UJI AKTIVITAS EKSTRAK KOLAGEN KULIT IKAN SALMON (*Salmo salar*) SEBAGAI INHIBITOR ACE1 BERDASARKAN STUDI *IN VITRO* DAN *IN SILICO*

SKRIPSI

diajukan untuk memenuhi salah satu syarat memperoleh Gelar Sarjana Sains
Program Studi Kimia

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PERNYATAAN

Dengan ini saya menyatakan bahwa skripsi dengan judul "Uji Aktivitas Ekstrak Kolagen Kulit Ikan Salmon (*Salmo salar*) sebagai Inhibitor ACE1 Berdasarkan Studi *In Vitro* dan *In Silico*" ini beserta seluruh isinya adalah benar-benar karya saya sendiri. Saya tidak melakukan penjiplakan atau pengutipan dengan cara-cara yang tidak sesuai dengan etika ilmu yang berlaku dalam masyarakat keilmuan. Atas pernyataan ini, saya siap menanggung risiko/sanksi apabila di kemudian hari ditemukan adanya pelanggaran etika keilmuan atau ada klaim dari pihak lain terhadap keaslian karya saya ini.

Bandung, 21 Agustus 2022

Yang Membuat Pernyataan

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ABSTRAK

Penggunaan inhibitor ACE1 dalam pengobatan COVID-19 pada pasien berkomorbid telah diteliti menunjukkan efek samping. Kulit ikan salmon sebagai produk samping industri perikanan memiliki kandungan protein kolagen cukup tinggi dan berpotensi menjadi alternatif penemuan inhibitor ACE1 baru berbasis peptida. Penelitian ini bertujuan untuk menguji aktivitas kolagen kulit ikan salmon (*Salmo salar*) sebagai inhibitor ACE1 berdasarkan studi *in vitro* dan *in silico*. Metode penelitian *in vitro* meliputi ekstraksi kolagen larut asam, karakterisasi menggunakan FTIR, UV, XRD, dan SDS-PAGE, serta penentuan inhibisi terhadap ACE1. Adapun metode pada studi *in silico* yang digunakan yaitu *molecular docking*. Ekstraksi kolagen memiliki kandungan protein 95,32±2,2 %. Spektrum FTIR mendeteksi gugus fungsi amida (A, B, I, II, III) dan imina prolin/hidroksiprolin yang mengindikasikan serapan khas kolagen. Karakterisasi UV menunjukkan serapan khas kolagen pada 230 nm. Berdasarkan data XRD dan SDS-PAGE, jenis rantai pada sampel adalah α-heliks dan rantai α. Pengujian inhibisi ACE1 dari sampel didapat sebesar 11,42%; 67,94%; 74,79%; 83,67%; dan 84,94% berturut-turut pada konsentrasi 10, 250, 500, 1000, dan 2000 ppm. Adapun studi *in silico* menunjukkan adanya interaksi antara ACE1 dan kolagen di luar sisi aktif dan afinitas sebesar -213,89 kkal/mol. Sementara itu, peptida aktif kolagen menunjukkan interaksi pada sisi aktif ACE1 dengan afinitas lebih baik dari lisinopril pada tiga peptida, yaitu WF (Trp-Phe), WYT (Trp-Tyr-Thr), dan PDPF (Pro-Asp-Pro-Phe) berturut-turut 9,1; 9,1; dan 8,8 kkal/mol. Dapat disimpulkan bahwa kolagen kulit ikan salmon memiliki aktivitas inhibisi ACE1 dengan prediksi mekanisme inhibisi non-kompetitif. Sebaliknya, peptida aktif dari kolagen kulit ikan salmon diprediksi memiliki potensi sebagai inhibitor kompetitif terhadap ACE1.

Kata Kunci: ACE1, *in vitro*, *in silico*, Kulit Ikan Salmon, Kolagen

ABSTRACT

The use of ACE1 inhibitors in COVID-19's comorbid patient treatment has been studied to have adverse effects. Salmon skin, as a by-product of the fishing industry, has potential due to its high collagen protein content and can be an alternative source for discovering new peptide-based ACE1 inhibitors. This study aims to determine the collagen activity of salmon skin (Salmo salar) as an ACE1 inhibitor based in vitro and in silico. In vitro research methods include extraction of acid-soluble collagen, characterization using FTIR, UV, XRD, and SDS-PAGE, and determination of ACE1 inhibition. The method used in the in silico study is molecular docking. The collagen extraction process gives protein content of 95.32±2.2 (%). FTIR analysis showed the spectrum of amide functional groups (A, B, I, II, III) and imine proline/hydroxyproline typical of collagen. UV characterization showed a typical absorption of collagen at 230 nm. Based on XRD and SDS-PAGE data, the sample chain type is alpha helix and alpha chain. The ACE1 inhibition test of the sample was 11.42%; 67.94%; 74.79%; 83.67%; and 84.94% at concentrations of 10, 250, 500, 1000, and 2000 ppm, respectively. The in silico study showed interactions between ACE1 and collagen outside the active site and an affinity of -213.89 kcal/mol. Meanwhile, the active peptide of collagen showed interactions at the active site of ACE1 with better affinity than lisinopril for three peptides, namely WF (Trp-Phe), WYT (Trp-Tyr- Thr), and PDPF (Pro-Asp-Pro-Phe) 9.1; 9.1; and 8.8 kcal/mol. It can be concluded that salmon skin collagen has ACE1 inhibitory activity with a mechanism that is predictable as non-competitive inhibition. In contrast, active peptides from salmon skin collagen were predicted to have potential as competitive inhibitors of ACE1.

Keywords: ACE1, collagen, in vitro, in silico, salmon skin

DAFTAR ISI

DAFTAR TABEL

DAFTAR GAMBAR

DAFTAR LAMPIRAN

DAFTAR ISTILAH, SINGKATAN, DAN LAMBANG

A : Alanin (Ala)

ACE1 : *Angiotensin Converting Enzyme I*

ASC : *Acid Soluble Collagen*

C : Sistein (Cys)

D : Asam aspartat (Asp)

E : Asam glutamat (Glu)

F : Fenilalanin (Phe)

G : Glisin (Gly)

GB : Giga Byte

GHz : Giga Hertz

H : Histidin (His)

HBD : *Hydrogen Bond Donor*

HBA : *Hydrogen Bond Acceptor*

I : Isoleusin (Ile)

K : Lisin (Lys)

L : Leusin (Leu)

M : Metionin (Met)

N : Asparagin (Asn)

P : Prolin (Pro)

PSC : *Pepsin Soluble Collagen*

Q : Glutamin (Gln)

R : Arginin (Arg)

RMSD : *Root Mean Square Deviation*

S : Serin (Ser)

T :Threonin (Thr)

V : Valin (Val)

W : Triptofan (Trp)

Y : Tirosin (Tyr)

DAFTAR PUSTAKA

Akita, M., Kono, T., Lloyd, K., Mitsui, T., Morioka, K., & Adachi, K. (2019). Biochemical study of type I collagen purified from skin of warm sea teleost Mahi mahi (Coryphaena hippurus), with a focus on thermal and physical stability. *Journal of Food Biochemistry*, *43*(11). https://doi.org/10.1111/jfbc.13013

Albini, A., Di, G., Douglas, G., Noonan, M., & Lombardo, M. (2020). The SARS-CoV-2 receptor , ACE-2 , is expressed on many different cell types : implications for ACE-inhibitor - and angiotensin II receptor blocker - based cardiovascular therapies. *Internal and Emergency Medicine*, *15*(5), 759–766. https://doi.org/10.1007/s11739-020-02364-6

Alves, A. L., Marques, A. L. P., Martins, E., Silva, T. H., & Reis, R. L. (2017). Cosmetic potential of Marine fish skin collagen. *Cosmetics*, *4*(4), 1–16. https://doi.org/10.3390/cosmetics4040039

Alves, A., Marques, A., Martins, E., Silva, T., & Reis, R. (2017). Cosmetic Potential of Marine Fish Skin Collagen. *Cosmetics*, *4*(4), 39. https://doi.org/10.3390/cosmetics4040039

Apweiler, R., Bairoch, A., Wu, C. H., Barker, W. C., Boeckmann, B., Ferro, S., Gasteiger, E., Huang, H., Lopez, R., Magrane, M., Martin, M. J., Natale, D. A., Donovan, C. O., Redaschi, N., & Yeh, L. L. (2004). UniProt : the Universal Protein knowledgebase. *Nucleic Acids Research*, *32*. https://doi.org/10.1093/nar/gkh131

Arora, P. K., & Chauhan, A. (2013). ACE INHIBITORS: A COMPREHENSIVE REVIEW. *International Journal of Pharmaceutical Sciences and Research*, *4*(2), 532–549.

Atef, M., Ojagh, S. M., Latifi, A. M., Esmaeili, M., & Udenigwe, C. C. (2020). Biochemical and structural characterization of sturgeon fish skin collagen (Huso huso). *Journal of Food Biochemistry*, *44*(8), 1–10. https://doi.org/10.1111/jfbc.13256

Avena-Bustillos, R. J., Olsen, C. W., Olson, D. A., Chiou, B., Yee, E., Bechtel, P. J., & McHugh, T. H. (2006). Water vapor permeability of mammalian and fish gelatin films. *Journal of Food Science*, *71*(4), 202–207.

https://doi.org/10.1111/j.1750-3841.2006.00016.x

Bachan Upadhyay, L. S., & Verma, N. (2013). Enzyme Inhibition Based Biosensors: A Review. *Analytical Letters*, *46*(2), 225–241. https://doi.org/10.1080/00032719.2012.713069

Baigi, K., & Stewart, W. F. (2015). Headache and migraine : a leading cause of absenteeism. In *Handbook of Clinical Neurology* (1st ed., Vol. 131, pp. 447–463). Elsevier B.V. https://doi.org/10.1016/B978-0-444-62627-1.00025-1

Bak, S. Y., Lee, S. W., Choi, C. H., & Kim, H. W. (2018). Assessment of the influence of acetic acid residue on type I collagen during isolation and characterization. *Materials*, *10*(12). https://doi.org/10.3390/ma11122518

Benet, L. Z., Hosey, C. M., Ursu, O., Oprea, T. I., Sciences, T., & Division, I. (2017). BDDCS, the Rule of 5 and Drugability. *Adv Drug Deliv Rev*, 89–98. https://doi.org/10.1016/j.addr.2016.05.007.BDDCS

Berman, H. M., Westbrook, J., Feng, Z., Gilliland, G., Bhat, T. N., Weissig, H., SHindyalov, I. N., & Bourne, P. E. (2000). The Protein Data Bank. *Nucleic Acids Research*, *28*(1), 235–242. https://doi.org/10.1093/nar/28.1.235

Bernstein, K. E., Khan, Z., Giani, J. F., Cao, D., Bernstein, E. A., Shen, X. Z., & Angeles, L. (2018). Angiotensin-converting enzyme in innate and adaptive immunity. *Nat Rev Nephrol*, *14*(5), 325–336. https://doi.org/10.1038/nrneph.2018.15.Angiotensin-converting

Biter, A. B., Pollet, J., Chen, W. H., Strych, U., Hotez, P. J., & Bottazzi, M. E. (2019). A method to probe protein structure from UV absorbance spectra. *Analytical Biochemistry*, *587*(June). https://doi.org/10.1016/j.ab.2019.113450

Boopathi, S., Poma, A. B., & Kolandaivel, P. (2020). Novel 2019 Coronavirus Structure , Mechanism of Action , Antiviral Drug Promises and Rule Out Against its Treatment. *Journal of Biomolecular Structure and Dynamics*, *0*(0), 1–10. https://doi.org/10.1080/07391102.2020.1758788

Borrajo, P., Pateiro, M., Gagaoua, M., Franco, D., Zhang, W., & Lorenzo, M. (2020). Evaluation of the Antioxidant and Antimicrobial Activities of Porcine Liver Protein Hydrolysates Obtained Using Alcalase, Bromelain, and Papain. *Applied Sciences*, *10*(2290). https://doi.org/10.3390/app10072290

Carvalho, A. M., Marques, A. P., Silva, T. H., & Reis, R. L. (2018). Evaluation of

the potential of collagen from codfish skin as a biomaterial for biomedical applications. *Marine Drugs*, *16*(12), 1–14. https://doi.org/10.3390/md16120495

Cascella, M., Rajnik, M., Cuomo, A., Dulebohn, S. C., & Napoli, R. Di. (2020). Features , Evaluation and Treatment Coronavirus (COVID-19). *StatPearls*.

Chen, J., Li, L., Yi, R., Xu, N., Gao, R., & Hong, B. (2016). Extraction and characterization of acid-soluble collagen from scales and skin of tilapia (Oreochromis niloticus). *LWT - Food Science and Technology*, *66*, 453–459. https://doi.org/10.1016/j.lwt.2015.10.070

Chuaychan, S., Benjakul, S., & Kishimura, H. (2015). Characteristics of acid- and pepsin-soluble collagens from scale of seabass (Lates calcarifer). *LWT - Food Science and Technology*, *63*(1), 71–76. https://doi.org/10.1016/j.lwt.2015.03.002

Coppola, D., Oliviero, M., Vitale, G. A., Lauritano, C., Ambra, I. D., Iannace, S., & Pascale, D. De. (2020). Marine Collagen from Alternative and Sustainable Sources : Extraction , Processing and Applications. *Marine Drugs*, *18*(214). https://doi.org/10.3390/md18040214

Daina, A., Michielin, O., & Zoete, V. (2017). SwissADME: A free web tool to evaluate pharmacokinetics, drug-likeness and medicinal chemistry friendliness of small molecules. *Scientific Reports*, *7*(March), 1–13. https://doi.org/10.1038/srep42717

Dave, D., Liu, Y., Clark, L., Dave, N., Trenholm, S., & Westcott, J. (2019). Availability of marine collagen from Newfoundland fisheries and aquaculture waste resources. *Bioresource Technology Reports*, *7*(June), 100271. https://doi.org/10.1016/j.biteb.2019.100271

Dawson, P., Al-jeddawi, W., & Remington, N. (2018). Effect of Freezing on the Shelf Life of Salmon. *International Journal of Food Science*, *2018*.

Derkach, S. R., Voron'ko, N. G., Kuchina, Y. A., & Kolotova, D. S. (2020). Modified fish gelatin as an alternative to mammalian gelatin in modern food technologies. *Polymers*, *12*(12), 1–10. https://doi.org/10.3390/polym12123051

Dhillon, A., Sharma, K., Rajulapati, V., & Goyal, A. (2017). Proteolytic Enzymes.

In *Current Developments in Biotechnology and Bioengineering: Production, Isolation and Purification of Industrial Products* (pp. 149–176). Elsevier B.V. https://doi.org/10.1146/annurev.bi.21.070152.000245

Dimitrov, I., Bangov, I., Flower, D. R., & Doytchinova, I. (2014). AllerTOP v.2 - A server for in silico prediction of allergens. *Journal of Molecular Modeling*, *20*(6). https://doi.org/10.1007/s00894-014-2278-5

Eberhardt, J., Santos-martins, D., Tillack, A. F., & Forli, S. (2021). AutoDock Vina 1.2.0: New Docking Methods, Expanded Force Field, and Python Bindings. *Journal of Chemical Information and Modeling*. https://doi.org/10.1021/acs.jcim.1c00203

Elezkurtaj, S., Greuel, S., Ihlow, J., Michaelis, E. G., Bischoff, P., Kunze, C. A., Sinn, B. V., Gerhold, M., Hauptmann, K., Heppner, B. I., Miller, F., Herbst, H., Corman, V. M., Martin, H., Radbruch, H., Heppner, F. L., & Horst, D. (2021). Causes of death and comorbidities in hospitalized patients with COVID - 19. *Scientific Reports*, *11*(4263), 1–9. https://doi.org/10.1038/s41598-021-82862-5

Fan, H., Wang, L., Liu, W., An, X., Liu, Z., He, X., Song, L., & Tong, Y. (2020). Repurposing of Clinically Approved Drugs for Treatment of Coronavirus Disease 2019 in a 2019-novel Coronavirus-related Coronavirus Model. *Chinese Medical Journal*, *133*(9), 1051–1056. https://doi.org/10.1097/CM9.0000000000000797

Fan, J., Fu, A., & Zhang, L. (2019). Progress in molecular docking. *Quantitative Biology*, *7*(2), 83–89.

Fu, Z., Akula, S., Thorpe, M., & Hellman, L. (2021). Marked difference in efficiency of the digestive enzymes pepsin, trypsin, chymotrypsin, and pancreatic elastase to cleave tightly folded proteins. *Biological Chemistry*, *402*(7), 861–867. https://doi.org/10.1515/hsz-2020-0386

Ge, B., Wang, H., Li, J., Liu, H., Yin, Y., Zhang, N., & Qin, S. (2020). Comprehensive Assessment of Nile Tilapia Skin (Oreochromis niloticus) Collagen Hydrogels for Wound Dressings. *Marine Drugs*, *18*(4), 178. https://doi.org/10.3390/md18040178

Gijsen, R., Hoeymans, N., Schellevis, F. G., Ruwaard, D., Satariano, W. A., & Bos,

G. A. M. Van Den. (2001). *Causes and consequences of comorbidity : A review*. *54*, 661–674.

Gornall, A. G., Bardawill, C. J., & David, M. M. (1949). Determination of serum proteins by means of the biuret reaction. *The Journal of Biological Chemistry*, *177*(2), 751–766. https://doi.org/10.1016/s0021-9258(18)57021-6

Gupta, S., Kapoor, P., Chaudhary, K., Gautam, A., Kumar, R., & Raghava, G. P. S. (2013). In Silico Approach for Predicting Toxicity of Peptides and Proteins. *PLoS ONE*, *8*(9). https://doi.org/10.1371/journal.pone.0073957

Hanwell, M. D., Curtis, D. E., Lonie, D. C., Vandermeersch, T., Zurek, E., & Hutchison, G. R. (2012). Avogadro: an advanced semantic chemical editor, visualization, and analysis platform. *Journal of Cheminformatics*, *4*(1), 17. https://doi.org/10.1186/1758-2946-4-17

Hariyanto, T. I., Japar, K. V., Damay, V., & Kwenandar, F. (2020). The Use of ACE inhibitor / ARB in SARS-CoV-2 Patients : A Comprehensive Narrative Review. *ASIAN JOURNAL OF MEDICAL SCIENCES*, *11*(6), 113–120. https://doi.org/10.3126/ajms.v11i6.29911

He, L., Lan, W., Zhao, Y., Chen, S., Liu, S., Cen, L., Cao, S., Dong, L., Jin, R., & Liu, Y. (2020). Characterization of biocompatible pig skin collagen and application of collagen-based films for enzyme immobilization. *RSC Advances*, *10*(12), 7170–7180. https://doi.org/10.1039/C9RA10794K

Homayun, B., Lin, X., & Choi, H. J. (2019). Challenges and recent progress in oral drug delivery systems for biopharmaceuticals. *Pharmaceutics*, *11*(3). https://doi.org/10.3390/pharmaceutics11030129

Houston, R. D., & Macqueen, D. J. (2018). Atlantic salmon (Salmo salar L .) genetics in the 21st century : taking leaps forward in aquaculture and biological understanding. *Animal Genetics*, *October*. https://doi.org/10.1111/age.12748

Hua, S. (2020). Advances in Oral Drug Delivery for Regional Targeting in the Gastrointestinal Tract - Influence of Physiological, Pathophysiological and Pharmaceutical Factors. *Frontiers in Pharmacology*, *11*(April), 1–22. https://doi.org/10.3389/fphar.2020.00524

Iswariya, S., Velswamy, P., & Uma, T. S. (2018). Isolation and Characterization of

Biocompatible Collagen from the Skin of Puffer Fish (Lagocephalus inermis). *Journal of Polymers and the Environment*, *26*(5), 2086–2095. https://doi.org/10.1007/s10924-017-1107-1

Jakhmola, S., Indari, O., Baral, B., Kashyap, D., Varshney, N., Das, A., Chatterjeeand, S., & Jha, H. C. (2020). Comorbidity Assessment Is Essential During COVID-19 Treatment. *Frontiers in Physiology*, *11*(August), 1–7. https://doi.org/10.3389/fphys.2020.00984

Jariashvili, K., Madhan, B., Brodsky, B., Kuchava, A., Namicheishvili, L., & Metreveli, N. (2012). Uv damage of collagen: Insights from model collagen peptides. *Biopolymers*, *97*(3), 189–198. https://doi.org/10.1002/bip.21725

Jin, R., Teng, X., Shang, J., Wang, D., & Liu, N. (2020). Identi fi cation of novel DPP – IV inhibitory peptides from Atlantic salmon (Salmo salar) skin. *Food Research International*, *133*(March), 109161. https://doi.org/10.1016/j.foodres.2020.109161

Jin, W., Zhang, W., Mitra, D., Mccandless, M. G., Sharma, P., Tandon, R., Zhang, F., & Linhardt, R. J. (2020). The Structure-Activity Relationship of The Interactions of SARS-CoV-2 Spike Glycoproteins with Glucuronomannan and Sulfated Galactofucan from Saccharina japonica. *International Journal of Biological Macromolecules*, *163*, 1649–1658. https://doi.org/10.1016/j.ijbiomac.2020.09.184

Khan, M. Y., & Kumar, V. (2018). Mechanism & inhibition kinetics of bioassay-guided fractions of Indian medicinal plants and foods as ACE inhibitors. *Journal of Traditional and Complementary Medicine*, *1*(12). https://doi.org/10.1016/j.jtcme.2018.02.001

Kim, S., Thiessen, P. A., Bolton, E. E., Chen, J., Fu, G., Gindulyte, A., Han, L., He, J., He, S., Shoemaker, B. A., Wang, J., Yu, B., Zhang, J., & Bryant, S. H. (2016). PubChem substance and compound databases. *Nucleic Acids Research*, *44*(D1), D1202–D1213. https://doi.org/10.1093/nar/gkv951

Laemmli, U. K. (1970). Cleavage of Structural Proteins during the Assembly of the Head of Bacteriophage T4. *Nature*, *227*.

Laurent, S. (2017). Antihypertensive drugs. *Pharmacological Research*, *124*, 116–125. https://doi.org/10.1016/j.phrs.2017.07.026

Lee, T., Cau, A., Cheng, P., Levin, A., Lee, T. C., Vinh, D. C., Lamontagne, F., Singer, J., Walley, K. R., Murthy, S., Patrick, D., Rewa, O. G., Winston, B. W., Marshall, J., Boyd, J., Tran, K., Kalil, A. C., Mcculoh, R., Fowler, R., … Russell, J. A. (2021). Systematic Review / Meta-analysis Angiotensin Receptor Blockers and Angiotensin-Converting Enzyme Inhibitors in COVID-19 : Meta-analysis / Meta- regression Adjusted for Confounding Factors. *CJC Open*, *3*(7), 965–975. https://doi.org/10.1016/j.cjco.2021.03.001

Li, C., Wang, J., Wang, Y., Gao, H., Wei, G., Huang, Y., Yu, H., & Gan, Y. (2019). Recent progress in drug delivery. *Acta Pharmaceutica Sinica B*, *9*(6), 1145–1162. https://doi.org/10.1016/j.apsb.2019.08.003

Li, P., Jia, J., Fang, M., Zhang, L., Guo, M., & Xie, J. (2014). In vitro and in vivo ACE inhibitory of pistachio hydrolysates and in silico mechanism of identified peptide binding with ACE. *Process Biochemistry*, *49*(5), 898–904. https://doi.org/10.1016/j.procbio.2014.02.007

Lin, R. B., He, Y., Li, P., Wang, H., Zhou, W., & Chen, B. (2019). Multifunctional porous hydrogen-bonded organic framework materials. *Chemical Society Reviews*, *48*(5), 1362–1389. https://doi.org/10.1039/c8cs00155c

Lipinski, C. A., Lombardo, F., Dominy, B. W., & Feeney, P. J. (2012). Experimental and computational approaches to estimate solubility and permeability in drug discovery and development settings. *Advanced Drug Delivery Reviews*, *64*(SUPPL.), 4–17. https://doi.org/10.1016/j.addr.2012.09.019

Los, H. M. (2020). Conservation and restoration. In *Ecology and Management of Inland Waters* (pp. 401–437). https://doi.org/10.1016/B978-0-12-814266-0.00026-X

Luo, Q., Chen, D., Boom, R. M., & Janssen, A. E. M. (2018). Revisiting the enzymatic kinetics of pepsin using isothermal titration calorimetry. *Food Chemistry*, *268*(February), 94–100. https://doi.org/10.1016/j.foodchem.2018.06.042

MacLeod, J. M., & Rosei, F. (2011). Directed Assembly of Nanostructures. In D. L. Andrews, G. D. Scholes, & G. P. Wiederrecht (Eds.), *Comprehensive Nanoscience and Technology* (pp. 13–68). Academic Press.

Mei, F., Liu, J., Wu, J., Duan, Z., Chen, M., Meng, K., Chen, S., Shen, X., Xia, G., & Zhao, M. (2020). Collagen Peptides Isolated from Salmo salar and Tilapia nilotica Skin Accelerate Wound Healing by Altering Cutaneous Microbiome Colonization via Upregulated NOD2 and BD14. *Journal of Agricultural and Food Chemistry*, *68*, 1621–1633. https://doi.org/10.1021/acs.jafc.9b08002

Menon, V. V., & Lele, S. S. (2015). Nutraceuticals and Bioactive Compounds from Seafood Processing Waste. In *Springer handbook of marine biotechnology* (pp. 1405–1425). Spinger.

Messerli, F. H., Mha, S. B., Mph, C. B., & Rimoldi, S. F. (2018). Angiotensin-Converting Enzyme Inhibitors in Hypertension. *Journal of the American College of Cardiology*, *71*(13), 1474–1482. https://doi.org/10.1016/j.jacc.2018.01.058

Minkiewicz, P., Iwaniak, A., & Darewicz, M. (2019). BIOPEP-UWM Database of Bioactive Peptides : Current Opportunities. *International Journal of Molecular Sciences*, *20*(5978).

Mooney, C., Haslam, N. J., Pollastri, G., & Shields, D. C. (2012). Towards the Improved Discovery and Design of Functional Peptides: Common Features of Diverse Classes Permit Generalized Prediction of Bioactivity. *PLoS ONE*, *7*(10). https://doi.org/10.1371/journal.pone.0045012

Moreno, H. M., Montero, M. P., Gómez-guillén, M. C., Fernández-martín, F., Mørkøre, T., & Borderías, J. (2012). Collagen characteristics of farmed Atlantic salmon with firm and soft fillet texture. *Food Chemistry*, *134*(2), 678–685. https://doi.org/10.1016/j.foodchem.2012.02.160

Morris, G. M., Huey, R., Lindstrom, W., Sanner, M. F., Belew, R., Goodsell, D. S., & Olson, A. J. (2009). AutoDock4 and AutoDockTools4: Automated Docking with Selective Receptor Flexibility. *Journal of Computational Chemistry*, *30*, 278502791. https://doi.org/10.1002/jcc.21256

Munawaroh, H. S. H., Gumilar, G. G., Nurjanah, F., Yuliani, G., Aisyah, S., Kurnia, D., Wulandari, A. P., Kurniawan, I., Ningrum, A., Koyande, A. K., & Show, P. L. (2020). In-vitro molecular docking analysis of microalgae extracted phycocyanin as an anti-diabetic candidate. *Biochemical Engineering Journal*, *161*(June), 107666. https://doi.org/10.1016/j.bej.2020.107666

Natesh, R., Schwager, S. L. U., Sturrock, E. D., & Acharya, K. R. (2003). Crystal structure of the human angiotensin-converting enzyme-lisinopril complex. *Nature*, *421*(6922), 551–554. https://doi.org/10.1038/nature01370

Neves, A. C., Harnedy, P. A., O'Keeffe, M. B., & Fitzgerald, R. J. (2017). Bioactive peptides from Atlantic salmon (Salmo salar) with angiotensin converting enzyme and dipeptidyl peptidase IV inhibitory , and antioxidant activities Keywords. *Food Chemistry*.

O'Boyle, N. M., Banck, M., James, C. A., Morley, C., Vandermeersch, T., & Hutchison, G. R. (2011). Open Babel: An open chemical toolbox. *Journal of Cheminformatics*, *3*(33), 1–14. https://jcheminf.biomedcentral.com/track/pdf/10.1186/1758-2946-3-33

OECD. (2017). Atlantic salmon (Salmo salar). In *SAFETY ASSESSMENT OF TRANSGENIC ORGANISMS IN THE ENVIRONMENT: OECD CONSENSUS DOCUMENTS Volume 7* (Vol. 7). OECD Publisher.

Oliveira, V. de M., Assis, C. R. D., Costa, B. de A. M., Neri, R. C. de A., Monte, F. T., Freitas, H. M. S. da C. V., França, R. C. P., Santos, J., Bezerra, R. de S., & Porto, A. L. F. (2021). Physical, biochemical, densitometric and spectroscopic techniques for characterization collagen from alternative sources: A review based on the sustainable valorization of aquatic by-products. *Journal of Molecular Structure*, *1224*. https://doi.org/10.1016/j.molstruc.2020.129023

Oliver, E., Mayor Jr, F., & D'Ocon, P. (2019). Beta-blockers: Historical Perspective and Mechanisms of Action. *Revista Española de Cardiología (English Edition)*, *72*(10), 853–862. https://doi.org/10.1016/j.rec.2019.04.006

Pagadala, N. S., Syed, K., & Tuszynski, J. (2017). Software for molecular docking : a review. *Biophysical Reviews*, 91–102. https://doi.org/10.1007/s12551-016-0247-1

Pal, G. K., Nidheesh, T., & Suresh, P. V. (2015). Comparative study on characteristics and in vitro fibril formation ability of acid and pepsin soluble collagen from the skin of catla (Catla catla) and rohu (Labeo rohita). *Food Research International*, *76*, 804–812. https://doi.org/10.1016/j.foodres.2015.07.018

Panatdasirisuk, W., & Vongsetskul, T. (2020). Bromelain Immobilized onto Diamine-functionalized Electrospun Polyvinyl Chloride Fibers as a Durable Heterogeneous Catalyst. *Fibers and Polymers*, *21*(10), 2224–2230. https://doi.org/10.1007/s12221-020-1247-0

PDSPKP, D. (2022). Statistik Impor Hasil Perikanan Tahun 2017 – 2021. In *Sekretariat Direktorat Jenderal Penguatan Daya Saing Produk Kelautan dan Perikanan*.

Pearman, N. A., Ronander, E., Smith, A. M., & Morris, G. A. (2020). The identification and characterisation of novel bioactive peptides derived from porcine liver. *Current Research in Food Science*, *3*, 314–321. https://doi.org/10.1016/j.crfs.2020.11.002

Pinzi, L., & Rastelli, G. (2019). Molecular Docking: Shifting Paradigms in Drug Discovery. *International Journal of Molecular Sciences*, *20*(18), 4331. https://doi.org/10.3390/ijms20184331

Poblador-plou, B., Carmona-Pírez, J., Ioakeim-skoufa, I., Poncel-Falcó, A., Bliek-Bueno, K., Pozo, M. C., Gimeno-Feliú, L. A., González-Rubio, F., Aza-Pascual-Salcedo, M., Bandrés-Liso, A. C., Díez-Manglano, J., Marta-Moreno, J., Mucherino, S., Gimeno-Miguel, A., Prados-Torres, A., & Group, E. (2020). Baseline Chronic Comorbidity and Mortality in Laboratory-Confirmed COVID-19 Cases : Results from the PRECOVID Study in Spain. *International Journal of Environmental Research and Public Health*, *17*(5171).

Pokhrel, S., & Chhetri, R. (2021). A Literature Review on Impact of COVID-19 Pandemic on Teaching and Learning. *Higher Education for the Future*, *8*(1), 133–141. https://doi.org/10.1177/2347631120983481

Qian, B., Tian, C., Huo, J., Ding, Z., Xu, R., Zhu, J., Yu, L., & Villarreal, O. D. (2019). Design and evaluation of four novel tripeptides as potent angiotensin converting enzyme (ACE) inhibitors with anti-hypertension activity. *Peptides*, *122*, 170171. https://doi.org/10.1016/j.peptides.2019.170171

Rice, P., Longden, L., & Bleasby, A. (2000). EMBOSS: The European Molecular Biology Open Software Suite. *Trends in Genetics*, *16*(6), 276–277. https://doi.org/10.1016/S0168-9525(00)02024-2

Ruyck, J. De, Brysbaert, G., Blossey, R., & Lensink, M. F. (2016). Molecular

Docking as a Popular Tool in Drug Design , an In Silico Travel. *Advances and Applications in Bioinformatics and Chemistry*, *9*, 1–11.

Salvatore, L., Gallo, N., Natali, M. L., Campa, L., Lunetti, P., Madaghiele, M., Blasi, F. S., Corallo, A., Capobianco, L., & Sannino, A. (2020). Marine collagen and its derivatives: Versatile and sustainable bio-resources for healthcare. *Materials Science and Engineering C*, *113*(April), 110963. https://doi.org/10.1016/j.msec.2020.110963

Santana, J. C. C., Gardim, R. B., Almeida, P. F., Borini, G. B., Quispe, A. P. B., Llanos, S. A. V., Heredia, J. A., Zamuner, S., Gamarra, F. M. C., Farias, T. M. B., Ho, L. L., & Berssaneti, F. T. (2020). Valorization of Chicken Feet By-Product of the Poultry Industry: High Qualities of Gelatin and Biofilm from Extraction of Collagen. *Polymers*, *12*(3), 529. https://doi.org/10.3390/polym12030529

Schrödinger, & DeLano, W. L. (2020). *PyMOL* (2.5.2). http://www.pymol.org/pymol

Schwede, T., Kopp, J., Guex, N., & Peitsch, M. C. (2003). SWISS-MODEL: An automated protein homology-modeling server. *Nucleic Acids Research*, *31*(13), 3381–3385. https://doi.org/10.1093/nar/gkg520

Selamassakul, O., Laohakunjit, N., Kerdchoechuen, O., Yang, L., & Maier, C. S. (2018). Isolation and characterisation of antioxidative peptides from bromelain-hydrolysed brown rice protein by proteomic technique. *Process Biochemistry*, *70*, 179–187. https://doi.org/10.1016/j.procbio.2018.03.024

Shaik, M. I., & Sarbon, N. M. (2020). A Review on Purification and Characterization of Anti-proliferative Peptides Derived from Fish Protein Hydrolysate. *Food Reviews International*. https://doi.org/10.1080/87559129.2020.1812634

Sharma, K. M., Kumar, R., Panwar, S., & Kumar, A. (2017). Microbial alkaline proteases: Optimization of production parameters and their properties. *Journal of Genetic Engineering and Biotechnology*, *15*(1), 115–126. https://doi.org/10.1016/j.jgeb.2017.02.001

Stani, C., Vaccari, L., Mitri, E., & Birarda, G. (2020). FTIR investigation of the secondary structure of type I collagen: New insight into the amide III band.

Spectrochimica Acta - Part A: Molecular and Biomolecular Spectroscopy, *229*, 118006. https://doi.org/10.1016/j.saa.2019.118006

Tacias-Pascacio, V. G., Castañeda-Valbuena, D., Morellon-Sterling, R., Tavano, O., Berenguer-Murcia, Á., Vela-Gutiérrez, G., Rather, I. A., & Fernandez-Lafuente, R. (2021). Bioactive peptides from fisheries residues: A review of use of papain in proteolysis reactions. *International Journal of Biological Macromolecules*, *184*(April), 415–428. https://doi.org/10.1016/j.ijbiomac.2021.06.076

Tedeschi, T., Anzani, C., Ferri, M., Marzocchi, S., Caboni, M. F., Monari, S., & Tassoni, A. (2021). Enzymatic digestion of calf fleshing meat by-products: Antioxidant and anti-tyrosinase activity of protein hydrolysates, and identification of fatty acids. *Foods*, *10*(4). https://doi.org/10.3390/foods10040755

Trott, O., & Olson, A. J. (2010). AutoDock Vina: Improving the Speed and Accuracy of Docking with a New Scoring Function, Efficient Optimization, and Multithreading. *Journal of Computational Chemistry*, *31*, 455–461. https://doi.org/10.1002/jcc.21334

Tylingo, R., Mania, S., Panek, A., Piątek, R., & Pawłowicz, R. (2016). Isolation and Characterization of Acid Soluble Collagen from the Skin of African Catfish (Clarias gariepinus), Salmon (Salmo salar) and Baltic Cod (Gadus morhua). *Journal of Biotechnology & Biomaterials*, *6*(2). https://doi.org/10.4172/2155-952X.1000234

Vinarov, Z., Abdallah, M., Agundez, J. A. G., Allegaert, K., Basit, A. W., Braeckmans, M., Ceulemans, J., Corsetti, M., Griffin, B. T., Grimm, M., Keszthelyi, D., Koziolek, M., Madla, C. M., Matthys, C., McCoubrey, L. E., Mitra, A., Reppas, C., Stappaerts, J., Steenackers, N., … Augustijns, P. (2021). Impact of gastrointestinal tract variability on oral drug absorption and pharmacokinetics: An UNGAP review. *European Journal of Pharmaceutical Sciences*, *162*(March). https://doi.org/10.1016/j.ejps.2021.105812

Volokitina, Z. V., Ionova, I. I., & Krasnova, I. S. (2020). Whey beverage with collagen hydrolysate from salmon skin and feijoa. *IOP Conference Series: Earth and Environmental Science*, *421*(3), 1–6. https://doi.org/10.1088/1755-

1315/421/3/032017

Walimbe, T., & Panitch, A. (2020). Best of Both Hydrogel Worlds : Harnessing Bioactivity and Tunability by Incorporating Glycosaminoglycans in Collagen Hydrogels. *Bioengineering*, *7*(156). https://doi.org/10.3390/bioengineering7040156

Xie, N. Z., Du, Q. S., Li, J. X., & Huang, R. B. (2015). Exploring strong interactions in proteins with quantum chemistry and examples of their applications in drug design. *PLoS ONE*, *10*(9), 1–19. https://doi.org/10.1371/journal.pone.0137113

Yan, Y., Zhang, D., Zhou, P., Li, B., & Huang, S. Y. (2017). HDOCK: A web server for protein-protein and protein-DNA/RNA docking based on a hybrid strategy. *Nucleic Acids Research*, *45*(W1), W365–W373. https://doi.org/10.1093/nar/gkx407

Yang, X., Chen, K., Liu, H., Zhang, Y., & Luo, Y. (2019). Purification and identification of peptides with high angiotensin-I converting enzyme (ACE) inhibitory activity from honeybee pupae (Apis mellifera) hydrolysates with in silico gastrointestinal digestion. *European Food Research and Technology*, *245*(3), 535–544. https://doi.org/10.1007/s00217-018-03223-7

Yu, Z., Chen, Y., Zhao, W., Li, J., Liu, J., & Chen, F. (2018). Identification and molecular docking study of novel angiotensin-converting enzyme inhibitory peptides from Salmo salar using in silico methods. *Journal of the Science of Food and Agriculture*, *98*(10), 3907–3914. https://doi.org/10.1002/jsfa.8908

Zamora-Sillero, J., Gharsallaoui, A., & Prentice, C. (2018). Peptides from Fish By-product Protein Hydrolysates and Its Functional Properties: an Overview. *Marine Biotechnology*, *20*(2), 118–130. https://doi.org/10.1007/s10126-018-9799-3

Zhao, W., Xue, S., Yu, Z., Ding, L., Li, J., & Liu, J. (2019). Novel ACE inhibitors derived from soybean proteins using in silico and in vitro studies. *Journal of Food Biochemistry*, *43*(9), 1–10. https://doi.org/10.1111/jfbc.12975

Zhao, Y., Wang, Z., Zhang, J., & Su, T. (2018). Extraction and characterization of collagen hydrolysates from the skin of Rana chensinensis. *3 Biotech*, *8*(3), 1–8. https://doi.org/10.1007/s13205-018-1198-y