

Profiler Plus™ Validation: Establishing Thresholds of Stutter Percentages and Allele Ratio at Heterozygous Loci for Use in Mixture Analysis

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Crime scene samples often consist of a mixture of bodily substances originating from more than one individual. If the minor contributor's component is more than 1-2% of the mixture, STR DNA typing will yield a combination profile. Interpretation of such profiles is a crucial component of forensic investigations as dissecting away a first profile (e.g. the victim's) allows for a second profile (e.g. the crime perpetrator's) to be identified. This isolated profile can then be compared to a suspect's profile, or searched for a match in a criminal offender DNA database. Mixtures with a 50:50 ratio of contributors are admittedly the most difficult to dissect without prior knowledge of the contributor's profiles. A minor profile of less than 20% can lead to difficulties if some of the peaks overlap the known position of the stutter of one of the major profile's peaks. Therefore, an assessment of the expected ranges of allele ratios at heterozygous loci (HR) and stutter percentages (%S) for a specific STR DNA typing system is essential for the reliable interpretation of mixtures.

To enable us to draft practical interpretation guidelines, we have processed a large number of samples (468 population database samples and 275 casework samples), as part of our in-house validation of the Profiler Plus™ kit. From this data, we tabulated HR values for all allele pairs and %S for all allele peaks. Stutter of peaks under 100rfu of peak height as well as casework samples exhibiting mixed profiles were excluded from this tabulation. HR and %S values were compiled using peak height and peak area; medians and standard deviations were calculated.

Peak height values produced tighter distributions of data points and proved more reliable than peak area. When HR and %S data was plotted against peak height and area, no upward shift in the cluster of data points was noted for data points exceeding 4500rfu (peak height). The range of values (height) obtained was:

	% Stutter Threshold Range (%S)		Allele ratios at Heterozygous loci (HR) (low/high intensity)	
	+2SD	+3SD	Median	Standard Deviation
Population Database	8.2 – 13.2%	10.5 – 16.3%	91 – 94%	6 – 8%
Casework	7.3 – 12.6%	9.4 – 15.9%	88 – 90%	10 – 13%

This data helped establish the interpretation guidelines currently being used in DNA typing casework at the RCMP.