ASSESSMENT OF DNA RETENTION ON PLASTIC SURFACES OF COMMERCIALLY AVAILABLE MICROCENTRIFUGE TUBES

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It is well documented that DNA can bind to the surface of polypropylene microcentrifuge tubes commonly employed during DNA extraction and storage. Depending on the ionic strength of the buffer, the amount of DNA absorbed to polypropylene ranges from 0.25 to 5 ng/mm^2. Furthermore, it has been demonstrated that DNA bound to the internal wall of plastic tubes nevertheless seems to be available for subsequent amplification by the PCR.

Earlier assessments of DNA adhesion to plastic polymers have been performed by quantitation of bound radiolabeled DNA. We report here a simple assay that will potentially allow for the measurement of DNA retention to tubes. Incubation of Applied Biosystems Genescan®-500 ROX internal lane standard provides a suitable range of sizes (35-500 bp) to detect adhesion of DNA to plastic tubes by fluorescence, following exhaustive removal of the solution prior to and following generous washing of the tube. Laser-induced emission from the ROX dye linked to the DNA fragments was detected with the FMBIO® Fluorescence Imaging System (Hitachi Genetic Systems) by direct scanning of the microcentrifuge tubes. We confirm that increasing ionic strength induces DNA binding, an important fraction of it, irreversibly. Our results suggest that certain tubes advertised as being non-retentive of DNA, are potentially more retentive than some others not advertised for non-retention. Silicone coating of tubes does not eliminate DNA binding. We find that non-ionic detergents Tween 20 and Triton X-100, at concentrations that do not interfere with the PCR, are very effective in preventing DNA adhesion to plastic matrices. However, our results indicate that these detergents are not useful in removing DNA once its binding has occurred to the plastic surface.

Fluorescence detection with the FMBIO® instrument allows for relative assessments of bound, dye-labeled DNA fragments. Further experiments using a quantitative (real-time) PCR assay will make possible the accurate measurement at picogram levels, of the kinetics and extent of loss from solution of human nuclear DNA as a consequence of its adhesion to the inner wall of storage tubes.