruments Ltd., USA).

#### 2.4.7. Root competition in the agroforestry field

To understand whether there was significant root competition for moisture and nitrogen between large intercropped trees and willows in the agroforestry field, a root barrier field experiment was set up in May 2007. Nine of the twelve plots in the agroforestry field (3 replications for each willow clone) were divided into two sections. The first section was the root barrier zone, which was a 10 m long area (one-fifth) of the plot, where a root barrier was installed on either side of this area of the plot to prevent root competition between the willows and the large intercropped tree rows. Trenches were dug using a mini-excavator, cutting all tree roots within those zones in the top 1 m of soil, and sheets of 1 mm thick flexible vinyl sheeting were buried to a depth of 1 m. The location of root barriers was selected to ensure that there was no significant difference in tree shading and light levels compared to the areas being used as a control for the root barrier zones. Soil moisture, soil available nitrogen, and willow yields (using the same methods described above) were measured in three random locations both within the root barrier zones and outside of the root barrier zones in the remaining area of the plot used as a control. Locations of control measurements were at least 10 m away from the root barrier zones, and measurements within the root barrier zones left a 1 m buffer around the periphery to reduce edge effects.

#### 2.5. Statistical analyses

All analyses were conducted using SAS v.9.1 (SAS Institute, Cary, NC, USA) with a Type I error rate of  $\alpha = 0.05$ . Variance analyses were conducted using repeated measures where necessary. Univariate analyses and a Shapiro–Wilk test of residuals were used to test assumptions of homoscedasticity, and log or square-root transformations were used to normalize residuals where necessary. Lund's test for outliers was used to detect outliers in all datasets (Lund, 1975). Significant differences between means in variance analyses were determined using Tukey's HSD test. For data sets with  $n < 30$  significant differences between means were determined using a *t*-test.

Correlation analyses were done between photosynthetic photon flux density and soil temperature, soil temperature and soil moisture content, and between soil moisture content and willow yields for each year. Field-average PPFD values at each of four time periods within a day (9 a.m., 12 p.m., 3 p.m., 6 p.m.) were correlated with season-average soil temperature values at those same times within each field. Average soil moisture content for each plot in each field was correlated with daily-average soil temperatures (for each field) corresponding to each of the soil moisture sampling periods.

### 3. Results

#### 3.1. Willow growth and yields

Yields were significantly higher in the agroforestry field in both years of the study (*F*-test,  $p < 0.05$ ) (Table 2). Yields for the agroforestry and control fields were 0.78 and 0.54 odt ha<sup>-1</sup>,

**Table 2**

Yields and percent survival of three willow clones grown in an agroforestry intercropping system with 20-year-old mixed tree species, and a control (willow monocropping) system.

Field and willow clone	2006 <sup>a</sup>		2007 <sup>b</sup>	
	Yield <sup>c</sup> (odt ha <sup>-1</sup> )	Percent survival	Yield <sup>c</sup> (odt ha <sup>-1</sup> )	Percent survival
Agroforestry	0.78 <sup>*</sup>	93 <sup>*</sup>	3.00 <sup>*</sup>	89 <sup>*</sup>
SV1	0.56b	90c	3.93a	90a
SX67	0.89a	96a	3.30a	90a
9882-41	0.89a	92b	1.77b	88a
Control	0.54 <sup>*</sup>	86 <sup>*</sup>	1.11 <sup>*</sup>	66 <sup>*</sup>
SV1	0.83a	95a	1.63b	79b
SX67	0.46c	81d	1.03b	63c
9882-41	0.33c	81d	0.68c	57d

Values represent least squares means where those with the same letters (a–d) are not significantly different according to Tukey's HSD test ( $p > 0.05$ ).

<sup>a</sup> Willows had 1-year roots and shoots.

<sup>b</sup> After coppice, willows had two-year roots and 1-year shoots.

<sup>c</sup> Yields in oven-dried tonnes (odt) per hectare.

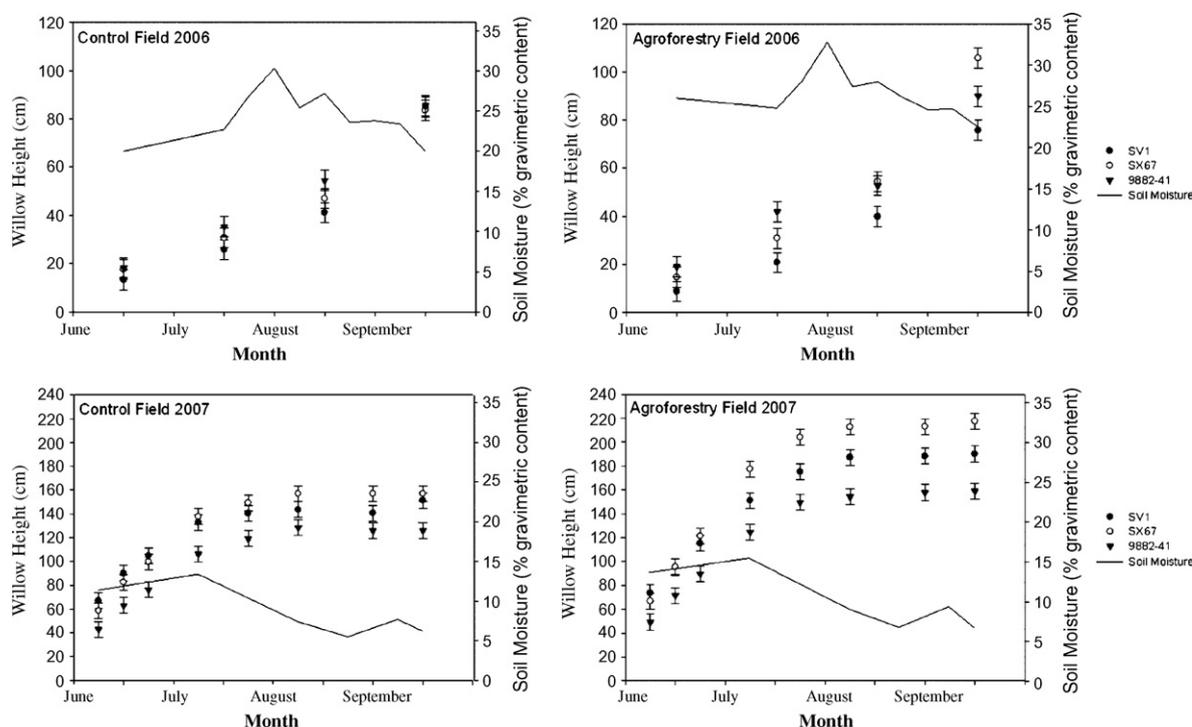
<sup>\*</sup> Field averages are significantly different according to an *F*-test ( $p < 0.05$ ).

respectively, in 2006, and 3.00 and 1.11 odt ha<sup>-1</sup>, respectively, in 2007. Percent survival was significantly higher in the agroforestry field in both years, and survival generally explained trends in yields between willow clones (Table 2). In 2006, yields between willow clones had the opposite pattern between fields, with SX67 = 9882-41 > SV1 in the agroforestry field, and SV1 > SX67 = 9882-41 in the control field. In 2007, yields between the clones were the same in both fields, with SV1 = SX67 > 9882-41. Yields for each plot in 2006 ranged from 0.48 to 1.37 odt ha<sup>-1</sup> in the agroforestry field, and 0.11 to 2.37 odt ha<sup>-1</sup> in the control field (se = 0.105). In 2007, yields for each plot ranged from and 1.06–5.89 odt ha<sup>-1</sup> in the agroforestry field, and 0.46–2.99 odt ha<sup>-1</sup> in the control field (se = 0.389). There was greater spatial variation in yields between plots in the control field in 2006 (range of 2.26 odt ha<sup>-1</sup> between plots compared to 0.89 odt ha<sup>-1</sup> in the agroforestry field), but in 2007, there was greater variation in the agroforestry field (range of 4.83 odt ha<sup>-1</sup> compared to 2.53 odt ha<sup>-1</sup> in the control field).

Willows in the agroforestry field were significantly taller by the end of the season in both years of the study (*F*-test,  $p < 0.05$ ) (Fig. 2). In 2006, there was no significant difference in the number of stems per willow between the two fields, but in 2007, there were significantly more stems in the agroforestry field, likely contributing to higher yields (*F*-test,  $p < 0.05$ ) (Table 3). Leaf area index (LAI) measured in August of each year showed significant differences between willow clones in the order 9882-41 > SV1 > SX67 in 2006, and in the order SV1 = SX67 > 9882-41 in 2007 (*F*-test,  $p < 0.05$ ) (Table 3).

#### 3.2. Photosynthetic photon flux density

Photosynthetic photon flux density was significantly lower throughout the day (9 a.m., 12 p.m., 3 p.m., 6 p.m.) in the agroforestry field compared to the control field in both years of the study (*F*-test,  $p < 0.05$ ) (Table 4). Between fields, there was a 250 µmol m<sup>-2</sup> s<sup>-1</sup> difference in daily-average PPFD values measured in August 2006, and a 169 µmol m<sup>-2</sup> s<sup>-1</sup> difference in PPFD averaged from May and July 2007. There was an increasing difference in PPFD values between fields throughout the day (9 a.m. to 6 p.m.) in 2006 and a decreasing difference in 2007, due to the changing angle of the sun and sunrise times throughout the growing season, and resulting effects on shading patterns in the agroforestry field (given the orientation of tree rows). Maximum differences in PPFD values between fields were about 400 µmol m<sup>-2</sup> s<sup>-1</sup>, and minimum differences were about 50 µmol m<sup>-2</sup> s<sup>-1</sup> (Table 4).



**Fig. 2.** Willow height and field-average soil moisture throughout the 2006 and 2007 growing seasons, where error bars represent standard errors. Note the different left-y-axis scales between years.

### 3.3. Photosynthetic light saturation curves

Photosynthetic light saturation curves generated for the three willow clones indicated that light saturation points were all significantly different, in the order 9882-41 > SV1 > SX67 ( $t$ -test,  $p < 0.05$ ) (Table 5). Light-saturated PPFD values for the three clones ranged from 1319 to 1394  $\mu\text{mol m}^{-2} \text{s}^{-1}$  (Fig. 3 and Table 5).

### 3.4. Soil temperature

Average soil temperatures from May to September in each field were significantly lower in the agroforestry field in both years, with a 0.4 °C difference in 2006 and a 2.7 °C difference in 2007 ( $F$ -test,  $p < 0.05$ ) (Table 4). In 2006, daily-average soil temperatures (May–September) between sampling locations in each field ranged from

22.1 to 23.5 °C in the agroforestry field, and 23.2 to 24.3 °C ( $se = 0.063$ ) in the control field. In 2007, daily-average temperatures ranged from 18.6 to 19.6 °C in the agroforestry field, and 21.2 to 22.3 °C in the control field ( $se = 0.051$ ). Soil temperatures were lower in 2007, despite warmer temperatures, due to greater canopy closure in the willow plots which shaded the soil. At peak daily soil temperatures (occurring at 05:30–06:00 p.m.), temperature differences were greater between the two fields compared to periods of increasing (2 p.m.) and decreasing (1 a.m.) temperatures, and compared to the lowest daily temperatures (8 a.m.) ( $t$ -test,  $p < 0.05$ ) (Fig. 4). Average differences in peak soil temperatures between fields were 1.2 °C ( $se = 0.074$ ) in 2006, and 0.8 °C ( $se = 0.065$ ) in 2007. Within the agroforestry field, there were significantly higher soil temperatures in the centre of alleyways compared to the edge of the alleyways just adjacent to large tree rows ( $F$ -test,  $p < 0.05$ ). Average soil temperature in the centre versus edge of alleyways were 23.3 °C and 22.1 °C ( $se = 0.36$ ), respectively.

### 3.5. Soil moisture

Soil moisture content was significantly higher in the agroforestry field in both years, with an average difference between fields (May–September) of 1.9% in 2006 and 1.4% in 2007 ( $F$ -test,  $p < 0.05$ ) (Table 4). The lowest values measured (across all sampling dates) in 2006 were 16.4% and 14.2% for the agroforestry and control fields, respectively, and the highest values were 38.2% and 36.6%, respectively. In 2007, the lowest values measured were 6.7% and 5.6% for the agroforestry and control fields, respectively, and the highest values were 16.3% and 13.5%, respectively. Within a single sampling date, the difference in soil moisture contents measured between plots in each field ranged from 0 to 9.7% in 2006, and 0 to 5.6% in 2007.

Averaged across all sampling dates, spatial variation in soil moisture within the agroforestry field was not significantly different between the two years of the study, with an average difference of 2.2% and 2.1% in 2006 and 2007, respectively ( $F$ -test,  $p > 0.05$ ). In the control field, there was significantly more

**Table 3**

Number of stems per individual and leaf area index (LAI) of three willow clones grown in an agroforestry intercropping system with 20-year-old mixed tree species, and a control (willow monocropping) system.

Field and willow clone	2006 <sup>a</sup>		2007 <sup>b</sup>	
	Number of stems	LAI	Number of stems	LAI
Agroforestry	1.6	1.37	17 <sup>*</sup>	1.94 <sup>*</sup>
SV1	1.2b	1.31c	16b	2.46a
SX67	1.8a	0.86d	18a	2.13a
9882-41	1.8a	1.94ab	17ab	1.24c
Control	1.8	1.56	10 <sup>*</sup>	1.64 <sup>*</sup>
SV1	1.8a	1.57bc	12c	1.85b
SX67	1.9a	0.88d	10d	1.79b
9882-41	1.6a	2.21a	9d	1.27c

Values represent least squares means where those with the same letters (a–d) are not significantly different according to Tukey's HSD test ( $p > 0.05$ ). Effects significant in an ANOVA were clone and field for number of stems in 2006, field and clone  $\times$  field for number of stems in 2007, clone for LAI in 2006, and field and clone  $\times$  field for LAI in 2007.

<sup>a</sup> Willows had 1-year roots and shoots.

<sup>b</sup> After coppice, willows had two-year roots and 1-year shoots.

<sup>\*</sup> Field averages are significantly different according to an  $F$ -test ( $p < 0.05$ ).

**Table 4**

Comparison of microclimate parameters in the agroforestry and control fields in 2006 and 2007.

Microclimate parameter	Field average	
	2006	2007
PPFD ( $\mu\text{mol m}^{-2} \text{s}^{-1}$ ) <sup>a</sup>		
Agroforestry field	1044 <sup>b</sup>	1176 <sup>b</sup>
9:00 a.m.	994 <sup>d</sup>	928 <sup>e</sup>
12:00 p.m.	1527 <sup>b</sup>	1628 <sup>b</sup>
3:00 p.m.	1378 <sup>c</sup>	1466 <sup>c</sup>
6:00 p.m.	277 <sup>f</sup>	680 <sup>f</sup>
Control field	1294 <sup>b</sup>	1345 <sup>b</sup>
9:00 a.m.	1048 <sup>d</sup>	1272 <sup>d</sup>
12:00 p.m.	1753 <sup>a</sup>	1845 <sup>a</sup>
3:00 p.m.	1692 <sup>a</sup>	1594 <sup>b</sup>
6:00 p.m.	687 <sup>e</sup>	730 <sup>f</sup>
Soil temperature ( $^{\circ}\text{C}$ ) <sup>c</sup>		
Agroforestry field	23.1 <sup>b</sup>	19.0 <sup>b</sup>
Control field	23.5 <sup>a</sup>	21.7 <sup>a</sup>
Soil moisture (%) <sup>c</sup>		
Agroforestry field	26.5 <sup>a</sup>	10.1 <sup>a</sup>
Control field	24.6 <sup>b</sup>	8.7 <sup>b</sup>
Soil nitrogen (%) <sup>c</sup>		
Agroforestry field	n/a	0.34 <sup>b</sup>
Control field	n/a	0.47 <sup>a</sup>
Foliar nitrogen (%) <sup>d</sup>		
Agroforestry field	n/a	2.69 <sup>a</sup>
Control field	n/a	2.69 <sup>a</sup>

Values represent least square means where those with the same letters (a–f) are not significantly different according to Tukey's HSD test ( $p > 0.05$ ). Letter comparisons are for values within each microclimate parameter and year. All displayed effects were significant in an ANOVA.

<sup>a</sup> Photosynthetic photon flux density (PPFD) values in 2006 are from one sampling date in August, and in 2007 are the average of two sampling dates in May and July.

<sup>b</sup> Values represent whole-day field averages (9 a.m. to 6 p.m.) and are significantly different between fields according to Tukey's HSD test ( $p < 0.05$ ).

<sup>c</sup> Soil temperature, moisture and soil nitrogen values are field-wide averages from May to September.

<sup>d</sup> Foliar nitrogen values are the average of two sampling dates in July and August.

**Table 5**

Mean light saturation points and light saturated photosynthetic photon flux density (PPFD) values for three willow clones based on photosynthetic light saturation curves measured on willows in the agroforestry and control fields in July 2007.

Willow clone	Light saturation point ( $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ )	Light saturated PPFD <sup>a</sup> ( $\mu\text{mol photons m}^{-2} \text{ s}^{-1}$ )
SV1	16.18a	1368
SX67	14.59c	1394
9882-41	18.30b	1319

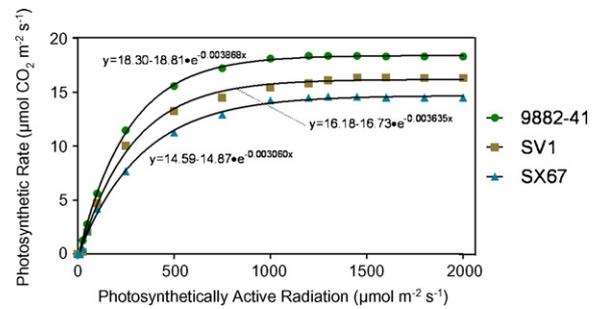
Values represent least squares means where those with the same letters (a–c) are not significantly different according to a *t*-test ( $p > 0.05$ ).

<sup>a</sup> Calculated from light curve equations in Fig. 3.

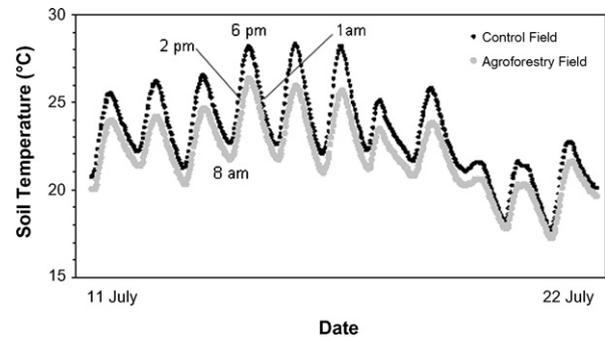
variation in soil moisture in 2006 with a range of 3.8% compared to 2.0% in 2007 ( $F$ -test,  $p < 0.05$ ). Between willow clones, average soil moisture levels occurring in plots for each clone in both fields were not significantly different ( $F$ -test,  $p < 0.05$ ), ranging from 25.1 to 25.9% ( $se = 0.52$ ) (9882-41 < SX67 < SV1) in 2006, and 9.1% to 9.6% ( $se = 0.34$ ) (9882-41 < SV1 < SX67) in 2007 (averaged across all sampling dates).

### 3.6. Soil and foliar nitrogen

Between fields in 2007, there was significantly lower soil available nitrogen content (nitrate and ammonium) in the agroforestry field compared to the control field with a difference



**Fig. 3.** Photosynthetic light saturation curves for three willow clones.  $R^2 = 0.99$  for all clones.



**Fig. 4.** Field-average soil temperatures over a 12 day period in July 2006. Daily maximum temperatures occurred at 6 p.m., minimum temperatures at 8 a.m., and temperatures intermediate to the high and low points occurred at 2 p.m. and 1 a.m. Data points represent an average of 12 values for each field, with standard errors of 0.024 and 0.026 for the agroforestry and control fields respectively. Trends indicate higher temperature peaks in the control field (at 6 p.m.), with an apparent 'buffering effect' in the agroforestry field.

of 0.13% (Table 4). However, measurements of foliar nitrogen found no significant differences between fields or willow clones ( $F$ -test,  $p < 0.05$ ).

### 3.7. Root competition in the agroforestry field

Soil moisture data was also used to test for an effect of intercropped trees on soil moisture in the agroforestry field in 2006, since samples were taken in a gradient across the alleyways. There was no significant effect of proximity to intercropped trees up to 2 m from the tree rows ( $F$ -test,  $p < 0.05$ ). However, the root barrier field experiment in 2007 indicated significantly higher moisture (0.9%) within the root barrier zones where root competition from intercropped tree rows was excluded (Table 6). The root barrier experiment also indicated no significant difference in soil or foliar nitrogen between the root barrier and control zones (Table 6).

**Table 6**

Influence of 1 m deep root barriers on soil moisture, nitrogen levels and willow yields in an agroforestry intercropping system with 20-year-old mixed tree species.

Location	Yield <sup>a</sup> (odt ha <sup>-1</sup> )	Soil moisture (%)	Foliar N (%)	Soil N (%)
Root barrier	3.6a	10.7a	2.62a	0.33a
Control	3.5a	9.8b	2.71a	0.36a

Values represent least square means across all sampling dates (yield was a single estimate) where those with the same letters (a and b) are not significantly different according to Tukey's HSD test ( $p > 0.05$ ). In an ANOVA, the location effect was significant for soil moisture.

<sup>a</sup> Yields in oven-dried tonnes (odt) per hectare.

### 3.8. Correlations

Average soil temperatures at 9 a.m., 12 p.m., 3 p.m. and 6 p.m. for each plot (May–September) were positively correlated with average PPF values at the same times of day in both years of the study ( $r = 0.82$  for 2006 and  $r = 0.93$  for 2007). Average soil moisture contents (May–September) were negatively correlated with soil temperatures on a plot basis in both fields ( $r = -0.54$  for 2006 and  $r = -0.41$  for 2007). Final yield values in both fields were significantly correlated with average (May–September) soil moisture contents on a plot basis ( $r = 0.48$  for 2006 and  $r = 0.72$  for 2007). Willow mortality in 2007 (a hot, dry season) was correlated with soil moisture levels on a plot basis in both fields ( $r = 0.89$ ).

## 4. Discussion

This study investigated the growth potentials of three willow clones (*S. dasyclados* SV1, *S. miyabeana* SX67 and *S. purpurea* 9882-41) on marginal (Class 3-4) land in both an agroforestry intercropping system and a conventional monoculture used as a control. Light (photosynthetic photon flux density), soil temperature, and soil moisture and nitrogen levels were compared between the two systems to understand whether differences in these parameters, as moderated by the presence or absence of large intercropped trees, were correlated with differences in willow growth and yields.

### 4.1. Clonal growth differences

The significantly different weather conditions during the two years of this study provided a unique situation to observe the response of the willow clones to varying environmental conditions. Clones SV1 and SX67 seemed the best suited to handle a range of environmental conditions, as they had the highest survival in both years, and produced the greatest yields at the end of 2007. A study by Labrecque and Teodorescu (2005) also found non-significant differences in yields between these two clones beyond the establishment year. On the other hand, 9882-41 appeared to be the most sensitive to moisture stress, as it experienced the greatest mortality during dry conditions in 2007. It also had higher yields in the agroforestry field in both years, where average soil moisture levels were higher.

The three clones had distinctly different growth strategies: 9882-41 had a greater number of stems but smaller average basal diameters and heights; SX67 grew tallest but had the least number of stems and intermediate basal diameters (and a greater range of diameters); and SV1 had the largest basal diameters, but intermediate heights and numbers of stems. A greenhouse experiment (data not shown) indicated that SV1 had the highest root:shoot ratio, which likely led to better establishment in the field in 2006 and allowed for better survival and growth during dry conditions in 2007. It was also qualitatively observed that some plots of SV1 had a large amount of sylleptic branching, which has been linked with higher yields (Robinson et al., 2004; Zeleznik, 2007). Sylleptic branching also results in a greater total leaf area over time, and faster canopy closure (Ceulemans et al., 1990) which makes this clone attractive from the standpoint of weed control. SV1 tended to have the greatest whole-tree leaf area, and the area of individual leaves was on average larger than the other clones. Studies on short rotation willow have found that varieties with larger individual leaf areas produce higher yields (Robinson et al., 2004), which is consistent with results of this study. SV1 had a greater stem angle at the base compared to the other clones, which reduced LAI values during early growth. At lower densities with little to no canopy closure, 9882-41 had a

higher LAI, as measured during the 2006 season. With higher stem densities and greater canopy closure, SV1 had a higher LAI, as measured in the 2007 season. SX67 had straight stems, but the whole-tree leaf area was intermediate to the other two clones, resulting in intermediate LAI values. Trends in LAI generally correlated with growth and yields between the clones, which is consistent with other studies of short rotation willow (Tharakan et al., 2005).

### 4.2. Microclimate differences between field setups

Willow survival was significantly higher in the agroforestry field during both years of this study, and patterns in survival generally explained differences in yields. However, willows in the agroforestry field were also significantly taller, indicating that growth was reduced in the control field—i.e. yield differences were not entirely explained by differences in mortality. There was an increase in mortality during 2007 due to lower precipitation throughout most of the season. Moisture stress caused wilting in almost half of the willows in one plot of 9882-41 in the control field (personal observation), and most plots experienced tip death during the most severe period of drought in July and August 2007, but recovery was apparent through new shoot growth in late September.

Weather differences between the two field seasons allowed for a better understanding of how soil temperature and moisture characteristics differed between the willow monoculture and the willow agroforestry intercropping system. There were significantly lower soil temperature peaks in the agroforestry field, which may have reduced evaporative losses from the soil, maintaining higher moisture levels. Differences would likely be more apparent during establishment and after coppice when the willow canopies are relatively open (which was the case in both years of this study). After full canopy closure, differences in soil temperatures and/or soil moisture content as a result of additional shading by an intercropped tree canopy in the agroforestry system would likely diminish.

There was similar variation in soil moisture and temperature between plots in the agroforestry field in both years, whereas there was much greater variation in the control field during 2006 and very little variation during 2007. There was also a much larger increase in mortality in the control field in 2007, suggesting that there was a moderating effect of intercropped trees on soil moisture which could be significant for willow survival and yields, especially under dry conditions. These findings are supported by studies of tree-intercropping systems with coffee shrubs in Mexico, where Lin (2007) found that increased shade resulted in smaller fluctuations in irradiance, air temperature and relative humidity, resulting in less water loss from the soil and smaller fluctuations in soil moisture content.

#### 4.2.1. Photosynthetic photon flux density

Willows in the agroforestry field were exposed to 16% lower daily average photosynthetic photon flux density, with an average difference between fields of  $210 \mu\text{mol m}^{-2} \text{s}^{-1}$ . However, this did not appear to have negative effects on willow growth, since yields were significantly higher in that field during both years of the study. Preliminary photosynthetic light saturation curve data indicated that photosynthesis might be reduced in the agroforestry field at 9 a.m., since average PPF levels were  $900\text{--}1000 \mu\text{mol m}^{-2} \text{s}^{-1}$ , below light-saturated PPF values of  $1319\text{--}1394 \mu\text{mol m}^{-2} \text{s}^{-1}$ . At 12 p.m. and 3 p.m., light levels in the agroforestry field ranged from  $1378$  to  $1628 \mu\text{mol m}^{-2} \text{s}^{-1}$ , still at or above the light saturated PPF values, indicating that there would likely be no difference in photosynthetic rates between the two fields. Other studies on *Salix* clones have reported light

saturated PPFD values of  $1350 \mu\text{mol m}^{-2} \text{s}^{-1}$ , which agrees well with those measured in this study (Patton and Jones, 1989). Robinson et al. (2004) reported light saturation points of  $13.29\text{--}16.27 \mu\text{mol CO}_2 \text{m}^{-2} \text{s}^{-1}$ , similar to values of  $14.59\text{--}18.30 \mu\text{mol CO}_2 \text{m}^{-2} \text{s}^{-1}$  measured in this study. Since light saturation curves were measured on willows in the centre of agroforestry alleyways (and unshaded individuals in the control field), no conclusions can be made regarding their shade tolerance.

#### 4.2.2. Soil temperature and moisture

PPFD was highly (positively) correlated with differences in soil temperature of  $0.4\text{--}2.7^\circ\text{C}$  between fields during the two years. Higher soil temperatures as a result of greater irradiance have been measured in a number of other studies (Carlson and Groot, 1997; Chen et al., 1993; Oliver et al., 1987). Differences in soil temperature can impact root growth and therefore plant production (McMichael and Burke, 1996), though direct relationships were not investigated in this study.

Soil temperatures were moderately (negatively) correlated with differences in average soil moisture content of  $1.4\text{--}1.9\%$  between fields in both years. Although soil temperatures are affected by soil moisture levels through their effects on soil heat capacity and conductivity (van Wijk, 1965), the greater correlations of light (PPFD) with soil temperature in this study indicated that irradiance had a much larger influence on soil temperatures than soil moisture content. These results support the hypothesis that increased shading by intercropped trees resulted in less moisture loss from the soil. Studies comparing forest clear-cuts to uncut areas have found that correlations between soil temperature and soil moisture content were not significant (Kim, 2008). However, in these types of forest gap studies, differences in soil moisture were likely more affected by relative water use by trees in the surrounding forest vs. regenerating growth, as well as increased precipitation reaching the soil surface in non-forested areas (Zirlewangen and von Wilpert, 2001; Ritter et al., 2005). In addition, heterogeneous regrowth in forest gaps leads to more complex relationships between irradiance and soil temperature and soil moisture content, and increased spatial variation in soil parameters (Bhatti et al., 2000; Ritter et al., 2005). It is therefore difficult to draw analogies between forest gap studies and an agroforestry intercropping scenario where there is a homogeneous understory of willow as well as a homogeneous, but sparse, overstory of large intercropped trees.

#### 4.3. Effects of microclimate differences on willow growth

Soil moisture content was moderately (positively) correlated with willow yields on a plot basis in both fields. This indicates that moderation of soil moisture levels by increased shading in the agroforestry field led to improved willow growth and yields. However, the fact that there were only moderate correlations between soil temperatures and soil moisture content, and between soil moisture content and yields, indicated that other site differences not accounted for in this study may have been significant for willow growth. Soil characteristics between the fields were not significantly different in the top 30 cm, and in one- and two-year-old willows, most of the rooting systems exist as fine roots, and are located in the top 30 cm of soil (Volk et al., 2001). However, there may have been deeper roots, where potential differences in subsoil structure and/or nutrient availability and hydrology may have affected growth. If there were not as many deep roots in the control field, this may have led to greater moisture stress during the dry season in 2007. No measurements of rooting distribution or density were taken in this study.

#### 4.4. Root competition in the agroforestry field

In the agroforestry field, the root barrier experiment indicated that a 2 m buffer between large intercropped trees and short rotation willow plots was enough to avoid significant root competition for soil moisture and nitrogen, at least for the first two years of growth. It is likely that lateral root growth, which can reach 4–5 m in willow and hybrid poplar plantations after two to three years (Hansen, 1981; Ericsson, 1984; Friend et al., 1991), may result in measurable competition between the willows and intercropped trees in future years. There may have been some competition for soil moisture from May to September, since soil moisture content was 0.9% higher in zones where competition was excluded, but this did not translate into higher yields at the end of the season. The difference in soil moisture between the agroforestry and control fields over the same time period was 1.4%, and this was correlated with growth differences between the two fields. This apparent contradiction (where only a 0.5% difference resulted in growth effects) may be explained by the fact that the field-average values did not reflect the large amount of spatial variation occurring both within and between fields across all sampling dates in each year. On a single sampling date, there was up to a 10% difference in soil moisture content between plots within each field, with a similar magnitude of difference when comparing plots between fields. Soil moisture content differences of that magnitude likely have the ability to influence growth on a daily or weekly basis, which is likely why the correlations between soil moisture content and yields were significant. These findings are supported by Lin (2007), who found that under more extreme conditions (hot and dry), shading of the soil by trees can reduce fluctuations of microclimate parameters more noticeably than reducing average values across an entire season.

### 5. Conclusion

Of the three willow clones used in this study, *S. dasyclados* SV1 and *S. miyabeana* SX67 produced the highest yields, and were best suited to withstand the range of environmental conditions during the course of this study. *S. purpurea* 9882-41 performed well under high soil moisture, and would be best suited for growth in wetlands or along riparian areas. The presence of large intercropped trees in a willow agroforestry intercropping setup resulted in lower photosynthetic photon flux density which was correlated with lower soil temperatures and higher soil moisture, contributing to higher willow yields. Results of this study suggest that moderate shading in an intercropping setup with 20-year mixed tree species (with tree rows spaced 15 m apart and oriented NW to SE) can result in a buffering effect on microclimate conditions, where there is less variation in soil temperatures and soil moisture contents across a range of weather conditions.

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