

# **Defining and Treating Autism: Considerations for Biomarkers in Research and Clinic**

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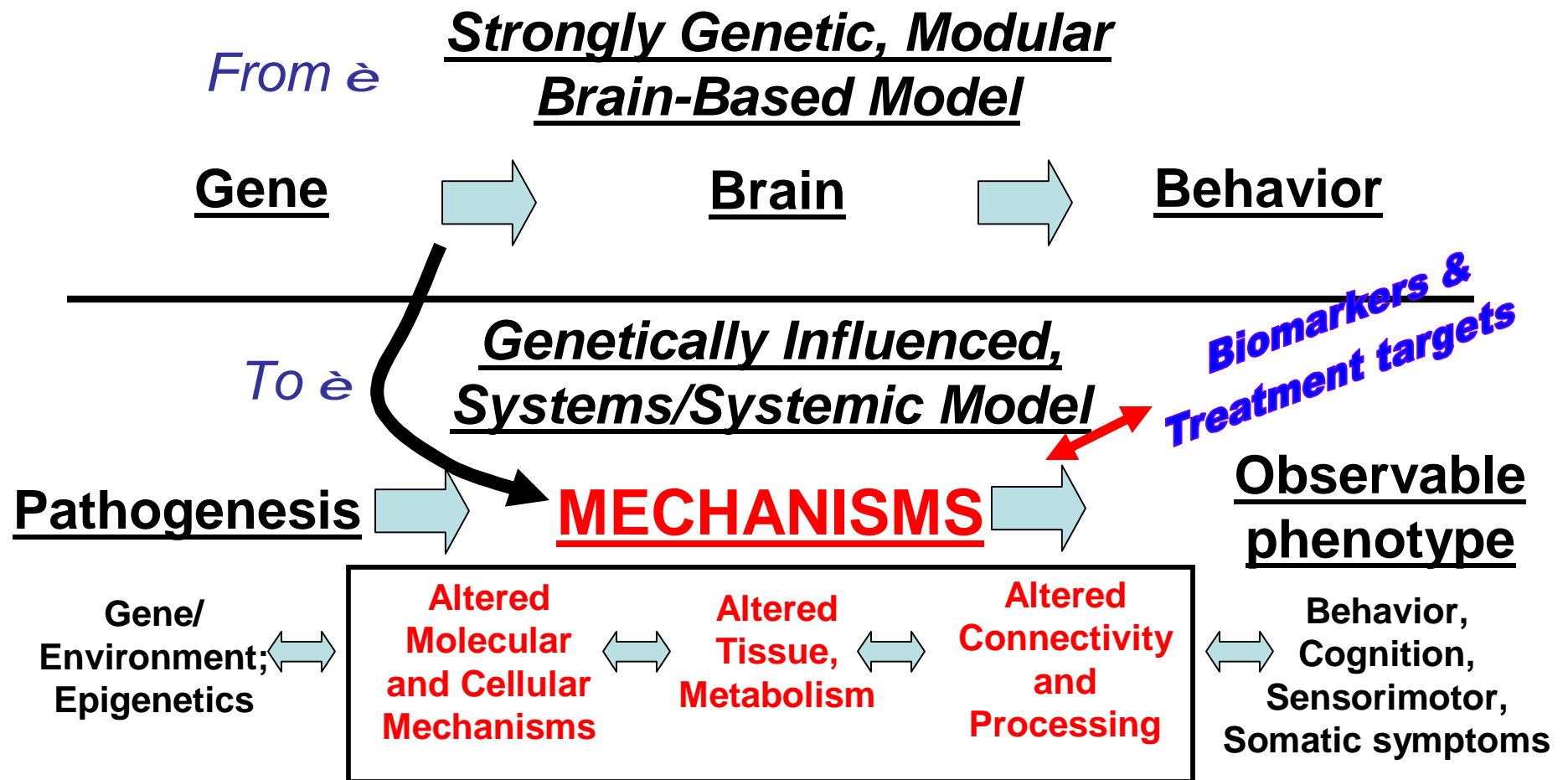
# Why biomarkers in autism?

- Diagnosis
- Identification of pathophysiological mechanisms
- Subgroup
- Identification of treatable features
- Predict/track treatment response

## **Overall NEEDS:**

- **Focus on BIOLOGY, PATHOPHYSIOLOGY**
  - **Coordinate measures across levels**

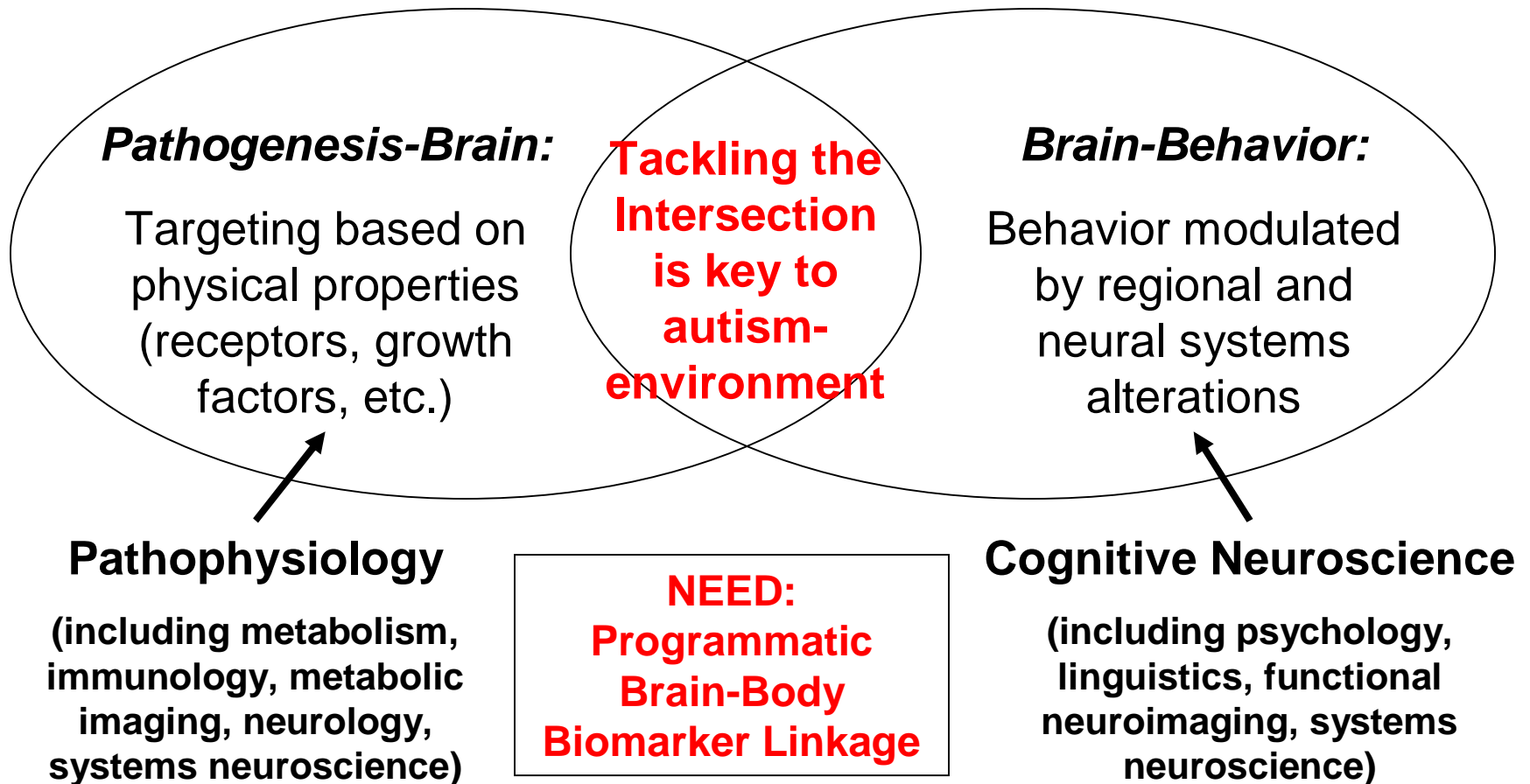
# Pathophysiological MECHANISMS: The “Middle-Out” Approach



# Multi-system from the start? Kanner 1943 on somatic symptoms

- Case 1:** “Eating has always been a problem .....” for him. He has never shown a normal appetite.”
- Case 2:** “...large and ragged tonsils.”
- Case 3:** diarrhea and fever following smallpox vaccination .... healthy except for large tonsils and adenoids.
- Case 4:** vomited a great deal during his first year... feeding formulas were changed frequently ... tonsils were removed...
- Case 5:** nursed very poorly ... quit taking any kind of nourishment at three months... tube-fed five times daily up to one year of age...At camp she slid into avitaminosis and malnutrition but offered almost no verbal complaints.”
- Case 7:** vomited all food from birth through the third month....
- Case 8:** feeding formula caused ...concern. ... colds, bronchitis, streptococcus infection, impetigo...
- Case 9:** none of the usual children’s diseases.” [? Overactive immune system?]
- Case 10:** frequent hospitalizations because the feeding problem ... repeated colds and otitis media
- Case 11:** was given anterior pituitary and thyroid preparations for 18 months

# Pathophysiology requires understanding the brain as an organ in the body that can get sick

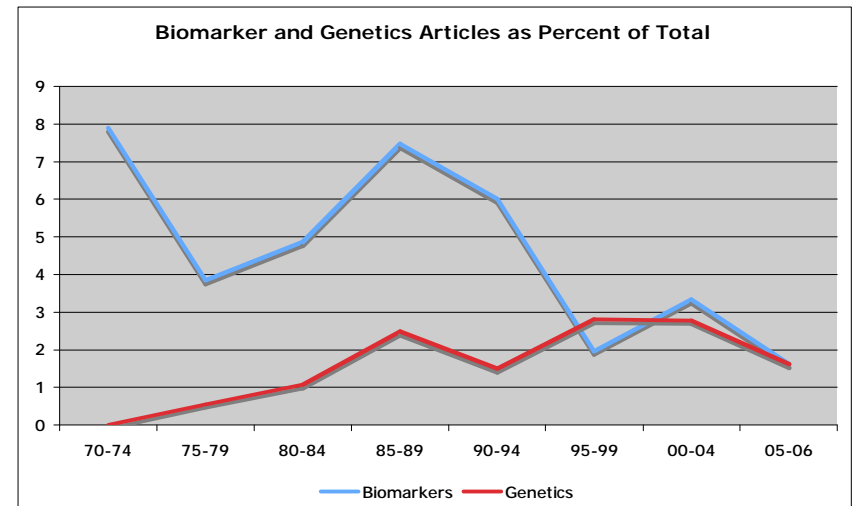


# Trends in Autism Biomarker Publications in *Journal of Autism and Developmental Disorders* (1970-2006)

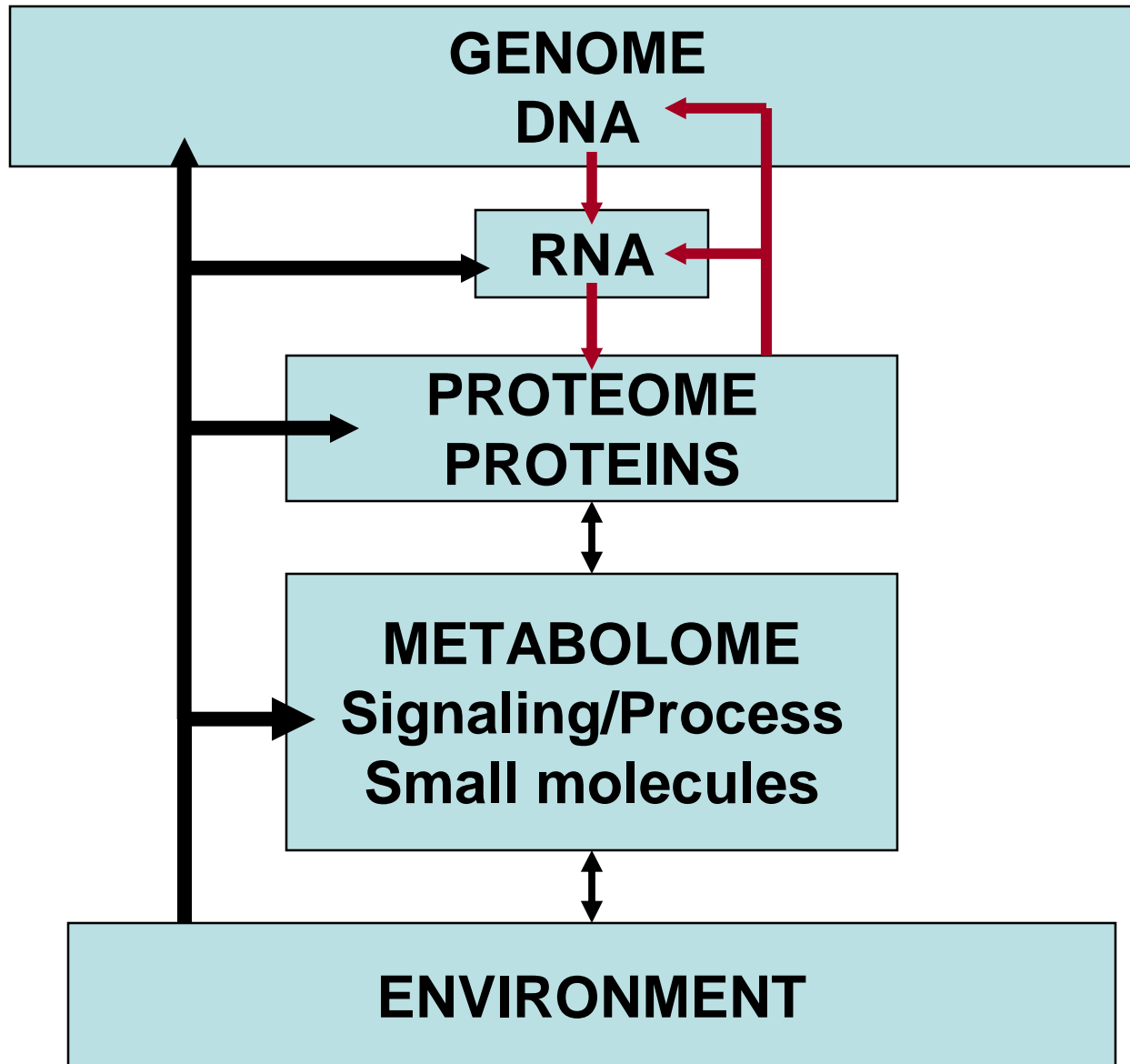
**In 36 years:  
78 articles out of 2004  
(average ~2/year)  
45 substances studied**

<b>Biomarkers Tested</b>	70-74	75-79	80-84	85-89	90-94	95-99	00-04	05-06
Autoantibodies	1							1
Free fatty acid response to insulin and glucose stimulation	1							
Hair amino acids	1							
Plasma and RBC cholinesterase activity	1							
Serotonin	3	2	1	6	3	1		2
Plasma dopamine-beta-hydroxylase		1	1	2				
Thyroid hormone		1	1					
Plasma elements		1		1				
Plasma AA		1					2	
Hemagglutination-inhibition antibody titer		1						
Plasma levels of folates, riboflavin, vitamin B6, and ascorbate		1						
Urine peptides			1	1				
CSF monoamine metabolites			1					
Plasma c-AMP and c-GMP			1					
Hair minerals			2					
Brain opioids				1				
Catecholamines				2				
CSF indoleacetic acid				1				
Homovanillic acid (HVA)				1				
Lactic Acid				1				
Plasma growth hormone response to hypoglycemia				1				
Plasma growth hormone response to oral l-dopa				1				
Platelet size and number				1				
Whole blood tryptophan				1				
Antiserotonin antibodies					1	1		
Growth hormone					1			
Immunoglobulins					1			
Plasma and urinary levels of biopterin, neopterin, and related pterins and plasma levels of folate					1			
Plasma norepinephrine					1			
Oxytocin					2			1
Plasma beta-endorphin					3			
ACTH							1	
Carnitine							1	
FMR1 protein							1	
N-acetyl galactosaminidase deficiency (Schindler disease)							1	
Neuropeptides and neurotrophins							1	
PKU							1	
Secretin							1	
Serum neural cell adhesion molecule (NCAM)							1	
Cortex S6 Ribosomal Protein Phosphorylation								1
Mitochondrial Markers								1
Urine Arginine Vasopressin (AVP)								1
CSF beta-endorphin						1		
IL-2 receptors						1		
Plasma androgens						1		

<b>Legend</b>	
	1
	2
	3
	6



# New Frontiers: THE METABOLOME CONCEPT



# Biomarker challenges in autism:

## *Critical issues*

- Variable severity
  - *Striking “good hair day, bad hair day” changes*
- Chronic features
  - *Oxidative stress, inflammation, metabolic perturbations are ongoing*
- Treatment-responsiveness
  - *Stable improvement can follow treatment*
- Multi-system—gut, immune, sleep, metabolism, more
  - *Is impact on brain primary, parallel or downstream?*
- Heterogeneity
  - *Where are commonalities & final common pathways?*
- Chronic pathophysiological features appear non-specific
  - *Some important potentially treatment-responsive features are non-specific to autism*

# Biomarker challenges in autism: *NOT a “static encephalopathy”*

- **Variable severity**

- *Transient improvement w fever* (Zimmerman A Pediatrics in press)
- *Spikes in function in stress or emotional situations*
- *Transient improvement on antibiotics* (Sandler, Finegold, Bolte, JCN 2000)
- *Improvement on allergy medications*
- *Variability in function related to food, allergen and toxic exposures*

- **Treatment-responsiveness**

- *Stable improvement can follow treatment*
- *Published reports of loss of diagnosis* (Fein D –Sutera, Kelley in JADD '06 & '07)
- *Recovery documentation studies in process*

**Neurobiological Implications:  
NEUROMODULATORS, not just wiring**

**Not just human metabolism:  
Abnormal Clostridial bacteria species  
in autistic children's stool.**

**What we NEED: **Extended Metabolome** for subtyping**

Finegold S, 2002

**9 Clostridial species in  
stool from autistic  
children not in controls**

**3 clostridial species in  
controls not in autistic  
children**

**Strong metabolic  
demands of abnormal  
species.**

**Abnormal gut flora  
*metabolism* can**

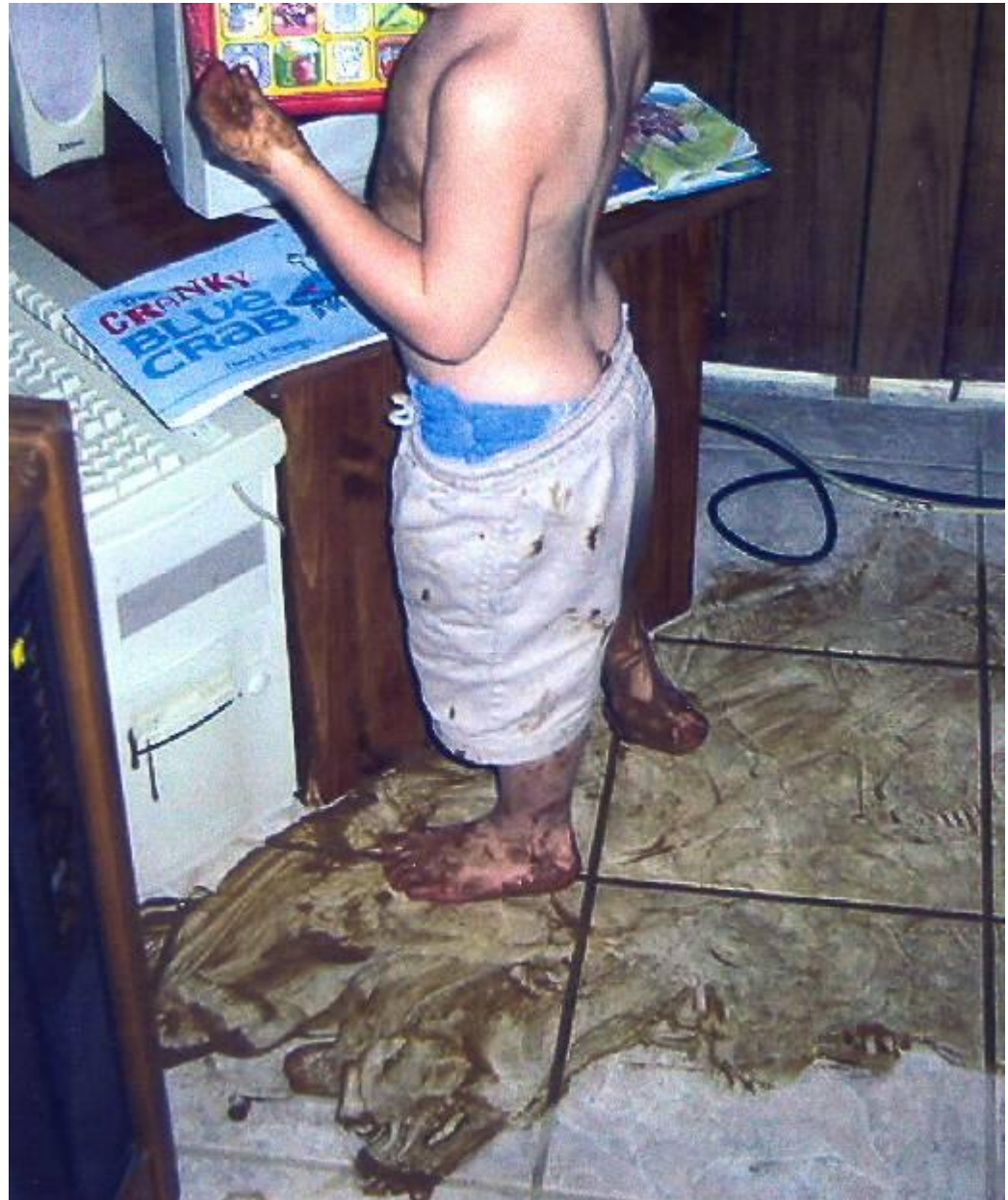
- **deplete vital nutrients**
- **alter metabolism of  
xenobiotics**

**This can cause or  
worsen metabolic  
stress. □ □**

**See also: Host and gut-microbial co-metabolome interaction  
J Nicholson, Nature Reviews Microbiology, 2005**

# The Every Day of Some Autisms

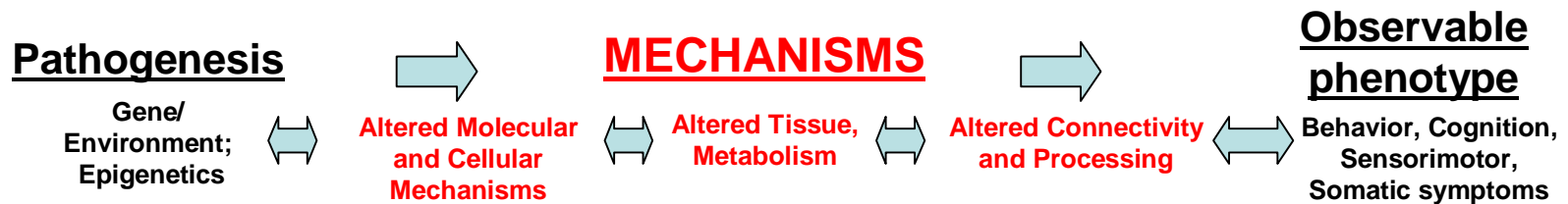
*What we need:*  
Clinical labs that will  
detect and report  
pertinent gut  
pathogens



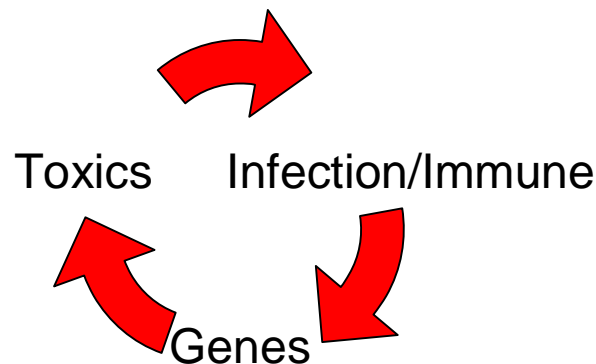
# Two reasons measurements need to be coordinated across levels

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## 1. Heterogeneity at multiple levels: Where is the commonality?



## 2. Clinical vicious circles with amplifying feedback loops across levels



# Conclusion I: **METABOLISM AS CORE FOCUS**

## What we know

- **Environmental factors perturb metabolism** even at low levels of exposure
- Some of the same mechanisms/pathways get hit, but spectrum and intensity of effects differs
  - **Multi-system and multi-level** impacts
- Metabolism is **target for biomedical treatments**

## What we need:

- Study how “Environmental perturbation of metabolism” has **different patterns and thresholds** than “inborn errors of metabolism”

## Conclusion II:

What we *know*:

Metabolism is complex and variable

What we *NEED*:

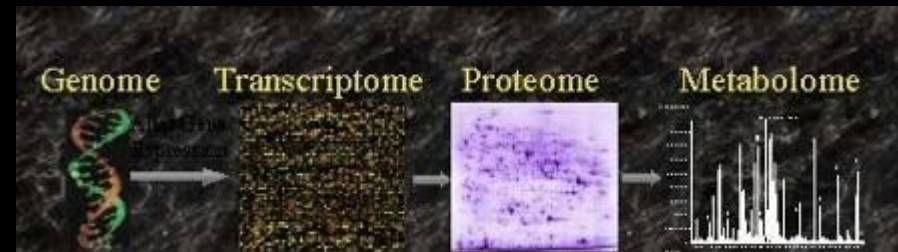
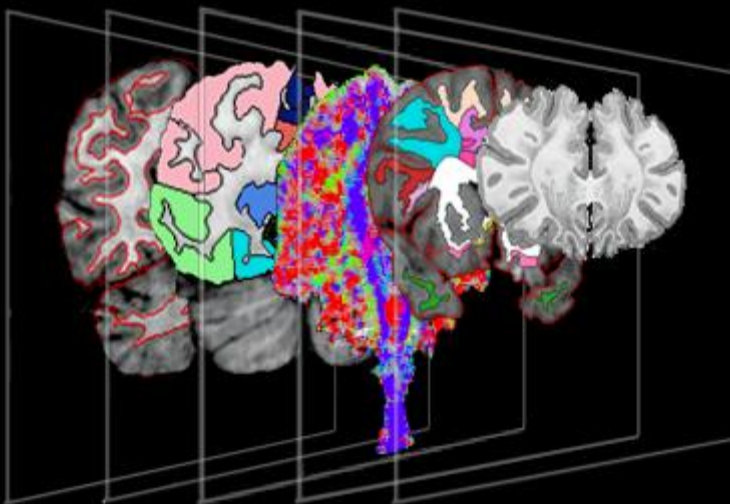
Strong **Infrastructural Support to meet the challenge**

- **Biomarker/metabolite consensus meetings**
  - Identify a **core of measures less sensitive to state and handling**
  - Consider **“Omics” and other profiles**
    - Metabolomics; Extended Metabolomics (gut metabolites)
    - Other –Omics—nutrigenomics, toxicogenomics
    - Organic and amino acid profiling
    - Cytokines, chemokines
  - **SOPs-Standard Operating Procedures**
  - Special focus on **environmentally responsive metabolism**
- **Repository with multi-center participation**
  - Encourage contributions from research projects with well-phenotyped subjects

What we know: Brain and metabolism are abnormal  
What we need: **Learn how metabolism modulates brain  
(and vice versa)**

*This requires **integration.***  
*Integration requires **infrastructure.***

- Brain *tissue* characterization
- High temporal resolution brain function measures (MEG, EEG)
  - closer to metabolically vulnerable synaptic (dys)function
- Systemic metabolic characterization
- Extended metabolome (gut microecology)



## Conclusion III:

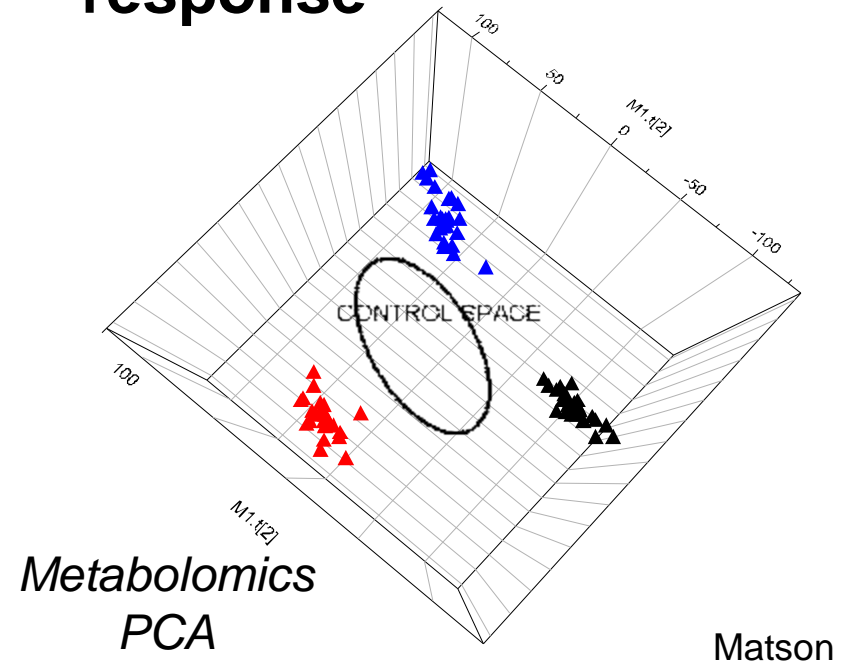
***What we know:*** **Change** and Treatment are possible  
***What we need:*** Better tools to track **treatment and change** biologically

### Repeated measures in same individuals to see what can change

- **Individuals to study**
  - Individuals over time
  - Children at risk for autism
  - Children undergoing treatment
  - Good-hair vs. bad-hair days

### Subgrouping

- Identify mechanisms
- Predict treatment response



Conclusion IV:

***What we know:***

**One biomarker for autism is unlikely.**

***What we need:***

***Profiles of Vulnerability and Treatability***

- **Environmental perturbation of metabolism is widespread and **can be missed by existing reference ranges****
- **Autism's **sensitive physiology** may mean trouble for the individual even when labs are within the population “normal” range**

***Beyond characterizing autism,  
the point is to treat it!***