



## Research paper

## Effects of cutting traits and competition on performance and size hierarchy development over two cutting cycles in willow



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## ABSTRACT

To assess if cutting traits (clone, fresh weight) in combination with strong competition from adjacent willows (planted in monoclonal stands and clonal mixtures) affected performance and size hierarchy development in willow during consecutive harvests, a field experiment was performed in central Sweden during 2008–2015. Cuttings from five *Salix* clones were planted in 1.8 m × 1.8 m plots, at a density of 197 500 cuttings ha<sup>-1</sup> in monoculture and in ten mixtures using two clones (i.e. 15 combinations). Maximum height was measured each year and the experiment was harvested after the growing seasons 2012 and 2014. Larger cuttings produced stools with lower death rate, higher maximum height and larger biomass production compared to smaller cuttings. The high competition pressure resulted in 9.2% stool death at harvest 2012 and 39% stool death at harvest 2014. Tora (*Salix schwerinii* × *Salix viminalis*) showed the highest performance and Jorr (*Salix viminalis*) and Gudrun (*Salix dasyclados*) the lowest. Planting system had less effect on stool death, height development and biomass production. Tora was the only clone producing more biomass when grown in mixtures compared to monoculture. The size hierarchy between individual stools was generally kept during the experimental period in both monocultures and mixtures, even if the height correlations decreased with year. To obtain a more even establishment of willow stands and lower the risk of a size hierarchy developing leading to large stool mortality and gaps in the stand, we recommend the use of larger and more even-sized cuttings.

## 1. Introduction

Establishment is considered the most important element in achieving a long-term yield in willow plantations [1]. A good establishment can be characterized by large shoots with higher survival expectancy and higher expected biomass production capacity [2], a quick coverage of the surface area to reduce competition from weeds, and a low variability in shoot size per unit area, which reduces the risk of developing size hierarchies. Cutting quality and characteristics have a considerable impact on survival and viability of the crop, particularly during the establishment phase. Larger cuttings have a larger carbohydrate pool available for allocation to roots and shoots [3], and a decreased susceptibility to soil desiccation [4]. A number of studies, Verwijst et al. [5,6], Edelfeldt [7] and Edelfeldt et al. [8–10], have shown that cutting size had considerable effect on early performance attributes. Increases in cutting length, diameter and/or weight led to higher survival, larger leaf area, longer shoots, higher number of shoots and higher biomass production. Burgess et al. [11], Rossi [12], Shield et al. [13] and Friedrich et al. [14] have shown similar results.

When growing willow at high densities, the plants are subjected to

an increased competitive pressure that augments self-thinning processes within the stand [15]. Fewer shoots will be produced for each stool, theoretically creating an even canopy. However, stools developing from larger cuttings with an initial competitive advantage may grow larger than other stools, developing into a size hierarchy which is preserved in remaining stumps and belowground roots during consecutive harvests [16,17]. Eventually the competitive pressure in this size hierarchy leads to stool mortality and gaps in the stand [16,18]. The reduced growth in these gaps may be compensated by competing willow plants colonizing the vacant space [19,20]. But if weeds get a foothold, or if the delayed growth results in a lower growth per unit area in the gap, there could still be production losses at harvest.

Growing willow in mixtures may potentially increase the initial advantage of some stools due to genotype differences in growth pattern, creating a faster developing size hierarchy than in monocultures. Clonal differences in early biomass production have been shown in several studies [6,21–26], and could have a large impact on the future plantation. On the other hand, susceptibility to leaf rust (*Melampsora* sp) and other diseases may be less in polyclonal plantations [19,20], as the spread of diseases may be slower when genotypes of varying

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susceptibility are introduced into the mixture [27]. Clone variations could be considerable, especially when comparing yield and resistance abilities (frost, pests, drought, etc.) between older and newer varieties. Mixtures have shown to be beneficial for biomass production in several studies [19,20,28], even if higher variation and stool death was reported in mixture stands. The remaining plants were able to compensate for the loss by occupying the vacant space. However, when clones of similar genotypes were planted, the difference between mixtures and monotypes were very small or non-existent, leading to the conclusion that there need to be sufficient genetic diversity for the mixture to function as a disease control strategy [29]. Mixtures may be beneficial to biomass production for other reasons than reduction of leaf rust or other diseases. Cowger and Weisz [30] noted that increased yields were still recorded in several cultivars even when disease effects had been accounted for.

Although there have been several studies comparing willow mixtures and monocultures [20,29], few have studied the early formation and development of size hierarchies, the effect of cutting qualities on size hierarchies, and the importance and preservation of size hierarchies in different clone mixtures.

The purpose of this study was to assess the relationship between cutting qualities and the hierarchy development and persistence over subsequent harvests. The influence of cutting traits (clone, fresh weight) on performance (survival, shoot development, biomass production) over two subsequent harvests was assessed in a highly competitive environment planted to induce the development of an early size hierarchy in both monoclonal and mixed stands. Our hypotheses are that:

1. Larger cuttings will produce stools with higher maximum shoot height and larger biomass production compared to smaller cuttings.
2. Shoot height development and biomass production will depend on clone.
3. Clones will produce stools with higher maximum shoot height and larger biomass production in two-clone mixtures than in monocultures.
4. Variation in performance will be higher in two clone mixtures than in monocultures.
5. Differences in performance will be augmented in two clone mixtures compared to monocultures, i.e. clones that perform well in monoculture will perform even better in mixtures when planted with less productive clones.
6. Differences in performance caused by effects in 1–5 will result in a size hierarchy development that will keep over successive harvests.

## 2. Materials and methods

### 2.1. Design and measurements

A field experiment was conducted between 2008 and 2015 at Ultuna close to Uppsala, Sweden (59° 48'N, 17°39'E) on a field with heavy clay soil. The field was plowed and harrowed a few days before planting. One-year old shoots (rods) from five *Salix* clones Gudrun (*Salix dasyclados*), Jorr (*Salix viminalis*), Olof (*Salix viminalis* x (*Salix schwerinii* x *Salix viminalis*)), Sven (*Salix viminalis* x (*Salix schwerinii* x *Salix viminalis*)) and Tora (*Salix schwerinii* x *Salix viminalis*) were harvested from a field plantation close to Uppsala on March 16, 2008 and put in the freezer at −6 °C. On June 23, the rods were taken from the freezer and a 5 cm long part of the base of each rod was removed to standardize the effects of drying, molds and other kinds of storage damage. The rods were first cut into eight 24 cm long parts, and then the top six cm were cut from each part, resulting in 18 cm long cuttings. Fresh weight of each cutting was measured.

The cuttings were planted in two replicate blocks situated next to each other. Each block consisted of four rows with four plots in each row. There was a 40-cm space between each plot within the block and

between the two blocks. Each plot (180 cm × 180 cm) consisted of eight rows with eight cuttings in each row, i.e. 64 cuttings plot<sup>−1</sup>, using a 20-cm space between the cuttings within each row and between the rows giving an overall planting density of 197 500 cuttings ha<sup>−1</sup> in the stand. Each plot was randomly assigned to be either a monoculture using one of the five clones, or to be one of the ten possible mixtures of two clones, resulting in 15 plot compositions. In plots with mixtures, cuttings of different clones were placed in a checkered pattern alternating the two different clones. Within plots, cuttings were randomly planted with regard to cutting weight. To create as similar conditions as possible for all plots, two filler plots were also planted using monocultures of Tora and Sven, resulting in a total of 32 plots. The data from these filler plots were not used in the analysis. The cuttings were planted by pushing them manually into the ground until about 5 cm protruded from the ground.

During the first year, the experiment was weeded regularly during the entire growing season. There was no weeding during the following years. Very little rust was observed on the shoots during the experimental period and no apparent wilting due to drought. There was a little grazing at the edges, mostly observed at the western edge. As the grazing damages were very minor, the effects on the willow were assumed to be negligible.

Standing height of the highest shoot (maximum height) of each cutting (stool) was measured after the growing seasons 2008–2010 and 2012–2014. Survival (i.e. cutting or stool death) was determined each year. The experiment was harvested in March 2013 (i.e. after the growing season of 2012), and in the winter of 2014–2015 (i.e. after the growing season of 2014). For simplicity, the harvests in this study will be called harvest 2012 and 2014, respectively. At each harvest, number of shoots and fresh weight including dead shoots were measured for each stool. Stool death, i.e., the occurrence of a dead stool, was noted. Dry weight was determined to be the most appropriate measurement of biomass production. To estimate the dry/fresh weight ratio, 70 shoots or more were randomly chosen from living shoots of each clone and year. From these, 20 cm were cut around the balance point. Fresh weight was measured on the cut pieces and they were put in the oven at 105 °C for 48 h before dry weight measurements.

### 2.2. Models and statistical analyses

All statistical tests were performed at significance level 0.05 in SAS 9.3 [31].

Dry/fresh weight ratios for each clone were estimated using a simple linear regression model with dry weight as dependent variable and fresh weight as explanatory variable for each harvest year and clone using the REG procedure of SAS. The ratios for Tora, Jorr, Olof, Sven and Gudrun were estimated to 0.451, 0.489, 0.472, 0.486 and 0.484, respectively, at harvest 2012, and 0.410, 0.468, 0.432, 0.428 and 0.453, respectively, at harvest 2014.

The effect of clone on initial cutting fresh weight was tested using one-way analysis of variance with the MIXED procedure.

To account for possible influence of edge effects, the stand was divided into edge rows and centre rows, edge rows consisting of the rows closest to the open field around the entire stand. The models were first tested by assigning the four outermost rows as potential separate edge rows. First and second edge showed differences between each other and compared to the centre rows. There were no significant differences between the third and fourth outermost edge rows and the centre rows for any performance attribute in either of the two harvests. Consequently, only the first and second edge rows were used in the models below.

Biomass production, maximum stool height and stool death were analyzed using edge effects (factor with two or three levels), clone (factor with five levels), cutting fresh weight (continuous covariate), planting system (factors with two levels: monoculture and mixture) and plot composition (factor with 15 levels) as explanatory variables. After

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