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Latest Developments in the Forensic Applications of MicroRNA analysis

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Forensic Biology Group
Body fluid identification

• Current tests are enzymatic, immunological, or histological
  – Most require some visualisation of stains before robust reporting
  – Nearly all have issues with sensitivity and specificity

• Relatively unchanged in decades

• Major capability shortfall in relation to DNA
Why is BFID necessary?

• Hierarchy of Propositions is a fundamental concept in forensic investigations
  – Offence Level (III)
    • Joe Bloggs raped Jane Smith
  – Activity Level (II)
    • Joe Bloggs had sexual intercourse with Jane Smith
  – Source Level (I)
    • Joe Bloggs’ semen was found on Jane Smith’s vaginal swabs
  – Analytical Level (O)
    • DNA profile matching that of Joe Bloggs found on Jane Smith’s vaginal swabs
What is microRNA?

DNA → MicroRNA gene → MicroRNA

Typical gene → Direct protein assembly → Protein

Messenger RNA → Binds to messenger RNA → Protein assembly is blocked
Why microRNA?

http://rnasilencing.wordpress.com
Co-extraction of DNA and miRNA

DNA extraction and washes using blood (n=15)

- Eluent
- Wash 1
- Wash 2
- Wash 3

Cq_40 - Cq hs-miR-205 and mmu-miR-451

DNA extraction step
DNA extraction on blood and saliva samples (n=90)
Body fluid mixtures

- Body fluid mixture common, particularly in sexual assaults

- Given sensitivity of microRNA analysis, mixed results are inevitable.

- Minimum criteria is that the RNA based test can actually identify the presence of mixtures
miR applications in blood and saliva mixtures

Mixtures (RNU44)

Mixtures (normalization)

ΔCq (Alg = 205 - 4.474051)

Mixture control

Blood : Saliva

Saliva : Blood

Mixing ratio/miR marker

Single-source controls

Endogenous controls

• Current work is exploring the appropriate endogenous controls; which are:
  – Human Specific
  – Equally expressed in all body fluids

• However, issues to date include:
  – Both factors are unlikely to be in the same marker, thus multiple markers required
  – The sensitivity and stability of markers must also be equivalent to that of the target markers
Current work

• Endogenous controls
• Specificity and Sensitivity

• Focus upon intimate body fluids
  – Variations within menstrual cycle
    • Including effects of contraceptive pill
  – Effect of vasectomies and infertility upon semen samples
Future work in miRNA analysis

• Age estimation

• Alteration in response to cytotoxicity
  – Potential application in long-interval toxicological analysis

• Comparison with mRNA and DNA methylation
The Forensic Genetics Research Group

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2. Kimberley Bexon
3. Donny van der Meer
4. Fathi Farag
5. Fisal Asaghiar
6. Charlotte Beever
7. Leander Stewart
8. Gabriela Roberts
Thank you for listening

Any questions?