

Technology Administration, U.S. Departme Commerce Email: john.butler@nist.gov

View of capillary array inside of ABI 3100



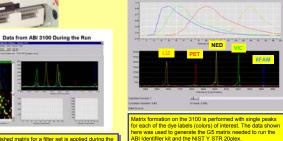
STR Typing with the ABI PRISM™ 3100 16-Capillary Genetic Analyzer

John M. Butler, Richard Schoske, and Margaret C. Kline

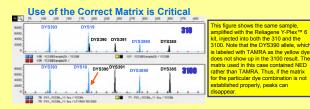
Acknowledgments

Biotechnology Division, National Institute of Standards and Technology, 100 Bureau Drive MS 8311, Gaithersburg, MD 20899-8311

We have had the ABI 3100 16-capillary system in our laboratory since April 2001 and have successfully analyzed a variety of STR typing kits including Promega's PowerPlex[®] 16 and Applied Biosystems' Identifiler[™] 16plex kits. These commercial kits and new research multiplex assays we have developed in-house use a variety of fluorescent dye combinations with both 4-dye and 5-dye chemistries. We have generated DNA fragment analysis matrices on the 3100 using various combinations of the following dyes: 5FAM, JOE, NED, ROX, 6FAM, HEX, FL, TMR, CXR, VIC, PET, and LIZ. Some of the challenges and solutions to obtaining successful results on the ABI 3100 will be presented. We have also performed direct comparisons of samples between the ABI 310 and 100 instruments.



The established matrix for a filter set is applied during the data collection so if there is a problem, the sample must be REINJECTED after a new matrix is established rather than applying a new matrix to any raw data as can be done on the ABI 310. In addition, the sizing algorithm is slightly different than what is used in the ABI 310.



Sample Preparation Options



cycling. We have developed protocols for preparing

plates for ABI 3100 analysis.



MWG Biotech RoboAmp 4200 is capable of liquid handling, plate movement, and integrated themat

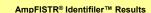
3100 96-well sample plates



We have developed Excel spreadsheets for direct input of plate record information to speed the sample entry process.

Each run (injections from 16 different samples) takes approximately 45-50 minutes for the entire cycle. Thus, 96 samples can be run in 5 hours on the 3100 instead of the 48 hours required for a single ABI 310.

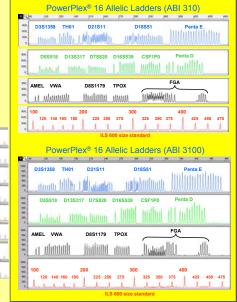
Data analysis with a Windows NT platform with GeneScan[®]3.7 and [®] Genotyper[®] 3.7 is essentially equivalent to the Macintosh versions and can be fairly labor-intensive depending on the data quality. Funding from the National Institute of Justice through the NIST Office of Law Enforcement Standards Rich Schoske is a graduate student at American University under the advisement of Dr. Jim Girard and is being funded by the United States Air Force, through the Air Force Institute of Technology Nadine Nassir (Promega Corporation) for PowerPiex 16 3100 matrix standards



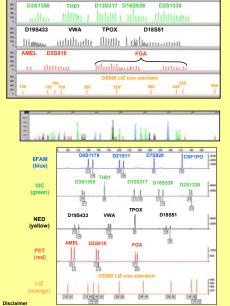
D7S820 CSF1PO

D21S11

D8S1170



We have performed a typing comparison with 192 PowerPlex 16 samples run on both the ABI 310 and ABI 3100.



Certain commercial equipment, reagents, and software are identified in order to adequately specify or describe the subject matter of this work. In no case does such identification imply recommendation or endorsement by the National Institute of Standards and Technology, nor does it imply that the equipment, reagents, or software are necessarily the best available for the purpose.