

Figure S1. The construction and verification of the modified mutants. (A) Schematic diagram of the mutants construct process; (B) The verification of QSRZ601 by PCR with primers *gcd* F1 and *gcd* R2. Lane 1-2: negative clone, 3-4: positive clone; (C) The verification of QSRZ602 by PCR with primers *gltA* F1 and *gltA* R2. Lane 1: negative clone, 2-4: positive clone; (D) The verification of QSRZ603 by PCR with primers *gltA* F1 and *gltA* R2. Lane 1-4: negative clone, 5: positive clone; (E) The verification of QSRZ604 by PCR with primers *hexR* F1 and *hexR* R1. Lane 1: positive clone; (F) The verification of QSRZ605 by PCR with primers *gcd* F1 and *gcd* R2. Lane 1: positive clone; (G) The verification of QSRZ606 by PCR with primers *gltA* F1 and *gltA* R2. Lane 1-2: negative clone, lane 3: positive clone; (H) The verification of QSRZ607 by PCR with primers *gltA* F1 and *gltA* R2. Lane 1: Positive clone, lane 2-4: negative clone. (I) The verification of *gltB* overexpression mutants. lane 1-2: P. *putida* QSRZ608 with primers *gltB* F1 and *gltB* R2; 3-4: The PCR verification of P. *putida* QSRZ609 with primers *gltB* F1 and *gltB* R2. (J): The verification of *phaD* overexpression mutants; Lane 1: The PCR

verification of QSRZ610 with primers *phaD* F1 and *phaD* R2; Lane 2: The PCR verification of QSRZ611 with primers *phaD* F1 and *phaD* R2.

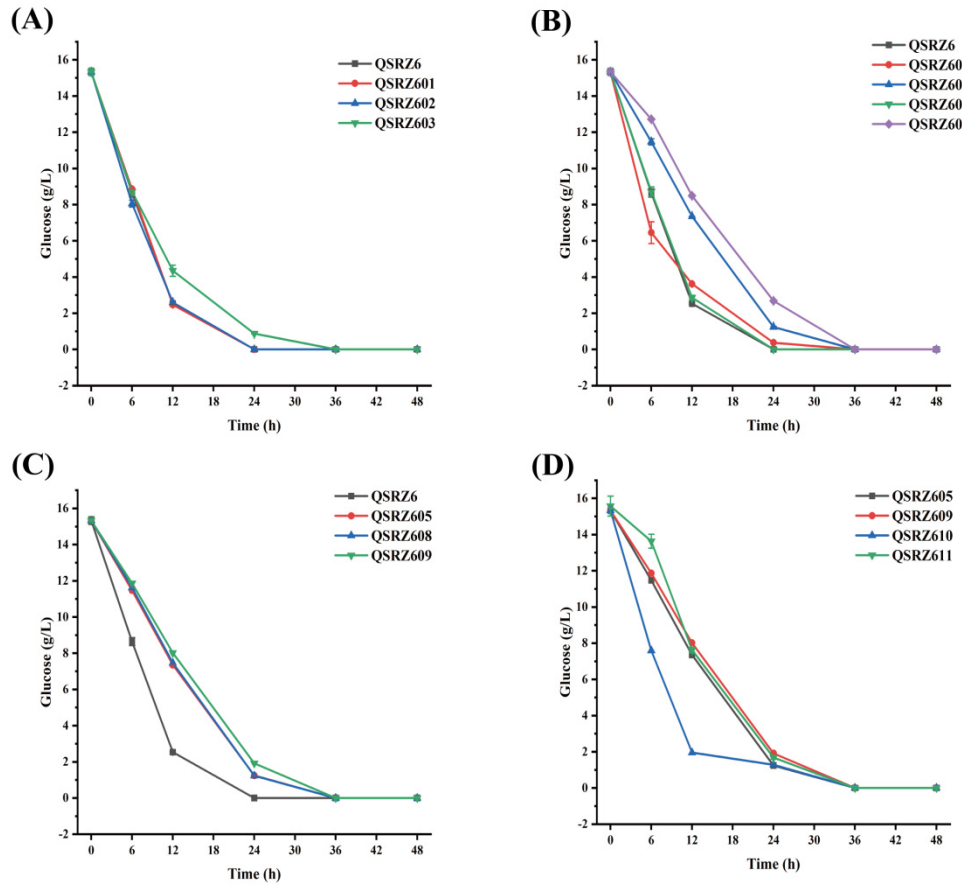


Figure S2 The amount of glucose remaining during the shake flask fermentation. (A) Residual sugar content of QSRZ6, QSRZ601, QSRZ602 and QSRZ603; (B): Residual sugar content of QSRZ6, QSRZ604, QSRZ605, QSRZ606 and QSRZ607; (C): Residual sugar content of QSRZ6, QSRZ605, QSRZ608 and QSRZ609; (D): Residual sugar content of QSRZ605, QSRZ609, QSRZ610 and QSRZ611.

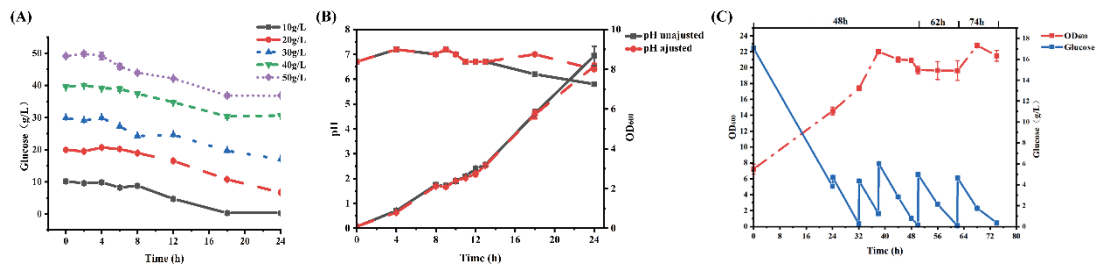


Figure S3 Optimization of glucose concentration, pH and feeding strategy. (A) Residual sugar content of QSRZ607 cultured in LBG medium with different

concentrations of glucose; (B) The cell growth of QSRZ607 in LBG medium with and without pH adjustment in culture; (C) the cell growth of QSRZ607 with a small amount of multiple feeding strategy.

Table S1 All primers in this study

Primers	Sequences (5'~3')
<i>gcd</i> F1	agctcgggtacccgggggatccGCCATGCCGTAGGCTTTGAC
<i>gcd</i> R1	agaacctacgatgagCTGCTCCCGAACCATTGAAAC
<i>gcd</i> F2	tcaatgggttcgggagCTCATCGTAGGTTCTCCGTC
<i>gcd</i> R2	cgacggccagtccaagcttACTTCAACAGCTTCCCTTGC
<i>glta</i> F1	agctcgggtacccgggggatccTTACCATAACCCAACTCCCACCAG
<i>glta</i> R1	ccagggcgaagatcaCAGCGCCCTCGATGACCAACT
<i>glta</i> F2	tcacgagggcgctgTGATCTTCGCCCTGGCACGTA
<i>glta</i> R2	cgacggccagtccaagcttATCGCCCCGTCCATCACTTCT
<i>hexR</i> F1	agctcgggtacccgggggatccGCCAATCCATTTTCGGTTCC
<i>hexR</i> R1	accgtgcaaaaagccCCGACATCTACATGCCAATG
<i>hexR</i> F2	gcatgtagatgtcggGGCTTTTGCACGGTCGTCT
<i>hexR</i> R2	cgacggccagtccaagcttGGCGTTGGCGTTGTCTTCTT
<i>glbB</i> F1	agctcgggtacccgggggatccCTGTGGTCGTGGTTGCCGTT
<i>glbB</i> R1	GCAGGCTAGTCGAAAGCTAG
<i>glbB</i> FP17	tttgactagcctgcGGGGATTTCGCGTGGCAGAAGA
<i>glbB</i> RP17	tagaaaacctccttaCGCCCATAACCACATTCCAGAC
<i>glbB</i> P17-F2	atgtggttatgggcgtaaggaggttttctaATGAAAACAGGTCTGTACCATCCC
<i>glbB</i> R2	cgacggccagtccaagcttCGATTTCCGAGGCGATGGTGA
<i>glbB</i> FP33	tttgactagcctgcCGGTCCGACATGAGTATTCC
<i>glbB</i> RP33	tagaaaacctccttatGGTGTTGCCCTCACTTGTTG
<i>glbB</i> P33-F2	agtgagggaacacctaaggaggttttctaATGAAAACAGGTCTGTACCATCCC
<i>phaD hexR</i> -F1	agctcgggtacccgggggatccGCCAATCCATTTTCGGTTCC
<i>phaD hexR</i> -R1	CCGACATCTACATGCCAATG
<i>phaD</i> FP33	gcatgtagatgtcggCGGTCCGACATGAGTATTCC
<i>phaD</i> RP33	tagaaaacctccttaGGTGTTGCCCTCACTTGTTG
<i>phaD</i> F	agtgagggaacacctaaggaggttttctaATGAAAACCCGCGATCGTAT
<i>PhaD</i> R	accgtgcaaaaagccACCCTGATCTGATACCGCGT
<i>phaD hexR</i> -F2	GGCTTTTTGCACGGTCGTCT
<i>phaD hexR</i> -R2	cgacggccagtccaagcttGGCGTTGGCGTTGTCTTCTT
<i>phaD</i> -qF	CCTGGAAATTGCCAACGAACTC
<i>phaD</i> -qR	ACAGCCAGTAATCCTCTGCGTC
16S-qF	CGGATCGCAGTCTGCAACTC
16S-qR	ACACCGTGGTAACCGTCCTC

Table S2 The CDW, PHAs content and production of QSRZ607 with different glucose concentrations

<b>Glc initial content</b> <b>Parameter</b>	<b>10 g/L</b>	<b>20 g/L</b>	<b>30 g/L</b>	<b>40 g/L</b>	<b>50 g/L</b>
CDW(g/L)	7.35±0.05e	9.13±0.13d	10.67±0.16a	10.08±0.03b	9.69±0.09c
PHAs(wt%)	46.76±0.98d	51.08±1.44c	56.20±1.17a	55.93±1.80ab	53.18±1.46bc
PHAs(g/L)	3.44±0.07e	4.66±0.13d	6.00±0.13a	5.63±0.18b	5.15±0.14c

Note: Different lowercase letters in the same industry indicate significant differences among different mutant strains ( $p<0.05$ ).

Table S3 The CDW, PHA content and production of QSRZ607 with two feeding strategy

<b>Feeding strategy</b>	<b>Single feeding to 10g/L</b>	<b>Multiple feeding to 4-6g/L</b>		
<b>Time(h)</b> <b>Parameter</b>	<b>48</b>	<b>48</b>	<b>62</b>	<b>74</b>
CDW(g/L)	11.58±0.03c	12.64±0.11b	12.97±0.03a	13.00±0.01a
PHAs(wt%)	58.34±2.74a	54.22±0.33b	56.00±2.16b	55.45±2.70b
PHAs(g/L)	6.75±0.32	6.85±0.09	7.26±0.27	7.20±0.35

Note: Different lowercase letters in the same industry indicate significant differences among different mutant strains ( $p<0.05$ ).