Supporting Data

Subproject 1: Large clostridial toxin complexes

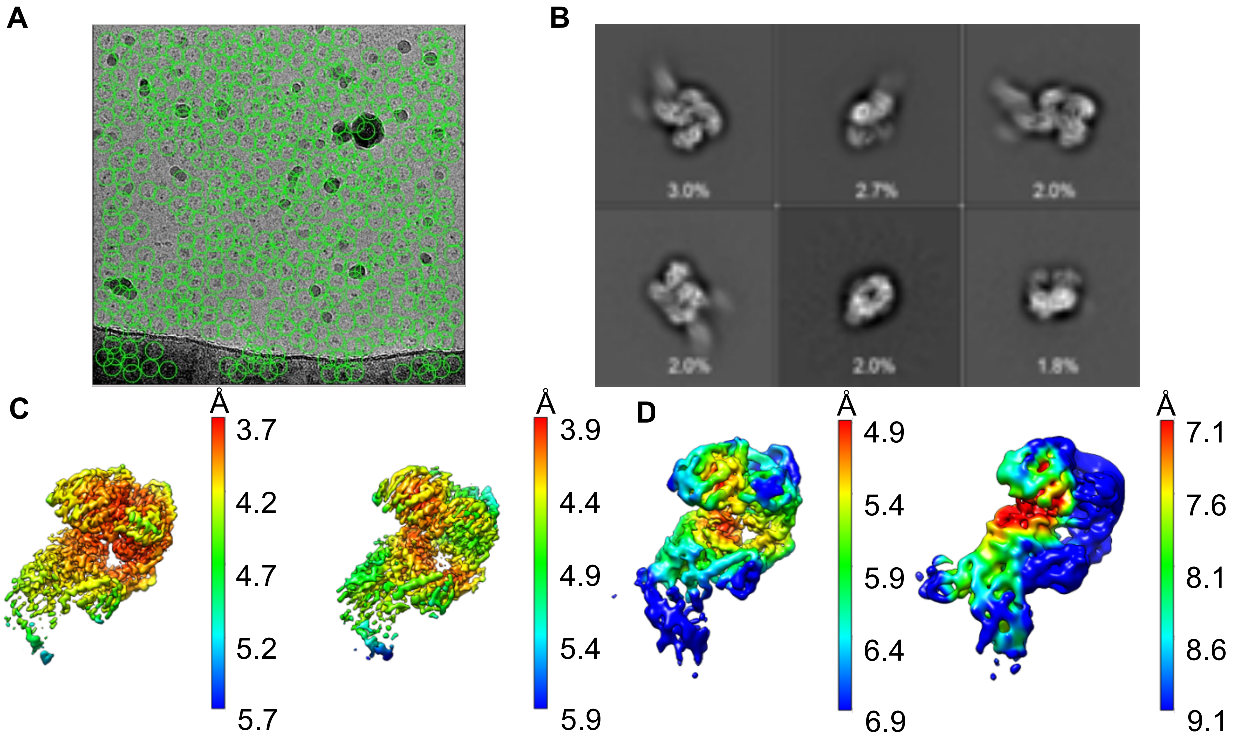

Fig. 1.1 Reconstructions of TcdB with and without the Fzd2 receptor. *A) We have obtained vitrification conditions of TcdB in complex with a number of host cell receptors, including Fzd2. B) From these data we observe all possible orientations as indicated by 2D classification. C) This analysis has led to high resolution reconstructions in the apo (left) and Fzd2 bound forms (right), D) though portions of the molecule are only present at low resolution. We may be able to address this with more data.*

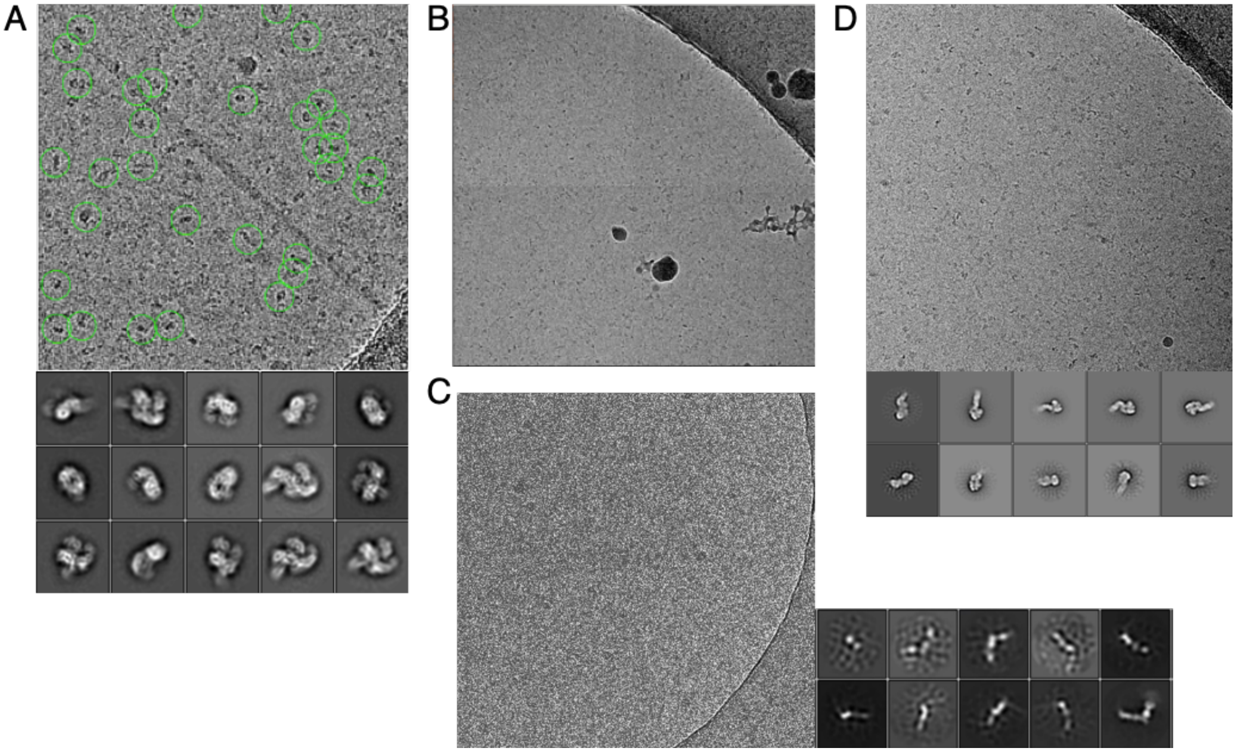

Fig. 1.2 Ongoing LCT structural projects. *A) Vitrification conditions for TcdB-NECTIN3-R2 complexes and preliminary class averages (lower panel). B) Vitrification conditions for TcdB-CSPG4. C) Vitrification conditions for TcdB-Bezlotoumab with preliminary class averages (lower right panel) D) Vitrification conditions for TpeL:LRP1 complex and negative stain class averages (lower panel).*

Subproject 2: Iota toxins

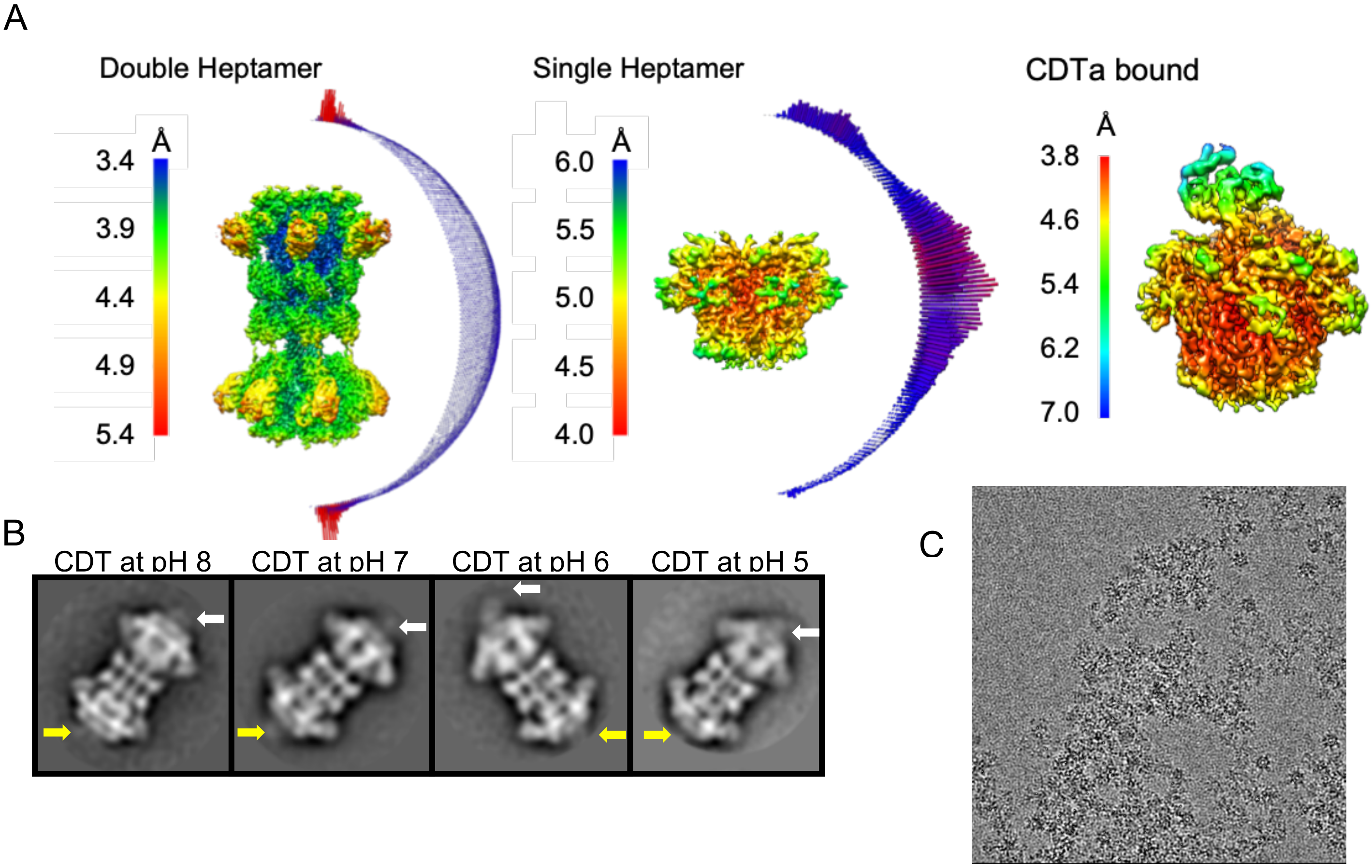

Fig. 2. Analysis of Iota Toxins. *A) Maps from three distinct forms of CDT have been reconstructed by cryo-EM. These include the CDTb double heptamer (left), CDTb single heptamer (center), and CDTa bound forms. All three have been reconstructed to better than 4 Å leading to near-atomic resolution models of each. B) We have shown that CDTa remains bound to the CDTb double heptamer at low pH, suitable for cryo-EM reconstruction. C) We have obtained vitrification conditions of the CDT complex in the presence of glycan receptor.*

Subproject 3: Regulation of TcdA/TcdB toxin production

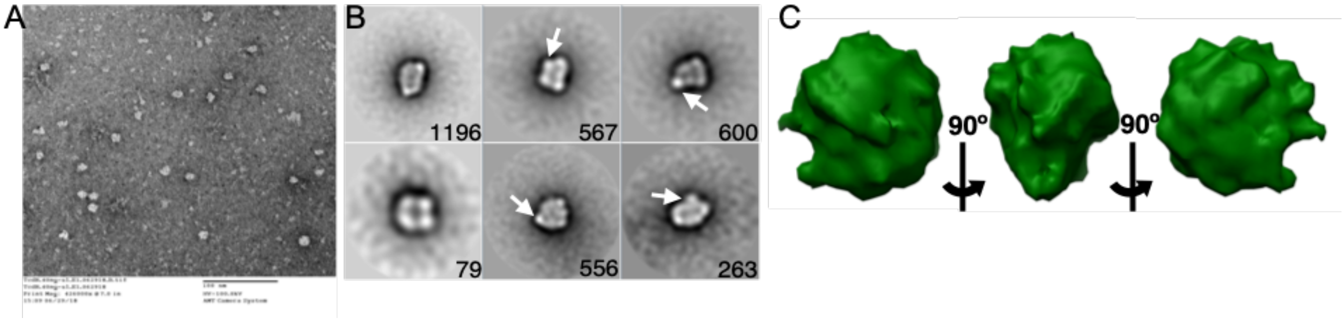

Fig. 3. A Negative stain structure of the TcdR sigma factor bound to RNA Polymerase. A. *Negative-stained TEM images TcdR/RNA polymerase at 40 ng/µL obtained at 44,000x using a 100 keV beam. Complexes are ~120 x 170 Å in size (scale bar 1000 Å). B. 2D alignments of 3,261 un-tilted particles with the number of particles per class indicated in black. Some classes exhibit defined protrusions (white arrows). A 3D model of TcdR bound to E. coli RNA polymerase generated from 3,261 particles. Model has a resolution of >20 Å due to the limitations of negative-stain EM.*