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Performance of 15 *Miscanthus* Genotypes at Five Sites in Europe

John C. Clifton-Brown,* Iris Lewandowski, Bengt Andersson, Gottlieb Basch, Dudley G. Christian, Jens Bonderup Kjeldsen, Uffe Jørgensen, Jørgen V. Mortensen, Andrew B. Riche, Kai-Uwe Schwarz, Koeyumars Tayebi, and Fernando Teixeira

ABSTRACT

Miscanthus **is a genus of high-yielding perennial rhizomatous grasses with C$_4$ photosynthesis. Extensive field trials of *Miscanthus* spp. biomass production in Europe during the past decade have shown several limitations of the most widely planted clone, *M.* × *giganteus* Greef et Deu. A 3-yr study was conducted at five sites in Europe (Sweden, Denmark, England, Germany, and Portugal) to evaluate adaptation and biomass production potential of four acquisitions of *M.* × *giganteus* (No. 1–4) and 11 other genotypes, including *M. sacchariflorus* (Maxim.) Benth. (No. 5), *M. sinensis* Andersson (No. 11–15), and hybrids (No. 6–10). At each site, three randomized blocks containing a 5- by 5-m plot of each genotype were established (except in Portugal where there were two blocks) with micropropagated plants at 2 plants m^{-2}. In Sweden and Denmark, only *M. sinensis* and its hybrids satisfactorily survived the first winter following planting. Mean annual yields across all sites for all surviving genotypes increased each year from 2 t ha^{-1} dry matter following the first year of growth to 9 and 18 t ha^{-1} following the second and third year, respectively. Highest autumn yields at sites in Sweden, Denmark, England, and Germany were 24.7 (*M. sinensis* hybrid no. 8), 18.2 (*M. sinensis* hybrid no. 10), 18.7 (*M.* × *giganteus* no. 3), and 29.1 t ha^{-1} (*M.* × *giganteus* no. 4), respectively. In Portugal, where irrigation was used, the top-yielding genotype produced 40.9 t ha^{-1} dry matter (*M. sinensis* hybrid no. 7). Highest-yielding genotypes in Sweden and Denmark were among the lowest yielding in Portugal and Germany, demonstrating strong genotype × environment interactions.**

M ISCANTHUS × *giganteus* was introduced to Europe in the 1930s by Aksel Olsen and was observed to have exceptionally vigorous growth (Linde-Laursen, 1993). In the late 1980s, interest in C$_4$ perennial rhizomatous grasses, such as *Miscanthus* spp. (Nielsen, 1987), switchgrass (*Panicum virgatum* L.) (Christian, 1994), *Cyperus* spp., and *Spartina* spp. (Potter et al., 1995), for biofuel production increased due to their high yield potential and rising energy prices. Since 1983, extensive field trials of *M.* × *giganteus* have been carried out in northern Europe, showing the capacity of this genotype for yields >20 t dry matter ha^{-1} year^{-1} (Nielsen, 1987; Schwarz et al., 1994).

There are several reasons why European-wide biomass production from a single genotype within the *Miscanthus* genus is inadequate. First, in northern Europe, a number of sites established with *M.* × *giganteus* failed to survive during the first winter (Jones and Walsh, 2001), principally due to insufficient freeze tolerance of the overwintering rhizome. Second, it is unlikely that one single clone is sufficient to fulfil all of the quality requirements of different uses (combustion and fiber). Third, *M.* × *giganteus*, being a sterile triploid (Greef and Deuter, 1993), must be propagated vegetatively, either with rhizome cuttings or by micropropagation, making establishment expensive compared with crops established from seed. Fourth, growing large areas of a single clone increases disease risk. A broad genetic base and the provision of different *Miscanthus* genotypes are required to overcome these limitations.

As part of the European *Miscanthus* Improvement Project, a *Miscanthus* gene pool was created by combining collections directly from Asia and material already made available in Europe by German, Danish, and Swedish breeders. *M. sinensis* is characterized by a tuft-forming rhizome with high shoot densities while *M. sacchariflorus* is characterized by a broad, creeping rhizome with thick tall stems. *M.* × *giganteus* shows an intermediate type of rhizome between *M. sinensis* and *M. sacchariflorus* and is most probably a natural hybrid of the two (Greef and Deuter, 1993; Hodkinson et al., 1997).

In this paper, we report on field trials planted with 15 *Miscanthus* genotypes, which can be broadly divided

J.C. Clifton-Brown and I. Lewandowski, Univ. of Hohenheim, Inst. for Crop Prod. and Grassl. Res. (340), D-70599 Stuttgart, Germany; B. Andersson, Svalöf Weibull, AB-S 268 81, Sweden; G. Basch, K. Tayebi, and F. Teixeira, Departamento de Fitotecnia, Universidade de Évora, Herdade da Mitra, P-7001 Evora/Codex, Portugal; D.G. Christian and A.B. Riche, Rothamsted Exp. Stn., Harpenden, Hertfordshire AL5 2JQ, United Kingdom; and J. Bonderup Kjeldsen, U. Jørgensen, J.V. Mortensen, and K.-U. Schwarz, Danish Inst. of Agric. Sci., Dep. of Soil Sci., Res. Cent. Foulum, P.O. Box 50, 8830 Tjele, Denmark. Current address of J.C. Clifton-Brown: Bot. Dep., Univ. of Dublin, Trinity College, Dublin 2, Ireland. This work was funded by the EU Contract no. FAIR3 CT-96-1392. Received 6 Oct. 2000. *Corresponding author (jcbrown@tcd.ie).

Table 1. *Miscanthus* genotypes used in field trials, showing the European *Miscanthus* Improvement project (EMI) number, genotype name, genotype group abbreviation, ploidy, and acquisition details.

EMI no.	Name	Group	Ploidy	Acquisition code	Acquired from	Additional Information
1	*M.* × *giganteus*	Gig	3n	LASEI1	Larsen, Denmark	No. 16.05 in Greef et al., 1997
2	*M.* × *giganteus*	Gig	3n	ILP53	Knoblauch, Hornum	No. 16.21 in Greef et al., 1997
3	*M.* × *giganteus*	Gig	3n	HAGA 56	Hagemann, Berlin	No. 17.02 in Greef et al., 1997
4	*M.* × *giganteus*	Gig	3n	GREIF63	Greifswald Bot. Gnd.	No. 17.03 in Greef et al., 1997
5	*M. sacchariflorus*	Sac	4n	MATEREC11	Deuter, Germany	Matumura et al., 1985
6	*M. sinensis* Hybrid	Sin-H	3n	GOFAL7	Deuter, Germany	Hybrid selected in a *M. sinensis* population
7	*M. sinensis* Hybrid	Sin-H	2n	BERBO42	Deuter, Germany	Hybrid of two *M. sinensis*
8	*M. sinensis* Hybrid	Sin-H	aneuploid	RH43	Deuter, Germany	Hybrid of *M. sacchariflorus* × *M. sinensis*
9	*M. sinensis* Hybrid	Sin-H	2n	JESEL78	Deuter, Germany	Hybrid of two *M. sinensis*
10	*M. sinensis* Hybrid	Sin-H	2n	RH81	Deuter, Germany	Hybrid of *M. sacchariflorus* × *M. sinensis*
11	*M. sinensis*	Sin	2n	88-110	Brander, Denmark	Collected in Honshu, Japan in 1983, selected 1988
12	*M. sinensis*	Sin	2n	88-111	Brander, Denmark	Collected in Honshu, Japan in 1983, selected 1988
13	*M. sinensis*	Sin	2n	90-5	Brander, Denmark	Collected in Honshu, Japan in 1983, selected 1990
14	*M. sinensis*	Sin	2n	90-6	Brander, Denmark	Collected in Honshu, Japan in 1983, selected 1990
15	*M. sinensis*	Sin	2n	SW217	Andersson, Sweden	Collected Hokkaido, Japan in 1990

into four genetic groups, at five locations: Sweden, Denmark, England, Germany, and Portugal. The objective was to screen these genotypes in different soil and climatic conditions for yield performance traits, including height, stem density, flowering time, autumn senescence rate, and yield. These measurements were used to identify the most suitable genotypes for the various regions of Europe and to improve our knowledge of the genetic base for the future development of *Miscanthus* spp. as a biomass crop.

MATERIALS AND METHODS

Genotypes

From the gene pool held by European breeders, 15 of the most promising genotypes were selected (Table 1). Four acquisitions of *M.* × *giganteus* genotypes (No. 1–4) were chosen because different crosses between *M. sacchariflorus* and *M. sinensis* were suspected to be in circulation all under the same name. Amplified fragment length polymorphism (AFLP) analysis had revealed small detectable differences between some of these acquisitions (Greef et al., 1997). One, *M. sacchariflorus* (No. 5), was selected from a collection described by Matumura et al. (1985). Five pure *M. sinensis* types were selected, four of which were collected by the Danish collector Poul Brander in 1983 on Honshu Island (No. 11–14) and one from central Hokkaido in 1990 by the Swedish collector Zandra Andersson (No. 15). Five *Miscanthus* spp. hybrids from crosses within *M. sinensis* and *M. sacchariflorus* (No. 6–10) were selected based on known differences in key physiological characteristics such as flowering time and autumn senescence time.

Field Trials

To ensure genetic and physiological uniformity at the five sites in Sweden, Denmark, England, Germany, and Portugal, all 15 genotypes were micropropagated by explants. Genotype no. 1–10 were propagated by Martin Deuter at TINPLANT (Klein-Wanzleben, Germany), and Genotype no. 11–15 were propagated at the Danish Institute of Plant and Soil Science (Aarslev, Denmark). After in vitro multiplication, the plants were grown for 8 to 10 wk in a temperate greenhouse in 5- by 5-cm peat pots before shipping to the field trials. Plant height at planting averaged 20 cm.

At each site, three randomized blocks containing a 5- by 5-m plot of each genotype were established (except in Portugal where there were two blocks). Plantlets were planted by hand between March and June 1997 at a density of 2 plants m^{-2}. Plots were irrigated after planting at all sites. In Portugal, irrigation was necessary throughout all growing seasons because of the limited rainfall during the growing season and sandy soils (Tables 2 and 3). Fertilizer was applied at rates equivalent to 60, 44, and 110 kg ha^{-1} yr^{-1} N, P, K, respectively, which, combined with residual nutrients, was estimated to saturate crop requirements (Lewandowski et al., 2000). Mechanical weed control was used between transplanting and September in 1997.

Plant survival during the first winter was quantified by counting the number of plants producing new shoots in spring 1998, and percentages of the total plants planted in 1997 were calculated. Five plants were tagged for regular measurements at the beginning of each growing season on plants in predetermined positions at least two rows from the edge of each plot. Tagged plants were in the middle of the plots to avoid border effects. Stem density and canopy height were monitored regularly within the growing season on the tagged plants. In each

Table 2. Site locations, planting dates in 1997, soil textures (sand, silt, and clay), soil bulk densities (bulk), and soil types to subgroup level in five countries.

Country	Location	Planting date	Soil characteristics (0–90 cm)				
			Sand	Silt	Clay	Bulk	Soil type†
			— % —			g cm^{-3}	
Sweden	55°60′ N, 14°00′ E	17 June	83.0	12.0	5.1	1.5	Aeric Endoaquept
Denmark	56°30′ N, 09°35′ E	03 June	63.1	25.8	11.2	1.5	Typic Fragiudalf
England	51°48′ N, 00°21′ W	22 May	41.5	29.2	29.4	1.7	Aquic Paleudalf
Germany	48°40′ N, 09°00′ E	21 May	8.2	51.7	40.2	1.5	Vertic Eutrudept
Portugal	38°43′ N, 09°13′ W	21 Mar.	76.2	15.7	8.1	1.6	Aquic Xerofluvent

† Soil Survey Staff, 1999.

Table 3. Mean air temperature and rainfall at the field sites in the five countries for the period April to September in 1997, 1998, and 1999; the long term (LT) mean for this period (>10 yr data during last 30 yr), and the annual LT (Ann.) mean.

	Air temperature					Rainfall				
	April to September				Ann. LT mean	April to September				Ann. LT mean
Country	1997	1998	1999	LT mean		1997	1998	1999	LT mean	
	°C					mm				
Sweden	13.4	12.5	13.8	13.0	7.9	297	459	416	359	696
Denmark	12.9	12.0	13.0	12.1	7.3	354	334	409	334	626
England	14.2	13.6	14.4	12.9	9.1	273	436	364	319	688
Germany	13.6	13.8	14.9	13.3	7.9	374	326	410	425	687
Portugal†	23.0	21.4	20.2	19.6	15.4	327	197	147	156	665

† In Portugal, irrigation between April and September was 300, 382, and 506 mm in 1997, 1998, and 1999, respectively.

autumn, *greenness* (an indication of the senescence) of the shoots was assessed on the tallest shoot of three of the tagged plants. If the length of the green part of a fully expanded leaf was >60%, then it was counted as green. If the green part was <60%, then it was counted as dead. Green leaf area was estimated by eye. Percentage greenness was calculated from the sum of green-scored leaves over the total number of leaves on a shoot × 100. Three shoots per plot were pooled to make a plot mean, and plot means were used as replicates ($n = 3$, except Portugal where $n = 2$).

Each autumn, a sample of 2 m² was harvested from within the central area of the plot (avoiding border effects) to assess the total aboveground productivity of each genotype. Cutting height for yield determinations was 5 cm. Moisture content was measured by drying at 80°C until constant weight had been achieved, and dry matter yield was calculated as tonnes of dry matter per hectare.

Statistical Analyses

When data were normally distributed, analysis of variance (ANOVA) was used to determine significant differences. For comparisons of shoot density, plant height, and yield among genotypes within a country at a particular sampling time, a minimum significant difference was calculated by the Tukey procedure at $P = 0.05$. Interactions among genotype, year, and country, where appropriate, are shown in the tables. For shoot greenness, plot means from the five selected plants were used from the three replicate plots for each genotype at each site to produce a standard error. Regression relationships between genotype yields in the different years were calculated across all countries. Plant height and shoot density in the third growing season were regressed with yield to assess the yield predictability using these simple growth parameters. All statistics were performed within Data Desk v.6 (Data Description, Ithaca, NY).

RESULTS AND DISCUSSION

Climatic Conditions during the Growing Season

Mean air temperatures and rainfall sums from April to September are given in Table 3 for all 3 yr of the field trials. The long-term means (>10 yr of data) and the whole year are also shown. Growing season air temperatures in 1997, 1998, and 1999 were generally warmer than normal for the five locations. Air temperatures in Portugal were at least 5°C higher than at the other locations.

Long-term annual rainfall at all sites was similar (626–696 mm), but rainfall distribution varied markedly with location. On average, the German site had relatively wet summers (425 mm) while Portugal had wet winters (509 mm). Consequently, in Portugal, natural water availability for crop growth was very limited, and irrigation was necessary.

Plant Survival

Established *Miscanthus* spp. stands grow from overwintering rhizomes in spring. At the more northern sites of Denmark and Sweden, nearly 100% and >50% of the plants of *M.* × *giganteus* and *M. sacchariflorus*, respectively, died during the first winter following planting, but losses of *M. sinensis* plants were only 1 to 16% (Table 4). In Denmark and Sweden, plant losses of *M. sinensis* hybrids (No. 6–10) varied from only 1% in Genotype no. 10 to 60% in Genotype no. 9. In these countries, winter soil temperatures in the first year following planting fell below −4.5°C. In England, Germany, and Portugal, where winter soil temperatures were warmer (> −2.8°C), all genotypes survived. These observations

Table 4. Plant losses and minimum soil temperature recorded at 5-cm depth in the field during the first winter (1997–1998) for the 15 *Miscanthus* genotypes at five sites.

	Genotype group and no.															
	Gig				Sac	Sin-H					Sin					Soil temp.
Country	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	
	% plant loss															°C
Sweden	99	93	97	99	50	17	52	6	50	2	6	5	16	5	5	−5.4
Denmark	100	98	96	96	67	1	8	6	60	1	1	10	15	6	1	−4.5
England	0	0	0	0	2	0	0	1	1	0	4	1	21	1	0	−1.2
Germany	0	1	0	1	1	0	5	0	1	0	1	0	0	1	5	−2.8
Portugal	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	NA

Table 5. Autumn yield for the first 3 yr (1997, 1998, and 1999) of 15 *Miscanthus* genotypes grown at five locations in Europe.

			Gig				Sac	Sin-H					Sin						
Country	Year	Date†	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	Mean	Tukey‡ MSD
			dry matter yield, t ha^{-1}																
Sweden	1997	12 Nov.	0.1	0.1	0.1	0.1	0.5	0.2	0.4	0.4	0.6	0.8	0.4	0.2	0.2	0.4	0.3	0.3	0.6
	1998	24 Nov.	0.0	0.0	0.0	0.0	0.0	5.0	3.8	9.5	5.4	11.4	5.2	5.0	6.7	3.9	3.1	3.9	4.4
	1999	18 Oct.	0.0	0.0	0.0	0.0	0.0	19.3	11.0	24.7	14.2	20.5	15.4	15.3	17.3	13.8	9.7	10.7	8.0
Denmark	1997	27 Nov.	0.7	0.8	1.1	0.8	0.5	0.9	1.1	0.3	1.4	1.4	0.4	0.3	0.3	0.6	0.2	0.7	0.7
	1998	19 Nov.	0.0	0.0	0.0	0.0	0.7	5.2	4.7	4.1	1.4	7.8	5.2	3.1	3.3	2.4	1.9	2.7	3.0
	1999	18 Nov.	0.0	0.0	0.0	0.0	1.4	16.4	10.4	15.9	0.9	18.2	15.0	12.4	11.2	9.9	6.8	7.9	5.1
England	1997	15 Nov.	0.6	0.8	0.9	0.8	0.4	1.4	1.2	0.2	1.7	0.8	0.1	0.1	0.2	0.1	0.2	0.6	0.8
	1998	24 Nov.	3.0	5.7	5.9	5.3	1.9	7.0	5.9	1.4	7.7	6.0	1.9	2.0	3.0	0.6	2.0	4.0	3.8
	1999	3 Nov.	13.8	16.8	18.7	14.6	11.1	15.7	17.7	6.5	15.8	14.0	8.0	7.0	10.9	4.6	5.9	12.1	7.4
Germany	1997	11 Nov.	3.2	3.4	3.3	3.0	2.9	2.8	1.1	1.0	3.0	1.9	0.7	0.6	1.0	0.5	0.5	1.9	2.0
	1998	23 Nov.	8.0	8.7	8.9	8.1	4.6	7.4	3.6	7.3	7.6	9.7	4.4	4.5	5.0	3.6	2.5	6.2	5.2
	1999	21 Nov.	22.8	24.3	25.7	29.1	12.6	20.0	17.0	19.2	10.3	19.1	12.8	10.9	12.3	10.4	9.1	17.0	15.3
Portugal	1997	27 Oct.	8.4	4.6	6.0	8.1	4.4	7.9	7.5	2.5	7.6	6.3	5.9	4.7	5.7	5.3	2.0	5.8	2.7
	1998	15 Oct.	26.9	25.5	30.3	25.6	15.1	18.4	27.2	10.8	20.1	13.6	10.6	10.2	16.3	11.6	8.8	18.1	8.5
	1999	6 Oct.	37.8	36.4	36.8	34.7	35.2	27.2	40.9	21.0	26.3	20.3	16.2	16.3	22.4	16.1	16.1	26.9	17.1

ANOVA All countries, genotypes, and years

Source of variation	df	P
Country	4	***
Genotype	14	***
Country × genotype	56	***
Year	2	***
Country × year	8	***
Genotype × year	28	***
Country × genotype × year	92	***

*** Significant at the 0.001 probability level. ANOVA's on the three years within each site showed genotype, year, and genotype × year differences were all highly significant (***) (not shown above).
† Harvest date.
‡ Minimum significant differences (MSD) between genotypes within a site and within a year were calculated by the Tukey test at *P* = 0.05.

are consistent with artificial freezing tests, which showed that rhizomes of *M. sinensis* hybrids could survive temperatures below −4.5°C, but rhizomes of *M.* × *giganteus* and *M. sacchariflorus* are killed at approximately −3°C (Clifton-Brown and Lewandowski, 2000). It appears, therefore, that temperatures in Sweden and Denmark were too cold for *M.* × *giganteus* and *M. sacchariflorus*. Consequently, *M. sinensis* and *M. sinensis* hybrids can be recommended for the regions where soil temperatures at 5-cm depth are likely to fall below −3°C.

Biomass Yield

Following planting in 1997, average biomass yield for all genotypes across all sites was only 1.9 t ha^{-1} in autumn (Table 5). Highest mean yields were in Portugal (5.8 t ha^{-1}) while lowest yields were in Sweden (0.36 t ha^{-1}). There were several reasons for this. First, late frosts in Sweden prevented planting before June, thus shortening the growing season by 2 mo compared with Portugal. Second, the plants were irrigated in Portugal, but in Sweden, growth was probably water limited because rainfall was abnormally low (Table 3). Third, growth temperatures in Portugal were higher throughout the season, promoting faster establishment.

In the second year, average yield for all sites and all surviving genotypes increased to 8.6 t ha^{-1}. In Portugal, *M.* × *giganteus* yields averaged 27 t ha^{-1}, but in England and Germany, where average air temperatures were approximately 5°C lower and plants were not irrigated, yields did not exceed 9 t ha^{-1}. *M. sinensis* hybrid no. 10

outyielded all other genotypes in Sweden and Denmark (11.4 and 7.8 t ha^{-1}, respectively). This indicates that *M. sinensis* hybrid no. 10 is better suited to low winter soil temperatures and shorter growing seasons than other genotypes in these trials.

In the third year, the highest-yielding genotypes in Germany and England were of *M.* × *giganteus*. In Portugal, the highest yield was obtained from *M. sinensis* hybrid no. 7, with autumn yield of 40.9 t ha^{-1}. *M. sinensis* genotypes (No. 11–15) produced a consistently lower yield than the highest-yielding genotype at any given location. Interestingly, at the higher latitudes in Sweden and Denmark, the yield gap between the *M. sinensis* genotype group (No. 11–15) and the other genotypes was smaller than at lower latitudes. This can be explained through lower overall yields at higher latitudes due to the shorter growing season.

Biomass yield interactions between year and genotype in any given country were significant at *P* < 0.001. Interactions among country, year, and genotype were also significant at *P* < 0.001. This shows that stand maturity increases yield significantly for all surviving genotypes. Equally, the different environmental conditions, particularly climate and soil, influence genotype yield performance (Table 5).

Comparisons between yields obtained in these field trials and other trials in Europe can only be made for the widely grown *M.* × *giganteus*. Trials in northern regions tend to mature slower than at southern latitudes, and ceiling yields of 15 t ha^{-1} are reached after only the fourth or fifth year (Lewandowski et al., 2000). Yields

Table 6. Correlations between yields in the first, second, and third growing seasons and the shoot densities and plant height using data from all sites and all genotypes except for those that died in Sweden and Denmark during the first winter following planting.

Variable 1	Variable 2	df	Slope	P	Constant	P	b^2
Yield year 1	Yield year 3	64	2.9	***	11.0	***	0.56
Yield year 2	Yield year 3	64	1.2	***	8.0	***	0.81
Yield year 3	Shoot density year 3	64	0.0	NS†	14.8	***	0.07
Yield year 3	Height year 3	64	0.1	***	−4.5	NS	0.54

*** Significant at the 0.001 probability level.
† NS, not significant at $P < 0.05$.

with irrigation in excess of 25 t ha^{-1} in southern Europe are often achieved in the second year with *M. × giganteus* (Clifton-Brown et al., 2001). Therefore, in warm climates, yields after the second growing season are a good indication of the potential ceiling yield from a genotype, but in cooler climates, it takes at least 3 yr to reach a ceiling. As expected, regression coefficients with third-year yield against first- and second-year yields were better for the second year than for the first year (Table 6).

Growing Season Performance Characteristics

The greatest yield variance from site to site was observed in the *M. sinensis* hybrid genotypes. For example, the lowest autumn yields in Sweden were for Genotype no. 7, the genotype that performed best in Portugal and England. For future breeding of higher yielding *Miscanthus* genotypes better suited to specific environments, it will be necessary to identify key growth characteristics that will increase the quantity of radiation intercepted and its conversion into harvestable dry matter over a growing season. Recent evidence suggests that photosynthetic efficiency per unit area of leaf has not been successful in identifying more productive genotypes of crop plants (Lawlor, 1995). Furthermore, within

a photosynthetic group, radiation conversion efficiency varies little (Monteith, 1978). Thus, yield from a given genotype will depend largely on the efficiency with which radiation is intercepted by the canopy leaves. This efficiency is dependent on a range of characteristics, including shoot emergence time from the overwintering rhizome and rate of canopy closure [a function of shoot density and leaf expansion rate (Clifton-Brown and Jones, 1997), flowering time after which growth stops and senescence begins]. These characteristics are detailed here for the third year following planting.

Genotypic differences in shoot emergence time in the spring were not observed within these field trials. Evidently, monthly measurements of plant height were insufficient to identify emergence differences at a site (data not shown). However, the monthly measurements did show the earlier start to the growing season in Portugal (data not shown). By the end of the third growing season, plants were tallest in Portugal (223 cm) and shortest in Denmark (120 cm) (Table 7).

Analysis of variance showed that shoot density and plant height differed significantly for all 15 genotypes within and between countries ($P > 0.001$) at the end of the growing season (Table 7). In those countries where *M. × giganteus* and *M. sacchariflorus* survived the first winter, plants were generally taller than the *M. sinensis* and *M. sinensis* hybrids. In contrast, shoot densities were higher in the *M. sinensis* and *M. sinensis* hybrids. Evidently, there is an antagonistic relationship between plant height and shoot density.

Genotypic variation in plant height appears to be associated with flowering time. The tallest genotypes at a site (e.g., *M. sacchariflorus* or *M. × giganteus*) tended to flower later than the earliest-flowering *M. sinensis* genotypes (No. 11–15). Indeed, it was only in Portugal where all of the genotypes reached flowering before autumn (Table 8). Thermal time required from emergence in spring to flowering of *M. × giganteus* in Portu-

Table 7. Shoot density and plant height at the end of the growing season for 15 *Miscanthus* genotypes grown at five locations in Europe.

Country	Gig 1	Gig 2	Gig 3	Gig 4	Sac 5	Sin-H 6	Sin-H 7	Sin-H 8	Sin-H 9	Sin-H 10	Sin 11	Sin 12	Sin 13	Sin 14	Sin 15	Mean	Tukey† MSD
										Shoot density, m^{-2}							
Sweden	0	0	0	0	0	75	55	162	55	137	127	131	119	108	100	71	36
Denmark	0	0	0	0	8	69	51	101	10	141	107	96	84	115	73	57	36
England	51	58	56	53	36	95	93	124	122	178	115	99	112	82	99	92	61
Germany	67	75	63	70	45	98	67	225	46	196	126	129	114	117	104	103	29
Portugal	88	82	75	95	82	165	113	270	133	273	252	196	168	226	284	167	57
										Plant height, cm							
Sweden	0	0	0	0	0	271	142	234	185	255	217	216	233	232	220	147	33
Denmark	0	0	0	0	184	196	165	182	112	180	167	160	172	151	136	120	22
England	221	245	247	225	251	199	183	120	163	178	155	140	142	104	141	181	36
Germany	273	278	261	283	308	192	178	172	127	183	175	175	175	177	160	208	47
Portugal	295	300	305	301	383	169	287	148	198	145	159	167	168	162	156	223	27

ANOVA

Source of variation	df	Shoot	Height
Country	4	***	***
Genotype	14	***	***
Country × genotype	56	***	***

*** Significant at the 0.001 probability level.
† Minimum Significant Differences (MSD) between genotypes within a site and within a year were calculated by the Tukey test at $P = 0.05$.

Table 8. Flowering date and shoot greenness (%) in autumn at the end of the third growing season (1999) for 15 *Miscanthus* genotypes grown at five locations in Europe.

Country	Genotype group and no.														
	Gig				Sac	Sin-H					Sin				
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
	Flowering date														
Sweden	†	†	†	†	†	8 Sept.	none	none	20 Sept.	8 Sept.	10 Aug.	10 Aug.	20 Aug.	2 Aug.	25 July
Denmark	†	†	†	†	†	15 Sept.	none	15 Oct.	15 Oct.	15 Sept.	20 Aug.	20 Aug.	25 Aug.	20 July	20 July
England	none	none	none	none	none	29 Sept.	none	29 Sept.	29 Sept.	29 Sept.	22 July	22 July	22 July	22 July	22 July
Germany	none	none	none	none	none	26 Aug.	20 Sept.	20 Sept.	20 Sept.	12 Aug.	13 July	30 June	13 July	30 June	30 June
Portugal	10 Sept.	13 Sept.	10 Sept.	10 Sept.	16 Sept.	6 July	10 Aug.	6 July	19 July	22 June	8 June	8 June	15 June	4 June	4 June
	Greenness (%)‡														
Sweden	†	†	†	†	†	0	46 ± 3	2 ± 2	15 ± 15	2 ± 2	0	0	0	0	0
Denmark	†	†	†	†	24 ± 7	8 ± 4	39 ± 2	0	60 ± 1	0	0	0	0	0	0
England	25 ± 16	0	16 ± 11	0	24 ± 4	0	52 ± 2	0	72 ± 2	0	0	0	0	0	0
Germany	17 ± 3	20 ± 0	15 ± 3	22 ± 7	15 ± 3	15 ± 3	62 ± 2	0	44 ± 4	2 ± 1	0	4 ± 2	22 ± 10	0	0
Portugal	53 ± 3	51 ± 2	49 ± 1	45 ± 2	40 ± 4	44 ± 2	60 ± 4	24 ± 6	56 ± 6	37 ± 3	43 ± 7	48 ± 2	42 ± 12	45 ± 5	48 ± 2

† Indicates plants did not survive the first winter following planting.
‡ Means shown are ±1 SE, $n = 3$, except Portugal where $n = 2$.

gal was >1800 degree days above a threshold of 10°C. In contrast, the *M. sinensis* genotypes (No. 11–15) only needed 400 to 600 degree days in Portugal. This effectively halves the length of the available growing season, approximately halving the yield (Tables 5 and 8). In Sweden and Denmark, where temperatures are lower, *M. sinensis* flowers later in the growing season so that less radiation is lost than at warmer sites. As with yield (Table 5), flowering time was most variable for the *M. sinensis* hybrid (No. 6–10) group within a site. For example, in Portugal, Genotype no. 7 was late flowering, and this coincided with the highest yield recorded throughout (Table 5).

In general, late flowering was associated with late senescence. For example, Genotype no. 7 senesced latest at all sites and was still relatively green compared with all other genotypes except Genotype no. 9 in late autumn (Table 8). Although early flowering leads to nearly complete senescence in northern climates, it did not lead to low greenness in Portugal. The *M. sinensis* hybrid genotypes no. 7 and 9 are *stay-green* types. Such stay-green traits are being investigated in other crops (Thomas and Howarth, 2000; Xu et al., 2000) with a view to increasing yield. However, in northern regions of Europe, the late-senescing genotypes (*M. sinensis* hybrids no. 7 and 9) yielded badly. Autumn frosts kill the green leaves of late-maturing genotypes at northern sites. This may lead to insufficient relocation of nutrients and assimilates from the aboveground shoots to the rhizomes in autumn, reducing both the overwintering capacity and regrowth potential in the following spring (Pude et al., 1997). Interestingly, overwintering of Genotypes no. 7 and 9 in Sweden and Denmark were the poorest of the *M. sinensis* hybrids (Table 4).

When genotypes that failed to overwinter in Sweden and Denmark are omitted (Table 4), plant height was the growing season performance characteristic most highly correlated with yield (Table 6). Greater height implies later flowering, and therefore, longer periods for which radiation is converted to biomass. Shoot densities were not found to be significantly related to yield (Table 6) for the genotypes tested. Exceptions to these general relationships included *M. sacchariflorus* (No. 5), which despite being the tallest genotype, had a low shoot density that lowered its yields below the highest-yielding genotypes (Table 7). *M. sinensis* hybrid genotypes (No. 6–10) showed the potential for breeding genotypes with appropriate characteristics for a wide range of environmental conditions. For future breeding, *M. sacchariflorus* will be an invaluable parent to create new taller, and therefore, higher yielding *M. sinensis* hybrid genotypes with rhizome freezing tolerance and late flowering time.

Practical Issues in Relation to *Miscanthus* spp. Production for Biomass

Without irrigation, *Miscanthus* spp. would not be productive in Portugal because of the combination of a low water-retentive sandy soil type, high evaporative demand, and low rainfall during the growing season. At the other four sites, no irrigation was used in 1999. Yields were highest at the German site, which had not only the most rainfall in 1999, but also a heavy clay soil. Because biomass production must be low cost and low input, it is unlikely that irrigation will be economic, and a combination of site and genotype selection will be important to ensure survival and adequate yields.

It is important to point out that biomass quality for combustion improves if the crop is harvested in early spring rather than in the previous autumn. Yields can, however, be as much as 30% less in the following spring due to death and detachment of leaves and stem tops (Clifton-Brown et al., 2000; Jørgensen, 1997).

There is a potential weed risk from the diploid *Miscanthus* genotypes in Europe because fertile seeds were produced at all sites where flowering occurred. To date, seedlings have not been found to spread far from the plots, which have had frequently mown paths. Current breeding programs for *Miscanthus* spp. are attempting to produce infertile hybrids (K.K. Petersen, personal communication, 2000).

CONCLUSIONS

New plantations with *M. × giganteus* and *M. sacchariflorus* are unlikely to be viable where winter soil temperatures fall below $-3°C$ at a depth of 5 cm. In England and Germany, *M. × giganteus* genotypes were among the top performers. These genotypes yielded well in Portugal, >34 t ha^{-1}, but the highest-yielding genotype in Portugal was the stay-green *M. sinensis* hybrid no. 7 (41 t ha^{-1} dry matter). The highest-yielding genotypes in Sweden and Denmark were the *M. sinensis* hybrids no. 6, 8, and 10. These results demonstrate that different *M. sinensis* hybrids can be found for a wide range of climatic conditions in Europe. In mid-Europe, *M. × giganteus* is still the genotype of preference. Plant height, which is largely controlled by flowering time, was a more important selection characteristic than shoot density.

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