

Original Articles

Biomass production in a high-density willow plantation through ten one-year rotation cycles in Central Argentina

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Abstract

High-density willows plantations are used for biomass production in several countries, but in Argentina, information about the adequate management for such plantations is scarce. This fact hinders the development of commercial plantations that could reduce the country dependence on fossil energy. The aim of this work was to analyse the productivity of a willow Short Rotation Coppice (SRC) plantation and to identify potential and actual biomass yields under local conditions. Our hypothesis was that water availability was major limitation for biomass yield in the area, so this factor was included in the trial design. The trial was planted in an agricultural soil in the Rolling Pampas region of Central Argentina, and its yield was measured for a period of 10 years. The factors analysed were irrigation (drip irrigation and rainfed), planting density (13,300 and 20,000 plants ha⁻¹) and two genotypes: *Salix alba* and a *S. matsudana* × *S. alba* hybrid. The trial was disposed in a split-split plot design, and the rotation length was one year. The irrigated treatment consistently produced more than the rainfed one, the yield of irrigated treatment ranged between 10.4-22.6 MG ha⁻¹, and between 2.9-17.6 MG ha⁻¹ for rainfed plots. Yield correlated with water supply during the summer months ($r_s = 0.79$). Biomass production was high in the first four years, but afterwards, yield steadily declined, both in irrigated and rainfed treatments. According to our results, to develop biomass SRC plantations with willows in Central Argentina, irrigation would be necessary during the summer months.

Keywords: short-rotation coppice, water supply, Rolling Pampas, *Salix* spp., irrigation and rainfed.

Introduction

Willows (*Salix* spp.) are suitable for the development of Short Rotation Coppice (SRC) biomass plantations, because of their fast growing, coppicing capability, and high output-input energy ratio (Clifton Brown et al., 2019; Rönnerberg-Wästljung et al., 2022). In some countries, commercial willow SRC plantations to produce biomass have existed for several years (Nord-Larsen et al. 2015, Castellano Albors et al. 2025, Stolarski 2025). In Argentina, this type of plantations has not been developed, although there are marginal areas for agriculture that could be used to produce lignocellulosic biomass (FAO, 2020). In the core forestry region of Northeast Argentina, some companies use wood residues (sawdust, wood shavings and similar) to power furnaces, and to produce chips for industrial and domestic use (Uasuf & Becker 2011; Uasuf & Hilbert 2012). However, for distances greater than 100 Km, biomass transport costs increased considerably (Uasuf & Hilbert, 2012). Hence the need to develop biomass plantations closer to the areas where the biomass will be consumed, to keep both cost and carbon emissions at a minimum (Yang et al., 2020). In Argentina, there are near 43,000 ha of planted willows, mostly used for fibre, pulp, and particleboard production (Secretaría de Agricultura, Ganadería y Pesca, 2025). For these plantations, there is extensive information on appropriate genotypes and silvicultural practices, in consequence, they could be developed into something like non-coppiced high-density Short Rotation Forestry plantations (SRF, Buchman et al., 2020; Stolarski, 2025). Regarding SRC systems, the situation is completely different due to the lack of knowledge about genotypes and management practices adequate for sustainable biomass production under local conditions.

Water supply is a major factor limiting productivity in SRC plantations under different climates and sites (Bergante et al., 2019; Njakou Djomo et al., 2015; Richard et al., 2019). In the case of willows, this is because they are sensitive to water stress compared to other tree species (Wikberg & Ögren, 2007). Despite this, there is genetic variation regarding drought tolerance between different willows species and hybrids (Bonosi et al., 2010; Doffo et al., 2017a). In consequence, it may be possible to select genotypes with a higher water stress tolerance for biomass production (Wikberg & Ögren, 2007; Bonosi et al., 2010).

There is ample evidence that plantation density, genotype and rotation length will affect yield of willows in SRC plantations. Usually, a higher initial plantation density will increase yield (Nord Larsen et al., 2015; Buchman et al., 2020; Wilkinson et al., 2007). But if densities are too high, the competition for resources will increase

plant mortality and in consequence, a decrease in biomass yield will occur (Stolarski et al., 2018). The length of the rotation has an impact too, a longer rotation increased both biomass yield (Stolarski et al., 2018) and the efficiency of energy production (Kulig et al., 2019).

The aim of this work was to analyse the productivity of a willow SRC plantation in an agricultural soil over a span of ten 1-year rotation cycles. During this period, we evaluated the effect of water supply, plantation density and genotype on biomass yield. We expected that these factors will be relevant for willow biomass plantations under the conditions of the agricultural area in the “Pampa Ondulada” (Rolling Pampas) region (SAGYP 1995). We analysed two willow genotypes bred for the area to produce pulp and paper, not biomass. Nevertheless, these clones show fast and erect growth, and vigorous re-sprouting capabilities in stool beds. These traits are selected in genotypes aimed for biomass production (Karp et al., 2011). Our hypothesis was that the irrigated treatment will provide a glimpse on the potential yield for high-density willow plantations in the area, while the rainfed treatment represented the actual yield, attainable in a commercial plantation without irrigation. The biomass yield of 2 years had already been published (Doffo et al., 2017a). In this work, we report eight more years of yield data, following the evolution of the plantation for a total of ten years. We think that this long-term evaluation of biomass productivity is necessary to ascertain the sustainability and economic viability of SRC willows plantations in this region.

Methods

Experimental design, plantation management and meteorological data. The field trial was planted in La Plata, Buenos Aires, Argentina (34°59'09" S; 57°59'42" W). The climate is temperate, with an average temperature of 17°C and a mean historical annual rainfall of 1,070 mm. The soil was a typic Argiudoll (Hurtado et al., 2006), with 26% clay, 46% silt and 28% sand. The soil nutrient content at plantation was adequate (Doffo et al., 2017a). The plantation was established over plastic mulch, therefore, weed control was unnecessary. Eight treatments were evaluated, which resulted from the combination of three factors with two levels in a split-split plot design. Three blocks were established following the natural slope of the terrain. Each block was further divided into two main plots according to the irrigation treatment: rainfed (abbreviated R) and drip irrigation (I). The main plots were 4 m apart to avoid lateral diffusion of the irrigation water. The main plots were divided into two sub-plots according to plantation density: low density (LD, 13,333 plants ha⁻¹) and high density (HD, 20,000 plants ha⁻¹). Each sub-plot was further

divided into two sub-sub-plots according to genotype: *S. matsudana* Koidz. × *S. alba* L. ‘Barrett 13-44 INTA’ (abbreviated Clone B), or *S. alba* L. ‘Yaguareté INTA – CIEF’ (abbreviated Clone Y). The blocks and plots were surrounded by a border of clone *S. alba* × *S. babylonica* L. ‘Ragonese 131-25 INTA’ (Figure 1). Each sub-sub-plot was formed by a total of 56 plants. In all plots, the distance between rows was 1m, the distance between plants in a row was 0.75 m in the low-density and 0.5 m in the high-density sub-plots. The size of the LD sub-plots was 48 m² and for the HD sub-plots 28 m².

The temperature and rain data were registered in an automated meteorological station (Davis Instruments) placed 200 m away from the plots (Figure 2). The growing season extended from August (sprouting) to May, when leaf abscission occurred. The rainfall data for the complete growing season was calculated from June to May the following year, and for summer as the sum of the months of December, January, and February (Figures 2 and 3). Actual rainfall was then compared to the sum of the average monthly rainfall for the period 1964-2014.

Drip irrigation was applied 3-4 times a week between November and April, if there was no rainfall during this period (Figure 2). The irrigation equipment had 2.48 emitters per square meter and with a working pressure of 0.5 bar allowed for the application of 2.51 mm hour⁻¹ of irrigation water. This was multiplied by the number of working hours of the irrigation system to estimate the amount of water supplied.

Irrigation maintained the topsoil at field capacity, while the rainfed plots experienced any drought period that might occur. The year of plantation received an unusually high amount of rain (356 mm above the historical mean for the area, Figure 2), in consequence irrigation was not applied. During the growing seasons 2017-18 and 2018-19 it was not possible to supply water to the drip irrigation system, then, for these years there were only clone and plantation density as factors.

There was mortality in the borders of the plots, and these plants were replaced. Plant survival inside the plots was 100% until the 2015-16 season, and from 2016 to 2021, the survival was above 90%. Up to then, plant mortality did not have a significant effect on yield estimates. But in 2022-2023, an extraordinary drought episode occurred, and the plant mortality in the non-irrigated plots increased. An assessment of plant survival was carried out in the spring of 2022. Sprouted plants were counted as alive, and non-sprouted plants as dead.

Yield determination. During the 10 years of the trial, all the plants were harvested and measured every season, thus the rotation length extended for a year. The harvest was carried out manually during the non-growing season (winter). The two central rows (24 plants) of each sub-sub-plot were cut, and the total fresh weight of all stools was determined with a 300 Kg dynamometer (error 0.1 Kg). From a subset of 3 randomly selected plants, the fresh and dry weight at

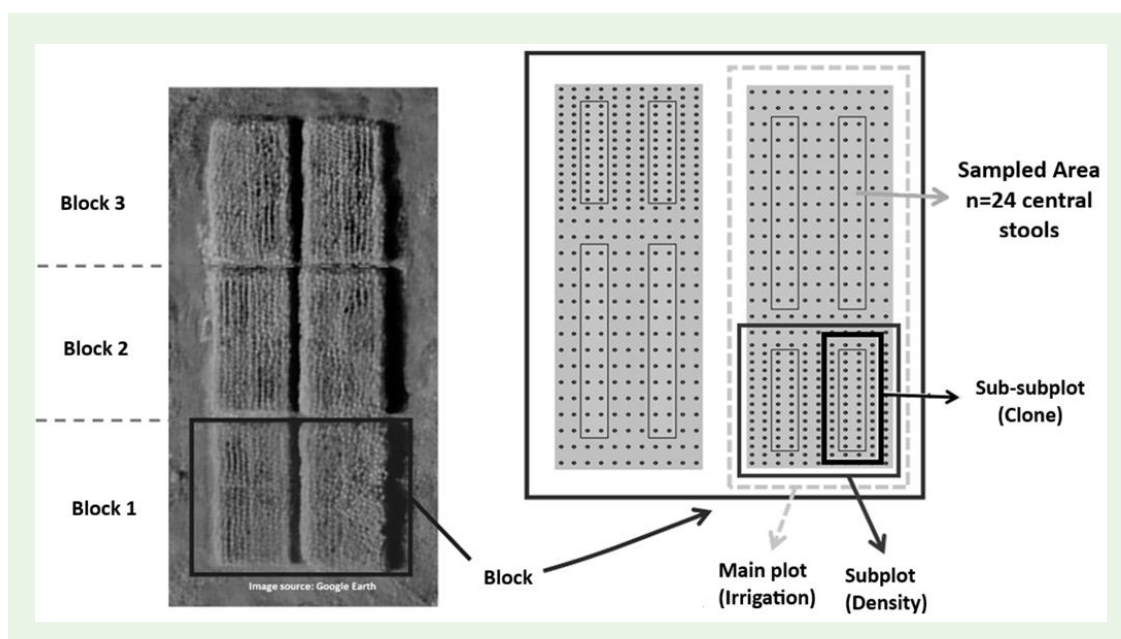


Figure 1. Layout of the split-split plot design.

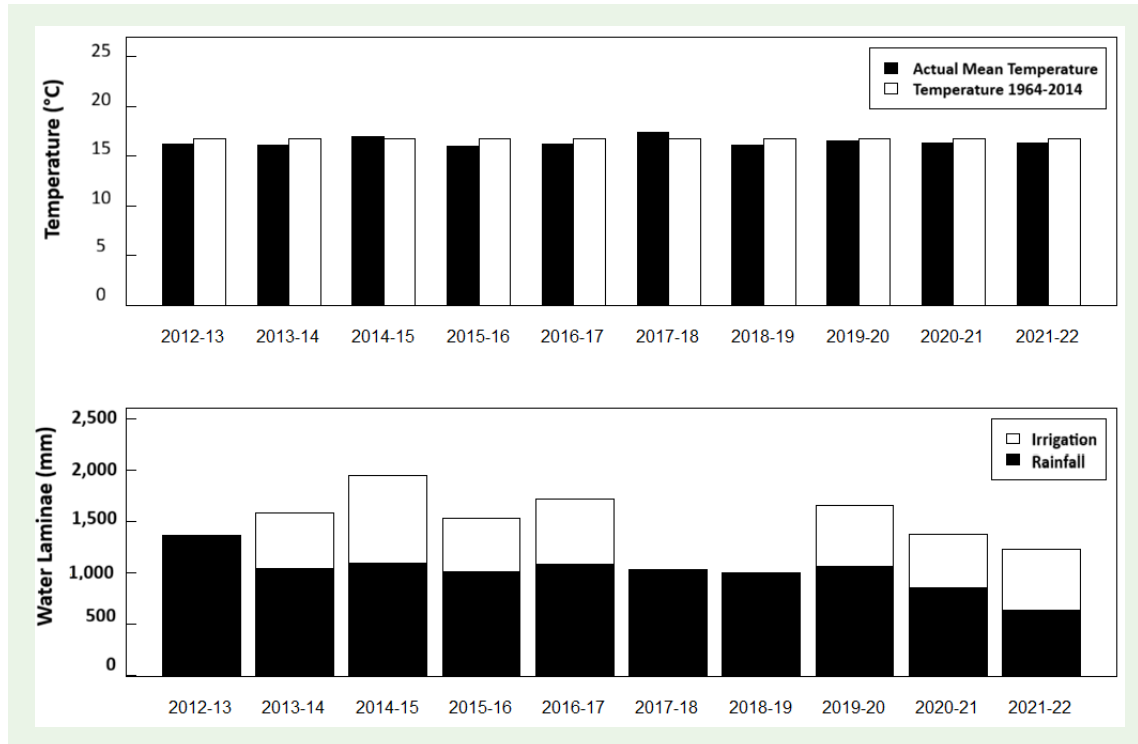


Figure 2. Yearly data for mean temperature (panel A, actual and historical between 1964 and 2014), and rainfall plus irrigation for the period of the trial (panel B). Each year represents a growing season (June to May of the following year).

105°C were determined, this dry weight to fresh weight ratio was used to determine the dry weight of the harvested biomass. Yield per hectare was calculated by extrapolating the measured plot yield.

Statistical Analysis. The data were analysed for each year growing season using a linear mixed model with the R version 4.0.3 (R CoreTeam 2020) using the *lmer* function from the *lme4* package (Bates *et al.* 2015), with irrigation, plantation density, and clone as fixed effects. The equation for the model was:

$$y_{ijkl} = m + a_i + \beta_j + c_k + d_{ij} + f_{ik} + g_{jk} + h_{ijk} + b_l + p_{il} + sp_{jil} + e_{ijkl} \quad [1]$$

Where m is the general mean, a_i is the i^{th} level associated to the main plots, β_j is the j^{th} level associated with the sub-plots within the main plots, c_k is the k^{th} level associated with sub-sub plots within the sub-plots, and d_{ij} , f_{ik} , g_{jk} and h_{ijk} the interactions. The random terms in the model were the block effects b_l , the plot effects p_{il} , the sub-plot effects sp_{jil} and the experimental error e_{ijkl} .

The mixed model was estimated through restricted maximum likelihood (REML), and the Satterthwaite method was used to

estimate the degrees of freedom. The data for the 2019-2020 season were transformed to natural logarithm to meet the normality requirements. The pairwise comparisons were carried out using the *emmeans* package with the Tukey HSD method (Lenth, 2022).

To evaluate the effect of time on yield, a second mixed model was adjusted with the same packages, with year and irrigation as fixed effects. In this second model, the years when irrigation was not applied were excluded from the analysis. The equation for this model was:

$$y_{ij} = m + a_i + \beta_j + d_{ij} + b_l + p_{il} + e_{ij} \quad [2]$$

Where m is the general mean, a_i is the i^{th} level associated to the main plots (irrigation), β_j is the j^{th} level associated with the year, and d_{ij} the interaction. The random terms in the model were the block effects b_l , the main plot effects p_{il} , and the experimental error e_{ij} .

The data for survival in 2022 did not comply with the normality requirements, in consequence were analysed with the Kruskal-Wallis non-parametric method followed by pairwise comparisons by the Wilcoxon (Mann-Whitney) test.

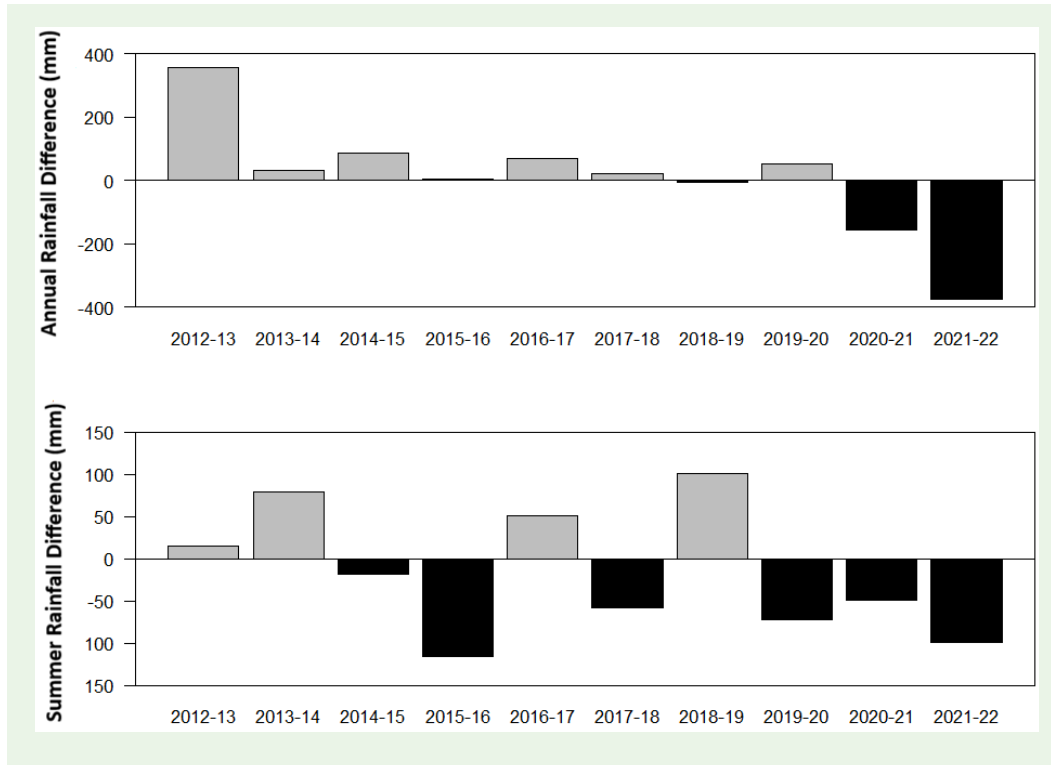


Figure 3. Difference between the annual historical (1964-2014) and the actual rainfall for the complete year (annual, panel A) and summer (December, January, and February, panel B) for the ten-year period analysed. Positive values indicate higher rainfall, whereas negative ones indicate a lower rainfall compared to the historical values for the period.

For correlation analysis, the Spearman Correlation Coefficient (r_s) was used, as the relationship between the variables was not clearly linear, albeit monotonous (Logan, 2010).

Results

The average mean temperature was slightly lower than the historical mean for most of the ten-year period, while the total amount of rainfall and its distribution were different over the years (Figure 2). The total amount of rainfall was usually similar, or above the historical value (Figure 3), except for the last two seasons (2020-2021 and 2021-2022) when the values were substantially lower because of the occurrence of La Niña dry period. If we focus on the summer months, the situation varies (Figure 3). Usually, the summer rainfall was below its historical values, even when the total annual precipitation was similar or above these values. For the most part of the ten-year period, summers were drier than average. The 2021-2022 season was exceptionally dry: annual rainfall was 376 mm below the historical average for the area.

Biomass data for all the years are shown in Figure 4. Over all the years when irrigation occurred, this factor was statistically significant (Table 1), and the yield of the irrigated treatments was higher than that of the rainfed ones. During the years 2013-2014 to 2016-2017, there were statistically significant differences between irrigated and non-irrigated treatments. Clone was significant in some years, but not in others, while planting density was significant only in the first harvest, and never again afterwards. In the years when the clone effect was significant, usually Clone B showed a higher yield than Clone Y. The exception was the first year, when Clone Y showed a higher yield. The higher yields occurred between the second and the fourth harvest. Afterwards, a steady decline in yield for all treatments, including the irrigated ones, was observed. This evolution of productivity is illustrated in Figure 6, depicting the yearly biomass for irrigated and rainfed treatments. In the later years (2020-2022), the decline in yield was statistically significant compared with the early years (Figure 6). In this analysis, both year and irrigation were significant, but their interaction was not (Table 2).

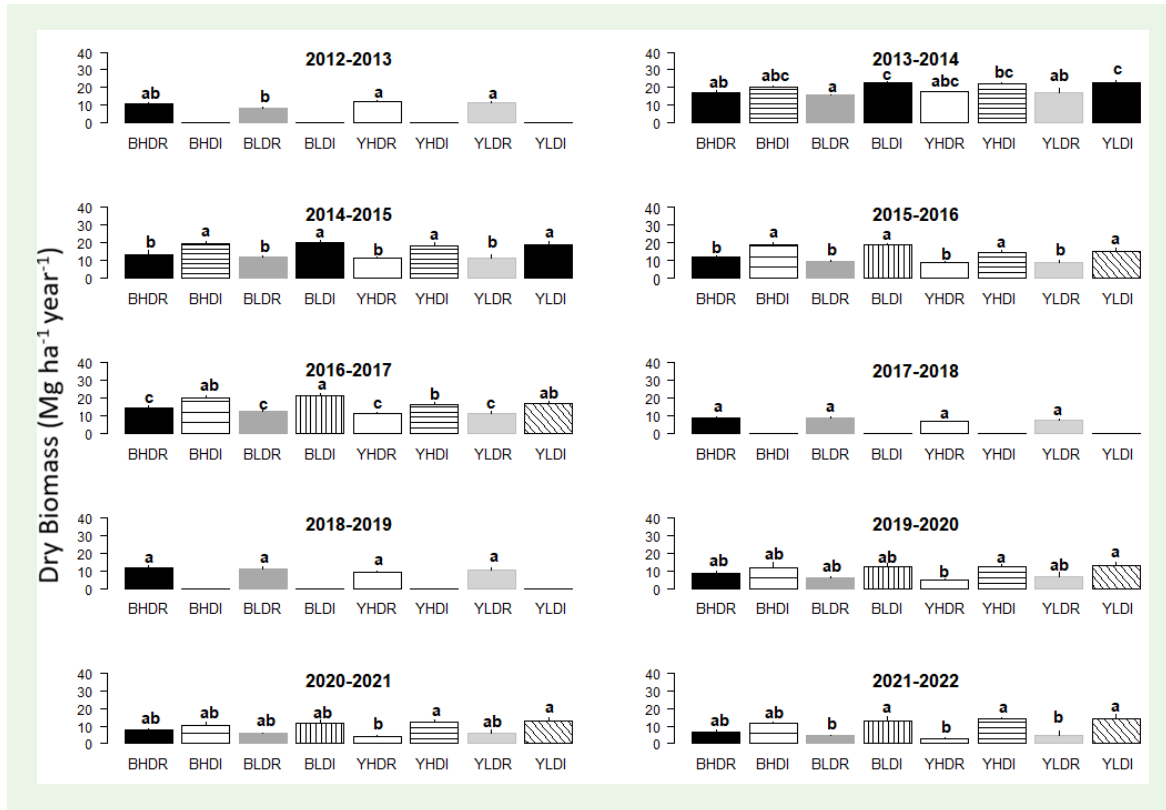


Figure 4. Dry biomass yield for the different years of the experiment. Vertical bars: Mean standard error. The values with the same letter do not differ at $P < 0.05$ according to the Tukey test. Treatments Key: B: Clone B, Y: Clone Y. HD: High density (20,000 plants ha^{-1}), LD: Low density (13,333 plants ha^{-1}). R: Rainfed, I: Irrigated. The biomass data for 2013-14 and 2014-15 have already been published (Doffo et al., 2017a).

Table 1. Mixed model analysis of biomass production for each year. Factors: Irrigation (I), planting density (D), clone (C). Code References: *** = $P < 0.001$; ** = $P < 0.01$; * = $P < 0.05$. ns = not significant. ---: Not determined this year.

Year	Irrigation	Density	Clone	IxD	IxC	CxD	IxDxC
2013	---	*	**	---	---	ns	---
2014	*	ns	ns	ns	ns	ns	ns
2015	*	ns	ns	ns	ns	ns	ns
2016	*	ns	**	ns	ns	ns	ns
2017	***	ns	***	ns	ns	ns	ns
2018	---	ns	ns	---	---	ns	---
2019	---	ns	ns	---	---	ns	---
2020	*	ns	ns	ns	ns	ns	ns
2021	*	ns	ns	ns	ns	ns	ns
2022	*	ns	ns	ns	ns	ns	ns

Table 2. Mixed model analysis of biomass production including year and irrigation as fixed effects. Code Reference: *** = $P < 0.001$; ** = $P < 0.01$; * = $P < 0.05$. ns= not significant. The model includes the data for the years 2013-2014 and 2014-2015 that have already been published. The years without irrigation were not included in the model.

Factor	Significance
Year	***
Irrigation	***
Year x Irrigation	ns

The plant survival was very high (above 90%), until the 2021-2022 season when an extreme drought event occurred, and plant mortality increased. Plant survival (PS) was assessed after the 2022 harvest (Figure 5). The survival of the irrigated sub-subplots was 99% on average, while in the rainfed treatments, it was higher in Clone B (96 and 91% for high and lower density) than in Clone Y (80 and 70% for high and lower density).

The correlation of biomass yield with water supply (rainfall plus irrigation, if any) was analysed for all the years, including those already

published (Figure 7). The correlation with water supply along the year and water supply in summer was 0.80 and 0.79, respectively, and highly significant. When the same relationships were calculated on clone basis, Clone Y showed a higher correlation than Clone B with water availability.

Discussion

Our results confirm that water availability is the crucial factor for yield of willow SRC in the Rolling Pampas of central Argentina, as have been shown for other regions (Bergante et al., 2010; Njakou Djomo et al., 2011). Every year that irrigation was applied, it had a significant effect on biomass yield (Figure 4, Figure 6 and Table 1). Of the other factors analysed, genotype was significant in some years, and density only in the year of plantation. In general, a higher plantation density increases yield (Wilkinson et al., 2007), but sometimes it depends on the cultivar analysed, and the length of the rotation (Kulig et al., 2019). Stolarski et al., (2018) tested doubling planting densities in willows (12,000; 24,000; 48,000; and 96,000 plants ha^{-1}); with a one-year rotation, biomass production peaked at 24,000 plants ha^{-1} , and the yield was lower in the other densities. Lack of response to density in our trial may be because the values we used (13,333 and 20,000 plants ha^{-1}) were not different enough to cause an impact in a one-year rotation. The yields of the first growing season (2012-2013) were lower compared to the other

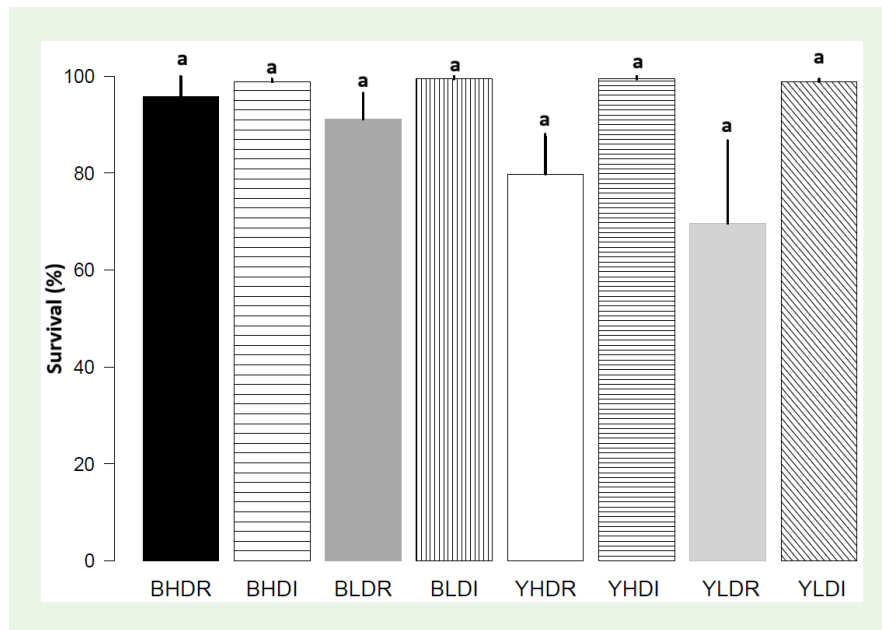


Figure 5. Plant survival (%) after the 2021-2022 season. Treatments Key: B: Clone B, Y: Clone Y. HD: High density (20,000 plants ha^{-1}), LD: Low density (13,333 plants ha^{-1}). R: Rainfed, I: Irrigated. Means with the same letter did not differ at $P < 0.05$.

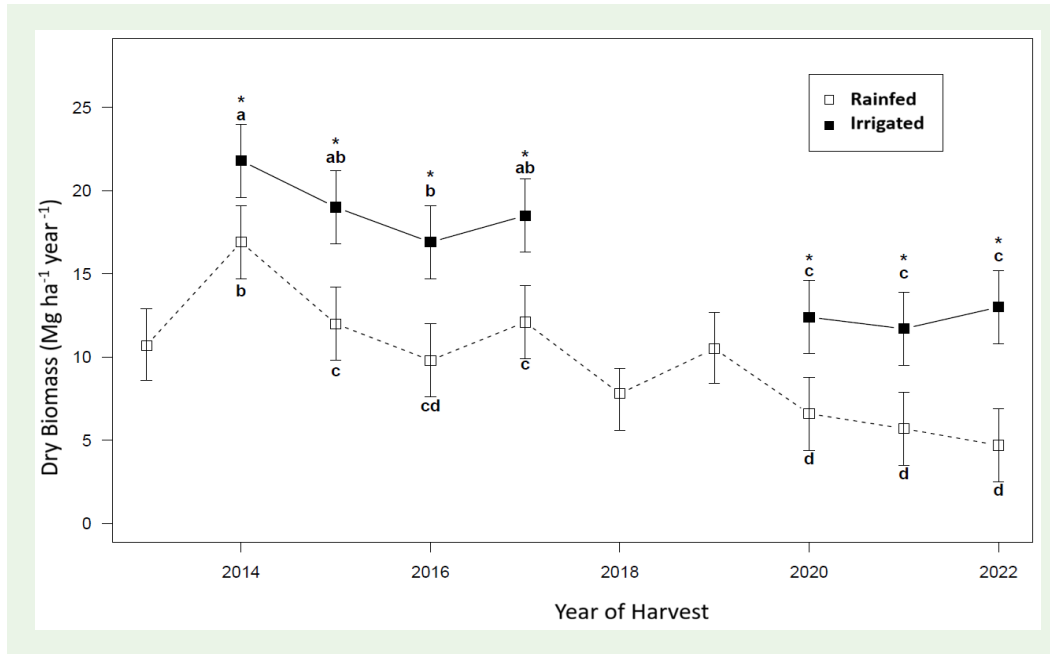


Figure 6. Dry biomass yield for rainfed and irrigated treatments over the different years of the experiment. Vertical bars: 95% confidence interval estimated with the Kenward-Roger’s method. Means with the same letter did not differ according to Tukey test. The non-irrigated years were not included in the analysis.

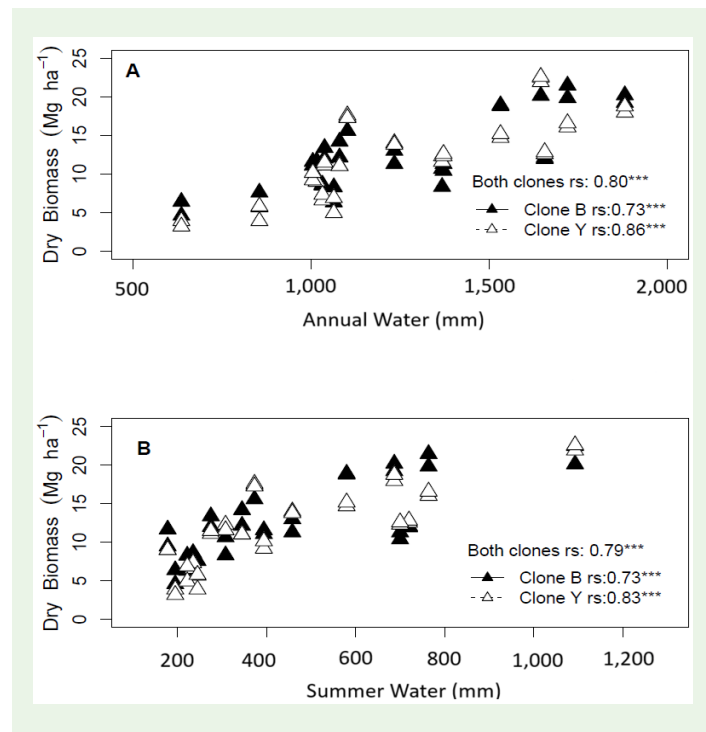


Figure 7. Spearman correlation coefficient (rs) of dry biomass vs. water supply for each clone and for both clones together. Panel A: Biomass vs. Annual water supply (year-round water supply, Rainfall plus irrigation from June to May). Panel B: Biomass vs. Summer water supply (Rainfall and irrigation in December, January, and February). *** = $P < 0.001$; ** = $P < 0.01$; * = $P < 0.05$.

years. This is to be expected since, at this point, the plants were developing a root system as they were originally planted as rootless stools. Over the following years, the plants increased their above ground growth, because they did not need to invest resources in the root system as in the first year of planting.

The historical average rainfall in the area is around 1,000mm per year, but the distribution along the year is quite uneven (Figure 3). The summer months (December, January, and February) are crucial because they are the months of maximum growth and higher evaporative demand. The correlation coefficient of yield with water supply of the whole growing season is similar with the correlation of yield with water received in the summer months (Figure 7). The occurrence of exceptional dry years because of La Niña episode, like 2021-2022 (Figure 3), took a heavy toll on the yield of non-irrigated plots. The reduction in yield was the result of physiological changes induced by drought (Wikberg & Ögren, 2007) and because of higher plant mortality in the rainfed treatment, especially in clone Y (Figure 5). Higher mortality in Clone Y was likely caused by its sensitivity to drought. Clone Y has a lower root to shoot ratio than Clone B (Doffo et al., 2017b), and this fact could hinder its ability to explore a larger volume of soil. In addition, Clone Y has vessels with a higher diameter than Clone B (Achinelli et al., 2018). Poplar plants with higher vessel size were more susceptible to embolism (Jacobsen et al., 2019), and the same could happen for willows.

During the first years, the rainfed annual yields were like willow plantations with genotypes improved for biomass production (Cunniff et al., 2015; Volk et al., 2017; Castellano Albors et al., 2025), but afterwards, yield diminished steadily (Figure 6). There is a strong correlation between biomass yield and plant survival (Nord Larsen et al., 2015), but the decline in yield over the years could not be explained by plant mortality in the irrigated treatments, since they had a 99% survival at the end of the tenth season (Figure 5). As there was no fertilization, one possibility is that the soil nutrients declined with the successive harvests, even though there was an ample initial supply at the site (Doffo et al., 2017a). Nutrient extraction with harvest was evaluated in the 2015-2016 season. There was a considerable extraction of N, Ca, K and P, and the irrigated treatment extracted more nutrients than the rainfed one (Doffo et al., 2024). But the decline in productivity with age could happen even in fertilized SRC plantations. Kopp et al., (2001) analysed a willow high-density plantation with yearly harvest for ten years, that was fertilized and irrigated. They found that biomass production peaked in the fifth year, but become erratic afterwards, and there was no difference in yield between fertilized and non-fertilized treatments. In this case, fertilization did not prevent the reduction in yield in the late years of the plantation, caused mainly by aging of the plants. It is

possible that something similar happened in our trial: the aging of the plants combined with the yearly harvest (a stressful situation for plants) likely caused the productivity reduction in the later years.

In Argentina, there are no commercial plantations of willows dedicated solely to biomass production. The biomass used for energy are residues of forest and agricultural industries (Uasuf & Becker, 2011; Roberts et al., 2015). The results of this work are important for an eventual development of SRC willows plantations in the future. In commercial biomass plantations, the actual yields will be lower than in this trial. For commercial plantations, the best agricultural soils and irrigation will not be used and weed control will be less efficient. With a one-year rotation, the yields in the rainfed plots are like SRC systems with improved genotypes for the first 4-5 years. But the decline in the following years put the biomass production in the lower end of the yield spectra (Njakou Djomo et al., 2015). It is likely that longer rotations will delay the steep decline in yield caused by the yearly harvest (Santucci et al., 2024; Castellano Albors et al., 2025), and the optimal rotation length for the area should be tested in the future. Other alternatives to explore in future experiments is fertilization and the use of genotypes with higher drought tolerance (Richard et al., 2019; Oliveira et al., 2020).

Ideally, the development of short rotation plantations should focus not only on biomass but in obtaining other materials and chemical compounds of interest for the industry, thinking of willows as biorefinery feedstocks (Baker et al., 2022; Stolarski, 2025). In addition, high density willow plantations could provide environmental benefits like carbon sequestration, soil protection from erosion and phytoremediation (Istenic & Bozic, 2021; Stolarski, 2025). The goal should be developing this kind of willow SRC systems as multipurpose plantations, not just as a source of biomass for energy.

Conclusions

Water supply is the main factor affecting willows biomass yield in the Rolling Pampas region of Argentina. If commercial SRC willows plantations are established in the area, it will be necessary to irrigate, at least during the summer months, in case of an exceptionally severe drought occurs.

Regarding the genotypes evaluated in this work, Clone B seems more adequate for SRC plantations because of its higher yield and survival than clone Y. But a more extensive evaluation of suitable material remains to be done. In addition, other rotations lengths should be tried to determine the ideal for the area, and to ameliorate the yield decline of the later years of the yearly harvested plantation.

Author contributions

FGA and VMCL designed the experiment. FGA, SM, MB, GND and MER performed the field measurements. FGA and VMCL analyzed the data. VMCL wrote the first draft of the manuscript, and all authors read and approved the final version.

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Data availability

The datasets generated during the current study are deposited at the CONICET institutional repository (<http://hdl.handle.net/11336/244393>).

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