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Molecular Marker Based Genetic Diversity Study of *Curcuma Caesia* Roxb. – A Mini Review

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Abstract

The availability of genetic resource provides an opportunity for plant breeders to develop new cultivars with desirable characteristics and these characteristics encompass various traits preferred by farmers (high yield potential, larger seed size, etc) as well as traits favored by breeders (pests and disease resistance). Genetic resources are also essential for conservation, as they provide a basis for identifying threatened species and developing conservation strategies. *Curcuma caesia* Roxb. (Zingiberaceae) is an underutilized perennial herb with bluish-black tuberous rhizome having the characteristic of aromatic and medicinal properties. There is an ample genetic diversity of *Curcuma* plants with medicinal importance around the world. Authentication and ensuring the genuineness of *Curcuma* are crucial aspects for marker-based analysis. Therefore, genetic variation serves as the base for the identification and characterisation of the plants. Molecular marker technology is widely used in genetic diversity analysis, germplasm resource identification, and molecular marker-assisted breeding technique. Multiple molecular markers have been used for characterization and identification of species for its sustainable utilization and conservation. The molecular investigation, which involves techniques like RAPD, SCAR, ISSR, and AFLP for fingerprinting, plays a crucial role in establishing molecular markers to assess the authenticity and diversity of plant species. Various marker analyses have demonstrated a significant degree of genetic variation among the species and these patterns tend to differ due to environmental influences and genetic factors. This review facilitates a complete information about the molecular-assisted genetic diversity study of *Curcuma caesia* Roxb. (Black turmeric) which may be used as a database while exploring the diversity analysis.

Keywords: *Curcuma Caesia*; RAPD; SSR; ISSR; EST- SSR

Abbreviations: ISSR: Inter Simple Sequence Repeats; AFLP: Amplified Fragment Length Polymorphism, SSR: Simple Sequence Repeats, RAPD: Random Amplified Polymorphic DNA; PIC: Polymorphic Information Content; UPGMA: Unweighted Pair Group with Arithmetic Mean; PCA: Principal Component Analysis; ESTs: Expressed Sequence Tags.

Introduction

The genus *Curcuma* belongs to the family *Zingiberaceae* is composed of 70-80 species of rhizomatous herbs. It is a taxonomically complicated genus and new species are yet to be explored. *Curcuma caesia* Roxb. is a perennial herb with bluish black rhizome and having extensive array of

pharmaceutical and medicinal applications [1]. Over few past decades, there has been a remarkable decline in the population of this plant. This decline can be attributed due to its overexploitation in various pharmaceutical and traditional medicines [2,3]. Diversity studies along with the structure of germplasm resources are used for accelerating different plant breeding program and helps in utilization and conservation of the species by finding taxonomic relations [4-6].

The use of molecular marker technology is prevalent in the analysis of genetic diversity, identification of germplasm resources, and molecular marker-assisted breeding [7]. Molecular markers such as Inter simple sequence repeats (ISSR), Amplified fragment length polymorphism (AFLP), Simple sequence repeats (SSR), Random amplified polymorphic DNA (RAPD) are mainly used for genetic diversity study [8]. Genetic diversity is an integral part for understanding evolutionary studies of the species, including, the intra specific regions which are not influenced by the environmental factors [7,9,10]. Therefore, this knowledge is crucial for knowing the genetics underlying biological diversity and can inform various fields like ecology, agriculture, and medicine [3]. To the best of knowledge, none of the review article on the molecular diversity study of *Curcuma caesia* is available in the public domain. Therefore, this review will help in preparing a molecular database of the widely used black turmeric based on its genomic organization.

RAPD Markers

The six accessions of *Curcuma caesia* were analysed for the genetic diversity by Ranemma, et al. 2017 [11]. A total of 15 primers were used and six of the primers (OPC-1, OPC-9, OPB-6, OPB-9, OPJ-1 and OPD-1) exhibited good amplification. In other hand, the remaining nine primers also showed medium amplification. The RAPD primers generated 74 bands among which 43 were polymorphic and 31 were monomorphic. 25% of polymorphism was shown in three primers (OPB-9, OPC-9, OPD-1, whereas, OPC-01 showed 20% of polymorphism followed by 16.6% of polymorphism in OPJ-1. The lowest percentage of polymorphism was shown by OPB-6 (12.5%). The polymorphism level of markers is compared by polymorphic information content (PIC). The highest PIC value was recorded in OPD-1. The genetic similarity between six accessions was calculated by preparing a matrix based on all amplified products, and a dendrogram was constructed using the Unweighted Pair Group with Arithmetic Mean (UPGMA) cluster analysis method with Jaccard's similarity coefficient. As a result, the accessions-1 and 4 showed the highest similarity of 64%, and the genetic distance between all six accessions ranged from 0.753 to 0.939, which was calculated using Jaccard's distance coefficient. Magar, et al. 2021 also analyzed genetic diversity of 22 accessions of *Curcuma amada*, *Curcuma longa* and *Curcuma caesia* using 10 RAPD markers. This study revealed 76.7% of genetic variability with RAPD markers [12]. Darwin software based dendrogram categorized accessions into three major clusters, at certain degree based on morphological characters. Based on the three clusters, *C. caesia* showed a distinct variability among the three species. RAPD analysis was performed on 30 accessions from five *Curcuma* species, namely *Curcuma latifolia*, *Curcuma malabarica*, *Curcuma manga*, *Curcuma Raktakanta*, and 13 conserved morphotypes of *Curcuma longa* by Hussain, et al. 2008 [13]. The RAPD data obtained in their study supported the morphological classification of these morphotypes. Furthermore, this study compared the efficiency of individual RAPD primers and demonstrated that RAPD markers were highly informative in distinguishing the germplasm of *Curcuma* species. On the other hand, Islam et al., 2005 reported that *Curcuma zedoaria* populations exhibit a high level of genetic diversity, possibly due to variations in ecological conditions. However, it should be noted that RAPD markers are known to exhibit lower levels of polymorphism compared to other molecular markers, such as microsatellites (also known as simple sequence repeats or SSRs) [14].

SSR Markers

Paw, et al. 2021 assessed a diversity study among the 78 high essential oil yielding (>1.5% w/w) of *C. caesia* using 45 SSRs. 53% of polymorphism were found among the lines. Variation among and within the population was analysed by AMOVA with 20% and 80 % respectively. A dendrogram was created using the Unweighted Pair Group with Arithmetic Mean (UPGMA) method to group the accessions into five clusters based on their geographical origins. These clusters were then verified using principal component analysis (PCA), which relied on the Jaccard's similarity coefficient. The findings indicated that the microsatellite markers could be effectively utilized to distinguish the genetic variations within the crop [15]. Akkaya and Buyukunal-Bal (2004) emphasized that markers with high polymorphic information content (PIC) tend to be more informative. The SSR markers CuMiSat-19 (PIC=0.98), CuMiSat-20 (PIC=0.97), CuMiSat-26 (PIC=0.97), CuMiSat-22 (PIC=0.96), and CuMiSat-27 (PIC=0.95) exhibited the highest PIC values in *Curcuma longa*. The PIC value reflects the diversity and frequency of alleles among the genotypes. The average PIC value for the markers in this study was 0.86, indicating that the SSR markers used were highly informative. A PIC value of 0.56 or higher is considered highly informative for genetic studies and can effectively distinguish the polymorphism rate of a marker at a specific locus [16].

EST- SSR Markers

Expressed sequence tags (ESTs) are valuable source for identifying simple sequence repeats (SSRs), which are useful in analyzing genetic diversity due to their abundance, polymorphism, and representation of functional regions in the genome. Sahoo, et al. 2021 reported a diversity study of nine *Curcuma* species by using 109 EST-SSR markers and resulted 33 polymorphic bands. These 33 polymorphic EST-SSRs were used to assess the distance and genetic resemblance among the *Curcuma* species. The value of Nei's gene diversity indicates the large scale production and conservation prioritization of *Curcuma caesia*. The population genetics resulted a genetic difference among the species. These SSR markers could also be utilized to identify morphologically similar *Curcuma* species [17].

ISSR Markers

ISSR markers are the PCR based markers that have been widely used in genetic studies due to their high level of polymorphism, reproducibility, and ease of use. Magar, et al. 2021 reported a genetic diversity study of 22 accessions of *C. amada, C. longa* and *C. caesia* collected from North East India using 42 ISSR. This study revealed a genetic variability of 78% [12]. The accessions were categorized into three major clusters using the Darwin software. The maximum genetic similarity was observed between *Curcuma amada* and *Curcuma longa*, while *Curcuma caesia* showed significant variability. Saha, et al. 2016 conducted ISSR fingerprinting to determine the genetic relationship of different species of *Curcuma* found in Tripura. The study focused on four species: *Curcuma amada, Curcuma caesia, Curcuma Longa*, and *Curcuma zedoaria*. Using 20 ISSR primers, able to generate 116 loci, which ranged in size from 200 to 5000 base pairs (bp). On average, each locus had 5.8 alleles and 1.6 effective alleles. The percentage of polymorphic bands, indicating genetic variation, was found to be 86.29, with an average of 5.15 polymorphic bands per primer. Their study revealed a high level of polymorphism and provided valuable insights into the genetic diversity within and between these species [18].

Conclusion

Curcuma caesia, also known as black turmeric, is native to Northeast India and Southeast Asia. It is known for its dark blue-black rhizomes, which are used in traditional medicine. The significant amount of genetic variation was attributed due to differences in geographical locations and climatic conditions. Marker-based genetic diversity studies play a crucial role in understanding the genetic variations within populations and their implications for various applications, such as breeding programs, conservation efforts, and evolutionary studies. By providing valuable information about allele diversity and frequency, these studies contribute to the development of strategies for genetic improvement, germplasm characterization, and population structure analysis. Overall, marker-based genetic diversity studies, particularly those employing highly informative markers like SSRs, RAPD offer powerful tools to unravel the genetic complexity within populations and aid in making informed decisions for various genetic applications. This information highlights the importance of preserving and studying the genetic diversity of this crop in order to develop strategies for its sustainable cultivation and conservation. The findings of this review are highly informative and will be useful to researchers seeking to develop effective strategies for the conservation and improvement of *Curcuma caesia.*

Conflicts of Interest

The authors declare that there is no conflict of interest regarding this study.

References

1. Singh S, Sahoo BC, Ray A, Jena S, Dash M, et al. (2021) Intraspecific chemical variability of essential oil of Curcuma caesia (Black Turmeric). Arabian Journal for Science and Engineering 46: 191-198.

2. Borah A, Paw M, Gogoi R, Loying R, Sarma N, et al. (2019) Chemical composition, antioxidant, anti-inflammatory, anti-microbial and in-vitro cytotoxic efficacy of essential oil of Curcuma caesia Roxb. leaves: An endangered medicinal plant of North East India. Industrial crops and products 129: 448-454.

3. Paw M, Gogoi R, Sarma N, Pandey SK, Borah A, et al. (2020) Study of anti-oxidant, anti-inflammatory, genotoxicity, and antimicrobial activities and analysis of different constituents found in rhizome essential oil of Curcuma caesia Roxb., collected from north east India. Curr Pharm Biotechnol 21(5): 403-413.

4. Jain A, Parihar DK (2019) Molecular marker based genetic diversity study of wild, cultivated and endangered species of Curcuma from Chhattisgarh region for in situ conservation. Biocatalysis and Agricultural Biotechnology 18(4): 101033.

5. Gepts P (2006) Plant genetic resources conservation and utilization: The Accomplishments and Future of a Societal Insurance Policy. Crop Science 46(5): 2278-2292.

6. Engelhardt KAM, Lloyd MW, Neel MC (2014) Effects of genetic diversity on conservation and restoration

potential at individual, population, and regional scales. Biological conservation 179: 6-16.

7. Agarwal M, Shrivastava N, Padh H (2008) Advances in molecular marker techniques and their applications in plant sciences. Plant Cell Reports 27: 617-631.

8. Ismail NA, Rafii MY, Mahmud TMM, Hanafi MM, Miah G (2016) Molecular markers: A potential resource for ginger genetic diversity studies. Mol Biool Rep 43(12): 1347-1358.

9. Nadeem MA, Nawaz MA, Shahid MQ, Dogan Y, Comertpay G, et al. (2018) DNA molecular markers in plant breeding: current status and recent advancements in genomic selection and genome editing. Biotechnology and Biotechnological Equipment 32(2): 261-285.

10. Mohanty S, Panda MK, Acharya L, Nayak S (2014) Genetic diversity and gene differentiation among ten species of Zingiberaceae from Eastern India. 3 Biotech 4(4): 383-390.

11. Ranemma M, Reddy SK (2017) Genetic Diversity and Similarity Studies in Different Accessions of Curcuma caesia Roxb. by RAPD Analysis. Trends in Biosciences 10(27): 5684-5689.

12. Magar SG, Meetei NT, Singh KN, Khanna VK, Chowdhury VK (2021) Genetic variability of Curcuma sp. accessions of North East Hill region of India using ISSR and RAPD markers. The Pharma Innovation 10(11): 2781-2787.

13. Hussain Z, Tyagi RK, Sharma R, Agrawal A (2008) Genetic diversity in in vitro-conserved germplasm of Curcuma L. as revealed by RAPD markers. Biol Plant 52: 627-633.

14. Islam MA, Meister A, Schubert V, Kloppstech K, Esch E (2006) Genetic diversity and cytogenetic analysis in Curcuma zedoaria (Christm) roscoe from Bangladesh. Genet Resour Crop Evol 54: 149-156.

15. Paw M, Borah A, Pandey SK, Baruah J, Begum T, et al. (2021) Simple sequence repeat marker based genetic diversity assessment amongst high essential oil yielding lines of Curcuma caesia Roxb. Genetic Resources and Crop Evolution 68(4): 1345-1358.

16. Akkaya MS, Bal EBB (2004) Assessment of genetic variation of bread wheat varieties using microsatellite markers. Euphytica 135: 179-185.

17. Sahoo A, Behura S, Singh S, Jena S, Ray A, et al. (2021) EST-SSR marker-based genetic diversity and population structure analysis of Indian *Curcuma* species: significance for conservation. Brazilian Journal of Botany 44: 411-428.

18. Saha K, Sinha RK, Basak S, Sinha S (2016) ISSR fingerprinting to ascertain the genetic relationship of Curcuma sp. of Tripura. Am J Plant Sci 7(2): 259-266.