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**DEVELOPMENT OF A SPECIFIC MOLECULAR MARKER TO
IDENTIFY THE COCONUT FORM BODIRI**

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DEVELOPMENT OF A SPECIFIC MOLECULAR MARKER FOR IDENTIFICATION OF COCONUT FORM BODIRI

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Coconut palm, referred to as the tree of life, is one of the most valuable plants to man as every part of it is utilized to make something useful to the community. Copra, oil and desiccated coconut are its major products while copra meal, activated carbon, shell charcoal, coir and coir dust are also important products of coconut. It is a heritage of people in many tropical countries to use coconut as a principal food commodity as well as a source of foreign exchange. Coconut is an important constituent of the daily diet of the average Sri Lankan providing approximately 25% of the calorific requirement (David, 1984)

The coconut in Sri Lanka was categorized into three distinct varieties, tall (*typica*), dwarf (*nana*) and thembili or king coconut (*aurantiaca*) (Liyanage, 1958). Tall coconuts are the most commonly grown and commercially exploited. They are predominantly cross-pollinating, late bearing and producing nuts that vary from medium to large in size (Liyanage, 1958). In general, tall coconuts are hardy and can thrive in a range of environmental conditions. Dwarf coconuts, mostly grown for ornamental and breeding purposes, are predominantly self-pollinating and less adapted to harsh environments. Dwarf coconuts produce small nuts in large quantities with distinct colour forms. The intermediate, thembili or king coconut is also characterized by the sweet nut water (Liyanage 1958).

The present study aims at resolving a practical problem that often arises in obtaining true types of coconut forms for various purposes *via* open pollination. Bodiri is one such coconut form, which need true propagation because pure bodiri has a significant economic value as a beverage coconut. The quality of the nut water of tender bodiri has no difference from that of

king coconut, the most popular natural beverage in the country. In addition bodiri produces profusely, with nuts per bunch averaging over 50. In order to utilize its potential, widespread cultivation of bodiri in home gardens is necessary and to accomplish this the biggest constraint is supplying of pure bodiri planting material in large quantities.

Bodiri is often self pollinating but cross pollination is not preventable because the country lacks pure stands of bodiri except for just one block containing approximately 100 palms in one of the field gene banks of the Coconut Research Institute(CRI). Therefore, any bunch of bodiri coconuts is certain to have a significant proportion of cross-pollinated nuts. On the other hand production of seeds by artificial pollination is not feasible because it require an enormous number of skilled labor and thorough supervision to carry out pollination and obtain a considerable number of legitimate seeds. Developing a molecular marker to identify pure bodiri at the seedling stage is certainly a more economical and an accurate alternative to supplying growers with pure planting material. This will save enormous time and costs in production of seeds and better prospect for growers who have to at least five years to cull the illegitimate plants otherwise. This study was introduced to develop a suitable molecular marker to distinguish bodiri from other common forms of coconut.

The leaf samples were obtained from the palms conserved in the field gene bank of the CRI located at the Bandirippuwa Estate, Lunuwila. The plant materials used for extraction of DNA were tender tissues of the upper coconut leaf, spear leaf. This is because high yield of DNA is expected from such young leaves. Leaves selected for isolation of DNA were clean and fresh with no signs of any fungal infections. 24 coconut leaf samples were taken for the study, among that 6 tall (*typica*), 6 dwarf (*nana*), 6 king coconut (*aurantiaca*), and 6 bodiri. The DNA was extracted from each coconut leaf sample and subjected to the PCR amplification using for microsatellite markers. Here pre designed primers are available for the PCR amplification (Perera et al, 1998). The primers used in the assay are CAC 8, CAC 2, CNZ 6, CNZ 44, CAC 4 CAC 20, CNZ 4, CNZ 10, CNZ 29, CNZ 37, and CNZ11A10. These primers showed different levels of polymorphism and all the primers aptly amplified DNA. Most of the primers revealed the presence of allele of both dwarf and tall in bodiri. The primer CAC 8 failed to amplify a band specific to bodiri but was very much capable for distinguish tall

variety from dwarf and kingcoconut. The CNZ6 was also a good primer to separate the tall variety and Dwarf variety. However it also failed to distinguish bodiri from other varieties, because it showed the same allele, which is present in the Dwarf. The CAC4 was a good primer to separate king coconut and tall and dwarf varieties from each other, but it also does not clearly distinguish the bodiri because both dwarf and tall alleles are present in the bodiri. However there is clear separation of bodiri and kingcoconut. CAC20 shows a great level of polymorphism by separating the dwarf from bodiri, but the similar alleles are found in both tall and dwarf. Thus there is no specific primer to separate the bodiri. So at the end of the result, we can separate the bodiri from other varieties using the three primers in combination. These primers are CAC 4 (fig 4.3), CAC 20 (fig 4.4), CNZ 6 (fig 4.2). As we discussed above the CAC4 primer can separate the kingcoconut from bodiri and CAC20 is the other primer, which separate the dwarf from bodiri. Then we can easily separate the tall from bodiri using the CNZ6 primer. Using these three primers in combination it is possible to separate the bodiri from other varieties. In this we used the eleven pre designed primers (Perera et al, 1998) and all the primers are amplified DNA aptly and most of them revealed the presence of allele of both dwarf and tall in bodiri. Examples for such primers are CAC 8, CAC 2, CNZ 4, CNZ 37, and CNZ 44.

These findings clearly exemplify the potential of molecular markers for separation of coconut individuals coming from open pollinated progenies to identify desired genotypes. In this case identification bodiri is so important due in part to the intention of the CRI to popularize this coconut genotypes as a natural beverage. In order to popularize bodiri in high industrially potential lands such as dry zone villages the requirement of pure bodiri seedlings is very high sometimes exceeding many more thousands. Therefore, the applicability of the above finding is very likely in the near future.