Neurological Disorders and Purinergic Metabolic Disruption

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Lesch-Nyhan Disease
Discovered by Michael Lesch and William Nyhan in 1964

Causes:

- Disruption of purine metabolism results from **deficiency of the activity of the enzyme hypoxanthine-guanine phosphoribosyl transferase (HPRT)**
  - All are **hyperuricemic**
    - Uric acid levels are between 5 and 10 mg/dL,
    - Normal children excrete less than 1 mg of uric acid
  - Most severe or complete phenotype
- Usually is carried by mother → **passed onto son**
  - **X-linked recessive** genetic disorder
  - Disease is present at birth
  - Rare cases in females - no symptoms
Clinical Presentation

- Impaired kidney functions (nephrolithiasis), acute gouty arthritis, tophi, painful crystalluria, hematuria, UTI → end-stage urate nephropathy
- Motor defects:
  - Involuntary movements (e.g. chorea, athetoid, dystonia)
  - Lose ability to sit, stand unassisted or to walk
  - 25% - Episodic spasm → cervical spinal cord injury, hips dislocation
  - Positive Babinski’ response ⇒ CNS damage
- Neurological impairment
  - Mental retardation (IQ: 40-70)
- Self-mutilating behaviors
  - Bite lips, tongue, fingers, cheeks, hands
  - Head banging
    - Loss of tissues and partial amputations
Progression

- 4–6 months = Normal
  - first sign = “orange sand” in the diapers
- Poor muscle control
  - Developmental delay
- Moderate intellectual disability
- Self-mutilating behaviors
- Scissoring the legs is common
- Most patients eat poorly, and most of them vomit
- Neurological symptoms - facial grimacing, involuntary writhing, and repetitive movements of the arms and legs similar to HD
- The life expectancy is reduced
  - With proper management, most males survive into their teens or 20's
Treatments

No cure

Common treatments - directed toward the specific symptoms:

- **Specialists:** Pediatricians, specialists who diagnose and treat skeletal disorders (orthopedists), physical therapists, and other professionals
- **Uric Acid level:** *allopurinol*
  - *has no effect on the neurological or behavioral symptoms*
- **Kidney stones:** *extracorporeal shock wave lithotripsy (ESWL)*
  - Patient is immersed in water and high energy shock waves are directed to the body in the area of the kidney stone.
  - The stone dissolves into small pieces, and these fragments are passed with the urine
- **Self-injury:** *physical restraint* at the hips, chest, and elbows
HPRT (hypoxanthine-guanine phosphoribosyl transferase) Enzyme
HPRT Enzyme Deficiency

- ↓ hypoxanthine and guanine = ↑ excretion of uric acid
- ↓ PRPP = ↑ de novo pathway, uric acid production
Lesch-Nyhan Disease

- HPRT activity in erythrocytes is very low or almost zero
- Three types:
  - Classic Lesch-Nyhan disease
  - Neurologic variants
  - Those with hyperuricemia
- No consistent correlation between enzyme activity and clinical phenotype measured in the red cell lysate
- Rough inverse correlation between HPRT activity and the severity of clinical manifestations in intact fibroblasts
Neurological variants

- Hyperuricemia with the typical extrapyramidal and pyramidal symptoms found in the classic Lesch-Nyhan patient
- Near normal intelligence
- Normal behavior
Partial HPRT Deficiency Phenotype

- 7.5% of enzyme activity
- Seen in a family with 4 affected members
- Atypical neurologic disease characterized by spasticity
- Positive Babinski’s response, but no dystonia or choreoathetosis
- Increased deep tendon reflexes
- Mild mental retardation
Molecular Genetics of Lesch-Nyhan

- Found on X-chromosome at position Xq2.6-Xq2.7
- 302 known mutations
  - Each family has a unique mutation
Types of Mutations Possible

- Every mutation type has been seen in this gene
- Examples include:
  - Point mutations or single base substitutions -> amino acid substitution
  - Nonsense point mutations -> stop codons and truncated proteins
  - Point mutations at splice sites -> problems with the mRNA
  - Small deletions (exon 4, exons 3, 6, and 7) and large deletions (exons 4 to 9, or the whole gene)
Phenotype Correlates to Residual Enzyme Activity

Exceptions reported where classic phenotypes have relatively sizable activities or mild variant phenotype with no demonstrable enzyme activities

○ Discrepancies between the activity of an aberrant enzyme structure when removed from its in vivo cellular environment
○ Mutations can change the kinetics of an enzyme
  ■ measured at different concentrations (us. greater) than one finds in vivo.
○ Some mutations are unstable
  ■ ie. two duplications recombine, restoring normal enzyme activity in some cells, creating a mosaic of normal and abnormal cells
Prognosis

● Analysis of the nature of mutation
  ○ Helpful when the mutation is known
  ○ Detection of heterozygotes and for prenatal diagnosis
  ○ Predict phenotype in a young presymptomatic infant born into a family with no precedent patients

● Enzyme analysis via the intact well method
  ○ Most reliable
Lesch-Nyhan Disease
Quandaries

Teva Bracha
Standard Inheritance of the Disease

- X-linked recessive inheritance
- Most commonly expressed in males
- Females may become carriers but will not have Lesch-Nyhan phenotype
An Unexpected Affected Female Patient in a Classical Lesch–Nyhan Family

- Family in Argentina with affected male and female siblings
- Caused by nonrandom inactivation of the paternal X chromosome in II-5

**FIG. 1.** Pedigree of the S-family. The proband (II-4) and his sister (II-5) had the classic Lesch–Nyhan phenotype, as indicated by the closed square and circle. The mother (I-1) and one sister (II-2) were clinically normal heterozygotes. III-3 was an affected boy, who died at 3 years of age.
Lesch–Nyhan disease in a female with a clinically normal monozygotic twin

- Mother and both monozygotic twin daughters are carriers for HPRT mutation
- Only one daughter displays LN phenotype
- Preferential X-inactivation in the affected daughter

Activity of enzymes involved in purine metabolism in intact fibroblasts

<table>
<thead>
<tr>
<th>Enzyme</th>
<th>Affected twin</th>
<th>Normal twin</th>
<th>Mother</th>
<th>Control^\text{b} mean ± SD</th>
</tr>
</thead>
<tbody>
<tr>
<td>HPRT activity</td>
<td>230</td>
<td>1606</td>
<td>1887</td>
<td>2066 ± 1468</td>
</tr>
<tr>
<td>GPR activity</td>
<td>330</td>
<td>1126</td>
<td>1705</td>
<td>2298 ± 663</td>
</tr>
</tbody>
</table>

^\text{a} Both the hypoxanthine (H)- and the guanine (G)-phosphoryltransferase activity of the HPRT enzyme were measured using ^14\text{C}-labeled precursors. Enzyme activity is expressed as pmol/100 nmol UV reactive material.

^\text{b} The data on control fibroblasts were obtained at the time that the kindred samples were assayed and reflected a new HPLC column and a new radioactivity flow detector.
Nonrandom/Preferential X-inactivation

- Mutation in the XIST region
  - XIST - gene that promotes X-inactivation
- Female monozygotic twins are often discordant for X-linked diseases
  - Segregation of twins occurs after X-inactivation
    - Happen to segregate by different active X-chromosome
  - Outside factors promote X-inactivation of paternal X-chromosome in affected twin
Basal ganglia dopamine loss due to defect in purine recycling

- Previous studies have shown 70-90% loss of dopamine in basal ganglia of LN patients
- Studied HPRT\(^+\) (normal) and HPRT\(^-\) mice
- Saw significantly decreased relative dopamine in the HPRT\(^-\) model compared to the normal mice

<table>
<thead>
<tr>
<th></th>
<th>Striatum</th>
<th>Midbrain</th>
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<tbody>
<tr>
<td></td>
<td>DA</td>
<td>DA</td>
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<tr>
<td>3–4 weeks</td>
<td></td>
<td></td>
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<tr>
<td>HPRT(^+)</td>
<td>85.9 ± 3.1</td>
<td>2.3 ± 0.1</td>
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<tr>
<td>HPRT(^-)</td>
<td>77.7 ± 3.4*</td>
<td>2.2 ± 0.3</td>
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<tr>
<td>3–6 months</td>
<td></td>
<td></td>
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<tr>
<td>HPRT(^+)</td>
<td>170.1 ± 4.8</td>
<td>2.0 ± 0.1</td>
</tr>
<tr>
<td>HPRT(^-)</td>
<td>84.5 ± 3.0**</td>
<td>1.3 ± 0.3</td>
</tr>
<tr>
<td>2 years</td>
<td></td>
<td></td>
</tr>
<tr>
<td>HPRT(^+)</td>
<td>157.7 ± 2.9</td>
<td>4.3 ± 0.3</td>
</tr>
<tr>
<td>HPRT(^-)</td>
<td>94.5 ± 2.0**</td>
<td>3.0 ± 0.4*</td>
</tr>
</tbody>
</table>
Defective Purine Recycling and Neuron Histology

- Found no significant histological differences between the two groups in their midbrains and striatum.
- Saw no difference in structure at the synaptic level.
Loss of Dopamine in Culture Model

- Used the MN9D cell line and derived HPRT\(^+\) and HPRT\(^-\) cells
  - MN9D cells are DA neurons
  - All HPRT\(^-\) cells had lower levels of dopamine than their HPRT\(^+\) counterpart
Purine Metabolism and Dopamine Synthesis
From Brain to Behavior
Road Map

- Gross Brain Differences
- Basal Ganglia & Cortical Loops Background
- Possible Neural Explanation of Symptomatology via Other Disorders
  - Tourette Syndrome (TS)
  - Huntington’s Disease (HD)
  - Obsessive Compulsive Disorder (OCD)
  - Attention Deficit-Hyperactivity Disorder (ADHD)
Gross Brain Differences

- Brain Volume < Controls by 20%
  - Basal Ganglia
  - Prefrontal Cortex
  - Limbic System
  - Frontal Temporal

- Decreased Dopaminergic Activity
  - Basal Ganglia
  - Prefrontal Cortex
Cortico-Basal Ganglia Circuits

(a) Motor circuit
(b) Associative circuit
(c) Limbic circuit
Basal Ganglia Movement Selection Oversimplified
Tourette Syndrome

- Too Many Motor Commands Initiated in Striatum
  - Dopaminergic Hyperactivity
- Smaller Striatum
Lesch Nyhan’s

- Smaller Striatum

- Dopaminergic Hypoactivity in Striatum
  - Motoric Hypoactivity With Late DA Loss
    - Ie. Parkinson’s
  - Motoric Hyperactivity With Early DA Loss
    - Theoretical Reorganization of Striatum
      - Shown in Mouse Models
    - DA Hypoactivity in LND Could Function Like Hyperactivity in TS
Lesch Nyhan’s

- Much Neuronal Loss in Basal Ganglia
  - Chorea Possibly from Damaged Indirect Pathway
Obsessive Compulsive Disorder

- Orbitofrontal & ACC Dysfunction
  - These Evaluate Stimuli and Responses
    - Overvalue Threat Leading to Obsession
    - Undervalue Cost of Compulsion
  - Exact Interaction with Basal Ganglia Unknown
    - Idea: Cortical Areas Don’t Bias Striatal Movement Selection Properly
  - Leads to Circuit Formation Supporting Compulsion as Habit
    - Long-term
Lesch Nyhan’s

- Smaller Orbitofrontal & ACC
- Habitual Self-Mutilation Despite Pain
- No Obsessions in LND
  - Smaller Orbitofrontal & ACC May Undervalue Resulting Pain
    - Too Little Inhibition of Urge in Striatum
Attention Deficit-Hyperactivity Disorder

- Smaller Striatum
  - Motoric Hyperactivity

- Decreased Frontal Volume & Dopaminergic Activity
  - Poor Response Inhibition
    - Hyperdirect Pathway
  - Poor Tonic Inhibition?
    - Monkeys with Severe Right IFC Ablation
      - Ballistic Movements
Lesch Nyhan’s

- Smaller Striatum
  - Motoric Hyperactivity

- Decreased Frontal Volume & Dopaminergic Activity
  - Smaller Frontal Lobe than ADHD
    - Could Result in Very Low Tonic Inhibition
      - Causing or Potentiating Chorea
    - Decreased Ability to Control Actions
      - Trouble Stopping Movements Causing Pain?
Summary

- Lots of Redundancy Built In
- LND Sufferers are Screwed