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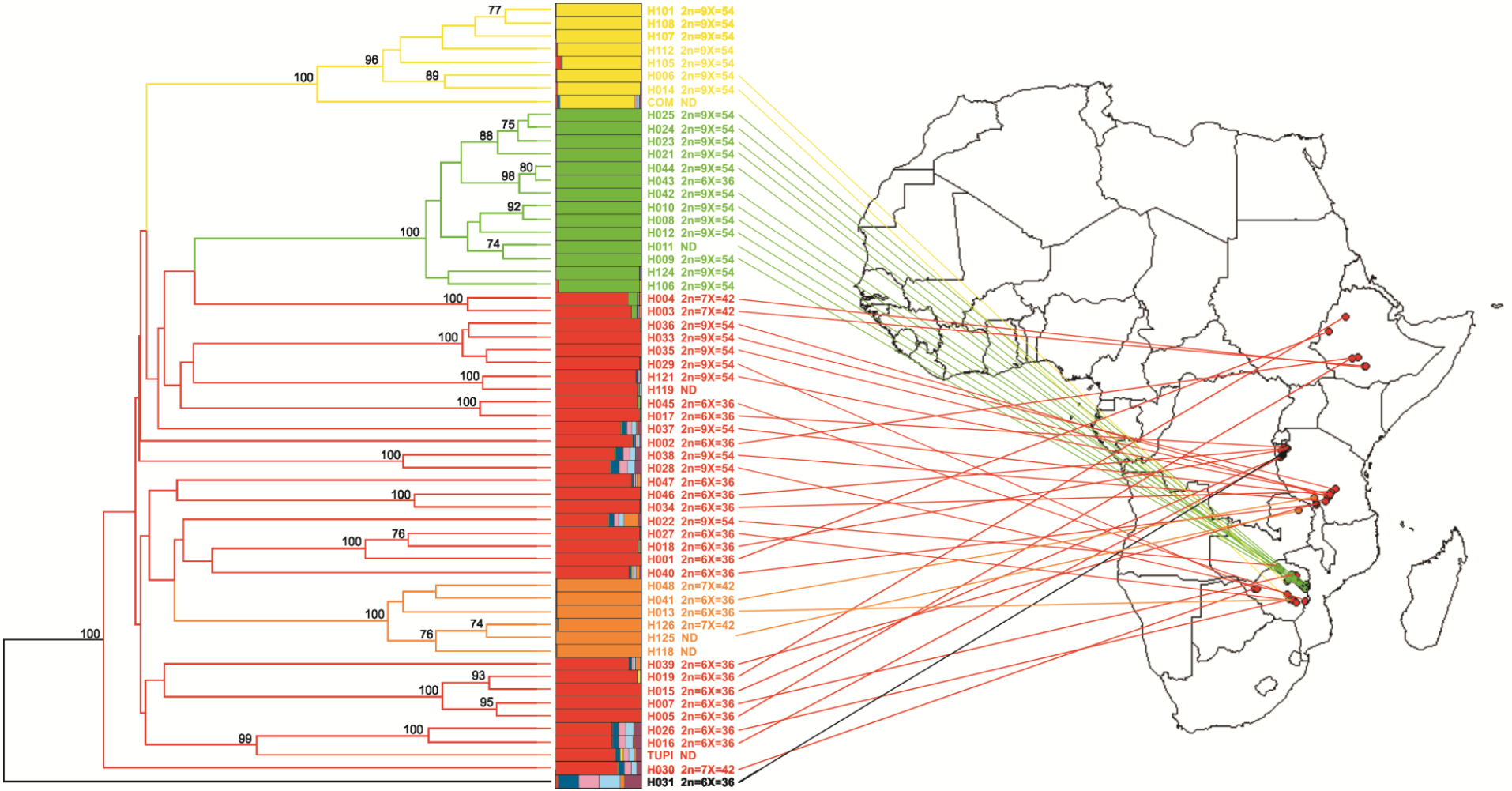
EVALUATION OF A GERMPLASM COLLECTION OF BRACHIARIA HUMIDICOLA USING MICROSATELLITES, MORPHOLOGICAL MARKERS, CYTOGENETICS AND GEOGRAPHICAL ORIGIN

Document prepared by experts from Brazil

Brachiaria humidicola (Rendle) Schweick. is a warm-season grass commonly used as forage in the tropics. Accessions of this species were collected in eastern Africa and massively introduced into South America in the 1980s. Several of these accessions form a germplasm collection at the Brazilian Agricultural Research Corporation (Embrapa). However, apomixis, ploidy, and limited knowledge of the genetic basis of this germplasm collection have constrained breeding activities. The objectives of this work were to access genetic variability in the 58 accessions of *B. humidicola* germplasm collection held at Embrapa using microsatellite markers and to compare the results with information on: (1) collection sites of the accessions; (2) reproductive mode and ploidy levels; and (3) genetic diversity revealed by morphological traits. The 58 accessions and 2 varieties of *B. humidicola* were genotyped with 27 microsatellite loci. Allelic profiles resulting from the combination of all loci revealed 60 distinct genotypes, thus indicating that this germplasm collection does not contain duplicated materials. Molecular data generated for these genotypes were subjected to two kinds of analyses. At first, we submitted data to a similarity-based method, by using the diversity software package DARwin to calculate a pair wise similarity matrix whose entries

gave the Jaccard's similarity between every pair of individuals. For the data from *B. humidicola* used in this study, distance-based methods could not be applied due to the impossibility to estimate allele frequencies for this material at microsatellite loci, owing to the high ploidy levels of the genotypes. The similarity matrix was than graphically represented by a UPGMA tree. Secondly, we used a model-based method, assuming that observations from each cluster were random draws from some parametric model. Individuals were allocated into clusters by estimating their likely membership coefficients for each cluster, using Bayesian statistics implemented in the software STRUCTURE. After running the analyses, the two methods showed to be complementary. Clusters formed using STRUCTURE software supported groups resulting from UPGMA tree, and revealed information that could not be accessed with the similarity-based grouping method. The sole sexual accession of this germplasm formed an out-group in the UPGMA tree, and revealed by Bayesian statistics a very particular genetic background in relation to all the other accessions, apomictics. The evaluated germplasm collection showed to be highly structured into four major groups. Genetic dissimilarities (which correspond to the arithmetical complement of similarities) did not correlate with either geographical distances of collections sites of accessions or genetic distances inferred from 24 morphological descriptors previously analyzed in this material. Additionally, the genetic structure identified in this collection did not linearly correspond to differences in ploidy levels reported elsewhere (accessions presenting 2n = 36, 42, and 54 chromosomes). The figure bellow is a comprehensive drawing to illustrate the relationship between the molecular results (similarity and Bayesian-based), cytogenetic data and geographical origin of evaluated materials.

Jungmann L, Vigna BBZ, Boldrini KR, Sousa ACB, Valle CB do, Resende RMS,
Pagliarini MS, Zucchi MI, Souza AP



UPGMA tree showing accessions and cultivars grouped by Jaccard's similarity, with Bootstrap values > 70% displayed in branches. Between the lines of branches and genotypes names, inserted bars represent allelic pools profile for each evaluated genotype (resulting from Bayesian analyses). Chromosome counts and ploidy levels are indicated at the right side of genotypes names. Accessions with geographic coordinates available in the Forages Database at CIAT (http://www.ciat.cgiar.org) are linked to collection sites, indicated on the African continent map using the software DIVA-GIS 5.2.0.2.

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