Review

UHN Microarray Centre

Using nanostrings for the quantification of RNA abundance in human AML samples

Review of: Payton JE, et al. High throughput digital quantification of mRNA abundance in primary human acute myeloid leukemia samples. J Clin Invest 2009, 119(6):1714

Acute promyelocytic leukemia (APL), a subtype (M3) of acute myeloid leukemia (AML), is characterised by the t(15;17) chromosomal translocation. This translocation results in the fusion of the retinoic acid receptor α gene to another gene, most commonly promyelocytic leukemia (PML), and this fusion protein has been shown to initiate APL in mouse models (1-4). Unlike most other AML subtypes, the initiating event of APL is known, making it a good model for the study of disease progression (5).

This paper reports a gene expression signature that is specific to M3 samples. This signature was not found in other AML subtypes and did not represent the expression pattern of normal promyelocytes. The experimental design of this study, which included profiling a large number of de novo AML samples and normal human myeloid samples, allowed researchers to define malignancy-specific gene expression signatures that were both unique and highly reproducible for M3 AML. By comparing with normal enriched promyleocyte samples, genes that were simply markers of the promyelocyte stage of myeloid development were filtered out (5).

The authors note that one shortcoming of many expression profiling studies involving AML is that only a small number of genes are validated in a small number of samples due to limiting clinical material and the labour-intensive and costly nature of qRT-PCR (5). This study, which used the Nanostring nCounter for validation, is the first to use a high throughput digital system to assay the expression of a large number of genes in 28 primary clinical samples. In this study, triplicate measurements of the expression levels of 46 genes were obtained using 300 ng of total RNA, using multiplexed reactions; about 10% of the RNA that would have been required for qPCR.

The findings of this study have implications for other types of cancer as well. For example, the amount of total RNA isolated from the tissue extracted using a fine needle biopsy would be sufficient for nCounter assays of hundreds of genes. This study found the nCounter method to be highly reproducible and an invaluable tool for biomarker measurement in low-abundance clinical samples.

References

1. Grisolano JL, *et al.* Altered myeloid development and acute leukemia in transgenic mice expressing PML-RAR alpha under control of cathepsin G regulatory sequences. Blood 1997, 89:376
2. Brown D, *et al*. A PMLRARalpha transgene initiates murine acute promyelocytic leukemia. PNAS 1997, 94:2551
3. He LZ, *et al*. Acute leukemia with promyelocytic features in PML/RARalpha transgenic mice. PNAS 1997, 94:5302
4. Westervelt P, *et al*. High-penetrance mouse model of acute promyelocytic leukemia with very low levels of PML-RARalpha expression. Blood 2003, 102:1857
5. Payton JE, *et al*. High throughput digital quantification of mRNA abundance in primary human acute myeloid leukemia samples. J Clin Invest 2009, 119(6):1714