QIAAMP DNA INVESTIGATOR WITHIN THE SOUTH AFRICAN POLICE SERVICE

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SAPS – FSL: BIOLOGY SECTION

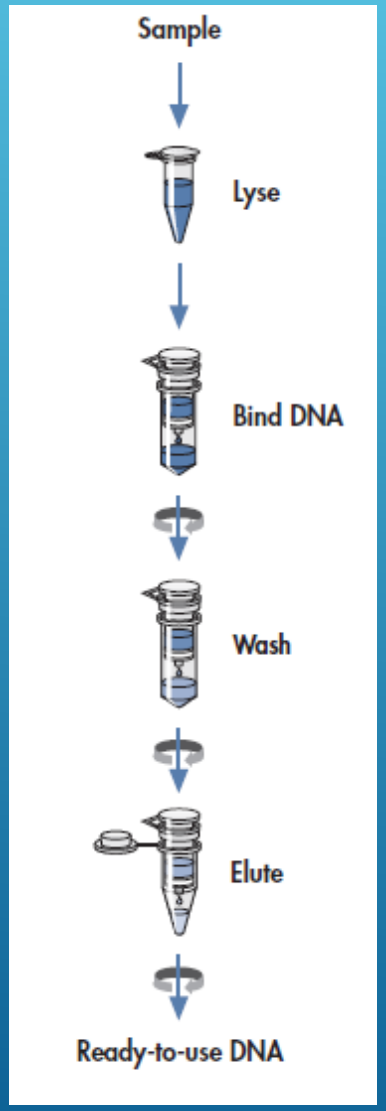
INTRODUCTION

- QIAGEN has a long association with the SAPS: FSL, 20 year relationship.
- MicroAmp replaced phenol chloroform.
- Custom extraction kits for the GSPS.
- Silica Membrane technology is the purification method for the “Manual DNA Process”.
- Laboratory is a high throughput environment.
- Use approximately 10 QIAGEN kits per day at Arcadia.
- Moved from MicroAmp to Investigator in 2014/2015.
- Stringent specifications for our consumables.

INTRODUCTION

- Validation conducted (In conjunction with QIAGEN).
- Reduced 12 Investigator Protocols to 5.
- Compression of the workflow to process more samples e.g. chewing gum.
- 5 protocols are:
  - “Universal Extraction” method
  - Nail & Hair
  - Tissue extraction
  - Sexual Assault (Differential)
  - Calcified tissue extraction

COMBINED PROTOCOLS



Sample Type	ATL	AL	AW1	AW2	Ethanol	ATE
Surface & Buccal Swabs	400	400	500	700	700	20-100
FTA & Guthrie Cards	280	300	500	700	700	20-100
Body fluid stains	300	300	500	700	700	20-50
Chewing gum	300	300	500	700	700	20-50
Paper etc.	300	300	500	700	700	20-50
Cigarette Butts	300	300	500	700	700	20-50
Nails & Hair	300	300	500	700	700	20-50
(Soft) Tissues	180	200	500	700	700	20-100
Bones and Teeth	360	300	600	700	700	20-50
Sexual Assault specimens	500	300	500	700	700	20-50

SAMPLE PREP

Sample Types
Blood
Semen
Hair
Nails
Envelopes
Cigarette butts
Saliva
Gum
Soft tissue
Bone

VALIDATION PARAMETERS

- Sensitivity
- Linearity
- Repeatability and Reproducibility
- “Concordance” –MicroAmp protocols were compared to the compressed Investigator protocols.

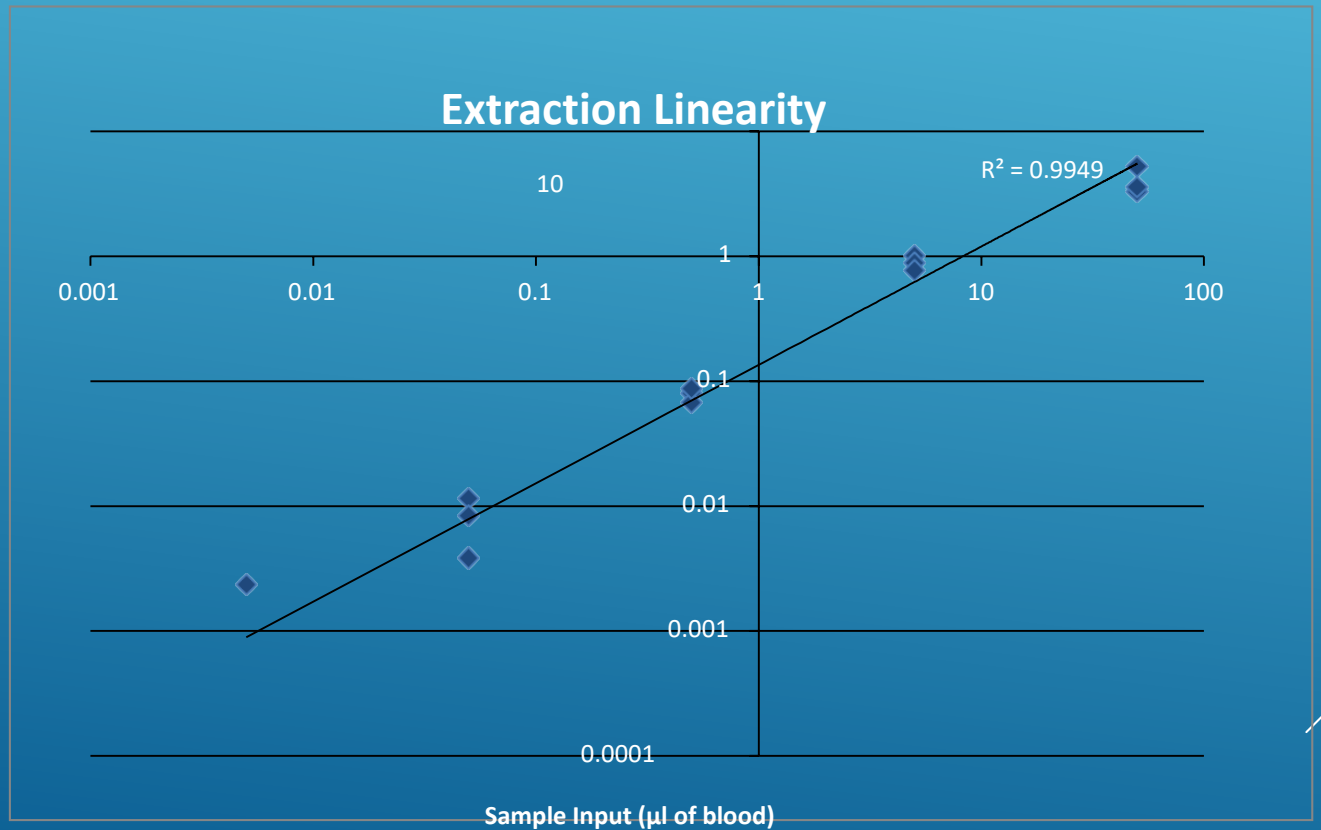
SENSITIVITY

- Sensitivity studies assess the range of sample inputs able to produce a reliable genotyping result.

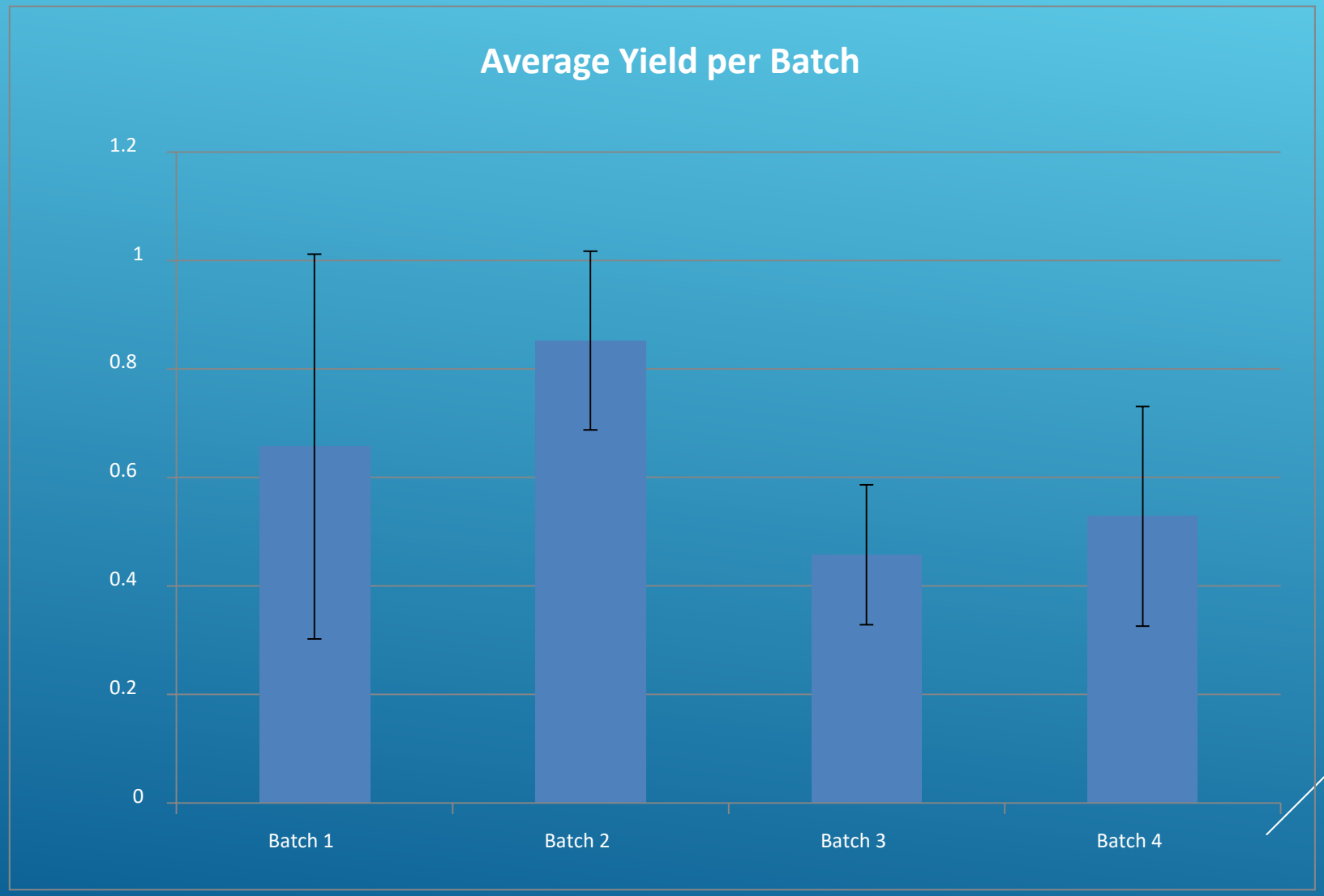
Sample ID	Details	Quantifiler Duo concentration (ng/µl)	Profile	Alleles recovered (%)
1.1	Neat blood	3.32736	FP	100%
2.1	Neat blood	5.261611	FP	100%
3.1	Neat blood	3.608488	FP	100%
1.2	1 in 10	1.005483	FP	100%
2.2	1 in 10	0.882098	FP	100%
3.2	1 in 10	0.772144	FP	100%
1.3	1 in 100	0.07962	FP	100%
2.3	1 in 100	0.067128	FP	100%
3.3	1 in 100	0.086244	FP	100%
1.4	1 in 1,000	0.011531	PP	76%
2.4	1 in 1,000	0.008391	PP	28%
3.4	1 in 1,000	0.003827	PP	24%
1.5	1 in 10,000	0	NP	0%
2.5	1 in 10,000	0.002351	NP	0%
3.5	1 in 10,000	0	NP	0%
Neg	NTC	0	NP	0%

EXTRACTION LINEARITY

- Linearity of a procedure is its ability to obtain results that are directly or indirectly proportional to the concentration of a compound in a sample within a given range.



REPRODUCIBILITY & REPEATABILITY



REPRODUCIBILITY & REPEATABILITY

- Reproducibility study assesses variation when two or more people process the same sample using the same technique.
- Repeatability studies asses the variation when one person processes the same sample with the same equipment multiple times.

REPRODUCIBILITY & REPEATABILITY

Batch	Sample ID	Quantifiler Duo Concentration (ng/µl)	Profile	Alleles recovered (%)	Batch
1	RR1.1	0.448476	FP	100%	RR1.1
	RR1.2	0.90958	FP	100%	RR1.2
	RR1.3	0	FP	100%	RR1.3
	RR1.4	0.792882	FP	100%	RR1.4
	RR1.5	0.475803	FP	100%	RR1.5
2	RR2.1	0.67114	FP	100%	RR2.1
	RR2.2	1.169815	FP	100%	RR2.2
	RR2.3	0.892636	FP	100%	RR2.3
	RR2.4	0.99286	FP	100%	RR2.4
	RR2.5	0.002351	FP	100%	RR2.5
3	RR3.1	3.608488	PP	90%	RR3.1
	RR3.2	0.393792	FP	100%	RR3.2
	RR3.3	0.616598	FP	100%	RR3.3
	RR3.4	0.319349	FP	100%	RR3.4
	RR3.5	0.498053	FP	100%	RR3.5
4	RR4.1	0.738131	FP	100%	RR4.1
	RR4.2	0.462726	FP	100%	RR4.2
	RR4.3	0.305147	FP	100%	RR4.3
	RR4.4	0.744292	FP	100%	RR4.4
	RR4.5	0.390634	FP	100%	RR4.5

Sample Type	Size / Amount	Purification Protocol	Comments
Blood on FTA collection paper	0.5cm^2	Universal Extraction method	INV >MicroAmp
Blood on Fabric	0.5cm^2	Universal Extraction method	INV = MicroAmp
Saliva from drinks cup	Area swabbed using wet swab	Universal Extraction method	INV = MicroAmp
Cigarette (manufactured)	1cm of paper from around filter	Universal Extraction method	INV = MicroAmp
Nails	1 x nail clipping cut into small sections	Isolation of Total DNA from Nail Clippings and Hair	INV >MicroAmp
Envelopes	0.5cm x 1cm section of gum area	Universal Extraction method	INV = MicroAmp
Cellular material from shirt collar	0.5cm^2 section	Universal Extraction method	INV >MicroAmp
Cellular swab from shirt collar	Wet swab of collar area	Universal Extraction method	INV = MicroAmp
Chewing Gum	~30mg	Universal Extraction method	INV <MicroAmp
Sperm from Epithelial (female blood) / Sperm mixture	50µl blood 10µl Semen	Isolation of DNA from Sexual Assault Specimens	INV >MicroAmp
Soft tissue (flesh)	~10mg	Isolation of Total DNA from Tissues	INV >MicroAmp
Bone	~100mg	Isolation of total DNA from Bones and Teeth	INV >MicroAmp

CURRENTLY.....

- Evaluating demineralization (EDTA) and Na-Ac/Membrane cleanup and concentration.
- “Compromised” bone samples *failing.*
- The same sample, when demineralized and concentrated/cleaned up produces full profiles.

ACKNOWLEDGEMENTS

- Technical Management – SAPS
- Whitehead Scientific
- Qiagen