

BAB V

KESIMPULAN DAN SARAN

V.1 Kesimpulan

1. Proses ekstraksi protein biji jarak menghasilkan protein murni bebas lemak dengan rendemen sebesar 68,36%. Ekstrak protein yang dihidrolisis enzimatis dengan menggunakan enzim tripsin menghasilkan nilai derajat hidrolisis sebesar 33,51%.
2. Fraksinasi dengan menggunakan resin penukar kation dielusi dengan menggunakan buffer pH 3-9 dimana fraksi pH 3 dan 4 diujikan pada bakteri *E.coli* dan *S.aureus* memiliki zona hambat yang paling besar diantara yang lain.
3. Nilai IC50 fraksi pH 3 dan pH 4 terhadap *S.aureus* masing-masing yaitu didapat nilai sebesar 4,25 dan 17,96 ppm. Sedangkan pengujian pH 3 dan 4 terhadap *E.coli* menghasilkan nilai sebesar 22,50 dan 25,74 ppm.
4. Hasil identifikasi peptida pada fraksi pH 3 menghasilkan urutan peptida AGVGIKPR, GAVVAPRK, GTPDDMGGAGAGNPPDDK dan ALQAPGR. Sedangkan pada fraksi pH 4 ditemukan urutan peptida AAGASG

V.2 Saran

Dari hasil penelitian ini didapat 3 urutan peptida pada fraksi pH 3 dan 1 urutan peptida pada fraksi pH 4 yang terbukti aktif antibakteri. Perlu dilakukan penelitian lanjutan dengan menggunakan metode *in silico* berupa docking molekuler maupun dengan metode *in vitro* dengan pemurnian peptida untuk menunjukan urutan peptida mana yang benar benar memiliki aktivitas.

DAFTAR PUSTAKA

- Adisasmito, A. W., dan Hadinegoro, S. R., 2004, Infeksi bakteri gram negatif di ICU anak : epidemiologi, manajemen antibiotik dan pencegahan, *Sari Pediatri*, 6(1), 32-39.
- Aditia, R. P., Desniar, Trilaksani, W., 2018, Aktivitas antioksidan dan antibakteri hidrolisat protein hasil fermentasi telur ikan cakalang, *JPHPI*, 21(1), 1-12.
- Adje, E. Y., Balti, R., Kouach, M., Dhulster, P., Guillochon, D., Nedjar-Arroume, N., 2011, Obtaining antimicrobial peptides by controlled peptic hydrolysis of bovine hemoglobin, *Int. J. Biol. Macromol.*, 49, 143-153.
- Adje, E. Y., Balti, R., Kouach, M., Dhulster, P., Guillochon, D., Nedjar-Arroume, N., 2013, Controlled enzymatic hydrolysis: a new strategy for the discovery of antimicrobial peptides, *Probiotics & Antimicro. Prot.*, 5, 176–186.
- Ahmad, A., Ahmad, E., Rabbani, G., Haque, S., Arshad, M., Hasan Khan, R., 2012, Identification and Design of Antimicrobial Peptides for Therapeutic Applications. *Curr. Protein Pept. Sci.*, 13(3), 211–223.
- Alamdari, E. K., Ehsani, M. R., 2017, Antimicrobial peptides derived from milk : A review, *J. Food sci. Technol.*, 7(1), 49-56.
- Ampou, E., Triyulianti, I., dan Nugroho, S., 2015, Bakteri asosiasi pada karang *Scleractinia* kaitannya dengan fenomena La-nina di pulau Bunaken, *Jurnal kelautan nasional*, 10(2), 55-63.
- Andrews, J. M., 2001, Determination of minimum inhibitory concentrations, *J. Antimicrob. Chemotherapy.*, 48, 5-16.
- Anonim, 2012, Buletin jendela data informasi kesehatan : penyakit tidak menular, kementerian kesehatan Republik Indonesia, ISSN 2088-270X.
- Ballatore, M. B., Bettiol, M. d. R., Braber, N. L. V., Aminahuel, C. A., Rosii, Y. E., Petroselli, G., Erra-Balsells, R., Cavaglieri, L. R., Montenegro, M. A., 2020, Antioxidant and cytoprotective effect of peptides produced by hydrolysis of whey protein concentrate with trypsin, *Food Chem.*, 319, 1-11.
- Berg, H.C., 2004, *E.coli* in motion, Springer-Verlag New York Inc.
- Besse, A., Peduzzi, J., Rebuffat, S., Carre-Mlouka, A., 2015, Antimicrobial peptides and proteins in the face of extremes: lessons from archaeocins, *Biochimie*, 30, 1-12.
- Betancourt, L. H., Bock, P. D., States, A., Timmerman, E., Perez-Riverol, Y., Sanchez, A., Besada, V., Gonzalez, L. J., Vandekerckhove, J., Gevaert, K., 2013, SCX charge state selective separation of tryptic peptides combined with 2D-RP-HPLC allows for detailed proteome mapping, *J. Proteom.*, 91, 164-171.
- Bilikova, K., Huang, S., Lin, I., Simuth, J., Peng, C., 2015, Structure and antimicrobial activity relationship of royalisin, an antimicrobial peptide from royal jelly of *Apis mellifera*. *Peptides*, 68, 190-196.
- Borrajao, P., Pateiro, M., Gagaoua, M., Franco, D., Zhang, W., and Lorenzo, J.M., 2020, Evaluation of the antioxidant and antimicrobial activities of porcine liver protein hydrolysates obtained using Alcalase, Bromelain, and Papain, *Appl. Sci.*, 10.

- Brogden, K. A., 2005, Antimicrobial peptides: pore formers or metabolic inhibitors in bacteria?, *Nat. Rev. Microbiol.*, 3, 238-250.
- Burgess, R. R., Deutscher, M. P., 2009, *Methods in enzymology: guide to protein purification*. 2nd ed. Vol. 436, San Diego, Academic Press.
- Burke, T. W. L., Mant, C. T. M., Black, J. A., Hodges, R. S., 1989, string cation-exchange high performance liquid chromatography of peptides : effect of non-specific hydrophobic interactions and linearization of peptide retention behavior, *J. Chromatogr. A.*, 476, 377-389.
- Burlingham, B. T., Widlanski, T. S., 2003, An Intuitive Look at the Relationship of K_i and IC_{50} : A More General Use for the Dixon Plot, *J. Chem. Edu.*, 80(2), 214-219.
- Cen, Q., Wang, Z., Tang, Z., Zhang, Y., Chen, T., Xue, D., Xu, M., Bai, X., Zhou, T., Shi, L., 2021, Initial purification of antimicrobial fermentation metabolites from *Paecilomyces cicadae* and its antimicrobial mechanism, *LWT*, 148, 1-9.
- Chakrabarti, S., Guha, S., and Majumder, K., 2018, Food-derived bioactive peptides in human health: Challenges and opportunities, *Nutrients*, 10, 1–17.
- Chambi, H.N.M., Lacerda, R.S., Makishi, G.L.A., Bittante, A.M.Q.B., Gomide, C.A., and Sobral, P.J.A., 2014, Protein extracted from castor bean (*Ricinus communis* L.) cake in high pH results in films with improved physical properties, *Ind. Crops Prod.*, 61, 217–224.
- Chen, S.C., Liu, J.W., Wu, X.Z., Cao, W., Wang, F., Huang, J.M., et al., 2020, Comparison of microdilution method with agar dilution method for antibiotic susceptibility test of *Neisseria gonorrhoeae*, *Infect. Drug Resist.*, 13, 1775–1780.
- Christenson, J. C., Korgenski, E. K., Relish, R. F., 2018, 286 - laboratory diagnosis of infection due to bacteria, fungi, parasites, and rickettsiae, *Principles and practice of pediatric infectious diseases (fifth edition)*, 1422-1434.
- Couto, N., Barber, J., Gaskell, S. J., 2011, Matrix-assisted laser desorption/ionisation mass spectrometric response factors of peptides generated using different proteolytic enzymes, *J. Mass. Spectrom.*, 46, 1233–1240.
- Dang, L., Damme, A. J. M. V., 2015, Toxic proteins in plants, *phytochemistry*, 117, 51-64.
- Danlami, J.M., Arsad, A., and Zaini, M.A.A., 2015, Characterization and process optimization of castor oil (*Ricinus communis* L.) extracted by the soxhlet method using polar and non-polar solvents, *J. Taiwan Inst. Chem. Eng.*, 47, 99–104.
- Demers-Mathieu, V., Gauthier, S.F., Britten, M., Fliss, I., Robitaille, G., and Jean, J., 2013, Antibacterial activity of peptides extracted from tryptic hydrolyzate of whey protein by nanofiltration, *Int. Dairy J.*, 28, 94–101.
- Desmachelier, P., Fegan, N., 2016, Pathogens in milk : *Escherichia coli*, Reference module in food sciences, 1-8.
- Desrini, S., 2015, Resistensi Antibiotik, Akankah Dapat Dikendalikan?, *JKKI*, 6(4), 2015

- Dias, J.M., Araújo, J.M., Costa, J.F., Alvim-Ferraz, M.C.M., and Almeida, M.F., 2013, Biodiesel production from raw castor oil, *Energy*, 53, 58–66.
- Doan, L. G., 2004, Ricin : Mechanism of toxicity, clinical manifestations, and vaccine development. A review, *J. Toxicol.*, 42(2), 201-208.
- Doellinger, J., Schneider, A., Hoeller, M., Lasch, P., 2020, Sample preparation by easy extraction and digestion (SPEED) - A universal, rapid, and detergent free protocol for proteomics based on acid extraction, *Mol. Cell. Proteom.*, 19(1), 209-222.
- Esmailpour, M., Ehsani, M. R., Aminlari, M., Shekarforoushd, Sh., Hoseini, E., 2017, Antimicrobial Peptides Derived from Goat's Milk Whey Proteins Obtained by Enzymatic Hydrolysis, *J. Food sci. Technol.*, 7(1), 65-72.
- Fernandes, P. and Martens, E., 2017, Antibiotics in late clinical development, *Biochem. Pharmacol.*, 133, 152–163.
- Forry, S. P., Madonna, M. C., Lopez-Perez, D., Lin, N. J., Pasco, M. D., 2016, Automation of antimicrobial activity screening, *AMB Express*, 6(20).
- Foster, T. J., Geoghegan, J. A., 2015, *Staphylococcus aureus*, *Mol. Microbiol.*, 655-675.
- Furtado, R. F., Alves, C. R., Moreira, A. C. O., Azevedo, R. M., Dutra, R. F., 2012, A novel xyloglucan film-based biosensor for toxicity assessment of ricin in castor seed meal, *Carbohydr. Polym.*, 89, 586-591.
- Gevaert, K., Vandekerckhove, J., 2000, protein identification methods in proteomics, *Electrophoresis*, 21, 1145-1154.
- Gnanamani, A., Hariharan, P., Paul-Satyaseela, M., 2017, *Staphylococcus aureus* : overview of bacteriology, clinical diseases, epidemiology, antibiotic resistance and therapeutic approach, *Frontier in Staphylococcus aureus*, Shymaa Enany and Laura E. Crotty Alexander, IntechOpen.
- Gottschalk, U., 2011, Overview of Downstream Processing in the Biomanufacturing Industry dalam *Comprehensive Biotechnology (Second Edition)*, reference module in life sciences, 3, 669-682.
- Gussakovsky, D., Neustaeter, H., Spicer, V., Krokhin, A. V., 2017, Sequence-Specific Model for Peptide Retention Time Prediction in Strong Cation Exchange Chromatography, *Anal. Chem.*, 89, 11795-11802.
- Guzman, M., Dille, J., Godet, S., 2012, Synthesis and antibacterial activity of silver nanoparticles against gram-positive and gram-negative bacteria, *Nanomedicine NANOMED-NANOTECHNOL.*, 8, 37-45.
- Ha, J.; Park, J.-Y.; Choi, Y.; Chang, P.-S.; Park, K.-M, 2021, Comparative Analysis of Universal Protein Extraction Methodologies for Screening of Lipase Activity from Agricultural Products, *Catalysts*, 11 (816), 1-12.
- Harscoat-Schiavo, C., Raminoso, F., Ronat-Heit, E., Vanderesse, R., Marc, I., 2010, Modeling the separation of small peptides by cation-exchange chromatography, *J. Sep. Sci.* 2010, 33, 2447–2457.
- Haslaniza H, Maskat, M. Y., Wan Aida, W. M., Mamot, S., 2010, The effects of enzyme concentration, temperature and incubation time on nitrogen content and degree of hydrolysis of protein precipitate from cockle (*Anadara granosa*) meat wash water, *Int. Food Res. J.*, 17, 147-152.

- Horvath, Gy., Bencsik, T., Acs, K., Kocsis, B., 2016, Chapter 12 - Sensitivity of ESBL-Producing gram-negative bacteria to essential oils, plant extracts, and their isolated compounds, *Antibiotic Resistance, mechanisms and new antimicrobial approaches*, 239-269.
- Jang, S. A., Kim, H., Lee, J. Y., Shin, J. R., Kim, D. J., Cho, J. H., Kim, S. C., 2012, Mechanism of action and specificity of antimicrobial peptides designed based on buforin IIb, *Peptides*, 34, 83-289.
- Jaziri, A.A., Brawijaya, U., and Brawijaya, U., 2017, Karakteristik protease dari ekstrak kasar khamir laut dan aktivitasnya dalam menghidrolisis protein ikan rucah,.
- Jiang, Z., Vasil, A.I., Hale, J. D., Hancock, R. E. W., Vasil, M. L., Hodges, R. S., 2007, Effects of Net Charge and the Number of Positively Charged Residues on the Biological Activity of Amphipathic α -Helical Cationic Antimicrobial Peptides, *peptidescience*, 90(3), 369-374.
- Jin, S. K., Choi, J. S., Yim, D., 2020, Hydrolysis condition of porcine blood proteins and antimicrobial effects of their hydrolysates, *Food. Sci. Anim. Resour.*, 40(2), 172-182.
- Jin, S.K., Choi, J. S., Yim, D. G., 2020, Hydrolysis Conditions of Porcine Blood Proteins and Antimicrobial Effects of Their Hydrolysates, *Food Sci. Anim. Resour.*, 40(2), 172-182.
- Johnson, J. R., Russo, T. A., 2002, Extraintestinal pathogenic *Escherichia coli* : “the other bad *E.coli*”, *J. Lab. Clin. Med.*, 139(3), 155-162.
- Kaur, J., Singh, P. K., 2020, Trypsin detection strategies : A review, *Crit. Rev. Anal. Chem.*, 1-20.
- Khirzin, M. H., Hilmi, M., Prastujati, A. U., Mawardi, N., Rahayu, R., 2020, Karakteristik hidrolisat gelatin tulang itik dengan enzim tripsin sebagai penghambat alfa amilase (α -amilase inhibitor), *Jurnal ilmiah INOVASI*, 20(3), 55-60.
- Kopaciewicz, W., Rounds, M. A., Fausnaugh, J., Regnier, F. E., 1983, Retention model for high-performance ion-exchange chromatography, *J. Chromatogr. A.*, 266, 3-2.
- Kusumaningtyas, E., Widiastuti, R., Dewantari Kusumaningrum, H., and Thenawidjaja Suhartono, M., 2015, Aktivitas Antibakteri Dan Antioksidan Hidrolisat Hasil Hidrolisis Protein Susu Kambing Dengan Ekstrak Kasar Bromelin, *J. Teknol. dan Ind. Pangan*, 26, 179–188.
- Lambert, R. J. W., Pearson, J., 2001, Susceptibility testing : accurate and reproducible minimum inhibitory concentration (MIC) and non-inhibitory concentration (NIC) values, *J. Appl. Microbiol.*, 88(5), 784-790.
- Lee, C. H., 2017, A Simple Outline of Methods for Protein Isolation and Purification, *Endocrinol Metab*, 32, 18-22.
- Lin, D., Tabb, D. L., Yates III, J. R., 2003, Large-scale protein identification using mass spectrometry, *Biochimica et Biophysica acta*, 1646, 1-10.
- Lord, M. J., Jolliffe, N. A., Marsden, C. J., Pateman, C. S. C., Smith, D. C., Spooner, R. A., Watson, P. D., Roberts, L. M., 2003, Ricin. Mechanisms of cytotoxicity, *Toxicol*, 22(1), 52-64.

- Mai-Prochnow, A., Clauson, M., Hong, J., Murphy, A. B., 2016, Gram positive and gram negative bacteria differ in their sensitivity to cold plasma, *Sci. Rep.*, 6, 1-11.
- Manadas, B., Mendes, V. M., English, J., Dunn, J. M., 2010, Peptide fractionation in proteomics approaches, *Expert Rev. Proteomics*, 7(5), 655-663.
- Martinez-Maqueda, D., Hernandez-Ledesma, B., Amigo, L., Miralles, B., Gomez-Ruiz, J. A., 2013, Extraction/fractionation techniques for proteins and peptides and protein digestion, dalam : Tolda, F., Nollet, L. proteomics in foods, Food microbiology and food safety, 2, Springer, Boston MA.
- Matsuzaki, K, 1999, Why and how are peptide^lipid interactions utilized for self-defense? Magainins and tachyplesins as archetypes, *Biochimica et Biophysica Acta*, 1462, 1-10.
- Mostovenko, E., Hassan, C., Rattke, J., Deelder, A. M., van Veelen, P. A., Palmblada, M., 2013, Comparison of peptide and protein fractionation methods in proteomics, *EuPA open proteomic I*, 30-37.
- Najafian, L., Babji, A. S., 2012, A review of fish-derived antioxidant and antimicrobial peptides : Their production, assessment and applications, *peptides*, 33, 178-185.
- Neshani, A., Zare, H., Eidgahi, M. R. A., Chichaklu, A. H., Movaghar, A., Ghazvini, K., 2018, Review of antimicrobial peptides with anti-helicobacter pylori activity, *Helicobacter*, 1-10.
- Nikolaev, E. N., Popov, I. A., Kononikhin, A. S., Indeykina, M. I., Kukav, E. N., 2012, High-resolution mass-spectrometry analysis of peptides and proteins, *Russian chemical reviews*, 81(11), 1051-1070.
- Novak, P, Havlicek, V., 2016, Protein Extraction and Precipitation dalam Proteomic Profiling and Analytical Chemistry (Second Edition), 51-62, Elsevier.
- Pelegrini, P. B., Farias, L. R., Saude, A. C. M., Costa, F. T., Bloch, C., Silva, L. P., Oliveira, A. S., Gomes, C. E. M., Sales, M. P., Franco, O. L., 2009, A Novel antimicrobial peptide from *Crotalaria pallida* seeds with activity against human and phytopathogens, *Curr Microbiol*, 59, 400-404.
- Piotto, S. P., Sessa, L., Concilio, S., Iannelli, P., 2012, YADAMP : yet another database of antimicrobial peptides, *Int. J. Antimicrob. Agents.*, 39, 346-351.
- Polito, L., Bortolotti, M., Battelli, M. G., Calafato, G., Bolognesi, A., 2019, Ricin : An ancient story for a timeless plant toxin, *Toxins*, 11(324), 1-16.
- Refdanita, Maksum, R., Nurgani, A., dan Endang, P., 2004, Pola kepekaan kuman terhadap Jakarta tahun 2001-2002, *Makara kesehatan*, 8(2), 41-48.
- Restiani, R., 2017, Hidrolisis Secara Enzimatis Protein Bungkil Biji Nyamplung (*Calophyllum inophyllum*) Menggunakan Bromelain, *J. Biota*, 1, 103-110.
- Rotilie, C.A., Fass, R.J., Prior, R.B., and Perkins, R.L., 1975, Microdilution technique for antimicrobial susceptibility testing of anaerobic bacteria, *Antimicrob. Agents Chemother.*, 7, 311-315.
- Salas, C.E., Badillo-Corona, J.A., Ramírez-Sotelo, G., Oliver-Salvador, C., 2015, Biologically Active and Antimicrobial Peptides from Plants, *Biomed Res. int.*, 1-11.

- Sari, R., Apridamayanti, P., Puspita, I. D., 2018, sensitivity of *Escherichia coli* bacteria towards antibiotics in patient with diabetic foot ulcer, *J. pharm. Sci. res.*, 5(1), 19-24.
- Senchyna, M., Jones, L., Louie, D., May, C., Forbes, I., Glasier, M., 2004, Qualitative and conformational characterization of lysozyme deposited on balafilcon and etafilcon contact lens materials, *Curr. Eye Res.*, 28(1), 25-36.
- Sheoran, I. S., Ross, A. R. S., Olson, D. J. H., Sawhney, V. K., 2009, Compatibility of plant protein extraction methods with mass spectrometry for proteome analysis, *Plant sci. J.*, 176, 99-104.
- Shi, W., Tang, Y., Sze, S., Zhu, Z., Wong, K., Shaw, P., 2016, Crystal Structure of Ribosome-Inactivating Protein Ricin A Chain in Complex with the C-Terminal Peptide of the Ribosomal Stalk Protein P2, *Toxins*, 8(296), 1-12.
- Sousa, N. L., Cabral, G. B., Vieira, P. M., Baldoni, A. B., Aragao, F. J. L., 2017, Bio-detoxification of ricin in castor bean (*Ricinus communis L.*) seeds, *Sci. Rep.*, 1-9.
- Sowa-Rogozinska, N., Sominka, H., Nowakowska-Golaca, J., Sandvig, K., Slominska-Wojewodzka, M., 2019, Intracellular transport and cytotoxicity of the protein toxin ricin, *Toxins*, 11(350), 1-37.
- Stein, A., Kiesewetter, A., 2007, Cation exchange chromatography in antibody purification: pH screening for optimised binding and HCP removal, *J. Chromatogr. B*, 848, 151-158.
- Stock, N. L., 2017, Introducing graduate students to high-resolution mass spectrometry (HRMS) using a hands-on approach, *J. Chem. Educ.*, 94(12), 1978-1982.
- Su y., 2011, Isolation and identification of pelteobagrin, a novel antimicrobial peptide from the skin mucus of yellow catfish (*Pelteobagrus fulvidraco*), *Comp Biochem Physiol B*, 158, 149-54.
- Suherman, B., Latif, M., Teresia, S., and Dewi, R.S.T., 2018, Potensi Kiotsan Kulit Udang Vannamei (*Litopenaus vannamei*) Sebagai Antibakteri Terhadap *Staphylococcus epidermis*, *Pseudomonas aeruginosa*, *Propionibacterium agnes*, dan *Escherichia coli* dengan Metode Difusi C, *Progr. Stud. Farm. Fak. Farm. Univ. Indones. Timur Mak.*, 14, 116-127.
- Tabb, D. L., Eng, J. K., and Yates, J. R., 2001, Protein Identification by SEQUEST, in: *Proteome Research: Mass Spectrometry*, James, P., Springer, New York.
- Taha, F. S., Mohamed, S. S., Wagdy, S. M., Mohamed, G. F., 2013, Antioxidant and Antimicrobial Activities of Enzymatic Hydrolysis Products from Sunflower Protein Isolate, *world appli. Sci. J.*, 21 (5), 651-658.
- Tavano, O. L., 2013, Protein hydrolysis using proteases: an important tool for food biotechnology, *J. Mol. Catal. B Enzym.*, 90, 1-11.
- Taylor, T. A., Unakal, C. G., 2021, *Staphylococcus aureus*, StatPearls Publishing LLC.
- Thapa, R. K., Diep, D. B., Tonnesen, H. H., 2020, Topical antimicrobial peptide formulation for wound healing : current developments and furture prospects, *acta biomater.*, 103, 52-67

- Theolier, J., Hammami, R., Labelle, P., Fliss, I., Jean, J., 2013, Isolation and identification of antimicrobial peptides derived by peptic cleavage of whey protein isolate, *J. Funct. Foods*, 5, 706-714.
- Trevino, S. R., Scholtz, J. M., Pace, C. N., 2008, Measuring and increasing protein solubility, *J. Pharm. Sci.*, 97(10), 4155-4166.
- Valgas, C., De Souza, S.M., Smânia, E.F.A., and Smânia, A., 2007, Screening methods to determine antibacterial activity of natural products, *Brazilian J. Microbiol.*, 38, 369-380.
- Verma, A. K., Chatli, M. K., Kumar, P., Mehta, N., 2017, Antioxidant and antimicrobial activity of protein hydrolysate extracted from porcine liver, *Indian J. Anim. Sci.*, 87(6), 711-717.
- Volmer, D. A., 2013, Principles of high-resolution MS dalam applications of high-resolution mass spectrometry in drug discovery and development, 6-20, Furture medicine Ltd.
- Walsh, C. dan Wencewicz, T. A., 2016, *Antibiotics : Challenges, Mechanisms, Oppoturnities*, ASM Press, Washington DC.
- Walsh, S.E., Maillard, J-Y., Russell, A. D., Catrenich, C. E., Charbonneau, D. L., Bartolo, R. G., 2003, Activity and mechanisms of action of selected biocidal agents on gram-positive and negative bacteria, *J. Appl. Microbiol.*, 94, 240-247.
- Wang, J., Song J., Yang, Z., He, S., Yang, Y., Feng, X., Dou, X., 2019, Antimicrobial peptides with high proteolytic resistance for combating gram-negative bacteria, *J. Med. Chem.*, 6, 2286-2304.
- Widodo, W. dan Sumarsih, S., 2007, *Jarak Kepyar*, Penerbit Kanisius, Yogyakarta.
- Setiawan, W., 2017, OPTIMASI PROSES PEMBUATAN BIODIESEL DARI MINYAK BIJI JARAK KEPYAR (*Ricinus communis* L.), 1, 45-54.
- Wiese, A., Brandenburg, K., Carroll, S. F., Rietschel, E. Th., Seydel, U., 1997, Mechanisms of action of bactericidal/permeability-increasing protein BPI on reconstituted outer membranes of gram-negative bacteria, *Biochemistry*, 36, 10311-10319.
- Wisuthiphaet, N., Konggruang, S., Chamcheun, C., 2015, Production of fish protein hydrolysates by acid and enzymatic hydrolysis, *J. Med. Biol. Eng.*, 4(6), 466-470
- Wizniewski, J. R., 2017, Filter-Aided Sample Preparation: The Versatile and Efficient Method for Proteomic Analysis, *Meth. Enzymol.*, 585, 15-27.
- Yamamoto, S., Miyagawa, E., 2000, Dynamic binding performance of large biomolecules such as γ -globulin, viruses and virus-like particles on various chromatographic supports, *Biotechnol. Prog.*, 16, 81-86.
- Yandri, A. S., 2011, Pengaruh modifikasi kimia terhadap titik isoelektrik pI enzim hasil modifikasi, *J. Sains MIPA*, 17(3), 92-98.
- Yeaman, M. R., Yount, N. Y., 2003, Mechanisms of antimicrobial peptides action and resistance, *Pharmacol. Rev.*, 55(1), 27-55.
- Yunita, M., dan Sukmawati, 2021, Edukasi bahaya resistensi bakteri akibat penggunaan antibiotik yang tidak rasional kepada masyarakat Desa Air Salobar, Indonesia Berdaya, 2(1), 1-6.

- Zambrowicz, A., Timer, M., Polanowski, A., Lube, G., Trziszka, T.,
Manufacturing of peptides exhibiting biological activity, *Amino Acids*, 44,
315-320.
- Zarai, Z., Kadri, A., Chobba, I. B., Mansour, R. B., Bekir, A., Mejdoub, H.,
Gharsallah, N., 2011, The in-vitro evaluation of antibacterial, antifungal and
cytotoxic properties of Marrubium vulgare L. essential oil grown in Tunisia,
Lipids in health and disease, 10 (161), 1-8.
- Zasloff, M., 2002, Antimicrobial peptides of multicellular organisms, *Nature*, 415,
389-396.