GENETIC DIVERSITY OF ELEPHANTGRASS ECOTYPES FOR BIOENERGY PRODUCTION

João Romero do Amaral Santos de Carvalho Rocha[a], Pedro Crescêncio Souza Carneiro[a], Ricardo Augusto Diniz Cabral Ferreira[a], Jailton da Costa Carneiro[b] and Juarez Campolina Machado[b*]

[a]Universidade Federal de Viçosa
[b]Embrapa Gado de Leite

[a]Avenida Peter Henry Rolfs s/n, Viçosa 36570-900, Brazil. joaoascrocha@gmail.com; carneiro@ufv.br; ricardo.cabral.agronomia@gmail.com
[b]Rua Eugênio do Nascimento 610, Juiz de Fora 36038-330, Brazil. +55 (32) 3311-7410/juarez.machado@embrapa.br; jailton.carneiro@embrapa.br

ABSTRACT: The aim of this work was to study the genetic diversity of elephantgrass ecotypes and recommend crosses for bioenergy production. The accessions of the Active Elephantgrass Germplasm Bank of Embrapa (BAGCE) classified in the standard ecotypes Cameroon and Napier were evaluated. Three evaluation cuttings were carried out. The 1st and 2nd cuttings were harvested at 250 days, and the 3rd, at 315 days. The genetic values were predicted by mixed model methodology via REML/BLUP. The Tocher clustering method was used to infer about the genetic diversity within the Cameroon and Napier ecotypes, based on the genetic dissimilarity matrix, obtained by the standardized Euclidean distance. The crosses recommendation was based on Tocher method using the matrix of genetic similarity. To the best of our knowledge this is the first study that investigate the genetic diversity of elephangrass ecotypes, aiming to select genitors and crosses to breed elephantgrass for bioenergy production. Our results indicate that elephantgrass accessions have high genetic diversity distributed in different clusters within each ecotype and crosses between Cameroon ecotypes are recommended to improve the elephantgrass for bioenergy purposes.
Keywords: biomass, combustion, genetic improvement.

1 INTRODUCTION

The Active Elephantgrass Germplasm Bank (BAGCE) maintained by Embrapa has 160 accessions. Of these, 101 accessions are of the specie *Pennisetum purpureum*, 19 accessions are of the species of the tertiary gene *pool* of the genus (*Pennisetum spp.*), and 40 accessions are of a work collection of *P. glaucum*. The pre-breeding expansion efforts of elephantgrass, such as the activities of characterization and evaluation of the germplasm favor its use as a bioenergy source.

The division of elephantgrass into four groups of standard ecotypes according to morphological variability was related by Lira et al. [1]: Cameroon - presents erect genotypes with thick stalks, broad leaves, upright clumps and late flowering; Napier - presents genotypes with thick/intermediate stalks, broad leaves, open clumps and intermediate flowering; Mecker - presents reduced height genotypes with thin stalks, thin and more numerous leaves, and early flowering; and Dwarf - presents lower height genotypes (up to 1.5 m high), and high leaf/stalk ratio. However, the study of genetic diversity insides of standard ecotypes considering multi-trait (*e.g.* morphological, agronomic and biomass quality traits) may guide us toward to crosses recommendation. Crosses recommendation is one of the hardest tasks in the genetic breeding. A successful cross is one that leads to a new cultivar. For this purpose, the parents should have high genetic value and genetic complementation.

Therefore, the objective of this research was to study the genetic diversity of elephantgrass ecotypes and recommend crosses for bioenergy production.

2 MATERIAL AND METHODS

2.1 Genetic material and experimental conduction

One hundred accessions of the Active Elephantgrass Germplasm Bank of Embrapa (BAGCE) were evaluated, of which 18 were classified in the Cameroon ecotype, 44 in the Napier ecotype, four in the Mercker ecotype, and the other accessions were classified as intermediate to the aforementioned groups. However, only the data from Cameroon and Napier groups were used to estimate the genetic diversity, due to the small number of accessions in the Mercker ecotype, and to the non-existence of accessions of the dwarf ecotype in this study. Besides, the ecotypes Mercker and Dwarf have low energy biomass production potential. Plots (1.5 m x 4 m) consisted of a single 4 m row. Rows were planted side by side, spaced 1.5 m apart. Plots were allocated in a simplex lattice design, with two replications. Three evaluation cuttings were carried out for this study. Aiming at using them as bioenergetic feedstock, 1st and 2nd cuttings were harvested at 250 days, and the 3rd, at 315 days.

2.2 Evaluated Traits

The following morpho-agronomic and biomass quality traits were measured: mean height (m); phenotypic vigor (1 to 5); stalk diameter (mm); total green biomass (Mg ha^{-1}); total dry biomass (Mg ha^{-1}); acid detergent fiber (g Kg^{-1}), neutral detergent fiber (g Kg^{-1}), cellulose (g Kg^{-1}), lignin (g Kg^{-1}); hemicellulose (g Kg^{-1}); cellulose/lignin ratio (-); *In vitro* digestibility of the dry biomass (g Kg^{-1}); nitrogen (g Kg^{-1}); ash (g Kg^{-1}); calorific value (MJ kg^{-1}) and dry matter concentration (g Kg^{-1}).

2.3 Statistical analyses

The genetic values for the 17 traits were predicted by mixed model methodology via REML/BLUP [2, 3]. The statistical model adopted was: $y = Xm + Zg + Wb + Op + Ti + \varepsilon$, where y is the data vector; m is the measurement-replication combination effect vector (fixed), added to the general mean; g is the genotypic effect vector (random); b is the block effect vector (random); p is the permanent environmental effect vector (random); i is the genotypes x cuttings interaction effect vector; and ε is the residue vector (random). X, Z, W, Q, and T are the incidence matrices for the effects. The Tocher clustering method [4]

was used to infer about the genetic diversity within the Cameroon and Napier ecotypes, based on the genetic dissimilarity matrix, obtained by the standard Euclidean distance of the genetic values. The crosses recommendation was based on Tocher method using the matrix of genetic similarity. The software Selegen-REML/BLUP [5] and R program [6] was used.

3 RESULTS AND DISCUSSION

3.1 Genetic variability

The genetic variability within the ecotypes Cameroon and Napier are presented in the Fig. 1 and 2.

3.2 Napier clustering

The genetic diversity analysis among the 44 Napier ecotypes revealed 14 clusters. The first cluster allocated 23 accessions (Figure 2). The Napier ecotypes have greater genetic variability compared with the Cameroon based on morpho-agronomic and biomass quality traits. According to Rocha et al. (2017) the Napier ecotypes showed greater genetic values for the biomass quality traits with exception of caloric value.

3.3 Cameroon clustering

Considering the variability of the morpho-agronomic and biomass quality traits evaluated in the 18 accessions within Cameroon ecotypes, seven clusters were observed by using the Tocher method (Figure 1). Seven accessions were allocated in the first cluster and they are the most similar.

According to Rocha et al. [7] Cameroon ecotypes presents the greatest aptitude to be used as bioenergetic raw material to energy co-generation due to the greater total dry biomass and calorific value. Therefore, it should be given focus on Cameroon ecotypes for energy co-generation genetic breeding.

3.4 Crosses recommendation

Traditionally, the Tocher clustering method uses the genetic dissimilarity matrix, thus the average distance intracluster must be lesser than the average distance intercluster [4]. On the other hand, if we consider the genetic similarity matrix, we will find in the same cluster the most divergent genotypes and the most similar ones in different clusters. In this way, we can recommend intercrossing of the selected genotypes considering the genetic variability among than. Therefore, the accessions Vruckwona Africano, BRS Capiaçu, Roxo Botucatu, IAC Campinas and Roxo do Itassú (Cameroon ecotypes) should be crossed in a complete diallel scheme

Thinking to improve the biomass quality of Cameroon ecotypes, we recommended elite accessions intercrosses in a partial diallel scheme (elite Cameroon ecotypes vs. elite Napier ecotypes).

It is important to comment that the diallel crosses is a good alternative to crosses scheme, because it could be used to obtain genetic parameters, estimates the effects of general and specific combining ability between parental and understanding the genetic control and inheritance of traits to develop strategies aimed at efficient selection and breeding [8, 9, 10].
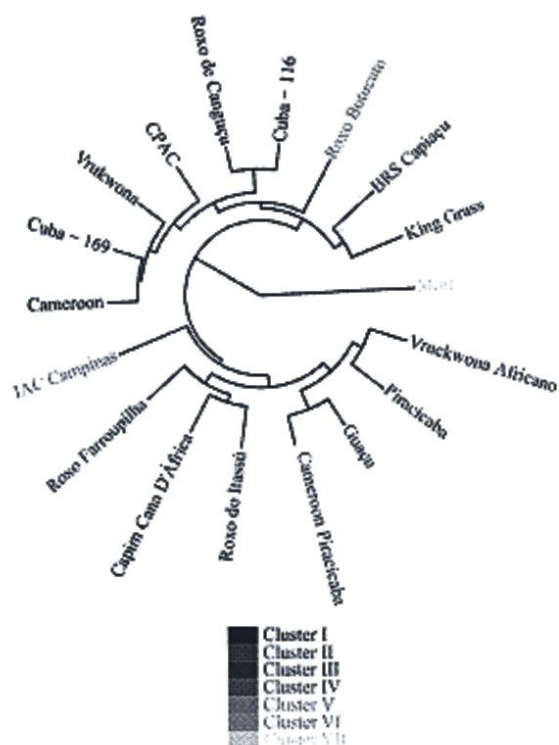
4 FIGURES

Figure 1: Dendrogram of 18 accessions of Cameroon ecotypes, based on average linkage clustering method, obtained from genetic values of morpho-agronomic and biomass quality traits. Different colors represent the clusters of similar accessions formed using the Tocher clustering method.
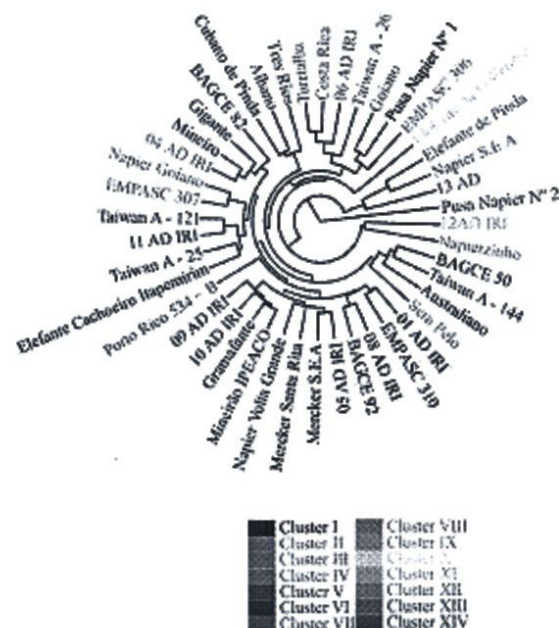

Figure 2: Dendrogram of 44 accessions of Napier ecotypes, based on average linkage clustering method, obtained from genetic values of morpho-agronomic and biomass quality traits. Different colors represent the clusters of similar accessions formed using the Tocher clustering method.

247

5 REFERENCES

[1] M.A. Lira, M.V. Cunha, A.V. Pereira, Melhoramento genético do capim-elefante. In: M.A. Lira, J.C.B. Dubeux Júnior, A.C.L. Mello. (Eds.). Capim-elefante: Fundamentos e Perspectivas. IPA/UFRPE, Recife (2010), pag. 48.

[2] H.D. Patterson, R. Thompson, Recovery of inter-block information when blocks sizes are unequal. Biometrika (1971), 58, 545–554.

[3] C.R. Henderson, Best linear unbiased estimation and prediction under a selection model. Biometrics (1975), 31, 423–447.

[4] C.R. Rao, Advanced statistical methods in biometric research. New York: John Wiley & Sons. (1952), pag. 390.

[5] M.D.V. Resende, Selegen-REML/BLUP: Sistema Estatistico e Seleção Genética Computadorizada Via Modelos Lineares Mistos. Embrapa Florestas, Colombo (2007), pag. 359.

[6] E. Paradis, J. Claude, K. Strimmer. APE: analyses of phylogenetics and evolution in R language. Bioinformatics (2004), 20: 289-290.

[7] J.R.A.S.C. Rocha, J.C. Machado, P.C.S. Carneiro, J.C. Carneiro, M.D.V. Resende, A.V. Pereira, J.E.S. Carneiro, Elephant grass ecotypes for bioenergy production via direct combustion of biomass. Ind. Crops Prod. (2017); 95: 27-32.

[8] N. Danehloueipour, G. Yan, H.J. Clarke, K.H.M Siddique, Diallel analyses reveal the genetic control of resistance to ascochyta blight in diverse chickpea and wild *Cicer* species Euphytica (2007); 154-195.

[9] J. Möhring, A.E. Melchinger, H.P. Piepho, REML-based diallel analysis. Crop Sci. (2011), 51: 470-478.

[10] T. Townsend, V. Segura, G. Chigeza, T. Penfield, A. Rae, D. Harvey, D. Bowles, I.A. Graham, The Use of Combining Ability Analysis to Identify Elite Parents for Artemisia annual F1 Hybrid Production. PLoS ONE (2013); 8 (4): e61989.

7 LOGOS

248