Fluorescent Amidite Matrix Standards

Product P/N 401546E
Insert P/N 4340041A
Printed in U.S.A.

For Research Use Only.
Not for use in diagnostic procedures.

These standards are used to generate the "multi-component matrix" required for four-color fluorescent fragment detection using the Model 672 GeneScan™ software on the model 373 DNA Sequencing System. The Model 672 *Analysis* software uses the multi-component matrix to automatically analyze four different-color fluorescent dye-labeled samples in a single gel lane.

The kit contains one tube each of the 6-FAM™, TET™, HEX, TAMRA™, and ROX™ matrix standards. The matrix standards contain 28 DNA fragments ranging in size from 55- to 14907 base pairs (negative gels) or from 37 to 1409 base pairs (denaturing acrylamide gels). Their concentrations are 0.5 nM for 6FAM and TET, 1.0 for HEX, 2.0 nM for TAMRA, and 4.0 nM for ROX (dye concentration per double-standard fragment). The standards contain a blue loading buffer for convenience in gel loading. They are buffered with 1x TBE containing added EDTA (7mM total). The standards are stable for one year at 2°C to 8°C (avoid freeze thaw cycles).

To make a matrix, run a set of four standards under the same gel conditions to be used for experimental samples. Note: the 6FAM and FAM cannot be accurately distinguished from one another by the instrument, and should not be used together. Similarly, HEX and JOE™ cannot be accurately distinguished from one another. Filter Set B must be used to distinguish TET from 6-FAM and/or HEX; note that this filter set cannot detect ROX (TAMRA can be used in place of ROX as the fourth dye: GeneScan™-2500 TAMRA, P/N 401545). Refer to the 373 Sequencing System User's Manual for gel preparation instructions , electrophoresis conditions, and more information on how dyes can be detected using Filter Sets A and B. Be sure to collect the data in the "GeneScan" mode. The matrix is calculated using the 672 GeneScan *Analysis* software.

Thoroughly mix the contents of each standard tube before use, and centrifuge to collect the liquid at the bottom.
Use a separate lane for each standard, and load alternate lanes, leaving the intervening lanes empty.

Denaturing acrylamide application:
For each standard, combine 2.5 µL of standard with 2.5 µL of formamide. Heat each sample at 90°C for two minutes to denature before loading. Load 5 µL per gel lane. *Note*: DNA samples must not be stored in formamide for more than a few hours.

Native gel application:
Load 5 µL per gel lane.

Note: For the phone number of the Applied Biosystems technical support in your area, consult your User's Manual.

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