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Xylella fastidiosa pil-chp operon is involved in regulating key structural genes of both type I and IV pili

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XfPilG... L R V M V I D D S K T I R R T A E T L L K R E G C E - V V T A I D G F E A L A K I A D Q K S Q I I F V D I M M P R L D G 72
L + + V + D D T + R R L L K G V A D G + A L K + + D M P + D G
Ec CheY... L K F L V V D D F S T M R R I V R N L L K E L G F N N V E E A E D G V D A L N K L Q A G G Y G F V I S D W N M P N M D G 65

XfPilG... Y Q T C A L I K N N N L F K S T P V I M L S S - - - K D G L F D K A R G R V V G S E Q Y L T K P F T 119
+ I + + + P V + M + + + K + + A + G + Y + K P F T
Ec CheY... L E L L K T I R A D G A M S A L P V L M V T A E A K K E N I I A A A Q - - - A G A S G Y V V K P F T 112
  
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Fig. 1: Sequence comparison of *X. fastidiosa pilG* and *E. coli cheY*. Conserved phospho-accepting residue in bold lettering and boxed.

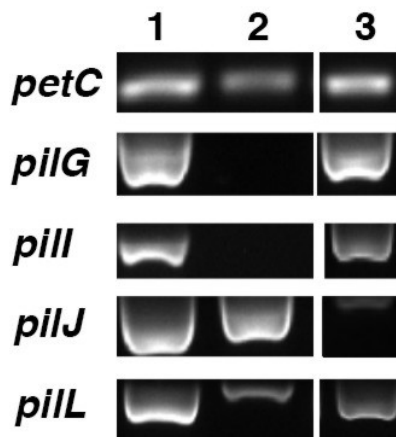


Fig. 2: *pilG*, *pilI*, *pilJ* and *pilL* gene expression in TM1, $\Delta pilG-I$, and $\Delta pilJ$ by reverse transcription (RT)-PCR. **1)** TM1 = *X. fastidiosa* wild-type strain Temecula 1, **2)** $\Delta pilG$ = TM1 deleted of *pilG* and also lacking expression of *pilI*, and **3)** $\Delta pilJ$ = TM1 deleted of *pilJ*. RT-PCR was performed twice with two independently extracted RNA samples for each strain and analyzed with 1 % agarose gel with same results. *petC* (PD1775) encodes a ubiquinol cytochrome C oxidoreductase cytochrome C1 subunit was included as the reference gene.



Fig. 3: Biofilm formation and cell-to-cell aggregation of TM1, $\Delta pilG-I$ and $\Delta pilJ$ cells in PD2 broth 3 d post-inoculation. Cells were grown for ~ 7 d on PWG plates, suspended in 30 mL of PD2 broth to an OD_{600} of ~ 0.013 and incubated at 28 °C, 185 rpm for 3 d. TM1 = *X. fastidiosa* wild-type strain Temecula 1; $\Delta pilG-I$ = TM1 deleted of *pilG* and also lacking expression of *pilI*; $\Delta pilJ$ = TM1 deleted of *pilJ*.

Table S 1

Primers

Purpose	Primer	Sequence (5' to 3')
<i>pilG</i> deletion	<i>pilGA</i>	CGCATCAGGATTATTTGCC
	<i>pilGB</i>	TTCGGCGCGCCGAATGCCAATATCCTCATGCGCA
	<i>pilGC</i>	TTCGGCGCGCCGAAGTACTGTTTCATCTGATGCG
	<i>pilGD</i>	GGCCGTCAACCAAAAACACA
<i>pilJ</i> deletion	<i>pilJA</i>	ACCTGACTGTTCATCTGATGCG
	<i>pilJB</i>	TTCGGCGCGCCGAATCTAAATATGCAAGACGGGACCG
	<i>pilJC</i>	TTCGGCGCGCCGAATGCTTCTCGGCTTGGAAAGGA
	<i>pilJD</i>	CGCAGCACGGATCTCGTTAA
<i>chpB</i> deletion	<i>chpBA</i>	TGGTTGTGTGATTGTTATTCTGG
	<i>chpBB</i>	TTCGTGCGCGCCGAACCTTACCTTGGACGCTATCATTCTC
	<i>chpBC</i>	CCTTAATTAAGGATTGGAACTACCTTCAATGAGCTATATC
	<i>chpBD</i>	CAAAACACATTCATCCCACCACATG
<i>chpC</i> deletion	<i>chpCA</i>	TGTGCTAGTGCTGGCTGGTAT
	<i>chpCB</i>	TTCGTGCGCGCCGAACCTTACATATCAATGTTTAGTCGGC
	<i>chpCC</i>	CCTTAATTAAGGATTCATGTGGTGGGATGAATGTGTTTT
	<i>chpCD</i>	GCTCCGATGAGGTAAAGCGAA
<i>pilG</i> mutant verification and RT-PCR	<i>pilG</i> verify- For	GTGCGCATGAGGATATTG
	<i>pilG</i> verify- Rev	CGGATGGCACTTAACAAC
<i>pilI</i> RT-PCR	<i>pilI</i> RT-PCR- For	ACCTGAATTTAGACAGGCTGCTGC
	<i>pilI</i> RT-PCR- Rev	TGCTGTGTAAGAGACTACGCCGTT
<i>pilJ</i> mutant verification and RT-PCR	<i>pilJ</i> verify- For	GGACGTTGATAATATCCTGG
	<i>pilJ</i> verify- Rev	CCTTAGCCGAAGTAGTATTG
<i>pilL</i> RT-PCR	<i>pilL</i> RT-PCR- For	AGGTGAGGAGGCGTTCTATAC
	<i>pilL</i> RT-PCR - Rev	CAGACAAGCATGAGACCAAGA
<i>chpB</i> mutant verification	<i>chpB</i> verify- For	CCCGAGTTGGCCTCAAATACT
	<i>chpB</i> verify- Rev	CCAGATTATCCCGAAGCACATA
<i>chpC</i> mutant verification	<i>chpC</i> verify- For	TGTGCTAGTGCTGGCTGGTAT
	<i>chpC</i> verify- Rev	GCTCCGATGAGGTAAAGCGAA
<i>dnaE</i> real-time RT-PCR	<i>dnaE</i> For	GCGAAGAACCATTCAATCA
	<i>dnaE</i> Rev	GTCCAGATGCGATACAGA
<i>dnaQ</i> real-time RT-PCR	<i>dnaQ</i> For	ATGGAGTTTCTAGCGGACAAGCCA
	<i>dnaQ</i> Rev	ATCACCAAAGTGTCAGCACGCTG
<i>mreD</i> real-time RT-PCR	<i>mreD</i> For	GCTGCGTAATACTTGGTTAT
	<i>mreD</i> Rev	TGACACTGGTAACGGAAG
<i>nuoA</i> real-time RT-PCR	<i>nuoA</i> For	TCAAGTAATAACGCACATCG
	<i>nuoA</i> Rev	ATTGATATTGATTGGCAGGTT
<i>petC</i> real-time RT-PCR	<i>petC</i> For	GTTGGATACTGTGCTTGTT
	<i>petC</i> Rev	CGATCACTCACATCATTACC
<i>fimA</i> real-time RT-PCR	<i>fimA</i> For	TGAACTAACAATTGCACGGCGGG
	<i>fimA</i> Rev	TACTGCCCAGCAATTGAACCTGC
<i>hxfB</i> real-time RT-PCR	<i>hxfB</i> For	TCATCGACATCAACGCCACATCA
	<i>hxfB</i> Rev	CGGTGACTTCAAAGCTGGCTTGTT
<i>pilA</i> real-time RT-PCR	<i>pilA</i> For	ATTATGTCGCCAGGTCTC
	<i>pilA</i> Rev	CCATCAGCCATACGGATT
<i>xadA</i> real-time RT-PCR	<i>xadA</i> For	TGATGTGAATGGACAGATGCGTCC
	<i>xadA</i> Rev	ATCGCACGCTGATAACCCAGAGAA