Leaf storage conditions and genomic DNA isolation efficiency in Ocimum gratissimum L. from Kenya

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Abstract:

Storage of plant tissues for DNA is important to avoid degradation of DNA. Preliminary studies were conducted on Ocimum gratissimum L. in order to establish the storage conditions for the collected samples before DNA extraction. Secondly, the aim was to determine the best protocol for the extraction of high quality DNA, which would later be used for molecular analysis. DNA was extracted from the samples one month after field sampling. During the DNA extraction, four protocols were used; the modified hexadecyltrimethyl ammonium bromide (CTAB) mini preparation method described by Doyle and Doyle (1990), with reductants either mercaptoethanol or dithiothreitol; the modified sodium dodecyl sulphate (SDS) mini preparation method of Edwards et al. (1991) with redundant either mercaptoethanol or dithiothreitol. The DNA was purified, treated with RNase, quantified and examined for intactness using gel electrophoresis method. Good quality and high yield DNA could only be extracted with the buffer containing the detergent SDS and the reducing agent dithiothreiotol.