International Journal of Advanced Multidisciplinary Research
ISSN: 2393-8870
www.ijarm.com
DOI: 10.22192/ijamr Volume 3, Issue 11 -2016

Research Article

DOI: http://dx.doi.org/10.22192/ijamr.2016.03.11.005

Identification of cotton (*Gossypium* spp.) genotypes by Single Sequence Repeat (SSR) Markers

S.Ezhilkumar* and S.Padmavathi

Department of Genetics and Plant Breeding, Faculty of Agriculture, Annamalai University, Annamalai Nagar, Tamil Nadu, India
*Corresponding Author: *sezhil_5000@yahoo.co.in*

Keywords

Cotton,
Genetic variability,
SSR,
DNA polymorphism.

Abstract

Cotton (*Gossypium* spp) is one of the most important commercial fiber and oil yielding crop in India. Genetic variability and relationship between varieties are of important for cotton breeding. SSR markers were used for identification and genetic diversity analysis of cotton genotypes. 10 cotton genotypes were subjected to SSR analysis using 10 SSR primers. PCR products were subjected to 1.0% agarose gel electrophoresis and banding patterns were compared. 10 SSR primers produced a total of 204 bands, with 35 polymorphic markers, which showed 17.2% polymorphism. All the 10 SSR primers produced scorable bands and only 5 primers produced polymorphic bands. The SSR primer BNL-3590 produced one unique marker in the cotton genotype G.Cot-20 which is used for identification. A dendrogram constructed from SSR data classified 10 cotton genotypes into two major clusters and the similarity between the genotypes varied with a range of 72 to 99%. Among the 10 SSR primers studied, 4 primers could identify the genotypes individually. SSR technique was thus found to be efficient method for detecting DNA polymorphism and useful for varietal identification in cotton genotypes.

Introduction

Cotton (*Gossypium* spp.) is the most important textile fiber crop and the second most important oil seed crop in the world. Cotton is considered as "king of fiber crop". It plays an important role in national and international economy. It is grown globally over 33.1 m ha with a total production of 116.67 million bales of 480 lb and the productivity of 760 per ha (Anon, 2014). India is the second largest cotton producer in the world next to China with cultivable area of 115.53 lakh ha with the production of 375 lakh bales of 170 kg and the productivity of 552 kg/ha (Anon, 2014). Cotton belongs to the family Malvaceae and the genus *Gossypium* with about 49 species (Percival and Kohel, 1990). Of the 49 species, only four species are cultivated in India.

Gossypium arboreum and *G. habaceum* belong to the old world diploid group, whereas, *Gossypium hirsutum* and *G. babadense* belongs to the new world tetraploid group. India is the only country in the world cultivating all the four cultivated species of cotton on commercial scale.

Crop improvement programmes in India have developed a large number of varieties in the last 30 years and originated from interspecific crosses of *G. hirsutum* resulted in the development of varieties with narrow genetic base.

Varietal characterization and identification are important for plant breeders, seed industry and seed certification

agencies with the objective of determining the extent to which a seed sample conforms to a given cultivar and assure the quality of seed marketed to the consumer. Traditionally, varieties are identified based on morphological features such as plant height, leaf size, leaf shape, leaf colour, pollen colour, anther colour, fruit size and fruit shape etc. Though the morphological markers are the indicative of genotypes, but represented by few loci only because, there is not a large enough number of characters are available and they can also be affected by environmental factors and growth practices. Biochemical markers can provide varietal profile, because the variation for these markers is ubiquitous. Though the proteins are the products of the primary transcripts of DNA, environmental factors can affect qualitative and quantitative levels of protein.

In recent years, limitations of morphological and biochemical markers has been overcome by molecular markers. Molecular marker is a powerful PCR based technique indicates the differences in the nucleic acid sequences at a particular location or locations in the genome. It has high discrimination power enabling detection of closely related genotypes. Molecular markers are independent of external factors with high rate of polymorphism. There are number of molecular marker technologies that are eventually used for varietal characterization *viz.*, Random amplified polymorphic DNAs (RAPD), Restriction fragment length polymorphism (RFLP), Amplified fragment length polymorphism (AFLP), Single sequence repeat (SSR) and Inter simple sequence repeat (ISSR).

Among all these techniques, simple sequence repeat (SSR) markers have been used for varietal identification in cotton genotypes. SSR is typically, the repeat units of 1-6 nucleotides and SSR analysis is performed by using pairs of specific primers flanking tandem arrays of microsatellite repeats. SSR markers are co-dominant, multi-allelic nature, relative abundance and good genome coverage. SSR variation is due to slippage of DNA polymerase during replication or unequal crossing over resulting in differences in the copy number of the core nucleotide sequence (Yu and Kohel, 1999). SSR markers are more discriminative, reliable and repeatable and they could potentially be standardized more easily for DUS testing.

The use of molecular markers for evaluation of genetic diversity is receiving much attention than morphological characterization. SSR marker provides an unlimited number of markers which can be used for various purposes like cultivar analysis and cultivar identification in most crop plants. DNA finger printing and genotyping of cotton varieties have already been conducted in cotton (Islam *et al*., 2012). Keeping in view the above, the present investigation was planned to study molecular characterization of cotton (*Gossypium* spp.) genotypes through SSR markers.

Materials and Methods

Plant materials

The experimental materials consisted of 10 genotypes of cotton which were collected from Central Institute for Cotton Research, Nagpur, India.

DNA extraction

Total genomic DNA was extracted by CTAB method (Saghai-Maroof *et al.*, 1989) with slight modification. Seeds of different varieties were sown in pots maintained in green house. Five grams of fresh young leaves were collected from 3-4 weeks old seedlings for DNA isolation. The leaf samples were ground into fine powder using liquid nitrogen in a pre-chilled pestle and mortar. The powder was transferred into 2 ml eppondorf tube containing 1 ml extraction buffer. The solution was vortexed for 30-40 seconds, centrifuged at 4000 rpm for 10 minutes. Supernatant was discarded and 600 µl lysis buffer was added. The solution was incubated at 65°C for 60 minutes in a water bath. Chloroform and isoamyl alcohol solution was prepared in 24:1 and added to the tubes at 500 ml per tube. The solution was inverted and centrifuged at 8000 rpm for 10 minutes. The aqueous supernatant solution was pipetted out and transferred to a fresh eppondorf tube. Ice chilled isopropanol was added at 10 ml to each tube. It was mixed by inverting and kept at 20°C for overnight. The solution was centrifuged at 8000 rpm for 10 minutes. Supernatant was decanted and the pellet was retained. The pellet was washed with 70% per cent alcohol for 30 minutes and centrifuged at 8000 rpm for 20 minutes recovering the pellet. The pellet was allowed to dry at room temperature until the alcohol is completely evaporated. The pellet was re-suspended in 20 µl of *Tris*-EDTA buffer and incubated at 4°C. SSR analysis was done individually with 10 SSR primers.

The amplification products along with 2 µl of loading dye (bromophenol blue) were separated on 1.0% agarose gel at 80 volts using 1X TAE buffer of pH 8.0 containing ethidium bromide (0.5 µl/1/10 ml of gel). The gels were viewed under UV-trans illuminator and photographed for documentation. A 1 kb DNA ladder was used as a molecular size standard.

SSR analysis

A total of 10 SSR primers with the following sequences were chosen randomly across the cotton marker database. The SSR primers used in the analysis were given in Table 1.

Table 1: List of SSR primers and their sequences used in the study

PRIMER	FORWARD	REVERSE
DPL0039	CCGTCATATCAGTGTCATACAGGT	AGTCGCTACAACTTCTTCCCTTC
BNL2495	ACCGCCATTACTGGACAAAG	AATGGAATTTGAACCCATGC
JESPR151	CTGGACTAAAAACCTTAACTGG	CTCGATTCTAACTCAATCACG
JESPR300	CGCATCACAAACCAAACAC	CGGAAAATGATGATGATGAAGAAG
BNL530	CGTAGGATGGAAACGAAAGC	GCCACACTTTTCCCTCTCAA
BNL2709	AGTGGACAAACAATCTTTTTGG	ACATAAGGAAAGGAACAAGAAGC
BNL3590	TCTTCCCTCTCTTTCTCTTTCG	ACACGGAAGACCAACCAAGT
BNL4029	CGAGGGATAGAGAGGACGAA	GTTTGATTATTATGCTTGTAAAGTTACC
BNL1061	GCTTGTCATCTCCATTGCTG	TAGCCCGGTTCATGTTCTTC
DPL0209	GAAGGAACCTCGTGATTATTTGAG	GACCGGTAGACAGAGATGAGAAAT

Polymerase chain reaction

SSR with 25 µl reaction volume was carried out in 200 µl tube. Reaction mixture contained 400 ng of genomic DNA, 2-5µl of reaction buffer with 1 mM $MgCl_2$, 200 µM of each dNTPs, 20 ng of SSR primer, 1 U Tag polymerase. The thermocycles was programmed as follows.

PCR programming for ISSR

An initial denaturing step of 5 minutes at 94°C, 40 cycles consisting each of denaturing step of 1 minute at 94°C, primer annealing step of 45 seconds at 40°C and primer extension step of 2 minutes at 72°C were given. Final step of 5 minutes at 72°C was given for polishing the ends of PCR products.

Agarose gel electrophoresis

The products of reactions were electrophoresed on 1.0% agarose gel containing 0.1 µg/µl of ethidium bromide for about 5 hours at 60 volts. Gel was photographed under UV light with gel documentation.

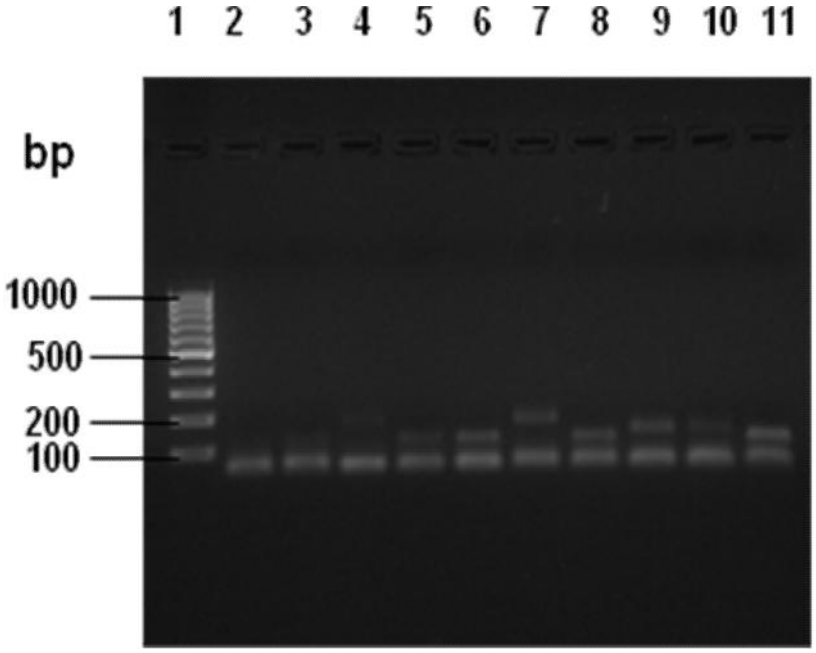
Data analysis

The molecular size of the each fragment was calculated using gene ruler 100 bp DNA ladder as standard. The DNA fragments amplified by a given primer was scored as present (1) or absent (0) for all the genotypes studied. Both monomorphic and polymorphic markers were included in the analysis and only reproducible fragments were considered. Coefficient of similarity among cultivars was calculated according to Nei and Li (1979). A dendrogram, based on these similarity coefficients was constructed by using Unweighted Pair Group Method of Arithmetic Means (UPGMA). The similarity matrix was analyzed by using NTSYS-PC 2.02 and clustered with UPGMA algorithm to determine the genetic relationship among the 10 genotypes of cotton.

Results

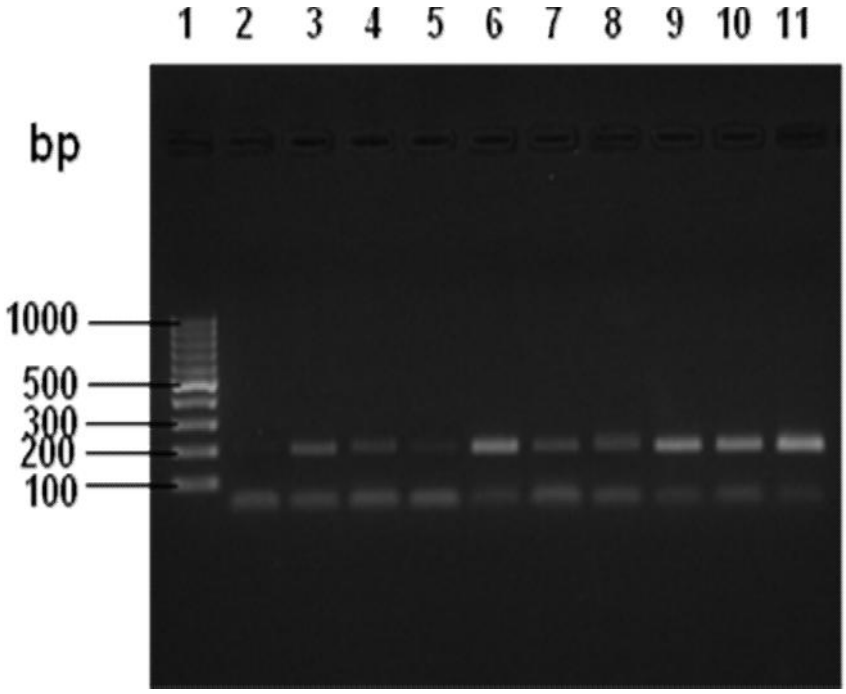
The results obtained in the present study are given below.

DNA of 10 cotton varieties was amplified with 10 SSR primer pairs. All the 10 genotypes with the 10 primers revealed a unique banding pattern and it can be used for varietal identification. Different primers produced a different level of polymorphism among the 10 different varieties studied (Figs. 1 and 2).



L1	Kanchana	L6	G.Cot-20
L2	Khandwa-2	L7	G.Cot-15
L3	Khandwa-3	L8	KC-3
L4	JK-4	L9	NH-545
L5	Anjali	L10	MCU-10

Fig 1. SSR analysis of 10 cotton genotypes with primer JESPR 2709



L1	Kanchana	L6	G.Cot-20
L2	Khandwa-2	L7	G.Cot-15
L3	Khandwa-3	L8	KC-3
L4	JK-4	L9	NH-545
L5	Anjali	L10	MCU-10

Fig 2. SSR analysis of 10 cotton genotypes with primer JESPR 151

Initially 10 SSR primers pairs were screened and all the 10 primers produced scorable bands. Out of 10 SSR primers, 5 SSR primer pairs were polymorphic one. A total of 204 DNA fragments were amplified. Out of 204 amplified fragments, 35 fragments were found to be polymorphic with 17.2% polymorphism. Average number of polymorphic loci amplified per primer was 3.5. The size of the amplified fragments varied with different primers. The size of SSR fragments was between 10 bp (BNL 2495, JESPR 300) to 400 (DPL 0039). The total number of markers and the number of polymorphic markers generated by each primer was given in Table 2.

Table 2. SSR bands and their characteristics generated by 10 SSR primers in 10 genotypes of cotton

Sl.No	Primers	Total number of bands	Number of polymorphic bands	Percentage polymorphism	Band size
1.	DPL0039	30	-	00.00	100-400
2.	BNL4029	10	-	00.00	100-100
3.	JESPR151	19	9	47.30	100-200
4.	JESPR300	20	-	00.00	100-200
5.	BNL530	20	-	00.00	100-200
6.	BNL2709	18	8	44.40	100-200
7.	BNL3590	20	1	05.00	100-500
8.	BNL2495	19	9	47.30	100-200
9.	BNL1061	30	-	00.00	100-200
10.	DPL209	18	8	44.40	100-200

The SSR primer DPL 0039 and BNL 1061 generated maximum number of bands (30) and the SSR primer BNL 2495 generated the least number of bands (10). Out of the five polymorphic primers, the primer JESPR 151 and BNL 2495 generated 9 bands each and the primer BNL3590 generated one polymorphic band. None of the primers was individually so informative as to differentiate all the studied genotypes. The SSR primer BNL 3590 produced one unique band in G. cot.-20 at 500 bp. The primer BNL 2495 produced one unique band at 200 pb, by the absence of specific band at 200 pb. Similarly the primer JESPR 151 could identify the genotype Kanchana by the absence of specific at 200 bp.

Cluster analysis

Nei and Lei (1979) coefficient method was used to generate similarity matrix (Table 3).

Table 3. Average estimates of genetic similarities between 10 cotton genotypes using 10 SSR primers

	kanchana	Khandwa -2	Khandwa -3	JK-4	Anjali	G.Cot-20	G.Cot-15	KC-3	NH5-45	MCU-10
Kanchana	1.0000									
Khandwa 2	0.7059	1.0000								
Khandwa3	0.8235	0.7647	1.0000							
JK-4	0.9333	0.6471	0.8750	1.0000						
Anjali	0.7895	0.7368	0.8421	0.7368	1.0000					
G.Cot-20	0.7895	0.7368	0.8421	0.7368	0.9000	1.0000				
G.Cot-15	0.6316	0.6667	0.7778	0.6667	0.8421	0.7500	1.0000			
KC-3	0.7895	0.7368	0.8421	0.7368	1.0000	0.9000	0.8421	1.0000		
NH-545	0.8333	0.7778	0.8889	0.7778	0.9474	0.9474	0.7895	0.9474	1.0000	
MCU-10	0.7368	0.6842	0.7895	0.6842	0.9474	0.8500	0.8889	0.9474	0.8947	1.0000

In ISSR analysis, the genotypes Anjali and NH-545, G. cot-20 and NH-545, KC-3 and NH-545, Anjali and MCU-10, KC-3 and MCU-10 showed maximum similarity of 95% followed by Anjali and G.Cot-20, G.Cot-20 and G.Cot-15 with of similarity of 90% among the 10 genotypes of cotton. The cotton genotypes Kanchana and G.Cot-15 showed least pair size similarity of 63% followed by Khandwa-2 and JK-4 with 64% among the 10 genotypes of cotton.

By using UPGMA cluster analysis, dendrogram was generated for SSR (Fig 3).

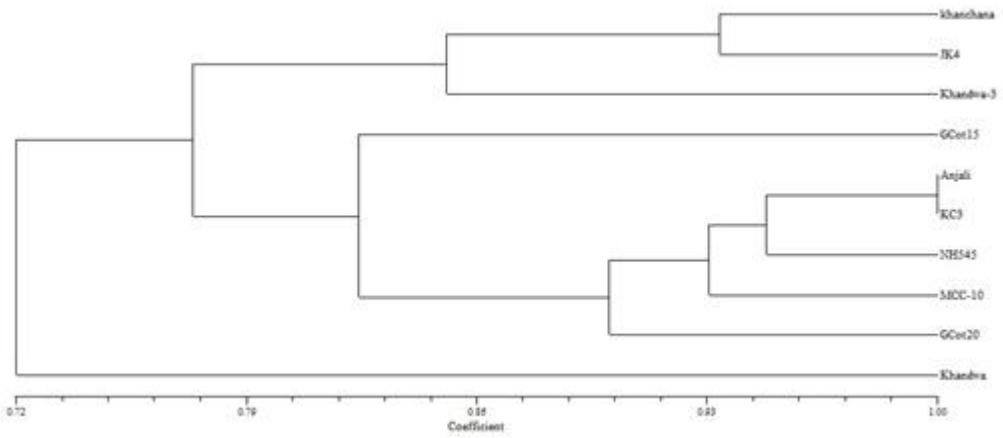


Fig. 3 UPGMA dendrogram showing relationship of 10 cotton genotypes by 10 SSR markers

In SSR analysis, the genotype Khandwa-2 was found to branch out from the rest of the genotypes. It showed 72% similarity with the rest of the genotypes studied. The dendrogram separated 10 cotton genotypes into two main clusters I and II with an average of 72% similarity. Main cluster I consisted of a single genotype Khandwa-2. The main cluster II consisted of 9 genotypes and these were divided into two sub cluster, II A and II B. The sub cluster II A consisted of 3 genotypes and it was divided into two sub subcluster II Ai and II Aii. The sub subcluster II Ai consisted two genotypes *viz.*, Khanchana, and JK-4 and the sub subcluster II Aii consisted of a single genotype Khandwa-3. The sub subcluster II Bi consisted of a single genotype G.Cot-15. The sub subcluster II Bii consisted of 5 genotypes *viz.*, Anjali, KC-3, NH-545, MCC-10 and G.Cot.-20. The similarity between genotypes varied with a range of 72 to 100%.

Unique identification of cotton genotypes

SSR markers were used for unique identification of 10 cotton genotypes. Unique markers were located across all the primers that individually identified each of the genotypes. Details of the genotype specific marker generated by different primers are given in Table 4.

Table 4. Unique markers present or absent 10 cotton genotypes

Marker	Present/Absent	Genotypes identified
JESPR151	(–)	Khanchana
BNL3590	(+)	G. Cot.-20
BNL2495	(–)	Khanchana
DPL209	(–)	G. Cot.-15 Khandwa-2

About 5 cotton genotypes were identified based on the presence or absence of unique marker. One genotype could be identified on the basis fof presence of single unique marker and other 4 genotypes were identified based on the absence of single marker present in all the genotypes. Genotype identified due to the presence of unique marker was G.Cot.-20 and genotypes identified by the absence of specific markers were Kanchana, G.Cot.-15 and Khandwa-2.

Discussion

Characterization of plant germplasm using molecular techniques has an important role in the management and utilization of plant genetic resources (Karp, 2002). It enhances on selection of diverse parents to widen the gene pool (Fu, 2006). These characterizations have provided useful information for understanding the genetic diversity and structure of cotton gene pools. Thin information could be incorporated into effective management of germplasm in cotton breeding program for control of genetic diversity.

SSR markers in large amount have raised more effort in molecular characterization of cotton germplasm. In the present study, Genetic diversity was observed in 10 cotton genotypes using 10 SSR primers. 10 SSR primers were able to show genetic diversity among the 10 cotton genotypes.

It has been reported in earlier studies that SSR markers target only one locus at a time and are useful source of observing molecular diversity (Lie *et al.,* 2006).

In the present SSR analysis, all the 10 SSR primers produced scorable bands. Out of the 10 SSR primers, five primers were polymorphic one. 10 primers generated a total of 204 bands. Among these, 35 were found to be polymorphic with 17.15% polymorphism. The SSR primers JESPR 151 and BNL 2495 gave the highest of 47.3% polymorphism, while the SSR primer BNL 3590 showed least of 5% polymorphism. Contradictory and similar research findings were reported by several authors. Lu and Myres (2002), Surgun *et al.* (2012), Beenish Ehsan *et al.* (2013).

Lu and Myres (2002) reported 13.5% polymorphism among the ten varieties of cotton. Sungun *et al.* (2012) reported 18.1% polymorphism between Turkish cotton genotypes. Amrean Ijaz *et al*., (2013 studied 20 genotypes of cotton using 31 SSR primers and showed 41.0% polymorphism. Dahab *et al*, (2013) studied 50 cotton genotypes using 93 SSR primers producing 147 polymorphic bands out of 241 total bands with 57.5% polymorphism, while Abdellatif *et al*, (2012) studied 28 cotton genotypes with 6 SSR primer pairs generating 44 polymorphic bands out of 52 total bands with 84.6% polymorphism. Sundar *et al.* (2014) studied SSR analysis of 156 cotton varieties with 38 SSR primers producing 86 polymorphic bands out of 120 total bands with 59.7% polymorphism. So, in the present study, a low level of polymorphism (17.2%) was observed and it may be due to narrow based genetic material used in the study that belonged to the same cotton species, which indicating the fact that more commonly cultivated genotype have relatively narrow genetic base, which may be true due to the fact that universal parents are usually used for the development of new varieties of cotton.

In the present study, the DNA fragments were amplified in the range of 100-500 bp. Average number of loci per genotype were about 20.4 and average number of loci per primer was 20.4. Average number of alleles per locus was 2.04. The average number of allies per locus of the present study was comparable with that of previous study in cotton. Bertini *et al.* (2006) reported 2.13 alleles per microsatellite locus using 31 primer pairs in 53 cotton varieties. Islam *et al.* (2012) reported 2.63 alleles/locus using 3 SSR primer pairs in 8 varieties of cotton (Candida *et al. (*2006) reported 2.13 alleles/locus using 31 SSR primer pairs in 53 cotton genotypes. Guiterrez *et al.* (2002) reported 2 alleles/locus using 60 primer pairs producing 139 alleles in 100 cotton genotypes. Liu *et al.* (2000) reported 5 alleles/locus using 15 primer pair in 62 cotton genotypes. Ehsan *et al.* (2013) reported 1.22 alleles/locus using 31 SSR primer sets in 20 genotypes with the band size of 100-500 pb. The possible reason for this result may be due to the less number of diverse cotton varieties used in this study.

Conclusion

The conclusion drawn from the present investigation are as follows:

1. 10 SSR primer pairs selected in the present study, generated 17.1% polymorphism. Five of 10 SSR primer pair produced polymorphism. So, these primers can be used for identification of these cotton genotypes.

2. SSR analysis produced very low level of polymorphism (17.1%) is due to narrow genetic base of genetic material studied.

3. The present study identified some unique primers for identification of cotton genotypes. The SSR primer BNL3590 could identify the genotype G. Cot. 20.

References

1. Anon., (2013). Annual report. All India Coordinated Crop Improvement Programme. 2013-14
2. Bertini, C.H.C.de Magalhaes., I. Schuster, T. Sediyama, G. Barros and M.A. Moreira. (2006).

Characterization and genetic diversity analysis of cotton cultivars using microsatellites. ***Geneics and Molecular Biology***, 29 (2): 321-329

3. Candida, H. C., D. M. Bertini, I. Schuster, T. Sediyama, E. G. D.Barrosi and M. A. Moreira. (2006). Characterization and genetic diversity analysis of cotton cultivars using microsatellites. ***Genetics and Molecular Biology,*** 29(2): 321-329
4. Ehsan, B., A. Haque, M. Younas, T. Shaheen, T. Huma, S. Sattar, S. Idrees and Z. Iqbal. 2013. Assessment of genomic diversity of cotton (*Gossypium hirsutum*) genotypes using simple sequence repeats markers through genetic analysis software. ***International Journal of Agriculture & Biology,*** 15(5): 968–972
5. Fu, Y.B. (2006). Genetics redundancy and distinctness of flax germplasm as revealed by RAPD dissimilarity. ***Plant Genetic Resource*s** 4, 177–124.
6. Gutierrez, O. A., S. Basu, S. Saha, J. N. Jenkins, D. B. Shoemaker, C. L. A. Cheatham and J. R. J. C. Mccarty, 2002. Genetic distance among selected cotton genotypes and its relationship with F_2 performance. ***Crop Science***, 42: 1841-1847
7. Ijaz, A., Sadia Ali, U. Ijaz, Smiullah and T. Shaheen. 2013. Molecular characterization of cotton using simple sequence repeat (*SSR*) markers and application of genetic analysis. ***International Journal of Genetics and Molecular biology,*** 5(4):49-53
8. Islam, M. N., Md. R. Molla, Md. M. Rohman, M. Hasanuzzaman, S. M. N. Islam and L. Rahman. 2012. DNA Fingerprinting and genotyping of cotton varieties using SSR markers. ***Not. Bot. Horti. Agrobo.,*** 40(2): 261-265
9. Karp, A. (2002). The new genetic era: will it help us in managing genetic diversity?, 43-56 p. In: Engels JMM, Rao VR, Brown AHD, Jackson MT (Eds.). Managing Plant Genetic diversity. International Plant Genetic Resources Institute, Rome, Italy.
10. Liu, S., R.G. Cantrell, McCarty-Jr JC, J.M.D. Stewart. 2000. Simple sequence repeat based assessment of genetic diversity in cotton race stock Accessions. ***Crop Science,*** 40:1459-1469.
11. Lu, H. J. and G. O. Myers. 2006. Genetic relationships and discrimination of ten influential upland cotton varieties using RAPD markers. ***Theoretical and Applied Genetics,*** 105:325–331
12. Nie, N. and W. Lei. 1979. Mathematiical model for studying genetic variation in terms of restriction endonuclease. ***Proc. Natl. Acad. Sci.*** 76:5269-5273
13. Percival, E. and R. J. Kobel. 1990. Distribution, collection and evaluation of *Gossypium*. ***Advances in Agronomy***, 44: 225-228.
14. Powell, W., M. Morgante, C. Andre, M. Hanafey, M.J. Vogel, S.V. Tingey and A. Rafalski, 1996. The comparison of RFLP, RAPD, AFLP and SSR (microsatellites) markers for germplasm analysis. ***Molecular Breeding***, 2: 225–238
15. Sheidai, M., Z. Noormohammadi, F. Shojaei - Jeshvaghani, S.G. Baraki, F. Farahani and O. Alishah. 2012. Simple sequence repeat (SSR) and inter simple sequence repeat (ISSR) analyses of genetic diversity in tissue culture regenerated plants of cotton. ***African Journal of Biotechnology***, 11(56): 11894-11900
16. Sundar, R., S. Shreekanth, H. M. Patil, P. Ranganath, S. M. Manjula and K. J. Pranesh. 2014. Genetic diversity analysis in cotton (*Gossypium hirsutum* L.*)* based on morphological traits and microsatellite markers. ***International Journal of Basic and Applied Biology,*** 1(1): 2349-5820
17. Surgun, Y., B. Col and B. Burun. (2003). 2012. Genetic diversity and identification of some Turkish cotton genotypes (*Gossypium hirsutum* L.) by RAPD-PCR analysis. ***Turkish Journal of Biology,*** 36: 143-150

Access this Article in Online

Website: www.ijarm.com

Subject: Agricultural Sciences

Quick Response Code

DOI:10.22192/ijamr.2016.03.11.005

How to cite this article:
S.Ezhilkumar and S.Padmavathi. (2016). Identification of cotton (*Gossypium* spp.) genotypes by Single Sequence Repeat (SSR) Markers. Int. J. Adv. Multidiscip. Res. 3(11): 62-69.
DOI: http://dx.doi.org/10.22192/ijamr.2016.03.11.005