RAPD Analysis of DNA Isolated from Turmeric Rhizomes Collected from Northeast India

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Abstract

Turmeric has been used as condiments and as medicine by traditional healers of northeast India. The rhizome contains various constituents including polyphenols, polysaccharides and alkaloids. A simple method was adapted for isolation, PCR amplification of DNA and the RAPD analysis was carried out. The results of the study are reported here.

Keywords: Turmeric; DNA; RAPD; Rhizomes; Northeast India

Introduction

Turmeric (Curcuma longa) is a rhizomatous herbaceous perennial plant of the ginger family, Zingiberaceae. India is a leading producer and exporter of turmeric in the world. The most important chemical components of turmeric are a group of compounds called curcuminoinds, which include curcumin and bisdemethoxycurcumin. The best studied compound is curcumin, which constitutes 3.14% (on average) of powder turmeric. Curcumin has antioxidant, anti-inflammatory, antiviral and antifungal actions.

There are different turmeric species available in northeast regions. Traditionally it is used for treatment of various physiological problems [1,2]. The northeast India has differences in topography and there are also vast differences in the climatic as well as in soil conditions in different areas of northeast regions. So there is a chance of genetic diversity as well as differences in curcumin content in the turmeric species grown in different environmental conditions.

The objective of this study was to study the genetic variation in samples of turmeric collected from three different states of northeast India. Total five samples were collected from and isolated DNA was amplified to study the variations. The results of the study in brief and the conclusions are reported here.

Materials and Methods

Plant samples were authenticated from Botanical Survey of India, Shillong, Meghalaya. Plant materials (5 accessions) were collected from mature rhizomes of turmeric from different areas of northeast India. Three states namely Arunachal Pradesh, Meghalaya and Assam having different topography. Three samples were collected from three districts of Assam namely Dibrugarh, Karbianglong and Tinsukia. The collected materials were immediately cleaned and the surface was sterilized. It was then transferred in the zip lock poly bags and stored at -20°C until DNA extraction.

DNA Isolation and PCR Amplification

Rhizomes were washed in sterile distilled water, scraped and sliced into thin pieces. The samples were then ground with the help of a mortar and pestle (4 g fresh tissue) in liquid nitrogen.

Modified CTAB (Cetyl Trimethyl Ammonium Bromide) method of Doyle and Doyle [3] was used for isolation of genomic DNA of turmeric. This protocol is suitable for isolating a good yield of good quality DNA from recalcitrant tissues such as rhizomes. DNA purity is a concern in extraction procedures.

RAPD Analysis

RAPD analysis of genome DNA was performed according to William et al. [4,5]. PCR products were visualized under UV light after electrophoresis in a 1% agarose gel, containing agarose, 1x TAE buffer and 3 μl/ml ethidium bromide for 50 minutes at 80V.

Results and Discussion

The present study revealed that the distinct genetic variation obtained from the various accessions of turmeric (Figure 1). Here in this study, we have successfully isolated DNA from fresh turmeric rhizomes and DNA purity was determined from the A260/280 ratio, averaged 1.83-1.89 in all samples. From RAPD study of different samples of Curcuma longa, it can be concluded that the good amplification was obtained with random decamer primer No.24 (GenNéi, Bangalore).

The RAPD analysis results demonstrated a wide genetic variation among the different turmeric accessions. A total of 5 distinct bands were recorded in which 1 band was monomorphic and 4 bands were polymorphic. The result also exhibited 80% polymorphism.
Figure 1: [A-D] Amplification of genomic DNA using primers 7,14,24 and 17.

Conclusion

This study reveals the genetic polymorphism among various accessions of turmeric available in northeast India. In this study only five accessions collected from different topographical area were used. Analysis of more accessions from different regions would provide genetic variations among the samples available in this region. Soil analysis and correlation of curcumin content with genetic variation would yield important scientific evidence and also would help in correlating the therapeutic efficacy of curcumin.

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