ABSTRACT

Agarwood plants (Aquilaria sp.) are known for the fragrant resinous wood. This economically important plant is used in the preparation of perfumes, traditional medicines and commercial products like joss sticks. Due to high demand, this valued agarwood plants population has been decreasing from their wild habitats over the years. The DNA markers are very useful in the precise identification of plant species and can be used in identification of individual species of Aquilaria. In addition, it is important to study endophytes of Aquilaria sp. in order to understand types of endophytes present in it. Plant endophytes are known to play an important role in the growth and development of their host plants. The main objective of this study was to identify DNA markers for the precise identification of 7 Aquilaria sp. and characterization of their bacterial endophytes. Total genomic DNA (gDNA) was successfully isolated from 7 species of Aquilaria; and by using 30 ng gDNA as template in each reaction, RAPD and microsatellite-polymerase chain reaction (PCR) analysis was carried out. All seven species of Aquilaria were analyzed by using 120 RAPD and 11 microsatellite primers to identify informative DNA markers for their precise identification. Surface decontaminated plant tissues (from leaf/petiole/stem) were used to isolate bacterial endophytes from 7 species of Aquilaria; and isolates were identified using PCR amplified 16S rDNA based method of bacterial identification. Ethyl acetate extracts from cell-free broth of isolates were screened for cytokinin-like compounds using cucumber cotyledon greening bioassay (CCGB). The results of RAPD and microsatellite-PCR analysis showed that 120 RAPD and 11 microsatellite primers gave 182 and 3 informative DNA markers, which could be useful in precise identification of 7 Aquilaria sp. Eighty one (81) endophytic bacterial isolates (EBIs) were isolated from 7 species of Aquilaria. The 16S rDNA based identification of EBIs suggests that they belong to 6 genera of bacteria. Result analysis suggests that there were 18 species of bacteria namely, Bacillus pumilus, B. licheniformis, B. arsenicus, B. tequilensis, B. subtilis, B. megaterium, B. aryabhattai, B. altitudinis, B. stratospericus, B. cereus, B. arbutinivorans, B. methylotrophicus, B. anthracis, Vibrio cholera, Rahnella aquatilis, Roseomonas mucosa, Acinetobacter radioresistens and Pantoea agglomerans within isolated 81 EBIs. Majority (92.2%) of EBIs were of Bacillus genus of bacteria. B. pumilus was the most dominant (36.4%) species within EBIs. The CCGB results showed that out of 81 EBIs, only 47 isolates broth extracts do have cytokinin-like activity. RAPD and microsatellite markers reported in

this study could be useful in identification of 7 studied *Aquilaria* sp. However, a suitable mechanism of EBIs usage needs to be identified to utilize them (leads from CCGB) for their applications in agriculture.