The Nitty Gritty of Autism and Gastrointestinal Disorders

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“The eleven children (eight boys and three girls) whose histories have been briefly presented offer, as is to be expected, individual differences in the degree of their disturbance, the manifestation of specific features, the family constellation, and the step-by-step development in the course of years.” ----- Kanner, Nerv. Child 2:217-250 (1943)
ASD Heterogeneity Is Not Well Characterized

<table>
<thead>
<tr>
<th>Domain</th>
<th>ASD</th>
</tr>
</thead>
<tbody>
<tr>
<td>social communication</td>
<td></td>
</tr>
<tr>
<td>language</td>
<td></td>
</tr>
<tr>
<td>repetitive, restrictive behaviors</td>
<td></td>
</tr>
<tr>
<td>sensory abnormalities</td>
<td>94%</td>
</tr>
<tr>
<td>developmental regression(b)</td>
<td>15%–40%</td>
</tr>
<tr>
<td>motor signs(c)</td>
<td>60%–80%</td>
</tr>
<tr>
<td>gross motor delay</td>
<td>5%–10%</td>
</tr>
<tr>
<td>sleep disturbance</td>
<td>50%</td>
</tr>
<tr>
<td>gastrointestinal disturbance(d)</td>
<td>4%–50%</td>
</tr>
<tr>
<td>epilepsy(e)</td>
<td>6%–60%</td>
</tr>
<tr>
<td>comorbid psychiatric diagnosis(f)</td>
<td>25–70%</td>
</tr>
</tbody>
</table>

adapted from Geschwind, Ann Rev Med  2009
For Example............................ASD Risk is Highly Heritable - But the Genetic Architecture is Heterogeneous
Biological Factors Contribute to the Expression of the Autisms and Other Co-Occurring Traits

Social Behavior

Communication

Restricted interests
Repetitive behaviors

?
Medical Co-Occurring Phenotypes

- Reflection of biological overlap in factors that impact brain and peripheral organ development
- Influence the functional status of children with ASD, which in turn will impact amenability to treatments and quality of life
Medical Co-Occurring Phenotypes

• Reflection of biological overlap in factors that impact brain and peripheral organ development

• Influence status of children with ASD, which in turn will impact amenability to treatments
Many Non-Biological Factors Invoked

• Diet
• Medications
• Parent over-reporting
• Behavioral issues
• Mercury/MMR/Toxins/Metals/Traffic/TV/Tylenol/Rain
Gut-Brain Interactions Influence Each Other

Cryan and Dinan, Nat Rev Neurosci 13 (2012)

Healthy status
- Normal behaviour, cognition, emotion, nociception
- Healthy levels of inflammatory cells and/or mediators
- Normal gut microbiota

Stress/disease
- Alterations in behaviour, cognition, emotion, nociception
- Altered levels of inflammatory cells and/or mediators
- Intestinal dysbiosis

Healthy CNS function

Abnormal CNS function

Healthy gut function

Abnormal gut function
Gut Microbiome in ASD

A) Autistic:
- Bacteroidetes: 51.548%
- Firmicutes: 37.254%
- Verrucomicrobia: 2.902%
- Proteobacteria: 0.601%
- Actinobacteria: 0.527%
- Other: 7.159%

B) Control:
- Bacteroidetes: 63.631%
- Firmicutes: 30.226%
- Verrucomicrobia: 1.812%
- Proteobacteria: 0.090%
- Actinobacteria: 0.468%
- Other: 3.773%

C) Sibling Control:
- Bacteroidetes: 44.326%
- Firmicutes: 44.012%
- Verrucomicrobia: 2.327%
- Proteobacteria: 1.037%
- Actinobacteria: 0.157%
- Other: 8.141%

Finegold et al Anaerobe 16 (2010)
• Exciting data from small samples are exciting data from small samples

• Co-occurring conditions don’t cause autism
Brain Architecture Differences Emerge Early in Development

Wolf et al. and the International Brain Imaging Science Network

American J. Psychiatry 169 (2012)
Brain Architecture Differences Emerge Early in Development

Wolf et al and the International Brain Imaging Science Network

American J. Psychiatry 169 (2012)
Medical Co-Occurring Phenotypes

- Reflection of biological overlap in factors that impact brain and peripheral organ development
- Influence the functional status of children with ASD, which in turn will impact amenability to treatments and quality of life
Biological overlap in factors (gene) that impact brain and peripheral organ development
MET Is a ‘Strong Candidate’ ASD Risk Gene

With the rapid increase in the number of genes potentially linked to ASD, we felt it would be useful to evaluate the strength of the evidence linking a given gene to the disorder. An expert panel of Advisors participated in the scoring process.

also AutsimKB: Xu et al, Nucleic Acids Research 2012
Pleiotropy – the control by a single gene of several distinct and seemingly unrelated traits
Two Children in Same Family with ASD – One with GI Condition is **Genetically Different**

![Graph showing MET 'C' Allele Frequency with categories: ASD+GI, ASD not GI, Parents, Non-ASD Sibs. The graph indicates a genetic difference between children with ASD and GI condition compared to those without.](image)
MET Expression in Injured Gut: Model Mouse

Normal Met Expression in Gut

Met Expression in Gut: 8 days of Colitis

Met Expression in Gut: 4 days of Colitis; 4 days repair
Gut Injury in the **Absence** of *Met* is More Severe
Evaluation, Diagnosis, and Treatment of Gastrointestinal Disorders in Individuals With ASDs: A Consensus Report

**Areas in Need of New Knowledge**

- Determine prevalence of gastrointestinal disorders in individuals with ASDs
- Develop screen for gastrointestinal disorders in individuals with ASDs that can be used by primary care and other providers
- Identify behaviors associated with gastrointestinal pain/distress in persons with ASDs
- Evaluate whether dietary restriction is efficacious for individuals with ASDs
- Identify role of abnormal gastrointestinal permeability in neuropsychiatric manifestations of ASDs
- Determine relationship of immune dysfunction to clinical symptoms that present in patients with ASDs
- Determine if alteration in gut microflora is associated with either gastrointestinal or neurobehavioral symptoms in patients with ASDs
- Clarify underlying pathophysiology and clinical aspects of ASDs
- Characterize genotype of individuals with ASDs and gastrointestinal disorders
- Identify genetic mutations that may be possible underlying causes of ASDs

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Richly Characterize GI Symptoms in Children with ASD Prospectively

75x2
+ ASD + GID → ADOS

75x2
+ ASD - GID → ADOS

75x2
- ASD + GID → ADOS

lots
- ASD - GID → SRS ~ QPQS
Published Findings from Nashville Site

RESEARCH ARTICLE   Autism Research 5 (2012)

Gastrointestinal Dysfunction in Autism: Parental Report, Clinical Evaluation, and Associated Factors

Phillip Gorrindo, Kent C. Williams, Evon B. Lee, Lynn S. Walker, Susan G. McGrew, and Pat Levitt
Table I. Characteristics of Study Participants

<table>
<thead>
<tr>
<th></th>
<th>ASD-GID (n = 40)</th>
<th>ASD-only (n = 45)</th>
<th>GID-only (n = 36)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age in years, mean (SD)</td>
<td>10.8 (3.7)</td>
<td>12.4 (3.4)</td>
<td>11.0 (3.4)</td>
</tr>
<tr>
<td>Male sex, % (n)</td>
<td>72.5 (29)</td>
<td>86.7 (39)</td>
<td>63.9 (23)</td>
</tr>
<tr>
<td>Ethnicity and race, % (n)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hispanic</td>
<td>12.5 (5)</td>
<td>0 (0)</td>
<td>2.8 (1)</td>
</tr>
<tr>
<td>Non-Hispanic white</td>
<td>77.5 (31)</td>
<td>86.7 (39)</td>
<td>88.9 (32)</td>
</tr>
<tr>
<td>Non-Hispanic black</td>
<td>7.5 (3)</td>
<td>8.9 (4)</td>
<td>8.3 (3)</td>
</tr>
<tr>
<td>Non-Hispanic other</td>
<td>2.5 (1)</td>
<td>4.4 (2)</td>
<td>0 (0)</td>
</tr>
<tr>
<td>ADOS Classification, % (n)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Autism</td>
<td>95.0 (38)</td>
<td>91.1 (41)</td>
<td>n/a</td>
</tr>
<tr>
<td>Autism spectrum</td>
<td>5.0 (2)</td>
<td>8.9 (4)</td>
<td>n/a</td>
</tr>
<tr>
<td>Nonverbal, % (n)a</td>
<td>30.0 (12)</td>
<td>6.7 (3)</td>
<td>0 (0)</td>
</tr>
<tr>
<td>BMI-for-age percentile, mean (SD)b</td>
<td>76.2 (30.2)</td>
<td>68.9 (31.0)</td>
<td>57.2 (36.3)</td>
</tr>
</tbody>
</table>

p = .03
High Agreement Between Parental Report and Physician’s Diagnosis for Any GID in ASD

<table>
<thead>
<tr>
<th></th>
<th>ASD-GID (n = 38)</th>
<th>GID-only (n = 36)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Parent-clinician agreement for diagnosis of any GID, %</td>
<td>92.1</td>
<td>88.9</td>
</tr>
</tbody>
</table>

These data do not support the claim that parents over-report presence of GID in ASD

Gastrointestinal Dysfunction in Children With & Without ASD, by Physician’s Evaluation and Parent’s Report


### Prevalence of Diagnosis (%)

<table>
<thead>
<tr>
<th>Condition</th>
<th>ASD-GID (n = 40(^A))</th>
<th>GiD-only (n = 36)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Constipation</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Irritable Bowel Syndrome</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Aerophagia</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Non-retentive Fecal Incontinence</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Adolescent Rumination Syndrome</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Functional Dyspepsia</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Functional Abdominal Pain &amp; Syndrome</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cyclic Vomiting Syndrome</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Reflux(^B)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Non-functional disorder(^B)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

\(^A\) Parent data were missing for two individuals, thus n = 38 for ASD-GID parental report; both individuals were evaluated by a physician

\(^B\) Parent-report instrument does not collect data on reflux or non-functional disorders
Increased Social and Language Impairment in Children With Co-Occurring GID & ASD

Supplemental Table 1: Classification of Medications by Potential Gastrointestinal Side Effects

<table>
<thead>
<tr>
<th>Without potential GI side effects</th>
</tr>
</thead>
<tbody>
<tr>
<td>amitriptyline, atenolol, benztropine, chlorpromazine, clonazepam, clonidine, diazepam, doxazosin, eszopiclone, haloperidol, hydroxyzine, levothyroxine, lithium, lorazepam, melatonin, zonisamide</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>With potential GI side effects, except constipation</th>
</tr>
</thead>
<tbody>
<tr>
<td>albuterol, amphetamine-dextroamphetamine, atomoxetine, baclofen, desmopressin, dexamfetamine, escitalopram, fluoxetine, lamotrigine, levetiracetam, lisdexamfetamine, methylphenidate, oxcarbazepine, sertraline, somatropin, topiramate, trazodone, valproic acid, divalproex sodium, ziprasidone</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>With potential GI side effects, including constipation</th>
</tr>
</thead>
<tbody>
<tr>
<td>aripiprazole, citalopram, clomipramine, clozapine, fluvoxamine, guanfacine, olanzapine, oxybutynin, paroxetine, pregabalin, quetiapine, risperidone</td>
</tr>
</tbody>
</table>
No Association Between Medications and Diagnosis of Constipation in Children With ASD

<table>
<thead>
<tr>
<th>Medications with potential constipating side effects</th>
<th>Unadjusted Odds Ratio</th>
<th>95% CI</th>
<th>P Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Medications with potential constipating side effects</td>
<td>1.04</td>
<td>0.42 - 2.64</td>
<td>NS</td>
</tr>
</tbody>
</table>

No Association Between Dietary Habits and ASD or GID Status

A Unique Subgroup of Children?

More severe functional impairments
(social-communication)
The Next Step:

Tummy Troubles

- More severe language impairment
- More severe autism symptoms
- Involvement of brain and gut functional problems
- More pathophysiology?
Reported Markers of Oxidative Stress and Autism
(126 publications)

- Rare mitochondrial mutations
- Anti-oxidant enzymes
- Free radicals
- *Arachadonic acid metabolites*
- Lipid peroxidation
- Impaired methylation
- Transsulfuration metabolite elevation
- Homocysteine elevation
- Reduced folate carriers
- Reduced Biotin
- Glutathione metabolism disruption
- Inflammatory marker elevation
- Nitric oxide elevation
F₂-Isoprostanes = **Gold Standard Marker of Oxidative Stress**!

kidney disease, cardiovascular disease, stroke, cancers, cigarette smoking, asthma, type 2 diabetes, Alzheimer’s Disease
Biomarker Correlate of Subgroup Severity
Elevated Marker of Oxidative Stress in Autism-GID
A Unique Subgroup of Children?

Patient Stratification For Treatment May Become A Reality
Perform Hierarchical Cluster Analysis of Medical Phenotypes

AGRE Sample
(n=420 probands with complete information on 18 medical characteristics)
Cluster 2 (n=66)

Abnormal growth pattern
Respiratory problems
Seizures
Gastrointestinal problems
Allergies
Asthma
Skin abnormalities
Teeth abnormalities
Movement abnormalities
Vision problems
Gait abnormalities
Coordination abnormalities
Sensory abnormalities
Visual
Tactile
Sleep disorder symptoms
Pain
Acoustic
Stereotypies
FOR THE FUTURE............

With an enrichment of minimally verbal children, who may or may not share a common medical condition, new methodologies to analyze clinician-child interactions -

- language sample of utterances, functional words
- vocal cues as predictors of clinician-child engagement
- timing and coordination of non-verbal interactions
- stereotypies
- multi-modal analyses of improvements post-treatments
New Study –
Treat GI Conditions to Test the Hypothesis that ASD Symptoms and Behavior Improve
Thank You!