

Supplementary information for

Functional characterization of a putative DNA methyltransferase, EadM, in *Xanthomonas axonopodis* pv. *glycines* by proteomic and phenotypic analyses

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Supplementary information includes

Supplementary Figure 1-4

Supplementary Table 1

*Supplementary Table 2 and 3 were provided in the separated files

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EZP46960      MKNQLIHGDALTVLPTLPAASFDALITDPPYASGGVHAASRQQAPSSKYVRSG---LHDD 57
EadM          MKNQLLQGDALITLPTLEANSFDALITDPPYASGGLTAAARARPPSTKYCRDGG---HAD 57
WP_031421399 MKNQLLQGDALITLPTLEANSFDALITDPPYASGGLTAAARARSPSTKYCRDGG---HAD 57
SOD53705      MKNQLLKGDALITLPTLEACSF DALITDPPYASGGLTAGARQKPPSEKYVQGGKSALHAD 60
*****:*****:**** * *****: *.:* : ** * :.* * *

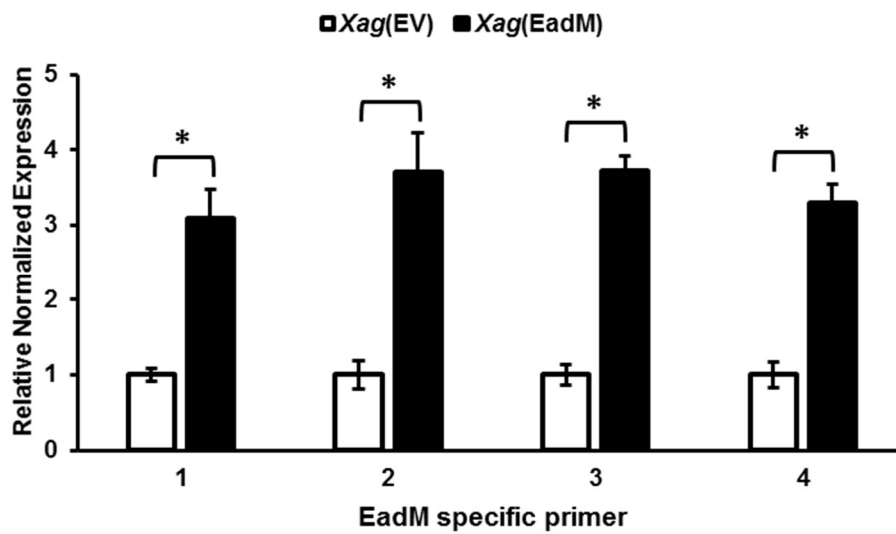
EZP46960      FVGDERDQRSHLAWMLWLAQC SRVLKDGAPVLLFTDWRQLPLTTDALQCAGFTWRGVAV 117
EadM          FVGDERDQRSHLKWMLWLSECARVLKDGAPVLLFTDWRQLPLTTDALQIAGFTWRGITV 117
WP_031421399 FVGDERDQRSHLKWMLWLSECARVLKDGAPVLLFTDWRQLPLTTDALQIAGFTWRGITV 117
SOD53705      FAGDERDQRSHLRWVMWLSECARLLKEGAPVCLFTDWRQLPLTTDALQCAGFTWRGITV 120
* ***** * * :*:*:*:*:*:***** *****: *

EZP46960      WDKTEGVRPQLGRFRNQAEYVWGSKGNMPLGRRAPVLPGVIRESVRKADKHHMTGKPTD 177
EadM          WDKTEGVRPQLGRFRNQAEYVWGSKGNMPLDRRAPVLPGVIREPVRKADKHHLTGKPTD 177
WP_031421399 WDKTEGVRPQLGRFRNQAEYVWGSKGNMPLDRRAPVLPGVIRESVRKADKHHLTGKPTD 177
SOD53705      WDKTEGVRPQLGRFRNQAEYVWGSKGSMP LQRRAPVLPGVIREPVRKADKHHMTGKPTA 180
*****:*****:*****.* ** *****: ***** *.:*****

EZP46960      LMRQLVRICEEGRILDPFAGSGTTLVAADQEGYSWTGIEMTEHYFGVAERRLPSE 233
EadM          LMRRLVRICESGGCVLDPFAGSGTTLVAAELEGYRWTGVEKTEHYATVAGSRICEI 233
WP_031421399 LMRQLVRICEAGGRVLDPFAGSGTTLIAAQLEGYNWTGVEITRHYASSAISRLSEL 233
SOD53705      LMRQLVRICEEGRVLDPFAGSGTTLVAAQLEGYSWLGCEMTHYVEVAGRLAAL 236
*.:***** * * :*****:*: ** * * * * * * * * * * * *

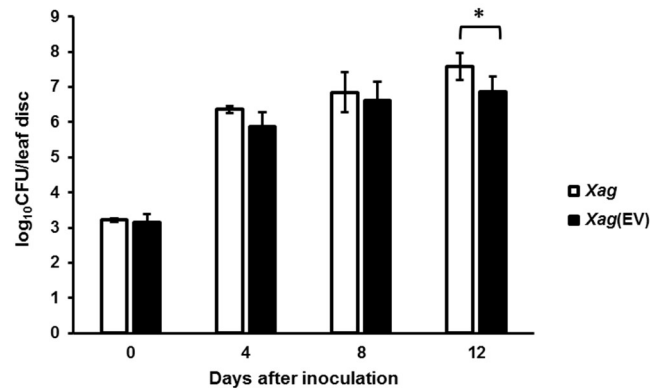
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Supplementary Figure 1. Sequence alignment of EadM and its homologs. The deduced amino acids of EadM were compared with its homologs in other bacteria belonging to the order Xanthomonadales, using the CLUSTALW program. EZP4690, WP_031421399, and SOD53705 are putative site-specific DNA methyltransferases in *Stenotrophomonas* sp. RIT309, *Xanthomonas euvesicatoria*, and *Pseudoxanthomonas wuyuanensis*, respectively. ‘*’, ‘:’, and ‘.’ indicate identical residues, conserved substitutions, and semi-conserved substitutions, respectively.

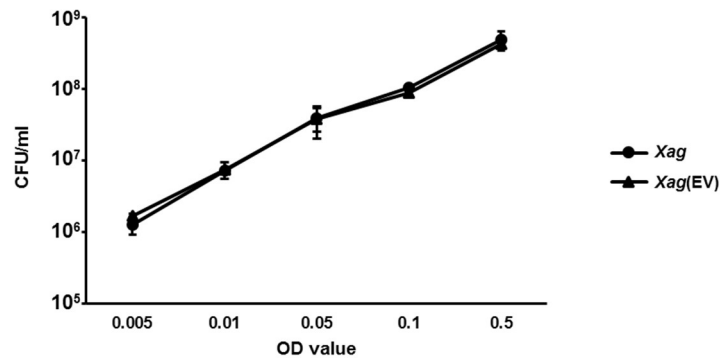


Supplementary Figure 2. Relative expression of *eadM* gene in *Xag* and *Xag(EV)*. *Xag* strains were harvested at 0.6 in an $OD_{600\text{ nm}}$ and total RNAs were extracted. The four *eadM*-specific primer sets were used for quantitative PCR. The 16S rRNA gene was used as a reference. Bars are the mean of three replicates \pm standard deviation. The asterisk indicates the statistical difference (t-test, $P < 0.05$).

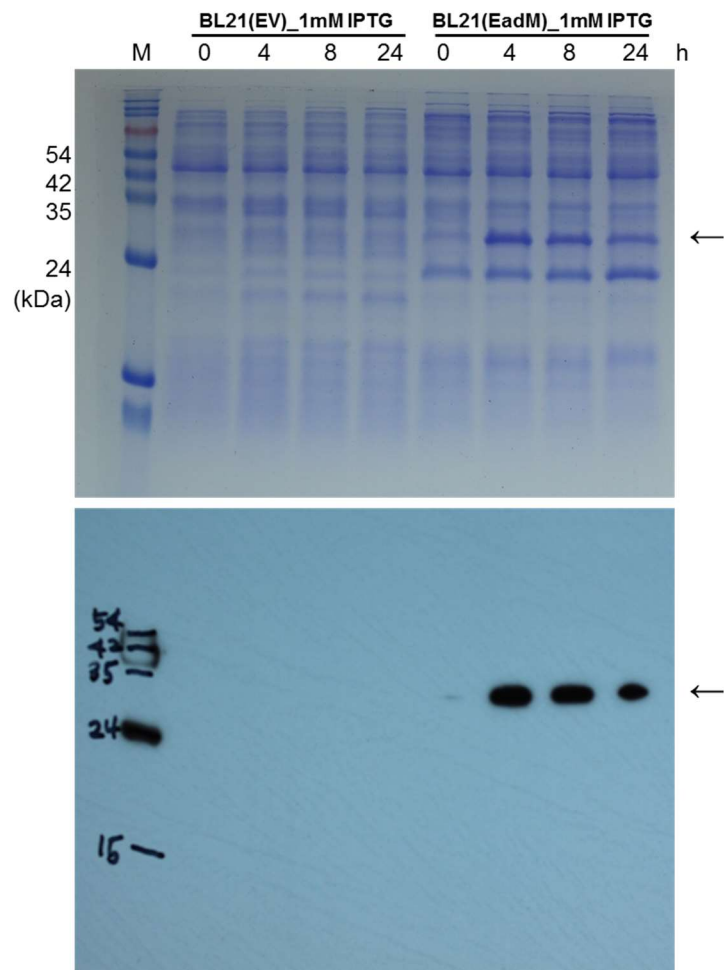
A



B



Supplementary Figure 3. Measurement of bacterial population in plants and viable cell numbers at different optical density values for *Xag* and *Xag*(EV). (A) Bacterial population of *Xag* (white), and *Xag*(EV) (black) were determined by the colony counting method at 0, 4, 8, and 12 days after inoculation. (B) The viable cell numbers of *Xag* (circle) and *Xag*(EV) (triangle) were quantified by the colony counting method at various OD values using a spectrophotometer. Different letters represent significant differences using the least significant difference test, $P \leq 0.05$. Error bars represent the mean of three biological replicates with the standard deviations. All experiments were repeated at least three times with three biological replicates.



Supplementary Figure 4. The full-length gel and blot used in Figure 6C. Arrows indicate EadM protein.

Supplementary Table 1. Proteins and peptide spectral matches between *Xag*(EV) and *Xag*(EadM) in three biological replicates from LC-MS/MS

Strains	1st		2nd		3rd		shared proteins in 3 biological replicates
	protein	PSM	protein	PSM	protein	PSM	
<i>Xag</i> (EV)	1100	47408	1077	47425	1087	47415	1029
<i>Xag</i> (EadM)	1140	50892	1148	50910	1154	50923	1071