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Figure S1. Comparison of the chemical structures of PHA and PTE

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R¹, alkyl group (C_nH_{2n+1}) or functionalized alkyl group; z, 1 to 4 -CH₂-; R², hydrogen,

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methyl or ethyl group.

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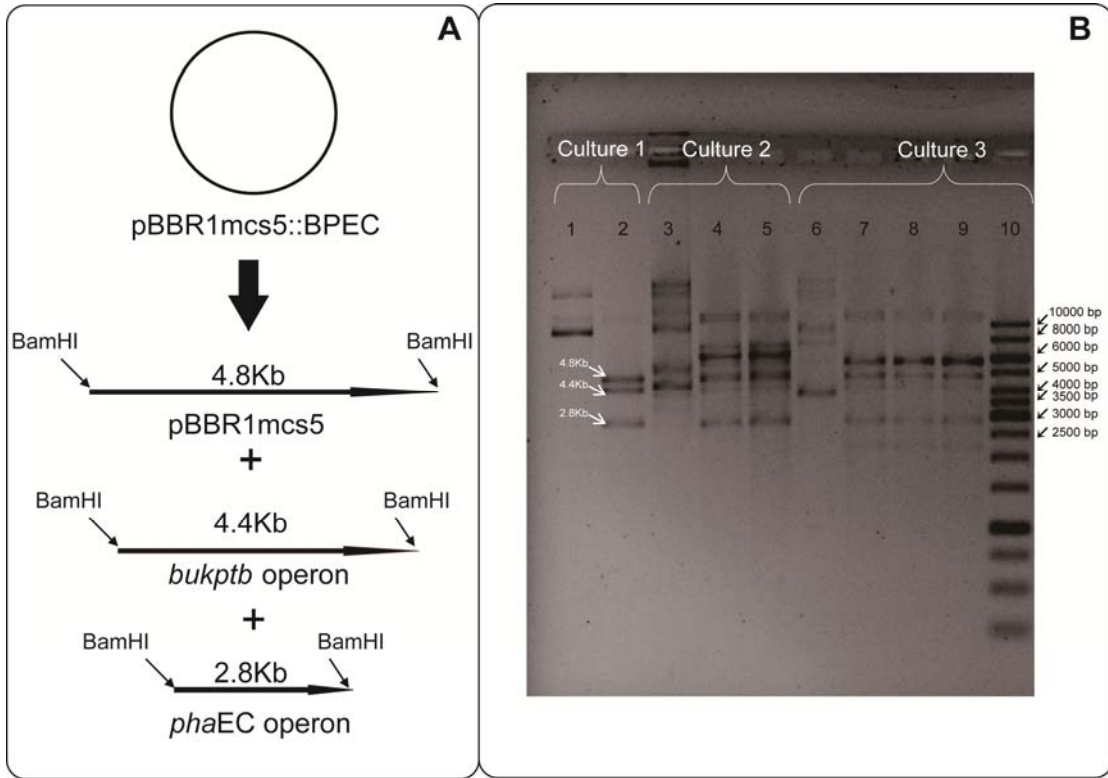
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23 Figure S2. Plasmid structural instability in strain SHX1

24 A) *Bam*HI cut pBBR1MCS5::BPEC consists of 4.8-kb pBBR1MCS5, 4.4-kb *bukptb*
 25 operon and 2.8-kb *phaEC* operon as indicated in the figure with arrows.

26 B) All plasmids were extracted from *A. mimigardefordensis* pBBR1MCS5::BPEC
 27 cultivated in MSM containing 0.5% (wt/vol) gluconate. Lanes 1, 3, and 6 show plasmid
 28 pBBR1MCS5::BPEC in a 1% (wt/vol) agarose gel. Lanes 2, 4, 5, 7, 8, and 9 show the
 29 fragments obtained from the same plasmid after restriction with *Bam*HI. Lanes 1 and 2
 30 are the normal sized plasmids without a structural rearrangement (compare Fig. S2A).

31 Plasmid was extracted after 4 h of cultivation. Lanes 3 to 9 represent plasmids extracted
 32 from the same culture, but after an incubation time of 12 h. This demonstrates the

33 instability of plasmids with structural rearrangement. Lane 10 shows the molecular
34 weight marker for agarose gel electrophoresis (GeneRuler™ 1-kb DNA Ladder).

35 Relevant fragment sizes are indicated.

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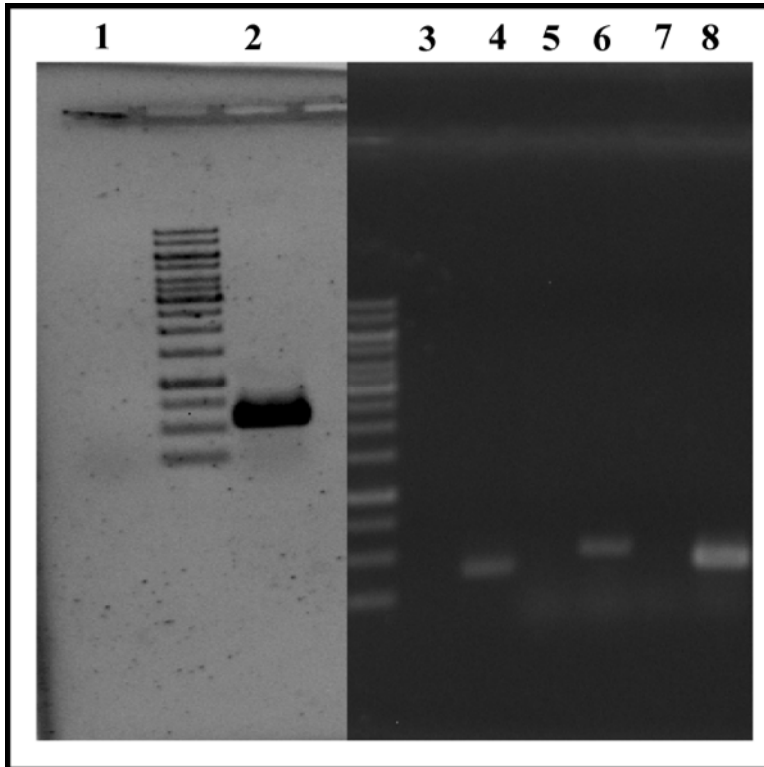
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49 Figure S3. Results of RT-PCR

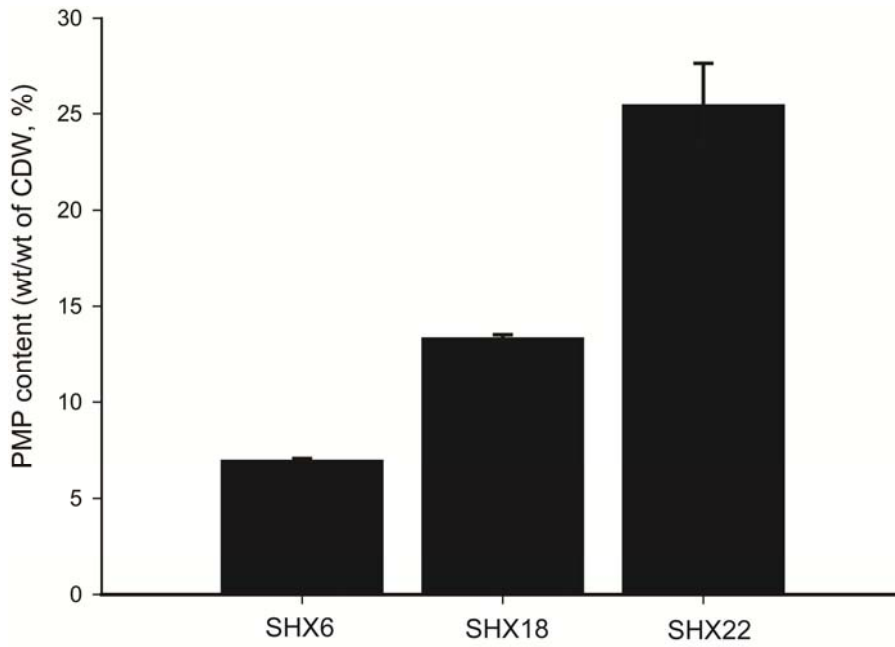
50 RT-PCR was carried out with RNA isolated from *A. mimigardefordensis* SHX5. Four
51 samples of their relevant genes in BPEC pathway gave positive signals by using a
52 RT-PCR kit (Qiagen). The samples in lanes 2, 4, 6, and 8 show the PCR signals of *ptb*,
53 *phaE*, *buk* and *phaC*, respectively. Samples in lanes 1, 3, 5, and 7 were DNA
54 contamination controls of the samples separated in lanes 2, 4, 6, and 8, respectively.

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61 Figure S4. Comparison of PMP production in three different strains cultivated under

62 the same conditions

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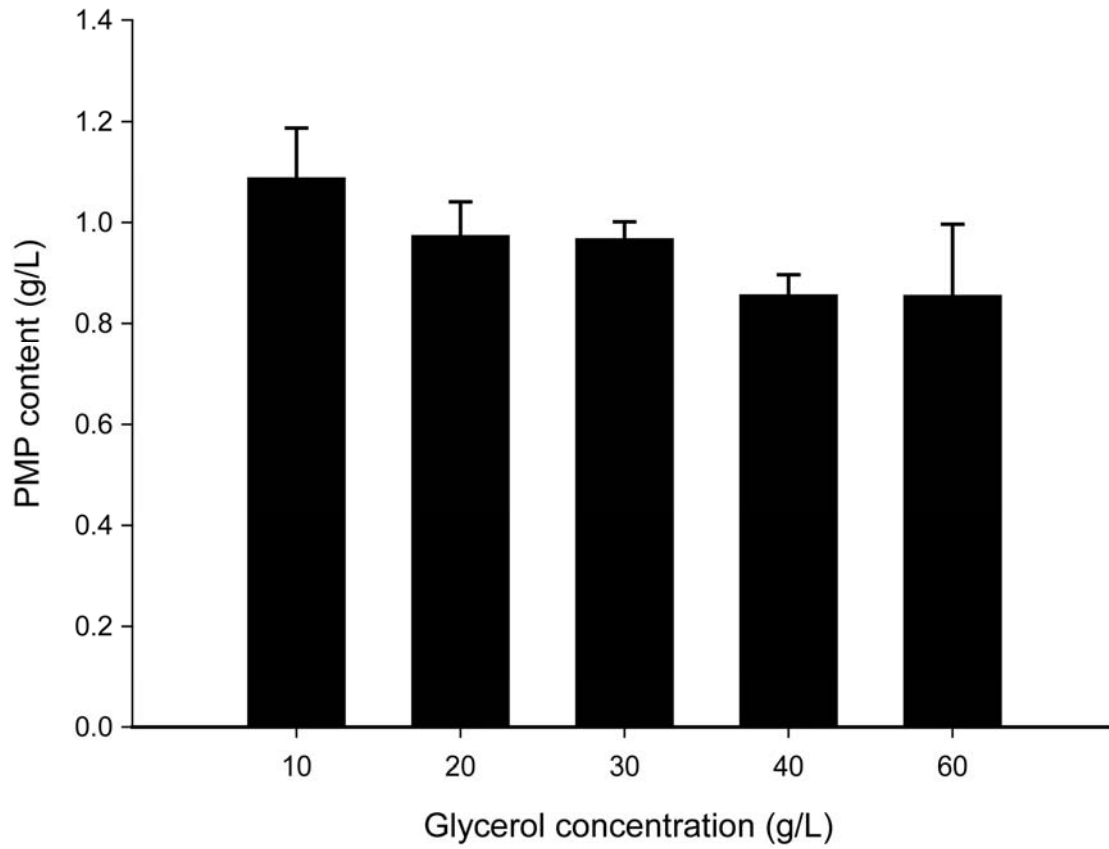
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75 Figure S5. Final PMP concentrations after cultivation of strain SHX22 in mineral salts

76 medium containing different glycerol concentrations

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84 TABLE S1. Primer used in this study

Oligonucleotides	Sequence	Purpose
M13 forward	GTAAAACGACGGCCAGT	Common primers for sequencing
M13 reverse	CAGGAAACAGCTATGAC	Common primers for sequencing
bukptb_Fr_EcoRI	GAATTCAGCGGCTGTGGATGGAGTT	Forward primer for insertion of <i>bukptb</i> operon into pBBR1MCS5
bukptb_Rev_EcoRI	GAATTCGGTGCAAATATTGGCATGTGG	Reverse primer for insertion of <i>bukptb</i> operon into pBBR1MCS5
recA_UFR_Fr	GGATCCACCAATCACTTCGTCTGC	Forward primer to clone the upstream flank of <i>recA</i>
recA_UFR_Rev	GAATTCGGAGGGTCCTTGTGACTG	Reverse primer to clone the upstream flank of <i>recA</i>
recA_DFR_Fr	GAATCTTTTTCCCAAGGGCAGGGCAGC	Forward primer to clone the downstream flank of <i>recA</i>
recA_DFR_Rev	GGATCCCCGAGCGTTATGGCGTTCAGTG	Reverse primer to clone the downstream flank of <i>recA</i>
recA_IN_Fr	GGACGACAAGCAATCCAAAG	Forward internal sequencing primer to verify deletion of <i>recA</i>
recA_IN_Rev	TCACTCATCCGCTTCCGTAG	Reverse internal sequencing primer to verify deletion of <i>recA</i>
recA_OUT_Fr	GGCAAGAAGCCATTCCGGTC	Forward external sequencing primer to verify deletion of <i>recA</i>
recA_OUT_Rev	TGGGATAACGCCAGCACAC	Reverse external sequencing primer to verify deletion of <i>recA</i>
mdo_UFR_Fr	CAACGGAGAGAAATACTGGATCAC	Forward primer to clone the upstream flank of <i>mdo</i>
mdo_UFR_Rev	AAAAAAGAATTCAATATGCTCCTTTGGTGTCG	Reverse primer to clone the upstream flank of <i>mdo</i>
mdo_DFR_Fr	AAGAATTCGACCGTTCCTATTGCTACATTCCAC	Forward primer to clone the downstream flank of <i>mdo</i>
mdo_DFR_Rev	GCCGGTTCCTGCACCGTTAC	Reverse primer to clone the downstream flank of <i>mdo</i>
phaC_UFR_Fr	GGATCCATAACGGCAAATCGCTGGGTC	Forward primer to clone the upstream flank of <i>phaC_{Am}</i>
phaC_UFR_Rev	GAATTCCTGTGGCACTCTCACCGTATAA	Reverse primer to clone the upstream flank of <i>phaC_{Am}</i>

phaC_DFR_Fr	GAATTCTCAGGCCAATACATCGTTT	Forward primer to clone the downstream flank of <i>phaC_{Am}</i>
phaC_DFR_Rev	GGATCCAGATAGGAACCCATCACGC	Reverse primer to clone the downstream flank of <i>phaC_{Am}</i>
phaC_inFr_Check	ATGAGCATTAGACAACAGTCGGAG	Forward internal sequencing primer to verify deletion of <i>phaC_{Am}</i>
phaC_inRev_Check	TCAGGCTTTAACTTTCACATAGTGC	Reverse internal sequencing primer to verify deletion of <i>phaC_{Am}</i>
phaC_frount_check	GAGCGCCTGGCTTTTGGTTATATAG	Forward external sequencing primer to verify deletion of <i>phaC_{Am}</i>
phaC_revout_check	TGCGGGATTGAACGGGAAC	Reverse external sequencing primer to verify deletion of <i>phaC_{Am}</i>
phaE_180_Fr_RT	ACCTGGTTCGCGACTTCATG	Forward primer of <i>phaE</i> probe using in RT-PCR
phaE_580_Rv_RT	AGCCGTTGTATTCGTTCAAGG	Reverse primer of <i>phaE</i> probe using in RT-PCR
phaC_325_Fr_RT	CGTCTATCTGATCGACTGGG	Forward primer of <i>phaC</i> probe using in RT-PCR
phaC_774_Rv_RT	GATGAACTGGCGGAAGGTC	Reverse primer of <i>phaC</i> probe using in RT-PCR
ptb_30_Fr_RT	CATGAAGGTAAAGAGCAAAG	Forward primer of <i>ptb</i> probe using in RT-PCR
ptb_390_Rev_RT	CTATCAGATGTCTCAGTTTC	Reverse primer of <i>ptb</i> probe using in RT-PCR
buk_35_Fr_RT	ATAATCAATCCTGGCTCGAC	Forward primer of <i>buk</i> probe using in RT-PCR
buk_550_Rv_RT	CTACTGAAGTACCTCCACC	Reverse primer of <i>buk</i> probe using in RT-PCR
phaCam_operon_Fr	GCTAAAGCTTACCAGCGATGACGGTGTTAAAC	Forward primer for insertion <i>phaC_{Am}</i> into pBBR1MCS5
phaCam_operon_Rev	AATGAAGCTTGAAGCTTCTTAGGCTACTTTCACGG	Reverse primer for insertion <i>phaC_{Am}</i> into pBBR1MCS5
phaCre_operon_Fr	AATCGGATCCC GCCATGCCATACATCAGGAAG	Forward primer for insertion <i>phaC_{Re}</i> into pBBR1MCS5
phaCre_operon_Rev	GATCGGATCCCTTTCAATGGAAACGGGAGGGAAC	Reverse primer for insertion <i>phaC_{Re}</i> into pBBR1MCS5

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88 Table S2. PMP and PHB production of strains SHX1 and SHX2 in the modified two

89 stage cultivation method

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Culture conditions	Strains	Polymer content		
		3HB (wt/wt of CDW, %)	3HV (wt/wt of CDW, %)	3MP (wt/wt of CDW, %)
Limited nitrogen	SHX1	14.2	3.4	below detection limit
	SHX2	12.1	2.2	below detection limit
Normal nitrogen	SHX1	below detection limit	below detection limit	1.7
	SHX2	3.5	below detection limit	below detection limit

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92 limited nitrogen condition: MS medium containing 0.5 g/L NH₄Cl

93 normal nitrogen condition: MS medium containing 1 g/L NH₄Cl

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