Evaluate the mixture profile for the following:

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| --- | --- |
| Peaks | Evaluate true allele vs stutter, pull-up, split peaks (-A), spikes, dye blobs, etc. |
| Stochastic Effects | Dropout, peak imbalance with a locus, ski-slope between loci, etc |
| Mixture Ratio | Determine mixture ratio for all loci within the profile containing allelic combinations suitable for determining mixture ratio; average results. |

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| Deconvolute the profile using the following parameters: | |
| PAT | Reported alleles must be above the established analytical threshold (universal or dye specific). |
| HET PHR | Expected PHR for heterozygote alleles is > 55%. Peak imbalance may occur and should be considered for lower RFU samples and at high molecular weight loci. |
| ST | When interpreting STR genotypes for samples of low RFUs, care must be taken to distinguish a homozygote peak from a heterozygote peak whose sister allele has dropped out (e.g., complete or partial dropout). As a general guideline, single peaks should not be called as homozygote genotypes unless the peak is > to the established ST. **Consider allele sharing (stacking) when assessing an allele as a possible homozygote peak.** |
| Mixture Ratio | Calculated genotype combinations should meet the expected mixture ratios. |

For mixture ratios ≥70:30, evaluate the mixture profile using the following guidelines:

|  |  |
| --- | --- |
| Total profile | Major Contributor > 70% |
| 4 alleles | No drop out even if one or more alleles are below ST. Deconvolute based on peak height ratio and mixture ratio. |
| 3 alleles | If one or more alleles are below ST, possibility of drop out. Deconvolute based on peak height and mixture ratio. In general, the genotype for any allele below ST is "X,ANY" (X=allele). An exception for the minor contributor would be in the instance of a major homozygote with two minor alleles where one or both are below ST. Consider allele sharing (stacking) when assessing an allele as a possible homozygote peak. |
| 2 alleles | If one or two alleles are below ST, possibility of drop out. If applicable, deconvolute based on peak height and mixture ratio. The genotype for any allele below ST is "X,ANY" (X=allele). Consider allele sharing (stacking) when assessing an allele as a possible homozygote peak. |
| 1 allele | If the allele is below ST, drop out is present. The locus is "INC" for statistical interpretation. |
|  | Minor Contributor ≤ 30% |
| 4 alleles | See above. |
| 3 alleles | See above. Consider allele sharing (stacking) when assessing an allele as a possible homozygote peak. Evaluate homozygote minor peak at ~2X ST. Account for possible stutter if allele is located in stutter position. |

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| 2 alleles | If one or two alleles are below ST, possibility of drop out. The genotype for any allele below ST is "X,any" (X=allele). Consider allele sharing (stacking) when assessing an allele as a possible homozygote peak. Evaluate homozygote minor peak at ~2X ST. Account for possible stutter if allele is located in stutter position. |
| 1 allele | The locus is "INC" for statistical interpretation. |

For mixture ratio <70:30, evaluate the mixture profile using the following guidelines:

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| --- | --- |
| Total profile | Mixture ratio < 70:30 |
| 4 alleles | No drop out even if an allele is below ST. All heterozygote genotype combinations included. |
| 3 alleles | If one or more alleles are below ST, possibility of drop out. Deconvolute based on peak height ratio. The genotype for any allele below ST is "X,ANY" (X=allele). Consider allele sharing (stacking) when assessing an allele as a possible homozygote peak. |
| 2 alleles | The locus is "INC" for statistical interpretation unless both alleles are ~2X ST. Consider allele sharing (stacking) when assessing an allele as a possible homozygote peak. |
| 1 allele | If the allele is less than ~2X ST, possible drop out is present. The locus is "INC" for statistical interpretation. |