

**What impact will the genomics field
have on the immediate or the near-
term development of biomarkers for
nervous system diseases?**

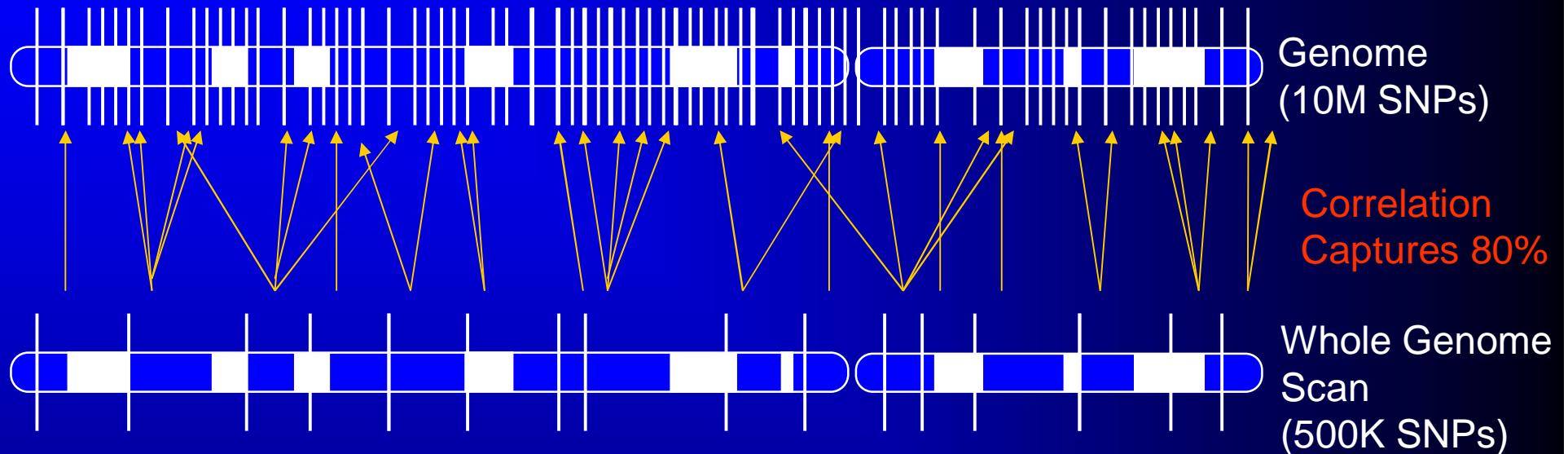
Allen D Roses, MD

IOM Biomarker Conference, 26 Feb 2007

Outline of presentation

- **Genome wide SNP scanning [gws] for:**
 - Human disease-specific target selection
 - Confirmation of gene-specific association studies
 - Hypothesis-free pathway analyses
- **Application of gws for efficacy/drug development**
 - Generation of drug-response profiles
 - Applications for companion diagnostic in drug development

Whole Genome Association Scan

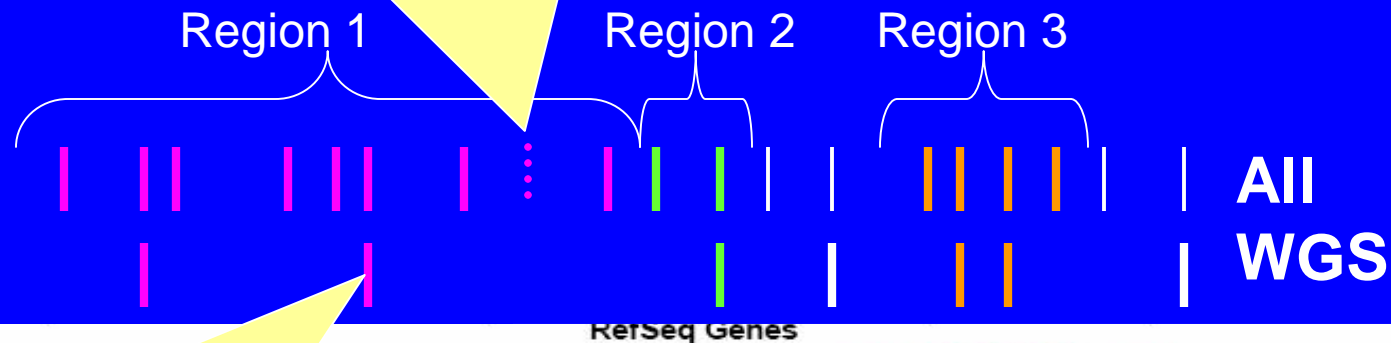


- If all 10 million SNPs in the genome were independent, we would need to examine every one of them.
- But because of the correlation (linkage disequilibrium) that exists between SNPs in close proximity to one another, only a small portion of SNPs are needed to provide information across the entire genome.
- Our current technology, using 500K SNPs, captures upwards of 80% of information across the genome

How do we use the WGS to detect disease association?

- The WGS allows us to not only examine genes directly but also to make use of regions of correlation (called Linkage Disequilibrium or LD) to find disease gene targets

Disease susceptibility SNP within gene – not genotyped

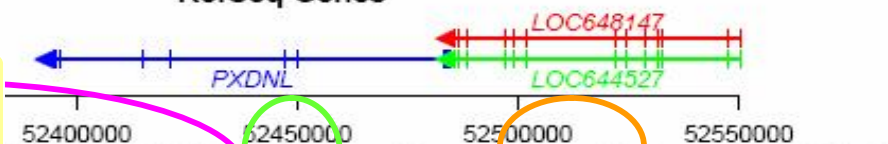


WGS SNP in Region 1, but not within the gene, can be used as a proxy for the disease SNP

Region of extended correlation

Region of extended correlation

Region of extended correlation



Chromosome Position (bp)

N SNPs = 56
Window = 278 kb
Vertical lines every 20 kb

LD matrix of r^2 values, white = 0, black = 1
Point types indicate LD clusters at $r^2 = 0.7$

Disease A Collection

- Genetic association to Disease A was investigated using the Affymetrix 500,000 SNP panel
- Subjects from 9 study sites - Caucasian
 - ~800 Disease A Cases
 - Ascertained by accepted clinical criteria
 - Positive diagnosis using prospective testing
 - Follow-up clinical data available confirming diagnosis
 - ~800 Disease A Controls
 - Ethnically matched, and when feasible gender- and age- matched (or older) to the cases
 - No history of usual symptoms
 - No impairment on clinical testing

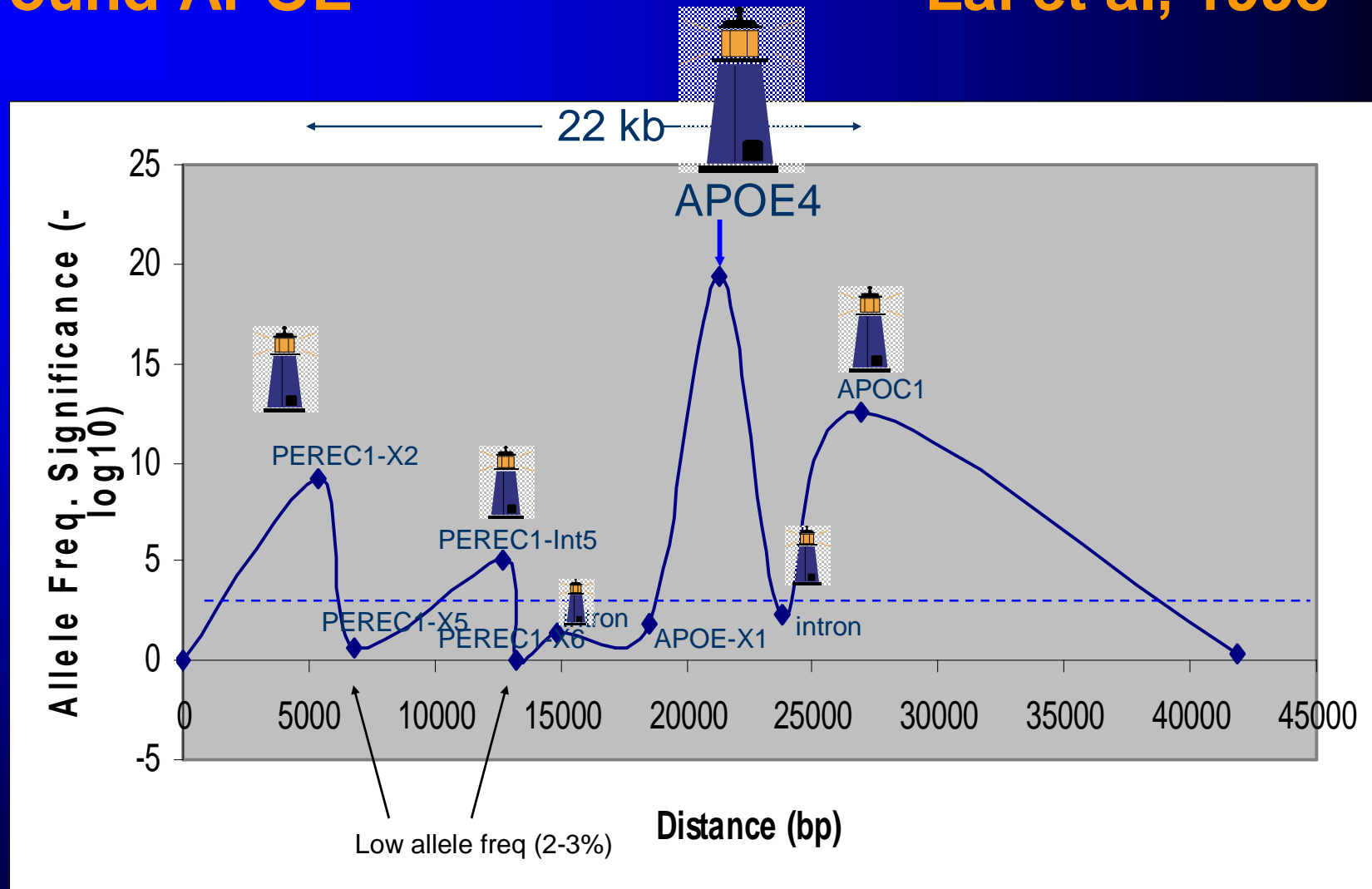


A published example – APOE within LD group

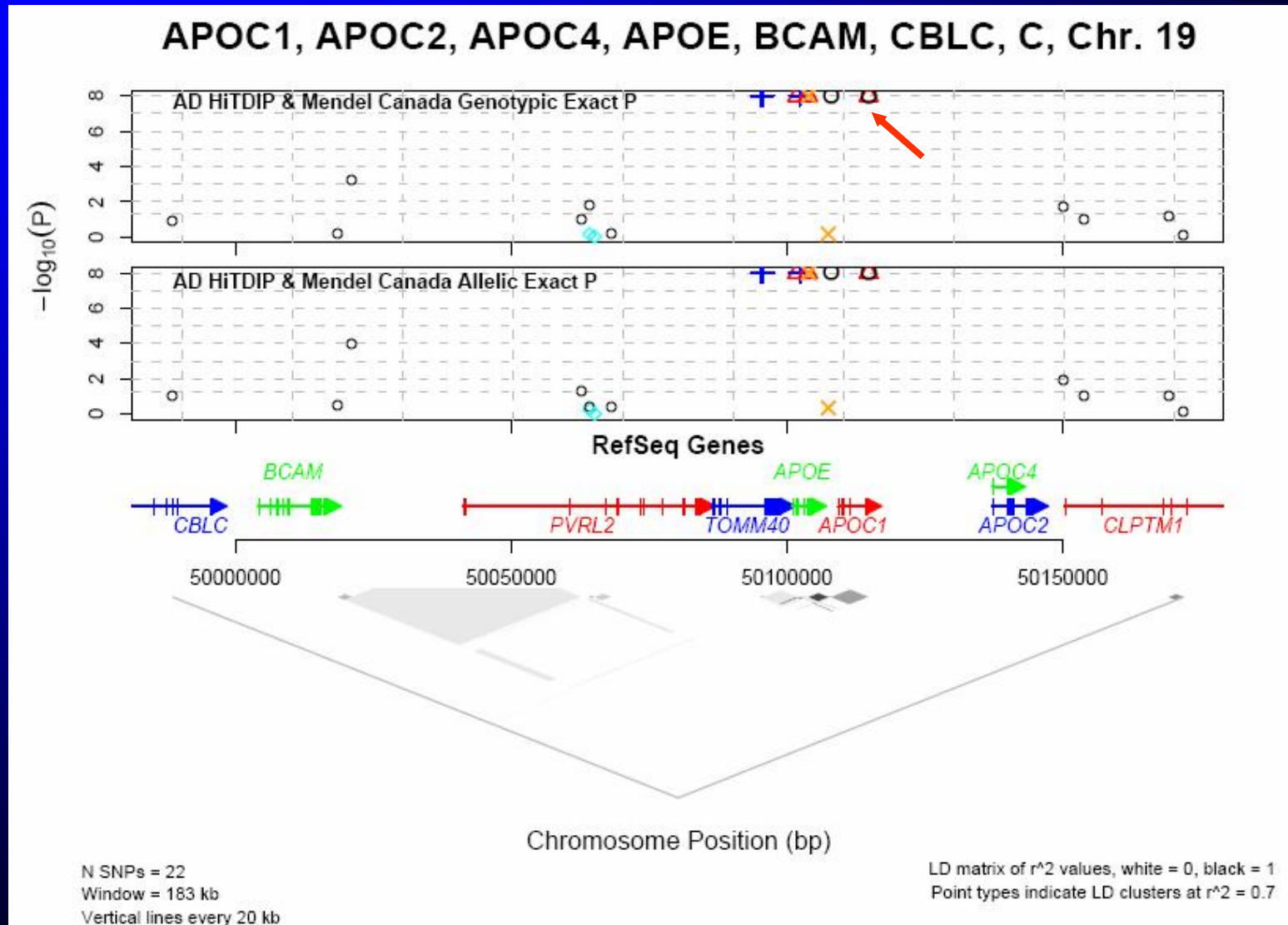
- The gws APOE HiTDIP association scans tested several SNPs in each of the three genes in the LD group. The p-values were all highly significant ($< E-8$)
- Other reported “associated” genes not confirmed
- New LD regions suggest association of other unreported genes

AD Association 1998: A high density SNP map around APOE

Lai et al, 1998

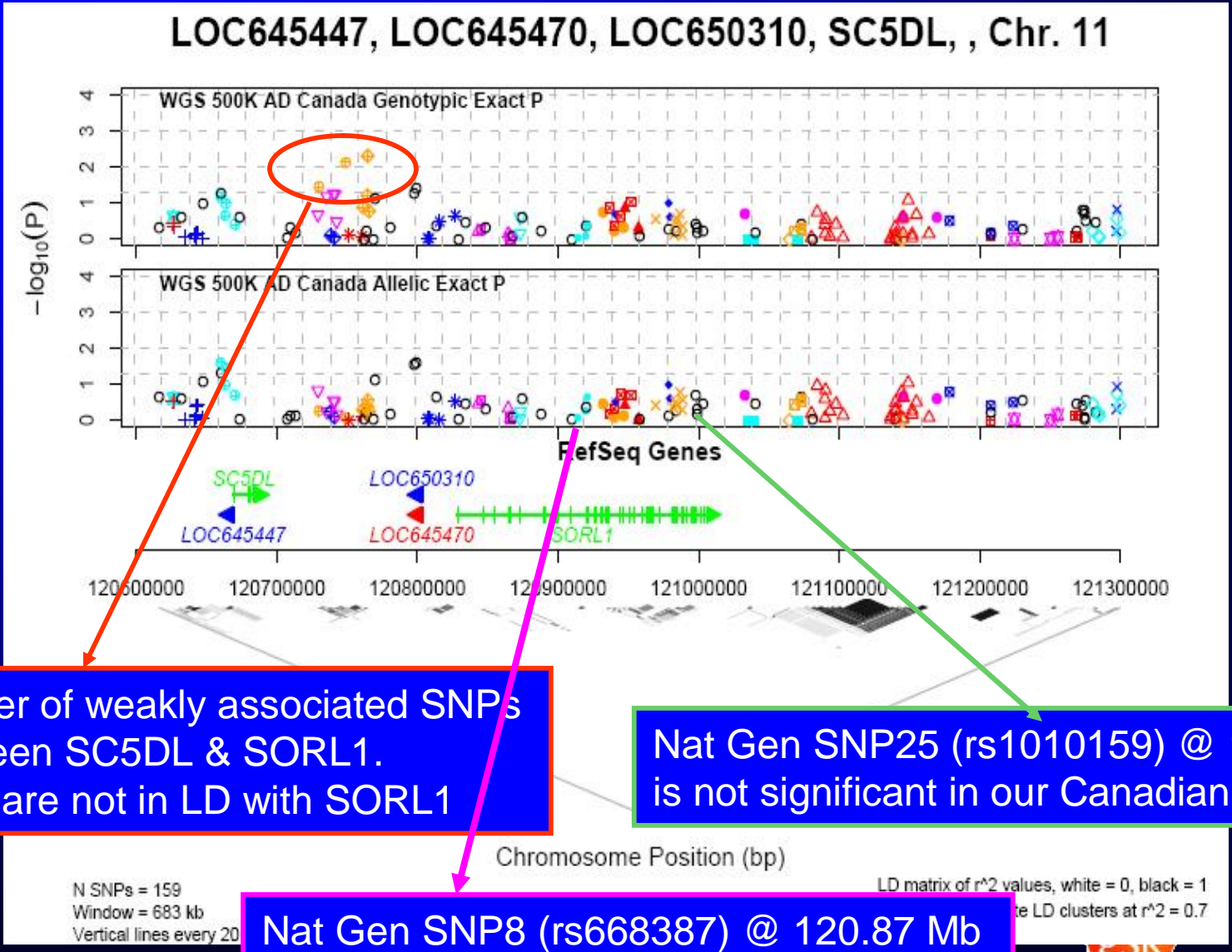


APOE in Alzheimer's disease in strong LD



Genome-wide scan in AD can be used to quickly assess newly reported association studies

SORL1 Region - Nature Genetics 1-07



Cluster of weakly associated SNPs between SC5DL & SORL1. They are not in LD with SORL1

Nat Gen SNP25 (rs1010159) @ 120.99 Mb is not significant in our Canadian AD data

Nat Gen SNP8 (rs668387) @ 120.87 Mb is not significant in our Canadian AD data

Conclusions from disease-target wgs

- Regions of reported associations, usually requiring years to assess by heterogenous confirmations, can be immediately confirmed – or *not* confirmed
- Very few reported gene associations for any disease have at least one major confirmation, with more than 97% of the literature having none. Fast data analysis is available for candidate genes with disease-specific gws
- **The hypothesis-free converse is more exciting, regions of well defined association by gws, supported by linkage disequilibrium data, can provide genetic specificity to target selections and pathway analyses**
- **Other genes in other diseases identified by gws**
 - **Ex: Schizophrenia, submitted for pub.**



Pharmacogenetic analyses of drug development programs with gws

- Alzheimer's disease example – efficacy PGx with rosiglitazone treatment



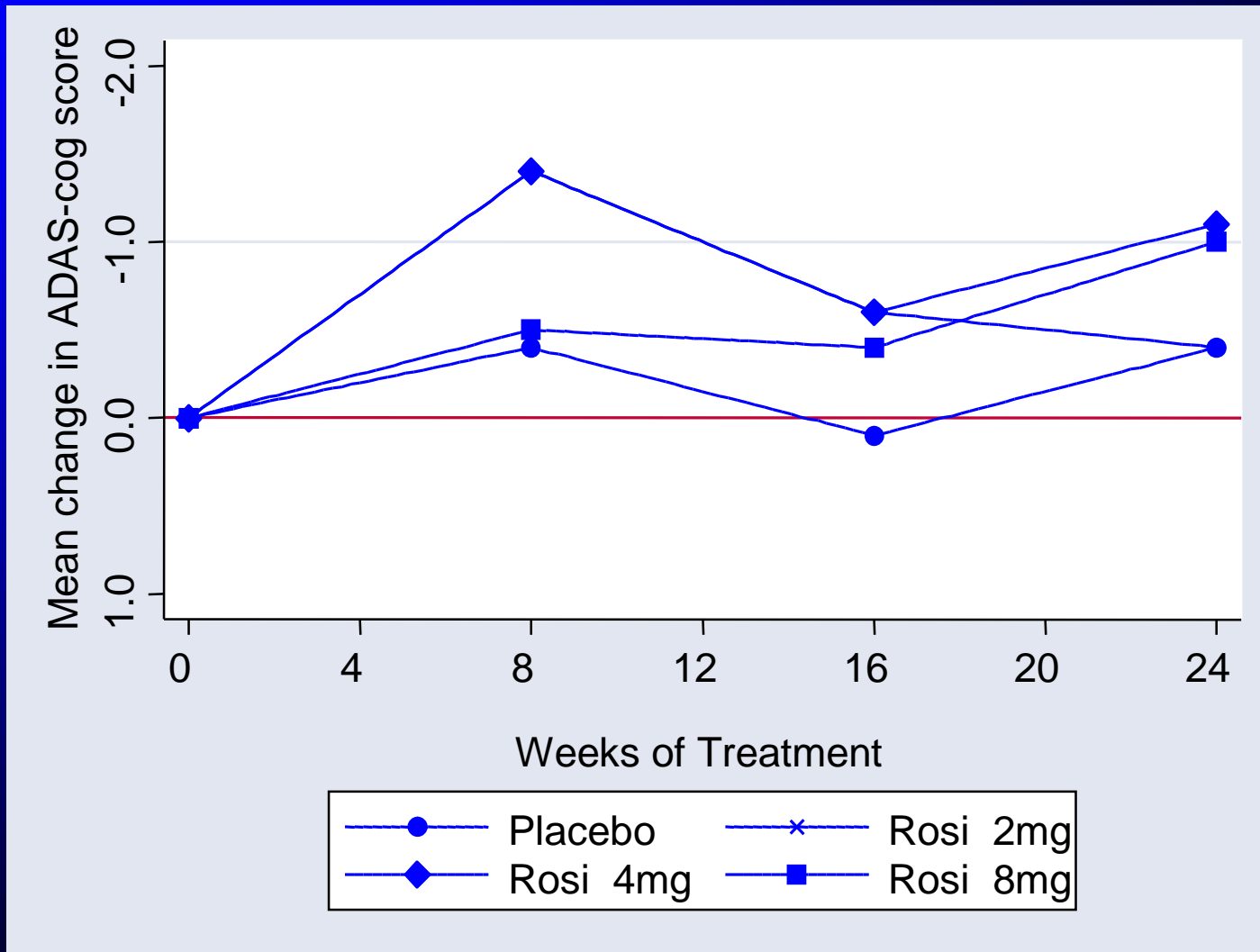
Based on validated use of **APOE** genotypes as a predictor of age of onset distribution of AD

More than 80 other candidate genes tested – including the usual popular favorites - **none** were associated with clinical response differentiation with rosiglitazone

PGX efficacy for AD with Rosiglitazone, a new drug directed against a mitochondrial energy pathogenesis

- Patients selected for inclusion in clinical trial with mild to moderate AD, not based on any genotyping
- Patients genotyped during the trial and, after analysis without these data, examined with respect to carriage of e4 allele [and other reported gene associations]
- **There was no positive clinical effect of treatment in ITT population**
- **In PGX analyses, e4- negative patients improved while e4+ carriers did not improve, but followed usual course of deterioration**
- Design of Phase III studies are powered using APOE genotype status

Model-adjusted Mean Change from Baseline in ADAS-cog: Intent To Treat population, Observed Cases

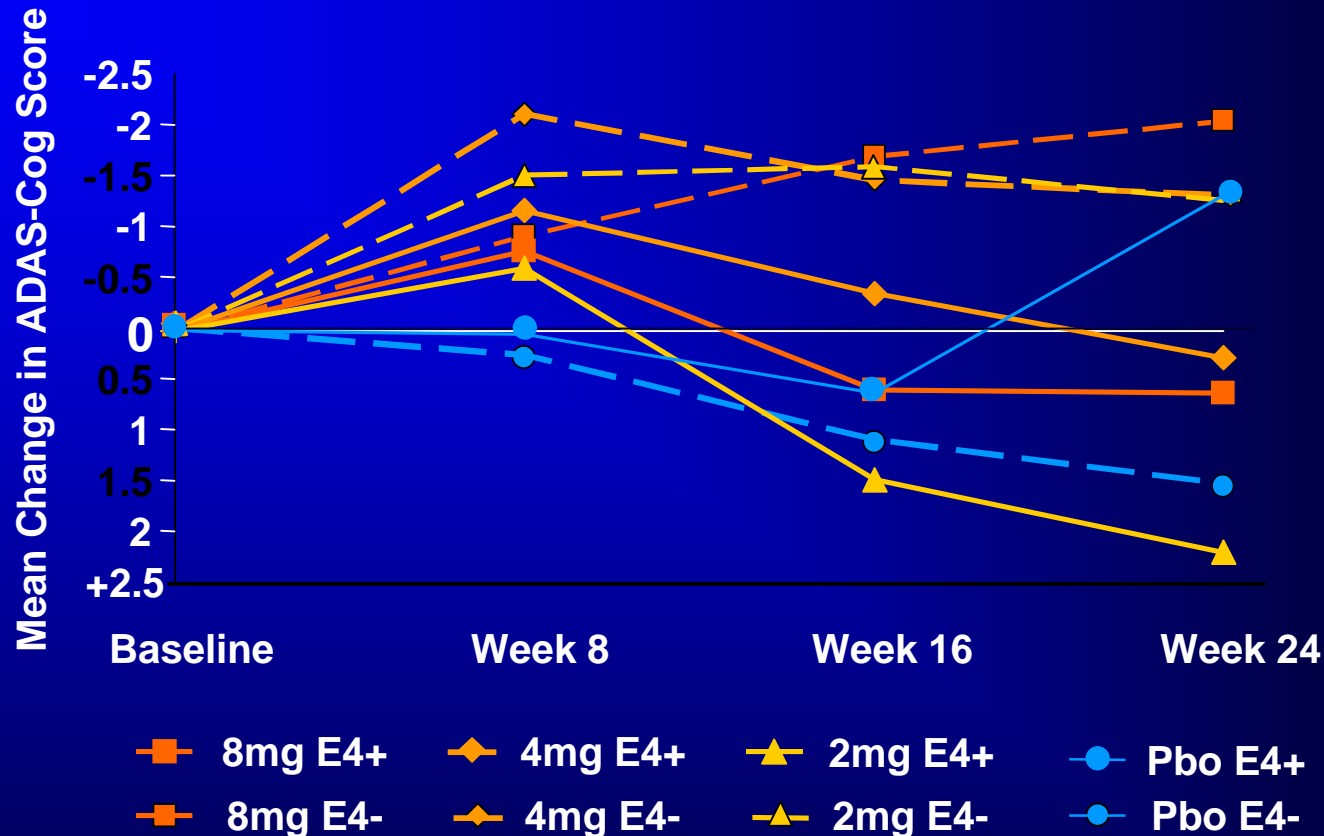


Pharmacogenetics makes the difference

- Failed study in ITT population
- Program would have been stopped
- Text book definition of **drug attrition**

- **However there was a prospective genetic hypothesis** for APOE susceptibility applied to the data

ADAS-Cog Score in APOE ε4-Negative, then APOE ε4-Positive Cohorts

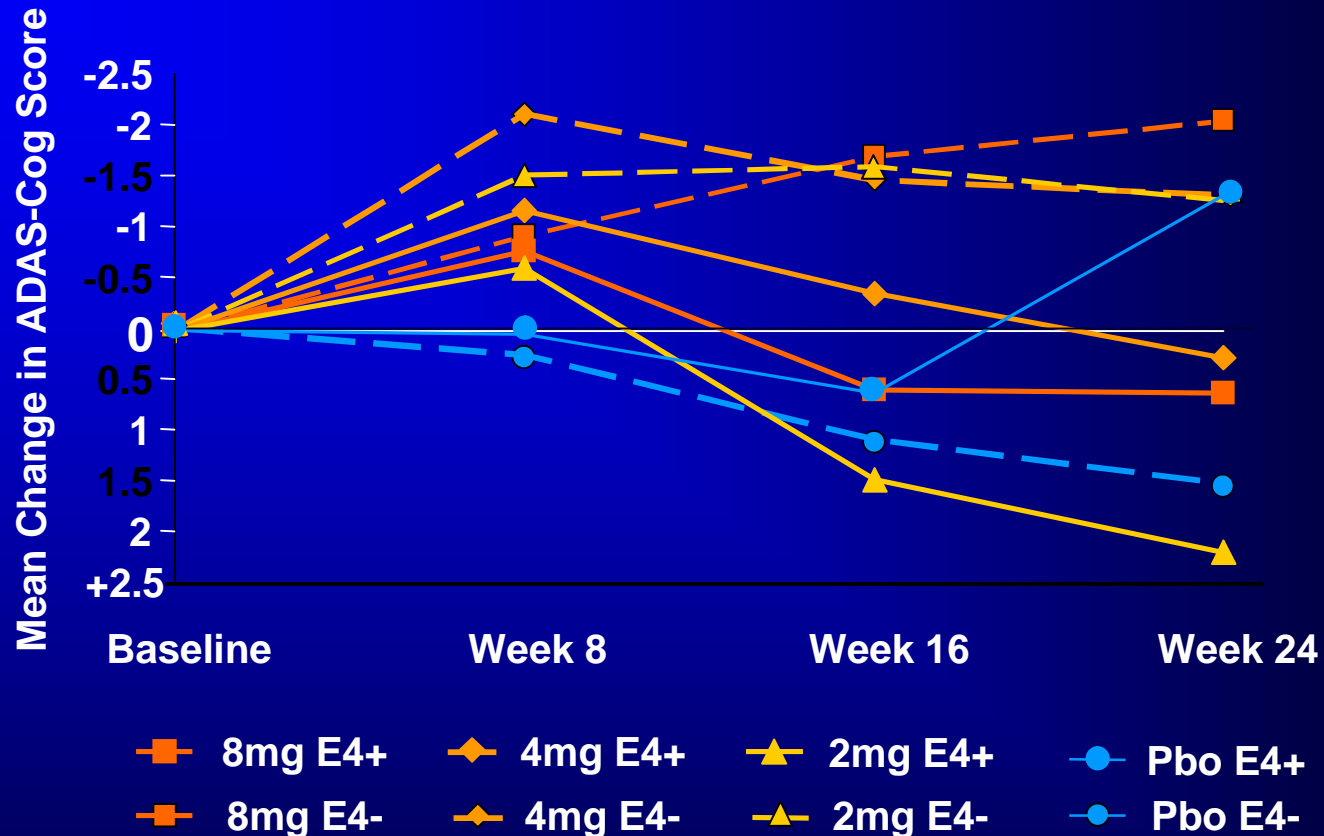


- Analysis of interaction between APOE carriage status and ADAS Cog change from baseline to Week 24 was significant ($P < 0.014$).
- Exploratory Testing Week 24 (p values not adjusted for multiplicity).
 - APOE e4-negative cohort RSG XR 8mg vs. placebo ($P < 0.024$)

Adapted from *The Pharmacogenomics Journal* 2006, online publication, 31 January 2006. doi:10.1038/sj.tpj.6500369.



ADAS-Cog Score in APOE ϵ 4-Negative and APOE ϵ 4-Positive Cohorts



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New Phase III trial designs

Phase IIB results:

“AD patients without an APOE4 allele responded better to rosiglitazone than patients who carry either 1 or 2 APOE4 alleles”

Genotype-specific Phase III trial designs (3 registration studies in progress):

AD patients have been screened at entry, and the placebo and dose response studies will be analyzed in groups of sufficient size to generate APOE genotype-specific response data [3-3; 3-4; 4-4; 2-3]

A framework for efficacy PGx using sequential, standardized genome wide analysis

- Sequential analyses in each separate trial can determine potential complex haplotypes to predict efficacy, or the need for different dosage forms.

Markers tested in every patient in each clinical trial	Hypotheses-derived from previous study	Phase of Clinical Development Program	# of patients in clinical trial <i>Approximation depending on type of clinical trial</i>	Expected # of association hits <i>Approximation based on current data in completed experiments</i>
~500,000	Free	Phase IIA Proof of Efficacy	~100	~ thousands
~500,000	~ thousands	Phase IIB	~500	~ hundred
~500,000	~ hundred	Phase III Registration1	~1,000s	~ tens
~500,000	~ tens	Phase III Registration2	~1,000s	< 20-30
~500,000	< 20-30	Phase III Other	~1,000s	< 10



Conclusions regarding standardized, sequential gws testing

- All patients, regardless of Phase of trial, receive exactly the same genome-wide testing
- Specific regions of the scans can be followed through the drug development program with a single testing platform *in silico*
- **Drugs can be saved from attrition**
- Ambiguous clinical responses can be associated with relatively few genetic markers by sequential gws
- Genetic markers can be evaluated for pathways and blood protein biomarkers