PEDIATRIC MULTIPLE SCLEROSIS CLINICAL AND TRANSCRIPTIONAL ASPECTS

MULTIPLE SCLEROSIS CENTER

Gurevich M.
PEDIATRIC MS

• Pediatric MS is defined as onset MS under the age of 18 years

• Pediatric MS occurs in about 3% of MS cases

• MS onset before puberty (<12 years) is defined as childhood and between 12-18 years is defined as juvenile Pediatric MS

• Phenotype: Relapsing-remitting MS >98%, Primary Progressive <2%

• As compared to adult patients, Pediatric MS is characterized by a more aggressive initial course, but better recovery and slower disease progression (Renoux C, et al. 2007)
GENDER EFFECTS IN ADULT MS

- Higher prevalence of MS in females

- Amelioration of MS during pregnancy (Finkelsztein A, 2011, meta-analysis of 22 reports, 13,141 women, ~70% suppression of RR)

- Sex hormones demonstrate anti-inflammatory and neuroprotective effects in MS clinical trials (Voskuhl RR, et al, 2016).


- Clinical phenotype in pediatric MS patients could be affected by pubertal transition.

- Effects of puberty on pediatric MS clinical presentation could be different in males and females.
OUR HYPOTHESIS

- Hormonal changes associated with puberty in Pediatric MS patients could ameliorate the disease.
- The expected effect of puberty could be gender dependent.
EFFECTS OF AGE AND GENDER ON DISEASE PROGRESSION IN PEDIATRIC MULTIPLE SCLEROSIS POPULATION

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METHODS

• Data of Pediatric MS patients followed prospectively in Sheba MS Center from 1995-2016 were analyzed. Patients were diagnosed according to 2010 McDonald criteria.
• Queried for onset of puberty by Tanner stage.

We examined the differences between childhood (< 12 years) and juvenile (12-18 years) MS patients as follows:
• Onset parameters:
  number and volume of brain MRI lesions, EDSS, EDSS presenting symptoms.
• Clinical outcome parameters:
  time to second relapse, time to EDSS 3.0, 5 years ARR, EDSS at 5, 10, 15 and 20 years from onset.
RESULTS
DISTRIBUTION OF PEDIATRIC MS PATIENTS BY AGE AT ONSET, N=133

Number of patients

Childhood 13%

Juvenile 87%

years
DISTRIBUTION OF PEDIATRIC MS PATIENTS BY GENDER

**Pediatric MS onset**
- N=133
- Onset: 15.0 ± 0.3 Y
- F:M 1.7

**Childhood MS onset**
- N=17
- Onset: 8.8 ± 0.7 Y
- F:M 0.9
  - Females: N=8
    - Onset: 8.6 ± 0.6 Y
  - Males: N=9
    - Onset: 9.0 ± 1.2 Y

**Juvenile MS onset**
- N=116
- Onset: 15.9 ± 0.1 Y
- F:M 1.9
  - Females: N=76
    - Onset: 15.9 ± 0.2 Y
  - Males: N=40
    - Onset: 14.8 ± 0.5 Y
**AGE EFFECT:** DISABILITY AT ONSET IS NOT AFFECTED BY AGE

Disability at onset

<table>
<thead>
<tr>
<th>EDSS</th>
<th>Childhood</th>
<th>Juvenile</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td></td>
<td></td>
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<tr>
<td>1</td>
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</tbody>
</table>

Proportion of EDSS functional systems

- Pyramidal: \( p < 0.01 \)
- Sensory: \( p < 0.03 \)
- Visual: \( p < 0.1 \)
- Cerebellar: \( p < 0.01 \)
- Brainstem: \( p < 0.01 \)
- Bladder: \( p < 0.01 \)

Number of EDSS functional systems

<table>
<thead>
<tr>
<th>Number</th>
<th>P = 0.03</th>
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<tr>
<td>0</td>
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<td>2</td>
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</table>

No differences in MRI parameters at onset
AGE EFFECT IN MALES: SEVERITY OF ONSET IS NOT AFFECTED BY PEDIATRIC MALE AGE

Disability at onset

<table>
<thead>
<tr>
<th>EDSS</th>
<th>Childhood</th>
<th>Juvenile</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td></td>
<td></td>
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<tr>
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<tr>
<td>4</td>
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</table>

Proportion of EDSS functional systems

- Pyramidal: p = 0.01
- Sensory: p = 0.02
- Visual: p = 0.5
- Brainstem: p = 0.01
- Cerebellar: p = 0.01
- Bladder: p = 0.5

No differences in MRI parameters at onset

Number of EDSS functional systems

- p = 0.01
AGE EFFECT IN FEMALES: DISABILITY AT ONSET
NOT AFFECTED BY AGE

No differences in MRI parameters at onset

Disability at onset

Proportion of EDSS functional systems

EDSS

<table>
<thead>
<tr>
<th>p</th>
<th>0.2</th>
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<tbody>
<tr>
<td>4</td>
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<tr>
<td>3</td>
<td></td>
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<tr>
<td>2</td>
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<tr>
<td>1</td>
<td></td>
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<tr>
<td>0</td>
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</tr>
</tbody>
</table>

Childhood | Juvenile

Pyramidal p=0.01

Sensory p=0.8

Visual p=0.05

Brainstem p=0.01

Cerebellar p=0.01

Pyramidal

60

40

20

0

Bladder p=0.01
GENDER EFFECTS CHILDHOOD MS: SEVERITY OF ONSET NOT AFFECTED BY GENDER IN CHILDHOOD MS

Disability at onset

Proportion of EDSS functional systems

No differences in clinical outcome parameters
GENDER EFFECTS IN JUVENILE MS: SEVERITY OF ONSET AND CLINICAL OUTCOME NOT AFFECTED BY GENDER IN JUVENILE MS

No changes in others onset and clinical outcome parameters
AGE EFFECT: BETTER CLINICAL OUTCOME IN JUVENILE PATIENTS

<table>
<thead>
<tr>
<th>Clinical parameters</th>
<th>Childhood MS N=17</th>
<th>Juvenile MS N=116</th>
<th>p value</th>
</tr>
</thead>
<tbody>
<tr>
<td>EDSS at 5 years</td>
<td>2.3 ± 0.6 (N=13)</td>
<td>1.5 ± 0.3 (N=64)</td>
<td>0.05</td>
</tr>
<tr>
<td>EDSS at 10 years</td>
<td>3.6±0.7 (N=12)</td>
<td>2.9 ± 0.4 (N=52)</td>
<td>0.1</td>
</tr>
<tr>
<td>EDSS at 15 years</td>
<td>5.2 ± 0.7 (N=12)</td>
<td>3.3 ± 0.4 (N=43)</td>
<td>0.01</td>
</tr>
<tr>
<td>EDSS at 20 years</td>
<td>5.7 ± 0.9 (N=8)</td>
<td>3.5 ± 0.1 (N=41)</td>
<td>0.01</td>
</tr>
</tbody>
</table>

Pediatric MS disease progression

No differences in other outcome parameters
AGE EFFECT IN MALES: JUVENILE MALES HAVE BETTER CLINICAL OUTCOME

No differences in other outcome parameters
CONCLUSIONS:

- In pediatric patients disability at onset is not affected by age or gender.
- Juvenile patients and especially juvenile males have better clinical outcome.

Our findings might be explained by the protective immunomodulatory effect of sex hormones during puberty.
GENE-EXPRESSION ANALYSIS OF PEDIATRIC PATIENTS WITH MULTIPLE SCLEROSIS TREATED WITH NATALIZUMAB

BRACHA ROBINSON
NATALIZUMAB

• Humanized monoclonal Antibody targeting a4 subunit of a4b1-integrin
• FDA approved therapy for adults
• In children similar efficacy to adults
  • Fewer relapses
  • No enhancing lesions on MRI
  • No association with PML (unlike adults)
• Effect on childhood development is not fully understood
OUR GOAL

- To determine whether Natalizumab induces changes in gene expression in pediatric MS similar to adult MS patients
- To determine whether Natalizumab treatment affects the expression of essential childhood and adolescent developmental genes
PATIENT CHARACTERISTICS

- 18 Patients treated with Natalizumab
  - Of these, 10 patients were sampled for gene expression analysis

<table>
<thead>
<tr>
<th></th>
<th>Total n=18</th>
<th>Female n=10</th>
<th>Male n=8</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age MS onset</td>
<td>13.9±0.8</td>
<td>13.8±1.0</td>
<td>14.1±1.2</td>
</tr>
<tr>
<td>Age Tx start</td>
<td>14.8±0.6</td>
<td>14.5±0.8</td>
<td>15.1±1.1</td>
</tr>
<tr>
<td>EDSS before Tx</td>
<td>1.67±0.27</td>
<td>1.95±0.36</td>
<td>1.31±0.38</td>
</tr>
<tr>
<td>Annual Relapse Rate before Tx</td>
<td>1.33±0.26</td>
<td>1.20±0.31</td>
<td>1.48±0.45</td>
</tr>
</tbody>
</table>
CLINICAL RESULTS: NATALIZUMAB INDUCES CLINICAL EFFICACY

Annual Relapse Rate

1 - before treatment
2 - one year after Natalizumab treatment

Annual EDSS Rate

Switch of Annual EDSS rate by Natalizumab treatment

Onset of treatment

p=3.7E-05

PCA of Blood gene expression

Red = Before treatment
Blue = After treatment
COMPARISON TO ADULTS (FUNCTIONAL ANALYSIS)
NATALIZUMAB ALTERS GENE EXPRESSION IN CHILDREN AND ADULTS IN SIMILAR WAYS

<table>
<thead>
<tr>
<th>Diseases or Functions Annotation</th>
<th>p-Value</th>
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<tbody>
<tr>
<td>differentiation of pre-B lymphocytes</td>
<td>3.05E-09</td>
</tr>
<tr>
<td>differentiation of B lymphocytes</td>
<td>5.34E-07</td>
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<tr>
<td>arrest in differentiation of B lymphocytes</td>
<td>1.37E-06</td>
</tr>
<tr>
<td>proliferation of B lymphocytes</td>
<td>4.23E-06</td>
</tr>
<tr>
<td>development of B lymphocytes</td>
<td>5.22E-06</td>
</tr>
<tr>
<td>development of pre-B lymphocytes</td>
<td>1.37E-05</td>
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<tr>
<td>expansion of B lymphocytes</td>
<td>0.000104</td>
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<td>proliferation of pre-B lymphocytes</td>
<td>0.000245</td>
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<td>lifespan of B lymphocytes</td>
<td>0.000486</td>
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<tr>
<td>differentiation of plasma cells</td>
<td>0.000611</td>
</tr>
<tr>
<td>development of follicular B lymphocytes</td>
<td>0.000835</td>
</tr>
</tbody>
</table>

It has been previously shown that in adults, Natalizumab alters expression of genes related to B cell activity.
WEIGHTED GENE CO-EXPRESSION NETWORK ANALYSIS (WGCNA)

- Developed by Steve Horvath and Peter Langfelder of UCLA in 2008
- This method creates clusters of highly correlated genes which can be related to external traits

Step 1: Construct a Network
Quantify connection and determine interaction patterns between genes

Step 2: Identify Modules
Find gene clusters

Step 3: Relate Modules to External Information
Find biologically interesting modules
WGCNA: GENE CLUSTERS

Each black line represents one gene.

Each color represents one cluster.

We merged highly-related clusters together.
WGCNA: COMPARISON TO EXTERNAL TRAITS

Names of clusters

Clinical traits

Red = positive correlation
Green = negative correlation
(p-values in parentheses)
Circles = p ≤ 0.05

Clusters that are correlated with treatment are not the same clusters that are correlated with age, gender, and Tanner scale → Natalizumab does not affect age-related genes.
CONCLUSIONS

Natalizumab treatment is an effective treatment in pediatric patients

Natalizumab does not affect gene expression modules that are associated with childhood development.