KAJIAN POTENSI PIGMEN MIKROALGA *Spirulina platensis* SEBAGAI KANDIDAT ANTIKANKER PAYUDARA MENGGUNAKAN PENDEKATAN *MOLECULAR DOCKING*

SKRIPSI

diajukan untuk memenuhi sebagian syarat memperoleh gelar

Sarjana Sains Program Studi Kimia

oleh

Adinda Saraswati

1606659

PROGRAM STUDI KIMIA

DEPARTEMEN PENDIDIKAN KIMIA

FAKULTAS PENDIDIKAN MATEMATIKA DAN ILMU PENGETAHUAN ALAM

UNIVERSITAS PENDIDIKAN INDONESIA

BANDUNG

2020

KAJIAN POTENSI PIGMEN MIKROALGA *Spirulina platensis* SEBAGAI KANDIDAT ANTIKANKER PAYUDARA MENGGUNAKAN PENDEKATAN *MOLECULAR DOCKING*

oleh

Adinda Saraswati

Sebuah skripsi yang diajukan untuk memenuhi salah satu syarat memperoleh gelar Sarjana Sains pada Fakultas Pendidikan Matematika dan Ilmu Pengetahuan Alam

ADINDA SARASWATI

KAJIAN POTENSI PIGMEN MIKROALGA *Spirulina platensis* SEBAGAI KANDIDAT ANTIKANKER PAYUDARA MENGGUNAKAN PENDEKATAN *MOLECULAR DOCKING*

disetujui dan disahkan oleh pembimbing:

Pembimbing I

Heli Siti Halimatul Munawaroh, Ph.D
NIP. 197907302001122002

Pembimbing II

Gun Gun Gumilar, S.Pd., M.Si.
NIP. 197906262001121001

Mengetahui,

Ketua Departemen Pendidikan Kimia

Dr. Hendrawan, M.Si.
NIP. 196309111989011001

ABSTRAK

Pigmen mikroalga *Spirulina platensis* dilaporkan memiliki aktivitas antikanker, antitumor, dan antiproliperatif. Namun, kajian interaksi molekulernya masih belum banyak diteliti. Pada penelitian ini, dilakukan kajian potensi pigmen *Spirulina platensis* sebagai kandidat antikanker payudara menggunakan simulasi *molecular docking.* Simulasi dilakukan untuk pigmen *C*-fikosianin, fikosianobilin, *trans*-β-karoten, 9-*cis*-β-karoten, *trans*-zeaxanthin, dan *trans*-diatoxanthin, terhadap empat reseptor hormon kanker payudara, yaitu: 1) *estrogen receptor alpha* (ERα) dan *progesterone receptor* (PR) untuk subjenis kanker payudara reseptor hormon positif; 2) *human epidermal growth factor receptor* 2 (HER2) untuk subjenis kanker payudara reseptor hormon negatif; dan 3) *epidermal growth factor receptor* (EGFR) untuk subjenis kanker payudara *triple-negative*. Tahapan penelitian yang dilakukan meliputi: preparasi protein, validasi metode *docking*, optimasi dan preparasi ligan, serta simulasi *docking* protein-ligan dan protein-protein. Hasil simulasi *docking* protein-ligan menunjukkan bahwa pigmen 9-*cis*-β-karoten membentuk kompleks dengan stabilitas tertinggi dengan ERα, HER2, dan EGFR, sedangkan pigmen *trans*-zeaxanthin membentuk kompleks dengan stabilitas tertinggi dengan PR. Harga ΔG pigmen 9-*cis*-β-karoten dengan ERα, HER2, dan EGFR secara berturut-turut sebesar: –10,2 *kkal/mol*; –9,7 *kkal/mol*; dan –9,1 *kkal/mol*, sedangkan untuk pigmen *trans*-zeaxanthin dengan PR adalah –7,8 *kkal/mol*. Pigmen fikosianobilin, *trans*-β-karoten, 9-*cis*-β-karoten, *trans*-zeaxanthin, dan *trans*-diatoxanthin mengikat di sisi aktif keempat reseptor, sedangkan *C*-fikosianin hanya mengikat di sisi aktif ERα, PR, dan EGFR. Analisis interaksi molekuler menunjukkan bahwa interaksi pigmen dengan reseptor di sisi pengikatan didominasi oleh gaya van der Waals dan interaksi hidrofobik. Simulasi *docking* protein-protein menunjukkan bahwa *C*-fikosianin menghasilkan harga afinitas pengikatan tertinggi dengan ERα (ΔG = –55,32 *kkal/mol*). Berdasarkan hasil penelitian, disimpulkan bahwa pigmen dari mikroalga *Spirulina platensis* berpotensi sebagai kandidat antikanker payudara.

Kata kunci: antikanker, *molecular docking*, pigmen, *Spirulina platensis*

ABSTRACT

Pigments of microalgae Spirulina platensis has been reported to exhibit anticancer, antitumor, and antiproliferative activities. However, the molecular interaction of pigments in the function of anticancers are still rare explored. In this study, a molecular docking simulation was carried out to screen the potential of pigments of Spirulina platensis as anti-breast cancer candidates. Molecular docking simulations was performed between C-phycocyanin, phycocyanobilin, trans-β-carotene, 9-*cis*-β-*carotene, trans-zeaxanthin, and trans-diatoxanthin pigments with four breast cancer hormone receptors i.e.* ERα (*estrogen receptor alpha*) *and* PR (*progesterone receptor*) *for receptor hormone positive breast cancer subtype,* HER2 (*human epidermal growth factor receptor* 2) *for receptor hormone negative breast cancer subtype, and* EGFR (*epidermal growth factor receptor*) *for triple-negative breast cancer subtype. Research stages include: preparation of protein*; *docking methods validation*; *and preparation of ligand*; *protein-ligand and protein-protein docking simulations. Protein-ligand docking simulations displayed that* 9-*cis*-β-*carotene formed the most stable complexes with* ERα, HER2, and EGFR, *while trans-zeaxanthin formed the most stable complex with* PR. *The free energy of binding* (ΔG) *of* 9-*cis*-β-*carotene and* ERα, HER2, *and* EGFR *were*: –10.2 *kcal/mol*; –9.7 *kcal/mol*; –9.1 *kcal/mol respectively, whereas the free energy of binding* (ΔG) *of trans-zeaxanthin, and* PR *were* –7.8 *kcal/mol. Phycocyanobilin, trans-β-carotene,* 9-*cis*-β-*carotene, trans-zeaxanthin, and trans-diatoxanthin binds to the active site of all four receptors, whereas C-phycocyanin binds to the active site of* ERα, PR, *and* EGFR. *Molecular interactions indicated that pigments and receptors interactions were dominated by van der Waals forces and hydrophobic interactions. Protein-protein docking simulations suggested that C-phycocyanin formed complex with the highest free energy of binding with* ERα (ΔG = –55.32 *kcal/mol*). *Research result shown that pigments of microalgae Spirulina platensis has potential as anti-breast cancer candidates.*

Keywords: *anticancer, molecular docking, pigments, Spirulina platensis*

DAFTAR ISI

DAFTAR PUSTAKA

Abd El-Hack, M. E., Abdelnour, S., Alagawany, M., Abdo, M., Sakr, M. A., Khafaga, A. F., Mahgoub, S. A., Elnesr, S. S., & Gebriel, M. G. (2019). Microalgae in modern cancer therapy: Current knowledge. *Biomedicine and Pharmacotherapy*, *111*, 42–50. doi: https://doi.org/10.1016/j.biopha.2018.12.069

Acharya, R., Chacko, S., Bose, P., & Lapenna, A. (2019). Structure Based Multitargeted Molecular Docking Analysis of Selected Furanocoumarins against Breast Cancer. *Scientific Reports*, 1–13. doi: https://doi.org/10.1038/s41598-019-52162-0

Aertgeerts, K., Skene, R., Yano, J., Sang, B. C., Zou, H., Snell, G., Jennings, A., Iwamoto, K., Habuka, N., Hirokawa, A., Ishikawa, T., Tanaka, T., Miki, H., Ohta, Y., & Sogabe, S. (2011). Structural analysis of the mechanism of inhibition and allosteric activation of the kinase domain of HER2 protein. *Journal of Biological Chemistry*, *286*(21), 18756–18765. doi: https://doi.org/10.1074/jbc.M110.206193

Allouche, A. (2012). Software News and Updates Gabedit — A Graphical User Interface for Computational Chemistry Softwares. *Journal of Computational Chemistry*, *32*, 174–182. doi: https://doi.org/10.1002/jcc

Arboleda, V. A., Quigley, C. A., & Vilain, E. (2015). Genetic Basis of Gonadal and Genital Development. In *Endocrinology: Adult and Pediatric* (Seventh Ed, Vols. 2–2). doi: https://doi.org/10.1016/B978-0-323-18907-1.00118-9

Arthur, D. E., & Uzairu, A. (2019). Molecular docking studies on the interaction of NCI anticancer analogues with human Phosphatidylinositol 4,5-bisphosphate 3-kinase catalytic subunit. *Journal of King Saud University - Science*, *31*(4), 1151–1166. doi: https://doi.org/https://doi.org/10.1016/j.jksus.2019.01.011

Athanasakoglou, A., & Kampranis, S. C. (2019). *Diatom isoprenoids : Advances and biotechnological potential*. *37*. doi: https://doi.org/10.1016/j.biotechadv.2019.107417

Baker, M. E., & Lathe, R. (2017). *The promiscuous estrogen receptor : evolution of physiological estrogens and response to phytochemicals and endocrine disruptors*. 1–20.

Banakar, V., Alam, Q., Pandit, A., Cladious, A., & Foundation, P. (2020). *INTERNATIONAL JOURNAL OF RESEARCH IN Spirulina , The Boon of Nature*. *11*(1), 57–62.

Baspinar, A., Cukuroglu, E., Nussinov, R., Keskin, O., & Gursoy, A. (2014). PRISM: A web server and repository for prediction of protein-protein interactions and modeling their 3D complexes. *Nucleic Acids Research*, *42*(W1), 285–289. https://doi.org/10.1093/nar/gku397

Bharathiraja, S., Seo, H., Manivasagan, P., Moorthy, M. S., Park, S., & Oh, J. (2016). In vitro photodynamic effect of phycocyanin against breast cancer

cells. *Molecules*, *21*(11), 1–12. https://doi.org/10.3390/molecules21111470

Brisken, C., Hess, K., & Jeitziner, R. (2015). Progesterone and overlooked endocrine pathways in breast cancer pathogenesis. *Endocrinology*, *156*(10), 3442–3450. https://doi.org/10.1210/en.2015-1392

Brylinski, M. (2018). Aromatic interactions at the ligand-protein interface: Implications for the development of docking scoring functions. *Chemical Biology & Drug Design*, *91*(2), 380–390. https://doi.org/10.1111/cbdd.13084

Bulusu, G., & Desiraju, G. R. (2020). Strong and Weak Hydrogen Bonds in Protein–Ligand Recognition. *Journal of the Indian Institute of Science*, *100*(1), 31–41. https://doi.org/10.1007/s41745-019-00141-9

Chaiklahan, R., Chirasuwan, N., & Bunnag, B. (2012). Stability of phycocyanin extracted from Spirulina sp.: Influence of temperature, pH and preservatives. *Process Biochemistry*, *47*(4), 659–664. https://doi.org/10.1016/j.procbio.2012.01.010

Chan, H. K., & Ismail, S. (2014). Side effects of chemotherapy among cancer patients in a Malaysian general hospital: Experiences, perceptions and informational needs from clinical pharmacists. *Asian Pacific Journal of Cancer Prevention*, *15*(13), 5305–5309. https://doi.org/10.7314/APJCP.2014.15.13.5305

Changavi, A., Shashikala, A., & Ramji, A. (2015). Epidermal growth factor receptor expression in triple negative and nontriple negative breast carcinomas. *Journal of Laboratory Physicians*, *7*(2), 79. https://doi.org/10.4103/0974-2727.163129

Chinnasamy, K., Saravanan, M., & Poomani, K. (2020). Evaluation of binding and antagonism/downregulation of brilanestrant molecule in estrogen receptor-α via quantum mechanics/molecular mechanics, molecular dynamics and binding free energy calculations. *Journal of Biomolecular Structure and Dynamics*, *38*(1), 219–235. https://doi.org/10.1080/07391102.2019.1574605

D'Ambrosio, C., Stigliani, A. L., & Giorio, G. (2016). Food from Genetically Engineered Plants. In *Genetically Modified Organisms in Food*. Elsevier Inc. https://doi.org/10.1016/b978-0-12-802259-7.00033-6

da Cunha Santos, G., Shepherd, F. A., & Tsao, M. S. (2011). EGFR Mutations and Lung Cancer. *Annual Review of Pathology: Mechanisms of Disease*, *6*(1), 49–69. https://doi.org/10.1146/annurev-pathol-011110-130206

Daeffler, K. N. M., Lester, H. A., & Dougherty, D. A. (2012). Functionally important aromatic-aromatic and sulfur-π interactions in the D2 dopamine receptor. *Journal of the American Chemical Society*, *134*(36), 14890–14896. https://doi.org/10.1021/ja304560x

Daniel, A. R., Hagan, C. R., & Lange, C. A. (2011). *Progesterone receptor action : defining a role in breast cancer*. 359–369.

Devillers, J. (2017). *Computational Design of Chemicals for the Control of*

Mosquitoes and Their Diseases. CRC Press. https://books.google.co.id/books?id=-zZDDwAAQBAJ

Du, X., Li, Y., Xia, Y., Ai, S., Liang, J., Sang, P., & Ji, X. (2016). *Insights into Protein – Ligand Interactions : Mechanisms , Models , and Methods*. *i*, 1–34. https://doi.org/10.3390/ijms17020144

Edwards, J. A. (2016). *Zeaxanthin : Review of Toxicological Data and Acceptable Daily Intake*. *2016*.

Eliassen, A. H., Liao, X., Rosner, B., Tamimi, R. M., Tworoger, S. S., & Hankinson, S. E. (2015). Plasma carotenoids and risk of breast cancer over 20 y of follow-up. *American Journal of Clinical Nutrition*, *101*(6), 1197–1205. https://doi.org/10.3945/ajcn.114.105080

Ellison, S. L. (2015). Carotenoids: Physiology. In *Encyclopedia of Food and Health* (1st ed.). Elsevier Ltd. https://doi.org/10.1016/B978-0-12-384947-2.00120-3

Ellmann, S., Sticht, H., Thiel, F., Beckmann, M. W., Strick, R., & Strissel, P. L. (2009). Estrogen and progesterone receptors: From molecular structures to clinical targets. *Cellular and Molecular Life Sciences*, *66*(15), 2405–2426. https://doi.org/10.1007/s00018-009-0017-3

Erijman, A., Rosenthal, E., & Shifman, J. M. (2014). How structure defines affinity in protein-protein interactions. *PLoS ONE*, *9*(10). https://doi.org/10.1371/journal.pone.0110085

Feng, Y., Spezia, M., Huang, S., Liu, B., Lei, Y., Du, S., Vuppalapati, A., Luu, H. H., Haydon, R. C., He, T., & Ren, G. (2018). ScienceDirect Breast cancer development and progression : Risk factors , cancer stem cells , signaling pathways , genomics , and molecular pathogenesis. *Genes & Diseases*, *5*(2), 77–106. https://doi.org/10.1016/j.gendis.2018.05.001

Fernandes, E., Figueira, F. S., Cañedo, A. D., Machado, K. S., Salgado, M. T. S. F., Silva, T. K., Wagner, E. F., Mattozo, F. H., Lima, É. A., Sales-neto, J. M., Ferreira, V. U., Comitre, A. A., Mascarenhas, S. R., Kalil, S. J., & Votto, A. P. S. (2018). Biomedicine & Pharmacotherapy C-phycocyanin to overcome the multidrug resistance phenotype in human erythroleukemias with or without interaction with ABC transporters. *Biomedicine & Pharmacotherapy*, *106*(June), 532–542. https://doi.org/10.1016/j.biopha.2018.06.145

Fernández-Rojas, B., Hernández-Juárez, J., & Pedraza-Chaverri, J. (2014). Nutraceutical properties of phycocyanin. *Journal of Functional Foods*, *11*(C), 375–392. https://doi.org/10.1016/j.jff.2014.10.011

Ferreira De Freitas, R., & Schapira, M. (2017). A systematic analysis of atomic protein-ligand interactions in the PDB. *MedChemComm*, *8*(10), 1970–1981. https://doi.org/10.1039/c7md00381a

Finn, R. S., Aleshin, A., & Slamon, D. J. (2016). Targeting the cyclin-dependent kinases breast cancers. *Breast Cancer Research*, 1–11. https://doi.org/10.1186/s13058-015-0661-5

Forli, S., Huey, R., Pique, M. E., Sanner, M. F., Goodsell, D. S., & Olson, A. J. (2016). Computational protein-ligand docking and virtual drug screening with the AutoDock suite. *Nature Protocols*, *11*(5), 905–919. https://doi.org/10.1038/nprot.2016.051

Fu, Y., Zhao, J., & Chen, Z. (2018). Insights into the Molecular Mechanisms of Protein-Ligand Interactions by Molecular Docking and Molecular Dynamics Simulation: A Case of Oligopeptide Binding Protein. *Computational and Mathematical Methods in Medicine*, *2018*. https://doi.org/10.1155/2018/3502514

Garg, S., Magro, M., & Serruys, P. W. (2011). *6.627 - Drug-Eluting Stents* (P. B. T.-C. B. Ducheyne (ed.); pp. 427–448). Elsevier. https://doi.org/https://doi.org/10.1016/B978-0-08-055294-1.00232-4

Gleiter, R. (1987). Pi-sigma interactions: Experimental evidence and its consequences for the chemical reactivity of organic compounds. *Pure and Applied Chemistry*, *59*(12), 1585–1594. https://doi.org/10.1351/pac198759121585

Gong, M., & Bassi, A. (2016). Carotenoids from microalgae: A review of recent developments. *Biotechnology Advances*. https://doi.org/10.1016/j.biotechadv.2016.10.005

Grune, T., Lietz, G., Palou, A., Ross, A. C., Stahl, W., Tang, G., Thurnham, D., Yin, S., & Biesalski, H. K. (2010). *b -Carotene Is an Important Vitamin A Source*. https://doi.org/10.3945/jn.109.119024.ants

Hammond, M. E. H., Hayes, D. F., Dowsett, M., Allred, D. C., Hagerty, K. L., Badve, S., Fitzgibbons, P. L., Francis, G., Goldstein, N. S., Hayes, M., Hicks, D. G., Lester, S., Love, R., Mangu, P. B., McShane, L., Miller, K., Osborne, C. K., Paik, S., Perlmutter, J., … Wolff, A. C. (2010). American society of clinical oncology/college of american pathologists guideline recommendations for immunohistochemical testing of estrogen and progesterone receptors in breast cancer. *Journal of Clinical Oncology*, *28*(16), 2784–2795. https://doi.org/10.1200/JCO.2009.25.6529

Handa, R. J., Larco, D. O., & Wu, T. J. B. T.-R. M. in B. S. (2014). *Steroid Hormone Action in Health and Disease*. Elsevier. https://doi.org/https://doi.org/10.1016/B978-0-12-801238-3.00099-4

Harbeck, N., Penault-Llorca, F., Cortes, J., Gnant, M., Houssami, N., Poortmans, P., Ruddy, K., Tsang, J., & Cardoso, F. (2019). Breast cancer. In *Nature Reviews Disease Primers* (Vol. 5, Issue 1). https://doi.org/10.1038/s41572-019-0111-2

Hefti, M. M., Hu, R., Knoblauch, N. W., Collins, L. C., Haibe-kains, B., Tamimi, R. M., & Beck, A. H. (2013). *Estrogen receptor negative / progesterone receptor positive breast cancer is not a reproducible subtype*. 1–13.

Helmestam, M., Andersson, H., Stavreus-Evers, A., Brittebo, E., & Olovsson, M. (2012). Tamoxifen modulates cell migration and expression of angiogenesis-

related genes in human endometrial endothelial cells. *American Journal of Pathology*, *180*(6), 2527–2535. https://doi.org/10.1016/j.ajpath.2012.02.026

Hempel, G. (2020). *Dose and therapy individualization in cancer chemotherapy*. 291–319. https://doi.org/10.1016/B978-0-444-64066-6.00013-7

Hewitt, S. C., & Korach, K. S. (2018). Estrogen receptors: New directions in the new millennium. *Endocrine Reviews*, *39*(5), 664–675. https://doi.org/10.1210/er.2018-00087

Hoseini, S. M., Khosravi-Darani, K., & Mozafari, M. R. (2013). Nutritional and Medical Applications of Spirulina Microalgae. *Mini-Reviews in Medicinal Chemistry*, *13*(8), 1231–1237. https://doi.org/10.2174/1389557511313080009

Hsieh-Lo, M., Castillo, G., Ochoa-Becerra, M. A., & Mojica, L. (2019). Phycocyanin and phycoerythrin: Strategies to improve production yield and chemical stability. *Algal Research*, *42*(June), 101600. https://doi.org/10.1016/j.algal.2019.101600

Hu, R., Hilakivi-Clarke, L., & Clarke, R. (2015). Molecular mechanisms of tamoxifen-associated endometrial cancer (Review). *Oncology Letters*, *9*(4), 1495–1501. https://doi.org/10.3892/ol.2015.2962

Hua, H., Zhang, H., Kong, Q., & Jiang, Y. (2018). Mechanisms for estrogen receptor expression in human cancer. *Experimental Hematology & Oncology*, 1–11. https://doi.org/10.1186/s40164-018-0116-7

Huey, R., & Morris, G. M. (2006). *Using AutoDock with AutoDockTools : A Tutorial*.

Iqbal, N., & Iqbal, N. (2014). *Human Epidermal Growth Factor Receptor 2 (HER2) in Cancers : Overexpression and Therapeutic Implications*. *2014*.

Isakoff, S. J. (2010). *Triple-Negative Breast Cancer*. *16*(1), 53–61.

Israel, B. B., Tilghman, S. L., Parker-Lemieux, K., & Payton-Stewart, F. (2018). Phytochemicals: Current strategies for treating breast cancer (review). *Oncology Letters*, *15*(5), 7471–7478. https://doi.org/10.3892/ol.2018.8304

Jacobsen, B. M., & Horwitz, K. B. (2012). Molecular and Cellular Endocrinology Progesterone receptors , their isoforms and progesterone regulated transcription. *Molecular and Cellular Endocrinology*, *357*(1–2), 18–29. https://doi.org/10.1016/j.mce.2011.09.016

Jeffrey, G. A. (1997). *An Introduction to Hydrogen Bonding*. Oxford University Press. https://books.google.co.id/books?id=ZRAFifo37QsC

Jiang, L., Wang, Y., Liu, G., Liu, H., Zhu, F., Ji, H., & Li, B. (2018). C-Phycocyanin exerts anti-cancer effects via the MAPK signaling pathway in MDA-MB-231 cells. *Cancer Cell International*, *18*(1), 1–14. https://doi.org/10.1186/s12935-018-0511-5

Jing, C., Qun, X., & Rohrer, J. (2014). *HPLC Separation of All-Trans- β -Carotene and Its Iodine-Induced Isomers Using a C30 Column*. *December*, 1–6.

Joshi, H., & Press, M. F. (2018). Molecular oncology of breast cancer. In *The Breast: Comprehensive Management of Benign and Malignant Diseases* (Fifth Edit). Elsevier Inc. https://doi.org/10.1016/B978-0-323-35955-9.00022-2

Kai, W., Yating, S., Lin, M., Kaiyong, Y., Baojin, H., Wu, Y., Fangzhou, Y., & Yan, C. (2018). Natural product toosendanin reverses the resistance of human breast cancer cells to adriamycin as a novel PI3K inhibitor. *Biochemical Pharmacology*, *152*, 153–164. https://doi.org/10.1016/j.bcp.2018.03.022

Kastritis, P. L., & Bonvin, A. M. J. J. (2013). On the binding affinity of macromolecular interactions: Daring to ask why proteins interact. *Journal of the Royal Society Interface*, *10*(79). https://doi.org/10.1098/rsif.2012.0835

Keskin, O., Gursoy, A., Ma, B., & Nussinov, R. (2008). Principles of protein-protein interactions: what are the preferred ways for proteins to interact? *Chemical Reviews*, *108*(4), 1225–1244. https://doi.org/10.1021/cr040409x

Khan, J. A., Tikad, A., Fay, M., Hamze, A., Fagart, J., Chabbert-buffet, N., Meduri, G., Amazit, L., Brion, J., Alami, M., Lombès, M., & Loosfelt, H. (2013). *A New Strategy for Selective Targeting of Progesterone Receptor with Passive Antagonists*. *C*, 1–16. https://doi.org/10.1210/me.2012-1328

Khoo, H., Prasad, K. N., Kong, K., Jiang, Y., & Ismail, A. (2011). *Carotenoids and Their Isomers: Color Pigments in Fruits and Vegetables*. 1710–1738. https://doi.org/10.3390/molecules16021710

Koníčková, R., Vaňková, K., Vaníková, J., Váňová, K., Muchová, L., Subhanová, I., Zadinová, M., Zelenka, J., Dvořák, A., Kolář, M., Strnad, H., Rimpelová, S., Ruml, T., Wong, R. J., & Vítek, L. (2014). Anti-cancer effects of blue-green alga Spirulina platensis, a natural source of bilirubin-like tetrapyrrolic compounds. *Annals of Hepatology*, *13*(2), 273–283. https://doi.org/10.1016/s1665-2681(19)30891-9

Koru, E. (2012). *Earth Food Spirulina (Arthrospira): Production and Quality Standarts*.

Krishnamurti, U., & Silverman, J. F. (2014). *HER2 in Breast Cancer : A Review and Update*. *21*(2), 100–107.

Le Goff, M., Le Ferrec, E., Mayer, C., Mimouni, V., Lagadic-Gossmann, D., Schoefs, B., & Ulmann, L. (2019). Microalgal carotenoids and phytosterols regulate biochemical mechanisms involved in human health and disease prevention. *Biochimie*, *167*, 106–118. https://doi.org/10.1016/j.biochi.2019.09.012

Lee, S., & Barron, M. G. (2017). *Structure-Based Understanding of Binding Affinity and Mode of Estrogen Receptor α Agonists and Antagonists*. 1–14. https://doi.org/10.1016/j.tiv.2007.08.004

Li, B., Chu, X., Gao, M., & Li, W. (2010). Apoptotic mechanism of MCF-7 breast cells in vivo and in vitro induced by photodynamic therapy with C-phycocyanin. *Acta Biochimica et Biophysica Sinica*, *42*(1), 80–89. https://doi.org/10.1093/abbs/gmp104

Li, H., Leung, K., Wong, M., & Ballester, P. J. (2016). *Correcting the impact of docking pose generation error on binding affinity prediction*. *17*(Suppl 11). https://doi.org/10.1186/s12859-016-1169-4

Mahajan, P., Suri, N., Mehra, R., Gupta, M., Kumar, A., Singh, S. K., & Nargotra, A. (2017). Discovery of novel small molecule EGFR inhibitory leads by structure and ligand-based virtual screening. *Medicinal Chemistry Research*, *26*(1), 74–92. https://doi.org/10.1007/s00044-016-1728-2

Maheswari, U. (2008). *Journal of Computer Science & Systems Biology Sequence and Structure Comparison Studies of Phycocyanin in Spirulina Platensis*. *January 2014*. https://doi.org/10.4172/jcsb.1000005

Makola, M. M., Dubery, I. A., Koorsen, G., Steenkamp, P. A., Kabanda, M. M., Du Preez, L. L., & Madala, N. E. (2016). The Effect of Geometrical Isomerism of 3,5-Dicaffeoylquinic Acid on Its Binding Affinity to HIV-Integrase Enzyme: A Molecular Docking Study. *Evidence-Based Complementary and Alternative Medicine*, *2016*(Ccd), 1–10. https://doi.org/10.1155/2016/4138263

Mani, S., Supriya, T., Shankar, M., Kavya Lalitha, S., Dastgiri, J., & Niranjan Babu, M. (2016). a Over View on Molecular Docking American Journal of Biological and Pharmaceutical Research a Over View on Molecular Docking. *American Journal of Biological and Pharmaceutical Research*, *3*(2), 83–89. www.mcmed.us/journal/ajbpr

Meng, X.-Y., Zhang, H.-X., Mezei, M., & Cui, M. (2012). Molecular Docking: A Powerful Approach for Structure-Based Drug Discovery. *Current Computer Aided-Drug Design*, *7*(2), 146–157. https://doi.org/10.2174/157340911795677602

Mezzomo, N., & Ferreira, S. R. S. (2016). *Carotenoids Functionality , Sources , and Processing by Supercritical Technology : A Review*. *2016*.

Morançais, M., Mouget, J., & Dumay, J. (2018). Proteins and Pigments. In *Microalgae in Health and Disease Prevention*. Elsevier Inc. https://doi.org/10.1016/B978-0-12-811405-6.00007-4

Motherwell, W. B., Moreno, R. B., Pavlakos, I., Arendorf, J. R. T., Arif, T., Tizzard, G. J., Coles, S. J., & Aliev, A. E. (2018). Noncovalent Interactions of π Systems with Sulfur: The Atomic Chameleon of Molecular Recognition. *Angewandte Chemie - International Edition*, *57*(5), 1193–1198. https://doi.org/10.1002/anie.201708485

Muchtaridi, M., Syahidah, H. N., Subarnas, A., & Yusuf, M. (2017). *Molecular Docking and 3D-Pharmacophore Modeling to Study the Interactions of Chalcone Derivatives with Estrogen Receptor Alpha*. 1–12. https://doi.org/10.3390/ph10040081

Mullins, J. (2012). Hyperconjugation: A more coherent approach. *Journal of Chemical Education*, *89*(7), 834–836. https://doi.org/10.1021/ed1011986

Munawaroh, H. S.H., Darojatun, K., Gumilar, G. G., Aisyah, S., & Wulandari, A. P. (2018). Characterization of phycocyanin from Spirulina fusiformis and its

thermal stability. *Journal of Physics: Conference Series*, *1013*(1). https://doi.org/10.1088/1742-6596/1013/1/012205

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

Murillo, A. G., Hu, S., & Fernandez, M. L. (2019). Zeaxanthin: Metabolism, properties, and antioxidant protection of eyes, heart, liver, and skin. *Antioxidants*, *8*(9), 1–18. https://doi.org/10.3390/antiox8090390

Nekoei, A., & Vatanparast, M. (2019). *π-Hydrogen bonding and aromaticity: a systematic interplay study*. 623–630. https://doi.org/10.1039/c8cp07003b

Pagels, F., Guedes, A. C., Amaro, H. M., Kijjoa, A., & Vasconcelos, V. (2019). Phycobiliproteins from cyanobacteria: Chemistry and biotechnological applications. *Biotechnology Advances*, *37*(3), 422–443. https://doi.org/10.1016/j.biotechadv.2019.02.010

Park, W. S., Kim, H. J., Li, M., Lim, D. H., Kim, J., Kwak, S. S., Kang, C. M., Ferruzzi, M. G., & Ahn, M. J. (2018). Two classes of pigments, carotenoids and c-phycocyanin, in spirulina powder and their antioxidant activities. *Molecules*, *23*(8), 1–11. https://doi.org/10.3390/molecules23082065

Patil, R., Das, S., Stanley, A., Yadav, L., Sudhakar, A., & Varma, A. K. (2010). Optimized hydrophobic interactions and hydrogen bonding at the target-ligand interface leads the pathways of drug-designing. *PloS One*, *5*(8), e12029. https://doi.org/10.1371/journal.pone.0012029

Pavlin, M., Spinello, A., Pennati, M., Zaffaroni, N., Gobbi, S., Bisi, A., Colombo, G., & Magistrato, A. (2018). A Computational Assay of Estrogen Receptor α Antagonists Reveals the Key Common Structural Traits of Drugs Effectively Fighting Refractory Breast Cancers. *Scientific Reports*, *8*(1), 1–11. https://doi.org/10.1038/s41598-017-17364-4

Petit-topin, I., Turque, N., Fay, M., & Gainer, E. (2009). *Met909 Plays a Key Role in the Activation of the Progesterone Receptor and Also in the High Potency of 13-Ethyl Progestins*. *75*(6), 1317–1324. https://doi.org/10.1124/mol.108.054312.induces

Prabakarana, G., Moovendhana, M., , A. Arumugam, a A. M., And, R. D., & Sampathkumar*a, P. (2018). *Quantitative Analysis of Phytochemical Profile in Marine Quantitative Analysis of Phytochemical Profile*. *8*(June), 562–565.

Pratama, M. R. F., Poerwono, H., & Siswandono, S. (2019). Design and molecular docking of novel 5-O-Benzoylpinostrobin derivatives as anti-breast cancer. *Thai Journal of Pharmaceutical Sciences*, *43*(4), 201–212.

Quiroga, R., & Villarreal, M. A. (2016). *Vinardo : A Scoring Function Based on Autodock Vina Improves Scoring , Docking , and Virtual Screening*. 1–18.

https://doi.org/10.1371/journal.pone.0155183

Ravi, M., Tentu, S., Baskar, G., Rohan Prasad, S., Raghavan, S., Jayaprakash, P., Jeyakanthan, J., Rayala, S. K., & Venkatraman, G. (2015). Molecular mechanism of anti-cancer activity of phycocyanin in triple-negative breast cancer cells. *BMC Cancer*, *15*(1), 1–13. https://doi.org/10.1186/s12885-015-1784-x

Ribas, J., Cubero, E., Luque, F. J., & Orozco, M. (2002). Theoretical study of alkyl-π and aryl-π interactions. Reconciling theory and experiment. *Journal of Organic Chemistry*, *67*(20), 7057–7065. https://doi.org/10.1021/jo0201225

Roy, K., Kar, S., & Das, R. N. (2015). Other Related Techniques. In *Understanding the Basics of QSAR for Applications in Pharmaceutical Sciences and Risk Assessment*. https://doi.org/10.1016/b978-0-12-801505-6.00010-7

Ruban, A. V, Lavaud, J., Rousseau, B., Guglielmi, G., & Etienne, A. (2004). *The super-excess energy dissipation in diatom algae : comparative analysis with higher plants The super-excess energy dissipation in diatom algae : comparative analysis with higher plants*. *February*. https://doi.org/10.1007/s11120-004-1456-1

Samaan, T. M. A., Samec, M., Liskova, A., & Kubatka, P. (2019). Paclitaxel ' s Mechanistic and Clinical E ff ects on Breast Cancer. *Biomolecules*, *9*(789), 1–22.

Sathasivam, R., & Ki, J. S. (2018). A review of the biological activities of microalgal carotenoids and their potential use in healthcare and cosmetic industries. *Marine Drugs*, *16*(1). https://doi.org/10.3390/md16010026

Seeliger, D., & De Groot, B. L. (2010). Ligand docking and binding site analysis with PyMOL and Autodock/Vina. *Journal of Computer-Aided Molecular Design*, *24*(5), 417–422. https://doi.org/10.1007/s10822-010-9352-6

Shareef, M., Ashraf, M. A., & Sarfraz, M. (2016). Natural cures for breast cancer treatment. *Saudi Pharmaceutical Journal*, *24*(3), 233–240. https://doi.org/10.1016/j.jsps.2016.04.018

Shiau, A. K., Barstad, D., Loria, P. M., Cheng, L., Kushner, P. J., Agard, D. A., & Greene, G. L. (1998). The Structural Basis of Estrogen Receptor/Coactivator Recognition and the Antagonism of This Interaction by Tamoxifen. *Cell*, *95*(7), 927–937. https://doi.org/10.1016/S0092-8674(00)81717-1

Siebenmorgen, T., & Zacharias, M. (2020). Computational prediction of protein–protein binding affinities. *Wiley Interdisciplinary Reviews: Computational Molecular Science*, *10*(3), 1–18. https://doi.org/10.1002/wcms.1448

Silverman, R. B., & Holladay, M. W. (2014). *Chapter 3 - Receptors* (R. B. Silverman & M. W. B. T.-T. O. C. of D. D. and D. A. (Third E. Holladay (eds.); pp. 123–163). Academic Press. https://doi.org/https://doi.org/10.1016/B978-0-12-382030-3.00003-9

Singh, A. (2016). *Structure, Synthesis, and Application of Nanoparticles* (pp. 19–

76). https://doi.org/10.1016/B978-0-12-801406-6.00002-9

Sohail, M. F., Rehman, M., & Webster, T. J. (2018). *Advancements in the oral delivery of Docetaxel : challenges , current state-of-the-art and future trends*. 3145–3161.

Sokka, I. K., Ekholm, F. S., & Johansson, M. P. (2019). Increasing the Potential of the Auristatin Cancer-Drug Family by Shifting the Conformational Equilibrium [Research-article]. *Molecular Pharmaceutics*, *16*, 3600–3608. https://doi.org/10.1021/acs.molpharmaceut.9b00437

Sommella, E., Conte, G. M., Salviati, E., Pepe, G., Bertamino, A., Ostacolo, C., Sansone, F., Prete, F. Del, Aquino, R. P., & Campiglia, P. (2018). Fast profiling of natural pigments in different spirulina (arthrospira platensis) dietary supplements by DI-FT-ICR and evaluation of their antioxidant potential by pre-column DPPH-UHPLC assay. *Molecules*, *23*(5). https://doi.org/10.3390/molecules23051152

Talero, E., García-Mauriño, S., Ávila-Román, J., Rodríguez-Luna, A., Alcaide, A., & Motilva, V. (2015). Bioactive compounds isolated from microalgae in chronic inflammation and cancer. *Marine Drugs*, *13*(10), 6152–6209. https://doi.org/10.3390/md13106152

Tanos, T., Rojo, L. J., Echeverria, P., & Brisken, C. (2012). ER and PR signaling nodes during mammary gland development. *Breast Cancer Research*, *14*(4). https://doi.org/10.1186/bcr3166

Trabert, B., & Mark E. Sherman, Nagarajan Kannan Stanczyk, and F. Z. (2019). Progesterone and breast cancer. *Obstetrics and Gynecology*, *68*(5), 715–717. https://doi.org/10.1210/endrev/bnz001

Traboulsi, T., El Ezzy, M., Gleason, J. L., & Mader, S. (2017). Antiestrogens: structure-activity relationships and use in breast cancer treatment. *Journal of Molecular Endocrinology*, *58*(1), R15–R31. https://doi.org/10.1530/JME-16-0024

Traxler, P., Allegrini, P. R., Brandt, R., Brueggen, J., Cozens, R., Fabbro, D., Grosios, K., Lane, H. A., Mcsheehy, P., Meyer, T., Tang, C., Wartmann, M., Wood, J., & Caravatti, G. (2004). *AEE788 : A Dual Family Epidermal Growth Factor Receptor / ErbB2 and Vascular Endothelial Growth Factor Receptor Tyrosine Kinase Inhibitor with Antitumor and Antiangiogenic Activity*. *222584*(30), 4931–4941.

Trott, O., & Olson, A. (2009). Software news and update 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

Vemula, P. K., Campbell, N. R., Zhao, F., Xu, B., John, G., & Karp, J. M. (2011). *4.421 - Self-Assembled Prodrugs* (P. B. T.-C. B. Ducheyne (ed.); pp. 339–355). Elsevier. https://doi.org/https://doi.org/10.1016/B978-0-08-055294-1.00135-5

Vo, T., Ngo, D., & Kim, S. (2015). Nutritional and Pharmaceutical Properties of Microalgal Spirulina. In *Handbook of Marine Microalgae*. Elsevier Inc. https://doi.org/10.1016/B978-0-12-800776-1.00019-4

Wadood, A., Ahmed, N., Shah, L., Ahmad, A., Hassan, H., & Shams, S. (2013). *In-silico drug design : An approach which revolutionarised the drug discovery process In-silico drug design : An approach which revolutionarised the drug discovery process*. *December 2016*. https://doi.org/10.13172/2054-4057-1-1-1119

Wahba, H. A., & El-Hadaad, H. A. (2015). Current approaches in treatment of triple-negative breast cancer. *Cancer Biology and Medicine*, *12*(2), 106–116. https://doi.org/10.7497/j.issn.2095-3941.2015.0030

Waks, A. G., & Winer, E. P. (2019). Breast Cancer Treatment: A Review. *JAMA - Journal of the American Medical Association*, *321*(3), 288–300. https://doi.org/10.1001/jama.2018.19323

Wan, S., & Coveney, P. V. (2011). *Rapid and accurate ranking of binding affinities of epidermal growth factor receptor sequences with selected lung cancer drugs*. *January*, 1114–1127.

Wee, P., & Wang, Z. (2017). Epidermal growth factor receptor cell proliferation signaling pathways. *Cancers*, *9*(5), 1–45. https://doi.org/10.3390/cancers9050052

Widowati, W., Jasaputra, D. K., Heriady, Y., Faried, A., Rizal, R., Widodo, W. S., Wibowo, S. H. B., Kusuma, H. S. W., Girsang, E., & Nyoman Ehrich Lister, I. (2019). Dietary flavonoids against various breast cancer subtypes: A molecular docking study. *ScienceAsia*, *45*(5), 452–457. https://doi.org/10.2306/scienceasia1513-1874.2019.45.452

World Health Organization. (2019). Indonesia Source GLOBOCAN 2018. *International Agency for Research on Cancer*, *256*, 1–2. http://gco.iarc.fr/

Xu, X., Peng, W., Liu, C., Li, S., Lei, J., Wang, Z., Kong, L., & Han, C. (2019). Flavone-based natural product agents as new lysine-specific demethylase 1 inhibitors exhibiting cytotoxicity against breast cancer cells in vitro. *Bioorganic and Medicinal Chemistry*, *27*(2), 370–374. https://doi.org/10.1016/j.bmc.2018.12.013

Yang, L., Tseng, H., Lin, C., Chen, L., Chen, S., Kuo, S., & Chen, D. (2012). *Breast Cancer Survival Benefit of Tamoxifen in Estrogen Receptor-Negative and Progesterone Receptor-Positive Low Grade Breast Cancer Patients*. *15*(3), 288–295.

Yewale, C., Baradia, D., Patil, S., & Bhatt, P. (2018). AC SC. *Journal of Drug Delivery Science and Technology*. https://doi.org/10.1016/j.jddst.2018.03.027

Yuan, S., Chan, H. C. S., & Hu, Z. (2017). *Using PyMOL as a platform for computational drug design*. *7*(April), 1–10. https://doi.org/10.1002/wcms.1298

Yun, C., Boggon, T. J., Li, Y., Woo, M. S., Greulich, H., Meyerson, M., & Eck, M. J. (2007). *Article Structures of Lung Cancer-Derived EGFR Mutants and Inhibitor Complexes : Mechanism of Activation and Insights into Differential Inhibitor Sensitivity*. *March*, 217–227. https://doi.org/10.1016/j.ccr.2006.12.017

Zaid, A. A. A., Hammad, D. M., & Sharaf, E. M. (2015). Antioxidant and anticancer activity of Spirulina platensis water extracts. *International Journal of Pharmacology*, *11*(7), 846–851. https://doi.org/10.3923/ijp.2015.846.851

Zhang, X., Spiegelman, D., Baglietto, L., Bernstein, L., Boggs, D. A., Brandt, P. A. Van Den, Julie, E., Gapstur, S. M., Giles, G. G., Giovannucci, E., Goodman, G., Hankinson, S. E., Helzlsouer, K. J., Horn-ross, P. L., Inoue, M., Jung, S., Khudyakov, P., Larsson, S. C., Lof, M., … Walter, C. (2012). *Carotenoid intakes and risk of breast cancer defined by estrogen receptor and progesterone receptor status : a pooled analysis of 18 prospective cohort studies 1 – 3*. *C*. https://doi.org/10.3945/ajcn.111.014415

Zhang, Y., Wang, Y., & Xue, J. (2018). Paclitaxel inhibits breast cancer metastasis via suppression of aurora kinase-mediated cofilin-1 activity. *Experimental and Therapeutic Medicine*, *15*(2), 1269–1276. https://doi.org/10.3892/etm.2017.5588

Zheng, L., Lin, V. C., & Mu, Y. (2016). *Exploring Flexibility of Progesterone Receptor Ligand Binding Domain Using Molecular Dynamics*. 1–27. https://doi.org/10.1371/journal.pone.0165824

Zheng, L., Xia, K., & Mu, Y. (2019). *Ligand Binding Induces Agonistic-Like Conformational Adaptations in Helix 12 of Progesterone Receptor Ligand Binding Domain*. *7*(May), 1–13. https://doi.org/10.3389/fchem.2019.00315

Zhu, C., Gao, Y., Li, H., Meng, S., Li, L., Francisco, J. S., & Zeng, X. C. (2016). Characterizing hydrophobicity of amino acid side chains in a protein environment via measuring contact angle of a water nanodroplet on planarpeptide network. *Proceedings of the National Academy of Sciences of the United States of America*, *113*(46), 12946–12951. https://doi.org/10.1073/pnas.1616138113