

Presentation 9 – Nancy Klimas

GWV – The Miami Experience

- Our Center is comparing GWV, CFS and deployed healthy GW veterans in a longitudinal study.
- The initial assessment is an extensive evaluation and blood and saliva collections pre-post an exercise challenge; the subjects are then followed over time to assess the trajectory and variability of the illness as it relates to biomarkers.

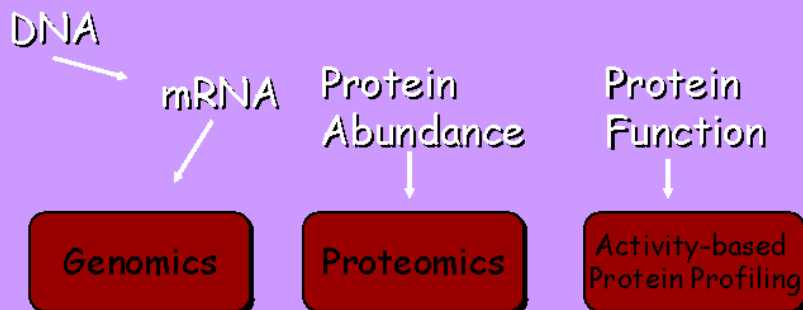
GWV and CFS comparative studies – The Miami experience

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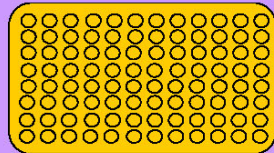
GWV - The Miami Experience

- Samples are then processed for use in the following:
- Genomics – 20,000 gene array pre post exercise; (proteomics to follow)
- Immune regulation – functional studies, cytokine
- Neuroendocrine studies
- Neuropeptide Y

Molecular Epidemiology Laboratory Strategy



Microarray Technology

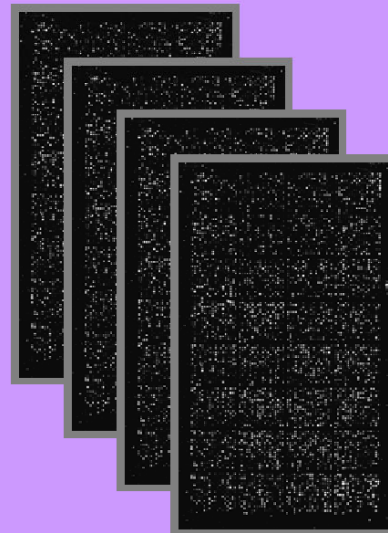
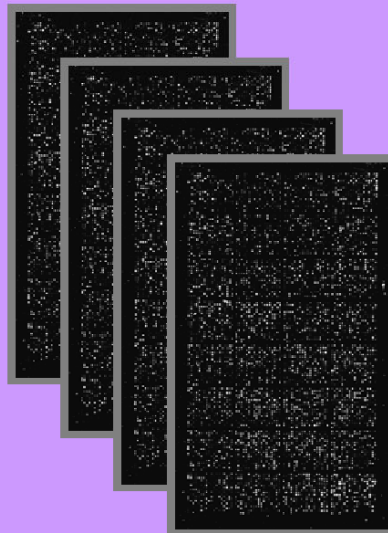


- A tool to measure the expression (mRNA) of genes
- An ordered array of spots (that represent genes) on a glass microscope slide

Atlanta Case Control Study Gene Expression

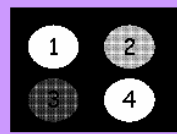
CFS Case Arrays

CFS Control Arrays

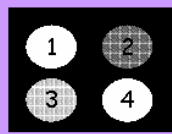


How do we use microarray data?

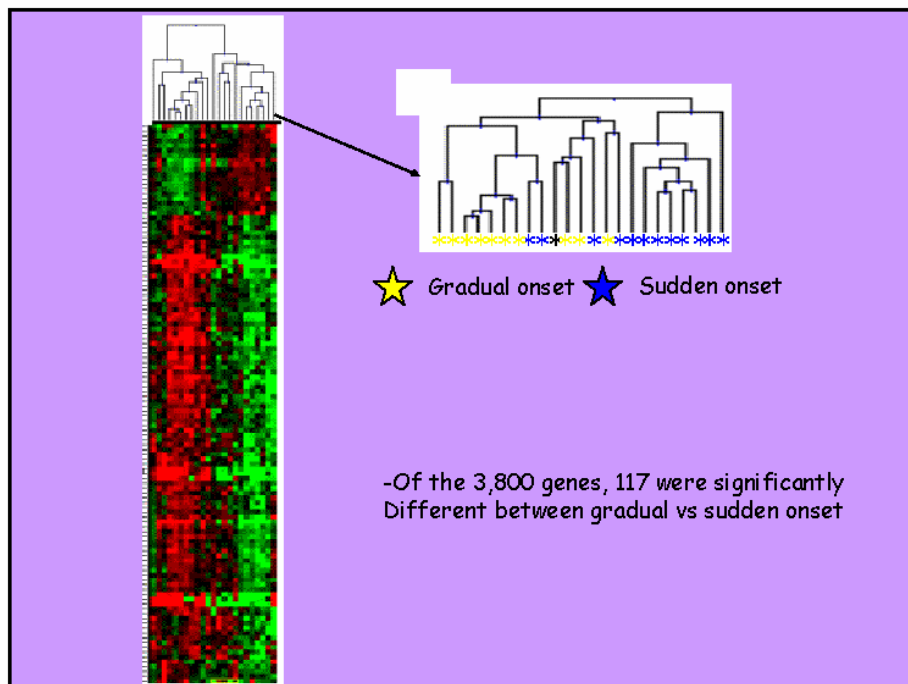
- Compare the intensity of one spot (CFS) to the intensity of the corresponding spot (control)



vs



	CFS	Cont	Ratio
1	100	100	1
2	60	30	2
3	20	80	4
4	100	100	1



Patterns of gene activation and protein expression

- Gene expression patterns break population into two groups, one with increases in gene expression involved in immune activation, another with lower levels of gene expression in areas reflecting metabolism

Whistler et al BMC Physiology 2005 (5)5 e- journal

CFS subtyping by gene expression

- “Molecular” evidence of a difference between suddenly & those with gradual illness onset
- Different profiles imply different pathophysiology
- Several immune, endocrine and metabolic genes and pathways involved
- Differentially expressed genes in RNA processing and metabolic pathways account for the differences between CFS and controls
- All gene ontology categories are required to distinguish someone with CFS from someone without (can’t just use one)
- There are hundreds of gene ontologies – CDC studies narrowed CFS classification to 26

What Did the Genes Teach Us?

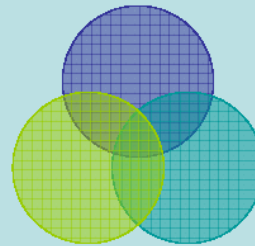
- CDC CFS study: Of 20,000 genes studied the activity of 26 genes did accurately predict which of six categories of chronic fatigue a patient had on the basis of symptoms and other clinical tests.
- Most of these genes are involved in immune system regulation, the HPA axis, and mitochondrial function.
- Studies of hormones and immune factors confirm these predictions
- Kerr's group: 35 genes in 25 pts/25 controls suggesting T cell activation, and perturbations of neuronal and mitochondrial function.

What Did the Genes Teach Us in CFS?

- In other analyses, involving 50 genes that some people inherit with seemingly minor "misspellings," five of the 500 genetic glitches that were tracked repeatedly correlated with an apparent susceptibility to chronic fatigue.
- Those five include genes that affect levels of serotonin and glutamate
- Additional studies of cortisol regulation pathway genes reveal mutations in regulatory pathway

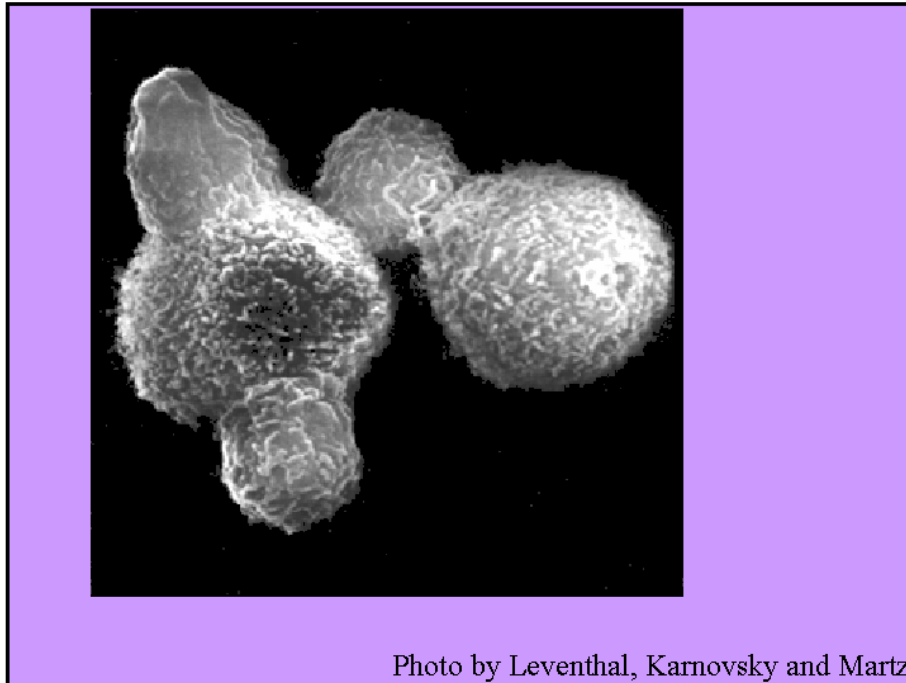
CFS is a Complex Illness

- CFS Illness represents alterations in complex systems of homeostasis
- Not a result of a single mutation or single environmental factor
- Arise from a combined action of many genes, environmental factors and risk-conferring behavior



GWVI - Miami

- Recruiting is underway
- Interim data available on immune studies pre post exercise challenge
- Gene array data is being performed at the CDC for comparability to their data set



GWV

- Impact of acute exercise on circulating populations of cells
- Selective shift in compartment of cell subpopulations

Conclusion

- 1. Gene array work and suggested proteomics offers a unique opportunity to better understand the complex underpinnings of multi symptom illness.
- 2. The peripheral blood compartment is uniquely suited to study neuro-immune regulation at the genomic level.
- 3. CFS and GWI share many commonalities, with what seems to be overlapping mechanistic studies of immune dysfunction.