### A

**A**

![Blot α-GFP](image)

<table>
<thead>
<tr>
<th></th>
<th>α-SedV</th>
<th>α-TlgB</th>
<th>α-PepA</th>
</tr>
</thead>
<tbody>
<tr>
<td>GFP-SedV</td>
<td>IP</td>
<td>IP</td>
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<tr>
<td>GFP-TlgB</td>
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<tr>
<td>GFP-PepA</td>
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</tbody>
</table>

### B

**B**

![Western Blot](image)

- **Qb**: HA3-Vti1
- **Qc**: HA3-Tlg1, HA3-Syn8, HA3-Vam7
- **R**: HA3-SynA, Ykt6-HA3, Nyv1-HA3

### C

**C**

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<tr>
<th></th>
<th>Vti1</th>
<th>Tlg1</th>
<th>Syn8</th>
<th>Vam7</th>
<th>SynA</th>
<th>Ykt6</th>
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</table>
A Direct or indirect interaction test

![Diagram showing the interaction between proteins Stx1 and Stx2 with SM1 and SM2, and the interaction between Vps33-HA3 and Vps45-HA3 extracts.]

- **IPs of Vps45-HA3 extracts**
  - Input TNT
  - IP, pre
  - α-TlgB, α-PepA
  - Lane: 1 2 3 4 5 6 7 8 9 10

- **IPs of Vps33-HA3 extracts**
  - Input TNT
  - IP, pre
  - α-TlgB, α-PepA
  - Lane: 1 2 3 4 5 6 7 8 9 10

B Direct interaction test

![Diagram showing the interaction between proteins Stx1 and SM1-HA3, and the interaction between TNT-synthesized proteins with SM1 and Stx1.]

- **IPs of TNT-synthesized proteins**
  - Input TNT
  - IP, pre
  - α-TlgB, α-PepA
  - Vps45 or Vps33

- **IPs of TNT-synthesized proteins**
  - Input TNT
  - IP, pre
  - α-TlgB, α-PepA
  - Vps45 or Vps33