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Review

A review of the genetic study of the energy crop *Miscanthus*

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ABSTRACT

Miscanthus is one of the top nine potential perennial energy crops. Its high production of biomass is largely due to its efficient C4 photosynthesis pathway and rhizome function, even in cool climates. As a relatively new industrial plant in Europe, it needs the support of both breeding programmes and biotechnology techniques. The paper aimed at synthetic resumption of genetic studies of the energy crop *Miscanthus*. Phylogenetics, genetic diversity, confirmation of hybridisation origin, genetic structure of natural populations, mapping, transferability of markers, molecular diagnostic and sequencing in genus *Miscanthus* were reviewed. Perspectives for the application to *Miscanthus* breeding of such molecular techniques as: hybrid creation with marker-aided selection, genome sequencing and synteny were also discussed.

1. Introduction

The interest in using biomass for energy production is constantly growing in Europe [1–5]. The countries of the EU are committed to producing an increasing proportion of their energy needs from renewable sources. According to the Polish Ministry of Economy, biomass has the best outlook for development among all potential renewable energy source technologies in Poland [6]. In a forecast of the structure of renewable energy sources [6] in 2010, 53% of energy will be generated from biomass. In the UK, recent suggestions say that about 350,000 ha of perennial energy crops will be used to supply energy by 2020 [7]. *Miscanthus* field experiments from many locations in Europe show that this is the crop with the highest energy production (MJ ha^{-1}) among all potential energy plants [8,9]. Additionally, it is characterised as having the highest energy-use efficiency (EUE). Because these crops efficiently produce biomass, these grasses may play an important role in sustainable agriculture of biomass fuel production in the near future [4,10]. The predictions, which are supported by modelling studies using MISCANFOR, showed that *Miscanthus* could provide 12% of the EU's primary energy needs by 2050 if 35% of the current arable land is used to cultivate *Miscanthus* [11]. Scenario of future changes in land use supporting biomass plantation presented Rounsevell et al. [12]. Today, *Miscanthus* acreage in Poland is over 4000 ha (Gostkowski – chairman of the Bio-Energia group of Praterm, pers. comm.). The 2009 report from Department for Environment, Food and Rural Affairs (DEFRA) says that about 12,700 ha of *Miscanthus* plantations exist in the UK with harvestable area of 9672 ha [13].

In a series of studies on agronomy (e.g., [14]), productivity (e.g., [15]) and utilisation (e.g., [16]), three *Miscanthus* species were identified as having the highest potential for biomass production. These species are *M. sinensis*, *M. sacchariflorus* and *M. x giganteus*. The most important species for biomass

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Table 1 – Molecular techniques used in *Miscanthus* species studies.

Molecular technique	Species	References
Mitochondrial DNA sequences	*M. sinensis*	[40]
Chloroplast restriction enzyme site mutation	*M. sinensis*	[41]
Isoenzymes	*M. sinensis* *M. x giganteus* *M. saccharifloru*	[50]
AFLP	*M. sinensis* *M. sacchariflorus* *M. x giganteus* Progeny of *M. sinensis* × *M. sacchariflorus*	[51]
Satellite	*M. sinensis*	[44]
rDNA-IGS sequences	*M. sinensis*	[68]
Inter-*Alu*-like species-specific sequences	*M. sinensis*	[45]
rDNA-IGS sequences	*M. sinensis*	[54]
RAPD	*M. sinensis*	[55]
rDNA-ITS sequences	*M. sinensis*	[46]
Mikrosatellite RFLP	*M. sinensis* *M. sinensis* *M. sacchariflorus* *M. x giganteus* *M. condensatus*	[65]
rDNA-ITS sequences	*M. sinensis* *M. floridulus*	[67]
RAPD	*M. sinensis* mapping population	[59]
AFLP ISSR	*M. sinensis* *M. floridulus* *M. nepalensis* *M. oligostachyus* *M. sacchaliflorus* *M. transmorrisonensis* *M. x giganteus*	[28]
AFLP GISH FISH rDNA-ITS sequences Plastid DNA sequences	*M. x giganteus*	[53]
Plastid DNA sequences rDNA-ITS sequences	*M. sinensis* *M. sacchariflorus* *M. transmorrisonensis* *M. ecklonii* *M. floridulus* *M. fuscus* *M. junceus* *M. nepalensis* *M. oligostachyus*	[47]
QTLs identification for total height, flag-leaf height and stem diameter	*M. sinensis* mapping population	[60]
QTLs identification for yield and its componets	*M. sinensis* mapping population	[61]
QTLs identification for Cl and K content	*M. sinensis* mapping population	[62]
QTLs identification for Ca, P and S content	*M. sinensis* mapping population	[63]
QTLs identification for flowering date	*M. sinensis* mapping population	[64]
Alcohol dehydrogenase one (*Adh1*) gene sequence Plastid DNA sequences	*M. sinensis* *M. condensatus*	[31]
α-*tubulin* gene sequence	*M. sinensis* *M. condensatus* *M. floridulus*	[49]
AFLP Mikrosatellite	*M. sinensis* *M. floridulus*	[38]
AFLP RFLP	*M. sinensis*	[36]
Mikrosatellite	*M. sinensis*	[56]

AFLP-amplified fragment length polymorphism; FISH – fluorescent *in situ* hybridisation; GISH – genomic *in situ* hybridisation; ISSR – internal-simple sequence repeat; QTL – quantitative trait locus; RAPD – random amplified polymorphic DNA; rDNA-IGS – intergenic spacer of ribosomal DNA; rDNA-ITS – internal transcribed spacer of ribosomal DNA; RAPD – random amplified polymorphic DNA; RFLP – restriction fragment length polymorphism.

production is *M. x giganteus* Greef et Deuter which is the triploid hybrid between *M. sinensis* and *M. sacchariflorus* [17,18]. However, *M. sinensis* and *M. sacchariflorus* seem to be the most important species to broaden the genetic base for successful breeding [19]. The new *M. sinensis* hybrids selected from the cross pollination of *M. sinensis* genotypes are characterised by a resistance to leaf senesce and showed no significant root biomass reduction together with no significant influence of low water supply on shoot production [20]. Breeding programmers could be interested too in an artificial polyploid creation in the *M. sinensis* genus. Preliminary studies of tetraploid forms in field experiments showed that some genotypes could have greater tuft weight, greater stem diameter, taller stems [21] and greater leaf width in comparison to the controls [22].

As a relatively new industrial plant in Europe, *Miscanthus* needs the support of both breeding programmes and biotechnology techniques. The paper aimed at synthetic resumption of genetic studies of the energy crop *Miscanthus*. In the Table 1 the molecular techniques used in the studies of 11 *Miscanthus* species, mapping population and interspecific hybrids were presented. Phylogenetics, genetic diversity, confirmation of hybridisation origin, genetic structure of natural populations, mapping, transferability of markers, molecular diagnostic and sequencing in genus *Miscanthus* were reviewed. At the end the discussion on possible ways for science investigations to support *Miscanthus* breeding was added.

2. Application of molecular techniques in *Miscanthus* studies

2.1. *Phylogenetics*

The genus *Miscanthus* belongs to the *Saccharum* complex along with *Erianthus*, *Narenga*, *Saccharum* and *Sclerostachya*. The first time the term "*Saccharum* complex" has been used by Mukherjee in 1957 [23] to group together five genera exhibited