

## **ABI Prism® 3100 Genetic Analyzer Instrument to Instrument Variation**

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In a high throughput sample processing environment it is important to eliminate as many variables as possible in order to process samples efficiently and achieve the highest possible quality of results with each sample. In pre-amplification stages, it is possible to standardize sample input with DNA extraction, quantification, and normalization of sample DNA concentration. It is also possible to standardize aspects of the post-amplification process.

We observed that the same amplified product run on different ABI 3100 Genetic Analyzers produced noticeably different peak heights for the same alleles. Some instruments were reproducibly “stronger” and others reproducibly “weaker”. We decided to perform a variability study with all 11 3100 instruments we use for STR genotyping analysis. It was observed that average peak height intensities for entire samples or across allelic ladder alleles varies as much as two fold across the 11 genetic analyzers. This compares with an inter-color variation that was as high as six-fold on some instruments, but not others. The relative instrument intensity tended to correlate with instrument age, but not specifically with laser age. Two of our “middle-aged” instruments have had laser replacements. This did not make them stronger with regard to relative RFU intensity. To counteract this effect, we have determined that we can modify injection conditions to overcome the instrument variability. To a first approximation, in the typical ranges used, a combination of injection voltage times injection time (i.e., kV-sec) is directly proportion to RFU strength for an individual instrument. Thus increasing the combined kV-sec used for injection on weaker instruments or decreasing it on stronger instruments allows adjustment so that all instruments provide approximately equivalent performance.