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MOLECULAR DOCKING OF BREAST CANCER RECEPTORS AGAINST ANTI-CANCER DRUG SOLASODINE

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ABSTRACT

Cancer is a deadly disease that occurs in skin, pancreas, breast, and other body parts. Breast cancer is nowadays more common and can develop from various cell lines like MCF-7, MDA-MB-231, BT-20, etc. Early diagnosis and treatment can reduce the frequency of fatalities due to breast cancer, but early cancer screenings are often postponed due to erroneous information, fear, and ignorance. In the present work, we used molecular docking to find out the effect of solasodine binding on different cancer receptors. Solasodine was extracted from black nightshade (*Solanum nigrum*), a weed that was long used against stomach ache in Ayurveda. The isolated solasodine was even used in ointments against skin cancer. Docking such a medicinally potent compound resulted in positive response against a few receptors like Farnesoid X, epidermal growth factor receptor and progesterone receptor. MCF-7 breast cancer cell line was chosen for further *ex-vivo* studies and the results of MTT assay showed significant anti-cancer effects at solasodine concentrations of 500 and 1000 µg mL^{-1}. These findings indicated that Solasodine was a vital candidate as drug against breast cancer.

Keywords: Autodock, breast cancer, discovery studio, Farnesoid x receptor, molecular docking, solasodine

INTRODUCTION

Cancer incidence is anticipated to increase significantly, with 27 million more cases forecast by 2040 (Arnold *et al*., 2022). There are over 2.3 million new instances of breast cancer each year. In almost 95% of countries, breast cancer is the second or main cause of mortality for women (Koo *et al*., 2017). It is estimated that around 4.4 million women are dying from cancer worldwide, making one million children as orphans, with breast cancer accounting for 25% of these deaths (Mattiuzzi and Lippi, 2019). Despite advances in the disease management, chemotherapy remains the most popular method of breast cancer treatment resulting in severe adverse effects like hair fall also (Curtis *et al*., 1992).

There is resurgence in the interest in solasodine-containing species of *Solanum*, not only for their ability in potential conversion to synthetic drugs but also for the anti-cancer properties they possess. *Solanum nigrum* was one such plant which was traditionally used in several ailments including stomach ache, cough, asthma. The isolated solasodine was even used in the formulation of ointments against skin cancer (Patel *et al*., 2013).

The method of molecular docking supports in predicting how a potential medicine will interact with a target protein. Molecular docking tools seem promising in drug discovery and design, and

forecast the binding affinity of small compounds to particular target proteins (Meng *et al*., 2011). SwissADME (*http://www.swissadme.ch/*) is used to study the solasodine drug-likeness by using the conical smile format and to predict the pharmacokinetic, drug-likeness, ADME parameters (adsorption, distribution, metabolism, and extraction) and physicochemical properties (Daina and Zoete, 2017). PubChem (*https://pubchem.ncbi.nlm.nih.gov/*) is a public database of chemical substances, including information on their biological activities and properties (Kim *et al*., 2023). Autodock uses PubChem to retrieve small molecules' 3D coordinates and properties for docking simulations. Similarly, SMILES Translator (*https://cactus.nci.nih.gov/translate/*) is a tool that converts chemical structures represented by linear notation known as SMILES (Simplified Molecular Input Stream Input System) into 3D structures that can be used in assembly simulations. Autodock uses SMILES Translator to generate 3D structures of small molecules from their SMILES representation. Protein Data Bank (PDB) (*https://www.rcsb.org/*) is a database of biological macromolecules, including proteins, nucleic acids, and complex assemblies, that provides access to search and download structural data and advanced tools for data visualization and analysis (Berman *et al*., 2003). Discovery Studio 2021 (Biovia Discovery Studio Client 21.1 developed by Dassault systems) include its powerful visualization and analysis capabilities, its ability to perform molecular docking simulations, its support for homology modeling and protein structure prediction, and its integration with other software tools and databases (Bhagyashree and Sachin, 2021). AutoDock Vina (developed by the Molecular Graphics Lab at The Scripps Research Institute) is well-known for its capability to perform quick and precise docking simulations (Eberhardt *et al*., 2021). Its efficient search algorithm can explore the conformational space of ligand and protein structures, producing reliable predictions of the binding affinity between them.

A naturally occurring substance called solasodine found in Solanaceae family of plants (Da-Ke *et al*., 2021), has previously been used in experiments on breast cancer cell lines so as to demonstrate its anticancer effects (Churiyah *et al*., 2020). Solasodine's pharmacokinetics and drug-likeness are evaluated using SwissADME (Daina and Zoete, 2017). A robust and user-friendly docking program called AutoDock Vina makes precise and effective predictions of ligand-protein interactions (Di *et al*., 2017).

The present study was aimed to investigate the probable binding sites/affinities and interactions between solasodine and the selected breast cancer receptors using molecular docking simulations like Autodock Vina 1.5.6. *Ex vivo* analysis using the MTT assay is a widely used method to evaluate the cytotoxicity of compounds against cancer cells. The MTT assay measures the ability to live cells to reduce a yellow tetrazolium salt to a purple formazan product, which is proportional to the number of viable cells. The MTT assay is a convenient and reliable method for screening compounds for their anticancer activity (Gowing *et al*., 2017).

MATERIALS AND METHODS

Plant collection and extraction

For the present study, fresh disease-free *Solanum nigrum* samples were collected from the dry and shady areas of Karur district of Tamil Nadu (India) and identified as per Jain and Rao (1976) method. Fresh leaves (100 g) were homogenized and extracted with 150 mL of 3% aqueous acetic acid and the resulting thick slurry was filtered through a filter cloth. The filtrate and washings were mixed with equal volume of ethanol, stirred for 2 h, centrifuged and the filtrate was discarded. The brown-coloured residue was re-suspended in ethanol (50 mL) and extracted for 2 h. The alcoholic concentrate was made alkaline with ammonia (pH 9.5) and kept in refrigeration. After 2 days, a precipitate was formed and separated by centrifugation. The precipitates from the combined extracts were extracted with hot 10 mL ethanol three times, which recovered most of the glycol

alkaloid from the precipitate. The ethanol extracts were refluxed with 0.5 to 1 g activated charcoal filtered through Buckner funnel. Concentrated HCl was added to the filtrate to make it 3N and refluxed for 1 h and the alcohol was removed by distillation. On cooling, the alkaloid was crystallized as a light brown solid (Sedhupathi *et al*., 2022). The extracted alkaloid was estimated for solasodine by dissolving 5 mg crystals in 5 mL 20% acetic acid. Then 5 mL acetate buffer and 1 mL methyl orange were added and the contents shaken for 10 sec. Then 5 mL chloroform was added, shaken well for 3 min and kept standing for a few min. The chloroform layers were withdrawn into dry test tubes, dried with small amount of anhydrous Na_2SO_4 and absorbance read on a spectrophotometer at 425 nm (Saini *et al*., 2007). This crystallized solasodine was preserved for use in cytotoxicity and further assays.

Molecular docking

The structure of solasodine compound was obtained from PubChem in SMILE format. The 3D structure for compound's SMILE format was generated using an online SMILE translator. The obtained 3D structure was optimized using the Discovery Studio software. The receptor structures were obtained from the Protein Data Bank (PDB). The breast cancer receptors as shown in Table 1 were selected based on their high expression levels in breast cancer. The receptor structures obtained from PDB were prepared by removing water molecules and adding hydrogen atoms using Discovery Studio software.

The solasodine compound was docked against twelve breast cancer receptors *viz*., estrogen receptors (ER), progesterone receptor (PR), mammalian target of rapamycin (mTOR), epidermal growth factor receptor (EGFR), androgen receptor (AR), mineralocorticoid receptor retinoid X receptor (RXR) alpha, peroxisome proliferator-activated receptor (PPAR), liver X receptor (LXR), vitamin D receptor (VDR), cyclin-dependent kinase 2 (CDK2) and farnesoid X receptor (FXR) using AutoDock Vina software. The docking results were visualized using Discovery Studio software (Samy and Xavier, 2015). The binding energies and interactions between the solasodine compound and the receptors were then analyzed to select a particular receptor to focus on cytotoxicity assay.

Ex vivo analysis

The MCF-7 cell line for breast cancer was obtained from Veterinary College, Veppery, Chennai and all the chemicals and media were procured from Sigma Aldrich, Mumbai and Hi-media Laboratories. MTT assay was followed as per the standard protocol (Riss *et al*., 2013). For this, the cells (1×10^5 well^{-1}) were plated in 24-well plates and incubated at 37°C with 5% CO_2 condition. After the cell reached the confluence, the various concentrations of solasodine were added and incubated for 24 hr. The solasodine was then removed from the well and washed with phosphate-buffered saline (pH 7.4). 100µl/well (5mg/ml) of 0.5% 3-(4,5-dimethyl-2-thiazolyl)-2,5-diphenyl--tetrazolium bromide (MTT) was added and incubated for 4 h. Then 1 mL DMSO was added in all the wells. The absorbance was measured with UV-spectrophotometer at 570 nm using DMSO as blank. The concentration required for 50% inhibition (IC_{50}) was determined graphically. The cell viability of MCF7 cell line was calculated using the formula:

$$\text{Cell viability (\%)} = \frac{\text{Absorbance at 570 nm of treated cells}}{\text{Absorbance at 570 nm of control cells}} x\ 100$$

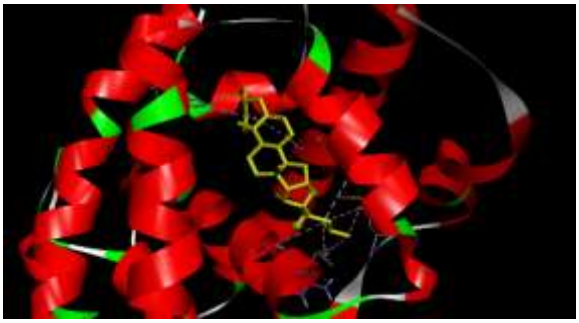
RESULTS AND DISCUSSION

Solasodine was extracted from the fresh leaves of *Solanum nigrum* and the crystals obtained were found to have 88.9% (w/w) solasodine as per spectrophotometric analysis. This was almost similar amount of solasodine obtained from *Solanum xanthocarpum* by Saini *et al*. (2007). The twelve

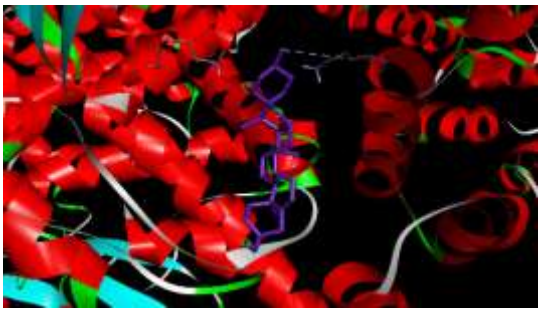
receptors used for molecular docking of solasodine were obtained from PDB and their details are given in Table 1. SwissADME revealed the physicochemical property of solasodine drug which was obtained from the conical smile format from PubChem (Table 2). The molecular docking analysis revealed that solasodine had best binding affinity towards Farnesoid X receptor (FXR) (Fig. 1). FXR is not only involved in breast cancer, but also in various diseases like non-alcoholic fatty liver disease, type 2 diabetes, and inflammatory bowel disease (IBD) (Albi *et al*., 2023).

Table 1: Receptors obtained from PDB and used for molecular docking of solasodine

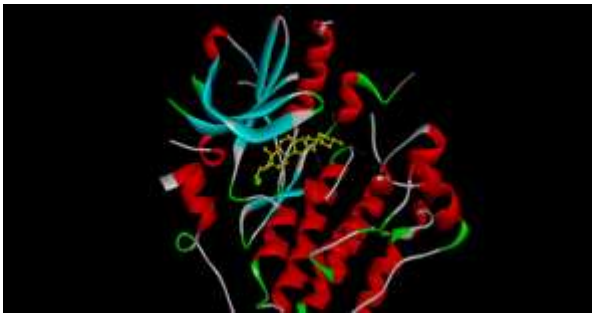
Receptors	PDB ID	Role in breast cancer	References
Estrogen receptors (ER)	7KBS	The expression of signaling elements of the insulin-like growth factor system is targeted by ER-, which stimulates the proliferation of breast cancer cells.	Hefti *et al*. (2013)
Progesterone receptor (PR)	1E3K	PR regulates estrogen receptor alpha and the mutation can result in breast cancer.	Hefti *et al*. (2013)
Mammalian target of rapamycin (mTOR)	4JVS	ErbB family receptors become more active or changes or mutations to PI3K signaling are caused by a mutation in Mtor.	Toomey *et al*. (2017)
Epidermal growth factor receptor (EGFR)	3POZ	EGFR overexpression results in large tumor size.	Masuda *et al*. (2012)
Androgen receptor (AR)	1E3G	AR is frequently expressed in breast cancer. Its mechanism is a little complex.	Yu *et al*. (2011)
Mineralocorticoid receptor	2AA2	MR has been shown to interplay with the progesterone receptor in breast cancer cells to promote cell adherence and growth inhibition.	Iacopetta *et al*. (2012)
Retinoid X receptor (RXR) alpha	1MVC	RXR involves in cell growth and differentiation, mutation results in breast cancer.	Crowe and Chandraratna (2004)
Peroxisome proliferator-activated receptor (PPAR)	2P54	Peroxisome proliferator-activated receptors, or PPARs, are transcription factors that control the expression of genes related to cell differentiation, inflammation, and lipid metabolism.	Crowe and Chandraratna (2004)
Liver X receptor (LXR)	3KFC	The LXR receptor, which is high in the breast cancer cell, maintains cholesterol metabolism.	Hutchinson *et al*. (2021)
Vitamin D receptor (VDR)	1DB1	VDR can control breast cancer cell behavior in invitro as well as in ex vivo studies.	Trivedi *et al*. (2017)
Cyclin-dependent kinase 2 (CDK2)	2B54	CDK control the spread of breast cancer cell, as it involves proliferation activity.	Fernandez *et al*. (1998)
Farnesoid X receptor (FXR)	3DCT	FXR is a promising target to inhibit aromatase in breast cancer therapy, especially it targets alternative promoter II, which is functional in breast tissue but not in other tissues such as bone.	Swales *et al*. (2006)



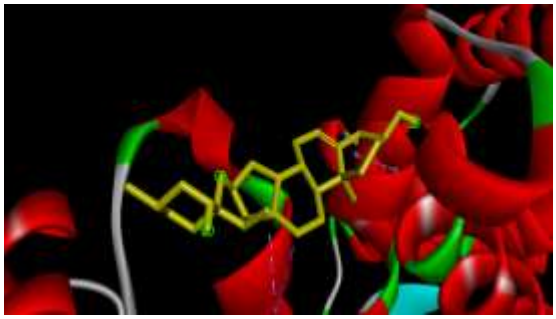
a. FXR with solasodine



c. EGFR with solasodine



b. mTor with solasodine



***d.* PR with solasodine**

Fig. 1: Docking results showing ligand-receptors binding interaction

Table 2: Physicochemical properties of solasodine obtained from SwissADME

Property	Value
Formula	$C_{27}H_{43}NO_2$
Molecular weight	413.64 g mol^{-1}
No. of heavy atoms	30
No. of aromatic heavy atoms	0
Fraction Csp3	0.93
No. of rotatable bonds	0
No. of H-bond acceptors	3
Num. H-bond donors	2
Molar refractivity	127.23
TPSA	41.49 $Å^2$

The binding energies of solasodine to the progesterone receptor (PR) and the epidermal growth factor receptor (EGFR) were tested and found to be -9.7 and -9.6 kcal mol^{-1}, respectively (Table 3). These numbers indicate that solasodine has a high affinity for these receptor bindings (Samy and Xavier, 2015; Renadi *et al*., 2023). Solasodine also displayed a modest propensity for binding to a number of additional molecular targets, in addition to EGFR and PR. The estrogen receptors, human epidermal growth factor 2, mTOR protein, androgen receptor, liver X receptor, vitamin D receptor, and cyclin-dependent kinase 2 were some of these targets.

Table 3: The binding affinity of solasodine with different receptors

Receptors	PDB ID	Binding affinities
Farnesoid X receptor (FXR)	3DCT	-10.7
Epidermal growth factor receptor (EGFR)	3POZ	-9.7
Progesterone receptor (PR)	1E3K	-9.6
Mammalian target of rapamycin (mTOR)	4JVS	-9.2
Estrogen receptors (ER)	7KBS	-9.0
Liver X receptor (LXR)	3KFC	-8.8
Androgen receptor (AR)	1E3G	-8.4
Cyclin-dependent kinase 2 (CDK2)	2B54	-8.3
Mineralocorticoid receptor	2AA2	-7.9
Retinoid X receptor (RXR) alpha	1MVC	-7.9
Vitamin D receptor (VDR)	1DB1	-7.7
Peroxisome proliferator-activated receptor (PPAR)	2P54	-7.5

Table 3 gives the binding energies (kcal mol^{-1}) of solasodine for these receptors. The 3D docking results of the first 4 receptors (Fig. 1) gives an account of the location of receptor binding with the solasodine and the bonding involved in it (Renadi *et al*., 2023).

Table 4: Cytotoxicity effect of solasodine on *MCF-7* cell line

Concentration (µg mL^{-1})	Dilutions	Absorbance (OD)	Cell viability (%)
1000	Neat	0.21	58.33
500	1:1	0.23	63.88
250	1:2	0.24	71.65
125	1:4	0.26	76.12
62.5	1:8	0.31	86.11
31.2	1:16	0.32	88.21
15.6	1:32	0.34	94.44
7.8	1:64	0.35	97.22
Cell control	-	0.36	100

The MTT assay of solasodine extracted from *S. nigrum* leaves against MCF-7 cancer cell line showed significant anti-cancer effects against MCF-7 breast cancer cells at concentrations of 500 and 1000 µg mL^{-1} (Table 4). The neat concentration of solasodine had an absorbance of 0.21 and a cell viability of 58.33%, while 1:1 dilution of solasodine had an absorbance of 0.23 and a cell viability of 63.88%. The results suggested that solasodine has a dose-dependent effect on the viability of MCF-7 cells, with higher concentrations leading to greater reductions in cell viability. Similar kind of cell decrease was observed earlier in MCF7 and other cancer cells by Chen *et al*. (2022). These findings suggest that solasodine has a broad spectrum of molecular targets and has potential as a therapeutic agent in cancer treatment. More research is needed to fully understand the mechanism of action of solasodine on the receptors and also to determine its efficacy and safety in clinical settings.

Conclusions: Solasodine is a natural product that showed promising anti-cancer activity in preclinical studies. Molecular docking studies suggest that solasodine has high binding affinities for FXR, EGFR, mTOR, PR, ER, and HER2, which are important targets in cancer development and its progression. The present study revealed the potential of solasodine as a therapeutic candidate for breast cancer treatment. It also emphasizes the value of molecular docking in drug discovery and development. Further studies in this field may result in the creation of innovative and potent treatments for breast cancer patients, ultimately improving the prognosis of condition.

Conflict of interest: The authors declare that there is no conflict of interest.

REFERENCES

Albi, E., Mandarano, M., Cataldi, S., Ceccarini, M.R., Fiorani, F., Beccari, T., Sidoni, A. and Codini, M. 2023. The effect of cholesterol in MCF7 human breast cancer cells. *International Journal of Molecular Sciences.* **24**(6): 5935. [*doi: 10.3390/ijms24065935*].

Arnold, M., Morgan, E., Rumgay, H., Mafra, A., Singh, D., Laversanne, M., Vignat, J., Gralow, J.R., Cardoso, F., Siesling, S. and Soerjomataram I. 2022. Current and future burden of breast cancer: Global statistics for 2020 and 2040. *Breast*, **66**: 15-23.

Berman, H.M., Henrick, K. and Nakamura, H. 2003. Announcing the worldwide Protein Data Bank. *Nature Structural Biology*, 10:980 [*https://doi.org/10.1038/nsb1203-980*].

Bhagyashree, L.J. and Sachin H.R. 2021. Drug designing in Discovery Studio. *Asian Journal of Research Chemistry*, **14**(2): 135-138.

Chen, J., Ma, D., Zeng, C., White, L.V., Zhang, H., Teng, Y. and Lan, P. 2022. Solasodine suppress MCF7 breast cancer stem-like cells via targeting Hedgehog/Gli1. *Phytomedicine*, **107:** 154448. (*https://doi.org/10.1016/j.phymed.2022.154448*).

Churiyah, C., Ningsih, S. and Firdayani, F. 2020. The cytotoxic, apoptotic induction, and cell cycle arrest activities of *Solanum nigrum* L. ethanolic extract on MCF-7 human breast cancer cell. *Asian Pacific Journal of Cancer Prevention,* **21**(12): 3735-3741.

Crowe, D.L. and Chandraratna, R.A. 2004. Open Access A retinoid X receptor (RXR)-selective retinoid reveals that RXR-α is potentially a therapeutic target in breast cancer cell lines, and that it potentiates antiproliferative and apoptotic responses to peroxisome proliferator-activated receptor ligands. *Breast Cancer Research,* **6**(5): R546-55. [*doi: 10.1186/bcr913*].

Curtis, R.E., Boice Jr, J.D., Stovall, M., Bernstein, L., Greenberg, R.S., Flannery, J.T., Schwartz, A.G., Weyer, P., Moloney, W.C. and Hoover, R.N. 1992. Risk of Leukemia after chemotherapy and radiation treatment for breast cancer. *The New England Journal of Medicine,* **326**(26): 1745-1751.

Daina, O.M. and Zoete, V. 2017. SwissADME: A free web tool to evaluate pharmacokinetics, drug-likeness and medicinal chemistry friendliness of small molecules. *Science Reports*, **7**(1): 42717. [*doi: 10.1038/srep42717*].

Da-Ke, Z., Yi Z., Sui-Yun, C. and Kennelly J.E. 2021. *Solanum* steroidal glycoalkaloids: Structural diversity, biological activities, and biosynthesis. *Natural Product Reports*, **38**(8): 1423 [*doi: 10.1039/D1NP00001B*].

Di M.E., Toti, D. and Polticelli, F. 2017. Docking App: A user-friendly interface for facilitated docking simulations with AutoDock Vina. *Journal of Computer Aided Molecular Design,* **31**(2): 213-218.

Eberhardt, J., Santos-Martins, D., Tillack, A.F. and Forli, S. 2021. AutoDock Vina 1.2.0: New docking methods, expanded force field, and python bindings. *Journal of Chemical Information and Modeling,* **61**(8): 3891-3898.

Fernández, P.L., Jares, P., Rey, M.J., Campo, E. and Cardesa, A. 1998. Cell cycle regulators and their abnormalities in breast cancer. *Journal of Clinical Pathology - Molecular Pathology*, **51**(6): 305-309.

Gowing, G., Svendsen, S. and Svendsen, C.N. 2017. *Ex vivo* gene therapy for the treatment of neurological disorders. Functional neural transplantation IV - Translation to clinical application, Part A. *Progress in Brain Research.* **230**: 99-132.

Hefti, M.M., Hu, R., Knoblauch, N.W., Collins, L.C., Haibe-Kains, B., Tamimi, R.M. and Beck, A.H. 2013. Estrogen receptor negative/progesterone receptor positive breast cancer is not a reproducible subtype. *Breast Cancer Research*, **15**(4): R68. [*doi: 10.1186/bcr3462*].

Hutchinson, S.A., Websdale, A., Cioccoloni, G., Røberg-Larsen, H., Lianto, P., Kim, B., Rose, A., Soteriou, C., Pramanik, A., Wastall, L.M., Williams, B.J., Henn, M.A., Chen, J.J., Ma, L.., Moore, J.B., Nelson, E., Hughes, T.A. and Thorne, J.L. 2021. Liver x receptor alpha drives chemoresistance in response to side-chain hydroxycholesterols in triple negative breast cancer. *Oncogene*, **40**(16): 2872-2883.

Lacopetta, D., Rechoum, Y. and Fuqua, S.A.W. 2012. The role of androgen receptor in breast cancer, *Drug Discovery Today: Disease Mechanisms*, **9**(1-2): e19-e27. [*doi: 10.1016/j.ddmec.2012.11.003*].

Jain, S.K. and Rao, R.R. 1976. *A Handbook of Field and Herbarium Methods.* Today and Tomorrow Printers and Publishers, New Delhi, India.

Kim, S., Chen, J., Cheng, T., Gindulyte, A., He, J., He, S., Li, Q., Shoemaker, B.A., Thiessen, P.A., Yu, B., Zaslavsky, L., Zhang, J. and Bolton, E.E. 2023. PubChem 2023 update. *Nucleic Acids Research,* **51**(D1): D1373-1380. [*doi:10.1093/nar/gkac956*].

Koo, M.M., von Wagner, C., Abel, G.A., McPhail, S., Rubin, G.P. and Lyratzopoulos, G. 2017. Typical and atypical presenting symptoms of breast cancer and their associations with diagnostic intervals: Evidence from a national audit of cancer diagnosis. *Cancer Epidemiology,* **48**: 140-146.

Masuda, H., Zhang, D., Bartholomeusz, C., Doihara, H., Hortobagyi, G.N. and Ueno, N.T. 2012. Role of epidermal growth factor receptor in breast cancer. *Breast Cancer Research and Treatment*, **136**(2): 331-345.

Mattiuzzi, C. and Lippi, G. 2019. Current cancer epidemiology. *Journal of Epidemiology in Global Health,* **9**(4): 217-222.

Meng, X.Y., Zhang, H.X., Mezei, M. and Cui, M. 2011. Molecular docking: A powerful approach for structure-based drug discovery. *Current Computer Aided Drug Design,* **7**(2): 146-157.

Patel, K., Singh, R.B. and Patel, D.K. 2013. Medicinal significance, pharmacological activities, and analytical aspects of solasodine: A concise report of current scientific literature. *Journal of Acute Disease*, **2**(2): 92-98. [*doi:10.1016/S2221-6189(13)60106-7*].

Renadi, S., Pratita, A., Mardianingrum, R. and Ruswanto, R. 2023. The potency of alkaloid derivatives as anti-breast cancer candidates: *In silico* study. *Journal Kimia Valensi*, **9**(1): 89-108.

Riss, T.L., Moravec, R.A., Niles, A.L., Duellman, S., Benink, H.A., Worzella, T.J. and Minor, L. 2013. Cell viability assays (eds. S. Markossian, A. Grossman, K. Brimacombe *et al*.) Assay Guidance Manual [Internet]. Bethesda (MD): Eli Lilly & Company and the National Center for Advancing Translational Sciences. (*https://www.ncbi.nlm.nih.gov/books/NBK144065/*).

Samy, G.B. and Xavier, L. 2015. Molecular docking studies on antiviral drugs for SARS. *International Journal of Advanced Research in Computer Science and Software Engineering*, **5**(3): 75-79.

Sedhupathi, S., Mathiyazhagan, S. and Venkatkumar, G. 2022. *In vitro* production of solasodine from *Solanum nigrum*. *International Research Journal of Modernization in Engineering Technology and Science*, **4**(06): 829-838.

Swales, K.E., Korbonits, M., Carpenter, R., Walsh, D.T., Warner, T.D. and Bishop-Bailey, D. 2006. The farnesoid X receptor is expressed in breast cancer and regulates apoptosis and aromatase expression. *Cancer Research,* **66**(20): 10120-10126.

Toomey, S., Eustace, A.J., Fay, J., Sheehan, K.M., Carr, A., Milewska, M., Madden, S.F., Teiserskiene, A., Kay, E.W., O'Donovan, N., Gallagher, W., Grogan, L., Breathnach, O., Walshe, J., Kelly, C., Moulton, B., Kennedy, M.J., Gullo, G., Hill, A.D., Power, C., Duke, D., Hambly, N., Crown, J. and Hennessy, B.T. 2017. Impact of somatic PI3K pathway and ERBB family mutations on pathological complete response (pCR) in HER2-positive breast cancer patients who received neoadjuvant HER2-targeted therapies. *Breast Cancer Research*, **19**(1): 87. [*doi: 10.1186/s13058-017-0883-9*].

Trivedi, T., Zheng, Y., Fournier, P.G.J., Murthy, S., John, S., Schillo, S., Dunstan, C.R., Mohammad, K.S., Zhou, H., Seibel, M.J. and Guise, T.A. 2017. The vitamin D receptor is involved in the regulation of human breast cancer cell growth via a ligand-independent function in cytoplasm. *Oncotarget*, **8**(16): 26687-26701. [*doi:10.18632/oncotarget.15803*].

Yu, Q., Niu, Y., Liu, N., Zhang, J.Z., Liu, T.J., Zhang, R.J., Wang, S.L., Ding, X.M. and Xiao, X.Q. 2011. Expression of androgen receptor in breast cancer and its significance as a prognostic factor. *Annals of Oncology*, **22**(6): 1288-1294.

Saini, V., Midhha, A., Gupta, S., Rathore, M.S., Wate, S.P. and Bhusari, K.P. 2007. A simple spectrophotometric method for determination of solasodine in *Solanum xanthocarpum* capsule formulations. *International Journal of Plant Science*, **2**(1): 128-129.