

1 Supplementary Information for

2 Motor properties of PilT-independent type 4 pilus retraction in
3 gonococci

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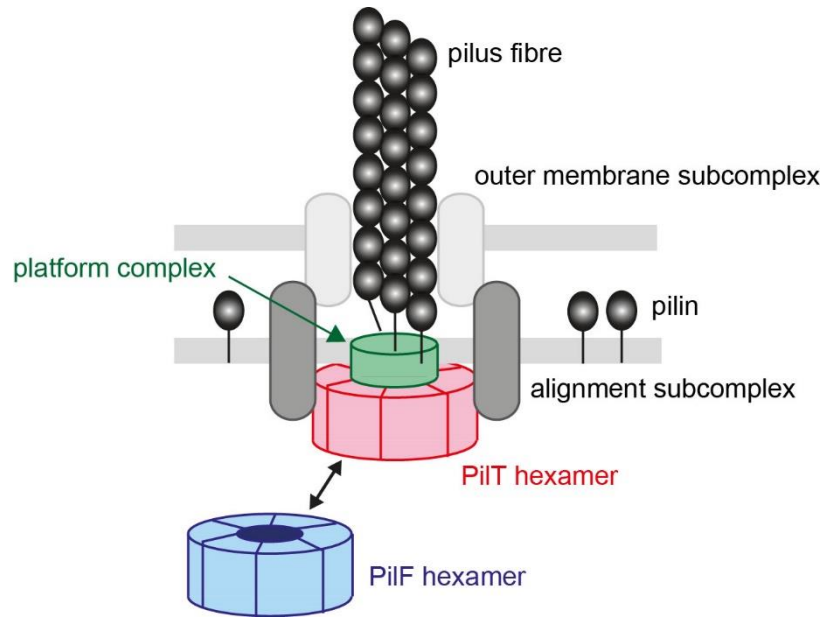
Strain	Genotype	Force Generation	Reference
$\Delta G4$ (Ng150)	<i>G4::aac</i>	yes	(1)
$\Delta pilT$ (Ng178)	<i>pilT::m-Tn3cm</i> <i>G4::aac</i>	yes	(2)
$\Delta pilT \Delta pilT2$ (Ng184)	<i>pilT2::kanR</i> <i>pilT::m-Tn3cm</i> <i>G4::aac</i>	yes	(3), this study
$\Delta pilT \Delta pilU$ (Ng185)	<i>pilU::ermC</i> <i>pilT::m-Tn3cm</i> <i>G4::aac</i>	yes	(3), this study
$\Delta pilT \Delta pilT2 \Delta pilU$ (Ng186)	<i>pilT2::kanR</i> <i>pilU::ermC</i> <i>pilT::m-Tn3cm</i> <i>G4::aac</i>	yes	(3), this study
$\Delta pilQ$ (Ng118)	<i>pilQ::m-Tn3cm</i> <i>igA1::P_{pilE} gfpmut3 ermC</i> <i>recA6ind(tetM)</i>	no	(1)

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8 **TABLE S1** Deletion of the gonococcal *pilT* paralogues does not inhibit T4P retraction.

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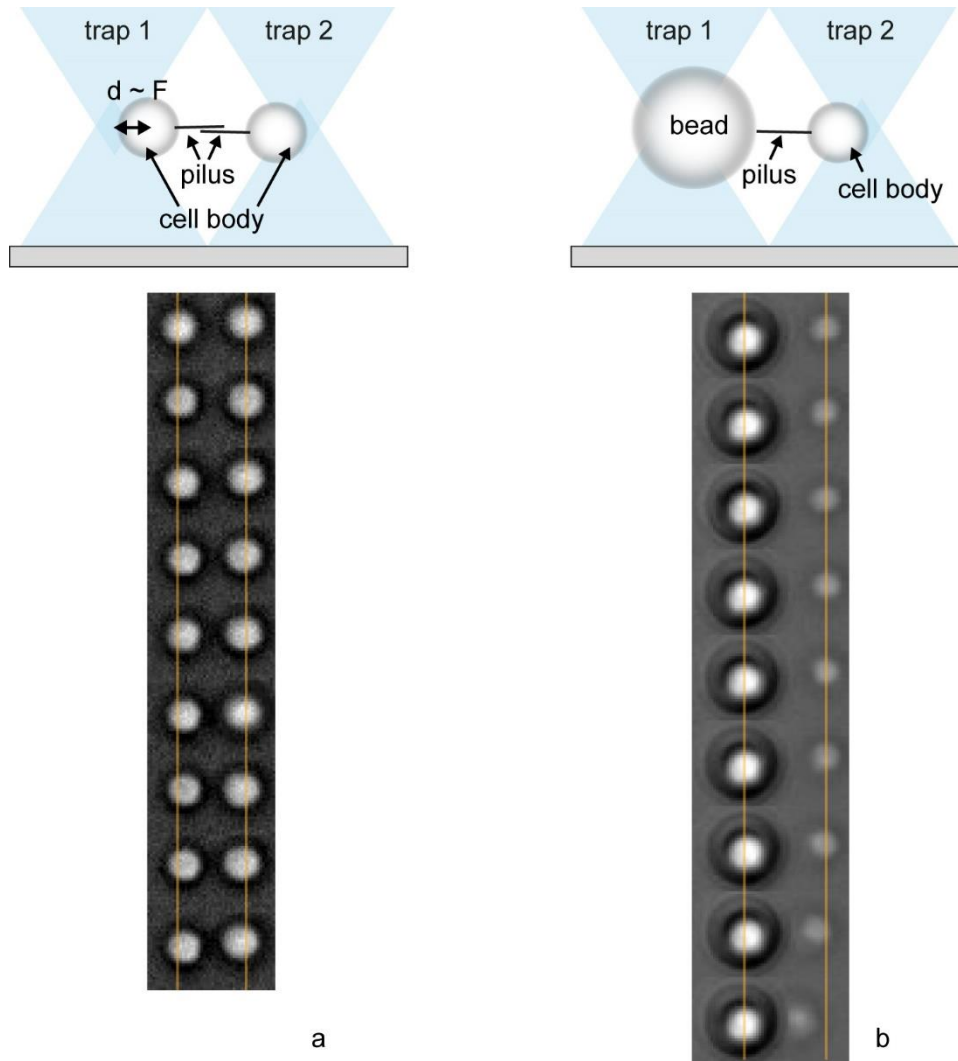
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12 **FIG S1** Hypothetical model of the T4P machine. The outer membrane subcomplex enables
 13 secretion of the pilus fibre. The alignment subcomplex is in contact with the outer membrane
 14 subcomplex and the motor subcomplex comprising the elongation ATPase PilF, the retraction
 15 ATPase PiT, and the platform complex formed by PilG. PilF and PiT form oblong, two-fold
 16 symmetric hexamers. Sequential ATP binding and hydrolysis causes a deformation wave
 17 running through the rings in opposite directions for PilF and PiT hexamers. This deformation
 18 wave couples to the platform complex, potentially causing platform rotation. Due to the helical
 19 shape of the pilus fibre, the direction of rotation determines whether pilins are inserted into or
 20 removed from the terminal end of the fibre.

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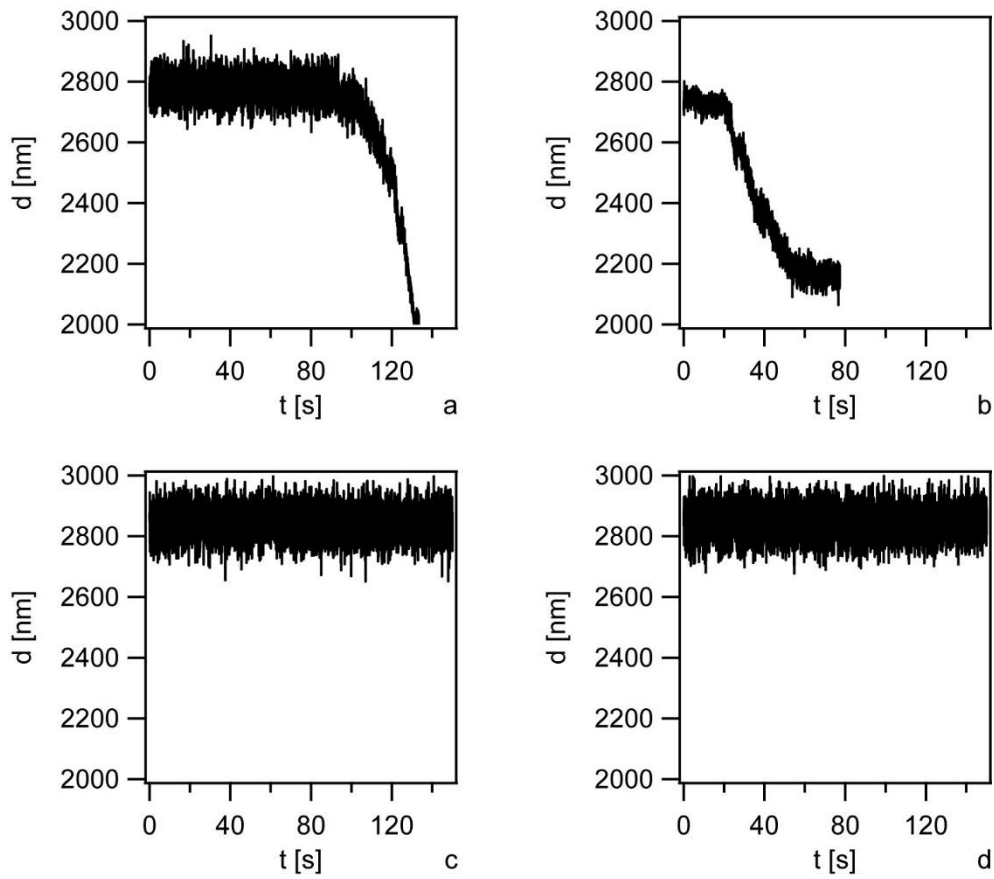


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23 **FIG S2** T4P retraction in $\Delta pilT$ strain (Ng178) in dual laser trap setup. a) A spherical
 24 gonococcus was trapped in each laser trap. Typical time lapse of two cells attracting each other.
 25 b) A bead was trapped in one trap and a spherical gonococcus in the other trap. Typical time
 26 lapse of a cells moving towards the bead. The laser trap exerts a stronger force on the bead than
 27 on the bacterium and therefore the deflection of the bacterium is higher. $\Delta t = 6.53$ s.

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31 **FIG S3** T4P retract in $\Delta pilT \Delta pilT2$ strain. T4P retraction was characterized in dual laser trap.
 32 Typical examples for deflection of bacteria in dual laser trap setup at a trap stiffness of 2 fN
 33 nm^{-1} . a) and b) $\Delta pilT \Delta pilT2$ (Ng184); c) and d) non-piliated $\Delta pilQ$ (Ng118).

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37 govern competition dynamics through positioning in bacterial colonies. *Scientific*
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- 39 2. **Welker A, Cronenberg T, Zollner R, Meel C, Siewering K, Bender N, Hennes M,**
40 **Oldewurtel ER, Maier B.** 2018. Molecular Motors Govern Liquidlike Ordering and
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43 gonococcal type IV pilus retraction and of twitching motility. *Mol Microbiol* **86**:857-
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