Growth Performance of Willow Clones in Short Rotation Coppice after Sewage Sludge Application

KATRIN HEINSOO [1*] AND IOANNIS DIMITRIOU [2]

[1] *Institute of Agricultural and Environmental Sciences, Estonian University of Life Sciences, Riia 181, 51014 Tartu, Estonia*

[2] *Department of Crop Production Ecology, Swedish University of Agricultural Sciences (SLU), Ecology Building, P.O. Box 7043, SE 750 07, Uppsala, Sweden*

* *e-mail: katrin.heinsoo@emu.ee; tel.: +37 27 311882; mobile: +37 25295325; fax: +37 27 383013*

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Abstract

The aim of the current study was to compare the growth performance of willow clones from different breeding programmes in short rotation coppice field trials established in different European countries when applied with sewage sludge. In general, willow clones from the European Willow Breeding Programme (clones “Resolution” and “Endeavour”) had lower survival rate, but higher aboveground shoot biomass than willow clones from the Swedish State Willow Selection Programme (clone 78183) and the Willow Breeding Programme of Agrobränsle AB (clones “Tora” and “Inger”). Sludge application did not have any significant impact on growth performance. The average aboveground biomass production of willows in Poland was larger than that in Estonia or Germany, and in the three-year-old plantations it was not correlated to the actual plant density.

Key words: biomass; production; short rotation coppice; sewage sludge; willow

Introduction

The use of bioenergy from agriculture is projected to increase in European countries in the near future and will play an important role to fulfil renewable energy and environmental commitments set by the EU (EEA 2006). Short Rotation Coppice (SRC) with willow (*Salix* sp.) for producing biomass for heat, electricity, or both, is considered as an energy-efficient bioenergy system to reduce greenhouse gas emissions (Styles and Jones 2007), and a promising way to reach European targets for renewable energy. The crop management of willow SRC, such as soil preparation, weed control, planting, fertilisation, more resembles that of agricultural crops than forestry. In order to fulfil the public demand for production of both food products and bioenergy raw materials, the agricultural bioenergy systems should be land-efficient producing maximal energy yield per hectare. Moreover, the production must be more cost-efficient than that of food crops, to convince farmers shifting their production from food crops towards cultivating bioenergy crops. The application of municipal sewage sludge to SRC plantations as nutrient source has been identified as one of the most attractive methods for achieving environmental and energy goals, while simultaneously increasing farmers’ income (Dimitriou and Rosenqvist 2011).

Municipal sewage sludge contains nutrients, mainly phosphorus (P) which is a finite source on earth and needs to be recycled, but also nitrogen (N) which is essential for high growth of agricultural crops. Recycling of sewage sludge in agriculture varies greatly among European countries; in some of them (e.g. Denmark, France, Ireland, Spain, the UK) more than 60% of sludge production was used in agriculture in 2006, but in others (e.g. Finland, Netherlands, Romania, Greece) almost no sewage sludge was recycled in agriculture (Anonymous 2008). The trends of sludge recycling in agriculture has been also varying, with countries as e.g. the UK increasing the application rates during the last decades and others such as the Netherlands who stopped the recycling of sludge to agricultural land due to concerns that contaminants in the sewage sludge will enter the food chain (Anonymous 2008). This variability on how sewage sludge is seen by the different European countries is also depicted in the differences in the legislation concerning the limits of nutrients or heavy metals allowed to be applied on agricultural soils and also heavy metals limits in the supplied sludge (BIOPROS 2008). Certain countries (e.g. Sweden, Denmark, the Netherlands) have much stricter legislation than the rest which in large follow the European Sewage Sludge Directive (86/278/EEC).

Willow SRC as an agricultural crop needs fertiliser to achieve high biomass production. At the same time it is a non-food non-fodder crop and therefore avoiding risks of contaminants entering the food chain if applied with sludge but with ability to take-up heavy metals, especially Cd, in their shoots (Aronsson and Perttu 2001, Dimitriou et al. 2006). The shoots are regularly harvested and consequently heavy metals are removed from the soil after each harvest. Several studies have also indicated that the amounts of heavy metals supplied with sludge are lower than the ones taken out with shoot harvest (Dimitriou et al. 2006, Baum et al. 2009). Therefore, the great potential of applying sewage sludge to SRC in different European countries to increase the profitability of SRC cultivation by decreasing fertilisation costs and increasing biomass production has been expressed (Dimitriou and Rosenqvist 2011). However, to ensure a sustainably sound practice applied in different locations or countries, willows need to respond positively in terms of growth with regard to the local conditions (e.g. climate, soil), plant material (e.g. clones used) and sewage sludge applied (e.g. quality, quantity).

This study examines the growth response of willow SRC plantations established in three European countries (Estonia, Germany and Poland), planted with five different willow clones and applied with two different application rates (doses) of sewage sludge. The adapted experimental design was similar in the different locations. The objectives of this study were to assess the growth response of willow SRC to the different application rates of sewage sludge and to evaluate the suitability of the different willow clones in the different countries to be further used for sewage sludge applications.

Materials and Methods

All three SRC plantations in the current study were specially designed for the current experiment in the premises of local farmers of three different countries. The geographical position and soil types of the selected sites are given in Table 1. All the selected fields have been previously used for annual agricultural crops. Common chemical weed control with glyphosate-based herbicide was performed 1-2 weeks before planting. The local weather conditions in the sites were presumed to be similar to those in the closest meteorological station (Table 2 and Figure 1).

Table 1. Characteristics of the studied SRC locations

Country	Geographical position	Soil type	Closest town for weather data
Estonia	58°29’18’’ N; 26°53’15’’ E	sandy loam (Phaeozem)	Tartu (25 km)
Germany	54°23’51’’ N; 10°23’06’’ E	sandy clay (Cambisol)	Kiel (20 km)
Poland	50°15’38’’ N; 17°52’36’’ E	silt loam (Cambisol)	Zakrzów; Kędzierzyn-Koźle (20 km)

Table 2. Monthly precipitation (mm) near the study sites

year	month	Estonia	Germany	Poland
2006	april	9.4	44.7	34.0
	may	34.4	70.4	54.8
	june	28.6	26.4	64.0
	july	18.8	105.8	4.8
	august	54.6	165.0	151.8
	september	20.4	29.3	17.8
	october	47.0	90.2	26.1
	november	18.2	52.2	59.1
	december	21.4	65.5	23.0
	SUM	252.8	649.5	435.4
2007	january	42.0	133.4	58.2
	february	10.6	62.2	n.a.
	march	14.8	59.3	35.4
	april	23.4	2.5	10.6
	may	72.4	89.8	52.2
	june	28.4	109.7	65.4
	july	72.2	109.9	70.4
	august	30.4	57.5	114.6
	september	31.4	108.7	108.8
	october	65.2	26.3	35.3
	november	23.2	41.1	63.5
	december	8.4	64.5	n.a.
	SUM	422.4	864.9	614.4
2008	january	19.6	66.2	n.a.
	february	32.4	38.1	n.a.
	march	14.4	59.0	39.8
	april	18.2	49.1	68.3
	may	20.4	6.5	60.7
	june	86.6	30.4	50.9
	july	53.8	71.5	82.6
	august	158.2	152.3	101.5
	september	29.6	30.4	83.2
	october	55.0	n.a.	25.9
	SUM	488.2	503.5	512.9

The planting material used in all studied SRC plantations originated from the same nurseries and was provided to farmers in spring 2006. In order to increase the genetic range of planting material five willow clones originating from different breeding programs were used. The selected clones included plant material from the Swedish State Willow Selection Programme (clone 78183 according to the Swedish clone numbering system (*Salix viminalis*)), the Willow Breeding Programme of the Swedish company Agrobränsle AB (clones “Tora” (*Salix schwerinii x S. viminalis*) and “Inger”

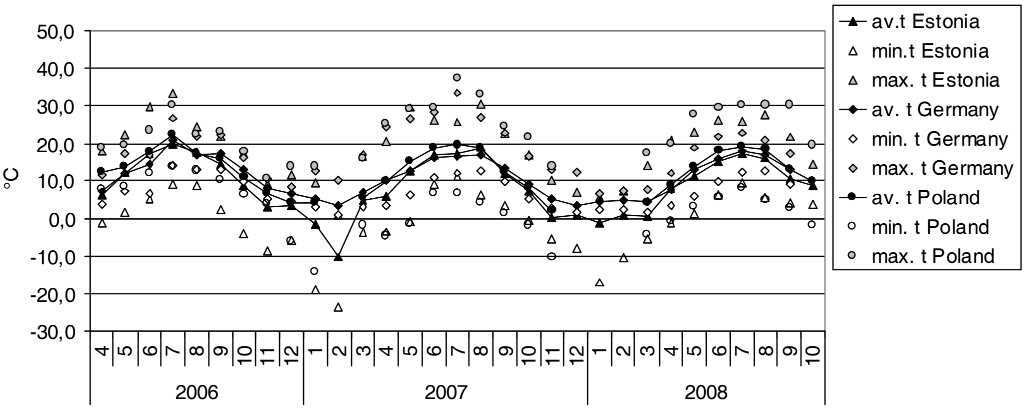

Figure 1. Temperature fluctuation close to studied SRC plantations in different countries during the study period. min. t - detected lowest month temperature; max. t –detected month highest temperature; av. t – monthly average based on data per hour

(*S. triandra x S. viminalis*)) and the European Willow Breeding Programme (clones “Resolution” (3rd generation hybrid of *S. schwerinii x S. viminalis*) and “Endeavour” (*S. schwerinii x S. viminalis*)).

In Germany and Estonia, each of the selected willow clones was manually planted in seven double rows with distance between rows 0.75 + 1.5 m and row length 60 m. The distance between plants in a row was 0.6 m. Due to different shape of available area in Poland each clone was planted in 21 double rows with row length of 20 m. The distance of plants between and within rows in Poland were the same as in Germany and Estonia.

For sludge application each plot was divided into three blocks of 20 m × 15.75 m. One block per plot remained as control area (P0) and the second and third block were applied manually with pre-treated sewage sludge received from local wastewater treatment plants with the approximate load of phosphorus of 60 (P60) and 120 (P120) kg per hectare, respectively. The location of each block inside the plot was chosen for each clone randomly. The same sludge application scheme was used in all studied plantations. However, as the chemical composition of pre-treated sewage sludge in different countries was variable, the amount of applied sludge was different in each country (Table 3). According to local legislations, the load of the critical chemical elements in our field experiments never exceeded the maximum allowed application rate limits for agricultural soils.

The survival of plants was measured each autumn during the study period 2006-2008 by counting the number of plants with green leaves in each double row and for every clone. For aboveground biomass estimations (leaves not included), 20 plants for each clone and sludge treatment were selected in each country during the first study year. For this reason in each plot, and counting from the edge, plants from number 6 to 15 of the third double row from both outward rows were labelled with plastic tags. If the plant was absent or dead, no other plant was selected instead, and therefore the same plants remained under investigation each year. For biomass growth estimations, the diameter of all living shoots of monitored plants at 55 cm from the ground (D55) was measured in autumn of each year if the shoot was taller than 70 cm. Shorter than 70 cm shoots were excluded from the measurements as their contribution to aboveground biomass production was considered low and accuracy of such measurements was poor (according to our previous experience).

For allometric relations in estimating aboveground biomass, destructive sampling of 30 shoots of each studied clone covering the total diameter range was performed in 2008 after the third vegetation period in Estonia. For each clone the parameters of the allometric relations of second order polynom between shoot diameter and its dry weight were calculated separately (adjusted R^2 between 0.76–0.99). The aboveground biomass in each plot was calculated by multiplying the data of average survival of plants of particular plot/

Table 3. Concentrations of chemical elements in the pre-treated sewage sludge of different countries and application rates to P120 plots. Sludge: Sludge supply for 120 kg P load per ha

	Estonia		Germany		Poland	
	chemical analysis (mg/kg)	load to the field if P=120kg (kg/ha)	chemical analysis (mg/kg)	load to the field if P=120kg (kg/ha)	chemical analysis (mg/kg)	load to the field if P=120kg (kg/ha)
N	22700	136	23100	98	30500	276
P	20000	120	28200	120	13250	120
K	3000	18	n.a.	n.a	n.a.	n.a.
Cd	2.5	<0.1	0.9	<0.1	4.7	<0.1
Cr	190.0	1.1	24.0	0.1	26.9	0.2
Cu	190.0	1.1	699.0	3.0	127.0	1.1
Hg	0.7	<0.1	0.3	<0.1	1.3	<0.1
Ni	53.0	0.3	15.0	0.1	27.5	0.2
Pb	51.0	0.3	16.0	0.1	77.5	0.7
Zn	720.0	4.3	478.0	2.0	1124.0	10.2
DM (%)	29.8		23.3		43.9	
Sludge supply (t /ha)		20.1		18.3		20.6

year with the average aboveground biomass of measured willows in the same plot after each vegetation period.

Statistical analyses and correlations were performed with the software package SAS 9.1 (SAS Institute Inc, Cary, NC, USA 2002-2008 version 9.1). Significance of different factors on the regressions between variables was detected with GLM Multiple Linear Regression test (MLR) Type III SS hypothesis. The general impact of different classes on data was detected with ANOVA procedure. To analyse further the differences in survival and aboveground biomass between different countries and clones, the Tukey's Honestly Significant Difference test (HSD) and Ryan-Einot-Gabriel-Welsch Multiple Range test (REGWQ) were used. Differences in survival and plant biomass of various clones in different countries were detected with the Least Square Means test (LSM). The confidence level of all analyses was set at 95%.

Results

The SAS GLM analysis revealed that sewage sludge application had no influence on the survival rate of plants of different willow clones over the study period. Therefore the survival rate data of all plots regardless of the fertilisation level were pooled and used for further statistical analyses on survival. In general, plant survival decreased with plantation age and was clone-specific (Figure 2). After the first vegetation period, three of five clones (78183, Tora and Inger) had similar survival rates which were larger than 88% of planted cuttings. The survival rates of plants from clones originating from the European Willow Breeding Programme were significantly lower (75 and 64-% for Resolution and Endeavour, respectively). After the first wintering the survival of plants of all clones decreased. At the end of the study period, the survival rate of all studied clones except Endeavour was significantly different compared to the planting year. The best performing clone when data for all countries were lumped together was 78183 followed by Tora and Inger.

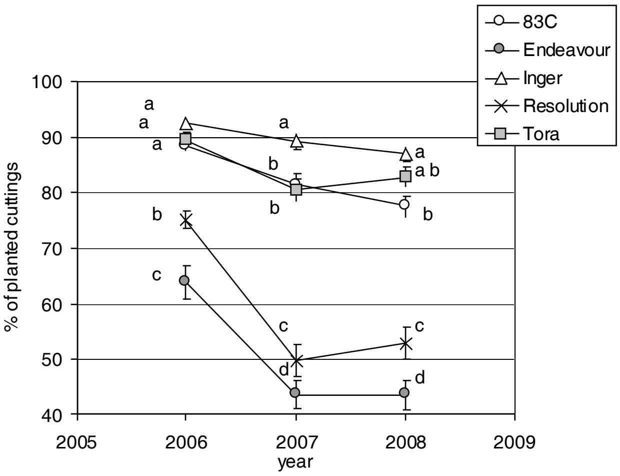

Figure 2. Average survival rate (%) of the different willow clones during study years. Vertical bars indicate standard error of the mean. Statistically significant differences between clones during the same year's dataset are labelled with different letters (n = 63)

The survival rate of different willow clones varied among countries (Table 4). However, the ranking order of different clones based on their survival was almost similar in all countries, with 78183, Tora and Inger having much higher survival rates than the other clones.

Table 4. Survival rate (% of planted cuttings) in the different countries over the different study years. The average values were calculated over years and willow clones. Statistically significant differences between countries are labelled with different letters (n = 21)

year	clone	Estonia		Germany		Poland	
2006	78183	81.4	a	92.7	b	91.6	b
	Endeavour	66.8	a	70.8	a	53.9	b
	Inger	86.6	a	96.8	a	94.0	a
	Resolution	77.4	a	70.2	a	77.8	a
	Tora	81.4	a	90.1	a b	97.3	b
2007	78183	70.4	a	85.3	b	88.9	b
	Endeavour	39.8	a	48.7	a	42.2	a
	Inger	79.8	a	95.5	b	92.4	b
	Resolution	46.3	a	31.8	b	71.1	c
	Tora	75.7	a	68.6	a	97.1	b
2008	78183	70.1	a	75.2	a	87.2	b
	Endeavour	39.6	a	50.1	a	40.8	a
	Inger	78.8	a	91.3	b	90.8	b
	Resolution	45.9	a	43.6	a	69.3	b
	Tora	75.0	a	77.1	a	96.6	b
	AVERAGE	**67.7**	**a**	**79.4**	**b**	**73.2**	**c**

Sludge application did not enhance the average aboveground biomass of studied willow SRC plots (Figure 3). No statistically significant differences were revealed using only annual datasets. Therefore for further aboveground biomass analyses the year factor was not included.

In general, the average aboveground biomass of studied plants varied between clones and at the end of the experiment statistically significant differences were detected (Figure 4). The number of measured plants during the first year of the study was small due to poor plant growth. The largest annual biomass increase of all studied plants of the different clones took place during the third growing season. Also the clon-

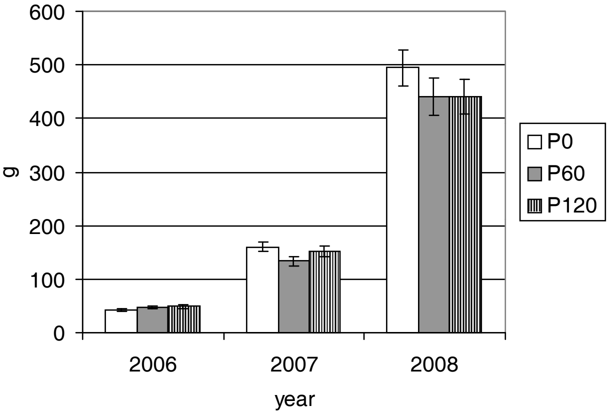

Figure 3. Average willow aboveground biomass (g) in plots with various application rates of sewage sludge during study years. P0 – plots without sludge application; P60 – plots with sludge application that correspond for P load 60 kg ha^{-1}; P120 – plots with sludge application that correspond for P load 120 kg ha^{-1}. Vertical bars indicate standard error of the mean (n = 98-232)

al differences in terms of aboveground biomass were the largest at the end of the third vegetation period. Therefore the biomass dataset of 2008 was used for more detailed analyses. When looking at the average aboveground biomass of individual plants per clone that year, Endeavour from the European Willow Breeding Programme demonstrated the highest yield and was followed by Resolution from the same programme (Figure 4). The three-year-biomass of other clones that were bred under the Swedish State Willow Selection Programme (78183) or by the Swedish company Agrobränsle AB (Tora, Inger) was in average much smaller (Figure 4).

The large variations in aboveground biomass of each particular clone in 2008 can be partly explained by the differences between countries. At the end of this year the average individual plant aboveground biomass in Poland was significantly higher (905.8 g) than in Germany and Estonia (108.2 and 154.0, respectively). Unlike the survival data, the ranking order of studied clones regarding their average aboveground biomass varied between countries and general trends could not be detected (Table 5).

In order to check if the large variability of average aboveground biomass was due to differences in actual plant density in SRC caused by differences in survival, we analysed the relations between average plant survival and average aboveground biomass in 2008. The general regression coefficient was not significant ($R^2 = 0.01$, $P = 0.69$). Also no statistically significant relations were detected when the data were analysed on country basis (Figure 5).

Aboveground biomass per hectare after three years growth period was very low in Estonian SRC

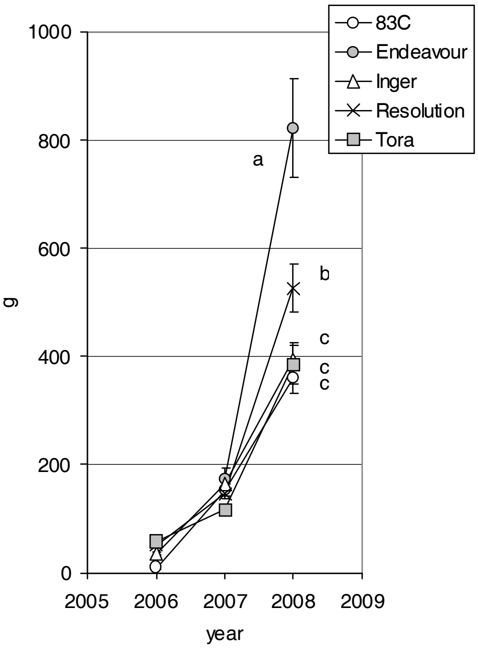

Figure 4. Average aboveground biomass (g) per plant for the different willow clones during the study. Vertical bars indicate standard error of the mean. Statistically significant differences between clones in 2008 dataset are labelled with different letters (for this year n = 77-173 depending on clone)

Table 5 The ranking order of studied willow clones according to their aboveground biomass at the end of third vegetation period in the different countries. Statistically significant differences between clones in the country datasets are labelled with different letters (n = 16-59; 15-58 and 37-59 for Estonia, Germany and Poland, respectively)

Estonia			Germany			Poland		
clone	biomass		clone	biomass		clone	biomass	
Endeavour	240.0	a	78183	242.6	a	Endeavour	1243.8	a
78183	194.2	a b	Resolution	213.5	a	Resolution	944.0	b
Inger	158.4	a b c	Inger	203.2	a b	Tora	833.2	b
Tora	117.0	b c	Endeavour	145.3	a b	Inger	802.9	b
Resolution	109.6	c	Tora	105.9	b	78183	703.4	b

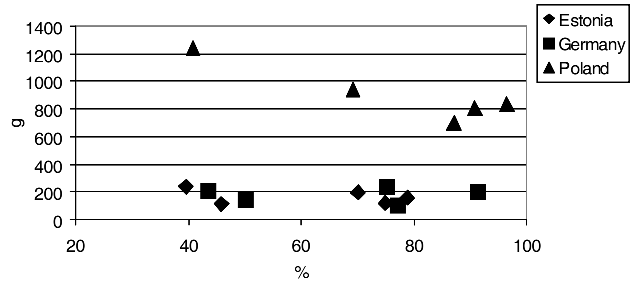

Figure 5. Relations between average survival (%) and average aboveground biomass (g) in studied countries in 2008

plantation (0.7-2.0 t Dry Matter (DM)/ha/yr depending on clone). In Germany it was slightly higher (1.0-2.8 t DM/ha/yr), and the highest biomass was measured in Poland (7.5-11.9 t DM/ha/yr). The detailed analyses confirmed that on average the aboveground biomass yield of willow SRC per area was more depending on plant average biomass than on survival rate (R^2 = 0.79 and 0.22, respectively). However, further analyses of the relations between plant survival and aboveground biomass on country basis did not confirm any significant impact of average aboveground biomass on clone basis and the data of Poland gave a controversial trend (Figure 6).

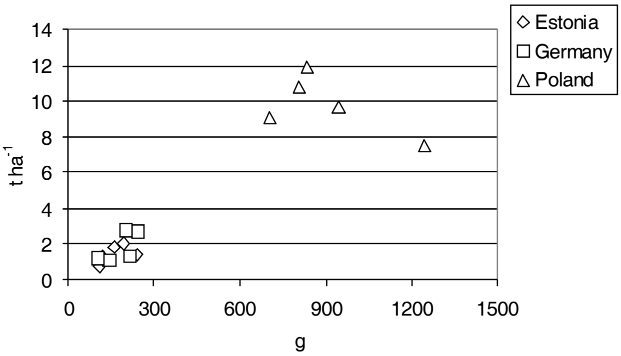

Figure 6. Relation between average aboveground biomass (g) and calculated willow aboveground biomass (t ha^{-1}) in studied countries in 2008

Discussion and conclusions

Sewage sludge application did not enhance willow growth in all locations, and no fertilisation effect was evident at least during the first three years after application while several other studies have reported higher growth of willows when sewage sludge is applied compared to non-fertilised controls (Hasselgren 1998, Labrecque and Teodorescu 2001, Heinsoo and Holm 2013). Although our results come in agreement with Dimitriou et al. (2006) revealing that applied fields with sewage sludge need to be compensated with mineral fertilisation with nitrogen to achieve higher biomass, at least in the short-term. Despite the rather high amounts of nitrogen applied with sewage sludge in our case (98 and 276 kg/ha in Germany and Poland, respectively), most of nitrogen seemed to be unavailable for plants, as it has been already reported (Adegbidi et al. 2003, Cogliastro et al. 2001), and additional fertilisation should be applied to achieve higher yields, at least during the first years after sludge application. Sewage sludge application had also no influence on plants survival, since survival rate changes were not related to the different treatments. This indicates that even without direct impact to the crop yield sewage sludge is an appropriate mean to enrich soil with nutrients and organic matter and has no negative impact to plant response when applied to willow SRC. Moreover, application of sewage sludge to exploited peatland area may promote its suitability for willow growth (Gradeckas 1997) and neutralize the growing substrate in root zone (Gradeckas et al. 1998).

Plant survival decreased with the age of the plantations and was clone-specific. Survival rate of willow SRC has been reported to decrease over time especially during later cutting cycles (from 23% in first cycle down to 72% in third cycle) (Nordh 2005). In our case, and for the plants of clones from the European Willow Breeding Programme, survival rates especially in Estonia and Germany were substantially lower than previously reported ones after three years of growing, indicating that high mortality rates can potentially be a problem. In Poland, the survival rates of plants from clones of European Willow Breeding Programme were higher than in other countries, but still they were significantly lower than the ones for plants of the Swedish State Willow Selection Programme clones. Therefore, in terms of establishment potential, clones from the European Willow Breeding Programme did not seem to be as suitable as the Swedish clones under the climatic conditions prevailing at the locations were our experiment took place.

However, in terms of aboveground biomass growth per individual plant, the opposite result concerning plant survival was observed. The average biomass of Endeavour clone was significantly higher than that of all others studied clones in Estonia and Poland, and average biomass of Resolution clone (also from European Willow Breeding Programme) was second best in Germany and Poland. This means that despite lower survival, in a number of cases the growth potential of plants from the European Willow Breeding Programme was better than that of plants of the other clones. According to our statistical analyses, this result was not depended on their lower density in the experimental plots that occurred due to higher mortality for Resolution and Endeavour, therefore not related to potential lower stress due to less competition between plants. Data analyses on country level checking any potential relation between survival and weight per individual plant did not reveal any clear trends, suggesting that as long as plants manage the site and climatic conditions and get established, plants from the European Willow Breeding Programme are in most cases performing better on individual tree level than plants from the rest of the clones.

In our experiment, this better plant growth resulted in higher yields per area in Estonia and Germany, showing that it was more important than survival rate, but this trend was not followed in Poland, where we measured significantly higher aboveground biomass per plant than in the other countries. Besides the strong influence caused by one outlier in our small-sized dataset, this might also be an indication that the local environmental conditions play a very important role on which response a plant from a certain clone might have in terms of growth. A number of factors related to local site and climatic conditions interfere and affect survival and growth when a crop plantation is introduced, and therefore our results that indicated differences between sites and clones were somewhat expected. Suitability tests on individual level (e.g. small-scale experiments in pot-trials) but also on stand level (e.g. larger-scale multi-annual field experiments) need to be conducted for choosing an appropriate clone when willow SRC is to be addressed in a certain area.

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РОСТ КЛОНОВ ИВЫ В КУЛЬТУРЕ С КОРОТКОЙ РОТАЦИЕЙ ПОСЛЕ УДОБРЕНИЯ С ОСАДКОМ СТОЧНЫХ ВОД

К. Хейнсоо и И. Димитрйоу

Резюме

Целью данной работы было сравнить показатели роста клонов ивы происходящих от различных программ разведения в культуре сортов с короткой ротацией, установленных в различных европейских странах, в условиях удобрения с осадком сточных вод. В общем, ивы из Европейской программы разведения (European Willow Breeding Programme, клоны “Resolution” и “Endeavour”) имели более низкую выживаемость, но более высокую надземную биомассу чем ивы из Шведской государственной программы (Swedish State Willow Selection Programme, клона 78183) или программы по компани Agrobrдnsle AB (клоны “Tora” и “Inger”). Прибавление сточных вод не оказало существенного влияния на рост. Средняя продуктивность надземной биомассы ивы в Польше было больше, чем в Эстонии или Германии, и в трех-летней плантации это не коррелировало с фактической густотой побегов в культуре.

Ключевые слова: биомасса, ива, осадка сточных вод, продуктивность, сорты с короткой ротацией