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Genetic Differentiation of within and among the Genome Groups of Banana (*Musa* spp.) Cultivars of Northeast India

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Genetic variation of 27 banana (*Musa* spp.) cultivars collected from North-eastern region of India belonging to five different genome groups (AA, BB, AAA, AAB and ABB) were studied using RAPD markers. Altogether 30 RAPD markers were tested, out of which nine were successful in generating consistent PCR products. Amongst the 99 PCR products generated, 7.07% were monomorphic and 92.93% were polymorphic bands. High level of genetic diversity values were observed comprising expected genetic heterozygosity (Ht) of 0.24, homozygosity (Hs) of 0.07, genetic differentiation (Gst) of 0.67 and gene flow (Nm) of 0.23. Analysis of molecular variations (AMOVA) also revealed high level of genetic variation among the cultivars of different genomic groups (70%) and within the same genome group (30%). Among the five genome groups studied, AA showed the highest genetic variation. The dendrogram and principal component analysis based on RAPD profiles revealed the successful differentiation and clustering of banana cultivars based on their genome groups except in one cultivar (Noneh-laphu, AA) which was clustered with cultivars belonging to BB group.

Key Words: Genome, Genetic differentiation, Musa, North-eastern region of India, RAPD

Abbreviations: RAPD: randomly amplified polymorphic DNA; UPGMA: unweighted pair group method with arithmetic averages; PIC: Polymorphism information content; AFLP: amplified fragment length polymorphism; CTAB: cetyltrimethyl ammonium bromide

Introduction

Bananas (*Musa* spp.), consisting of both dessert and cooking types, are giant herbaceous monocotyledonous plants that belong to the family Musaceae of the order Zingeberales (Simmonds and Shepherd, 1955). The Musaceae family consist of two genera *Musa* and *Ensete* where *Musa* differs from *Ensete* in producing suckers around the parent plants (Simmonds and Shepherd, 1955; Ħakkinen, 2013). The present day edible and seeded bananas are believed to be derived from hybridization between wild diploid of *M. acuminata* (AA genome) and *M. balbisiana* (BB genome) contributing various levels of ploidy and genomic constitution such as AA, BB, AB, AAA, AAB, ABB, ABBB, etc. (Simmonds and Shepherd, 1955). The North-eastern region (NER) of India has been considered as a rich hub of natural banana diversity where *M. balbisiana* from Indian subcontinent meet *M. acuminata* from South-East Asia (Molina and Kudagamage, 2002). However, the vast genetic resource of wild and cultivated banana in the region remains relatively unexplored and untapped for scientific studies.

Knowledge on the genetic diversity and genetic relationships among the various wild and cultivated bananas will provide valuable information on the genetic status and can be utilized for breeding and sustainable production. The use of morphological data for determination of ploidy levels and genome classification in banana has been supplemented with the use of molecular data (Pollefeys *et al.,* 2004; Pachuau *et al.,* 2014). Among the molecular markers, RAPD, AFLP and microsatellites are frequently used for the characterization of banana (Wong *et al.,* 2002; Onguso *et al.,* 2004). The use of RAPD makers has the advantage of being efficient and inexpensive without requiring prior knowledge of the genome (Bhat *et al.,* 1995). It requires very small amount of genomic DNA without the need for blotting and radioactive detection and are moderately reproducible (Williams *et al.,* 1990; Cipriani *et al.,* 1996; Aagaard *et al.,* 1998). Thus, the present study was undertaken to analyze the genetic variation of banana cultivars of NER of India using RAPD markers.

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Materials and Methods

A total of 27 banana cultivars which were collected from different parts of the NER state of Manipur and previously classified into five genome groups (AA, AAA, BB, ABB and AAB) by Pachuau *et al*. (2014) were used in the RAPD analysis. The details of the cultivars are given in Table 1.

Genomic DNA Isolation

Genomic DNA was extracted from the young cigar leaf tissues using modified CTAB method (Thangjam *et al.*, 2003). The leaves were grounded in 700 µl extraction buffer (1M Tris HCl, pH 8.0; 5M NaCl; 0.5 M EDTA; 10% sodium dodecyl sulphate) and the mixture was treated with 5 µl of proteinase K (20 mg/ml) for 1 h at 36°C. To this cell lysate, 500 µl of CTAB buffer [10% CTAB; 1M Tris HCl pH 8.0; 0.5M EDTA; 5M NaCl; 5% PVP (Polyvinyl pyrolidine] was added and incubated for 2 h at 65°C. The suspension was extracted thrice with equal volume of phenol, chloroform, isoamyl alcohol (24:25:1). The DNA was precipitated using isopropanol, sodium acetate and 70% ethanol; 10 µL RNase (10µg/ml) was added to each 50 µL DNA and incubated for 30 min at 37°C. Purity and quantity of DNA were measured by using Biophotometer plus (Eppendorf, Germany) by taking their absorbance at 260 nm and 280 nm and verified by running on 0.8% agarose electrophoresis to check the quality of DNA along with 1kb plus DNA ladder.

RAPD-PCR and Data Analysis

PCR of the isolated DNA from the 27 banana samples were carried out in a 25µl volume reaction. Each reaction tube contained 50 ng of genomic DNA, 2.5 mM $MgCl_2$, 0.5 mM dNTP mix, 2.5 mM buffer, 1U

Table 1. Details of banana (*Musa* spp.) cultivars from Manipur state of North-eastern India used for genetic diversity study

S. No.	Collector No.	Local cultivar name	Collection site (place, district)	Scientific name	Genome group*
1.	TRSMN- 01	Ragunshen-masung-ahumnaibi	Khongsang, Tamenglong	*Musa* sp.	ABB
2.	TRSMN- 02	Heijao-angouba	Keithelmanbi, Imphal West	*Musa* sp.	ABB
3.	TRSMN- 03	Champa-colla- manbi	Jiribam, Imphal East	*Musa* sp.	AAB
4.	TRSMN- 04	Champa-colla-angangbi	Kangbai, Churachandpur	*Musa* sp.	ABB
5.	TRS MN- 05	Teralaphoi	Pangai, Imphal East	*Musa* sp.	ABB
6.	TRS MN- 06	Luine	Noneh, Imphal West	*M. balbisiana*	BB
7.	TRS MN- 07	Champa-colla	Khumbong, Imphal west	*Musa* sp.	AAB
8.	TRSMN- 08	Korbot	Khuga river, Bisnupur	*M. acuminata*	AA
9.	TRSMN- 09	Noneh-laphu	Noneh, Imphal West	*M. acuminata*	AA
10.	TRSMN- 10	Morpi	Kangbai, Churachandpur	*Musa* sp.	ABB
11.	TRS MN- 11	Hei	Khumbong, Imphal West	*Musa* sp.	ABB
12.	TRSMN- 12	Grandnaine	Keithelmanbi, Imphal West	*M. acuminata*	AAA
13.	TRSMN- 13	Jahaji	Keithelmanbi, Imphal West	*M. acuminata*	AAA
14.	TRSMN- 14	Changbi-mara-chatpi	Sekmai, Imphal East	*M. acuminata*	AA
15.	TRS MN- 15	Hangou	Pangei, Imphal East	*Musa* sp.	ABB
16.	TRSMN- 16	Masung-ahum-naibi	Mayai Leikai, Thoubal	*Musa* sp.	ABB
17.	TRSMN- 17	Meitei-hei	Pangei, Imphal East	*Musa* sp.	ABB
18.	TRSMN- 18	Des-laphu	Sekmai, Imphal East	*M. acuminata*	AAA
19.	TRS MN- 19	Yensang-chabi	Jiribam, Imphal East	*Musa* sp.	ABB
20.	TRSMN- 20	Morshell	Noneh, Imphal West	*Musa* sp.	ABB
21.	TRSMN- 21	Nagunsen	Tipaimukh, Churachandpur	*Musa* sp.	AAB
22.	TRSMN- 22	Chingchup	Bamdyar, Imphal West	*M. acuminata*	AAA
23.	TRSMN- 23	Changbi	Yurembam, Imphal West	*M. balbisiana*	BB
24.	TRSMN- 24	Mizoram-laphu	Monlom, Churachandpur	*M. acuminata*	AAA
25.	TRSMN- 25	Kharam-laphu	Kharam, Imphal West	*M. balbisiana*	BB
26.	TRSMN- 26	Utteibi	Bamon Leikai, Thoubal	*Musa* sp.	ABB
27.	TRSMN- 27	Ngalai	Mata, Churachandpur	*Musa* sp.	ABB

* based on Pachuau *et al.* (2014)

Taq polymerase (Bangalore Genei, India) and 10 pmol of each primer. The RAPD primers (UBC Set # 5) were obtained from University of British Columbia, Vancouver, Canada. The amplification was carried out using a thermal cycler (Applied Biosystems Geneamp PCR System 9700) programmed at 94°C for 4 min., followed by 35 cycles consisting of 94°C for 1 min., 36°C for 50 sec. and 72°C for 2 min. The final extension step was programmed at 72°C for 5 min. Amplified PCR products were separated by electrophoresis along with 100bp standard (100 bp DNA ladder) on 1.8% agarose gel prepared in 1X TAE buffer. DNA banding profiles were visualized and photographed using Gel Documentation System (G:BOX Chemi XT4 version 1.2.5.0, Syngene).

The amplified DNA fragments were treated as a separate character and scored as a discrete variable, using 1 to indicate presence and 0 for absence. Accordingly, rectangular binary data matrix was generated from the RAPD profiles. The efficiency of each primer were studied by evaluating band characteristics such as number of bands (NB), number of polymorphic bands (NPB), percentage of polymorphic bands (PPB).

Polymorphism information content (PIC_i) of a band was calculated according to Anderson *et al.* (1993) as follows:

$$PIC_i = 1 - \sum_j f_{ij}^2 \sum_j f_{ij}^2$$

Where f_{ij} is the frequency of the j^{th} pattern of the i^{th} band (dominant markers have two patterns for a band as being present and absent). Then, the PIC of each primer was calculated as:

$$PIC = 1/n \sum_{i=1}^{n} PIC\ i \sum_{i=1}^{n} PIC\ i$$

where n is the NPB for that primer.

Resolving power (Rp) was calculated according to formula of Prevost and Wilkinson (1999):

$$RP = \sum_{i=1}^{n} IBi \sum_{i=1}^{n} IBi$$

Where Informativeness of a band (BI_i) was calculated as:

$$BI_i = 1 - (2 \times | 0.5 - p)$$

where *p* is the proportion of the all accessions containing the band.

Further we calculated mean resolving power (MRP) for each primer as:

$$MRP = 1/n\ RP$$

Marker index (MI) was calculated as product of PIC and effective multiplex ratio (EMR), which is defined as the product of the fraction of polymorphic loci and the number of polymorphic loci.

MI of each primer was calculated according to Milbourne *et al.,* (1997) as

$$MI = PIC^* EMR$$

Genetic Diversity Analyses

Considering different genomes as a group, a total of five groups were used in the genetic analysis. It was carried out using software PopGene (version 1.32) (Yeh *et al.,* 1997). Nei's genetic distance (Nei, 1978) was calculated by using the programme RAPDDIP, a part of PopGene, between all the studied groups of banana. Shanon Population Index was also calculated; the larger the index, larger is the genetic diversity.

Total heterozygosity (Ht) is the expected heterozygosity, calculated as 1 minus the sum of the average allele frequencies over populations. G_{st}, a measure of population genetic differentiation was calculated (Nei, 1978) as:

$$G_{st} = \frac{Ht - HsHt - Hs}{Ht \quad Ht}$$

Where Hs and Ht are the expected heterozygosity within subpopulations and for the total population.

Effective migration rates (Nm) were estimated based on genetic differentiation (G_{st}) ($Nm=0.5(1-G_{st})/G_{st}$) inbreeding rates (McDermott and McDonald, 1993). An analysis of molecular variance (AMOVA) was performed by GenAlEx ver. 6.501 (Peakall and Smouse, 2006) to assess the overall distribution of molecular variance among and within different banana groups.

Genetic Relationships, Cluster Analysis and Principal Component Analysis

A pairwise comparison of the 27 samples was done based on simple matching (SM) matrix to show the similarity coefficient between the samples. A dendrogram, summarizing the genetic relationships among all samples was constructed using the same matrix employing UPGMA method. Principal component analysis (PCA) was carried out to assess the genetic distances among the groups. These analyses were conducted using the software NTSYS-pc version 2.20a (Numerical Taxonomy and Multivariate Analysis for Personal Computer, Rohlf, 2000).

Results

A total of 30 UBC-RAPD primers (set # 5) were initially screened with the DNA isolated from 27 banana samples for their amplification. Out of the 30 primers only nine primers (UBC-414, UBC-417, UBC-425, UBC-433, UBC-438, UBC-445, UBC-447, UBC-449 and UBC-450) produced clear and reproducible banding patterns in all the banana samples and two primers (UBC-440 and UBC-441) produced monomorphic bands thus, nine primers were used for further experiments. Fig. 1 shows the amplification pattern of 27 samples using UBC-438 primer.

Genetic Variation Revealed by RAPD Analysis

The nine UBC-RAPD primers generated a total of 99 bands ranging from 250-3000 bp molecular weight in the 27 tested samples (Table 2). The primer UBC-438 produced the highest number of bands (17) whereas UBC-417 generated the lowest number (6) of scorable bands. Out of the 99 bands, 92 were found polymorphic (92.93%). Polymorphic information content (PIC) was found highest in primer UBC-433 (0.59) while UBC-417 has least PIC value of 0.38. The resolving power (Rp) which indicates ability to distinguished groups was found highest in UBC-425 (6.2) and the lowest in UBC-433 (1.00). The MRP ranged from 0.25 – 0.83. MI, which is the discriminating power for a marker was found highest in UBC-438 (7.48) and lowest in UBC-433 (1.06). Among the nine primers, 5 primers (UBC-414, UBC-438, UBC-445, UBC-425 and UBC- 447) showed 100% polymorphism.

Analysis of Genetic Diversity within and among Genome Groups

The total heterozygosity was calculated as 0.245, a rich genetic variation among sample which is evident from a high value of G_{st}, *i.e.* 0.676, indicating a very great genetic differentiation. Effective migration rates (Nm) was observed to be 0.238, depicting a very limited gene flow (Table 3).

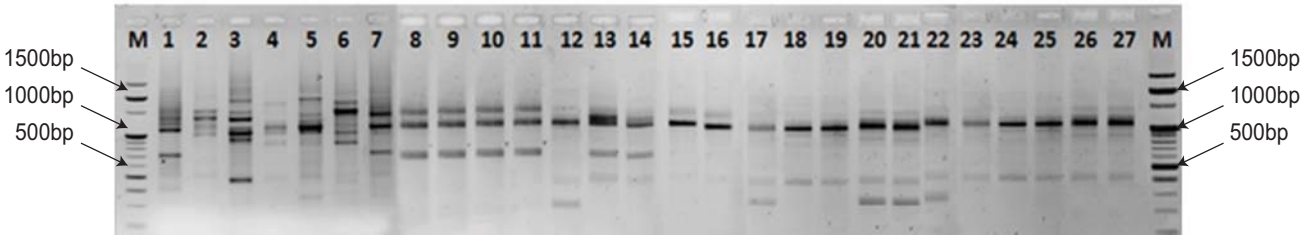

Fig. 1. RAPD-PCR profile of the 27 banana cultivars of Manipur using UBC primer #445.
M: 100bp marker; 1: Korbot; 2: Noneh-laphu; 3: Changb-imara-chatpi; 4: Luine; 5: Changbi; 6: Kharam-laphu; 7: Grandnaine; 8: Jahaji; 9: Des-laphu; 10: Chingchup; 11: Mizoram-laphu; 12: Champa-colla-manbi; 13: Champa-colla; 14: Nagunen; 15: Ragunshen-masung-ahum-naibi; 16: Heijao-angouba; 17: Champa-colla-angangbi; 18:Teralaphoi; 19: Morpi; 20: Hei; 21: Hangou; 22: Masung-ahum naibi; 23: Meitei-hei; 24:Yensang-chabi; 25: Morshell; 26: Utteibi; 27: Ngalai

Table 2. Details of RAPD–PCR amplification obtained from the 27 banana cultivars

UBC- RAPD Primer set #5	Sequence (5′→3′)	Band size (bp)	Total band (No.)	PMB	% PM	PIC	R_P	MRP	EMR	MI
UBC-414	AAGGCACCAG	500-2000	9	9	100	0.39	3.0	0.33	9	3.51
UBC-417	GACAGGCCAA	600-1600	6	3	50	0.38	2.5	0.83	1.5	0.57
UBC-425	CGTCGGGCCT	300-3000	14	14	100	0.46	6.2	0.44	14	6.44
UBC-433	TCACGTGCCT	800-2000	5	3	60	0.59	1.0	0.33	1.8	1.06
UBC-433	TCACGTGCCT	800-2000	5	3	60	0.59	1.0	0.33	1.8	1.06
UBC-438	CGTCGGGCCT	300-3000	17	17	100	0.44	5.1	0.3	17	7.48
UBC-445	TAGCAGCTTG	250 -2000	16	16	100	0.32	4.8	0.3	16	5.12
UBC-447	CAGGCTCTA	500-2000	10	10	100	0.50	3.04	0.30	10	5.00
UBC-449	GAGGTTCAAC	400-1500	10	9	90	0.50	2.5	0.27	8.1	4.05
UBC-450	CGGAGAGCCC	400-1600	12	11	91.6	0.52	2.88	0.25	10.08	5.24
			99	92†	92.93*					

*Mean; †Total
PMB: Polymorphic band; % PM: Percentage of polymorphic band; PIC: Polymorphic information content; ABA: Average band; Rp: Resolving power; EMR: Effective multiplex ratio; MI: Marker index.

Analysis of Molecular Variance (AMOVA) among the Groups

The genetic variation generated by the nine RAPD markers within the five banana groups in the present study were 52.53% in AA group, 24.24% in BB group, 10.10% in AAB group, 4.04% in AAB group and 23.3% in ABB group (Table 4). Among the groups, AA exhibits the highest variability with 52.53% of percentage polymorphic band (PPB), 1.52 allelle number (*Ae*), 1.3 effective allele number (*Ao*), 0.18 Nei's genetic diversity (*He*) and 0.28 Shannon index (I). The least variability was found in AAB group with 4.04 % of PPB, 1.04 allelle number (*Ae*), 1.02 effective allele number (*Ao*), 0.01 Nei's genetic diversity (*He*) and 0.02 Shannon index (I). The estimates of Nei's genetic identity and genetic distance of pairwise comparison between the groups pairs (Table 5) showed that the highest identity existed between AA and BB group with a value of 0.84 and the least identity exhibited between AAB and BB group (0.69). The overall genetic variation was done in the present study using analysis of molecular variance (AMOVA) among five groups. The significant levels of differentiation (Φ_{PT}) was calculated with 1000 permutation among the groups and it showed that the genetic differentiation among the banana cultivars belonging to the five genome groups was statistically significant (P<0.001) (Table 6). Of the total molecular variance, 70% variation was found in among the groups and 30% variation within the populations (Fig. 2).

Genetic Relationships, Cluster Analysis and Principal Component Analysis

A pairwise comparison of the 27 samples shows that the similarity coefficient ranged from 0.55 between the cultivars 'Kharam-laphu' and 'Champa-colla-manbi' to 1.0 between 'Jahaji' and 'Grand naine'. The dendrogram obtained by using (UPGMA) revealed clustering of the 27 banana cultivars into three major clusters (Fig. 3). The first major cluster has two of the cultivars belonging to AA genome (Korbot and Changbi-mara-chatpi) while

Table 3. Details of genetic differentiation obtained from RAPD analysis of the 27 banana cultivars

Samples	Primers	Overall, Ht	Overall, Hs	Overall, Gst	Overall Nm
27	9	0.245†±0.025*	0.0793†± 0.003*	0.676 *	0.238*

*Standard deviation; † Mean

Overall Ht: heterozygosity; Hs: expected within population heterzygosity; G_{st}: genetic differentiation; Nm: gene flow from Gst.

Table 4. Analysis of genetic variation generated by RAPD markers in the 27 banana cultivars belonging to 5 genome groups

Genome group	Percentage polymorphic band (PPB)	Allele number (Ao)	Effective allele number (Ae)	Nei's gene diversity (He)	Shannon index (I)
AA	52.53	1.52	1.30	0.18	0.28
BB	24.24	1.24	1.17	0.09	0.14
AAA	10.10	1.10	1.06	0.03	0.05
AAB	4.04	1.04	1.02	0.01	0.02
ABB	23.30	1.23	1.09	0.05	0.09

Table 5. Measure of Nei's (1972) genetic identity (above diagonal) and genetic distance (below diagonal) between the 27 banana cultivars belonging to the 5 genome groups studied

Genome group	AA	BB	AAA	AAB	ABB
AA	****	0.84	0.80	0.77	0.80
BB	0.16	****	0.74	0.69	0.74
AAA	0.21	0.29	****	0.77	0.78
AAB	0.25	0.37	0.25	****	0.80
ABB	0.22	0.29	0.24	0.22	****

Table 6. Details of AMOVA between the banana groups generated from the RAPD profiles of banana cultivars studied

Source	df	SS	MS	Est. var.	Percent	PT	p value
Among genome groups	4	217.293	54.323	10.598	70	0.703	0.001
Within genome groups	22	98.410	4.473	4.473	30		

df: degree of freedom; SS: sum of square; MS: mean of square; Est.var: estimated variance; percent: percentage of variance.

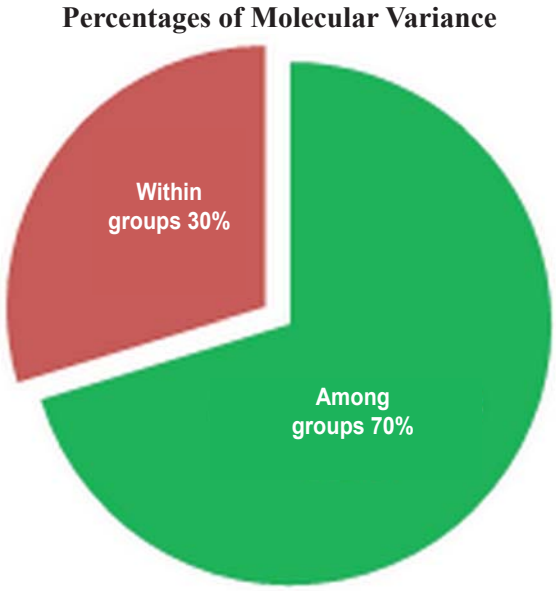

Fig. 2. Distribution of molecular variance within and among the genome groups obtained from the RAPD data

the second major cluster has been grouped into three minor clusters with the first minor cluster comprises of five triploid (AAA) cultivars ('Grandnaine', 'Jahaji', 'Des-laphu', 'Chingchup' and 'Mizoram-laphu') which are all AAA genome group, the second minor cluster comprises three AAB cultivars ('Champa-colla-manbi', 'Champa-colla' and 'Nagunsen') and the third minor cluster with 13 ABB cultivars ('Ragunsen-masung-ahum-naibi', 'Heijao-angouba', 'Champa-colla-angangbi', 'Teralaphoi', 'Morpi', 'Hei', 'Hangou', 'Masung-ahum-naibi', 'Meitei-hei', 'Yensang-chabi', 'Morshell', 'Utteibi' and 'Ngalai').

The third major cluster was subdivided into two clusters- the first minor cluster represented a lone AA genome cultivar ('Noneh-laphu') while the second cluster has three cultivars of BB genome ('Luine', 'Changbi' and 'Kharam-laphu').

The principal component analysis represented by the 2-D projection diagram (Fig. 4) was also in accordance with the dendrogram, projecting 'Noneh-laphu' (AA) near to BB genome group cultivars ('Changbi', 'Luine', 'Kharam-laphu') and far from other AA genome cultivars- 'Korbot' and 'Changbi-mara-chatpi'.

Discussion

In the present study the selected nine RAPD markers generated a total of 99 bands from the 27 selected banana cultivars, with an average of 14.14 bands per primer. Out of the 99 bands amplified, 7.07% were monomorphic and 92.93% were polymorphic bands. The generation of high level of polymorphic bands indicated the existence of a large genetic variability in the banana samples studied. Among the primers, UBC-438 produced the highest number of bands (17) and was the best in terms of detecting polymorphism (100%) with the Rp of 5.1 and MI of 7.48 respectively.

Analysis of the RAPD profiles revealed a high degree of genetic diversity as evident from the difference between the values of expected heterozygosity (Ht)

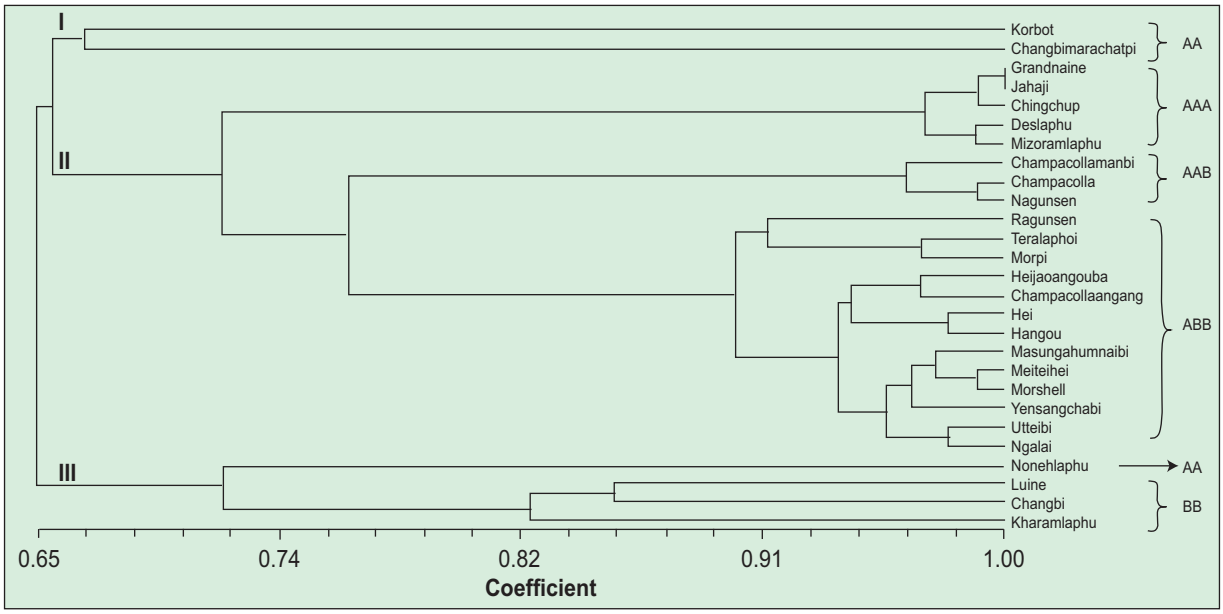

Fig. 3. Dendrogram obtained from the analysis of 5 different genome groups using RAPD data from 27 banana cultivars

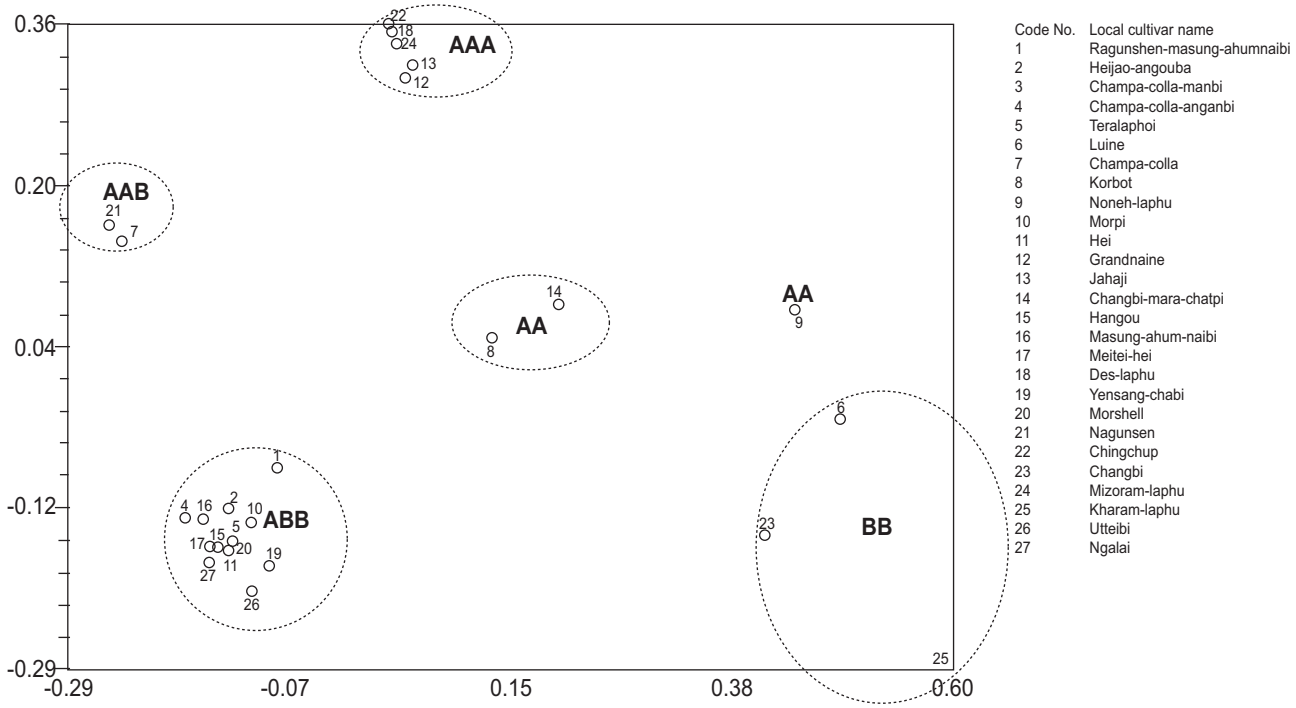

Fig. 4. 2-D view of Principal component analysis (PCA) based RAPD data using similarity (SM) coefficient of the different 27 banana cultivars

with 0.24 and homozygosity (Hs) of 0.07. The genetic differentiation (G_{st}) value of 0.67 and gene flow (Nm) value of 0.23 revealed the existence of high genetic differentiation and low gene flow among the cultivars of different genome groups studied. Similar observation has been reported by Resmi *et al.,* (2016) in banana cultivars from Southern India with a G_{st} value of 0.69. Lamare and Rao (2015) reported higher proportion of genetic variation among 25 *M. acuminata* genotypes from Meghalaya state of NER of India with a G_{st} value of 0.451. The source of difference may be attributed to its parthenocarpic nature and wide range of ecological condition within the distribution area and its adaptability to various environmental conditions. Studies on 40 cultivars of *M. balbisiana* by Oreiro *et al.* (2006) also revealed high levels of genetic differentiation with Ht values of 0.411. The analysis of molecular variance showed that high level of genetic variation occurred among the groups (70%) than within the groups (30%). Among the five genome groups studied, AA group showed the highest genetic variation with high Shannon index (I) value of 0.28, Nei's gene diversity (He) 0.18, effective allele number (Ae) 1.30 and PPB 52.5%. Similar observation has been reported by Resmi *et al.* (2011; 2016). Creste *et al.* (2003) postulated that high level of genetic diversity in AA diploid cultivars may be because of limited fertility and heterozygosity for chromosome structural abnormalities, maintained by vegetative propagation. Grapin *et al.* (1998) also reported that heterozygous condition mostly occurred in microsatellite loci in cultivated diploids. Ge *et al.* (2005) reported that heterozygosity may have occurred due to somatic mutation that accumulates genetic variation. Nei's genetic identity showed that the highest similarity occurred between the two wild types (AA groups and BB groups) with a value of 0.84 and lowest between BB groups and AAB groups (0.69). Similar observation has been made by Mukundakumar *et al.* (2013) in wild *Musa* with high level of genetic diversity (H=0.47, Shannon index=0.66, Ae No.=1.89). In the present study, analysis of the genetic relationship between the 27 cultivars based on RAPD markers revealed that, with the exception of 'Noneh-laphu' (AA) which is grouped along with BB group, all the remaining cultivars were clustered according to their genome groups which is not conforming to the findings of Pillay *et al.* (2000). The inclusion of the A genome containing cultivar, 'Noneh-laphu' (AA) within cluster of BB genome and its wide separation from other AA genome group cultivars is, however, explained by the fact that 'Noneh-laphu' may have undergone repeated selection/mutation that resulted in genome far from its ancestral diploid progenitors

(Bhat and Jarret, 1995; Resmi *et al.*, 2016). This finding is supported by the fact that Noneh laphu shares many similar morphological characters like plant height, male bud and presence of seed in fruit compared to remaining AA genome group cultivars like 'Korbot' and 'Changbi-mara-chatpi'.

The present study showed the usefulness of RAPD markers for the analysis of genetic variability, the distribution of variability and the genetic relationship present within the *Musa* germplasm of NER of India, which is an important region of banana diversity and origin. The ability of this marker to differentiate and analyse the cultivars of A and B genomes efficiently will be useful for characterization and understanding of the potential banana genetic resources for breeding and conservation programmes.

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