



**VANDERBILT KENNEDY CENTER
FOR RESEARCH ON HUMAN DEVELOPMENT**

Environmental Neurogenomics and Autism

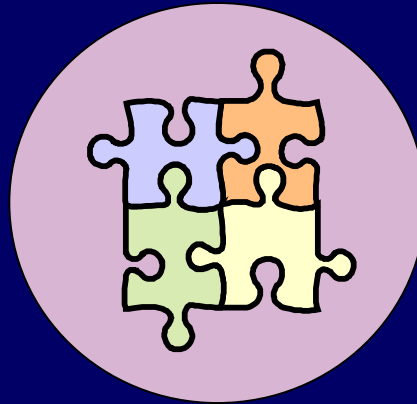
**Pat Levitt, Ph.D.
Director, Vanderbilt Kennedy Center**



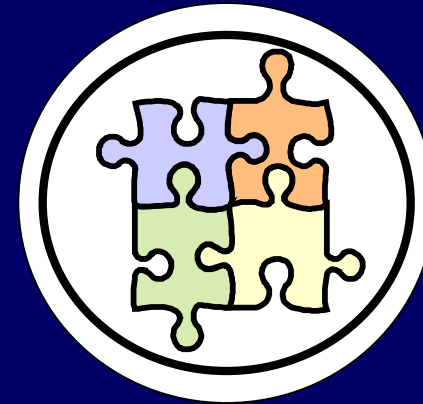
Complex Genetic Disorders Are Complex



Non-risk alleles



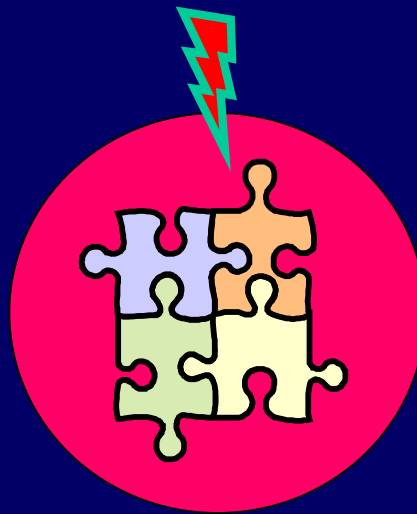
Combination of risk alleles –
INTERMEDIATE PHENOTYPE



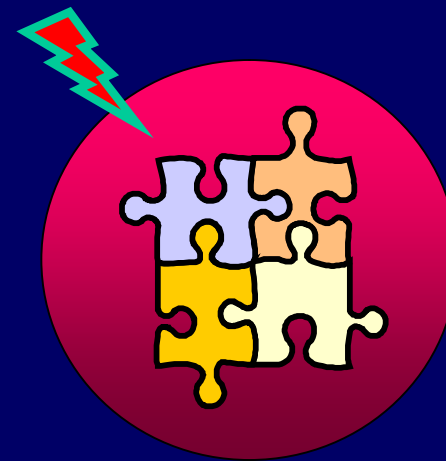
Incomplete penetrance with
combination of risk alleles



Non-risk alleles & environmental
factors



Combination of risk alleles &
environmental factors - AUTISM



Subset of risk alleles &
environmental factors - SPECTRUM

Keep in mind.....

**Genes are involved in the assembly of
specialized cells to perform specific functions**

Keep in mind.....

**Thus, there are no 'social behavior' or
'communication' genes.....**

Keep in mind.....

Gene mutations cause directly or increase the risk for the altered assembly of cells, tissues and systems that underlie specific functions.....

What We Know –

Highest Heritability Of Any DSM-IV Disorder (.6-.9)

- **Linkage Studies**

- **Look for excess sharing of *genomic regions on chromosomes* that track with the disease**

- **Allelic Association Studies**

- **Look for excess sharing of *alleles (SNPs; microsatellites)* at a *single locus***

- **Copy Number Variation (CNV)**

- **Look for *sub-microscopic alterations in chromosome structure* - deletions & duplications (sporadic)**

What We Know – Previous/current caveats



- **Ascertainment bias**
- **Sample size – studies uncorrected for Type 1 error**
- **Accuracy/completeness of phenotypes**
- **Technical QC**
- **Lack of assigned gene/variant function**

What We Know - Linkage

- Recent Autism Genome Project Consortium (>1100 families) -
11p; additional data filtering includes **2q, 7q**

Nature Genetics 39:319 (2007)

- Many other reported linkage regions - Chr 1q, 2p, 3q, 6q, 17q

What We Know - Submicroscopic Copy Number Variations (CNVs)

- AGP - ~10% have shared, 'detrimental' CNVs (microdeletions or duplications)
- CSHL - ~10% have increased CNVs compared to controls

What We Know - Rare Mutations

- **Loss-of-function –**

Neurexins1/3, Neuroligins4/5, pTEN, SERT, CNTNAP2, SHANK3, Ca_v⁺⁺, GABARg1

What We Know - Genetic Syndromes with Co-Occurring ASD Diagnosis

- **Fragile X (FRM1)**
- **Rett Syndrome (MeCP2)**
- **Angelman Syndrome (Ube3a)**
- **Tuberous Sclerosis (TSC1,2)**
- **Timothy Syndrome (Ca_v1.2)**
- **Smith-Lemli Opitz (Dhcr7)**

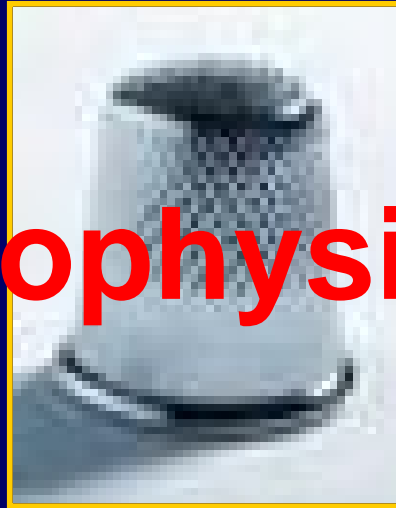
What We Know – Common Risk Alleles - Association

- Risk Alleles (non-functional) – *Neurexin1,3, b₃-Integrin, GABRb₃, Slit1, En2*
- Risk Alleles (functional) – *reelin, MET, b2-AdrR*

Genetic Contributions to ASD

- How much of the risk is due to direct impact of mutations on brain development?
- How much of the risk is due to direct impact of mutations on peripheral functions that influence brain development?
- How much of the risk is due to genetically established sensitivities to environmental perturbations?
- How much of the phenotypic heterogeneity of individuals with ASD are influenced combinatorially through genetic and/or environmental factors?

Opportunities - Making the Link



Pathophysiology

Making the Gene-Environment Translation – Experimental Links



Nature ~~versus~~ Nurture

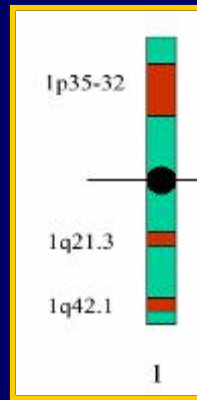
**Assuming connectivity is a key issue.....
.....What ASD isn't**

Genetic versus Environmental

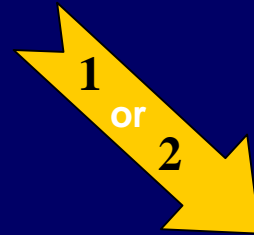
What ASD Is.....

Genetic and Environmental

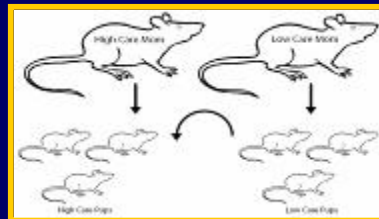
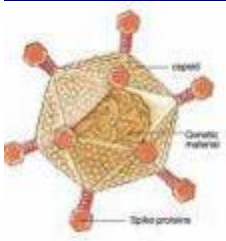
What is Needed - Translational Approaches & Technical Strategies



Precise Characterization of Mutations/Variants of 'Risk' Genes



Early Environmental Influences (Infections/Toxins/Stressors)



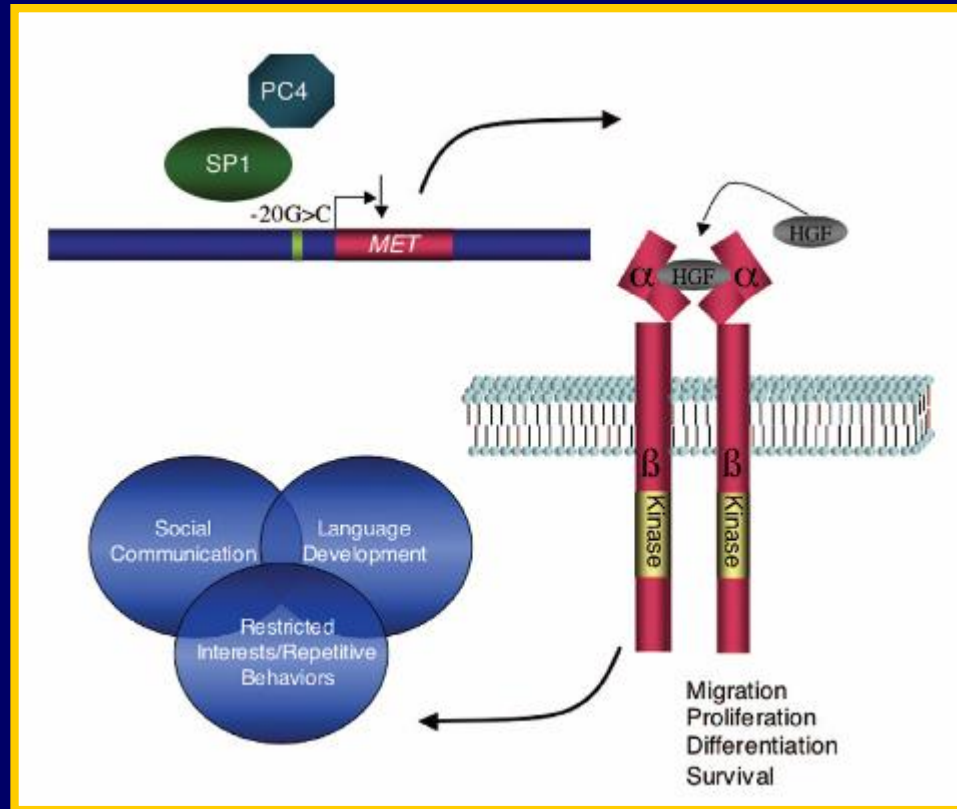
Defects in gene expression & early life stressors - linking to neurodevelopmental time courses?

A Model Strategy

MET and ASD



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- 5' SNP association - $p = 5 \times 10^{-6}$ (Bonferroni corrected)
- Relative Risk ~2.25 fold
- Common polymorphism – reduces transcription 2.5-fold

Genetic Vulnerability & Environmental Risk – What Can Be Done

Exp Toxic Pathol 2002, 53: 161-168
URBAN & FISCHER
<http://www.urbanfischer.de/journals/exptoxpat>

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Transplacental effects of 2,3,7,8-tetrachlorodibenzo-p-dioxin on the temporal modulation of Sp1 DNA binding in the developing cerebral cortex and cerebellum

TULU L. NAYYAR¹, NASSER H. ZAWIA², and DARREYL B. FLOOD¹

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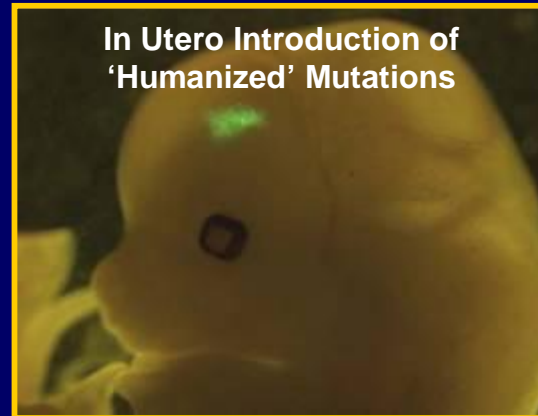
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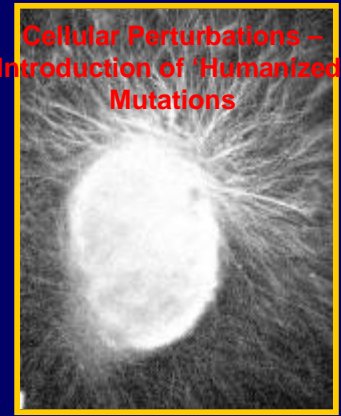
**'MET Humanized' mice -
Candidate Environmental Factor
Characterizations**



**In Utero Introduction of
'Humanized' Mutations**

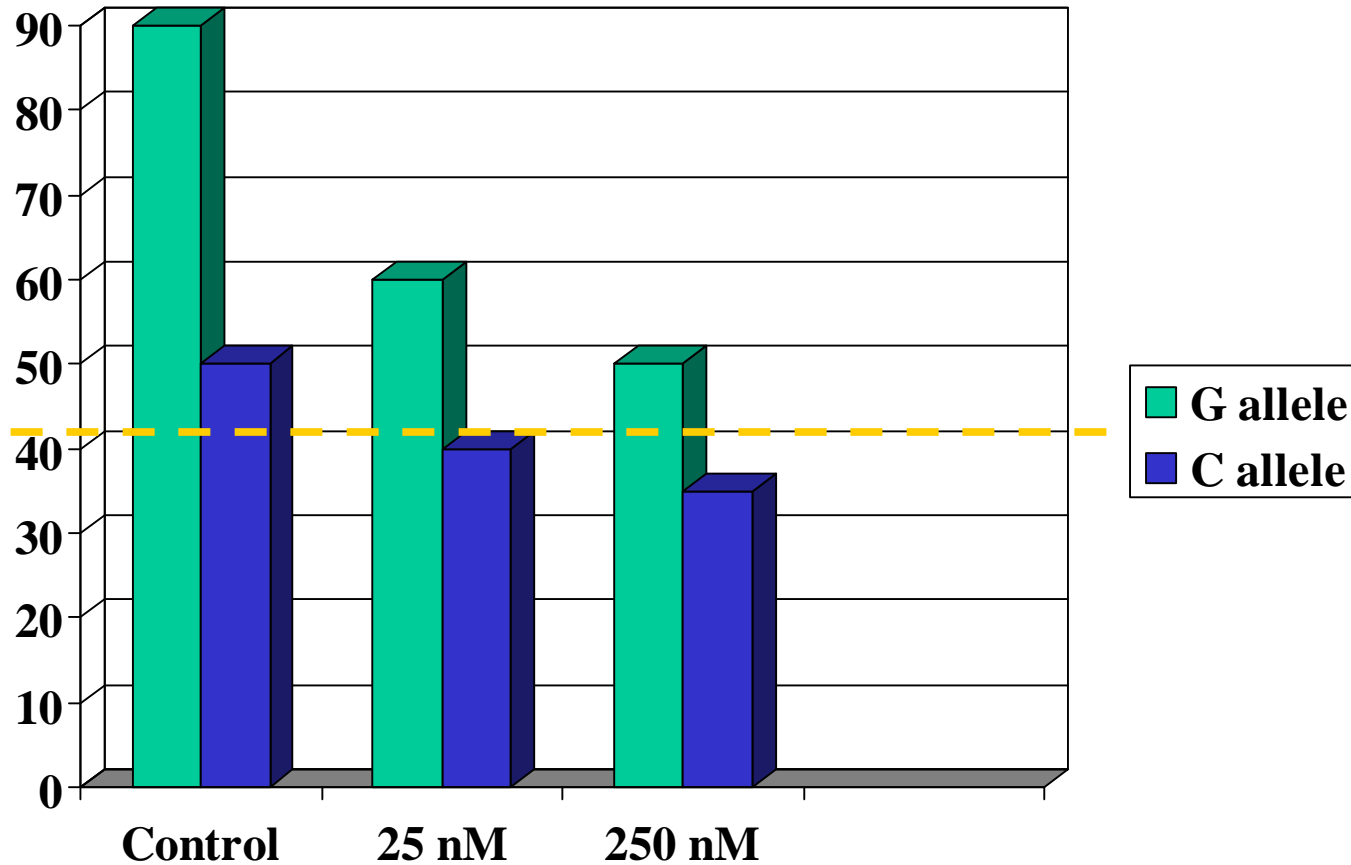


**Cellular Perturbations -
Introduction of 'Humanized'
Mutations**



Genetic Vulnerability & Environmental Risk – What Can Be Done

Benzo-a-pyrene Impact on *MET* Gene Transcription



Hypothetical
Disorder
Threshold

[BaP]

Campbell, Hood and Levitt, unpublished

What We Need

- **Increased ascertainment to improve stratification studies**
- **Deep sequencing to identify more functional variants**
- **Functional characterization of variants**
- **Wise investments in model systems – make them biologically and pathophysiologically relevant**
- **Primate expression patterns (timing and location) of genes**