Aus dem Institut für Pflanzenbau und Grünlandwirtschaft

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Giant Reed (Arundo donax L.) Network
Improvement biomass quality
Final report FAIR-CT-96-2028
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GIANT REED (*ARUNDO DONAX*) NETWORK

- Improvement, Productivity and Biomass Quality -

Final Report

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Objectives

Introduction of Giant Reed (Arundo donax L.), a high yielding, non-food plant, into EU agriculture for energy and/or pulp production, as well as for the construction of building materials.

Actions of the Institute within the project

Task 2: Adaptation in N-W EU countries

Sub-task 2.2: Evaluation of Giant Reed in NW EU countries

Sub-task 4.1: Application of micropropagation method on Giant Reed and cost assessment of propagation technique

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Summary

The native growth areas of Giant Reed (Arundo donax L.) are located in southern European regions (Greece, Italy, southern France, Spain, Portugal) and other Mediterranean countries but it grows also in other subtropical parts of the world. Arundo donax is a perennial, potentially high yielding non-food crop naturally growing along river banks, creeks and generally moist soils but also successfully on relatively dry and infertile soils such as roadsides and is used to mark field sites. Because of high biomass production and high cellulose content this plant might be a very promising crop for pulp, paper and energy use. Plant cultivation could contribute to a reduction of wood material requirements and thus would preserve native woodlands in arid areas. Giant reed is harvested in several countries for paper making, musical reeds and thatching material for roofs. It was introduced into California (USA) in the past century because of high potential of protection against soil and riverbank erosion due to high amount of rhizomes. The plant could influence climatic conditions and has the potential for CO_2 neutral production and use.
For Europe new market niches such as bio-energy and renewable industrial resources might be an interesting, forward-looking research field for Giant reed cultivation. Agricultural plant cultivation cycles could be widened and give new economical chances and income for rural populations.

This report reflects the activities and results of the research work carried out by the Institute of Crop and Grassland Science within the European Research Framework "Giant Reed (Arundo donax L) Network - Improvement, Productivity and Biomass quality" in the period 1997 - 2001. Two major aspects are considered, the adaptability of species under field conditions in northern Germany and the assessment of biomass productions as well as effects on the environment.

Rhizomes of ten Giant reed populations of different originally locations in southern France (Nimes and Bizet), north-eastern Italy (Rabuise and Torviscosa), southern Italy and Sicily (Caltagirone and Fondachello), central and northern Greece (Messolonghi and Ionannina) and southern Greece and Crete (Attiki and Hania) were planted in experimental field plots of 12 m^2 size with three replications. In every year 12 plants in the centre of the plot (4.8 m^2) were monitored for winter survival, plant growth and height, shoot number, shoot diameter, production of leaves and finally biomass yield in January.

Establishment of Arundo donax at the field site in Braunschweig by rhizomes was successfully for all populations. More than 86 - 100% of the plants survived frost conditions during winter period. Highest plant failures were observed for southerly populations Fondachello and Caltagirone, while plants of population Torviscosa (north eastern Italy) showed no failures during all years. Some replanting was necessary in the fields so that plants established in 1997 and 1998 were monitored separately in the following years.

Results show different growth and yields for both plant ages. Shoot development, shoot diameter, height and dry matter yield was higher in plants established in 1997 than for those established in 1998.
The populations with originally more northerly locations (Torviscosa, Rabuise) established in 1997 showed less number of shoots/plant and growth height than populations with more southerly original location (Attiki, Hania). Plant height on average was between 2 and 2.5 m. Highest plants on average were found in population Hania. Single plants of this population reached more than 4 m height. Between 5 and 18 shoots/plant were recorded with shoot diameter between 1.8 and 2.6 cm.

A biomass yield between 7 and 22 t dry matter /ha with an average of 14 t/ha was measured. Yields increased during the years but showed a high variation. Maximum yield of 25 t DM were obtained from population Hania.
Arundo donax did not finish physiological maturation until frost in autumn and did not flower in every year. Translocation of assimilates into the rhizomes is not finished before frost and rhizomes might be therefore sensitive against lower temperatures during winter. Biomass with 45 - 50 % dry matter content were harvested in January. Drying is therefore necessary before storage.

No weed competition as well as diseases and pests were observed during the vegetation periods. The plants could stand heavy rain and strong winds without lodging.

For the first time long-term field trials with giant reed were successfully established in northern Germany. Giant reed showed promising biomass yields compared with Miscanthus giganteus and sufficient high quality of ligno-cellulosic material for energy and fibre production.

1. Introduction

Giant Reed (*Arundo donax* L.) is a perennial, potentially high yielding non-food crop and is a wild grown plant in southern European regions (Greece, Italy, southern France, Spain, Portugal) and other Mediterranean countries. It grows also in other subtropical parts of the world like in India, China and in southern USA and some genotypes are also adapted to cooler climate conditions. Under natural site conditions giant reed is usually found along river banks, creeks and generally moist soils but it grows also successfully on relatively dry and infertile soils such as roadsides and is used to mark field sites (Dalianis, 1996, Wynd et al., 1948, Tucker, 1990, Sharma et al., 1998, Günes and Saygin, 1996).

Giant Reed is one of the largest C3-grass species reaching up to 14 m height with stem diameter up to 3.5 cm. The plant species develops clumps but can also spread vigorously due to its long woody rhizomes and can invade natural plant communities under certain environmental conditions. When planted, several buds are mobilised and up to 10 stems per rhizome may emerge until the end of the first growing period.
In southern France Giant Reed produced 20-25 t/ha dry matter (Toblez, 1940). Other authors reported dry matter yields of 35 t/ha in northern Italy (Matzke, 1988) while also 53 t/ha dry matter were measured in wild stands of Giant Reed in Turkey (Gunes and Saygin, 1996). First results from field plots in southern Germany showed in the years 1989 and 1990 dry matter yields between 7 and 26 t/ha (Schweiger and Oster, 1991; Oster and Schweiger 1992).

The dry matter content of harvested biomass of Giant Reed varies in Mediterranean climates between 36 and 49 % (Dalianis, 1996). Drying in the field after clipping resulted in a reduction of moisture up to a final moisture content of 15 % after 10 days (Günes and Saygin, 1996).

The stems and foliage of Giant Reed contains between 45 and 65 % of cellulose and hemicellulose and between 13 and 25 % lignin dependent on the plant age (Arnoux 1974; Faix et al, 1989, Pascoal Neto et al., 1997). The fibre length of Giant Reed is comparable with those obtained from wood material (Fagus, Picea abies) (Faix and Bremer, 1988). The ash content is about 3 % (Schweiger & Oster, 1991; Pascoal Neto et al. 1997). First experiments to assess suitability for energy production showed an energy content of 17 MJ/kg (heating value) (Faix et al, 1988). The total energy yield of Giant Reed fields was estimated between 425 and 561 GJ/ha.

Giant Reed can be cultivated like Miscanthus giganteus, an other C4-grass species producing high biomass yields. The water requirement of Giant Reed is about 300-700 mm in the vegetation period and a transpiration coefficient of approximately 200 l/kg DM was calculated (Chiaramonti et al. 2000). These data are comparable with Miscanthus growth requirements (Transpiration coefficient: 250 l/kg DM, Water requirement 500-700 mm/vegetation period). For plant cultivation not more than 100 kg nitrogen/ha and 150 kg P_2O_5 as well as 200 kg K_2O are recommended for fertilisation (Dalianis, 1996, El Bassam, 1998).

Giant Reed cultivations are less effected by insects and pests (Dalianis, 1996). Several substances in the plant such as noxious chemicals like tri-terpines, sterols, cardiac glycosides, curare-mimicking indols and hydroxamic acids might protect this species from damage due to insects (Jackson and Nunez, 1964; Chandhuri and Ghosal, 1970; Ghosal et al., 1972; Zuniga et al, 1983). It remains green also during summer drought and can withstand accidental fire sweeping across giant reed plantations (Dalianis, 1986).

Like Miscanthus, Giant Reed could be a source for energy, paper and building materials. Stems were harvested in several countries for paper making, musical reeds and other industrial puposes (Perdue, 1958). In 1820, it was introduced into California from Mediterranean countries for erosion control in drainage canals and was also used as thatching material for roofs of buildings (Hoshovsky, 1987). Subsequent planting have been made for the production of reeds for a variety of musical instruments including bassoons and bagpipes (Bell, 1998).

2. Aims of the Arundo donax productivity-Network

The aim of the Arundo productivity network is to establish *Arundo donax* (Giant Reed) under climatic conditions of N-W European countries. Because of the provenance of *Arundo donax* this plant is adapted to climatic conditions of southern European regions which are without risk of frost. Only certain genotypes seem to be adapted to cooler climates and can be grown in N-W European countries such as the United Kingdom and Germany (Dalianis, 1996). The contribution of the Federal Agricultural Research Centre (FAL) is to select different Giant Reed genotypes, which are resistant against frost under winter conditions of northern regions of Europe and are able to produce high dry matter yields.

3. Material and Methods

3.1 Soil features at the research centre in Braunschweig, northern Germany

The soil of the Federal Research Centre of Agriculture is a sandy loam in the depth of 0 – 50 cm and a loamy sand deeper than 50 cm (Table 1) with low organic matter content and is by nature of a moderate soil fertility.

Table 1: Grain-size distribution

Depth [cm]	Sand (%)	Silt (%)	Clay (%)	Soil type
0 – 25	34	57	9	sandy loam
25 – 50	33	55	12	sandy loam
50 – 60	64	28	8	loamy sand
60 – 80	75	14	11	loamy sand

The top soil reacts nearly neutral with a pH of 6.5. The C/N-ratio amounts are mostly around 10. Ground water stands in a depth of 7 m. Soil content of nutrients is middle-good. Yearly fertilisation with calcium and potassium fertilisers is usual. Additional fertilisation with phosphorus is not necessary. Irrigation of the experimental plots is possible.

3.2 Meteorological data

The meteorological data for monthly averages in the years 1997 to 2000 are given in the Appendix (Table 1 - 4). Additionally data of the years 1998 – 2000 are shown in Figure 1. In 1997, there were no extreme features during the vegetation period but both August and September 1997 were drier months than the average of the last 30 years (65 and 47 mm) (Table 2; Appendix, Table 1).

The weather conditions of the year 1998 were much different to the long term means most of the time (Figure 1, Appendix Table 2). In January and February 1998 moderate temperatures with a minimum of –12.9 °C (02. Jan.) near the soil and minimum temperature of –11.2 °C in 2 m height (01. Jan.) were measured. The mean air temperatures of January and February were 3.5 °C respectively 5.0 °C were higher than the mean temperatures of the last 30 years. Nevertheless the mean temperatures during the vegetation period were lower than the 30 year mean, the mean temperature of the whole year 1998 was 0,8 °C higher because of the warm months January and February. The cold summer was additionally characterised by high precipitation which was nearly 20 % higher than the mean of the last 30 years and a reduced

sunshine duration of more than 11 % (Figure 1, Appendix Table 2). Only in August some irrigation of 15 mm was necessary.

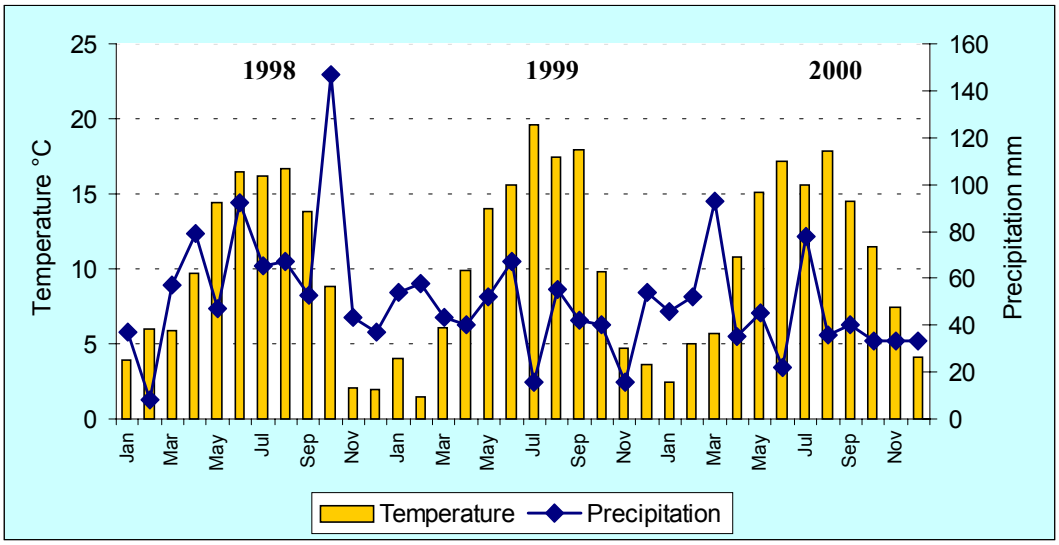


Figure 1: Weather conditions at the field site in Braunschweig during the years 1998 - 2000

Table 2: Comparisons of the major weather components in the years 1997 - 2000

	1997	1998	1999	2000
Air-Temperature °C	9,5	9,7	10,4	10,6
Sunshine duration h	1820	1342	1730	1569
Solar radiation J/cm2	1067	930	1085	997
Precipitation mm	587	732	536	544
Evaporation mm	624	517	689	637
Difference mm	-37	+215	-153	-93

Also in the year 1999 the weather conditions differed widely from the long term means (Figure 1, Appendix Table 3). In January and February 1998 moderate air temperatures with a minimum of –10.4 °C in February (Appendix Table 3). Lowest temperature near the soil was measured with –16.5 °C in February. The mean air temperatures of January 1999 and February 1999 were 4.0 °C respectively 1,5 °C. February 1999 was colder than February 1998 with a difference of 4.5 °C of the mean temperature. The mean temperature of the whole year was 1.5 °C higher than the mean temperature of the 30 year mean. The summer was characterised by low precipitation and high evaporation. The driest month in 1999 was July with a loss of water of 120 mm/m^2. Therefore the field was irrigated with 50 mm in this

month. Nevertheless global solar radiation and temperatures were high during the summer growing of Arundo was limited obviously in July and August because of water stress.

The year 2000 showed quite normal weather conditions (Figure 1 and Appendix Table 4). Higher temperatures than the long term mean were measured in May and June which resulted in a quite high water deficit in these months. The vegetation period finished in November with the first night frost (Appendix table 4). For the period 1997 – 2000 very moist weather conditions were measured in 1998 and a very dry year was identified in 1999 (Table 2).

3.3 Field trials

The experiment was established in the field as randomised block design with three repetitions and 10 populations. The experimental plots were established with an area of 3 x 4 m respectively 12 m^2. The inner part of the plot is the area of experimental value with 4.8 m^2 and 12 plants (plant density is 2.5 plants/m^2). 18 marginal plants are surrounding the area of experimental value of every plot (Figure 2).

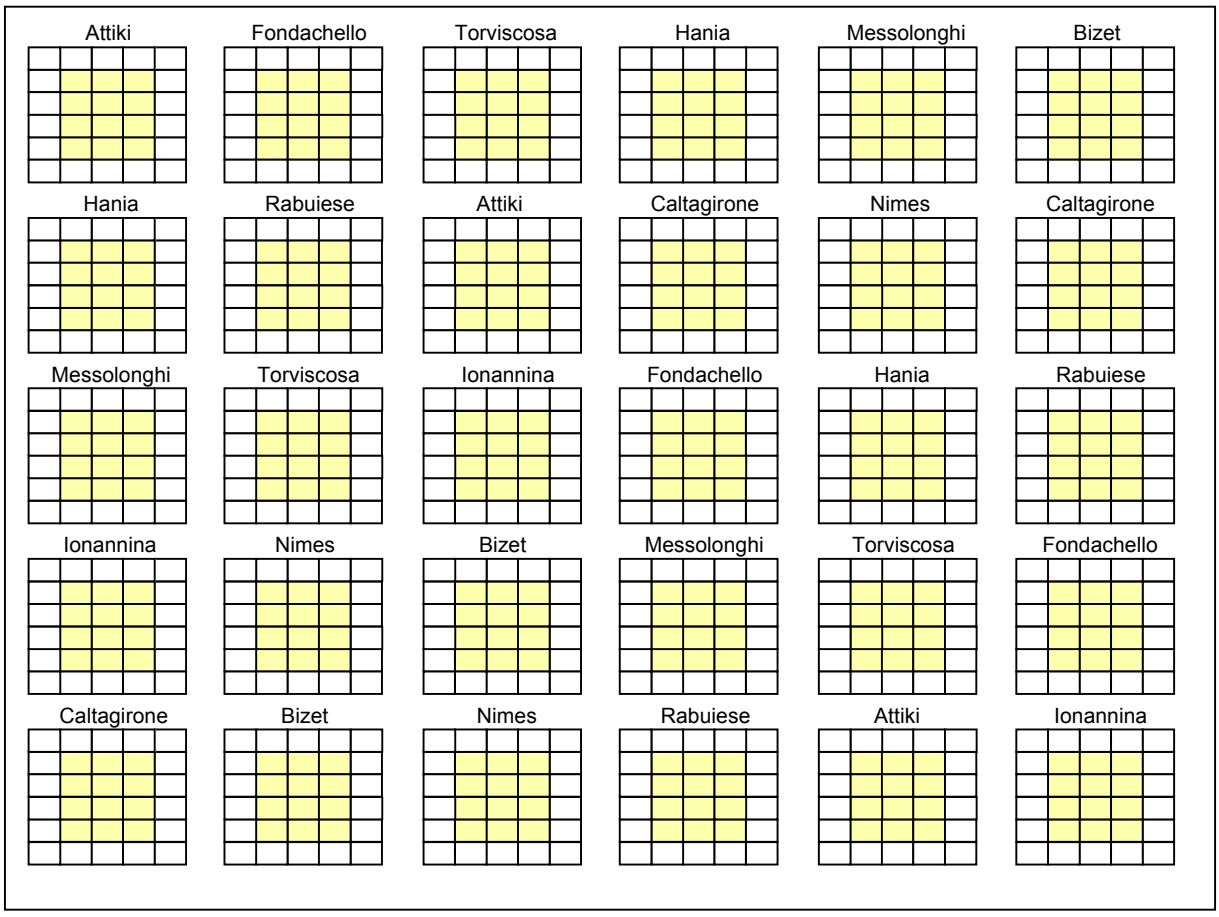


Figure 2: Field plan of the Arundo trials at FAL. Yellow marked plants are belonging to the area of experimental value (plot centre)

The total area of the experimental field is more than 1,600 m^2 (900 m^2 for adaptation examinations, 700 m^2 for additional tests by the FAL). The net experimental area is 144 m^2 as described in the technical annex of the project. Nitrogen fertilisation was applied as a solid ammonium/urea fertiliser deposit (BACHER, 1997). Every plant of *Arundo donax* in the field was given 5 g of nitrogen as deposit adjacent to the plant root. This is equivalent to 125 kg N/ha. The other nutrients in the soil were in sufficient supply.

During the experimental period several replanting of Arundo donax populations were necessary because of late delivery in the first year and limited survival during winter. Because of the two ages of Arundo plants, population planted in 1997 respectively 1998 were monitored separately in all years for the plot centre (12 plants). Additionally in 1999 further measurements were undertaken with 4 single marked plants for each establishment year chosen across the whole plot (Figure 3).

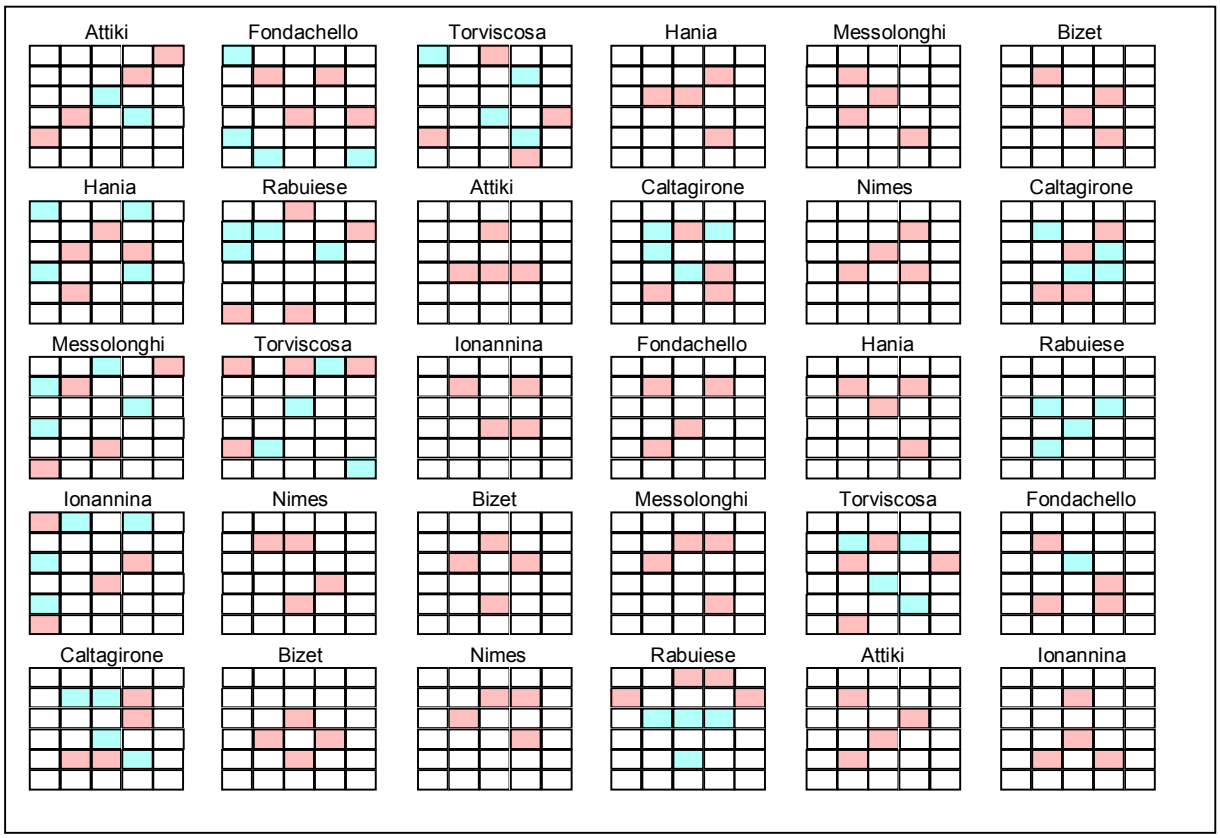


Figure 3: Additional measurements of Arundo donax planted in 1997 (blue) and 1998 (red) chosen across the whole plot in the year 1999

Measurements in the plot centres are obtained from the average of three replications (i.e. 3 x 12 plants), the results from additional measurements in 1999 were calculated as average from 12 selected plants for each population.

The Arundo donax plants were delivered from project partners in Greece, Italy and France.

Table 3: Arundo donax plants delivered from project partners

Location	Institution	Country
Attiki	CRES	South Greece
Hania	CRES	Crete Greece
Messolonghi	AUA	Central Greece
Ionannina	AUA	North Greece
Caltagirone	CETA/IAGCE	South Italy, Sicily
Fondachello	CETA/IAGCE	South Italy
Rabuise	CETA	North-East Italy
Torviscosa	CETA	North-East Italy
Nimes	INRA	South France
Bizet	INRA	South France

3.4 Establishment of *Arundo donax* stem-cuttings and rhizomes in the greenhouse

The Giant Reed genotypes were planted as described in the report of the Kick-off meeting in February 1997. Immediately after the delivery of the stem-cuttings and the rhizomes from the Southern-European partners CRES, AUA, CETA they were planted in plastic bags (16 cm diameter) in the greenhouse. For the first several weeks greenhouse conditions were 20° C during the day and 16° C at night. Two weeks after the delivery and planting of the last stem-cuttings, the greenhouse temperature was increased to 24° C during the day and 20° C at night. The plants were irrigated twice a day. After sprouting, stem-cuttings and rhizomes which were pre-cultivated in the greenhouse were transplanted into the field. Rhizome of the French partner INRA were delivered very late in November 1997 and were planted in plastic bags (10 l) in the greenhouse over the winter.

Statistical analysis was not able due to different plant ages and inhomogene plant numbers within the plot centres due to failures. Due to this fact only variation and standard deviation was given in the tables in the Appendix.

4 Results

In 1997 many work was done for establishment of delivered plant and rhizome material in the greenhouse. First planting actions in the plot centre did not happen before 25.6.1997. Therefore the vegetation period was quite short. Only the sprouting of Arundo donax as well as the number of tillers were monitored. No yield could be determined from the plot centres in the first year 1997 as there were to few plants in the plot centres.

4.1 Establishment of stem cuttings and rhizomes in the greenhouse and in the field in 1997

In order to test the best establishment of Arundo donax plantation the sprouting of stem cuttings as well as of rhizome material was grown in the greenhouse. The delivery of stem-cuttings of *Arundo donax* from South-European partners started in April 1997. First growth of the buds of stem-cuttings was observed 12-16 days after planting in plastic bags, other buds needed about three or four weeks. The stem-cuttings had an unsatisfactory sprouting capacity (Table 4) between 0 % (population Attiki) and 33 % (population Fondachello).

Table 4: Sprouting capacity of stem-cuttings of different *Arundo donax* populations established in 16 cm plastic bags under greenhouse conditions

Population	Sprouting capacity
Attiki	0 %
Hania	14 %
Messolongi	2 %
Ioannina	13 %
Caltagirone	15 %
Fondachello	33 %
Rabuiese	6 %
Torviscosa	15 %

Sprouting capacity of the delivered rhizome material (6-12 rhizomes were additionally delivered with stem-cuttings) was about 30 %. After adaptation of plants to field conditions sufficient sprouted stems and rhizomes were planted in the field outside of the plot centre (Table 5). The sprouting of late delivered rhizome material from INRA in November (Nimes and Bizet) was much better with 95%.

Table 5: Number of stem-cuttings/rhizomes planted outside the plot centre as marginal plants on 19.6.1997

Population	Number of plants
Attiki	9
Hania	10
Messolongi	9
Ioannina	9
Caltagirone	4
Fondachello	13
Rabuiese	15
Torviscosa	20

4.2 Plant development in the field in 1997

The plot centres were planted with newly delivered rhizomes of the populations on 25th June (Attiki and Hania), 1st July (Messolonghi and Ionannina), 2nd July (Rabuise and Torviscosa) and 21th July (Caltagirone and Fondachello). The sprouting capacity was recorded on four dates in August and September (Table 6). Rhizomes of populations Rabuiese and Torviscosa had the best sprouting capacity in the field with nearly 95 %.

Table 6: Sprouting of Arundo donax populations in the plot centre (36 rhizomes in 3 repetitions)

	Counting dates				Sprouting capacity
Population	4.8.1997	13.8.1997	12.9.1997	30.9.1997	
Attiki	2	2	2	2	6 %
Hania	1	1	1	1	3 %
Messolonghi	1	1	1	1	3 %
Ionannina	0	0	0	0	0
Caltagirone	3	11	15	15	42 %
Fondachello	0	9	10	10	28 %
Rabuise	31	33	33	33	92 %
Torviscosa	33	34	34	34	94 %

Because of the insufficient sprouting capacity of rhizomes of populations Attiki, Hania, Messolonghi, Ionannina, Caltagirone and Fondachello additional rhizomes were delivered and planted into the field in spring 1998.

In 1997 the average tiller number of all plants in the plots were counted and the results are shown in Table 7.

Table 7: Average number of tillers/plant of field established *Arundo donax* stem-cuttings and rhizomes (winter 1997)

Population	Marginal plants (rhizomes and cuttings)	Plants in the plot centre (rhizomes)
Attiki	9	2 – 4
Hania	10	1 – 2
Messolongi	9	1 – 3
Ioannina	13	1 – 3
Caltagirone	11	1 – 2
Fondachello	13	1 – 2
Rabuiese	12	3 – 8
Torviscosa	11	3 – 6

The growth of *Arundo donax* was abruptly interrupted by frost temperature when all parts of the plants were green (24.10.–26.10.1997). Nevertheless, *Arundo* of rhizomes and stem-cuttings delivered in April reached an average height of more than 1.7 m in the first year.
Some plants of population Ionannina grew to more than 3.2 m high at the end of September. Also several plants of populations Attiki and Hania reached a height of nearly 2.8 m. The average of heights of the marginal plants of all plots is shown in Figure 4.

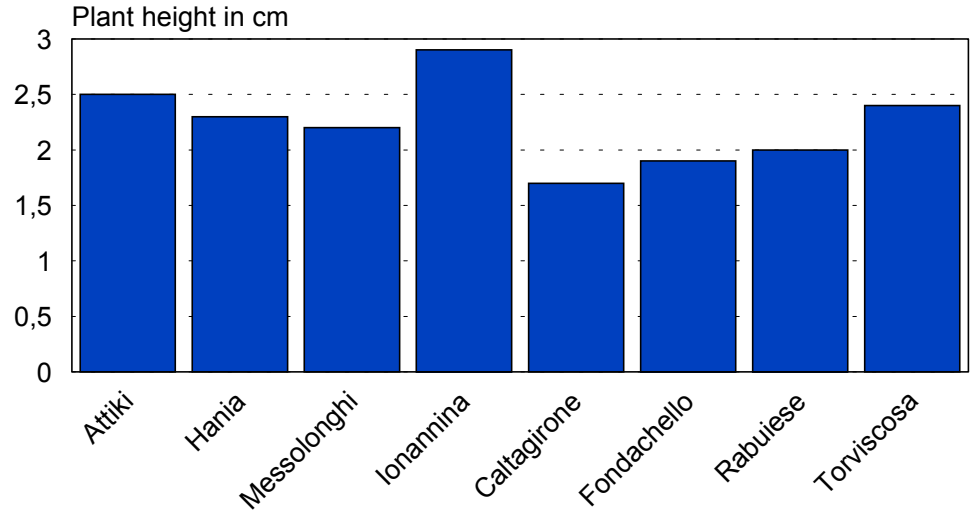


Figure 4: Average height of Arundo donax populations planted in marginal areas of the plots in 1997

4.3 Plant growth and monitoring results in the years 1998 – 2000

4.3.1 Re-sprouting and winter survival during the years 1998 and 2000

During spring 1998 the sprouting of Arundo donax was excellent. None of the Arundo plants established in 1997 was lost because of winter killing. This might be an effect of moderate temperatures during January and February 1998. First sprouts of Arundo genotypes were visible in the first week of April (06.04.1998), one week earlier than the sprouting of Miscanthus giganteus at Braunschweig site. During the vegetation period in 1998 new rhizomes of populations Attiki, Hannia, Messolonghi, Ioannina, Caltagirone and Fondachello were planted in June into gaps within the centres of the plots.

The resprouting of all Arundo genotypes in spring 1999 was nearly three weeks later than in spring 1998. None of the Arundo plants established in 1997 was lost because of winter killing during the first (97/98) and second winter (98/99), but only a small number of 1998 planted Arundo was lost during winter 98/99 (Table 8). Winter survival rate of in 1998 established Arundo of all populations was about 93 %.

During the winter 1999/2000 also several rhizomes of Arundo donax populations planted in 1997 died back in the centre of the plots (Table 8). After three years the percentage of all plants in the plot centres surviving cold winter conditions decreased from Torviscosa (100 %), Messolonghi and Nimes (94 %), Ionannina and Bizet (92 %), Attiki and Caltagirone and Rabuise (89 %), Hania (86 %) to Fondachello (83 %).

Table 8: Numbers of 1997 and 1998 planted Arundo donax lost during winter 1998/99 and 1999/2000

Population	Winter 1998/99	Winter 1999/2000	
	Arundo planted 1998	Arundo planted 1997	Arundo planted 1998
Attiki	94 %	100 %	91 %
Hania	97 %	100 %	90 %
Messolonghi	100 %	100 %	94 %
Ionannina	100 %	Not planted	92 %
Caltagirone	95 %	93 %	86 %
Fondachello	93 %	83 %	86 %
Rabuise	100 % (1997)	89 %	Not planted
Torviscosa	100 %	100 %	100 %
Nimes	94 %	Not planted	92 %
Bizet	100 %	Not planted	94 %

4.3.2 Growth development during the vegetation periods 1998, 1999 and 2000

The growth height of Arundo donax populations was monitored every 4 weeks during the vegetation period separately for plants planted in 1997 and 1998. A mean was calculated for both plant ages and presented for the year 1998 in Figure 5.

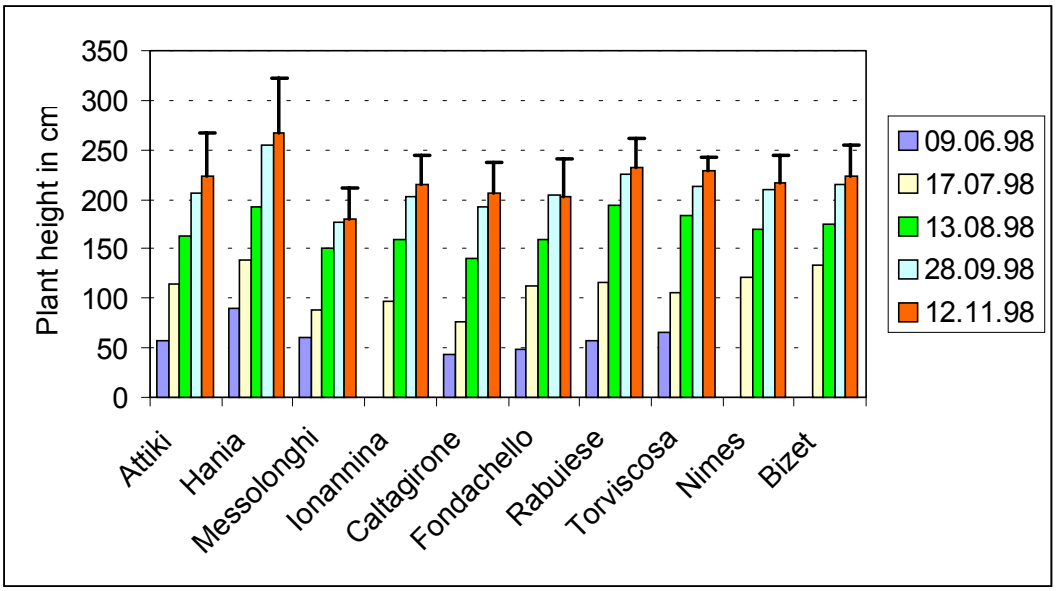


Figure 5: Average height of Arundo donax population grown in 1998 and maximum height (Average of 3 replications, plot centre)

The maximum plant height measured in November separated for plant age is given in Figure 6.

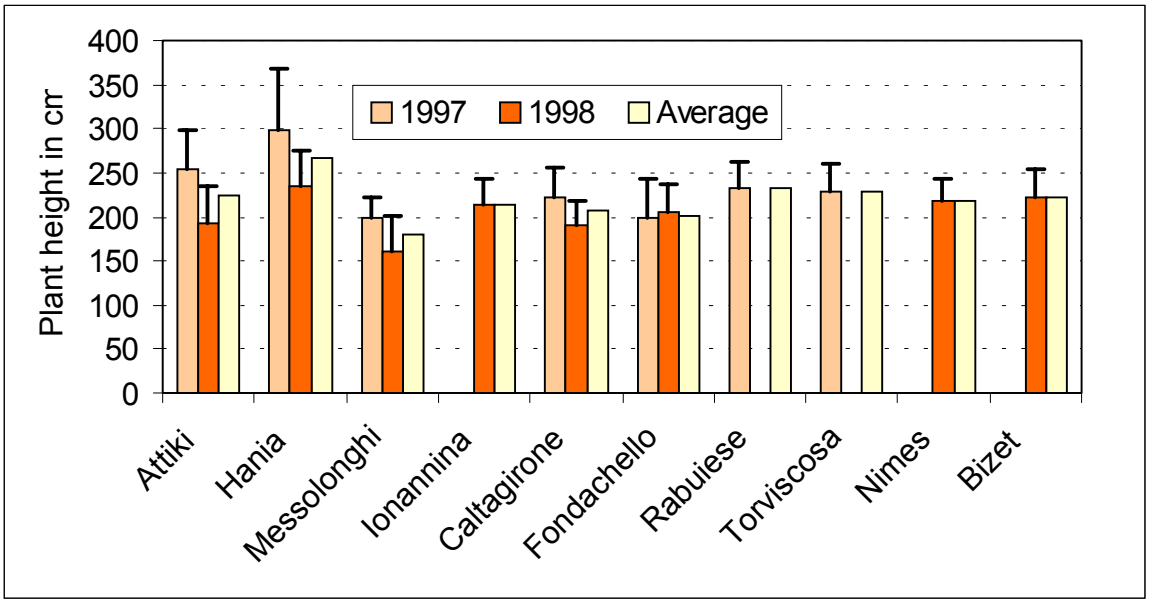


Figure 6: Height of Arundo donax population established in 1997 and 1998 in the year 1998 (Average and maximum of 3 replications and average of all plants)

The average height of Arundo donax population Hania for the whole plot was 267 cm. Arundo donax of population Hania planted 1997 and 1998 reached maximum heights of 369 respective 275 cm (average of all plots). Some single plants reached more than 4 m height (Figure 7). Older plants were on average higher than Arundo donax planted in 1998 (Figure 6 and Appendix Figures 1 - 4). Smallest plants were observed for population Messolonghi. For every population and growth year detailed figures about shoot length for Arundo populations planted in 1997 and 1998 are given in the Appendix (Figures 1 - 4).

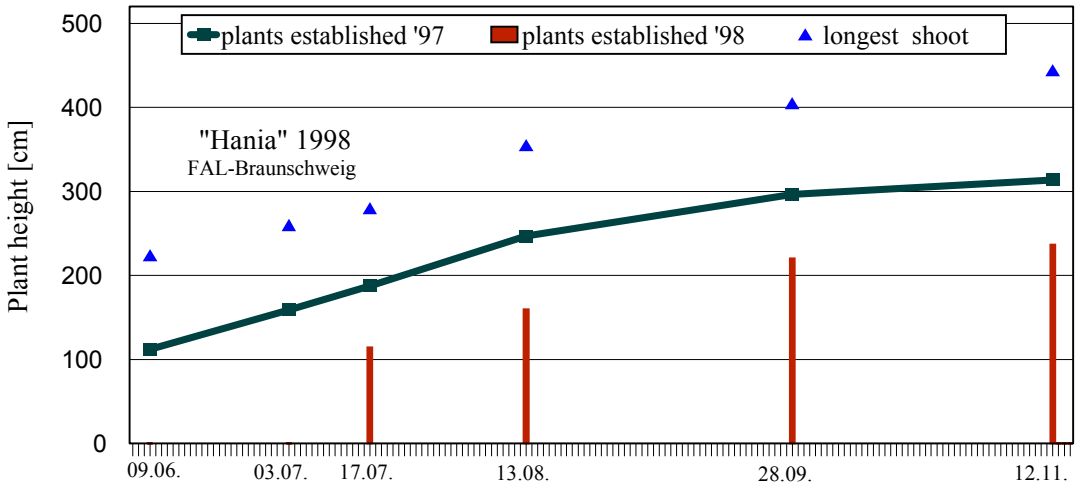


Figure 7: Plant height of population Hania in the year 1998 (Average of plants established in 1997 and 1998 as well as longest shoot)

Arundo donax grew continuously till the beginning of July with growth rates up to 2-3 cm/day. At the first measurement (09.06.) the Greek populations Hania, Messolongi and Ioannina established in 1997 reached a mean height of 100 cm and longest shoots heights of 200 cm (figure 7 and Appendix, Figures 1-4). All other genotypes reached heights between less than 50 cm (Caltagirone) and more than 80 cm (Torviscosa) (Appendix, Figures 2 and 3). Cold temperatures had a growth-retarding effect of all genotypes during June 1998. Growth rates slowed down between August and September.

The mean heights of Arundo donax planted in 1998 were in the average 80 cm less than the mean heights of the one year earlier established plants with the exception of the genotypes "Caltagirone" and "Fondachello" with a less height of 20 cm respectively 50 cm. The first frost of autumn 1998 occurred at 22nd of November. The vegetative phase of Arundo genotypes was finished without producing flowers.

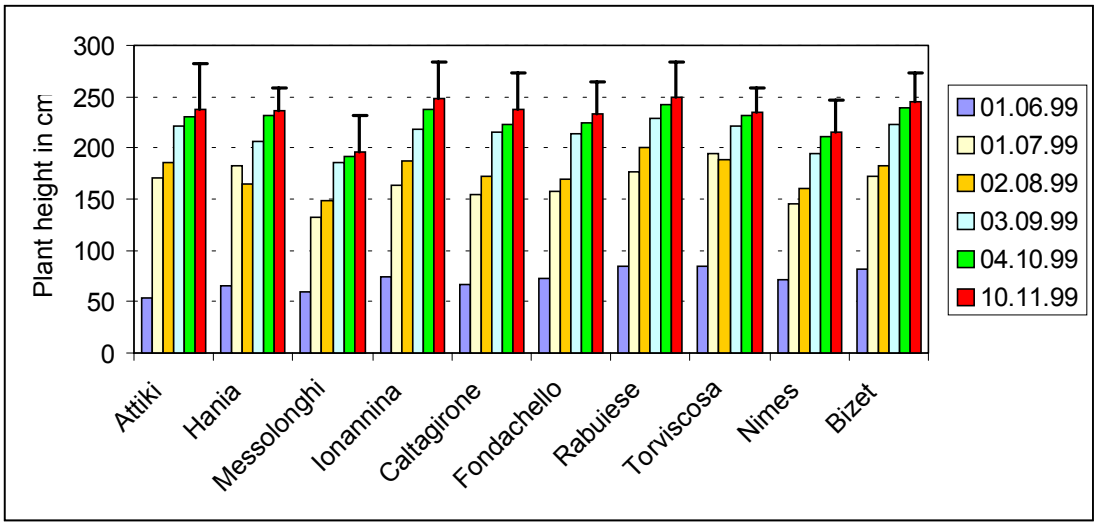


Figure 7: Average height of Arundo donax population grown in 1999 and maximum height (Average of 3 replications, plot centre)

In 1999 the average height measured over the whole plots was 250 cm in November (Figure 7). The maximum height was on average about 283 cm for populations Attiki, Ionannina and Rabuise. Single plants in all populations reached more than 3 m (Figure 7 and Appendix, Figures 5 - 14).

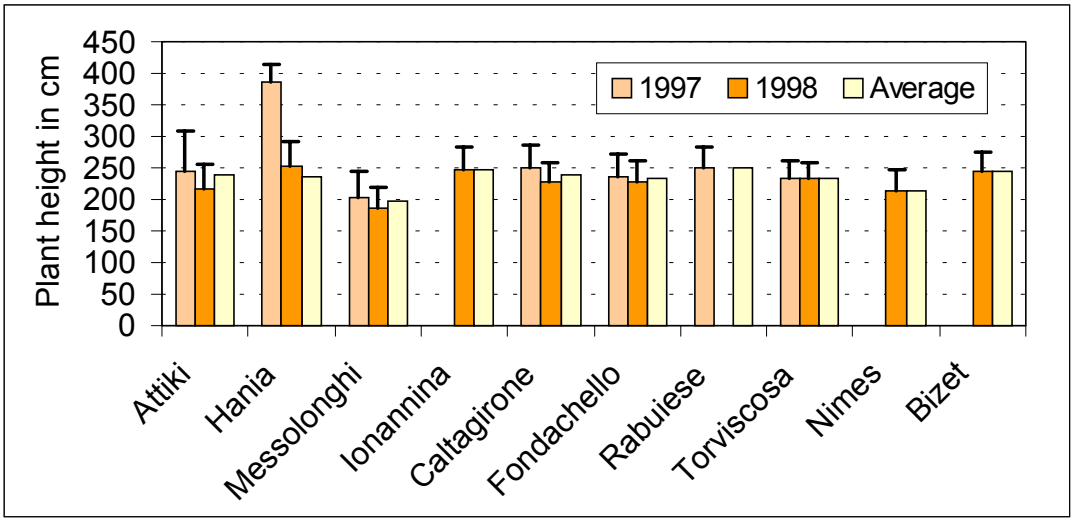


Figure 8: Height of Arundo donax population in the year 1999 separated between Plants established in 1997 and 1998 (Average of 3 replications)

Also in 1999 Arundo donax planted in 1997 reached larger heights than plants of the year 1998 (Figure 8). For Hania heights of more than 4 m were measured for plants established in 1997. Detailed growth data separated between the planting years are given in the Appendix (Figures 5 – 14). Additionally measurements were carried out for single plant chosen across the whole plot (Appendix Figure 5 - 14). For population Hania growth data are displayed in Figure 9 and 10)

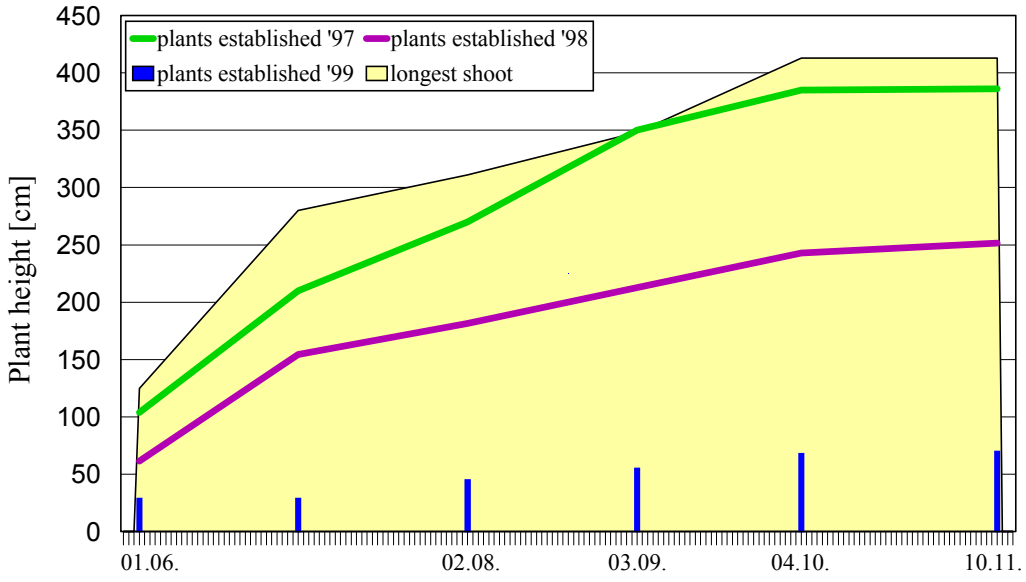


Figure 9: Growth of plants established in 1997, 1998 and 1999 of Arundo donax, population Hania in the year 1999 (plot centre, average of 3 replications)

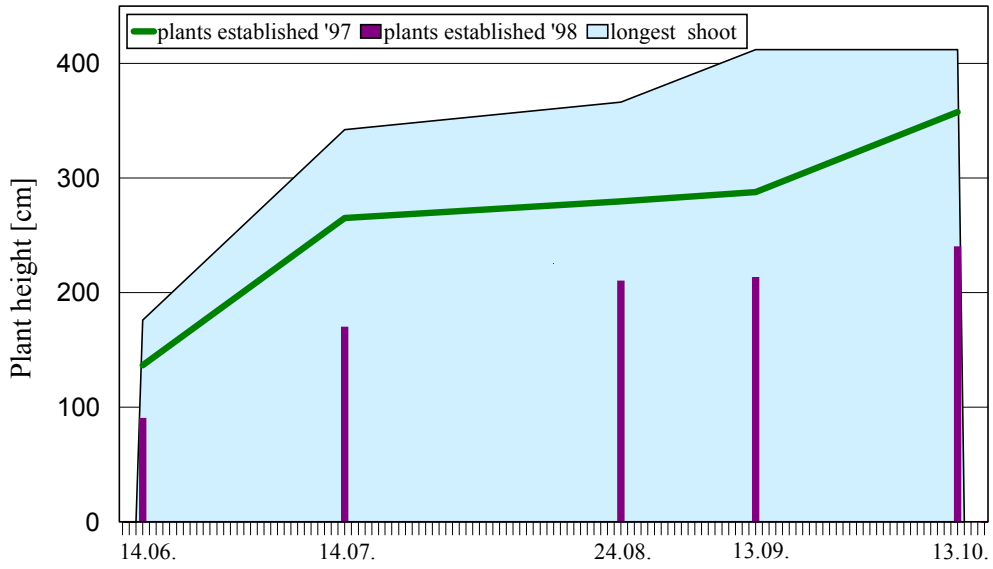


Figure 10: Selected Arundo donax plants of population Hania (average of 4 plants selected and marked across the whole plot)

Only minor differences of growth rates and height were detected when measurements obtained in the plot centre were compared with those of 4 selected plants chosen across the whole plot size (Figure 9 and 10; Appendix Figures 5-14).

In 1999 re-sprouting of Arundo started nearly 3 weeks later than in 1998 like the other grasses too. At the first measurement in June plants established in 1997 of all genotypes reached a mean height of between 60 cm and 100 cm and longest shoots heights of 150 cm and more (Appendix Figures 5 – 14). Because of the negative water-balance (beginning in April/May) with a maximum in July the growth off all Arundo genotypes stagnated during July and August (Figure 7). After this period growth rates increased again and all populations reached maximum heights of more than 3 meters in October and November. The vegetative phase of Arundo genotypes was finished without producing flowers.

In the year 2000 Arundo donax grew continuously until August. The growth rates slowed down from September to November and the growth period finished in November with the first Frost (Figure 11). As in the years before plants did not flower in this year. The average height was for population Hania in November 249 cm with maximum at 307 cm. Smallest populations in 2000 were Messolonghi (average height 166 cm in November) and Nimes (154 cm) (Figure 11).

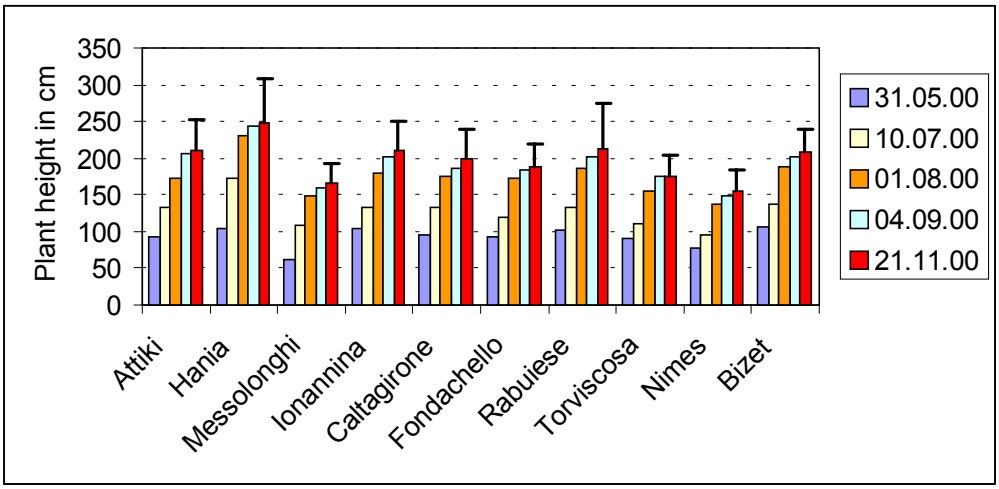


Figure 11: Average height of Arundo donax population grown in 1999 and maximum height (Average of 3 replications, plot centre)

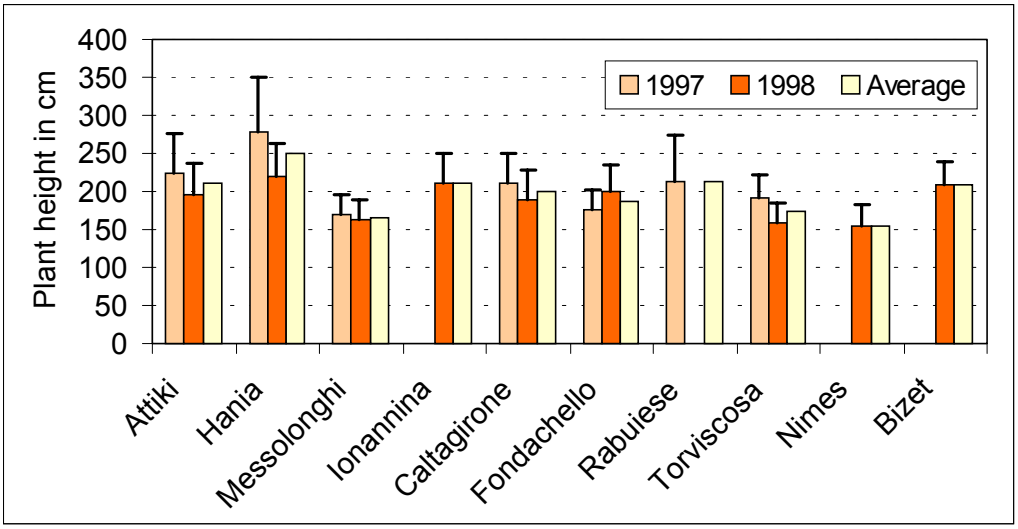


Figure 12: Height of Arundo donax population established in 1997 and 1998 grown in 1999 and maximum height (Average from 3 replications, whole plot centre)

Also in 2000 plants established in 1997 reached on average heights 10 – 50 cm higher than plants established in 1998. The maximum height on average was reached from population Hania of plants established in 1997 (349 cm high) (Figure 12). Plant growth decreased when compared with the year before. Further data of all populations are given in the Appendix (Figures 15-18). As an example data of average plant height for Arundo donax population Hania for the vegetation period 2000 is given in Figure 13.

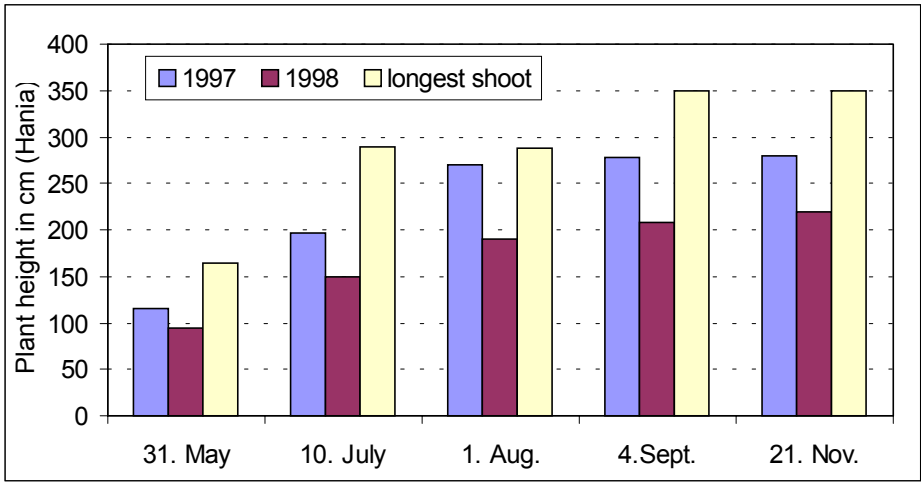


Figure 13: Height of Arundo donax population Hania in the year 2000
(average of plants established in 1997 and 1998, longest shoot, 3 replications)

4.3.3 Number of shoots of Arundo donax populations in the years 1998 – 2000

The number of shoots in the year 1998 were counted in June, July and August (Table 9). At the first date of measurement plants of the genotypes "Hania", "Messolongi", "Ioannina" and "Fondachello" produced between 11 and 13 shoots per plant and the genotypes "Caltagirone", "Rabuiese", and "Torviscosa" between 5 and 9. The number of shoots per plant increased continuously between the first and the third counting date. The increase of number of shoots between the first and the second date was higher than the increase between the second and the third date. In the year 1998 established plants showed between 3 and 4 shoots per plant in July and between 4 and 6 in August. The production rate of new shoots decreased throughout the remaining growing season but was not finished before the first frost.

Table 9: Number of shoots per plant of the 1997 and 1998 established Arundo populations in the year 1998

Genotype	12.06.98		03.07.98		19.08.98	
	97	98	97	98	97	98
Attiki	5	-	7	3	10	5
Hania	13	-	16	4	19	5
Messolongi	13	-	17	4	17	6
Ioannina	11	-	19	3	21	5
Caltagirone	5	-	5	3	9	6
Fondachello	11	-	12	3	13	5
Rabuiese	8	-	11		12	
Torviscosa	9	-	12	4	13	5
Nimes	-	-	-	4	-	6
Bizet	-	-	-	3	-	6

Similar results were obtained in the following years (Appendix, Table 5 and 6). In Table 10 the years 1999 and 2000 tiller numbers obtained at the end of the vegetation period are shown.

Table 10: Shoot numbers of Arundo donax populations planted in 1997 and 1998 for the years 1999 and 2000

Population	8. 11. 1999	8. 11. 1999	21. 11. 2000	21. 11. 2000
	1997	1998	1997	1998
Attiki	15	7	18	7
Hania	15	7	12	6
Messolonghi	13	8	15	8
Ionannina		8		8
Caltagirone	11	7	9	6
Fondachello	8	7	8	7
Rabuise	11		13	
Torviscosa	10	5	11	8
Nimes		10		7
Bizet		10		11

The number of shoots was higher in populations established in 1997 than in those established in 1998. No significant difference between the years was observed (Table 10). Highest tiller number was produced from population Attiki and Hania. Plants established in 1997 were not recorded in Population Ionannina in the years 1999 and 2000.

4.3.4 Shoot diameter of Arundo donax populations in the years 1998 and 2000

The shoot diameters of plants established in 1997 and 1998 were measured several times during the vegetation period. Measurements took place in a height of 10-15 cm from soil among two basal knots. In August 1998 all in 1997 established Arundo populations had shoot diameters of more than 2 cm. Shoot diameters of in 1998 established populations were between 1,5 cm ("Torviscosa") and 2,2 cm ("Nimes") (Figure 14 and Appendix Figure 19).

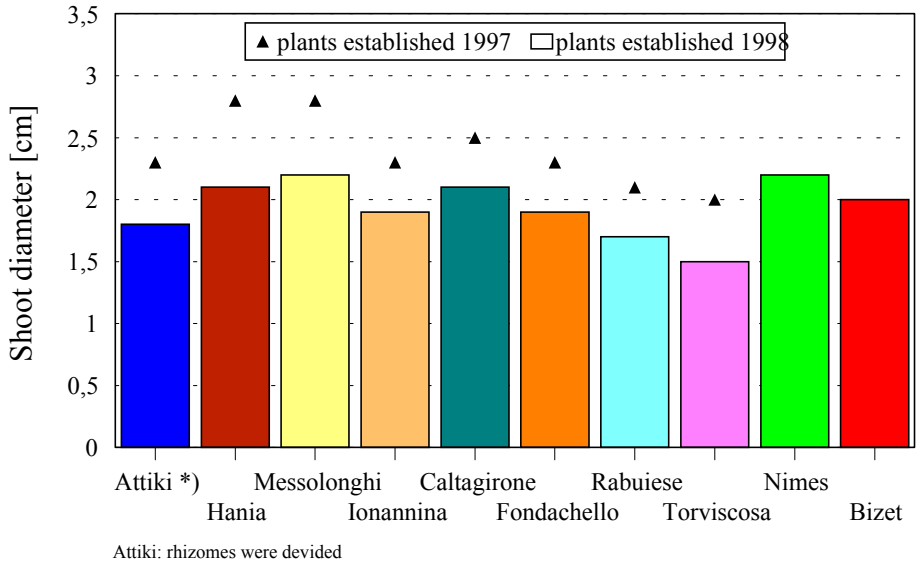


Figure 14: Shoot diameter of Arundo donax populations on 13th August 1998

The shoot diameter measured at the end of the vegetation period in the years 1999 and 200 is given in Table 11)

Table 11: Shoot diameter in cm of Arundo donax populations established in 1997 and 1999 at the end of the vegetation period in 1999 and 2000 (average of 3 replications)

Population	8. 11. 1999	8. 11. 1999	21. 11. 2000	21. 11. 2000
	1997	1998	1997	1998
Attiki	2,5	2,0	2,6	1,9
Hania	2,5	2,0		2,0
Messolonghi	2,3	2,1		2,2
Ionannina		2,0		2,0
Caltagirone	2,5	2,2	2,4	2,4
Fondachello	2,2	1,8	1,7	2,0
Rabuise	2,1		2,0	
Torviscosa	1,9	1,8	1,9	1,7
Nimes		2,0		1,9
Bizet		1,9		1,8

No significant differences in shoot diameter between both years were observed. Maximum shoot diameter was measured with 2,7 cm for shoots of population Hania in 1999 and 2,6 cm for Attiki and Caltagirone in 2000. Also shoot diameter increased during the vegetation period (Appendix, Table 7 and 8). At the end of the vegetation period the average shoot diameter of Arundo donax populations established in 1997 had with 2,3 cm (1999) and 2,1 cm (2000) slightly greater diameter than those established in 1998 with 2 cm (1999 and 2000).

4.3.5 Leaf number counted in the years 1999 and 2000

The leaf number was counted on single marked plants (4 plants for each establishment year) in the plot centre as well as from marginal plants. In 1999 both the plant height as well as the leaf number of marked stem was recorded (Figure 15). The leaf number increased during the vegetation periods continuously until Frost in autumn 1999.

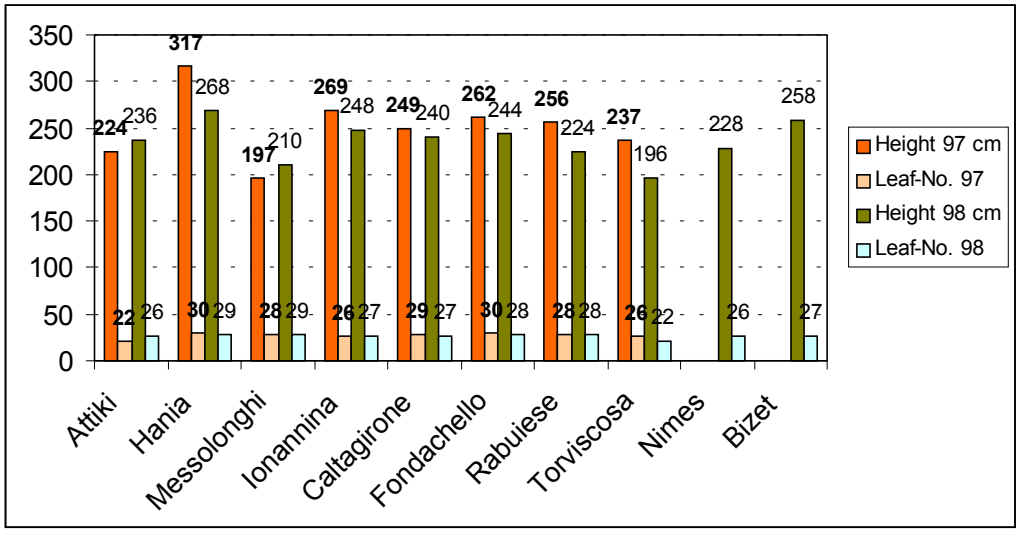


Figure 15: Stem height and leaf number of Arundo donax populations established in 1997 and 1998 (average of 8 plants in the plot centre in October 1999)

The population Hania had longest stems up to 317 cm and the highest leaf number of 30 leaves for plants established in 1997 (Figure 15). Also population Fondachello had 30 leaves but the stem length was with 262 cm shorter than Hania. All populations established in 1998 reached lower heights and less leaves than those established in 1997. The leaf number of plants established in 1998 decreased from 29 (Hania, Messolonghi) to 22 (Torviscosa).

The average leaf number for both plant ages was between 29 and 24 leaves/plant (Figure 16). The length of internodes was longest in populations of Hania and Ionannina the shortest was found in population Messolonghi (Figure 16).

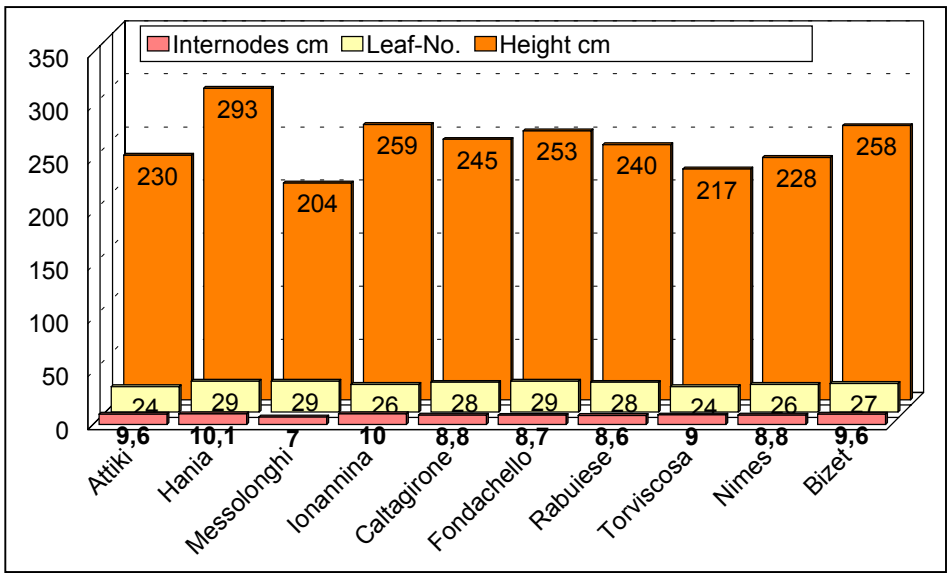


Figure 16: Average plant height, leaf number and internode length of Arundo donax populations in October 1999 (whole plot centre)

In the year 2000 four plants were chosen across the whole plot for counting leaf number. The plant was similar to height displayed in Figure 12 (2000).

Table 12: Leaf number of 4 selected Arundo donax plants in the year 2000

Population	16.6.2000	16.6.2000	16.6.2000	21.11.2000	21.11.2000	21.11.2000
	1997	1998	Average	1997	1998	Average
Attiki	13	12	12	23	20	22
Hania		13	13		24	24
Messolonghi		13	13		23	23
Ionannina		13	13		22	22
Caltagirone	15	14	15	25	22	23
Fondachello	12	13	12	19	21	20
Rabuise	14		14	25		25
Torviscosa	13	14	14	22	23	23
Nimes		11	11		21	21
Bizet		13	13		21	21

Maximum leaf number was reached with 25 in population Caltagirone and Rabuise at the end of the vegetation period in November 2000 (Table 12). This was less than in the year before. No differences between populations ages were recognised in 2000. The leaf number increased

until August 2000. Only up to 2 further leaves appeared until November 2000 (Figure 17 and Appendix, Table 8).

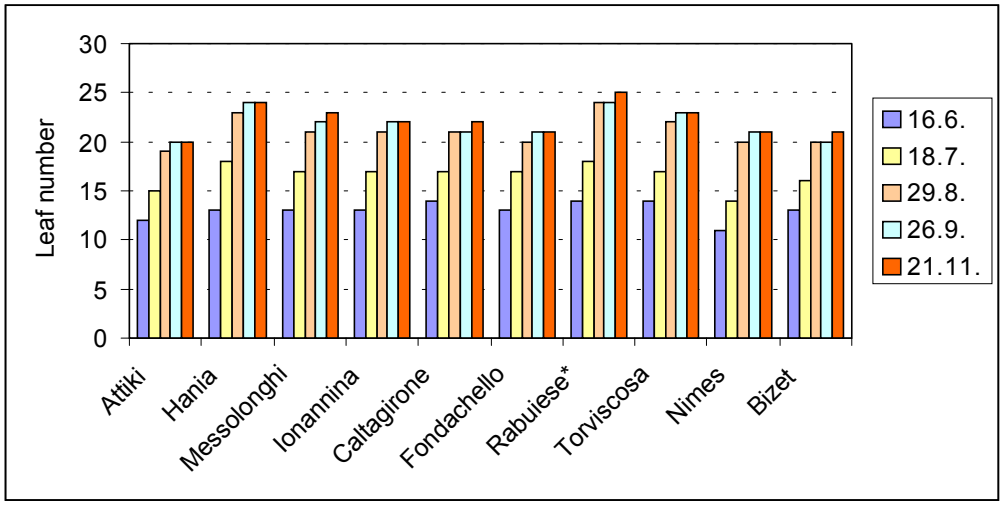


Figure 17: Leaf number of Arundo donax established in 1998 in the year 2000
(4 plants selected across the whole plot, *Rabuise: established in 1997)

4.3.6 Fresh and dry matter yield of Arundo donax in the years 1998 - 2000

The Arundo donax populations were harvested on 1th January 1999; 6th January 2000 and 16th January 2001 and fresh matter, dry matter yield as well as dry matter content were determined.

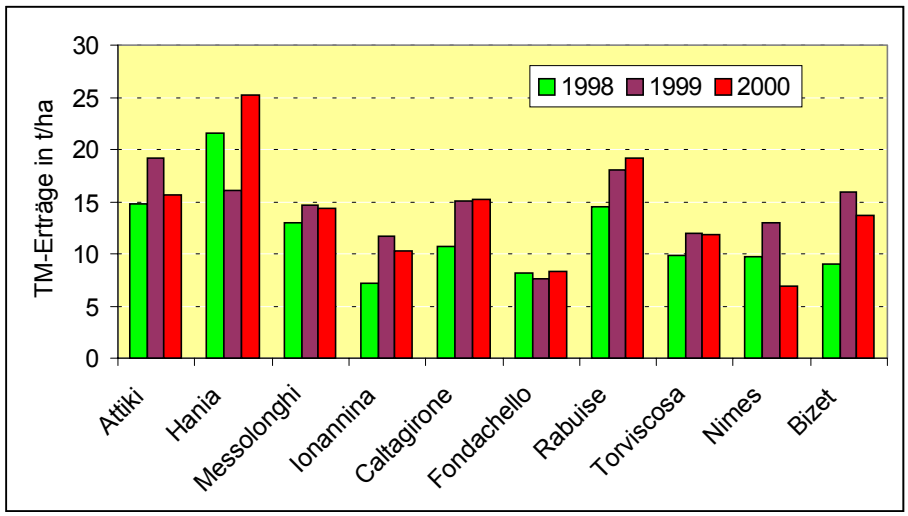


Figure 18: Average dry matter yield in the years 1998 and 2000 for Arundo populations
(plot centre, 3 replications)

The average dry matter yield was highest for population Hania but also for Attiki and Rabuise (Figure 18). During the years 1998 - 2000 a large yield variation between 5 and 9 t/ha was observed. While in the first (1998) and third year (2000) Hania reached highest yield, in the colder second year (1998) Attiki had the highest yield. Cold weather conditions in 1998 obviously decreased the yield of population Hania and Fondachello. On average, population Fondachello showed the lowest yields in all years. During the years 1998 to 2000 there was an continuous yield increase measured in population Rabuise.

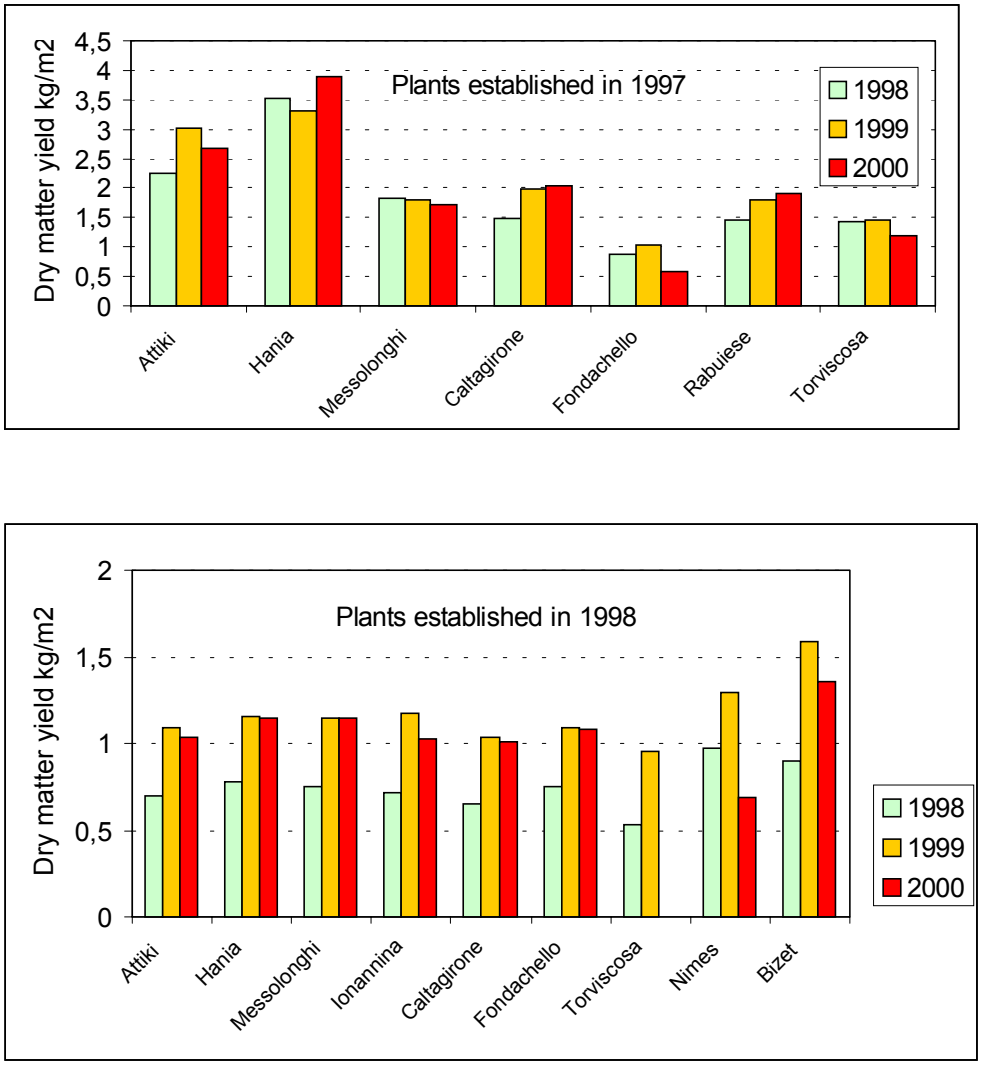


Figure 19: Dry matter yield measured in Arundo donax plants established in 1997 and 1998 (plot centre, 3 replications)

In the plot centre Arundo donax plant of different age were planted. The measured dry matter yield in kg/m^2 showed higher yield for Arundo donax established in 1997 compared with those established in 1998 (Figure 19). While there was a steady increase in dry matter

production in nearly all populations established in 1997, plants established in 1998 increased dry matter production only in population Bizet in the year 2000. Dry matter production decreased during the three years in population Messolonghi, Fondachello, Torviscosa (for plants established in 1997) and Nimes (plants established in 1998) (Figure 19).

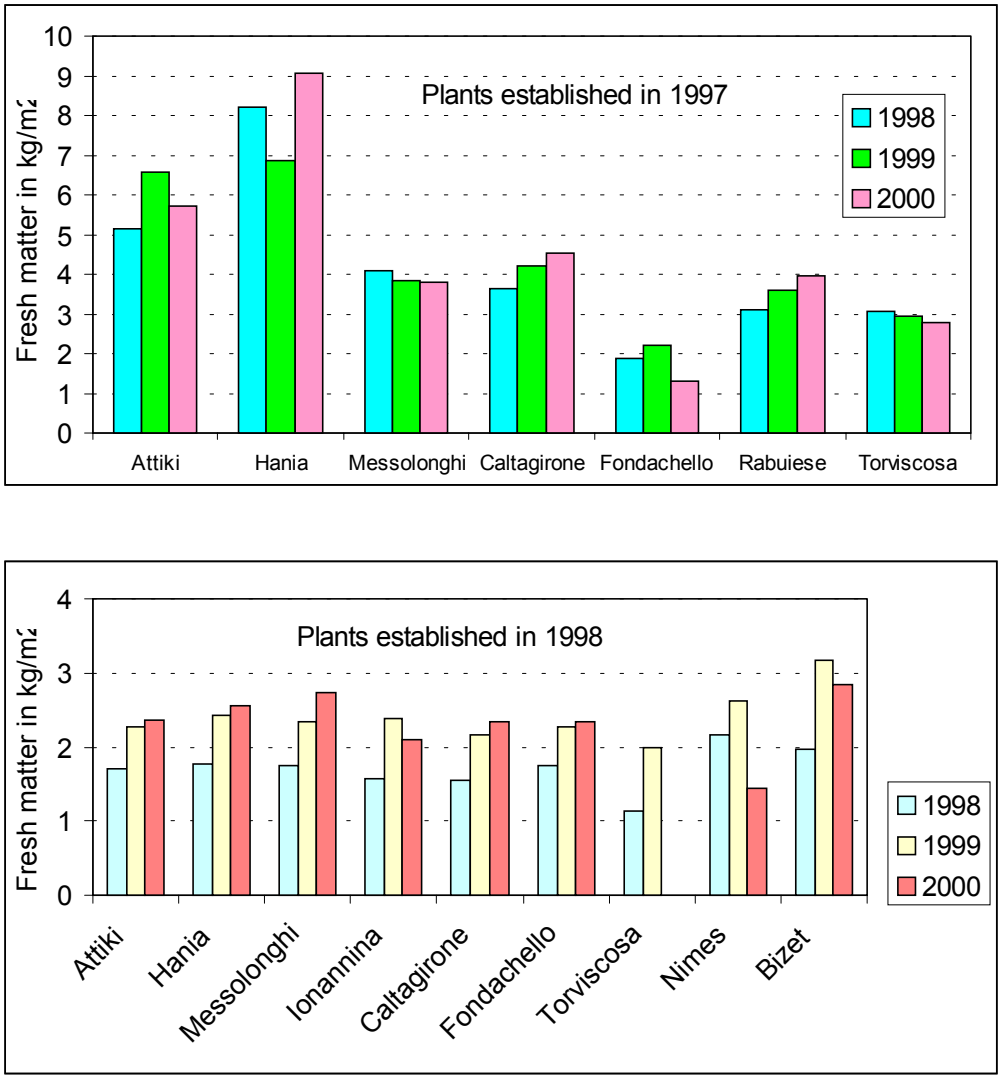


Figure 20: Fresh matter yield of Arundo donax populations in the years 1998 - 2000

The maximum fresh matter yield was reached form Population Hania (plants established in 1997) with 9 kg/m^2 (Figure 20). The variation in fresh matter yields was between 2 (Fondachello) and 6,5 kg/m^2 (Attiki) for the other populations for plants established in 1997. Highest fresh matter yield of plants established in 1998 was reached from population Bizet with 3 kg/m^2 in 1999.

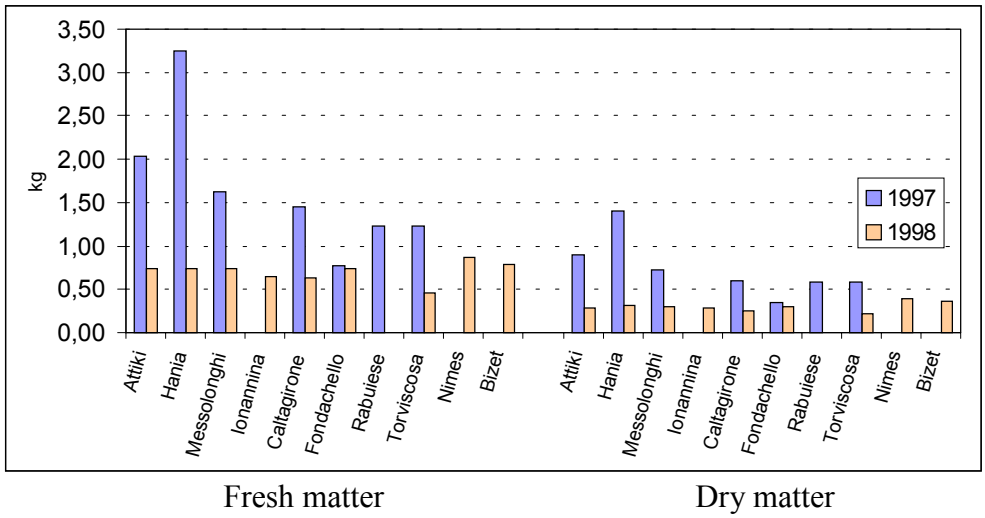


Figure 21: Fresh matter and dry matter weight of Arundo donax plants in the year 1998 (single plant weight of all plants in he plot centre)

The fresh matter weight of single plants decreased in 1998 from population Hania, Attiki, Messolonghi, Caltagirone, Rabuise, Torviscosa to Fondachello for plants established in 1997 (Figure 21). Only small differences in fresh matter weight were observed for plants established in 1998.

Table 13: Dry matter content of fresh biomass harvested in 1998, 1999 and 2000

Year	1998		1999		2000	
Pl. established	In 1997	in 1998	in 1997	in 1998	in 1997	in 1998
Population						
	%	%	%	%	%	%
Attiki	44	41	46	48	47	44
Hania	43	44	48	48	43	45
Messolonghi	45	43	47	49	45	42
Ionannina		46		49		49
Caltagirone	41	42	47	48	45	43
Fondachello	46	43	47	48	45	46
Rabuise	47		50		48	
Torviscosa	47	47	49	48	43	
Nimes		45		49		48
Bizet		46		50		48

The dry matter content of the fresh matter harvested in the years 1998, 1999 and 2000 varied between 41 and 50 % with a mean of 46 %. No differences between plants established in 1997 and 1998 were detected. In additionally experimental plots Arundo donax plants grown in the year 1999 were not harvested in January 2000. These plants survived the winter and produced

new shoots out of the leaf shoulders. During the vegetation period 2000 many plants died back. The dry matter content increased up to 71 % in these stems.

Due to reconstruction of the laboratory building of the institute only dry matter samples of the harvest in 1998 could be analysed for nitrogen, phosphorus, potassium, calcium and magnesium content (Table 14).

Table 14: Nutrient content of Arundo donax harvested in 1998

Population	N	P	K	Mg	Ca
	%	mg/ 100 g	mg/ 100 g	mg/ 100 g	mg/ 100 g
Planted 1997					
Attiki					
Hania					
Messolonghi					
Ionannina					
Caltagirone	1,6	149 (0,15 %)	1300 (1,3 %)	127 (0,13 %)	454 (0,45 %)
Fondachello	1,4	152	1700 (1,7 %)	98 (0,1 %)	432
Rabuise	1,4	151	1300	129	549 (0,55 %)
Torviscosa	1,3	140	1300	115	471
Nimes					
Bizet					
Planted 1998					
Attiki	1,5	172	1300	140	479
Hania	1,5	147	1400 (1,4 %)	107 (0,1 %)	448
Messolonghi	1,9	186 (0,19 %)	1400	147 (0,15 %)	542 (0,54 %)
Ionannina	1,7	149	1300	129	469
Caltagirone	1,9	176	1300	137	475
Fondachello	1,8	161	1300	131	500
Rabuise	1,4	134 (0,13 %)	1500	126	495
Torviscosa	1,5	158	1300	99	428 (0,43 %)
Nimes	1,6	145	1200 (1,2 %)	129	508
Bizet	1,5	142	1300	111	505

Because the plant material was relatively green when harvested, high content of nitrogen and potassium was found in the material. No differences between the plant ages were observed.

5. Discussion

During the research project adaptation, cultivation and biomass production of Arundo donax under nearly every climatic condition in Europe was evaluated. First results were obtained for cultivation under climatic conditions in northern Germany.

While Arundo donax is a seedless plant, it must be propagated by stem cuttings or rhizomes. The results of the first year clearly showed that propagation by stem cuttings in northern Germany was unsuccessful. Also propagation with rhizomes caused problems because of failures in sprouting. Similar problems due to failures in cultivation of Arundo donax rhizomes have also been reported from research trials in southern Germany (Oster and Schweiger, 1992). Also in those research trials several replanting actions with Arundo donax rhizomes were necessary. Therefore very inhomogene stock of plants appeared in the plots.

The winter survival of rhizomes established in 1997 during winter 1997/98 was excellent. Survival rate decreased during the years 1998 - 2000. While during the first winter 1998/99 between 0 and 7 % of all plants did not survive, this number increased in the second winter 1999/2000 to 6 - 14 %. This failures were higher than results reported from Miscanthus giantess rhizomes planted at the same site with 0,1 - 1,2 % between the years 1989 and 1998 (El Bassam et al., 2000) but proportion of failures can increase up to 10 % on clay soils (Schwarz et al., 1995). In southern Germany, Arundo donax plant losses between 10 and 25 % have been reported for the years 1989-1991 (Oster and Schweiger, 1992). Nevertheless also the establishment of Arundo donax by stem cuttings, a more cheaper and less labour intensive method, might be successful under warmer climatic conditions existing in Mediterranean countries. Plant survival of between 70 and 82 % have been reported from Jodice et al (1995). Arundo donax could not finish the vegetative circle under climatic conditions in northern Germany because flowering did not happen. Translation of assimilates into the rhizomes was not finished before the first frost. Therefore rhizomes might have been more sensitive against lower temperatures during winter. For Miscanthus growth disturbances due to lack of water supply, insufficient soil ventilation and damage due to strong winds are also supposed to be a cause of failures in establishment and winter survival. Furthermore, Rhizomes of Miscanthus can be attacked by fungi (Fusarium, Phoma, Phytium) (El Bassam, 1994).

The original location might influence growth parameters like number of shoots, height and dry matter yield (Christou et al. 2000). The original growth locations of Arundo donax populations Messolonghi, Ionannina as well as Rabuise and Torviscosa are in northern parts of Greece and Italy. Other populations like Caltagirone and Fondachello grow in southern Italy (Sicily). Hania grows on Crete the most southern location of all populations. The original location of Bizet and Nimes originally is in southern France.

Under native site conditions more than 50 stems per m^2 are quite common (Dalianis, 1996). Maximum shoot number was counted in plants established in 1997. From these populations in all years the northern populations Rabuise and Torviscosa showed reduced shoot development between 10 and 13 shoots/plant (up to 33 shoots/m^2) compared with populations of more southern original location such as Hania and Attiki (15 – 18 shoots/plant respective up to 45 shoots/m^2). These differences appeared mostly between populations established in 1997. Hania produced the highest number of shoots more than populations from Sicily (Caltagirone and Fondachello). Screening trials with 200 Arundo donax populations in Italy, France and Greece also showed a reduction of shoots/m^2 in populations grown in northern Italy and Greece with a mean of 7 - 8 shoots/m^2 , when compared with results obtained in southern parts in Italy, Greece and France (mean 10 – 15 shoots/m^2) (Christou, 2000). In southern Germany, 16 shoots per plant (43 plants/m^2) were counted in the third productions year of Arundo donax in 1991 (Oster and Schweiger, 1992). Compared with Miscanthus giganteus trials in the years 1993-97 at several locations in Germany (El Bassam et al. 2000), rhizomes of Arundo donax reached a nearly equal shoot density.

During al years the diameter of the thickest shoot was measured. Also this parameter was lowest for Torviscosa (1,5 – 1,9 cm), a more northerly location, and highest in southerly populations such as Attiki and Hania (2,0 – 2,6 cm). Some plants established in 1997 reached 3 cm in diameter in 1998. Plants established in 1997 on average had 1 – 5 mm thicker stems than those established in 1998. The diameter of populations from southern France (Nimes and Bizet) had little differences in 1998, when compared with populations of Sicily (Caltagirone and Fondachello) but had thinner shoots in 1999. The stem diameter increased some what during the vegetation period and between the first an second year but there were no differences recorded between the second and third growing seasons. The stem diameter was higher than recorded from partners in Italy, France and Greece in 1998 and 1999 (mean diameter 1,0 – 2,2 cm) (Christou et al, 2000).

Plant height was dependent on climatic conditions and was influenced also by the original location of the population. During the first months of the growing seasons large growth rates of 2-3 cm/day were measured, a rate which was also observed from Oster and Schweiger (1992) in southern parts of Germany. The growth length increased until September, October in each year but varied between the populations. Although cold temperatures and high precipitation characterised weather conditions during summer 1998, maximum height of most populations increased when compared with the previous year. Due to dry summer temperatures in 1999 growth rates slowed down in July and August. Effects due to wind and lodging were not observed also in the wet year 1998. Arundo donax seemed to have high resistance against strong winds comparable with Miscanthus (Rutherford and Heath, 1992). Highest growth was obtained from population Hania. Heights of more than 4 m were measured from single plants. The average height was measured between 2 and 2.5 m for all years. An average height of 328 cm was reached in the third vegetation period in 1991 in experimental plots located in southern Germany but several single shoots reached also heights of more than 4 m (Oster and Scheiger, 1992). Heights of more than 5 m have been reported form Christou et al. (2000a) for sites in southern Europe.

The trend of higher biomass yield of populations of originally southern location and lower yields of those originally found in northern locations could also be observed at the Braunschweig location. Hania had the highest fresh and dry matter yield on average (25 t DM/ha) for plants established in 1997. Fondachello, although this plant originally grows in Sicily, had the lowest biomass production comparable with Torviscosa, a population originally growing in northern Italy. Biomass yields was comparable with the average yield production obtained also in southern Greece and Italy but did not reach maximum yields (31 - 34 t DM/ha) (Christou et al., 2000). Mean yield for the Braunschweig site in the years 1998 - 2000 was 15 t DM/ha. For other sites, mainly in southern parts of Germany, dry matter yields between 8 and 26 t DM/ha have been reported (8-15 t/ha at Würzburg, Main river valley: Kolb et al., 1990; 22 t/ha at Braunschweig: El Bassam and Dambroth, 1991; 17-26 t/ha at Forchheim, upper Rhine-valley: Oster and Schweiger, 1992). The proportion of stems material of the harvested biomass was between 70 and 77 % of the dry matter in yields obtained from Oster and Schweiger (1992).

The average dry matter content in all three year was 46 % (range 41 - 50 %) and did not increase 50 % in all years. Only minor leaf losses were observed and the rest of the plants remained green until the first frost. Also plants did not die back completely as re-sprouting out of the leaf angles were observed in the following spring in plants not harvested in the previous year. Drying of harvested biomass is necessary in order to get a storable material for later industrial uses. Also under Mediterranean climate conditions dry matter contents between 40 and maximum 58 % were measured (Christou et al., 2000). Drying in the field might reduce moisture content up to 15 % under Mediterranean climate (Günes and Saygin, 1996) but depends on harvest date. The moisture content did not decrease much as observed in Miscanthus giganteus (51% in September/October and 80 % at the end of February/March) (El Bassam and Jakob. 1997, Oster and Schweiger, 1992). Only plants which died back after 1.5 years of growth reached 60-70 % dry matter content.

Only for the harvested biomass in 1998 nutrient content was analysed. According to these data the mean nutrient uptake of all Arundo donax populations in the year 1998 was 190 kg N/ha (range 135 - 323 kg/ha), 43 kg P_2O_5 (29 - 70 kg/ha) and 194 kg K_2O (range 140 - 263 kg/ha) for 12 t DM/ha on average. Of all elements, potassium content was very high in the dry matter (1,3 - 1,5 %) but less than in harvested biomass of Miscanthus (El Bassam and Jakob, 1997). Oster and Schweiger (1992) calculated for mean nutrient uptake of Arundo donax cultivations in southern Germany 150 kg N/ha; 60 kg P_2O_5 and 440 kg K_2O for dry matter production of 20 t/ha. Nitrogen content (1,6 %) was relatively high in the biomass because of nearly green plant material was harvested in all years. The nitrogen uptake was higher than the fertilised nitrogen amount. Dalianis (1996) recommends a nitrogen fertilisation up to 100 kg/ha especially in nitrogen poor soils.

Trials with different amounts of nitrogen fertiliser supply have not been established at the Braunschweig site. First results of trials with different nitrogen fertilisation in southern Germany showed an increase in dry matter yield from 16.4 (100 kg N/ha application) to 32.9 t DM (200 kg N/ha) but results were also affected by replanting actions and inhomogene plant stock in the plots (Oster and Schweiger, 1992). In Greece fertilisation on 40 as well as 120 kg N/ha caused no respond in yield differences but there were some increasing effects of higher irrigation rates (Dalianis, 1996; Christou, 1998). Other research results indicate that Arundo donax can endure a wide range of water table levels in the soil and thus can stand also summer drought (Rezk and Edany, 1979).

The plants were fertilised with 5 g nitrogen as deposit adjacent to the plant root (125 kg N/ha). This technique was applied in order to minimise nitrogen losses due to leaching, to reduce amount of nitrogen fertilisers, to maintain a continuous nitrogen supply for the plant and to gain a better adaptation to real nitrogen uptake (Bacher (1996). This method usually is applied for growing vegetables and called CULTAN (Bacher, 1996a; Pohen and Thelen, 1996), but also can be successfully applied for permanent bio-energy crops such as Arundo donax.

6. Conclusions

Arundo donax of orginally locations in southern Europe (France, Italy and Greece) can be successfully grown also under colder climatic conditions in northern Germany.

The establishment of cultivations is best carried out by rhizomes but up to 14 % failures during winter were observed. Two different plant ages were established in the plot centres with unequal numbers which made statistical analysis impossible.

Arundo donax did not reach physiological maturity as the plant grew in height and developed leaves until the first frost in autumn. Almost green plant material was harvested in January each year. As translocation of assimilates into rhizomes is interrupted after the first frost long term weakening of sprouting potential of the rhizomes in spring might occur. The only population with 100 % winter survival during all years was population Torviscosa. This population originally grows in regions of north-eastern Italy.

The shoot number increased up to 18 shoots/plant and showed dependencies to original growth locations as shoot number in populations of southern locations (Hania, Caltagirone, Attiki were higher than of those with northern original location (Torviscosa, Rabuise).

These differences disappeared for biomass yield as only population Hania reached up to 25 t DM/ha. Dry matter yield increased during the first two years but decreased in the third year. High variation within the population and between different populations were recorded.

The dry matter yield is comparable with those of Miscanthus. Arundo donax had higher moisture content in the biomass yield and must be dried before storage. Minor leaf losses were observed but plants did not flower and completely die back during winter in northern Germany.

Application of the CULTAN nitrogen fertilisation method is not only suitable for cultivations of vegetables but can also be adapted to perennial bio-energy crops.

7. Micropropagation of Miscanthus and Arundo donax

7.1 Introduction

Giant Reed (Arundo donax) and Miscanthus have high biomass yield potential and needs low inputs. It is therefore considered to be a very promising energy and fibre crop in Europe. The multiplication of species by seeds and reproductive organs is difficult. Under natural site conditions Arundo donax and Miscanthus spread outwards through their rhizomes' growth. For agricultural purposes it could be propagated either by rhizomes or stem cuttings (Dalianis, 1996).

Micropropagation is regarded as a cheap method of vegetative propagation of plants in order to get a large number of plantlets within short time. It has many advantages than rhizome division or stem cutting. There are three in vitro techniques of multiplication used: a) from nodal segments, b) from meristems, c) from callus.

Multiplication out of nodal segments are reported to be affected by high levels of contamination together with adverse effects of phenol exudates on shoot growth from the lateral buds (Rutherford and Heath, 1992).

Meristematic cultures can also used to induce callus out of small pieces of the growing shoot tip (apical dome plus one or two leaf pimordia) from which new plantlets can be derived. It might be an acceptable method of producing large numbers of Miscanthus and Arundo donax plantlets (Daniel and Baumann, 1987).

In Germany, PICCOPLANT laboratories have reportedly developed a commercially system of micropropagation for Miscanthus sinensis with the potential to produce 2 million plants a year. This system should also be adaptable for micropropagation of Arundo donax plantlets. This report reflects the contribution of subcontractor PICCOPLANT together with the Institute of Crop and Grassland Science in the development of methods of "artificial" seed production by somatic embryogenesis and an efficient in vitro culture system offers the chance of propagation with the advantage of achieving a large number of plantlets with reasonable low costs useful for breeding purposes.

7.2 Methods

7.2.1 Sterile induction of Arundo donax populations and genotypes

For the induction of sterile in vitro cultures nodes were taken from different positions of the plants and sterilised with several applications of 0,5% NAOCL. Detergents were taken to have better adhesion of the sterilant. After that, several washings with 80% ethanol and sterile water were done. The sterile pieces were put onto MS medium supplemented with BAP. In weekly intervals the plants were checked for contaminations and survival-rate of the plant tissue.

7.2.2 In vitro rejuvenation of Arundo donax

To get juvenile plant material the tissue had to be treated with different plant hormones. The **media optimisation** was done on the basis of MS media. The cultivation in the growth room took place with: 16 hours light, 8 hours darkness, 500 lux, 25°C.

7.2.3 Suspension culture with Arundo donax populations

Those varieties with successful induction of organogenic calli were taken into suspension culture. For the small calluses the same media in a liquid form were taken. They were cultivated in an Erlenmeyer flask on a shaker. The starting volume was 10 ml and it was possible to increase the volume up to 300 ml.

7.2.4 Callus culture of Miscanthus and Arundo donax

Plant material of Miscanthus x giganteus, Miscanthus sinensis genotype 'Goliath' and Arundo donax were obtained from field plantations. Young nodal segments of the shoot tip and inflorescences were used as explants. The plant material was surface sterilized with 80 % alcohol. The nodale segments were cut into 3 mm long pieces. All the inflorescences were originally 10-60 mm long. Three methods were compared in order to achieve the best callus formation: 1. Single racemes of inflorescences. 2. Single racemes of inflorescences, cut into pieces 5-8 mm long. 3. Full inflorescences cut into four quarters. These explant types were then placed on the callus induction medium.

The callus induction medium was MS basal medium (Murashige & Skoog, 1962) supplemented with 30 g l^{-1} sucrose, 3.4 g l^{-1} gelrite, 750 mg l^{-1} $MgCl_2 \cdot 6H_2O$ and a varying concentrations of 2,4-dichloro-phenoxyacetic acid (2,4-D). The chosen concentrations were

4.5, 12.5, 22.5, 32.5, and 45 µM. In all media used in this study the pH was adjusted to 5.7 prior to autoclaving. Explants were incubated in darkness at 23°C in 90 mm glass Petri dishes.

7.2.5 Axillary bud culture and shoot induction in Arundo donax and Miscanthus populations

Plant material of 14 different Miscanthus x giganteus origin were obtained from field plants. The plant material was surface sterilized with 80 % alcohol. Axillary buds were placed on a shoot inducing nutrient solution. These medium was a modified MS basal medium (Murashige & Skoog, 1962) supplemented with 20 g l^{-1} sucrose, and 0,3 mg l^{-1} 6-Benzylaminopurin for the shoot induction. In the nutrient solution was the pH adjusted to 5,7 prior to autoclaving. Explants were incubated with a day length of 16 h at 21 °C in glass culture dishes. During the whole culture duration (10th.-60th day, every 10th days) the nutrient content of the culture medium (N, P, K, Ca, Mg) of 2 genotypes were determined by Kjeldahl-method, AAS and photometer.

7.2.6 Root induction of Miscanthus and Arundo donax

After the shoot propagation they were transferred into a nutrient solution for root formation. These solution was a modified MS basal medium (Murashige & Skoog, 1962) supplemented with 20 g l^{-1} sucrose, and 0,5 mg l^{-1} Indole-3-Butyric acid for root formation. The pH was in the nutrient solution adjusted to 5,7 prior to autoclaving. Shoots were incubated in glass test tubes with a day length of 16 h at 21 °C.

7.3 Results

The induction of sterile in vitro cultures from nodes from different populations and genotypes of Arundo donax was affected by several problems. High concentrations of phenolic compounds were produced by freshly cut tissue. As these phenolics are toxic to the tissue the media had to be removed frequently, washed and replaced by fresh liquid. Also sterilisation process was very difficult as most of the plants had very high contamination rates with endogenous bacteria. Even after several months internal bacterial contamination came out of the tissue. Therefore plant material with high contamination rates was tested on different antibiotic concentrations. Those antibiotics killing the bacteria were as well damaging the plants. Only by inducing as many nodes as possible some minimal survival rate was achieved (Table 1, page 40).

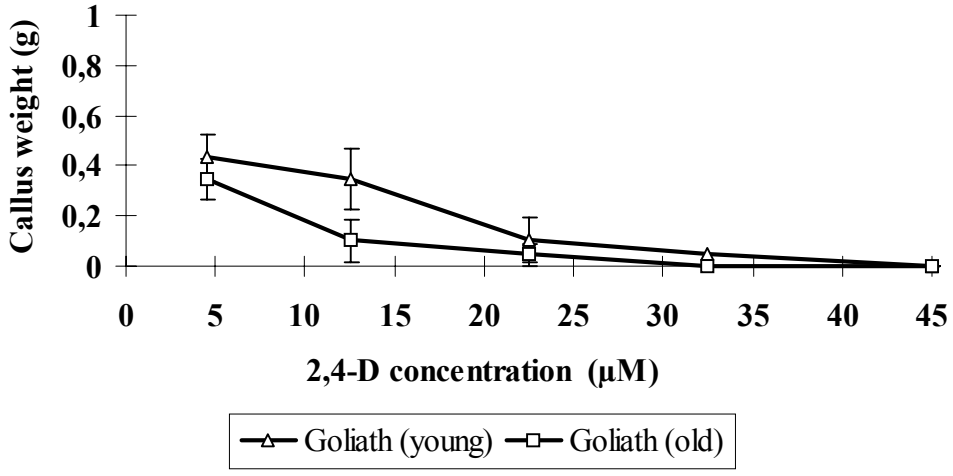
The overall multiplication rate after treatments with different plant hormones was between 2 and 2,5.

7.3.1 Induction of organogenic callus

The rejuvenation and media optimisation could be done with every offspring of Arundo donax populations and genotypes. The first problems did arise with the induction of organogenic callus formation (Table 1, page 40). This was possible with 5 clones only. It was achieved by giving higher auxin concentrations. At the same time too high auxin concentration could lead to mutations which do badly influence the regeneration capacity of the callus. Because of this reason only work with extremely low auxin concentrations was carried out.

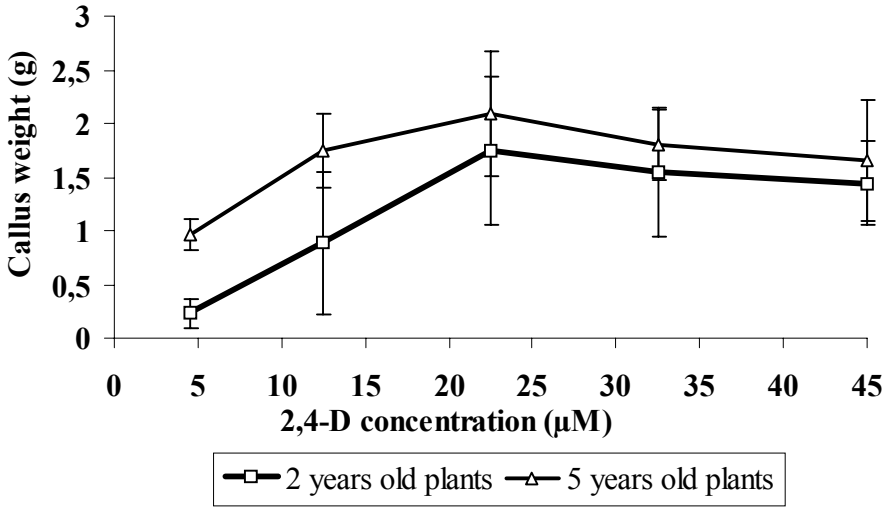
After 12 weeks of culturing the nodal segments of Miscanthus genotypes and Arundo donax, the best auxin concentrations for the different Miscanthus genotypes for optimum callus formation could be determined. The genotypes 'Goliath' showed the highest callus formation on a medium containing 4.5 µM 2,4-D. The highest concentrations of 2,4-D did not lead to callus formation by 'Goliath'. The results were improved by using younger nodal segments instead of older ones (Figure 1).

Figure 1. Influence of different 2,4-D concentrations on the callus formation rate of Miscanthus sinensis 'Goliath'. (Bars represent ± standard errors)



Miscanthus x giganteus showed the highest callus formation rate on a medium containing 22.5 µM 2,4-D. The best results were obtained with nodal segments from a 5 and 6 years old plantation of Miscanthus x giganteus (Figure 2).

Figure 2: Influence of different 2,4-D concentrations on the callus Formation rate of Miscanthus x gigantheus (Bars: standard errors)

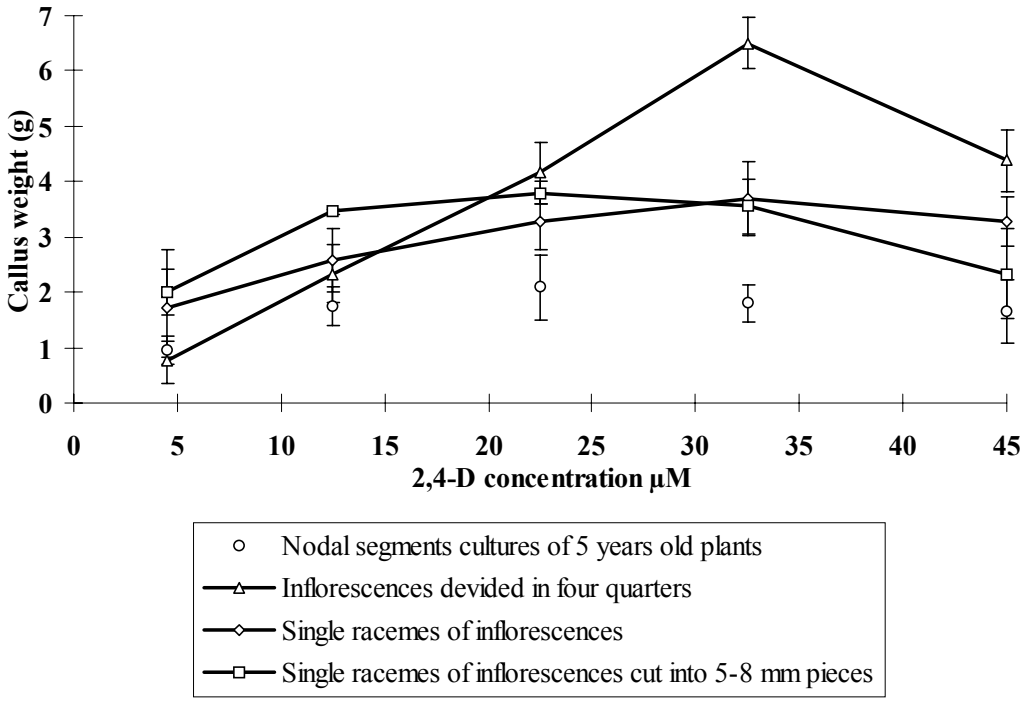


Considerable differences have been observed when different explant types of the same genotype were compared for their callus producing potential.

Explant establishment frequently requires special procedures to overcome problems associated with exudation of polyphenols. In many species (especially woody plants) as well as Miscanthus sinensis, the medium darkens within minutes, hours, or days, after transfer of the explants to culture vessels. Often, if left unattended, a lethal browning of the explants will occur. Consequently, researchers and commercial technicians must devote considerable time and expense to successfully obtain healthy growing cultures. These in vitro exudates have been identified as tannins or oxidized polyphenols. Plant phenols are, in part, synthesised through shikimic acid pathway, and are present in abundant quantities in plants (Michael E.C. and John E.P., 1986). The callus formation of Miscanthus sinensis nodal segments was strongly inhibited by polyphenole compounds (Toth and Mix-Wagner, 1998).

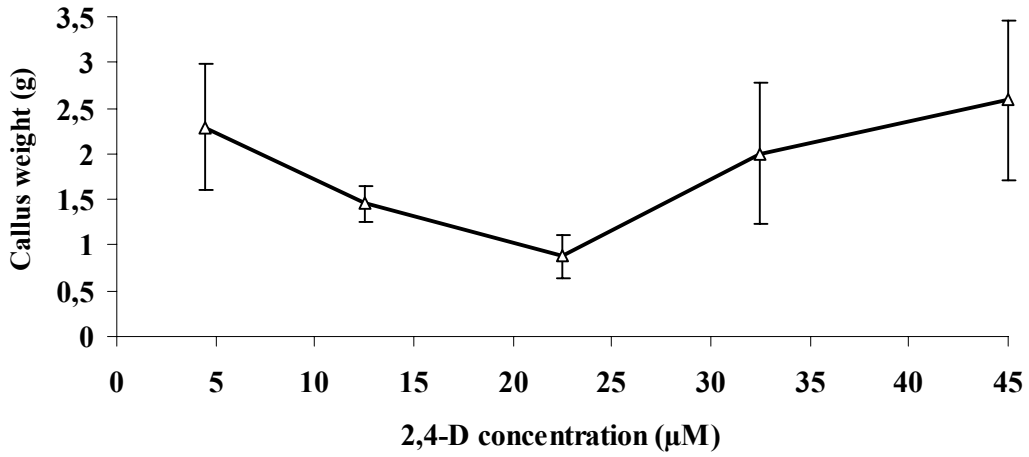
The inflorescences of Miscanthus x giganteus produced a higher callus weight than nodal segments. A concentration of 32.5 µM 2,4-D in the culture medium led to the best callus formation rate. Additionally, the use of 10 mm long inflorescences devised into four quarters could improve this callus formation rate significantly (Figure 3).

Figure 3: Influences of different 2,4-D concentrations on the callus formations rate of inflorescence culture of Miscanthus x giganteus in comparison to nodal segment cultures (Bars: standard errors)



Inflorescences in contrast to nodal segments did not show any browning during culturing. Nodal segments of Arundo donax showed better callus formation compared with all tested Miscanthus genotypes. Interestingly, the highest callus formation rate was achieved by the lowest (4.5 µM) and highest (45 µM) 2,4-D concentration tested when nodal segments were obtained in October. But high 2,4-D concentrations led to callus browning during further culturing under high 2,4-D concentrations in the culture medium and, in the end, the callus died (Figure 4). The nodal segment cultures, which were obtained in June showed a lower callus formation rate. High concentrations restricted callus formation.

Figure 4: Influence of different 2,4-D concentrations on the callus formation rate of Arundo donax (Bars: standard errors)



7.3.2 Induction of shoot clusters

The cells and cell aggregates from the suspension cultures with Arundo donax populations were once again regenerated on agar media. Of every variety a suspension culture could be established could and successfully used to form shoot clusters. In the older cultures the capacity to regenerate did not diminish. The shoot clusters from organogenesis were much more vigorous than those from the tissue culture. They were showing a very high growth response. The tissue was very bushy and juvenile (Table 1).

7.3.3 Hardening in the greenhouse

The plantlets of Arundo donax from tissue culture were pretty easy in hardening. The hardening effect of plantlets from shoot clusters was always successful. Long term observations will be necessary to follow the growth and phenotypic appearance of the plants whether later mutations might appear. The older plants remained more juvenile for approximately one year. This lead to a very bushy plant with many soft shoots. Only later with the formation of rhizomes strong shoots are generated out of these rhizomes (Table 1).

Table 1: Summary of results of micropropagation of Araundo donax genotypes
(after PICCOPLANT; Appendix Table 13)

	Growth	In vitro	Induction of	Suspension	Induction of	Tissue culture
	Greenhouse	Rejuventation	Callus	culture	Shoot clusters	offspring
Population						
Germany						
Clone A	+++	+++	+++	+++	+++	+++
Clone G	+++	+++	+	+	++	+++
Clone K	+++	+++	+++	+++	+++	+++
Messolonghi	+++	++	+	++	+++	+
Attiki	+	+	0	0	0	+
Chama	++	+	0	0	0	+
Pyrgos	+	+	0	0	0	+
Rhizoloum Karditza	+++	+	++	++	+	+++
Pyrus Evron	++	++	+	+	+	++
Fondachello	+	+	0	0	0	+
Torviscosa	+	++	0	+	0	+
Sommuries	+	+	+	0	++	+
Caltagirone	++	+	0	+	0	+
Altedo	++	++	+		+	+

+++: very good response; ++ good response; +: bad response; 0: no response of population

7.4 Discussion

There were different results obtained from both plant species. At the Institute of Crop and Grassland Science, different explant types of Miscanthus and Arundo donax were used from in vitro propagated plants. The explants were cultured on Murashige & Skoog, (MS 1962) medium supplemented with varying concentrations of 2,4-dichlorophenoxyacetic acid (4.5-31.7 µM). Explants were incubated in darkness at 25°C (Holme and Petersen, 1996).

The best callus induction percentages were obtained in explants from immature inflorescences or shoot apices. The addition of 2,4-D to the medium is required for induction calli. However, the concentrations of 2,4-D used, significantly affected callus induction only on leaf explants from greenhouse-grown or in vitro-grown shoots. The maximum callus formation rate on leaf explants from in vitro-propagated shoots was observed at a concentration of 22.6 µM 2,4-D as well as in the case of nodal segments from field plantations of Miscanthus x giganteus using a 2,4-D concentration of 22.5 µM.

Immature inflorescence culture from greenhouse-grown plants in comparison to inflorescence cultures from field plants of Miscanthus x giganteus both showed the highest callus formation rate on a medium 2,4-D concentration of 32.5 µM.

The increase in callus induction rate with increasing length of immature inflorescences is consistent with results obtained for other grasses and is explained by the many active meristematic cells contained within developing inflorescences (Brettel et al., 1980; Cai and Butler, 1990). Although no optimum was found at any length of inflorescence, some of the smaller inflorescences had the capacity to induce a very high percentage of embryogenic callus. The production of higher percentages of embryonic callus from smaller inflorescences has also been observed in many other grasses including sorghum (Berett et al., 1980). However, the embryonic callus formation on the different explant types at different developmental stages of Miscanthus was independent on the 2,4-D concentration (Bhaskaran and Smith, 1990).

In vitro propagation with different Arundo donax populations at PICCOPLANT only showed successful 2 German clones and Rhizoloum, Karditza, Messolonghi and Sommuries. From these populations also shoot clusters could be induced after callus formation in suspension cultures. A reason might be the delivered bad plant material, which needed to be further propagated in the greenhouse before starting in vitro cultures with node material. Comparable results were obtained from stem material of all delivered Arundo donax populations at the Institute of Crop and Grassland Science. Propagation with stem material under greenhouse

conditions failed completely for all populations. In the field trial therefore rhizome material was successfully used.

7.5 Conclusions:

Large numbers of Miscanthus genotypes can be propagated by in vitro culture. Differences in callus formation were observed between genotypes. For Arundo donax propagation is dependent on delivered plant material. Further research concerning in vitro propagation is necessary.

8. Acknowledgements

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10. Appendix

Tables of meteorological data in the years 1997 – 2000:

Table 1: Meteorological data in 1997 (Monthly average):

	Air temperature (°C) in 2m height			Sunshine duration (h)		Global solar radiation (J/cm2)		Precipitation (mm)		Water-Balance	
			Month	Month		Month		Month		mm.	
	max.	min.	mean	max.	Total	mean	Total	max.	Total		
January	8,4	-21,5	3,9	7,2	64	262	8115	5,0	13		4
February	13,7	-4,7	6,0	8,9	88	485	13567	21,7	81		61
March	18,5	-1,9	5,9	10,8	116	781	24210	14,1	55		29
April	16,5	-2,7	9,7	12,5	189	1410	42285	16,5	43		-5
May	27,7	1,8	14,4	14,5	219	1792	55547	19,9	77		8
June	29,4	5,3	16,5	14,7	273	2166	64973	21,2	60		-36
July	27,0	10,9	16,2	14,6	216	1809	56064	16,6	71		-11
August	32,7	11,1	16,7	13,9	269	1820	56416	15,3	25		-114
Sept.	30,0	1,8	13,8	11,8	195	1246	37381	4,4	14		-59
October	21,9	-6,1	8,8	9,5	122	649	20132	9,5	42		7
Nov.	15,7	-2,5	2,1	7,8	44	242	7268	21,2	55		39
Dec.	12,7	-11,3	2,0	5,6	25	144	4460	9,7	65		51
Summary	33	-22	9,5	14,7	1820	1067	390418	21,7	587		-37

Table 2: Meteorological data in 1998 (Monthly average):

	Air temperature (°C) in 2m height			Temp. near soil (°C)		Sunshine duration (h)		Global solar radiation (J/cm2)				Precipitation (mm)		Evaporation (mm)		Diff. (mm)
			Month	Month		Month		Month				Month		Month.		
	max.	min.	mean	min.	mean	max.	total	max.	min.	mean	total	max.	total	max.	total	
January	14,3	-8,4	3,9	-7,5	0,8	7,3	61,1	561	64		6863	9,3	37,1	1,9	17,7	19,4
February	15,5	-11,2	6,0	-12,9	1,5	7,2	84,1	767	82	480,9	13465	2,6	8,2	1,7	21,8	-13,8
March	19,9	-3,8	5,9	-7,0	1,6	11,5	108,7	1735	197	844,9	26193	17,8	57,4	2,8	29,8	27,6
April	20,3	-0,3	9,7	-3,2	4,6	12,7	101,7	2377	344	1100,8	33023	8,2	78,8	3,4	43,8	35,0
May	27,6	5,0	14,4	3,3	7,6	14,9	227,1	2900	570	1884,0	58403	22,9	47,0	7,2	95,2	-48,2
June	30,0	5,8	16,5	4,7	11,3	12,6	175,1	2773	765	1775,0	53251	18,5	91,7	5,7	71,1	20,6
July	31,8	8,8	16,2	7,0	11,3	14,6	145,8	2697	501	1543,5	47847	13,3	64,8	6,7	67,9	-3,1
August	33,9	9,0	16,7	6,2	10,9	14,2	186,5	2608	349	1543,5	47850	16,1	67,1	8,6	91,7	-24,6
Sept.	22,6	4,5	13,8	2,7	8,7	9,8	91,2	1613	266	885,2	26556	17,5	52,7	2,3	37,4	15,3
October	18,4	1,8	8,8	-0,8	5,8	8,8	58	987	102	430	13316	61,9	147	1,6	21	126
Nov.	12,5	-7,8	2,1	-9,7	-0,9	5,5	37	525	43	254	7622	13,0	43	0,7	8	35
Dec.	12,3	-10,1	2,0	-16,1	-1,2	7,1	62	426	46	195	6035	11,5	37	1,2	11	26
Total			9,7		5,1		1342			930	340424		732		517	215
difference to the 30 year mean			+0,8		+1,7		-172				-10866		+113		-24	

Table 3: Weather data of the year 1999 (monthly average)

	Air temperature (°C) in 2m height			Temp. Near soil (°C)		Sunshine duration (h)		Global solar radiation (J/cm2)				Precipitation (mm)		Evaporation (mm)		Diff. (mm)
			Month	Month		Month		Month				Month		Month.		
	max	min	mean	Min	mean	max	total	max	min	mean	total	max	Total	max	total	
January	14.7	-7.6	4.0	-9.5	0.5	7.1	49	574	32	224	6941	9.9	54	1.5	14	40
Febr.	10.8	-10.4	1.5	-16.5	-2.6	7.1	42	957	51	422	11826	14.2	58	1.1	7	51
March	19.7	-1.8	6.1	4.3	1.2	11.3	104	1779	224	792	24551	14.5	43	2.9	28	15
April	19.7	-0.9	9.9	-4.2	3.8	14.0	179	2551	443	1464	43917	12.2	40	4.5	59	-19
May	29.0	2.5	14.0	-1.0	6.5	15.1	248	2896	810	1990	61679	17.3	52	6.8	96	-44
June	27.0	7.2	15.6	5.1	8.7	14.9	257	2950	1104	2158	64733	26.4	67	5.6	87	-20
July	32.9	9.6	19.6	6.5	12.4	15.0	253	2809	834	2048	63496	3.6	16	9.0	136	-120
August	30.1	7.2	17.5	4.3	10.8	14.2	180	2565	475	1504	46616	13.7	55	8.4	102	-47
Sept.	30.3	8.2	17.9	4.7	10.3	13.1	215	2078	335	1316	39469	16.3	42	7.2	102	-60
October	18.8	-1.2	9.8	-4.0	5.3	9.4	105	1034	178	656	20331	11.5	40	1.8	31	9
Nov.	15.9	-5.6	4.7	-8.3	1.2	8.7	63	730	68	296	8884	3.4	16	1.4	14	2
Dec.	10.8	-4.1	3.6	-6.7	0.8	6.5	35	347	42	155	4650	10.7	54	1.1	13	41
max min	32.9					15.1		2950				26.7		9.0		
Mean		-10.4		-16.5					32							
Total			10.4		4.9					1085						
							1730				397093		536		689	-153
difference to the 30 year mean			+ 1.5		+ 1,5		+216				+ 45803		- 83		-148	

Table 4: Weather data in the year 2000

	Air temperature (°C) in 2m height			Temp. near soil (°C)		Sunshine Duration (h)		Global solar radiation (J/cm2)				Precipitation (mm)		Evaporation (mm)		Diff. (mm)
			Month	Month		Month		Month				Month		Month.		
	max.	min.	Mean	min.	Mean	max.	Total	max.	min.	Mean	Total	max.	total	max.	Total	
January	10,3	-7,4	2,5	-10,3	-0,9	8	71	586	45	248	7696	11,7	46	1,0	12	34
February	15,9	-2,9	5,0	-5,5	1,3	10	76	1077	146	467	13546	11,7	52	1,7	20	32
March	13,8	-1,0	5,7	-2,4	2,4	9	66	1486	124	657	20361	12,8	93	1,3	19	74
April	24,3	-2,7	10,8	-5,7	3,9	13	162	2464	295	1382	41466	9,5	35	5,0	70	-35
May	29,0	6,1	15,1	2,2	7,4	15	274	2854	831	1986	61576	11,0	45	8,7	110	-65
June	33,8	6,8	17,2	5,4	10,3	16	228	2966	888	2051	61526	4,4	22	10,4	114	-92
July	24,8	7,6	15,6	5,1	11,1	11	106	2769	564	1388	43027	23,3	78	4,3	63	15
August	31,4	7,3	17,8	4,7	11,2	13	217	2245	443	1640	50846	13,8	36	7,7	101	-65
Sept.	25,0	3,1	14,5	0,3	9,0	11	124	1581	400	982	29455	13,9	40	3,3	51	-11
October	20,1	3,7	11,5	1,4	6,6	9	109	1183	138	625	19379	6,1	33	2,2	39	-6
Nov.	15,2	1,4	7,5	-1,6	2,4	6	71	671	68	325	9741	6,6	33	1,5	23	10
Dec.	14,8	-7,4	4,1	-9,9	0,6	7	66	349	57	209	6487	6,7	33	1,0	15	18
Mean/			10,6		5,4					997						
Total							1569				365106		544		637	-93

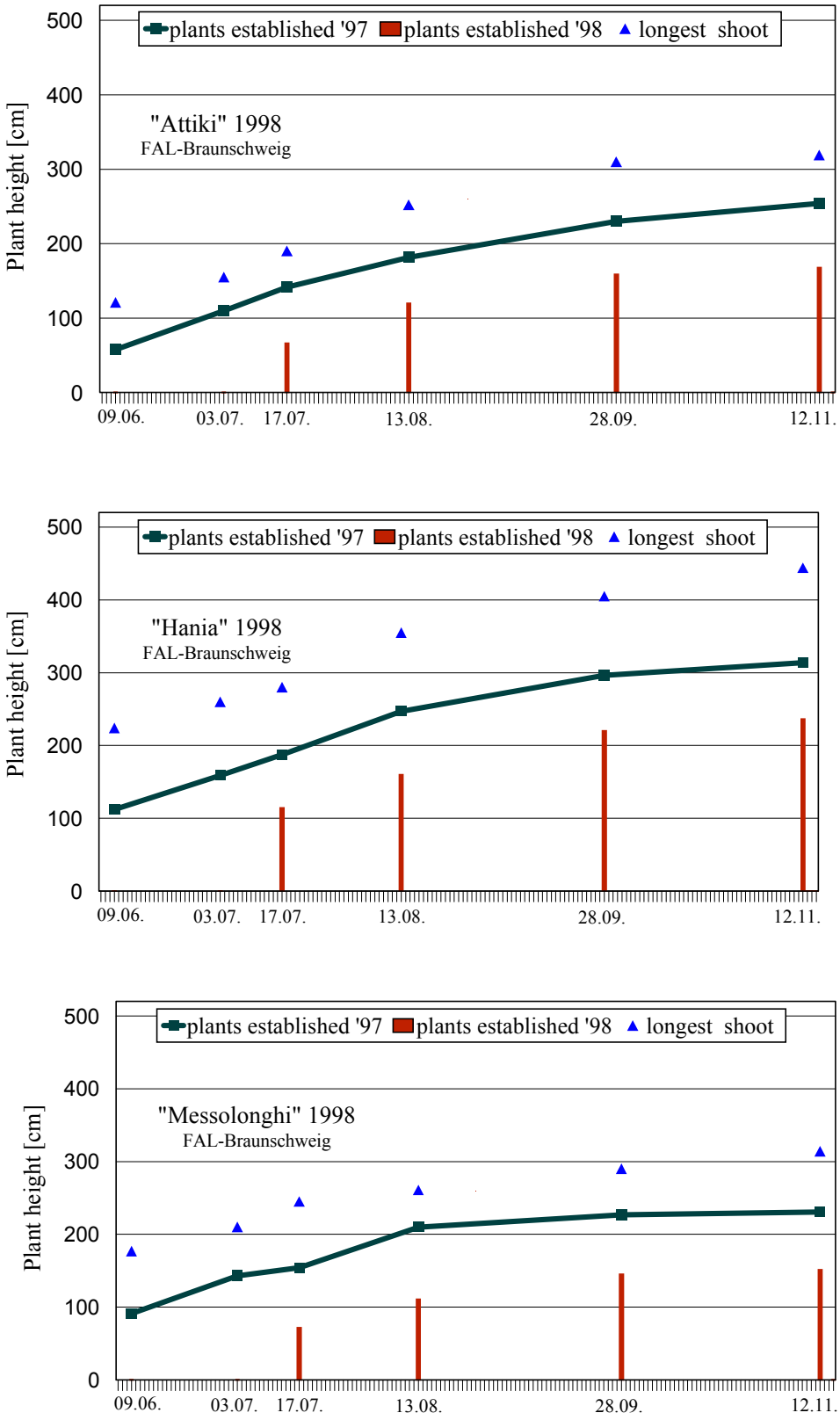


Figure 1: Average plant growth in the year 1998 separated for Arundo donax planted in 1997, 1998 as well as length of longest shoot in the plots

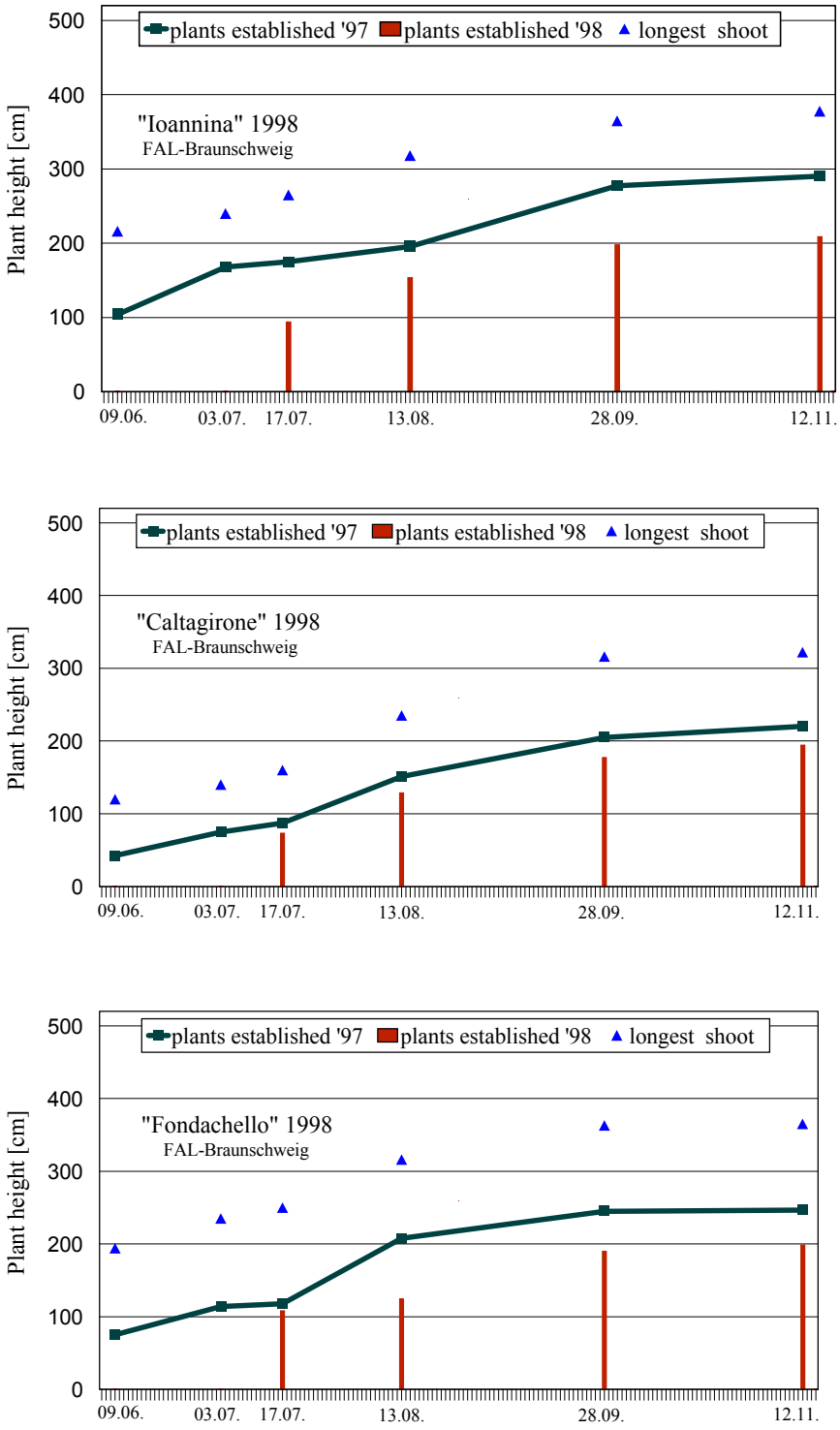


Figure 2: Average plant growth in the year 1998 separated for Arundo donax planted in 1997, 1998 as well as length of longest shoot in the plots

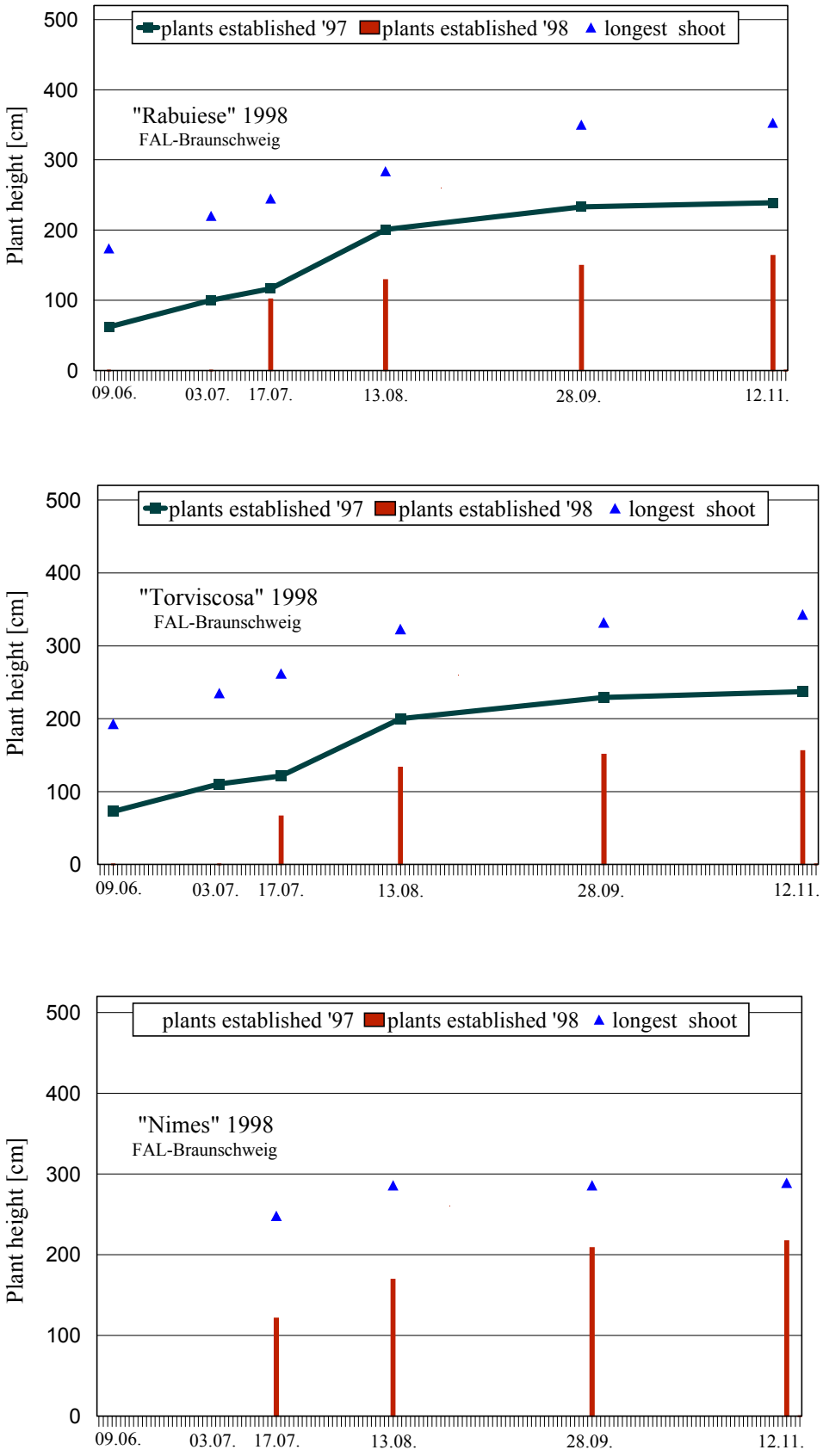


Figure 3: Average plant growth in the year 1998 separated for Arundo donax planted in 1997, 1998 as well as length of longest shoot in the plots

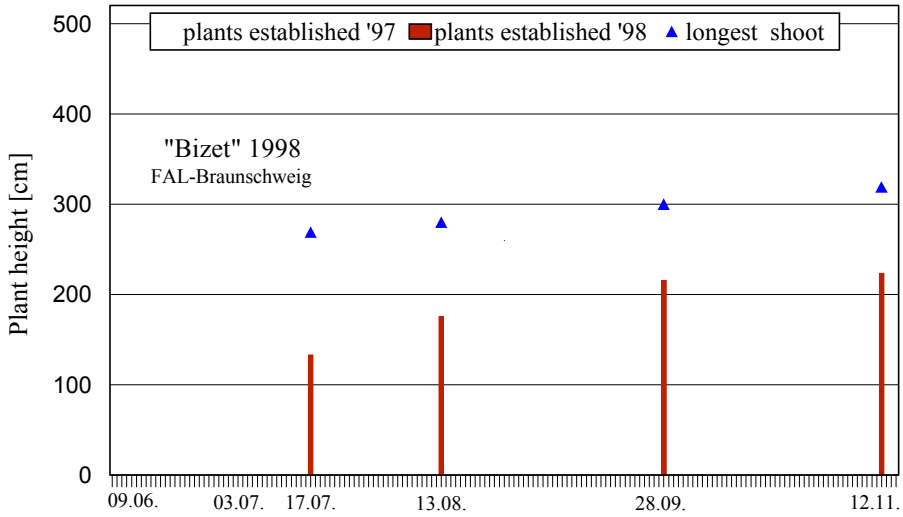
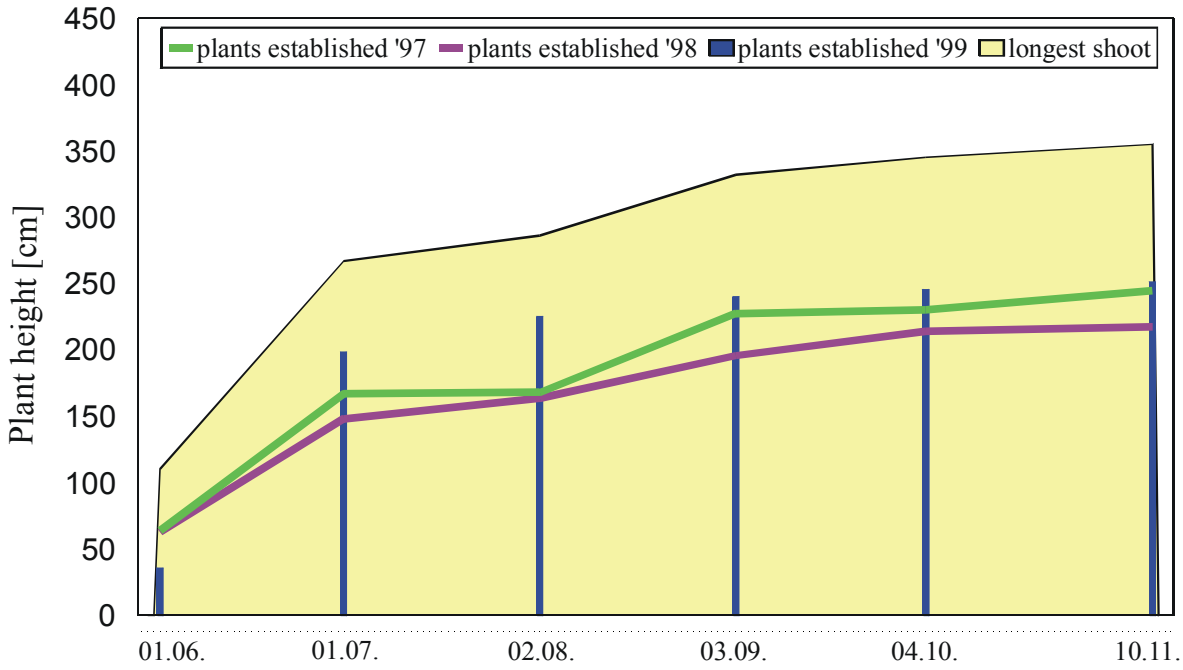


Figure 4: Average plant growth in the year 1998 separated for Arundo donax planted in 1997, 1998 as well as length of longest shoot in the plots

Plant growth of Arundo donax planted in 1997 and 1998 monitored in the year 1999 (A: Monitoring of the plot centre; B: Monitoring of 4 single plant chosen across the whole plot)

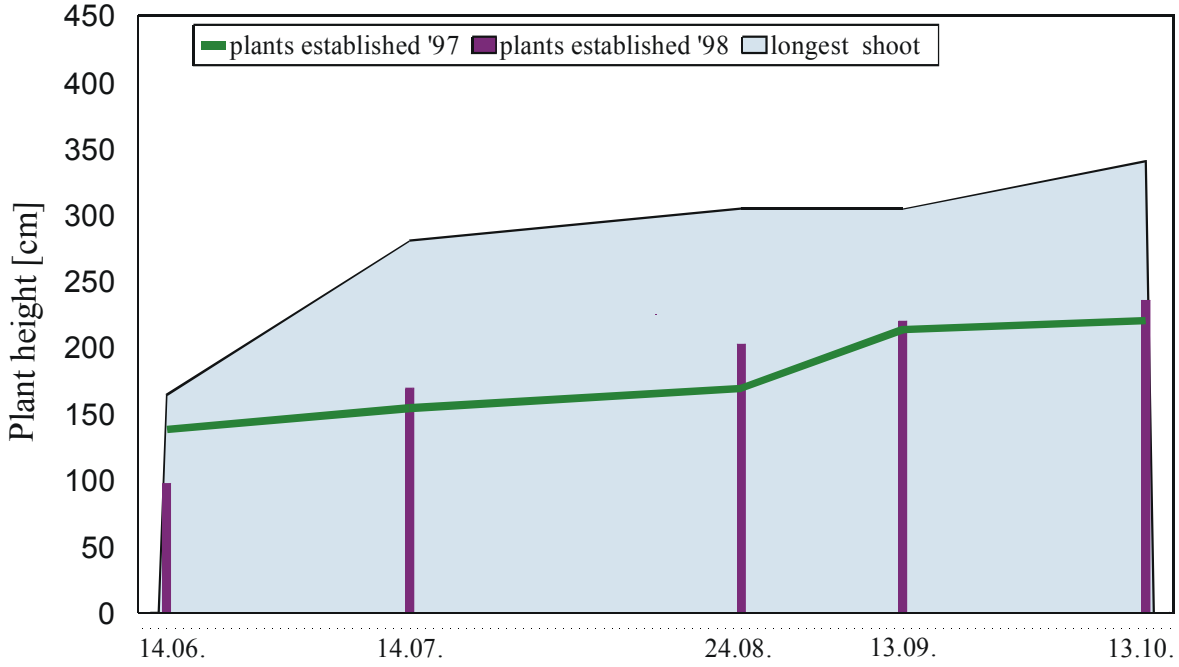
see following pages

A



Plant height of the Arundo genotype "Attiki" during the growing season 1999
FAL-Braunschweig
(Centre of the plot)

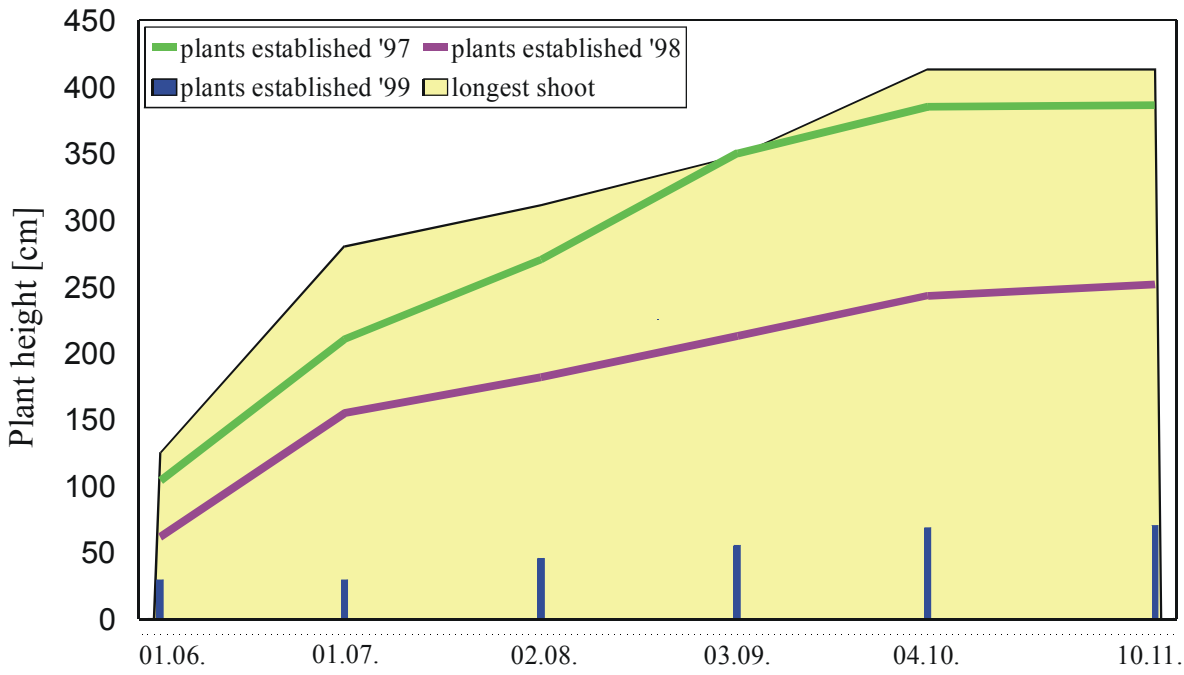
B



Plant height of the Arundo genotype "Attiki" during the growing season 1999
FAL-Braunschweig

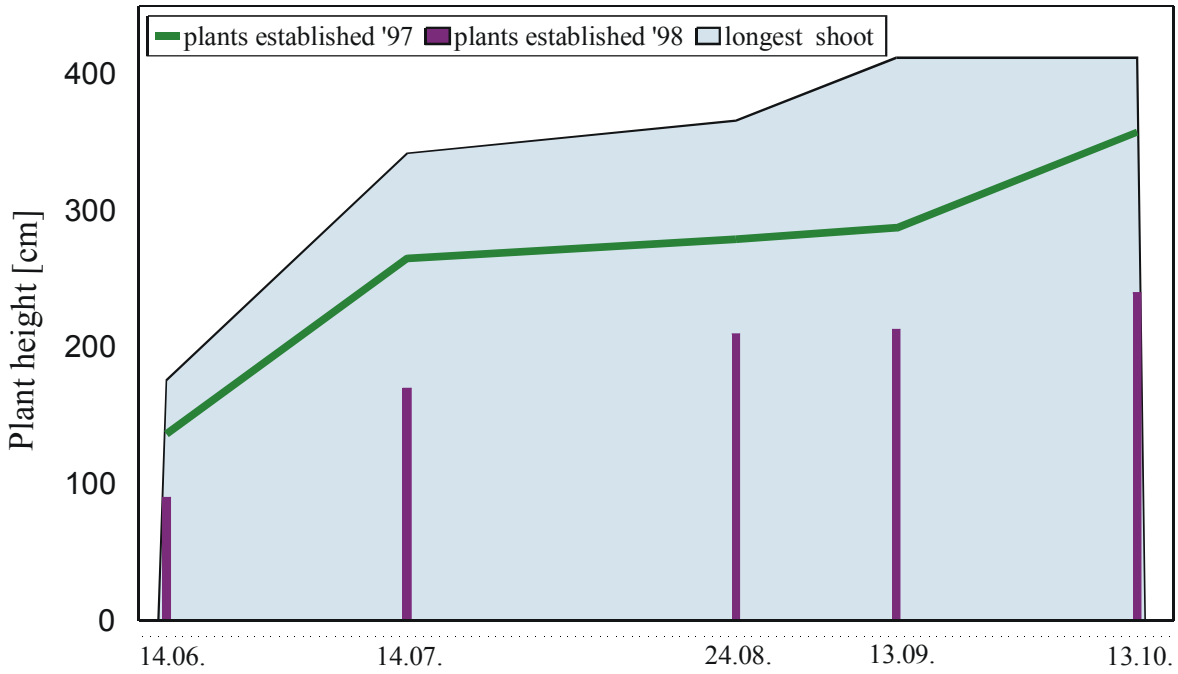
Figure 5: Average height of Arundo donax planted in 1997 and 1998 as well as longest shoot
(A: plot centre; B: 4 marked plants chosen across the whole plot)

A



Plant height of the Arundo genotype "Hania" during the growing season 1999
FAL-Braunschweig

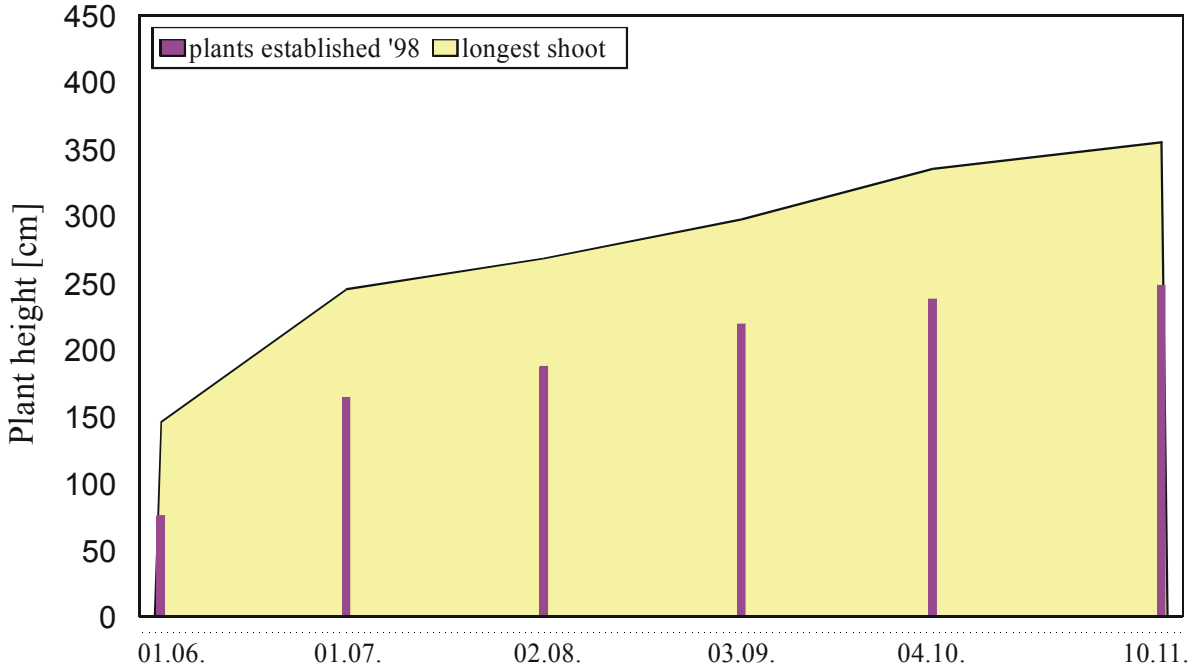
B



Plant height of the Arundo genotype "Hania" during the growing season 1999

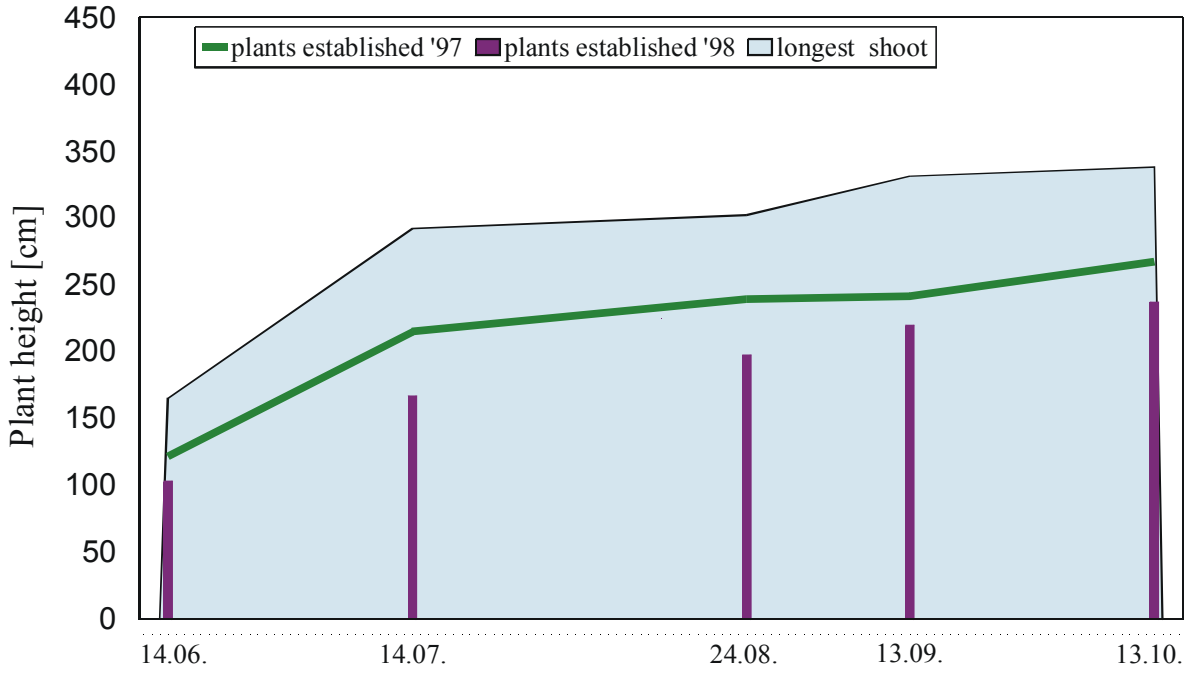
Figure 6: Average height of Arundo donax planted in 1997 and 1998 as well as longest shoot (A: plot centre; B: 4 marked plants chosen across the whole plot)

A



Plant height of the Arundo genotype "Ionannina" during the growing season 1999
FAL-Braunschweig

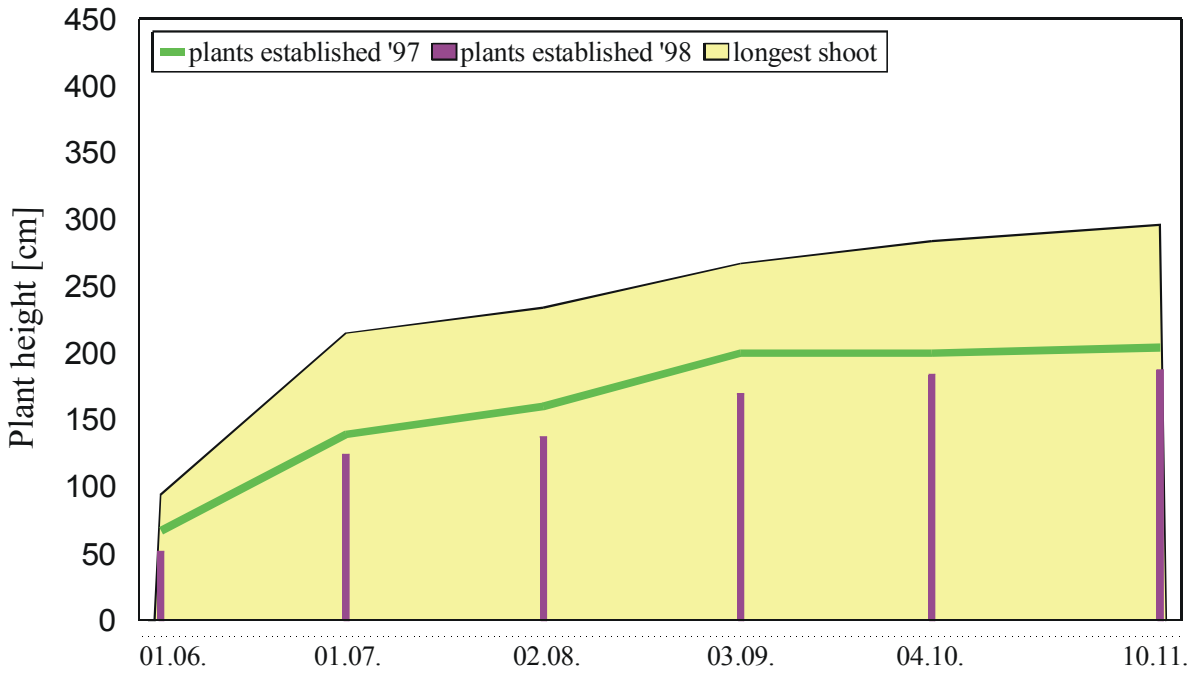
B



Plant height of the Arundo genotype "Ionannina" during the growing season 1999
FAL-Braunschweig

Figure 7: Average height of Arundo donax planted in 1997 and 1998 as well as longest shoot (A: plot centre; B: 4 marked plants chosen across the whole plot)

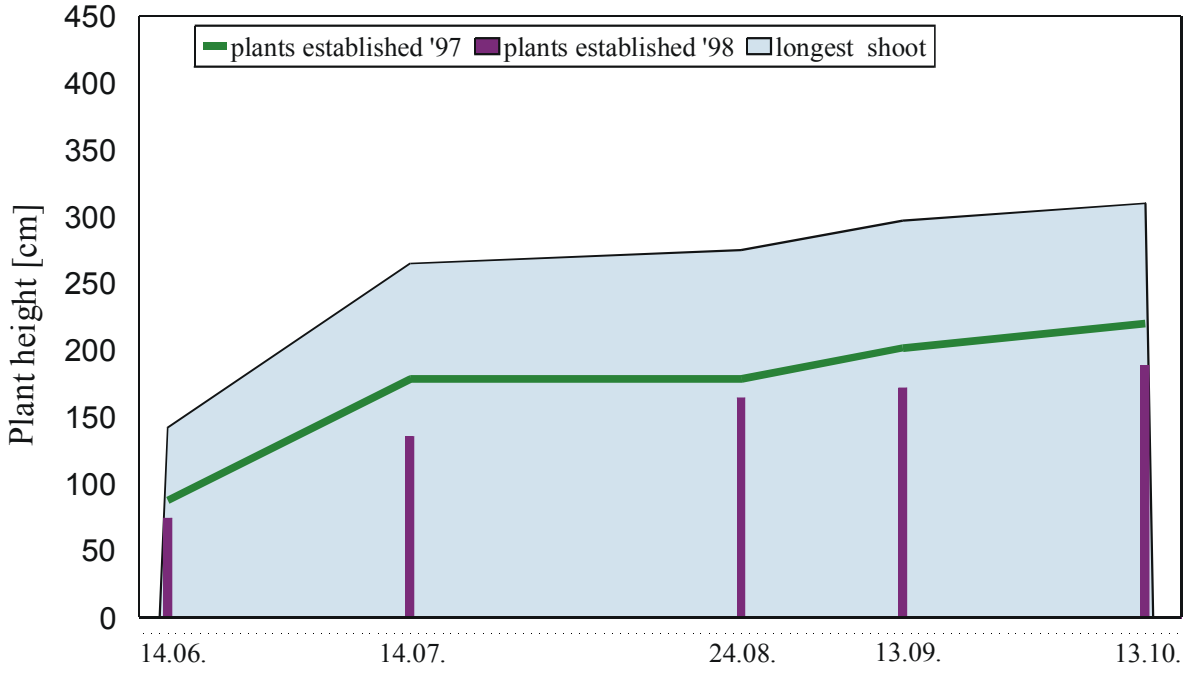
A



Plant height of the Arundo genotype "Messolonghi" during the growing season 1999

FAL-Braunschweig

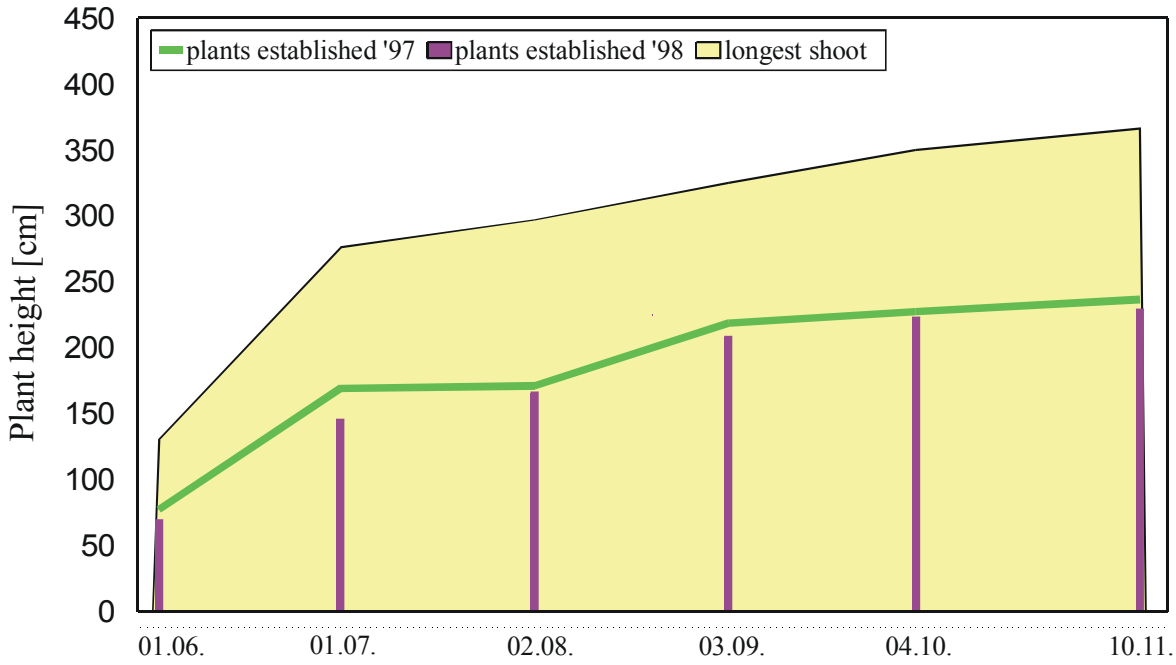
B



Plant height of the Arundo genotype "Messolonghi" during the growing season 1999

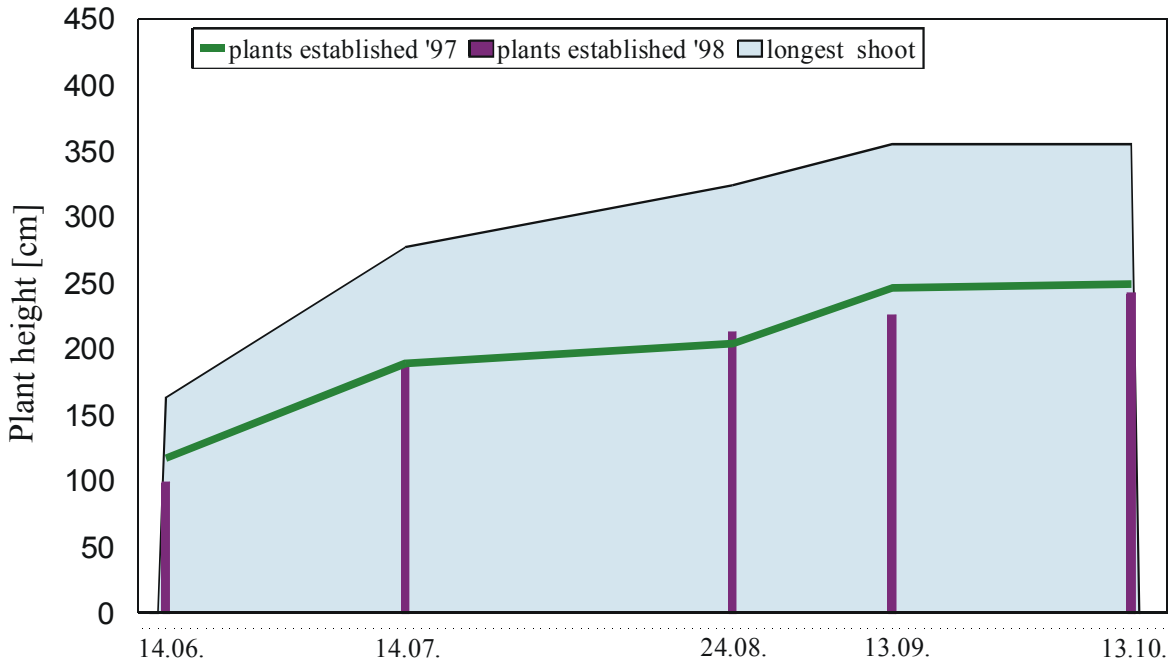
Figure 8: Average height of Arundo donax planted in 1997 and 1998 as well as longest shoot (A: plot centre; B: 4 marked plants chosen across the whole plot)

A



Plant height of the Arundo genotype "Fondachello" during the growing season 1999
FAL-Braunschweig

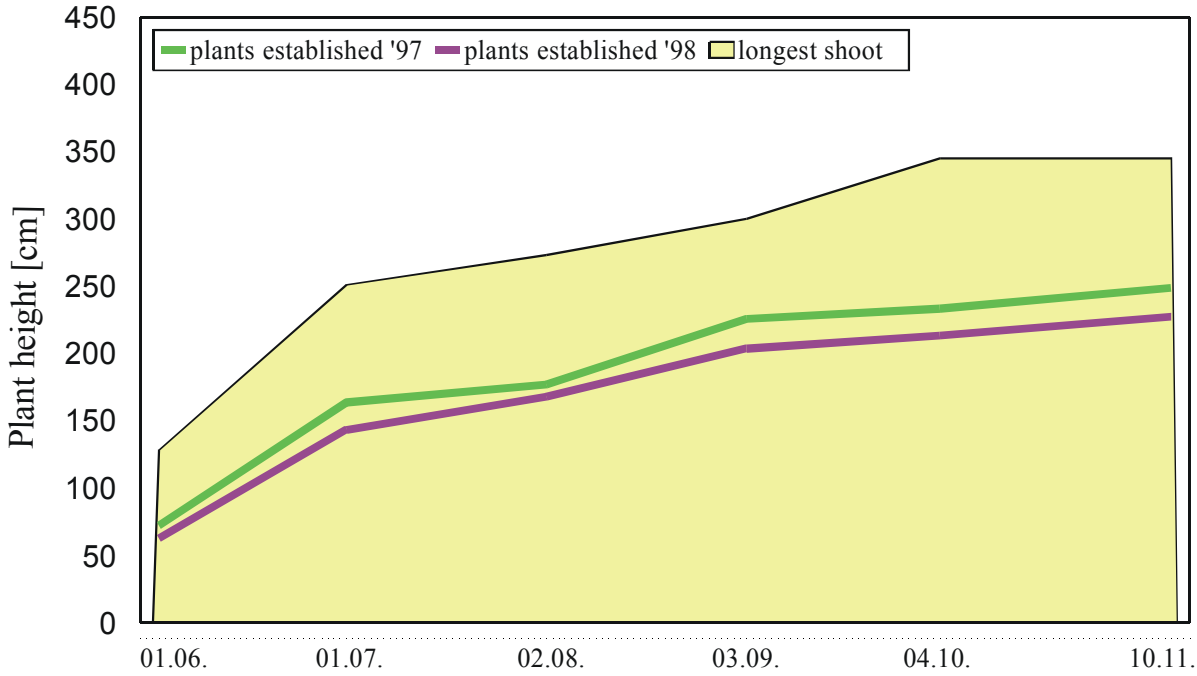
B



Plant height of the Arundo genotype "Fondachello" during the growing season 1999
FAL-Braunschweig

Figure 9: Average height of Arundo donax planted in 1997 and 1998 as well as longest shoot (A: plot centre; B: 4 marked plants chosen across the whole plot)

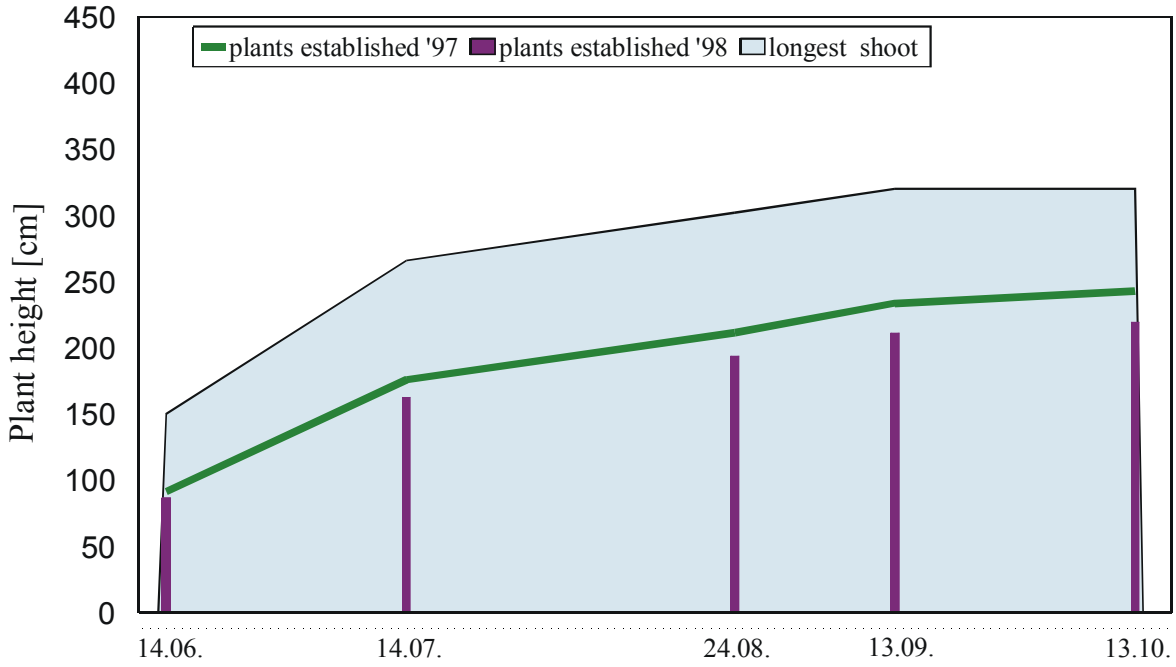
A



Plant height of the Arundo genotype "Caltagirone" during the growing season 1999

FAL-Braunschweig

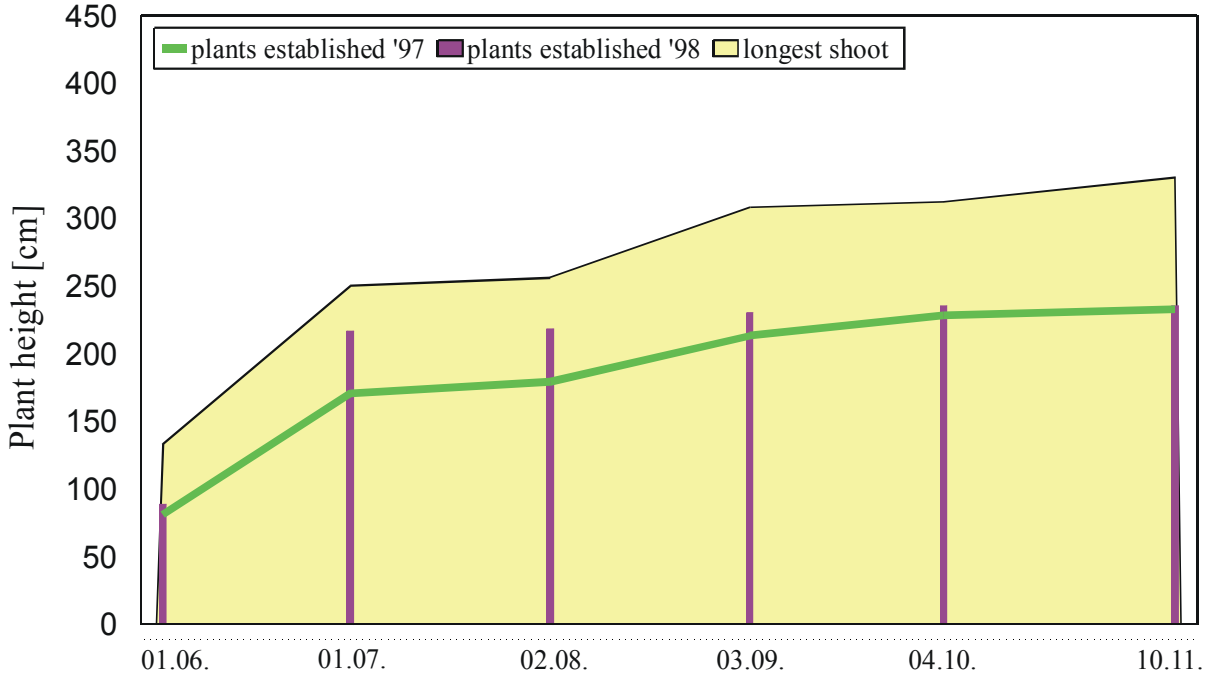
B



Plant height of the Arundo genotype "Caltagirone" during the growing season 1999

Figure 10: Average height of Arundo donax planted in 1997 and 1998 as well as longest shoot (A: plot centre; B: 4 marked plants chosen across the whole plot)

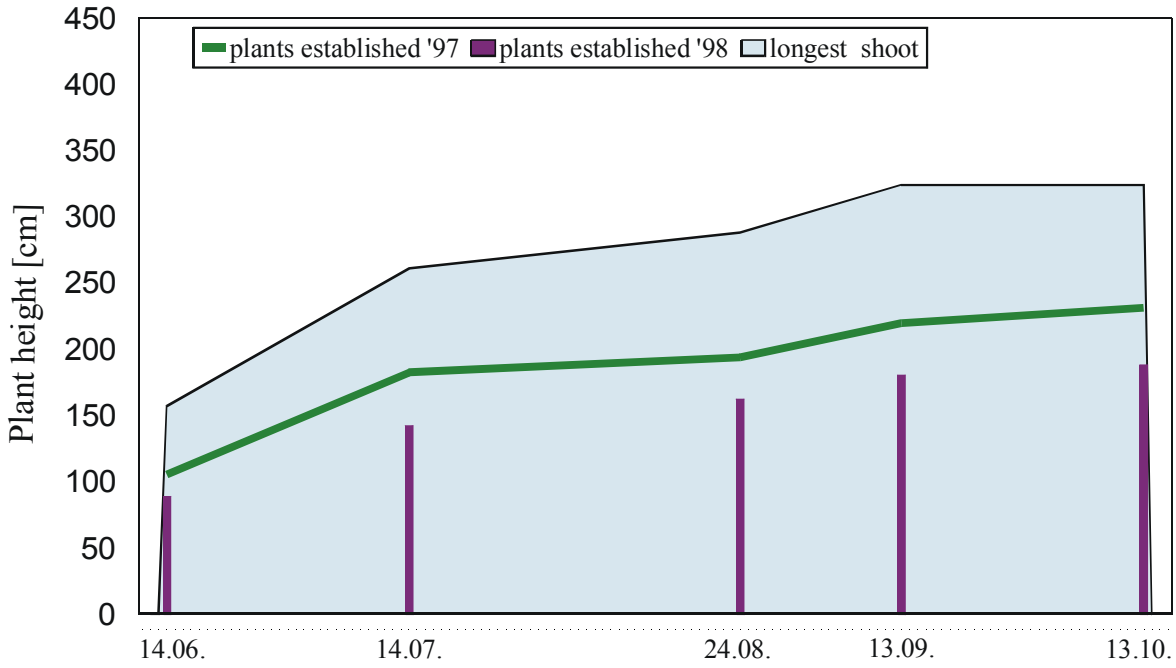
A



Plant height of the Arundo genotype "Torviscosa" during the growing season 1999

FAL-Braunschweig

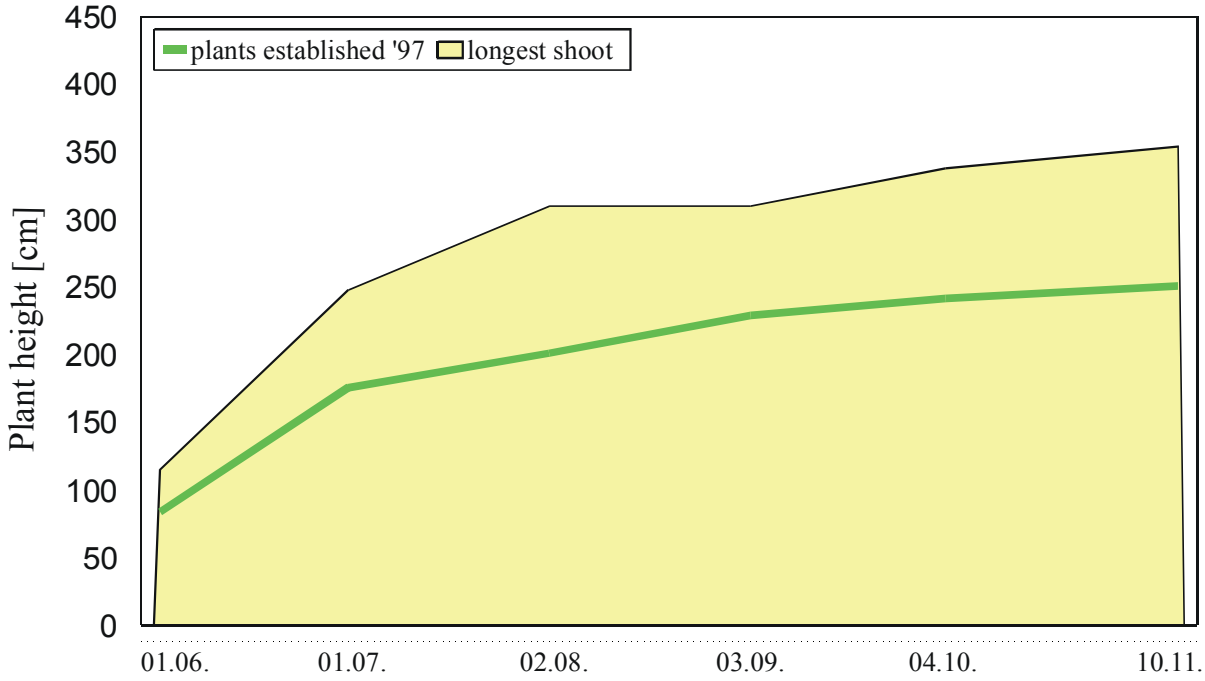
B



Plant height of the Arundo genotype "Torviscosa" during the growing season 1999

Figure 11: Average height of Arundo donax planted in 1997 and 1998 as well as longest shoot (A: plot centre; B: 4 marked plants chosen across the whole plot)

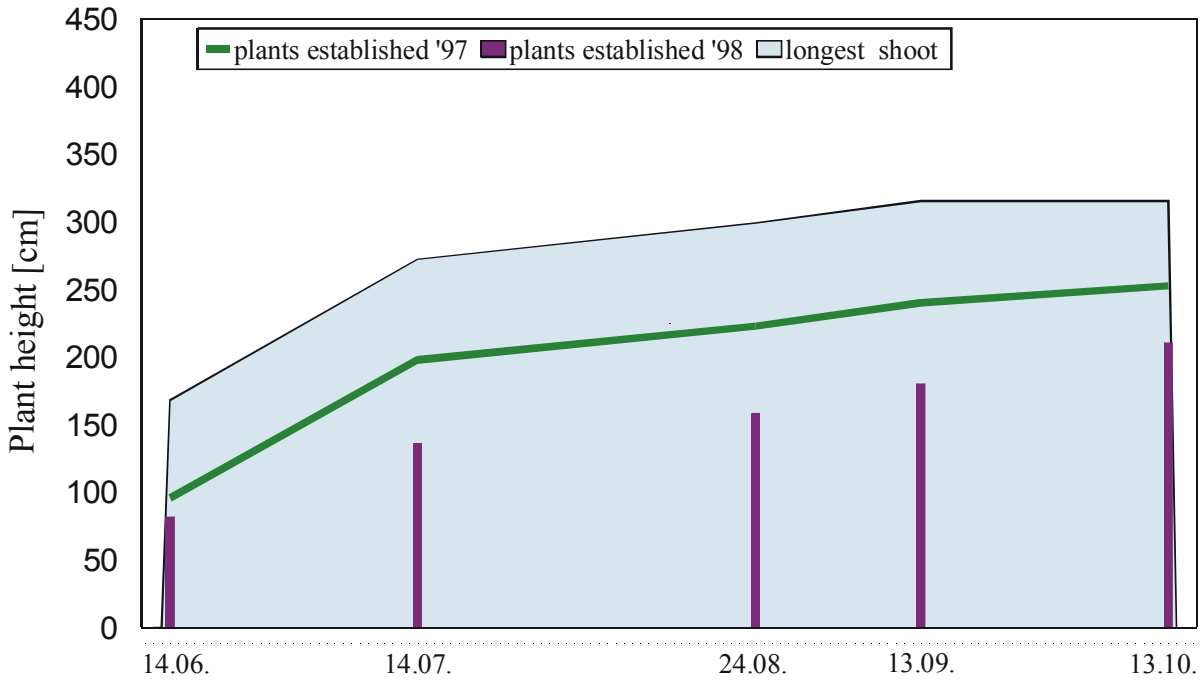
A



Plant height of the Arundo genotype "Rabuiese" during the growing season 1999

FAL-Braunschweig

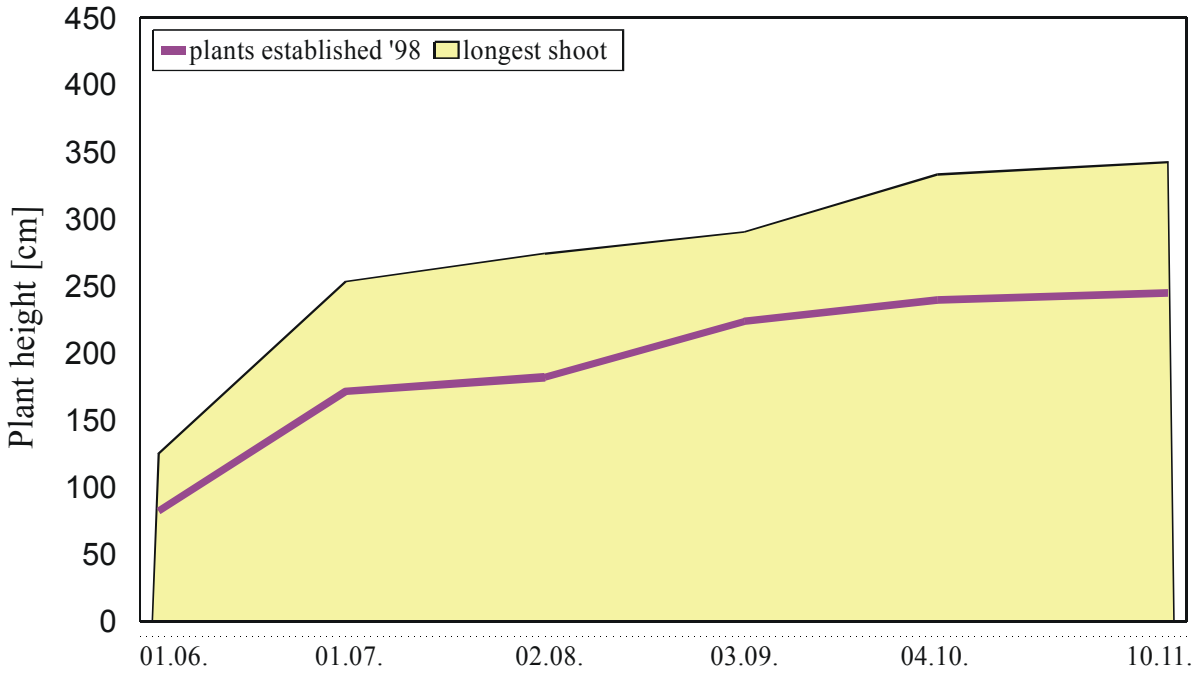
B



Plant height of the Arundo genotype "Rabuiese" during the growing season 1999

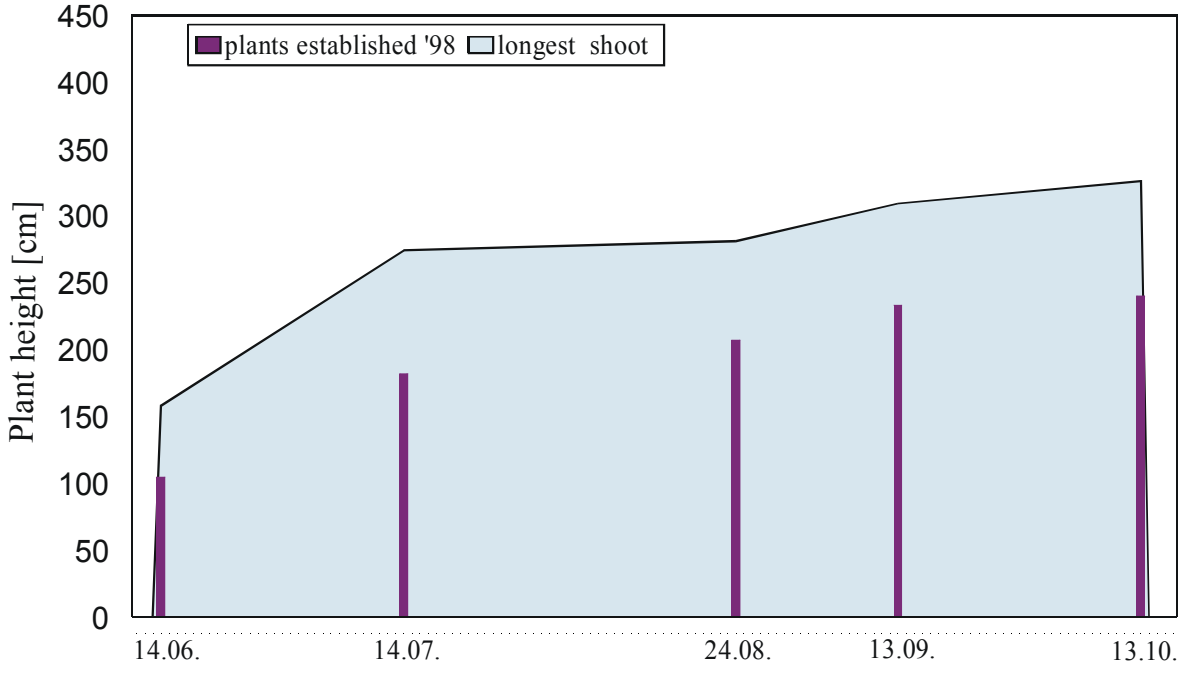
Figure 12: Average height of Arundo donax planted in 1997 and 1998 as well as longest shoot (A: plot centre; B: 4 marked plants chosen across the whole plot)

A



Plant height of the Arundo genotype "Bizet" during the growing season 1999
FAL-Braunschweig

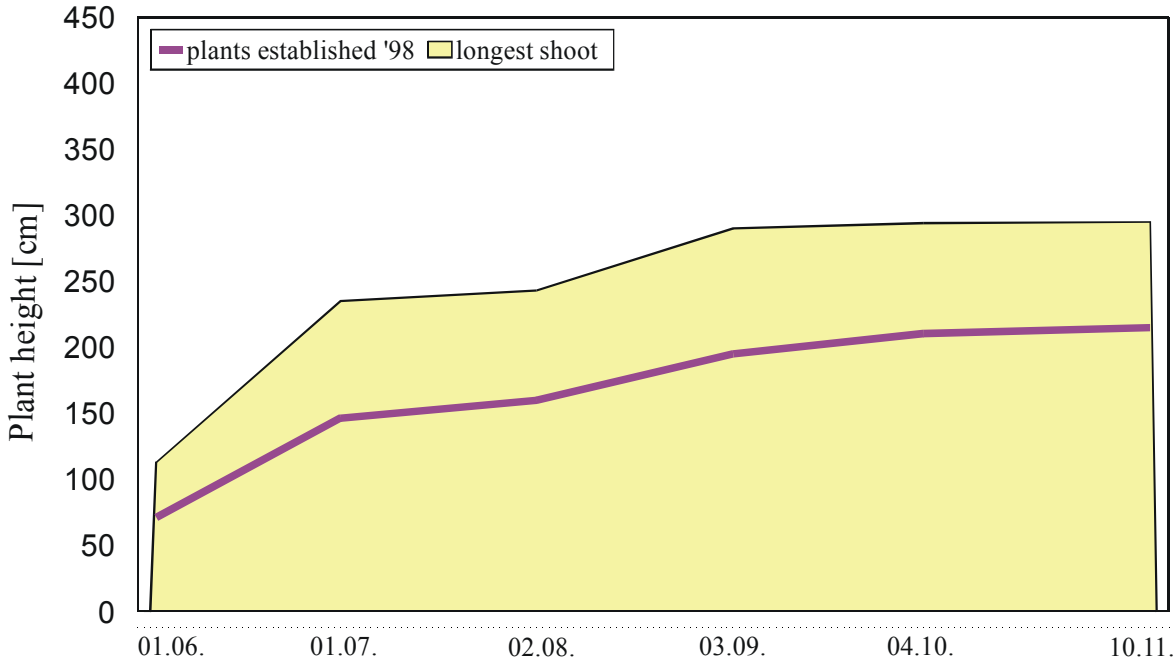
B



Plant height of the Arundo genotype "Bizet" during the growing season 1999
FAL-Braunschweig

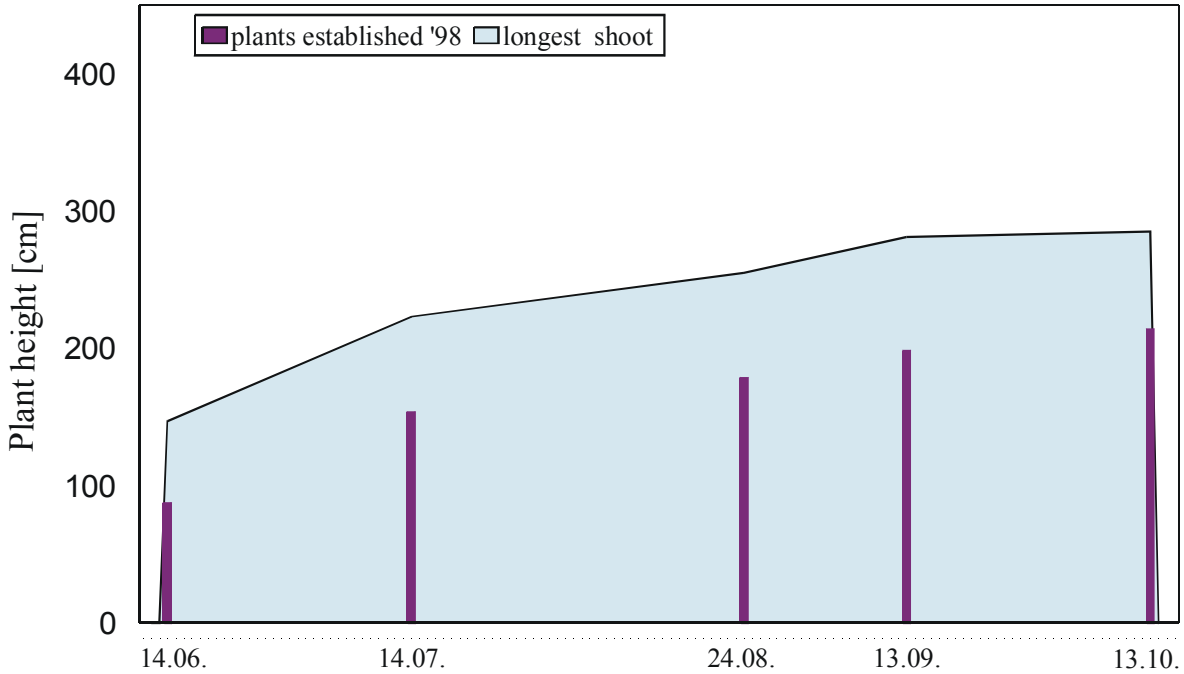
Figure 13: Average height of Arundo donax planted in 1997 and 1998 as well as longest shoot
(A: plot centre; B: 4 marked plants chosen across the whole plot)

A



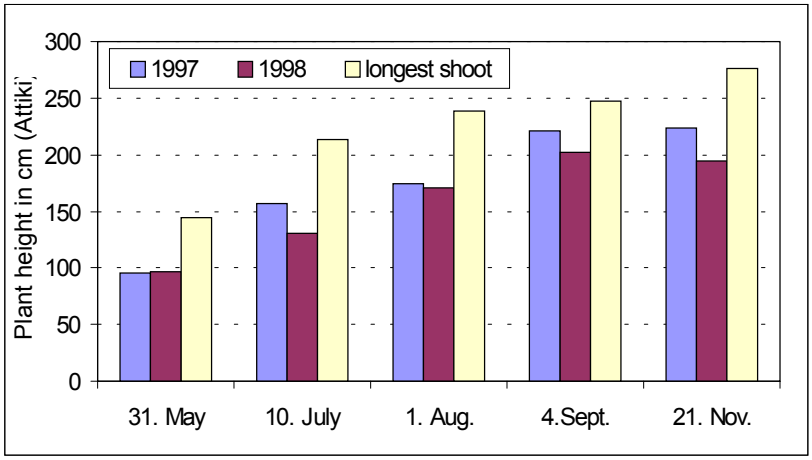
Plant height of the Arundo genotype "Nimes" during the growing season 1999
FAL-Braunschweig

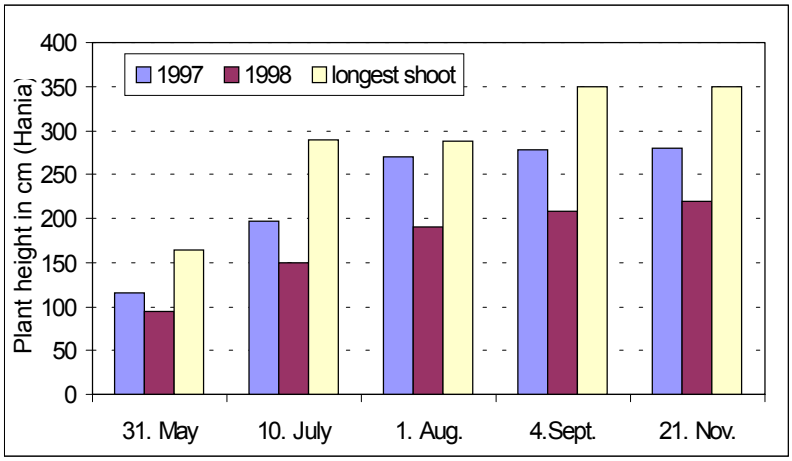
B



Plant height of the Arundo genotype "Nimes" during the growing season 1999

Figure 14: Average height of Arundo donax planted in 1997 and 1998 as well as longest shoot (A: plot centre; B: 4 marked plants chosen across the whole plot)





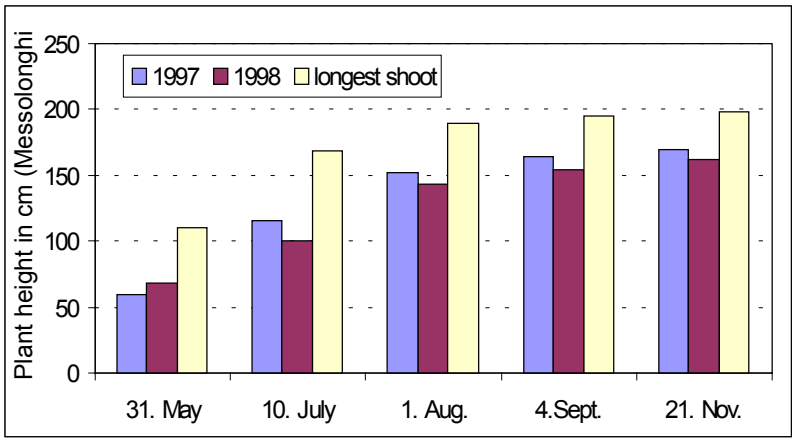
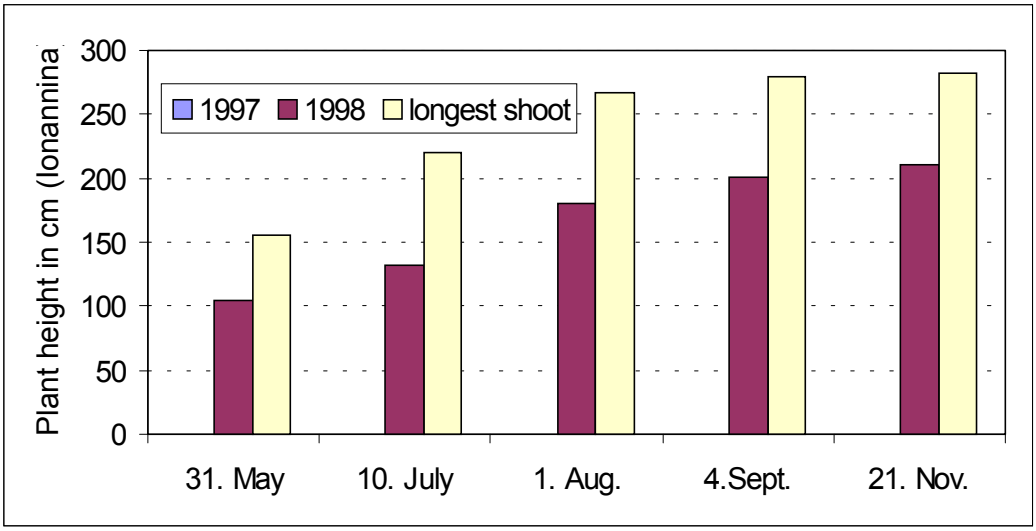
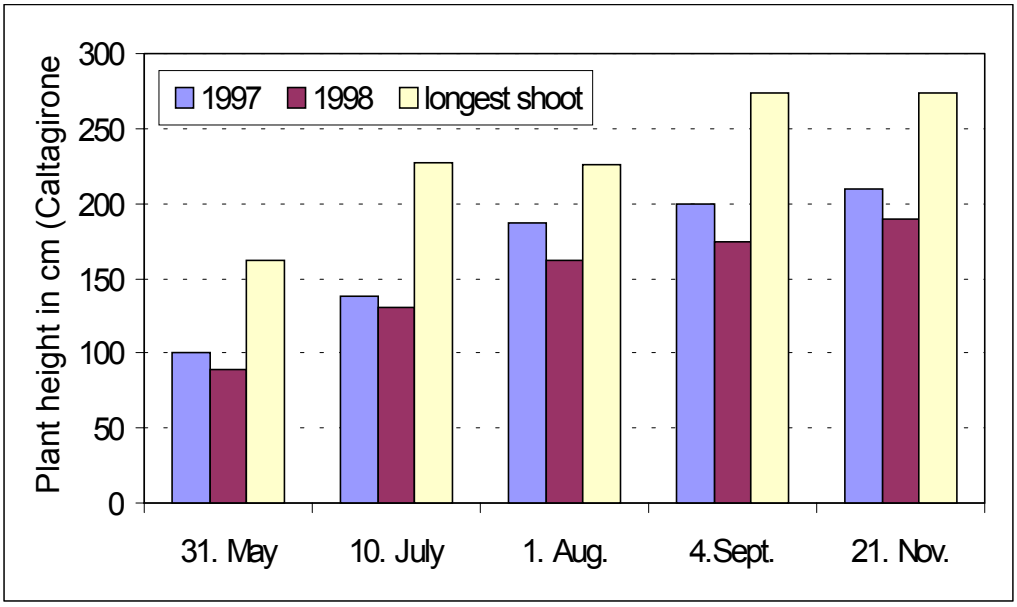


Figure 15: Average plant height of Arundo donax populations established in 1997 and 1998 (year 2000; plot centre)





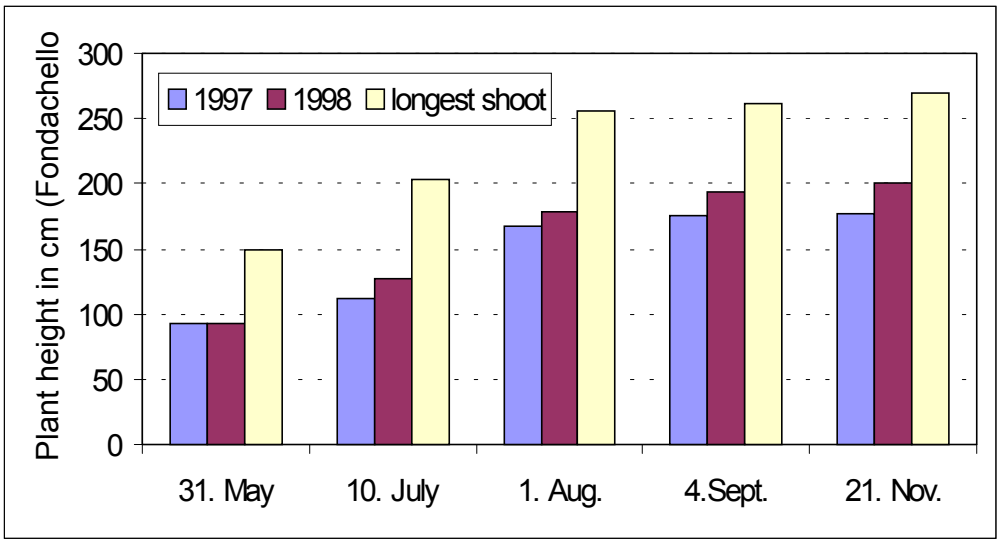
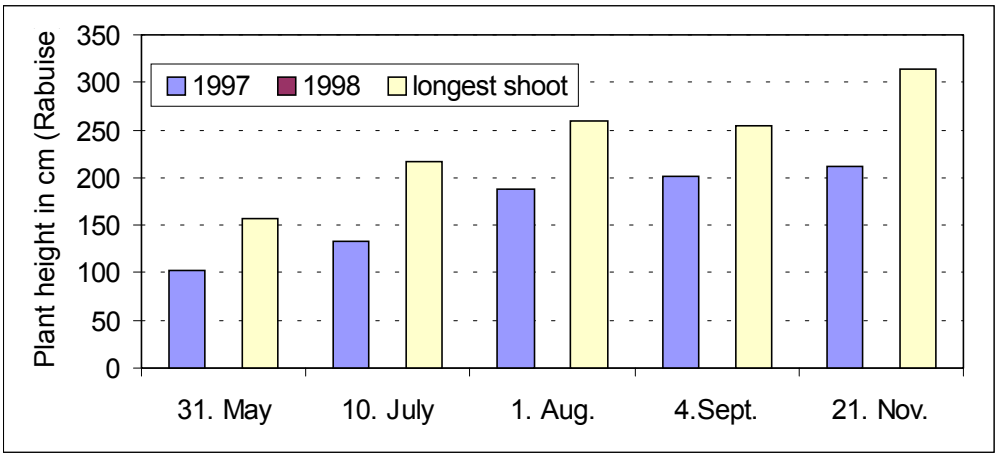
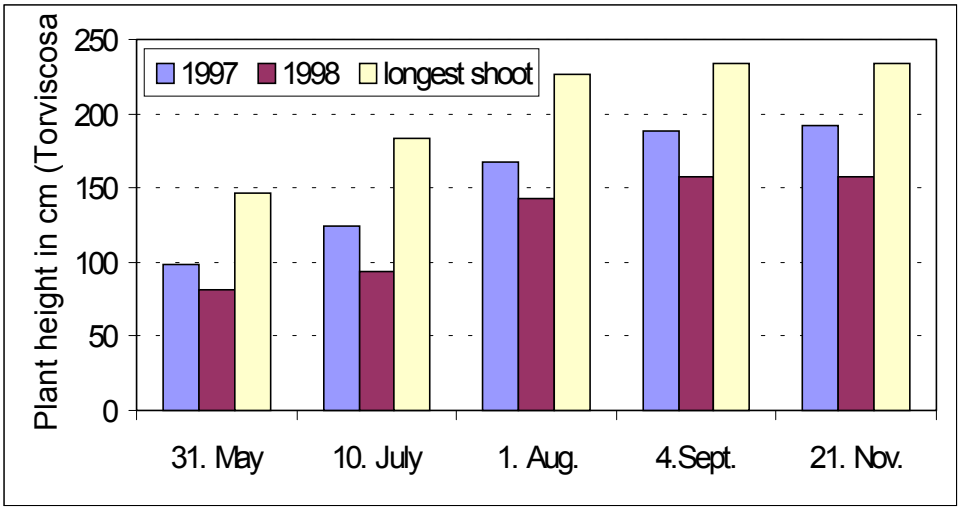


Figure 16: Plant height of Arundo donax established in 1997 and 1998 (year 2000, plot centre)





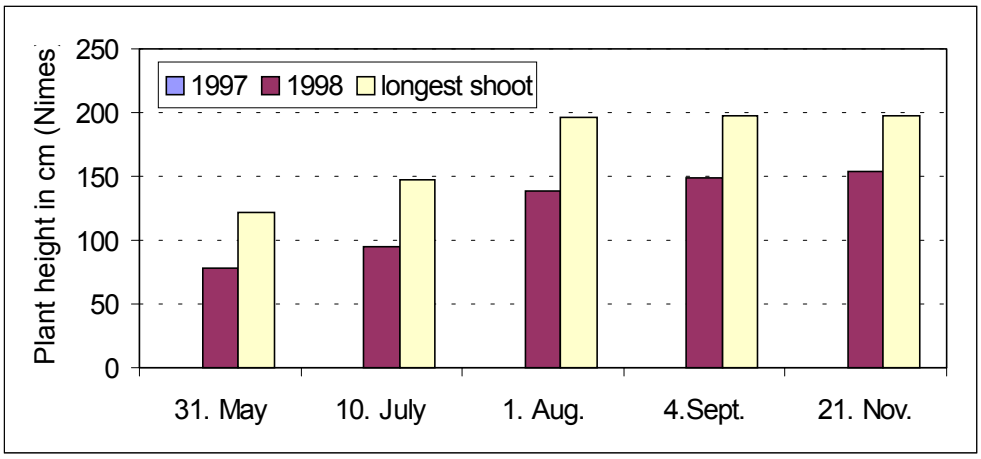


Figure 17: Height of Arundo donax populations planted in 1997 and 1998 (year 2000, plot centre)

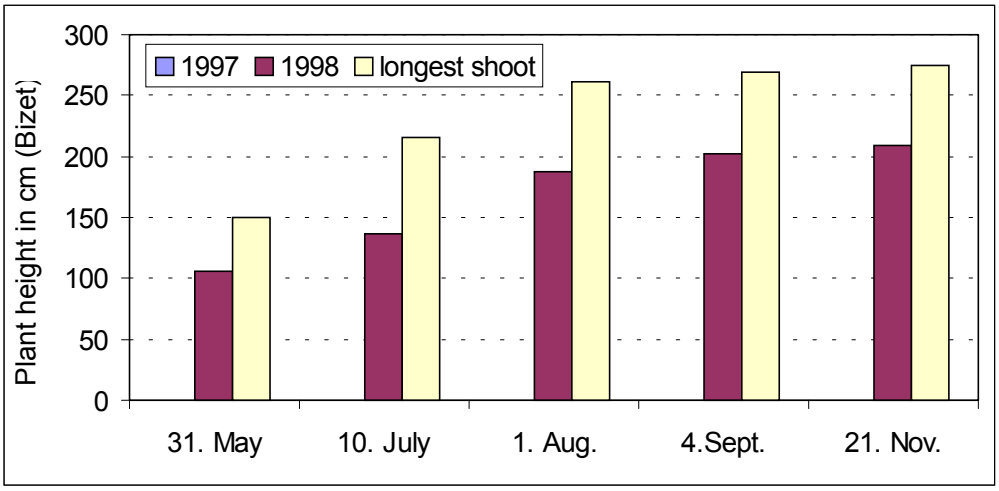


Figure 18: Height of Arundo donax plants established in 1997 and 1998 (year 2000, plot centre)

Table 5: Number of shoots of different Arundo donax populations in 1999 (Plants established in 1997 and 1998)

Population							Average*
Est. 1997	1.6.	1.7.	5.8.	3.9.	5.10.	8.11.	8.11.1999
Attiki	9	10	12	12	14	15	11
Hania	8	9	12	12	13	15	8
Messolonghi	7	7	11	13	13	13	11
Ionannina							8
Caltagirone	6	7	8	10	10	11	9
Fondachello	6	8	8	8	8	8	8
Rabuise	7	9	9	10	11	11	11
Torviscosa	8	8	9	9	10	10	8
Nimes							10
Bizet							10
Average	**7**					**12**	**9**
Est. 1998							
Attiki	5	6	6	7	7	7	
Hania	4	5	5	6	6	7	
Messolonghi	5	7	7	8	8	8	
Ionannina	4	6	7	8	8	8	
Caltagirone	3	5	5	6	7	7	
Fondachello	5	5	6	7	7	7	
Rabuise							
Torviscosa	3	4	4	5	5	5	
Nimes	5	6	7	9	10	10	
Bizet	4	6	7	9	10	10	
Average	**4**					**8**	

* overall average between Plants established 1997 and 1998

Table 6: Number of shoots of different Arundo donax populations in 2000
(Plants established in 1997 and 1998)

Population							Average*
Est. 1997	29. 5.	3. 7.	31. 7.	4. 9.	21. 11.		21. 11.
Attiki	14	18	18	18	18		11
Hania	10	11	11	12	12		9
Messolonghi	14	15	15	15	15		12
Ionannina							8
Caltagirone	8	8	9	9	9		8
Fondachello	4	4	8	8	8		8
Rabuise	11	12	12	13	13		13
Torviscosa	9	10	10	11	11		10
Nimes							7
Bizet							11
Average	**10**				**12**		**10**
Est. 1998							
Attiki	6	6	6	6	7		
Hania	5	6	6	6	6		
Messolonghi	7	7	8	8	8		
Ionannina	6	7	8	8	8		
Caltagirone	5	6	6	6	6		
Fondachello	7	7	7	7	7		
Rabuise							
Torviscosa	7	8	8	8	8		
Nimes	6	6	7	7	7		
Bizet	9	10	10	10	11		
Average	**6**				**8**		

* Average of plants established in 1997 and 1998

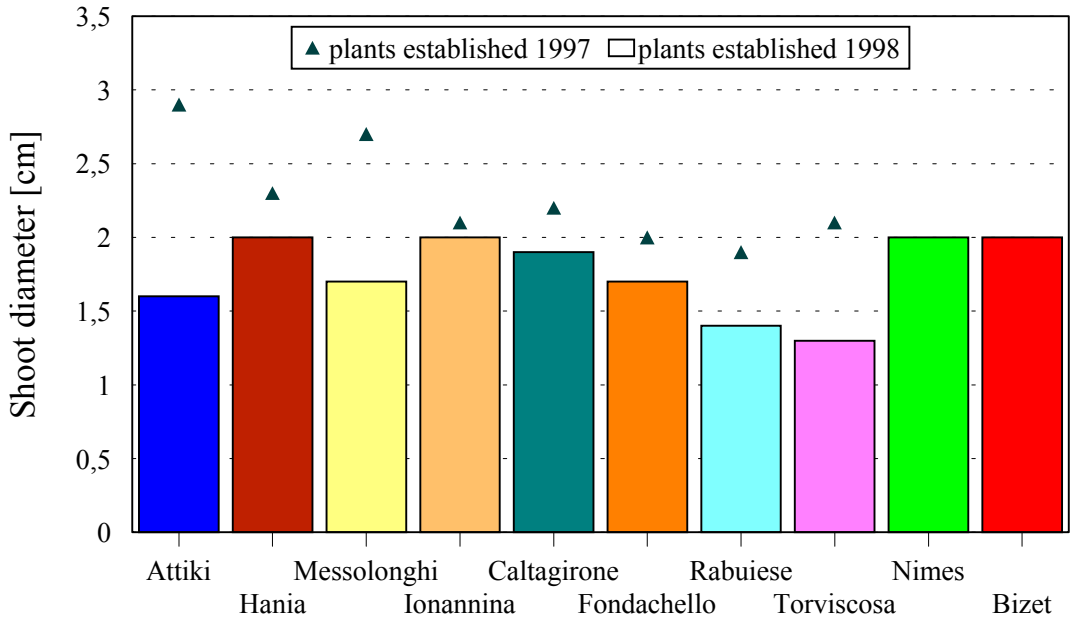


Figure 19: Shoot diameter of Arundo donax populations on 12th June 1998

Table 7: Shoot diameter in cm of different Arundo donax populations in 1999
(Plants established in 1997 and 1998, thickest shoot)

Population							Average
Est. 1997	1. 6.	1. 7.	5. 8.	3. 9.	6. 10.	8. 11.	8. 11.
Attiki	2,6	2,0	2,5	2,7	2,6	2,5	2,3
Hania	2,7	2,0	2,8	3,1	3	2,5	1,8
Messolonghi	2,0	2,3	2,2	2,6	2,6	2,3	2,2
Ionannina							2,0
Caltagirone	2,2	2,3	2,3	2,7	2,7	2,5	2,4
Fondachello	1,9	1,9	1,9	1,8	2,1	2,2	2,0
Rabuise	1,8	1,9	1,9	2,1	2,2	2,1	2,1
Torviscosa	1,9	1,9	1,9	2,0	2,0	1,9	1,9
Nimes							2,0
Bizet							1,9
Average	**2,2**					**2,3**	**2,1**
Est. 1998							
Attiki	2,1	2,0	2,0	2,1	2,1	2,0	
Hania	1,8	2,0	1,8	2,0	2,1	2,0	
Messolonghi	2,2	2,1	2,1	2,2	2,3	2,1	
Ionannina	1,8	1,8	1,8	1,9	2,0	2,0	
Caltagirone	2,1	2,1	2,0	2,3	2,3	2,2	
Fondachello	1,9	1,8	1,8	2,1	2,0	1,8	
Rabuise							
Torviscosa	2,0	1,9	1,9	2,0	1,9	1,8	
Nimes	1,9	1,8	1,8	2,1	2,0	2,0	
Bizet	1,9	1,9	1,8	1,9	1,9	1,9	
Average	**2,0**					**2,0**	

Table 8: Shoot diameter in cm of different Arundo donax populations in 2000
(Plants established in 1997 and 1998, thickest shoot)

Population							Average
Est. 1997	16. 6.	18. 7.	29. 8.	26. 9.		21. 11.	21. 11.
Attiki	2,2	1,7	2,0	2,0		2,6	2,1
Hania							2,0
Messolonghi							2,2
Ionannina							2,0
Caltagirone	2,2	2,1	2,4	2,4		2,4	2,4
Fondachello	1,5	1,5	1,5	1,6		1,7	1,8
Rabuise	1,8	1,6	1,8	1,9		2,0	2,0
Torviscosa	1,8	1,7	1,9	2,0		1,9	1,8
Nimes							1,9
Bizet							1,8
Average	**1,9**					**2,1**	**2,0**
Est. 1998							
Attiki	1,9	1,8	2,0	2,0		1,9	
Hania	1,9	1,8	2,0	2,0		2,0	
Messolonghi	2,1	1,9	2,1	2,1		2,2	
Ionannina	1,7	1,6	1,8	1,8		2,0	
Caltagirone	2,1	1,9	2,1	2,2		2,4	
Fondachello	1,7	1,7	1,8	1,8		2,0	
Rabuise							
Torviscosa	1,9	1,6	1,9	2,0		1,7	
Nimes	1,6	1,5	1,7	1,8		1,9	
Bizet	1,6	1,5	1,7	1,7		1,8	
Average	**1,8**					**2,0**	

Table 9: Leaf number of 4 Arundo donax plants selected across the whole plot in the year 1999 (longest shoot, average of all selected plants)

Population					
	15. 6.	16. 7.	24. 8.	15. 9.	15.10.
Attiki	11	15	20	23	24
Hania	10	17	22	25	29
Messolonghi	11	17	20	25	29
Ionannina	11	16	20	23	26
Caltagirone	11	17	21	24	28
Fondachello	12	17	21	24	29
Rabuise	11	16	20	23	28
Torviscosa	11	15	18	22	24
Nimes	9	15	19	24	26
Bizet	10	16	21	25	27
Average	**10**				**27**

Table 10: Leaf number of 4 Arundo donax plants selected across the whole plot in the year 2000

Population							Average*
Est. 1997	16. 6.	18. 7.	29. 8.	26. 9.		21. 11.	21. 11.
Attiki	13	17	22	22		23	22
Hania							24
Messolonghi							23
Ionannina							22
Caltagirone	15	19	24	25		25	23
Fondachello	12	14	18	19		19	20
Rabuise	14	18	24	24		25	25
Torviscosa	13	17	21	22		22	23
Nimes							21
Bizet							21
Average	**13**					**23**	**22**
Est. 1998							
Attiki	12	15	19	20		20	
Hania	13	18	23	24		24	
Messolonghi	13	17	21	22		23	
Ionannina	13	17	21	22		22	
Caltagirone	14	17	21	21		22	
Fondachello	13	17	20	21		21	
Rabuise							
Torviscosa	14	17	22	23		23	
Nimes	11	14	20	21		21	
Bizet	13	16	20	20		21	
Average	**13**					**22**	

Table 11: Growth height of Arundo donax population and standard deviation in cm

Population	Average			Maximum		
	1998	1999	2000	1998	1999	2000
Attiki	224 ±38	238 ±26	211 ±29	266 ±41	282 ±36	253 ±20
Hania	267 ±36	236 ±75	249 ±45	322 ±51	259 ±70	307 ±56
Messolonghi	180 ±23	196 ±28	166 ±10	212 ±12	231 ±25	193 ±10
Ionannina	214 ±6	248 ±24	211 ±31	244 ±5	283 ±20	251 ±33
Caltagirone	207 ±41	238 ±29	200 ±16	238 ±46	273 ±29	240 ±19
Fondachello	202 ±19	233 ±26	188 ±21	240 ±16	265 ±28	219 ±27
Rabuise	233 ±15	250 ±30	212 ±30	262 ±23	283 ±30	274 ±39
Torviscosa	228 ±10	234 ±9	175 ±16	242 ±8	259 ±12	204 ±17
Nimes	217 ±9	215 ±21	154 ±27	244 ±10	246 ±29	183 ±25
Bizet	223 ±20	245 ±3	209 ±28	255 ±21	274 ±6	239 ±30

Table 12: Dry matter yield in t/ha and standard deviation of populations established in 1998 (average of 3 replications = plots)

Population			
	1998	1999	2000
Attiki	7,0 ±2	10,9 ±2	10,4 ±4
Hania	7,8 ±4	11,6 ±5	11,5 ±5
Messolonghi	7,5 ±3	11,5 ±4	11,5 ±2
Ionannina	7,2 ±3	11,7 ±6	10,3 ±4
Caltagirone*	14,9 ±3	19,8 ±4	20,3 ±7
Fondachello	7,5 ±4	10,9 ±6	10,8 ±6
Rabuise*	14,5 ±1	18,0 ±6	19,1 ±10
Torviscosa*	14,4 ±5	14,5 ±5	11,9 ±1
Nimes	9,7 ±1	12,9 ±3	6,9 ±1
Bizet	9,0 ±2	15,9 ±1	13,6 ±3

* of plants established in 1997

Table 13: Summary of results of micropropagation of Arundo donax populations and genotypes (PICCIPLANT, original page)

	growth response	sterile	in vitro	media	Induction of	Suspension	Induction of		
	in the green - house	induction	rejuventation	optimisation	organogenic callus	culture	shoot clusters	tissue culture	suspension culture
Populations								offspring	offspring
German clones									
A	+++	+++	+++	V	+++	+++	+++	+++	++
G	+++	++	+++	V	+	+	++	+++	+
K	+++	+++	+++	V	+++	+++	+++	+++	++
Messolonghi	+++	+++	++	V	+	++	+++	+	+++
Allithi (Attiki?)	+	+	+	V	0	0	0	+	0
Chama	++	++	+	V	0	0	0	+	0
Pyrgos	+	+	+	V	0	0	0	+	0
Rhizoloum Karditza	+++	++	+	V	++	++	+	+++	+
Pyrus Evron	++	++	++	V	+	+	+	++	+
Fondachello	+	+	+	V	0	0	0	+	0
Torviscosa	+	+	++	V	0	0	0	+	0
Sommuries	+	+	+	V	+	+	++	+	+
Caltagirone	++	++	+	V	0	0	0	+	0
Altedo	++	++	++	V	+	+	+	+	+

+++: very good response; ++ good response; + bad response; 0: no response