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Full length Research Paper

Analysis of genetic diversity in accessions of *Irvingia gabonensis* (Aubry-Lecomte ex O'Rorke) Baill

G. N. Ude[1,2]*, C.O. Dimkpa[2], P. O. Anegbeh[3], A. A. Shaibu[2], A. Tenkouano[2], M. Pillay[4] and Z. Tchoundjeu[5]

[1]Department of Natural Sciences, Bowie State University, 14000 Jericho Park Road, Bowie, MD 20715, USA. [2]Crop Improvement Division, Plantain and Banana Improvement Program, International Institute of Tropical Agriculture, Onne, PMB 008 Nchia Eleme, Rivers State, Nigeria. [3]World Agroforestry Centre (ICRAF), IITA Station, Onne, PMB 008 Nchia Eleme, Rivers State, Nigeria.
[4]International Institute of Tropical Agriculture, P.O. Box 7878, Kampala, Uganda. [5]ICRAF, BP 2067, Nkolbisson, Yaounde, Cameroon.

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Amplified fragment length polymorphism (AFLP) was used to assess genetic diversity and relationships among 15 accessions of *Irvingia gabonensis* collected from Cameroun, Gabon, and Nigeria. Twelve AFLP+3 primers produced 384 polymorphic fragments. Average genetic distance (AGD) between the 15 accessions was 58.7% (32-88%). AGD and range of genetic distance among accessions from Cameroun, Nigeria and Gabon were 62% (53-76%), 52% (32.3 – 84.8%) and 50% (45-53%), respectively, indicating more genetic diversity in Cameroun than Nigeria and Gabon. The unweighted pair-group method of the arithmetic average (UPGMA) and principal coordinate analysis (PCO) showed a clear distinction between the Gabon and Nigeria accessions into two separate clusters, with accessions from Cameroun overlapping them. Principal coordinate analysis (PCO) indicated a closer relationship between accessions from Cameroun and Gabon. In general the Cameroun germplasm appears to be a bridge between the genetically isolated Nigeria and Gabon accessions. This overlap of Gabon and Nigerian accessions by the accessions from Cameroun may be an indication that Cameroun is the center of diversity of *I. gabonensis* and also the primary source of original materials grown in the other countries. More collection in Cameroun is necessary to ensure the optimum collection and preservation of the existing genetic diversity in *I. gabonensis*.

Key words: *Irvingia gabonensis*, accession, amplified fragment length polymorphism, genetic diversity.

INTRODUCTION

Irvingia gabonensis is an economically important fruit tree native to moist tropical forests in west and central Africa (Harris, 1996; Lowe et al., 2000). As part of the effort to conserve the gene pool of *I. gabonensis* against deforestation, the World Agroforestry Centre (ICRAF) and International Institute of Tropical Agriculture (IITA), Onne, Nigeria have engaged in intensive germplasm collection activities that led to the establishment of three *ex-situ* field gene-banks in Nigeria and Cameroun. Currently, all effort is geared towards genetic characterization of the accessions for the purpose of enhancing their application in crop improvement and tree domestication (Tchoundjeu et al., 1998; Anegbeh, 2000).

Earlier studies have quantitatively assessed the tree to tree variation in *I. gabonensis* (Leakey et al., 2000; Atangana et al., 2002; Anegbeh et al., 2003). Knowledge of the diversity of the genetic base as well as the intra-specific distribution of genetic variation within the native ranges of *I. gabonensis* (Lowe et al., 2000) is essential for

*Corresponding Author's E- mail: gude@bowiestate.edu. Tel: (301) 860 3347. Fax: (301) 860 3346.

Table 1. Accessions and collection centers of *Irvingia gabonensis*.

S/No	Accession name	Local name	Collection site	State/Country
1	BM3	Ogbolo	Isua Akoko	Ondo/Nigeria
2	A29	Ogbono	Iva valley	Enugu/Nigeria
3	A19	Ogbono	Onne	Rivers/Nigeria
4	N36	Ogbolo	Ipesi akoko	Ondo/Nigeria
5	A22	Ogbono	Obubra	Cross River/Nigeria
6	CAM10	Bareko	Schouam	Cameroun
7	CAM11	Koumadjab	Payo	Cameroun
8	CAM12	Banque	Souap	Cameroun
9	G27	Ndock	Misi	Gabon
10	N18	Ogbolo	Lona ayangba	Kogi/Nigeria
11	B12	Ogbolo	Aredo farm	Edo/Nigeria
12	G19	Ndock	Adan bine 1	Gabon
13	G10	Mouiba	Nyali bangono 1	Gabon
14	G9	Mouiba	Douano	Gabon
15	N43	Ogbolo	Olubi mamu	Oyo/Nigeria

producing appropriate conservation and sustainable utilization strategies. Random amplified polymorphic DNA analysis identified 'Hot Spots' of genetic diversity of *I. gabonensis* in southern Nigeria (West Africa), southern Cameroun and central Gabon (Central Africa) (Lowe et al., 2000). However, knowledge of the full extent of genetic diversity in *I. gabonensis* populations in West and Central Africa is still very scanty (Lowe et al., (2000). Therefore, the objective of this paper is to assess the distribution of genetic variation and relationships among some accessions of *I. gabonensis* collected from Nigeria, Cameroun and Gabon using the amplified fragment length polymorphism (AFLP) technique.

MATERIALS AND METHODS

Plant materials

Fifteen accessions of *Irvingia gabonensis* representing germplasm collection from various sites in Nigeria (West Africa), Cameroun and Gabon (Central Africa) were used in this study. The accessions are maintained in the field genebanks of ICRAF established at the International Institute of Tropical Agriculture, Onne station, Nigeria (Table 1).

DNA extraction and AFLP procedure

About 10 g of leaf tissues per sample, collected from the field in liquid nitrogen, were ground with a mortar and pestle. DNA extraction, AFLP procedure and data analysis were as described by Ude et al. (2002). DNA fingerprints were generated with 12 *Eco*R1+3 and *Mse*1+3 pairs of primers (E-AGC/M-CTT; E-AAC/M-CAT; E-AAG/M-CAA; E-ACA/M-CTG; E-ACT/M-CTG; E-ACC/M-CTA; E-ACG/M-CAG; E-AGG/M-CAC; E-AGC/M-CTG; E-ACT/M-CTA; E-AAG/M-CAC; and E-ACA/M-CTT) obtained from the GIBCO BRL commercial AFLP kit. Using a Hoefer SQ3 vertical gel sequencer, the bands were separated electrophoretically in polyacrylamide gels according to the procedure of Lin et al. (1996). A dendrogram was constructed from the matrix of similarity coefficients, using the unweighted pair-group method of the arithmetic average (UPGMA). Genetic distances (GD%) were obtained by subtracting the similarity indices from 1 and multiplying the outcome by 100 [(1-Sij) x 100] (Kim et al., 1992). Neighbour-joining and multidimensional principal co-ordinate analyses (PCO) were used to reveal relationships among the 15 accessions in a scatter-plot.

RESULTS AND DISCUSSION

Three hundred and eighty four polymorphic fragments were obtained with the 12 AFLP primers used in this study. The average number of fragments per primer pair was 32 (range, 14 - 41). The genetic dissimilarity coefficients between accessions varied widely, ranging from 0.32 to 0.84 with an average of 0.59. The UPGMA analysis was able to discriminate between all the 15 genotypes. The dendrogram (Figure 1) divided the 15 accessions into two major genetic groups (Clusters A and B). With the exception of N43, cluster A contained all the other Nigerian accessions and CAM 10, an accession from Cameroun. Cluster B was comprised of four accessions from Gabon, N43 from Nigeria and CAM11 and CAM12 from Cameroun.

The accessions from Cameroun were found to be the most diverse with an average genetic distance of 61.5% (range, 55.5 – 76%). The accessions from Nigeria and

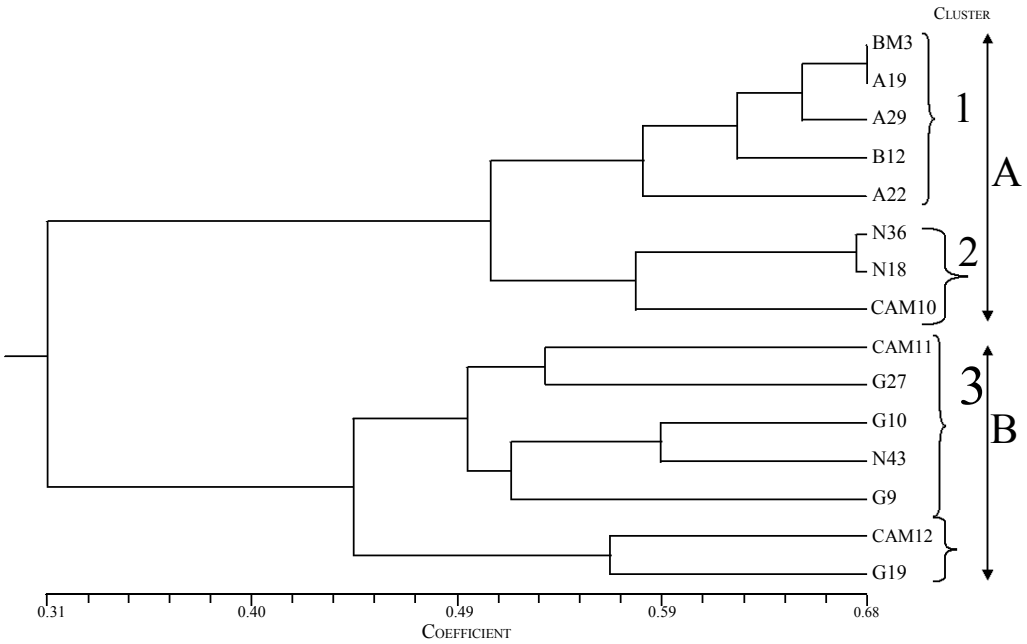

Figure 1. UPGMA-based dendrogram of genetic relationships of 15 accessions of *Irvingia gabonensis* using 384 AFLP polymorphic markers.

Table 2. Genetic distance matrix of *Irvingia gabonensis* accessions.

	BM3	A29	A19	W36	A22	CAM10	CAM11	CAM12	G27	N18	B12	G19	G10	G9
A29	33.3													
A19	32.3	37.1												
W36	51.9	51.4	59.2											
A22	39.7	43.7	46.6	41.7										
CAM10	42.5	45.5	48.3	40.7	48.8									
CAM11	70	73.6	62.5	87.6	84.3	76.1								
CAM12	46.9	50.7	43.6	64.6	60.3	52.5	55.8							
G27	65.8	71	62.4	83.8	81	68.8	46.6	55.3						
N18	50.8	50.2	58.7	32.8	44.9	44.4	88.1	62.9	82.2					
B12	32.9	41.5	39.8	47.2	39	44.1	76.1	52.1	72.7	50.2				
G19	49.8	54.4	42.1	67.1	64.4	55.7	56.8	43.7	52.4	67.2	55.2			
G10	66	70.2	59.7	83.6	80.5	69	47.8	54.8	47.4	81.6	73.2	53.2		
G9	63.4	66.9	62.3	80.6	77.6	69	51.5	52.8	50.3	77.7	68	52.9	45.2	
N43	69	71.1	65	84.8	83.5	73.1	53.3	58.8	50.3	83.3	74.6	58.1	41.4	51

Gabon showed a narrower genetic diversity with almost identical AGD of 51.6% (range, 32.3 - 84.8), and 50.2% (range, 45.2 - 53.2), respectively (Table 2). The PCO analysis data (Figure 2) showed that the Cameroun accessions (CAM11 and CAM12) clustered more closely with Gabon accessions than with accessions from Nigeria. Overall, the clustering pattern of the genotypes in the PCO analysis corresponds with the dendrogram derived from UPGMA (Figures 1 and 2).

The AGD of 59% among the 15 studied accessions supports the observations of Lowe et al. (2000) that Cameroun, Nigeria and Gabon harbour significantly high

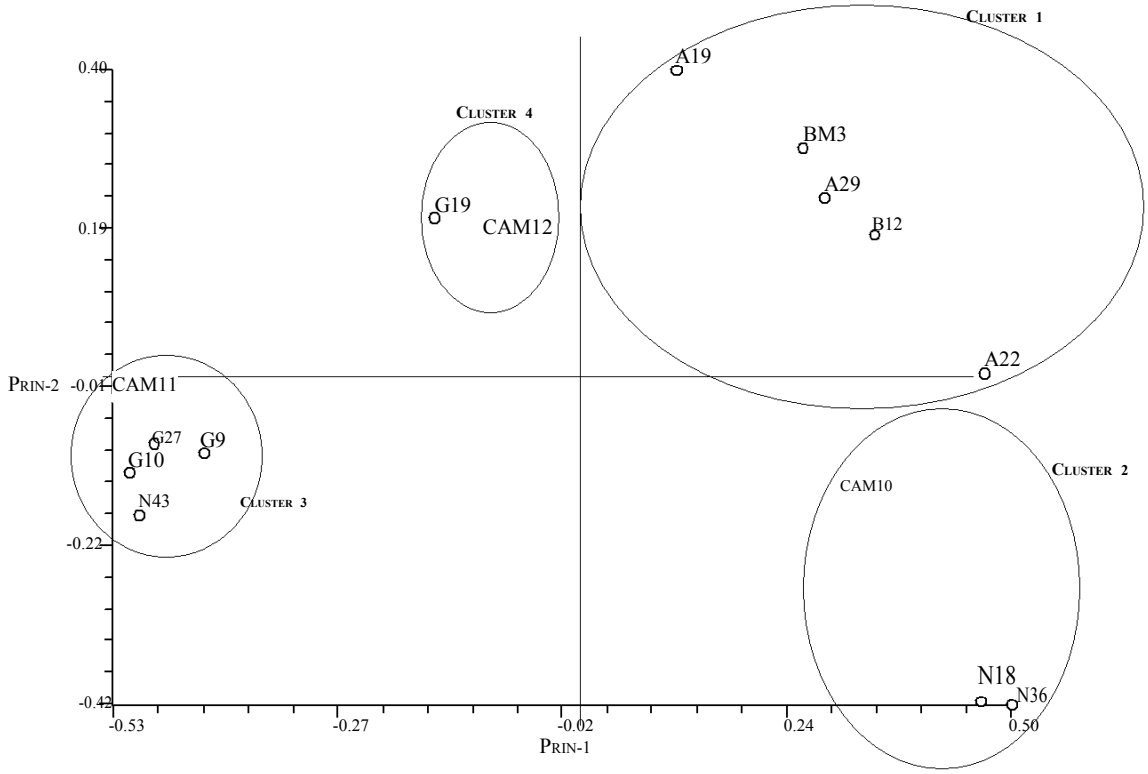

Figure 2. Principal Co-ordinate analysis of accessions of *Irvingia gabonensis* using 384 AFLP polymorphic markers.

level of genetic diversity of *I. gabonensis.*. However, the wider diversity among the Cameroun germplasm coupled with its overlap of Gabon and Nigeria accessions suggest Cameroun to be both the center of diversity of *I. gabonensis* and also the primary source of original materials grown in the other countries. Priority should be given to collection of germplasm from Cameroun in order to ensure optimum protection of the existing genetic diversity within *I. gabonensis.*

The agreement between the RAPD results of Lowe et al. (2000) and the AFLP data generated in this study suggests that AFLP has no advantage over RAPDs in identifying intraspecific variation within *I. gabonensis*. However, the smaller sample size of this study (15) compared to 130 accessions used by Lowe indicates that AFLP has more discriminatory power than RAPDs in analyzing genetic variation within *I. gabonensis*.

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