Expression Profiling Distinguishes Melanomas from Dysplastic Nevi and Normal Skin

U133 Plus 2.0 GeneChip assay of specimens
- Background subtraction: 50
- ANOVA (p< 0.05)
- Multiple testing correction
  - Westfall & Young permutation
  - False discovery rate (q < 0.05)
- 117 differentially expressed genes selected
- Hierarchical clustering analysis
  - log2-transformed, mean-centered for arrays and genes
  - cluster with complete linkage metric
- D: Dysplastic nevi (N=38)
- M: Melanoma (N=15)
- N: Normal Skin (N=12)

Results:
- Melanomas distinguished from normal skin
- At least 3 major groups of dysplastic nevi
- Suggests existence of low and high grade nevi

Study Methods
- Following informed consent, the suspicious pigmented lesion was tape stripped four times using EGIR and then biopsied as per standard of care. Normal, uninvolved skin was tape stripped to serve as a negative control. All biopsies underwent primary and central review for histopathology. Total RNA was isolated from the tapes using MELT (Ambion, Inc.) and assessed for quality by Experion (Bio-Rad, Inc.) analysis. The yield of RNA was approximately 1 ng, as determined by quantitative RT-PCR of the specimen for β-actin gene expression. Total RNA (200-500 pg) was then amplified using the WT-Ovation Pico RNA Amplification System (NuGen, Inc.) and assayed for gene expression profile using the U133 plus 2.0 GeneChip (Affymetrix, Inc.).

Further characterization of DE genes between melanoma and dysplastic nevi
- to distinguish melanoma from dysplastic nevi
- starting with 117 genes previously defined
- FDR (0.05)
- Fold change: 2x

5 Genes Discern Melanoma Perfectly From Dysplastic Nevi

Stepwise binary logistic regression (top 10 genes out of 98 genes)

Summary
- EGIR-based tape stripping can be used to differentiate melanoma from dysplastic nevi
- Some of the differentially expressed genes between melanoma and dysplastic nevi are relevant to the biological functions of melanoma:
  - Preliminary results indicate 5 genes discriminate melanoma from dysplastic nevi with 100% accuracy
  - Gene expression profile of stratum corneum is altered, either directly or indirectly, by the presence of melanoma
- Need total of 25 – 30 melanoma samples for a robust melanoma classifier based on power and sample size assessments
- Need additional 15 – 20 melanoma samples to test robustness of the melanoma classifier

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