

Reproducibility and Accuracy of Cross Platform RTQ-PCR Testing for the Detection of mRNA from Surface Squamous Cells Overlying Pigmented Skin Lesions

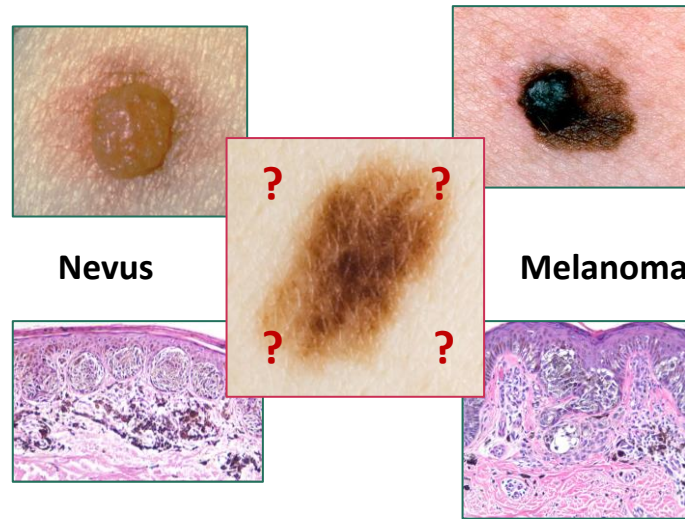
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- Molecular diagnostics in dermatology can reduce unnecessary biopsies for suspicious lesions
- RTQ-PCR is reliable for multi-gene expression signature
- Multi-gene expression assay was migrated and validated across RTQ-PCR platforms

1 Non-invasive adhesive patch for sampling pigmented skin lesions

A clinical pathologic problem solved by a molecular signature assay

Clinical and pathologic identification of pigmented skin lesions depends on visual pattern recognition, with less than acceptable sensitivity and specificity for melanocytic neoplasms.



5 We developed an accurate and reproducible expression signature

2 Specimen preparation

Partition tissue samples into <10 mg pieces

Digest tissue in MELT digestion master mix

Purify RNA with magnetic beads

96-well plate

Single tube

- Blind
- Wash
- Elute

(Optional) On-bead DNase digestion

- DNase digestion
- Repurify

reverse transcription RTQ-PCR

RNA cDNA Quantitation

4 The measurements are reproducible

Model of real time quantitative PCR plot

LifeTech QuantStudio vs Biotrove NT

$$y = 0.9933x + 1.3708$$

$$R^2 = 0.9804$$

3

OpenArray Format
2,688 reactions per array

Old platform
Biotrove NT

New platform
QuantStudio 12K Flex (LifeTechnologies)