Biobased Products and Energy Crops

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Approaches to improve the implementation and expansion of *Miscanthus* production

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Chapter 1 General Introduction

1.1 Background

Nowadays, bioenergy contributes roughly 10% (approximately 50 EJ yr^{-1}) to the world primary energy supply in both traditional and modern utilization ways [1-3]. The traditional bioenergy produces products used in small-scale sector with low conversion efficiency (10-20%), e.g. burning firewood, crop residue and dung cake for residential heating and cooking, while the modern bioenergy is supplied on large-scale in forms of combustible solid biomass (e.g. chips, pellets), liquid biofuels (e.g. bioethanol, biodiesel) and gaseous fuels (e.g. biogas, synthesis gas) [2]. Although the current consumption of modern bioenergy is still small amount (only 10 EJ yr^{-1} [4]), it already grows steadily since mid-2000s. For example, the modern bioenergy consumption in Germany has increased to 0.74 EJ in 2012 from 0.53 EJ in 2006 [5-6].

In last decade, first-generation energy crops, i.e. agricultural crops for energy use (e.g. maize, rapeseed), drove the increase of modern bioenergy [4]. However, due to their food and feed purposes, the production of first-generation bioenergy (i.e. first-generation energy crops generated bioenergy) causes a concern of 'food vs. fuel' conflict. Many analyses [7-10] link the growth of first-generation bioenergy to rising food prices. For example, Baier *et al.* [7] found that the worldwide price of maize and soybean increased, respectively, by 17% and 14% in response to the growth of bioethanol and biodiesel production over the period 2006-2008. Rosegrant [8] modelled the price of maize in 2020 would be 41% higher than that in 2006 under the scenario of 'aggressive first-generation bioenergy growth'. The rising food prices may result in food insecurity as expressed by an increase in the number of undernourished people, who are so poor that they cannot afford the budget of enough safe and nutritious food for a healthy life. The first-generation bioenergy is now more expensive and not economically competitive to fossil fuels [9, 11]. All these concerns stimulate a requirement of producing bioenergy from cheap and abundant non-food materials.

In nature, lignocellulosic biomass makes up the majority of non-food materials. It is therefore of great importance of the lignocellulosic feedstock for future bioenergy production. In general, the lignocellulosic feedstock mainly includes crop residues, woody wastes, by-products and biomass of dedicated lignocellulosic energy crops (later referred to as 'DLE crops') [13]. The DLE crops are grassy or woody plants specifically grown for energy purpose. Although large quantities of crop residues, wood wastes and by-products are currently available for generating bioenergy, these amounts will not satisfy the feedstock demand of future bioenergy industry [14-15]. For example, to meet the German government's target of 11% share of bioenergy in its 2020 gross energy consumption [16], the demand for biomass is expected to be 1.45 EJ yr^{-1}, while the total crop residues, wood waste and by-products only amount to 0.76 EJ yr^{-1} [17]. A possible way to close the gap between feedstock supply and demand is using the biomass of DLE crops. Compared to arable crops, the DLE crops normally have higher utilization efficiencies of light, water and nutrient, and more robust tolerances to environmental stresses [18-19], which can support their productions on non-prime agricultural land. Additionally, the DLE crops are not grown for food/feed purposes. Both indicate the use of DLE crops would reduce the threat of bioenergy production on food security, in particular when they are cultivated on non-arable land. The biomass of DLE crops not only can be burned for thermal application or generating bio-electricity, but also can be fermented for biogas and bio-ethanol. All these indicate an important role of DLE crops for future bioenergy industry.

The lignocellulosic plants are generally categorized as woody or grassy (herbaceous). Compared to woody plants, the grassy plants get more interests for bioenergy use. There are two reasons mainly responsible for this: firstly, the grassy plants are more appropriate as feedstock for bio-ethanol (the most favoured bioenergy type) fermentation [20]; secondly, as the main arable crops are also herbaceous, switching current agricultural practices to produce grassy plants is easier [21]. Despite a wide range of grass species are available for bioenergy use, not all of them can meet the industry requirements for both good quality and high yield. According to projects of screening grasses for bioenergy production in the USA and Europe, perennial reed canary grass (*Phalaris arundinacea* L.), giant reed (*Arundo donax* L.), switchgrass (*Panicum virgatum* L.) and miscanthus (*Miscanthus* spp.) were selected out as promising dedicated grassy lignocellulosic energy crops (later referred to as 'DGLE crops') [22]. Unlike annual grasses, productions of the perennial DGLE crops only need soil tillage in the establishment year. Without long periods tilling, soil erosion risk will be reduced. Furthermore, rhizome systems of the perennial DGLE crops can recycle the nutrients, which can reduce the nutrient inputs for their productions. Within perennial grasses, reasons for the selection of above four DGLE crops mainly include their high and reliable productivities across a wide range of environmental conditions, low production requirements, good energy-related qualities and broad genetic variability [22]. Reed canary grass is a cool-season (C3) grass and adapts well to low temperature, while this is not the case for the warm-season (C4) miscanthus and switchgrass. Therefore, in cold regions, reed canary grass shows higher yield and safer overwintering than miscanthus and switchgrass [22-23]. Similarly, in dry regions without irrigation (e.g. Mediterranean region), giant reed performs better than miscanthus and switchgrass due to its higher water use efficiency [24]. With these two exceptions, the above two promising C4 DGLE crops especially the miscanthus normally have higher biomass yields, higher nutrient and water use efficiency than that of the C3 reed canary grass and giant reed [22, 24-25]. Therefore, researches and commercial utilization of lignocellulosic plants for bioenergy production focus on miscanthus in most European countries.

1.2 What is *Miscanthus*?

Miscanthus is a genus of perennial, rhizomatous, giant grass that belongs to the subtrib *Saccharinae Grisebach* of the tribe *Andropogoneae* in the family of *Poaceae*. It originates from East Asia and now has a worldwide distribution from tropical Southeast Asia to temperate Europe [26-28]. As a genus with subtropical origin, miscanthus has C4 photosynthetic pathway and requires warm season, short-day condition for growing and flowering [29-30]. According to the taxonomic classification in 'Flora in China' [27], worldwide, there are 14 species included in the miscanthus genus with a basic chromosome number of 19; however, only four species are of interests for biomass production [30], namely *Miscanthus sinensis* Andersson, *Miscanthus sacchariflorus* (Maxim.) Hackel, *Miscanthus lutarioriparius* Liu S. L. Chen and *Miscanthus floridulus* (Lab.) Warburg ex Schumann et Lauterbach. In addition, a natural hybrid *Miscanthus* × *giganteus* Greef et Deuter is already commercially used for biomass production in

Europe [31]. In the following sections, descriptions of above four promising species and the natural hybrid *M. × giganteus* will be provided, including the botanical, morphological characteristics and yield potentials (Table 1.1). [27-28, 30, 32-34]

M. sinensis is a perennial grass growing in temperate regions with thin, densely tufted stems and short rhizomes [27-28]. Stems are 0.5-4.0 m tall and 3-10 mm in diameter near the base [30]. Each stem is made up of a series of alternating nodes and internodes. The internodes have hard cortex and soft pith. The majority of internode is closely surrounded by hairy leaf sheath. Leaf blades are linear with length of 20-140 cm and width of 0.5-1.2 cm [28, 30]. Leaf has serrated margin and apparent white midrib. Panicle includes 10-100 racemes, which are 10-30 cm long. Spickelets have awns which are 3-13 mm long and are not easy to fall off from panicles after mature. In the climatic condition of Hunan China, the flowering time is in July to December and peaks at middle October [30, 33]; the annual aboveground biomass yield vary in a range of 5.1-24.0 odt (oven dried ton) ha^{-1} [30]. Rhizomes are thin-stemmed and short.

M. sacchariflorus is chiefly growing in temperate and cold-temperate regions with erect, thick and widely spreading stems [27-28]. Stems are 0.6-3.0 m tall and 5-10 mm in diameter near the base [28, 30]. Nodal buds or branches are enclosed between culm and sheath on lower nodes (1st-5th node from the bottom). The lanceolate leaf blades are 19-85 cm long and 0.4-2.0 cm wide with rough margins. Panicle is slightly pendulous and consists of 12-60 racemes on a short axis. Also in Hunan, the flowering time is in September to October; and annual biomass yield can reach 3.0-13.5 odt ha^{-1} or more [30, 34]. Rhizomes are strong, long, creeping and covered by scale-like sheath blades.

M. lutarioriparius is an endemic species in Middle-east China and mainly grows in the costal area around Dongting Lake [27-28, 30]. Plants are generally characterized to be tall and woody. The normal stems are 4-7 m tall with a stem-base diameter of 15-30 mm [30]. Stems are hollow and easily branching. Nodal buds and aerial roots are normally enclosed on basal stems. Leaves and panicles are similar to that of *M. sacchariflorus.* In Hunan conditions, its flowering time is in October to November and annual biomass yield varies in the range of 10.5-33.0 odt ha^{-1} [30]. Rhizomes are also strong, long, creeping and mainly distribute close to soil surface with a depth of 5-20 cm.

Table 1.1 Comparison of ecological demands, morphological characteristics and biomass yield potentials of the four promising miscanthus species (*Miscanthus sinensis* Andersson, *Miscanthus sacchariflorus* (Maxim.) Hackel, *Miscanthus lutarioriparius* Liu S. L. Chen & *Miscanthus floridulus* (Lab.) Warburg ex Schumann et Lauterbach) and the single commercial hybrid clone *Miscanthus × giganteus* Greef et Deuter.

	M. sinensis	*M. sacchariflorus*	*M. lutarioriparius*	*M. floridulus*	*M. × giganteus*
Ecological demands	Temperate climate Not tolerant to prolonged flooding and drought periods	Temperate & cold-temperate climate Not tolerant to prolonged flooding and drought periods	Warm-temperate climate Tolerant to 2-3 month flooding	Tropical, subtropical & warm-temperate climate Not tolerant to prolonged flooding and drought periods	Tropical, subtropical & warm-temperate climate Not tolerant to prolonged flooding and drought periods
Morphological characteristics	Thin and densely tufted stems with a plant height of 0.5-4.0 m and base diameter of 3-10 mm	Erect and widely spreading stems with a plant height of 0.6-3.0 m and base diameter of 5-10 mm	Thick and widely spreading hallow stems with a plant height of 4-7 m and base diameter of 15-30 mm	Loosely tufted stems with a plant height of 1.5-4.7 m and base diameter of 6-15 mm	Thick and widely spreading stems with a plant height of 2.5-3.5 m
Flowering time	July to December	September to October	October to November	June to August	No information
Biomass yield (odt ha^{-1} yr^{-1})	5.1-24	3-13.5	10.5-33	6-31	No information
Citation	[27-28, 30-33]	[27-28, 30-32, 34]	[27-28, 30-32]	[27-28, 30-32, 34]	[27-28, 30-32, 35-36]

Note: Flowering time and biomass yield (yield of aboveground biomass harvested in autumn) were both measured in the climatic condition of Hunan China.

M. floridulus is a species preferring warm condition and mainly grows in tropical, subtropical and warm-temperate regions [27-28, 30]. Stems are loosely tufted, pith filled, 1.5-4.7 m tall and 6-15 mm in diameter near the base [27-28, 30]. Stem surface is covered by wax powder, in particular at the base of leaf blade. Leaf blade is flat, broad and drooping with a length of 50-104 cm and width of 0.7-5.0 cm [30]. Leaf blades do not fall off and still keep green (i.e. evergreen) through the whole winter period. The panicle is 30-50 cm long and consists of more than 100 racemes that are 10-20 cm long. The flowering time in Hunan is from June to August. The annual aboveground biomass yield is 6.0-31.0 odt ha^{-1} [30, 34]. Rhizomes are thick but short.

M. × *giganteus* is a natural allotriploid involving diploid *M. sinensis* and tetraploid *M. sacchariflorus* [35]. It was firstly found in Japan, but now mainly distributed in Europe [28]. Recently, it has been introduced to many countries as a promising energy crop, e.g. China, the USA, also including its origin Japan [36]. The widely spreading stems are not very tall yet strong with a plant height of 2.5-3.5 m. Stem nodes are without hairs. Branches and root primordia at lower nodes are sometimes observed [28]. Leaf blades are linear with a length of approximately 50 cm and width of 3.0 cm. After flowering, the inflorescence becomes fan-like panicle with a length of 30 cm. However, in long-day regions, it does not flower [29]. The belowground system is extensive, including top layer rhizomes (mainly in the upper 0-25cm soil) and deep roots (even to 2 m depth). Rhizomes are often oval to round in diameter and only slightly creepy. The full establishment generally takes 3-5 years. After that, a stable biomass yield level can be kept for another 15 years with an average spring harvest yield up to 25 odt ha^{-1} in central European conditions and up to 30 odt ha^{-1} in southern European conditions [22, 24]. Additionally, as a sterile species with no seed production, *M.* × *giganteus* is evaluated to be non-invasive [25].

Table 1.2 Photosynthetic characteristics, reported biomass yield (annual aboveground biomass yield), typical biomass yield, lower heating value and typical energy yield of four promising dedicated lignocellulosis crops in Europe.

Crops	Latin Name	Photosynthetic pathway	Reported biomass yield (odt ha^{-1} yr^{-1})	Typical biomass yield (odt ha^{-1} yr^{-1})	Lower heating value (MJ kg^{-1})	Typical energy yield (GJ ha^{-1} yr^{-1})	Citation
Miscanthus	*Miscanthus × giganteus*	C4	2-49	20-25	17.6	352-440	[22, 36-40]
Switchgrass	*Panicum virgatum* L.	C4	5-23	12-18	17.0	204-306	[22, 41-42]
Giant reed	*Arundo donax* L.	C3	3-37	15-20	16.8	252-336	[22, 43-44]
Reed canary grass	*Phalaris arundinacea* L.	C3	7-13	10-12	16.4	164-197	[22, 45-48]

1.3 Advantages of miscanthus as dedicated lignocellulosic energy crops

1.3.1 High yield potential of biomass and energy

Among the four promising DGLE crops, miscanthus typically shows the highest yield potential of both biomass and energy (Table 1.2). In European conditions, the aboveground biomass yields harvested in autumn (later referred to as 'biomass yield') of above three promising miscanthus species (without *M. lutarioriparius*) and the commercial clone *M.* × *giganteus* were observed to vary in a wide range of 2-49 odt ha^{-1} yr^{-1}, depending on species, genotypes, environmental conditions and management practices [22, 36-40]. *M.* × *giganteus* generally produces the highest biomass yield among all the species and genotypes and then its biomass yield is used for the comparison with other crops. In some cases, the miscanthus biomass yield may be lower than that of the other three promising DGLE crops (switchgrass, giant reed & reed canary grass). For example, in Denmark, reed canary grass can yield more biomass than *M.* × *giganteus* (9 vs. 5 odt ha^{-1} yr^{-1}) because the cold winter there kills most miscanthus plants [49]; in central Italy, giant reed has higher biomass yield than *M.* × *giganteus* (37.7 vs. 28.7 odt ha^{-1} yr^{-1}), which is benefited from the high water use efficiency of giant reed [50]. However, a typical biomass yield (the stable yield in a good but not poor or excellent condition) of 20-25 odt ha^{-1} yr^{-1} is reached by the well-established *M.* × *giganteus* in long periods [38-39]. In contrast, for switchgrass, giant reed and reed canary grass, which are also growing in Europe, a lower typical biomass yield than miscanthus is found to be 12-18, 15-20 and 10-12 odt ha^{-1} yr^{-1} (Table 1.2), respectively.

For energy yield potential, the typical calorific value (here expressed by the lower heating value-LHV) of miscanthus (17.6 MJ kg^{-1}) is also higher than that of switchgrass (17.0 MJ kg^{-1}), giant reed (16.8 MJ kg^{-1}) and reed canary grass (16.4 MJ kg^{-1}) (Table 1.2). Together with the highest biomass yield potential, miscanthus produces the highest energy yield among the four promising DGLE crops. Taking the typical biomass yield shown in Table 1.2, the energy yield of miscanthus grown in European conditions is estimated to be 352-440 GJ ha^{-1} yr^{-1}, which is approximately 26%, 36% and 55% higher than that of giant reed, switchgrass and reed canary grass, respectively (Table 1.2).

1.3.2 Low input requirements for production

Typically, agronomic practices summarized in Table 1.3 are recommended for the production of maize (the representative first-generation energy crop) and the above four promising DGLE crops. Compared to the other four crops, miscanthus production generally requires less agronomic input items and application rate (per hectare) of each item.

For crop production, land preparation is generally considered essential for good establishment, easy crop management and high yield. Therefore, the land preparation (e.g. ploughing, harrowing) is also recommended for the productions of above four promising DGLE crops (Table 1.3), but only in the first year because they are perennial. In contrast, production of the annual maize needs the land preparation at the beginning of each growing season. Compared to miscanthus, productions of seeded switchgrass and reed canary grass require more times harrowing and rolling for a better seedbed [56-58]. Despite more planting material (per hectare) is required by miscanthus, this high input can be compensated by its longer life-years (see Table 1.3) and higher biomass yield than the other three DGLE crops.

Following planting/sowing, irrigation and weed control are important for good establishment. For the seeded maize, irrigation application after sowing is mostly recommended in each growing season, especially in southern Europe [51]. In contrast, for switchgrass and reed canary grass, irrigation is only recommended in the establishment year because at that time their small seeds do not contain enough water for germination and seedlings' roots cannot take up water from deep soil for continuous growth [56-58]. For miscanthus cultivation, irrigation is only applied in the first year when plantlets are transplanted or direct planting rhizomes in dry condition (e.g. southern Europe) [54]. With respect to weed control, it is necessary in the initial phase of crops' establishment because the initial short and weak plants compete poorly with uncontrolled weeds for light, water, nutrients and space. However, from the second or third year onwards, the high and dense canopy of miscanthus can suppress the weed interference; therefore, weed control is not recommended from then on [53-55]. In contrast, weed control for maize production is needed in every growing season [51].

Table 1.3 Agronomic practices typically recommended for producing the representative first-generation energy crop maize (*Zea mays* L.) and the most promising dedicated lignocellulosic energy crops miscanthus (*Miscanthus* spp.), switchgrass (*Panicum virgatum* L.), giant reed (*Arundo donax* L.) and reed canary grass (*Phalaris arundinacea* L.).

Items	Maize	*Miscanthus*		Switchgrass		Giant reed		Reed canary grass	
	1st – 20th year	1st year	2nd - 25th year	1st year	2nd - 15th year	1st year	2nd-20th year	1st year	2nd - 20th year
Land preparation	For every year: Ploughing once + Harrowing once + Rolling once	Ploughing once + Harrowing once	Ne	Ploughing once + Harrowing twice + Rolling twice	Ne	Ploughing once + Harrowing once	Ne	Ploughing once + Harrowing twice + Rolling twice	Ne
Planting	20-25 kg seeds ha^{-1}	16,000 rhizomes ha^{-1}	Ne	8-10 kg seeds ha^{-1}	Ne	12,500 rhizomes ha^{-1}	Ne	15-20 kg seeds ha^{-1}	Ne
Irrigation	0-900 m^{3} ha^{-1} yr^{-1}	0-600 m^{3} ha^{-1} yr^{-1}	Ne	600 m^{3} ha^{-1} yr^{-1}	Ne	900-1400 m^{3} ha^{-1} yr^{-1}	500-800 m^{3} ha^{-1} yr^{-1}	600 m^{3} ha^{-1} yr^{-1}	Ne
Weeding	For every year: Applying pre-emergent herbicide once and post-emergent herbicide once/twice	Applying pre-emergent herbicide once and post-emergent herbicide once	Ne	Applying pre-emergent herbicide once and post-emergent herbicide once	Ne	Applying pre-emergent herbicide once	Ne	Applying pre-emergent herbicide once	Ne
Fertilization	100-230 kg N ha^{-1} yr^{-1} 70-110 kg P ha^{-1} yr^{-1} 185-265 kg K ha^{-1} yr^{-1}	Ne	For 2nd -4th year: 50-70 kg N ha^{-1} yr^{-1}	Ne	50-100 kg N ha^{-1} yr^{-1}	100 kg N ha^{-1} yr^{-1}	100 kg N ha^{-1} yr^{-1}	40-50 kg N ha^{-1} yr^{-1} 15 kg P ha^{-1} yr^{-1} 50 kg K ha^{-1} yr^{-1}	50 kg N ha^{-1} yr^{-1} 5 kg P ha^{-1} yr^{-1} 20 kg K ha^{-1} yr^{-1}
Pest & Disease control	Applying fungicide with seed treatment + Applying soil insecticides + Spraying insecticides	Ne	Ne	Sometimes applying bactericide for controlling rust	Sometimes applying bactericide for controlling rust	Sometimes applying insecticide for controlling *Sesamia* spp.	Ne	Sometimes applying pesticide for controlling aphids	Ne
Citation	[51-52]	[22, 53-57]		[56-58]		[22, 56]		[22, 56-57]	

Ne-not needed.

Although fertilizer requirements of any crop vary with soil type and soil fertility, typically, the fertilizer requirements of miscanthus production are less than half that of the maize production (Table 1.3). There are two main reasons for the low fertilizer requirements by miscanthus: firstly, due to its perennial characteristics, nutrients in above-ground biomass can be annually recycled to below-ground rhizome after mature and then reused in the following growing season, i.e. nutrients re-translocation [59]; and secondly, its extensive and deep root system can take off nutrients from a deep and large area of soil [53]. In addition, in the establishment year, no fertilizer application is recommended for the productions of miscanthus and switchgrass because the soil nutrients should be sufficient for the nutrient offtakes by their small and short one-year-old plants; and fertilization (mainly nitrogen) will promote weed growth, i.e. increase the weed interference [53-54]. In the subsequent life-years, nitrogen application is mostly only recommended between the second and fourth growing seasons for miscanthus. That is because after the full establishment (i.e. from the 4th growing season onwards), miscanthus can host nitrogen-fixation organisms that can balance the nitrogen input requirements [60]. To date, no insect pests and diseases are found to seriously infest plant growth and reduce biomass yield of miscanthus [53, 61]. It is therefore not necessary to apply the pest and disease control during the miscanthus production. In contrast, a high number of pests and diseases may damage the plant growth of maize and result in significant yield reduction [51]. For example, if the European corn borer (*Ostrinia nubilalis* Hbn.) is not controlled, a yield loss of 5-30% can happen for the maize production in Europe [52]. Therefore, the pest and disease control is of great importance and essential during the maize production. Additionally, rust, *Sesamia* spp. and aphid have been found affecting the production of switchgrass, giant reed and reed canary grass [56-58]. To reduce their damage risks, there is a possible demand of pests and diseases control in the production of switchgrass, giant reed and reed canary grass.

1.3.3 Environmental benefits

Effects of miscanthus production on environmental aspects are various and not yet adequately understood. Several studies [62-66] have been conducted to review and evaluate environmental impacts of miscanthus plantation and their results show both detrimental and positive impacts. Critically, large-scale conservation of land use for miscanthus production may disrupt regional hydrologic cycles especially in dry areas [62-63]; the conversion of natural forest for cultivating miscanthus may cause deforestation [12]; and miscanthus establishment with tillage can immediately cause soil erosion, soil carbon release and nutrient loss [62-66]. Even though, based on current knowledge, miscanthus production in long periods has more beneficial than harmful impacts on environmental aspects [62, 64]. It is summarized that the production of miscanthus has outstanding positive effects on GHG mitigation, soil quality and biodiversity [63-66].

Theoretically, the CO_2 released by miscanthus derived bioenergy equal that absorbed by biomass during photosynthesis, i.e. carbon neutral - net carbon emission equal zero. Then the theoretical GHG mitigation potential by miscanthus-based bioenergy is equivalent to the sum of GHG emissions that produced by the combustion of substituted fossil energy. However, in fact, miscanthus biomass production and conversion require the inputs of energy (e.g. fossil fuels) and production materials (e.g. fertilizer, herbicide), both of which have net GHG emissions [66]. The miscanthus plantation can also sequester carbon into soil by below-ground rhizomes and roots with an annual amount of approximately 4% of the biomass yield [67-70]. This sequestrated carbon can balance the carbon losses by the production inputs. Therefore, production and utilization miscanthus biomass have the potential of saving GHG emissions. However, the achievable GHG mitigation potential of miscanthus will vary, heavily depending on biomass production system and conversion technology that used [65]. Meyer & Lewandowski [71] found that the per-hectare GHG savings of miscanthus biomass used for bioethanol production is generally higher than that for combustion because higher per-hectare yield is available for processing into bioethanol than combustion (delaying the harvest to winter for combustion use reduces the yield by about 25% [22]). In the case of burning miscanthus for heat and electricity, the potentially per-hectare GHG savings of aboveground biomass is estimated to be 31.7 t $CO_{2\ eq}$ ha^{-1} yr^{-1} (or 90.1 kg $CO_{2\ eq}$ GJ^{-1} yr^{-1}) based on the typical biomass yield of 20 odt ha^{-1} yr^{-1} [71].

Concerning soil quality, the short-term effects of miscanthus cultivation is dependent on the initial land use. When cultivating miscanthus on arable land, the establishment generally has no net short-term effect on soil quality because similar agricultural practices as arable crops are used [72]. While converting grassland to miscanthus establishment, the agricultural practices (e.g. ploughing, fertilizing) will immediately result in losses of organic carbon and nitrogen [72-74] and increase the soil erosion risk [66]. However, the long-term miscanthus cultivation is expected to have benefits of enhancing soil organic matter (SOM) content, improving soil texture and reducing soil erosion risk [62-64]. Due to the absence of annual tillage (the most damaging on SOM content) and high input of residues (fallen leaves and senescent rhizomes/roots), the productive miscanthus can potentially enhance the SOM accumulation. In Germany, Kahle *et al.* [69] found that the 4-9 year old miscanthus plot could accumulate 11.7 t ha^{-1} yr^{-1} more SOM than the neighbouring grassland. The SOM components (e.g. lipids, sterols) play an important role in soil aggregate formation and stability [69]. Therefore, the soil texture can be improved along with the enhancement of SOM by miscanthus cultivation [74]. Additionally, the reduced machinery use by miscanthus production compared with annual cropping system are likely to result in benefits of improving soil structure. After full establishment, miscanthus field could exhibit lower erodibility potential than annual crops field owing to the stronger soil conservation ability by extensive rhizome, deep roots and greater interception of rainfall by large canopy [75]. Smeets *et al.* [66] predicted a reduction of soil erosion rate from 10.5 t ha^{-1} yr^{-1} to 2.6 t ha^{-1} yr^{-1} when grain maize were replaced by miscanthus in the condition with annual precipitation of 400 mm.

Similarly, effects of miscanthus cultivation on biodiversity are also dependent on time horizon (i.e. short-term and long-term effect) and initial land use [62, 76]. In a short-term after planting, miscanthus establishment is expected to have no net effect on fauna biodiversity compared to annual food crops because they use similar agricultural practices (also the main cause for biodiversity losses) in the establishment phase. In contrast, a higher diversity of the ground flora between rows was found in miscanthus field in the first three years after planting when the canopy is not closed [77]. In a long period, the miscanthus production is under a low agrochemicals input and less disruption system than that of annual food crops (Table 1.3). What's more, leaves falling [22] creates an increased litter layer in the miscanthus field. Both create an optimum condition for the development of soil micro-organisms and soil faunas (e.g. earthworm) [78-79]. Also in a long term, an increased diversity of wildlife populations (e.g. insects, birds and mammals) was detected in the miscanthus field [77, 80] compared to the annual crop field. This can be attributed to the great ground cover diversity and long-standing period of miscanthus plants, both of which provide a natural shelter for the animals, especially in winter. When converting initial biodiversity-rich ecosystems (e.g. grassland) to miscanthus cultivation, the establishment disturbance would result in biodiversity losses immediately [62]. However, due to the above benefits of miscanthus cultivation on biodiversity increasing, a new biodiversity equilibrium with increased species is still expected, but needs confirmation by long-term experiment.

1.3.4 Diverse genetic resources for breeding and genetic improvement

Worldwide, especially in East Asia (the region of miscanthus origin), miscanthus is found to include diverse genetic resources (i.e. wide genetic diversity) at both phenotypic and molecular levels [30, 33, 81-83]. In China -- the distribution and diversity centre of miscanthus, seven species are found distributing widely at both latitudinal (18°-47° N) and altitudinal (0-3600 m above sea level) level [30]. Also, a wide range of ecological adaptation was found by natural population, e.g. from the dry Loess Plateau to seasonal flooding Dongting Lake area, from subtropical Hainan Island to cold temperate Northeast Plain. Through analyzing data of 388 *M. sinensis* accessions collected across China, vast phenotypic diversity (e.g. variation of 15 agronomic traits of 20.8-82.8%) and genetic diversity (e.g. Shannon diversity index of 0.020-1.522) were both identified [33]. These resources compose a valuable gene pool for miscanthus breeding and genetic improvement. The rich genetic resources give a possibility to select varieties directly from wild germplasms with desirable traits. The wide genetic diversity indicates variations of alleles that are suited for various environments including the adverse environments (e.g. flooding, dry, salt-alkaline) [84], which can be used to develop resistant varieties for these stressful conditions [81]. For example, the miscanthus breeding group in Hunan Agricultural University directly selects two natural hybrid varieties (crossed within *M. lutarioriparia*) from the collected germplasms, namely Xiangnandi No.1 & 2 carrying resistances to 3-months-long flooding and high biomass yield potential (average autumn yield of 30 odt ha^{-1} yr^{-1} in Hunan). The resistance varieties give a potential to expand the miscanthus production on non-arable land.

1.4 Current status of miscanthus production and application

Currently, the European countries and China are the world centres for miscanthus production and application. Interests in miscanthus research and commercialization are growing from other countries, such as the USA, Canada and South Korea [31-32, 36, 85-87]. The presently commercialized miscanthus plants are cultivated in Europe, but wildly growing in China. The reported applications of miscanthus mainly include horticulture (ornamental plant), animal husbandry (fodder, animal bedding material), combustion for heat and electricity generation, paper-making, pickle-making (using the spring emerged young shoots) and building material [88-91].

In Europe, research on miscanthus utilization concentrates on bioenergy use, which already took off in the early-1980s. However, the commercial production and utilization just began from 2006 and the production scale increased sharply since 2008 [38, 92]. To date, without countries in cold Northern Europe (e.g. Finland), almost all the European countries have miscanthus plantations, but mainly in the UK, Germany and France. According to the 2014 statistics data [93], there are approximately 40,000 ha miscanthus established in Europe with 17,000 ha, 15,000 ha and 3,500 ha in the UK, Germany and France, respectively. In the UK, the largest application of miscanthus biomass is in co-firing with coal for electricity generation [79]; while in Germany, thermal application (e.g. house heating) is the main utilization [91]. The applications of generating biogas and cellulosic ethanol are under research without commercial production so far [94-95]. Large proportion of the already established miscanthus in Europe is *M.* × *giganteus* because it is the single commercial clone with stable and high biomass yield in the European conditions. Due to sterility of the dominating *M.* × *giganteus*, the commercially miscanthus establishment is mainly achieved by direct rhizomes planting with an establishment cost of 3,000-3,600 € ha^{-1} [96]. Based on the recently released miscanthus production calculator by Terravesta [97], the current British miscanthus production system can produce a net revenues of 515 £ ha^{-1} yr^{-1} (approximately 720 € ha^{-1} yr^{-1}).

Personally, China has the largest miscanthus growth area worldwide of approximately 100,000 ha (communication with Dr. Liang Xiao). The vast majority of these plants are wildly growing *M. lutarioriparius* in the costal area around Dongting Lake with a biomass yield potential of 12 t ha^{-1} yr^{-1} (air dried weight). To date, the largest application of these plants is papermaking and the preferred pattern of production organization is termed as land-leasing model. This organization model is a form in which land tenure (here is the administrative office of Dongting Lake Natural Reserve, i.e. the government) leases the miscanthus land for a specific period of time (e.g. 10 years) to a company under a contracted land price (unpublished investigation data of approximately 2,600 CNY ha^{-1} yr^{-1}) and then the company self-organizes field management (average cost of 750 CNY ha^{-1} yr^{-1}), biomass harvest (average cost of 1,500 CNY ha^{-1} yr^{-1}) and transportation (cost of 0.5 CNY t^{-1} km^{-1} by cargo boat). In addition, contracting farming model is sometimes applied where the government organizes the miscanthus production and then sells biomass to the contracting company with an average price of 600 CNY t^{-1} (air dried weight). However, since 2006, the government shut down many papermaking companies due to the serious water pollution caused by the wastewater from papermaking procedure [98]. One new concept of using the wild miscanthus is pickle-making, i.e. pickling the young shoots (20-30 cm tall) collected in early spring. In 2014, there are approximately 5,300 t pickled miscanthus shoots produced with a production value of 0.5 billion CNY [99]. The main limitation on the development of miscanthus pickle-making industry is the short harvest period (only 2-3 weeks) of the young shoots. Nevertheless, the miscanthus pickle-making is still a vigorously promoted industry by local government with ambitions to produce 20,000 t pickled miscanthus shoots in 2015 [99]. To data, no energy-related commercial utilization of miscanthus is reported in China.

1.5 Dissertation topics and objectives

Although benefits of miscanthus plantation have been widely accepted, the large-scale production is still not realized as expected. There are a number of factors responsible for the slow expansion of miscanthus production. From a macroscopical aspect, the miscanthus biomass market is currently limited. There is no market using miscanthus biomass to produce cellulosic ethanol and biogas because the relevant technologies are not yet commercially available [37]. Although some power plants burn miscanthus biomass for electricity [37, 100], the market consuming miscanthus through combustion is small and the possible displacement of expensive miscanthus biomass by cheap agricultural residues makes it difficult to develop this small market. As a new crop, farmers have no experiences and technologies to plant and manage miscanthus, finally resulting in a low farmers' acceptance. From a technical perspective, inefficient and expensive propagation techniques, lack of varieties adapting to various environments are the two main limiting factors [29, 55, 101-104]. In addition, lack of land available for growing miscanthus (i.e. land use dilemma) also hampers the miscanthus production [104]. Against these limiting factors, this dissertation focus on reducing the limitations by inefficient propagation techniques and lack of genotypes/varieties with resistance to environmental stresses, and addressing the land use dilemma.

1.5.1 Improve the propagation techniques

According to the European miscanthus production experiences, the inefficient and expensive propagation is a bottleneck factor that presently limits the expansion of miscanthus production [29, 102]. Although steady progress is being made in propagating new plants from stem cuttings, seeds and micropropagated plantlets [29, 54-55, 105], to date these approaches are not mature enough for commercialization and almost all the commercial miscanthus establishment is performed by direct planting rhizomes [57, 101]. Due to 3-5 years are required to grow rhizomes and a big rhizome size (15-20 cm long) is required to germinate, only an annual division efficiency (i.e. multiplication ratio) of 1:10 can be achieved by the direct planting rhizome [53, 106]. The damage risk of current harvest technologies on rhizomes quality may furtherly reduce this division efficiency less. Because of the low multiplication ratio, not enough plant materials for expanding miscanthus cultivation can be supplied. Additionally, the current rhizome production has low mechanization level and needs high labour input, resulting in a high production cost [53]. The high production cost together with low multiplication ratio cause a high rhizome price of around 0.12 € per cutting [107], which, in turn, causes a high establishment cost and low profit of current miscanthus production [66, 108-109]. The low revenue of miscanthus production hinders its acceptance by farmers. What's more, the farmers' subjective perception of the financial risk resulting from the high one-off investment for establishment hinders the miscanthus acceptance [110]. The low farmers' acceptance dampens their enthusiasm to grow miscanthus. Therefore, the first aim of this dissertation is to improve the current techniques for efficient and economically feasible propagation.

1.5.2 Enlarge land reserve for expanding miscanthus production by marginal land

At the present stage, there are approximately 1.4 billion ha 'spare agricultural lands' (lands that are suitable but currently not used for agricultural productions) worldwide could be allocated for producing energy crops, including miscanthus [111]. Two-thirds of these 'spare agricultural lands' are concentrated in Latin Americans and Africa countries. The main components of these 'spare agricultural lands' are left fallow (uncultivated agricultural land for crop rotation), policy driven set-aside areas and unused pasture [111-112]. The fallow rotated out of production for agronomic purposes, e.g. maintaining the soil fertility. Therefore, farmers would like to plant non-commercial crops designed purely for soil quality improvement, but not the energy crops on fallow [112]. For the set-aside areas, farmers prefer to produce annual energy crop (e.g. rapeseed) but not the perennial miscanthus because annual crops offer them the flexibility of changing crops [113]. In some populated countries (e.g. China), the agricultural lands are not legally allowed to be converted to plant non-food energy crops [114-115]. All these together reduce the 'spare agricultural lands' that are actually available for planting miscanthus to be 900-1,400 million ha worldwide [111]. On the other hand, even if all the 'spare agricultural lands' are used to plant miscanthus, their production potential (200 EJ yr^{-1} if average biomass yield reaching 8 t ha^{-1} yr^{-1}) may still not satisfy the material demand of future bioenergy industry (e.g. 250 EJ yr^{-1} in 2050 globally [116]). With the world population growing, some current 'spare agricultural lands' will be reused to produce food. The land use dilemma for miscanthus production will be more serious in the future.

A practical solution to the land use dilemma for miscanthus production is cultivating miscanthus on barren land with natural condition which is not well suited to agricultural production but suitable for growing plants with resistance to environmental stresses [117]. Here, 'marginal land' is used as shorthand for this kind of land, mainly including shoal/bottomland, grassland, saline and alkaline land, and bare land. Several countries, e.g. Australia, Canada, China and India, have adopted policies mandating using marginal land for producing non-food energy crops [118]. The China's policies are partially adamant that only marginal land can be used for planting non-food energy crops because its per capita agricultural area is quite low (only 40% of the world average) [114-115]. It is therefore urgent and necessary to explore the marginal land potential for expanding the miscanthus production in China. In addition, China has almost all the marginal land types and its miscanthus production on marginal land can set a compensative example for other countries. Therefore, in this dissertation, the marginal land potential for miscanthus production is evaluated in the case study of China.

1.5.3 Selection of dedicated genotypes for marginal land

M. × *giganteus* is the single clone that is used commercially and its high yield potential can only be guaranteed in regions with a minimum monthly-averaged winter air temperature of -3.5 °C (sensitive to frost) and annual precipitation of 600 mm (sensitive to drought) [40, 119]. There are two main limitations by the above tough environmental requirements on the expansion of *M.* × *giganteus* production [37, 61, 120]: the poor frost tolerance constitutes an obstacle to expand production to cold areas; and the high water requirement to ensure good establishment and satisfactory yield limits expanding the production to dry areas. The low winter temperature in cold area damages the young *M.* × *giganteus* plants, which cannot emerge in following spring (i.e. high overwintering mortality). This is the critical limitation for expanding *M.* × *giganteus* production to Northern Europe currently. In limited water supply conditions, growth and yield of both above-ground and below-ground parts of *M.* × *giganteus* would be reduced [40, 61, 121]. For example, in the dry Davis (American) conditions [121], a reduction of 98% in total biomass yield (both above-ground and below-ground biomass) was observed by the rainfed plants compared to the irrigated plants. The reduced below-ground growth may result in high overwintering mortality and the limited above-ground yield would reduce the net revenue of miscanthus production [37]. For these concerns, large-scale plantation of *M.* × *giganteus* is also not yet achieved in dry areas. In addition, the *M.* × *giganteus* also does not adapt well to many stressful conditions such as stagnant water soil, saline-alkali soil [122], which inhibits the miscanthus production expanding to marginal land areas.

As mentioned above, the marginal lands will be central to energy crops' productions (including miscanthus), while the current available genotypes cannot survive and grow well on most marginal lands. To resolve this conflict, selection of dedicated genotypes/varieties with good adaptation to marginal conditions is required. Environmental stresses of different marginal lands are generally not alike, e.g. seasonal flooding for bottomland vs. high salt content for salinity land. It is therefore furtherly required to select dedicated genotypes/ varieties for specific marginal land types. Also due to the stressful environments, conventional practices of miscanthus establishment on arable land cannot be directly applied to marginal land. Then effective methods of miscanthus establishment on different marginal lands also need to be developed. Among all the available marginal land types, grassland has the largest area suitable for growing energy crops (including miscanthus) because: (1) grassland has the largest terrestrial area, which is 40.5% (approximately 5.3 billion ha) of the global land area [123]; (2) compared to other marginal land types, the environmental stresses of grassland are mild and a large proportion of grassland is suitable for growing energy crops; (3) due to the intensification of livestock farming and use of arable forage crops, there is an increasing area of grassland which is no longer used for animal husbandry but can be used for growing energy crops [124]. For these reasons, it is primarily urgent to investigate and screen optimal energy crop species/genotypes and related agricultural practices for the bioenergy use of grassland. *Miscanthus* is considered an important energy crop for biomass production. Therefore, in this dissertation, we try to screen optimal genotypes and effective practices for the miscanthus establishment and management on grassland land – the representative marginal land type.

1.6 Formal structure of this dissertation

To achieve above objectives, several field trials, farmer surveys and modelling approaches were carried out. The results gained during investigating each study goal are used to prepare one scientific article. Then the body of present thesis consists of two published articles (**Chapter 2 & Chapter 3**) and one submitted article (**Chapter 4**).

In **Chapter 2**, a literature review was performed with aims of presenting the currently available miscanthus propagation options and the best practices for each available option. Farmers were interviewed to clarify the currently practical farm experience regarding miscanthus production and collect problems encountered in their miscanthus production, which could lead research questions. Additionally, field trials were conducted to improve the propagation system of direct rhizome planting to be more efficient and to explore the potential of seeds propagation system in south-western Germany conditions. To enlarge land reserve for miscanthus production, **Chapter 3** was designed to assess the marginal land potential for the miscanthus production in a case study of China. In this chapter, Geographic Information System (GIS) techniques, model simulation were adopted to identify the productive marginal areas in China for miscanthus and to estimate their biomass and bioenergy production potential. Results from **Chapter 3** show that grassland is one of the main marginal land types exploitable for miscanthus production in China. Not only in China but also worldwide, grassland is the most important marginal land type to expand miscanthus production. However, there is lacking of optimal genotypes and agricultural practices for the miscanthus establishment on grassland. Therefore, establishment and management practices for miscanthus establishment on C3 grassland were investigated in **Chapter 4** of this thesis. Through conducting three field trials, effects of miscanthus genotype and propagation method, grassland pre-disturbances and cutting frequencies on miscanthus establishment and growth were assessed. Finally, the best practices for miscanthus establishment and management on grassland are shown. In addition, **Chapter 1** contextualizes this thesis by introducing the general background and reasons why we designed this study; **Chapter 5** discusses the main findings of this thesis in a broader context; **Summary & Zusammenfassung** finalizes this thesis by summarizing the main findings.

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Chapter 2 Present and future options for the improvement of *Miscanthus* propagation techniques

Based on the analyses in Chapter 1 that the currently inefficient and uneconomic propagation techniques limit the expansion of miscanthus production, this chapter aims to investigate the potential ways to improve the propagation techniques. To achieve this objective, a literature review was performed with aims of presenting the currently available propagation and establishment options and the best practices for each available option; an economic estimation was conducted to investigate factors which contribute to the high establishment costs; farmers were interviewed for the existing problems which should be addressed in further research; and field trials were conducted to improve the rhizome propagation system to be more efficient by minizing the rhizome size and to explore the potential of seeds propagation system in south-western Germany conditions.

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Present and future options for *Miscanthus* propagation and establishment

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ABSTRACT

Several species of the genus *Miscanthus* are characterized by high biomass yields and low input requirements, and there is increasing interest in their commercial use for bioenergy production. However, the lack of inexpensive and effective propagation and establishment techniques is currently limiting the potential of miscanthus as a commercial bioenergy crop. In this review, through an evaluation of previous studies, results of our own field trials, experiments and farmer surveys, we concluded that there are five main approaches that can be used for miscanthus establishment. First is direct rhizome planting which is relatively mature, easily realized and inexpensive (1904–3375.7 € ha^{-1}); therefore it is the method mostly preferred by farmers. However, in the long term, its low dividing efficiency (1:10) will cause a conflict between the demand for and supply of rhizomes for large-scale plantations. Compared to the direct rhizome planting, an increased multiplication ratio (1:30) has been realized using rhizome- or stem-derived plantlets. However, due to higher labour and energy inputs required for the pre-growing of plantlets, their establishment cost reduction potential is limited, with estimated costs of between 4240.8 € ha^{-1} and 4400.8 € ha^{-1}. The seed-setting rate of miscanthus (*Miscanthus sinensis*) is very low (0.0–28.7%) under the climatic conditions of south-west Germany, making commercial seed production difficult. The high multiplication ratio (1:960) and fast bulk-up production potential achieved by micropropagation provide an opportunity to reduce the costs of this currently most expensive establishment method (6320.8 € ha^{-1}). The cheapest method could be direct seed sowing (1508.5 € ha^{-1}) if it will become feasible in future. Additionally, the recently developed CEED technology may become a good alternative, if it is not too expensive. For all the propagation methods considered, new technologies and research efforts are required to reduce the material production costs and simultaneously increase the multiplication ratio.

Contents

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1. Introduction

The perennial rhizomatous grass miscanthus has been identified as a leading candidate energy crop for heat, electricity and transport fuel production under European conditions due to the combination of its high energy yields and low input (fertilizer, pesticides, energy) requirements [1,2]. As a crop with the C4 photosynthetic pathway, miscanthus exhibits a high rate of net photosynthesis and water-use efficiency even under the relatively cold conditions of temperate climates [3,4]. These result in high harvestable biomass yields generally varying from 15 to 20 oven-dry ton (odt) ha^{-1} in temperate regions and up to 44 odt ha^{-1} in southern Europe [5]. High contents of lignin (approx. 25%) and carbohydrates with high calorific value (approx. 38% of cellulose and 24% of hemicelluloses) further contribute to the energetic quality of miscanthus biomass [6]. A deep rooting system and an effective mechanism of nutrient (N, P and K) relocation from aboveground plant parts to belowground rhizomes lead to low nitrogen (N) input requirements [7,8]. *Miscanthus* has very few natural pests and diseases, and usually no pesticide application is necessary [9–11]. Heat and power production from miscanthus biomass exhibits a high energy output/input ratio ranging from 15:1 to 32:1 depending on the farming system [1,12,13]. This ratio is much higher than for the processing of traditional energy crops, such as ethanol production from wheat (9:1) and biodiesel from rapeseed (4:1) [1,14].

However, there are still very few large-scale plantations of miscanthus in the world and the estimated total area in Europe is only 38,300 ha [15]. By the end of 2011, 9,000 ha of miscanthus had been established in the UK and 3,000 ha in Germany for the co-firing of biomass with coal and thermal applications [16,17]. The area of cropland used to grow miscanthus is also increasing in other countries. In 2011, the Biomass Crop Assistance Program (BCAP) projects planted 6,600 ha of miscanthus in the United States [18]. The key bottleneck for large-scale production of miscanthus is the high biomass production costs due mainly to the lack of inexpensive and effective propagation and establishment systems [19-21]. Previous studies [19,22–31] have shown that various plant materials can be used for miscanthus propagation, in particular rhizomes, terminal and nodal buds, seeds, nodal stem cuttings and immature tissues, such as inflorescences and leaves (Fig. 1).

Presently, only one clone, *Miscanthus* × *giganteus* Greef et Deuter, is grown commercially. The main establishment technique for *M.* × *giganteus* is the harvest and direct planting of rhizomes into the field (referred to in the following sections as 'rhizome planting'). The growing of plants from rhizomes in the greenhouse and their subsequent transplanting into the field (referred to in the following sections as 'rhizome-derived plants') has recently been gaining favour in North America and is used by farmers to replace failed plants [32]. *M.* × *giganteus*, as a triploid infertile clone, cannot be directly established via seeds [33]. The results of trials with the fertile species *Miscanthus sinensis* Andersson and *Miscanthus sacchariflorus* Bentham suggest that the two establishment methods of direct sowing of seeds (referred to in the following sections as 'seed sowing') and the transplanting of plantlets grown from seeds in module trays (referred to in the following sections as 'seed-propagated plants') are possible and may be less expensive than rhizome planting [19,34,35]. Another vegetative production method, deriving new plants from nodal stem sections (referred to in the following sections as 'stem-derived plants'), has been proved possible for the establishment of *M.* × *giganteus* and may be feasible for other genotypes, in particular species with stem buds such as *M. sacchariflorus* [24,25,27,30,31]. Micropropagation technologies developed to produce progeny plants from tissues are also available for miscanthus and include regenerating plants from somatic embryos formed in callus culture and direct shoot regeneration with in-vitro tillering from apical rhizome meristems or axillary nodes [29,36,37]. More recently, a propagation system called CEED (Crop, Expansion, Encapsulation and Delivery System) has been developed for *M.* × *giganteus*. In this system encapsulated plant material is put into the soil, from which plants emerge. Commercial *M.* × *giganteus* CEEDs have been provided by New Energy Farms since spring 2014 [38]. Even though there are so many options for propagating miscanthus, nearly all commercially available miscanthus plants are currently produced by the methods of rhizome planting, which is easily achieved and relatively inexpensive, and micropropagation, which provides theoretically unlimited propagation possibilities. However, to support the promising future of miscanthus, more studies need to be conducted to lower establishment failure, increase the reproduction rate and reduce establishment costs by optimising current propagation technologies or exploiting other approaches.

The aims of this study are to provide an overview of the state-of-the-art of miscanthus propagation methods including factors that hamper cost reduction and establishment success and to evaluate the different propagation systems with regard to technologies and costs. For this purpose a literature review was performed, farmers propagating and selling rhizomes were interviewed (Farmers), and field trials (Trials) and greenhouse experiments (Exp) were conducted. Trials and experiments were designed to investigate the factors affecting the establishment and development of rhizome-propagated plants, and to estimate the potential of seed production in south-west Germany.

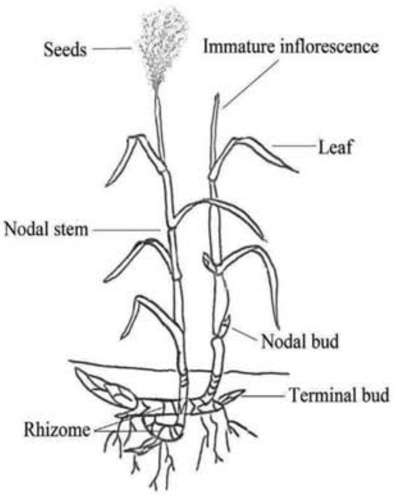

Fig. 1. *Miscanthus* plant parts suitable for propagation.

2. Materials and methods

2.1. Literature review

A thorough computerized search of published research materials on methods of *Miscanthus* propagation was conducted using the following keywords: 'miscanthus propagation', 'rhizome cutting', 'rhizome-derived plug', 'stem cutting', 'miscanthus seed', 'micropropagation', 'miscanthus cost' and 'miscanthus morphological performance'. The documents reviewed included books and published peer-reviewed research articles, conference papers, dissertations, project reports and academic presentations accessed via the internet. The search was limited to materials published in English, German and Chinese; when two or more articles included the same data set, only the original (first-published) article was used. In total, 80 articles and other publications were identified in this review. Each publication was systematically searched for the following information: plant material source, technologies used to propagate and establish miscanthus in the field, establishment operations and their costs, field performance of the plants, e.g. establishment and overwintering survival, morphological traits and aboveground biomass yield. This information was summarized to present a best option for each propagation system and to identify the existing problems which should be addressed in further research.

2.2. Field trials and germination tests

2.2.1. Field trial on rhizome propagation (Trial 1)

In May 2011 a field trial was established at the University of Hohenheim (48°42′N, 9°13′E), Stuttgart, south-west Germany. The experimental site is located at an elevation of 400m on a 3° south-facing slope with a long-term annual average rainfall of 698 mm and annual average air temperature of 8.8 °C; the soil of the experimental field is silty loam. The experiment was set up in a randomized complete block design with three factors and four replications. The factors examined were (1) genotypes, (2) rhizome harvest time and (3) rhizome size (length of rhizome cuttings). Fresh rhizomes of *M. sinensis* and *M.* × *giganteus* were manually collected from 14-year-old mother plants grown at Ihinger Hof (IHO) experimental station (48°45′N, 8°56′E) of the University of Hohenheim. The rhizomes were harvested with 20-day intervals between 10 March and 20 April 2011. The rhizomes were washed from soil and cut into cuttings of either 3 or 6 cm length. The rhizome cuttings were placed in plastic bags and stored in a climate chamber at 4 °C for 20, 40 and 60 days, depending on the harvest date (i.e. the rhizomes harvested earlier were stored longer). On 16 May 2011, all the rhizome cuttings were taken to the field and planted manually in 48 1 × 1 m plots with a planting density of 10 plants m^{-2} and a depth of 5–10 cm. In 2011 and the following two growing seasons, no fertilizer and irrigation were applied. In September 2011 and 2012 the number of plants alive in each plot was counted to calculate the plant survival percentage. At the end of the second growing season (2012), the shoot number, and the length and diameter of the highest shoot were measured for each plant in every plot and the plot biomass was harvested at a level of 5 cm above ground and weighed. To ascertain the dry weight, about 400 g fresh biomass samples were taken from each plot. The samples were weighed, dried to constant mass (seven days in the oven at 60 °C) and weighed again to obtain the dry weight.

2.2.2. Seed-setting of miscanthus in the field (Exp 1)

The seed-setting rates of two *M. sinensis* hybrids and three *M. sinensis* genotypes were assessed. The hybrids were represented by GOFAL7 and RH81 genotypes produced by crossing two different *M. sinensis* populations and an *M. sacchariflorus* × *M. sinensis* hybrid, respectively. *M. sinensis* was represented by three genotypes: 88-111, 90-6 and SW217. The field trial was established in a completely randomized block design with 3 block replicates in 1997 by hand-planting of in-vitro cloned plants. The detailed information on each genotype can be found in the publications by Clifton-Brown [39], Clifton-Brown and Lewandowski [40] and Iqbal and Lewandowski [41]. In late September 2012, when the seeds were ripe, 15 large complete inflorescences (panicles) were collected from each plot. The number of florets was counted on three randomly selected inflorescences per genotype in each of the three replicates and threshed by hand to obtain the seeds. The seeds were then cleaned and the number per panicle counted. The length and width of 10 seeds randomly selected from the mixed seeds of the three sampled panicles of each genotype in each replicate were measured with a calliper.

2.2.3. Seed germination test (Exp 2)

For an effective seed propagation system, good seed germination (seed quality) is of the upmost importance. In this experiment, seed germination tests were carried out in a climate chamber. Because the hybrids GOFAL7 and RH81 produced no viable seeds in the field, only the seeds from the open-pollinated *M. sinensis* genotypes (88-111, 90-6 and SW217) were used for the germination test. All the available seeds were placed in sterile plastic Petri dishes (Ø 9 cm) on two layers of filter paper (Munktell, Grade 3 hw). For the genotypes 90-6 and SW217 there were 40 seeds per Petri dish with 4 replications and for 88-111 there were 15 seeds per Petri dish also with 4 replications. The filter paper was moistened with 4 ml distilled water per Petri dish. No further water was added during the whole germination process. The Petri dishes were randomised and placed in a climate chamber at 25 °C without light. The germinated seeds were counted at 24 h intervals over a 7-day period. Germination was considered to have occurred once the radicle had protruded beyond the seed coat by at least 1 mm.

2.3. Farmer surveys

In order to collect more information on practical farm experience of miscanthus propagation, a questionnaire was developed for miscanthus farmers. Surveys were conducted in spring 2013 by email, face-to-face interview or on the phone. In total, three German, one British, one Austrian and two Canadian farmers from the 'Ontario Soil and Crop Improvement Association' participated in the survey. The survey questions aimed to clarify the current status of miscanthus propagation and establishment regarding (1) the farmer's practical experience of the optimal propagation and establishment methods, (2) problems encountered that could lead to research questions and (3) the economic performance of miscanthus production.

2.4. Establishment cost estimation

To evaluate the commercial potential of each available propagation system, an economic estimation of the establishment costs was conducted using 2012 prices converted into euro (€) by the Gross Domestic Product Deflator Index. These are made up of: (a) the opportunity cost of land; (b) the cost of production material inputs including fertilizer, herbicide, planting materials and irrigation water; (c) the fixed machine costs including depreciation, repair, maintenance, insurance (0.5% of purchasing price per year [42]) and storage (1.75% of purchasing price per year [42]); and (d) the variable machine costs which were divided into fuel, labour and lubricant costs. To estimate the field operation costs for miscanthus establishment, the literature data were used (Table 1). Production cost calculations were based on the unit price of each input item derived from literature data [10,34,42,43,46–48], government (Eurostat) [49] or non-governmental organisation (Fertecon) [50] databases and an on-line shop (Amazon)

Table 1
Field operations for the cost assessment of different miscanthus establishment methods, including rhizome planting, rhizome-derived plants, stem-derived plants, seed sowing and micropropagation.

Items	Rhizome planting	Rhizome-derived plants	Stem-derived plants	Seed sowing	Micropropagation
Planting density	20,000 cuttings ha^{-1}	16,000 plants ha^{-1}	16,000 plants ha^{-1}	100 seeds m^{-2} (equal to 4.8 kg ha^{-1} coated seeds)	16,000 plants ha^{-1}
Land preparation	Ploughing once Power harrowing twice	Ploughing once Power harrowing twice	Ploughing once Power harrowing twice	Ploughing once Power harrowing 3 times	Ploughing once Power harrowing twice
Herbicide application rate and time	4 l ha^{-1} Glyphosate before ploughing	4 l ha^{-1} Pendimethalin before ploughing	4 l ha^{-1} Pendimethalin before ploughing	3.52 l ha^{-1} Atrazine before ploughing 1.75 l ha^{-1} 2,4-D in July–August	4 l ha^{-1} Pendimethalin before ploughing
Fertilizer	P 44 kg ha^{-1} K 110 kg ha^{-1}	P 26 kg ha^{-1} K 100 kg ha^{-1}	P 26 kg ha^{-1} K 100 kg ha^{-1}	P 10 kg ha^{-1} K 60 kg ha^{-1}	P 26 kg ha^{-1} K 100 kg ha^{-1}
Rolling frequency and time	Rolling once after planting	–	–	Rolling twice before and after seeding, respectively	–
Irrigation	–	400 m^3 ha^{-1}	400 m^3 ha^{-1}	600 m^3 ha^{-1}	400 m^3 ha^{-1}
Sources	[40,42–43]	[22,42,44]	[22,27,42]	[19,34,43,45]	[22,27,42]

[51]. The exchange rates used for price conversion of sterling and dollars against the euro were 1.23 and 0.78 [52], respectively.

Based on the above estimations, establishment costs (C_e) were calculated using Eq. (1):

$$C_e = C_{\mathrm{lop}} + C_{\mathrm{pm}} + \sum_{m=1}^{M} \left[C_{\mathrm{fm}}(m) + C_{\mathrm{vm}}(m) \right] \tag{1}$$

where C_{lop} represents the land opportunity cost, measured as the average foregone profits from converting land from rapeseed, wheat or barley production to miscanthus and equals 414.3 € ha^{-1} [46]; C_{pm} is the production material costs including fertilizer, herbicide, irrigation water and planting material; C_{fm} and C_{vm} are the fixed and variable machine costs of m^{th} machine operations including ploughing, harrowing, planting, rolling and spraying fertilizer and herbicide, respectively.

Labour costs (C_{la}) of each operation are calculated using Eq. (2):

$$C_{\mathrm{la}}(m) = 1.1 \times W_m \times N_m \tag{2}$$

where W_m and N_m are the working hours per person and number of workers required for the m^{th} operation; and 1.1 is the coefficient for the unproductive time required for travelling and servicing [53].

The total costs of repair and maintenance (C_{rm}) and lubrication (C_{lu}) are estimated using Eqs. (3) and (4), respectively [54,55].

$$C_{\mathrm{rm}} = \sum_{m=1}^{M} \left[H_m \times Rf_1 \times PP_m \times (Lt_m/1000)^{Rf_2} / Lt_m \right] \tag{3}$$

$$C_{\mathrm{lu}} = \sum_{m=1}^{M} (0.021 + 0.00059 \times MP_m) \times UP \tag{4}$$

where H_m represents the working hours required per hectare of the m^{th} machine; Rf_1, Rf_2 are the repair and maintenance coefficient 1 and coefficient 2 obtained from AAEA [55]; PP_m represents the purchase price of the m^{th} machine and Lt_m its lifetime; MP_m and UP represent the maximum power (kW) of the m^{th} machine and unit price of the lubricant (€ l^{-1}), respectively.

2.5. Statistical analysis

All statistical analyses were performed in SAS v9.2 (SAS Institute, Cary, NC, USA). The data from the authors' own trials and experiments are presented as mean values ± standard error (SE). In analysing the data from Trial 1, general linear modelling (GLM) was employed to determine the effects of the miscanthus genotypes, the rhizome harvest time and rhizome size and the interactions of these on the measured traits, e.g. plant survival percentage, morphological characteristics. To compare the differences of the measured traits among the 12 treatment combinations, the 'CONTRAST' statement in PROC GLM was used. Main and interaction effects were significant at $P < 0.05$ and Fisher's least significant difference multiple comparison tests at the $P < 0.05$ level was used in the means comparison. Data from Exp 1&2 were analysed and compared using a one-way analysis of variance (ANOVA) followed by Duncan's multiple range tests at the 5% confidence level. A highly variable variance among treatments in terms of percentage data (e.g. 27.5% vs 92.5%) was observed. To ensure a normal distribution of the tested data, all the percentage data were log transformed before analysis.

3. Results

Different parts of the miscanthus plant can be used for propagation purposes (Fig. 1). This review focuses on the four main propagation systems: rhizome-, stem- and seed-based propagation, and micropropagation.

3.1. Rhizome-based propagation systems

A rhizome is a horizontal, subterranean shoot/stem with active meristems such as nodes and buds [56]. The miscanthus rhizome is a branched underground stem with multiple active nodes and terminal buds that can give rise to roots and new shoots. The rhizome morphology varies among miscanthus genotypes. *M. sacchariflorus* forms broad trailing and thick-stemmed rhizomes. The rhizome of *M. sinensis* is thin-stemmed and short; *M.* × *giganteus* has a thick-stemmed rhizome, which is often oval to round and only slightly trailing [4,57]. The establishment success of rhizome-propagated miscanthus stands depends on the genotype, the rhizome harvest time, the harvest technology, the storage time between harvest and planting, the storage conditions, the rhizome size, the planting technology and also on the weather and soil conditions of the planting site (Tables 2 and 3).

The results of the authors' field trial on miscanthus establishment via rhizomes (Trial 1) showed significant effects of genotype, rhizome size, harvest time and interactions thereof on plant survival percentage, morphological development and biomass yield (Table 2). Generally, for both genotypes (*M.* × *giganteus* and *M. sinensis*) the 2011 plant survival percentage from 6 cm rhizomes was significantly ($P < 0.001$) higher than that from 3 cm rhizomes. For *M.* × *giganteus*, the average 2011 plant survival percentage of rhizomes harvested at the latest date (late April) was about 29% higher than that of those harvested at the middle date (early April) and 28% higher than that of the early harvest date (mid-March). Late rhizome harvest is therefore preferable for *M.* × *giganteus* whereas an early harvest is preferable for *M. sinensis* (Table 2). Overall *M. sinensis* showed fewer winter failures

Table 2
Effects of genotypes (G), rhizome size (RS) and rhizome harvest time (RH) on miscanthus establishment and morphological performance, Hohenheim field trial on rhizome establishment (Trial 1).

Treatment combination	Plant survival percentage in 2011 (%)	Plant survival percentage in 2012 (%)	Shoot number (m^{-2})	Shoot length (cm)	Single shoot diameter (cm)	Biomass yield (g m^{-2})
Gig-3-1	**75.0 ± 23.8**abc	57.5 ± 5.0cd	75.67 ± 6.81bcd	155.93 ± 42.21bc	0.89 ± 0.11b	**2061.2 ± 162.2**a
Gig-3-2	50.0 ± 23.1de	40.0 ± 11.5e	52.67 ± 11.85de	**191.68 ± 48.04**a	0.83 ± 0.14c	1527.4 ± 214.3bc
Gig-3-3	27.5 ± 22.2e	20.0 ± 14.1f	35.33 ± 7.57e	151.44 ± 30.03c	0.84 ± 0.10bc	722.0 ± 228.1ef
Gig-6-1	**82.5 ± 12.6**ab	**75.5 ± 5.8**ab	**81.00 ± 7.00**abc	170.47 ± 25.98b	**0.96 ± 0.10**a	**2242.2 ± 160.6**a
Gig-6-2	50.0 ± 16.3de	50.0 ± 16.3de	58.33 ± 9.87cd	162.23 ± 46.93bc	0.86 ± 0.14bc	1565.6 ± 277.5bc
Gig-6-3	**75.0 ± 10.0**abc	65.0 ± 5.8bcd	64.67 ± 17.90cd	168.08 ± 20.47bc	0.87 ± 0.11bc	**1892.3 ± 274.6**ab
Sin-3-1	60.0 ± 14.1bcd	57.5 ± 12.6cd	60.33 ± 11.93cd	161.73 ± 21.47bc	0.63 ± 0.07d	1007.1 ± 79.6ef
Sin-3-2	55.0 ± 12.9cd	55.0 ± 12.9cde	34.33 ± 3.21e	154.78 ± 22.02bc	0.64 ± 0.07d	695.0 ± 133.3ef
Sin-3-3	**75.0 ± 5.8**abc	70.0 ± 8.2bc	67.33 ± 9.29bcd	159.76 ± 34.21bc	0.56 ± 0.08ef	1001.3 ± 91.7ef
Sin-6-1	72.5 ± 9.6bcd	70.0 ± 8.2bc	**100.67 ± 13.01**a	154.81 ± 14.32bc	0.63 ± 0.08d	1498.6 ± 496.1bcd
Sin-6-2	**82.5 ± 9.6**ab	**75.0 ± 5.8**ab	**89.67 ± 6.43**ab	154.20 ± 24.70bc	0.61 ± 0.11de	1151.3 ± 193.7cde
Sin-6-3	**92.5 ± 5.0**a	**87.5 ± 5.0**a	**100.33 ± 27.47**a	131.44 ± 22.92d	0.55 ± 0.07f	1095.0 ± 227.3def
Source of variation						
G	0.0053	< 0.0001	0.0026	< 0.0001	< 0.0001	< 0.0001
RS	< 0.0001	< 0.0001	< 0.0001	0.0858	0.1823	< 0.0001
RH	0.0577	0.0271	0.0020	0.0054	< 0.0001	< 0.0001
G × RS	0.9242	0.2037	0.0019	0.0586	0.0090	0.4686
G × RH	0.0005	< 0.0001	0.0105	0.0963	0.0029	0.0106
RS × RH	0.0913	0.0410	0.6726	0.0675	0.3973	0.1350
G × RS × RH	0.0346	0.0396	0.0954	< 0.0001	0.7989	0.0010

Note: Treatment combination refers to genotypes (Gig=*M.* × *giganteus*, Sin=*M. sinensis*) followed by rhizome cutting size (3=3 cm, 6=6 cm) and rhizome harvest time before planting date (1=20 days, 2=40 days, 3=60 days); For the shoot number, length, diameter and biomass yield, mean values marked with the same letter are not significantly different at the 0.05 level of probability; Figures marked in bold indicate the best performance without significant differences.

Table 3
Overview of the factors influencing miscanthus rhizome establishment and best propagation options.

Types of rhizome propagation systems	Factors influencing establishment	Alternatives tested	Best options	Source
Direct rhizome planting	Genotype	*M.* × *giganteus* *M. sinensis*	*M.* × *giganteus* (Central & South Europe) *M. sinensis* (North Europe)	[5,22,26,58–60]
	Age of mother plant	1 year, 5 years and 9 years	3–5 years	[10,60]; Farmers
	Rhizome harvest time	November to June	End of March/Beginning of April (Central Europe)	[10,61–63] Farmers; Trial 1
	Harvest technology	Two-step procedures with plough, shaking lifter, digger or rotor-tiller for lifting and cutting the rhizome and picking by potato harvester or stone picker; One-step procedure with combined rope-chain harvester	Two-step procedure: cutting of rhizome via plough or rotor-tiller and picking of rhizomes with potato harvester	[10,44] Farmers
	Planting time	March to June (Central Europe & North Canada) & Autumn	March to April, as quickly as possible after rhizome harvest (Central Europe)	[10,61,64] Farmers
	Rhizome storage	−1–7 °C, 60–100% Humidity	3–4 °C, > 70% Humidity, dark, wrapped in coco fibre bags with moist miscanthus chips or plastic bags	[26,65] Farmers; Trial 1
	Rhizome size	20–80 g per cutting 3–20 cm per cutting	5–15 cm or 60–75 g, fist-sized, at least 3 buds per rhizome cutting	[10,26,61] Farmers & Trial 1
	Planting depth	5–20 cm	10–20 cm	[10,26,61]; Farmers
	Planting density	10,000–80,000 cuttings ha^{-1}	For biomass production: 12,000–16,000 cuttings ha^{-1} with row distance of 70–115 cm in Central and South Europe; 40,000 cuttings ha^{-1} in North Europe; For rhizome production: > 40,000 cuttings ha^{-1} in Central Europe	[1,44,59,66,67] Farmers & Trial 1
	Weed control	Various types of herbicide, i.e. fluroxypyr, glyphosate, isoproturon	Roundup by glyphosate until two weeks before planting; Harrowing (Striegeln) until plants reach a height of 50 cm; Maize herbicides (Terano®, Callisto®)	[10,61,66,68] Farmers
Rhizome-derived plants	Rhizome size	N/A	At least 2 cm	Farmers
	Greenhouse period	N/A	2–3 months to a plant height of 30 cm	[69,32]; Farmers
	Greenhouse conditions	N/A	20 °C with light in the day; 15 °C dark at night	[22]
	Planting density	N/A	12,000 plantlets ha^{-1} (Central Europe)	Farmers

N/A represents no information available.

than M. × *giganteus* in all treatments indicating higher winter hardiness of *M. sinensis*. This difference was especially pronounced when 3 cm rhizomes and early harvest were used (Table 2). With regard to the plant morphological development, reducing rhizome size from 6 cm to 3 cm led to a significant ($P < 0.001$) decrease in the shoot number in both genotypes: by 10–45% in M. × *giganteus* and 33–62% in *M. sinensis*, depending on the harvest time. There was no significant effect of the harvest time ($P = 0.6736$) on the shoot number, especially when 6 cm rhizomes were used (Table 2). The shoot length of the plants was significantly ($P < 0.001$) affected by the genotype and harvest time but not by the rhizome size ($P = 0.0858$). An average plant height (all treatments pooled) was 167 cm for M. × *giganteus* and 153 cm for *M. sinensis*. A similar significant effect of genotype ($P < 0.001$) was observed on shoot diameter, with thicker shoots produced by M. × *giganteus*. Early rhizome harvest had a significant negative effect on shoot diameter ($P < 0.001$), in particular for *M. sinensis*. In general, the biomass yield declined significantly ($P < 0.001$) with the reduction in rhizome size and early rhizome harvest. Biomass yield of all the treatments ranged from 7.2 to 22.4 odt ha^{-1} for M. × *giganteus* and was significantly higher than that of *M. sinensis* (6.9–14.9 odt ha^{-1}), with the highest yield observed in treatments of 6 cm rhizomes harvested in late April for both genotypes.

Currently the procedure for rhizome propagation most practised by farmers is: (a) harvesting the aboveground biomass; (b) separating the rhizomes (while still in the soil) from the deep roots; (c) breaking up the rhizome mat (still in the soil) into pieces of smaller size; (d) ridging the mixture of the small rhizome cuttings and soil, and then separating the mixture; (e) grading and sorting the harvestable cuttings; cutting larger pieces to a suitable size, often performed manually; (f) planting rhizome sections with a potato planter [1,10,67]. All these operations can be conducted using conventional agricultural machines, such as a vegetable subsoiler to separate the rhizomes and roots, a rotavator for rhizome chopping, a potato harvester or stone picker to separate the rhizomes and soil. The farmer survey showed that the rhizome harvest in the field can also be performed using a combined rope-chain harvester which can remove rhizomes from the soil and cut them simultaneously (Table 3), but a two-step procedure for the rhizome harvest has the advantage that more soil is left in the field. The disadvantages of the unspecialized equipment are the high level of harvest damage (destroyed rhizome buds) and the different sizes of the rhizomes obtained. Specialised equipment for rhizome harvest and planting has been developed by the UK company ADAS Ltd. (Agricultural Development Advisory Service), but farmers are not content with the establishment success (Farmers). The farmers' survey also revealed the importance of optimal soil and weather conditions for the rhizome harvest: the soil should not be too moist and the air temperature should not be too high. Weed control, either chemical or mechanical (Table 3), is also essential for good establishment. Because nitrogen (N) fertilizer supports weed growth, N application is not recommended during the year of establishment. Due to the relocation of nutrients and carbohydrates to the rhizomes in autumn, it is recommended to harvest the rhizomes for propagation purposes in late winter or early spring [7,8,10]. Although autumn harvest and planting of rhizomes is also possible, the establishment success is usually low [64]. After harvest the rhizomes can be stored at a temperature of 3–4 °C for several weeks without significant viability reduction (Table 3). Optimal planting depths are 10 to 20 cm depending on the planting site [10,26,61].

An overview of the best options for rhizome propagation based on the scientific literature and farmers' experience is given in Table 3. For the rhizome-based propagation and establishment of M. × *giganteus* and *M. sinensis* under Central Europe conditions, the optimal procedure is: (a) separating rhizomes from roots using an under-cutter; (b) breaking rhizomes into pieces of 5–15 cm with more than three viable buds from 3- to 4-year-old mother plants in late March or early April by rotavator; (c) harvesting and collecting the rhizome pieces with a potato or flower bulb harvester; (d) putting rhizomes into storage (within four hours of being removed from the soil) at 4 °C, darkness and 75% humidity; (e) in March, planting 2–5 cm rhizome pieces into pots under greenhouse conditions of 20 °C/15 °C (day/night); (f) in mid-May, planting rhizome cuttings at a depth of 10 cm and density of 16,000–20,000 cuttings ha^{-1} with a modular potato planter or recently developed specialized machines, or transplanting pre-grown plantlets with a height of 30 cm into the field at a density of 12,000 plantlets ha^{-1} using a vegetable transplanter.

Although there are many different approaches to the propagation of miscanthus via rhizomes, these have mainly been developed by farmers who are often reluctant to publicize the methods which form the basis for their successful business. This is also true of the recently developed CEED system. For all rhizome propagation methods, the major problems are high failure rates due to poor rhizome quality and pest attack. There are reports of freshly planted rhizomes attacked by wireworm larvae, microtus (Farmer) or rabbits [10], especially when miscanthus was established after grassland conversion. The lack of specialised harvesting and planting machinery leads to low rhizome quality and different rhizome sizes which make planting difficult. This often results in an establishment success of only 40% or less for a large-scale plantation (Farmers), whereas under optimal conditions up to 90% establishment is possible. To improve the rhizome quality for planting, some farmers break or sort the rhizomes manually. This procedure is labour-intensive and needs to be mechanized.

The costs for miscanthus establishment via rhizomes are reported in the literature [43,70–73] and by the Farmer survey to vary from 1,904 to 3,006 € ha^{-1}. The lower end can only be achieved if the farmers use rhizome cuttings harvested from their own fields. The field operations for miscanthus establishment procedures are summarized in Table 1. The costs calculated for these establishment procedures (Table 1) are 3,375.7 € ha^{-1} for the method of direct rhizome planting and 4,400.8 € ha^{-1} for rhizome-derived plants (own data shown in Table 4) [43,70–73]. The highest costs of rhizome planting method were planting material costs (2,400.0 € ha^{-1}), followed by land costs (414.3 € ha^{-1}), variable machine costs (235.6 € ha^{-1}), agro-chemicals (212.6 € ha^{-1}) and fixed machine costs (113.2 € ha^{-1}) (Table 4), indicating the importance of the reduction of plant material costs in order to reduce establishment costs. That is also true for the method of rhizome-derived plants.

3.2. *Stem-based propagation systems*

Nodal stem cuttings (Fig. 1) removed from parent plants are capable of growing a new clone through rooting. Propagation via stem cuttings is practised in some agricultural crops, ornamental plants and forest species, e.g. sugarcane [74], bamboo [75], *Jatropha* [76]. In particular herbaceous plants with stems that remain soft and succulent throughout their life can be easily reproduced from stem cuttings [77]. For miscanthus, a plant with strong lignified stems, commercial propagation via stem cuttings has not been developed. Once miscanthus stem pieces are cut from a parent plant, they quickly deteriorate, especially in warm, dry conditions, and cannot easily root under field conditions [24,77]. Therefore, stem sections should first be pre-grown in optimal environmental conditions and then transplanted into the field.

Table 5 summarizes how the genotype, age and development stage of the mother plant, cutting characteristics and environmental conditions affect shoot emergence from the nodal stems of miscanthus. Good stem germination success (up to 97%) has been achieved under controlled conditions with the genotypes *M. sacchariflorus* and M. × *giganteus*, but not with the other miscanthus species [25,78,79]. Apart from the genotype effect, germination success of stem cuttings can be markedly impacted by the node position, the development stage at which the stem is cut and the size of the cuttings [24,25,80]. More mature and hardened basal nodes (1st–4th node from the stem base) showed a higher emergence success than the younger ones, with the best results observed from the 3rd node [27]. Based on the studies reviewed, it seems that the stem emergence potential is not influenced by the age of the mother plant, but is highly dependent on the cutting time [24,25,27,80]. Nodes taken later in the growing season tend to develop roots more easily than cuttings taken earlier in the season [25,80]. The size of the cuttings is also important: new shoots emerge more easily from longer cuttings than from small stem sections [25,80].

In a controlled environment, a temperature of 30 °C and humidity over 60% were optimal for the stem cuttings to produce roots and shoots [27,81–83]. Research studies on grass species including miscanthus showed that high photon flux densities encourage stem cuttings to shoot and increases the outgrowth of axillary buds [27,83,84]. Based on their successful use in other species [76,85], application of artificial growth stimulants such as indole-3-butyric acid (IBA), 1-naphthalene acetic acid (NAA) or abscisic acid (ABA) may help to increase rooting and sprouting ability. The studies by Defra [81,82] on miscanthus showed that pre-treatment with 4-indol-3-ylacetic acid (IAA) increased and in some cases even doubled the percentage frequency of root and shoot development from stem cuttings, in particular for the 3rd and 4th nodes.

Due to the lower input of stem harvest when compared with rhizome harvest, a cheaper stem-derived plantlet price of 0.19 € plant^{-1} was used for establishment cost calculation [86]. If the field operations shown in Table 1 are used, the costs for the establishment of miscanthus stands via stem-derived plantlets are 4,240.8 € ha^{-1} (own data shown in Table 4), which is more expensive than that of the rhizome planting (3,375.7 € ha^{-1}) but cheaper than method of rhizome-derived plants (4,400.8 € ha^{-1}). Here too it is the plant material which causes the highest costs of all the cost items (listed in Table 4). In future, the establishment of miscanthus via stem cuttings could become an easy and inexpensive option for self-propagation by farmers if the pre-treatment of miscanthus stems and rooting and emergence success can be optimized. However, this method is currently at an early stage of development and no mature technologies are available for the establishment of miscanthus from stems directly in the field or for its establishment in pots. The development of such propagation systems requires further research. The question of how miscanthus stem cuttings can be stored effectively without losing their propagation ability should also be addressed as the period between the optimal harvest time of stem cuttings (late summer) and planting time (following spring) is so long.

3.3. Micropropagation systems

Micropropagation is a technique that manipulates small quantities of axenic plant material under favourable conditions to form new clonal offspring [87]. It has proven to be a good approach for disease-free plant production, germplasm conservation, to facilitate international germplasm exchange and to produce large number of

Table 4
Estimated miscanthus establishment costs for different establishment options, including rhizome planting, rhizome-derived plants, stem-derived plants, seed sowing and micropropagation.

Cost items	Rhizome planting (€ ha^{-1})	Rhizome-derived plants (€ ha^{-1})	Stem-derived plants (€ ha^{-1})	Seed sowing (€ ha^{-1})	Micropropagation (€ ha^{-1})
Land costs	414.3	414.3	414.3	414.3	414.3
Agro-chemicals	212.6	171.3	171.3	98.4	171.3
Planting materials	2,400.0	3,200.0	3,040.0	406.6	5,120.0
Fixed machine costs	113.2	133.0	133.0	109.1	133.0
Variable machine costs	235.6	359.8	359.8	311.4	359.8
Irrigation	0.0	112.4	112.4	168.7	112.4
Total costs	**3,375.7**	**4,400.8**	**4,240.8**	**1,508.5**	**6,320.8**

Note: The fertilizer and herbicide input are combined in agro-chemical costs; the fixed machine costs include depreciation, repair and maintenance, insurance and storage costs; the variable machine costs were divided into fuel, labour and lubricant costs; the irrigation costs include the cost of water, labour and energy.

Table 5
Overview of factors influencing the establishment of stem-propagated miscanthus and the best propagation options.

Factors influencing establishment	Alternatives tested	Best options	Source
Genotype	*M. × giganteus*, *M. sinensis* and *M. sacchriflorus*	*M. × giganteus* and *M. sacchriflorus*	[25,30,31,78,79]
Age of mother plant	1-year and 20-year old plants	No significant influence	[24,25,27]
Development stage/ cutting stage	May to September	Later growing season (September)	[24,25,27,80,81]
Size of stem cuttings	5 cm With one node and 10 cm with two nodes	10 cm Nodal stem cuttings	[25]
Node position	1st to 17th Basal node	Node 1–4 (most mature nodes)	[24,25,27,81]
Pre-growing temperature	15, 20, 25 and 30 °C	30 °C	[27,81,82]
Pre-growing humidity	N/A	Humidity > 60% is required	[82,83]
Pre-growing light	Light/dark period of 0 h/24 h and 16 h/8 h	No significant influence but longer lighting period is better	[27,83]
Growth stimulants	Indole-3-butyric acid (IBA), 1-naphthalene acetic acid (NAA) & 4-indol-3-ylacetic acid (IAA)	1-Naphthalene acetic acid (NAA) and 4-indol-3-ylacetic acid (IAA)	[81,82]

N/A represents no information available.

clones from agronomic crops [88]. Previous studies have shown that micropropagation of miscanthus can be achieved by direct shoot regeneration from nodal segments (nodal stem or rhizome) and apical meristems followed by in-vitro rooting or by callus culture through inducing somatic embryogenesis from immature inflorescences, leaf sections or shoot apices [29,37,89,90,91]. After root induction, plantlets with roots are transplanted into pots for hardening under controlled conditions and later planted in the field. A cold acclimatization before planting improves the cold tolerance of micropropagated plants [91,92].

Currently miscanthus is not commercially propagated through callus culture because the three-step procedure of micropropagation requires higher labour input and expenses compared to the other methods. The step of somatic embryogenesis can also lead to mutations and genetic alterations of the crop [93,94]. Hence, the micropropagated miscanthus plants currently available on the market are propagated via direct in-vitro shoot induction from nodal stems or terminal buds. Since micropropagation methods and culture media compositions are not made public by companies, little can be found in the literature on micropropagation of miscanthus via direct shoot regeneration. Table 6 gives an overview of the available information.

For micropropagation through callus culture, the success of callus induction mainly depends on the genotype, the kind of plant material used and the development stage of the material. Overall higher callus induction rates were observed for *M. × giganteus* than for *M. sinensis* [96]. In several studies immature inflorescences showed higher regeneration capacity than tissue from leaves and shoot apices; no embryogenic calluses were formed by root tissue [36,37,96,103,104]. Callus induction and shoot regeneration success were lower when older plants were taken as a source [36,96]. An appropriate mixture and concentrations of growth regulators are required to regenerate shoots and induce roots efficiently, e.g. 2,4-Dichlorophenoxy acetic acid (2,4-D), 6-Benzylaminopurine (BAP) and indole-3-butyric acid (IBA). Components which prevent browning of explants (L-cysteine-HCL, proline, activated charcoal) are also considered essential [29,37,91,93,99,101]. A full protocol for the media and phytohormones applied for the steps of embryogenic callus production, shoot regeneration and rooting is provided by Lewandowski [37]. To perform year-round propagation via callus, effective callus storage methods are necessary. Storage on solid rooting medium is more successful than that on proliferation medium. The best option is to store calluses in cell suspension cultures with low nitrogen and abscisic acid (ABA) concentration [92,102].

Table 6

Factors that influence micropropagation systems including direct shoot induction and callus culture and best propagation options.

Type of micropropagation method	Factors influencing plant induction efficiency	Alternatives tested	Best options	Source
Direct shoot induction and in-vitro tillering	Genotype	*M. × giganteus, M. sinensis, M. sacchariflorus*	*M. sinensis* required young shoot explants; *M. × giganteus* required non-detached axillary buds	[22,29,37,95]
	Explant sources	Young shoot, isolated and non-detached axillary buds		
	Sterilized material	N/A	Ethanol, NaOCl, calcium hypochloride, sodium hypochlorid	[29,37,95]
	Medium type	Solidified and Liquid MS medium	Solidified medium for shoot induction; Liquid medium for in vitro tillering and root induction	[22,29,95]
	Method of preventing browning of explants	Pre-soaking in solution containing citric acid/ascorbic acid; adding L-cysteine-HCl/polyvinylpyrrolidone/active charcoal to cultivation medium	L-Cysteine-HCl supplemented to medium, especially for the genotypes of *M. sinensis* Silberfeder and *M. × giganteus*	[29]
	Growth regulator	N/A	6-Benzylaminopurine (BAP), 1-naphthaleneacetic (NAA), indole 3-acetic acid (IAA)	[22,29,37,95]
	Cultivation conditions	N/A	24–26 °C/20 °C and 16 h light/8 h dark for the day/night period	[29,95]
	In-vitro tillering/rooting medium status	N/A	Liquid medium including $20\ \mathrm{g\,l^{-1}}$ sugar, $4.3\ \mathrm{g\,l^{-1}}$ MS medium with vitamins (Duchefa Biochemie BV, the Netherlands) and $0.3\ \mathrm{mg\,l^{-1}}$ BAP	Unpublished data
Callus culture	Genotype	*M. × giganteus, M. sinensis* & *M. sacchriflorus*	A higher callus induction rate by *M. × giganteus*	[22,91, 92,96–98]
	Explant sources	Immature inflorescences, leaf sections, shoot apices, rhizome buds and root tissue	Immature inflorescences	[36,37,96]
	Explant development stage	Immature inflorescences in two developmental stages with length of 0.1–2.5 cm and 2.6–5 cm, respectively	A young explant, e.g. 0.5–1.5 cm inflorescence, is required	[29,96]
	Sterilized material	N/A	Ethanol, NaOCl, calcium hypochloride, sodium hypochloride	[28,29,37,96,99]
	Medium type	B5, SH, Jensen 25–8 and MS medium	Liquid MS medium with suitable nitrogen and phosphorus content	[89,96,99,100]
	Method of preventing browning of explants	N/A	Adding L-cysteine-HCl, proline or activated charocoal to the medium	[37,92,101]
	Growth regulator	BAP, 2,4-D, IBA, 2.4.5-T, kinetin, zeatin	BAP, 2,4-D, IBA	[37,89,91,93,99]
	Callus storage condition	Temperature of 8, 12, 16, 20 °C with photosynthetic photon flux densities of 5, 10, 20 $\mu\mathrm{mol\,m^{-2}\,s^{-1}}$ on rooting medium and proliferation medium, respectively	Temperature of 8–16 °C with photosynthetic photon flux densities of 20 $\mu\mathrm{mol\,m^{-2}\,s^{-1}}$ on rooting medium	[102]
	Cultivation condition	N/A	24–26 °C with 65% humidity in the dark for callus induction; 24–26 °C with a 16 h photoperiod for plant regeneration	[28,91,96,99]

N/A represents no information available for comparison.

Studies on direct shoot induction and in-vitro tillering methods also show the effect of genotype, explant source, medium type and addition of particular medium components (e.g. growth regulators) on plant regeneration. For efficient regeneration of *M. sinensis* and *M. sacchariflorus*, use of young shoot explants gives better results; while non-detached axillary buds with stem segments (20 mm long) are better material for *M.* × *giganteus* in-vitro initiation (Table 6). Addition of L-cysteine-HCl to the shoot regeneration medium is recommended to prevent explant browning of *M. sinensis* and *M.* × *giganteus* [37,29,105]. The authors' own practical experience has shown that an appropriate medium for miscanthus direct shoot induction can be a liquid medium containing 20 g l^{-1} sugar, 4.3 g l^{-1} MS medium [106] with vitamins (Duchefa Biochemie BV, the Netherlands) and 0.3 mg l^{-1} BAP. As with rhizome propagation, micropropagation of miscanthus is currently implemented but best propagation procedures are not made public. More research needs to be carried out on the development of less expensive micropropagation options, in particular the reduction of the manual labour required and automation of the propagation process.

Based on 2000 prices, the 2012 converted price for micropropagated plantlets should be around 0.32 € plantlet^{-1} [67]. Following the summarized field operations in Table 1, the costs for the establishment of miscanthus stands via micropropagated plantlets are calculated as 6,320.8 € ha^{-1} (own data shown in Table 4). This is the most expensive of the available propagation systems. The largest contribution to these high establishment costs comes from the plant material input (5,120.0 € ha^{-1}), which is 2.1, 1.6, 1.7 and 12.6 times greater than that of rhizome cuttings, rhizome-derived plants, stem-derived plants and seeds, respectively (Table 4).

3.4. Seed propagation system

Seeding is the most widely used method of crop establishment on account of its high production efficiency and low costs. However seeding is not yet applied in commercial miscanthus production because the most widely grown genotype *M.* × *giganteus* is a triploid hybrid that does not produce seed. Seed propagation is however possible in *M. sinensis* and a newly developed *M.* × *giganteus* cultivar which can produce viable seeds [107,108]. In field trials in the UK, for example, *M. sinensis* seeds showed good germination after six days and could even be established in the field without pelleting [19]. Seed establishment of *M. sinensis* can be achieved under a wide set of environmental conditions, with air temperatures ranging from 5.3 to 30 °C and soils with pH values between 4.3 and 8.5 [109,110]. The miscanthus species suitable as parental lines for seed production are primarily *M. sinensis* and the *M. sacchariflorus* subspecies 'Robustus' [111]. These are adapted to a temperate climate and can flower and set seeds in these conditions. Other interesting parent materials for seed propagation could be the *M. lutarioriparia* and *M. floridulus*, which are both characterized by high seed setting rates and a biomass yield potential that reaches up to that of *M.* × *giganteus* [112].

Improving seed production in miscanthus should be based on understanding the factors which can affect seed setting: the genotype effect, cross compatibility and the environmental conditions in which the plants are grown and seeds are ripened [67,113]. Previous research has shown that a higher seed setting rate is achieved by *M. sinensis* than *M. sacchariflorus* [113]. Interspecific hybrids, e.g. the natural interspecific triploid hybrid *M.* × *giganteus* ($2n=3x=57$), are usually characterized by low pollen fertility and low or no seed formation [33,114,115]. The differences in pollen quality between genotypes may be responsible for the different seed setting rates in miscanthus. For a better fertility rate, a large growth area with a high diversity of genotypes is often more beneficial than a small isolated stand [113]. Short-day conditions and good water and nitrogen supply are important for floral induction and seed formation in miscanthus [113,116].

In this study the seed setting rate of five different miscanthus genotypes from a 15-year old field trial in south-west Germany was assessed. It was found that *M. sinensis* produced viable seeds with a seed setting rate ranging from 0.4 to 28.7%, with individual panicles containing an average of 4.4 (genotype 88-111) to 190.9 (genotype SW217) seeds (Table 7). The highest seed counts were observed for the open-pollinated *M. sinensis* genotype SW217. No fertile seeds were produced by the hybrids (GOFAL 7 and RH 81) in Exp 1 (Table 7). When panicles were covered by pollination bags for self-pollination, few or no seeds were produced (seed setting rate less than 0.58%) by the *M. sinensis* genotypes as well as by the hybrids, indicating a possible self-incompatibility in *M. sinensis* (unpublished data). No significant differences in seed size were observed between the genotypes. The seeds of *M. sinensis* were small with an average length of 2.5 mm and width of 0.92 mm. Under controlled conditions in the climate chamber, the *M. sinensis* seeds showed a high germination rate which varied significantly ($P < 0.05$) from 88.1% to 98.3% depending on the genotype (Table 7).

To improve seed establishment in the field, the transplanting of seed-derived plantlets can be practised. This method enables field establishment rates of 60–100%, based on the authors' own field trial experience (unpublished data). Compared to plants derived from the traditional method of rhizome planting, seed-propagated plants usually produce fewer, smaller and thinner shoots; therefore the biomass yield of seed-propagated plants is expected to be lower than that of the rhizome-derived plants in the first growing season.

It is known that miscanthus seeds have low dormancy and air moisture additionally decreases seed viability during storage [117]. To sustain the viability of fertile seeds over a long period, mature seeds should be kept in cool, dry conditions. Hsu [118] and Xi and Zhang [35] found that, when stored at room temperature, the germination ability of *M. sinensis* seeds decreases dramatically after six months and most genotypes of this species showed poor germination after two years. This is also true for *M. sacchariflorus*, *M. lutarioriparius* and for hybrids [35]. Therefore, when seeds need to be stored for longer than six months, they should be kept in a cold chamber, in particular *M. sacchariflorus* which shows shorter seedlife than *M. sinensis* and *M. lutarioriparius* [35].

To date, seed establishment of miscanthus is not used for commercial planting, therefore no best practice can be identified. Previous studies of Aso [109], Clifton-Brown [110], and Christian [119] on seed germination in *M. sinensis* show that no germination takes place at temperatures below 5.3 °C, whereas good emergence can be achieved at temperatures of 20 to 30 °C. Several other studies showed no effect of the size of miscanthus seeds on the final seed germination rate, but larger seeds germinated earlier than smaller ones [109,119,120].

Table 7
Seed-setting rate and seed germination of different genotypes grown in south-west Germany (Exp 1&2).

Species	Genotype	Spikelet No./Panicle	Seed setting rate/Panicle (%)	Seed length (mm)	Seed width (mm)	Seed germination ratio (%)
M. sinensis hybrids	GOFAL 7	1046.2 ± 256.6a	0.0	–	–	–
	RH81	667.5 ± 229.8b	0.0	–	–	–
M. sinensis	88-111	1033.8 ± 221.2a	0.4 ± 0.1c	2.28 ± 0.19a	0.86 ± 0.12a	98.3 ± 2.9a
	90-6	577.4 ± 180.2b	12.5 ± 0.1b	2.65 ± 0.27a	0.93 ± 0.16a	92.7 ± 1.7b
	SW217	665.4 ± 219.5b	28.7 ± 3.7a	2.44 ± 0.21a	0.96 ± 0.08a	88.1 ± 3.8c

Mean values marked with the same letter are not significantly different at the 0.05 level of probability.

Application of the artificial growth stimulants gibberellic acid (GA3) and BAP increases the seed germination rate in miscanthus [109,121]. Several field studies have shown that seed germination and rooting potential are highly dependent on seedbed quality, including water content, pH and heavy metal concentrations [19,109,122,123]. Drilled seeds germinate better than those sown on the soil surface [19]. Work on another small-seed C4 grass, switchgrass (*Panicum virgatum* L.), has also shown that germination on rolled soil is significantly higher than on tilled and unrolled soil [45]. Close contact between the soil and the seeds seem to play an important role in the germination and early growth of miscanthus, which may explain the poor germination of unpeeled and coated seeds [19,124].

Due to the small size of miscanthus seeds, which results in low water-absorbing ability, there is a high risk of seedlings dying from desiccation, especially in warm, dry conditions. Drilling seeds into soil, followed by rolling and irrigation could form a close seed-to-soil hydraulic contact leading to better germination and seedling survival. Weed control is important when establishing miscanthus via seeds to minimize the competition faced by the newly established small seedlings. Another reason for low seed viability could be seed damage during threshing and cleaning, which for breeding and small-scale experiment purposes is usually performed manually. Initial seed-coating experiments have shown that larger, more uniform seeds could be sown directly using standard sowing machinery [19,121]. Seed coating could be beneficial for miscanthus seed establishment, e.g. by improving the water and nutrient supply to the emerging seedlings. However, further research needs to be conducted to develop the optimal coating materials and methods for miscanthus seeds. In addition, for commercial seed production seed harvesting and threshing procedures will have to be optimised and mechanised.

Based on the comparison of establishment costs between rhizome and seed propagation systems provided by Panter [34], a price of 84.7 € kg^{-1} pelleted seeds was calculated. Combined with the establishment procedures shown in Table 1, the costs calculated for the establishment of miscanthus via seed are 1,508.5 € ha^{-1} (own data shown in Table 4), which makes the seed-based system one of the cheapest miscanthus propagation options. To realize the economic potential of the seed propagation system in future, breeding new fertile varieties with biomass yields and economic profits at least as high as those of the traditional *M.* × *giganteus* will be necessary.

4. Discussion

Since no apparent yield differences between the propagation variants were reported from the 3rd growing season onwards [22,30], the propagation systems do not vary in the management, harvest, storage and transportation phases, but for the most part only in the establishment phase. Hence, the major factors determining the competitiveness of each system are the multiplication ratio, establishment costs and complexity of the problems discussed in Table 8. Although the propagation methods described above are an improvement on initial technologies, there are still

Table 8

Strengths, weaknesses and problems of the main optional miscanthus propagation methods: direct rhizome planting (RD), rhizome-derived plants (RP), stem-derived plants (SP), direct seed sowing (SD) and micropropagation (MP).

Propagation methods	Strengths	Weaknesses	Problems
RD	• Availably commercial production with mature technology • High establishment (85%) and winter survival success (> 90%) potential • Medium establishment costs (3,375.7 € ha^{-1}); • Farmer can use self-propagated plant material for establishment.	• Low parental plant division efficiency (1:10) • Labour-intensive for sorting and cutting of rhizomes • Short rhizomes storage period after harvest (max. 4 months)	• Risk of diseases transmitting and spreading from infected mother plants • Potential gap between demand for and supply of rhizomes for large-scale plantation • High damage rate (40%) may be caused by unsuitable harvest technology and storage conditions
SP & RP	• Young material origin (1 to 2 years old) is available • High establishment (> 95%) and winter survival (> 90%) success potential • Enhanced multiplication ratio (1:30 for RP & 1:120 for SP) with reduced establishment costs of RP (2,640.8 € ha^{-1})	• Care in the field is needed for good establishment (initial irrigation, weeding) • Increased labour, energy input for pre-grown plantlets	• Low germination and establishment success possible in unsuitable conditions with low temperature and desiccation
SD	• New varieties with good performance can be bred and current equipment can be used • Extremely high multiplication ratio (1:1,172) • Potentially lowest costs (1,508.5 € ha^{-1}) of all methods • Seeds can be stored for a long time (max. 2 years) • Undamaged donor plants • Hybrid without seed production is less invasive	• Geographical restrictions (short day and high temperature) for seed production • More care in the field after drilling is needed for good establishment (initial irrigation, weeding) • Low field germination (< 10%) and potentially emerging plants can die due to desiccation and competition from weeds	• Technical challenges involved in seed harvesting, threshing and pelleting • Problems of genetic diversity and production of parental lines for breeding not yet overcome
MP	• Better control over the plants diseases • Fast bulk-up and year-round operation is available • Also extreme high multiplication ratio (1:960) • Seedlings can be maintained for a long time (> 1 year) • Undamaged donor plants	• Currently the most expensive method (6,320.8 € ha^{-1}) • Care in the field is needed for good establishment (initial irrigation, weeding)	• Possible low rate of winter survival in cold regions

some weaknesses which need to be addressed. The following sections discuss the technological advancements in miscanthus propagation and future requirements for their enhancement, addressing the two main questions: "What are the future options for the propagation and establishment of miscanthus?" and "What type of research should be conducted to reduce establishment costs and thus improve the potential of each system?"

During the early phases of miscanthus research in Europe (mid-1980s to early 1990s) propagation work focused on macropropagation using manually-propagated rhizomes [22,125,126]. At that time, the Danish Research Centre Foulum began research on the mechanization of rhizome propagation; however, results were not published in the scientific literature. In the mid-1990s, the earliest commercial rhizome planter was developed by the former company BICAL and Hvidsted Energy Forest [127,128]. The relevant development steps in rhizome propagation were performed in practice by farmers and private companies (e.g. Tomax Ltd., Portlaw Co., ADAS Ltd.), who developed automatic rhizome planters and mechanical rhizome harvesters [10,32]. Researchers contributed to the development in rhizome propagation by increasing the multiplication ratio through reducing rhizome cuttings' size [10,26,62]. Nowadays farmers widely apply rhizome planting methods for miscanthus establishment and several farmers and companies (e.g. The New Energy Farm) offer both miscanthus rhizome materials and planting/establishment services. The multiplication ratio of current rhizome-based propagation systems has been improved. A recent technical novelty in rhizome propagation is the CEED technology, which has increased the division efficiency to 1:30 by using 'micro-rhizomes' and enhanced the establishment success by using coatings [38]. Because the technology of this propagation method was developed by private entities, details have not been published or otherwise made public. However, the multiplication efficiency is still low [10,60] and this accounts for the slow increase in cultivation area and the large gap between the demand for and supply of rhizomes for large-scale plantations, despite rhizome planting generally being characterised by a high establishment and winter survival success (85% and 90%, respectively) [129]. Another concern is that virus-free rhizome production has not yet been a subject of research or farmers' attention. The rhizome planting procedure currently used could lead to disease transmission through infected mother plants. It is therefore expected that in future standards will be set for virus-free rhizome planting material sold commercially. In addition, the different methods of rhizome harvesting and planting presently used lead to significant variation in rhizome quality. From the authors' own communication with farmers it is clear that quality control and sorting of rhizomes is performed with various input levels of manual labour. However, it is questionable whether more efficient rhizome harvest and propagation equipment will be developed in the near future as the miscanthus market is currently not large enough to make it profitable for a machinery manufacturer. It is therefore likely that farmers will continue to develop or adapt their own equipment. Thus rhizome planting may remain the preferred establishment method for agricultural trials and small-scale plantations for the foreseeable future, especially as long as *M.* × *giganteus* dominates the commercial miscanthus market. In summary, the present situation is that rhizome planting is characterized by 'farmer-made' methods that work well but are not documented (as farmers do not want to disclose their competitive knowledge).

The main reason for developing propagation methods based on pre-grown plantlets, such as rhizome- or stem-derived plants, is the need to reduce the cost of planting material by increasing the multiplication ratio and establishment survival. The possibility of propagating miscanthus using stem segments was investigated and confirmed in 1988 by German LWG (Bayerische Landesanstalt für Weinbau und Gartenbau) and BFH (Bundesforschun- gsanstalt für Forst- und Holzwirtschaft) [61]. With the initial technologies, the stem-propagation method was characterized by low rooting ability and high input requirements due to manual harvest and planting. The need to store the stem-propagated plants during winter renders this method inefficient and uneconomic. Hence little attention has been paid to the improvement of stem-propagation methods. It was not until 2007, when the insufficient quality of rhizome material became apparent, that stem propagation was again considered as a possible option for miscanthus propagation [24]. Following recent improvements, the multiplication ratio has been increased to 1:120 [130] and establishment survival percentage to 80% [130,131]. Additionally, application of growth stimulators could increase the stem-rooting potential especially with nodal cuttings from the 3rd nodes onward. This means not only establishment survival but also the multiplication ratio could be increased, thus reducing establishment costs and increasing the competitiveness of the stem-derived plant method. Due to higher labour and energy inputs required for the pre-growing of plantlets, the estimated establishment costs of the stem-derived plant method are higher than those of rhizome planting at 4,240.8 € ha^{-1}. These high input requirements mean there is only limited cost reduction potential for rhizome- or stem-derived plants. Moreover, the high sensitivity of the plantlets to environmental stresses, such as low temperature and desiccation, calls for greater field care after planting, further limiting the cost reduction potential. The same is true for rhizome-derived plantlets. Therefore, the current methods of transplanting pre-grown plantlets derived from rhizomes or stem segments can only be used on a small scale, for example in research trials.

Initially miscanthus micropropagation was developed and documented in public research and then further developed by private companies. The enhanced but unpublished technologies used by these companies led to the micropropagated plants becoming cheaper than rhizome-propagated plants and dominating the miscanthus planting material supply in the early 1990s [37]. But the cost of rhizome-propagated plants soon began to fall as rhizome propagation was mechanised [126,128]. By contrast, the level of manual work required in micropropagation remained high and little was done to mechanise this propagation method. From 2000 onwards, micropropagated plants were more expensive than rhizome-propagated plants [67] and no further improvements were made in miscanthus micropropagation by commercial companies. Although some research efforts focused on the optimization of the culture media for callus induction and shoot regeneration [28,29,36,37,132,133], little work was done on automated micropropagation [134]. Hence to date, micropropagation still requires high labour and energy input for the multi-step plant regeneration and cultivation processes. For these reasons, micropropagated plantlets are expensive, limiting the practical large-scale application of micropropagation. Future research should therefore be focused on reducing labour costs, e.g. by developing robots or automated transfer of in-vitro plants to pots. Although the most expensive, miscanthus micropropagation is characterized by a very high multiplication ratio of 1:960 [29,96]. Other characteristics of micropropagation are its theoretically unlimited production capacity and long material storage period. These could ensure the fast bulk-up and year-round production of healthy plants. Another concern is poor winter survival of micropropagated plants, which leads to considerable financial risk [91,135]. Therefore a further line of research could be the improvement of cold tolerance in micropropagated plants. Artificial seeds from micropropagated somatic embryos could become a good alternative for miscanthus propagation in the future but a great deal of research still needs to be done in this area. In summary, miscanthus micropropagation has increasingly become a propagation method used for scientific and breeding purposes, but which finds little application in commercial production.

Compared to the initial situation where miscanthus propagation via seeds was impossible on account of the sterility of the only commercially available genotype, *M.* × *giganteus*, today the prospect of seed establishment has significantly improved. This is mainly due to the breeding of new lines and to improved field management methods [19,34,108]. Theoretically, the seed propagation system provides the highest multiplication ratio (1:1,172) and lowest establishment costs (1,508.5 € ha^{-1}) of all the available methods and the seed-propagated plants can produce similar biomass to the rhizome-propagated plants after two growing seasons [108], indicating a promising future for this propagation system. Another advantage of the seed propagation method is that new varieties with good performance can be bred for different climate zones, which could end the present dominance of *M.* × *giganteus* on the commercial miscanthus market. This could also reduce the invasiveness potential of miscanthus because few fertile seeds can be derived by miscanthus hybrids due to their poor pollen quality [19,114]. However, in relatively dry conditions, seed sowing is currently still considered unreliable and is not practiced for miscanthus propagation [136–139] because of low field germination rates and high seedling mortality. The small seeds do not contain much water or carbohydrates for germination and development and the seedlings' short roots limit their ability to absorb water from deep soil, making the risk of low seed germination in the field ($<10\%$) high. Young seedlings often die from desiccation or weed competition. This problem could be overcome by either raising plantlets from seeds in the greenhouse or coating seeds. In future more attention should be paid to the development of seed coating material. Direct drilling of coated seeds would be cheaper and more flexible (no need to pre-grow plants under controlled conditions) and therefore could become the preferred option for farmers. The coating material can be enriched with fungicide, growth stimulators and nutrients to support germination and initial plant growth. Micro-nutrients necessary for germination can be supplied in the optimal dose. The coating can also be made water-absorbent to protect the seedlings from desiccation. These advantages of coating material are probably also used by the recently developed CEED technology, which is coating vegetative plant material instead of seeds.

Although there have been some reports of successful miscanthus field seed sowing, almost all of this success has been achieved with the relatively low-yielding *M. sinensis* [19,107,109,119,139] rather than the highly productive hybrids. This indicates the lack of commercial hybrids as a further limitation to the development of seed propagation. Work on breeding commercial varieties has only recently begun and needs to overcome the bottlenecks of low genetic diversity. Flowering synchronization of parental lines is also a problem which needs to be overcome because miscanthus is largely self-incompatible [113]. It is therefore expected that in future more effort will be made in miscanthus breeding programmes to broaden the gene pool and to develop parental lines for targeted crosses. Breeding work carried out in China and America has shown that hybrids between *M. sinensis* and *M. lutarioriparia* can be successfully produced via seeds, indicating that the seed sowing method may soon be available for commercial use [34,124]. In Europe, there are miscanthus breeding programs at Aberystwyth University and Wageningen University. These programs aim to genetically improve miscanthus, to increase biomass yield and composition with regard to energetic use and ethanol production, to improve the tolerance to abiotic and biotic stress and to reduce propagation costs and increase seed-setting rates [140,141].

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Chapter 3 Assessment of marginal land potentials for the *Miscanthus* production - a case study of China

The analyses in Chapter 1 also show that the expansion of miscanthus production is facing a land use dilemma, i.e. lack of land available for growing miscanthus. A potential way to address the challenge is cultivating miscanthus on barren land with natural condition which is not well suited to agricultural production but suitable for growing plants with resistance to environmental stresses, i.e. marginal land. To assess the marginal land potential for the miscanthus production, a case study of China was conducted here. In the present study, Geographic Information System (GIS) techniques, model simulation were adopted to identify the productive marginal areas in China for miscanthus and to estimate their biomass and bioenergy production potential.

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Assessment of the production potentials of *Miscanthus* on marginal land in China

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ABSTRACT

Miscanthus is characterized by high biomass production potential and low input requirements and therefore considered a leading candidate for second-generation energy crops. Because China has limited agricultural land resources, development of its miscanthus-based bioenergy industry must rely on the use of marginal land. This study focuses on the assessment of the production potential of miscanthus on China's marginal land, which in this context is defined as land presently not used for agricultural production, residential purposes and other social uses. Geographic Information System (GIS) techniques and model simulation are adopted to identify the productive marginal areas for miscanthus and to estimate their biomass and bioenergy production potential. The results show that although a large marginal area of $17{,}163.54 \times 10^4$ ha is available for producing miscanthus, due to the limitation of low winter temperatures and low precipitation levels in some areas, the total suitable marginal area is only 769.37×10^4 ha. The Monteith radiation yield model was used to determine the potential miscanthus yield in Chinese climatic conditions. The simulation gave actual harvestable yield levels on arable land of 18.1–44.2 t ha^{-1} yr^{-1}. Taking the environmental stresses of marginal conditions into account then gave an achievable miscanthus yield potential on marginal land of 2.1–32.4 t ha^{-1} yr^{-1} (average for the different marginal lands). Based on these achievable yield levels, the total biomass production potential on the entire marginal area is $13{,}521.7 \times 10^4$ t yr^{-1}; the bio-electricity generation and total greenhouse gas saving potential from these biomasses are 183.9 TW h yr^{-1} and $21{,}242.4 \times 10^4$ t CO_2 eq. yr^{-1}, respectively. The spatial distribution of the suitable marginal areas shows that they are mainly concentrated in the central part of Northeast China and the Loess Plateau. Both regions are recommended as priority development zones for the Chinese miscanthus-based bioenergy industry.

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1. Introduction

The substitution of fossil energy by bioenergy is a promising and multifunctional way to protect national energy security, slow down climate change and improve rural economy [1]. This is also of great interest and significance for China, which has a large gap between the demand for and supply of fossil energy. Additionally, bioenergy development is also considered a new way to promote rural economic development in China [2,3]. Hence, the Chinese government has gone to great effort to advance its bioenergy industry. In the past decade, the Chinese bioenergy market has been dominated by bio-ethanol produced from grains that had been stored for more than 3 years [4,5]. In 2008, approximately 1.5 million tons bio-ethanol were produced from maize and wheat, accounting for 79% of total biofuel production, but offsetting only 0.4% of the total annual fossil fuel consumption [6]. Even though only a small proportion of fossil fuels is replaced, there is still concern about effect on national food security [7]. For this reason, in 2007 the Chinese government stopped approving and constructing new grain-based bio-ethanol production programmes and facilities. At the same time a ‘non-food’ principle, which stipulates the use of non-food biomass for bioenergy generation and non-farm land for energy crop production, was set for the future bioenergy industry [8,9].

In China, non-food biomass is generally divided into the following categories: agricultural residues, forest biomass (including firewood and forest residues), manure, municipal waste and industrial waste (mainly organic wastewater). Their estimated yield potentials in Mt yr^{-1} are 728–750 [10,11], 200–220 [8,12–13], 220–280 [8,13,14], 155 [10,15] and 48,240 [10], respectively. However, this large potential can only partly be exploited due to utilization for other purposes and other limiting factors. For example, if the demand for straw for papermaking and animal feed is subtracted, the agricultural residue potential remaining for the bioenergy industry is limited to 314 Mt yr^{-1} [11], less than half of the overall potential. Furthermore, once factors such as harvest and transportation limitations are taken into account, the final amount of agricultural residues available for bioenergy production will be even lower [16,17]. This is also true for forest biomass potential, which is reduced to 28.1 Mt yr^{-1} for the bioenergy industry through the competitive use of firewood by rural residents and also due to harvest and transportation limitations [8,10]. Finally, the total annually acquirable quantity of non-food biomass for bioenergy production in China is approximately 210 Mtoe (million tons of coal equivalents) [8,10]. This amount is not sufficient to satisfy the feedstock demand of the future Chinese bioenergy industry given its aspiration to achieve 5% of gross energy consumption by 2020 (4700 Mtoe) [18]. Therefore, cultivation of energy crops is required to close the gap between biomass feedstock supply and demand. Considering the ‘non-food’ principle, energy crop production in China should be based on non-food energy crops and marginal land traditionally not considered agricultural land [8,9].

Miscanthus (*Miscanthus spp.*) is a promising non-food energy crop, which has origin and widespread distribution in China [19,20]. In Europe, miscanthus has been researched and developed as energy crop for over two decades and is currently used mainly for heat and electricity generation [21]. In China, crop residues are presently the main feedstock for bio-electricity generation. Lignocellulosic materials such as miscanthus biomass are currently still not used for bio-ethanol production because the technology for cellulosic ethanol production is not yet commercially available [22]. For these reasons, there has so far been no commercial effort in China to develop miscanthus as a feedstock for bioenergy generation. However, the following outstanding characteristics could make miscanthus a promising energy crop for Chinese farmers. The perennial rhizomatous miscanthus has a lifetime of over 20 years, and lower energy demands and production costs than annual energy crops such as sweet sorghum (*Sorghum bicolour* (L.) Moench) [21,23,24]. Additionally, miscanthus is characterized by a nutrient relocation mechanism [25,26], high water-use efficiency [27,28] and low susceptibility to diseases and pests [29,30]. Therefore, miscanthus has low demand for fertilizer and pesticides. Due to its C4 photosynthetic pathway, it has a high biomass production potential. In addition, its ability to grow in marginal conditions, such as drought and salinity [31,32], would make miscanthus the ideal crop for the future Chinese ‘non-food’ bioenergy industry.

According to an evaluation by the Chinese Ministry of Agriculture, there are 27 million ha marginal land that could be used for the cultivation of energy crops [33]. If all these resources were used to produce 1st generation energy crops (i.e. starch and sugar plants), the estimated bio-ethanol generation potential would be 74 million tons [34], assuming these crops produced economic yields on marginal land. However, not all these marginal land areas will actually be suitable for planting energy crops (including miscanthus) due to environmental stresses such as drought and cold. In addition, the areas of marginal land suitable for growing specific crops differ according to the varying environmental requirements of these crops [35,36]. For example, there are 4 million ha marginal land (mainly located in Northeast China) suitable for sweet sorghum production [37], while only 0.015 million ha (mainly located in South China) suitable for cassava (*Manihot esculenta* Crantz.) production [38]. For the most promising non-food energy crop-miscanthus, there are as yet no studies on its production potential on marginal land in China. Therefore, the objectives of this study are to assess the biomass and bioenergy production potential and the greenhouse gas (GHG) saving potential of miscanthus grown on marginal land in China. For this purpose, the marginal land area available and suitable for cultivating miscanthus in China is identified and yield potentials of miscanthus species well adapted to these land areas are modelled. Finally, the bioenergy generation and GHG saving potentials are assessed, working on the assumption that all biomass is combusted for bio-electricity generation.

2. Methodology

The biomass and bioenergy production and GHG saving potentials of miscanthus on marginal land in China were assessed in four steps:

Step 1: Identification of available marginal land (defined in Table 1). The spatial distribution (Fig. 1) and total area of marginal land available for the cultivation of miscanthus were identified.

Step 2: Identification of miscanthus yield zones. Firstly, the environmental requirements for the continuous growth of several promising miscanthus species were identified. Secondly,

Table 1
Definitions used in this study for land and yield categorization.

Items	Definition
Suitable land for bioenergy plants	Land that is neither temporarily nor permanently used for agricultural production and with natural conditions rendering it suitable for growing bioenergy plants. This includes fallow land and suitable marginal land.
Marginal land	Land that is not used for agricultural production, residential purposes and other social uses.
Available marginal land	Marginal land types that could be used for miscanthus cultivation without conflicting with political factors (e.g. laws and governmental strategic planning) and environment protection concerns (e.g. ecological functions and services of marginal land should not be destroyed by miscanthus cultivation).
Miscanthus production area	Area with natural conditions of Temp ≥ −23 °C and Prec ≥ 400 mm that can support continuous biomass production of miscanthus.
Suitable marginal land for miscanthus	Available marginal land that is characterized by natural conditions of Temp ≥ −23 °C and Prec ≥ 400 mm, i.e. the available marginal land within the miscanthus production area.
Harvestable yield	Peak aboveground biomass yield (i.e. autumn yield) that is continuously produced by miscanthus grown on arable land in given climatic conditions.
Achievable yield on marginal land	Harvestable yield remaining after subtracting of aboveground biomass losses due to the constraints of marginal factors, e.g. drought, cold.

Note: Temp = minimum monthly-averaged lowest air temperature; Prec = averaged annual accumulated precipitation.

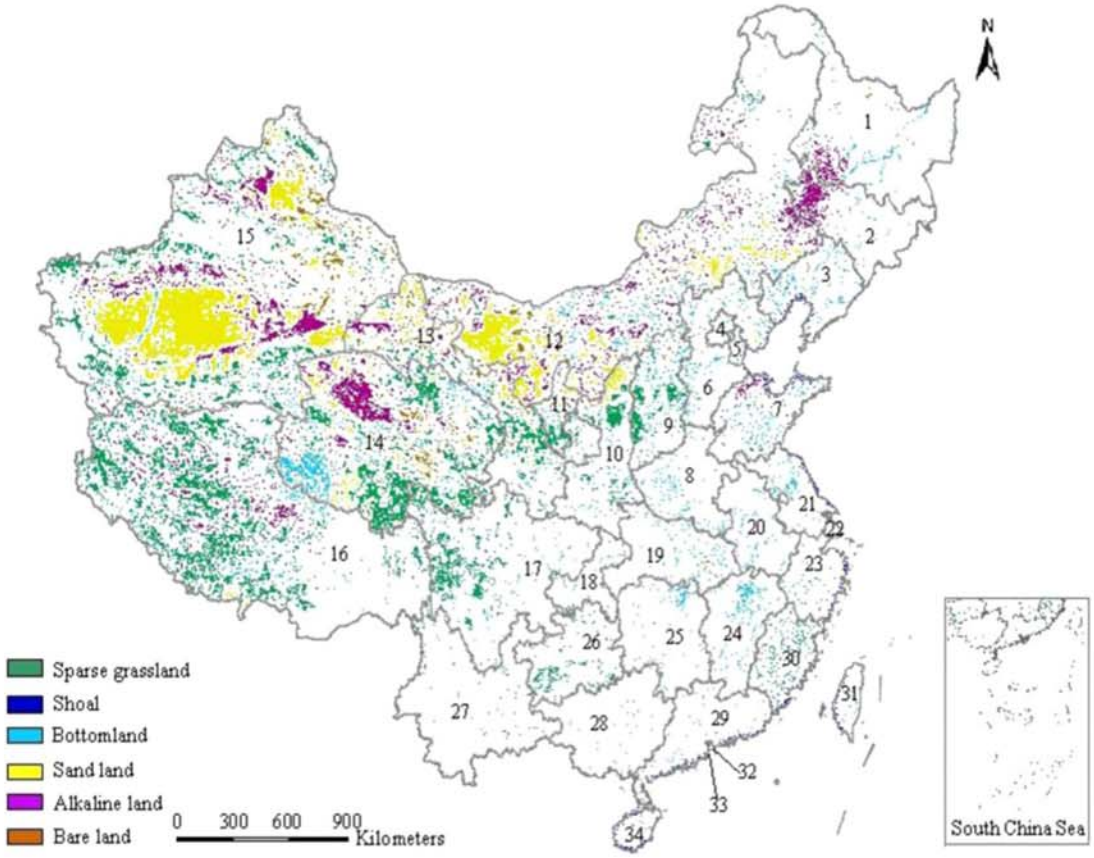

Fig. 1. Spatial distribution of available marginal land areas for miscanthus production across China. Numbers 1–34 represent the administrative division of China: 1 – Heilongjiang, 2 – Jilin, 3 – Liaoning, 4 – Beijing, 5 – Tianjin, 6 – Hebei, 7 – Shandong, 8 – Henan, 9 – Shanxi, 10 – Shaanxi, 11 – Ningxia, 12 – Inner Mongolia, 13 – Gansu, 14 – Qinghai, 15 – Xinjiang, 16 – Tibet, 17 – Sichuan, 18 – Chongqing, 19 – Hubei, 20 – Anhui, 21 – Jiangsu, 22 – Shanghai, 23 – Zhejiang, 24 – Jiangxi, 25 – Hunan, 26 – Guizhou, 27 – Yunnan, 28 – Guangxi, 29 – Guangdong, 30 – Fujian, 31 – Taiwan, 32 – Hongkong, 33 – Macau, 34 – Hainan.

yield potentials of the most adapted species for each climatic condition were simulated using a radiation yield model and subsequently adjusted down to actual harvestable yields (defined in Table 1) by measured field yields. The final output of this step was the modelled harvestable yield zonation (Fig. 2), i.e. the spatial distribution of the miscanthus production area (defined in Table 1) with varying actual harvestable yield levels across China.

Step 3: Identification and assessment of suitable marginal land for miscanthus (defined in Table 1). Spatial distribution (Fig. 3) and area of suitable marginal land were identified by overlaying the distribution maps of available marginal land (Fig. 1) and modelled harvestable yield zonation (Fig. 2).

Step 4: An evaluation of bioenergy (represented by bio-electricity in this study) production and GHG saving potential of miscanthus on suitable marginal land was performed based

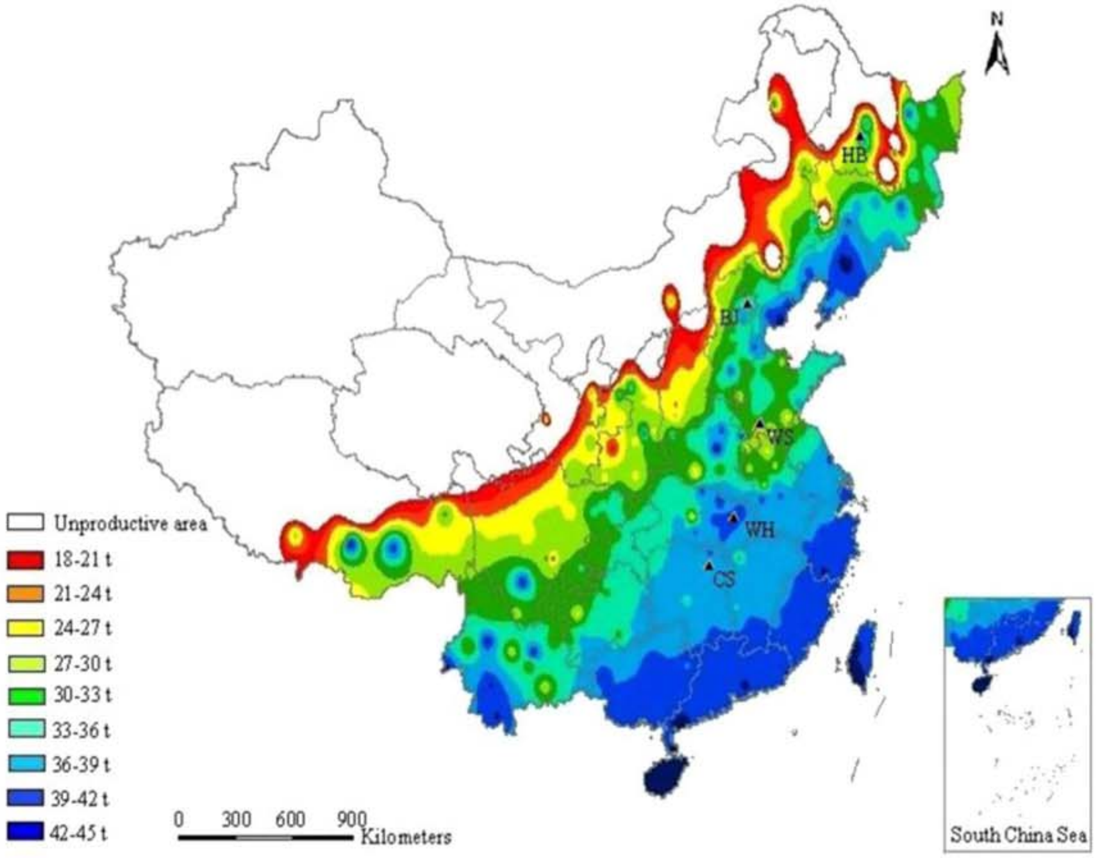

Fig. 2. Zonation of miscanthus yield across China. Here the yield potentials (t ha^{-1} yr^{-1}) were the harvestable yields that simulated using a radiation yield model and subsequently adjusted to actual harvestable yields in arable condition according to the measured field yields (yields of sites represented by black triangles on the map). CS=Changsha in Hunan Province; WH=Wuhan in Hubei Province; WS=Weishan in Shandong Province; BJ=Beijing; HB=Harbin in Heilongjiang Province.

on the achievable yield potential (defined in Table 1) in marginal conditions, the conversion ratio of biomass to bio-electricity and the carbon sequestration potential of the biomass conversion pathway of combustion.

2.1. Identification of available marginal land for miscanthus production

According to the definition by the Chinese Ministry of Agriculture, suitable land for bioenergy plants (as defined in Table 1) includes winter-fallowed paddy land and marginal land [3]. Because winter-fallowed paddy land is only available seasonally, perennial crops such as miscanthus cannot be cultivated here. Hence, cultivated land resources for miscanthus are restricted to marginal land only. According to the land-use classification system of the Chinese Academy of Science [39,40], marginal land resources include: shrub land (system code: sc.22), sparse forest land (sc.23), sparse grassland (sc.33), shoal (sc.44), bottomland (sc.45), sand land (sc.61), Gobi desert (sc.62), alkaline land (sc.63), wetland (sc.64), bare land (sc.65) and bare rock land (sc.66). The definition of each above-mentioned marginal land type is shown in Table 2 and this land-use classification system was adopted in this study. Taking into consideration the limitations of environmental and geographic conditions (shown in Table 2) of some marginal land types and the strategic course of development of the Chinese bioenergy industry, six marginal land types available for cultivating miscanthus were finally selected from the above 11 resource types. These are: (1) sparse grassland (sc.33), (2) shoal (sc.44), (3) bottomland (sc.45), (4) sand land (sc.61), (5) alkaline land (sc.63) and (6) bare land (sc.65).

Datasets of the distribution of the six chosen land types were taken as 1 km × 1 km grid datasets for 2000 from Data Sharing Infrastructure of Earth System Science (http://www.geodata.cn/). These were generated by rasterizing the original satellite images at a scale of 1:1,000,000 from Landsat™ and CBERS-1 (China–Brazil Earth Resource Satellite 1) and then interpreted by the Data Centre of Resource and Environmental Science (RESDC), Chinese Academy of Sciences [46,47]. To analyse the datasets, a uniform coordinate system is required. Hence, before further progressing in this study, all the raster data were firstly converted into a uniform format using Albers Conic Equal Area projection system with original longitude 105°E, double standard parallel of 27°N and 45°N, Beijing 1954 geodetic datum and Krassovsky ellipsoid. Then the total area of each land type was calculated using the tool Summary Statistics (Analysis) in ArcGIS 10.1 (ESRI, Redlands, California).

2.2. Identification of suitable marginal land for miscanthus production and yield potentials

The distribution of suitable marginal land was determined from the overlap of available marginal land and the miscanthus productive area using the overlay analysis in ArcGIS 10.1. The miscanthus production area is defined as the region with natural conditions that can support continuous biomass production after establishment (Table 1). Of all factors, the minimum monthly-averaged lowest air temperature (Temp) and annual accumulated precipitation (Prec) are the most critical for miscanthus growth

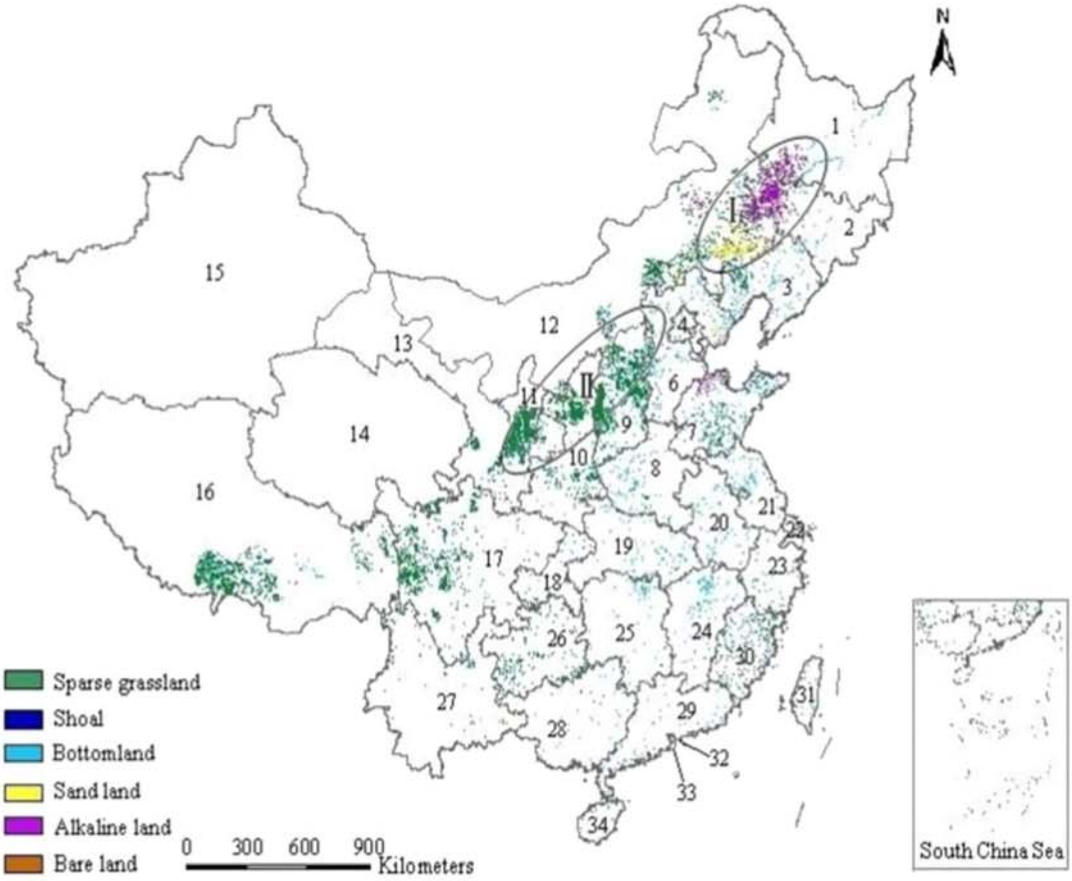

Fig. 3. Spatial distribution of suitable marginal land areas for miscanthus across China. Regions I and II marked on the map are the main regions where suitable areas for miscanthus production on marginal land are concentrated. Numbers 1–34 represent the administrative division of China: 1 – Heilongjiang, 2 – Jilin, 3 – Liaoning, 4 – Beijing, 5 – Tianjin, 6 – Hebei, 7 – Shandong, 8 – Henan, 9 – Shanxi, 10 – Shaanxi, 11 – Ningxia, 12 – Inner Mongolia, 13 – Gansu, 14 – Qinghai, 15 – Xinjiang, 16 – Tibet, 17 – Sichuan, 18 – Chongqing, 19 – Hubei, 20 – Anhui, 21 – Jiangsu, 22 – Shanghai, 23 – Zhejiang, 24 – Jiangxi, 25 – Hunan, 26 – Guizhou, 27 – Yunnan, 28 – Guangxi, 29 – Guangdong, 30 – Fujian, 31 – Taiwan, 32 – Hongkong, 33 – Macau, 34 – Hainan.

[48,49]. Combining literature [50–56] and our own field investigation [57] data, the miscanthus production area is characterized by $\text{Temp} \geq -23\,^{\circ}\text{C}$ and $\text{Prec} \geq 400$ mm.

Previous field trials conducted in Europe indicate that different species/genotypes have different adaptation abilities and biomass production potentials in different climate conditions [53]. Therefore the most suitable species/genotypes should be selected for each specific region to guarantee the maximum productivity. Taking this into consideration, the entire production area in our study was finally divided into three production zones for four potential species (Table 3). According to results of previous evaluations of miscanthus germplasm across China by Yi [54] and Xiao [57], only *Miscanthus sinensis* Andersson, *Miscanthus floridulus* (Labillardière) Warburg ex K. Schumann & Lauterbach, *Miscanthus sacchariflorus* (Maximowicz) Hackel and *Miscanthus lutarioriparius* L. Liu ex Renvoize & S. L. Chen are considered suitable for biomass production in China. In general, *M. lutarioriparius* has higher biomass yield and higher environmental stress sensitivity than *M. sinensis* and *M. sacchariflorus* (in descending order). More specifically, *M. lutarioriparius* requires warm and wet conditions and has the highest yield potential at over 38 t $\text{ha}^{-1}\,\text{yr}^{-1}$ [54,58]. In cold and dry conditions, both *M. sinensis* and *M. sacchariflorus* perform best [52,53,56]. *M. floridulus* is characterized by a moderate yield potential, which is lower than that of *M. lutarioriparius* but higher than that of *M. sinensis* and *M. sacchariflorus* [54]. Based on the biological characteristics of these species, the three production zones were then sub-divided. Firstly, within the entire production area, the most productive zone (Zone 1 in Table 3) was allocated to *M. lutarioriparius* based on the environmental requirements ($\text{Temp} > 1\,^{\circ}\text{C}$ & $\text{Prec} > 1000$ mm) for continuous growth. Then within the remaining production areas after the exclusion of Zone 1, areas with $-3\,^{\circ}\text{C} < \text{Temp} < 1\,^{\circ}\text{C}$ and $760\text{ mm} < \text{Prec} < 1000$ mm (Zone 2 in Table 3) were allocated to the moderately productive *M. floridulus*. The remaining production area with Temp ranging from $-23\,^{\circ}\text{C}$ to $-3\,^{\circ}\text{C}$ and Prec ranging from 400 to 760 mm was defined as the zone suitable for *M. sinensis* and *M. sacchariflorus* (Zone 3 in Table 3). The Temp and Prec values (shown in Table 3) were allocated to the different zones according to the literature data [50–56] and the natural distribution zones of each genotype found in our own field investigations [57]. For *M. sinensis*, a minimum soil temperature at a depth of 5 cm (Tso), as opposed to the required air temperature (Temp), was reported in the literature [53,55]. To determine the corresponding Temp value for *M. sinensis*, Eq. (1) was used.

$$\text{Tso} = 0.0088 \times \text{Temp}^2 + 0.6658 \times \text{Temp} + 2.214 \quad \left(R^2 = 0.9252, n = 1314\right) \tag{1}$$

Compared to other energy crops, such as sweet sorghum, the biomass yield of miscanthus shows wider variation in different conditions [53,59,60]. If only one average yield figure were used to calculate the total biomass production potential, as is the case in other studies [37,61], it would give a larger error. Therefore, to

Table 2
Chinese marginal land classification system [39,40] and selection of available marginal land types for miscanthus production.

Land type	System code (sc.)	Description	Is it available for miscanthus production?	Reasons for exclusion from available marginal land types for miscanthus production
Shrub land	sc.22	Land covered by woody vegetation less than 2 m high with canopy cover more than 40%	No	This land was given strategic priority for the development of Chinese biodiesel industry [41,42] because non-food biodiesel plants, i.e. shrubs and trees [43–45], can be grown here.
Sparse forest	sc.23	Land dominated by trees with canopy cover between 10% and 30%	No	Same as shrub land
Sparse grassland	sc.33	Land covered by herbaceous vegetation with canopy cover between 5% and 20%	Yes	–
Shoal	sc.44	Land between the high tide level and low tide level	Yes	–
Bottomland	sc.45	Land between normal water level and flood level	Yes	–
Sand land	sc.61	Land covered by sandy soil with vegetative cover of less than 5%	Yes	–
Gobi desert	sc.62	Land covered by gravel with vegetative cover of less than 5%	No	Water shortage makes this land unsuitable for miscanthus production.
Alkaline land	sc.63	Land covered by sparse vegetation with saline accumulation on the top soil layer	Yes	–
Wetland	sc.64	Permanently or seasonally water-saturated land covered by herbaceous or woody vegetation	No	The high ecological value of wetland is more important than bioenergy production.
Bare land	sc.65	Bare exposed soil with less than 5% vegetative cover	Yes	–
Bare rock	sc.66	Bare exposed rocks with less than 5% vegetative cover	No	Lack of soil makes this land unsuitable for miscanthus production.

achieve a more precise estimation of the biomass production potential, in our study specific yield data were taken for each given site. The miscanthus yield potential was calculated using the Monteith radiation model [62] adapted by Liu et al. [63] as follows Eq. (2):

$$Y_n = \frac{S_n P_n}{S_{ck} P_{ck}} Y_{ck} \tag{2}$$

where Y_{ck} and Y_n are the above-ground biomass yield in the control site (ck) and any given site (n) near the control site; S and P are the annually accumulated photosynthetic active radiation and precipitation for each site. The control site is the site where miscanthus yields from arable land are measured. $S = \varepsilon H_e$, where ε is the hourly active radiation converted to biomass by plants, which is considered to be constant in our model; and H_e is the number of efficient sunshine hours during the growing season. The optimized model includes the adjustment based on measured yields in arable conditions (i.e. the yield of the control site). Hence, the final output Y_n is the actual harvestable yield potential in arable conditions.

The growing season is defined as the period between growth start (when mean daily temperatures ≥ 10 °C) and first autumn frost or flowering (considered as the end of vegetative growth). In our field observation, the longest growing season was approximately 210 days. It is thus reasonable to conclude that H_e equals the annually accumulated active sunshine hours H_t at the sites with less than 210 active days (days with mean daily temperature ≥ 10 °C); while for the sites with more than 210 active days, H_e is estimated using the logistic relationship between H_e and H_t described by Eq. (3).

$$H_e = \frac{H_{max}}{1 + ae^{-b(H_t - H_{min})}} \tag{3}$$

where H_{max} is the upper limit of H_e, which is not considered a variable in this study; a and b are the equation variables.

Finally, the miscanthus yield potential of any given site (Y_n) can be simulated based on the climate characteristics (including $H_{t\text{-}ck}$ and P_{ck}) and biomass yield of the control site (Y_{ck}) within each productive zone using Eq. (4).

$$Y_n = \frac{\left(1 + ae^{-b(H_{t-ck} - H_{min})}\right) P_n}{\left(1 + ae^{-b(H_{t-n} - H_{min})}\right) P_{ck}} Y_{ck} \tag{4}$$

where H_{min} is the lower limit of accumulated active sunshine hours and set as the accumulated value of the first 120 growing days within each control site. The value of each equation variable is shown in Table 3.

Based on reports on miscanthus yield across China, Changsha [54] and Wuhan [64] were set as control sites for Zone 1 and Zone 2, respectively. For Zone 3, Harbin [65] was taken as control site for the sites in the provinces Heilongjiang, Jilin, Liaoning, Inner Mongolia and Tibet; Beijing [66] for sites in Hebei, Shaanxi, Ningxia and Gansu; and Huishan [67] for sites in Shandong, Henan and Shaanxi. Based on climate data (1981–2010) obtained from the China Meteorological Data Sharing Service System (http://cdc.cma.gov.cn/), the yield potential of a total of 722 sites (the meteorological observation stations) was estimated using Eq. (4). Assuming that water availability would not limit biomass production where Prec was more than 1000 mm, P_n of the Zone 1 model was equal to its P_{ck}, i.e. $P_n/P_{ck} = 1$. For sites in Zone 4, considered unproductive sites, the yield potential was set to zero. Finally, a map (Fig. 2) of miscanthus production areas across China with corresponding actual harvestable yield potentials was generated based on the 722 modelled harvestable yield potentials using the inverse distance weighted (IDW) analysis of ArcGIS 10.1.

Table 3
Variables of equations used to predict the miscanthus yield potential in different climatic conditions.

Zone	Required climatic conditions	Optimal genotypes	Variables for radiation yield model Eq. (4)						Scope of the equation application
			$H_{t\text{-}ck}$	a	b	H_{min}	P_{ck}	Y_{ck}	
1	Temp > 1 °C Prec > 1000 mm	*Miscanthus lutarioriparius*	1300	0.785	0.00210	560	N/A[b]	38.0	All sites within the required climatic condition area across China
2	−3 °C < Temp < 1 °C 760 mm < Prec < 1000 mm	*Miscanthus floridulus*	1500	0.803	0.00027	670	1315	41.5	All sites within the required climatic condition area across China
3	−23 °C < Temp < −3 °C 400 mm < Prec < 760 mm	*Miscanthus sacchariflorus* & *Miscanthus sinensis*	1300	0.182	0.00129	1100	550	37.5	Sites within the required climatic condition area in the provinces of Heilongjiang, Jilin, Liaoning, Inner Mongolia and Tibet
			1600	0.684	0.00155	950	570	39.0	Sites within the required climatic condition area in the provinces of Beijing, Hebei, Shanxi, Ningxia and Gansu
			1600	0.684	0.00155	950	800	43.8	Sites within the required climatic condition area in the provinces of Shandong, Henan and Shaanxi
4	Temp < −23 °C Prec < 400 mm	Unproductive area not used for miscanthus production	N/A	N/A	N/A	N/A	N/A	N/A	All sites within the required climatic condition area across China

Note: Temp = the minimum monthly-averaged lowest air temperature; Prec = averaged annual accumulated precipitation; N/A = no data available.

2.3. Assessment of miscanthus production potential on suitable marginal land

As previously mentioned, the miscanthus yield calculated by the radiation model is the actual harvestable yield potential in arable conditions. On account of the increased environmental stress compared to arable land, the achievable yield (defined in Table 1) on marginal land would be expected to be lower than that modelled here. For example, due to the extreme low soil nutrient content (10.5 mg/kg N, 3.95 mg/kg P) of sand land, which was incapable of supporting regular plant growth, the final miscanthus aboveground biomass yield decreased from the arable level of 27.5 t ha^{-1} yr^{-1} to 2.3 t ha^{-1} yr^{-1} [68]. This indicates that when cultivating miscanthus on sand land only 8.4% of the yield achievable on arable land can be expected. Hence, in this study the achievable yield on each marginal land was estimated and used to evaluate the potential biomass production, bioenergy generation and GHG saving. The achievable yield on each marginal land type was calculated by multiplying the actual harvestable yield by the percentage of achievable yield in marginal conditions. There was no data available in the literature for yields on bare land. Generally, bare land produces similar but slightly higher biomass than sand land [69]. For this reason, our study adopted a figure of 9% of the harvestable yield as the achievable yield on sand land and bare land together. On shoal and alkaline land, where the miscanthus yield decreased from arable level of 36 t ha^{-1} yr^{-1} [70] to 21.9 t ha^{-1} yr^{-1} [71], 60% of the harvestable yield was achieved. For *M. lutarioriparius* growing wildly on bottomland, a similar yield level to the harvestable yield was reported [58], suggesting a percentage of 100% for bottomland. The figure for sparse grassland was calculated as 69% based on the *M. sinensis* harvestable yield on arable land of 5.7 t ha^{-1} yr^{-1} [72] and achievable yield on the natural grasslands of north-east Japan of 3.9 t ha^{-1} yr^{-1} [73].

These calculations assume electricity generation as the best technology for the application of miscanthus biomass. The bioenergy potential of miscanthus on marginal land was expressed as the amount of bio-electricity generation and standard coal replacement. *Miscanthus* can only be stored and combusted when dry. Therefore it is usually harvested after the winter. Pre-harvest losses during winter are 15–25% of the aboveground biomass [74,75]. Hence, it was assumed here that only 80% of the aboveground biomass is available for bio-electricity generation. About 1,700 kW h electric power is generated per ton miscanthus biomass with an energy content of 17.5 GJ t^{-1} [76] and an energy conversion efficiency of 0.35 from thermal to electricity [77]. The standard coal equivalent was calculated using a standard coal energy content of 29.3 GJ t^{-1}.

Although the captured carbon in aboveground biomass will be released to the atmosphere in the combustion process, 0.16–1.12 t ha^{-1} [78–81] carbon are still sequestrated in the soil by the below-ground rhizomes and roots. Even taking into account the carbon emissions from the fossil energy used for biomass production (e.g. establishment, management, harvest and transportation), miscanthus production still has a carbon sequestration potential [82,83]. This potential was found to be positively correlated with the aboveground biomass yield. Based on the relationship between miscanthus yield and GHG savings described in [83], and the estimated achievable yield on marginal land, the average per-hectare GHG savings by miscanthus on bare land, sand land, alkaline land, sparse grassland, shoal and bottomland were calculated to be 1.8, 2.4, 21.6, 28.0, 35.5 and 52.3 t CO_2 eq. ha^{-1} yr^{-1}, respectively. The average GHG saving potentials were used to calculate the total GHG savings by miscanthus on suitable marginal land.

3. Results

3.1. Available marginal land for miscanthus production in China

The final characteristics of the available marginal land areas (as defined in Table 1) which could be used for miscanthus cultivation without conflicting with government policy or environment protection concerns, and their spatial distribution across China, are shown in Table 4 and Fig. 1.

The results indicate that the available marginal land areas for miscanthus production are mainly located in Northwest China with a total area of 17,163.54 × 10^4 ha (Table 4). These are predominantly composed of sparse grassland and sand land, accounting for 53.74% (9224.49 × 10^4 ha) and 33.31% (5716.81 × 10^4 ha) of the total available marginal land area, respectively. Even though the shoal and bare land only account for 0.35% (59.71 × 10^4 ha) and 1.70% (291.87 × 10^4 ha) of the total available area, taken together they still represent a large potential area of 351.58 × 10^4 ha that could be exploited for miscanthus production.

As shown in Fig. 1, the marginal land resources of sparse grassland, sand land, alkaline land and bare land are mainly located in Northwest China, including the provinces of Xinjiang, Tibet, Inner Mongolia, Qinghai and Ningxia. A second predominant area of alkaline land can be seen in the border zone of Inner Mongolia, Jilin and Heilongjiang in Northeast China. There are a number of scattered but concentrated areas of bottomland in the regions of South and Southeast China and areas of shoal interspersed along with the coastline.

3.2. Yield potential and suitability of marginal land for miscanthus production in China

From Fig. 2 it can be seen that the boundary of the miscanthus production area shows a distinct northeast-southwest trend, almost consistent with the 400 mm isohyet of China. Virtually the entire area of Northwest China is unsuitable for miscanthus cultivation due to an annual accumulated precipitation of less than 400 mm, in some regions even lower than 10 mm. This indicates that drought is a more critical constraint than cold temperatures for miscanthus production in China. Within the production area, the actual harvestable yield generally increases from the northwest to the south-east with the highest potential lying on the islands of Hainan and Taiwan (Fig. 2). In addition, there is high yield potential in some parts of Northeast China, mainly in east Liaoning where solar radiation and precipitation are both relative high. The highest and lowest harvestable yield potentials of the entire production area are, respectively, 44.2 and 18.1 $t\ ha^{-1}\ yr^{-1}$ with an overall average potential of 26.0 $t\ ha^{-1}\ yr^{-1}$. Taking the constraints of marginal conditions on biomass production into account, the average levels of achievable yield potential for each marginal land type are as follows: 22.3 $t\ ha^{-1}\ yr^{-1}$ for shoal, 32.4 $t\ ha^{-1}\ yr^{-1}$ for bottomland, 2.4 $t\ ha^{-1}\ yr^{-1}$ for sand land, 17.8 $t\ ha^{-1}\ yr^{-1}$ for sparse grassland,14.0 $t\ ha^{-1}\ yr^{-1}$ for alkaline line and 2.1 $t\ ha^{-1}\ yr^{-1}$ for bare land (Table 5).

Table 4

Available marginal land and suitable marginal land areas for miscanthus production across China.

Marginal land types	Available marginal land areas (10^4 ha)	Percentage of total available area (%)	Suitable marginal land areas (10^4 ha)	Percentage of total available area (%)
Sparse grassland	9,224.49	53.74	319.23	1.86
Shoal	59.71	0.35	6.55	0.04
Bottomland	505.17	2.94	114.56	0.67
Sand land	5,716.81	33.31	49.59	0.29
Alkaline land	1,365.49	7.96	275.56	1.61
Bare land	291.87	1.70	3.88	0.02
Total	17,163.54	100.00	769.37	4.49

From the available marginal land identified, suitable marginal land for miscanthus cultivation was differentiated by considering its environmental requirements. The total suitable marginal land area for miscanthus is 769.37×10^4 ha, accounting for only 4.49% of the total available area (Table 4). Sparse grassland remains domination with a suitable area of 319.23×10^4 ha (1.86%). Alkaline land surpasses sand land, ranking second among all the land types with a suitable area of 275.56×10^4 ha (1.61%). Bare land is the land type with the smallest suitable area (3.88×10^4 ha), accounting for only 0.02% of the total available area. Based on the spatial distribution of these areas shown in Fig. 3, it is concluded that there are two main regions with a concentration of suitable marginal land for miscanthus cultivation. One is the central part of Northeast China (Region I in Fig. 3), including east Inner Mongolia, northwest Jilin and southwest Heilongjiang. The marginal land in this region is mainly composed of alkaline land and sand land. The other is the Loess Plateau area (Region II in Fig. 3), including southeast Gansu, south Ningxia, north Shaanxi and central Shanxi. The marginal land in this region is mainly composed of sparse grassland.

3.3. Productivity and GHG savings potential of the suitable marginal land

Results shown in Table 5 indicate that approximately $13{,}521.7 \times 10^4\ t\ yr^{-1}$ aboveground biomass could be produced from miscanthus on the entire suitable marginal land area. The land types with the highest biomass production potentials are sparse grassland, alkaline land and bottomland with yield potentials of 5682.9×10^4, 3852.3×10^4 and $3711.7 \times 10^4\ t\ yr^{-1}$, respectively. Assuming that 80% of the produced biomass (20% losses in winter) is combusted for bio-electricity generation, this corresponds to a total bio-electricity generation of 183.9 $TW\ h\ yr^{-1}$. The bio-electricity potential varies between biomasses from different land types. Sparse grassland, alkaline land and bottomland are still the land types with the highest electricity generation potential, together accounting for 98% of the total potential on the entire suitable marginal land area. Assuming a standard coal energy content of 29.3 $GJ\ t^{-1}$, a total of 6460.9×10^4 t standard coal (Table 5) could be replaced by this biomass for electricity generation.

Table 5

Potentials of biomass production, bio-electricity generation, standard-coal equivalent and greenhouse gas (GHG) savings from miscanthus grown on the entire suitable marginal land area across China.

Marginal land type	Average achievable yield ($t\ ha^{-1}\ yr^{-1}$)[a]	Biomass production potential ($10^4\ t\ yr^{-1}$)[b]	Bio-electricity generation potential ($TW\ h\ yr^{-1}$)[c]	Standard-coal equivalent potential ($10^4\ t\ yr^{-1}$)[d]	GHG-saving potential ($10^4\ t\ CO_2$ eq. yr^{-1})[e]
Sparse grassland	17.8	5682.9	77.3	2715.4	8934.4
Shoal	22.3	146.2	2.0	69.9	232.6
Bottomland	32.4	3711.7	50.5	1773.5	5993.5
Sand land	2.4	120.5	1.6	57.6	118.1
Alkaline land	14.0	3852.3	52.4	1840.7	5956.9
Bare land	2.1	8.0	0.1	3.8	6.9
Total	–	13521.7	183.9	6460.9	21,242.4

[a] The average achievable yield for each marginal land type was calculated by multiplying the average harvestable yield by the percentage of achievable yield in each marginal condition.

[b] Biomass production potential was calculated by multiplying the average achievable yield on each marginal land type by the corresponding suitable marginal land area shown in Table 4.

[c] Approximately 1700 kW h electric power is generated per ton miscanthus biomass with an energy content of 17.5 $GJ\ t^{-1}$ [76] and an energy conversion efficiency of 0.35 from thermal to electricity [77].

[d] Standard-coal equivalent was calculated based on an energy content of standard coal of 29.3 $GJ\ t^{-1}$.

[e] The per-hectare GHG savings from miscanthus cultivation on bare land, sand land, alkaline land, sparse grassland, shoal and bottomland were estimated at 1.8, 2.4, 21.6, 28.0, 35.5 and 52.3 $t\ CO_2$ eq. $ha^{-1}\ yr^{-1}$, respectively, according to a model of Meyer and Lewandowski [83].

In terms of GHG savings, the results indicate that the mitigation potential of miscanthus cultivation on the entire suitable marginal land area would be approximately $21{,}242.4 \times 10^4$ t CO_2 eq. yr^{-1} (Table 5). Sparse grassland is also the marginal land type with the highest GHG savings potential at 8934.4×10^4 t CO_2 eq. yr^{-1}. Due to the higher per-hectare GHG savings from miscanthus on bottomland than on other marginal land types (see Section 2.3), the total GHG savings for bottomland (5993.5×10^4 t CO_2 eq. yr^{-1}) are the second highest, closely followed by alkaline land (5956.9×10^4 t CO_2 eq. yr^{-1}). This is despite the fact that the suitable bottomland area is less than half the suitable alkaline land area (Table 4). If these three marginal land types (sparse grassland, bottomland and alkaline land) alone were used for miscanthus cultivation, 98.3% ($20{,}884.8 \times 10^4$ t CO_2 eq. yr^{-1}) of the total GHG mitigation potential could still be achieved.

4. Discussion

From our analyses, it is concluded that China has abundant suitable marginal land for miscanthus cultivation. Development of a miscanthus-based bioenergy industry on this land would provide opportunities for the Chinese government to reduce its energy security risk, dependence on fossil energy carriers and imported oil, carbon intensity (i.e. CO_2 emission per unit of GDP) and to improve its rural economy. Here in this study it was estimated that a total of $13{,}521.7 \times 10^4$ t DM yr^{-1} miscanthus biomass could be produced on 769.37×10^4 ha suitable marginal land. This biomass could replace 6460.9×10^4 t standard coal annually, accounting for 1.7% of the 2013 Chinese gross energy consumption [84]. If all the biomass was combusted for electricity generation, a total of 183.9 TW h bio-electricity would be generated, equivalent to 3.5% of the 2013 Chinese gross electricity consumption [84]. Alternatively, this biomass could be used to produce 3428.9×10^4 t yr^{-1} cellulosic ethanol (using the conversion rate given by Zhao et al. [85]), offsetting 6.9% of the 2013 Chinese gasoline consumption [84]. The GHG mitigation potential of electricity production from miscanthus biomass grown on marginal land in China is $21{,}242.4 \times 10^4$ t CO_2 equivalents annually. This mitigation potential could contribute to a 14.7% reduction in carbon intensity by 2020 relative to 2005 [86]. This is approximately one third of the CO_2 reduction goal of 40 – 45% set by the Chinese government. At an average carbon trading price of 32 CNY t^{-1} CO_2 [87], the total revenue from carbon mitigation trading would be 6.37 billion CNY. Sale of the biomass could also bring farmers additional income totalling 24.3 billion CNY based on an average biomass price of 180 CNY per dry ton [88]. *Miscanthus* production on marginal land would also create additional employment opportunities in rural areas.

In the following sections the various implications of and requirements for the implementation of the theoretical miscanthus biomass production potential on marginal land are discussed.

4.1. Potential advantages and disadvantages of establishing miscanthus on marginal land

From an ecological point of view, the production and energetic use of miscanthus biomass can strongly contribute to climate change mitigation, as has been discussed above. There are other advantages, but also potential disadvantages.

If marginal land is to be used for crop production, the perennial character and high nutrient-use efficiency of miscanthus make it the most environmentally beneficial option. The noteworthy ecological benefits of miscanthus cultivation are the reduction of soil erosion and the increase of soil organic carbon content and biodiversity [82,89,90]. Due to its deep and extensive below-ground system of roots and rhizomes and also its perennial nature, miscanthus has, once established, the potential to reduce the soil erosion risk [82,91]. As soil erosion rates are negatively correlated to vegetation cover [82,92], the establishment of miscanthus on marginal land with low vegetation cover, such as sparse grassland, sand land or bare land, is expected to considerably reduce erosion. For this reason, miscanthus cultivation is recommended for the Loess Plateau area of China, which is characterized by a high risk of soil erosion [93,94]. However, soil erosion risk reduction can only be achieved after the establishment phase when rhizomes and roots are well developed. Soil tillage before establishment and the low planting density of about two plants m^{-2} create a very high initial soil erosion risk in miscanthus plantations. To minimize this risk the applicability of conservation tillage techniques for establishment or the undersowing of cover crops should be assessed. The soil organic carbon content of miscanthus plantations can increase by 0.16–1.63 t ha^{-1} yr^{-1} [78–81,95], mainly due to leaf litter and the development of the below-ground root and rhizome system [96]. These organic matter inputs support the maintenance of soil structure and reduce the soil erosion risk [90,97,98]. These mechanisms give miscanthus plantations the potential to upgrade the soil quality of marginal land [99,100]. Compared to annual crops, misanthus plantations are characterized by a lower soil disturbance frequency and fewer herbicide and pesticide requirements [101]. In addition, the vertical and horizontal habitat structure created by the standing miscanthus plants can provide more ecological niches than annual crops for birds, insects, reptiles and mammals, resulting in an increased diversity of wildlife populations [102–104]. Therefore, miscanthus plantations are preferable to annual crop production systems for marginal areas in China. In addition to the above ecological benefits, miscanthus establishment on marginal land can also provide economic benefits, i.e. income. For example, in Europe there is a continuously increasing area if grassland no longer used for animal husbandry [105]. Although they are no longer in production, these grasslands still require conservation measures, which may incur high management costs, to maintain their positive functions for soil carbon storage [106]. The introduction of miscanthus cultivation on these unused grasslands would combine grassland conservation with energy crop production. These positive ecological and economic features make miscanthus a good choice of energy crop to cultivate on marginal land.

However, the production of miscanthus on marginal land that is currently not cultivated will also have an impact on vegetation of that land. This may be of concern particularly for land originally rich in biodiversity. There are concerns that (1) miscanthus cultivation may reduce the biodiversity because the disturbance of establishment may destroy the original species [107]; (2) shading from the dense miscanthus canopy may inhibit the growth of native plants [108]; and (3) the allelopathic inhibitory effect on other species seen in *M. transmorrisonensis* and *M. floridulus* [109,110] may also be characteristic of other miscanthus species. The ecological impact assessment should also include the potential risk of newly introduced miscanthus genotypes to become invasive. There are generally two mechanisms by which miscanthus can become invasive: (a) spreading by seed, (b) spreading by the outgrowth of above- or below-ground shoots (vegetative propagation). In European miscanthus breeding programmes the former was observed in *M. sinensis* genotypes and the latter in *M. sacchariflorus*. By contrast, neither form of proliferation has been observed in more than 20 years' field research of *M.* × *giganteus* in either Europe or Australia [111]. *M.* × *giganteus* is a triploid natural hybrid between *M. sacchariflorus* and *M. sinensis* [112], and a model genotype for miscanthus breeding activities in Europe. As short-day plants [113,114], miscanthus species do not usually produce seeds or even flower in long-day conditions (e.g. high latitude areas of the Northern Hemisphere). To reduce the invasion risk by seed spreading, the fertile species *M. sinensis* and *M. sacchariflorus* are proposed for North and Northeast China, which are generally characterized by long-day conditions. *M. lutarioriparius* and *M. floridulus* are species which occur naturally in the areas for which they are proposed in this study.

4.2. Economical and practical barriers to miscanthus production on marginal land

In this study the theoretical potentials of miscanthus production on marginal land were assessed. The implementation of these potentials however requires the consideration of the following aspects:

A. Competition for marginal land from other uses
One of the main limitations to the development of a Chinese miscanthus industry is the challenge of land shortage because more and more agricultural land is required to meet the increasing food and feed demand. This land shortage can be remedied by exploiting marginal land. The cultivation of miscanthus on suitable marginal land is evaluated to have immense potential for improving energy security and reducing GHG emissions. However, currently there are still a number of constraints to fully exploiting this potential. The main limitation is the uncertainty in the availability of the suitable marginal land due to competition from other usages such as land reclamation for the cultivation of food crops [47] or other energy crops [37,38,115].

B. Accessibility of marginal land and availability of transport infrastructure
Marginal land areas are often poorly accessible due to the lack of roads or other infrastructure for transporting machinery needed for crop management or transporting and storing the produced biomass. Consequently there are currently several areas in China that would be difficult to access for miscanthus production. Therefore the implementation of bioenergy production on marginal land requires investment in infrastructure or the installation of biomass to bioenergy processing plants nearby.

C. Availability of infrastructure and technology for bioenergy
At present, bioenergy is still an emerging industry in China. Some small-scale plantations of sweet sorghum [116,117] and *Jatropha curcas* L. [6,41,118] have been established for pilot biofuel production programmes but no commercial miscanthus plantations have yet been established for bioenergy production. This is mainly due to the immature technology for the conversion of miscanthus biomass to bioenergy. Combined heat and power (CHP) technology for biomass combustion is still in the development stage in China [119]. Also there are no facilities yet available for producing cellulosic ethanol. To achieve the implementation of miscanthus-based bioenergy production on marginal land, widely available CHP plants for biomass combustion would need to be built. Another option is to co-fire miscanthus in existing coal plants [120]. In China, the existing coal-fired power plants currently require 1591 Mt coal annually for combustion [121]. Based on this capacity, the miscanthus biomass produced on the entire suitable marginal land area could be completely co-combusted at a mixing rate of 8.5%.

D. Profitability and acceptance of miscanthus production
As a totally new crop that has never been cultivated before, miscanthus production would create a real challenge for Chinese farmers due to the divergence from traditional agronomy practices. Additionally, in order for miscanthus to be adopted by farmers, the attainable profit needs to be at least as high as that from traditional crops. However, currently the profit from miscanthus production is lower than the net revenue from traditional crops produced in China, for example 14,760 CNY ha^{-1} for maize (*Zea mays* L.) [117]. Also consistently higher production costs have been reported for miscanthus compared to other energy crops such as sweet sorghum [23,24]. Although land-use costs of marginal land are generally lower than for good agricultural land, miscanthus production costs on a per-ton basis are still high on account of the low yields on marginal land [122,123].

E. Availability of productive genotypes and efficient management systems for miscanthus production

The lack of dedicated varieties, efficient establishment techniques and specialist equipment will limit the development of the Chinese miscanthus industry [101], as has been the case with the development of miscanthus production in Europe. Therefore, the implementation of the large production potential requires the development of miscanthus varieties adapted to the various marginal land conditions in China and of crop management systems that are efficient and economically viable.

4.3. Recommendations for miscanthus breeding

As marginal areas will be central to China's future bioenergy programme, it is essential to breed a series of varieties with good adaptation to various marginal conditions. On account of the natural climate conditions and land characteristics of the two priority development zones (Regions I & II marked in Fig. 3), future miscanthus breeding programmes should focus on the following aspects: (1) breeding of dedicated varieties with high tolerance to low winter temperatures and soil salinity for marginal land lying in Northeast China; and (2) breeding of varieties characterized by drought tolerance and ability to compete with grasses found on the marginal land areas of the Loess Plateau. Although the distribution of bottomland is more scattered, its high biomass productivity and GHG mitigation potentials also render it potentially important for Chinese miscanthus production. This would however necessitate the breeding of varieties with flooding tolerance. While the time frames of breeding programmes are generally long, the fact that China is the distribution centre of wild miscanthus germplasm resources [54] could shorten the breeding process by selecting productive wild species/genotypes or even natural hybrids to overcome the current shortage of commercial varieties. Moreover, as discussed above, the miscanthus establishment techniques currently applied in Europe are not recommended for cultivation on marginal land on account of their potential adverse effects relating to soil erosion and biodiversity reduction. Therefore, in addition to the selection of suitable genotypes, an economical and ecologically-friendly practice guideline for miscanthus cultivation on marginal land should also be developed.

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Chapter 4 Establishment and management miscanthus on marginal land-a case study on grassland in south-west Germany

The results from Chapter 3 show that grassland is one of the main marginal land types exploitable for miscanthus production in China. This is also true for the European countries because along with the intensification of livestock farming, increasing areas of European grassland are no longer used for animal husbandry and then are recommended for bioenergy use. However, the grassland currently is not applicable for growing miscanthus due to the lack of optimal genotypes and effective practices. The aim of this study was then to investigate effective practices for miscanthus establishment and management on C3 grassland, including accessing effects of genotypes, propagation method and pre-planting grassland disturbance on miscanthus establishment and effects of grassland cutting frequency on maintenance of the miscanthus-improved grassland.

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4.1 Introduction

Use of bioenergy is widely considered a promising way to reduce greenhouse gas (GHG) emissions, mitigate climate change and relieve the conflict between increasing energy demands and depleting fossil resources [1]. However, the sharp increase in biofuel production from food-based feedstock (e.g., corn, rapeseed) in the last decades has raised concerns that using food crops for bioenergy may conflict with food security [2-4]. On the other hand, further expansion of agricultural areas for high-input food crop production may lead to an increase in water pollution, forest destruction and soil degradation [5-6]. To address these concerns, non-food bioenergy production chains should be enhanced and promoted [7-9].

Non-food biomass resources include agricultural residues and by-products, forest biomass (firewood and forest waste), and biomass from dedicated energy crops [8]. Despite the vast potential of crop residues, forest biomass and by-products for bioenergy production, the amounts available will be insufficient to satisfy the feedstock demand of the future bioenergy industry [8, 10-12]. By contrast, dedicated energy crops can provide high quantities of biomass with good feedstock quality, which could be used to close the gap between the biomass supply and demand [13-14]. The lignocellulosic perennial C4 grass miscanthus (*Miscanthus spp.*) has been identified as one of the most promising dedicated energy crops with high yield potential and cellulose content, and good biomass combustion quality [15-17]. However, in Europe, only small areas of *Miscanthus × giganteus* (approximately 40,000 ha) are currently grown commercially. These are used for the generation of electricity and heat [18-19]. The main limitation to the expansion of miscanthus production is the lack of land available for its cultivation. In densely populated countries with only a small area of agricultural land per capita (e.g. China and India), there is very little or no arable land available for additional crops [9, 20-21]. In Europe, farmers prefer to use any surplus cropland for annual food crops rather than perennials such as miscanthus, because this gives them the flexibility to change the crop depending on market prices. Also, the total bioenergy production potential of miscanthus is still limited (5.8 EJ yr^{-1}) [22].

Miscanthus cultivation on non-agricultural land could potentially address this land-use dilemma.

Grassland has traditionally been used for fodder production and as pasture for livestock. However, the intensification of livestock farming and increasing use of forage crops grown on arable land in recent years has led to large areas of grassland no longer being used for animal husbandry [23]. These abandoned grasslands are among the non-agricultural land resources which could be used for miscanthus cultivation, especially grasslands on soils with low nutrient (in particular nitrogen) content which cannot provide sufficient net revenue from fodder production [24-25]. Nevertheless, these grasslands still require proper conservation management, such as regular mowing, to prevent succession leading to loss of biodiversity and reduction in grassland area [26-27]. However, grassland maintenance incurs high costs. In order to gain income from such grassland, increasing areas are being converted to arable land [23, 28]. The large-scale conversion of grassland to cropland will, in the long term, lead to negative consequences such as desertification, reduction of soil carbon sequestration and loss of biodiversity [28-29]. To preserve grassland's ecological functions, new concepts for the profitable utilization of these unused grasslands are necessary. One solution could be the production of bioenergy from grassland biomass, providing both economic (income) and ecological benefits and preventing grassland succession [30-36]. Cultivating miscanthus on grassland could be a sound, multi-functional way to increase the cultivation area of miscanthus and, at the same time, gain income from unused grasslands. *Miscanthus* is known for its high resource-use efficiency, in particular low nitrogen requirements [37-38]. In addition, the introduction of warm-season (C4) species could also potentially enhance the total dry matter yield of the cool-season (C3) grassland, through the complementary characteristics of C3 and C4 plants' growth rates [39-40]. *Miscanthus* cultivation on grassland could also lower production costs through reducing land opportunity costs [41-43].

Biomass from natural grasslands with wild-growing miscanthus has been successfully burned for electricity generation in Japan [44], showing that the use of mixed grassland/miscanthus biomass for bioenergy production is technologically feasible.

Grassland has important ecological functions and is protected in many European countries. Often it is undesirable or even legally prohibited to convert grassland into bioenergy cropland to avoid biodiversity loss and soil carbon sequestration reduction through tilling practices. For this reason, conventional practices for miscanthus establishment on arable land cannot be directly applied to its establishment on grassland. However, no-till establishment could potentially combine grassland maintenance with miscanthus production. Therefore, effective no-till methods for establishing miscanthus in highly competitive grassland plant communities need to be developed.

In scholarly articles, miscanthus is often described as a crop requiring regular weed control during the establishment stage, suggesting that it is a low competitor against weed species [38, 41]. Other studies have also shown that miscanthus establishment and overwintering survival, annual biomass yield and production costs are highly dependent on the genotype and propagation method [45-49]. In order to effectively establish miscanthus on grassland, genotypes need to be identified which can withstand competition and regular cutting. Reducing competition in grassland, e.g., through mowing or herbicide spraying, is known to be beneficial for the establishment of introduced plants [40, 50-52], whereas regular mowing of grassland vegetation may increase grassland productivity [53-54]. Therefore, in this study three field trials were conducted with the following aims: (1) to investigate the possibility of no-till establishment and cultivation of miscanthus on low- and high-productivity grassland under European conditions; (2) to assess the effects of genotype and propagation method on miscanthus establishment and growth on grassland; (3) to test how different grassland pre-treatments (methods of removing grassland vegetation to reduce initial competition) affect miscanthus establishment and growth; and (4) to assess the effects of different grassland management practices (cutting frequency) on biomass yield of grassland with introduced miscanthus. Based on the results of these trials, recommendations are made on effective practices of miscanthus cultivation on grassland and the maintenance of miscanthus-improved grassland.

4.2 Material and methods

4.2.1 Field sites characteristics

Three field trials were established on experimental grassland of the University of Hohenheim, Stuttgart, Germany. The first trial was established in May 2011 at the university's campus site in Hohenheim (UHO, 48˚42' 53.72″ N, 9˚12' 40.24″ E, 409 m a.s.l.). The other two field trials were established in May 2012, one on high-productivity grassland (48˚44' 36.07″ N, 8˚55'01.03″ E, 465 m a.s.l.) and the other on low-productivity grassland (48˚44' 39.82″ N, 8˚55'47.36″ E, 480 m a.s.l.) at the university's experimental station Ihinger Hof (IHO). All three sites are located in south-west Germany and have similar climate characteristics, but differ in soil conditions and productivity (Table 4.1). The UHO and IHO field sites are characterized by an annual average rainfall of 698 and 693 mm and a mean daily temperature of 8.8 and 8.1 °C, respectively (based on 1991-2011 data). During the experimental period (2011-2013), detailed climatic data were collected from the meteorological stations at each field site. These are presented in Fig. 4.1.

At the two IHO grassland sites, aboveground biomass of three grassland plots (2.55 m^2 each) was harvested in late May 2012, before the miscanthus was planted, and the yield of each plot was recorded. Soil core samples were then taken at five randomly selected locations at each IHO grassland site at depths (layers) of 0-30 cm, 30-60 cm and 60-90 cm. The plant-available nitrogen (NO_3-N and NH_4-N) content in each soil layer was analysed according to the methods described in the study of Übelhör [55]. In autumn 2012, the aboveground biomass yield of the same grassland plots was measured again. The spring and autumn yield combined constitute the grassland productivity. The average biomass yield per plot, reflecting the corresponding grassland productivity, is shown in Table 4.1. No soil analysis was performed at the UHO field site but an approximate plant-available nitrogen content was estimated based on a vegetation analysis. Grasslands with low nitrogen content are known to have a high legume/grass proportions [56-57]. According to vegetation analysis data, the legume/grass proportions of the IHO high-productivity, IHO low-productivity and UHO grasslands were recorded as 0.002, 0.264 and 0.048, respectively. The available nitrogen content of the soil at the UHO site is therefore expected to lie between those of the IHO low-productivity and high-productivity sites, i.e. between 18.6 and 46.2 kg ha^{-1}, but nearer to the high-productivity soil conditions (see Table 4.1). This is also true for the grassland productivity of the UHO grassland.

Table 4.1 Details of the grassland study sites including location, climate characteristics, soil conditions and grassland productivity at the University of Hohenheim (UHO) and Ihinger Hof Experimental Station (IHO)

Study site	UHO	IHO high-productivity site	IHO low-productivity site
Site location			
Geographical coordinates	48°42' 53.72″ N 9°12' 40.24″ E	48°44' 36.07″ N 8°55'01.03″ E	48°44' 39.82″ N 8°55' 47.36″ E
Altitude (m a.s.l.)	409	465	480
Climate characteristics [a]			
Mean daily temperature (°C)	8.8	8.1	8.1
Annual precipitation (mm)	698	693	693
Soil parameters			
Soil texture	Silty loam	Silty clay	Silty clay
Available N content (kg ha^{-1}) [b]			
Soil layer 0-30 cm	N/A	33.6	15.6
Soil layer 30-60 cm	N/A	6.7	2.1
Soil layer 60-90 cm	N/A	5.9	0.9
Grassland productivity (odt ha^{-1}) [c]	N/A	9.6	5.6

a – Means of 1991 to 2011 data;

b – Average of NH_4-N and NO_3-N content in each soil layer collected at five randomly selected places before miscanthus planting;

c– Combined aboveground biomass yield harvested in 2012 spring and autumn;

N/A = no data available

4.2.2 Experimental design

Field trial at the University of Hohenheim (*UHO Experiment*)

The *UHO Experiment*, conducted from 2011 to 2013, included the factorial combinations of three establishment regimes (Er = grassland pre-treated by removing existing vegetation), two propagation methods (Prop) and two miscanthus genotypes (Geno). All the factorial treatments were arranged in a split-plot design with two factorial sub-plot factors and four replications per treatment (Table A.4.1). The establishment regimes were applied at the plot level, the combinations of propagation methods and genotypes at the sub-plot level. The total experimental area of 240 m^2 (4 × 60 m) was divided into 12 plots for the three main treatments (Er in Table 4.2) and four replicates. Each 20 m^2 (4 × 5 m) plot was divided into four 5 m^2 (2 ×2.5 m) sub-plots.

The four combinations of propagation methods and genotypes (see Table 4.2) were randomly arranged as sub-plots within the plots.

In April 2011, 120 rhizomes of each selected miscanthus genotype were manually uprooted from the 14-year-old mother plants grown at the IHO experimental station and cleaned of soil. For each genotype, half the rhizomes were cut into approximately 20-cm-long cuttings. These were put into plastic bags and stored in a cooling chamber at 3.5 °C. The other half of the rhizomes were cut into pieces of approximately 50 g each and planted into pots in the greenhouse. In early May 2011, the three grassland pre-treatments (Er in Table 4.2) were applied and then the cold-stored rhizome cuttings and pre-grown miscanthus plantlets (approximately 20 cm tall) were planted manually into the pre-treated grassland. Five miscanthus cuttings/plantlets were planted into each 2.5 × 2 m sub-plot in rows, with a distance of 0.75 m between the rows and 0.5 m between the planting positions within the rows, equivalent to a planting density of 10,000 plants ha^{-1}. The plantlets and rhizomes were watered once after planting; no further irrigation was applied. The trial was not fertilized during the entire experimental period. In early June 2011, i.e. one month after planting, the establishment survival (percentage of living plantlets or percentage of plants emerged from rhizomes) was assessed for each sub-plot. In October 2013, at the end of the third growing season, the number of living plants in each sub-plot was counted again to calculate the final survival rate, defined as the percentage of living plants from the plantlets or rhizomes initially planted. No measurements were taken and no harvests were performed in the establishment year (2011). In 2012, the grass and miscanthus were harvested once in late autumn. In the third growing season (2013), two harvests were performed: one in-season green harvest in late May and one end-of-season harvest in late October. For each harvest, first all the miscanthus plants were harvested individually by hand; then a 1 × 0.5 m quadrat of grass was harvested manually in the centre of each sub-plot. Both miscanthus and grass were harvested at a height of 5 cm above ground (common cutting height for a mowing machine or a grass harvester).

Field trials at Ihinger Hof Experimental Station (*IHO Experiment*)

The *IHO Experiment* comprised two separate, simultaneously implemented trials, one on high-productivity and the other on low-productivity grassland. For both trials, a split-split-plot design with four block-replicates and the same factorial combinations were adopted (Table A.4.2). Each main plot occupied 30.6 m^2 (6.8 × 4.5 m) and was treated by one of the two establishment regimes (see Table 4.2). The secondary treatments consisted of three different cutting frequencies (later called 'cutting regimes' or Cr), which were applied to the 10.2 m^2 (6.8 × 1.5 m) sub-plots within each main plot. Within each sub-plot, three different miscanthus genotypes were planted into sub-sub-plots of 2.55 m^2 (1.7 × 1.5 m) each. Additionally, one sub-sub-plot of pure grassland (without miscanthus planted) was used as a control for biomass yield comparisons. For both trials, three *Miscanthus sacchariflorus* Bentham genotypes CSA-435, CSA-322 and CSA-334 (see Table 4.2) were chosen because *M. sacchariflorus* may be more competitive in grassland conditions (supported by the more extensive rhizome system [58]) than *Miscanthus × giganteus* Greef et Deuter and *Miscanthus sinensis* Andersson. CSA-435 (later referred to as 'tall woody' genotype) is characterized by tall plants with a small number of thick, widely spreading shoots (Table 4.3) and is expected to be a strong competitor in grassland conditions. The genotypes CSA-322 and CSA-334 (later referred to as 'grassy' genotypes) have multiple, slim, fascicular shoots (Table 4.3) and may withstand frequent cutting. In addition to these three main genotypes, one standard 'tall grassy' *M. sinensis* clone 'Goliath' and one 'tall woody' *M. sacchariflorus* genotype JSA-742 (later referred to as 'two additional genotypes') were included in the trials but, due to the lack of plant material, only planted in the sub-plots treated by one end-of-season cutting (i.e. Cr1 in Table 4.2).

In April 2012, rhizomes from the 4-year-old mother plants of the genotypes CSA-435, CSA-334, JSA-742 and 'Goliath' of the genetic collection of Julius Kühn Institute in Braunschweig, Germany, were cleaned of soil, cut into equal pieces of 8.5 cm length and planted into 10 × 10 × 10 cm pots in the greenhouse. The genotype CSA-322 was propagated *in vitro*, then also planted into 10 × 10 × 10 cm pots and kept for two months in the same greenhouse as the rhizome-propagated plantlets. In late May 2012, the grassland pre-treatments Er1 and Er4 (see Table 4.2) were applied at both experimental sites at IHO; then five pre-grown plantlets (25-30 cm tall for rhizome-derived plantlets; 15-20 cm tall for micropropagated plantlets) were manually transplanted from the pots to each sub-sub-plot. The distance between the plants within the plot was 0.71 m, resulting in a planting density of 19,607 plants ha^{-1}. Irrigation was only applied once after planting; no fertilization was applied during the experiment. One month after planting, in late June 2012, the living miscanthus plants in each sub-sub-plot were counted and the establishment survival rate was calculated in the same way as in the *UHO Experiment*. In October 2012, plant height and shoot number of the three main miscanthus genotypes (CSA-322, CSA-435 and CSA-334) were measured. To ensure successful establishment, the native grasses and miscanthus were not harvested in 2012. In April 2013, before the emergence of new shoots, miscanthus plants of the three main genotypes were harvested by hand to assess biomass yield (formed in the establishment year 2012). Then, in the growing season, the three cutting regimes (Cr1, Cr2 & Cr3 in Table 4.2) were applied. In October 2013, the living plants in each sub-sub-plot were counted to calculate the final survival percentage. The final survival percentages could not be recorded for miscanthus in the Cr3-treated sub-plots. By the end of the season, the frequent cutting in Cr3 had left the miscanthus plants too small to be accurately distinguished from other herbaceous species. After the living plants had been counted, the miscanthus and grass were harvested at a sub-sub-plot level and the total biomass yield (combined yield of miscanthus and grassland species) of each sub-sub-plot was recorded.

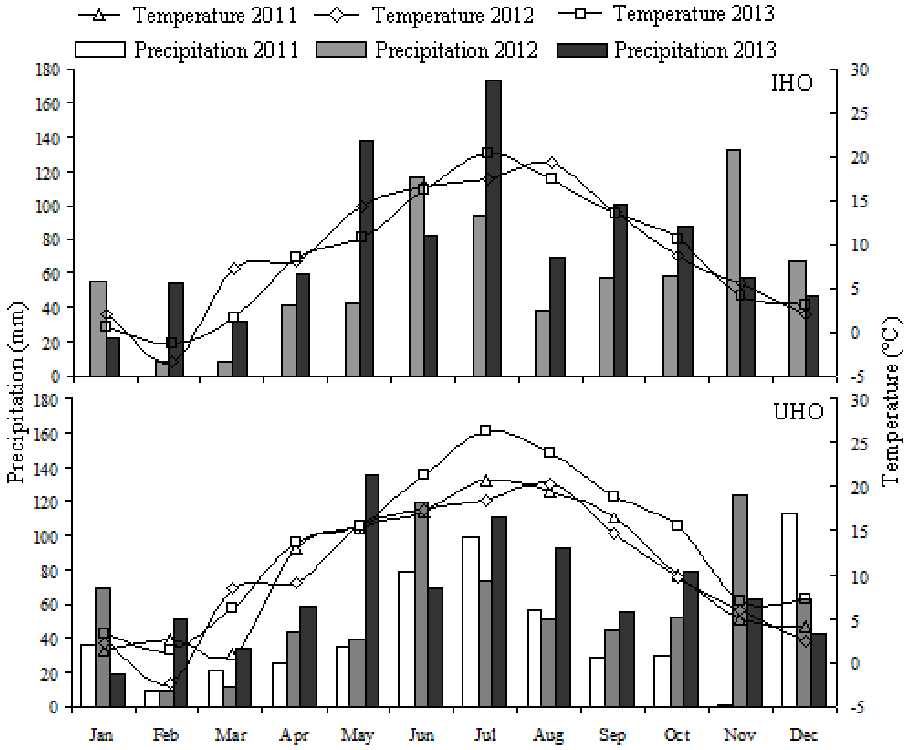

Fig. 4.1 Monthly average temperature and precipitation in 2011, 2012 and 2013 at the Experimental Station Ihinger Hof (IHO) and at Hohenheim (UHO), south-west Germany.

Table 4.2 Summary of the experimental treatments used in the field trials at the University of Hohenheim (*UHO Experiment*) and Ihinger Hof Experimental Station (*IHO Experiment*).

Field Trial	Experimental treatment types	Included treatments	Treatment application schedule
UHO Experiment	Establishment regimes (Er) [a]	Er1, Er2 & Er3	Only in the establishment year 2011
	Propagation methods (Prop) [b]	Rd & Rp	Only in the establishment year 2011
	Genotypes	*M. × giganteus* & *M. sinensis*	Planting in the establishment year 2011
	Cutting regimes (Cr) [c]	Cr1 & Cr2	Cr1 in 2012; Cr2 in 2013
IHO Experiment	Grassland types (Site)	High-productivity grassland & Low-productivity grassland	2012-2013
	Establishment regimes (Er)	Er1 & Er4	Only in the establishment year 2012
	Cutting regimes (Cr)	Cr1, Cr2 & Cr3	Starting from the second growing season 2013
	Main genotypes Additional genotypes	*M. sacchariflorus*: CSA-322, CSA-435 & CSA-334 *M. sinensis* ‘Goliath’ & *M. sacchariflorus* ‘JSA-742’	Planting in the establishment year 2012

a – Er indicates grassland pre-treatment prior to miscanthus planting by removing existing vegetation: Er1 = cutting the existing grassland vegetation to a height of 5 cm; Er2 = Er1 + soil tillage in 20 cm-wide bands to a depth of 3-5 cm and with a distance of 0.75 m between bands; Er3 = Er1 + whole-plot herbicide (glyphosate) spraying; Er4 = Er1 + spraying herbicide (Motivell Forte & Glyphosate) in stripes of 20 cm width with a distance of 0.71 m between stripes;

b – Rd = direct planting of rhizome cuttings into the field; Rp = transplanting rhizome-derived plantlets into the field;

c – Cr indicates the frequency of vegetation cutting/mowing during one growing season: Cr1 = only one end-of-season cutting in October; Cr2 = one in-season cutting in early June + one end-of-season cutting in October; Cr3 = one in-season cutting in early June + one in-season cutting in early August + one end-of-season cutting in October

Table 4.3 Morphological characteristics, biomass yield and geographic origin of miscanthus genotypes used in the trials at Ihinger Hof Experimental Station (*IHO Experiment*).

Species	Genotypes	Geographical origin (Latitude)	Canopy height[a] (cm)	Dry mass yield[a] (kg/plant)	Morphological classification[b]
M. sinensis	Goliath	Japan	300	2.65	Tall-grassy
M. sacchariflorus	CSA-322	China (45.2° N)	180	1.86	Tall-grassy
M. sacchariflorus	CSA-435	China (32.2° N)	230	1.13	Tall-woody
M. sacchariflorus	CSA-334	China (45.5° N)	150	1.89	Short-grassy
M. sacchariflorus	JSA-742	Japan (35.8° N)	245	3.91	Tall-woody

a - measured on the 4-year-old mother plants in early March 2012 at Julius Kühn Institute nursery in Braunschweig, Germany.

b - Tall-grassy = tall plants with slim and fascicular shoots; Tall-woody = tall plants with a few thick and widely spreading shoots; Short-grassy = short plants with slim and fascicular shoots.

4.2.4 Statistical analyses

Analysis of variance for the effects of establishment regime (Er), cutting regime (Cr), genotype (Geno), propagation method (Prop) and their interactions was performed using the MIXED procedure in SAS 9.4 (SAS Institute, Cary, NC, USA). Fisher's Protected LSD test was used to compare the treatment means; the differences were regarded as significant if $P<0.05$.

In the *UHO Experiment*, the data were analysed as a split-plot design with establishment regimes designated as the whole plot, and combinations of propagation methods and genotypes designated as the sub-plots. The total biomass yield was recorded for two growing seasons (2012 and 2013). However, since the cutting regimes were different in these two seasons (i.e. years cannot be treated as replicates), the variance analysis was performed separately for the 2012 and 2013 total biomass yield.

In the *IHO Experiment*, the cutting regimes were not applied in the first growing season; therefore the data collected in the establishment year, including establishment survival, plant height, shoot number and miscanthus yield, were analysed as a split-plot design. Here the establishment regimes were designated as whole plots and genotypes were designated as sub-plots. Statistical analyses of the data on final survival and total biomass yield collected in the second growing season were conducted as a split-split-plot design described in Section 4.2.2. In the statistical analysis of the total biomass yield, the pure grassland was also treated as a factor applied at sub-sub-plot level (as factor 'genotype'). Because the two additional miscanthus genotypes 'Goliath' and JSA-742 were only planted in the Cr1 treatment and were not fully randomised with the other genotypes, these were not included in the ANOVA analyses; instead the comparisons of 'Goliath' and JSA-742 with the other genotypes were conducted using the CONTRAST procedure in SAS. The data from the high-productivity and low-productivity grasslands were compared in the ANOVA analyses by setting site (i.e. different grassland types) as an ordered categorical factor.

4.3 Results

4.3.1 *Miscanthus* establishment and survival

In the *UHO Experiment*, the miscanthus establishment survival averaged 93.4%, with 6.7% higher survival (propagation methods pooled) observed for *M. sinensis* than for *M.* × *giganteus* (P=0.025 for the main effect of Geno). The establishment survival of *M.* × *giganteus* was significantly influenced by the propagation method: Rp-propagated plants (see Table 4.2) showed 20.0% higher establishment survival than Rd-propagated plants (100% vs. 80%). This difference was not observed for *M. sinensis* (P=0.002 for interaction of Geno × Prop). In the *IHO Experiment*, miscanthus showed similar, high (over 85%) establishment survival on both tested grasslands (Fig. 4.2). *Miscanthus* establishment survival was also not significantly affected by the establishment regime, but was significantly affected by the genotype (P=0.029 for the main effect of Geno). On average, the 'short' CSA-334 exhibited 10.8% and 9.6% lower establishment survival than the 'tall' CSA-322 and CSA-435, respectively. The other two 'tall' genotypes, 'Goliath' and JSA-742, were also observed to have 3.8% (P>0.05) and 8.8% (P<0.05) higher establishment survival than the 'short' CSA-334 (Er1 & Er4 and two grassland sites pooled).

In the *UHO Experiment*, the miscanthus final survival (assessed after three growing seasons) averaged 65.5% and was not significantly affected by genotype, establishment regime or propagation method (Table 4.4). In the *IHO Experiment* however, the final survival (assessed after two growing seasons) was affected by grassland site (low-productivity grassland vs. high-productivity grassland) and genotype, but not by establishment and cutting regimes (Table 4.4). On average, across all treatments, the final survival of miscanthus planted on the IHO low-productivity grassland significantly (P=0.025 for the main effect of site) exceeded that on the high-productivity grassland (84.7% vs. 74.4%). All 'tall' genotypes showed significantly higher final survival than the 'short' CSA-334 (Fig. 4.2) at both IHO sites. This difference was particularly pronounced on the IHO high-productivity grassland, where the final survival of the 'tall' genotypes (CSA-322, CSA-435, JSA-742 & 'Goliath' pooled) was on average 1.2 times higher than that of CSA-334.

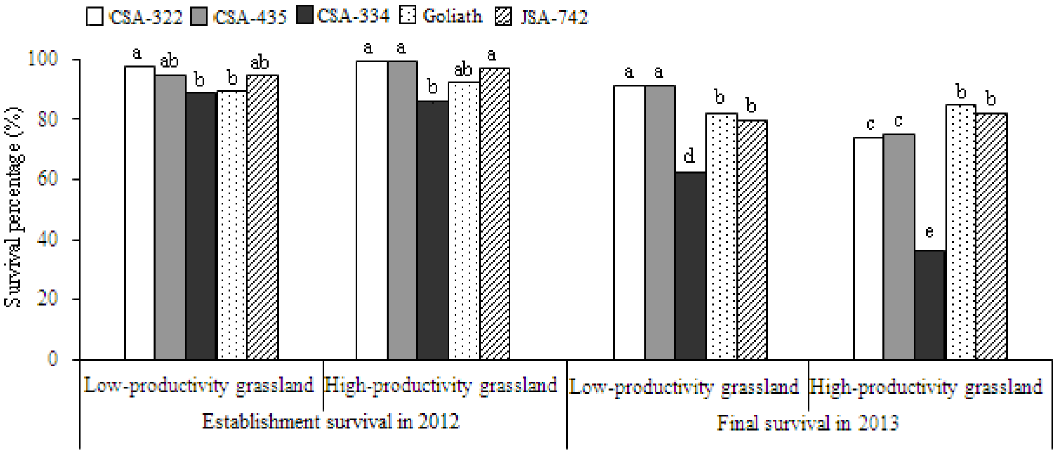

Fig. 4.2 The establishment survival (percentage of living plantlets to initially planted plantlets in the establishment year 2012) and final survival (percentage of living plantlets in 2013 to initially planted plantlets) of four *M. sacchariflorus* genotypes (CSA-322, CSA-435, CSA-334 & JSA-742) and one *M. sinensis* genotype (Goliath) planted on the Ihinger Hof (IHO) low-productivity and high-productivity grasslands. Different letters between any treatment means within each growing season indicate least significant differences at P<0.05 level.

Table 4.4 Analysis of variance (P values) for the effects of establishment regime (Er), cutting regime (Cr), genotype (Geno), propagation method (Prop) and their interactions on miscanthus establishment survival, final survival, establishment year's performance (plant height, shoot number and miscanthus biomass yield) and total biomass yield (grass and miscanthus) on the nitrogen-rich and nitrogen-poor grassland (i.e. two sites) at Ihinger Hof (IHO) and 'Goldener Acker' grassland at the University of Hohenheim (UHO).

Variation source	Establishment survival		Final survival		Plant height	Shoot number	Miscanthus yield	Total biomass yield		
	IHO	UHO	IHO	UHO	IHO	IHO	IHO	IHO	UHO-2012	UHO-2013
Site	0.931	Nt	**0.025**	Nt	**<0.001**	**0.006**	**0.006**	**<0.001**	Nt	Nt
Er	0.222	0.189	0.474	0.331	**<0.001**	**<0.001**	**0.001**	0.476	0.075	0.056
Cr	Nt	Nt	0.229	Nt	Nt	Nt	Nt	**<0.001**	Nt	Nt
Geno	**0.029**	**0.025**	**0.036**	0.312	**<0.001**	**<0.001**	**<0.001**	0.979	0.859	0.678
Prop	Nt	**0.002**	Nt	0.275	Nt	Nt	Nt	Nt	0.686	0.143
Site × Er	0.838	Nt	0.596	Nt	**<0.001**	0.097	0.073	0.867	Nt	Nt
Site × Geno	0.573	Nt	0.186	Nt	0.207	**0.004**	**0.001**	0.275	Nt	Nt
Er × Geno	0.765	0.697	0.666	0.383	0.891	**<0.001**	**0.012**	0.977	0.233	0.053
Prop × Geno	Nt	**0.002**	Nt	0.984	Nt	Nt	Nt	Nt	0.655	0.701

Nt means not tested in this study. Factor interactions not included in the variation source list did not significantly affect any recorded traits in this study. Figures marked in bold indicate significant effects ($p<0.05$) of the tested factors/interactions on the recorded traits.

4.3.2 Miscanthus performance and yield on IHO grasslands in the first growing season

In the *IHO Experiment*, the miscanthus was observed to have lower plant height ($P<0.001$ for the main effect of site) and biomass yield ($P=0.006$), but higher shoot number ($P=0.006$), on low-productivity than on high-productivity grassland in the first growing season (three main genotypes pooled). The miscanthus plants grown on high-productivity grassland were on average 10.8% taller than on low-productivity grassland (63.3 vs. 57.1 cm. Fig. 4.3A). This difference was however only observed in Er1 (existing vegetation cut but not sprayed). CSA-332 showed a significantly higher shoot number when grown on low-productivity grassland than on high-productivity grassland (Fig. 4.3B). This difference was not seen in the other genotypes. Only one genotype of the three tested, the ‘woody’ CSA-435, showed a significant biomass yield increase on high-productivity grassland compared to the low-productivity site (Fig. 4.3C). The CSA-334 and CSA-332 genotypes had similar yields at both sites.

The establishment regime and miscanthus genotype also had a significant effect on the morphological traits assessed (Table 4.4). The miscanthus planted on the Er4-treated grassland showed on average (three main genotypes pooled) 11.0% taller plants, 38.5% more shoots and 51.9% higher biomass yield than on the Er1-treated grassland. The difference in miscanthus plant height between Er1 and Er4 was only significant on the IHO low-productivity grassland (Fig. 4.3A; $P<0.001$ for the interaction of site × Er). Shoot number and miscanthus biomass yield were significantly increased by Er4 at both IHO sites (no significant interaction of site × Er). The difference in biomass yield corresponded to the significant increase in average shoot number per plant from 4 on Er1-treated to 5.9 on Er4-treated marginal grassland (Fig. 4.3B).

In grassland conditions, ‘woody’ miscanthus genotypes showed better morphological performance than ‘grassy’ genotypes, and ‘tall’ better than ‘short’ ones. Of the three main genotypes on both IHO grasslands, the ‘woody’ CSA-435 was characterized by taller plants (Fig. 4.3A) and higher biomass yield (Fig. 4.3C) than the ‘grassy’ CSA-322 and CSA-334. However, the ‘tall grassy’ CSA-322 had a 4.6 times higher shoot number than the other genotypes. Nevertheless, the per-hectare biomass yield of the ‘woody’ CSA-435 was on average 1.2 times higher than that of the ‘tall grassy’ CSA-322 and 3.6 times higher than the yield of the ‘short grassy’ CSA-334 (Fig. 4.3C).

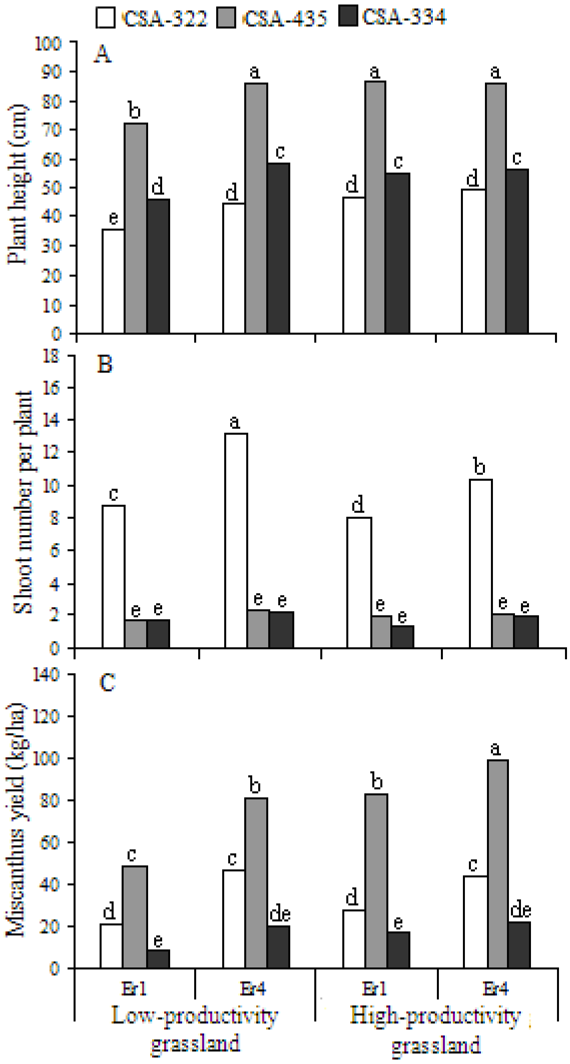

Fig. 4.3 Effects of establishment regimes (Er1 & Er4) on the first season's plant height (A), stem number (B) and aboveground biomass yield (C) of three *M. sacchariflorus* genotypes (CSA-322, CSA-435, CSA-334) planted on the Ihinger Hof (IHO) low-productivity and high-productivity grasslands. Er1 = grassland pre-treated by cutting the existing vegetation to a height of 5 cm; Er4 = Er1 + spraying herbicide in stripes of 20-cm width. Different letters between any treatment means within each measured trait indicate least significant differences at P<0.05 level.

4.3.3 Total biomass yield

In the *UHO Experiment*, the total biomass yield (combined yield of miscanthus and grassland species) was not affected by miscanthus-related factors (Table 4.4), including genotype and propagation method, because the contribution of the miscanthus to the total biomass yield was quite low (2%-4%) in the first three growing seasons. However, the total biomass yield was visibly higher in 2013 than that in 2012 (see Fig. 4.4). This may be a result of the more frequent cutting regime Cr2 applied in 2013 than Cr1 in 2012. In addition, the effect of establishment regime on the total biomass yield of the second ($P=0.075$ in 2012) and third ($P= 0.056$ in 2013) growing seasons was marginally significant. A decrease in total biomass yield (Fig. 4.4) was observed with the increase in grassland pre-disturbance. In the second growing season (2012), the total biomass yield reached 3.9 t ha^{-1} yr^{-1} on the least treated grassland (Er1), 2.7 t ha^{-1} yr^{-1} on the intermediate (Er2) and 2.6 t ha^{-1} yr^{-1} on the most intensely (Er3) treated grassland. However, these differences became less apparent (Fig. 4.4) with time: in the third year (2013), the total biomass yield of the Er1-, Er2- and Er3-treated UHO grassland averaged (miscanthus genotypes and propagation methods pooled) 8.1, 7.9 and 7.4 t ha^{-1} yr^{-1}, respectively.

In the *IHO Experiment*, the introduction of miscanthus did not generally affect the total biomass yield of the grassland: the mixed grassland/miscanthus and pure grassland plots showed no significant ($P=0.979$ for the main effect of Geno) difference in total biomass yield in the second growing season (see Fig. 4.5). However, the total biomass yield of the plots with 'Goliath' and JSA-742 miscanthus genotypes was on average 1.2 t ha^{-1} yr^{-1} higher ($P<0.05$) than that of the pure grassland plots on the high-productivity grassland (Fig. 4.5). In addition, a significant effect of grassland site ($P<0.001$) was observed for the second year's total biomass yield. The total biomass yield from the high-productivity grassland was almost twice that of the low-productivity grassland (Fig. 4.5). No differences in total biomass yield were observed between the establishment regimes at either IHO site, but the total biomass yield increased significantly ($P<0.001$ for the main effect of Cr) with increase in cutting frequency (Fig. 4.5). On average, the total biomass yield of the Cr2-treated grassland was nearly twice that of the Cr1-treated grassland (7.2 vs. 4.1 t ha^{-1} yr^{-1}). However, the Cr3-treated grassland showed only slightly higher biomass yield than the Cr2-treated grassland.

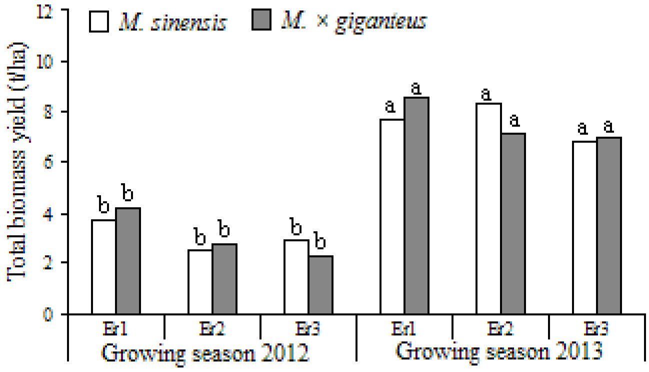

Fig. 4.4 Total biomass yield of miscanthus and grasses from grassland at the University of Hohenheim (UHO) in the second (2012) and third (2013) growing season. Er1 = grassland pre-treated by cutting the existing vegetation to a height of 5 cm; Er2 = Er1 + soil tillage in 20-cm-wide bands to a depth of 3-5 cm; Er3 = Er1 + whole-plot herbicide spraying; Rd =direct planting of rhizome cuttings into field; Rp = transplanting rhizome-derived plantlets into field. Different letters indicate least significant differences at P<0.05 level.

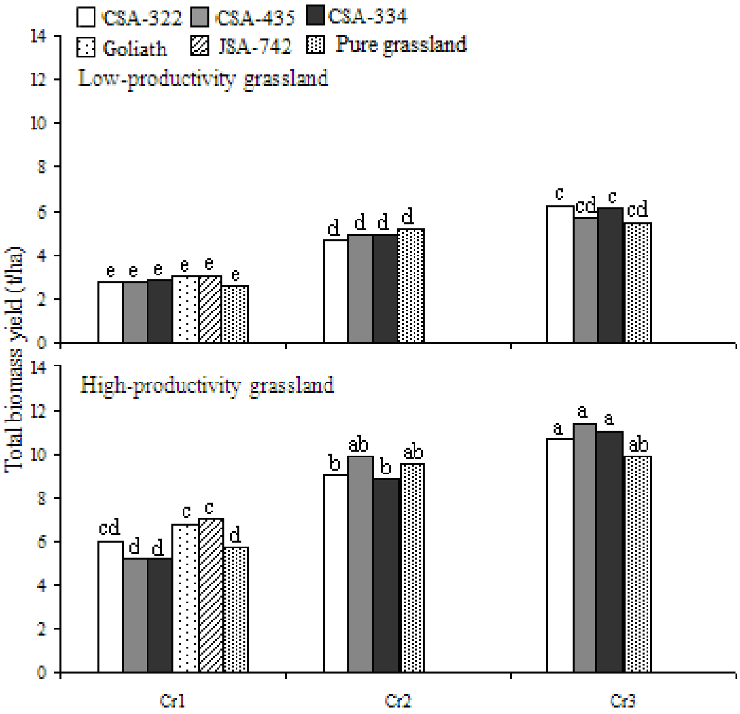

Fig. 4.5 Total biomass yield of miscanthus and grass from the Ihinger Hof (IHO) low-productivity and high-productivity grasslands with and without (i.e. pure grassland) established miscanthus in the secondgrowing season. Differences between four *M. sacchariflorus* genotypes (CSA-322, CSA-435, CSA-334 & JSA-742) and one *M. sinensis* genotype (Goliath) are shown here. Cr1 = only one end-of season cutting in October; Cr2 = one in-season cutting in early June + one end-of-season cutting in October; Cr3 = one in-season cutting in early June + one in-season cutting in early August + one end-of-season cutting in October. Different letters between any treatment means indicate leastsignificant differences at P<0.05 level.

4.4 Discussion

The first aim of this study was to establish miscanthus on C3 grassland with a no-till approach and assess the effects of genotype and propagation method on its establishment and growth. We found that over 80% of the plants survived in the establishment year on all tested grasslands. However, when final survival was assessed after two or three growing seasons, significantly fewer miscanthus plants were found on the high-productivity than on the low-productivity grassland. This could be the result of stronger competition from the existing grassland plant community on high-productivity soil where a denser and taller grass canopy with less gaps and correspondingly lower under-canopy light intensity were observed (expressed by a higher leaf area index (LAI) of 5.6 compared to 3.4 in the low-productivity grassland). Competition for light is considered one of the most important drivers of species elimination in grassland communities [59-61]. Lower light availability under the canopy and stronger competition from the existing vegetation may have led to higher in-season death of miscanthus plants on the high-productivity site in our trials. The differences in the existing grassland plant communities (species richness and ratios of grasses, legumes and forbs) may also play a role [62-64]. Grass species and miscanthus, both belonging to the *Poacea* family, may have similar resource requirements [65-66], therefore competition for these resources is likely to be higher on the high-productivity site (dominated by grasses) than on low-productivity grassland (with more diverse plant communities) when miscanthus is introduced.

Our results also showed the differences between miscanthus genotypes in their final survival on grassland: overall, 'tall woody' plants were more successful than 'short grassy' genotypes. 'Tall grassy' *M. sinensis* survived better than 'short grassy' *M. sacchariflorus*. This may be related to the stronger ability of larger, taller plants (tall and/or woody genotypes) to capture resources for growth and overwintering [37, 67-69]. Hence, the competitive tall and woody genotypes could be more suitable for miscanthus establishment on grassland.

We found that Rp-propagated plants had higher survival rates than Rd-propagated plants in the establishment year, which is consistent with previous studies [48, 70]. This difference may be due to less favourable rooting opportunities for rhizomes in the field (Rd) with water limitation and competition, compared to greenhouse conditions (Rp) [45, 70]. However, this effect was only apparent in the first year after planting; thus the propagation method did not affect the final survival of miscanthus on grassland. The *in vitro* propagated CSA-322 showed shorter plant height but higher shoot number than the other (rhizome-propagated) genotypes. This is in line with other studies that show *in vitro* propagation leads to an increase in shoot production but shorter and thinner shoots [45, 48].

The second aim of this study was to test how different grassland pre-treatments affect miscanthus establishment and growth. Competition for water, light and nutrients from the surrounding grassland vegetation normally limits the growth of introduced plants [50, 71-72]. However, low-competition conditions can be created in grassland through grazing, mowing or burning, to support the establishment and growth of introduced plant species [71, 73-75]. In this study, we applied four grassland pre-treatments varying in intensity from cutting the existing vegetation (Er1) to the complete removal of vegetation by herbicide (Er3). We found that more intense pre-treatments (herbicide spraying in strips or whole-plot spraying) supported the growth of miscanthus: the plants in such treatments were taller and accumulated more above-ground biomass. The grassland light conditions were similar in high-productivity and low-productivity grasslands after pre-treatment (LAI of 2.34 vs. 2.14). With similar light interception, most of the miscanthus genotypes tested grew equally well on high-productivity and low-productivity grassland, irrespective of the soil nitrogen content. Thus, competition for light seemed to be the main limitation on miscanthus growth in our trials, pointing to the importance of grassland pre-treatment for successful miscanthus establishment. The grass canopy closed more slowly in the plots treated with herbicide than in the plots where the existing vegetation was merely cut (visual observations). Thus, low-competition conditions for miscanthus growth were maintained longer in the herbicide-treated plots. Other studies have found similar effects of herbicide spraying on the growth of species introduced into grassland [50, 74-76].

A further aim of this study was to assess the effects of different grassland management practices (cutting frequency) on the biomass yield of miscanthus-improved grassland. The total biomass yields of Cr3-treated plots were only slightly higher than those of Cr2-treated plots, whereas the yields from both treatments (Cr3 and Cr2) were significantly higher than those of the Cr1-treated plots. This is consistent with earlier studies, which show that in-season cutting increases grassland productivity compared to a single end-of-season harvest [53, 77]. The native C3 grasses on the Cr1-treated plots started senescing from mid-June onwards (visual observation), resulting in a lower total annual biomass yield compared to the other cutting regimes. Increasing the number of in-season harvests from one (Cr2) to two (Cr3) only slightly increased the total biomass yield, as native C3 grasses have limited growth in summer [78-79]. In the *UHO Experiment*, the total biomass yield was higher in 2013 than in 2012 (see Fig. 4.4). This yield increase could have been potentially driven by several factors: larger (older) miscanthus plants, more advantageous weather conditions, and Cr2 applied in 2013 (only a single end-of-season harvest was performed in 2012). Because miscanthus yields and weather conditions (see Fig. 4.1) did not differ significantly between the two growing seasons, it can be speculated that the higher total biomass yield in 2013 was the result of the more frequent cutting in 2013 than in 2012. Despite the positive effect of multiple in-season harvests on grassland productivity, we observed a reduction in end-of-season miscanthus plant size, especially in the Cr3 treatments. Therefore, two harvests, one in late spring and one in late autumn (Cr2), seem to be the most suitable for the maintenance of miscanthus-improved grassland.

The total biomass yield of the mixed grassland/miscanthus stands was at times slightly higher, but overall similar to the yield of pure grassland plots, possibly due to the low contribution of miscanthus to the total biomass yield in the second growing season. As expected, the total biomass yield from high-productivity grassland was significantly higher than that from low-productivity grassland because the yields of the dominant native grasses were positively affected by nitrogen availability [80]. Interestingly, the miscanthus biomass yield assessed after the first growing season did not differ between low-productivity and high-productivity soils, except for the one genotype CSA-435. This indicates that there could be genotypic differences in the response of miscanthus to nitrogen and that most of the tested genotypes can grow equally well on both high-productivity and low-productivity grassland. Although more intense grassland disturbance (herbicide spraying) was beneficial for miscanthus survival and growth in grassland, we observed lower total biomass yield from the plots completely sprayed with herbicide than those not sprayed or sprayed in strips in the *UHO Experiment* in both the second and third growing seasons. Thus, when choosing the method of miscanthus establishment on grassland, these beneficial effects of grassland pre-disturbance should be balanced with the preservation of the original vegetation. Our results showed that the intermediate pre-disturbance Er4 (cutting existing vegetation to a height of 5 cm and spaying herbicide in strips) may be the most suitable to maintain biomass yields.

This study showed that miscanthus can be established on no-till C3 grassland and grassland productivity can be increased by introducing miscanthus, particularly if the competitive 'tall woody' miscanthus genotypes are used. Further multi-year experiments are necessary to draw conclusions on the long-term yield increase potential of miscanthus-improved grassland. Grasslands act as carbon sinks and assist nitrogen fixation and erosion prevention [81]. These ecological functions are often more important than the economic value of grassland. Therefore, the exploitation of grassland for miscanthus production would only be beneficial if its establishment does not negatively affect the ecology of the grassland. Future research should therefore include the assessment of possible changes in biodiversity and soil carbon sequestration on miscanthus-improved grassland.

4.5 Conclusion

This study showed that high miscanthus establishment success (over 80%) can be achieved on both high-productivity and low-productivity C3 grassland with no-till establishment and application of grassland pre-planting disturbance. In some cases the mixed grassland/ miscanthus stands had a slightly higher total biomass yield than pure grassland, indicating that introducing miscanthus could potentially improve grassland productivity. Our findings imply that competitive miscanthus genotypes with tall, thick shoots would be a better choice for establishment on grassland than genotypes with short, thin shoots, regardless of the species. With regard to propagation methods, our results revealed that transplanting rhizome-derived plantlets can lead to higher establishment success compared to direct rhizome planting for *M.* × *giganteus*. For *M. sinensis* however, the above two propagation methods led to an equally high establishment success. Our findings indicate that intermediate pre-treatment of grassland, i.e. cutting the existing vegetation to a height of 5 cm followed by spraying herbicide in narrow strips, is the most advantageous and could improve miscanthus establishment without negatively influencing grassland productivity. Similarly, the intermediate cutting regime (two cuts per season in spring and autumn) appears to be the most suitable for the maintenance of miscanthus-improved grassland because more frequent cutting increased grassland productivity but reduced miscanthus plant size.

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Appendix materials

Table A.4.1 Field map of trial on the ‘Goldener Acker’ grassland at the University of Hohenheim (*UHO Experiment*).

Rep1						**Rep 2**						**Rep 3**						**Rep 4**					
Er2		**Er3**		**Er1**		**Er3**		**Er1**		**Er2**		**Er1**		**Er3**		**Er2**		**Er2**		**Er3**		**Er1**	
Rd-S	Rd-G	Rd-G	Rp-G	Rd-G	Rd-S	Rp-S	Rd-S	Rd-G	Rp-G	Rd-G	Rp-S	Rd-G	Rd-S	Rp-S	Rd-S	Rd-G	Rp-G	Rd-G	Rp-G	Rd-G	Rd-G	Rd-G	Rp-S
Rp-S	Rp-G	Rp-S	Rd-S	Rp-S	Rp-G	Rd-G	Rp-G	Rp-S	Rd-S	Rd-S	Rp-G	Rp-G	Rp-S	Rd-G	Rp-G	Rd-S	Rp-S	Rp-S	Rd-S	Rp-G	Rp-S	Rd-S	Rp-G

Rep = Replication;

Er indicates grassland pre-treatment prior to miscanthus planting by removing existing vegetation: Er1 = cutting the existing grassland vegetation to a height of 5 cm; Er2 = Er1 + soil tillage in 20 cm-wide bands to a depth of 3-5 cm and with a distance of 0.75 m between bands; Er3 = Er1 + whole-plot herbicide (glyphosate) spraying;

Rd = direct planting of rhizome cuttings into field; Rp = transplanting rhizome-derived plantlets into field;

S = *Miscanthus sinensis*; G = *Miscanthus× giganteus.*

Table A.4.2 Field map of trials on the high-productivity and low-productivity grassland at Ihinger Hof (*IHO Experiment*).

(1) Experimental design of the field trial on high-productivity grassland.

Rep 1						**Rep 2**						**Rep 3**						**Rep 4**					
Er1			**Er4**			**Er4**			**Er1**			**Er4**			**Er1**			**Er1**			**Er4**		
Cr 3	**Cr 1**	**Cr 2**	**Cr1**	**Cr2**	**Cr3**	**Cr1**	**Cr3**	**Cr2**	**Cr2**	**Cr1**	**Cr3**	**Cr1**	**Cr3**	**Cr2**	**Cr3**	**Cr1**	**Cr2**	**Cr3**	**Cr1**	**Cr2**	**Cr3**	**Cr2**	**Cr1**
1	4	3	1	3	1	2	4	1	4	1	3	2	4	1	2	4	1	1	3	2	1	3	2
2	1	2	3	1	4	4	3	2	2	3	2	3	1	4	4	2	4	2	4	1	3	4	1
3	2	1	4	2	3	3	1	4	3	2	4	4	2	3	1	3	3	3	1	3	4	2	4
4	3	4	2	4	2	1	2	3	1	4	1	1	3	2	3	1	2	4	2	4	2	1	3
	5		5			6				6		5				6			6				5
	6		6			5				5		6				5			5				6

(2) Experimental design of the field trial on low-productivity grassland.

Rep 1						**Rep 2**						**Rep 3**						**Rep 4**					
Er4			**Er1**			**Er1**			**Er4**			**Er4**			**Er1**			**Er4**			**Er1**		
Cr 2	**Cr 1**	**Cr 3**	**Cr2**	**Cr3**	**Cr1**	**Cr2**	**Cr3**	**Cr1**	**Cr3**	**Cr2**	**Cr1**	**Cr3**	**Cr2**	**Cr1**	**Cr1**	**Cr3**	**Cr2**	**Cr1**	**Cr2**	**Cr3**	**Cr1**	**Cr3**	**Cr2**
3	1	2	4	1	3	1	4	2	4	1	3	1	3	2	4	2	4	2	3	4	3	2	3
2	3	4	2	3	1	3	2	3	1	4	2	3	4	3	1	3	2	4	1	2	4	3	1
1	4	3	3	4	2	4	1	4	3	2	4	4	1	1	3	4	1	1	4	3	1	4	2
4	2	1	1	2	4	2	3	1	2	3	1	2	2	4	2	1	3	3	2	1	2	1	4
	6				6			5			6			5	6			6			5		
	5				5			6			5			6	5			5			6		

Rep = Replication;

Er indicates grassland pre-treatment prior to miscanthus planting by removing existing vegetation: Er1 = cutting the existing grassland vegetation to a height of 5 cm; Er4 = Er1 + spraying herbicide (Motivell Forte & Glyphosate) in stripes of 20 cm width with a distance of 0.71 m between stripes;

Cr indicates the frequency of vegetation cutting during one growing season: Cr1 = only one end-of-season cutting in October; Cr2 = one in-season cutting in early June + one end-of-season cutting in October; Cr3 = one in-season cutting in early June + one in-season cutting in early August + one end-of-season cutting in October.

Number 1-6 represents the genotypes used in the trials: 1 = *M. sacchariflorus* CSA-435; 2 = *M. sacchariflorus* CSA-322; 3 = *M. sacchariflorus* CSA-334; 4 = pure grassland without planting miscanthus; 5 = *M. sinensis* ‘Goliath’; 6 = *M. sacchariflorus* JSA-742.

The 1 m paths (grassland) between the experimental plots are not shown.

Chapter 5 General Discussion

As described in Chapter 1, there are many problems surrounding miscanthus production. These issues constitute a serious of barriers that limit the implementation and expansion of miscanthus production. In previous chapters of this thesis, one technical limitation of the inefficient propagation technique was mitigated through minimizing the rhizome size and exploring the seeds propagation potential. The land-use dilemma was alleviated by exploring the marginal land production potential. Additionally, constrains of lack of genotypes and agronomic practices for the miscanthus establishment on marginal land were improved by field trials on grassland (the most important marginal land type with a huge potential). In addition to constrains improved in this thesis, there are still many other barriers. The present chapter aims to discuss further opportunities in upscaling issues in a broader context, focusing on issues in terms of technical, economical and financial, social and political, environmental aspects. Due to the different national conditions, the miscanthus production in different countries should not be limited by same issues all the time, especially in terms of social and legislative problems. Germany is a pioneering country with extreme ambition to expand miscanthus production; and China has a great potential and increasing interest to implement miscanthus production. Therefore, the following discussion only considers the further barriers and opportunities for the miscanthus production expansion in Germany and implementation in China.

5.1 Further technical barriers and opportunities

Technical constrains are usually the core issues that could derive many other barriers. For both Germany and China, the technical barriers mainly include lack of appropriate conversion techniques, economic propagation techniques, efficient equipments for planting/harvesting, various varieties and agronomic practices for the miscanthus establishment in different site conditions (especially marginal conditions).

Lack of appropriate conversion techniques restricts the development of miscanthus market. Due to the biomass fermentation techniques for bio-ethanol and biogas are not commercially mature, the current energetic application of biomass is limited to CHP (combined heat and power) and heating [1]. That is also true for miscanthus biomass.

However, due to the high ash alkalinity and low melting temperature of miscanthus biomass, the current biomass boilers (mostly designed for woody feedstock) are not compatible with miscanthus [2]. Plantation of SRC (short rotation coppice) is then more favoured [3], so the development of miscanthus is restricted. Further work needed towards improving boilers that suit miscanthus biomass well. Compared to SRC, miscanthus has a better fermentation quality (higher cellulose and lower lignin content) [4] and bioethanol is the most favoured bioenergy type. Future work should furtherly develop the fermentation techniques to produce bioethanol from miscanthus biomass.

The inefficient propagation techniques result in expensive planting materials (rhizome cuttings & plantlets), which are mainly responsible for the currently high upfront establishment costs [5]. In Chapter 2, the division efficiency and cost of rhizome propagation were improved by reducing rhizome size to 6-cm-long. Accordingly, the miscanthus establishment cost could be reduced to 1,800 € ha^{-1}. Even though, this lowered cost is still in excess of the expected cost by farmers (at least not higher than that of traditional crops). Further work should continue to improve the propagation techniques for cheap planting materials. However, there is not much space left for lowering the rhizome price by reducing size because the 6-cm length is close to the minimum size of rhizomes that can germinate after directly planting into field (Chapter 2). More future attentions to optimize the propagation should give to develop the most promising seed-based propagation system that has the highest multiplication ratio and lowest cost potential. Due to the current commercial clone of *M.* × *giganteus* is sterile [6], the primary task for developing the seed-based propagation system is breeding fertile varieties. For breeding programme, specialist varieties for each application (i.e. conversion technique) should be considered. As short-day plants [7-8], miscanthus species do not usually produce seeds or even flower in long-day conditions (e.g. in most of Germany and North China). The miscanthus seeds production is then a challenge, especially for Germany. The direct seed sowing is also unreliable presently, requiring further work towards developing safe seed establishment techniques.

Currently, the mechanization level of miscanthus production is low (Chapter 2). The used unspecialized equipments (e.g. modified potato planter for planting rhizome) are characterized to have low efficiency and high labour requirement [9]. Both finally contribute to a high miscanthus production cost and small margin. Even though, the machinery manufacturers would not invest in improving the mechanization for miscanthus production because the miscanthus market is currently not large enough to make it profitable for them. Government support is then required to encourage machinery suppliers to invest in the development of specialist equipments. According to literature review [5, 9-12], it is required to develop equipments for rhizome harvesting and planting, seeds threshing, coating and sowing, tissue culture plantlets cutting and transplanting, biomass harvesting. As mentioned before, the marginal land areas will be central to future miscanthus production. The irregular shaps and sometimes small-area (e.g. the edge area of a field) of marginal land may cause more turnings during field operations [13]. Therefore, it would be better to design these equipments as small-size devices.

Up to now, *M.* × *giganteus* is the single commercial clone available for the miscanthus production in Germany. The concern of potential outbreaks of diseases and pests, which is derived by the small number of varieties, may hamper confidences and interests of conservative farmers for miscanthus uptake [14]. More importantly, the *M.* × *giganteus* does not adapt well to stressful conditions, inhibiting the expansion of miscanthus production to stressful conditions (Chapter 1). In Chapter 4, the optimal genotype and effective agronomic practices for the miscanthus establishment on grassland were assessed. However, the environmental stresses differ between marginal land types. This means the experiences of establishing miscanthus on grassland should not be fully applicable for other marginal lands. Therefore, suitable genotypes and effective establishment methods for the other marginal lands still need to be developed [15]. This is more challenging for China because in comparison with Germany, China has more different marginal land types and environmental stresses are complex and changing [16-17]. The environmental stresses generally include poor soil fertility, drought, salinity, flooding, low temperature and contaminated soil. For a specific region, it may be subjected to only one of the above stresses or duple-stresses or even multi-stresses. Therefore, the selection criteria of future breeding programmes should be specified based on the environmental stresses of target area. In general, due to large areas of marginal land locating in dry Northwest China (Chapter 3), it is therefore crucial to exploit varieties with combined tolerance to drought and the other soil stress, e.g. saline soil, contaminated soil.

5.2 Economical and financial barriers and opportunities

In order for miscanthus to be adopted by farmers, the attainable profit of miscanthus production needs to be at least as high as that of traditional farming. However, the current miscanthus production in Germany is not economically competitive compared to the productions of traditional crops. Based on agronomic assumptions for rhizome establishment (shown in Chapter 2), plants management and biomass harvest (shown in Smeet *et al.* [18]), the currently annualized farm-to-gate cost of miscanthus production in Germany is calculated to be 2,230 € ha^{-1}. With a typical biomass yield of 20-25 odt ha^{-1} and biomass purchasing price of 90 € odt^{-1} [19], the current miscanthus production could generate a net margin of -430-20 € ha^{-1} yr^{-1}. In contrast, a net return of 475 € ha^{-1} yr^{-1} could be generated by the production of winter wheat and 205 € ha^{-1} yr^{-1} by winter rape in Germany [20]. In addition, the current miscanthus establishment procedure needs a high one-off investment, while it is difficult for most farmers to find upfront capitals for this investment [2]. It is even worse that there are low or no incomes during the intermittent years between planting and first harvest, which may create a ‘cash flow’ problem for most farmers. No farmers would like to participate in the uneconomic and financially risky miscanthus production. These are also the case in China. The miscanthus farm-to-gate production cost in China is estimated to be around 2,800 CNY ha^{-1} yr^{-1} (approximately 400 € ha^{-1} yr^{-1}) [21]. Under a typical biomass yield scenario of 15 t ha^{-1} yr^{-1}, the miscanthus production in China could generate a return of 6,200 CNY ha^{-1} yr^{-1} (approximately 885 € ha^{-1} yr^{-1}). Although the miscanthus production in China is more profitable than that in Germany, it is still lower than the traditional crops productions, e.g. a margin of 14,760 CNY ha^{-1} yr^{-1} (approximately 2,100 € ha^{-1} yr^{-1}) by the maize production [22]. With the increase of Chinese labour price, the competitiveness of miscanthus production in China is declining. For both countries, future works should drive down production costs, increase profits and develop grants and financial incentives for miscanthus production. The potential approaches to reduce the production cost mainly include improving the propagation techniques and developing the mechanization of production process. For increasing profits, a possible way is increasing the biomass yield potential by breeding. In addition, another potential approach is increasing the miscanthus economic value by cascade utilization [23-24], which uses same biomass in multiple successional applications as that product after its first use is used as feedstock for other additional uses. As a new concept, implementation of the miscanthus cascade utilization need to design a cascade utilization pathway and then transfer the processes into practice [25].

5.3 Social and political barriers and opportunities

In Germany, the government has launched a series of policies, active plans to improve the energy crops production (including miscanthus) [26], while farmers' negative attitude (social constrain) towards participating in miscanthus production is the main social and political barrier [27]. As a new crop, the miscanthus production involves a break from traditional agricultural practices [9]. For example, the rhizome propagation of miscanthus is totally different from the seeds propagation of traditional crops. Most farmers would not accept miscanthus at present because growing miscanthus is really a challenge for them. Also, the instability of small market damage farmers' confidence because farmers need a mature and reliable market to encourage their uptake of miscanthus. In addition, the current miscanthus production is uneconomic, financially risky and can block the farmers' land for more than 20 years without flexibility of changing crops. It stands to reason that most farmers would not shift growing profitable annual crops to growing less economic miscanthus on fertile land. It is not so bad that farmers would like to grow miscanthus on marginal land, which could not produce sufficient net revenues to be deemed worthwhile by producing food/feed [14]. However, there is not much such land available for growing miscanthus in Germany as that 200,000 ha unused grassland (grassland excluded that used for agriculture use) [28-30] and 500,000 ha unused land (mainly waste land and former mining land) [29] constitute most of the German marginal land reserve. What's worse, according to the German law, the grassland conversion into crop field (including energy crop) is forbidden at present [31-32]. It is therefore necessary to explore other possibilities that can provide space to grow miscanthus. A suggestion put forward is integrating miscanthus production into farming system or ecological protection system and using the highway and roadside land for miscanthus production [16]. In addition, it is also necessary to take measures to encourage farmers to accept and participate in the miscanthus production. For example, an educational programme should be promoted to train the farmers/landowners for good understanding of miscanthus production. Financial support programmes, which can provide financial capital for the miscanthus establishment and address the ‘cash flow’ problems in the first 3-4 growing seasons, should be gave a primary attention.

In comparison with Germany, farmers’ miscanthus acceptance is even lower in China as that there is no commercially cultivated miscanthus in China now. The political but not social constrains are the main barriers for the implementation of miscanthus production in China. On the national level, miscanthus is not in the list of energy crops propped up focally by the Chinese government. Due to the tough task of keeping safe food-supply, the Chinese government stays cautious to develop energy crops all the time. In addition, there are amounts of agricultural (740 Mt yr^{-1} [33]) and forestry wastes (200 Mt yr^{-1} [34]) available for bioenergy use in China. Therefore, most Chinese regulations and polices are always deflected to use waste biomass for developing bioenergy industries [35-36]. Until recently, support for developing non-food energy crops is officially confirmed by the ‘12th Five-Year Plan for Bioenergy Development (2011-2015)’. However, development of non-food biodiesel plants (e.g. *Jatropha carcas* L. & *Elaeis guineensis*) is listed as the primary programme and miscanthus is still not mentioned in this active plan. *Miscanthus* related industry as a quite new concept, without government support, it is unlikely that enterprisers would invest and participate in the miscanthus production. According to the ‘non-food’ principle set for the Chinese bioenergy industry, only growing non-food plants on marginal land is legal. *Miscanthus* is a promising non-food energy crop and China does have large areas of marginal land available and suitable for growing miscanthus (Chapter 3). However, it is argued that the legal criterion of marginal land is not clear now and farmers cannot determine whether it is legal or not when they grow miscanthus on land what they think is marginal [37]. This uncertainty may discourage farmers. Therefore, miscanthus needs to be legislative classified to the non-food energy crops group with development priority. A miscanthus promotion scheme is required to give confidence to potential farmers, entrepreneurs or agents. In this scheme, a training and education programme should be gave a priority. Also urgently, specific regulations of using marginal land to grow energy crops need to be introduced.

5.4 Environmental barriers and opportunities

Environmental concerns are the important barriers that limit using marginal land for miscanthus production [2]. Marginal lands usually locate in the fragile ecological region. Their ecological functions are more important than the economic values. The exploitation of marginal lands for miscanthus production would only be beneficial if miscanthus establishment does not negatively affect their ecological functions. However, there remains a lack of evidence on the environmental impacts of growing miscanthus on marginal land. Currently, the conclusion that miscanthus production in long periods has more beneficial than harmful impacts on environmental aspects was made based on comparison with the cultivation of traditional annual crops [2, 38-39]. It may be doubt whether this is also true for the miscanthus establishment on marginal land because some marginal lands already have initial ecological functions which are vulnerable that may be hurt by the miscanthus establishment [40]. For example, due to the shading of miscanthus canopy, there is a concern that the grassland may degrade after miscanthus establishment [41]. Further research is therefore required to test the real environmental impact of miscanthus production on marginal land.

Despite some effective approaches that can improve the miscanthus production found in this thesis, there is a long way needs to go to achieve a large area of miscanthus cultivation because the production is still constrained by many other technical, economical issues as described above. Among all the mentioned barriers, the technical issues are the basic and core constrains that subsequently derive many other barriers. Due to the current biomass conversion techniques are not mature enough to warrant a big biomass demand, the farmers’ confidence and interests to grow miscanthus are damaged. The inefficient and uneconomic propagation techniques directly result in high planting material price, expensive production cost and then finally cause a small net return, which is the main economic issue. Lack of various varieties and efficient agronomic practices make the miscanthus production as uncertain and risky (usually means daunting prospect) which may discourage farmers. Further research is therefore recommended to focus on improvements appropriate to technical issues as described above. This is not only applicable for Germany and China, but also for the other countries.

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Summary

Several species within the miscanthus genus (*Miscanthus spp.*) are characterized by high biomass yields and low production input requirements. This raised increasing interests in their applications for bioenergy. However, to date, only small areas of *Miscanthus × giganteus* (approximately 40,000 ha) are commercially grown and used for generating electricity and heat in Europe, where miscanthus has been developed as bioenergy crop for more than decade. Reviewing state-of-the-art revealed four main factors limiting the implementation of miscanthus production. These are inefficient and expensive propagation techniques, land use dilemma (i.e. lack of land available for growing miscanthus), lack of varieties/genotypes adapted to various and especially to stressful environmental conditions and lack of efficient agronomic practices for miscanthus establishment. Against these limiting factors, this thesis aims to (1) evaluate the different propagation systems with regard to technologies and costs, and improve the preferred rhizome propagation techniques; (2) address the land use dilemma through exploring marginal land (i.e. non-arable land with ability to grow plants with tolerance to environmental stresses) for miscathus production; (3) and screen optimal genotypes and effective practices for establishing and managing miscanthus on marginal land in a case study on grassland.

To achieve the first objective, a review, our own field trials and farmer surveys were performed. Direct seed sowing was found to be the cheapest propagation method (1,508.5 € ha^{-1} overall establishment costs) and micro-propagation the most expensive (6,320.8 € ha^{-1}). Direct rhizome planting is the farmers' most preferred and most applied establishment method and has moderate establishment cost of 1,904-3,375.7 € ha^{-1}. However, it goes along with the lowest propagation efficiency (1:10) and consequently restricts the availability of propagation material for large-scale plantations. However, the multiplication ratio can be increased by reducing the rhizome size. Field trial results showed that 6-cm length is close to the minimum size of rhizome that can germinate after directly planting into field. Compared to the traditionally used macro-rhizome, the multiplication ratio of the improved rhizome propagation (using 6-cm rhizomes) is tripled. In addition, the multiplication ratio can also be increased by transplanting rhizome- or stem-derived plantlets. However, due to higher labour and energy inputs

required for the pre-growing of plantlets, their establishment cost reduction potential is limited, with estimated costs of 4,240.8-4,400.8 € ha^{-1}. Direct seed sowing as the cheapest method is presently only possible for *Miscanthus sinensis* and not yet practical under German conditions. In addition, the seed-setting rate of *M. sinensis* is very low (0.0-28.7%) under the climatic conditions of south-west Germany, making commercial seeds production difficult. For all the propagation methods considered, more research efforts are still required to reduce the material production costs and simultaneously increase the multiplication ratio.

For the second objective, the production potential of miscanthus on marginal land in China was assessed. Because China has limited agricultural land resources and its non-food bioenergy policy (it is only allowed to grow energy crops on marginal land) is adamant, there is a desideration for exploiting its marginal land potential. In this study, Geographic Information System (GIS) techniques, model simulation were adopted to identify the productive marginal areas for miscanthus and to estimate their biomass and bioenergy production potentials. The results show that in China there are large marginal land areas of 17,163.54 × 10^{4} ha available for growing miscanthus. However, due to limitation by low winter temperatures and low precipitation levels in some areas, the total marginal area suitable for growing miscanthus is only 769.37 × 10^{4} ha. The Monteith radiation yield model was used to determine the potential miscanthus yield in Chinese climatic conditions. The simulation gave the actual harvestable yield levels on arable land of 18.1-44.2 odt ha^{-1} yr^{-1}. Taking the environmental stresses of marginal conditions into account an achievable miscanthus yield potential on marginal land of 2.1-32.4 odt ha^{-1} yr^{-1} was calculated (varying between different marginal land types). Based on these achievable yield levels, the total miscanthus production potential on the entire suitable marginal land areas is 13,521.7 × 10^{4} odt yr^{-1}; the corresponding bio-electricity generation and total greenhouse gas saving potentials are 183.9 TW h yr^{-1} and 21,242.4 × 10^{4} t CO_2 eq. yr^{-1}, respectively. The spatial distribution of the suitable marginal areas shows that they are mainly concentrated in the central part of Northeast China and the Loess Plateau. Both regions are recommended as priority development zones for the Chinese miscanthus-based bioenergy industry. However, implementation of this huge marginal land potential is currently constrained by many barriers, e.g.

concerns on potential ecological effects, competition for marginal land from other uses, lack of high yield varieties in marginal conditions.

Lack of varieties with suitability to marginal conditions and efficient agronomic practices for the establishment on marginal land are the main barriers that limit using marginal land for miscanthus production. Therefore, stress tolerant varieties need to be selected and methods of effective establishment of miscanthus on marginal land need to be developed. Worldwide, grassland is the most important marginal land type because it has the largest terrestrial area and mild environmental stresses for growing energy crops (including miscanthus). However, it is undesirable or even legally prohibited to convert grassland into bioenergy cropland to avoid biodiversity loss and soil carbon being reduced by tilling practices. Hence, no-till establishment practices for miscanthus establishment and maintenance on grassland are investigated here under the third objectives. Our study demonstrates that miscanthus can be successfully cultivated on both good (nutrient-rich) and marginal (nutrient-poor) grassland using the proposed agronomic practices and an increased grassland productivity may be achieved through the establishment of suitable miscanthus genotypes. The recommended agronomic practices are summarized as following. *Miscanthus* genotypes with tall, thick shoots perform better than those with short, thin shoots. Better establishment is achieved when rhizome-derived plantlets are transplanted into pre-disturbed grassland. The grassland pre-disturbance of low vegetation cutting (5 cm) and herbicide spraying in narrow stripes is recommended for its beneficial effect on miscanthus establishment without significant negative effects on grassland productivity. Two harvests, one in late spring and one in late autumn, are optimal to achieve a high grassland yield.

In this thesis, the limitation of the inefficient propagation technique was mitigated through minimizing the rhizome size and exploring the seeds propagation potential. The land-use dilemma was alleviated by exploring the marginal land production potential. Additionally, constrains of lack of genotypes and agronomic practices for the miscanthus establishment on marginal land were improved by field trials on grassland (the most important marginal land type with a huge potential).These results can improve the implementation and expansion of miscanthus production. However, in addition to constrains improved in this thesis, the miscanthus production is currently constrained by many other technical, economic and financial, social and political, environmental issues. It is unlikely that the implementation and expansion will achieve without mitigating these constrains. Further research and support should address these barriers in an integrate manner.

Zusammenfassung

Mehrere Arten innerhalb der Gattung Miscanthus (*Miscanthus spp.*) zeichnen sich durch hohe Biomasseerträge und eine effiziente Ressourcennutzung aus. Daher steigt das Interesse an ihrer Nutzung als Rohstoff für die Bioenergieerzeugung. Dennoch wird in Europa, wo innerhalb der letzten Jahrzehnte die Nutzung von Miscanthus als Bioenergiepflanze entwickelt wurde, bis heute nur der Genotyp *M.* × *giganteus* in geringem Umfang (ca. 40.000 ha) kommerziell angebaut und zur Erzeugung von Strom und Wärme genutzt. Anhand der Überprüfung des aktuellen Wissensstands kristallisieren sich im Wesentlichen vier Hauptursachen heraus, die die Ausweitung des Miscanthusanbaus begrenzen. Neben der ineffizienten und dadurch sehr teuren Vermehrung von Miscanthus spielt vor allem der Mangel an Land, welches für den Anbau von Miscanthus verfügbar ist, eine Rolle. Zusätzlich fehlen einerseits geeignete Sorten beziehungsweise Genotypen, die an verschiedene Umweltbedingungen - vor allem auf marginalen Standorten - angepasst sind, und andererseits effiziente Verfahren zur Etablierung von Miscanthus. Ziel dieser Dissertation ist es, Lösungen für die oben genannten limitierenden Faktoren zu finden, um den weiteren Ausbau des Miscanthusanbaus zu ermöglichen. Dies soll geschehen durch (1) eine Evaluierung der vorhandenen Vermehrungsverfahren hinsichtlich der verschiedenen Technologien und jeweiligen Kosten sowie durch die Verbesserung des Verfahrens der Rhizomvermehrung; (2) die Erforschung marginaler Standorte, d.h. zur Zeit ungenutzte landwirtschaftliche Nutzflächen, die potenziell für den Anbau von stresstoleranten Kulturpflanzen in Frage kämen, auf ihre Eignung für den Anbau von Miscanthus zu überprüfen; (3) sowie die Selektion optimaler Genotypen und effizienter Verfahren für die Etablierung und Bewirtschaftung von Miscanthus auf marginalen Standorten in einer Fallstudie auf Grünland.

Um das erste Ziel zu erreichen, wurden neben einer Literaturstudie und einer Umfrage unter landwirtschaftlichen Betrieben auch eigene Feldversuche durchgeführt. Es wurde festgestellt, dass Direktsaat das günstigste Vermehrungsverfahren ist (1.508,5 € ha^{-1} Gesamtetablierungskosten) und In-vitro-Vermehrung das teuerste (6.320,8 € ha^{-1}). Das von landwirtschaftlichen Betrieben bevorzugte und dadurch auch am häufigsten

angewandte Verfahren ist die direkte Pflanzung der Rhizome. Diese Methode ist zwar verhältnismäßig kostengünstig (1.904-3.375,7 € ha^{-1}), hat aber auch die geringste Vermehrungseffizienz (1:10), wodurch nicht ausreichend Vermehrungsmaterial für den großflächigen Miscanthusanbau zur Verfügung gestellt werden kann. Allerdings kann durch eine Verkleinerung der Rhizome die niedrige Vermehrungseffizienz bei der direkten Pflanzung der Rhizome verbessert werden. Die im Rahmen dieser Arbeit durchgeführten Feldversuche haben gezeigt, dass eine Mindestgröße der Rhizome von etwa 6 cm erforderlich ist, um ein Austreiben der Rhizome nach der Direktpflanzung nicht zu beeinträchtigen. Die Vermehrungseffizienz des bislang praxisüblichen Verfahrens der direkten Pflanzung der Rhizome kann durch die Verwendung von 6 cm langen Rhizomen verdreifacht werden. Des Weiteren kann durch das Verpflanzen von aus Rhizomen oder Stängeln gewonnenen Jungpflanzen die Vermehrungseffizienz weiter erhöht werden. Doch die Etablierungskosten sind mit geschätzten 4.240,8 – 4.400,8 € ha^{-1} auch wesentlich höher, da dieses Verfahren arbeits- und energieintensiver ist. Das günstigste Vermehrungsverfahren, die Direktsaat, ist bislang nur mit *Miscanthus sinensis* möglich, jedoch nicht unter den klimatischen Bedingungen Süddeutschlands, wo außerdem die Samenbildungsrate sehr niedrig ist (0,0 bis 28,7 %) und somit nicht für eine kommerzielle Saatgutproduktion vor Ort ausreichen würde. Folglich sind für alle hier berücksichtigten Vermehrungsmethoden weitere Forschungsanstrengungen notwendig, um sowohl die Produktionskosten zu senken als auch das Multiplikationsverhältnis zu erhöhen.

Für das zweite Ziel wurde das Produktionspotenzial von Miscanthus auf Grenzertragsflächen in China berechnet. China hat nur begrenzte landwirtschaftliche Nutzflächen zur Verfügung und seine Non-Food-Bioenergiepolitik legt fest, dass nur marginales Land für den Anbau von Energiepflanzen genutzt werden darf. In dieser Studie wurde das Geographische Informationssystem (GIS) sowie eine Modellsimulation genutzt, um marginale Standorte in China für den Anbau von Miscanthus zu identifizieren, sowie ihre Biomasse- und Bioenergiepotenziale abzuschätzen. Die Ergebnisse zeigen, dass in China theoretisch 17.163,54 × 10^4 ha marginales Land für den Anbau von Miscanthus zur Verfügung stehen. Aufgrund der Einschränkungen durch niedrige Temperaturen im Winter und geringe

Niederschlagsmengen, umfassen davon jedoch die Flächen, die auch praktisch für den Anbau von Miscanthus geeignet sind, nur 769,37 × 10^4 ha. Ein Strahlung-Ertrags-Modell nach Monteith wurde verwendet, um den potenziellen Miscanthusertrag unter den klimatischen Bedingungen in China zu bestimmen. Die Simulation ergab, dass der potenzielle Ertrag auf Ackerland in China zwischen 18,1 und 44,2 t Trockenmasse (TM) ha^{-1} $Jahr^{-1}$ liegt. Wenn die Umweltbedingungen auf den Marginalstandorten berücksichtigt werden, ergibt sich für verschiedene Grenzertragsflächen ein durchschnittliches Ertragspotenzial für Miscanthus von 2,1 bis 32,4 t TM ha^{-1} $Jahr^{-1}$. Basierend auf diesen modellierten Erträgen, ist das Biomassepotenzial von Miscanthus hochgerechnet auf die gesamten geeigneten marginalen Flächen 13.521,7 × 10^4 t TM $Jahr^{-1}$. Ausgehend von diesem Biomassepotenzial ergibt sich eine theoretische Stromerzeugung von 183,9 TWh $Jahr^{-1}$ und somit eine theoretische Treibhausgaseinsparung von insgesamt 21.242,4 × 10^4 t CO_2eq. $Jahr^{-1}$. Die Untersuchung der räumlichen Verteilung der geeigneten Flächen zeigt, dass sich diese vor allem auf den zentralen Teil von Nordostchina und das Löss-Plateau konzentrieren. Beide Regionen bieten sich daher als prioritäre Entwicklungszonen für die chinesische miscanthus-basierte Bioenergieindustrie an. Allerdings ist die Nutzung dieses großen Potenzials an marginalem Land zur Miscanthusnutzung in der Praxis derzeit aus mehreren Gründen nur eingeschränkt möglich. So gibt es beispielsweise Probleme hinsichtlich der ökologischen Auswirkungen der Ausweitung des Miscanthusanbaus, des Wettbewerbs um marginale Flächen, in dem der Miscanthusanbau mit anderen Verwendungsmöglichkeiten konkurriert, sowie zusätzlich des Mangels an für diese Grenzertragsflächen geeigneten Hochertragssorten.

Das Fehlen von Genotypen, die an die Bedingungen marginaler Standorte angepasst sind, und ein Mangel an effizienten praxistauglichen Etablierungsverfahren verhindern bislang die Nutzung marginaler Standorte durch den Anbau von Miscanthus.

Daher müssen stresstolerante Genotypen identifiziert werden sowie effektivere Methoden zur Etablierung von Miscanthus auf marginalen Standorten entwickelt werden. Weltweit stellen hierfür Grünlandflächen die bedeutendsten Marginalstandorte dar, da sie zum einen die größte Landfläche bieten und zum anderen noch relativ milde

Stressfaktoren für den Anbau von Energiepflanzen (einschließlich Miscanthus) aufzeigen. Um den aus klima- und umweltschutztechnischen Gründen unerwünschten und oft gesetzlich verbotenen Grünlandumbruch zu vermeiden, wurden im dritten Teil dieser Arbeit direkte Etablierungsverfahren von Miscanthus auf Grünlandstandorten untersucht, die keine Bodenbearbeitung benötigen. Die Studie ergab, dass Miscanthus in einem speziellen Anbauverfahren sowohl in guten (nährstoffreichen) als auch in marginalen (nährstoffarmen) Grünlandbeständen integriert werden kann, wobei die Produktivität des Grünlands durch geeignete Miscanthus-Genotypen sogar verbessert werden kann. Die hierfür empfohlenen Anbautechniken können wie folgt zusammengefasst werden. Miscanthusgenotypen mit hohen, dicken Trieben entwickelten sich besser als solche mit kurzen, dünnen Trieben. Die Etablierung kann ferner dadurch optimiert werden, indem aus Rhizomen gezogene Jungpflanzen gepflanzt werden. Grundsätzlich ist es empfehlenswert, vor der Etablierung des Miscanthus einen niedrigen Schnitt des Grünlands (5 cm) mit einer anschließenden Herbizidbehandlung in schmalen Streifen zu kombinieren. Diese Bewirtschaftungsweise hat zum einen eine positive Wirkung auf die Etablierung der Miscanthusbestände und zum anderen geringe negative Auswirkungen auf die Produktivität des Grünlandes. Um hohe Grünlanderträge zu erzielen, sollte das Grünland in jedem Jahr zweimal geschnitten werden, im späten Frühjahr und im Spätherbst.

In dieser Thesis konnte die geringe Effizienz der Vermehrungsverfahren von Miscanthus durch eine Verwendung kleinerer Rhizome sowie durch die Erforschung der Möglichkeit Miscanthus über Samen zu vermehren verbessert werden. Das Landnutzungs-Dilemma konnte durch eine Untersuchung des Produktionspotenzials marginaler Standorte klarer eingegrenzt werden, und in Feldversuchen konnten zusätzlich Genotypen und Anbautechniken aufgezeigt werden, die eine direkte Etablierung (ohne Bodenbearbeitung) von Miscanthus auf Gründlandstandorten, und somit auf den bedeutendsten marginalen Standorten ermöglichen. Diese Ergebnisse können dabei helfen, die Einführung und den Ausbau des Miscanthusanbaus weiter voranzutreiben. Allerdings gibt es neben den Limitationen, die in dieser Arbeit diskutiert werden, viele weitere technische, ökonomische, soziale, politische und ökologische Belange, ohne deren Berücksichtigung eine zunehmende Einführung und

Ausweitung des Miscanthusanbaus unwahrscheinlich bleibt. Zusätzlich bedarf es daher weiterer Forschung, in der diese Probleme im Gesamtzusammenhang betrachtet und bearbeitet werden.

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