



# Interpretation Errors Detected in a NIST Interlaboratory Study on DNA Mixture Interpretation in the U.S. (MIX13)

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# NIST Interlaboratory Studies on DNA Mixture Interpretation

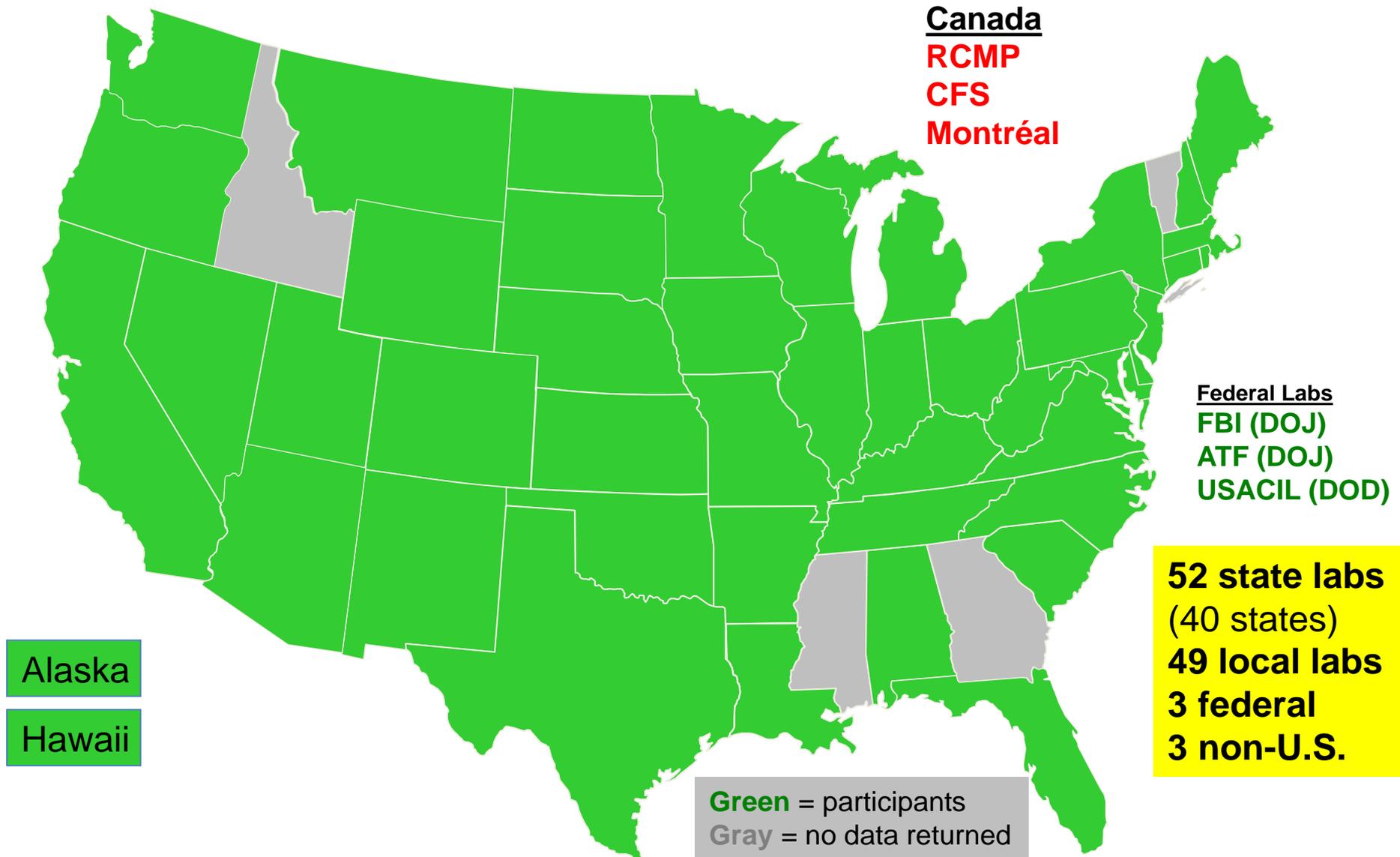
# Summary of DNA Mixture Interlaboratory Studies

<b>Study</b>	<b>Year</b>	<b># Labs</b>	<b># Samples</b>	<b>Mixture Types</b>
MSS 1	1997	22	11 stains	ss, 2p, 3p
MSS 2	1999	45	11 stains	ss, 2p, 3p
MSS 3	2000-01	74	7 extracts	ss, 2p, 3p
MIX05	2005	69	4 cases (.fsa)	only 2p
MIX13	2013	108	5 cases (.fsa)	2p, 3p, 4p

- Other recent studies
  - UK Regulator
  - DFSC (next presentation)

# MIX13 Participants from 108 Laboratories

46 states had at least one lab participate



Due to the number of laboratories responding and the federal, state, and local coverage obtained, this MIX13 interlaboratory study can be assumed to provide **a reasonable representation of current U.S. forensic DNA lab procedures across the community**

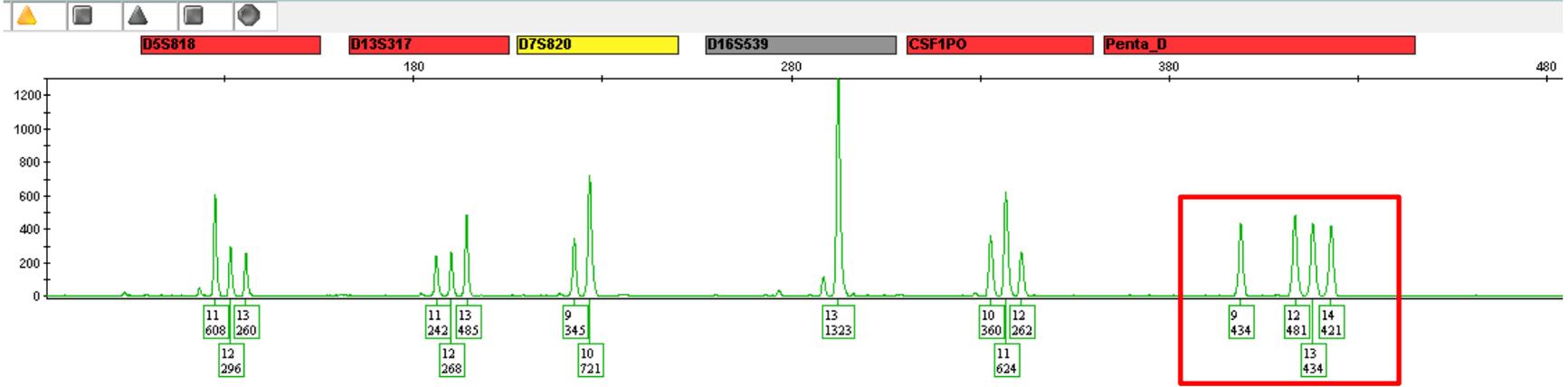
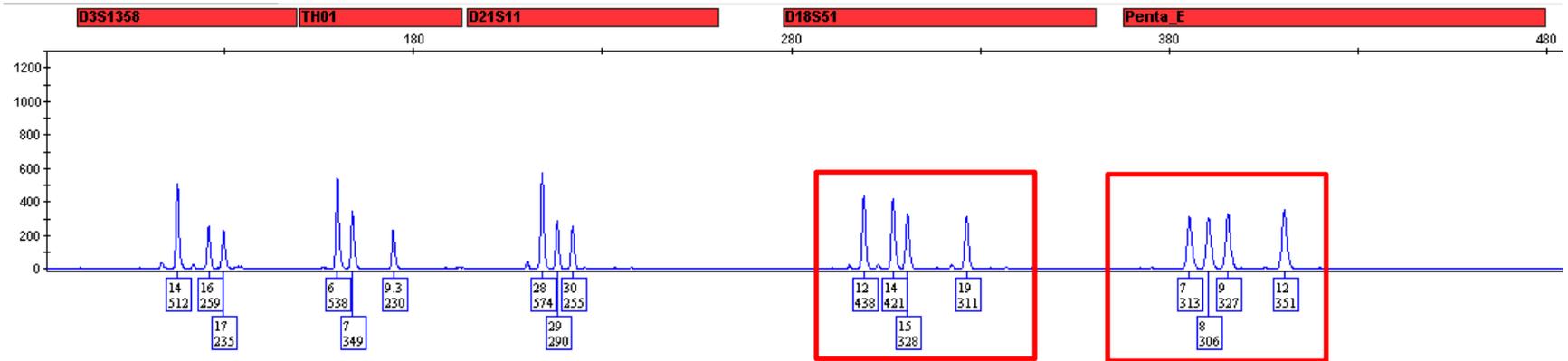
# Purpose of MIX13 Cases

	Challenge provided to study responses
Case 1	~1:1 mixture (2-person)
Case 2	<b>Low template</b> profile with potential dropout (3-person)
Case 3	Potential <b>relative</b> involved (3-person)
Case 4	Minor component (2-person)
Case 5	Complex mixture (>3-person) with <b># of contributors</b> ; inclusion/exclusion issues

# MIX13 Study (Case 01)

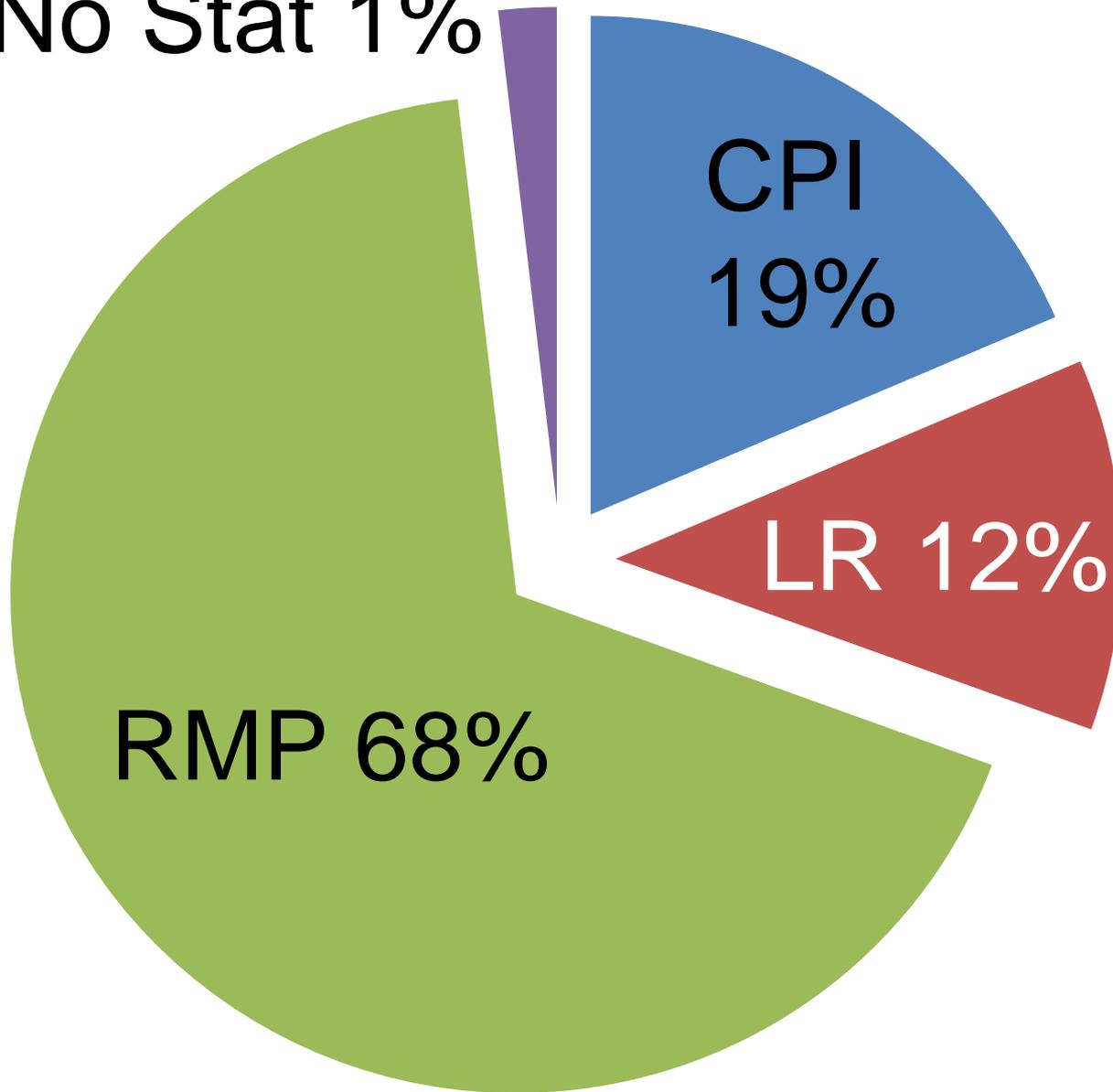
- Summary – Mock sexual assault, 2 person 50:50 mixture, all alleles above a ST of 150 RFU.
- Purpose – How many labs would consider the victim's profile and determine genotypes (deconvolution) for a mRMP statistic?

# Case 01 – PP16HS



All alleles are above a ST of 150RFU "Indistinguishable"

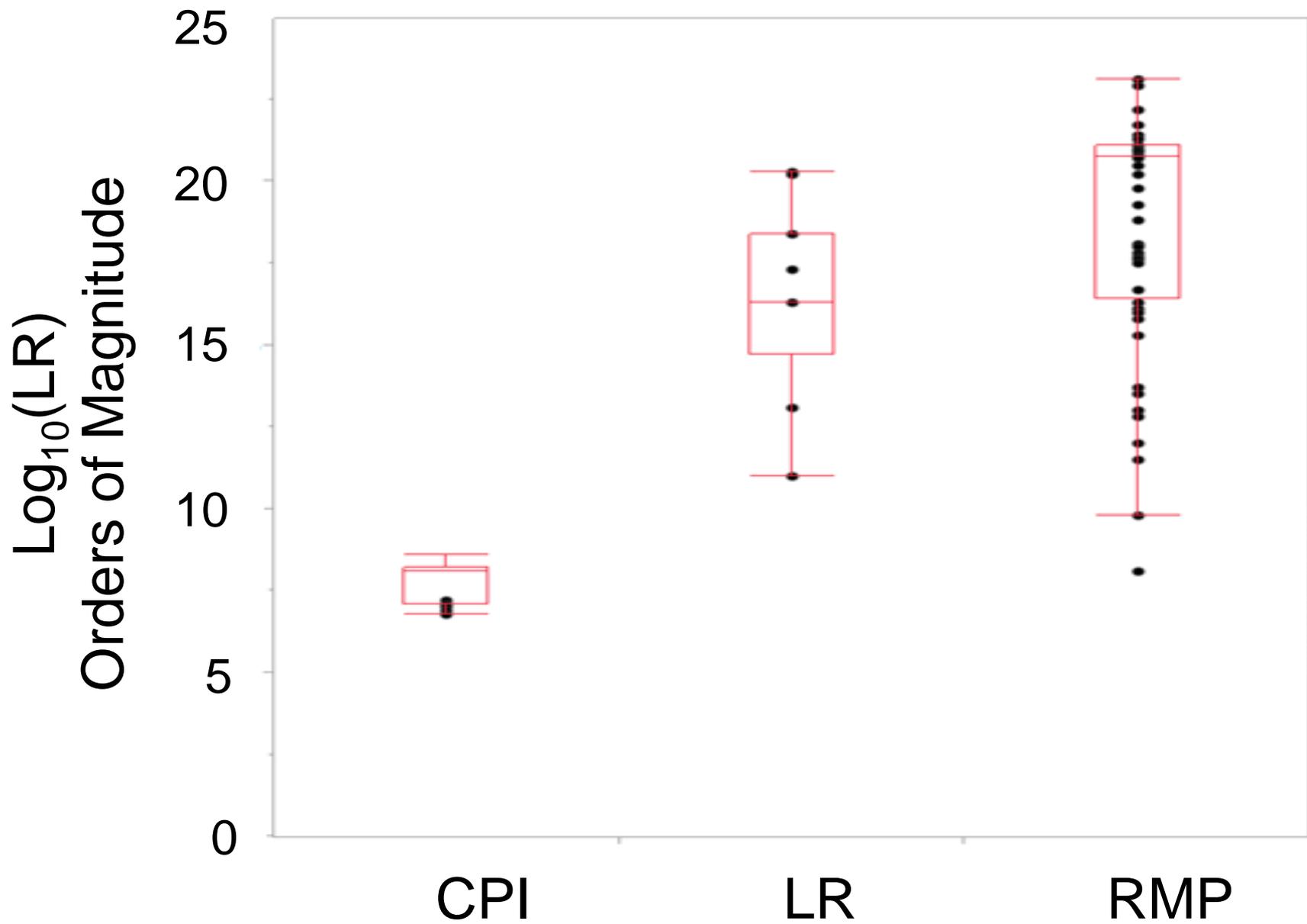
No Stat 1%



CPI  
19%

LR 12%

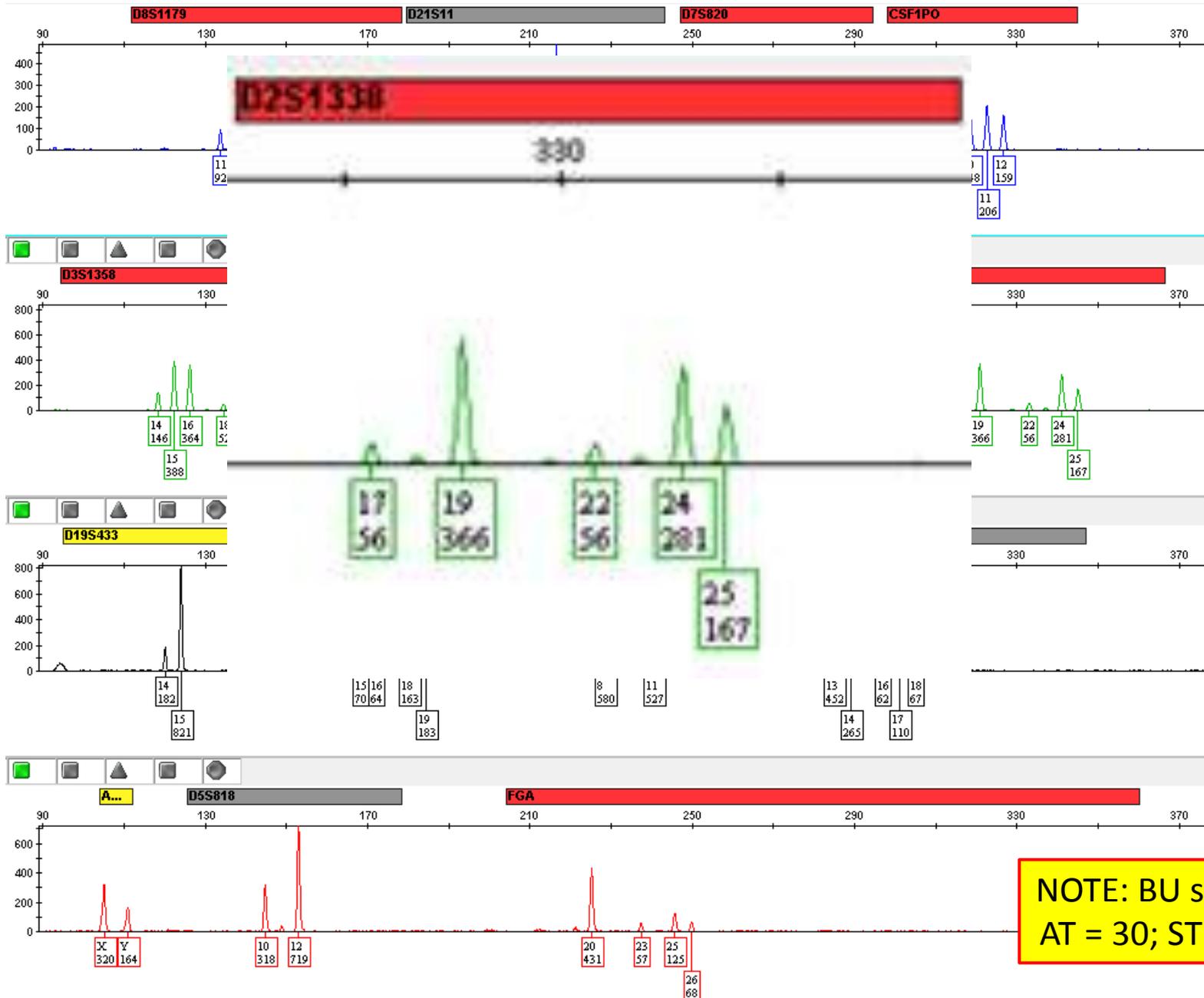
RMP 68%



# Conclusions

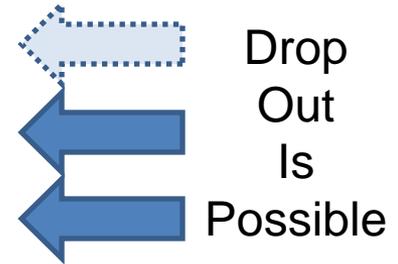
- No false exclusions
- Wide range of variation in stats reported for labs that inferred genotypes (RMP or LR).

# Case 02 – IDFile



# MIX13 Study (Case 02)

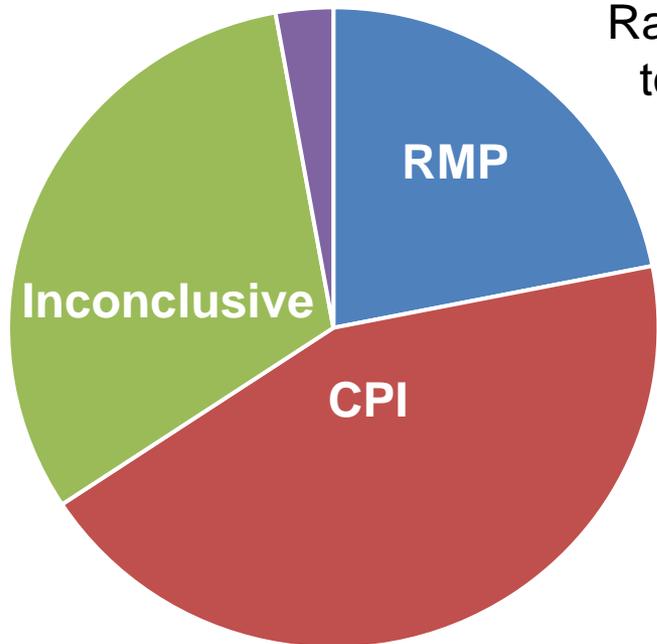
	Individual	Included?	Ratio
212 pg	Suspect A	Yes	6
53 pg	Suspect B	Yes	1.5
35 pg	Suspect C	Yes	1
	Suspect D	No	--



Total Input DNA = 300 pg

Suspect 2A

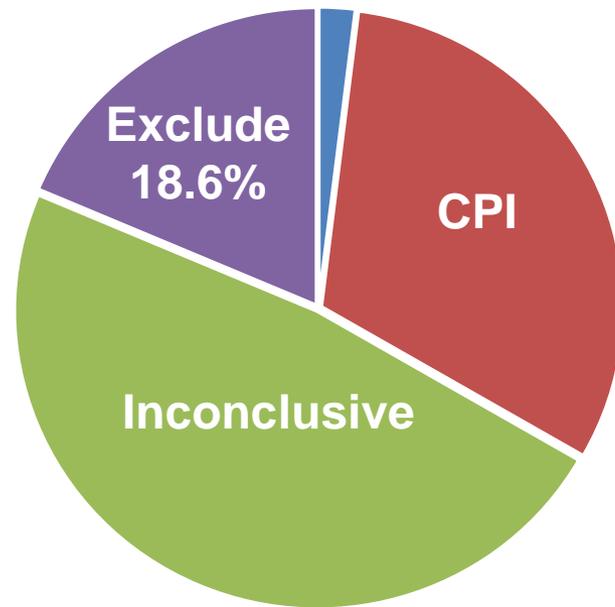
**Exclude (2.8%)**



Range = 100M  
to 1.5 Quad

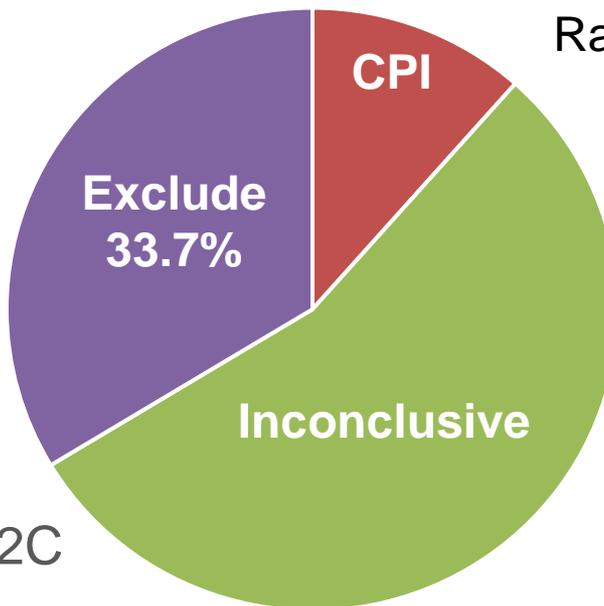
**RMP**

Suspect 2B



Range = 2.8 to 15K

Suspect 2C



# Concerns with Case 02

- One lab included Suspect D (False Inclusion).
- A substantial number of labs falsely excluded the two low-level contributors.
- Major concern with labs using CPI for contributor #3...

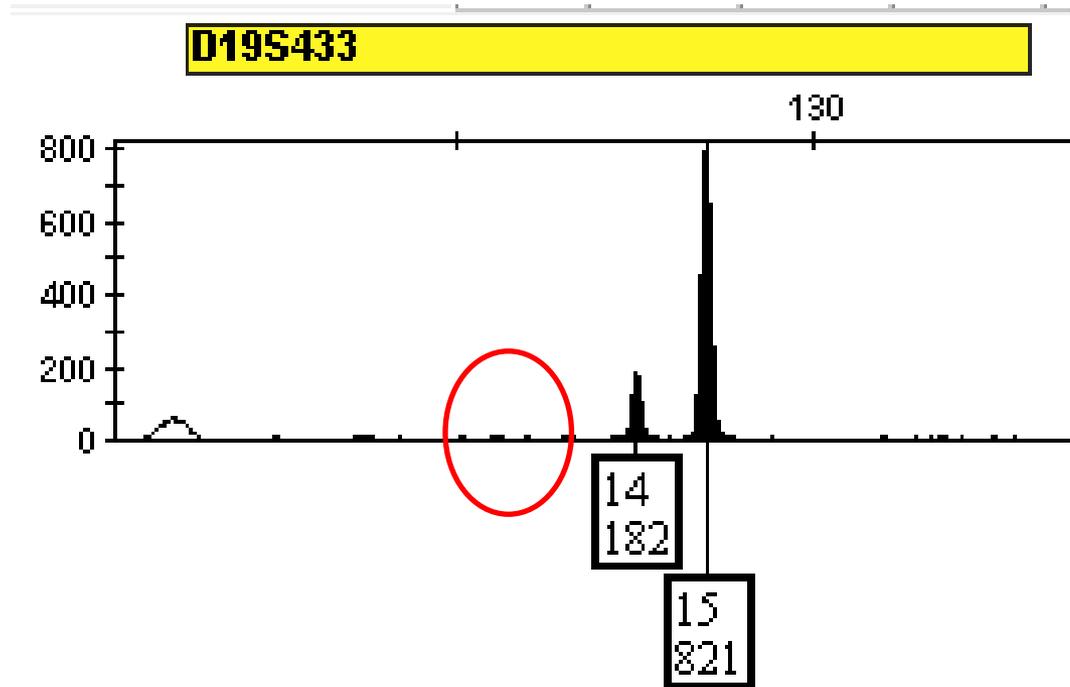
# CPI with Suspect C

## Contributors

A = 15, 15

B = 14, 15

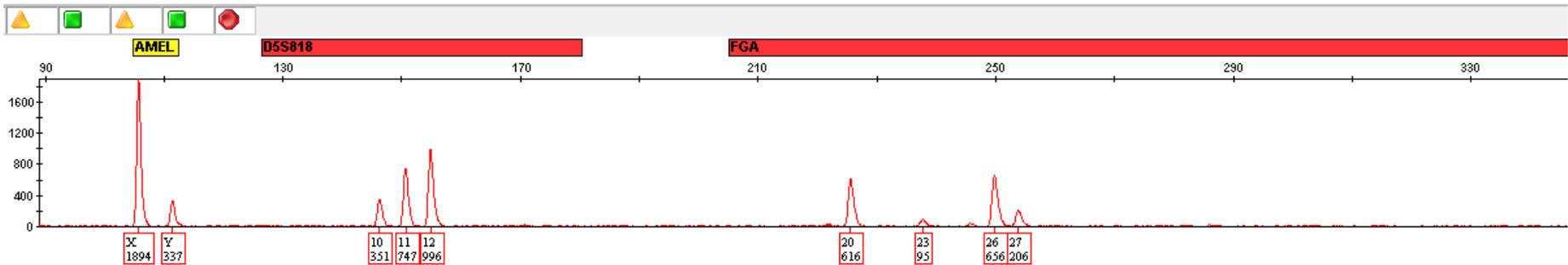
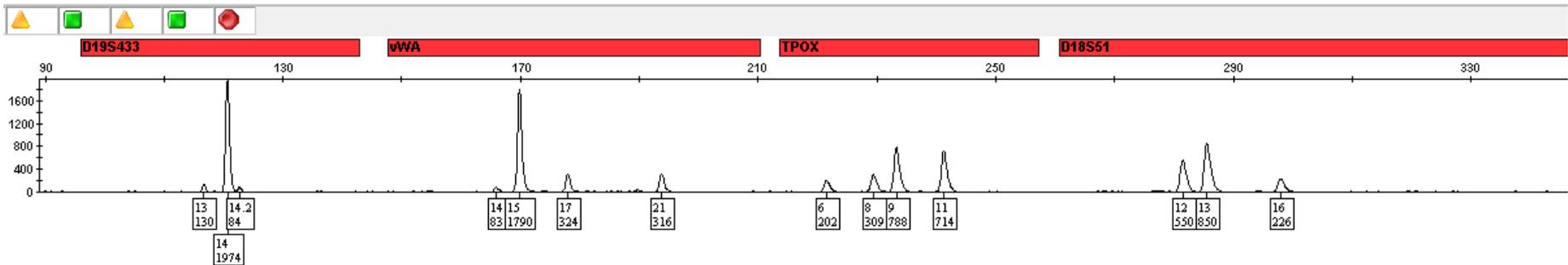
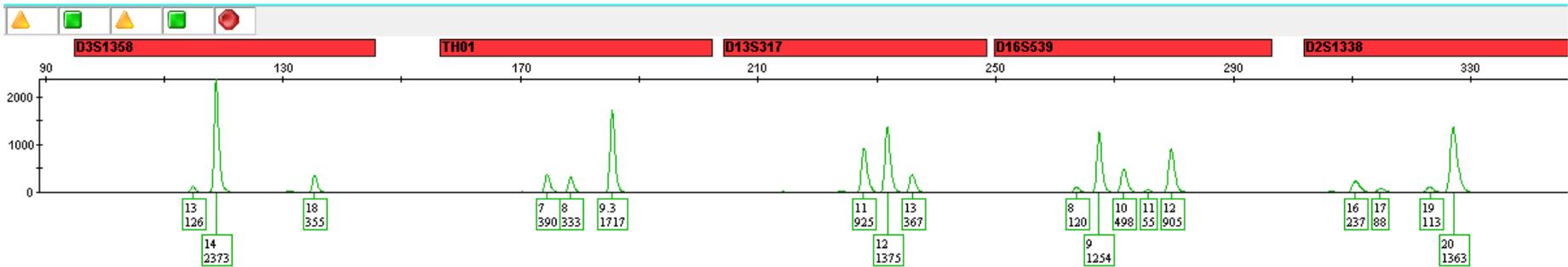
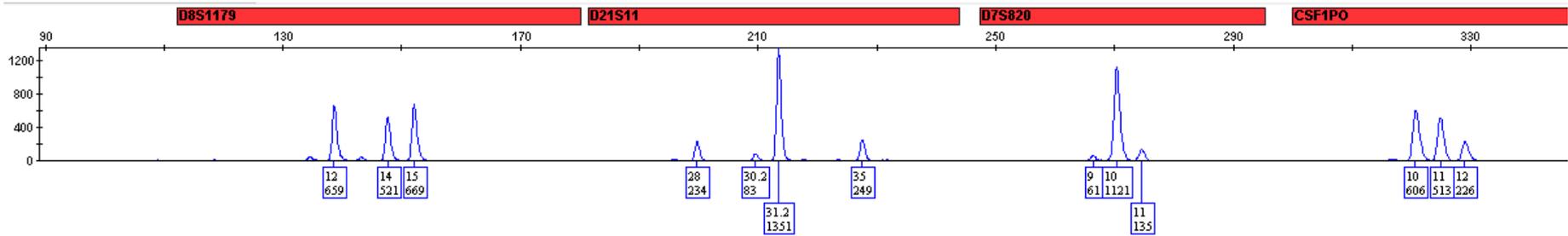
C = **12**, 14



**15 of 108 labs used CPI to include Suspect C (13.8%)**

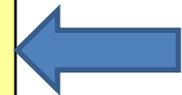
**4 of these 15 (26.6%) used D19 as a locus for CPI**

# Case 03 – IDPlus



# Case 03 – Two Suspects

Individual	Inclusion?	Ratio
Victim	Included	7
Boyfriend (CP)	Included	2
Suspect 3A (Brother)	Included	1
Suspect 3B (Friend)	Excluded	--

 Drop-out Possible

# The Brothers

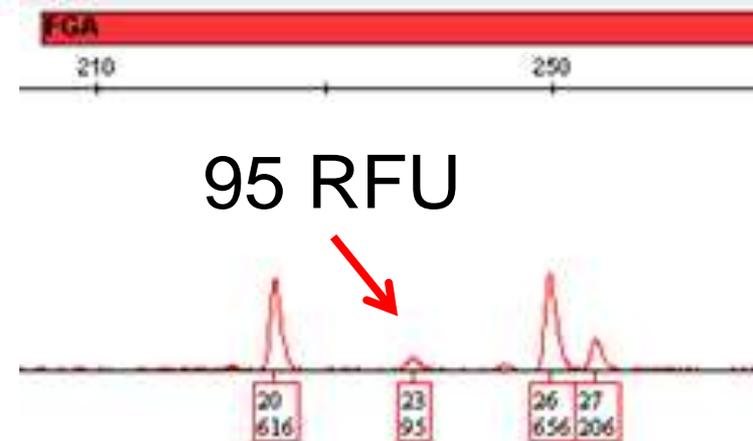
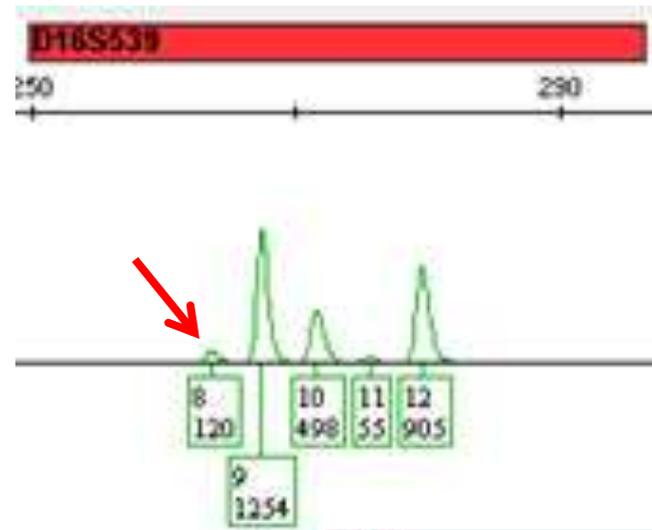
Markers	D8S1179	D21S11	D7S820	CSF1PO	D3S1358	TH01
Victim 03A	12,15	31.2,31.2	10,10	10,11	14,14	9.3,9.3
Cons Partner	14,14	28,35	10,11	10,12	14,18	7,8
Suspect 03A	14,15	28,35	10,11	12,12	14,18	7,8

Markers	D13S317	vWA	TPOX	D18S51	D5S818
Victim 03A	11,12	15,15	9,11	12,13	11,12
Cons Partner	12,13	17,21	6,8	13,16	10,12
Suspect 03A	12,13	17,21	8,9	13,16	10,12

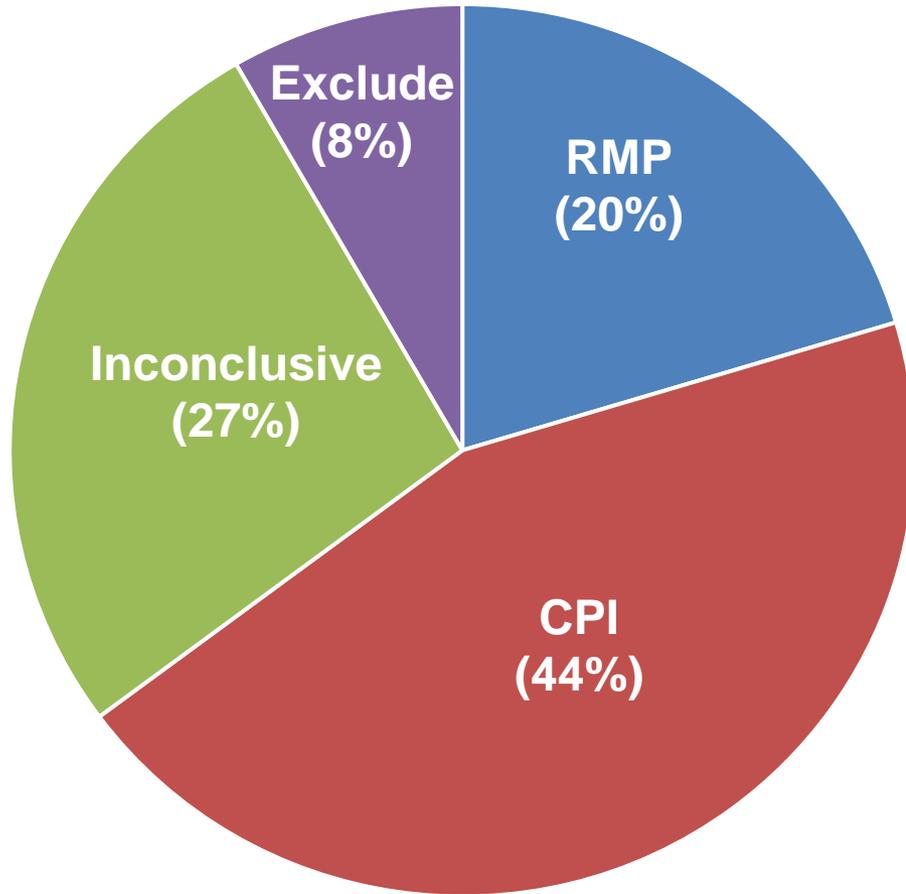
# The Brothers

For D16 and FGA – two alleles of the suspected brother are present in the epg

Markers	D16S539	FGA
Victim 03A	9,12	20,26
Cons Partner	10,10	26,27
Suspect 03A	<b>8,9</b>	<b>23,27</b>



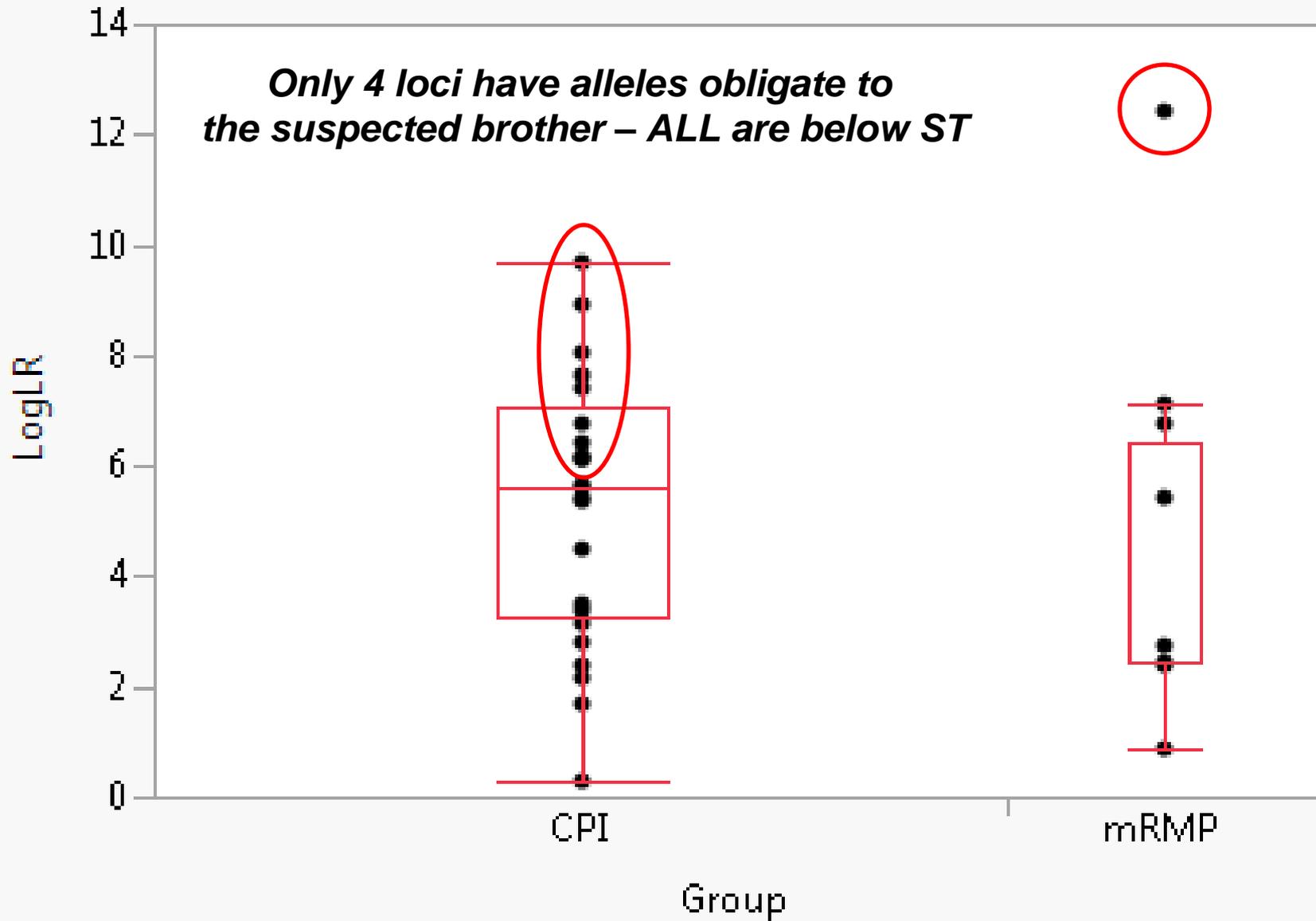
## Case 03



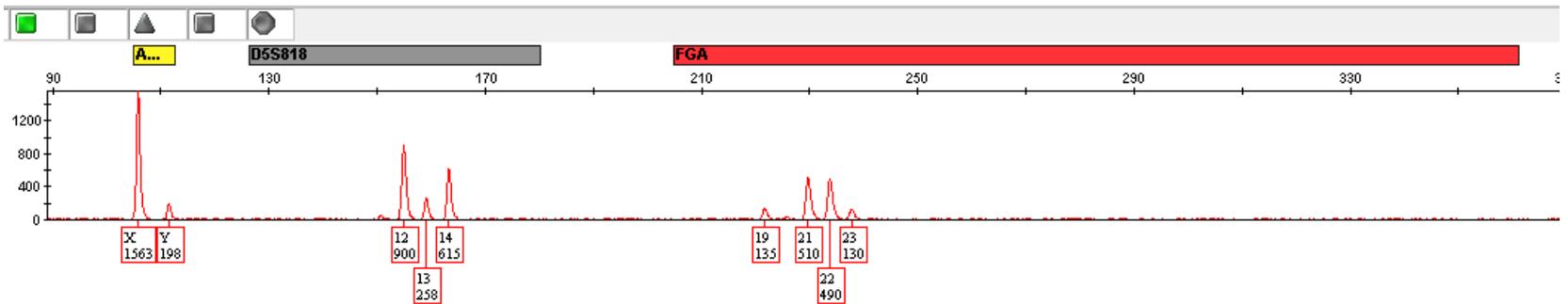
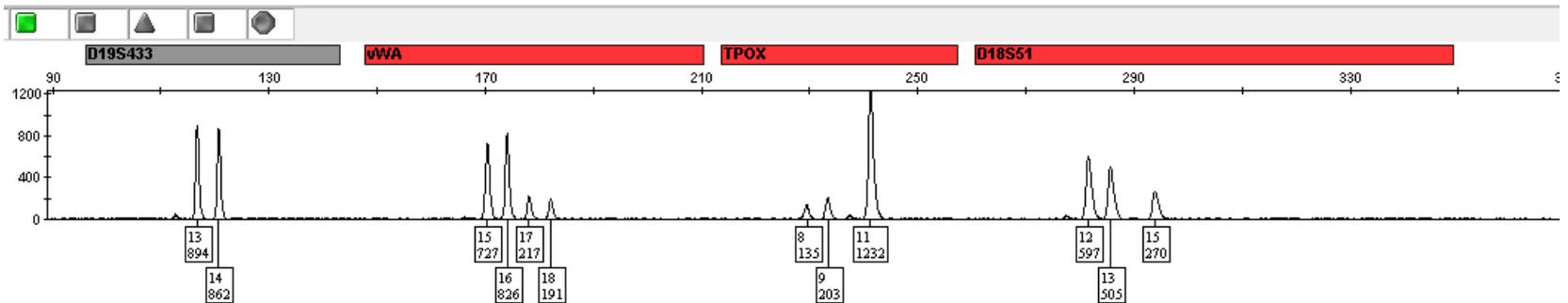
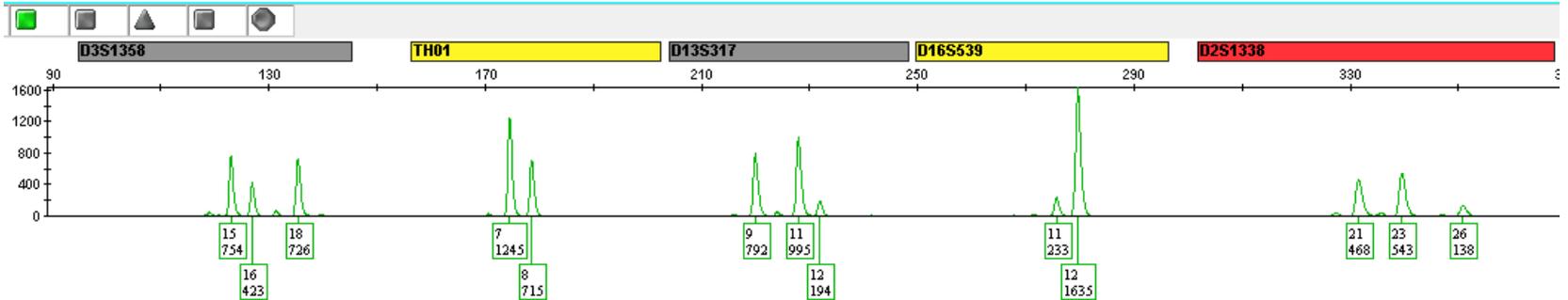
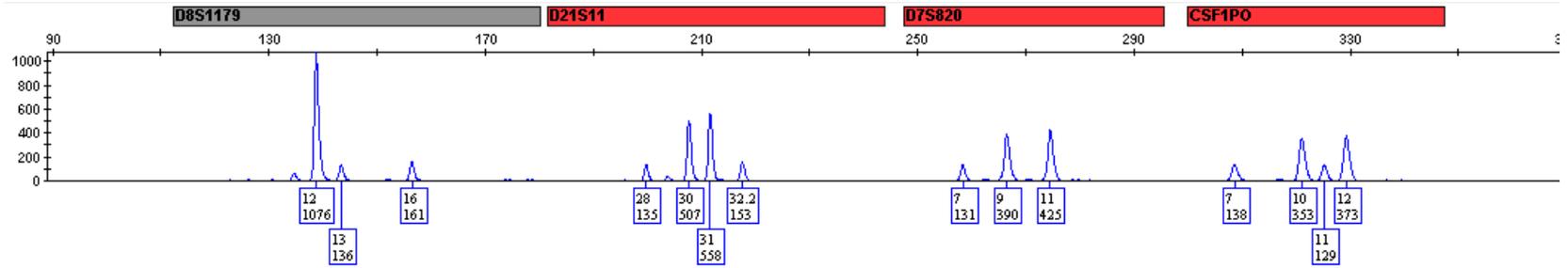
9 labs – false exclusion

1 lab – false inclusion

# Statistical Concerns



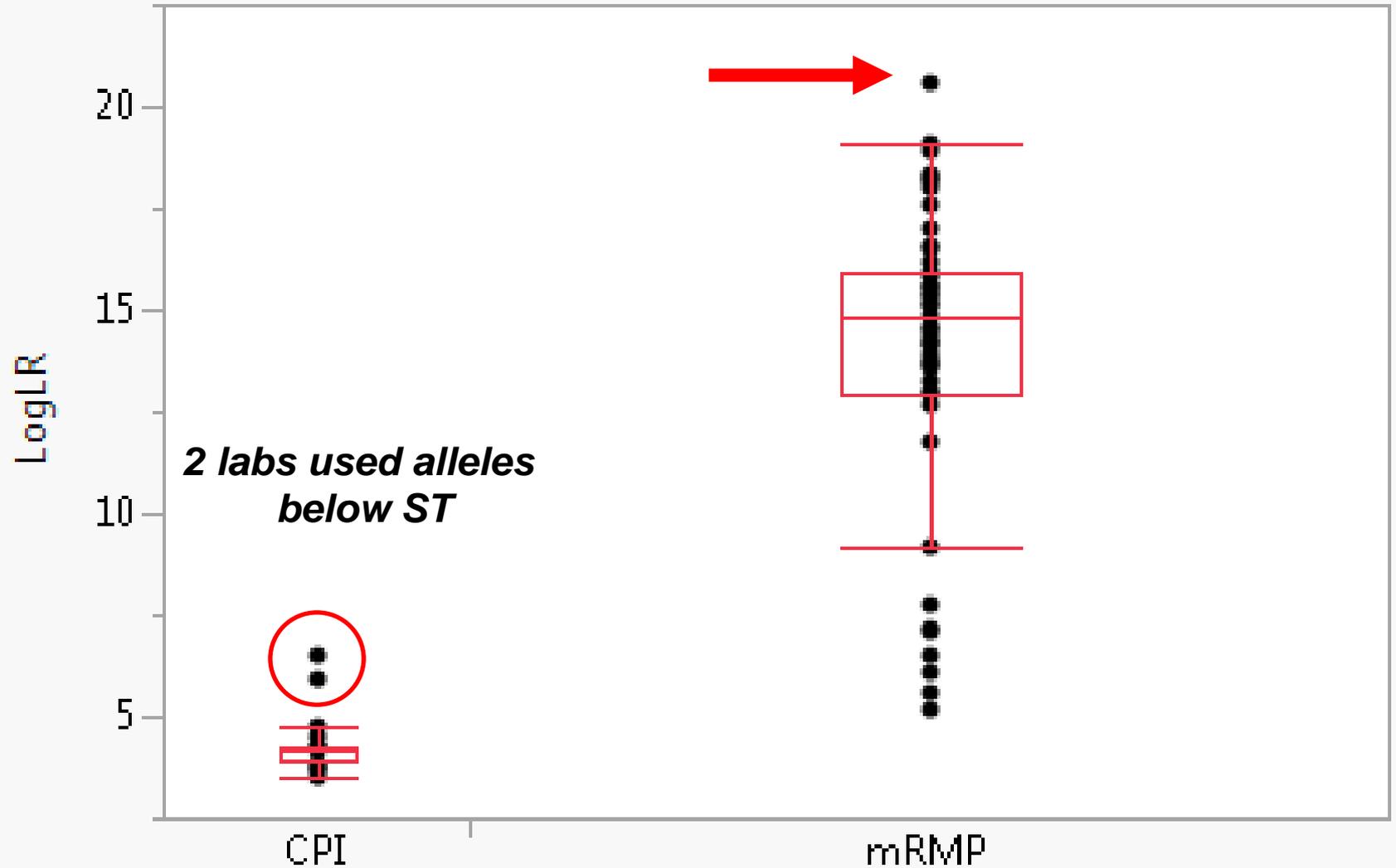
# Case 04 – IDPlus



# MIX13 Study (Case 04)

- Summary – Mock sexual assault, 2 person **3.5:1** mixture, minor component has alleles below the ST of 150 (required by all labs!)
- Purpose – How many labs would attempt to separate the two components?
- Good News! No false exclusions.

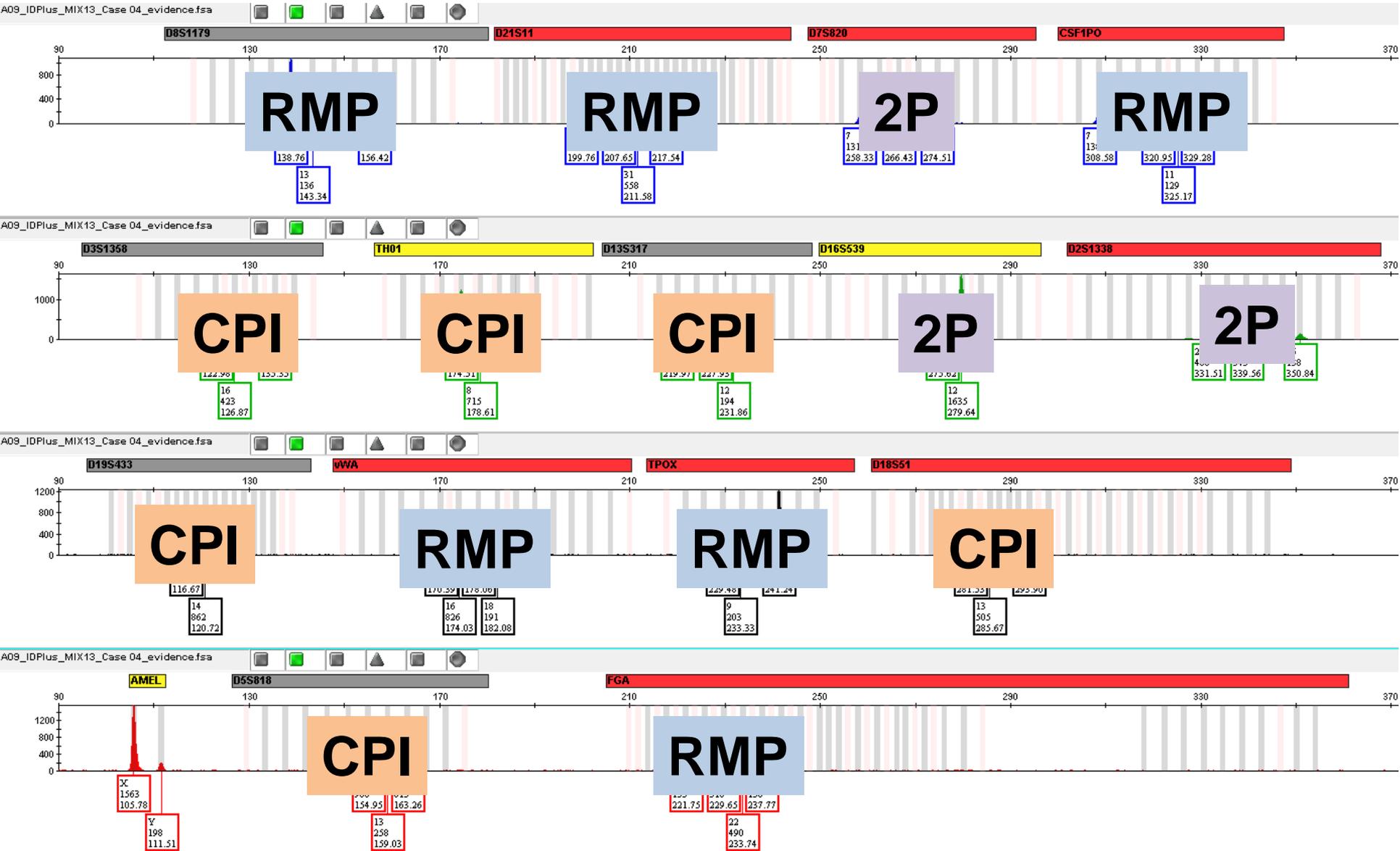
# Statistical Concerns



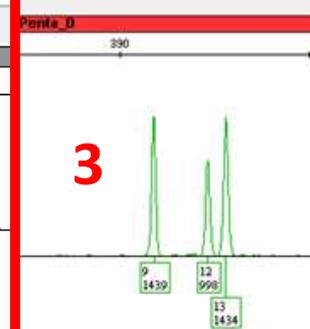
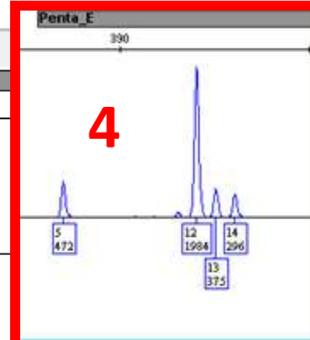
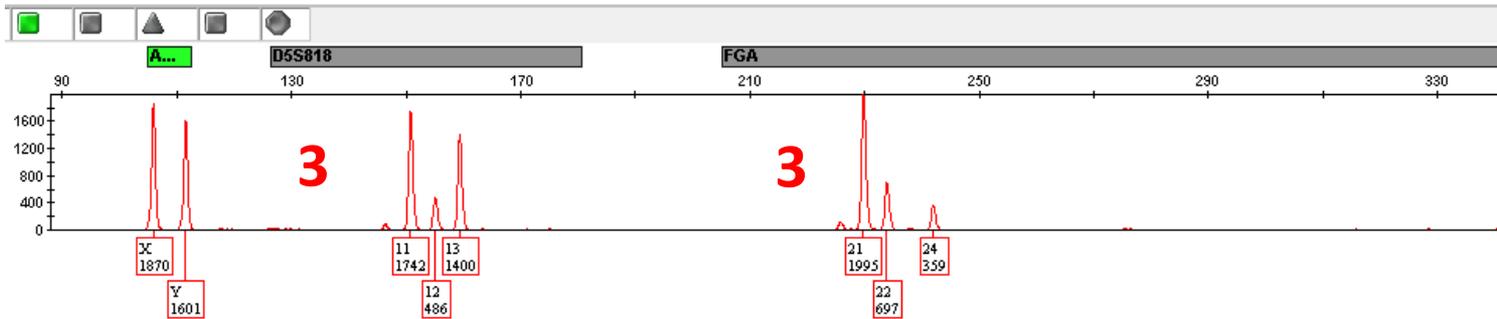
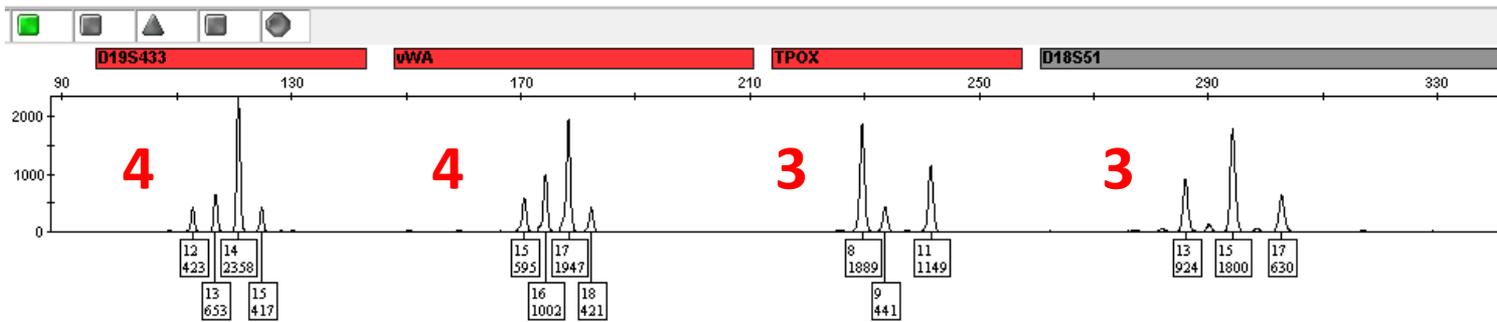
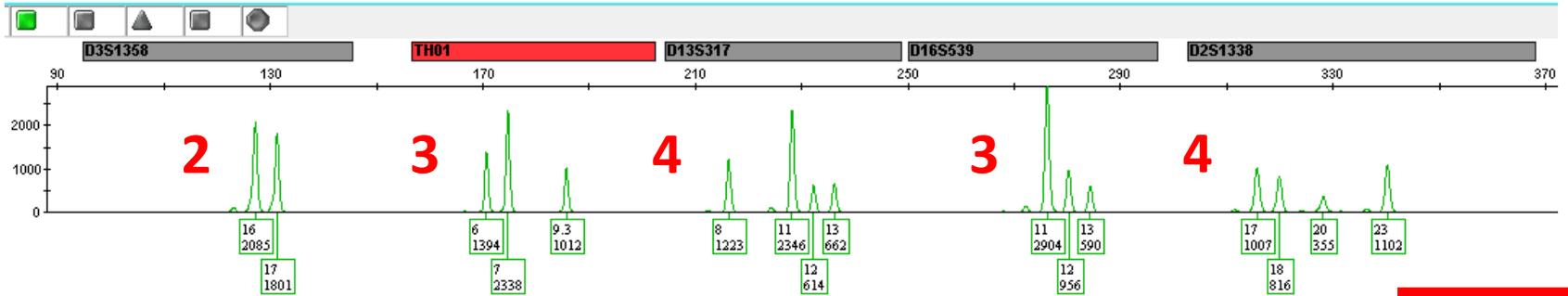
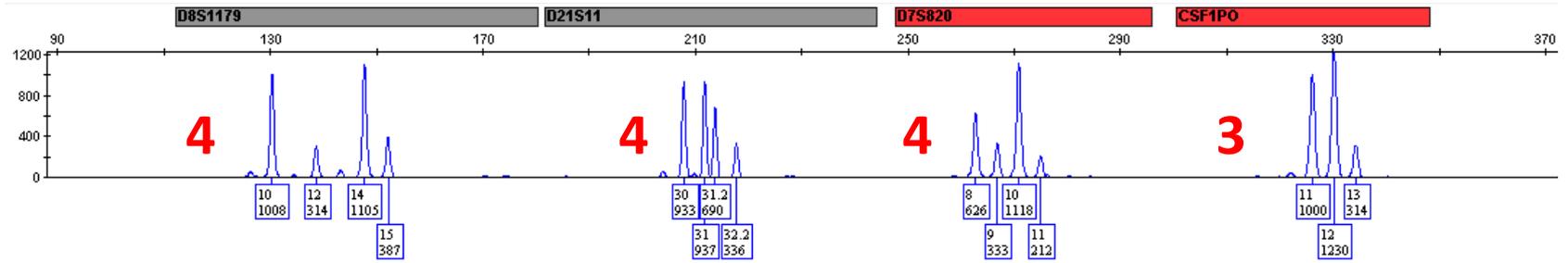
# More Statistical Concerns

- SWGDAM 2010 Autosomal STR Guidelines
- 4.6.2. It is not appropriate to calculate a composite statistic using multiple formulae for a multi-locus profile. For example, the CPI and RMP cannot be multiplied across loci in the statistical analysis of an individual DNA profile because they rely upon different fundamental assumptions about the number of contributors to the mixture.

# One Lab's Interpretation

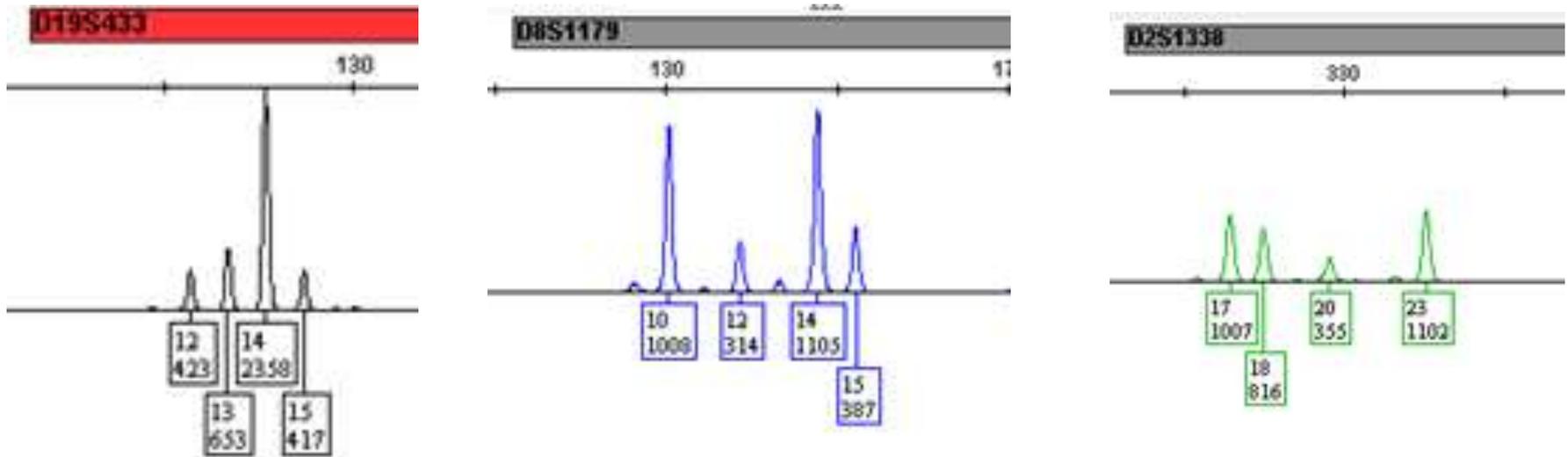


# Case 05 – IDPlus



# No more than 4 alleles at a locus

- Suggests a 2 person mixture



- Peak Height information does not agree

# MIX13 Study (Case 05)

- Summary – Mock bank robbery with ski mask evidence (touch DNA), 4 person **1:1:1:1** mixture.
- However – this mixture had no more than 4 alleles at any locus (appears as a 2p mixture). 2 of the 4 contributors were provided along with a non-contributor.
- Purpose – How many labs would consider this mixture as too complex to interpret?

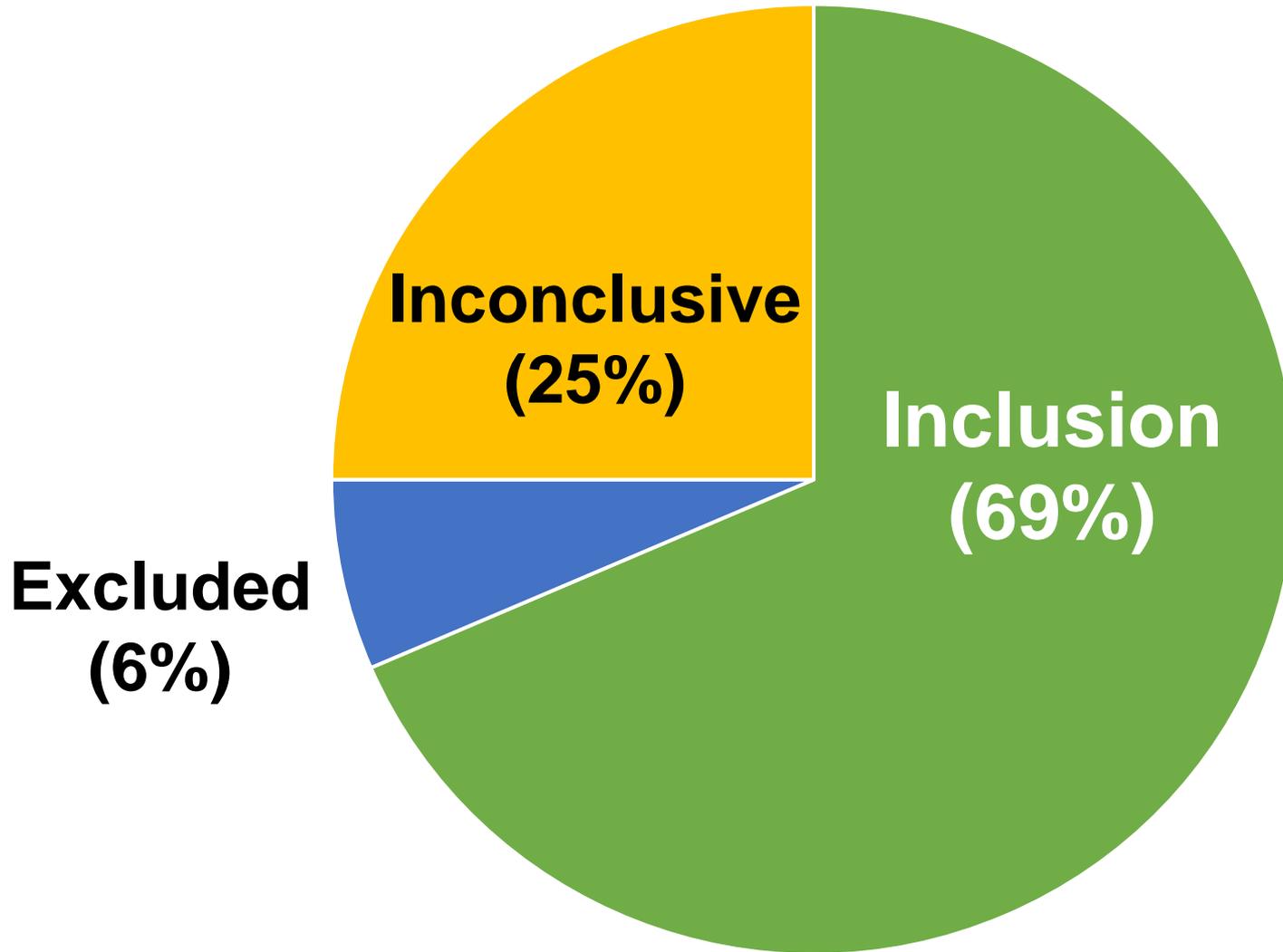
# Case 05 – 3 Suspects

Individual	
Suspect 5A	Included
Suspect 5B	Included
Suspect 5C	Not in the mixture

# Suspects A and B

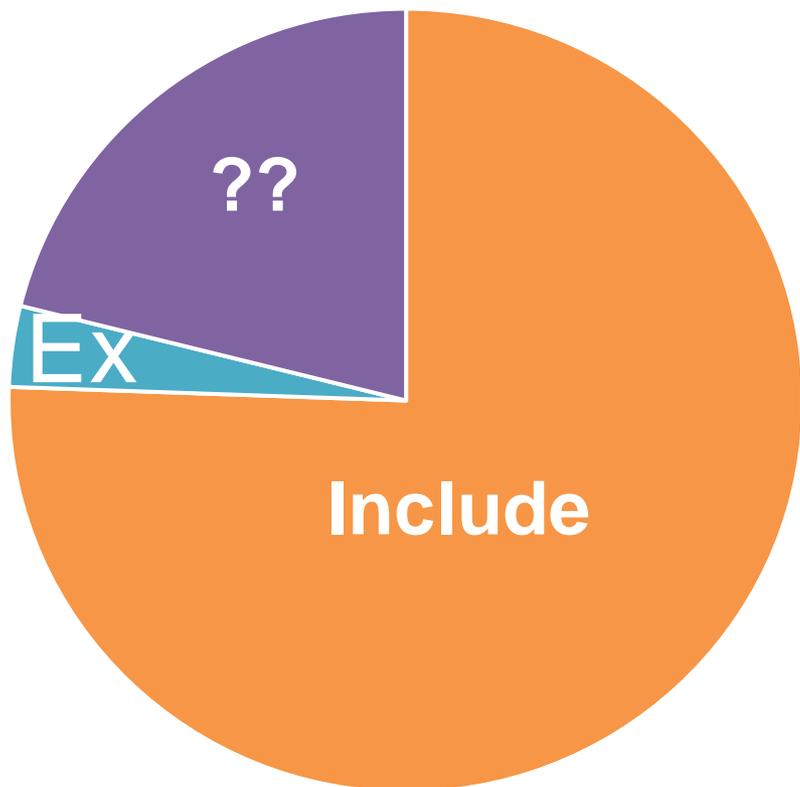
- Two labs falsely excluded suspects A, B and C due the complexity of the profile.

Case 05 – Suspect C (not in mixture)



# Exclusions by Kit

Identifiler Plus



3 of 90 labs

PP16 HS



4 of 18 labs

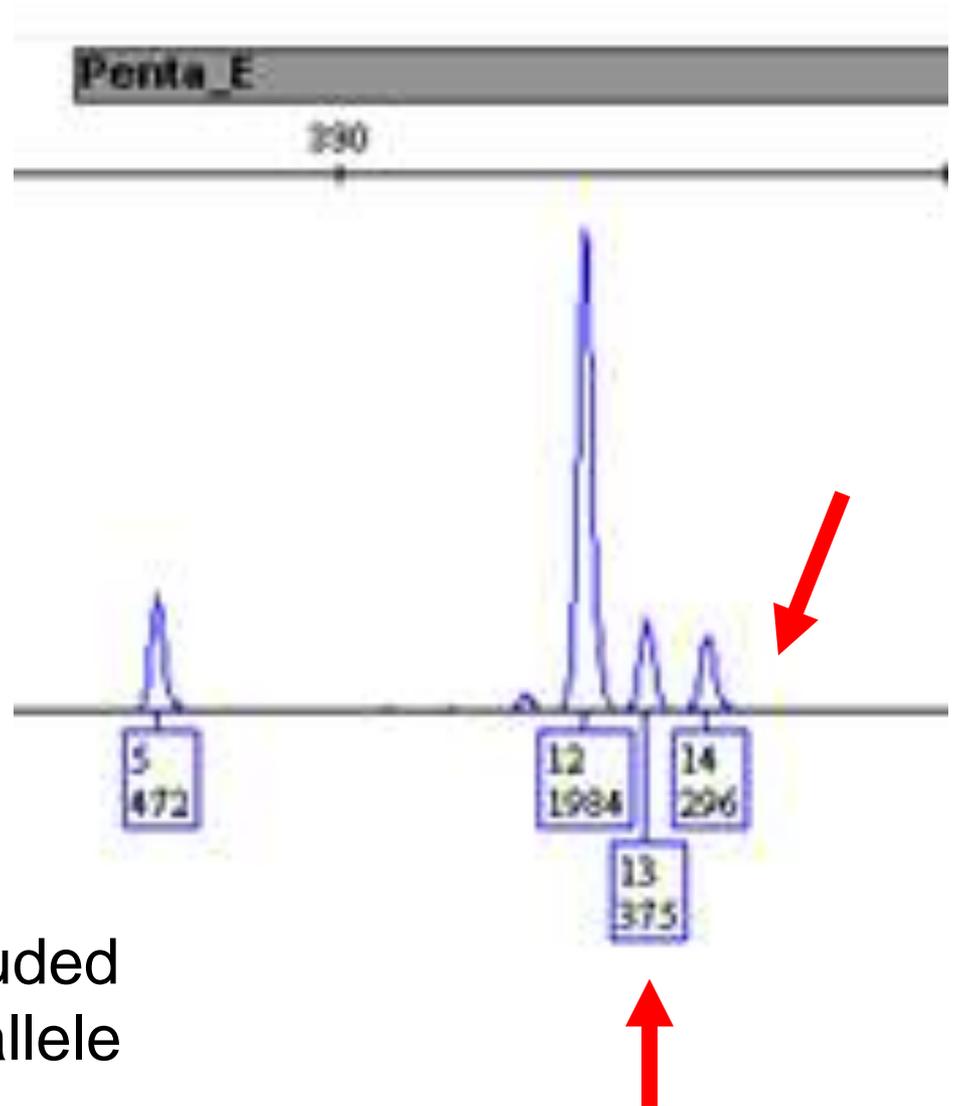
# The Challenge at Penta E

## Contributors

A = 12, 14

B = 5, 12

C = 12, **15**



Only 4 of 18 PP16 labs excluded based upon the missing 15 allele

# Concluding Thoughts

- Despite the improvements in protocols and interpretation guidelines since 2010, mixture interpretation is still all over the place.
- Some of this is a consequence of using a statistical approach that is inappropriate for complex mixture interpretation – CPI is often being used as a substitute for interpretation, and has the risk of including a non-contributor.
- Software solutions can greatly assist in the statistical evaluation and removal of bias.

# Thank you for your attention

## Acknowledgments

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