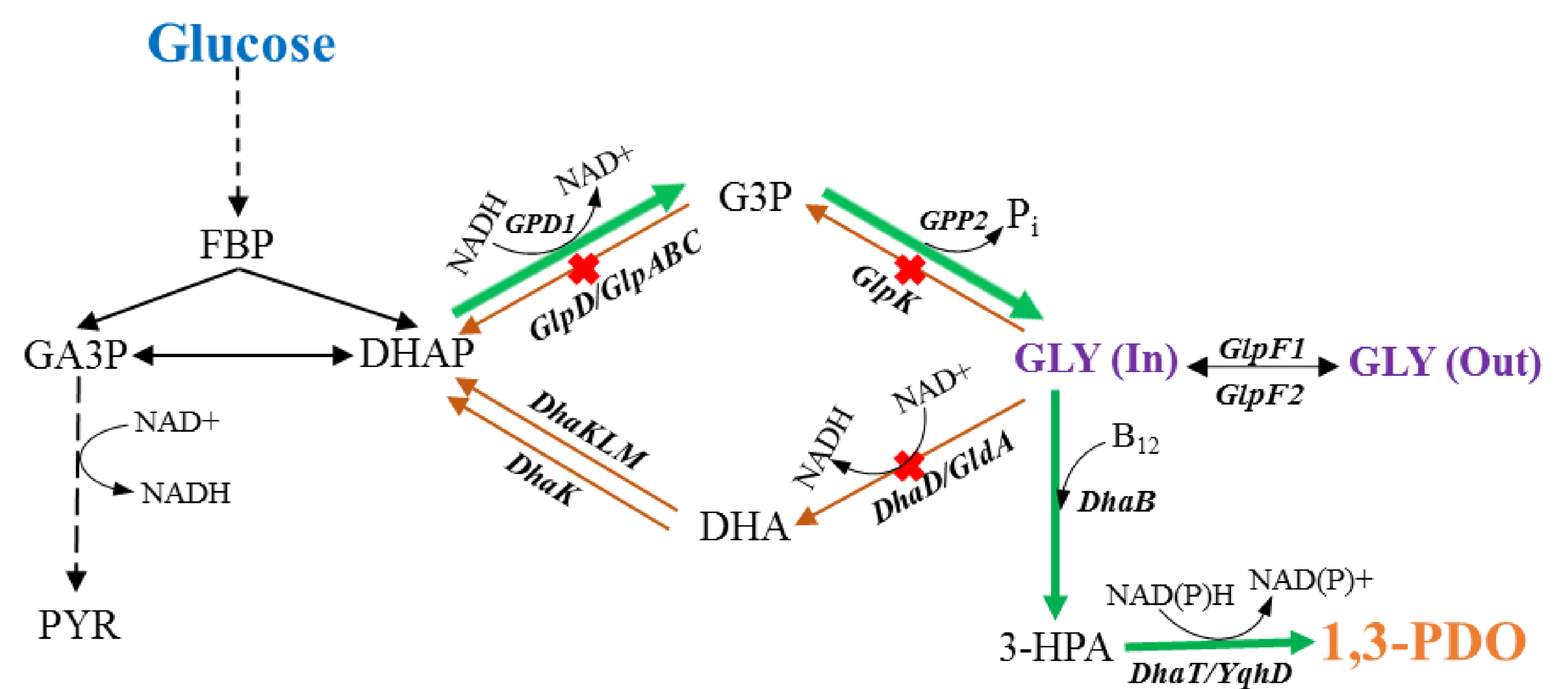


INTRODUCTION

- 1,3-propanediol (1,3-PDO) important precursor for polytrimethylene terephthalate (PTT) synthesis.
- No natural microorganisms that could directly convert glucose into 1,3-PDO have been found so far.
- *K. pneumoniae* J2B
 - ❖ Natural producer of 1,3-PDO from glycerol
 - ❖ Coenzyme B₁₂ producer: Cofactor of glycerol dehydratase
- We aimed to produce 1,3-PDO from glucose using *K. pneumoniae* J2B by introducing synthetic glycerol biosynthesis pathway.
- Glycerol-3-phosphate dehydrogenase (GPD1) and glycerol-3-phosphate phosphatase (GPP2) of *S. cerevisiae* were well-studied for production of glycerol from glucose.

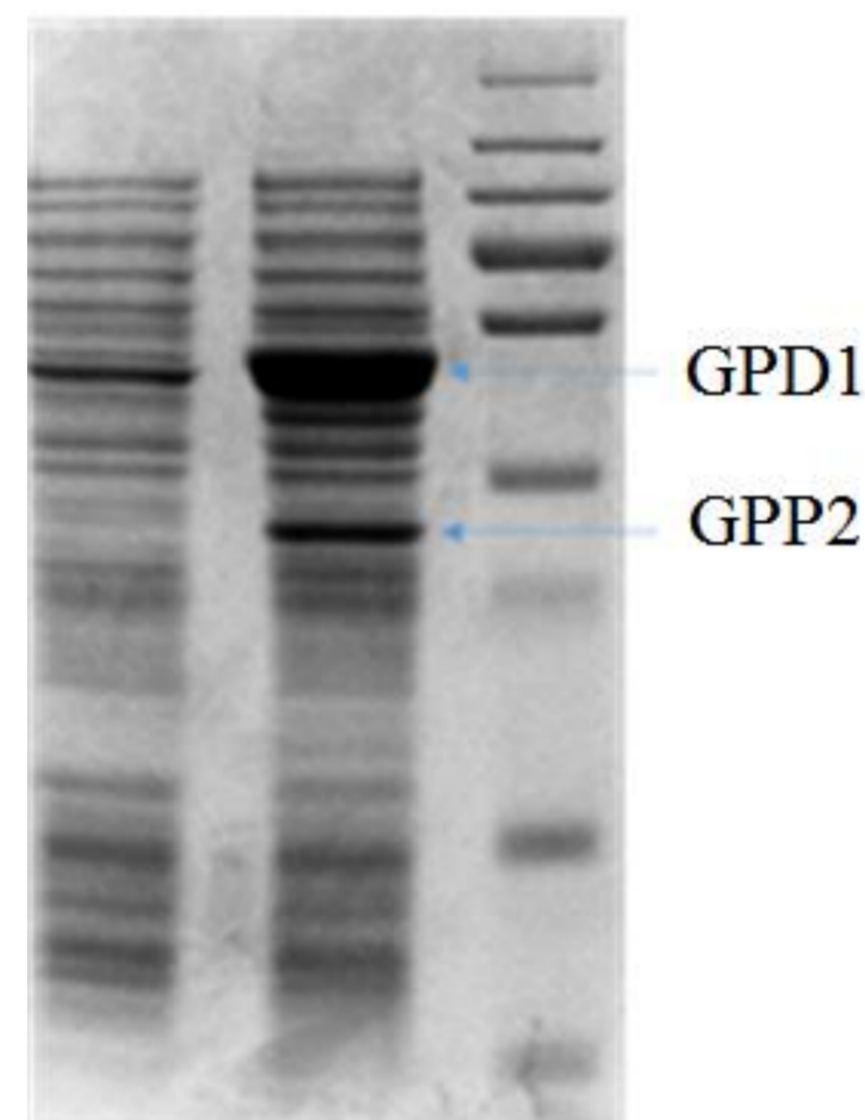
APPROACH

- Overexpression of *Saccharomyces cerevisiae* glycerol-3-phosphate dehydrogenase (GPD1) and glycerol-3-phosphate phosphatase (GPP2), and deletion of glycerol oxidative pathway.

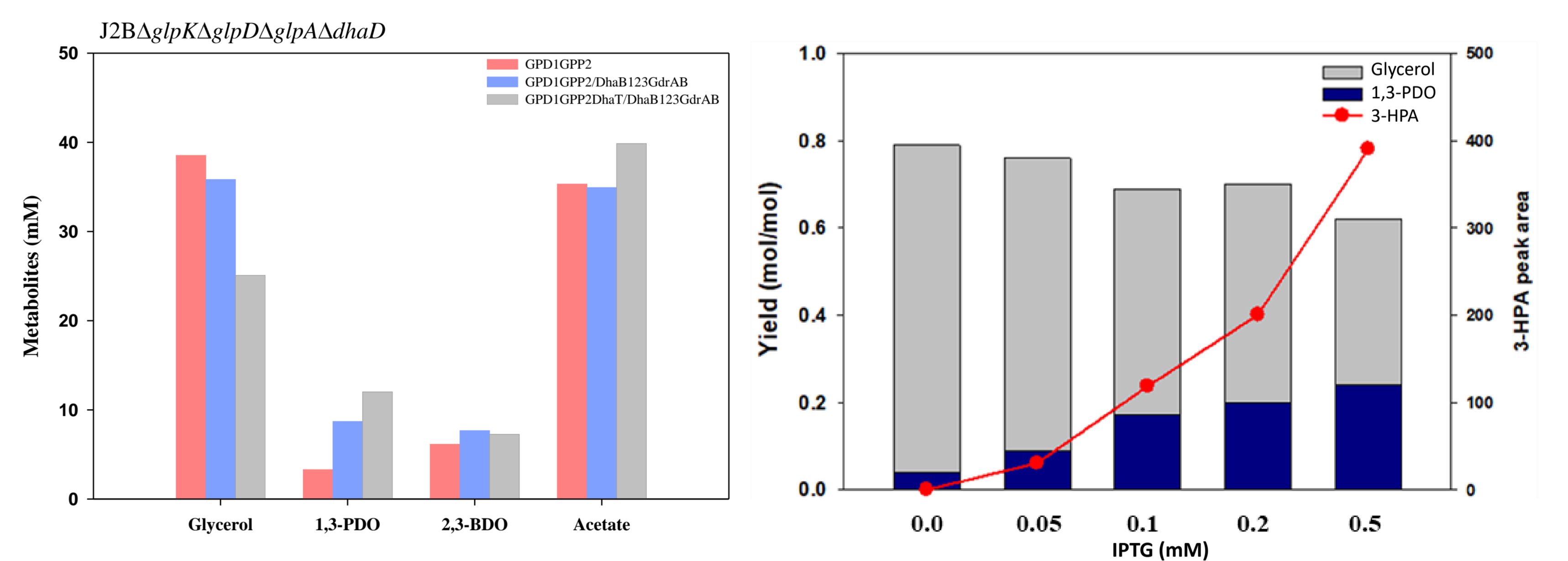


RESULTS

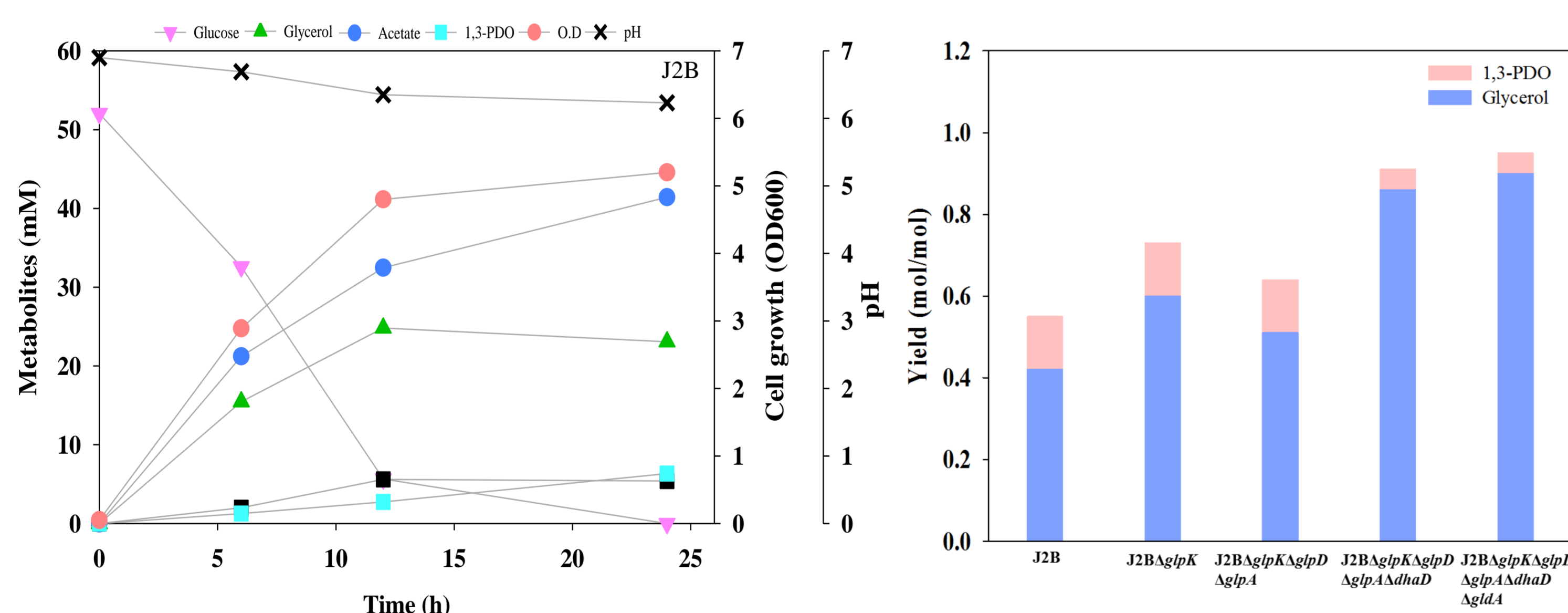
Overexpression of GPD1 and GPP2 in *K. pneumoniae* J2B



Effect of overexpression of glycerol dehydratase and oxidoreductase

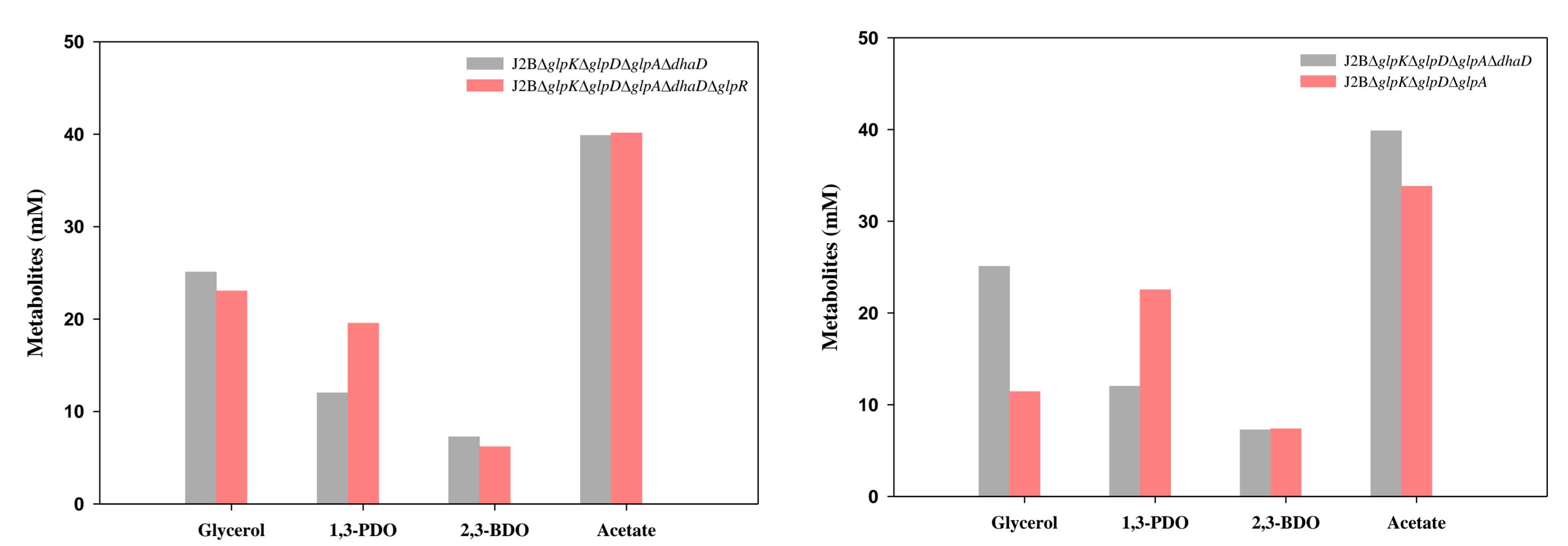


Improvement in production of glycerol by *K. pneumoniae* J2B deletion mutants



- ✓ With overexpression of GPD1 and GPP2, glycerol was produced.
- ✓ Deletion of glycerol oxidative pathway further improved the glycerol production.
- ✓ However, 1,3-PDO production was very low.
- ✓ Low 1,3-PDO production: coenzyme B₁₂ supply / expression of *dha* regulon / overflow metabolism.

Enhancement in production of 1,3-PDO by improving glycerol transport and NADH supply



- ✓ Improvement of glycerol transport increased the 1,3-PDO production.
- ✓ Increment in NADH availability also improved the 1,3-PDO production.
- ✓ Acetate accumulation was significant in all strains.

CONCLUSION

- *Klebsiella pneumoniae* J2B was engineered to produce glycerol by introducing the glycerol biosynthetic enzymes GPD1 and GPP2 from *S. cerevisiae*.
- The glycerol yield was improved by deleting glycerol oxidative pathway.
- The resulting strains successfully produced glycerol and 1,3-PDO.
- To improve the production of 1,3-PDO, further modification of the host strain is needed.

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ACKNOWLEDGEMENT

This study was supported financially by the Korean Ministry of Education, Science and Technology through the Advanced Biomass R&D center (ABC Grant No. 2011-0031361), KAIST, Korea. In addition, the authors are grateful for the financial assistance provided by the BK21 Plus Program for Advanced Chemical Technology at Pusan National University.