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RESEARCH ARTICLE

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In Silico Analysis Reveals MDM2 as a Potential Target of Ursolic Acid for Overcoming Tamoxifen Resistance in Breast Cancer

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Abstract

Objective: Ursolic acid (UA) has been proven to inhibit various cancer signaling pathways; however, the involvement of UA in overcoming tamoxifen resistance remains unclear and needs further investigation. This study aims to discover the potential gene targets and explore how ursolic acid interacts with those genes to restore sensitivity to tamoxifen. **Methods:** Analyzing gene expression data from GeneCards and Swisstargetprediction for UA related genes, and the Gene Expression Omnibus (GEO) for tamoxifen resistance genes. DEGs were analyzed for functional annotation and molecular pathways using DAVID v6.8, continued with constructing a protein-protein interaction (PPI) network to highlight crucial genes associated with tamoxifen resistance using STRING-DB and Cytoscape. Genetic alteration analysis using cBioportal for target validation and consideration. Molecular docking was done using Autodock4 and PyMoL for visualisation. **Results:** The KEGG pathway and PPI network suggest that *MDM2, STAT3, TGFB1*, and MAPK1 were indicated as potential target genes of UA. Genetic alteration analysis further confirms that *MDM2* has the highest alteration, which becomes potentially targeted by UA. Molecular docking analysis confirms that UA can target *MDM2* by targeting the N-terminus site on 4HBM and 5ZXF structure. The binding energy of UA is -5.36 for 4HBM and -8.71 for 5ZXF, with all RMSD values below 2. This result shows that UA has a lower docking score than the native ligand for the 5ZXF structure. Additionally, *MDM2* is mainly involved in the PI3K-Akt pathway, which plays a role in the chemotherapy resistance mechanism. **Conclusion:** *MDM2* has become a potential target for UA to reverse the tamoxifen resistance mechanism in breast cancer.

Keywords: Ursolic acid-Tamoxifen Resistant- breast cancer- *In silico*- *MDM2*

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Introduction

Breast cancer is one of the most common cancers in women worldwide and has become one of the leading causes of death [1]. Among them, estrogen-positive (ER+) breast cancer has the most frequent case, with over 70-75% of patients [2]. This indicates that cancer developments are highly influenced by hormonal activity. So tamoxifen is usually given as the first line of therapy for ER+ breast cancer since it was classified as a selective estrogen receptor modulator (SERM) [3]. The clinical trial data demonstrated that women who took tamoxifen for 10 years experienced a 25% lower risk of breast cancer recurrence and nearly a 30% reduction in breast cancer mortality compared to those who took it for only 5 years [4]. However, with a high dosage and a more extended period of consumption, the effectiveness of tamoxifen was decreased by the development of a resistance mechanism. The resistance mechanisms associated with tamoxifen treatment often involve alterations in cell signaling, such as PI3K-Akt and MAPK pathways, changes in ER signaling, and activation of various oncogenic pathways, including those mediated by *MDM2* [5, 6]. *MDM2* is an oncogene that inhibits *p53* activity, increasing cell proliferation and survival.

Ursolic acid (UA), a natural triterpenoid found in various fruits and herbs, has garnered significant attention for its potential therapeutic effects against breast cancer [7]. Recent studies showed the UA's ability to inhibit tumour growth, induce apoptosis, and enhance the

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efficacy of conventional treatments like tamoxifen [8, 9]. However, the emergence of tamoxifen resistance in breast cancer patients poses a significant challenge to effective treatment. This report will explore the potential of ursolic acid as a therapeutic agent to overcome tamoxifen resistance in breast cancer by explicitly targeting the MDM2 protein. Moreover, molecular docking analysis will confirm the specific interaction between UA and MDM2 which will give us better understanding through the mechanism. These properties are particularly relevant in tamoxifen resistance, as MDM2 are often cause *p53* failure and implicated in disease recurrence.

The investigations will focus on how UA affects MDM2 expression and its downstream effects on *p53* signaling, aiming to elucidate a novel approach to combating tamoxifen resistance in breast cancer therapy. In silico approach was used to analyze gene expression data from the Gene Expression Omnibus (GEO) database. Differentially Expressed Genes were analyzed for functional annotation, such as Gene Ontology (GO) and molecular pathways based on the Kyoto Encyclopedia Genes Genomes (KEGG), which continued with constructing a protein-protein interaction (PPI) network and genetic alteration to highlight crucial genes associated with tamoxifen resistance. Molecular docking was done to find specific targets of resistance-related proteins. By targeting *MDM2*, UA may restore *p53* function and promote apoptotic pathways in tamoxifen-resistant breast cancer cells.

Materials and Methods

Data Collection and Processing

The tamoxifen-resistant microarray data were obtained from GEO datasets by NCBI with accession number GSE67916 [10] Those contained two samples of Tamoxifen-resistant and Tamoxifen-sensitive MCF-7 cells, and the data is normally distributed. GEO2R was used as a data analysis tool which provided at the Gene Expression Omnibus database written in R language. Data cutoffs were specifically determined (p-value < 0.05, log-fold change > 1.5, and log-fold change < −1.5) to select significant DEGs [11]. UA target genes microarrays were obtained from genecards by typing "Ursolic acid Target Genes" and from swisstargetpredictions by inputting the canonical SMILES. Both datasets were screened for finding the differentially expressed genes (*DEGs*).

Functional annotation and pathway analysis

All the DEGs were then run for GO and KEGG pathway enrichment analyses using DAVID v6.8 with a p-value < 0.05 as the cutoff value [12]. The GO analysis was analyzed based on biological processes, cellular components, and molecular function. The gene count is used as the basis for the order.

Construction of Protein-Protein Interaction (PPI) network

DEGs were then constructed into The PPI network using STRINGDB v11.0 [13] with a confidence score > 0.400 and visualized using Cytoscape Software v3.8.0 [14]. The top 10 hub genes were screened using the CytoHubba Plugin under default settings and selected according to their degree score. The included genes were designated as Ursolic acid target genes (*UTGs*).

Genetic alterations findings

Genetic alterations were run for the selected UTGs using cBioPortal for Cancer Genomics [15], using 15 studies in breast cancer as query genes. The breast cancer study included the most significant genetic alterations for a clinical case and was chosen for further analysis.

Molecular Docking Analysis

Molecular confirmation was performed on a computer that specifically has an Intel Core i5-10th Gen processor, WindowsX operating system, and 6 GB of RAM. Autodoc4 was used for docking simulation, RMSD, and binding affinity calculation. While the binding interaction and visualization are done using PyMoL. The PDB IDs of *MDM2* were searched in rcsb.org and found to be 4HBM and 5ZXF. The UA structure (Figure 1A) was drawn using Marvin Sketch, subjected to a conformational search, and prepared using Autodoc4 using the ligand menu. For docking simulation settings, the Genetic Algorithm (GA) was used as the parameters, with the number of GA runs of 50 and a population size of 300. The output file was set to a Lamarckian GA to get the binding energy, RMSD value, number of hydrogen bonds, and the amino acid residues. The protein-ligand interaction was then visualized using PyMoL to see the hydrogen bond and

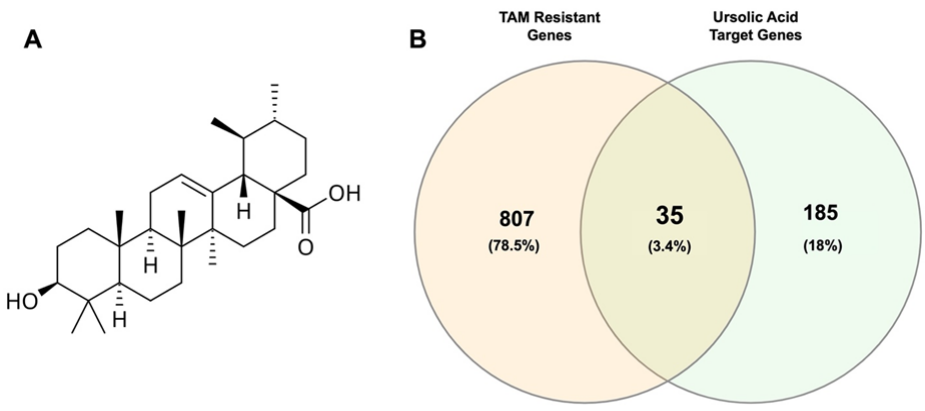

Figure 1. (A) The Chemical Structure of Ursolic Acid (UA). (B) Venn Diagram of TAMR Gene and UA Target Gene

the distance between them.

Results

Data collection and processing

The microarray data collected from GEO datasets, along with the cutoff value, resulted in 842 DEGs of tamoxifen-resistant genes. This consists of 546 upregulated genes and 433 downregulated genes. Moreover, *UA* target genes from integrating gene cards and Swisstargetprediction resulted in 220 genes. Based on the overlap in the Venn diagram, 35 genes that potentially can be targeted by UA are called ursolic acid target genes (*UTG*) (Figure 1B). Those represent the over- and underexpressed tamoxifen-resistant gene that responds to the presence of *UA*.

Gene Ontology and KEGG Pathway

Functional annotation analyses on gene ontology revealed that 35 genes are involved in 192 biological processes, 31 cellular components, and 46 molecular functions. Afterward, pathway analysis showed that all the *UTGs* were engaged in 114 signaling pathways. The results of the top GOs involved in cancer development are summarized in Table 1. The signalling pathways frequently involved in breast cancer signalling processes are shown in Table 2. Overall, the results show that all the *UTGs* are associated with cancer, particularly several genes involved in the tamoxifen-resistant mechanism.

Analysis of The PPI network and Determination of Hub Genes

To better understand the interaction among the *UTGs*, the construction of PPI networks was done. Among all

Tabel 1. Top 10 GO Based on Gene Count

ID	Term	Count	p-value
	Biological Process		
GO:0045944	Positive regulation of transcription by RNA polymerase II	11	1.90E-05
GO:0010628	Positive regulation of gene expression	9	1.10E-06
GO:0045893	Positive regulation of DNA-templated transcription	9	1.40E-05
GO:0007165	Signal transduction	9	6.40E-04
GO:0006357	Regulation of transcription by RNA polymerase II	9	3.40E-03
GO:0009410	Response to xenobiotic stimulus	8	1.40E-07
GO:0006915	Apoptotic process	8	3.70E-05
GO:0000122	Negative regulation of transcription by RNA polymerase II	8	7.40E-04
GO:0008285	Negative regulation of cell population proliferation	7	6.10E-05
GO:0043066	Negative regulation of apoptosis process	7	1.40E-04
	Cellular Component		
GO:0005737	Cytoplasm	24	5.70E-07
GO:0005634	Nucleus	23	1.10E-05
GO:0005829	Cytosol	21	5.30E-05
GO:0005654	Nucleoplasm	18	3.30E-05
GO:0016020	Membrane	15	1.60E-02
GO:0005886	Plasma membrane	15	3.70E-02
GO:0032991	Protein-containing complex	10	7.80E-07
GO:0005739	Mitochondrion	10	3.70E-04
GO:0000785	Chromatin	8	1.90E-03
GO:0005783	Endoplasmic reticulum	8	2.20E-03
	Molecular Function		
GO:0005515	Protein binding	33	1.70E-06
GO:0042802	Identical binding protein	19	1.90E-11
GO:0003677	DNA binding	9	4.70E-04
GO:0005524	ATP binding	9	3.00E-03
GO:0019899	Enzyme binding	8	2.10E-06
GO:0001228	DNA-binding transcription activator activity, RNA polymerase II-specific	8	1.30E-05
GO:0003700	DNA-binding TF activity	8	2.30E-05
GO:0000978	RNA polymerase II cis-regulatory region sequence-specific DNA binding	8	4.30E-03
GO:0000981	DNA-binding TF activity, RNA polymerase II-specific	8	6.00E-03
GO:0061629	RNA polymerase II-specific DNA-binding TF binding	7	5.10E-07

*TF, transcription factor

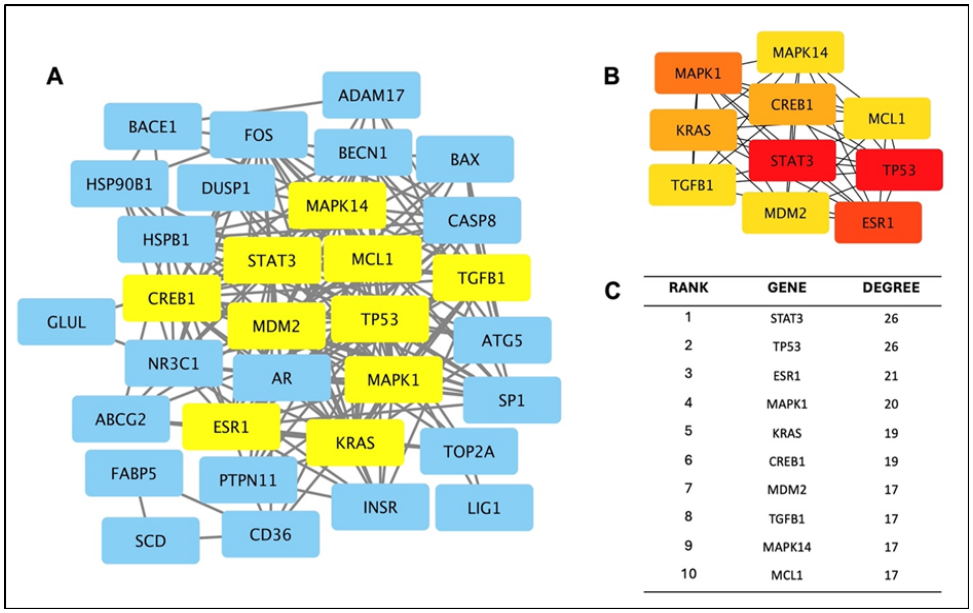

Figure 2. (A) PPI Network of UTG that Potentially Overcome Tamoxifen Resistance Analyzed by STRING-DB. (B) Hub Genes of UTG Analyzed by Cytohubba. (C) List of Top 10 UTG Based on Their Degree.

35 genes, 33 genes are detected as encoding proteins in Homo sapiens (humans) which are represented as nodes and the interaction as edges (Figure 2A.). The result was 33 nodes and 195 edges in total with PPI enrichment value of <1.0e-16 and an average of local clustering coefficient of 0.707. The result indicates that the number of observed interactions (edges) among the proteins is significantly higher than expected if the proteins were randomly selected from a larger pool. This means the proteins may work together in cellular functions or pathways, enhancing our understanding of their roles in cancer development and tamoxifen-resistant mechanisms in particular [16]. For instance, if a specific path shows a low p-value in enrichment analysis, it suggests that the proteins involved are likely to contribute to that pathway's function [17]. Further, the clustering coefficient manifests that all the genes are part of functional modules or complexes. Those indicate that they may work together in the same pathways or biological processes [18]. This is particularly relevant in cellular functions where coordinated action is essential. In studies involving specific diseases or biological processes, a high average local clustering coefficient can indicate potential targets for therapeutic intervention or highlight critical pathways involved in disease mechanisms [19], which in this case is tamoxifen-resistant. The works are then exported to Cytoscape 3.8.0 to Identify the gene with

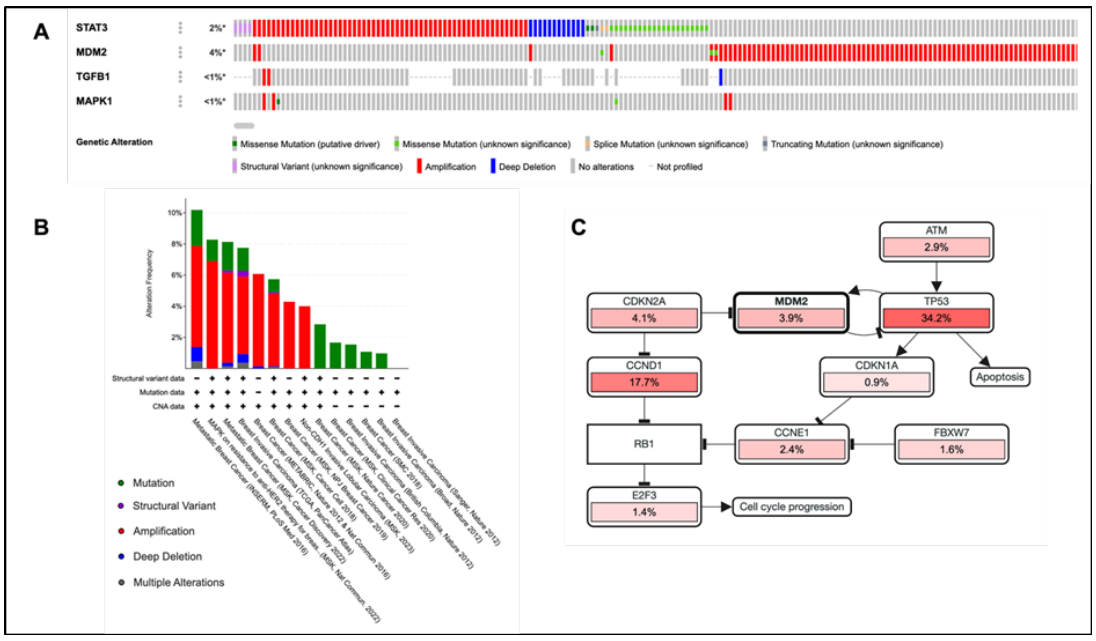

Figure 3. (A) Oncoprints summary of genetic alterations in *STAT3, MDM2, TGFB1,* and *MAPK1.* (B) Frequency of alterations in STAT3, MDM2, TGFB1, and MAPK1 in a genomic dataset obtained from 15 studies of breast cancer. (C) Pathway related to Breast Cancer Based on Altered Hub Genes as Analyzed Using the cBioPortal for Cancer Genomics Database.

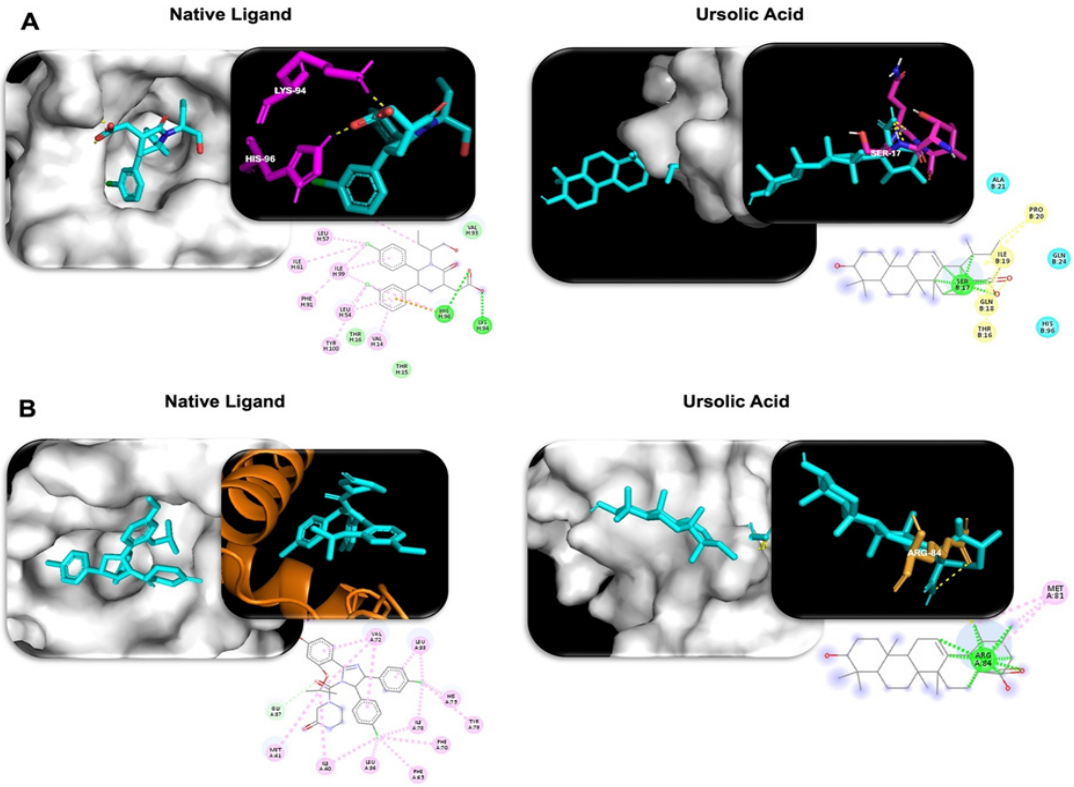

Figure 4. Docking Visualization of Both MDM2 Model Structure Targeted by UA and Native ligand. (A) 4HBM Model Structure which the UA has a Higher Binding Energy. (B) 5ZXF Model Structure, which the UA has a Lower Binding Energy.

Table 2. KEGG Pathway Related to Cancer

Term	p-Value	Genes
Pathways in Cancer	4.7E-7	*TGFB1, STAT3, FOS, ESR1, HSP90B1, AR, CASP8, SP1, MDM2, BAX, MAPK1, KRAS, TP53*
MAPK Signaling Pathway	2.6E-5	*ATF2, TGFB1, DUSP1, INSR, HSPB1, MAPK1, KRAS, FOS, MAPK14, TP53*
PI3K-Akt Signaling Pathway	4.4E-4	*ATF2, CREB1, INSR, MDM2, MAPK1, KRAS, TP53, HSP90B1, MCL1*
FoxO Signaling pathway	1.1E-4	*TGFB1, INSR, STAT3, MDM2, MAPK1, KRAS, MAPK14*

the most influence on the network. The result is shown in Figure 2A, in which the yellow colour indicates a protein degree above 15.

The analysis continued with the determination of hub genes using the CytoHubba plugin on Cytoscape. This extension could arrange the top 10 genes with the most significant impact based on their degrees. Hub genes are determined for all those 10 genes, with a Red-Yellow colour indicating a Strong-Weak interaction (Figure 2B). The 10 highly potential UTGs were then designated as potential target genes (*PTGs*) based on their involvement in several signalling pathways, as listed on Table 2. *MDM2, STAT3, TGFB1*, and *MAPK1* were further analyzed for potential mutation. *MDM2* (Mouse Double Minute 2) is a gene that encodes an E3 ubiquitin ligase, primarily known as a natural inhibitor of *p53* tumour suppressor. It binds to *p53*, promoting proteasomal degradation through ubiquitination and inhibiting its transcriptional activity [20]. STAT3 (Signal Transducer and Activator of Transcription 3) is a transcription factor that mediates cellular responses to cytokines and growth factors. It translocates to the nucleus, regulating gene expression in cell growth, survival, and differentiation [21]. *TGFB1* (Transforming Growth Factor Beta 1) is a cytokine that is responsible for several cellular processes, such as cell growth, differentiation, and immune responses [22]. *MAPK1* (Mitogen-Activated Protein Kinase 1), also known as *ERK2* involved in the MAPK signaling pathway to transfer signals from cell surface receptors to the nucleus, affecting various cellular functions, including proliferation, differentiation, and survival [23].

Analysis of Genetic Alteration of PTGs

Since cancer development is highly related to genetic mutations, this study seeks to determine how the alteration of the PTGs affects multiple breast cancer studies, i.e., The Cancer Genome Atlas (TCGA) [24]. Using the cBioportal database, which includes 15 breast cancer studies, the results showed several parameters such as mutations, amplifications, deep deletions, and multiple alterations

Table 3 Docking Result of the Interactions between UA and Native Ligand to Each MDM2 Model Structure.

PDB ID	Native Ligand				Ursolic Acid			
	Binding Energy	RMSD	H-Bond	AA	Binding Energy	RMSD	H-Bond	AA
4HBM	-11.72	1.35	1	Leu54	-5.79	1.11	1	Leu54
5ZXF	-6.56	0.69	0	-	-8.71	0.29	1	Gln51

regarding all the PTGs (Figure 3B).

The genetic alteration for each PTG is represented in oncoprint data, as shown in Figure 3A. The *MDM2* gene has excessive change with 4% alterations, with amplification and mutation having the highest changes. Other PTGs also have alterations, such as STAT3 (2%), *TGFB1* (1%), and *MAPK1* (<1%). The highest percentage of alteration indicates that *MDM2* may serve as essential genes for tamoxifen-resistant mechanisms in breast cancers, potentially providing treatment strategies, prognosis, or a guide to diagnosis. It can be prioritized and confirmed for further research to develop targeted therapies. Furthermore, the result also gives several pathways that are strongly related to the alteration of *MDM2*. The cell cycle and *TP53* pathway are mentioned, and both play a crucial role in breast cancer development (Figure 3C). The *MDM2* has a strong potential to be targeted as a protein target for UA to overcome tamoxifen-resistant mechanisms as their crucial involvement.

Molecular Docking

Previous analysis showed that *MDM2* has a high potential to be targeted with ursolic acid as it has the highest gene to be altered and plays a vital role in several carcinogenesis pathways. *MDM2* is also an E3 ubiquitin ligase that targets the tumor suppressor protein *p53* for degradation [25]. In normal conditions, *p53* regulates the cell cycle and promotes apoptosis in response to DNA damage or stress signals [26]. However, in tamoxifen-resistant breast cancer cells, elevated levels of *MDM2* can lead to decreased *p53* activity, allowing cells to evade cell cycle arrest and apoptosis induced by tamoxifen treatment [27, 28]. Targeting that gene could potentially reduce breast cancer cell resistance and restore the effectiveness of tamoxifen. Therefore, *MDM2* was chosen to be analyzed further through molecular docking to confirm its interaction with UA.

The preparation of ligands and receptors was done using AutodockTools. Two *MDM2* model structures, with PDB IDs of 4HBM and 5ZXF, were used for the docking target as well as its native ligand. The native ligand is 0Y7 ({(3R,5R,6S)-5-(3-chlorophenyl)-6-(4-chlorophenyl)-1-[(2S)-1-hydroxybutan-2-yl]-2-oxopiperidin-3-yl} acetic acid for 4HBM and NUT (4-({(4S,5R)-4,5-bis(4-chlorophenyl)- 2-[4-methoxy-2-(propan-2-yloxy)phenyl]-4,5-dihydro-1H-imidazol-1-yl}carbonyl)piperazin-2-one) for 5ZXF. The grid box was set at the same point as each native ligand using grid point spacing of 0.500 Å and x,y,z-points of 50,40,40, respectively. The binding energy at the 4HBM structure for UA was two times higher than those with native ligands. The lower binding energy indicates a better docking result, which means those ligands have less effort to bind to the protein target. But the unique thing is that both 0Y7 and UA ligand have the same interaction with Leu54 to form a hydrogen bond. On the other hand, at the 5ZXF structure, UA has a lower binding energy than the native ligand, which means UA has a better affinity to target *MDM2*. This result was also confirmed by the hydrogen bond formed on UA and 5ZXF via Gln51, where native ligands do not have any (Table 3).

Discussion

Several analyses, including molecular docking confirmation of *UA* target genes for overcoming tamoxifen resistance, revealed *MDM2* as a robust potential gene target. Previously, gene enrichment analysis showed that the gene ontology (GO) of biological processes for several genes is intensely embroiled in the regulation of transcription by RNA polymerase II. That regulation is significantly linked to drug-resistance mechanisms, which are strongly associated with the *MDM2* gene in breast cancer. For tamoxifen-resistant breast cancer, the positive regulation of transcription by RNA polymerase II may be enhanced due to overexpression of *MDM2* [29]. The enhanced regulation will lead to the escalation of the gene transcription that promotes cell survival and proliferation while inhibiting apoptotic pathways, thus contributing to drug resistance [30]. The cytoplasm, nucleus, and cytosol were identified as the primary locations of those genes which *MDM2* is mostly located in the nucleus, but it also has functions in the cytoplasm. The high number of *MDM2* will lead to enhanced degradation of *p53*, which means it will inhibit apoptosis of tumor cells [6]. Proteasomal degradation processes will occur in both compartments, allowing cancer cells to evade apoptosis even when exposed to tamoxifen. Shortly, *MDM2*, as a transcriptional regulator within the nucleus, diminishes *p53*'s ability to activate apoptosis and cell cycle arrest genes, promoting a survival advantage for tamoxifen-resistant cells [31].

Furthermore, enrichment analysis of the KEGG pathway showed that most of the genes are involved in the mechanism of cancer, such as the MAPK pathway, PI3K-Akt pathway, and FoxO pathway (Table 2). Research indicates that targeting *MDM2* can activate the MAPK (TAB1/TAK1/p38) pathway, which reverses drug resistance, including doxorubicin resistance in breast cancer cells. These have proven to enhance apoptosis and cell cycle arrest, counteracting the survival advantage conferred by *MDM2* overexpression [32]. The PI3K-AKT signaling pathway is often activated in tamoxifen-resistant breast cancer, and *MDM2* has been implicated in this pathway. *MDM2* can interact with components of the PI3K-AKT pathway, promoting cellular responses that favor survival and proliferation even in tamoxifen [30].

All those gene interactions were then found using PPI network construction, and the top 10 genes with the most influence on the mechanism were listed based on their degrees. Some hub genes are chosen for genetic alteration analysis depending on their involvement in the KEGG pathway. These reveal *MDM2* with the highest alteration, which is also involved in the PI3K-Akt pathway. Phosphatidylinositol 3-kinase (PI3K) catalyzes the production of phosphoinositides, leading to the activation of Akt/PKB serine-threonine kinase. Activated Akt phosphorylates *MDM2* on serine 166 and 186, facilitating its nuclear translocation [33]. Once inside the nucleus, phosphorylated *MDM2* enhances *p53* degradation through ubiquitin-dependent pathways [34].

The molecular involvement of *MDM2* concerning *p53* within the PI3K-Akt pathway plays a pivotal role in mediating tamoxifen-resistance in breast cancer. Through phosphorylation-induced nuclear entry and enhanced degradation of *p53*, *MDM2* suppresses pro-apoptotic functions necessary for effective chemotherapy response [35]. This emphasizes how *MDM2* interacts with *p53* and it is modulated through phosphorylation events triggered by the PI3K-AKT pathway. This ultimately contributes to the tamoxifen resistance mechanisms in breast cancer cells.

UA has been widely explored and known to have the ability to inhibit various carcinogenesis pathways, including the PI3K-Akt and MAPK signaling pathways [36]. UA treatment inhibits the phosphorylation of EGFR, ERK1/2, p38 MAPK, and JNK, which are the key proteins in the EGFR/MAPK pathway. This inhibition correlates with the observed growth inhibitory effects and suggests that UA acts by disrupting this critical signaling pathway [37]. Treatment with UA in colorectal cancer has been shown to enhance the efficacy of doxorubicin by blocking the Akt signaling pathway while simultaneously activating the Hippo signaling pathway. This dual action reduces tumor growth and improves cancer cell apoptosis rates [38]. Furthermore, the inhibition of the PI3K-Akt pathway by UA is linked to its ability to downregulate the downstream effectors associated with tumor progression, thereby providing a multifaceted approach to combating cancer [9].

Confirmation by molecular docking is aligned with all those studies where UA can target both *MDM2* model structures. Despite those two models representing similar regions of the *MDM2* protein, distinct differences make them unique, and it is a concern to use those two structures for better understanding. The 4HBM model covers residues 6-125 to represent the N-terminus of *MDM2* [39], providing a broader representation of the N-terminal domain. The binding energy of UA (-5.79) is two times higher than the native ligand (-11.72). Therefore, UA still has the potency to inhibit the *p53* binding on *MDM2*, compared to that without UA. On the other hand, the 5ZXF model spans residues 24-110, focusing on the core binding region critical for *p53* interaction, which is essential for *p53* attachment (Figure 4A). Targeting this region will possibly decrease the *p53* attachment to the *MDM2* binding site and retain the *p53* activation. The binding energy of UA (-8.71) with 5ZXF was lower than the native ligand (-6.56), which means UA has more affinity to target the 5ZXF structure of *MDM2* and potentially prevent the degradation of the *p53* protein (Figure 4B). Moreover, using multiple structures ensures that one specific model does not bias docking results. If the ligand consistently binds well across both structures, it strengthens the hypothesis that it has a potency for therapeutic candidates.

Figure 4. Docking Visualization of Both *MDM2* Model Structure Targeted by UA and Native ligand. (A) 4HBM Model Structure which the UA has a Higher Binding Energy. (B) 5ZXF Model Structure, which the UA has a Lower Binding Energy.

This in silico study gives us valuable insights into UA's potential and therapeutic mechanism. However, several limitations in this research, which may not fully capture the complexity of biological systems, must be acknowledged. Therefore, further experimental validation, including in vitro and in vivo studies, to validate the bioinformatics predictions and enhance our understanding of the mechanisms at play. It is crucial to confirm that UA can overcome tamoxifen-resistant breast cancer by targeting *MDM2*.

In summary, this study reveals that *MDM2* has become a potential target for *UA* to reverse the tamoxifen-resistant mechanism in breast cancer patients. This particular gene is mainly involved in the PI3K-Akt pathway, which plays a role in the chemotherapy resistance mechanism. Molecular docking results confirm that UA could target both the N-terminus which focuses on the *p53* binding domain region of the *MDM2* structure. Further research is needed to confirm these findings, especially with morphological and physiological matter conditions.

Author Contribution Statement

YSJ was responsible for the data collection, data analysis, writing original draft, and finalizing the manuscript. DNS was responsible for data collection and data analysis. ANZ was responsible for analysis. DDPP was responsible for conceptualization, supervision, writing review, and editing.

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Ethical Issue

All the analyses were conducted using publicly available de-identified datasets to ensure participant privacy. The use of these datasets adheres to ethical

standards, and proper acknowledgments have been provided to the data sources.

Data Availability

The data supporting this study's findings are available from the corresponding author upon reasonable request.

Declaration of Conflicts of Interest

The authors declare that they have no competing financial or any other conflict of interests that could have appeared to influence the work reported in this paper.

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