

**International Conference on Food Security
through Agriculture & Allied Sciences
(FAAS-2019)**

SOUVENIR



27-29 May, 2019

Venue

Tribhuvan University, Kathmandu, Nepal

**International Conference on Food Security through Agriculture & Allied Sciences
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SOUVENIR CUM LEAD/ABSTRACT PROCEEDINGS BOOK

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Theme 1 : Breeding and Biotechnological innovations

Optimization of genomic DNA isolation protocol from emerging leaves of small cardamom [*Elettaria cardamomum* (L.) Maton] for genetic diversity analysis

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ABSTRACT

Elettaria cardamomum (L.) Maton popularly known as Indian cardamom or small cardamom is mostly cultivated in the Southern Western Ghats (SWG) at higher altitudes ranging from 900 to 1400 msl. DNA isolation in small cardamom leaves have various difficulties as their DNA is highly viscous due to the presence of proteins, polysaccharides and polyphenols. Therefore, present study aimed to develop a rapid DNA isolation and PCR amplification protocol from the cardamom leaves (just emerged to unfold). In this study, we report a simple but rapid protocol for the isolation of DNA by CTAB extraction method, in which combined application strategy of 2-mercapto ethanol, polyvinyl pyrrolidone (PVP), sodium metabisulphate and NaCl (5M) was devised and used for removing polyphenol and polysaccharides. Also, ammonium acetate (7.5M) used to remove the DNA bounded cellular and histone proteins. This procedure is highly suitable for emerging leaf tissues from which an excellent quality of genomic DNA was achieved with high yield as well as free from contamination and development of brownish yellow colour. Current protocol yields pure DNA as documented from bio-spectrophotometer readings, where the A260/280 ratio was 1.6-1.9, indicating the absence of contaminants. In 0.8% agarose gels, the DNA forms a thick intact single band of high molecular weight confirming good quality. The DNA yield from emerging leaves of 15 promising cardamom accessions ranged from 100-155 µg/µl. In addition to that, our results emphasized that this procedure can be suitable to detect the genetic variability through molecular marker amplification. The isolated DNA has been successfully used for genetic diversity analysis of small cardamom using different inter simple sequence repeats (ISSR) markers.

Keywords: Cardamom, DNA isolation, PCR, ISSR markers, Diversity analysis.