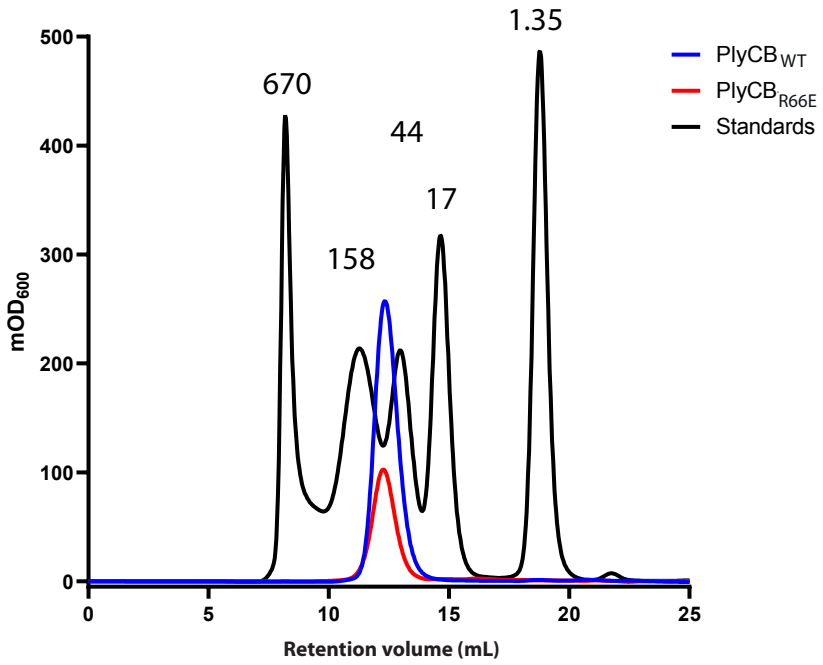
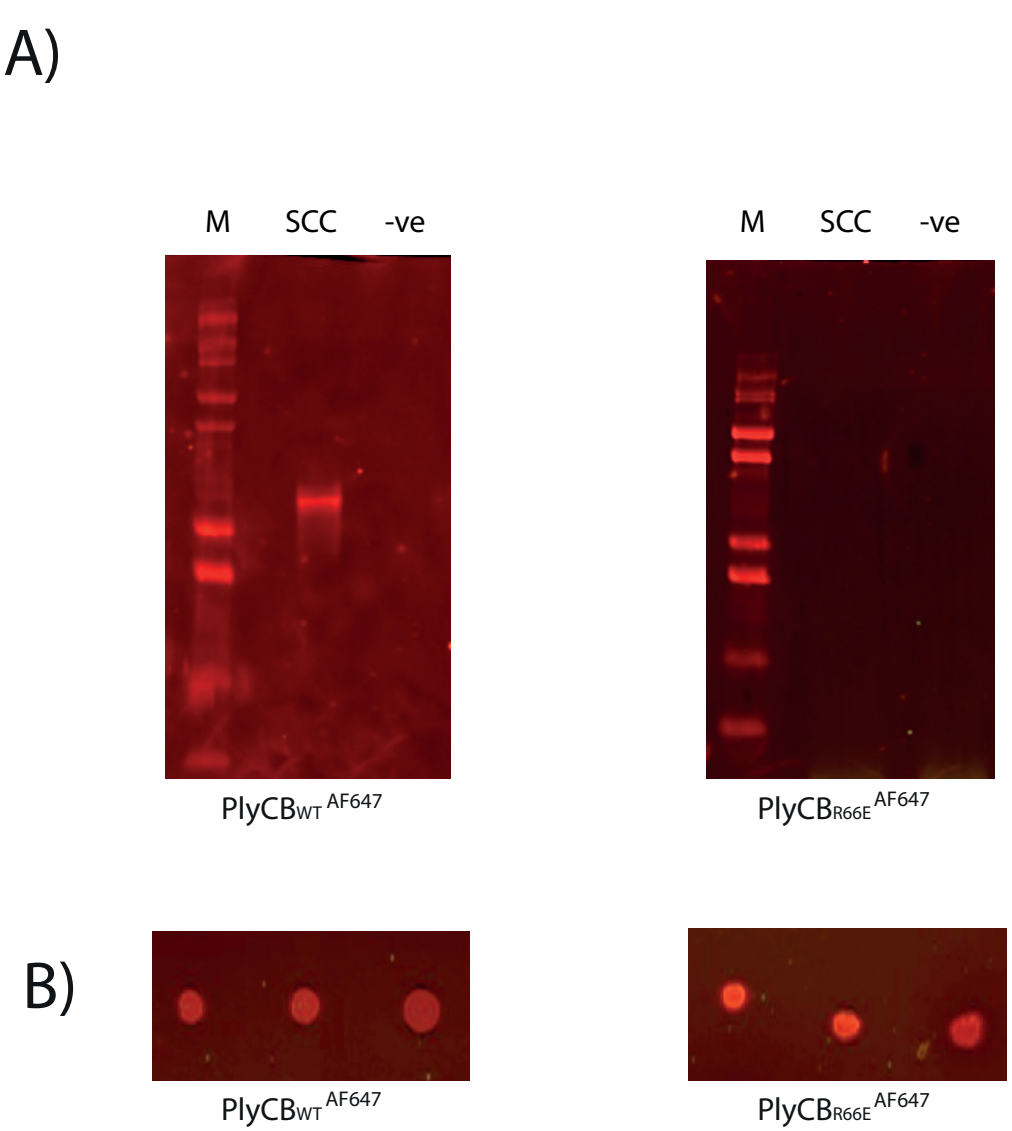
A)

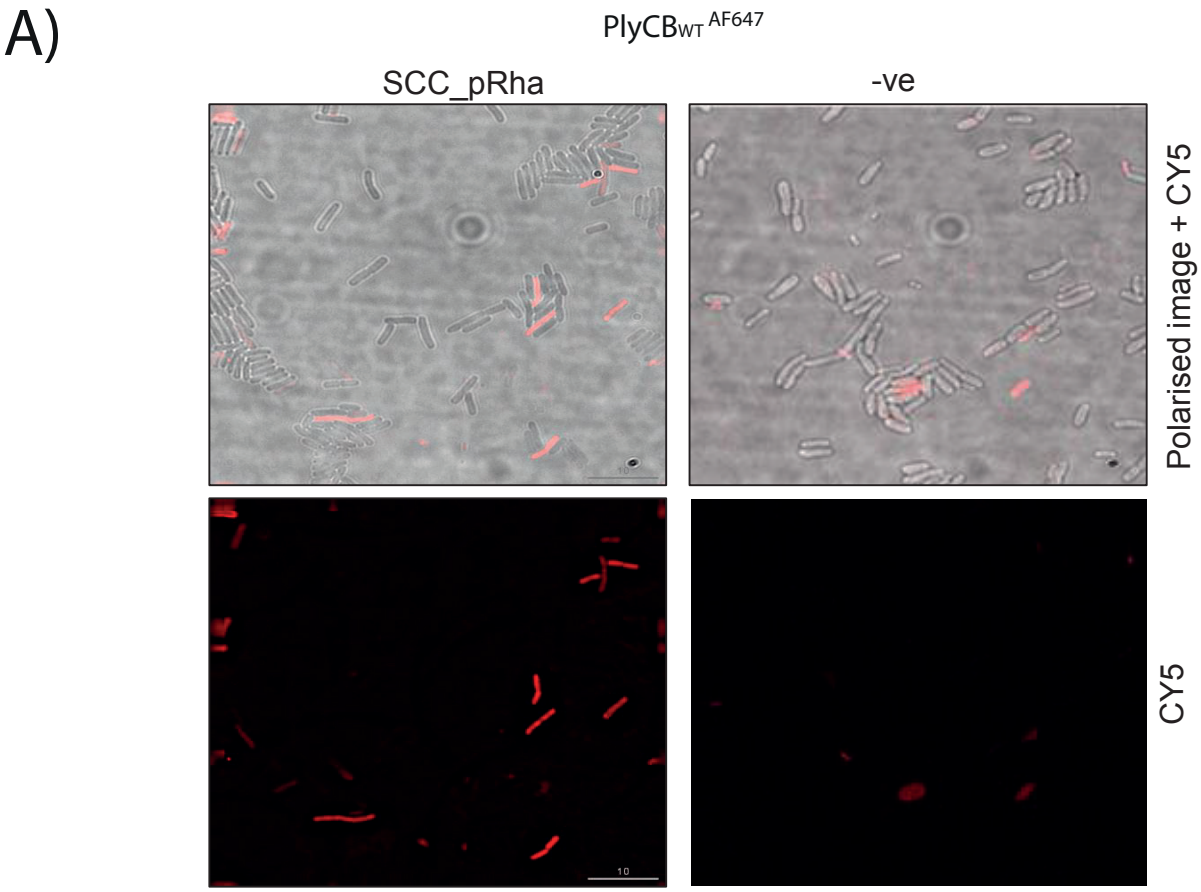


SF 1) Gel filtration purification of WT and R66E-PlyCB proteins. $PlyCB_{WT}$ (blue) and $PlyCB_{R66E}$ (red) proteins elute at a rentention volume of 12 mL. The PlyCB octamer has a theoretical mass of 63.8 kDa, with a monomeric mass of 7.9 kDa. Gel filtration standards are shown in kDa and include bovine thyroglobulin (670 kDa, void volume), bovine gamma-globulin (158 kDa), chicken ovalbumin (44 kDa), horse myoglobin (17 kDa), and vitamin B12 (1.35 kDa).



SF 2)

PlyCB binds to pRha in *E. coli* cell lysate whilst PlyCB$_{R66E}$ does not. A) Representative membrane blots showing that only PlyCB wildtype protein binds to total *E. coli* lysate when the pRha is expressed from the SCC gene cluster. PlyCB$_{R66E}$ fails to bind to pRha. **B)** Control dot blots showing that both PlyCB proteins are fluorescently labelled and detected when spotted directly onto nitrocellulose membrane (1:1000 dilution - identical to Figure A).



SF 3)

PlyCB binds to pRha expressing *E. coli* cells analysed using microscopic analysis: Representation of microscopic graph shows defined elongated PlyCB$_{WT}$ stained *E. coli* bacterial cells suggesting that PlyCB binds specifically to SCC_pRha cells but not to *E. coli* cells lacking pRha expression (-ve) detected using CY5 channel.