Higher C-peptide, higher neutrophil and lower NK peripheral counts at T1D onset – biomarkers for a longer remission phase?

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INTRODUCTION

Type 1 diabetes (T1D) is an autoimmune disease resulting from the interaction of environmental, genetic and complex immunologic mechanisms. Besides the known loss of balance between regulatory and effector T cells, other immune cell populations are involved.

Human T cell development through distinct phases with particular immunologic and metabolic features, in the remission phase, a partial and transient restoration of insulin production occurs.

Existing metabolic and immune biomarkers do not reflect the stages or severity of disease. Immunologic characterization throughout disease development is essential to identify biomarkers that allow the individualization and monitoring of immune modulation strategies.

OBJECTIVE

To identify clinically useful biomarkers for longer remission phase.

METHODS

Prospective evaluation of children with T1D, along disease development
Children aged 2–18 years, 1+ positive auto-Ab
Exclusion criteria: other autoimmune disease, allergic disease, infection.
Patients and age-matched controls PB samples were analysed by flow cytometry.
Metabolic data were prospectively collected.

In this data subset, relations between cellular populations, metabolic data and remission phase duration were analysed. Statistical significance: p<0.05

RESULTS

Table 1: Characteristics of study population

<table>
<thead>
<tr>
<th>Age (years)</th>
<th>Sex (M:F)</th>
<th>positive AIA</th>
<th>positive IA2</th>
<th>positive GAD</th>
<th>IDAA1C (mean, SD)</th>
<th>Direct fasting C-peptide (ng/ml, median, IQR)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Patients (n=28)</td>
<td>10 (5)</td>
<td>14/14</td>
<td>24/28</td>
<td>9/28</td>
<td>10/28</td>
<td>12.7 (2.4)</td>
</tr>
<tr>
<td>Controls (n=20)</td>
<td>10 (5)</td>
<td>14/14</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>

New-onset C-peptide level was positively related to remission time (r=0.389; p=0.05)

Relative neutrophil count at onset was positively related to remission duration (r=0.412; p=0.03)

Inversely, NK count in T1 was negatively related to remission phase duration (r=-0.538; p=0.003)

DISCUSSION AND CONCLUSIONS

In our DTI cohort, both neutrophils and NK were significantly decreased in T1D children at disease onset, compared to controls. As disease progressed, neutrophils increased to values comparable to controls, but NK remained below controls (unpublished data).

Higher peripheral neutrophil levels at disease onset may signal less intense pancreatic infiltration and therefore a less severe initial beta-cell mass destruction. This may reflect a more preserved residual insulin production (higher C-peptide levels) at disease onset and thus a longer remission phase.

The role of NK on T1D pathogenesis is intriguing. Data on NK involvement in insulitis are diverse, from scarce to massive infiltration in viral infection setting. Considering a pathogenic role, lower NK counts may predict a longer remission phase because of functional and/or quantitative defects or previous immune cellular exhaustion. Admitting a beneficial role in insulitis, lower peripheral counts may be due to increased protective pancreatic migration.

STUDY DESIGN

T1D PATIENTS

<table>
<thead>
<tr>
<th>Study phase</th>
<th>C-peptide (ng/ml)</th>
<th>NK cells (% of total WBC)</th>
<th>Neutrophils (% of total WBC)</th>
</tr>
</thead>
<tbody>
<tr>
<td>T1 – onset</td>
<td>0.00-0.20</td>
<td>0.00-0.40</td>
<td>0.50-0.80</td>
</tr>
<tr>
<td>T2 – remission</td>
<td>0.20-0.80</td>
<td>0.40-0.80</td>
<td>0.50-0.80</td>
</tr>
<tr>
<td>T3 – established remission</td>
<td>0.80-1.00</td>
<td>0.80-1.00</td>
<td>0.80-1.00</td>
</tr>
</tbody>
</table>

Children with new-onset C-peptide levels >0.4 ng/ml at T1 had higher neutrophil counts (p=0.03)

At remission phase entrance, children with lower new-onset C-peptide (≤0.4 ng/ml) had significantly lower neutrophil levels (p=0.02), higher Th1 (p=0.04) and total IFN-producing cells (p=0.05)

Neither Th17, Tc17, Th1, Tc1 nor Treg related significantly with the remission phase.

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RESULTS

Table 2: Timespan between disease stages

<table>
<thead>
<tr>
<th>Disease stage</th>
<th>Days (mean ± SD)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Diagnos – T1</td>
<td>46±2 days</td>
</tr>
<tr>
<td>T1 – T2</td>
<td>111±45 days</td>
</tr>
<tr>
<td>T2 – T3</td>
<td>279±118 days</td>
</tr>
<tr>
<td>T1 – T3</td>
<td>338±104 days</td>
</tr>
</tbody>
</table>

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