

Tropentag, October 9-11, 2007, Witzenhausen

"Utilisation of diversity in land use systems: Sustainable and organic approaches to meet human needs"

An Affordable and Effortless Method for High-Throughput DNA Extraction from Animal Tissues

Tarik Rabie¹, Mohamed Mandour², Hassan Zeweil³

Abstract

A very simple and inexpensive method for high-throughput DNA extraction from animal tissues has been developed. The procedure contains three steps (digestion, Incubation, and centrifugation) and it is compatible with the normal eppendorf tubs which are commonly used in the routine laboratory work. A small piece of tissue (from 10—400 mg) was used to isolate DNA. It yields enough templates DNA for polymerase chain reaction (PCR) amplifications (at least two complete 96 well PCR plates). Enormous number of sample are used to establish the final procedure for DNA isolation from chicken, duck, and quail for different kind of tissues such as Heart, Liver, Spleen, Pancreas, Kidney, Stomach, Gizzard, Oviduct, Ovary, Testes, Bronchi, Lung, brain, and Pituitary gland. DNA purity and concentration have been measured by NanoDrop® Spectrophotometer and the standard curve was drawn. Both microsatellite (MCW0166, MCW0317 - derived from chicken linkage map-, SCU00100, and SCU00101-not published) and STS markers (Randomly selected STS marker from National Center for Biotechology Information, NCBI) have been used to examine the isolated DNA by normal PCR. Agarose gel electrophoresis (different concentrations ranged as 1, 1.5, and 2%) has applied successfully, followed by 8% and 10% polyacrylamide gel to emphasise the viability of extracted DNA for several molecular biology applications. DNA marker ranged from 50bp-1000bp was used as a guide for the PCR product size resulted in variable range of band sizes. Therefore, this application method is expected to facilitate studies that require high-throughput DNA isolation for PCR amplification, such as genotyping by microsatellite markers for mapping and genetic diversity studies, as well as mutant screening in poultry.

Keywords: DNA isolation, large-scale screen, microsatellites, polymerase chain reaction, poultry, tissues

Contact Address: Tarik Rabie, Suez Canal University, Faculty of Agriculture, Animal Production Department, 41522 Ismailia, Egypt, e-mail: tarik.rabie@gmail.com

¹Suez Canal University, Faculty of Agriculture, Animal Production Department, Equpt

² Alexandria University, Faculty of Veterinary Medicine, Department of Animal Husbandry and Animal Wealth Development, Egypt

³ Alexandria University, Faculty of Agriculture (Saba Basha), Department of Animal and Fish Production, Egypt