

Supporting Information

Seeger *et al.*,

**The biosynthetic genes for prenylated phenazines are located
at two different chromosomal loci of *Streptomyces
cinnamomensis* DSM 1042**

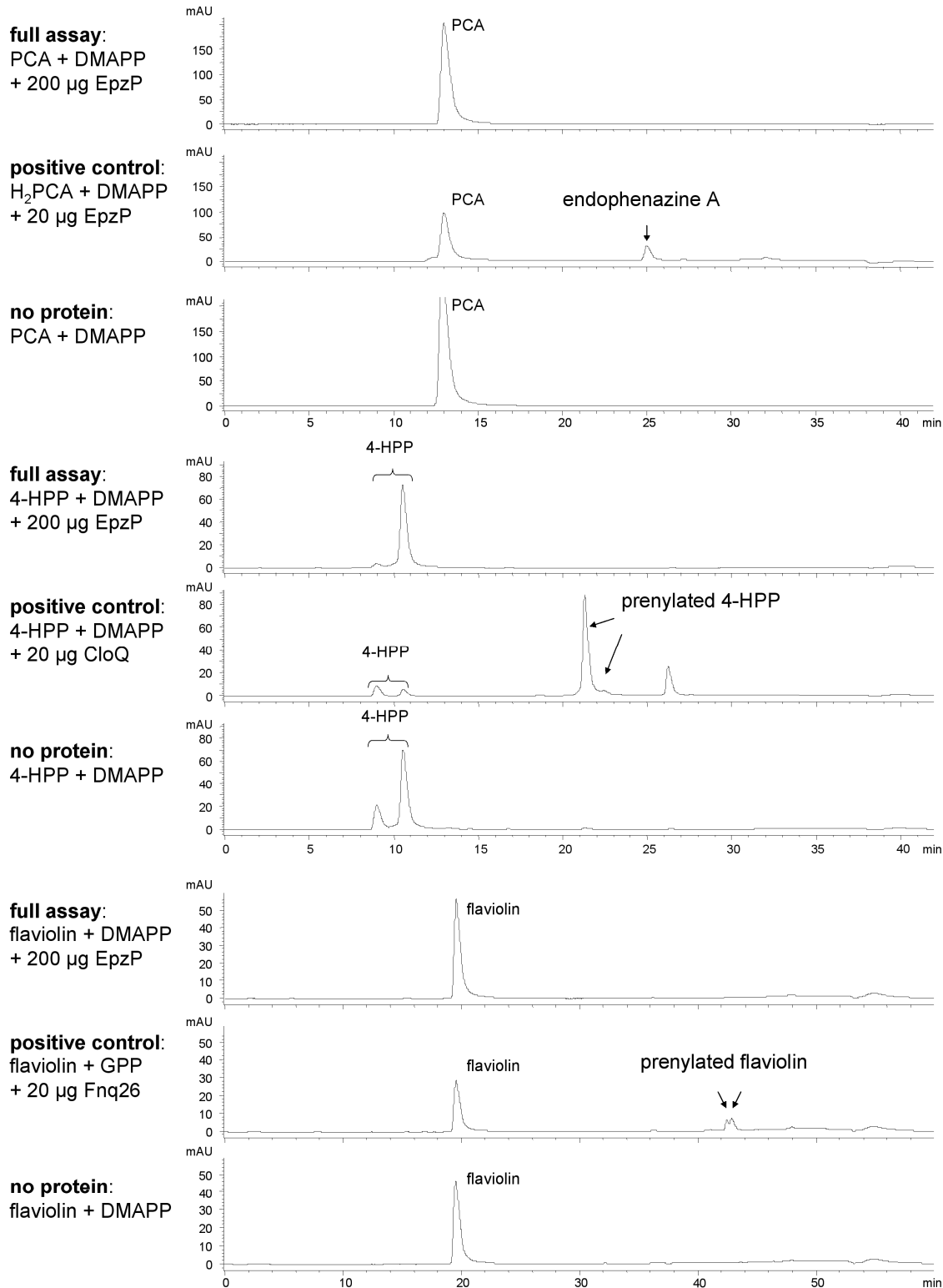


Figure S1: Activity of EpzP with different substrates.

EpzP prenylated 9,10-dihydro-phenazine-1-carboxylic acid (H_2 PCA), but not phenazine-1-carboxylic acid (PCA), 4-hydroxyphenylpyruvate (4-HPP) or flaviolin. The prenylation of flaviolin with GPP by Fnq26 results in two structurally different prenylated products (Haagen et al., 2007). 4-HPP and its prenylated product show two peaks due to keto-enol tautomerism.

Detection in HPLC was carried out at 365 nm for PCA and endophenazine A, 308 nm for 4-HPP and its products, and 306 nm for flaviolin and its products. All assays contained 0.4 mM aromatic substrate and 0.4 mM isoprenoid substrate and were incubated for 30 min at 30°C. EpzP assays were performed with 500 mM NaCl and 100 mM TAPS pH 7.5, the assay with CloQ with 2 mM MgCl₂ and 75 mM Tris-HCl pH 7.5 and the assay with Fnq26 with 2 mM MgCl₂ and 100 mM TAPS pH 8.5, respectively.

endophenazine A formation		
	(nmol)	(%)
0 mM NaCl, 0 mM MgCl ₂	6.2	100
500 mM NaCl, 0 mM MgCl ₂	6.2	100
500 mM NaCl, 2 mM MgCl ₂	6.7	108
500 mM NaCl, 10 mM MgCl ₂	6.9	111
500 mM NaCl, 20 mM MgCl ₂	4.1	66
0 mM NaCl, 0 mM MgCl ₂ , 10 mM EDTA	4.8	77

Table S1: Influence of MgCl₂ and EDTA on the activity of EpzP.

Endophenazine A formation was assayed with 0.4 mM H₂PCA, 1 mM DMAPP, 10 µg EpzP in 100 mM TAPS pH 7.5 (100 µl, 30 min, 30°C).

References

Haagen, Y., Unsöld, I., Westrich, L., Gust, B., Richard, S. B., Noel, J. P., and Heide, L. (2007) A soluble, magnesium-independent prenyltransferase catalyzes reverse and regular C-prenylations and O-prenylations of aromatic substrates, *FEBS Lett.* **581**: 2889-2893.