

DEVELOPMENT OF AN AUTOMATED DNA CHARACTERIZATION PROCEDURE FOR USE IN DNA MICROARRAY PREPARATION

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Detection and quantification of small amounts of DNA, such as PCR products, are extremely important in a wide variety of biological applications. A problem frequently encountered while attempting a gene expression analysis or the quantitation of a PCR amplification yield is the unreliable automation of experiments. The inaccurate data occurs because there are often variances in the amounts and/or concentrations of the samples. Therefore, an automated quantitation of probes for use in DNA microarrays was attempted using a Packard MultiPROBE II EX (MP11) robotic liquid handling system and a Perkin Elmer HT Soft 7000 Plus Bio Assay Reader. A standard curve that was comprised of known concentrations of DNA was first obtained through hand pipetting. This standard curve was then prepared using automated procedures on the MP11 with a known amount of a fluorescent intercalating dye called picogreen. Precise readings of the liquid's fluorescence yielded a standard curve. Refinement of the procedure produced a reliable standard curve that allows for the determination of PCR products' concentrations by correlating the fluorescent readings with those of the standards. This achievement was significant in that the automated quantitation of the PCR amplification yields will allow for the rapid characterization of the large numbers of PCR products needed to prepare high density DNA arrays.