Biomass production of *Populus nigra* L. clones grown in short rotation coppice systems in three different environments over four rotations

Vojtěch Benetka, Kateřina Novotná, Petra Štochlová

Three clones of black poplar and a hybrid poplar clone, "NE-42", were trialled in a short rotation coppice system. The trial was replicated in three sites located in the Czech Republic with distinctly different soils and climates which were judged to be respectively favorable, unfavorable and marginal for growing black poplar. In the marginal and unfavorable sites the planting density was 2222 trees ha^{-1} and in optimal conditions 7407 trees ha^{-1}. The aim of the trial was: (1) to test the performance of black poplar clones as a possible replacement for commercial hybrid poplars in areas where the genetic integrity of wild black poplar populations is under threat; (2) to test the range of conditions in which it is possible to grow black poplar and its suitability for utilizing marginal agricultural land; and (3) to measure the potential yield of black poplar clones grown in these conditions. During four harvests at three-year intervals, the following parameters were measured: plant mortality, number of shoots, thickness of shoots, the total cross-sectional area (TCA) of all shoots, the dry matter weight of individual plants (DMIP) and the dry matter yields per hectare. The differences observed between "NE-42" and the best black poplar clone decreased as conditions became more favorable. During the fourth harvest at the unfavorable site for growing poplars, the yield of dry biomass was 11.7 t ha^{-1} yr^{-1} for "NE-42" compared to 3.7 t ha^{-1} yr^{-1} for the best black poplar clone. In marginal conditions the yields were 11.8 and 9.9 t ha^{-1} yr^{-1} respectively, and in the favorable conditions there was no statistically significant difference, being 15.9 and 13.2 t ha^{-1} yr^{-1}, respectively. The higher yield of "NE-42" was due to the higher proportion of thicker shoots or, alternatively, lower plant mortality. The observed TCA was highly correlated with DMIP ($r_s = 0.87$) and dry biomass yield ($r_s = 0.48$). It was demonstrated that black poplar can be successfully grown in marginal conditions on land which otherwise would not be especially suitable for agricultural production, and also in areas where the genetic purity of native populations of black poplar is threatened by the spread of commercially grown hybrid poplars.

Keywords: Growing Conditions, Fast-growing Trees, NE-42, Number of Shoots, Stool Mortality, Yield

Introduction

The cultivation of fast-growing trees in short rotation coppice (SRC) systems has increased in recent years mainly because it provides a source of renewable energy. Today such kind of farming covers thousands of hectares in Europe (Venendaal et al. 1997, Langeveld et al. 2012), with the most commonly grown species being willows (*Salix*) and poplars (*Populus* - Herve & Ceulemans 1996, Verwijst 2001, Langeveld et al. 2012). This trend has also been accompanied by extensive plant breeding (Stanton et al. 2010, Karp et al. 2011), but the selection of new cultivars has been based almost entirely on inter-specific crosses. In Europe new poplar hybrids are derived mainly from crosses between *P. nigra* and *P. deltoides*

Bart. ex Marsh. (Bisoffi & Gullberg 1996) or *P. nigra* and *P. maximowiczii* Henry (Stanton et al. 2010). Such hybrids are capable of successfully pollinating and breeding with wild populations of *P. nigra*, and consequently are a threat to the genetic purity of this indigenous species (Cagelli & Lefèvre 1995, Benetka et al. 2002, Smulders et al. 2008). The spread of these hybrids and the contamination of *P. nigra* gene pools has lead in certain places to restrictions being placed on their cultivation. This raises the question if it is possible or not to develop commercially acceptable clones of pure-bred black poplars to grow in place of the undesirable interspecific hybrids. According to several published results, many clones derived from genetically pure species of the

genus *Populus* show yields comparable with the inter-specific hybrid clones. In trials with the species *P. trichocarpa* Torr. & Gray, the pure species had similar yields to the hybrids (Rogers & Stettler 1989). In other trials using *P. nigra* and *P. trichocarpa*, their biomass yields over four successive harvests were remarkably higher than in hybrid clones such as *P. trichocarpa* × *P. deltoides* and *P. deltoides* × *P. trichocarpa* (Dillen et al. 2011).

Species of the genus *Populus* and their hybrids can be regarded as having two main different strategies for biomass formation as for the number of shoots per plant. The first group includes *P. trichocarpa* and *P. trichocarpa* × *P. deltoides*, which eliminate small shoots rapidly to leave just a few large dominant shoots. The second group, comprising genotypes like *P. nigra* and *P. trichocarpa*× *P. balsamifera* L., eliminate small shoots slowly, so maintaining many small shoots per stool (Laureysens et al. 2005, Al Afas et al. 2008). Furthermore, differences in the proportion of lateral shoots contributing to yield in *P. nigra* and hybrid clones have been observed as early as in the second and third harvests, when the proportion of lateral shoots can be significant (Laureysens et al. 2005, Benetka et al. 2007). In situations where there is a lower overall number of shoots it can be expected that these will be thicker. Moreover, after each harvest there is an increase in the number of shoots per plant (Strong 1989, Al Afas et al. 2008, Dillen et al. 2011) and therefore the resource demands on the site will also increase, boosting the intra-specific plant competition. However, this is off-set by the reduction of plant shoots or even plant numbers due to pathogens, shading or increased competition for resources (the self-thinning effect), which then opens up the area to be better utilized by the remaining plants (Dillen et al. 2011). The number of plants per unit area after the

first and second rotations can be significantly reduced by plant mortality (Ceulemans & Deraedt 1999, Laureysens et al. 2005). Large differences in mortality among the studied genotypes have been observed in experiments conducted over several rotations (Al Afas et al. 2008, Dillen et al. 2011).

SRC systems are a long-term culture which may last for up to 25 years (Dillen et al. 2011). The expression of the various yield traits, which together contribute to the final yield of biomass, is dependent on the interaction of any given genotype and its environment. It is therefore important to evaluate the performance of genotypes in the different conditions in which they will be grown. The main environmental factors to consider are levels of available light, water and soil fertility.

P. nigra is the principal pioneer tree species that colonizes river floodplains, wastelands and other exposed sites, where the moist, sandy soil exposed after seasonal flooding provides an ideal seedbed (Dickmann & Kuzovkina 2008). Based on an analysis of ecological requirements of *P. nigra*, optimal conditions for its growth in the Czech Republic would be provided by sites with an average annual temperature of 7.5 to 9.0 °C, a temperature in the growing season (April to September) above 14 °C, with hydromorphic or semi-hydromorphic soils and a water table at a depth of 0.5 to 1.2 m, or at a depth of only 1.0 m where dense vegetation prevents the full penetration of rain water. Heavy clay soils are unsuitable. Colder locations are less favorable, and those with an annual average temperature below 6 °C and temperatures during the growing season below 12 °C are unsuitable (Mottl & Dušek 1991). However in Italy, for example, optimal conditions for hybrid poplar cultivation are a mean annual temperature of 8.5 to 17 °C and a summer precipitation of 100-150 mm (Di Matteo et al. 2012).

Fast-growing trees in SRC systems are predominantly grown in areas of intensive agriculture. This lead to a competition between the production of food and other agricultural commodities on the one hand, and the production of biomass from fast-growing trees on the other. Trees can also be grown in conditions which are marginal for conventional agricultural activities, but perhaps the real question here is whether they can still achieve satisfactory yields in such conditions.

The aim of this study was to appraise the individual yield parameters contributing to the total yield of biomass, and compare the biomass yields of several clones of black poplar with that of the hybrid clone "NE-42", when grown in three distinctly different environments for a total of four rotations. Further, to determine the environments in which black poplar can be successfully established and grown.

Materials and methods

Locality, soil and climatic conditions

The experiment was established in three sites in the Czech Republic, each with different soil and climatic conditions (Tab. 1).

The first site (Smilkov - SM) represents natural conditions which can be regarded as marginal but typical for the main areas where SRC systems are used. The second site (Bystrice - BY) is located in unfavorable soil and climate conditions. After a four year delay, the third trial (Rosice - RO) was established in optimal soil and climate conditions typical for a flood plain ecosystem and therefore most favorable. Compared to the other two localities, trees were established here using more dense plantings.

Materials

Four clones of *P. nigra* (designated 107, 210, 301 and 302), of the total of 29 included in a wider trial, can be found growing in all three trial localities. These clones are the offspring of individuals found in local subpopulations (Benetka et al. 2007). The first three clones of *P. nigra* (107, 210, 301) have already been shown to be healthy and possessing good yield potential, and have been recommended for cultivation in the Czech Republic (Benetka et al. 2011b). Clone 302, on the other hand, is regarded as being one of the least productive. Clone "NE-42", an interspecific hybrid of *P. maximowiczii* × *P. trichocarpa*, was used as a control.

Organization and treatments

Densities of 2222 plants ha^{-1}, with a spacing of 3 × 1.5 m, were employed in the less favorable (SM) and the unfavorable (BY) localities; in the RO site (favorable conditions) the planting density was increased to 7407 plants ha^{-1}, with a tighter spacing of 2 × 0.67 m. Plant densities were chosen with regard to the soil and climatic characteristics of these three localities (Tab. 1). Rainfall during the growing season (months IV-IX) was only measured at the SM site. In the localities BY and RO a crucial role was played by groundwater.

The experiment was established in 1998 at the SM and BY sites, while the RO site was planted in 2002. A randomized block design with four replications was used, with four plants of each clone per replication in the BY locality and with five plants per replication in the SM site. In the RO locality the experiment had five replications with six plants per replication. Bordering rows were planted around each experimental plot. The plots were also fenced to prevent browsing by wild animals.

The planting material was obtained by cultivating 0.2 m-long hardwood cuttings, 5 to 6 mm in thickness; the resulting one-year old saplings were planted out manually, with those planted at the SM site being watered in. Using one-year old saplings, the planting densities at the beginning of the experiment were the same for each investigated clone at any given site. All plants were clipped to a height of 0.4 m after planting out. The SM

Tab. 1 - Description of the study localities.

Characteristics	Year	BY	SM	RO
Latitude (N)		49° 21'	49° 36'	50° 03'
Longitude (E)		12° 48'	14° 36'	15° 42'
Altitude (m a.s.l.)		551	515	219
Climatic region		moderately cold and moist	moderately warm and moist	warm and moderately dry
Mean temperature > 10°C (num. of days)		120-140	140-160	160-170
Mean ann. temperature [°C]		5.7	6.8	8.5
Soil type		Histosol	Cambisol	Alluvial soils
pH/CaCl$_2$		4.95	6.75	6.68
Plot status before the experiment		lay fallow	farmed up	lay fallow
Influenced by neighboring river		yes	no	yes
Sum of rainfall	1999	-	352	-
(Apr-Sep [mm])	2000	-	393	-
	2001	-	501	-
	2002	-	548	-
	2003	-	265	-
	2004	-	369	-
	2005	-	415	-
	2006	-	635	-
	2007	-	355	-
	2008	-	284	-
	2009	-	401	-
	2010	-	506	-
	2011	-	346	-

site was also always fertilized after harvest, with an NPK (1:1:1) growth fertilizer at a rate of 150 kg ha^{-1}, and a herbicide applied in the spring following each harvest. Twice each summer the weeds were cut and the soil disced between the rows. At the BY and RO sites discing was omitted.

The first harvest at the BY and SM sites was carried out in February 2001, the second harvest was in March 2005, the third in March 2008 and the fourth in March 2011. At the RO site the first harvest was carried out in February 2005, the second in February 2007, the third in March 2010 and the fourth in January 2013. Plants were pruned by hand to a height of 0.1 m above the ground for the first harvest, but in subsequent harvests they were pruned to 0.2 m above the ground.

Measurement

At the end of each growing season before harvesting, growth was measured by taking the diameter of each main stem at a height of 0.5 m from the ground. Before the second, third and fourth harvests, all shoots thicker than 0.01 m were counted and their diameters measured using a digital caliper. Shoots were classified on the basis of their diameter as being "weak" (10-43 mm) or "strong" (44-95 mm). The shoot with the highest diameter was considered as a main stem. Before the fourth harvest the combined total cross-sectional area of all measured shoots (TCA) was calculated and expressed as the sum total area in mm^2 per plant. The TCA per 100 m^2, which also indirectly took account of plant mortality, was expressed in m^2.

During harvest the fresh weight biomass produced jointly from all the plants of a given clone in each replicate was weighed. To measure dry weights at the same time, a representative sample of the harvested material, including the main and lateral shoots, was taken from each replicate and weighed separately. The size of these samples ranged from 400 to 1200 g, depending on the thickness of the shoots. The samples were dried at 105 °C until their weights stabilized. Total dry matter yields per unit area were calculated from the weight of harvested fresh biomass obtained from a given replicate multiplied by the appropriate value for the per cent dry matter and calculated for a given unit area (ha) per year. Yield is a function of the number of plants actually harvested and takes the plant mortality into account. The average dry matter weights of individual plant (DMIP) were expressed in kg yr^{-1}.

Statistical analysis

The analysis of variance (ANOVA, MANOVA) or its non-parametric homologous tests was carried out using the Statistica 8.0 statistical software package (StatSoft Inc., Tulsa, OK, USA). Clones, replicates, localities and rotations were used as independent

variables, and parameters such as total dry matter yields per unit area, DMIP, number of shoots per plant, TCA, and diameters of the strongest shoots were used as dependent variables. When a statistical difference was found, a *post-hoc* comparison was carried out using Duncan's test or multiple comparisons of mean ranks. Significant differences between individual rotations or localities were established on the basis of *t*-tests. Spearman's coefficient of rank correlation was calculated between TCA and the parameters DMIP and total biomass production. All the results are presented using a significance level of α = 0.05.

Results

Since some of the results based on the first two harvests have already been published elsewhere (Benetka et al. 2007), the results presented here are mainly those derived from the third and fourth harvests. Significantly different performances of the clones in each of the three different localities was confirmed during the fourth harvest for each of the variables considered, with the exception of the dry matter yields per unit area.

Stool mortality

The number of shoots and plants per unit area fell during the course of the four rotations, and significantly differed among sites and clones (Tab. 2).

At the locality with marginally suitable soils and climatic conditions (SM), and with less dense vegetation, mortality of clones was low (0-5%), and such losses were the result of mechanical damage incurred during the year of planting. Only for clone 302, which has been shown to be unsuitable for growing in SRC, losses were high, reaching 20%. There were no significant differences between "NE-42" and the black poplar clones.

In locality BY mortality of clone "NE-42" was lower (6%), though not statistically significant. Before the fourth harvest mortality in the black poplar clones ranged from 19 to 50%, although in clone 302 it was 94%. The high losses were the result of unsuitable soils and climate.

In the locality RO (with dense vegetation), losses were observed for all clones, and by the time of the fourth harvest, mortality in the black poplar clones ranged from 40 to 90%. In "NE-42" the mortality was 36%. An increase in mortality was always observed after harvest, and was particularly significant after the third harvest (Tab. 3). At this time, the surviving plants completely shaded the ground. The dead tree stumps were also infected by *Cylindrocarpon, Fusarium* and *Verticillium* fungi (Pešková & Soukup, personal communication), which in certain circumstances can act as facultative parasites, invading weakened plants.

Tab. 2 - Percentage of surviving plants in each site before harvests.

Locality	BY				SM				RO			
Harvest	I	II	III	IV	I	II	III	IV	I	II	III	IV
Rotation length [yrs]	3	4	3	3	3	4	3	3	3	2	3	3
Clone	**%**				**%**				**%**			
107	100	58	50	50	95	95	95	95	93	80	80	60
210	100	100	88	81	100	100	100	100	97	93	60	23
301	83	83	75	67	95	95	95	95	97	93	77	10
302	56	44	13	6	92	88	80	80	83	60	50	29
NE-42	100	100	94	94	100	100	100	100	97	90	87	63

Tab. 3 - A comparison of successive harvests in clones of black poplar and "NE-42" in each locality (*t*-tests). (*): significant differences (p<0.05).

Site	Clone	DMIP			dry matter yield			stool mortality		
		I & II	II & III	III & IV	I & II	II & III	III & IV	I & II	II & III	III & IV
BY	107	0.225	0.231	0.562	0.360	0.270	0.607	0.132	0.815	1.000
	210	0.037*	0.036*	0.307	0.037*	0.075	0.142	1.000	1.000	1.000
	301	0.005*	0.106	0.560	0.008*	0.065	0.247	1.000	0.725	0.725
	NE-42	0.000*	0.870	0.400	0.000*	0.796	0.487	1.000	0.356	1.000
SM	107	0.000*	0.029*	0.492	0.000*	0.029*	0.863	1.000	1.000	1.000
	210	0.000*	0.310	0.947	0.000*	0.310	0.947	1.000	1.000	1.000
	301	0.002*	0.249	0.649	0.005*	0.348	0.732	1.000	1.000	1.000
	NE-42	0.000*	0.112	0.431	0.000*	0.112	0.431	1.000	1.000	1.000
RO	107	0.007*	0.014*	0.203	0.119	0.010*	0.937	0.111	1.000	0.094
	210	0.003*	0.056	0.077	0.002*	0.254	0.042*	0.667	0.024*	0.023*
	301	0.293	0.288	0.401	0.204	0.773	0.054	0.545	0.161	0.001*
	NE-42	0.007*	0.385	0.529	0.007*	0.418	0.825	0.242	0.545	0.011*

Tab. 4 - Four yield traits and differences among clones before the fourth harvest. (N): number of observations.

Parameter	Clone	BY		SM		RO	
		N	mean ± SE	N	mean ± SE	N	mean ± SE
Number of shoots	107	5	13.4 ± 2.6 [b]	18	28.4 ± 1.6 [b]	18	6.6 ± 0.8 [a]
	210	13	12.5 ± 2.1 [b]	20	36.5 ± 2.0 [a]	7	8.6 ± 0.8 [a]
	301	8	22.4 ± 2.3 [a]	19	40.5 ± 1.6 [a]	3	6.7 ± 3.8 [a]
	NE-42	15	10.2 ± 1.2 [b]	20	21.9 ± 1.1 [c]	19	6.0 ± 1.4 [a]
Diameter of the main shoot [mm]	107	5	49.2 ± 3.4 [b]	18	57.8 ± 1.6 [b]	18	61.2 ± 4.7 [a]
	210	13	39.0 ± 2.8 [c]	20	44.1 ± 1.6 [c]	7	53.3 ± 3.9 [a]
	301	8	37.0 ± 2.7 [c]	19	47.1 ± 1.6 [c]	3	52.7 ± 11.7 [a]
	NE-42	15	74.3 ± 3.1 [a]	20	65.0 ± 1.9 [a]	19	53.7 ± 4.3 [a]
Total cross-sectional area of all shoots per plant [mm²]	107	5	8214 ± 1659 [ab]	18	15478 ± 4479 [a]	18	7042 ± 1074 [a]
	210	13	5466 ± 826 [b]	18	14063 ± 3872 [a]	7	6717 ± 1237 [a]
	301	8	7598 ± 1021 [ab]	19	14371 ± 3381 [a]	3	5449 ± 3280 [a]
	NE-42	15	15677 ± 1973 [a]	20	15974 ± 3942 [a]	19	5878 ± 964 [a]
Total cross-sectional area of all shoots per 100 m² [m²]	107	5	0.16 ± 0.05 [b]	18	0.33 ± 0.02 [a]	18	0.34 ± 0.06 [a]
	210	13	0.10 ± 0.02 [b]	18	0.31 ± 0.02 [a]	7	0.17 ± 0.05 [a]
	301	8	0.12 ± 0.02 [b]	19	0.30 ± 0.02 [a]	3	0.07 ± 0.04 [a]
	NE-42	15	0.33 ± 0.04 [a]	20	0.35 ± 0.02 [a]	19	0.29 ± 0.05 [a]
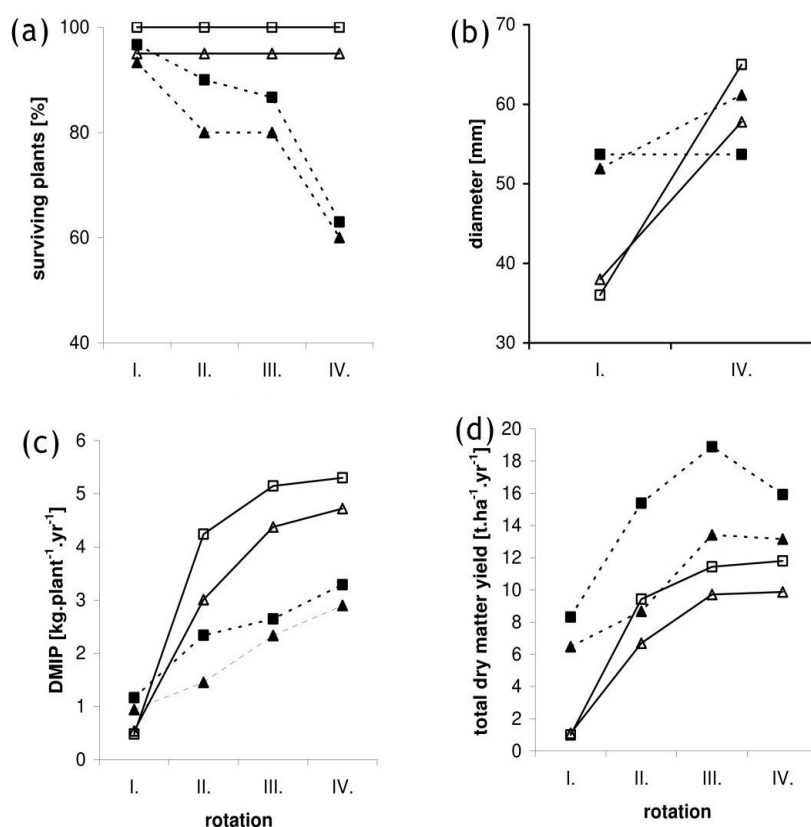
Fig. 1 - Time course of stool survival rate (a), diameter of the strongest shoot (b), dry matter weight of individual plants (c) and total dry matter yield per unit area (d) over the four rotations of the coppiced the best *P. nigra* clone 107 (triangle) and "NE-42" (square) in SM (continuous line) and RO site (dotted line).

Tab. 5 - Mortality of shoots during the forth rotation with respect to the dead plants.

Clone	Number of shoots per plant						Shoot mortality [%]		
	after the III harvest			before the IV harvest					
	BY	SM	RO	BY	SM	RO	BY	SM	RO
107	18.7	51.9	8.3	11.2	28.4	5.4	40.2	45.2	35.0
210	16.3	44.8	9.3	12.5	36.5	4.3	23.7	18.5	53.8
301	27.4	59.3	7.8	22.4	40.5	1.1	18.3	31.7	85.7
NE-42	16.1	28.5	6.6	10.2	21.9	4.6	36.8	23.3	30.9

Clone 302 was dropped from subsequent assessments due to the unacceptably high mortality observed in all sites.

Number of shoots

The number of shoots per plant was always higher in the black poplar clones compared to "NE-42" before harvest in each locality but only statistically significantly higher in the SM locality, before the second, third and fourth harvests. In locality BY, with less suitable conditions, the average number of shoots was significantly lower than in SM. In the locality RO the number of shoots before the second harvest was low and increased very slowly in following harvests (Tab. 4).

The number of shoots developed after the third harvest in locality SM was roughly double compared to the BY site (Tab. 5). In the interval between the third and fourth harvests the number of shoots in SM and BY was reduced by a third, and in RO by half, although in locality BY the temperature and soil conditions were less favorable than RO with similar soil-water availability. As a result of the higher planting density the light conditions were worse in locality RO, which probably had an influence on the production of a lower number of shoots and their higher mortality.

The results show that the plants are able to utilize a larger area for the formation of a higher number of shoots, especially in *P. nigra*.

Shoot diameter

Leading shoot diameter before the first harvest correlated with the quality of natural conditions in *P. nigra* (Fig. 1b). During the fourth harvest in locality BY with sufficient ground water but unfavorable soil and temperature conditions, the leading shoot diameter was significantly greater in "NE-42"

compared to the black poplar clones. Also, in the site SM with less available ground water and average rainfall but with higher levels of nutrients (see Tab. 1), the differences between "NE-42" and the black poplar clones were significant. In locality RO with accessible ground water, higher temperatures and fertile soils, the leading shoot diameter in "NE-42" was the same as the black poplar clones (Tab. 4).

The average shoot diameter was 30.4 mm in RO, where the number of shoots was lower, 25.9 mm in BY and 21.4 mm in SM, where the number of shoots was higher.

The proportion of stronger shoots in any given locality was always lower in black poplar (based on average values for the three clones) compared to "NE-42" (Tab. 6), although in the most favorable soils and temperatures the differences were reduced. This shoot diameter difference is also related to the yield parameters.

Total cross-sectional area of shoots (TCA)

The TCA of all shoots was measured before the fourth harvest (Tab. 4). In locality BY there was a significantly higher average TCA in "NE-42" (15.677 mm^2) than in the black poplar clone 210 (5.466 mm^2), while in SM the average TCA values in all clones were similar, ranging from 14.063 to 15.974 mm^2. In RO the best black poplar clone, 107, had an average TCA value of 7.042 mm^2 and "NE-42" had an average TCA of 5.878 mm^2. Just as with the leading shoot diameter, in unfavorable conditions of soil and temperature in the presence of adequate water, "NE-42" performed better than black poplar regarding TCA. Using Spearman's coefficient of rank correlation the very strong positive correlation of TCA and DMIP ($r_s = 0.871$, $p < 0.001$) and moderate positive correlation of TCA and dry matter yield ($r_s = 0.475$, $p = 0.001$) was confirmed.

Dry matter weight per individual plant (DMIP)

In the locality RO, the DMIP increased almost linearly among harvests (Fig. 1c) and the increment between first and second harvest was significant in almost all clones (Tab. 3). No difference in DMIP was found among all clones in any harvest (Tab. 7). When comparing localities SM and BY after the first harvest, non-significantly higher average values for DMIP were found in all clones at the locality SM with exception of "NE-42". During the second and subsequent harvests, significantly higher average values were found in the locality SM compared to BY in all black poplar clones. By the fourth harvest, the DMIP of black polar clones in SM was twofold higher than in BY. However, the DMIP of the best black poplar clone in each locality was only significantly

lower than "NE-42" for the second and fourth harvests at the BY site.

Biomass production

"NE-42" gave the highest biomass yields in each of the four harvests in each of the three sites (Tab. 8). During the fourth harvest the difference in biomass produced by "NE-42" and the best black poplar clone was signifi-

cant in sites BY and SM. However, in locality RO the yield from clone 107 during the fourth harvest was not significantly different, being 13.2 t ha^{-1} yr^{-1} compared to 15.9 t ha^{-1} yr^{-1}. In the locality BY the black poplar yields were low and showed no significant differences in growth from the the second to the fourth harvest (Tab. 3). In the SM site yields from the second harvest onwards were

Tab. 6 - Percentage of weak (diameter 10-43 mm) and strong shoots (44-95 mm) per clone in each locality. ($\varnothing$): diameter [mm].

Locality	Clone	Percentage of shoots	
		$\varnothing$ 10-43 mm	$\varnothing$ 44-95 mm
BY	clones of black poplar	95.3	4.7
	NE-42	61.5	38.5
SM	clones of black poplar	89.6	10.4
	NE-42	82.4	17.6
RO	clones of black poplar	80.8	19.2
	NE-42	76.3	23.7

Tab. 7 - Dry matter weight of individual plants [kg yr^{-1}] for each harvest.

Rotation	clone	BY		SM		RO	
		N	mean ± SE	N	mean ± SE	N	mean ± SE
I rotation	107	2	0.14 ± 0.00 [a]	4	0.54 ± 0.10 [a]	5	0.94 ± 0.12 [a]
	210	4	0.34 ± 0.08 [a]	4	0.57 ± 0.07 [a]	5	1.05 ± 0.10 [a]
	301	3	0.19 ± 0.02 [a]	4	0.43 ± 0.04 [a]	1	0.69 [a]
	NE-42	4	0.63 ± 0.22 [a]	4	0.48 ± 0.09 [a]	5	1.16 ± 0.09 [a]
II rotation	107	2	0.99 ± 0.49 [b]	4	3.01 ± 0.34 [a]	5	1.45 ± 0.08 [a]
	210	4	1.00 ± 0.23 [b]	4	3.29 ± 0.15 [a]	5	1.61 ± 0.09 [a]
	301	3	1.54 ± 0.24 [b]	4	3.57 ± 0.62 [a]	5	1.44 ± 0.25 [a]
	NE-42	4	4.41 ± 0.40 [a]	4	4.24 ± 0.45 [a]	5	2.34 ± 0.31 [a]
III rotation	107	1	3.22 [a]	4	4.37 ± 0.33 [ab]	5	2.33 ± 0.27 [a]
	210	4	1.81 ± 0.19 [a]	4	3.59 ± 0.23 [b]	4	1.98 ± 0.15 [a]
	301	3	2.61 ± 0.45 [a]	4	4.50 ± 0.38 [ab]	5	1.90 ± 0.31 [a]
	NE-42	4	4.23 ± 0.99 [a]	4	5.15 ± 0.17 [a]	5	2.64 ± 0.63 [a]
IV rotation	107	2	2.24 ± 0.69 [b]	4	4.72 ± 0.33 [ab]	5	2.90 ± 0.31 [a]
	210	4	1.45 ± 0.26 [b]	4	3.62 ± 0.32 [c]	3	2.59 ± 0.25 [a]
	301	3	2.30 ± 0.19 [b]	4	4.31 ± 0.10 [bc]	3	2.56 ± 0.93 [a]
	NE-42	4	5.52 ± 1.02 [a]	4	5.30 ± 0.05 [a]	5	3.29 ± 0.70 [a]

Tab. 8 - Total dry matter yields per unit area [t ha^{-1} yr^{-1}] in black poplar clones and in the hybrid clone "NE-42"; expressed with respect to the percentage of surviving plants.

Rotation	Clone	BY		SM		RO	
		N	mean ± SE	N	mean ± SE	N	mean ± SE
I rotation	107	2	0.3 ± 0.0 [a]	4	1.1 ± 0.1 [a]	5	6.5 ± 0.8 [ab]
	210	4	0.8 ± 0.2 [a]	4	1.3 ± 0.2 [a]	5	7.5 ± 0.7 [ab]
	301	3	0.3 ± 0.1 [a]	4	0.9 ± 0.1 [a]	1	4.3 [b]
	NE-42	4	1.4 ± 0.5 [a]	4	1.0 ± 0.2 [a]	5	8.3 ± 0.7 [a]
II rotation	107	2	1.9 ± 1.4 [b]	4	6.7 ± 0.8 [a]	5	8.7 ± 1.0 [a]
	210	4	2.2 ± 0.5 [b]	4	7.3 ± 0.3 [a]	5	10.9 ± 0.4 [a]
	301	3	2.7 ± 0.5 [b]	4	7.6 ± 1.6 [a]	5	9.8 ± 1.5 [a]
	NE-42	4	9.8 ± 0.9 [a]	4	9.4 ± 1.0 [a]	5	15.4 ± 1.8 [a]
III rotation	107	1	7.2 [a]	4	9.7 ± 0.7 [ab]	5	13.4 ± 1.0 [a]
	210	4	3.8 ± 0.5 [a]	4	8.0 ± 0.5 [b]	4	9.6 ± 1.1 [a]
	301	3	4.1 ± 0.2 [a]	4	9.5 ± 1.0 [ab]	5	10.5 ± 1.7 [a]
	NE-42	4	9.1 ± 2.4 [a]	4	11.4 ± 0.4 [a]	5	18.9 ± 3.7 [a]
IV rotation	107	2	3.7 ± 2.8 [b]	4	9.9 ± 0.5 [b]	5	13.2 ± 3.0 [ab]
	210	4	2.5 ± 0.5 [b]	4	8.0 ± 0.7 [c]	4	5.4 ± 1.2 [ab]
	301	3	3.3 ± 0.5 [b]	4	9.1 ± 0.6 [bc]	2	3.2 ± 1.2 [b]
	NE-42	4	11.7 ± 2.6 [a]	4	11.8 ± 0.1 [a]	5	15.9 ± 4.1 [a]

significantly greater and gradually increased over time, even though the differences were not statistically significant. Only in clone 107 was the yield significantly increased from the second to the third harvest in SM and RO (Tab. 3). In this clone, as in "NE-42", the strong shoots made up a significant proportion of the TCA. In "NE-42" after the second harvest biomass did not significantly increase. During the first two harvests in RO the yields of all clones were much higher than in the other two locations, which can be explained by the higher initial planting densities and, during the first harvest, also the higher individual plant weights. In subsequent harvests the yields increased less rapidly and during the fourth harvest in all clones yields actually fell (Fig. 1d), due to high mortality.

Discussion

The interaction of environmental conditions on yield traits and biomass produced by black poplar compared to the hybrid clone "NE-42" was evaluated at three localities. Differing natural conditions in these sites allowed the study of a wide range of reactions of the individual genotypes. Two localities represented marginal areas in the distribution of black poplar, where the planting of allochthonous species is usually not allowed (*e.g.*, national parks). In these situations it is anticipated that only pure clones of black poplar will be grown in SRC systems. The third locality represents flood plain areas which are suitable for growing black poplar because it is tolerant of long-term flooding (Mottl 1989).

The hybrid "NE-42" showed higher yield than the most productive black poplar clone in all three environments. It must be said, however, that this hybrid clone is the result of a selection process involving several thousand seedlings derived from planned crossings (Stout & Schreiner 1933), while the black poplar clones were chosen from a group of 200 individuals collected from wild populations (Benetka et al. 2007). However, several new clones of black poplar now exist which are the result of deliberate crossings, and in early trials have given higher yields than "NE-42" (Benetka et al. 2012), so black poplar clones with satisfactory yield potential are expected to be available soon.

In the locality with a low average temperature and low pH, and nutrient-poor peaty soils (BY), the black poplar clones had low yields, while "NE-42" achieved yields comparable to those in locality SM with its medium quality soils and higher average annual temperatures. In these more favorable conditions the yields of the best black poplar clones approached those of "NE-42". In the most favorable conditions (locality RO), there was no significant difference between the yields of the best black poplar clones and

the hybrid. This means that black poplar did not perform well only in the very unfavorable conditions regarding soils and temperatures, while "NE-42" demonstrated greater adaptability and performed well even under unfavorable conditions. It would appear that here the character of one of the parent species (*Populus trichocarpa*) is being expressed (Smith 1957).

Total yields per unit area are determined by the number of plants per unit area and the weight of individual plants. The initial numbers of plants falls over time due to mortality, although observed mortality was lower than in stands with higher initial plant densities (Laureysens et al. 2005). The effect of varying initial plant densities on subsequent mortality was observed in our experiment also.

The weight of individual plants is positively correlated with various component parameters such as height, thickness and the number of shoots (Pontailler et al. 1997, Rae et al. 2004, Benetka et al. 2007). As has been shown, height is correlated with thickness of the main stem (Nelson et al. 1981), and so only one of these parameters needs to be measured in order to estimate yields. Differences in the thickness of stems between "NE-42" and the black poplar clones was less pronounced in the more favorable growing conditions. The number of shoots increased during the four harvests in this study, although the number of shoots regenerating from the cut stumps also depends on the amount of light, which is in turn affected by plant density (Atwood et al. 2008). Competition between shoots for light during canopy closure is believed to affect stool survival (Dillen et al. 2011). The poplar genotypes differ with regard to their light-use efficiency and so also in the extent to which their photosynthesis is optimized in dense canopies (Green et al. 2001).

Even though the weight of individual plants was great in locality SM, the amount of biomass yield per hectare was not corresponding with it. The maximum yield of biomass was probably limited here by the overall low density of plants, as reported by Enquist et al. (2009) and Weiner & Freckleton (2010).

In comparing the yield potential of the hybrid "NE-42" and the best black poplar clones used in these trials, it can be supposed that black poplar can be successfully grown even in marginal areas for the natural occurrence of black poplar, that is, in areas with low average temperatures and less fertile soils, but not however in areas with low temperatures and peaty soils (Mottl & Dušek 1991), where the hybrid clone "NE-42" is more successful.

The TCA measured just before the fourth harvest was highly correlated with DMIP and also correlated with the yield of biomass

on individual plots. As early as the second harvest a correlation was confirmed between the number of shoots and also the thickness of the main shoot and biomass yields (Benetka et al. 2007). The correlation between the height of plants, thickness of shoots and other parameters and biomass yields is well-known from many studies and has been used for the non-destructive estimation of total biomass (Zianis et al. 2005, Felix et al. 2008). These correlations could be used to estimate DMIP and biomass yields during the selection process in breeding programs, when the first selection is made on the basis of visual assessments. However, breeding programs must also take into account resistance to important diseases, principally the leaf rust *Melampsora larici-populina*, which can significantly lower final yields (Benetka et al. 2011a) with its damaging effects on plant growth (Nelson et al. 1981), affecting the amount of light penetrating the canopy and so the competition among plants.

It can be concluded that in the selection of new black poplar clones it is important to consider the genotypes which produce thick, strong shoots rather than large numbers of weaker shoots, even though black poplar yields tend to be based on the production of large numbers of weaker shoots (Laureysens et al. 2005). The possibility of successfully making such a selection in black poplar is confirmed by the marked genetic diversity evident in the number and strength of its shoots (Benetka et al. 2012).

Conclusions

The three best black poplar clones planted in three different sites had in all four harvests a lower yield than the hybrid clone "NE-42" which was used as a standard for comparison. These differences were less marked in the locality with better natural growing conditions and were not significantly different at the site with the most favorable growing conditions. The higher yield of the hybrid clone is evidently related to the fact that it has been produced by selecting from the progeny of an interspecific cross, in which the aim was to take advantage of the phenomenon of hybrid vigor. The black poplar clones, however, were derived by taking material from a wild population and were not the product of any kind of selective breeding program. These clones showed less adaptability to unfavorable temperature and soil conditions compared to "NE-42", whose greater adaptability can be attributed to the characteristics of one of its parent species, *P. trichocarpa*.

A clear correlation was shown between the total cross-sectional area of shoots on the one hand and the dry weight of individual plants and the dry weight of harvested biomass on the other. This relationship could prove useful when making the first selection

in any breeding program designed to produce higher performing clones of black poplar.

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