



Y-Screening and Direct Amplification of Sexual Assault Samples

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Abstract #46

Introduction

Screening for sperm in sexual assault samples is a laborious process with limited sensitivity in comparison to male DNA detection by amplification.

Direct amplification of casework samples reduces both cost (labor & reagent) as well as the for potential DNA loss during purification, a special concern with low template samples.

The prototype Casework Direct Solution Kit rapidly generates lysates from casework samples that may be amplified with the PowerQuant[®] System to screen sexual assault samples for male DNA and to normalize human template for STR amplification with one of the PowerPlex[®] Systems.

Rapid, Simple Method

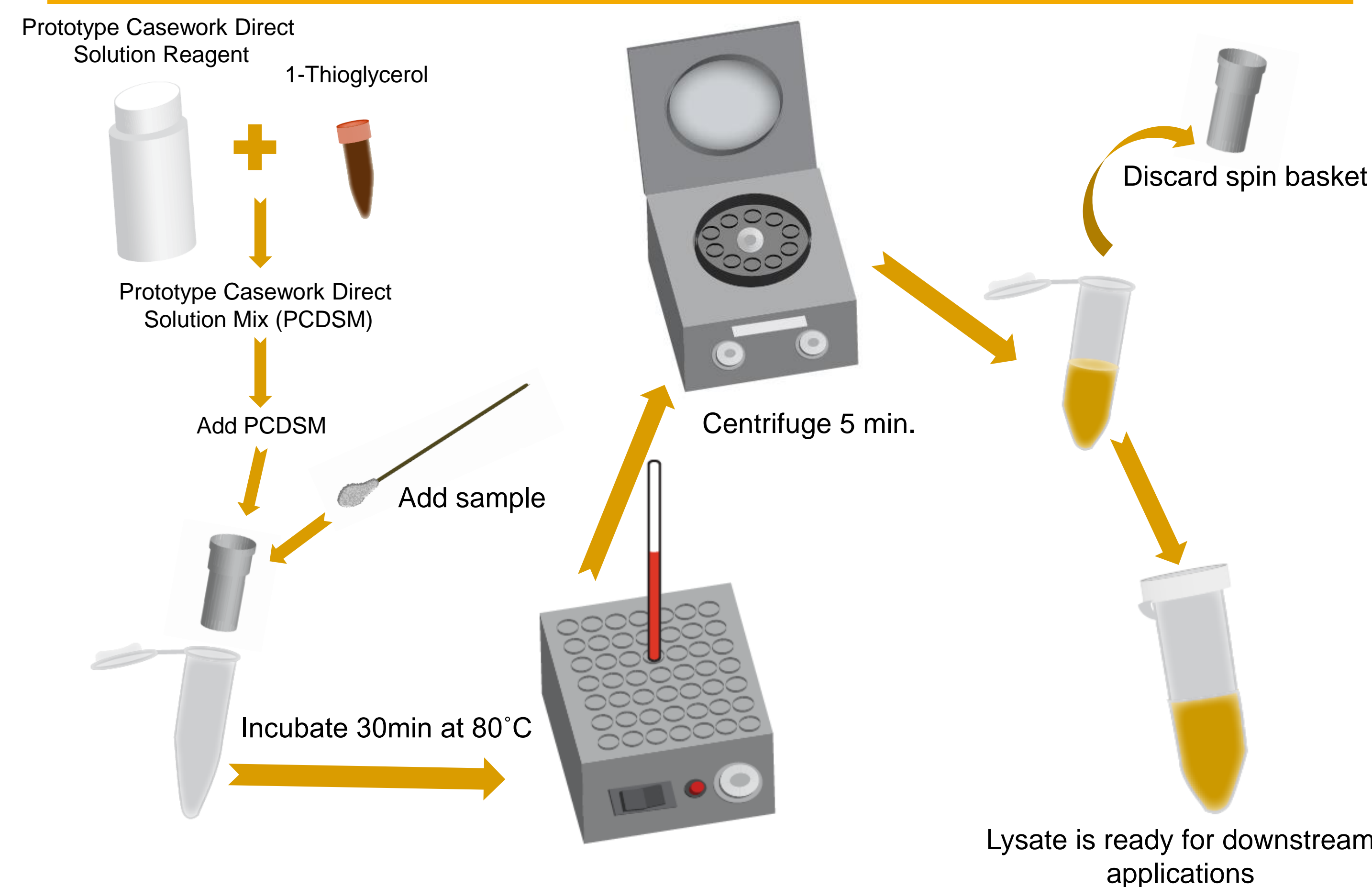


Figure 1: Prototype Casework Direct Solution Kit method.

Sensitivity: Detection of Male DNA in the Presence of High Female DNA

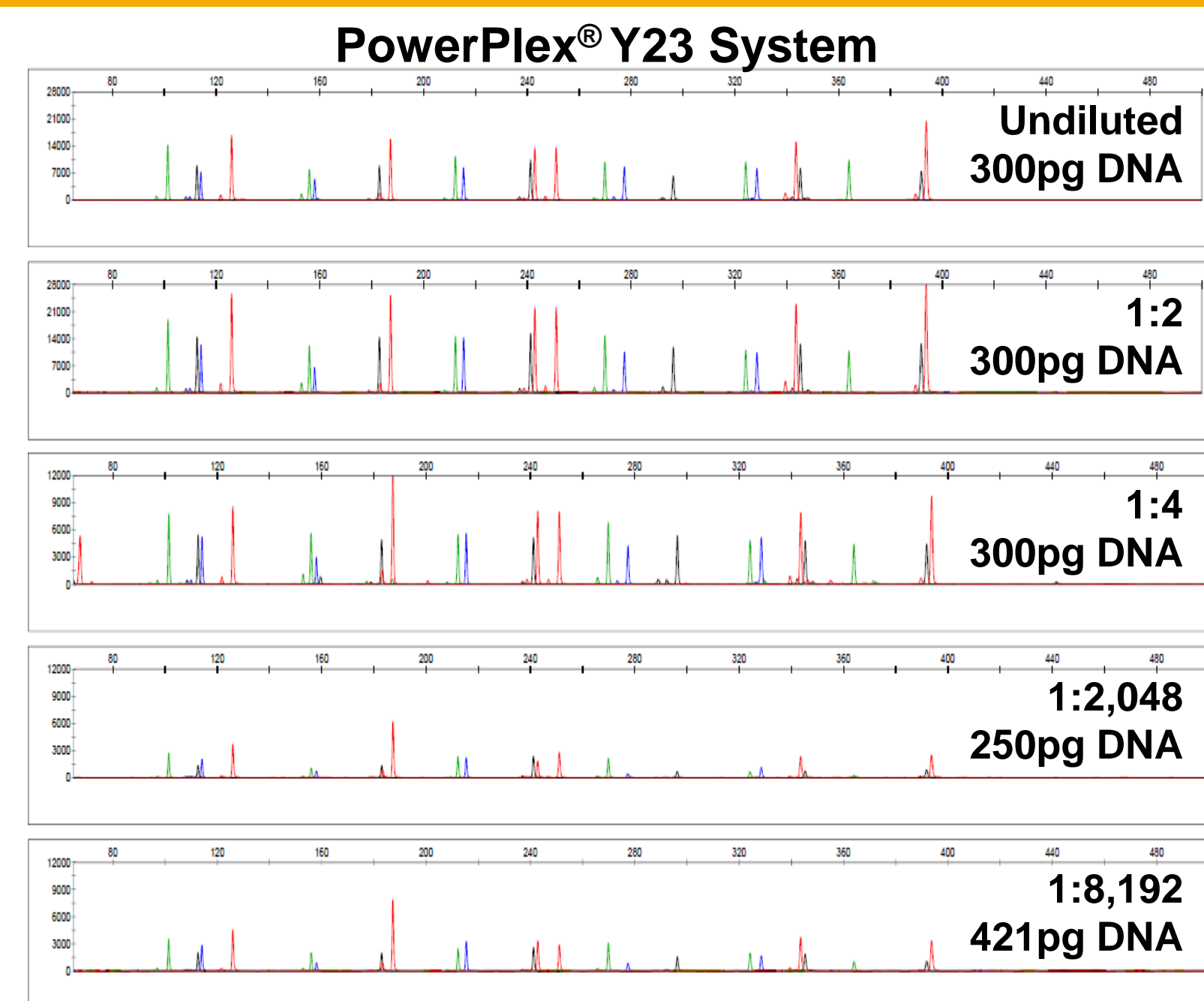
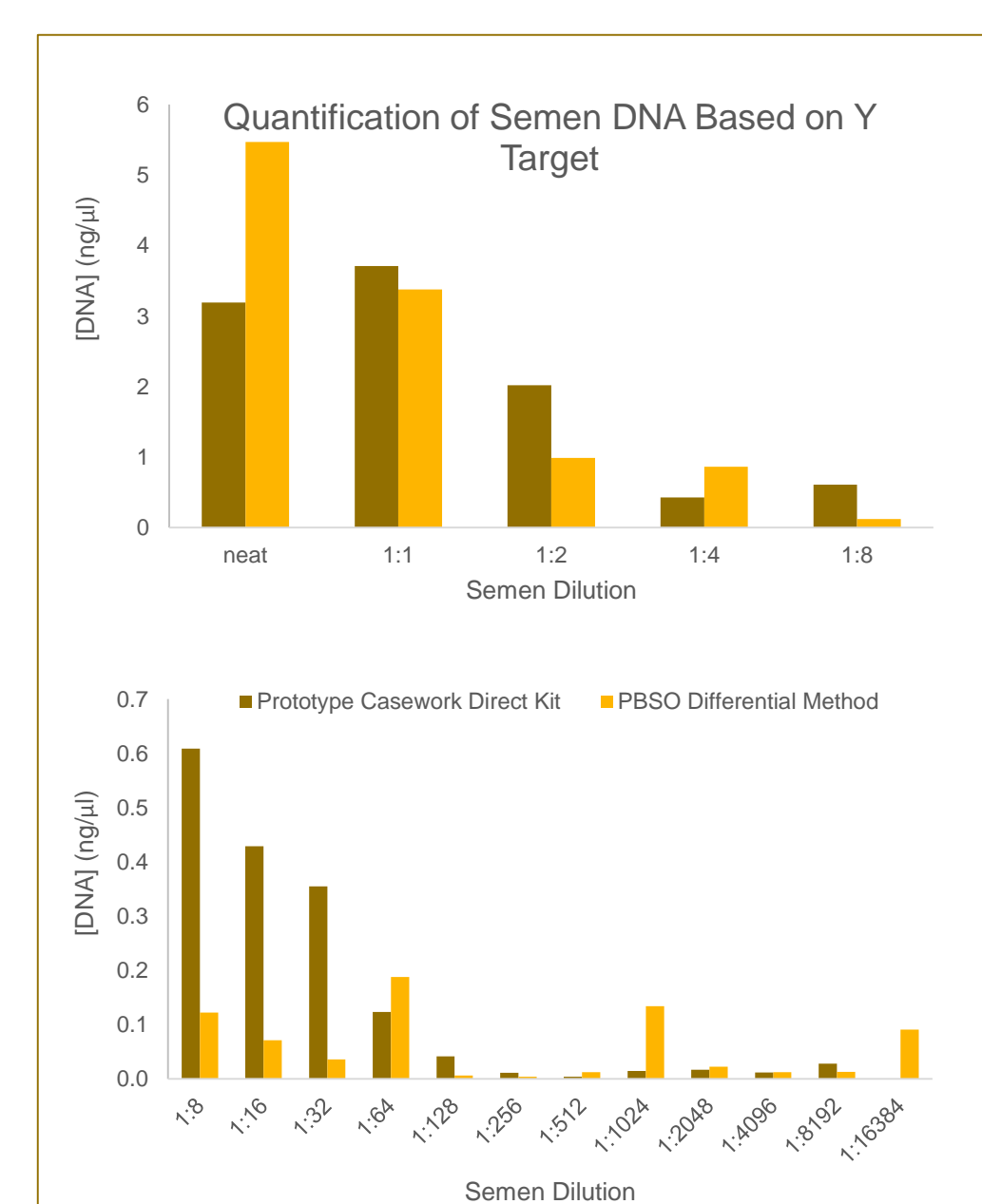


Figure 2. STR Semen Profiles in the Presence of High Female Background. Donor vaginal swabs were spotted with 10μl donor semen serially diluted. Swab tips (1/4) were processed with either 400μl PCDSM or laboratory's validated method. PowerPlex[®] Y23 System data shown for PCDSM-processed samples. Data collected on an Applied Biosystems[®] 3500 Genetic Analyzer with a 1.2kV 24sec. injection.

Post-Coital Swabs: Quantification of Human DNA with the PowerQuant[®] System

Hours Post-Coitus	Prototype Casework Direct Solution Kit			Non-Sperm Fraction			Sperm Fraction		
	Autosomal (ng/μl)	Y (ng/μl)	[Auto]/[Y]	Autosomal (ng/μl)	Y (ng/μl)	[Auto]/[Y]	Autosomal (ng/μl)	Y (ng/μl)	[Auto]/[Y]
5	6.22	1.94	3.2	9.00	0.06	163.0	1.78	1.83	0.97
24	14.59	0.19	77.5	12.89	<0.01	7169.7	0.10	0.05	1.97
48	23.34	0.03	721.5	12.12	<0.01	50840.9	0.04	0.01	6.01
72	22.57	0.02	1050.0	21.59	<0.01	39292.7	0.16	0.04	4.22
96	6.09	0.01	638.3	17.16	N.D.	N.D.	0.14	0.02	8.98

Table 1: DNA Quantification of Post-Coital Swabs. Swab tips (1/4) were processed in 400μl PCDSM, or laboratory in-house differential extraction method.

Post-Coital Swabs: PowerPlex[®] Fusion and Y23 System STR Profiles from PCDSM Lysate

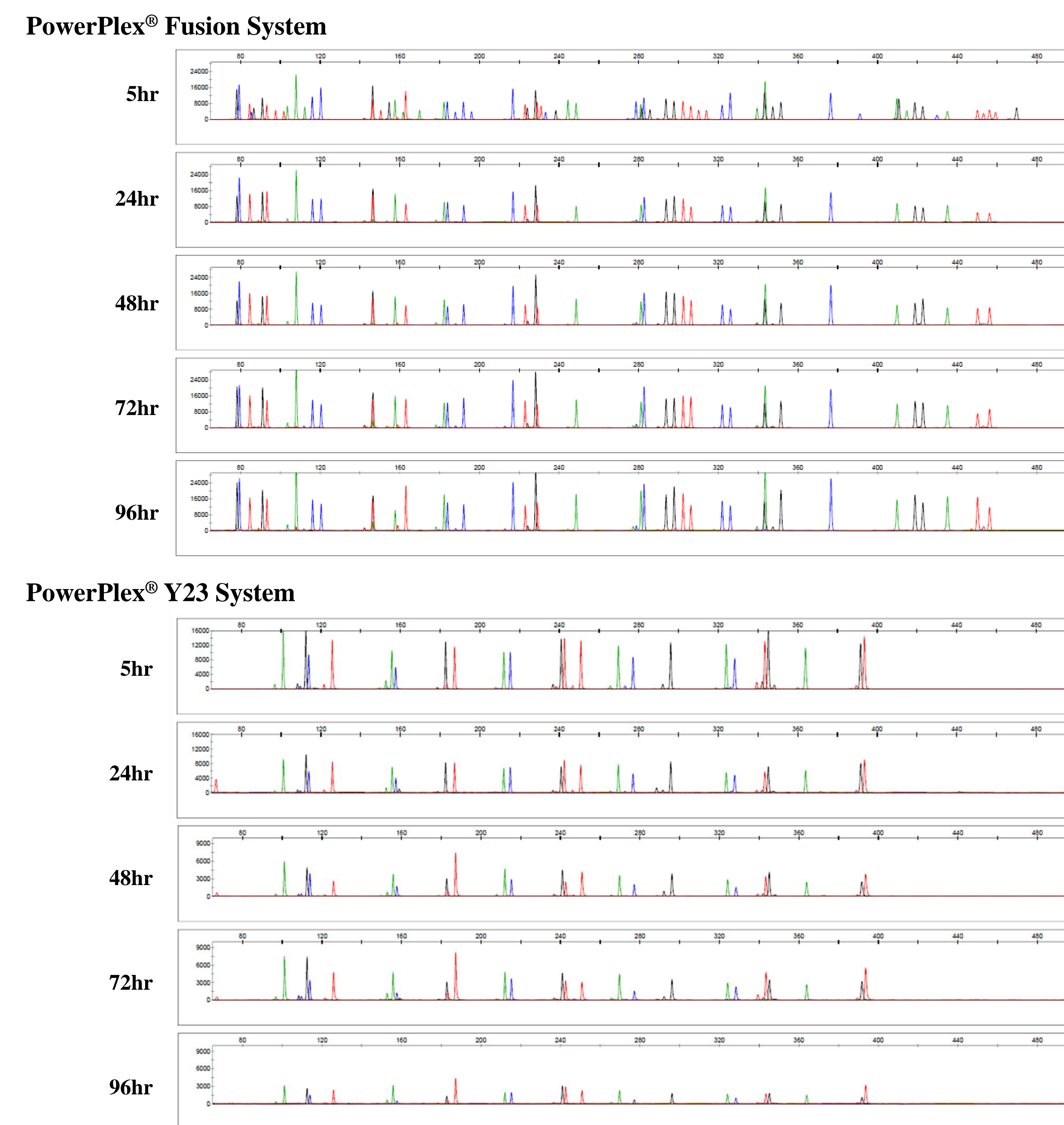


Figure 3. PowerPlex[®] Fusion and Y23 System Profiles. Normalized to 300pg template, based on Autosomal and Y Target DNA concentrations listed in Table 1. Data collected on Applied Biosystems[®] 3500 Genetic Analyzer with a 1.2kV 24sec. Injection.

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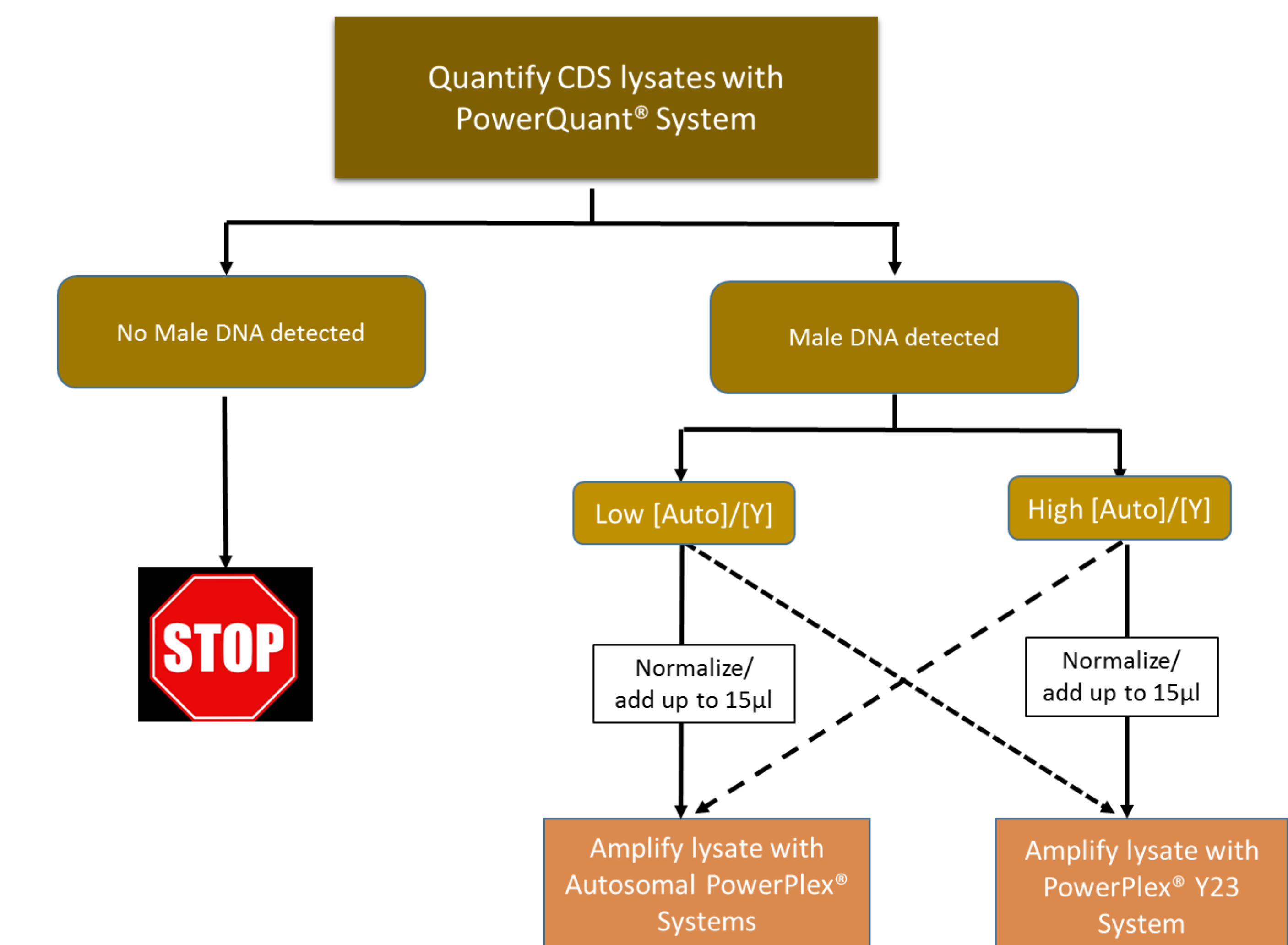
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Post-Coital Swabs: Correlation with Presumptive Tests

Time Post Coitus	Presumptive Tests		Prototype Casework Direct Kit		Differential Non-Sperm Fraction		Differential Sperm Fraction	
	Acid Phosphatase	Microscopic Evaluation	♂ STR Profile	♂ Y Haplotype	♂ STR Profile	♂ Y Haplotype	♂ STR Profile	♂ Y Haplotype
5hr	+	+	+	+	+	+	+	+
24hr	+	+	Partial	+	-	20/23	+	+
48hr	+	+	one locus	+	-	11/23 Drop-in	+	+
72hr	+	+	-	+	-	14/23 Drop-in	+	+
96hr	-	+	-	+	-	-	+	+

Table 2: Presumptive Tests & STR results of Post-Coital Swabs. In-house presumptive test results were compared to PowerPlex[®] Fusion & PowerPlex[®] Y23 System profiles generated from PCDSM lysates and in-house differential extraction method

Decision Tree for Processing of Sexual Assault Samples



Summary

- Fast**
 - Generate amplification-ready lysates in less than one hour including set-up time.
- Sensitive**
 - PowerPlex[®] Y23 System profiles obtained from 1:8,192 dilution of semen with high female background

Correlates with presumptive tests & differential extraction
• PowerPlex[®] Y23 System profiles obtained from Prototype Casework Direct Solution Kit lysates and sperm fraction with acid phosphatase negative/microscopy positive samples.

Available for sale in December 2016 as "Casework Direct Kit, Custom".