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Diversity analysis of sweet potato (*Ipomoea batatas* [L.] Lam) germplasm from Burkina Faso using morphological and simple sequence repeats markers

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Collecting and characterizing plant material has been basic for crop improvement, and diversity has long been seen as vital for rational management and use of crops. Thirty (30) morphological characters and thirty (30) simple sequence repeat (SSR) markers were used to assess the diversity among 112 sweet potato (*Ipomoea batatas* [L.] Lam) cultivars in Burkina Faso and to develop a core collection. Eight morphological characters were able to differentiate the 112 accessions and to identify 11 duplicates while 28 SSR markers were more informative in discriminating the accessions and to identify five duplicates. The diversity assessment using the two approaches revealed high diversity with a coefficient of 0.73 using the phenotypic data, while moderate diversity with a coefficient of 0.49 was obtained using the SSR markers. These results show no correlation between the two approaches (with dissimilarity index of 0.95). A core collection was constituted using the SSR based data while the eight discriminative phenotypic descriptors will be used in the identification of cultivars.

Key words: Accessions, genetic diversity, germplasm, molecular markers, morphological characters, simple sequence repeat, sweet potato.

INTRODUCTION

Sweet potato (*Ipomoea batatas* [L.] Lam), a hexaploid crop (2n = 6X = 90) is one of the most economically important crops in the world. In Burkina Faso, the major production areas are near the borders with Mali, Ghana,

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Abbreviations: SSR, Simple sequence repeat; **PCR,** polymerase chain reaction; **PIC,** polymorphic information content; **PT,** plant type; **GC,** ground cover; **VID,** vine internode diameter; **VIL,** vine internode length; **PVC,** predominant vine colour; **SVC,** secondary vine colour; **VTP,** vine tip pubescence; **GOL,** general outline of leaf; **LLN,** leaf lobes number; **LLT,** leaf lobes type; **MLS,** mature leaf size; **ALVP,** abaxial leaf vein pigmentation; **PL,** petiole length; **PP,** petiole pigmentation; **SCLL,** shape of central leaf lobe; **MLC,** mature leaf colour ; **ILC,** immature leaf colour; **FH,** flowering habit; **PSC,** predominant skin colour; **IPSC,** intensity of predominant skin colour; **SSC,** secondary skin colour; **PFC,** predominant flesh colour; **SFC,** secondary flesh colour; **DSFC,** distribution of secondary flesh colour; **SRF,** storage root formation; **SRS,** storage root shape; **LPSR,** latex production in storage roots; **OSR,** oxidation in storage roots; **SRSD,** storage root surface defects; **SRCT,** storage root cortex thickness; **UPGMA,** unweighted pair group method using arithmetic average.

Togo and Benin suggesting that important exchanges of planting material has occurred between these neighbouring countries. Cultivar names differ from one location to another, therefore placing limitations on accurate identification on locally available sweet potato germplasm that is vital to the rational management and use of the crop. Collection, characterization and maintenance of local germplasm are the bases of varietal improvement (Mok and Schmiediche, 1998).

Morphological characterization has been used extensively on various crop plants diversity assessments in many places of the world (Bos et al., 2000; Kaplan, 2001; Lacroix et al., 2005; Li et al., 2009; K'Opondo, 2011). Despite the environmental influences on plant morphology, this direct inexpensive and easy to use method of estimations was perceived as the strongest determinant of the agronomic value and taxonomic classification of plants (Li et al., 2009) and the first step in the assessment of plant diversity. On sweet potato, this tool has been used successfully to analyse genetic diversity necessary for the germplasm conservation, to reduce accession number by identification and elimination of duplicates and to enhance crop breeding (Huaman, 1992; Mok and Schmiediche, 1998; Tairo et al., 2008; Li et al., 2009; Karuri et al., 2009; Yada et al., 2010a).

According to La Bonte (2002), when trait expression is environmentally unstable or difficult to evaluate, molecular markers become more useful than traditional phenotypic evaluations. During the last decade a lot of molecular information has been accumulated and used for genetic diversity assessment on sweet potato germplasm (Jarret et al., 1992; Kowyama et al., 1992; Jarret and Austin, 1994; Bruckner, 2004; Tseng et al., 2002; Hu et al., 2003; He et al., 2006; He et al., 2007; Soegianto et al., 2011). The most widely used molecular marker procedures for population genetic analysis of both animals and plants during the past few years are the simple sequence repeat (SSR) markers or microsatellites (Shih et al., 2002; Veasey et al., 2008; Zhang et al, 2001; Karuri et al., 2010; Yada et al, 2010b; Li et al., 2009) (Weising et al., 1995). These markers are highly polymorphic, co-dominant, and can easily be detected on high-resolution gels.

Limited success has been achieved with morphological diversity analysis alone (Yada et al., 2010a). Therefore, to optimize the characterization efficiency, morphological characterization has now been combined with molecular techniques. SSR markers have been used in combination with morphological descriptors to analyse genetic diversity in sweet potato germplasm and useful core collections have been developed using this combination (Li et al., 2009; Karuri et al., 2010).

The objective of this research was to quantify the diversity in sweet potato germplasm collected in Burkina Faso using morphological descriptors and SSR molecular markers.

MATERIALS AND METHODS

Collection of plant materials

One hundred and forty-four (140) sweet potato accessions (Table 1) were collected from December 2008 to January 2009 and January 2010 from the main production areas located in the Cascades, Western, Central-West, Southern, Central-South, Central-East and Eastern regions of Burkina Faso using the method described by Huaman (1991). One hundred and seven (107) accessions survived and were maintained at the INERA research station of Kamboinse located in the centre of the country in the Soudanian zone characterized by an annual rainfall ranged from 600 to 1100 mm. Three varieties introduced from the International Potato Center (CIP) East Africa CIP-440001 (known as Resisto), CIP-199062-1 and TIB-440060, one from China (TN-Leo) and Tiebele-2 an orange fleshed sweet potato of unknown origin were added and used as control.

Morphological characterization

The experiment

The 112 accessions were grown at the INERA station of Kamboinsé during the rainy season, from July to October 2009. Based on the records of the first year, the experiment was replicated from July to October 2010 and the materials were planted in groups of relatedness to allow further morphological comparisons between those accessions which were morphologically alike. Planting was done on ridges of 3 m long with distance between ridges of 1 m. On each ridge, 11 cuttings were planted at a spacing of 30 cm. The fields were maintained by frequent weeding. NPK (14-23-14) fertilizer was applied 21 days after planting when the cuttings were well established. Additional watering was done by irrigation to complement rainfall.

Data collection

Morphological data were collected 60 days after planting based on the average of three measurements from the middle portion of the main stem as recommended by Huaman (1992). Qualitative characters were scored using a scale of 0 to 9. The following variables were scored: Plant growth characteristics: plant type (PT), ground cover (GC); mature vine characteristics: vine internode diameter (VID), vine internode length (VIL), predominant vine colour (PVC), secondary vine colour (SVC), vine tip pubescence (VTP); mature leaf characteristics: general outline of leaf (GOL), leaf lobes number (LLN), leaf lobes type (LLT), mature leaf size (MLS), abaxial leaf vein pigmentation (ALVP), petiole length (PL), petiole pigmentation (PP), shape of central leaf lobe (SCLL), mature leaf colour (MLC), immature leaf colour (ILC); flowering habit (FH); Storage root characteristics: predominant skin colour (PSC), intensity of predominant skin colour (IPSC), secondary skin colour (SSC), predominant flesh colour (PFC), secondary flesh colour (SFC), distribution of secondary flesh colour (DSFC), storage root formation (SRF), storage root shape (SRS), latex production in storage roots (LPSR), oxidation in storage roots (OSR), storage root surface defects (SRSD), storage root cortex thickness (SRCT). Measurements were done on three plants chosen randomly from the 11 plants per plot and averaged for the variable.

Data analysis

The computer program Genstat 14th edition was used to analyse the morphological data. Stepwise discriminant analysis was

Table 1. List of accessions collected in Burkina Fasoand the varieties introduced used for the characterisation.

Code	Name	Site	Number	Code	Name	Ssite	Number	Code	Name	Site
BF1	Unknown	Koubri	38	BF49	Dagouam	Mantiagogo	75	BF93	Massakoun-Gbeman	Beregadougou
BF2	Unknown	Koubri	39	BF51	Bagre	Tiebele/Tigalo	76	BF94	Unknown	Banfora
BF3	Unknown	Koubri	40	BF52	Unknown	Garango	77	BF95	Wosso-Gbe 2	Sourou
BF4	NangnouNoondo	Koubri	41	BF53	Unknown	Garango	78	BF97	Diabo Local	Diabo
BF7	Unknown	Koubri	42	BF54	Unknown	Garango	79	BF98	Garango	Diabo
BF8	Unknown	Koubri	43	BF55	Unknown	Garango	80	BF99	Sawiyague	Lo-Longo
BF9	Gelwango	Tingandgo	44	BF56	Unknown	Garango	81	BF100	NalougourouNono	Tiebele
BF10	Tiébélé	Tingandgo	45	BF57	Unknown	Maoda	82	BF108	Bobo rouge	Reo
BF11	Patate	Tingandgo	46	BF58	Unknown	Maoda	83	BF112	ShiraJaa	Reo
BF12	Saafaré	Tingandgo	47	BF59	Nakalbo	Koupela	84	BF114	Dayejopouri	Goundi
BF13	Tiébélé	Tingandgo	48	BF60	Unknown	Koupela	85	BF115	Dayepoan	Goundi
BF14	Jaune 2	Kombissiri	49	BF61	Unknown	Koupela	86	BF116	Kokonetioulou	Poun
BF15	Patate	Kombissiri	50	BF62	Unknown	Maoda	87	BF117	Dayebioun	Poun
BF16	Bananbato	Kombissiri	51	BF63	Fandaga	Badara	88	BF119	Dayepouan	Poun
BF17	Saafaréblanc	Kombissiri	52	BF64	Wosso	Badara	89	BF120	Dayebioun	Poun
BF18	Saafaré rose	Kombissiri	53	BF65	Unknown	Badara	90	BF126	Zimien-botouhin	Mboa
BF19	Jaune 1	Kombissiri	54	BF66	Unknown	Badara	91	BF127	Zipo-kouka	Mboa
BF20	Nayiré	Yale	55	BF67	Unknown	Badara	92	BF128	Zipo-botouhin	Mboa
BF21	Nayiré	Yale	56	BF68	Unknown	Oradara	93	BF129	Zimien-kouka	Mboa
BF23	Nayi-mina	Sagalo	57	BF71	Denbaya	Oradara	94	BF130	Ziro-dodobo	Mboa
BF24	Nayir-vapapao	Sagalo	58	BF72	Fardan-wouleman	Oradara	95	BF131	Nagnou-pla	Komsaya
BF25	Nayir-sian	Sagalo	59	BF74	Wosso-Gbe	Sourou	96	BF132	Nagnou-ziè	Komsaya
BF27	Nayir-po	Leo	60	BF75	Djakani	Sourou	97	BF133	Unknown	CREAF
BF32	Kabakourou	Leo	61	BF77	Gambagre	Sikorla	98	BF135	Nankansongo	Lolongo
BF33	Nayir-papao	Sissili	62	BF78	Badara	Sikorla	99	BF136	Nankanpongo	Lolongo
BF34	Kabakourou	Sissili	63	BF80	Massako-fing	Sikorla	100	BF137	Iloropongo	Lolongo
BF35	Nayir-manan	Sissili	64	BF81	Massoko 2	Sikorla	101	BF138	Nayoumondo-1	Kombissiri
BF36	Nayir-mian	Sissili	65	BF82	Bagayogo	Sikorla	102	BF139	Nayournondo-2	Kombissiri
BF38	Unknown	Kombissiri	66	BF83	Massakoun-Gnin	Sitiena	103	BF140	Djacané	Sarkandiara
BF40	Unknown	Kombissiri	67	BF85	Massakoun 2	Sitiena	104	BF141	Sèguè-Bana	Sarkandiara
BF41	Unknown	Kombissiri	68	BF86	Massakoun-Plaa	Kiribina	105	BF142	Ouagnougui	Gonsin
BF42	Nankan-poupiou	Lo	69	BF87	Wosso-Gbe	Banfora	106	BF144	Unknown	Sikorla

Table 1. Contd.

BF43	Nankan-pongo	Lo	70	BF88	Fandaga-Woule	Banfora	107	BF145	Unknown	Ouagadougou
BF44	Nankan-soungo	Lo	71	BF89	Fandaga-Gbeman	Banfora	108	TN.LEO	TN.LEO	Introduced
BF45	BinagaNapouni	Mantiagogo	72	BF90	Wosso-Woule	Banfora	109	CIP-199062-1	CIP 199062-1	Introduced
BF46	Nanlougourou	Mantiagogo	73	BF91	Wosso-Woule	Banfora	110	TIB-440060	TIB	Introduced
BF47	Manga	Mantiagogo	74	BF92	Massakoun-Woule 2	Beregadougou	111	TIEBELE.2	TIEBELE.2	Tiebele/Tigalo
							112	CIP-440001	Resisto	Introduced

performed to select a subset of variables that best discriminate among the classes. The Wilks' Lamda criterion was used to measure the variable contribution to the discriminatory power of the model as described by Daulfrey (1976); least contribution leads to removal of the variable.

The significant level of retaining or adding a discriminative variable was 0.15. Subsequently, principal component analysis was applied to examine the structure of the correlations between variables. The null hypothesis that any r_{ij} was equal to zero was tested by computing the ratio of the explained variance to the unexplained variance. The eigenvalues and eigenvectors of the correlation matrix were derived, and the eigenvectors scaled by the square root of the corresponding eigenvalues to produce the matrix of component loadings. The eigenvalues and their associated eigenvectors, the correlation matrix are used to reduce the number of variables in the statistical analyses (Daulfrey, 1976).

A graphical display of the genetic relationships was also computed by principal coordinate analysis using the Rogers Tanimoto dissimilarity index of DARwin5.0.158 software. Cluster analyses were performed to group observations together using the method of Euclidian distance. Data points with the smaller distances between them were grouped together. A dendrogram was plotted from these computed clusters as a graphical relationship among accessions. From the dendrogram duplicates, samples were identified as a result of complete similarity between accessions.

Molecular characterization

Leaf sampling procedure

Leaf sampling was done as recommend by the DNALandmarks, a Canadian biotechnology laboratory, where the molecular work was done. Using 96-wells blocks, two leaf discs of 5 mm diameter were harvested from young leaves of each accession using a whole paper punch and put into a specific well position.The block was then placed inside a plastic bag with 50 g of silica gel and kept for 24 h to dry.

DNA extraction and SSR amplification

DNA extraction and amplification were done using an internal protocol at DNALandmarks laboratory in Canada. After extraction, the quality of the DNA was tested on 1% agarose gel. The DNA samples were then diluted to 25 ng/ul. The diluted DNA samples were then used for polymerase chain reaction (PCR) amplification with 30 SSR markers which sequences were provided by the International Potato Center (Table 2). PCR reactions were performed following an internal protocol of DNALandmarks with minor modifications (Ghislain et al., 2009). Forward primers were tailed with a M13 primer and the M13 primer (CACGACGTTGTAAAACGAC) labelled with one of the four fluorescence dyes (6FAM, PET, NED or VIC) for multiplexed PCR products detection using the ABI3730xl apparatus. The PCR conditions consisted of an initial denaturation at 95°C for 15 min, annealing at 60°C for 1 min and 72°C followed by 35 cycles of 94°C for 1 min, annealing at 60°C for 1 min, and 72°C for 1 min. This was followed by a final extension step of 20 min at 72°C and a halt at 4°C. The allele sizes were scored using GeneMapper software. Multiple peaks were detected due to the polyploidy nature of sweet potato. Any peak with the peak height greater than one sixth of the highest peak was scored. Allele size was calculated by subtracting 19 (M13 primer length) from the peak size. The raw data were provided for the further analysis. Failed samples were repeated one to two times.

Data analysis

The polymorphic information content (PIC) that is the importance of each SSR marker in distinguishing between accessions was determined (Weir, 1996) as:

$$PIC = 1 - \sum P_i^2$$

Where, P_i is the frequency of the i^{th} allele.

Each SSR fragment was treated as binary matrix in which band presence was coded as present or absent by 1 and 0, respectively. Based on the binary matrix, Jaccard'sdissimilarity index was computed as follows. A graphical display of the genetic relationships was also computed by principal coordinate analysis. Subsequently,

Table 2. The 30 SSR primers used for the genotyping of the 112 sweet potatoaccessions.

Marker	Primer sequences from client	Forward_primer with M13 tailed *
IbL16_F	GTCTTGCTGGATACGTAGAACA	cacgacgttgtaaaacgacGTCTTGCTGGATACGTAGAACA
IbL16_R	GGGAGAAGTAAGAGAACCGATA	-
IbL32_F	GGGATGAAGGAGAGAATGAGTA	cacgacgttgtaaaacgacGGGATGAAGGAGAGAATGAGTA
IbL32_R	TTGAAAACCTAGAGAGAAAGGG	-
IbL46_F	CTGAAATTAGGGATTGAAGAGG	cacgacgttgtaaaacgacCTGAAATTAGGGATTGAAGAGG
IbL46_R	TCCAATCACTCCTTGTTTTCTC	-
IbO2_F	TGTGGATCTGTTCTTTGAACC	cacgacgttgtaaaacgacTGTGGATCTGTTCTTTGAACC
IbO2_R	TTCCATGTGGAGTGTGAAGTAT	-
IBS100_F	TGCTATAGTTACGTGGACGAAG	cacgacgttgtaaaacgacTGCTATAGTTACGTGGACGAAG
IBS100_R	TTTAATGCTGATGTGGATGC	-
IBS12_F	CAGTTATCAATTCCCACCTACC	cacgacgttgtaaaacgacCAGTTATCAATTCCCACCTACC
IBS12_R	TTGCTGTGTTATAGGCTTTGTC	-
IBS134_F	CTTCAATCACCTGAAACTCTGA	cacgacgttgtaaaacgacCTTCAATCACCTGAAACTCTGA
IBS134_R	AATATCGCTATGTTCTTGGGaC	-
IBS137_F	TcAACAGACGTCTTCACTTACC	cacgacgttgtaaaacgacTcAACAGACGTCTTCACTTACC
IBS137_R	TCGATAGTATGATGTGAATCGC	-
IBS139_F	CTATGACACTtCTGAGAGGCAA	cacgacgttgtaaaacgacCTATGACACTtCTGAGAGGCAA
IBS139_R	AGCCTTCTTGTTAGTTTCAAGC	-
IBS144_F	TCGAACGCTTTCTACACTCTT	cacgacgttgtaaaacgacTCGAACGCTTTCTACACTCTT
IBS144_R	CTGTGTTTATAGTCTCTGGCGA	-
IBS147_F	TGTGTACATGAGTTTGGTTGTG	cacgacgttgtaaaacgacTGTGTACATGAGTTTGGTTGTG
IBS147_R	GAAGTGCAACTAGGAAACATGA	-
IBS156_F	TTGATTCCACTATGACTTGAGC	cacgacgttgtaaaacgacTTGATTCCACTATGACTTGAGC
IBS156_R	ACACCAACCCTTATATGCTTTC	-
IBS166_F	TCCGTCTTTCTTCTTCTTCTTC	cacgacgttgtaaaacgacTCCGTCTTTCTTCTTCTTCTTC
IBS166_R	ATACACTAACTGCATCCAAACG	-
IBS18_F	GCCAAGGATGAAGGATATAGAa	cacgacgttgtaaaacgacGCCAAGGATGAAGGATATAGAa
IBS18_R	ACAAcCAAACTAGCTAAAAGCC	-
IBS19_F	TCCTATGAGTGCCCTAAGAATC	cacgacgttgtaaaacgacTCCTATGAGTGCCCTAAGAATC
IBS19_R	CTCCTTCGTCTTCTTCTTcTTC	-
IBS199_F	TAACTAGGTTGCAGTGGTTTGT	cacgacgttgtaaaacgacTAACTAGGTTGCAGTGGTTTGT
IBS199_R	ATAGGTCCATATACAATGCCAG	-
IBS24_F	AGTGCAACCATTGTAATAGCAG	cacgacgttgtaaaacgacAGTGCAACCATTGTAATAGCAG
IBS24_R	TCCTTTCtTcATCATGCACtAc	-
IBS33_F	ATCTCTtCATACcAATCGgAaC	cacgacgttgtaaaacgacATCTCTtCATACcAATCGgAaC
IBS33_R	CaATgaTAGCGGAGATTGAAG	-
IBS72_F	CTACTCTCTGCTGGTTTATCCC	cacgacgttgtaaaacgacCTACTCTCTGCTGGTTTATCCC
IBS72_R	CTAGTGGTCTCTCTTCCTCCAC	-
IBS82_F	GACATAATTTGTGGGTTTAGGG	cacgacgttgtaaaacgacGACATAATTTGTGGGTTTAGGG
IBS82_R	GAAATGGCAGAATGAGTAAGG	-
IBS84_F	CAAAGATGAAGCAAGTAAGCAG	cacgacgttgtaaaacgacCAAAGATGAAGCAAGTAAGCAG
IBS84_R	ACTAATGTTGATCTACGGACCC	-
IBS85_F	AACTACTCATGGGGAGAACAAC	cacgacgttgtaaaacgacAACTACTCATGGGGAGAACAAC
IBS85_R	CTAACGAAAGTTTGGACATCTG	-
IBS86_F	AGAAACTGAAAACTAAGCTCGC	cacgacgttgtaaaacgacAGAAACTGAAAACTAAGCTCGC
IBS86_R	GCTATGCGTTTACAGAAACAAG	-
IBS97_F	GTTACCAGGAATTACGAACGAT	cacgacgttgtaaaacgacGTTACCAGGAATTACGAACGAT
IBS97_R	CTCTCTACAAAAACTCACAGCG	-
IbU13_F	GCAACCAATCTACAGCAAACTA	cacgacgttgtaaaacgacGCAACCAATCTACAGCAAACTA
IbU13_R	CAGATAAAGTCCCCATTTCTTC	-
IbU20_F	GGAGAGCAAGTGGAGAAAGTAT	cacgacgttgtaaaacgacGGAGAGCAAGTGGAGAAAGTAT

Table 2. Contd.

IbU20_R	ACTCCTAGACCCACAATTGAAC	-
IbU31_F	CCGCAGAAAAAGTTCAGATT	cacgacgttgtaaaacgacCCGCAGAAAAAGTTCAGATT
IbU31_R	GCAACTTTTCTTCTTCCGTAAC	-
IbU33_F	TTTGAAGAAGATGAGAGCGAC	cacgacgttgtaaaacgacTTTGAAGAAGATGAGAGCGAC
IbU33_R	TCAGAAAGACGATACACTAGAGAGA	-
IbU4_F	GGCTGGATTCTTCATATTTAGC	cacgacgttgtaaaacgacGGCTGGATTCTTCATATTTAGC
IbU4_R	GCTTAATGGATCAGTAACACGA	-
IbU6_F	GGGGTAGAGAGAAGAGAGTGAC	cacgacgttgtaaaacgacGGGGTAGAGAGAAGAGAGTGAC
IbU6_R	CCAGGTGAGAGTGTCTTTCAA	-

Table 3. Selected morphological characters by The STEPDISC procedure.

Step	Entered	Partial R-square	F value	Pr> F	Wilks' Lambda	Pr< Lambda	Average squared canonical correlation	Pr> ASCC
1	Predominant Flesh Color (PFC)	0.8498	299.81	<.0001	0.15022095	<.0001	0.42488952	<.0001
2	Leaf Lobe Number (LLN)	0.4128	36.90	<.0001	0.08821579	<.0001	0.62429365	<.0001
3	Leaf Lobe Type (LLT)	0.1204	7.12	0.0013	0.07759628	<.0001	0.65429960	<.0001
4	Mature Leaf Size (MLS)	0.1035	5.94	0.0036	0.06956707	<.0001	0.66236509	<.0001
5	Vine Tip Pubescence (VTP)	0.0711	3.91	0.0232	0.06461809	<.0001	0.67647456	<.0001
6	Storage Root Surface Defects (SRSD)	0.0525	2.80	0.0655	0.06122257	<.0001	0.68154041	<.0001
7	Petiole Pigmentation (PP)	0.0514	2.71	0.0716	0.05807721	<.0001	0.69485966	<.0001
8	Storage Root Formation (SRF)	0.0508	2.65	0.0759	0.05512967	<.0001	0.69785323	<.0001

Number of observation =109, Variables in the analysis=30, Class level=3, Significance level to enter=0.15, Significance level to stay=0.15.

a dendrogram was generated with the unweighted pair group method using arithmetic average (UPGMA) algorithm of DARwin5.0.158 software (Perrier et al., 2003 and Perrier and Jacquemoud-Collet, 2006).

RESULTS AND DISCUSSION

Morphological characterization

Discriminant analysis

Eight morphological traits with sufficient discriminative power to differentiate the accessions were identified based on their significant p-value forWilk's Lambda ($P < 0.0001$) and p-values for the average squared canonical correlations ($P < 0.0001$) (Table 3). These were: PFC, LLN and LLT (Figure 3), MLS, VTP, SRSD, PP and SRF. The correlation matrices from Table 4 shows that these eight descriptors were not correlated with one another; this therefore indicates that using them will not create redundancy in the measurements. The F values revealed that the PFC and the LLN, respectively, with 299.81 and 36.90 had the greatest discriminating power associated with highly significant F values. Among the 22 variables discarded were the PSC commonly used by farmers to identify cultivars; the FH very important in breeding and other visible traits such as PT, MLC, ILC, GOL, and PVC.

Principal component analysis

Four principal components (PC) were identified which accounted for 67.22% of the total variation among the accessions (Table 5).The first PC accounted for 23.08% whereas the second, the third and the forth PC axes accounted respectively for 18.08, 13.32 and 12.73%. The first PC with reference to its high loadings (Table 6) was positively associated with traits such as leaf lobe number and predominant flesh colour. The second PC was associated with storage root characteristics (predominant flesh colour, storage root surface defects); the third with leaf characteristics (mature leaf size and petiole pigmentation) as well as with storage root formation, while the forth was associated with traits related to stems (leaf lobe type, petiole pigmentation and vine tip pubescence).

Cluster analysis

From the hierarchical cluster analysis, leaf lobe number, leaf lobe type, petiole pigmentation, vine tip pubescence, predominant flesh colour, storage root formation, storage surface defectand storage root surface defect showed a high polymorphism of 0.75 within the 112 sweet potato accessions (Figure 1).

Table 4. Correlation matrix for the 8 morphological traits used to distinguish the 112 sweet potato accessions.

Parameter	VTP	LLN	LLT	MLS	PP	PFC	SRF
Vine tip pubescence (VTP)							
Leaf lobe number (LLN)	0.1666						
Leaf lobe type (LLT)	0.0035	0.1039					
Mature leaf size (MLS)	0.2159	0.1699	-0.1856				
Petiole pigmentation (PP)	-0.0089	-0.2091	0.0254	0.0133			
Predominant flesh color (PFC)	0.1763	0.2690	0.0976	0.1906	-0.1685		
Storage root formation (SRF)	-0.2387	-0.2462	0.1031	-0.0850	0.0980	-0.0117	
Storage root surface defects (SRSD)	0.0157	0.2267	0.1249	-0.0096	-0.1464	0.2728	0.1879

Table 5. Eigenvalues of the correlation matrix**.**

Eigen values	Difference	Proportion	Cumulative
1.84627182	0.39948563	0.2308	0.2308
1.44678619	0.38093347	0.1808	0.4116
1.06585272	0.04728026	0.1332	0.5449
1.01857246	0.26427255	0.1273	0.6722
0.75429991	0.04126424	0.0943	0.7665
0.71303567	0.06889746	0.0891	0.8556
0.64413821	0.13309520	0.0805	0.9361
0.51104301		0.0639	1.0000

Table 6. Eigenvectors from the eight principal component axes used to classified the 112 sweet potato accessions.

Parameter	Prin1	Prin2	Prin3	Prin4	Prin5	Prin6	Prin7	Prin8
Vine tip pubescence (VTP)	0.366	-.311	0.090	0.433	-.641	0.248	0.240	0.208
Leaf lobe number (LLN)	0.528	0.038	-.231	0.030	0.564	0.177	0.125	0.548
Leaf lobe type (LLT)	0.058	0.448	-.271	0.650	0.085	-.386	0.275	-.262
Mature leaf size (MLS)	0.313	-.343	0.575	-.074	0.298	-.309	0.395	-.328
Petiole pigmentation (PP)	-.306	-.137	0.413	0.601	0.362	0.314	-.351	0.050
Predominant flesh color (PFC)	0.484	0.209	0.248	0.022	-.162	-.397	-.686	0.075
Storage root formation (SRF)	-.255	0.498	0.512	-.084	-.129	-.127	0.316	0.534
Storage root surface defects (SRSD)	0.306	0.522	0.201	-.121	-.017	0.625	0.038	-.432
Eigen value	1.846	1.447	1.066	1.019	0.755	0.713	0.644	0.511
% Variation	23.08	18.08	13.32	12.73	9.43	8.91	8.05	6.39
Cumulative %	23.08	41.16	54.49	67.22	76.65	85.56	93.61	100

The accessions were grouped into eleven (11) clusters based on their average linkage and the Euclidean test. Clusters IV, VIII, IX and XI can be considered as outliers as they contained only one accession each, BF90, BF120, BF81 and BF137, respectively. Cluster I consisted of 37 accessions, cluster II had 11 accessions, cluster III had 10 accessions, cluster V of 6 accessions, cluster VI and VII had 21 accessions each, whereas cluster X had two accessions. Cluster II and cluster III were entirely constituted by orange fleshed accessions mostly with three leaf lobes, while the other clusters did not show any distinguishable relationship or pattern. The three East African OFSPs:Resisto (CIP 440001) belonged to cluster II while CIP-199062-1 and TIB-440060 belonged to cluster III. Cluster I was associated mostly with accessions with yellow flesh and a leaf lobe number of nine except for BF16 and BF42 which had 13 and 11 leaf lobes, respectively. Cluster V was constituted by accessions with white flesh and seven leaf lobes, cluster VI had individuals characterized by white flesh and one leaf lobe while cluster VII had white flesh with a very divergent number of leaf lobes ranging from one to

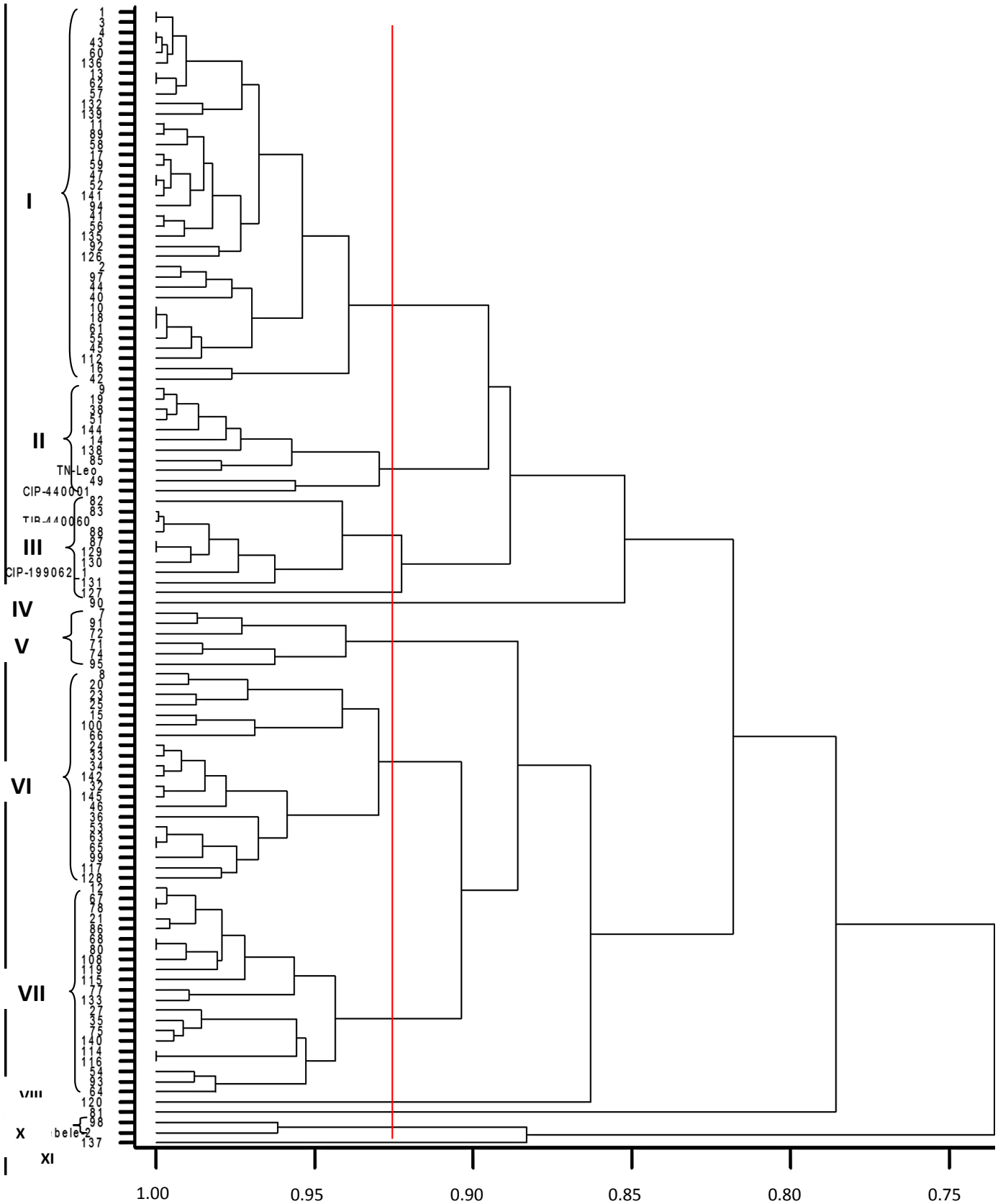

Figure 1. Dendrogram of the 112 sweet potato accessions revealed by average linkage cluster analysis based on the eight discriminant phenotypic characters.

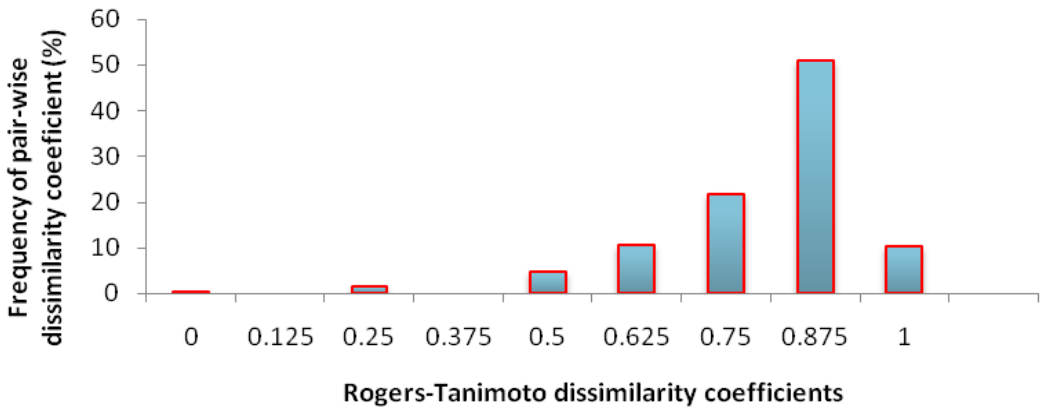

Figure 2. Rogers-Tanimoto Dissimilarity index using: Mean = 0.73, Min value = 0, Max value = 1 (DARwin5.0.158).

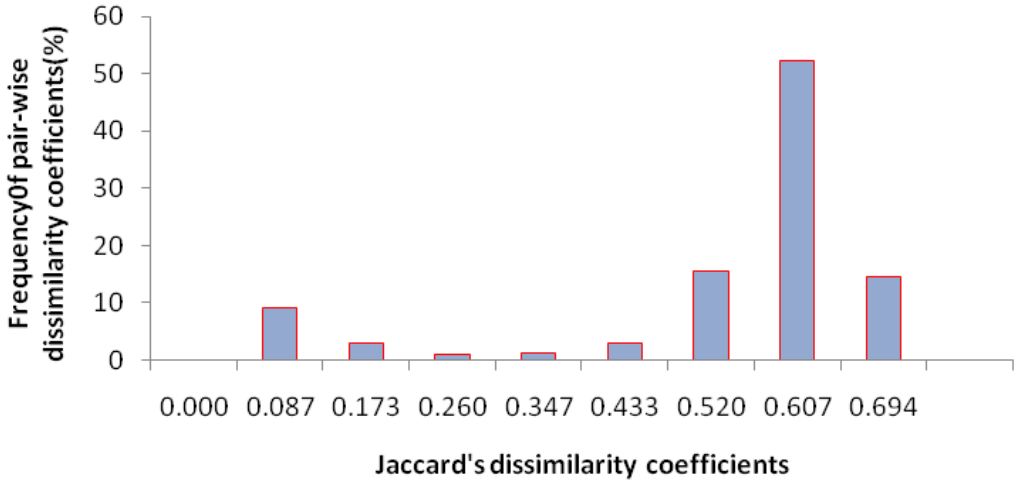

Figure 3. Jaccard's dissimilarity index using: Mean = 0.49, Min value = 0, Max value = 0.69 (DARwin5.0.158).

five with most of the accessions having five leaf lobes. The Rogers-Tanimoto pairwise dissimilarity coefficients computed as single and modality data using DARwin 5.0.158 revealed a dissimilarity index ranging from 0 to 1 with an average value of 0.73 (Figure 2) suggesting a very high diversity among these 112 accessions. Most of accessions had dissimilarity indices ranging from 0.75 to 0.875 explaining 72.51% of the total frequency of dissimilarity with a maximum pair-wise dissimilarity of 1.

Identification of duplicates

From the hierarchical cluster analysis (Figure 4), duplicates were identified. Accessions BF1 and BF3 from two close villages in the central region were identical. Accession BF13 from the central south was identical to accession BF62 from the Eastern region; two accessions BF78 and BF67 from the "Hauts-Bassins" region were identical as well as accessionsBF129 from the "Hauts-Bassins" and BF87 from the "Cascades". BF80 and BF68 from the Hauts-Bassins were also identical as were BF65 and BF63 from the same region. BF10 and BF18 from the central south and BF61 from the Eastern region were also identical. BF116 and BF114 from the Central west were morphologically identical as were BF52 and BF47 from the Central South.

Molecular characterization

Number of alleles detected

Among the 30 SSR markers, 27 were detected between one to six alleles while the remaining three markers detected between seven to eight alleles.

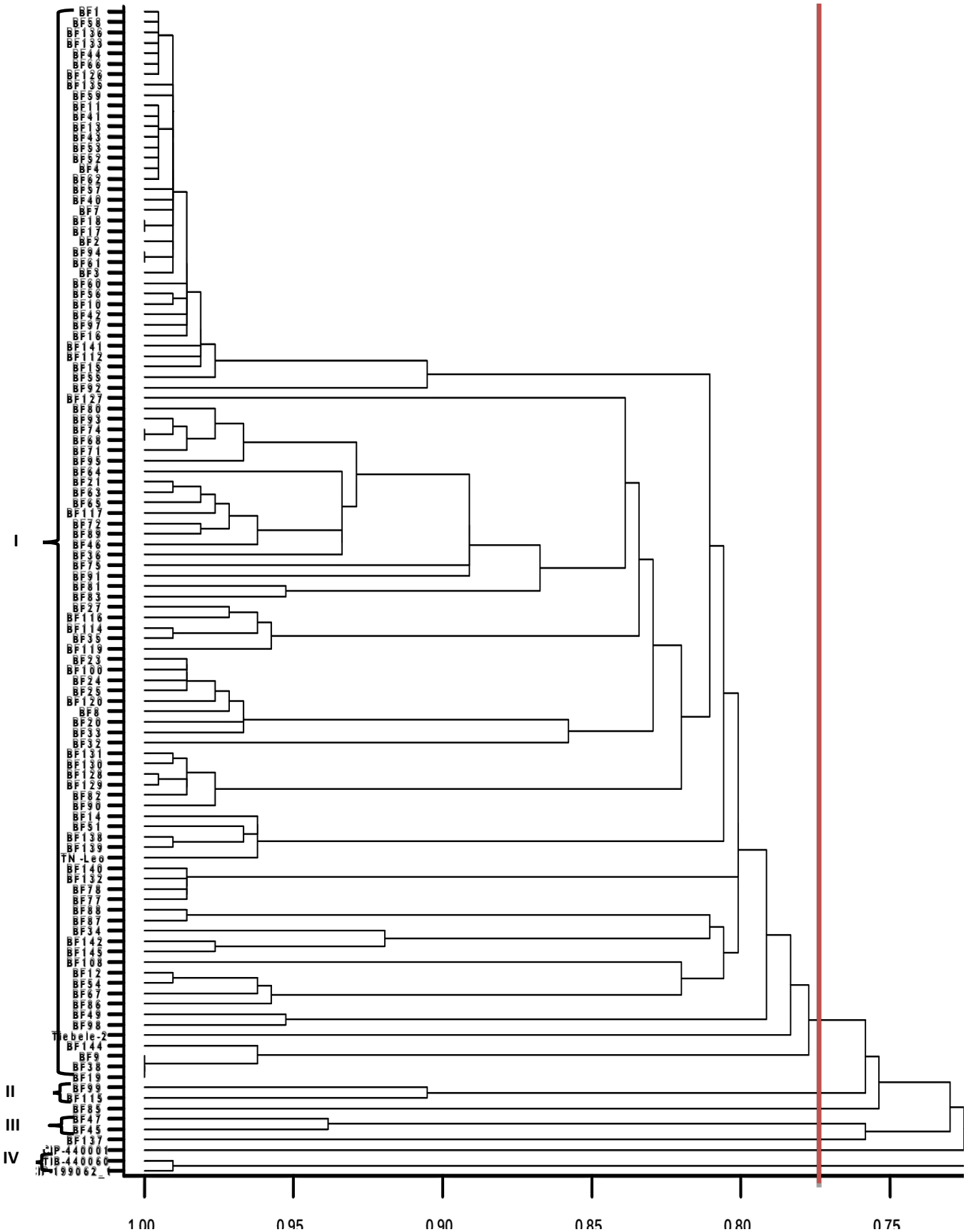

Figure 4. SSR UPGMA based dendrogram of 112 sweetpotato accessions from Burkina Faso.

Table 7. Markers, number of alleles per locus, total number of alleles and PIC for 30 SSR.

Marker	Number of alleles per locus	Total alleles	PIC
IBL16	8	232	0.715
IbL32	5	374	0.762
IbL46	7	252	0.713
IbO2	10	644	0.881
IBS12	8	267	0.782
IBS18	7	324	0.774
IBS19	7	322	0.796
IBS24	7	329	0.795
IBS33	5	313	0.734
IBS72	4	253	0.746
IBS82	7	382	0.764
IBS84	6	433	0.789
IBS85	8	240	0.776
IBS86	7	301	0.726
IBS97	7	346	0.744
IBS100	6	347	0.771
IBS134	4	287	0.708
IBS137	7	337	0.781
IBS139	12	431	0.873
IBS144	8	374	0.812
IBS147	8	336	0.788
IBS156	6	133	0.283
IBS166	10	232	0.739
IBS199	12	441	0.841
IbU4	9	370	0.813
IbU6	8	395	0.819
IbU13	6	333	0.786
IbU20	1	111	0.000
IbU31	4	135	0.547
IbU33	7	329	0.763
Mean			0.727

The SSR marker IbO2 detected one to six alleles from 61 samples, seven alleles from 48 samples and eight alleles from three samples. The markers IBS139 and IBS199 detected one to six alleles each from 111 samples and seven alleles from one sample. The samples BF32 and BF99 showed between seven to eight alleles by the three SSR markers.

Polymorphic information content (PIC)

The thirty SSR markers revealed the usefulness of a marker in distinguishing between accessions with PIC values ranging from 0.00 for IbU20 to 0.881 for IbO2with an average of 0.727 (Table 7). Except for two SSR markers that had PIC values lower than 0.50 (IbU20 with 0 and IBS156 with 0.283), twenty eight (28) markers had high power of polymorphism (PIC>0.50). The high PIC values observed in this study indicated that the twenty eight SSR markers used were informative.

Genetic dissimilarity analyses and identification of duplicates

The frequency of pair-wise dissimilarity coefficients of the 112 sweet potato accessions based on the Jaccard's coefficient is shown in Figure 3. These SSR-basedpair-wise dissimilarity coefficients ranged from 0 to 0.69 with a mean of 0.49 suggesting a relatively moderate diversity among the 112 sweet potato accessions. Most of the dissimilarity coefficients were between 0.52 and 0.69 explaining 82.35% of the total frequency.

Nine accessions were identified with a pair-wise

dissimilarity of 0 and therefore were considered as duplicates. This observation is confirmed by the dendrogram (Figure 4) generated using the unweighted pair group method (UPGMA). Thus, BF61 and BF94 with yellow flesh which were collected from“Cascades” and the Central-East region, respectively, were genetically identical; BF17 and BF18, two yellow fleshed accessions collected in two different communities in the Bazega province (Central-South region), were identical. BF38, BF19 and BF9, three orange fleshed accessions from the Bazega province, were also identical and different from the OFSP introduced from CIP-Eastern Africa.BF74 and BF68, with white flesh from the Kenedougou province, constituted a unique accession. After removing the duplicates, the initial number of 112 accessions was reduced to 107. These 107 sweet potato accessions will constitute a national core collection of sweet potato germplasm.

Comparison between morphological and SSR data

Using the morphological characters, the 112 accessions were grouped into 11 clusters with dissimilarity indices ranging from 0 to 1 with a mean of 0.73 suggesting a very high genetic diversity among the accessions. The use of the morphological data reduced the number of accessions from 112 to 101. Conversely, using the SSR-based analysis, 7 clusters were obtained.The dissimilarity indices ranged from 0 to 0.69 with a mean of 0.49, therefore, showing a relatively moderate diversity among the 112 accessions. The accession numbers were reduced from 112 to 107 using SSR markers. The accessions BF87 and BF88; BF63 and BF65; BF114 and BF116 identified as group of duplicates by morphological descriptors were closely related (nested on the dendrogram) using the SSR markers. Except for the groups of duplicates BF47 and BF52; BF67 and BF78 that were seen far away by the SSR markers, the other morphologically duplicates accessions belonged to the same molecular cluster.

In the other side, the duplicates identified using the SSR marker procedure BF17 and BF18 belonged to the same morphological cluster, as did BF94 and BF61. The duplicates BF74 and BF68 were seen morphologically far away, while BF9, BF19 and BF38 identified as the same accessions by the molecular procedure were found nested closely on the morphological dendrogram. The consensus between the morphological and the molecular based treeswas performed by using the strict rule consensus method consisting of simple counts of the frequency of occurrence of clusters in the set of trees (Perrier Perrier and Jacquemoud-Collet, 2006). It was observed that between the two trees, 4.7% of the clusters were in agreement. This weak consensus between the two trees suggested that there was no correlation between the morphological and the molecular data.

The Quartet tree distance estimate used as a measure of dissimilarity between the two trees was 0.95 demonstrating the absence of correlation between the two approaches used in the genetic diversity estimation.

DISCUSSION

The high diversity (mean of 0.73) detected within the 112 accessions regarding dissimilarity coefficient values suggests that the sweet potato accessions used in the current work would be a good source of selection for sweet potato breeding materials. Diversity studies have been done on sweet potato using morphological descriptors in various parts of the world and similarities or differences have been ascribed to sample size, number and type of descriptors used, the origin of accessions and the method of analysis. Using forty morphological descriptors in Uganda on 1256 accessions, 20 discriminatory descriptors were identified (Yada et al., 2010a). These 20 descriptors contained seven of the eight descriptors identified in this present study. Predo-minant skin color, commonly used in identification of cultivars in farmers’ fields in Burkina Faso was not useful in differentiation among the accessions. Contrary to the results of this present work, Yada et al. (2010a) found this descriptor as discriminatory. In Kenya, Karuri et al. (2010) identified two descriptors (general outline of leaf, and, the shape of central leaf lobe) that differentiated among 89 accessions and separated them into two clusters. Karuri et al. (2010) found in agreement with the results of the current work, that flower habit was not significantly discriminative. High diversity index was also observed in a population of sweet potato in Kenya (Karuri et al., 2010), Uganda and India (Vimala and Hariprakash, 2011) using morphological traits. However, Tairo et al. (2008) observed lowdiversity of 0.52 among 280 sweet potato accessions in Tanzania.

Considering that SSR-based data are more accurate than the morphological data, the moderate diversity obtained in this study suggests that high priority should be given to further collect and/or introduce divergent materials, since variation in the collections is needed for a successful breeding program. Results from similar studies using SSR markers in sweet potato diversity analysis have been reported and most of the differences in results have been ascribed to sample size, the number of SSR markers used and the source of materials. Moderate genetic diversity values have been reported in Uganda (Yada et al., 2010b) among 192 accessions using 10 SSR markers; Gichuru et al. (2006) also reported low diversity in East African sweet potato cultivars while Soegianto et al., (2011) in Java reported similarity ranging from 15 to 78% between Indonesian accessions. Considering Eastern Africa as the second zone of diversity of sweet potato after the Central America (Villordon et al., 2007), one would expect a high

diversity. The reason for the low diversity has been attributed to narrow geographic zone of collection of the cultivars. High SSR-based diversity has been noticed by Veasyet al. (2008) in Brazil, in Taiwan by Shih et al. (2002) and in China by Li et al. (2009) where the Jaccard's coefficient of similarity ranging from 0.400 to 0.938 was observed.

The weak agreement between the morphological based tree and the SSR based tree was also confirmed by different duplicates identified by each of these approaches. The findings of the present study are in agreement with those of Karuri et al. (2010) in Kenya who compared morphological and SSR-based evaluation of diversity.

A low correlation of -0.05 was observed between the two data sets. Further studies have reported low correlation between morphological and molecular markers in many crops (Koehler-Santos et al., 2003; Ferriol et al., 2004; Bushehri et al., 2005). The suggested reasons were that it could be a result of the independent nature of morphological and molecular variations. According to Vieira et al. (2007), this low correlation could also be due to the fact that a large portion of variation detected by molecular markers is non-adaptive as compared with phenotypic characters, which are influenced by the environment. The core collection obtained using the SSR markers' approach will be used for breeding purposes but the identified eight phenotypic characters will be used for the physical identification of the cultivars within the core collection.

Conclusion

Findings of the present study reveal that sweet potato germplasm in Burkina Faso presented moderate to high diversity based on molecular and phenotypic assessment approaches. The results obtained will serve as a guide for the basis germplasm management and improvement in the Burkina Faso and in the Sahelian zone of West Africa. However, further diversity is needed that can be achieved through introduction or more collection. The power of eight morphological descriptors and 28 SSR markers in the differentiation of cultivars was identified and could be useful in subsequent studies. Despite the poor correlation between morphological and molecular markers, both techniques can be use defectively in sweet potato characterization. The constitution of core collection will be done based on the SSR based data, but the eight phenotypic characters will be useful in distinguishing the cultivars in the field.

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