

## Supplementary Information

**Table S1.** Table of measured enzyme activities.

<b>Construct</b>	<b>Assay Description</b>	<b>Activity</b>
pRC41-4 (WT)	crude +MV evolution assay	4.0 ± 0.8 (nmol H <sub>2</sub> ) mg <sup>-1</sup> ·min <sup>-1</sup>
	crude -MV evolution assay	0.018 ± 0.004 (nmol H <sub>2</sub> ) mg <sup>-1</sup> ·min <sup>-1</sup>
pCM012 (G1)	crude +MV evolution assay	9.8 ± 1.1 (nmol H <sub>2</sub> ) mg <sup>-1</sup> ·min <sup>-1</sup>
	crude -MV evolution assay	0.14 ± 0.03 (nmol H <sub>2</sub> ) mg <sup>-1</sup> ·min <sup>-1</sup>
pIY087 (I1)	crude +MV evolution assay	10.1 ± 1.7 (nmol H <sub>2</sub> ) mg <sup>-1</sup> ·min <sup>-1</sup>
	crude -MV evolution assay	0.14 ± 0.05 (nmol H <sub>2</sub> ) mg <sup>-1</sup> ·min <sup>-1</sup>
pIY088 (I2)	crude +MV evolution assay	11.0 ± 3.1 (nmol H <sub>2</sub> ) mg <sup>-1</sup> ·min <sup>-1</sup>
	crude -MV evolution assay	0.25 ± 0.05 (nmol H <sub>2</sub> ) mg <sup>-1</sup> ·min <sup>-1</sup>
pIY089 (G2)	crude +MV evolution assay	9.4 ± 0.9 (nmol H <sub>2</sub> ) mg <sup>-1</sup> ·min <sup>-1</sup>
	crude -MV evolution assay	0.67 ± 0.19 (nmol H <sub>2</sub> ) mg <sup>-1</sup> ·min <sup>-1</sup>
pIY099 (Δ15)	crude +MV evolution assay	9.9 ± 4.1 (nmol H <sub>2</sub> ) mg <sup>-1</sup> ·min <sup>-1</sup>
	crude -MV evolution assay	0.18 ± 0.06 (nmol H <sub>2</sub> ) mg <sup>-1</sup> ·min <sup>-1</sup>
pIY098 (Δ22)	crude +MV evolution assay	10.0 ± 1.8 (nmol H <sub>2</sub> ) mg <sup>-1</sup> ·min <sup>-1</sup>
	crude -MV evolution assay	0.17 ± 0.02 (nmol H <sub>2</sub> ) mg <sup>-1</sup> ·min <sup>-1</sup>
pIY107 (G1)	Tandem-Purification +MV assay	2.09 ± 0.36 (μmol H <sub>2</sub> ) mg <sup>-1</sup> ·min <sup>-1</sup>
	Tandem-Purification -MV assay	12.1 ± 2.2 (nmol H <sub>2</sub> ) mg <sup>-1</sup> ·min <sup>-1</sup>
	<b>Relative -MV/+MV activity</b>	<b>5.8 ± 1.4 × 10<sup>-3</sup></b>
pIY108 (G2)	Tandem-Purification +MV assay	0.40 ± 0.05 (μmol H <sub>2</sub> ) mg <sup>-1</sup> ·min <sup>-1</sup>
	Tandem-Purification -MV assay	9.2 ± 2.2 (nmol H <sub>2</sub> ) mg <sup>-1</sup> ·min <sup>-1</sup>
	<b>Relative -MV/+MV activity</b>	<b>2.3 ± 0.6 × 10<sup>-2</sup></b>
pIY109 (G1-Fd)	Tandem-Purification +MV assay	0.85 ± 0.05 (μmol H <sub>2</sub> ) mg <sup>-1</sup> ·min <sup>-1</sup>
	Tandem-Purification -MV assay	7.8 ± 0.9 (nmol H <sub>2</sub> ) mg <sup>-1</sup> ·min <sup>-1</sup>
	<b>Relative -MV/+MV activity</b>	<b>9.1 ± 1.2 × 10<sup>-3</sup></b>
pIY110 (G2-Fd)	Tandem-Purification +MV assay	0.70 ± 0.07 (μmol H <sub>2</sub> ) mg <sup>-1</sup> ·min <sup>-1</sup>
	Tandem-Purification -MV assay	37.1 ± 9.6 (nmol H <sub>2</sub> ) mg <sup>-1</sup> ·min <sup>-1</sup>
	<b>Relative -MV/+MV activity</b>	<b>5.3 ± 1.4 × 10<sup>-2</sup></b>
pIY094 (G2, E217Q) *	crude +MV evolution assay	2.0 ± 0.2 (nmol H <sub>2</sub> ) mg <sup>-1</sup> ·min <sup>-1</sup>
pIY038 (T7, G1) *	crude +MV evolution assay	8.5 ± 1.4 (nmol H <sub>2</sub> ) mg <sup>-1</sup> ·min <sup>-1</sup>
	crude -MV evolution assay	0.25 ± 0.06 (nmol H <sub>2</sub> ) mg <sup>-1</sup> ·min <sup>-1</sup>
	crude +MV evolution assay (included pACYC empty vector)	5.6 ± 0.2 (nmol H <sub>2</sub> ) mg <sup>-1</sup> ·min <sup>-1</sup>
pIY042 (T7, ALL+) *	crude +MV evolution assay (included pACYC empty vector)	0.15 ± 0.05 (nmol H <sub>2</sub> ) mg <sup>-1</sup> ·min <sup>-1</sup>
pIY062 (T7, DDE) *	crude +MV evolution assay	6.8 ± 0.86 (nmol H <sub>2</sub> ) mg <sup>-1</sup> ·min <sup>-1</sup>
	crude -MV evolution assay	0.34 ± 0.05 (nmol H <sub>2</sub> ) mg <sup>-1</sup> ·min <sup>-1</sup>
pIY063 (T7, DDE, E261K, I1) *	crude +MV evolution assay	6.1 ± 0.6 (nmol H <sub>2</sub> ) mg <sup>-1</sup> ·min <sup>-1</sup>
	crude -MV evolution assay	0.17 ± 0.02 (nmol H <sub>2</sub> ) mg <sup>-1</sup> ·min <sup>-1</sup>

**Table S1. Cont.**

<b>Construct</b>	<b>Assay Description</b>	<b>Activity</b>
pIY068 (T7, DDE, E261K, I2) *	crude +MV evolution assay	$7.6 \pm 4.2$ (nmol H <sub>2</sub> ) mg <sup>-1</sup> ·min <sup>-1</sup>
	crude -MV evolution assay	$0.54 \pm 0.08$ (nmol H <sub>2</sub> ) mg <sup>-1</sup> ·min <sup>-1</sup>
pIY069 (T7, DDE, E261K, G2) *	crude +MV evolution assay	$7.1 \pm 2.2$ (nmol H <sub>2</sub> ) mg <sup>-1</sup> ·min <sup>-1</sup>
	crude -MV evolution assay	$1.2 \pm 0.1$ (nmol H <sub>2</sub> ) mg <sup>-1</sup> ·min <sup>-1</sup>
pIY070 (T7, DDE, I1) *	crude +MV evolution assay	$11.0 \pm 1.1$ (nmol H <sub>2</sub> ) mg <sup>-1</sup> ·min <sup>-1</sup>
	crude -MV evolution assay	$0.33 \pm 0.1$ (nmol H <sub>2</sub> ) mg <sup>-1</sup> ·min <sup>-1</sup>
pIY071 (T7, DDE, G2) *	crude +MV evolution assay	$8.9 \pm 1.7$ (nmol H <sub>2</sub> ) mg <sup>-1</sup> ·min <sup>-1</sup>
	crude -MV evolution assay	$0.65 \pm 0.05$ (nmol H <sub>2</sub> ) mg <sup>-1</sup> ·min <sup>-1</sup>

Asterisked constructs are not presented in figures but are referenced in the main text.

**Table S2. List of Primers used in this study.**

<b>Name</b>	<b>Description</b>	<b>Sequence</b>
IY170HynSF	forward primer upstream of the BamHI enzyme site in hynS	GTATTATTGCCATCGGTACC
IY171HynSR	reverse primer downstream of the AgeI enzyme site in hynL	CGAGTGATAGGATCGACGAC
IY100p18F	pUC18 Forward primer	AAAAGCAGCTTTTACCAAGCCCGTAATCATG GTCATAGCTGTTTC
IY100p18R	pUC18 Reverse primer	GAGATCAGGGAAACGCTTAAACCACTGGCCG TCGTTTTAC
IY102FdR1PF	D216R, E217K Forward	GTTTAAGCGTTTCCCTGATCTCCGGAAGTTAC AACGCCCACTCGC
IY103FdR1PR	D216R, E217K Reverse	GAGATCAGGGAAACGCTTAAAC
IY104FdR2PF	E227K, D231R Forward	GCCCACTCGCTTTTTTTGGTAAGAGCATCTGC CGGAGCATCCATGACAGATGTTATCG
IY105FdR2PR	E227K, D231R Reverse	ACCAAAAAAAGCGAGTGGGC
IY106FdR3PF	E240K Forward	GTTATCGACGCCATTTTTTAAGCAACGTAA ATTTGCAAAATCG
IY107FdR3PR	E240K Reverse	AAAAAATGGGCGTCGATAAC
IY108FdR4PF	E261K Forward	CTTGGTTGTAAAGGGCCC
IY109FdR4PR	E261K Reverse	GGGCCCTTACAACCAAGCTTGAACAAACAC CACCCATT
IY110FdR5PF	E268K Forward	ACCTTTAACGCATGTGCAAC
IY111FdR5PR	E268K Reverse	GTTGCACATGCGTTAAAGGTCTTGGGCCCTTT ACAACCAAG
IY112FdR6PF	E297K, D299R, D302R Forward	AAAAGCAGCTTTTACCAAGCC

Table S2. *Cont.*

Name	Description	Sequence
IY113FdR6PR	E297K, D299R, D302R Reverse	GGCTTGGTAAAAGCTGCTTTTCCGCCAGAAC CGGGGCTTAGAGCAACCAAGACACGG
IY600DDDEF	ΔDDE forward	GGGGCAAAAAATGGGTGGTG
IY601DDDER	ΔDDE reverse	CACCACCCATTTTTTGCCCCAAACGATTTTGC AAATTTACGTTG
IY602Mut2F	ΔDDE I1 (E261K E268Q) forward	AAACTTGGTTGTAAAGGGCCCCAAACCTTTA ACGCATGTGCAACGGT
IY603Mut2R	ΔDDE I1 (E261K E268Q) reverse	TTGGGGCCCTTTACAACCAAGTTTGAACAAA CACCACCCATTTTTTG
IY664Rd2F	I1 (E261Q E268Q) forward	CAACTTGGTTGTAAAGGGCCCCAAACCTTTA ACGCATGTGCAACGGT
IY665Rd2R	I1 (E261Q E268Q) reverse	TTGGGGCCCTTTACAACCAAGTTGGAACAAA CACCACCCATTTTTTG
IY630Rd3Mut1F	I2/G2 common forward	GATGTTATCGACGCCCATTTTTT
IY631Rd3Mut1R	I2 (E227Q) reverse	AAAAAATGGGCGTCGATAACATCTGTCACAG ATGCTTTGACCAAAAAAAGCGAGTGGG
IY633Rd3Mut2R	G2 (E227Q D231H) reverse	AAAAAATGGGCGTCGATAACATCTGTCACAG ATGCTTTGACCAAAAAAAGCGAGTGGG
IY720E240QF	E240Q forward	CAGATGTTATCGACGCCCATTTTTTCAGCAAC GTAAATTTGCAAAATC
IY721E240QR	E240Q reverse	CTGAAAAAATGGGCGTCGATAACATCT GATAGGATCGACGACTAACCGGTTGTTACTT GCTGTATTTTCCATGAGCCTTGCCTATTATCT TAGGCTTGGTAAAAGCTGCTTTTATCC
IY743dCR	HynS CTA <sub>22</sub> reverse	GTGATAGGATCGACGACTAACCGGTTGTTAC TTGCTGTATTTTCCATGAGCCTTGCCTATTAT CTTATTTGTACCACTCCCATGGACCCA
IY747dC2R	HynS CTA <sub>15</sub> reverse	GAGTGATAGGATCGACGACTAACCGGTTGTT ACTTGCTGTATTTTCTGCGTGATGGTGGTGGT GATGTGCCATGAGCCTTGCCTATTATC
IY699HHisR	His <sub>6</sub> -HynL reverse	CCAGCACCACCGCCAGAACCGCCACCTGAGT TTTTCCAGCATGCTTCTG
IY667HynSR	HynSΔ <sub>22</sub> -linker-PetF reverse	GAGCCTTGCCTATTATCTTATTTTTCGAACTG CGGGTGGGACCAAGCGCTACGGTAGAGGTCT TCTTCTTTGTGGGTTTCGATCGTCAC
IY747PetFR	PetF-strep-(hynSL intergenic region) reverse	CGGTTCTGGCGGTGGTGCTGGTGGTGGCTCT GGTGGCGGTGCAACCTACAAGGTTACGCT CGGATGGCCTTTTTGCGTTTTCTACTACTAGTT GACAATTAATCATCCGGCTCGTATAATGTGT GGAATTGTGAGCGGATA
IY668PetFF	PetF forward primer	TTGTAAAACGACGGCCAGTGAATTCTAGTAT TTCTCCTCTTCTAGTATGTGTGAAATTGTTA TCCGCTCACAAATCCACA
P1	Used for assembly of pRC41-4	
P2	Used for assembly of pRC41-4	

Table S2. *Cont.*

Name	Description	Sequence
T1	Used for assembly of pRC41-4	AACAGCTATGACCATGATTACGCCACATGCG AGAGTAGGGAAGCTG
T2	Used for assembly of pRC41-4	GATGATTAATTGTCAACTAGTAGTAGAAACG CAAAAAGGCC
AMR-1	Used for assembly of pRC41-4	GAATTCGAGCTCAAGGAGGAATAACATATGC ATGCGAGAGTAGGGAAGCTG
AMR1-2	Used for assembly of pRC41-4	AtgccggtactgccggcctcttgcgggatCATGCGAGAGTA GGGAAGCTG
AMR-2	Used for assembly of pRC41-4	CAAAAAGCCGCTACCAATAATGGAAGCCATC TAGTATTTCTCCTCTTTCTAGTATGTGTG
AMR-3	Used for assembly of pRC41-4	CACACATACTAGAAAGAGGAGAAATACTAG ATGGCTTCCATTATTGGTAGC
AMR3-2RBS-F	Used for assembly of pRC41-4	CAATTTACACAGGAAACAGACCATGGAggag gaactagATGGCTTCCATTATTGGTAGC
AMR-4	Used for assembly of pRC41-4	TGATGCCTGGCAGTTCCTACTCTCGCATGTT AGCTATGAGGTGCTGGAATT
AMR-12	Used for assembly of pRC41-4	TGATGCCTGGCAGTTCCTACTCTCGCATGTC AATCCTCCTTAGCCGTTT
AMR-13	Used for assembly of pRC41-4	TCTTCTGATGAAACGGCTAAGGAGGATTGAC ATGCGAGAGTAGGGAAGCTG
AMR-14		CGCCATTTCAATCCTCCTTAGCCGTTTCATCT AGTATTTCTCCTCTTTCTAGTATGTGTG
AMR-15		CACACATACTAGAAAGAGGAGAAATACTAG ATGAAACGGCTAAGGAGGAT
AMR20-3		CCGGCCACGATGCGTCCGGCGTAGAGGATCT TAACGAACTTTTAGACGCAAAC
AMR20-4		TGTTATTCCTCCTTGAGCTCGATTAACGAACT TTTAGACGCAAAC
AMR-22		AATGACTTTCATGGGGATAGCGAGACACATC TAGTATTTCTCCTCTTTCTAGTATGTGTG
AMR-23		CACACATACTAGAAAGAGGAGAAATACTAG ATGTGTCTCGCTATCCCCA
AMR-32		TGATGCCTGGCAGTTCCTACTCTCGCATGTT AGACTCCGCTATTTTGTAACGA
AMR-33		TCTCAGTCGTTACAAAATAGCGGAGTCTAAC ATGCGAGAGTAGGGAAGCTG
AMR-34		GTCTCTTAGCAACGATTCAACTGTACATCT AGTATTTCTCCTCTTTCTAGTATGTGTG
AMR-35		CACACATACTAGAAAGAGGAGAAATACTAG ATGTACAGTGTTGAATCGTTGCTA
AMR-44		TGCATGCCTGCAGGTGCACTCTAGAGGATCT CAGCAGATCCTTGGAAGC

**Table S3.** List of Plasmids used in this study.

<b>Plasmid</b>	<b>Description</b>	<b>Construction Notes</b>
pRC41-4	WT hydrogenase, 4× TRC-driven	See Methods section
pCM012	G1 hydrogenase, 4× TRC-driven	BamHI/AgeI digest of pRC41-4, gibson assembly with IY170HynSF/IY171HynSR amplicon of pIY007 Reference: [12]
pIY038	G1 hydrogenase, T7-driven	See reference: [12]
pIY041	HynS C-terminus inserted into pUC19	SmaI digest of pUC19, blunt-end ligation with IY170HynSF/IY171HynSR amplicon of pIY007
pIY042	ALL+ substitutions in pUC19 insert of HynS	Three-stage PCR: stage 1: IY170HynSF, IY103FdR1PR; IY102FdR1PF, IY107FdR3PR; IY106FdR3PF, IY111FdR5PR; IY110FdR5PF, IY171HynSR amplicons of pIY041 stage 2: IY170HynSF, IY105FdR2PR; IY104FdR2PF, IY109FdR4PR; IY108FdR4PF, IY113FdR6PR; IY112FdR6PF, IY171HynSR amplicons of pIY041 stage 3: IY170HynSF, IY171HynSR amplicons of stage 2 amplicons, IY100p18F/IY100p18R amplicons of pIY041 gibson assembly of stage 3 amplicons
pIY043	ALL+, T7-driven. This construct is essentially pIY038 with the following substitutions: D224R, E225K, E235K, D239R, E248K, E269K, E276K, E305K, D307R, D310R	BamHI/AgeI digest of pIY038, gibson assembly with IY170HynSF/IY171HynSR amplicon of pIY042
pIY062	G1, DDE, T7-driven	BamHI/AgeI digest of pIY038, gibson assembly with IY170HynSF, IY601DDDER; IY600DDEF, IY171HynSR amplicons of pIY038
pIY063	I1, DDE, E261K, T7-driven	BamHI/AgeI digest of pIY038, gibson assembly with IY170HynSF, IY603Mut2R; IY602Mut2F, IY171HynSR amplicons of pIY062
pIY068	I2, DDE, E261K, T7-driven	BamHI/AgeI digest of pIY038, gibson assembly with IY170HynSF, IY631Rd3Mut1R; IY630Rd3Mut1F, IY171HynSR amplicons of pIY063
pIY069	G2, DDE, E261K, T7-driven	BamHI/AgeI digest of pIY038, gibson assembly with IY170HynSF, IY633Rd3Mut2R; IY630Rd3Mut1F, IY171HynSR amplicons of pIY063
pIY070	I1, DDE, T7-driven	BamHI/AgeI digest of pIY038, gibson assembly with IY170HynSF, IY665Rd2R; IY664Rd2F, IY171HynSR amplicons of pIY062

**Table S3. Cont.**

<b>Plasmid</b>	<b>Description</b>	<b>Construction Notes</b>
pIY071	G2, DDE, T7-driven	BamHI/AgeI digest of pIY038, gibson assembly with IY170HynSF, IY633Rd3Mut2R; IY630Rd3Mut1F, IY171HynSR amplicons of pIY070
pIY087	I1, 4× TRC-driven	BamHI/AgeI digest of pCM012, gibson assembly with IY170HynSF, IY665Rd2R; IY664Rd2F, IY171HynSR amplicons of pCM012
pIY088	I2, 4× TRC-driven	BamHI/AgeI digest of pCM012, gibson assembly with IY170HynSF, IY631Rd3Mut1R; IY630Rd3Mut1F, IY171HynSR amplicons of pIY087
pIY089	G2, 4× TRC-driven	BamHI/AgeI digest of pCM012, gibson assembly with IY170HynSF, IY633Rd3Mut2R; IY630Rd3Mut1F, IY171HynSR amplicons of pIY087
pIY094	G2, E240Q, 4× TRC-driven	BamHI/AgeI digest of pCM012, gibson assembly with IY170HynSF, IY721E240QR; IY720E240QF, IY171HynSR amplicons of pIY089
pIY098	G1, HynS CTΔ22, 4× TRC-driven	BamHI/AgeI digest of pCM012, gibson assembly with IY170HynSF, IY743dCR amplicon of pRC41-4
pIY099	G1, HynS CTΔ15, 4× TRC-driven	BamHI/AgeI digest of pCM012, gibson assembly with IY170HynSF, IY747dCR amplicon of pRC41-4
pIY107	G1, HynS-strep, His <sub>6</sub> -HynL, 4× TRC-driven	BamHI/AgeI digest of pCM012, gibson assembly with IY170HynSF, IY699HHisR amplicon of pIY086. Please see [12]
pIY108	G2, HynS-strep, His <sub>6</sub> -HynL, 4× TRC-driven	BamHI/AgeI digest of pCM012, gibson assembly with IY170HynSF, IY665Rd2R amplicon of pIY089 and IY664Rd2F, IY171HynSR amplicon of pIY107
pIY109	G1, HynS CTΔ22::PetF-strep, His <sub>6</sub> -HynL, 4× TRC-driven	BamHI/AgeI digest of pCM012, gibson assembly with two-stage PCR amplicon. stage 1: IY170HynSF, IY667HynSR amplicon of pRC41-4; IY668PetFF, IY747PetFR amplicon of <i>S. elongatus</i> gDNA stage 2: IY170HynSF, IY699HHisR amplicon of both stage 1 amplicons
pIY110	G2, HynS CTΔ22::PetF-strep, His <sub>6</sub> -HynL, 4× TRC-driven	BamHI/AgeI digest of pCM012, gibson assembly with two-stage PCR amplicon stage 1: IY170HynSF, IY667HynSR amplicon of pIY089; IY668PetFF, IY747PetFR amplicon of <i>S. elongatus</i> gDNA. stage 2: IY170HynSF, IY699HHisR amplicon of both stage 1 amplicons

Full sequences are available in supplemental file sequences.gbk, pRC41-4 is available in reference [12].

LOCUS pCM012 19895 bp DNA circular UNA 10-Jul-2010

## DEFINITION

## FEATURES

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promoter	5616..5678 /label=PTRC
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ORIGIN

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LOCUS pIY038 18706 bp DNA circular UNA 10-Jul-2010

DEFINITION

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DEFINITION
FEATURES      Location/Qualifiers

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## ORIGIN

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3181 aaaaataaac aaataggggt tccgcgcaca tttccccgaa aagtgccacc tgacgtctaa
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LOCUS      pIY043      18706 bp      DNA      circular UNA 10-Jul-2010

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DEFINITION

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## ORIGIN

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LOCUS pIY062 18697 bp DNA circular UNA 10-Jul-2010

DEFINITION

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LOCUS      pIY063      18697 bp      DNA      circular UNA 10-Jul-2010

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DEFINITION

FEATURES

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## ORIGIN

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LOCUS pIY068 18697 bp DNA circular UNA 10-Jul-2010

DEFINITION

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LOCUS pIY069 18697 bp DNA circular UNA 10-Jul-2010  
DEFINITION

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## ORIGIN

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LOCUS pIY070 18697 bp DNA circular UNA 10-Jul-2010

DEFINITION

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LOCUS pIY071 18697 bp DNA circular UNA 10-Jul-2010

DEFINITION

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LOCUS pIY087 19895 bp DNA circular UNA 10-Jul-2010

DEFINITION

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19261 GCCAACTTAC TTCTGACAAC GATCGGAGGA CCGAAGGAGC TAACCGCTTT TTTGCACAAC
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19621 CCCTCCCGTA TCGTAGTTAT CTACACGACG GGGAGTCAGG CAACTATGGA TGAACGAAAT
19681 AGACAGATCG CTGAGATAGG TGCCTCACTG ATTAAGCATT GGTAACGTGC AGACCAAGTT
19741 TACTCATATA TACTTTAGAT TGATTAAAA CTCATTTTT AATTTAAAAG GATCTAGGTG
19801 AAGATCCTTT TTGATAATCT CATGACCAA ATCCCTAAC GTGAGTTTTT GTTCCACTGA
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LOCUS      pIY089      19895 bp      DNA      circular UNA 10-Jul-2010

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DEFINITION

FEATURES

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misc_feature    complement(773..913)
                /feature_type="Transfer Origin"
                /label=oriT
Region          1060..1908
                /label=NSI-up
CDS             2682..3622
                /cds_type=ORF
                /label=SpSm
terminator      3882..3978
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CDS             4302..5384
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promoter        5616..5678
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CDS             5706..6824
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                /label=orf2
CDS             6839..7393
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terminator      complement(7825..8075)
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CDS             8163..9161
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                /label=IY631Rd3Mut2R
mutation        8841..8843
                /label=E235Q
mutation        8850..8852
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mutation        8853..8855

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mutation        /label=IY630Rd3MutF
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                9015..9017
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CDS             /label=hynL-RBS
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CDS            /label=PTRC
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CDS            /label=hypC
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                /cds_type=ORF
CDS            /label=hypA
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CDS            /label=hypF
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terminator /label=hypE  
 complement (17829..18079)  
 Region /label=rrnBt  
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 /label=NSI-dn

## ORIGIN

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121 CAGCAGAGCG CAGATACCAA ATACTGTCCT TCTAGTGTAG CCGTAGTTAG GCCACCACTT
181 CAAGAACTCT GTAGCACCGC CTACATACCT CGCTCTGCTA ATCCTGTTAC CAGTGGCTGC
241 TGCCAGTGGC GATAAGTCGT GTCTTACCGG GTTGGACTCA AGACGATAGT TACCGGATAA
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LOCUS

pIY094

19895 bp

DNA

circular UNA 10-Jul-2010





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LOCUS pIY098 19826 bp DNA circular UNA 10-Jul-2010

DEFINITION

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mutation      8850..8852
                /label=H238C
mutation      9015..9017
                /label=P293C
primer        complement(9065..9154)
                /label=IY743dCR
mutation      9089^9090
                /label=delta-C
RBS           9096..9101
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CDS           9110..10987
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CDS           11396..11605
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241 TGCCAGTGGC GATAAGTCGT GTCTTACCGG GTTGGACTCA AGACGATAGT TACCGGATAA
301 GGCGCAGCGG TCGGGCTGAA CGGGGGGTTT GTGCACACAG CCCAGCTTGG AGCGAACGAC
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LOCUS      pIY099      19850 bp      DNA      circular UNA 10-Jul-2010

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DEFINITION

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241 TGCCAGTGGC GATAAGTCGT GTCTTACCGG GTTGGACTCA AGACGATAGT TACCGGATAA
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LOCUS pIY107 19952 bp DNA circular UNA 10-Jul-2010

## DEFINITION

## FEATURES

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terminator	3882..3978 /label=T4t
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CDS	5706..6824 /cds_type=ORF /label=orf2
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mutation	9015..9017 /label=P293C
RBS	9198..9203 /label=hynL-RBS
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ORIGIN

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LOCUS pIY108b 19952 bp DNA circular UNA 10-Jul-2010

DEFINITION

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LOCUS pIY109 20225 bp DNA circular UNA 10-Jul-2010

DEFINITION

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LOCUS pIY110 20225 bp DNA circular UNA 10-Jul-2010

DEFINITION

FEATURES

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CDS 8163..9467
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19561 GCTGCCATAA CCATGAGTGA TAACACTGCG GCCAACTTAC TTCTGACAAC GATCGGAGGA  
19621 CCGAAGGAGC TAACCGCTTT TTTGCACAAC ATGGGGGATC ATGTAACTCG CCTTGATCGT  
19681 TGGGAACCGG AGCTGAATGA AGCCATACCA AACGACGAG GTGACACCAC GATGCCTGCA  
19741 GCAATGGCAA CAACGTTGCG CAAACTATTA ACTGGCGAAC TACTTACTCT AGCTTCCCAG

19801 CAACAATTAA TAGACTGGAT GGAGGCGGAT AAAGTTGCAG GACCACTTCT GCGCTCGGCC  
19861 CTTCCGGCTG GCTGGTTTAT TGCTGATAAA TCTGGAGCCG GTGAGCGTGG GTCTCGCGGT  
19921 ATCATTGCAG CACTGGGGCC AGATGGTAAG CCCTCCCGTA TCGTAGTTAT CTACACGACG  
19981 GGGAGTCAGG CAACTATGGA TGAACGAAAT AGACAGATCG CTGAGATAGG TGCCTCACTG  
20041 ATTAAGCATT GGTAAGTGTG AGACCAAGTT TACTCATATA TACTTTAGAT TGATTTAAAA  
20101 CTTCATTTTT AATTTAAAAG GATCTAGGTG AAGATCCTTT TTGATAATCT CATGACCAAA  
20161 ATCCCTTAAC GTGAGTTTTT GTTCCACTGA GCGTCAGACC CCGTAGAAAA GATCAAAGGA  
20221 TCTTC

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