Differential risk of DQ2.5 and DQ2.2 to celiac disease is related to sustained gluten antigen presentation

Lars-Egil Fallang\textsuperscript{1*}, Elin Bergseng\textsuperscript{1*}, Kinya Hotta\textsuperscript{2}, Axel Berg-Larsen\textsuperscript{1},

Chu-Young Kim\textsuperscript{2} & Ludvig M. Sollid\textsuperscript{1}

1. Centre for Immune Regulation, Institute of Immunology, University of Oslo and Oslo University Hospital - Rikshospitalet, 0027 Oslo, Norway
2. Department of Biological Sciences, National University of Singapore, Singapore 117543

* These authors have contributed equally to this work
**Supplementary Figure 1. Critical role of DQ\(\alpha_{22}\) in the dissociation rate of the CLIP1 peptide from DQ2.** Dissociation of CLIP1 peptide from thrombin-treated water soluble DQ2.5 (■), DQ2.2 (●), DQ2.5 Y\(\alpha_{22}\)F (□) and DQ2.2 F\(\alpha_{22}\)Y (○) molecules (1.6 \(\mu\)M) in the presence of excess competitive high affinity peptides (35 \(\mu\)M). The off-rates are presented in Table 2. Peptide release was determined by MALDI-TOF MS analysis comparing the intensity of the isotopic peaks of added indicator peptides with those of the released peptides. Data from the mean of at least two independent experiments were fitted to a one-phase exponential decay function (\(Y = As \times \exp(-sx)\)) and values for \(t_{1/2}\) were calculated.
**Supplementary Figure 2.** DQ\(\alpha_{44,47-51}\) residues do not alter HLA-DM mediated peptide dissociation from DQ2. Dissociation of CLIP1 peptide from thrombin-treated water soluble DQ2.5 (square), DQ2.2 (circle) and DQ2.5 \(\alpha_{44,47-51}\) (triangle) molecules (1.6 µM) in the presence of excess competitive high affinity peptides (35 µM), and in the presence (a) or absence (b) of HLA-DM (6 µM). The off-rates are presented in Supplementary Table 1. Peptide release was determined as in Supplementary Figure 1. Data are representative of at least two independent experiments.
Supplementary Figure 3. Critical role of DQα22 in the dissociation rate of the DQ2-γ-I peptide from DQ2. Dissociation of DQ2-γ-I peptide from thrombin-treated water soluble DQ2.5 (■), DQ2.2 (●), DQ2.5 Yα22F (□) and DQ2.2 Fα22Y (○) molecules (1.6 μM) in the presence of excess competitive high affinity peptides (35 μM). The off-rates are presented in Table 2. Peptide release was determined as in Supplementary Figure 1. Data are representative of at least two independent experiments.
Supplementary Figure 4. DQα22 affects kinetic stability of most DQ2 ligands. Thrombin-treated water soluble DQ2.5 (■), DQ2.2 (●), DQ2.5 Yα22F (□) and DQ2.2 Fα22Y (○) molecules were loaded with the non-gliadin derived fluorescent dye-labeled peptides (a) P2019 (YLTFLPSADEIYD), (b) P198 (KPLLIIAEDVEGEY), or (c) P1538 (AAIAAVKEEAF) and allowed to dissociate in the presence of an excess of unlabeled competitive high affinity peptide. Data from one of at least two independent experiments were fitted to a one-phase exponential decay function ($Y = A_s x \exp(-sx)$) or two-phase exponential decay function ($Y = A_r x \exp(-k_rx) + A_s x \exp(-k_sx)$) and values for $t_{1/2}$ were calculated. The off-rates are presented in Table 3.
Supplementary Table 1. CLIP1 peptide dissociation in the absence and presence of HLA-DM.

<table>
<thead>
<tr>
<th>Constructs</th>
<th>Dissociation t_{1/2} (h) (95% CI)</th>
<th>In absence of DM</th>
<th>In presence of DM</th>
</tr>
</thead>
<tbody>
<tr>
<td>sDQ2.5-CLIP1</td>
<td>144 (110-208)</td>
<td>9.0* (6.4-15.5)</td>
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<tr>
<td>sDQ2.2-CLIP1</td>
<td>4.8 (4.0-6.0)</td>
<td>0.3 (0.19-0.47)</td>
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<tr>
<td>sDQ2.5 α44,47-51-CLIP1</td>
<td>80 (56-141)</td>
<td>5.1 (4.4-6.2)</td>
<td></td>
</tr>
</tbody>
</table>

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