

### CYP199A4 Supplementary data

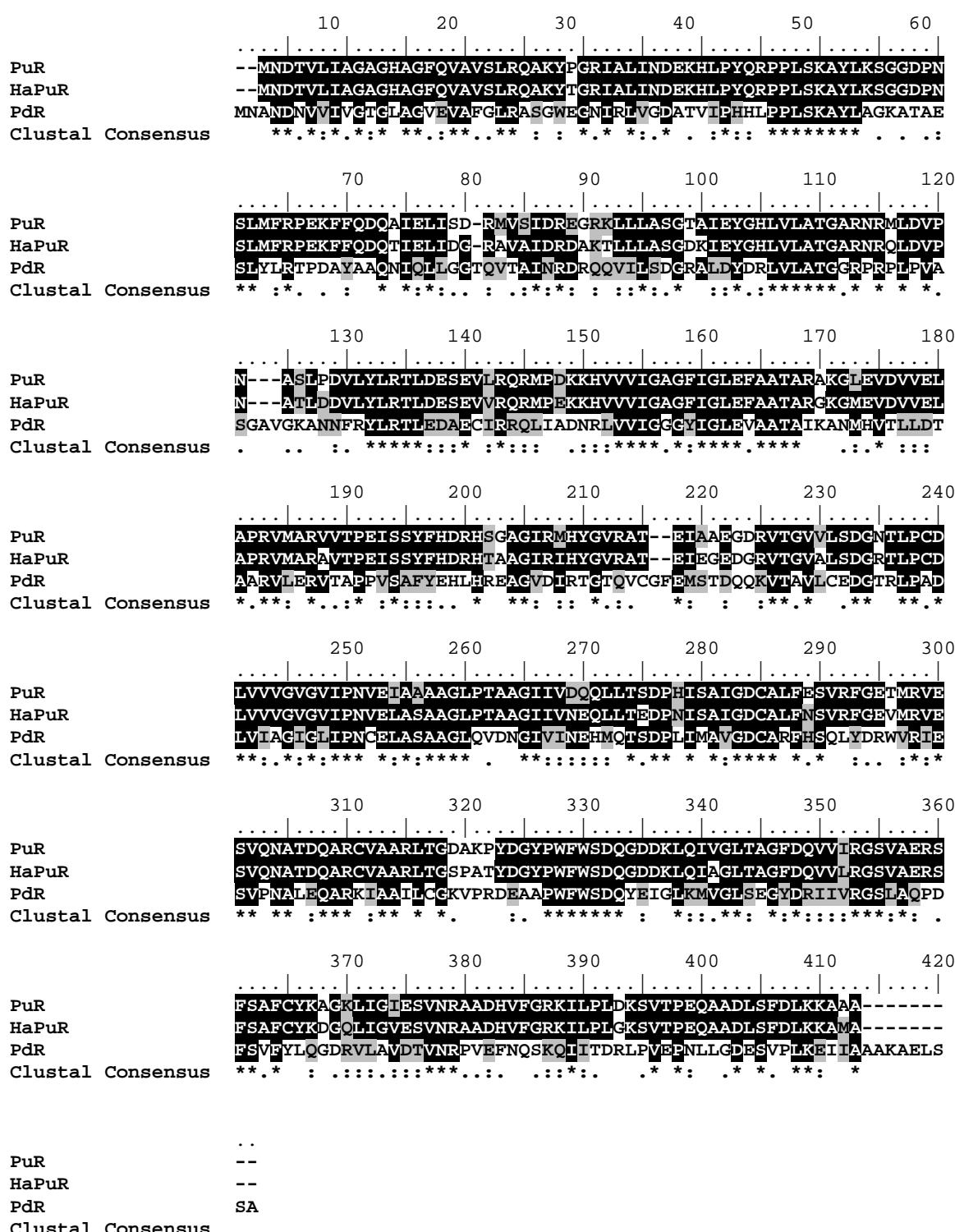
**Table S1.** Sequence similarities and alignment between the Class I CYP systems from *R. palustris* HaA2 and *R. palustris* CGA009.

CYP199A4/HaPux/HaPuR		CYP199A2/Pux/PuR		
		Identities	Positives	Gaps
CYP199A4		356/412 (86%)	383/412 (92%)	2/412 (0%)
HaPux		74/106 (69%)	91/106 (85%)	1/106 (0%)
HaPuR		357/405 (88%)	374/405 (92%)	0/405 (0%)

**Table S2.** Spectral properties of CYP199A2, HaPux and HaPuR from *R. palustris* HaA2.

	Absorbance (nm)	$\epsilon$ (mM <sup>-1</sup> cm <sup>-1</sup> )
<b>CYP199A4</b>	279	48.6
	357	27.9
	419	103.1
	538	9.4
	569	10.7
<b>HaPux</b>	280	18.7
	316	14.1
	416	11.2
	457	9.1
<b>HaPuR</b>	172	65.3
	383	9.6
	455	10.0
	482	8.2

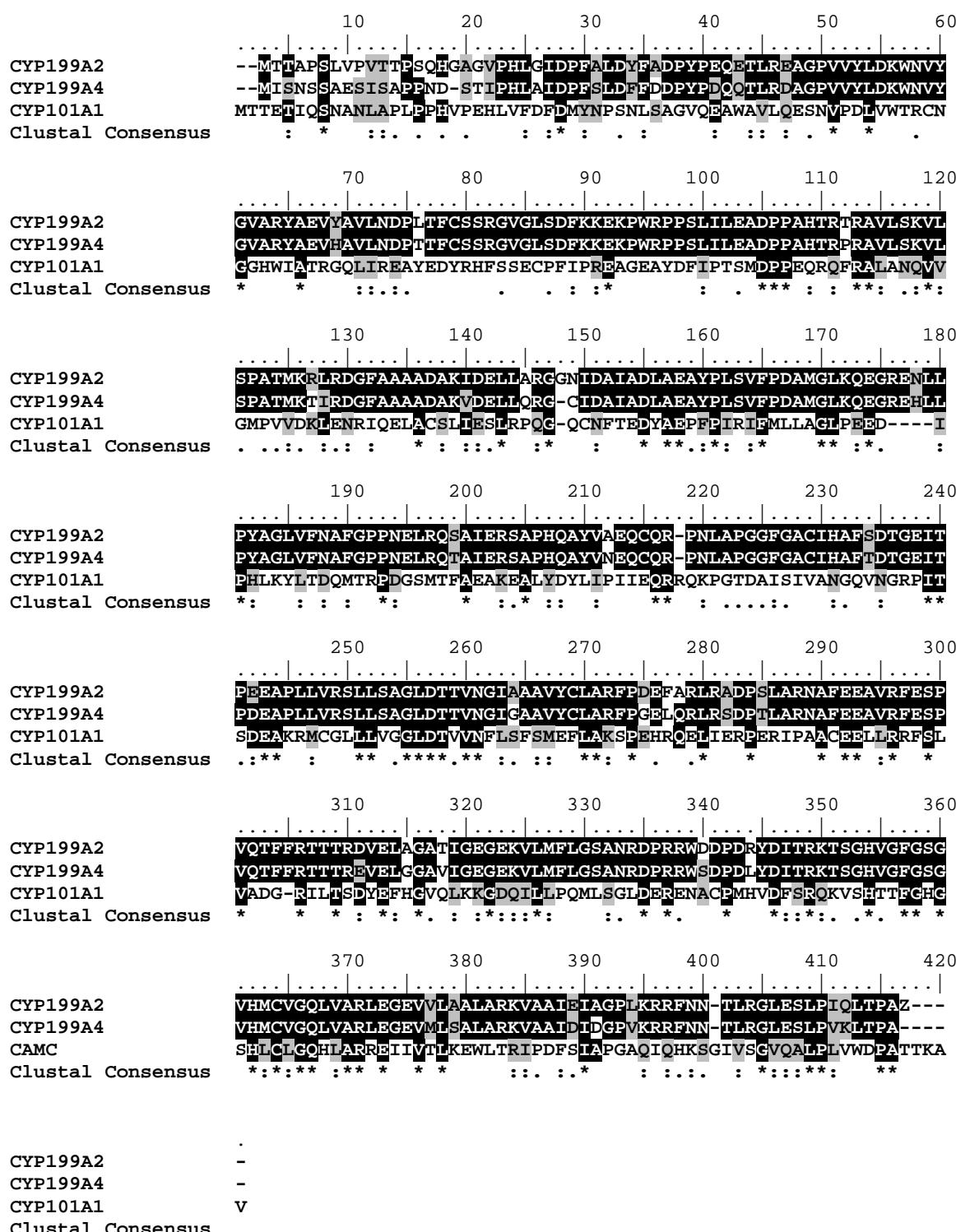
**Figure S1.** Sequence alignment of HaPuR, PuR and PdR from *R. palustris* HaA2, *R. palustris* CGA009 and *P. putida* respectively.



**Figure S2.** Sequence alignment of HaPux, Pux and Pdx from *R. palustris* HaA2, *R. palustris* CGA009 and *P. putida* respectively.

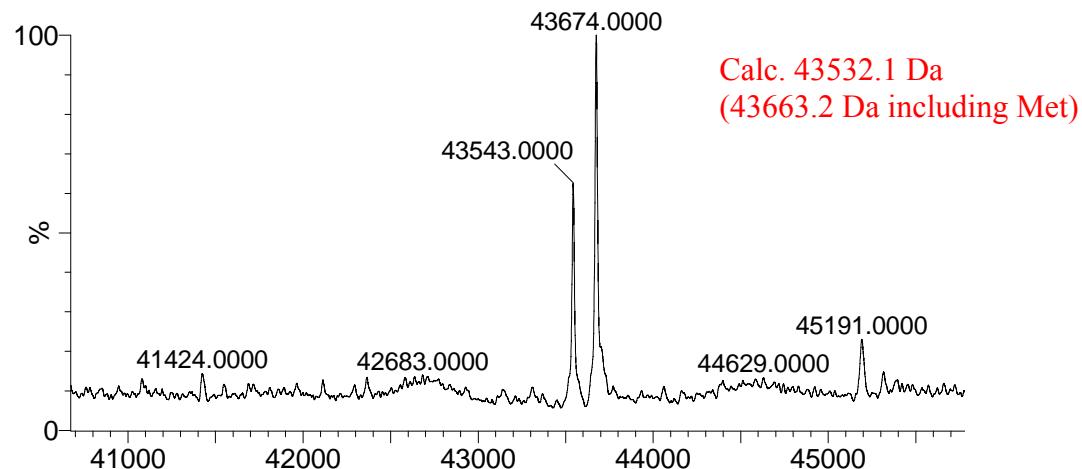
10 20 30 40 50 60  
Pux MPSITFILPDGERRTTEAAVGDTAMYAALSGLLDGVVAECGGNAVCATCHVYVEHG-LEK  
HaPux MPSITFIHPDGRSEIVDAAIIGDSAMEAAINHGIDSIVAECGGNAVCATCHVYVDDLWLAK  
Pdx MSKVYYVSHDGTRRELVDAGVSLMOAAAVSNGIYDIVGDCGGSASCATCHVYVNEAFTDK  
Clustal Consensus \*...: :: \* . : .. \* : \* \*\*: .. : .. : \*\*\* . \*\*\*\*\*: . \*  
  
70 80 90 100  
Pux LPAVAADEDDLLLDGTAAERLPNSRLSCQIKLSSLDGLILRIPDRQV  
HaPux LPPVDANEEDDLLLDGTASDRLPNSRLSCQIKIAPELDGLVLRIPEROT  
Pdx VPAANEREIGMLECVTAELKPNSRLCCQIIMTPELDGIVVDVPDRQW  
Clustal Consensus : \* .: \* : : \*\*\*\*\*.\*\*\* :: : \*\*\*: : : \*:\*\*

**Figure S3.** Sequence alignment of CYP199A4, CYP199A2 and CYP101A1 from *R. palustris* HaA2, *R. palustris* CGA009 and *P. putida* respectively.

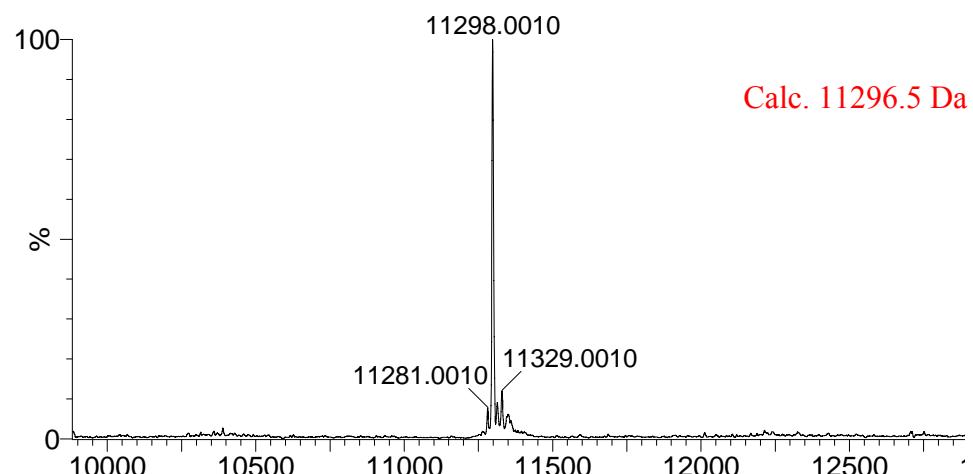


**Figure S4.** Electrospray mass spectra of (a) HaPuR (b) HaPux and (c) CYP199A4.

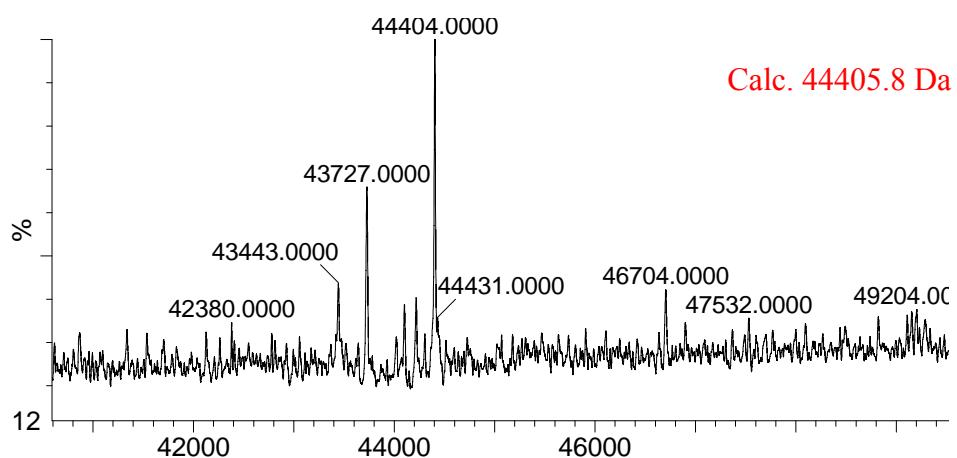
(a)



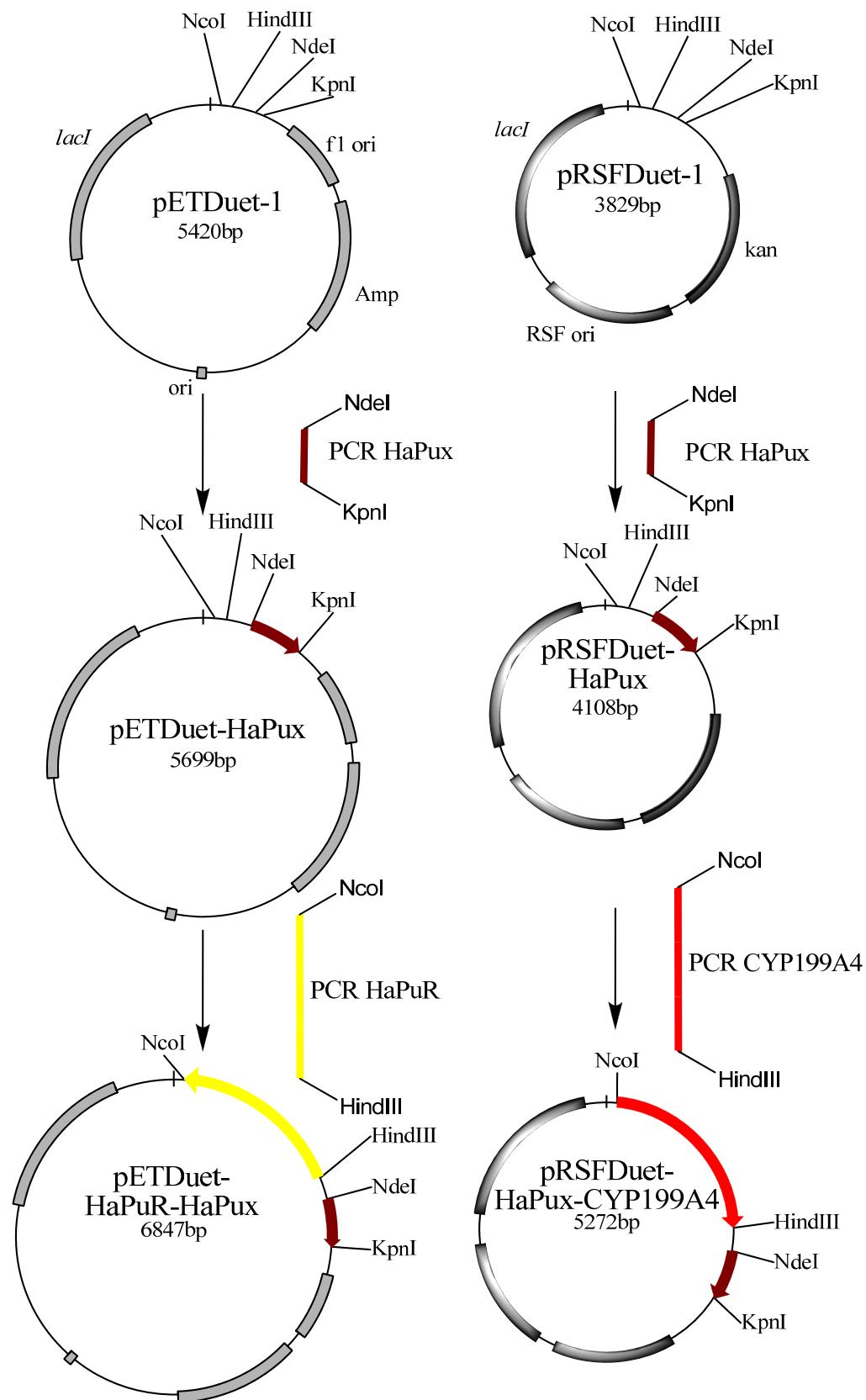
(b)



(c)



**Figure S5.** Construction of the plasmids of the whole cell oxidation system.



**Figure S6.** Example of NADH turnover data using CYP199A4 (0.5  $\mu$ M), HaPux and HaPuR in a 1:10:1 ratio.

