IMPROVING THE WATER USE EFFICIENCY OF SHORT ROTATION COPPICE (SRC) WILLOWS

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Contractor
IACR Rothamsted

with
Cranfield University, ADAS and European Willow Breeding Partnership

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EXECUTIVE SUMMARY

Introduction

The UK has undertaken to reduce its emissions of Greenhouse gases, and to meet this commitment has undertaken to ensure that 10% of its electricity requirements will be met by renewable energy sources by 2010 (DTI, 2000). Of these, biofuels will be an important component, especially willow short rotation coppice (SRC). However, the high water requirements of SRC means that water stress may reduce biomass yields to below commercially viable levels in the drier eastern areas of the country. Increasing the water use efficiency and/or the drought resistance of willow would have the potential both to improve the productivity in wetter areas and to extend the agroclimatic range over which it would be feasible to grow these crops.

A better understanding of the extent and nature of varietal differences in the responses of willow SRC to water stress is required in order to identify the potential for breeding superior willow varieties, which use water more efficiently, and/or are more drought resistant.

This work should help, in the longer term, to increase the productivity of biofuel willow through the identification of drought resistant and water efficient varieties. These would be important to help increase the production and profitability of SRC grown commercially in the drier areas of the UK, and also elsewhere in Europe.

Project Aims and Objectives

This principal aim of this research project was to determine the range of water use efficiency and drought resistance within existing willow varieties, in order to identify the potential for improving productivity through plant breeding. The project planned to examine the water use of both a number of near-market varieties and evaluate simple techniques for early drought tolerance screening in the breeding programme.

The work was undertaken at two levels:

1. field observations of water use by medium scale plots of willow cultivars, to determine the patterns of water use, its role in determining crop yield, and the importance of irrigation in maximising yield; and
2. detailed observation of crop physiological responses of willow to drought in order to develop an understanding of the crop traits associated with improved water use efficiency and drought resistance.

Background

This project was jointly funded by the European Willow Breeding Partnership (EWBP) and the DTI. With the closure of the EWBP in 2002, the project funding was transferred to DEFRA and is now a component of the Willow breeding project co-ordinated by Dr Angela Karp at Rothamsted. This report therefore covers only the preliminary results obtained during the first experimental season. During this period the primary activities were related to initiating the two experimental approaches involving field-scale experiments conducted by ADAS and detailed physiological studies by Cranfield University. The project was proceeding on schedule but DTI funding ceased before much of the data collected during the first year had been analysed. The results presented here are therefore only indicative and should be treated with caution.

Summary of Methodology Adopted

The two strands of this project both involved growing a range of willow varieties in order to quantify responses to water stress.

In the meso-scale experiment established at Gleadthorpe, 12 varieties of released or near release varieties bred at Long Ashton and elsewhere were planted in two blocks. In the establishment year, both blocks were irrigated to encourage growth. Plant height, establishment rate and biomass production were recorded.

In the detailed experiments established at Silsoe, 50 varieties representing a range of genetic material including parent species were planted in pots and also in a small field experiment. The pots were subject to periodic water stress episodes whilst the field experiment was irrigated to avoid water stress. A range of morphological and physiological parameters were measured to identify whether any would provide a quick indicator of drought tolerance or high water use efficiency for use in subsequent breeding programmes. Parameters measured included: leaf length, width and area; specific leaf area; stem height; biomass production and chlorophyll fluorescence.

Conclusions and Recommendations

Few conclusions can be drawn from the first year of a three-year project and this project is no exception. There are, however, clear differences in the performance of the varieties included in the two sets of experiments. The next phase of the project, funded by DEFRA under the biomass breeding programme, should help to identify how a range of high-yielding willow varieties respond to water stress.

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1 INTRODUCTION

1.1 Technical background

The UK has undertaken to reduce its emissions of greenhouse gases, and to meet this commitment has undertaken to ensure that 10% of its electricity requirements will be met by renewable energy sources by 2010. Biofuels, especially willow short rotation coppice (SRC), have the potential be an important component as they are a less weather dependent energy source than wind, wave or solar power. However, the high water requirements of SRC means that water stress may reduce biomass yields to below commercially viable levels in the drier eastern areas of the country. Increasing the water use efficiency and/or the drought resistance of willow would have the potential both to improve the productivity in wetter areas and to extend the agroclimatic range over which it would be feasible to grow these crops.

A better understanding of the extent and nature of varietal differences in the responses of willow SRC to water stress is required in order to identify the potential for breeding superior willow varieties, which use water more efficiently, and/or are more drought resistant.

This work should help, in the longer term, to increase the productivity of biofuel willow through the identification of drought resistant and water efficient varieties. These would be important to help increase the production and profitability of SRC grown commercially in the drier areas of the UK, and also elsewhere in Europe.

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1.3 Scope of the report

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2 METHODOLOGY

2.1 Field scale experiments

The varieties selected for the field experiment comprise promising new clones already included in other variety trials conducted as part of the EWBP breeding programme.

Table 1. Varieties selected for field scale trial on water use efficiency in willow at Gleadthorpe

Code	Variety
A	78313
B	Tora
C	Stott
D	LA960048
E	LA960340
F	LA960365
G	LA970048
H	LA970164
I	LA970024
J	LA980125
K	LA980132
L	LA980221
M	LA980414
N	LA980442
O	LA980447

The field site at ADAS Gleadthorpe was established on sandy soil. The land was sown with grass in the preceding autumn to establish a working guard area.

The field plots were laid out in February 2002. These followed the plot design provided by Long Ashton. Two areas, each approximately 60 m by 24 m were laid out in parallel. The intention is irrigate one and to leave the other unirrigated. However, for the first year, both received irrigation to ensure establishment.

Plate 1. Irrigation of newly planted willow varieties at Gleadthorpe, March 2002.

The plots were sprayed with herbicide (glyphosate) on 1 March, and the willows planted on 13 and 14 March 2002. The willow material for varieties J and K in the wet block, and varieties M and I for both blocks was supplied separately by Murray Carter, and were planted later, on 26 March 2002. For each "row" the varieties were randomised separately. The layout of varieties was:

Table 2. Gleadthorpe Willows variety trial - Randomisation plan

Rep	Wet Block	Dry block
I	NAODMGJECBKDHLF	LEOAHNJKIGDMBCF
II	OEJLHDCFKIGANMB	NKJLBOMIAHDCFGE
III	ANIHBMDLFEOCKJG	DILFNCJGAEMHBKO

Following planting, the plots were irrigated using a spray irrigator, on three occasions in April, applying an estimated 120 mm of water. As a result, good establishment of the willows was achieved.

To support the water use studies, daily meteorological data were recorded using the synoptic station at ADAS Gleadthorpe, about 400 m from the trial plots.

During the first season, three evaluations were undertaken: crop height for all plants was surveyed in July, and crop height for a representative sub-sample was again recorded in October. Lastly, the weight of biomass production on each plot was recorded at cut-back in January 2003. All of these assessments were, however, primarily concerned to record the rate of establishment of the crops, and are not necessarily expected to relate to the final biomass production.

2.1.1 Soil water monitoring

Neutron probe access tubes were installed in the plots in April 2002. One access tube was installed in each of the 15 varieties along the middle row of the two plots, so making a total of 30 access tubes. These tubes were installed by hand auguring using the standard ADAS procedures, down to a target depth of 1.5 m.

In addition, access tubes were also installed in the grass discard at either end of the plots and in the central: inter-plot gap which was kept fallow. These additional tubes give a point of reference for comparison with the results from the willows. Readings were taken every week during the summer and every other week after 1 November at 0.1 m depth increments down to the base of the tube.

In the first year there were no differential irrigation treatments and the results presented are pooled data from all willow plots.

2.2 Lysimeter studies

2.2.1 Selection of clones

Selection of varieties for inclusion in the experiment was undertaken in conjunction with Kevin Lindegaard, the willow breeder based at Long Ashton (Table 3). Cuttings from selected varieties were taken in mid-February 2002 and stored in a controlled-temperature room at 5 °C prior to planting.

Table 3. Varieties selected for initial screening in pot and field experiments

Code	Clonal No.	Pedigree (female parent first)
1	LA940044	115/34 S. viminalis Bowles Hybrid x 036/01 S. candida
2	LA960048	115/34 S. viminalis Bowles Hybrid x 110/02 S. bebbiana
3	LA960326 Beagle	115/70 S. viminalis Astrid x S.viminalis ?
4	LA970048	109/03 S. schwerinii x 115/27 S. viminalis Readers Red
5	LA970136	109/03 S. schwerinii x 115/10 S. vim Vigorous
6	LA970164	109/03 S. schwerinii x 115/65 S. vim Jorr
7	LA970184	033/18 S. dasyclados Aud x 075/01 S. dasyclados Kotens
8	LA970217	127/02 S. schwer x vim Tora x 037/01 S. caprea (x aurita?)
9	LA970249	115/71 S. vim SW880514 x 052/01 S. cordata Purpurescens
10	LA970416	115/68 S. viminalis Jorunn x 033/15 S. burjatica Lapin
11	LA970485	041/03 S. x dasyclados x 055/01 S. discolor
12	LA970504	041/03 S. x dasyclados x 033/19 S. dasyclados Loden
13	LA970523	041/03 S. x dasyclados x 024/02 S. x capreola
14	LA970534	115/11 S. viminalis Romanin x S. caprea in fence LARS plot 12
15	LA970573	LA940140 S. x mollissima x 087/01 S. miyabeana
16	LA970617	LA940147 S. x mollissima x 003/01 S. aegyptiaca
17	LA970654	062/01 S. glaucophylloides x 003/01 S. aegyptiaca
18	LA970766	062/01 S. glaucophylloides x 037/01 S. caprea (x aurita?)
19	LA980024	109/03 S. schwerinii x 127/01 S. vim x schwer Bjorn
20	LA980030	109/03 S. schwerinii x 77056 S. dasyclados
21	LA980038	127/02 S. schwer x vim Tora x 033/15 S. burjatica Lapin
22	LA980125	127/02 S. schwer x vim Tora x 001/01 S. udensis Shrubby willow ex China
23	LA980132	LA940140 S. x mollissima x 001/01 S. udensis Shrubby willow ex China
24	LA980172	127/02 S. schwer x vim Tora x 041/14 S. dasyclados 81090
25	LA980190	127/02 S. schwer x vim Tora x 087/01 S. miyabeana
26	LA980200	109/03 S. schwerinii x 055/01 S. discolor
27	LA980221	109/03 S. schwerinii x 115/38 S. viminalis Yellow Osier
28	LA980230	LA940140 S. x mollissima x 127/01 S. vim x schwer Bjorn
29	LA980266	125/01 S. schwer x vim x smith V7535 x 003/01 S. aegyptiaca
30	LA980279	127/02 S. schwer x vim Tora x 055/01 S. discolor
31	LA980289	125/01 S. schwer x vim x smith V7535 x 115/65 S. viminalis Jorr
32	LA980309	062/01 S. glaucophylloides x 001/01 S. udensis Shrubby willow ex China
33	LA980348	115/34 S. vim Bowles Hybrid x 001/01 S. udensis Shrubby willow ex China
34	LA980372	115/34 S. vim Bowles Hybrid x 106/01 S. sachalensis Sekka

Code	Clonal No.	Pedigree (female parent first)
35	LA980402	125/01 S. schwer x vim x smith V7535 x 106/01 S. sachalenensis Sekka
36	LA980414 Resolution	LA960231 (S. vim x schwer) x vim Quest x SW930812 (S. vim x schwer) x vim
37	LA980442 Endurance	102/01 S. redheriana x 77056 S. dasyclados
38	LA980496	041/03 S. x dasyclados x 115/10 S. vim Vigorous
39	033/20	S. dasyclados 77056
40	001/01	S. udensis Shrubby willow ex China
41	003/01	S. aegyptiaca
42	037/01	S. caprea (x aurita?)
43	055/01	S. discolour
44	062/01	S. glaucophylloides
45	087/01	S. miyabeana
46	102/01	S. rehderiana
47	109/03	S. schwerinii
48	115/34 Bowles Hybrid	S. viminalis
49	034/10 Ashton Stott	S. viminalis x burjatica
50	127/02 Tora	S. viminalis x schwerinii

The selection comprised 41 varieties, including recent promising clones and industry standards Ashton Stott, Bowles Hybrid and Tora, and nine parents. The nomenclature for the varieties bred at Long Ashton is LAYYXXXX where YY is the year of selection and XXXX is the variety number. Varieties are named when they are chosen for release, e.g. LA980414 has been named Endurance.

2.2.2 Pot experiment

The pot experiment was located at Cranfield University's Silsoe Campus in Bedfordshire. Silsoe is in one of the driest areas of England receiving, on average, around 600 mm y^{-1} rainfall. The experiment was designed to determine the comparative performance of the wide range of willow genotypes in response to water stress during establishment.

The objective of this experiment was to screen the 50 *Salix sp* to detect responses to drought in a range of morphological and physiological characteristics.

In April 2002, 50 varieties were planted in pots in a randomised block design with four replicates. The pots were laid out at 0.5 x 0.5 m. The plants were allowed to establish under fully irrigated conditions. Episodes of water stress were then initiated over the summer months.

Plate 2. Arrangement of pots for detailed study of 50 willow varieties prior to emergence – April 2002.

Plate 3. Pot with newly planted willow cutting planted centrally to a depth of ca 0.2 m.

2.2.3 Field experiment

The field experiment was established in sandy loam soil on the Silsoe Campus farm, some 500 m from the pot experiment.

The objective of this experiment was to provide data on the development and biomass production of 50 *Salix* varieties to compare with results from the pot experiment.

In April 2002, 50 varieties were planted in plots of 4 cuttings in a square arrangement spaced at 0.5 x 0.5 m. Plots were separated by a single guard row of a willow variety not included in the experiment (Q683).

The experiment was laid out in a randomised block design with three replicates giving a total of 150 plots. There was considerable variation in the diameter of the cuttings available for planting so they were sorted into three size categories (large, intermediate and small) and assigned to the blocks. The plots were irrigated during the summer in order to minimise water stress.

2.2.4 Measurements

Stem height

The length of the tallest stem from the top of the cutting was measured weekly in both the pot and field experiments.

Leaf shape and specific weight

Three leaves from each cultivar were collected on 27/09/2002 in Rep III of the field experiment. The leaves were sampled from the middle of the main stem. The petioles of the leaves were removed. The length (L), width (W) and area (A) were estimated for each leaf using a DeltaT leaf scanner. The leaf shape was characterised by the ratio (R)

$$R = \frac{L}{W} \qquad \text{Equation 1}$$

Each leaf was oven dried and weighed (ODw). The specific weight was calculated as follows.

$$SW = \frac{A}{ODw} \qquad \text{Equation 2}$$

Relative water retention

Bertholdsson (personal communication 2002) indicated that the Relative Water Retention capacity (RWR) could be used to screen clones for increased drought tolerance. Based of a study of six clones, Bertholdsson (personal communication 2002) found that RWR was correlated with stomatal conductance (Rs), ΔC^{13} discrimination and with WUE determined in a greenhouse experiment. His results suggested that clones with a high RWR performed relatively better than others during dry years.

Samples of three or four leaves of a similar age were taken from each willow tree and sealed in a plastic bag until the fresh weights (W0) were measured. In a greenhouse the leaves were left to dry on paper for 4 h and then weighed again (W4). Finally the leaves were oven dried for 48 at 80 °C and weighed (W48). RWR was calculated as:

$$RWR = \frac{(W0 - W4)}{(W0 - W8)} \qquad \text{Equation 3}$$

Early results using this method were not consistent within the same cultivar or variety. It appeared that the consistency was due to variation in the first drying period. The method was therefore, modified slightly in order to increase the replicability of the air drying treatment. For the first experiment followed the procedure suggested by Bertholdsson (2002), but for the second set of measurements the leaves were dried on a bench in laboratory and finally for the third set, the leaves were dried in an fan oven at 25 °C.

Stem biomass

At the end of the 2002 growing season both the field and pot experiments were cut back to ground level. The stems were dried and stem biomass determined.

Chlorophyll fluorescence

Photosynthetically active radiation (PAR) absorbed by chlorophyll drives photochemistry, is dissipated by non-photochemical processes – mostly as heat, or is re-emitted as fluorescence. These processes compete with each other so that if photochemistry or non-photochemical processes are inactive, fluorescence will be high and *vice versa* (Maxwell & Johnson, 2000). By measuring chlorophyll fluorescence in leaves the effects of stresses such as drought and heat can be monitored. The nomenclature used to describe photosynthesis and fluorescence is presented in Table 4.

Table 4. Definitions of chlorophyll fluorescence nomenclature adapted from Van Kooten and Snel, (1990).

Fluorescence intensity indicator		Description
F	Fluorescence intensity	Actual fluorescence at any time
Fo	Minimal fluorescence (dark)	Fluorescence intensity with all PSII reaction centres open while the photosynthetic membrane is in a non-energised state, i.e., dark adapted.
Fs	Fluorescence at steady state	Period within which the fluorescence intensity does not change while the external circumstances remain constant
Fm	Maximal fluorescence (dark)	Fluorescence intensity with all PSII reaction centres closed all non-photochemical quenching processes are at a minimum. Maximum fluorescence level in the dark adapted state
F'm	Maximal fluorescence (light)	Fluorescence intensity with all PSII reaction centres closed in a light adapted state.
F'o	Minimal fluorescence (light)	Fluorescence intensity with all PSII reaction centres open in a light adapted state.
Fv	Variable fluorescence (dark)	Maximum variable fluorescence in the state when all non-photochemical processes are at minimum (Fm-Fo)
F'v	Variable fluorescence (light)	Maximum variable fluorescence in a light adapted state (F'm-F'o)

In Figure 1, the fluorescence indicators, described in Table 4, are annotated on a light response curve for *Ficus benjamina*.

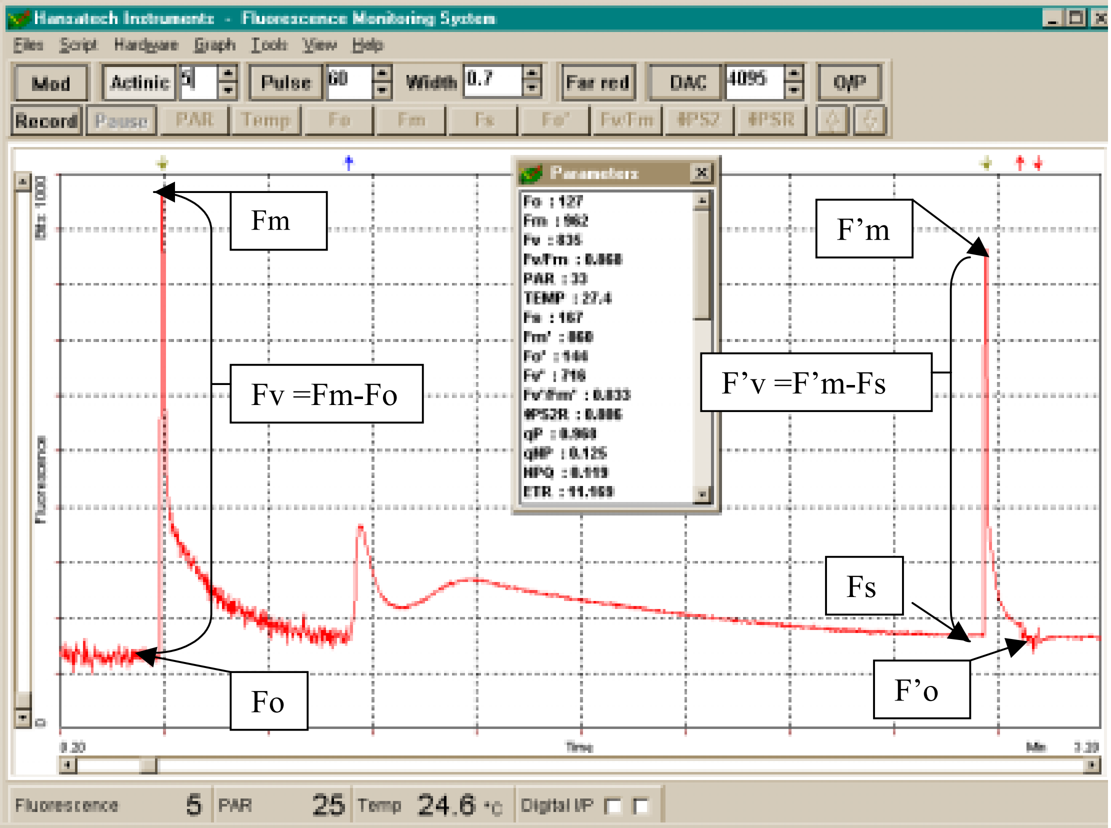

Figure 1. Two phases light response curve for *Ficus benjamina*, from the dark-adapted state, to light-adapted state. Light adaptation at 300 µmol m^{-2} s^{-1}. Indicators measured in bits and time in minutes.

2.2.4.1 Material and methods

A Fluorescence Monitoring System (FMS2, Hansatech Instruments Ltd, Kings Lynn, UK) was used to determine the dark adapted fluorescence of the 50 willow varieties grown in pots. The leaf clips provided by the manufacturer for dark adaptation were assumed to be identical. These were attached to individual leaves and left for about 40 minutes before sampling to allow dark adaptation of the photosynthesis systems.

Each variety was sampled between 27 and 29 times over the period 3 to 6 September 2002. On each day the same leaves were sampled from each variety but the monitoring order for the varieties was randomised. The results presented are the mean values over all days.

3 RESULTS

3.1 Field studies

3.1.1 Establishment

The survey undertaken in July 2002 showed some variation in plant performance (Table 5).

Table 5 Gleadthorpe Willows Trial: Summary of crop performance statistics recorded on July 21 2002, except stem biomass which was recorded in January 2003.

Variety code	Variety	stems	Height (m)	Missing plants[1]	Success rate	Stem biomass (kg ha^{-1})
A	78313	2.2	0.88	1	99.7%	3580
B	Tora	1.5	1.07	5	98.4%	3150
C	Stott	1.7	0.65	2	99.4%	2680
D	LA960048	1.6	0.79	4	98.7%	2670
E	LA960340	1.9	0.96	2	99.4%	3720
F	LA960365	1.9	1.04	6	98.1%	4580
G	LA970048	2.0	0.88	3	99.1%	3080
H	LA970164	1.5	0.89	6	98.1%	2750
I	LA970024	1.9	1.10	2	99.4%	4350
J	LA980125	1.9	0.63	82	74.1%	1280
K	LA980132	2.1	0.92	10	96.8%	2850
L	LA980221	1.9	1.00	3	99.1%	4150
M	LA980414	2.0	1.28	3	99.1%	5580
N	LA980442	1.9	0.72	3	99.1%	2920
O	LA980447	1.9	0.69	4	98.7%	3000
	Mean	**1.9**	**0.90**	**9.1**	**97.1%**	**3,356**
	S.e.m.	0.055	0.05	5.24	1.66%	263.1

[1] Plants that failed to produce any growth.

Statistical analysis showed that the differences between varieties were significant. There were two major reasons for this: firstly, different varieties had different growth patterns. In particular some had a large number of low thick stems, others a small number of tall stems. Secondly, variety LA980125 established very poorly. This variety was planted late (due to problems with the supply of the plant material), just before a particularly dry period, and it is considered that this might have affected the establishment. Whether the poor performance of this variety was due to the late establishment or other factors can only be resolved in the subsequent years.

3.1.2 Crop Yield

The crop was cut back in January 2003 and the mass of stem biomass per plot was recorded (Table 5). The yields were all small, and reflect the fact that this was only an establishment year. They remain therefore an indication of the effectiveness of the varieties in their first year.

Plot Uniformity Analysis.

The initial measurement of the crop in July was used to identify any potential non-uniformity in the plots. To this end, the data were analysed in terms of their position within the field: by row (up and down the field); and by line (across the field). The analyses showed that although there were differences between both rows and lines (reflecting the different varietal compositions of the rows) there were no systematic differences within the plots (Figure 2). It was thus concluded that the plots could be treated as uniform, and that any subsequent differences will be most likely to reflect the variation between plants, than any systematic variation in field properties. This reflects the previous experimental performance of this field reported by the farm manager.

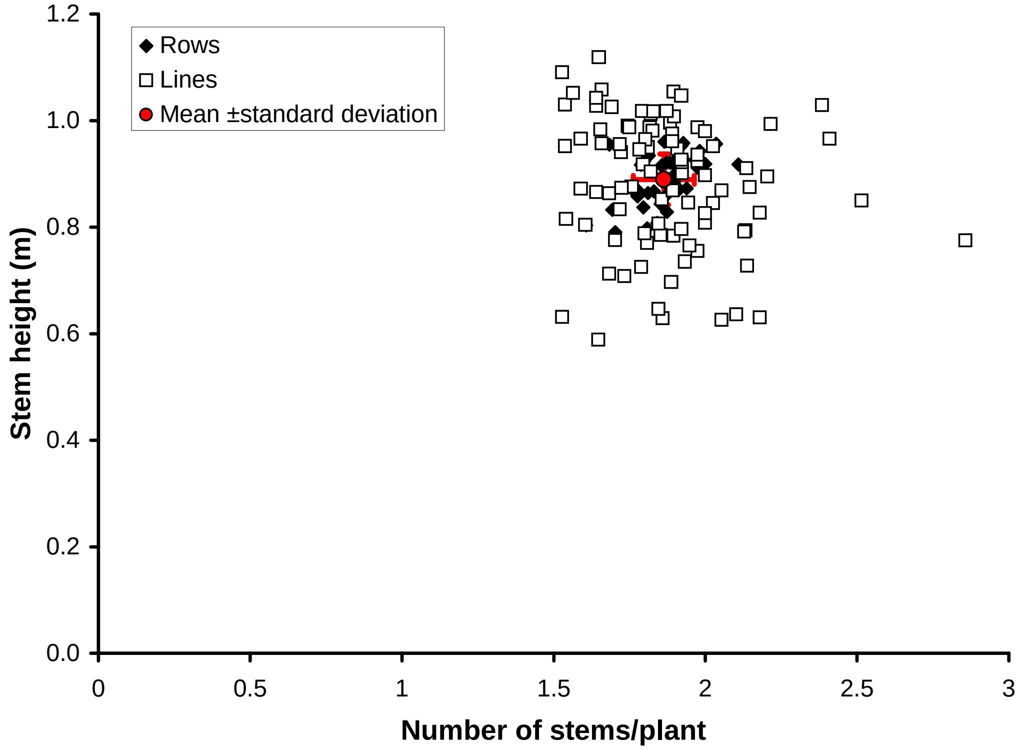

Figure 2. Relationship between stem number and stem height in July 2002 for rows and lines in the Gleadthorpe willow irrigation trial.

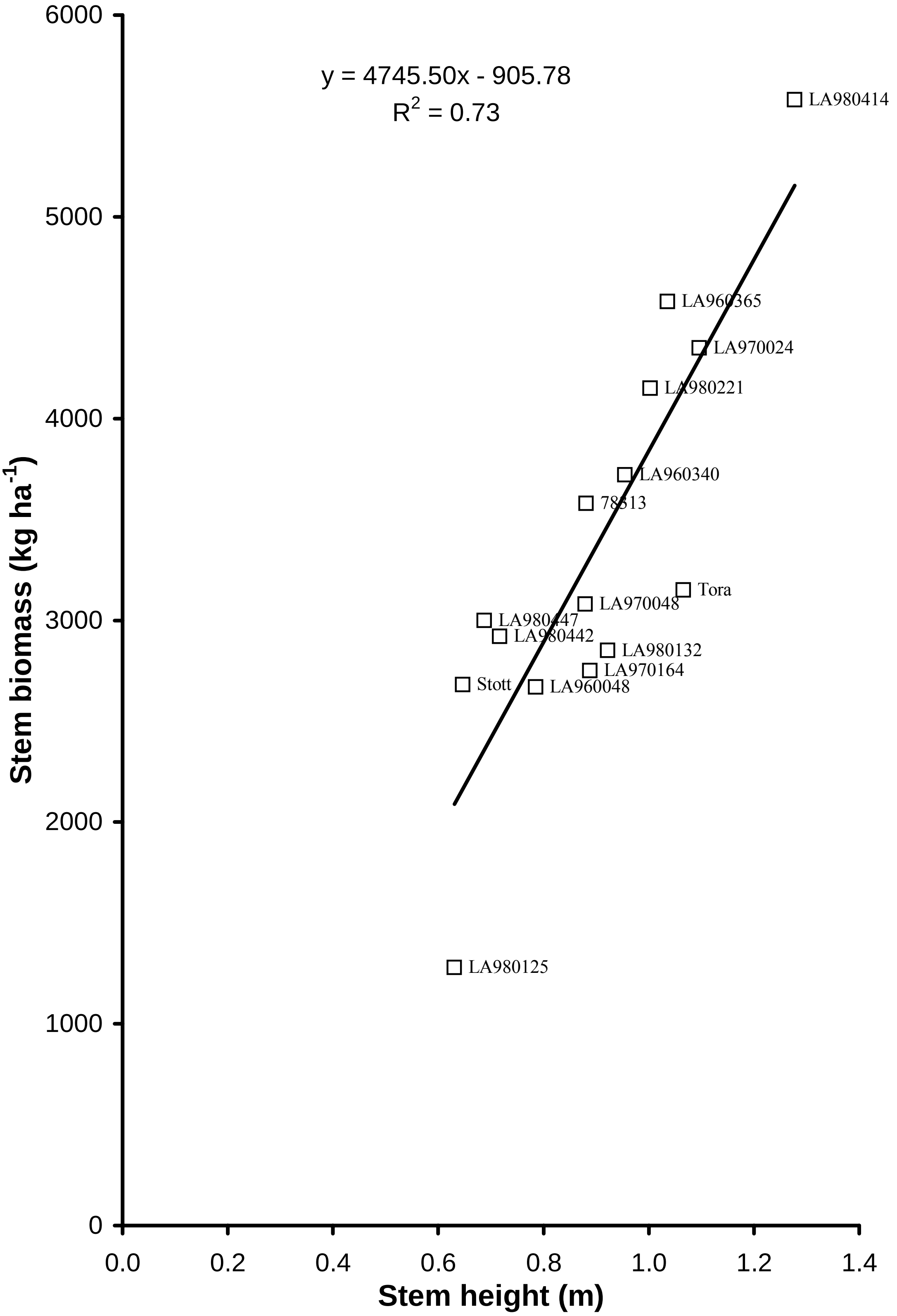

Figure 3. The relationship between stem height in July 2002 and stem biomass at cutback in January 2003: Gleadthorpe willow irrigation trial.

The relationship between stem height in July and biomass in January accounted for 73% of the observed variation with 4.7 t ha^{-1} increase in biomass for each 1 m increase in stem height (Figure

3). Variety LA980414 had the tallest stems and the greatest biomass, outperforming the current standards Tora and Ashton Stott. The very low biomass yield of LA 980125 was related to the poor establishment in the field.

3.1.3 Water balance studies

Water monitoring

Results from the soil water monitoring programmes show that the profile water content under willow varied through time with minima occurring in mid-July and mid-October (Figure 4). These minima indicate that about 100 mm of water had either drained or been abstracted from the soil profile by the willows.

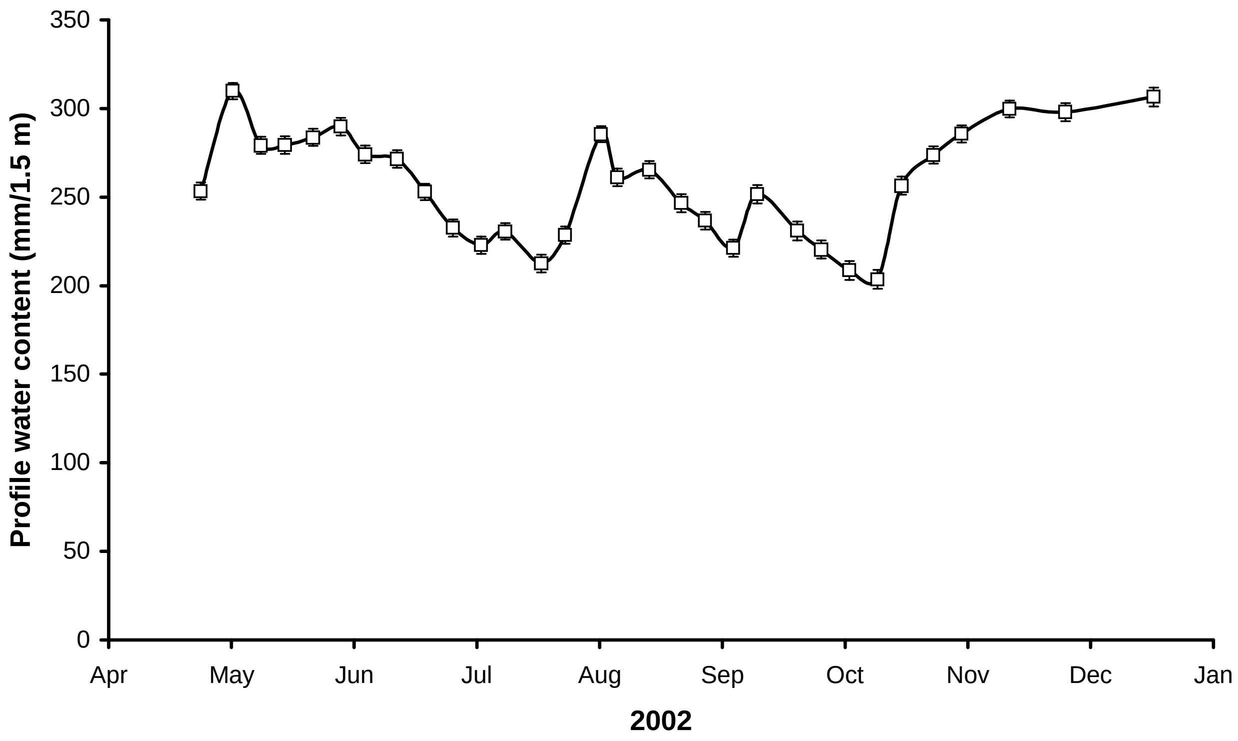

Figure 4. Profile water content under willow through the first season of growth for Gleadthorpe irrigation trial. Error bars show the standard error of the mean (n = 30).

Comparing the volumetric water contents down the soil profile for willow in May and October shows that the water content had decreased at all depths to 1.3 m (Figure 5). This indicates either drainage down the profile or that willow roots had reached this depth by the end of the summer.

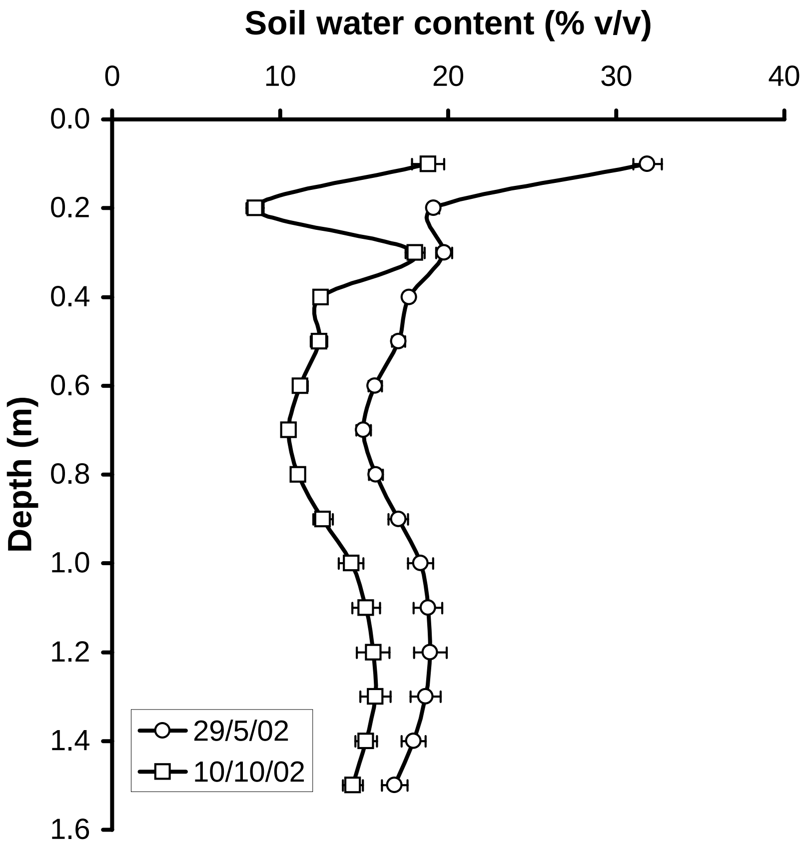

Figure 5. Volumetric water content under willow on 29 May and 10 October 2002: Gleadthorpe willow irrigation trial. Error bars show the standard error of the mean (n = 30).

3.2 Lysimeter studies

3.2.1 Stem height

There was a wide range in the stem height amongst the willow varieties (Figure 6). Generally the parent genotypes had the shortest stems and produced the least biomass. The relative performance of the varieties in the post compared with in the field was an indication of how they were able to withstand the stress (e.g. drought, rooting volume, temperature) that the plants in pots were exposed to.

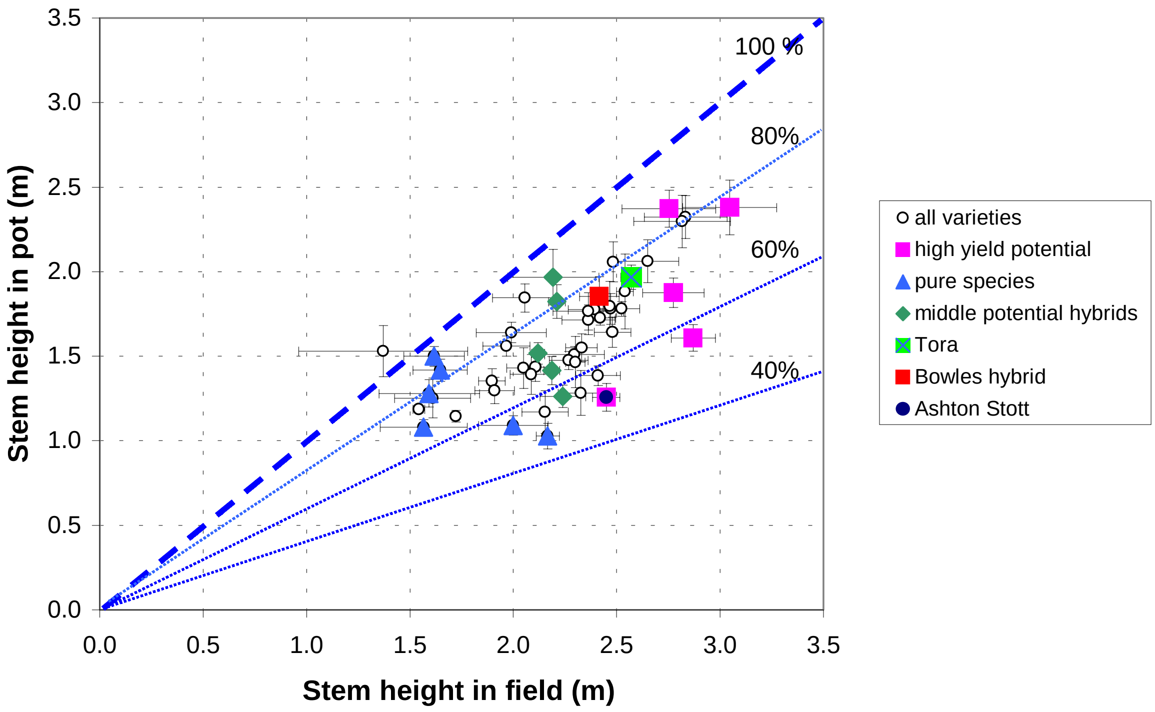

Figure 6. The relationship between maximum stem height of 50 varieties of willow grown in the field and in pots (greater water stress) during 2002. Lines show 40%, 60%, 80% and 100% relative achievement for pots vs field grown plants.

The relationship between stem height in the field and in pots is an indicator of the genotype x environment interactions. For all varieties, stem height in the field accounted for 52% of the variation observed in the pots. On average, stems in the pots were only 73% as high as those in the field, as a result of the greater stress imposed on plants grown in pots.

3.2.2 Leaf shape and specific leaf area

The varieties included in this experiment had a wide range of leaf sizes (Figure 7). The mean leaf width over all varieties was 35±2.0 mm and mean length 170±4.4 mm. The leaf length to width ratio is used as a field tool for separating willow species and was closely related to leaf width with a log-linear relationship accounting for 85% of the observed variation (Figure 8).

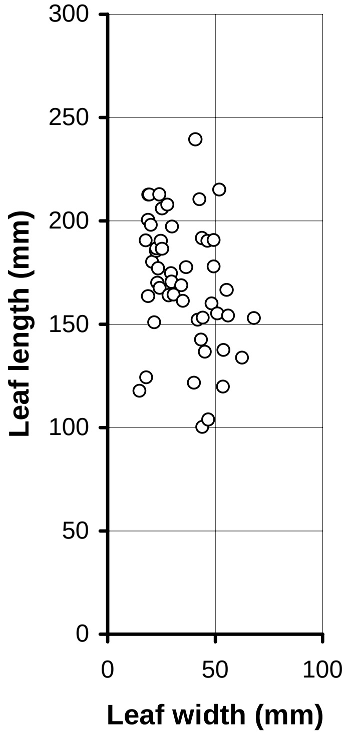

Figure 7. Relationship between leaf length and width for 50 willow varieties.

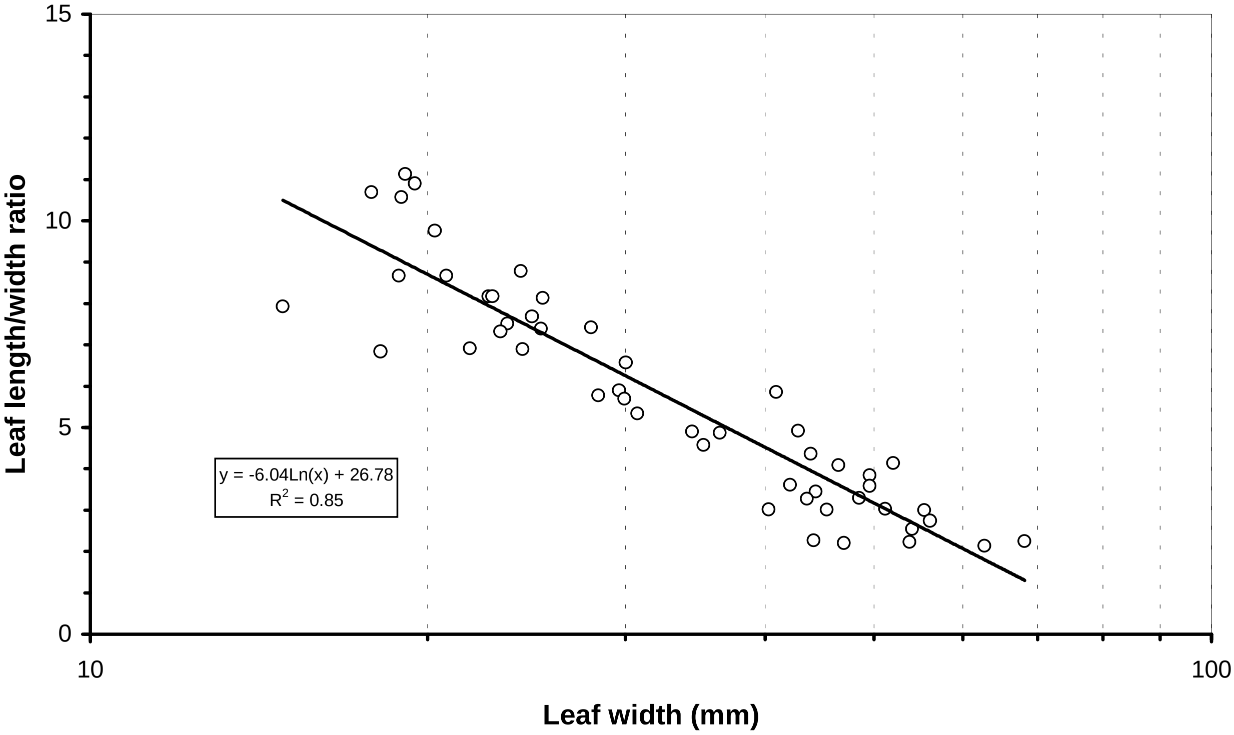

Figure 8. Relationship between leaf length/width ratio and leaf width for 50 willow varieties.

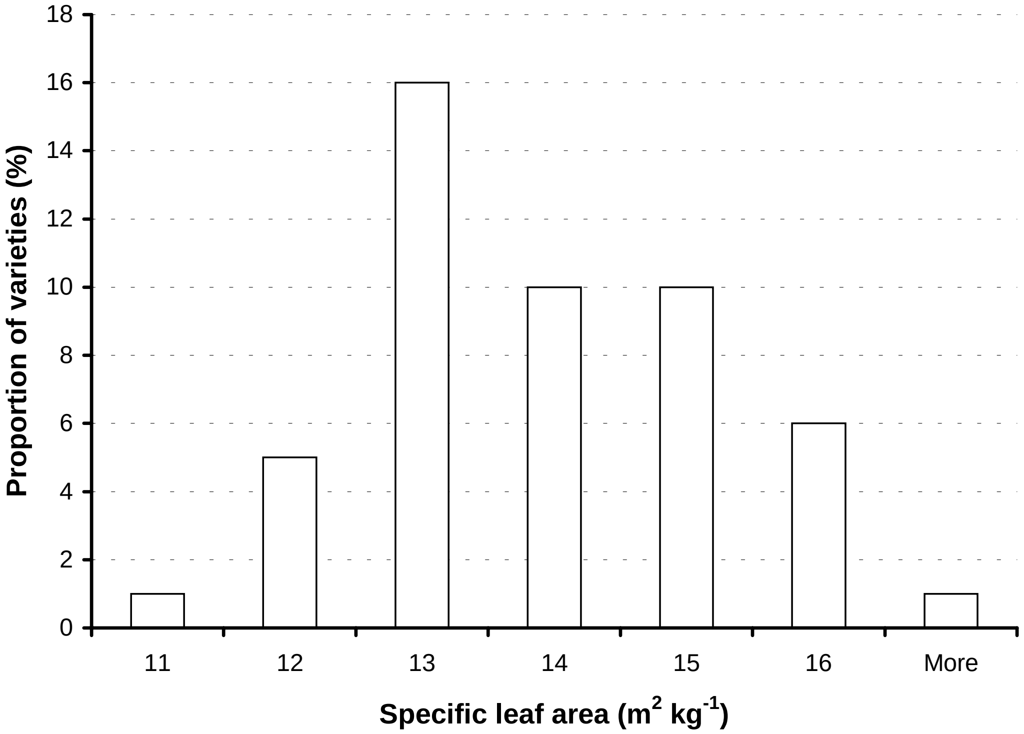

Figure 9. Frequency distribution of specific leaf area for 50 willow varieties.

Specific leaf area (SLA) is sometimes used as an indicator of drought resistance and, within the selected varieties, ranged from less than 11 to more than 16 m^2 kg^{-1} (Figure 9).

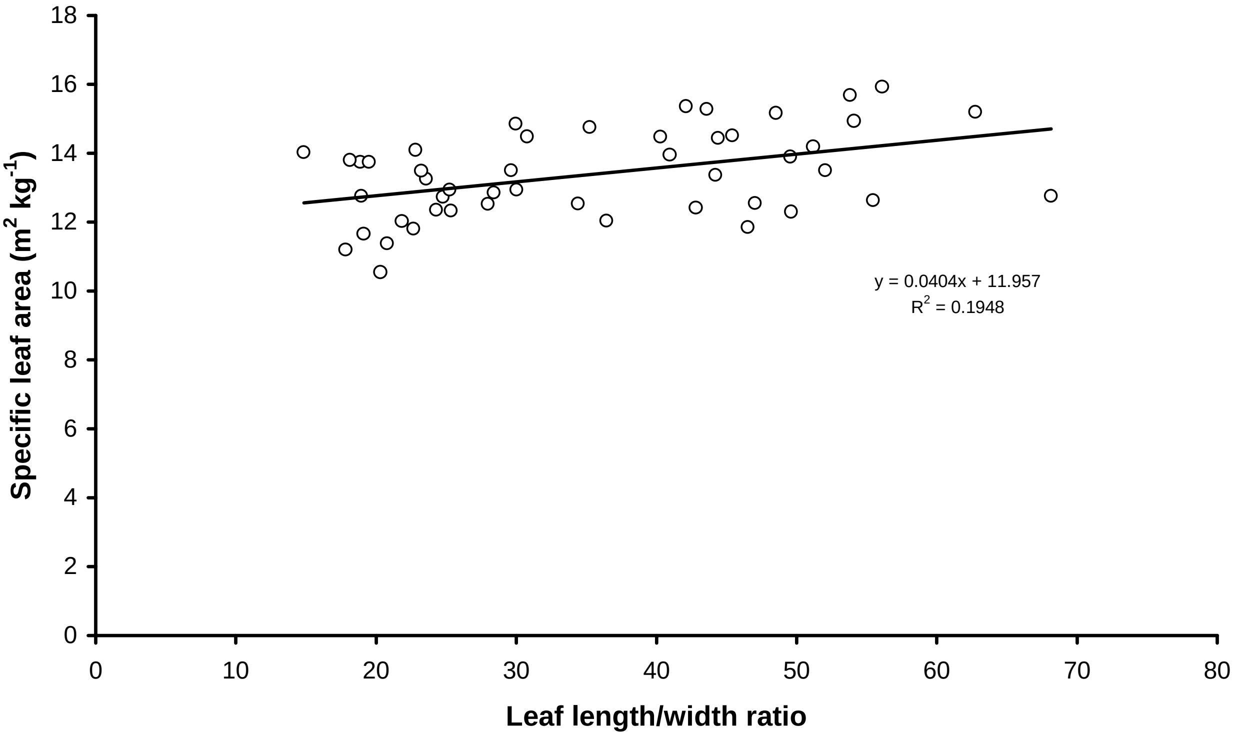

Figure 10. The relationship between leaf length/width ratio and specific leaf area (SLA) for 50 willow varieties.

Leaf length/width ratio accounted for less than 20% of the variation observed in SLA, with a slight positive correlation showing that longer, narrower leaves tended also to be thinner.

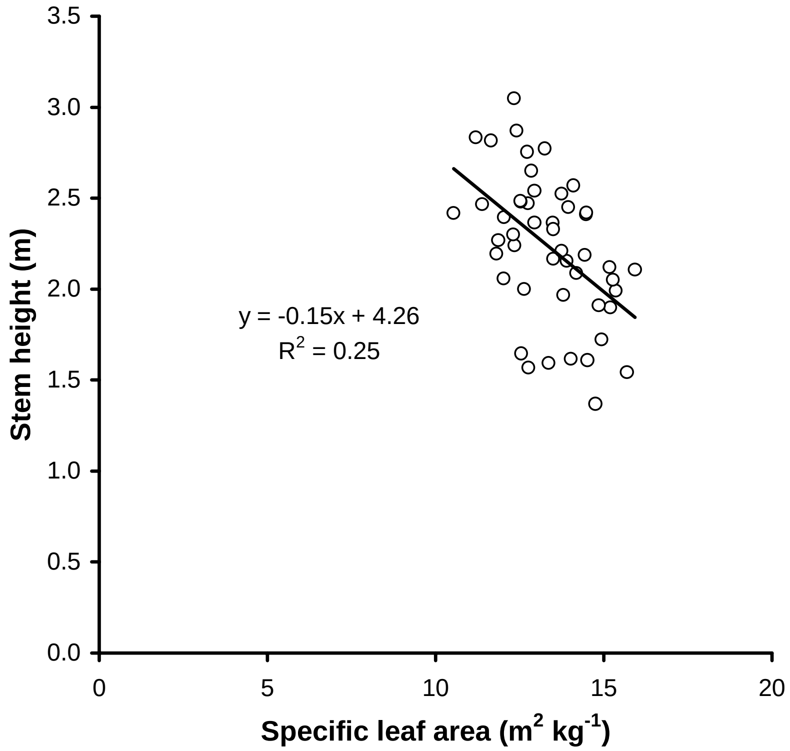

Figure 11. Relationship between stem height in the field experiment after one growing season and specific leaf area for 50 willow varieties.

Stem height tended to decrease with increasing SLA but the relationship only accounted for 25% of the observed variation (Figure 11).

3.2.3 Biomass production

When dry stem biomass was measured at the end of the season, a similar pattern was observed to that shown in stem height (Figure 12). There was, however, a wider range of relative performance than with height, probably related to the number and diameter of stems. Four varieties produced more dry matter in the pots than in the field but this was related to poor field establishment in these varieties. In general the parent material showed the best relative performance but since their overall performance was very low then the stress these varieties experienced was probably less severe than those with a larger leaf area and hence water use.

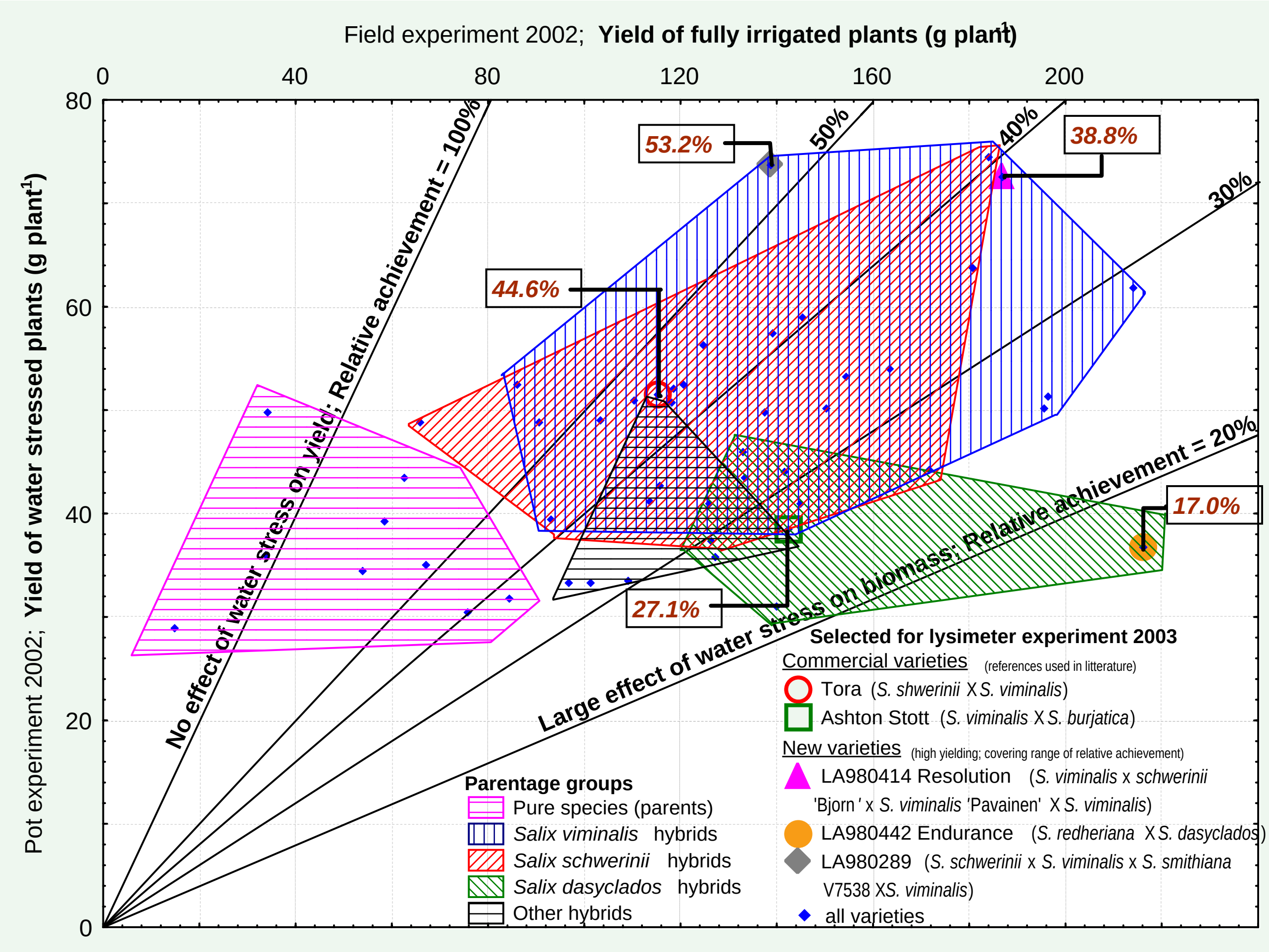

Figure 12. The relationship between biomass production of 50 varieties of willow in the field and in pots (greater water stress) during 2002. Lines show 20%, 40%, 60%, 80% and 100% relative achievement for pots vs field grown plants.

Nine varieties produced more than 160 g plant^{-1} whilst the majority of the hybrids produced between 80 and 160 g plant^{-1}. Plants grown in pots and exposed to water stress mostly produced less than 50% of the biomass of the same variety in the field. Amongst the leading new varieties, Endurance (LA980442) had the best performance in the field but stem biomass production was severely curtailed in the pots. By contrast, LA980289 produced an average amount of stem biomass but also performed relatively well in the pots.

3.2.4 Chlorophyll fluorescence

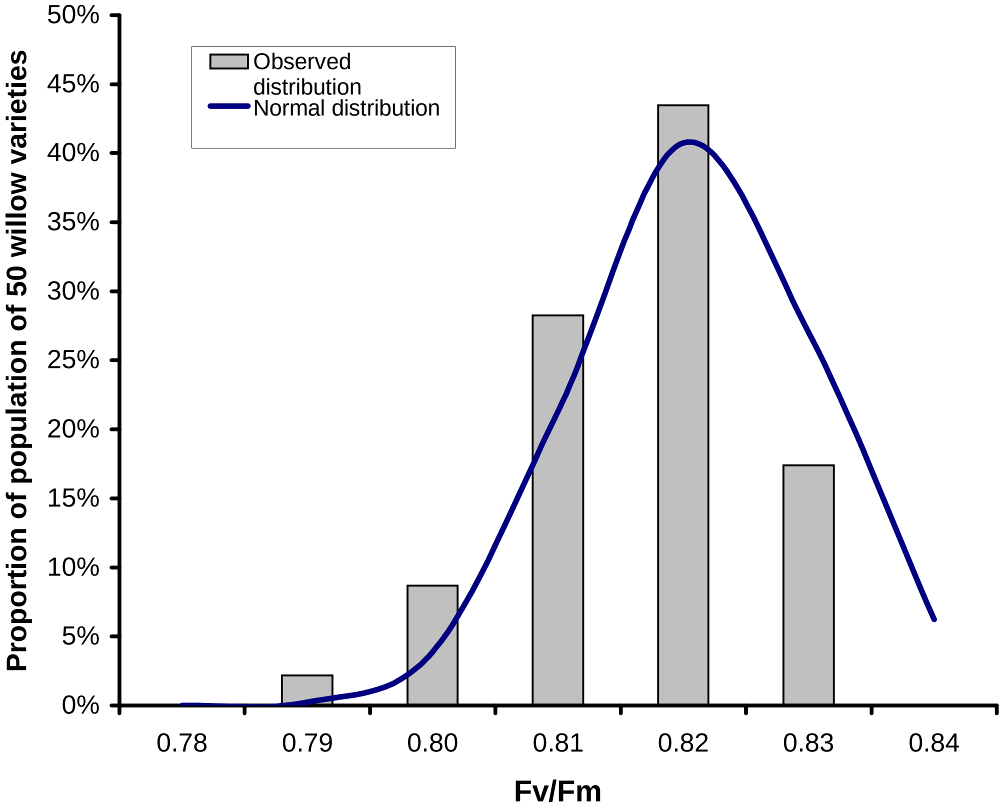

Figure 13. Frequency distribution of the ratio of variable to maximum dark adapted fluorescence (Fv/Fm) for 50 willow varieties grown in pots with episodic drought stress during 2002.

The range of Fv/Fm values observed for the 50 varieties included in this experiment was small and showed little sign of any damage to Photosystem II.

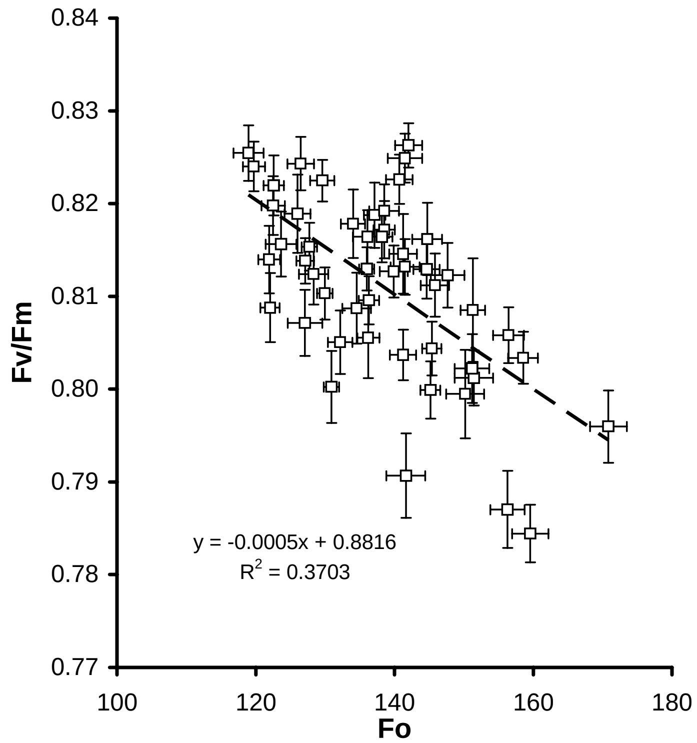

Figure 14. The relationship between the ratio of variable to maximum dark adapted fluorescence (Fv/Fm) and minimum dark adapted fluorescence (Fo) for 50 willow varieties grown in pots and subjected to episodic water stress during 2002.

Relating Fv/Fm to the minimum dark adapted fluorescence (Fo) separates out the varieties to some extent. The four varieties with lowest Fv/Fm values had fairly high Fo values. The two with the lowest Fv/Fm were Bowles hybrid (*S. viminalis*) and *S. glaucophylloides* the other two had these varieties as the female parent. This may indicate a lower photosynthetic efficiency in these clones under drought conditions.

4 DISCUSSION

4.1 Field studies

Initial results from the field-scale experiment at Gleadthorpe indicate that there is likely to be a wide range in stem biomass production by the different varieties. Their relative performance under water stress has not yet been tested as differential irrigation will only be applied in the year after cutback.

The irrigation regime was intended to assist establishment of the experiment during the year after planting. Soil water monitoring, however, identified that the soil became quite dry over two periods during the summer. This may well have had an effect on the leaf area duration of the plants and on stem extension but these parameters were not measured.

As the plants developed through the season they began to extract an increasing amount of water and from a greater depth in the profile. By October, the roots appeared to be extracting water to a depth of about 1.3 m, similar to that observed in experiments elsewhere, though changes in soil water content at this depth may also be attributable to drainage of water from the profile.

4.2 Detailed studies

Establishing the 50 selected varieties in pots and in a field experiment allowed comparative performance to be compared with and without stress. The stress experienced by plants in pots was inevitably a compound effect including temperature (small black pots meant that soil temperature was quite high during summer months), water stress and constriction of the roots. In addition, the relative level of stress was related to the size of plant and the leaf area as larger plants used the available soil water more rapidly than those with a small leaf area.

Despite the wide range of genetic material there was a reasonably good agreement between stem height in the pots and the field. About half of the observed variation could be accounted for by the stem height in the field experiment. The remaining variation could have been related to genotypic differences in drought tolerance of the willow varieties.

Stem biomass of willow varieties grown in pots was more affected by stress than was stem height. In the meso-scale experiment, variety LA980414 now named “Resolution”, produced around twice as much biomass in the first year of growth as the two standard varieties Stott and Tora. Resolution also performed well in the detailed experiments but only out yielded the two standards by between 30 and 60%. By contrast, LA980442 (now named “Endurance”) was the highest yielding variety in the detailed experiments but yielded about the same as Tora and Stott at Gleadthorpe. This may be as a result of water stress during the establishment phase since, in the detailed experiments, “Endurance” had the greatest relative reduction in biomass as a result of water stress.

The preliminary screening of all 50 willow varieties using chlorophyll fluorescence suggests that there was evidence of reduced photosystem function under water stress conditions in only a few varieties. Circumstantial evidence from the same experiment indicated that, even when part of a leaf was necrotic as a result of severe water stress, the remainder showed no signs of damage to photosystem II and hence greater fluorescence. These results agree with previous research on chlorophyll fluorescence in willow (Ögren, 1990).

5 CONCLUSIONS

It is difficult to draw any firm conclusions after one year of a three year project. Preliminary results show that, as expected, there is a wide range of biomass production from the willow varieties included in the experiments reported here and that the relative performance of stressed and unstressed plants also varied widely.

The next two years of the project will allow these preliminary results to be confirmed on the field scale experiment and further detailed physiological measurements to be made on five varieties selected from the original group of 50 grown under stressed and unstressed conditions in lysimeters.

6 ACKNOWLEDGMENTS

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7 REFERENCES

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Ögren, E. 1990. Evaluation of chlorophyll fluorescence as a probe for drought stress in willow leaves: *Plant Physiology* 93:1280–1285.

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APPENDICES

A: Field Experiment: Experimental Layout

B: Detailed experiments: Experimental layout

A: Field Experiment: Experimental Layout

Willow species x drought 2002

HUNGAR HILL (BETWEEN RESERVOIR AND CONCRETE PAD)

N

fence line

12 m

WET BLOCK

12 m 0.7 m 1.3 m **12m disca**

0.5 m

2 m

6.5

XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX
XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX	XX
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N ↑

Willow species x drought 1st year 2002

HUNGAR HILL (BETWEEN RESERVOIR AND CONCRETE PAD)

DRY BLOCK

0.7m 1.3m

X X	X X	X X	X X	X X	X X	X X	X X	X X	X X	X X	X X	X X	X X	X X	X X	X X
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X D	D D	D I	II	IL	L L	L F	F F	F N	N N	N C	C C	C J	J J	J G	G G	G A
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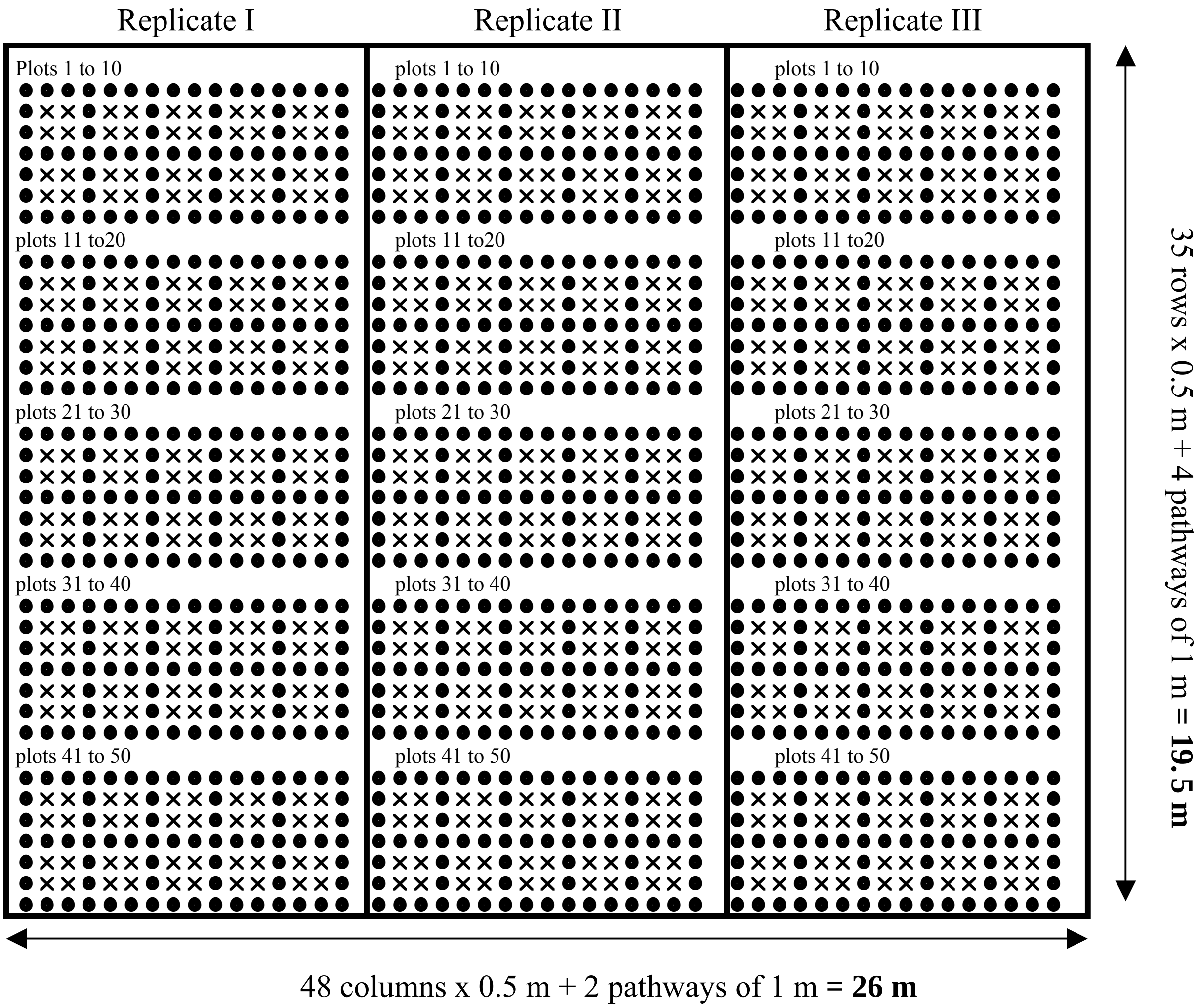
A	78183	J	LA980125
B	Tora	K	LA980132
C	Stott	L	LA980221
D	LA960048	M	LA980414
E	LA960340	N	LA980442
F	LA960365	O	LA980447
G	LA970048		
H	LA970164	X	GUARD PLANT
I	LA970024		

B: Detailed experiments: Experimental layout



● Guard row Q683 ✕ Selected variety

Between each block of 10 plots, a pathway of 1 m was left uncropped to allow access to each plot. Total area = 507 m^2.