

DAFTAR PUSTAKA

- Ahmad, T., Aadil, R. M., Ahmed, H., Rahman, U. ur, Soares, B. C. V., Souza, S. L. Q., Pimentel, T. C., Scudino, H., Guimarães, J. T., Esmerino, E. A., Freitas, M. Q., Almada, R. B., Vendramel, S. M. R., Silva, M. C., & Cruz, A. G. (2019). Treatment and utilization of dairy industrial waste: A review. *Trends in Food Science and Technology*, 88(12), 361–372. <https://doi.org/10.1016/j.tifs.2019.04.003>
- Albuquerque, P. B. S., & Malafaia, C. B. (2018). Perspectives on the production, structural characteristics and potential applications of bioplastics derived from polyhydroxyalkanoates. *International Journal of Biological Macromolecules*, 107(PartA), 615–625. <https://doi.org/10.1016/j.ijbiomac.2017.09.026>
- Alshehrei, F. (2017). Biodegradation of Synthetic and Natural Plastic by Microorganism. *Journal of Applied & Environmental Microbiology*. *Journal of Applied & Environmental Microbiology*, 5(1), 8–1(1), 8–19. <https://doi.org/10.12691/jaem-5-1-2>
- Aristya, G. R., Lin, Y. J., Chang, J. S., Chang, J. J., & Yen, H. W. (2022). Polyhydroxybutyrate (PHB) production from crude glycerol by genetic engineering of *Rhodotorula glutinis*. *Bioresource Technology Reports*, 18(April), 101048. <https://doi.org/10.1016/j.biteb.2022.101048>
- Arumugam, A., Senthamizhan, S. G., Ponnusami, V., & Sudalai, S. (2018). Production and optimization of polyhydroxyalkanoates from non-edible *Calophyllum inophyllum* oil using *Cupriavidus necator*. *International Journal of Biological Macromolecules*, 112, 598–607. <https://doi.org/10.1016/j.ijbiomac.2018.02.012>
- Asiri, F. (2024). Polyhydroxyalkanoates for Sustainable Aquaculture: A Review of Recent Advancements, Challenges, and Future Directions. *Journal of Agricultural and Food Chemistry*, 72(4), 2034–2058. <https://doi.org/10.1021/acs.jafc.3c06488>
- Awoyera, P. O., & Adesina, A. (2020). Plastic wastes to construction products: Status, limitations and future perspective. *Case Studies in Construction Materials*, 12, e00330. <https://doi.org/10.1016/j.cscm.2020.e00330>
- Barati, F., Asgarani, E., Gharavi, S., & Soudi, M. R. (2021). Considerable increase in Poly(3-hydroxybutyrate) production via phbC gene overexpression in *Ralstonia eutropha* PTCC 1615. *BioImpacts*, 11(1), 53–57. <https://doi.org/10.34172/BI.2021.07>
- Batumalaie, K., Khalili, E., Mahat, N. A., Huyop, F. Z., & Wahab, R. A. (2018). A statistical approach for optimizing the protocol for overexpressing lipase KV1 in *Escherichia coli*: purification and characterization. *Biotechnology and Biotechnological Equipment*, 32(1), 69–87. <https://doi.org/10.1080/13102818.2017.1407670>
- Bauer, R. J., Zhelkovsky, A., Bilotti, K., Crowell, L. E., Evans, T. C., McReynolds, L. A., & Lohman, G. J. S. (2017). Comparative analysis of the end-joining activity of several DNA ligases. *PLoS ONE*, 12(12), 1–20. <https://doi.org/10.1371/journal.pone.0190062>
- Benachour, A., Auffray, Y., & Hartke, A. (2007). Construction of plasmid vectors for screening replicons from gram-positive bacteria and their use as shuttle cloning vectors. *Current Microbiology*, 54(5), 342–347. <https://doi.org/10.1007/s00284-006-0358-1>
- Bernadus, Z. G., Fatimawali, F., & Kolondam, B. (2019). Transformasi Plasmid yang Mengandung Gen *merB* pada *Escherichia coli* BL21(DE3). *Pharmakon*, 8(1), 196. <https://doi.org/10.35799/pha.8.2019.29254>
- Bharti, S., & Swetha, G. (2016). Need for Bioplastics and Role of Biopolymer PHB: A Short Review. *Journal of Petroleum & Environmental Biotechnology*, 07(02), 2–4. <https://doi.org/10.4172/2157-7463.1000272>
- Bilás, R., Szafran, K., Hnatuszko-Konka, K., & Kononowicz, A. K. (2016). Cis-regulatory elements used to control gene expression in plants. *Plant Cell, Tissue and Organ Culture*, 127(2), 269–287. <https://doi.org/10.1007/s11240-016-1057-7>



- Brabcová, K. P., Jamborová, Z., Michaelidesová, A., Davidková, M., Kodaira, S., Šefl, M., & Štěpán, V. (2019). Radiation-Induced Plasmid DNA Damage: Effect of Concentration and Length. *Radiation Protection Dosimetry*, 186(2–3), 168–171. <https://doi.org/10.1093/rpd/ncz196>
- Bragança, C. R. S., Colombo, L. T., Roberti, A. S., Alvim, M. C. T., Cardoso, S. A., Reis, K. C. P., de Paula, S. O., da Silveira, W. B., & Passos, F. M. L. (2015). Construction of recombinant *Kluyveromyces marxianus* UFV-3 to express dengue virus type 1 nonstructural protein 1 (NS1). *Applied Microbiology and Biotechnology*, 99(3), 1191–1203. <https://doi.org/10.1007/s00253-014-5963-5>
- Bugnicourt, E., Cinelli, P., Lazzeri, A., & Alvarez, V. (2014). Polyhydroxyalkanoate (PHA): Review of synthesis, characteristics, processing and potential applications in packaging. *Express Polymer Letters*, 8(11), 791–808. <https://doi.org/10.3144/expresspolymlett.2014.82>
- Carr, I. M., Robinson, J. I., Dimitriou, R., Markham, A. F., Morgan, A. W., & Bonthron, D. T. (2009). Inferring relative proportions of DNA variants from sequencing electropherograms. *Bioinformatics*, 25(24), 3244–3250. <https://doi.org/10.1093/bioinformatics/btp583>
- Castillo, T., Flores, C., Segura, D., Espín, G., Sanguino, J., Cabrera, E., Barreto, J., Díaz-Barrera, A., & Peña, C. (2017). Production of polyhydroxybutyrate (PHB) of high and ultra-high molecular weight by *Azotobacter vinelandii* in batch and fed-batch cultures. *Journal of Chemical Technology and Biotechnology*, 92(7), 1809–1816. <https://doi.org/10.1002/jctb.5182>
- Chakravarthi, B. V. S. K., Singh, S., Kamalraj, S., Gupta, V. K., & Jayabaskaran, C. (2020). Evaluation of spore inoculum and confirmation of pathway genetic blueprint of T13 α H and DBAT from a Taxol-producing endophytic fungus. *Scientific Reports*, 10(1), 1–11. <https://doi.org/10.1038/s41598-020-77605-x>
- Chen, G. Q., Chen, X. Y., Wu, F. Q., & Chen, J. C. (2020). Polyhydroxyalkanoates (PHA) toward cost competitiveness and functionality. *Advanced Industrial and Engineering Polymer Research*, 3(1), 1–7. <https://doi.org/10.1016/j.aiepr.2019.11.001>
- Chen, G. Q., & Jiang, X. R. (2018). Engineering microorganisms for improving polyhydroxyalkanoate biosynthesis. *Current Opinion in Biotechnology*, 53, 20–25. <https://doi.org/10.1016/j.copbio.2017.10.008>
- Cheng, Y.-L., Lee, C.-Y., Huang, Y.-L., Buckner, C. A., Lafrenie, R. M., Dénomée, J. A., Caswell, J. M., Want, D. A., Gan, G. G., Leong, Y. C., Bee, P. C., Chin, E., Teh, A. K. H., Picco, S., Villegas, L., Tonelli, F., Merlo, M., Rigau, J., Diaz, D., ... Mathijssen, R. H. J. (2016). PCR Techniques and Their Clinical Applications. *Intech*, 11(4), 13. <https://www.intechopen.com/books/advanced-biometric-technologies/liveness-detection-in-biometrics>
- Chinaglia, S., Tosin, M., & Degli-Innocenti, F. (2018). Biodegradation rate of biodegradable plastics at molecular level. *Polymer Degradation and Stability*, 147(October 2017), 237–244. <https://doi.org/10.1016/j.polymdegradstab.2017.12.011>
- Colussi, P. A., & Taron, C. H. (2005). *Kluyveromyces lactis* LAC4 promoter variants that lack function in bacteria but retain full function in *K. lactis*. *Applied and Environmental Microbiology*, 71(11), 7092–7098. <https://doi.org/10.1128/AEM.71.11.7092-7098.2005>
- Cranenburgh, R. M. (2004). An equation for calculating the volumetric ratios required in a ligation reaction. *Applied Microbiology and Biotechnology*, 65(2), 200–202. <https://doi.org/10.1007/s00253-004-1577-7>
- Crépin, L., Lombard, E., & Guillouet, S. E. (2016). Metabolic engineering of *Cupriavidus necator* for heterotrophic and autotrophic alka(e)ne production. *Metabolic Engineering*, 37, 92–101. <https://doi.org/10.1016/j.ymben.2016.05.002>



- Di Felice, F., Micheli, G., & Camilloni, G. (2019). Restriction enzymes and their use in molecular biology: An overview. *Journal of Biosciences*, 44(2). <https://doi.org/10.1007/s12038-019-9856-8>
- Dietrich, K., Oliveira-Filho, E. R., Dumont, M. J., Gomez, J. G. C., Taciro, M. K., Silva, L. F. da, Orsat, V., & Rio, L. F. D. (2020). Increasing PHB production with an industrially scalable hardwood hydrolysate as a carbon source. *Industrial Crops and Products*, 154(December 2019), 112703. <https://doi.org/10.1016/j.indcrop.2020.112703>
- El-kadi, S. M., Elbagory, M., El-zawawy, H. A. H., & El-shaer, H. F. A. (2021). Biosynthesis of Poly- β -Hydroxybutyrate (PHB) from Different. *Polymers*, 13, 1–20.
- Engler, C., Kandzia, R., & Marillonnet, S. (2008). A one pot, one step, precision cloning method with high throughput capability. *PLoS ONE*, 3(11). <https://doi.org/10.1371/journal.pone.0003647>
- Eraslan, K., Aversa, C., Nofar, M., Barletta, M., Gisario, A., Salehiyan, R., & Goksu, Y. A. (2022). Poly(3-hydroxybutyrate-co-3-hydroxyhexanoate) (PHBH): Synthesis, properties, and applications - A review. *European Polymer Journal*, 167(November 2021), 111044. <https://doi.org/10.1016/j.eurpolymj.2022.111044>
- Evode, N., Qamar, S. A., Bilal, M., Barceló, D., & Iqbal, H. M. N. (2021). Plastic waste and its management strategies for environmental sustainability. *Case Studies in Chemical and Environmental Engineering*, 4(8), 1–8. <https://doi.org/10.1016/j.csee.2021.100142>
- Farid, N. F. S. M., Ariffin, H., Mamat, M. R. Z., Zahari, M. A. K. M., & Hassan, M. A. (2015). Non-solvent pretreatment of poly(3-hydroxybutyrate) for improved bio-based crotonic acid production. *RSC Advances*, 5(1), 33546–33553.
- Favaro, L., Basaglia, M., & Casella, S. (2019). Improving polyhydroxyalkanoate production from inexpensive carbon sources by genetic approaches: a review. *Biofuels, Bioproducts and Biorefining*, 13(1), 208–227. <https://doi.org/10.1002/bbb.1944>
- Fernandes, M., Salvador, A., Alves, M. M., & Vicente, A. A. (2020). Factors affecting polyhydroxyalkanoates biodegradation in soil. *Polymer Degradation and Stability*, 182, 109408. <https://doi.org/10.1016/j.polymdegradstab.2020.109408>
- Filiciotto, L., & Rothenberg, G. (2021). Biodegradable Plastics: Standards, Policies, and Impacts. *ChemSusChem*, 14(1), 56–72. <https://doi.org/10.1002/cssc.202002044>
- Gaffar, S., & Sumarlin, S. (2021). Analisis sekuen mtDNA COI Pari Total Biru yang didaratkan di Tempat Pendaratan Ikan Kota Tarakan. *Jurnal Harpodon Borneo*, 13(2), 80–89. <https://doi.org/10.35334/harpodon.v13i2.1835>
- Ganapathy, K., Ramasamy, R., & Dhinakaran, I. (2018). Polyhydroxybutyrate production from marine source and its application. *International Journal of Biological Macromolecules*, 111, 102–108. <https://doi.org/10.1016/j.ijbiomac.2017.12.155>
- Ghaly, A. E., Dave, D., Budge, S., & Brooks, M. S. (2010). Fish spoilage mechanisms and preservation techniques: Review. *American Journal of Applied Sciences*, 7(7), 846–864. <https://doi.org/10.3844/ajassp.2010.859.877>
- Gruss, A., & Ehrlich, S. D. (1988). Insertion of foreign DNA into plasmids from gram-positive bacteria induces formation of high-molecular-weight plasmid multimers. *Journal of Bacteriology*, 170(3), 1183–1190. <https://doi.org/10.1128/jb.170.3.1183-1190.1988>
- Gupta, S. K., & Shukla, P. (2016). Advanced technologies for improved expression of recombinant proteins in bacteria: perspectives and applications. *Critical Reviews in Biotechnology*, 36(6), 1089–1098. <https://doi.org/10.3109/07388551.2015.1084264>
- Ha, S., Shin, B., & Park, W. (2018). Lack of glyoxylate shunt dysregulates iron homeostasis in pseudomonas aeruginosa. *Microbiology (United Kingdom)*, 164(4), 587–599. <https://doi.org/10.1099/mic.0.000623>



- Hagrosta, A., Goudarzi, H., Ghalavand, Z., Faghihloo, E., & Langroudi, R. (2020). *Molecular Cloning of a New Bi-Functional Fusion Protein of Clostridium perfringens Type A Alpha and Clostridium septicum Alpha Toxin Genes in E. coli* (pp. 1–8).
- Hammer, S. K., & Avalos, J. L. (2017). Harnessing yeast organelles for metabolic engineering. *Nature Chemical Biology*, 13(8), 823–832. <https://doi.org/10.1038/nchembio.2429>
- Hassan, M. A., Bakhiet, E. K., Hussein, H. R., & Ali, S. G. (2019). Statistical optimization studies for polyhydroxybutyrate (PHB) production by novel Bacillus subtilis using agricultural and industrial wastes. *International Journal of Environmental Science and Technology*, 16(7), 3497–3512. <https://doi.org/10.1007/s13762-018-1900-y>
- Hausjell, J., Weissensteiner, J., Molitor, C., Halbwirth, H., & Spadiut, O. (2018). *E. coli* HMS174(DE3) is a sustainable alternative to BL21(DE3). *Microbial Cell Factories*, 17(1), 1–9. <https://doi.org/10.1186/s12934-018-1016-6>
- Inui, M., Suda, M., Kimura, S., Yasuda, K., Suzuki, H., Toda, H., Yamamoto, S., Okino, S., Suzuki, N., & Yukawa, H. (2008). Expression of *Clostridium acetobutylicum* butanol synthetic genes in *Escherichia coli*. *Applied Microbiology and Biotechnology*, 77(6), 1305–1316. <https://doi.org/10.1007/s00253-007-1257-5>
- Jiang, G., Hill, D. J., Kowalczyk, M., Johnston, B., Adamus, G., Irorere, V., & Radecka, I. (2016). Carbon sources for polyhydroxyalkanoates and an integrated biorefinery. *International Journal of Molecular Sciences*, 17(7). <https://doi.org/10.3390/ijms17071157>
- Joshi, S. J., & Geetha, S. J. (2019). Recent advances in biotechnology. *Recent Advances in Biotechnology*, 2(1), 1–290. <https://doi.org/10.14429/dsj.51.2297>
- Jung, H. M., Im, D. K., Lim, J. H., Jung, G. Y., & Oh, M. K. (2019). Metabolic perturbations in mutants of glucose transporters and their applications in metabolite production in *Escherichia coli*. *Microbial Cell Factories*, 18(1), 1–14. <https://doi.org/10.1186/s12934-019-1224-8>
- Khalid, S., Zia-ur-Rehman, M., Ali, S. A., Hameed, U., Khan, F., Ahmed, N., Farooq, A. M., & Haider, M. S. (2017). Construction of an infectious chimeric geminivirus by molecular cloning based on coinfection and recombination. *International Journal of Agriculture and Biology*, 19(4), 629–634. <https://doi.org/10.17957/IJAB/15.0310>
- Kocharin, K., Chen, Y., Siewers, V., & Nielsen, J. (2012). Engineering of acetyl-CoA metabolism for the improved production of polyhydroxybutyrate in *Saccharomyces cerevisiae*. *AMB Express*, 2(1), 1–11. <https://doi.org/10.1186/2191-0855-2-52>
- Kumar, G., Bakonyi, P., Kobayashi, T., Xu, K. Q., Sivagurunathan, P., Kim, S. H., Buitrón, G., Nemestóthy, N., & Bélafi-Bakó, K. (2016). Enhancement of biofuel production via microbial augmentation: The case of dark fermentative hydrogen. *Renewable and Sustainable Energy Reviews*, 57, 879–891. <https://doi.org/10.1016/j.rser.2015.12.107>
- Kusnadi, H., Izzati, N., Nuryana, I., & Laksmi, A. (2023). Kloning Gen Penyandi Trehalosa Sintase dari *Arthrobacter psychrolactophilus* di *Escherichia coli* DH5- α . 1(1), 21–29.
- Lalonde, J. N., Pilania, G., & Marrone, B. L. (2024). Materials designed to degrade: structure, properties, processing, and performance relationships in polyhydroxyalkanoate biopolymers. *Polymer Chemistry*, 16(3), 235–265. <https://doi.org/10.1039/d4py00623b>
- Lee, Y. R., Nur Fitriana, H., Lee, S. Y., Kim, M. S., Moon, M., Lee, W. H., Lee, J. S., & Lee, S. (2020). Molecular profiling and optimization studies for growth and phb production conditions in rhodobacter sphaeroides. *Energies*, 13(23), 1–14. <https://doi.org/10.3390/en13236471>
- Li, M., Liu, Q., Teng, Y., Ou, L., Xi, Y., Chen, S., & Duan, G. (2019). The resistance mechanism of *Escherichia coli* induced by ampicillin in laboratory. *Infection and Drug Resistance*, 12, 2853–2863. <https://doi.org/10.2147/IDR.S221212>



- Li, Z. J., Qiao, K., Liu, N., & Stephanopoulos, G. (2017). Engineering *Yarrowia lipolytica* for poly-3-hydroxybutyrate production. *Journal of Industrial Microbiology and Biotechnology*, 44(4–5), 605–612. <https://doi.org/10.1007/s10295-016-1864-1>
- Lopes, C., dos Santos, N. V., Dupont, J., Pedrolli, D. B., Valentini, S. R., de Carvalho Santos-Ebinuma, V., & Pereira, J. F. B. (2019). Improving the cost effectiveness of enhanced green fluorescent protein production using recombinant *Escherichia coli* BL21 (DE3): Decreasing the expression inducer concentration. *Biotechnology and Applied Biochemistry*, 66(4), 527–536. <https://doi.org/10.1002/bab.1749>
- Lopez-Arenas, T., González-Contreras, M., Anaya-Reza, O., & Sales-Cruz, M. (2017). Analysis of the fermentation strategy and its impact on the economics of the production process of PHB (polyhydroxybutyrate). *Computers and Chemical Engineering*, 107, 140–150. <https://doi.org/10.1016/j.compchemeng.2017.03.009>
- Lozano Terol, G., Gallego-Jara, J., Sola Martínez, R. A., Martínez Vivancos, A., Cánovas Díaz, M., & de Diego Puente, T. (2021). Impact of the Expression System on Recombinant Protein Production in *Escherichia coli* BL21. *Frontiers in Microbiology*, 12(6), 1–12. <https://doi.org/10.3389/fmicb.2021.682001>
- Madeira-Jr, J. V., & Gombert, A. K. (2018). Towards high-temperature fuel ethanol production using *Kluyveromyces marxianus*: On the search for plug-in strains for the Brazilian sugarcane-based biorefinery. *Biomass and Bioenergy*, 119(August), 217–228. <https://doi.org/10.1016/j.biombioe.2018.09.010>
- Markl, E., Grünbichler, H., & Lackner, M. (2018). PHB - Bio Based and Biodegradable Replacement for PP: A Review. *Novel Techniques in Nutrition & Food Science*, 2(5), 206–209. <https://doi.org/10.31031/ntnf.2018.02.000546>
- Martin, W. F. (2020). Older Than Genes: The Acetyl CoA Pathway and Origins. *Frontiers in Microbiology*, 11(June), 1–21. <https://doi.org/10.3389/fmicb.2020.00817>
- Mazur, P., Katkov, I. I., Katkova, N., & Critser, J. K. (2000). The enhancement of the ability of mouse sperm to survive freezing and thawing by the use of high concentrations of glycerol and the presence of an *Escherichia coli* membrane preparation (Oxyrase) to lower the oxygen concentration. *Cryobiology*, 40(3), 187–209. <https://doi.org/10.1006/cryo.2000.2238>
- McAdam, B., Fournet, M. B., McDonald, P., & Mojicevic, M. (2020). Production of polyhydroxybutyrate (PHB) and factors impacting its chemical and mechanical characteristics. *Polymers*, 12(12), 1–20. <https://doi.org/10.3390/polym12122908>
- Mierziak, J., Burgberger, M., & Wojtasik, W. (2021). 3-Hydroxybutyrate As a Metabolite and a Signal Molecule Regulating Processes of Living Organisms. *Biomolecules*, 11(3), 1–21. <https://doi.org/10.3390/biom11030402>
- Moshood, T. D., Nawadir, G., Mahmud, F., Mohamad, F., Ahmad, M. H., & AbdulGhani, A. (2022). Biodegradable plastic applications towards sustainability: A recent innovations in the green product. *Cleaner Engineering and Technology*, 6, 100404. <https://doi.org/10.1016/j.clet.2022.100404>
- Mozumder, M. S. I. (2015). *Optimization of a two-phase fermentation process for the production of polyhydroxybutyrate (PHB) from organic and inorganic (industrial waste) substrate.*
- Muhammadi, Shabina, Afzal, M., & Hameed, S. (2015). Bacterial polyhydroxyalkanoates-eco-friendly next generation plastic: Production, biocompatibility, biodegradation, physical properties and applications. *Green Chemistry Letters and Reviews*, 8(3–4), 56–77. <https://doi.org/10.1080/17518253.2015.1109715>
- Mukherjee, A., & Koller, M. (2023). Microbial PolyHydroxyAlkanoate (PHA) Biopolymers—Intrinsically Natural. *Bioengineering*, 10(7), 1–16. <https://doi.org/10.3390/bioengineering10070855>



- Müller-Santos, M., Koskimäki, J. J., Alves, L. P. S., De Souza, E. M., Jendrossek, D., & Pirttilä, A. M. (2021). The protective role of PHB and its degradation products against stress situations in bacteria. *FEMS Microbiology Reviews*, 45(3), 1–13. <https://doi.org/10.1093/femsre/fuaa058>
- Najim, S. M., Al-Noor, J. M., & Al-Waely, W. A. (2015). Extraction of crude peptone from fish wastes for use as a nitrogen source in microbiological media. *Global Journal of Fisheries and Aquaculture Researches*, 2, 29–37.
- Nandy, S. K., & Srivastava, R. K. (2018). A review on sustainable yeast biotechnological processes and applications. *Microbiological Research*, 207(August 2017), 83–90. <https://doi.org/10.1016/j.micres.2017.11.013>
- Nath, P. C., Sharma, R., Debnath, S., Sharma, M., Inbaraj, B. S., Dikkala, P. K., Nayak, P. K., & Sridhar, K. (2023). Recent trends in polysaccharide-based biodegradable polymers for smart food packaging industry. *International Journal of Biological Macromolecules*, 253(P8), 127524. <https://doi.org/10.1016/j.ijbiomac.2023.127524>
- Neuman, N., Kostic, M., Scheffmann, P., & Gibson, T. (2014). Contents, Ed. Board. *Trends in Biochemical Sciences*, 39(8), i. [https://doi.org/10.1016/s0968-0004\(14\)00112-1](https://doi.org/10.1016/s0968-0004(14)00112-1)
- Noor, S., Pramono, H., & Aziz, S. (2014). Deteksi Keragaman Spesies Bakteri Metanogen Rumen Sapi Menggunakan Kloning Gen 16S rRNA dan Sekuensing. *Scripta Biologica*, 1(4), 1–8.
- Nora, L. C., Westmann, C. A., Martins-Santana, L., Alves, L. de F., Monteiro, L. M. O., Guazzaroni, M. E., & Silva-Rocha, R. (2019). The art of vector engineering: towards the construction of next-generation genetic tools. *Microbial Biotechnology*, 12(1), 125–147. <https://doi.org/10.1111/1751-7915.13318>
- Nouban, F., & Abazid, M. (2017). The principle and application of new PCR technologies. *IOP Conference Series: Earth and Environmental Science*, 8(2), 68–74. <https://doi.org/10.1088/1755-1315>
- Olavarria, K., Carnet, A., van Renselaar, J., Quakkelaar, C., Cabrera, R., Guedes da Silva, L., Smids, A. L., Villalobos, P. A., van Loosdrecht, M. C. M., & Wahl, S. A. (2021). An NADH preferring acetoacetyl-CoA reductase is engaged in poly-3-hydroxybutyrate accumulation in *Escherichia coli*. *Journal of Biotechnology*, 325(July 2020), 207–216. <https://doi.org/10.1016/j.jbiotec.2020.10.022>
- Partridge, S. R., Kwong, S. M., Firth, N., & Jensen, S. O. (2018). Mobile genetic elements associated with antimicrobial resistance. *Clinical Microbiology Reviews*, 31(4). <https://doi.org/10.1128/CMR.00088-17>
- Patigu, R. F., Wijayanti, P., Sebastian, A., & Purwestri, Y. A. (2021). Optimization of heat shock temperature and time on the transformation of pRGEB32 into *Escherichia coli* DH5 α . *Jurnal Biologi Tropis*, 21(3), 632–640. <https://doi.org/10.29303/jbt.v21i3.2811>
- Rajan, K. P., Thomas, S. P., Gopanna, A., & Chavali, M. (2019). Polyhydroxybutyrate (PHB): A standout biopolymer for environmental sustainability. *Handbook of Ecomaterials*, 4, 2803–2826. https://doi.org/10.1007/978-3-319-68255-6_92
- Rianti, E. D. D., Tania, P. O. A., & Listyawati, A. F. (2022). Kuat medan listrik AC dalam menghambat pertumbuhan koloni *Staphylococcus aureus* dan *Escherichia coli*. *Bioma : Jurnal Ilmiah Biologi*, 11(1), 79–88. <https://doi.org/10.26877/bioma.v11i1.9561>
- Rivera-Briso, A. L., & Serrano-Aroca, Á. (2018). Poly(3-Hydroxybutyrate-co-3-Hydroxyvalerate): Enhancement strategies for advanced applications. *Polymers*, 10(7), 1–28. <https://doi.org/10.3390/polym10070732>
- Roberts, L. A., & Shell, S. S. (2023). A research program-linked, course-based undergraduate research experience that allows undergraduates to participate in current research on

- mycobacterial gene regulation. *Frontiers in Microbiology*, 13(January). <https://doi.org/10.3389/fmicb.2022.1025250>
- Rodríguez-Beltrán, J., DelaFuente, J., León-Sampedro, R., MacLean, R. C., & San Millán, Á. (2021). Beyond horizontal gene transfer: the role of plasmids in bacterial evolution. *Nature Reviews Microbiology*, 19(6), 347–359. <https://doi.org/10.1038/s41579-020-00497-1>
- Roohi, Zaheer, M. R., & Kuddus, M. (2018). PHB (poly-β-hydroxybutyrate) and its enzymatic degradation. *Polymers for Advanced Technologies*, 29(1), 30–40. <https://doi.org/10.1002/pat.4126>
- Rosano, G. L., & Ceccarelli, E. A. (2014). Recombinant protein expression in *Escherichia coli*: Advances and challenges. *Frontiers in Microbiology*, 5(4), 1–17. <https://doi.org/10.3389/fmicb.2014.00172>
- Rotinsulu, S., Fatimawali, F., & Tallei, T. E. (2019). Transformasi Plasmid yang Mengandung Gen *merB* pada Bakteri *Escherichia coli* TOP-10. *Pharmakon*, 8(2), 290. <https://doi.org/10.35799/pha.8.2019.29294>
- Rujnić-Sokele, M., & Pilipović, A. (2017). Challenges and opportunities of biodegradable plastics: A mini review. *Waste Management and Research*, 35(2), 132–140. <https://doi.org/10.1177/0734242X16683272>
- Saini, P., Beniwal, A., Kokkiligadda, A., & Vij, S. (2018). Response and tolerance of yeast to changing environmental stress during ethanol fermentation. *Process Biochemistry*, 72(July), 1–12. <https://doi.org/10.1016/j.procbio.2018.07.001>
- Sakihama, Y., Hidese, R., Hasunuma, T., & Kondo, A. (2019). Increased flux in acetyl-CoA synthetic pathway and TCA cycle of *Kluyveromyces marxianus* under respiratory conditions. *Scientific Reports*, 9(1), 1–7. <https://doi.org/10.1038/s41598-019-41863-1>
- Sakthiselvan, P., & Madhumathi, R. (2019). Optimization on Microbial Production of Polyhydroxybutyrate (PHB): A Review. *International Journal of Research and Analytical Reviews*, 6(1), 243–251.
- Saleem, J., Adil Riaz, M., & Gordon, M. (2018). Oil sorbents from plastic wastes and polymers: A review. *Journal of Hazardous Materials*, 341, 424–437. <https://doi.org/10.1016/j.jhazmat.2017.07.072>
- Samori, C., Martinez, G. A., Bertin, L., Pagliano, G., Parodi, A., Torri, C., & Galletti, P. (2022). PHB into PHB: Recycling of polyhydroxybutyrate by a tandem “thermolytic distillation-microbial fermentation” process. *Resources, Conservation and Recycling*, 178(11), 106082. <https://doi.org/10.1016/j.resconrec.2021.106082>
- Sanderson, H., McCarthy, M. C., Nnajide, C. R., Sparrow, J., Rubin, J. E., Dillon, J. A. R., & White, A. P. (2023). Identification of plasmids in avian-associated *Escherichia coli* using nanopore and illumina sequencing. *BMC Genomics*, 24(1), 1–14. <https://doi.org/10.1186/s12864-023-09784-6>
- Sandström, A. G., Muñoz de las Heras, A., Portugal-Nunes, D., & Gorwa-Grauslund, M. F. (2015). Engineering of *Saccharomyces cerevisiae* for the production of poly-3-d-hydroxybutyrate from xylose. *AMB Express*, 5(1). <https://doi.org/10.1186/s13568-015-0100-0>
- Schadeweg, V., & Boles, E. (2016). N-Butanol production in *Saccharomyces cerevisiae* is limited by the availability of coenzyme A and cytosolic acetyl-CoA. *Biotechnology for Biofuels*, 9(1), 1–12. <https://doi.org/10.1186/s13068-016-0456-7>
- Schwarzenbach, H., Da Silva, A. M., Calin, G., & Pantel, K. (2015). Data normalization strategies for microRNA quantification. *Clinical Chemistry*, 61(11), 1333–1342. <https://doi.org/10.1373/clinchem.2015.239459>
- Segawa, M., Wen, C., Orita, I., Nakamura, S., & Fukui, T. (2019). Two NADH-dependent (S)-3-hydroxyacyl-CoA dehydrogenases from polyhydroxyalkanoate-producing

- Ralstonia eutropha*. *Journal of Bioscience and Bioengineering*, 127(3), 294–300. <https://doi.org/10.1016/j.jbiosc.2018.08.009>
- Segeritz, C. P., & Vallier, L. (2017). Cell Culture: Growing Cells as Model Systems In Vitro. In *Basic Science Methods for Clinical Researchers*. Elsevier Inc. <https://doi.org/10.1016/B978-0-12-803077-6.00009-6>
- Sha, Y., Ge, M., Lu, M., Xu, Z., Zhai, R., & Jin, M. (2025). Advances in metabolic engineering for enhanced acetyl-CoA availability in yeast. *Critical Reviews in Biotechnology*, 45(4), 904–922. <https://doi.org/10.1080/07388551.2024.2399542>
- Sharma, R., Garg, P., Kumar, P., Bhatia, S. K., & Kulshrestha, S. (2020). Microbial fermentation and its role in quality improvement of fermented foods. *Fermentation*, 6(4), 1–20. <https://doi.org/10.3390/fermentation6040106>
- Sharma, V., Sehgal, R., & Gupta, R. (2021). Polyhydroxyalkanoate (PHA): Properties and Modifications. *Polymer*, 212(September 2020), 123161. <https://doi.org/10.1016/j.polymer.2020.123161>
- Shearer, R. F., & Saunders, D. N. (2015). Experimental design for stable genetic manipulation in mammalian cell lines: Lentivirus and alternatives. *Genes to Cells*, 20(1), 1–10. <https://doi.org/10.1111/gtc.12183>
- Shen, R., Yin, J., Ye, J. W., Xiang, R. J., Ning, Z. Y., Huang, W. Z., & Chen, G. Q. (2018). Promoter Engineering for Enhanced P(3HB-co-4HB) Production by *Halomonas bluephagenesis*. *ACS Synthetic Biology*, 7(8), 1897–1906. <https://doi.org/10.1021/acssynbio.8b00102>
- Singh, V., Haque, S., Niwas, R., Srivastava, A., Pasupuleti, M., & Tripathi, C. K. M. (2017). Strategies for fermentation medium optimization: An in-depth review. *Frontiers in Microbiology*, 7(JAN). <https://doi.org/10.3389/fmicb.2016.02087>
- Sirohi, R., Prakash Pandey, J., Kumar Gaur, V., Gnansounou, E., & Sindhu, R. (2020). Critical overview of biomass feedstocks as sustainable substrates for the production of polyhydroxybutyrate (PHB). *Bioresource Technology*, 311(March), 123536. <https://doi.org/10.1016/j.biortech.2020.123536>
- Stevenson, J., Krycer, J. R., Phan, L., & Brown, A. J. (2013). A practical comparison of ligation-independent cloning techniques. *PLoS ONE*, 8(12), 8–14. <https://doi.org/10.1371/journal.pone.0083888>
- Suea-Ngam, A., Bezingue, L., Mateescu, B., Howes, P. D., Demello, A. J., & Richards, D. A. (2020). Enzyme-Assisted Nucleic Acid Detection for Infectious Disease Diagnostics: Moving toward the Point-of-Care. *ACS Sensors*, 5(9), 2701–2723. <https://doi.org/10.1021/acssensors.0c01488>
- Tang, H. J., Neoh, S. Z., & Sudesh, K. (2022). A review on poly(3-hydroxybutyrate-co-3-hydroxyhexanoate) [P(3HB-co-3HHx)] and genetic modifications that affect its production. *Frontiers in Bioengineering and Biotechnology*, 10(December). <https://doi.org/10.3389/fbioe.2022.1057067>
- Tang, R., Weng, C., Peng, X., & Han, Y. (2020). Metabolic engineering of *Cupriavidus necator* H16 for improved chemoautotrophic growth and PHB production under oxygen-limiting conditions. *Metabolic Engineering*, 61(January), 11–23. <https://doi.org/10.1016/j.ymben.2020.04.009>
- Tao, Z., Yuan, H., Liu, M., Liu, Q., Zhang, S., Liu, H., Jiang, Y., Huang, D., & Wang, T. (2023). Yeast Extract: Characteristics, Production, Applications and Future Perspectives. *Journal of Microbiology and Biotechnology*, 33(1), 151–166. <https://doi.org/10.4014/jmb.2207.07057>
- Tarrahi, R., Fathi, Z., Seydibeyoğlu, M. Ö., Doustkhah, E., & Khataee, A. (2020). Polyhydroxyalkanoates (PHA): From production to nanoarchitecture. *International*

- Thapa, C., Shakya, P., Shrestha, R., Pal, S., & Manandhar, P. (2019). Isolation of Polyhydroxybutyrate (PHB) Producing Bacteria, Optimization of Culture Conditions for PHB production, Extraction and Characterization of PHB. *Nepal Journal of Biotechnology*, 6(1), 62–68. <https://doi.org/10.3126/njb.v6i1.22339>
- Vashishth, A., & Tehri, N. (2015). The role of recombinant DNA technology for human welfare. *International Journal of Research in Biological Sciences*, 5(4), 35–39.
- Wagner, J. C., Goldfless, S. J., Ganesan, S. M., Lee, M. C. S., Fidock, D. A., & Niles, J. C. (2013). An integrated strategy for efficient vector construction and multi-gene expression in *Plasmodium falciparum*. *Malaria Journal*, 12(1), 1–2. <https://doi.org/10.1186/1475-2875-12-373>
- Wahyuni, F. D., Sidiq, M. A., Seprianto, S., & Saraswati, H. (2022). Transformasi Plasmid Rekombinan pRI_101-AN Membawa Sisispan Gen *cryIII* melalui *Agrobacterium tumefaciens*. *Berita Biologi*, 21(1), 71–78. <https://doi.org/10.14203/beritabiologi.v21i3.4256>
- Wang, Y., Yin, J., & Chen, G. Q. (2014). Polyhydroxyalkanoates, challenges and opportunities. *Current Opinion in Biotechnology*, 30, 59–65. <https://doi.org/10.1016/j.copbio.2014.06.001>
- Wehantouw, A., Ginting, E., & Wullur, S. (2017). Identifikasi sirip ikan hiu yang didapat dari pengumpul di Minahasa Tenggara menggunakan DNA Barcode. *Jurnal Pesisir Dan Laut Tropis*, 5(1), 62. <https://doi.org/10.35800/jplt.5.1.2017.15007>
- Wiharyani, R., Hardianto, D., Kusumaningrum, H. P., & Budiharjo, A. (2014). Kloning Gen *pcbC* dari *Penicillium chrysogenum* ke dalam Plasmid pPICZA untuk Pengembangan Produksi Penisilin G. *Bioma: Berkala Ilmiah Biologi*, 16(1), 33. <https://doi.org/10.14710/bioma.16.1.33-38>
- Wu, H., Chen, J., & Chen, G. Q. (2016). Engineering the growth pattern and cell morphology for enhanced PHB production by *Escherichia coli*. *Applied Microbiology and Biotechnology*, 100(23), 9907–9916. <https://doi.org/10.1007/s00253-016-7715-1>
- Yang, C., Hu, S., Zhu, S., Wang, D., Gao, X., & Hong, J. (2015). Characterizing yeast promoters used in *Kluyveromyces marxianus*. *World Journal of Microbiology and Biotechnology*, 31(10), 1641–1646. <https://doi.org/10.1007/s11274-015-1899-x>
- Ylinen, A., de Ruijter, J. C., Jouhten, P., & Penttilä, M. (2022). PHB production from cellobiose with *Saccharomyces cerevisiae*. *Microbial Cell Factories*, 21(1), 1–13. <https://doi.org/10.1186/s12934-022-01845-x>
- Zeka, F., Vanderheyden, K., De Smet, E., Cuvelier, C. A., Mestdagh, P., & Vandesomepele, J. (2016). Straightforward and sensitive RT-qPCR based gene expression analysis of FFPE samples. *Scientific Reports*, 6(1), 2–11. <https://doi.org/10.1038/srep21418>
- Zhang, B., Ren, L., Wang, Y., Xu, D., Zhang, S., Wang, H., Wang, H., Zeng, X., Xin, B., & Li, F. (2020). Glycerol production through TPI1 defective *Kluyveromyces marxianus* at high temperature with glucose, fructose, and xylose as feedstock. *Biochemical Engineering Journal*, 161(April), 107689. <https://doi.org/10.1016/j.bej.2020.107689>
- Zhang, B., Zhu, Y., Zhang, J., Wang, D., Sun, L., & Hong, J. (2017). Engineered *Kluyveromyces marxianus* for pyruvate production at elevated temperature with simultaneous consumption of xylose and glucose. *Bioresource Technology*, 224, 553–562. <https://doi.org/10.1016/j.biortech.2016.11.110>
- Zhang, H., Ma, S., Dou, X., Chen, R., Lu, H., Chi, Z., Xue, S., Li-Beisson, Y., & Kong, F. (2024). Harnessing Algal Peroxisomes for Efficient Poly Hydroxybutyrate Production. In *ACS Sustainable Chemistry and Engineering* (Vol. 12, Issue 8). <https://doi.org/10.1021/acssuschemeng.3c07974>



Strategi Produksi Polyhydroxybutyrate melalui Overekspresi Gen Pengkode PHB pada *Kluyveromyces marxianus* IFO 1735

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- Zhang, M., Kurita, S., Orita, I., Nakamura, S., & Fukui, T. (2019). Modification of acetoacetyl-CoA reduction step in *Ralstonia eutropha* for biosynthesis of poly(3-hydroxybutyrate-co-3-hydroxyhexanoate) from structurally unrelated compounds. *Microbial Cell Factories*, 18(1), 1–12. <https://doi.org/10.1186/s12934-019-1197-7>
- Zhang, Q., Wu, D., Lin, Y., Wang, X., Kong, H., & Tanaka, S. (2015). Substrate and product inhibition on yeast performance in ethanol fermentation. *Energy and Fuels*, 29(2), 1019–1027. <https://doi.org/10.1021/ef502349v>