Triple threats:
Tracking antigen-specific T cells in a case of concurrent autoimmunity, infectious disease and possible malignancy

Dr Grace Thompson
Mr AT

- 50 year old Somalian refugee
  - 3-4 months of worsening abdominal pain and distension
  - Fevers and night sweats
  - Nausea and vomiting
  - LOW 10kg

Background
- Coeliac disease
  - Diagnosed 1 year prior
  - Strongly positive tissue transglutaminase antibodies
Examination

- Febrile 38.3
- Oral candidiasis
- Abdominal distension impression of palpable nodularity
  No lymphadenopathy or organomegaly
Investigations
Further Investigations

- HIV EIA – Positive
- HIV western blot – positive.
- Peripheral blood lymphocyte subsets
  - Total lymphocyte count 0.7
  - CD3+: 651 - 93% (58-84%)
  - CD4+: 7 – 1% (30-61%)
  - CD8+: 637 - 91% (12-45%)
  - CD4/CD8 ratio: 0.01 (0.72-4.18)
  - CD19+: 7 – 1% (6-22%)
  - CD16/56+: 35 – 5% (6-28%)
Severe AIDS
- small bowel and omental infiltrative disease
- Relative CD8 lymphocytosis

- ?Opportunistic infection
  - Extensive microbiological investigations unable to find an infective cause
- ?Lymphoma
  - Enteropathy associated T cell lymphoma
  - HIV/AIDS related lymphoma
- ?Diffuse infiltrative lymphocytosis syndrome
• Immunophenotyping
  (omentum, lymph node, peripheral blood, bone marrow)
  • Absent or few B lymphocytes with a predominance of CD8+ T cells. Expressing normal T cell markers.

• Histopathology
  • Mixed inflammation no definite neoplasia

• TCR gene rearrangements
  (omentum, lymph node, peripheral blood)
  • Two possible TCR products on capillary electrophoresis
  • High background
• 8 weeks into hospital admission still no diagnosis
• Commenced on HAART and anti-fungal therapy
  • Virological response (11749 – 282 copies/ml)
• No improvement in abdominal disease
  • Ongoing weight loss
  • Requiring total parental nutrition

• TCR sequencing using next generation sequencing...
Next generation sequencing by Illumina
• Gluten challenge following GFD
• Induced antigen specific CD4 T cells in circulation expressing α4β7, CD38
• Induced CD8 T cells (αEb7+/CD103+, CD38+)
• Not detected in healthy patients post oral challenge, peak at day 6
- Induced CD8+ T cells sorted and sequenced TCR

<table>
<thead>
<tr>
<th>V-gene</th>
<th>CDR3 motif</th>
<th>celiac clones</th>
<th>healthy clones</th>
<th>Significance</th>
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<td>TRBV7-9</td>
<td>CxxxxGN</td>
<td>12/36</td>
<td>12/9584</td>
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<td>47/1144</td>
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</table>

- One of which corresponded to the CD8 αβ TCR in our case
TCR diversity

- Thymic VDJ recombination generates approximately $10^9$ unique TCR
- Next generation sequencing methods allow
  - capture TCR diversity
  - true clonality
- TCR’s are unique to an individual
- Public TCR’s are frequently observed in multiple individuals
- Described in a variety of immune responses including:
  - Infectious diseases, Malignancy, Autoimmunity
- Identification of which may have potential diagnostic and therapeutic implications
Back to the case

- CD8 lymphoproliferation not driven by HIV or malignancy BUT coeliac disease
- Identification of a potential coeliac disease public TCR
- Commenced on strict gluten free diet and oral prednisolone
- Small bowel and omental thickening resolved, able to tolerate diet and come of total parental nutrition
Conclusions

• Tremendous advance in our ability to capture TCR diversity through next generation sequencing

• Allowed identification of coeliac disease as the driver of a CD8 lymphoproliferative disorder

• Applications of TCR sequencing
  • Lymphoma detection
    • Cancer immune responses, minimal residual disease and acute allograft rejection.
  • Potentially in investigating infection responses and autoimmunity
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