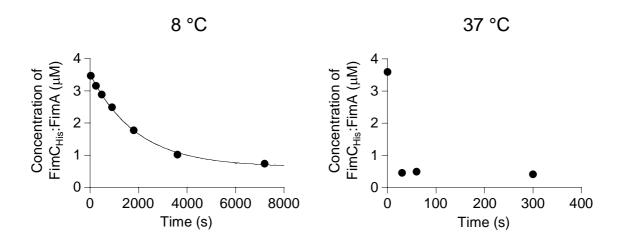
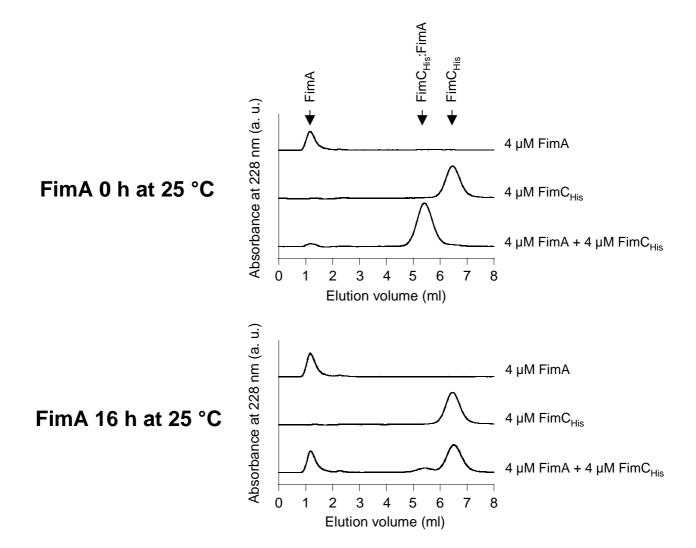


**Supplementary Fig 1** FimC<sub>His</sub> is an active variant of FimC. *Escherichia coli* strains expressing functional type 1 pili (e.g. strain W3110) bind to mannose displayed on the surface of yeast cells through FimH, the adhesive pilus subunit at the tip of each pilus (well 1). Strain W3110 $\Delta$ fimC does not express FimC and, as a consequence, bears no type 1 pili. Therefore, no agglutination is observed (well 2). Agglutination is restored if the same strain carries the plasmid pfimC (well 3) or pfimC<sub>His</sub> (well 4).

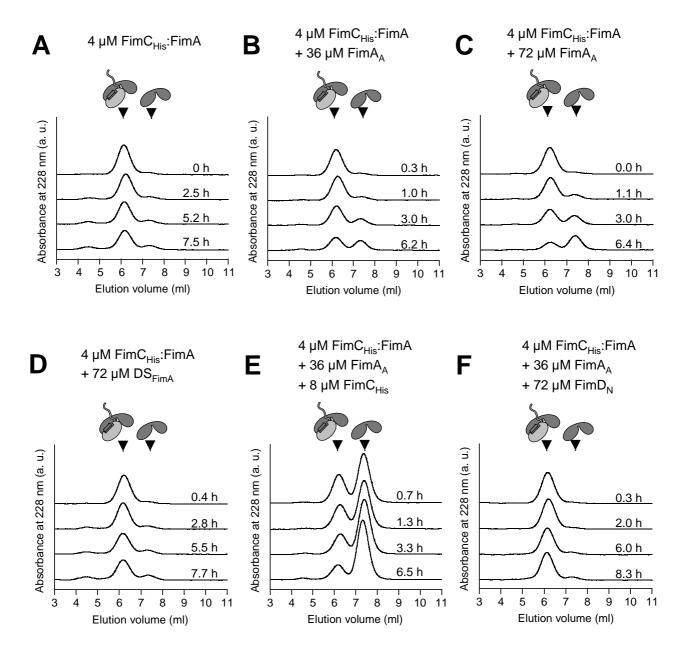
Plasmid pfim $C_{His}$  encodes FimC with a C-terminal (His)<sub>6</sub>-tag. It was obtained by site-directed mutagenesis (QuikChange, Stratagene) using pfimC (Hermanns, U. et al., *Biochemistry* **39**, 11564-11570 (2000)) as template. Strain W3110 $\Delta$ fimC (Nishiyama, M. et al., *EMBO J.* **24**, 2075-2086 (2005)) was transformed with pfimC or pfim $C_{His}$  and grown in LB at 25 °C to an OD<sub>600</sub> (optical density at 600 nm) of 0.5. The cells were centrifuged and resuspended in PBS to an OD<sub>600</sub> of 1.0. Of these suspensions, 9 volumes were mixed with 1 volume yeast in PBS (50 mg dry weight/ml) and agglutination was examined after 5 minutes. W3110 and W3110 $\Delta$ fimC without plasmid were included as positive and negative controls, respectively.



**Supplementary Fig 2** Dissociation dynamics of the FimC<sub>His</sub>:FimA complex at 8 °C and 37 °C. Reactions were performed at the indicated temperature as described for Fig 2. The initial concentrations were 4  $\mu$ M for the FimC<sub>His</sub>:FimA complex and 36  $\mu$ M for FimC. The analyses (solid lines) yield time constants ( $\tau$  values) of 1900±100 s and < 6 s at 8 °C and 37 °C, respectively.



Supplementary Fig 3 Formation of the FimC<sub>His</sub>:FimA complex from the individual, purified components. Momomeric FimA was obtained after dissociation of purified type 1 pili with the denaturant guanidinium chloride (8.2 M) and subsequent removal of the denaturant on a desalting column. The subunit FimA was then mixed with an equal molar amount of the chaperone FimC<sub>His</sub> and incubated at 25 °C for 1 minute (in 20 mM Tris/HCl, pH 8.0) before complex formation was analyzed by fast ion exchange chromatography. Upper panel: Freshly purified FimA associates with FimC<sub>His</sub> during the dead time of the experiment (~1 min). Lower panel: Only a small fraction of FimA formed the FimC<sub>His</sub>:FimA complex when freshly prepared FimA was incubated alone (at 25 °C in 20 mM Tris/HCl, pH 8.0) for 16 hours prior to mixing with FimC<sub>His</sub>. Consequently, monomeric FimA looses its ability to bind FimC<sub>His</sub> during storage in physiological buffer.



**Supplementary Fig 4** Representative elution profiles that were obtained during the kinetic analysis of DSE at 25 °C. (**A-F**) The FimC<sub>His</sub>:FimA complex (4  $\mu$ M) was incubated either alone, with FimA<sub>A</sub>, with FimA<sub>A</sub> and FimC<sub>His</sub>, or with DS<sub>FimA</sub>, a synthetic peptide corresponding to the donor strand of FimA. After the specified reaction times, samples were withdrawn and analyzed by fast ion exchange chromatography to measure the consumption of FimC<sub>His</sub>:FimA and the generation of FimC<sub>His</sub>. The initial protein concentrations and the elution volumes of the binary FimC<sub>His</sub>:FimA complex and monomeric FimC<sub>His</sub> are indicated.

Individual peak areas in the elution profiles (**A-E**) and (**F**) were quantified and translated into the protein concentrations shown in Fig 2A-E, and Fig 3A, respectively.