obtained fold change values was investigated and discussed with regard to non-specific binding of labeled species in the sample solution. The customized cDNA chips were used in studies within pharma drug development and the results were compared to RT-PCR.

Bunker, Christopher

Macrophage function in lipoprotein/cholesterol metabolism and atherosclerosis

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We have established a cDNA microarray platform for use in research focused on atherosclerosis and heart disease. Our objective is the elucidation of signalling pathways with an impact on cholesterol metabolism regulation. Specifically, we aim to discover novel genes and their encoded products that could serve as drugs, drug targets or diagnostic markers for this indication. Our specific focus is on modulating gene function to promote cholesterol efflux from peripheral tissues (reverse cholesterol transport) via HDL, thereby alleviating the potentially deleterious accumulation and oxidation of cholesterol, most notably in macrophage foam cells, that is strongly correlated with atherosclerosis.

Macrophages mediate a limited-specificity immune response that includes recognition of 'foreign' lipid derivatives. Macrophages display a diverse set of 'scavenger receptors' that bind bacterial lipopolysaccharides (LPS), phospholipids inappropriately exposed on apoptotic cells and oxidized forms of LDL cholesterol. The resulting signal transduction events and transcriptional programs are overlapping but distinct. Whereas LPS induces a transcriptional program constituting an inflammatory response, the binding and phagocytosis of apoptotic cells results in a non- (or anti-) inflammatory response. It appears that oxidized LDL or oxidized cholesterol metabolites stimulate an inflammatory response that becomes chronic and pathological as they accumulate to high levels in macrophage foam cells.

A full understanding of the transcriptional programs and signalling cascades mediated by related scavenger receptor ligands will enable the identification of unique components of each signalling program, which may provide opportunities for drug development. In addition to targeting genes involved in an abherent inflammatory response to cholesterol accumulation, target genes may regulate cholesterol metabolism and have an impact on efflux mechanisms.

Our current microarray analyses cover approximately 50% of the human genome (~57,000 cDNAs from IMAGE, distributed by Genome Systems). We are collecting data on gene expression in cultured and primary monocytes/macrophage and those accompanying foam cell formation and stimulation by bacterial lipids and apoptotic cells. A future objective is the analysis of monocyte/macrophage-specific arrays for a more targeted gene discovery effort. Novel candidate genes will be confirmed and validated in physiological assays of cholesterol metabolism and macrophage function.

Bushnell, Steven

GECKO: a software system for the analysis and organization of gene expression information

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The advent of large-scale gene expression technology in pharmaceutical drug discovery has precipitated an acute need for bioinformatics systems and methods for data analysis. These include methods for: data storage; integration of LIMs information; assessment of chip quality; normalization; metrics for the confidence of fold changes; integration with annotation information; and clustering approaches for the analysis of temporal and time series information. We present a suite of tools, called 'gene expression computation and knowledge organization' (GECKO), developed at the Hoechst-ARIAD Genomics Center, LLC, for the analysis of high-throughput (thousands of scans per year) gene expression data from both oligonucleotide (Affymetrix) and spotted cDNA (Molecular Dynamics) technology. As quality metrics are central to meaningful data interpretation, we also present a method, grounded in a Bayesian framework, for estimating the distribution of fold changes from inputs of experimental noise and show how this can be used to establish a '*P*-value' to represent the statistical significance of fold change.

Buxbaum, Joseph

RNA profiling in neuropsychiatric disorders

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There are several critical requirements for successful identification (or exclusion) of specific alterations in expression profiles in brain samples from individuals with neuropsychiatric disorders. These requirements include antemortem neuropsychiatric assessment, short post-mortem intervals, preservation of RNA and protein integrity, and, because of likely heterogeneity, large numbers of cases and controls. The Department of Psychiatry of the Mount Sinai School of Medicine has been collecting brain samples from antemortem assessed subjects since 1985. The original focus was on Alzheimer disease (AD), but was expanded a decade ago to include psychiatrically ill patients, especially those with schizophrenia. By collaborating with—and providing clinical services for—long-term care facilities, the Brain Bank has established a system whereby residents of the facilities are followed for long periods and tested annually or semi-annually with an extensive array of neuropsychiatric instruments. Post-mortem intervals (PMI) are short (one-half less than seven hours and a one-sixth under three hours), and tissue from half the brain is immediately snap frozen for RNA and other studies. The remaining tissue is paraformaldehyde fixed and examined for a large battery of neuropathological changes in a blinded manner. Recently, RNA profiling, first using differential display and now using microarrays, has been undertaken in both the AD and schizophrenia cohort.

In AD we have been studying a large cohort (n=79) of cases chosen to particularly represent early cognitive decline (clinical dementia rating of 0–2), with the expectation that early markers of dementia will be identified. The sample cohort was selected from a group of 278 consecutively, rapidly autopsied subjects (between 1986 and 1997) who had been residents of the Jewish Home and Hospital in Manhattan and the Bronx, New York. A multi-step approach was applied to the assignment of clinical dementia rating (CDR) score based on cog-