Comparative Evaluation of Six Extraction Methods for DNA Quantification and PCR Detection in Cocoa and Cocoa-Derived Products

Lam Thi Viet Ha[*a,b,c], Lore Vanlerberghe[b], Ha Thanh Toan[c], Koen Dewettinck[a] and Kathy Messens[b]

[a] Laboratory of Food Technology and Engineering, Department of Food Safety and Food Quality, Faculty of Bioscience Engineering, Ghent University, Coupure links 653, 9000 Gent, Belgium

[b] Laboratory AgriFing, Department of Applied Biosciences, Faculty of Bioscience Engineering, Ghent University, Valentin Vaerwyckweg 1, 9000 Gent, Belgium

[c] Biotechnology Research and Development Institute, Cantho University, Campus II, 3/2 Street, Cantho city, Vietnam

[*]Corresponding author: Lam Thi Viet Ha

Laboratory of Food Technology and Engineering, Department of Food Safety and Food Quality, Faculty of Bioscience Engineering, Ghent University, Coupure links 653, 9000 Gent, Belgium

Telephone number: 0084-945 227 097

Email: ltvha@ctu.edu.vn

Abstract

Six methologies for extracting DNA in cocoa and cocoa-derived products (cocoa leaves, cocoa beans, cocoa powder, cocoa butter, cocoa mass, and dark chocolate) are described. These DNA extraction methods included 4 commercial kits DNeasy Plant kit, DNeasy Plant kit with polyvinyl polypyrrolidone (PVPP), QIAamp DNA Stool kit, Wizard Genomic DNA kit and 2 CTAB based methods, CTAB-Proteinase K procedure and CTAB-SDS (sodium dodecyl sulfate) procedure. The DNA yield, purity, and quality for 6 different cocoa matrices are discussed. The yield and purity were determined through spectrophotometry. Three different conventional PCR reactions were used to assess the quality of the DNA extracted. The Wizard Genomic DNA kit and the CTAB-Proteinase K method were found to be the best for DNA quality and PCR detection in cocoa leaves. The CTAB-Proteinase K procedure is highly recommended for cocoa beans, while the Wizard Genomic DNA kit revealed the best PCR performance when applied to cocoa powder. The cocoa butter matrix gave the lowest DNA yield, purity, and amplification results for all the examined extraction methods. The QIAamp DNA Stool kit and the CTAB-SDS method showed the best result for PCR detection of cocoa mass. The Wizard Genomic DNA kit and the CTAB-SDS method showed the best PCR performance for dark chocolate. In general, the two CTAB based methods, namely the CTAB-Proteinase K and the CTAB-SDS were suitable for the DNA extraction of all types of cocoa-derived products with the exeption of cocoa butter. This evaluation can be especially useful for the detection of DNA in cocoa derived samples.

Keywords: Cocoa, Cocoa-derived products, Deoxyribonucleic Acid extraction, Polymerase Chain Reaction