D-34: Identification of random decanucleotide primers for characerization of coconut *(Cocos nucifera* L.) germplasm using random amplified polymorphic DNA

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Coconut Research Institute has taken serious measures to manage coconut germplasm by means of establishing *ex-situ* gene banks. Apart from measurable morphological, reproductive and agronomical plant attributes, molecular marker descriptors are also now being used to strengthen germplasm management. Among all molecular assays, RAPD analysis is easy and fast for genetic assessment of plants less benefited by DNA technology. Early studies have shown that RAPDs can clearly discriminate coconut varieties and forms accurately and hence this study plan to identify a set of decanucleotide primers for precise characterization of coconut germplasm.

DNA isolated from tall and dwarf coconut types using the procedure described by Doyle and Doyle (1990) and quantified by 'Genequant' (Pharmacia) were repeatedly tested with sixty random primers from Operon Technologies in the polymerase chain reaction using *Taq* polymerase (Promega) and dNTP (Pharmacia) in a Perkin Elmer programmed for 94°C for 1 min, 36°C for 1 min and 72°C for 2 min and 45 cycles. Amplified DNA was assayed in 2% agarose gels under UV.

The primers generated a total of 448 fragments averaging 7 - 8 (range: 1-14) bands per primer, sizes ranging from 225 bp to 2,500 bp. Among amplified bands 326 (73%) were temporally consistent and 138 (42%) clearly exhibited polymorphism among tall and dwarf types. Judging by 186 consistent bands 225 primers were preferentially listed for assessment of the genetic diversity of coconut accessories in *ex-situ* gene banks.

In spite of few spurious bands generated by random primers the RAPD technique is evident here as a fast and easy procedure for characterization of coconut germplasm. The 25 primers, apart from characterization of *ex-situ*

coconut accessions can also be used in prioritization of new accessions avoiding duplications in widening of gene banks and selection of core populations for germplasm utilization in future breeding programmes.

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