African Journal of Biotechnology Vol. 8 (12), pp. 2730-2739, 17 June, 2009
Available online at http://www.academicjournals.org/AJB
DOI: 10.5897/AJB09.184
ISSN 1684–5315 © 2009 Academic Journals

Full Length Research Paper

SSR markers reveal diversity in Guinea yam (*Dioscorea cayenensis*/*D. rotundata*) core set

J. E. Obidiegwu[1,2,3]*, M. Kolesnikova-Allen[4], E. E. Ene-obong[3], C. O. Muoneke[3] and R. Asiedu[1]

[1]International Institute of Tropical Agriculture, Carolyn House, 26 Dingwall Road, Croydon CR9 3EE, UK.
[2]National Root Crops Research Institute, Umudike, PMB 7006, Umuahia, Abia State, Nigeria.
[3]Department of Agronomy, Michael Okpara University of Agriculture, Umudike, Abia State, Nigeria.
[4]Biotechnology Unit, Tun Abdul Razak Research Centre Brickendonbury Hertford, UK SG13 8NL.

Accepted 23 February, 2009

The genetic diversity of 219 accessions of Guinea yam germplasm from Benin, Congo, Côte d' Ivoire, Equatorial Guinea, Gabon, Ghana, Nigeria, Sierra Leone and Togo was accessed using 15 microsatellite loci. High diversity of 0.677 was found among the accessions. An allelic average of 8.06 and polymorphic information content (PIC) value of 0.65 was observed for the markers. The observed heterozygosity value of 0.563 suggests that spontaneous hybridization must have contributed to the ancestry of some of the accessions and improvement by farmers must have been far more often by selection of somatic mutants. The twenty distinct cluster groups generated by the radial phylogram shows that *Dioscorea cayenensis* and *D. rotundata* are distinct species with intermediate hybrid forms. There was no relationship between relatedness of the accessions and their geographical area of origin. This study contributes to an increased understanding of the genetic organisation of the core germplasm.

Key words: Core germplasm, *Dioscorea cayenensis/D. rotundata*, genetic diversity, microsatellite.

INTRODUCTION

Dioscorea cayenensis and *D. rotundata* (also known as Guinea yams) are the most popular and economically important yams in West and Central Africa where they are indigenous and represent the largest depository of biodiversity, as a result of centuries of large domestication, production, trade and consumption (Degras, 1993). The name Guinea yam does not only signify their intensive cultivation and great importance in the socio-cultural life of the people of this region, but also suggests similarities in various aspects of their botany and agriculture (Akoroda and Chheda, 1983). The diversity in Guinea yam provides plant breeders with the necessary options to develop, through selection and breeding, new and more productive crops that are resistant to virulent pests and diseases, and adapted to changing environments. The International Institute of Tropical Agriculture (IITA) has established a core set of *Dioscorea* germplasm based on morphological descriptors (Mahalakshmi et al., 2007). This core set offers a good starting point when searching for new traits (Vaughan, 1991).

The extent of genetic diversity and relationship in the established Guinea yam core set in the IITA germplasm has not been investigated using DNA based markers. Molecular marker information can help monitor the level of genetic diversity in breeding materials and assist breeders to more efficiently choose genetically diverse parents for breeding scheme. Such diversity assessment could provide a means for identifying potential gaps in the species collection and further guiding target collecting missions. Molecular markers such as restriction fragment length polymorphism (RFLP), random amplified polymorphic DNA (RAPD), amplified fragment length polymorphism (AFLP) and simple sequence repeat (SSR) have been applied in yams for taxonomic, phylogenetic, diversity and mapping studies (Terauchi et al., 1992; Terauchi and Kanoma, 1994; Asemota et al., 1996; Ramser et al. 1996, 1997; Mignouna et al. 1998, 2002a,

*Corresponding author. E-mail: ejikeobi@yahoo.com. Tel.: +234-70-35029566.

b, 2003; Dansi et al., 2000; Egesi et al., 2006; Scarcelli et al., 2006; Tamiru et al., 2007; Tostain et al., 2007). In this study we used SSR markers to assess the diversity of the core collection of Guinea yams held in trust by IITA.

MATERIALS AND METHODS

Plant materials

The 219 accessions of Guinea yams collected from 10 countries of West and Central Africa are listed in Table 1. These are part of the core collection of *Dioscorea* germplasm held in trust by the IITA genebank. The accessions were planted in 30 cm size pots filled with sterilized loamy soil and maintained in a screen-house at the IITA, Ibadan Nigeria.

DNA extraction and quantification

Genomic DNA was extracted from fresh leaf apex of young leaves using modified CTAB procedure (Mignouna et al., 1998). The quality and concentration of DNA was assessed by gel electrophoresis using 1% agarose with known concentrations of undigested lambda DNA (Sigma, St. Louis, MO, USA). Quantification of DNA was done using a spectrophotometer (Beckman Coulter DU530) at 260 nm. Extracts were diluted in water to obtain DNA concentrations of 25 ng/µl.

Polymerase chain reaction and fragment analysis

A total of fifteen SSR primer pairs were used in the study (Table 2). PCR reaction was conducted in a 20 µl volume in a 96-well microtiter plate using an automated thermal cycler (model: Peltier Thermal Cycler 200). The reaction volume contained 25 ng of template DNA, 100 µM each of dNTP, 2.5 mM $MgCl_2$, 0.5 µM each of fluorescently labelled forward primer and unlabelled reverse primer, 1X reaction buffer and 2 units of Taq DNA polymerase (Invitrogen). The forward primer was 5´- labeled with one of the four fluorochromes PET, 6-FAM, NED and VIC. The PCR programme consisted denaturation at 94°C for 4 min, followed by 34 cycles of 94°C for 30 s, 51 or 58°C for 1 min and 72°C for 1 min, with a final extension step at 72°C for 7 min. Capillary electrophoresis with a semi-automated system ABI PRISM 3100 Genetic Analyser was used to separate amplified PCR products. Samples for amplified product separation were prepared by adding 1 µl of diluted PCR products to 9.4 µl formamide and 0.1 µl GenSize-500 LIZ. This was dispensed in ABI 96-well plates and were denatured at 94°C for 5 min and allowed to cool down on ice.

Data analyses

Observed allelic data were binned into discrete units and SSR fragment sizes were called using Genemapper v. 3.7 software (Figure 1). The fragment sizes in base pairs for each genotype across SSR markers were converted to binary data where alleles were transformed into presence (1) or absence (0) of an SSR band. Missing data accounted for less than 5% (that is, marker × genotype) of the entire data set. The genetic diversity parameters (Table 2) such as number of alleles per locus, percent of polymorphic loci, observed heterozygosity and gene diversity were estimated with FSTAT v. 2.9.3 software (Goudet, 2002). The tree structure (Figure 2) of the genetic diversity was constructed using DARwin 5.0 software.

RESULTS AND DISCUSSION

A total of 121 alleles were amplified with 15 SSR loci analyzed in 219 accessions, with the number of alleles observed per locus varying from 6 to 9 alleles (Table 2). The observed heterozygosity of 0.563 on average, varied from 0.276 (Dpr3F12) to 0.750 (Dab2D06). A total gene diversity of 0.677 was observed according to Nei diversity indices (Nei, 1973) for the accessions. Polymorphism was observed in all fifteen microsatellite loci analysed (Table 2). Polymorphic information content (PIC) ranged from 0.37 (Da1A01) to 0.80 (Dpr3D06). Average PIC value was 0.65. The UPGMA- derived radial phylogram constructed for the studied accessions provides an overview of the diversity structure (Figure 2) resulting into twenty distinct clusters groups. Accessions from different countries were fairly represented within each cluster.

In our study genetic diversity was detected in acessions with an average of 8.06 alleles per locus. Gene diversity of 0.677 on average was also found. The results demonstrate a genetic polymorphism in the studied germplasm from Benin, Congo, Côte d' Ivoire, Equatorial Guinea, Gabon, Ghana, Nigeria, Sierra Leone and Togo and high potential for genetic improvement. These findings suggest that morphological descriptors earlier used to develop this core set were discriminatory enough to capture as much diversity. Polymorphism was observed at all fifteen microsatellite loci analysed (Table 2). Polymorphic information content (PIC) ranged from 0.37 (Da1A01) to 0.80 (Dpr3D06) with an average value of 0.65. Tostain et al. (2007) also found SSR markers as discriminatory enough in diversity studies of yam. The 0.563 value for observed heterozygosity in this vegetative propagated crop is expected due to the fact that yams are dioceous and implies that spontaneous hybridization must have contributed to the ancestry of some of the accessions and improvement by farmers must have been far more often by selection of somatic mutants.

Accessions from different countries were grouped together in the thirteen clusters (Figure 2). There was no relationship between relatedness of the accessions and their geographical area of collection. This could be due to the fact that cultivars must have been distributed over great distances as clones in the course of human migration. *D. cayenensis* accessions clustered specifically in group 11 and 13 with overlapping mixtures with *D. rotundata* in group 10. This trend seems to uphold the view that Guinea yams are two distinct but related species and perhaps share a common secondary gene pool as proposed by Akoroda and Chheda (1983). Mignouna et al. (1998) showed that the varietal groups of *D. cayenensis* were genetically distant from those of *D. rotundata*. The overlapping *D. cayenensis* and *D. rotundata* in cluster group 10 could be regarded as hybrid derivatives as a result of natural hybridization. This view

Table 1. List and origin of the 219 accessions of Guinea yam (*D. cayenensis/D. rotundata*) analysed in the present study.

Genebank accession number	Local/cultivar name	Country of origin
TDr 3964	Tantoumani	Benin
TDr 3780	Noukpassi	Benin
TDr 3873	Bebeterou	Benin
TDr 3956	Gbera	Benin
TDr 3995	Gouroko	Benin
TDr 3886	Ouroutanai	Benin
TDr 1974	Be 110	Benin
TDr 3800	Unknown	Benin
TDr 3866	Kee	Benin
TDr 3953	Gomin	Benin
TDr 3854	Dourokonou	Benin
TDr 3994	Asnan	Benin
TDr1935	BE 116	Benin
TDr 3875	Soagona	Benin
TDr 4087	Unknown	Burkina Faso
TDr 3698	Mboru	Congo
TDr 2246	C.V 1784	Côte d'Ivoire
TDr 2159	C.V 204	Côte d'Ivoire
TDr 1888	IC 42	Côte d'Ivoire
TDr 2010	IC 35	Côte d'Ivoire
TDr 1983	IC 4	Côte d'Ivoire
TDr 1915	IC 16	Côte d'Ivoire
TDr 1913	IC 22	Côte d'Ivoire
TDr 1873	IC 11	Côte d'Ivoire
TDr 3496	C.V.1149	Côte d'Ivoire
TDr 1877	IC 23	Cote d Ivoire
TDr 2235	C.V 1746	Cote d Ivoire
TDr 2650	EQ-89 - 23	Equatorial Guinea
TDr 2656	EQ-89 - 23	Equatorial Guinea
TDr 3311	Pasandjo	Ghana
TDr 1907	GH 57	Ghana
TDr 2877	Kokoaseasobayere	Ghana
TDr 2008	GH 73	Ghana
TDr 3419	Butugu	Ghana
TDr 1966	Gh 66	Ghana
TDr 3402	Sakawa	Ghana
TDr 3426	Duobara	Ghana
TDr 2769	Kangba	Ghana
TDr 3323	SO/89/055	Ghana
TDr 3373	Aso Bayere	Ghana
TDr 3413	Nananto	Ghana
TDr 1986	GH 69	Ghana
TDr 1957	GH 72	Ghana
TDr 1892	GH 75	Ghana
TDr 3334	Jatiba	Ghana
TDr 4178	35 Dembatikou	Guinea
TDr 4159	60 Wakourouba	Guinea
TDr 4176	90 Waaman	Guinea
TDr 4181	61 Wakoula	Guinea

Table 1. contd.

TDr 4188	91 Ketikou	Guinea
TDr 2205	85/0016	Nigeria
TDr 2206	Gbongi	Nigeria
TDr 2766	Zania 87/0020	Nigeria
TDr 3683	P.Y.T. TDr 89/00613	Nigeria
TDr 4065	Unknown	Nigeria
TDr 2611	BN 218	Nigeria
TDr 3983	Asuikwu	Nigeria
TDr 2352	Verilly 93 (129)	Nigeria
TDr 2127	Kwiadu	Nigeria
TDr 2439	HVTD 87/0245	Nigeria
TDr 2374	Agatu	Nigeria
TDr 4072	Unknown	Nigeria
TDr 2079	Eochojah	Nigeria
TDr 3681	P.Y.T. TDr 89/01161	Nigeria
TDr 3312	A.Y.T. II TDr 87/00203	Nigeria
TDr 4156	Ayin	Nigeria
TDr 3578	P.Y.T. TDr 89/00183	Nigeria
TDr 2228	Sagbe egbor	Nigeria
TDr 2077	PYT (1986) 85/00410	Nigeria
TDr 2256	614	Nigeria
TDr 2613	Ht 86/0178	Nigeria
TDr 2435	Kaba-ex-arigadi 87/0123	Nigeria
TDr 3669	P.Y.T. TDr 89/00518	Nigeria
TDr 2424	I.Y.T 1986.26	Nigeria
TDr 3631	P.Y.T. TDr 89/01442	Nigeria
TDr 2347	R.10-102-21	Nigeria
TDr 2427	Dokara	Nigeria
TDr 2724	87/0/66 PYT 1986	Nigeria
TDr 2251	Iroko	Nigeria
TDr 2121	IYT 388	Nigeria
TDr 2084	PYT 121	Nigeria
TDr 2312	Ede ojah	Nigeria
TDr 2694	86/0094	Nigeria
TDr 2144	Unknown	Nigeria
TDr 2107	IYT 470	Nigeria
TDr 2217	HT-188	Nigeria
TDr 2126	Zmabor	Nigeria
TDr 2050	152(85)0109	Nigeria
TDr 2155	Okunmodu	Nigeria
TDr 2480	U.M.Y.P./8403	Nigeria
TDr 2031	PYT (1986)89/019	Nigeria
TDr 2178	Lasinrin	Nigeria
TDr 2049	PYT (1986)85-006	Nigeria
TDr 3675	P.Y.T. TDr 89/00765	Nigeria
TDr 3673	P.Y.T. TDr 89/01105	Nigeria
TDr 2083	Sotinrin alapa	Nigeria
TDr 2332	87/0066/29	Nigeria
TDr 2681	Alebu	Nigeria
TDr 3682	P.Y.T. TDr 89/00101	Nigeria
TDr 3654	P.Y.T. TDr 89/00572	Nigeria
TDr 3658	P.Y.T. TDr 89/00779	Nigeria

Table 1. contd.

TDr 2060	Agunmoka	Nigeria
TDr 2103	PYT-531	Nigeria
TDr 2119	87/0175	Nigeria
TDr 2205	85/0016	Nigeria
TDr 2276	Unknown	Nigeria
TDr 2386	86/00094	Nigeria
TDr 2434	HRTD 6	Nigeria
TDr 2455	ODO 87/0088	Nigeria
TDr 2557	Uregbe 87/0147	Nigeria
TDr 2752	Rosm 8458	Nigeria
TDr 3001	Dan anacha	Nigeria
TDr 3303	A.Y.T II 87/00102	Nigeria
TDr 3589	P.Y.T 89/00034	Nigeria
TDr 3603	P.Y.T 89/00790	Nigeria
TDr 3857	Amara	Nigeria
TDr 2259	IYT Bulk 158 (1986)	Nigeria
TDr 2077	PYT 85/00410	Nigeria
TDr 2177	Orin 338	Nigeria
TDr 1946	466	Unknown
TDr 4166	Hekpebulie	Sierra Leone
TDr 4187	Ngaobue	Sierra Leone
TDr 1529	Kjatiba KN 46	Togo
TDr 1787	Koukou (0-5)	Togo
TDr 2537	AOO-011	Togo
TDr 1706	Kratsi A22	Togo
TDr 1762	Kpena KN 35	Togo
TDr 2923	Kpayere KN-50 1280	Togo
TDr 1862	Alassorakoukou	Togo
TDr 1714	Aroukpe	Togo
TDr 3546	Tchabisot-125 585	Togo
TDr 1637	Kalamofo BL9	Togo
TDr 1521	Abononojawite	Togo
TDr 2992	Tila 2B-52 - 1034	Togo
TDr 1808	Alassora T-84	Togo
TDr 1709	Kpandov 2R 13	Togo
TDr 1747	Lili 2B9	Togo
TDr 1847	Naka BH 49	Togo
TDr 1577	Gnidou 25	Togo
TDr 3437	Tinougdjat 1350	Togo
TDr 2903	Tifiou A-83-969	Togo
TDr 2023	29	Togo
TDr 2910	Sousou KN-98 1128	Togo
TDr 3471	Atan-A-44 - 885	Togo
TDr 2774	Tila 2B 52	Togo
TDr 1591	Craysi	Togo
TDr 2949	Kplindjo IIB-78-1061	Togo
TDr 3508	Kplingjo IIB-63 - 1045	Togo
TDr 1721	Sakoro S-75	Togo
TDr 1753	AOO 589 B	Togo
TDr 1615	Lotossou 63	Togo
TDr 1492	Duo Wetanom	Togo
TDr 3517	Djigbri A-59 -948	Togo

Table 1. contd.

TDr 2489	PDM 110	Togo
TDr 1483	Oko-fasse	Togo
TDr 1611	Toukla	Togo
TDr 2914	Alassora 606	Togo
TDr 1574	Keke KN 42	Togo
TDr 1491	Edja	Togo
TDr 2492	BN 47	Togo
TDr 1612	Kpagnina A87	Togo
TDr 1849	Bakpanatene T-98	Togo
TDr 1733	Kotokolsot 113	Togo
TDr 1622	Kpofo 146	Togo
TDr 2534	33	Togo
TDr 1860	Datala 820	Togo
TDr 1261	Keressi 246	Togo
TDr 1519	Laboco KN 59	Togo
TDr 2927	Kplindjo 2B-78 1060	Togo
TDr 3459	Kratsi 208	Togo
TDr 2471	1538-18	Togo
TDr 2969	Boraha BH-48 - 1141	Togo
TDr 1730	Bakou A34	Togo
TDr 2527	BN 340	Togo
TDr 1688	BN 307	Togo
TDr 1501	Kapaza A 52	Togo
TDr 1998	1021	Togo
TDr 3489	Kanitiki D-3 - 1150	Togo
TDr 3009	Kratsi BL-23 – 483	Togo
TDr 1842	Fulakde KN 63	Togo
TDr 1798	Lamlau 2B C2	Togo
TDr 1756	Abafo He D-48	Togo
TDr 1739	Lou B-7	Togo
TDr 1737	Fasse-He-Bl 35	Togo
TDr 1699	Aridji S-74	Togo
TDr 1673	Korokoro 2B 12	Togo
TDr 1565	Adoworo BH 49	Togo
TDr 1512	Keke T113	Togo
TDr 1478	Foulande KN 75	Togo
TDr 2234	Nwana III 337	Togo
TDr 2493	BN 337	Togo
TDr 2547	S-Tdr	Togo
TDr 2704	Larboko 1039 IIB 56	Togo
TDr 2776	Tombre 634	Togo
TDr 2961	Keressi 246	Togo
TDr 2986	Tsiboto 97	Togo
TDr 3473	Kratsi 338	Togo
TDr 3481	Yabayi BH-15-1108	Togo
TDr 3493	Sarmata 2B-4-1028	Togo
TDr 3506	Djatouba 1400	Togo
TDr 3513	Bakou 875	Togo
TDr 3516	Bayere KN-44-1274	Togo
TDr 1855	Kpadjoli B-14	Togo
TDr 1738	Afi Ratwa KN-22	Togo
TDc 3967	N'keni	Benin

Table 1. contd.

TDc 2809	BE 109	Benin
TDc 3704	Unknown	Congo
TDc 3712	Unkown	Congo
TDc 2817	C.V. 1730	Cote d Ivoire
TDc 2794	IC 14	Cote d Ivoire
TDc 2831	Obou bi kwae	Ghana
TDc 2828	Sebor	Ghana
TDc 3840	Lasinrin	Nigeria
TDc 3807	Yellow	Nigeria
TDc 2815	HVTD 2/12/89	Nigeria
TDc 2811	BO 6	Togo
TDc 2790	282	Togo
TDc 2798	Okuaoo 1389	Togo
TDc 3839	Iganganran	Uknown

Table 2. Primer sequences (forward/reserve) used in the SSR analyses and their respective size annealing temperature (*Ta*), number of alleles per locus (*A*), observed heterozygosity (*Hobs*) and polymorphic information content (PIC).

Microsatellite name	5' to 3' Primer sequence	T_a (ºC)	*A*	*Hobs*	PIC
Da1F08	AATGCTTCGTAATCCAAC -F CTATAAGGAATTGGTGCC -R	51	6	0.532	0.64
Dab2C05	CCCATGCTTGTAGTTGT -F TGCTCACCTCTTTACTTG -R	51	9	0.520	0.63
Dab2D06	TGTAAGATGCCCACATT -F TCTCAGGCTTCAGGG -R	51	9	0.770	0.67
Dab2D08	ACAAGAGAACCGACATAGT -F GATTTGCTTTGAGTCCTT -R	51	8	0.602	0.74
Dab2E07	TTGAACCTTGACTTTGGT -F GAGTTCCTGTCCTTGGT -R	51	9	0.746	0.77
Dab2E09	AACATATAAAGAGAGATCA -F ATAACCCTTAACTCCA -R	51	8	0.496	0.66
Dpr3B12	CATCAATCTTTCTCTGCTT- F CCATCACACAATCCATC -R	51	9	0.647	0.62
Dpr3D06	ATAGGAAGGCAATCAGG -F ACCCATCGTCTTACCC -R	51	9	0.714	0.80
Dpr3F12	TCCCCATAGAAACAAAGT -F TCAAGCAAGAGAAGGTG -R	51	9	0.239	0.59
Dpr3F04	AGACTCTTGCTCATGT -F GCCTTGTTACTTTATTC -R	51	6	0.526	0.66
Da1A01	TATAATCGGCCAGAGG - F TGTTGGAAGCATAGAGAA -R	51	7	0.315	0.37
YM5	AATGAAGAAACGGGTGAGGAAGT -F CAGCCCAGTAGTTAGCCCATCT -R	58	6	0.765	0.67
YM13	TTCCCTAATTGTTCCTCTTGTTG -F GTCCTCGTTTTCCCTCTGTGT -R	58	3	0.398	0.57
YM15	TACGGCCTCACTCCAAACACTA -F AAAATGGCCACGTCTAATCCTA -R	58	7	0.448	0.70
YM26	AATTCGTGACATCGGTTTCTCC -F ACTCCCTGCCCACTCTGCT -R	58	9	0.724	0.61

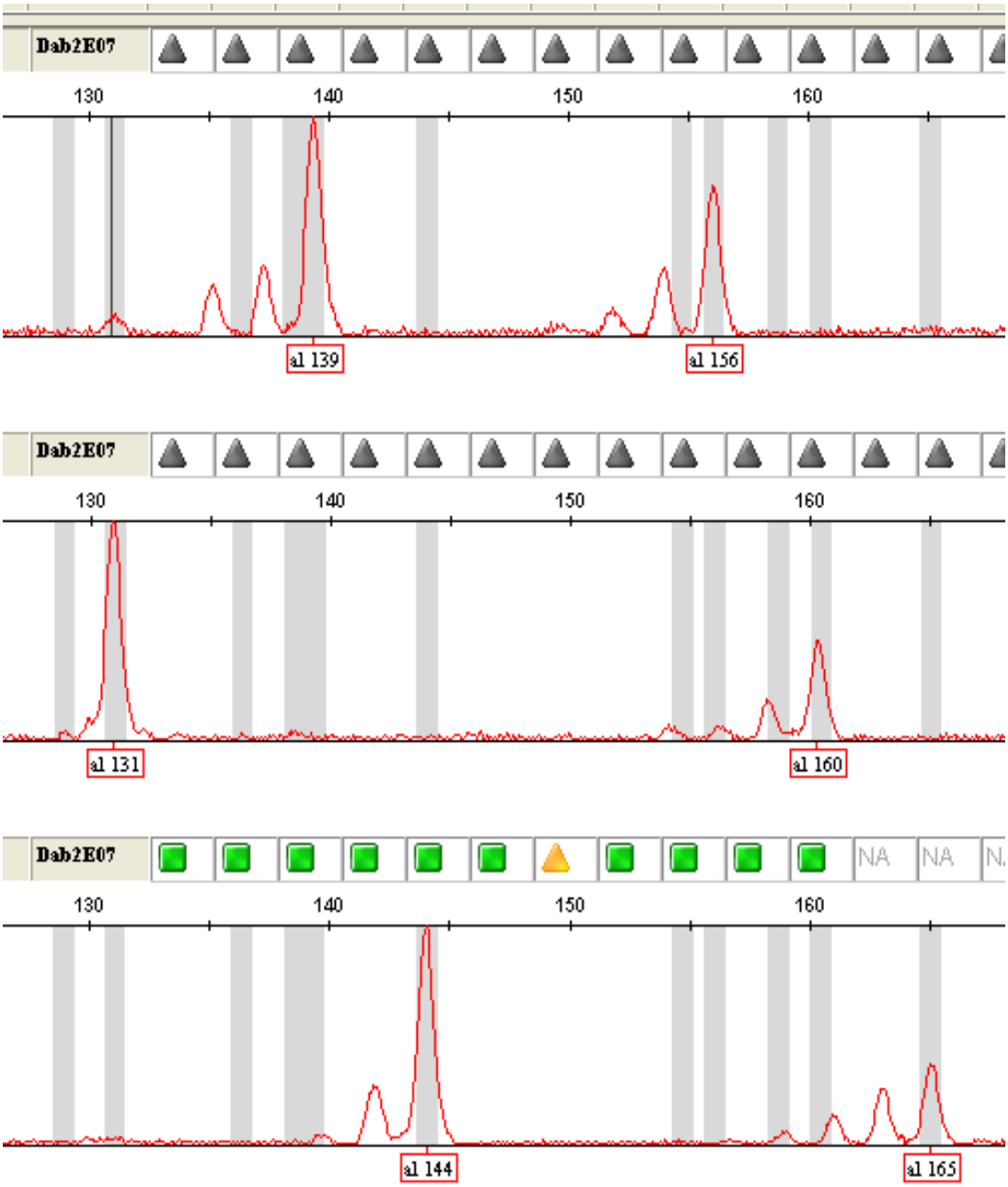

Figure 1. Examples of SSR profiles obtained for *3* yam accessions with marker *Dab2E07* using analysis software GeneMapper v. 3.7 (Applied Biosystems, USA).

is in agreement with Hamon and Touré (1990) who recognized some intermediate accessions. Taxonomic relationship between *D. cayenensis* and *D. rotundata* (Guinea yams) has often been a subject of controversy and speculation. The existences of numerous vernacular names specifying a given cultivar (Dansi et al., 1999; Mignouna et al., 1998), and the existence of many intermediate forms (Akoroda and Chheda, 1983; Hamon and Touré, 1990; Mignouna et al., 2002c), have complicated their exact classification.

Our study contributes to an increased knowledge of the taxonomic classification and genetic arrangement of core set of Guinea yams.

ACKNOWLEDGEMENTS

This study would not have been possible without the support of Generation Challenge Program (GCP) in Project Number 3*d* of sub-programme 1. The technical assistance of Athanson Blessing, Hammed Adeola and Olatunji Aina of the Central Biotechnology Laboratory (CBL),

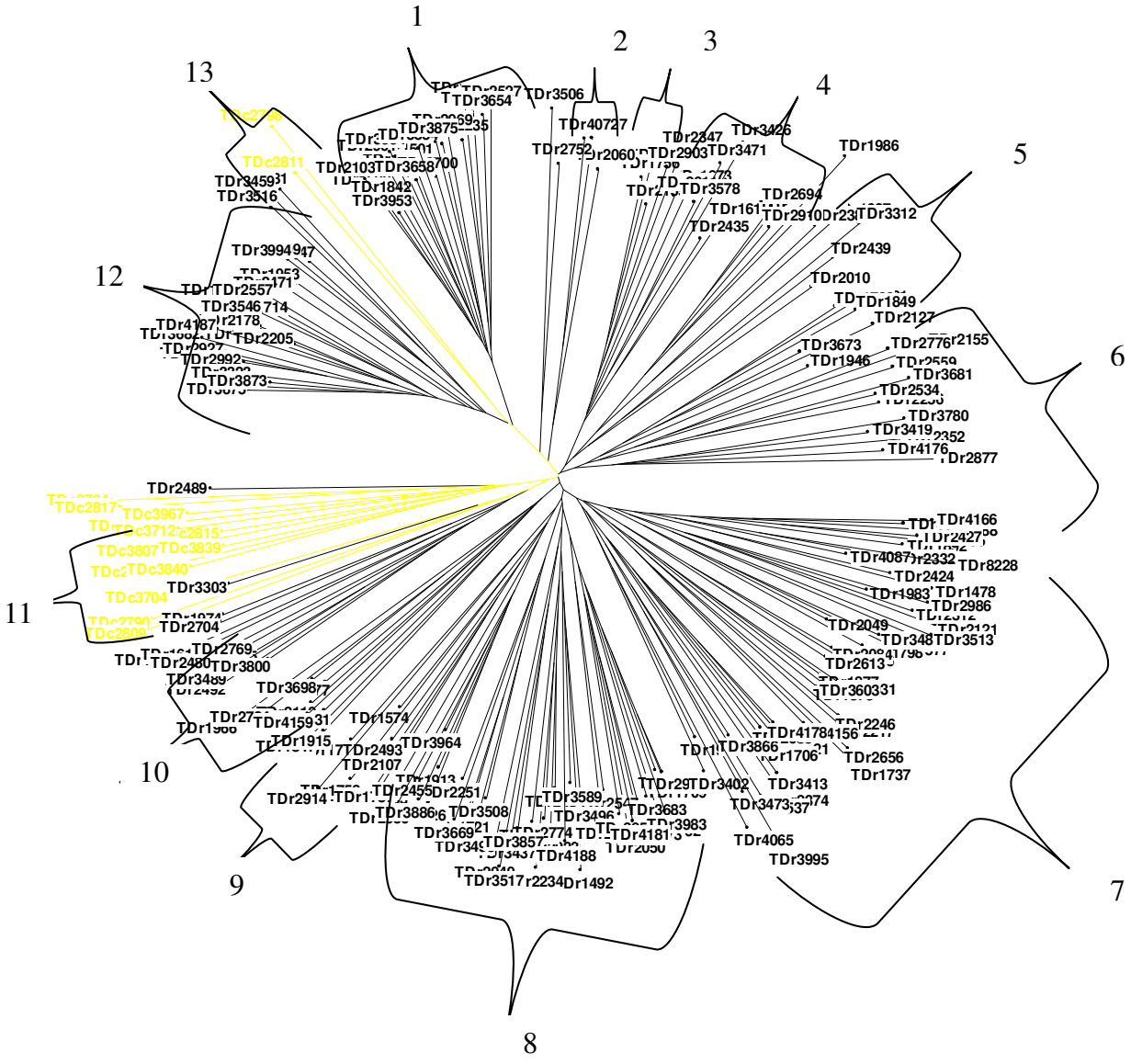

Figure 2. Radial phylogram of 219 accessions of Guinea yam based on Outweighed Neighbour joining cluster analysis.

International Institute of Tropical Agriculture (IITA), Ibadan is acknowledged.

REFERENCES

Akoroda MO, Chheda HR, (1983). Agro-botanical and species relationships of Guinea yams. Trop. Agric. (Trinidad)60: 242-248.

Asemota HN, Ramser J, Lope´z-Peralta C, Weising K, Kahl G (1996). Genetic variation and cultivar identification of Jamaican yam germplasm by random amplified polymorphic DNA analysis. Euphytica, 92: 341-351.

Dansi A, Mignouna HD, Zoundjihekpon J, Sangare A, Asiedu R, Quin FM (1999). Morphological diversity, cultivar groups and possible descent in cultivated yams (*Dioscorea cayenensis-Dioscorea rotundata* complex) of Benin Republic. Genet. Resour. Crop Evol. 46: 371-388

Dansi A, Mignouna HD, Zoundjihe´kpon J, Sangare A, Ahoussou A, Asiedu R (2000). Identification of some Benin Republic's Guinea yam (*Dioscorea cayenensis/Dioscorea rotundata* complex) cultivars using Randomly Amplified Polymorphic DNA. Genet. Resour. Crop Evol. 47: 619-625.

Degras L (1993). The Yam: a tropical root crop. MacMillan Press Ltd, London, UK

Egesi CN, Asiedu R, Ude G, Ogunyemi S, Egunjobi JK (2006). AFLP marker diversity in water yam (Dioscorea alata L.). Plant Genet. Resour. 4(3): 181-187

Goudet J (2002). Fstat vol. 2.9.3. Lausanne: Institute of Ecology.

Hamon P, Touré B (1990). The classification of the cultivated yams (*Dioscorea-cayenensis-rotundata* complex) of West Africa. Euphytica, 47(3): 179-187.

Mahalakshmi V, Ng N, Obidiegwu J, Ogunsola D, Lawson M, Ortiz R (2007). Development of West African yam *Dioscorea* spp. core collection. Genet. Resour. Crop Evol. (in press, doi: 10.1007/s10722-007-9236-3).

Mignouna HD, Ellis NTH, Knox MR, Asiedu R, Ng QN (1998). Analysis of genetic diversity in Guinea yams (*Dioscorea* spp.) using AFLP fingerprinting. Trop. Agric., (*Trinidad)* 75: 224-229.

Mignouna HD, Mank RA, Ellis THN, Van den Bosh N, Asiedu R, Ng

SYC, Peleman J (2002a). A genetic linkage map of Guinea yam (*Dioscorea rotundata* Poir.) based on AFLP markers. Theor. Appl. Genet. 105: 716-725.

Mignouna HD, Mank RA, Ellis NTH, van den Bosch N, Asiedu R, Abang MM, Peleman J (2002b). A genetic linkage map of water yam (*Dioscorea alata* L.) based on AFLP markers and QTL analysis for anthracnose resistance. Theor. Appl. Genet. 105: 726-735.

Mignouna HD, Dansi A, Zok S (2002c). Morphological and isozymic diversity of the cultivated yams (*Dioscorea cayenensis*/*Dioscorea rotundata* complex) of Cameroon. Genet. Resour. Crop Evol. 49: 21-29.

Mignouna HD, Abang MM, Fagbemi SA (2003). A comparative assessment of molecular marker assays (AFLP, RAPD and SSR) for white yam (*Dioscorea rotundata)* germplasm characterisation. Ann. Appl. Biol. 142: 269-276.

Nei M (1973). Analysis of gene diversity in subdivided populations. Proceed. National Acad. Sci. USA, 70: 3321-3323.

Ramser J, Lope´z-Peralta C, Wetzel R, Weising K, Kahl G (1996). Genomic variation and relationships in aerial yam (*Dioscorea bulbifera* L.) detected by random amplified polymorphic DNA. Genome, 39: 17-25.

Ramser J, Weising K, Lope´z-Peralta C, Terhalle W, Terauchi R, Kahl G (1997). Molecular marker based taxonomy and phylogeny of Guinea yam (Dioscorea rotundata-D. cayenensis). Genome, 40: 903-915.

Scarcelli N, Tostain S, Mariac C, Agbangla C, Da O, Berthaud J, Pham J (2006). Genetic nature of yams (*Dioscorea* spp.) domesticated by farmers in Benin (West Africa). Genet. Resour. Crop Evol. 53: 121-130

Tamiru M, Becker HC, Maass BL (2007). Genetic diversity in yam germplasm from Ethopia and their relatedness to the main cultivated *Dioscorea* species assessed by AFLP markers. Crop Sci. 47: 1744-1753.

Terauchi R, Konuma A (1994). Microsatellite polymorphism in *Dioscorea tokoro*, a wild yam species. Genome, 37: 794-801.

Terauchi R, Chikaleke VA, Thottapily G, Hahn SK (1992). Origin and phylogeny of Guinea yams as revealed by RFLP analysis of chloroplast DNA and nuclear ribosomal DNA. Theor. Appl. Genet. 83: 743-751.

Tostain S, Agbangla C, Scarcelli N, Mariac C, Daınou O, Berthaud J, Pham JL (2007). Genetic diversity analysis of yam cultivars (*Dioscorea rotundata* Poir.) in Benin using simple sequence repeat (SSR) markers. Plant Genetic Resources: Characterization and Utilization 5(2): 71-81

Vaughan DA (1991). Choosing rice germplasm for evaluation. Euphytica, 54: 147-154.