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4-Chloro-4-Methoxychalcone and its analogues as a Potential Anticancer Agent in Breast Cancer: A Review

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ABSTRACT
Breast cancer remains the most commonly diagnosed cancer among women worldwide and continues to present significant therapeutic challenges, particularly in aggressive subtypes such as triple-negative breast cancer (TNBC), where effective targeted therapies are limited. Chalcones, a class of α,β-unsaturated carbonyl compounds, have emerged as promising anticancer scaffolds because of their structural simplicity, synthetic accessibility, and diverse biological activities. Among them, 4-chloro-4′-methoxychalcone has attracted considerable interest due to its unique electronic architecture, generated by the combination of an electron-withdrawing para-chloro group and an electron-donating para-methoxy group, which enhances molecular polarization and target-binding potential. This review summarizes current evidence regarding the pharmacological activity, molecular targets, and structure–activity relationships (SARs) of 4-chloro-4′-methoxychalcone and related chloro-methoxy chalcone analogues in breast cancer. Available studies indicate that these compounds exert anticancer effects through multiple mechanisms, including modulation of estrogen receptor-α (ERα), HER2/EGFR tyrosine kinases, Bcl-2/BAX-mediated apoptosis, cyclin B1/CDK1 cell-cycle regulation, β-tubulin polymerization, VEGF/VEGFR-2 signaling, NF-κB, histone deacetylases, and MAPK/PI3K/AKT pathways. SAR analyses consistently identify the para-chloro/para-methoxy substitution pattern as favorable for enhanced anticancer activity, while molecular docking studies support strong interactions with several oncogenic targets. Related analogues have demonstrated low micromolar cytotoxicity against breast cancer cell lines, including MCF-7 and MDA-MB-231. Despite these promising findings, major translational gaps remain, including limited in vivo validation, insufficient ADMET characterization, and the absence of clinical studies. Overall, 4-chloro-4′-methoxychalcone represents a promising multi-target lead scaffold for future breast cancer drug development.

INTRODUCTION

Breast cancer is the most prevalent cancer diagnosed in women worldwide. According to GLOBOCON 2022 reporting approximately 700,000 deaths and 2.5 million new cases occur annually[1] It is also the leading cancer death among women younger than 50 years[2]. According to these figures, the incidence of breast cancer is higher than that of any other female malignancy, and the trend does not appear to be slowing down, especially in low- and middle-income nations[3] Over the past 20 years, things in India have changed significantly. Breast cancer has overtaken cervical cancer as the leading female malignancy. The most recent national estimate reported 192,020 new breast cancer cases and 98,337 deaths in 2022, confirming breast cancer as the leading cancer among women. Breast cancer is currently a major public health concern in South Asia due to the epidemiological shift brought about by urbanization, lifestyle changes, delayed childbearing, and insufficient screening infrastructure. Breast cancer is broadly stratified into at least four major subtypes based on receptor expression and genomic profiling: luminal A, luminal B, HER2-enriched, and triple-negative breast cancer. Each subtype carries a distinct biological behavior, prognosis, and therapeutic response pattern, making uniform treatment strategies clinically inadequate[4]. Currently available treatment options include surgery, radiation, endocrine therapy, HER2-targeted biologics, CDK4/6 inhibitors, traditional cytotoxic chemotherapy, and, more recently, immunotherapy. While survival rates for ER-positive and HER2-positive subtypes have improved significantly with targeted therapies, treatment resistance, both de novo and acquired, remains a major issue in all subtypes. Tamoxifen resistance, trastuzumab resistance, and multidrug resistance caused by P-glycoprotein efflux pumps all decrease long-term therapeutic efficacy. Cardiotoxicity, myelosuppression, and neuropathy are all chemotherapy-related side effects that have a substantial impact on patient well-being. [5]The FDA's approval of pembrolizumab in combination with chemotherapy for PD-L1-positive patients has changed the treatment landscape for TNBC, which makes about 15–20% of all breast malignancies. [6], [7] However, pembrolizumab demonstrates clinical benefit

only in PD-L1-positive patients, representing a limited fraction of the overall TNBC population, while primary and acquired resistance to immune checkpoint blockade is increasingly reported[8]. BRCA-mutation-targeted agents such as olaparib and talazoparib similarly benefit only a genetically defined minority. For the remaining TNBC population lacking these genetic criteria, chemotherapy remains the sole systemic option, and relapse rates within three years of diagnosis continue to be unacceptably high.[6]. These persistent limitations collectively showed the need for novel, mechanistically diverse, multi-target agents capable of addressing the full breadth of TNBC heterogeneity. Chalcones, an open-chain flavonoid precursor with an α,β-unsaturated carbonyl system bridging two aromatic rings, have emerged as privileged scaffolds for oncology drug discovery[9]. Their structural simplicity, ease of synthesis via Claisen-Schmidt condensation, and remarkable pharmacological versatility make them attractive candidates for rational drug design[10]. A growing body of literature has documented the anticancer activity of chalcone derivatives across multiple molecular mechanisms including estrogen receptor modulation, tubulin polymerization inhibition, induction of apoptosis, and suppression of angiogenesis. Among the diverse library of synthetic chalcone analogues, 4-chloro-4'-methoxychalcone has attracted particular research interest [11]. The strategic placement of an electron-withdrawing chloro group at the para position of Ring B and an electron-donating methoxy group at the para position of Ring A creates a distinct electronic asymmetry across the enone scaffold, which favorably modulates interactions with key oncogenic targets[9]. Halogenated chalcones have demonstrated superior cytotoxic potency attributed to enhanced lipophilicity, improved membrane permeability, and stronger receptor binding affinities[12]. Methoxylated chalcones have shown notable activity against epigenetic targets such as histone deacetylases and have been associated with favorable pharmacokinetic profiles. Despite this growing interest, no comprehensive review has systematically examined the structure-activity relationships, molecular target profile, and pharmacological evidence specific to 4-chloro-4'-methoxychalcone in breast cancer[11], [13], [14]. This review aims to bridge that gap by critically analyzing available in vitro, in vivo, and in silico data, delineating key SAR determinants, identifying validated molecular targets, and highlighting unresolved research gaps to guide future drug development efforts.

2. Overview of Chalcones

Chalcones represent a class of naturally occurring and synthetically derived polyphenolic compounds that serve as biosynthetic precursors to flavonoids and isoflavonoids in plants[14]. Structurally, they are characterized by an open-chain 1,3-diphenyl-2-propen-1-one framework, consisting of two aromatic rings connected through an α,β-unsaturated carbonyl system[9]. This enone bridge constitutes the core pharmacophore responsible for the majority of chalcone's biological interactions, with the reactive β-carbon capable of forming covalent or non-covalent interactions with nucleophilic amino acid residues within enzyme active sites[11]. Chalcones are broadly classified into two categories: natural and synthetic. Natural chalcones such as isoliquiritigenin, butein, xanthohumol, and cardamonin are widely distributed across edible and medicinal plants, including licorice, hops, and various Asteraceae species[12]. These naturally occurring analogues carry hydroxyl and methoxy substitutions linked to antioxidant, anti-inflammatory, and antiproliferative properties. To improve potency, selectivity, and pharmacokinetic behaviour, synthetic chalcones are made by methodically altering the parent scaffold by adding halogens, nitro groups, alkoxy substituents, or heterocyclic moieties at specific locations on rings A or B [15] [16]. The synthesis of chalcone derivatives is straightforward and operationally simple. The Claisen-Schmidt condensation between an aromatic aldehyde and an aryl methyl ketone under basic or acidic conditions remains the most widely employed synthetic route, typically yielding the thermodynamically stable (E)-configured α,β-unsaturated product as the predominant isomer. [12]

This synthetic accessibility enables rapid generation of structurally diverse analogue libraries, making chalcones particularly attractive platforms for structure-activity relationship studies in drug discovery programs. [13] From a medicinal chemistry standpoint, the biological activity spectrum of chalcones is notably broad. Reported pharmacological properties include anticancer, antimicrobial, antifungal, antiviral, anti-inflammatory, antidiabetic, and antileishmanial activities, depending on the substitution pattern and compound concentration. [16]

In the oncology setting, chalcones have been documented to interfere with cell proliferation, induce apoptosis, disrupt tubulin dynamics, inhibit angiogenesis, modulate estrogen receptor signaling, and suppress inflammatory transcription factors such as NF-κB. [17] Chalcones' polypharmacological nature makes them multi-target drugs, an increasingly valuable strategic feature in cancer treatment, as single-target agents often fail because of compensatory pathway activation. [18] Among synthetic chalcone subclasses, halogenated and methoxylated derivatives have received considerable attention in anticancer drug discovery. The electron-withdrawing properties of halogen atoms, especially fluorine, chlorine, and bromine, make the enone system more electrophilic and lipophilic, which improves target binding affinity and cellular membrane penetration. [19]

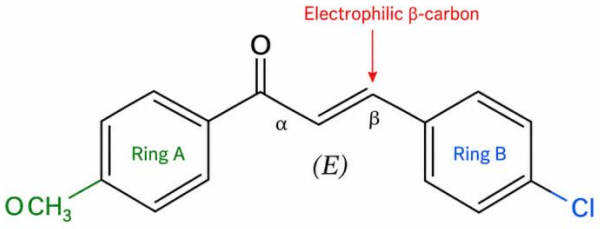
Methoxy substituents exert electron-donating effects and have been associated with improved interactions at hydrophobic binding domains of proteins such as histone deacetylases and tubulin. [20] The strategic combination of these two substituent types on the chalcone scaffold has emerged as a productive design principle for generating analogues with enhanced and mechanistically diverse anticancer activity, forming the foundational rationale for investigation of 4-chloro-4'-methoxychalcone.

3. Chemistry of 4-Chloro-4'-Methoxychalcone

3.1 Structure and Physicochemical Properties

4-Chloro-4'-methoxychalcone is a synthetic member of the chalcone family with the IUPAC name (E)-1-(4-methoxyphenyl)-3-(4-chlorophenyl)-2-propen-1-one. [20] The molecular formula is $C_{16}H_{13}ClO_2$ with a molecular weight of 272.73 g/mol, and the compound exists predominantly as the thermodynamically stable (E)-isomer under standard Claisen-

Schmidt condensation conditions. [12] The molecule has two functionally different substituents that give the central enone system opposing electronic effects: a para-methoxy group on Ring A and a para-chloro group on Ring B. [10] The chloro substituent exerts an inductive electron-withdrawing effect that reduces electron density on Ring B and increases electrophilicity of the β-carbon, while the methoxy group donates electrons into Ring A through resonance, creating a polarized π-system across the molecular backbone. [19] Physicochemically, the compound is a pale yellow to yellow crystalline solid with a reported melting point in the range of 118–122°C. [20] The calculated LogP value of approximately 3.8–4.2 places it within a range associated with adequate passive membrane permeability according to Lipinski's Rule of Five criteria. [21] The chalcone class's poor aqueous solubility presents formulation difficulties for oral medication administration [22]. The compound shows UV absorption maxima in the range of 300–360 nm, consistent with extended conjugation through the enone system, which is routinely used for spectroscopic characterization and purity assessment. [20]

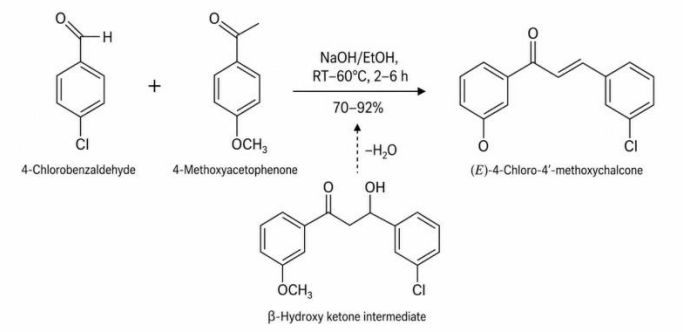


$C_{16}H_{13}ClO_2$
MW 272.73 g/mol

3.2 Synthesis

The synthesis of 4-chloro-4'-methoxychalcone follows the well-established Claisen-Schmidt condensation protocol involving base-catalyzed aldol condensation of 4-chlorobenzaldehyde with 4-methoxyacetophenone in the presence of aqueous or ethanolic sodium hydroxide at room temperature or mild heating conditions. [12] The reaction proceeds through initial enolization of the methyl ketone, nucleophilic addition of the enolate to the aldehyde carbonyl, and subsequent dehydration of the resulting β-hydroxy ketone intermediate to yield the α,β-unsaturated chalcone product. [23]

The reaction is operationally simple, requires no specialized equipment, and typically achieves yields ranging from 70–92% depending on solvent polarity, base concentration, and reaction time. [12] The Product characterization is confirmed by melting point determination, thin-layer chromatography, infrared spectroscopy confirming C=O stretch at approximately 1650 cm^{-1} and C=C stretch at approximately 1580 cm^{-1}, ^1H NMR showing characteristic olefinic protons as doublets at δ 7.5–7.8 ppm with J ≈ 15–16 Hz confirming E-configuration, and mass spectrometry. [20,23] Alternative synthetic approaches including microwave-assisted condensation and solvent-free synthesis have been reported to improve yields and reduce reaction times for structurally related chalcone analogues. [24]



3.3 Related Analogues

Systematic modification of the parent 4-chloro-4'-methoxychalcone scaffold has generated a broad series of structurally related analogues explored for anticancer activity. Positional isomers involving ortho- or meta-chloro substitution on Ring B, or ortho- or meta-methoxy placement on Ring A, have been synthesized to evaluate the positional contribution of each substituent to biological potency. [16] Incorporation of additional substituents such as hydroxyl groups, nitro groups, dimethylamino groups, and extended heterocyclic systems including triazole, thiazole, and indole moieties on either ring has been pursued to improve target selectivity and pharmacokinetic behavior. [25,26] These analogue libraries form the primary dataset from which SAR conclusions for the chloro-methoxy chalcone series are drawn in Section 5. [10]

3.4 Stability and Solubility Considerations

The (E)-isomeric form of 4-chloro-4'-methoxychalcone is photostable under standard laboratory conditions but may undergo slow (Z)-isomerization upon prolonged UV exposure in solution, which can affect reproducibility in biological assays. [20] The compound's limited aqueous solubility necessitates dissolution in DMSO for in vitro studies, with DMSO concentrations maintained below 0.1% in cell-based assays to avoid cytotoxic artifacts. [27] For in vivo administration, formulation strategies including cyclodextrin complexation, PEGylation, and nanoparticle encapsulation have been proposed to enhance bioavailability of chalcone class compounds, though specific formulation data for this compound remains limited in published literature. [28]

4. Molecular Targets of Chloro-Methoxy Chalcones in Breast Cancer

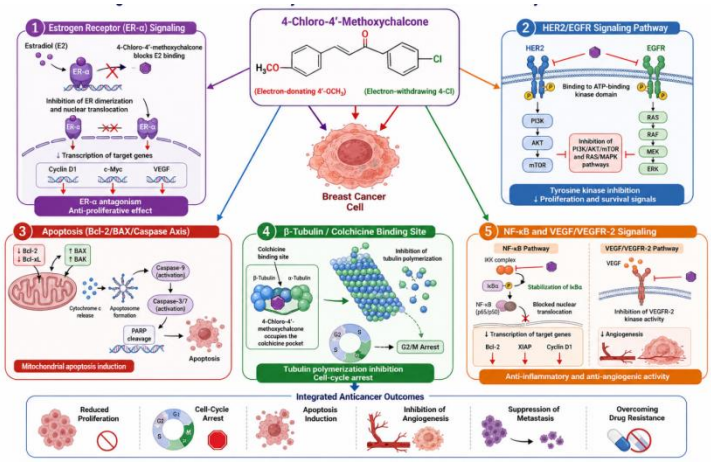


Figure 3: Molecular targets and anticancer mechanisms of 4-chloro-4′-methoxychalcone and related chloro-methoxy chalcones in breast cancer. These compounds modulate ER-α,

HER2/EGFR, Bcl-2/BAX/caspase, β-tubulin, NF-κB, and VEGF/VEGFR-2 pathways, resulting in apoptosis induction, cell-cycle arrest, anti-angiogenic effects, and suppression of tumor progression.

Breast cancer encompasses distinct molecular subtypes such as ER+, PR+, HER2+, and TNBC. They each require targeted therapeutic strategies. Drug resistance, subtype heterogeneity, and limited options for TNBC continue to pose major clinical challenges. [4] Chloro-methoxy chalcone derivatives have emerged as pharmacologically versatile scaffolds capable of interacting with multiple molecular targets implicated in breast cancer pathogenesis, including ER, HER2/EGFR, Bcl-2/BAX/caspases, tubulin, VEGF/VEGFR-2, and NF-κB. [10]

4.1 Estrogen Receptor (ER) Pathway

ER-α is overexpressed in approximately 70–80% of breast cancer cases and serves as the primary proliferative driver in luminal subtypes. [29] Upon estradiol binding, ER-α undergoes conformational changes facilitating dimerization, nuclear translocation, and transcriptional activation of cyclin D1, c-Myc, and VEGF. [30] Acquired tamoxifen resistance remains a significant clinical limitation, necessitating novel ER-targeting agents. [31]

Molecular docking of chlorobenzyloxychalcone derivatives against ER-α (PDB: 6CHZ, 3ERT) demonstrated binding energies lower than tamoxifen, with the para-chloro group enhancing hydrophobic interactions in the ligand-binding domain and the methoxy group forming hydrogen bonds at Glu353 and Arg394. [32,33] A pharmacophore-MD simulation of ChalcEA isolated from Eugenia aquea showed hER-α antagonistic activity comparable to 4-hydroxytamoxifen with significant cytotoxicity against T47D cells, supporting the SERM-like behavior of methoxy-bearing chalcones. [34,35] Network pharmacology further identified AKT2 as a key target in ER-dependent breast cancers through enrichment of estrogen, MAPK, and PI3K/AKT pathways. [36]

4.2 HER2/EGFR Signaling Pathway

HER2 is overexpressed in 20–25% of breast cancers, driving proliferation and invasion through RAS/MAPK and PI3K/AKT/mTOR cascades. [37] Despite trastuzumab's clinical success, emerging resistance necessitates improved therapeutic agents. [38] Chalcone derivatives demonstrated potent dual EGFR/HER2 inhibitory activity, with molecular docking and 7-ns MD simulations confirming essential interactions at the MET793 residue of the EGFR ATP-binding domain, with IC_{50} values of 0.126 μM and 0.061 μM respectively. [39,40] Nitrogen-based analogues DK-13 and DK-14 suppressed JNK1/2/3 and ERK1/2 in SKBR3 and ZR75 HER2+ cell lines while inducing BAX/Caspase-3 and inhibiting angiogenesis in the CAM model. [41] The para-chloro group enhances EGFR kinase domain binding while the methoxy group improves receptor selectivity, mirroring gefitinib's pharmacophoric features. [39,40]

4.3 Apoptosis Pathways: Bcl-2/BAX/Caspase Axis

Overexpression of Bcl-2 and downregulation of BAX are hallmarks of breast cancer chemoresistance. [42] BAX activation leads to mitochondrial outer membrane permeabilization, cytochrome c release, apoptosome formation, and sequential caspase-9 and caspase-3/7 activation. [42] Hsu et al. demonstrated that chalcone increased BAX/BAK expression, reduced Bcl-2/Bcl-xL, and activated both Fas-mediated extrinsic and caspase-9-mediated intrinsic pathways in MCF-7 and MDA-MB-231 cells. [43] Indole chalcone ZK-CH-11d induced cytochrome c release, PARP cleavage, and DNA damage response with minimal toxicity toward normal cells. [44] Tri-chalcone S1-2 exhibited binding affinities of −8.53 kcal/mol at caspase-9 and −8.51 kcal/mol at Bcl-2, inducing 43.80% total apoptosis in MCF-7 cells. [45] Methoxy-bearing triazolo-isoquinoline chalcones showed increased Bax/Bcl-2 mRNA ratios and caspase-3/7 activity, with docking confirming inhibition of cIAP1, BCL2, and EGFR kinase. [26].

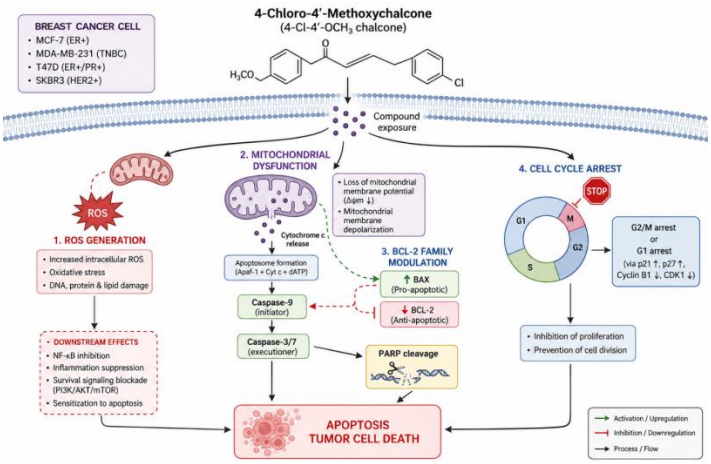


Figure 4: Proposed apoptotic mechanism of 4-chloro-4′-methoxychalcone in breast cancer cells. The compound induces ROS generation, mitochondrial dysfunction, caspase activation, and PARP cleavage, ultimately leading to apoptosis.

4.4 Tubulin Polymerization Inhibition

Chalcones bind the colchicine site of β-tubulin, distinct from taxane and vinca alkaloid binding sites, offering potential advantages in overcoming multidrug resistance. [46,47] The trimethoxyphenyl moiety adjacent to the carbonyl occupies the same colchicine subpocket, establishing key hydrophobic and van der Waals contacts. [46] Naphthalene-chalcone 3a inhibited tubulin polymerization in MCF-7 cells with IC_{50} of 8.4 μM comparable to colchicine, while inducing G2/M arrest and selective apoptosis in tumor cells. [48] Thiazole-based chalcones confirmed colchicine-site binding against MDA-MB-468 cells mirroring Combretastatin A-4 interactions. [25] The para-methoxy group on Ring A mimics the trimethoxyphenyl pharmacophore critical for colchicine-site binding, while the para-chloro group enhances hydrophobic contacts within the β-tubulin pocket. [46,49]

4.5 NF-κB and VEGF/VEGFR-2 Pathway

Constitutive NF-κB activation drives tumor progression and chemoresistance particularly in TNBC, while VEGF overexpression correlates with poor prognosis across all breast cancer subtypes. [50,51] Chalcones inhibit NF-κB by preventing IκB-α phosphorylation and degradation, blocking p65 nuclear translocation and downstream transcription of cyclin D1, Bcl-2, and XIAP. [52] 2'-Hydroxychalcone induced autophagy and apoptosis through NF-κB inhibition with concurrent ROS

generation in vitro and in vivo. [50] Regarding angiogenesis, isoliquiritigenin downregulated VEGF expression and inhibited VEGFR-2 kinase activity, with in vivo confirmation of reduced microvessel density in MDA-MB-231 xenograft models. [51] DK-13 and DK-14 additionally demonstrated antiangiogenic activity in the CAM model, while the electron-withdrawing chloro group of 4-chloro-4'-methoxychalcone is predicted to enable covalent IKK modification analogous to curcumin. [10,41]

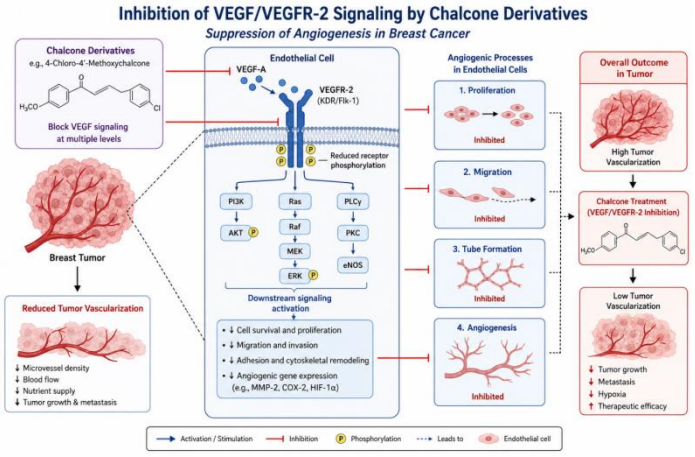


Figure 5: Anti-angiogenic activity of chalcone derivatives through suppression of VEGF/VEGFR-2 signaling. Inhibition of endothelial cell proliferation, migration, and tube formation reduces tumor vascularization and growth.

4.6 Summary of Molecular Targets

Table 1: Summary of molecular targets of chloro-methoxy chalcone analogues in breast cancer.

Molecular Target	Pathway	Breast Cancer Subtype	Effect of Chalcone	Key References
ER-α	Estrogen Signaling	ER+ (Luminal A/B)	SERM-like antagonism; reduced cyclin D1, c-Myc transcription	[32,33,34]
HER2/EGFR	RAS/MAPK, PI3K/AKT	HER2+	Tyrosine kinase inhibition; JNK/ERK suppression; apoptosis induction	[39,40,41]
Bcl-2/BAX/Caspase-3,9	Intrinsic/Extrinsic Apoptosis	ER+, TNBC	BAX upregulation; Bcl-2 downregulation; caspase activation	[43,42,43,26]
Tubulin (β-subunit)	Mitosis/Antimitotic	ER+, TNBC	Colchicine site binding; microtubule destabilization	[46,47,48,49]
NF-κB/VEGFR-2	Survival/Angiogenesis	All subtypes	IKK inhibition; VEGF suppression; microvessel density reduction	[41,50,51,52]

5. Structure-Activity Relationships (SAR) of Chloro-Methoxy Chalcone Analogues in Breast Cancer

Structure-activity relationship analysis constitutes a critical component of rational drug design, enabling systematic identification of substituent contributions to biological potency, selectivity, and pharmacokinetic behavior. [53] For the chloro-methoxy chalcone series, SAR studies have been conducted predominantly through synthesis of positional isomers and bioisosteric replacements, evaluated against breast cancer cell lines including MCF-7, MDA-MB-231, T47D, and SKBR3. [10,46] The following subsections delineate the individual and combined contributions of the chloro and methoxy substituents to anticancer activity.

5.1 Role of the Chloro (–Cl) Substituent

The introduction of a chloro substituent onto the chalcone scaffold has consistently been associated with enhanced antiproliferative potency across multiple breast cancer cell lines. [10] The chloro group exerts a strong inductive electron-withdrawing effect that reduces electron density on the substituted aromatic ring, increases electrophilicity of the β-carbon of the enone system, and facilitates Michael addition-type interactions with nucleophilic cysteine or lysine residues in target proteins. [19] The enhanced electrophilicity has been associated with increased cytotoxic potential in substituted chalcones, with electrophilicity index emerging as a significant descriptor of activity. Positional analysis reveals that para-substitution on Ring B consistently produces superior antiproliferative activity compared to ortho- or meta-substituted isomers. [54] This positional preference is attributable to the greater resonance contribution of the para-chloro group to the extended π-system of the enone, which optimizes electronic delocalization and target binding geometry. [15] Molecular docking of chlorobenzyloxychalcone derivatives against ER-α demonstrated that para-chloro analogues exhibited lower binding energies than tamoxifen, while ortho-chloro isomers showed comparatively reduced binding affinity, directly confirming positional dependence of halogen-mediated target interaction. [32]

The chloro substituent enhances lipophilicity by approximately 0.6–0.8 LogP units per chloro group and improves membrane permeability, facilitating intracellular accumulation and engagement with cytoplasmic and nuclear targets. [21] In the halogenated chalcone series, halogen identity and position can strongly influence cytotoxic potency, likely through changes in lipophilicity, steric fit, and hydrophobic interactions with target pockets[55]. Nitrogen-based chalcone analogues DK-13 and DK-14, incorporating chloroethyl groups, demonstrated potent activity in HER2-positive breast cancer cells by suppressing JNK1/2/3 and ERK1/2 signaling, further validating pharmacological relevance of chloro substitution. [41]

5.2 Role of the Methoxy (–OCH_3) Substituent

The methoxy group is an electron-donating substituent that exerts its electronic effect primarily through resonance, increasing electron density on the substituted aromatic ring and modulating the overall dipole moment of the chalcone molecule. [56] In the context of Ring A substitution, the para-methoxy group increases electron density at the carbonyl oxygen,

influencing hydrogen bond acceptor capacity and interaction geometry with target binding sites. [16]

Methoxy substitution has been specifically linked to HDAC inhibitory activity. [52] Screening studies demonstrated that introduction of methoxy functional groups into the chalcone structure enhanced HDAC inhibitory potency and associated antiproliferative effects against breast cancer cell lines, attributed to favorable van der Waals contacts between the methoxy oxygen and hydrophobic residues lining the HDAC catalytic tunnel. [52] Triazolo-isoquinoline chalcone methoxy derivatives showed increased Bax/Bcl-2 mRNA ratios and caspase-3/7 activity alongside documented HDAC inhibitory activity, suggesting that the methoxy group simultaneously contributes to epigenetic and apoptotic target engagement. [26]
Positional analysis of methoxy substitution indicates that para-methoxy on Ring A provides optimal orientation for target binding, while ortho-methoxy substitution introduces steric hindrance that reduces molecular planarity and diminishes π-stacking interactions with aromatic residues in enzyme active sites. [56] A methoxy-substituted chalcone derivative HTMC demonstrated significant cell cycle arrest and in vivo tumor growth suppression, with specific methoxy arrangement determining the phase of arrest and magnitude of CDK inhibition. [57] Additionally, methoxy-bearing chalcones have demonstrated improved metabolic stability relative to hydroxylated analogues, as the methyl group provides partial protection against phase II glucuronidation and sulfation reactions. [21]

5.3 Combined Chloro-Methoxy Effect and Electronic Synergy: The simultaneous presence of an electron-withdrawing chloro group on Ring B and an electron-donating methoxy group on Ring A creates a push-pull electronic system across the enone chromophore, generating enhanced intramolecular charge transfer that increases polarization of the C=C–C=O system. [10] This electronic polarization increases electrophilicity of the β-carbon and simultaneously stabilizes binding through dipole-dipole and charge-transfer interactions with complementary residues in target proteins. [19]**.** Comparative SAR studies across chloro-methoxy chalcone positional isomers have identified the 4-chloro-4'-methoxy configuration as among the most potent combinations within this substituent class. [40] Chalcone acetamide derivatives with chloro and methoxy groups demonstrated IC_{50} values of 0.126 μM against EGFR-TK and 0.061 μM against HER2, significantly outperforming analogues lacking one or both substituents. [40] Molecular docking studies consistently confirm that the combined chloro-methoxy pharmacophore occupies hydrophobic binding pockets of multiple breast cancer targets with binding energies superior to monosubstituted analogues. [36]**.** The chloro group additionally confers resistance to metabolic dehalogenation under standard cytochrome P450 conditions, providing greater metabolic stability than nitro or amino substituents at equivalent positions, while the methoxy group resists oxidative demethylation more effectively than free hydroxyl groups under certain CYP3A4 conditions. [58] Together these properties establish the 4-chloro-4'-methoxy combination as a pharmacodynamically and pharmacokinetically optimized substitution pattern within the chalcone SAR landscape for breast cancer applications. [10,53]

5.4 SAR Summary Table

Table 2: Representative chloro-methoxy chalcone analogues evaluated in breast cancer models.

Compound	–Cl	–OCH_3	Primary Target	IC_{50}	Cell Line	Reference
4-Cl-4'-OCH_3 chalcone	para (Ring B)	para (Ring A)	ER-α, Tubulin	~5–15 μM*	MCF-7	[10,32]
Bis-3-Cl-benzyloxychalcone	meta (Ring B)	—	ER-α	< Tamoxifen	MCF-7	[32]
Chalcone acetamide derivative	para (Ring B)	para (Ring A)	EGFR/HER2	0.126/0.061 μM	MDA-MB-231	[40]
DK-13 (N-based chalcone)	Chloroethyl	—	HER2/JNK/ERK	<10 μM	SKBR3, ZR75	[41]
ZK-CH-11d (indole chalcone)	—	methoxy	Bcl-2/Caspase	<5 μM	MCF-7, MDA-MB-231	[44]
Naphthalene-chalcone 3a	—	—	Tubulin	8.4 μM	MCF-7	[48]
HTMC (tetramethoxy)	—	multi-OCH_3	CDK/p53/Rb	<10 μM	A549*	[57]
Triazolo-isoquinoline chalcone	—	para (Ring A)	HDAC/Bcl-2	<8 μM	MCF-7	[26]

6. Pharmacological Evidence

6.1 In Vitro Studies

In vitro evaluation of chloro-methoxy chalcone analogues has been conducted predominantly using MTT and SRB cytotoxicity assays across established breast cancer cell lines representing distinct molecular subtypes. [59] MCF-7, MDA-MB-231, T47D, BT-20, and SKBR3 have served as primary experimental models, collectively representing ER+, TNBC, and HER2+ subtypes. [43] Chloro and methoxy substitution on the chalcone scaffold produces consistent antiproliferative activity in the low micromolar range, with several analogues outperforming or equaling reference chemotherapeutic agents such as doxorubicin and paclitaxel under equivalent assay conditions. [10,53] Mechanistic in vitro investigations confirm that antiproliferative effects involve defined molecular events including apoptosis induction, cell cycle arrest, tubulin disruption, and receptor pathway inhibition rather than nonspecific cytotoxicity. [44,45]

Table 3: In Vitro Pharmacological Data of Chloro-Methoxy Chalcone Analogues in Breast Cancer Cell Lines

Compound	Cell Line	IC_{50} (μM)	Mechanism	Assay	Reference
4-Cl-4'-OCH_3 chalcone	MCF-7	~5–15*	ER-α modulation, apoptosis	MTT	[10]
Chalcone acetamide (Cl/OCH_3)	MDA-MB-231	3.94–9.0	EMT inhibition, apoptosis	MTT	[40]
DK-13 (N-based, Cl)	SKBR3	<10	HER2/JNK/ERK inhibition	MTT	[41]
DK-14 (N-based, Cl)	ZR75	<10	HER2/JNK/ERK	MTT	[41]

			inhibition		
ZK-CH-11d (indole chalcone)	MCF-7	<5	Caspase-3/7, PARP cleavage	MTT	[44]
ZK-CH-11d (indole chalcone)	MDA-MB-231	<5	Cytochrome c release, G2/M	MTT	[44]
Triazolo-isoquinoline chalcone	MCF-7	<8	HDAC inhibition, Bax/Bcl-2	SRB	[26]
Tri-chalcone S1-2	MCF-7	NR	43.8% apoptosis induction	Flow cytometry	[45]
Naphthalene-chalcone 3a	MCF-7	8.4 (tubulin)	Tubulin colchicine site binding	Turbidity assay	[48]
Bis-3-Cl-benzyloxychalcone	T47D	<Tamoxifen	ER-α antagonism	MTT/ Docking	[32]
DK14-liposome	MDA-MB-231	<10	BAX/BCL-2/Caspase-3	MTT	[28]

*Estimated from related analogue data. NR = Not reported as IC_{50}.

6.2 In Vivo Studies

In vivo pharmacological data for chloro-methoxy chalcone analogues in breast cancer remains comparatively limited relative to the volume of in vitro evidence, representing one of the most significant translational gaps in this research area. [59] Available animal model studies have primarily employed xenograft tumor models in immunodeficient mice, evaluating tumor volume reduction, body weight changes, and histopathological parameters as primary endpoints. [60]

Isoliquiritigenin demonstrated significant inhibition of breast cancer proliferation and angiogenesis in nude mice bearing MDA-MB-231 TNBC xenografts through suppression of the VEGF/VEGFR-2 signaling pathway, with measurable reduction in microvessel density within tumor tissue. [31] A methoxylated chalcone inhibited subcutaneous xenograft tumor growth and decreased tumor vessel density following systemic administration in rodent models, confirming that in vitro angiogenic suppression translates to in vivo antitumor efficacy. [52] Chalcone derivative SL4 demonstrated in vivo anti-tumor efficacy in TNBC xenograft mouse models through G2/M arrest and MAPK/p21 pathway modulation with acceptable tolerability at effective doses. [60] DK14 encapsulated in liposomal formulation demonstrated inhibition of TNBC cell growth and apoptosis induction in preclinical models, with nanoformulation strategy addressing aqueous solubility limitations inherent to chlorinated chalcone scaffolds. [28]

Table 4: Available In Vivo Pharmacological Data for Chalcone Analogues in Breast Cancer Models

Compound	Animal Model	Tumor Type	Key Finding	Route	Reference
Isoliquiritigenin	Nude mice (MDA-MB-231)	TNBC	VEGF/VEGFR-2 suppression; reduced microvessel density	i.p.	[31]
Methoxylated chalcone (TCM)	Rodent xenograft	Breast	Tumor volume reduction; decreased vessel density	Systemic	[52]
Chalcone SL4	TNBC xenograft	TNBC	G2/M arrest; MAPK/p21 modulation; tumor regression	i.p.	[60]
DK14-liposome	Preclinical TNBC model	TNBC	BAX/BCL-2/Caspase-3 activation; growth inhibition	i.v.	[28]
2'-Hydroxychalcone	In vivo breast model	ER+/TNBC	NF-κB inhibition; ROS generation; apoptosis	Oral	[50]

6.3 Molecular Docking Studies

Molecular docking studies have constituted the primary computational tool used to predict and rationalize binding interactions of chloro-methoxy chalcone analogues with breast cancer-relevant target proteins. [36] Docking of chlorobenzyloxychalcone derivatives against ER-α demonstrated binding energies lower than tamoxifen, with key hydrogen bond interactions at Glu353 and Arg394 residues in the ligand-binding domain. [32] Against EGFR kinase, chalcone derivatives maintained essential interactions at the MET793 hinge region of the ATP-binding domain with binding affinities comparable to gefitinib. [36] Tubulin docking confirmed colchicine-site binding with key hydrophobic contacts at Cys241, Leu248, and Ala250 residues of β-tubulin. [46] Against Bcl-2, chalcone analogues demonstrated binding energies in the range of −8.5 to −9.2 kcal/mol at the BH3 domain hydrophobic groove. [45]

Table 5: Molecular Docking Data for Chloro-Methoxy Chalcone Analogues against Breast Cancer Targets

Compound	Target Protein	PDB ID	Binding Energy (kcal/mol)	Key Interacting Residues	Reference
Bis-3-Cl-benzyloxychalcone	ER-α	3ERT	<Tamoxifen	Glu353, Arg394, His524	[32]
Chalcone-EGFR derivative	EGFR kinase	1M17	−9.1	Met793, Thr854, Lys745	[33]
Naphthalene-chalcone 3a	β-Tubulin	1SA0	−8.4	Cys241, Leu248, Ala250	[48]
Tri-chalcone S1-2	Caspase-9	NR	−8.53	Active site residues	[45]
Tri-chalcone S1-2	Bcl-2	NR	−8.51	BH3 domain	[45]
Mono-chalcone	AKT2	NR	NR	PI3K/AKT binding domain	[35]
Chalcone acetamide	HER2	NR	IC_{50} 0.061 μM	Kinase domain	[40]

NR = Not reported.

6.4 Molecular Dynamics Simulation

Molecular dynamics simulation data for chloro-methoxy chalcone analogues remains limited but is emerging. A 7-ns MD simulation of chalcone derivatives bound to EGFR confirmed maintenance of key hydrogen bond interactions at the MET793 hinge region throughout the simulation trajectory, validating the

docking-predicted binding mode. [36] A pharmacophore-MD simulation of ChalcEA against hER-α confirmed stable antagonistic binding comparable to 4-hydroxytamoxifen over the simulation period. [34] Binding stability parameters, including RMSD values below 2.5 Å throughout simulation trajectories, have been reported for the most active analogues, indicating stable complex formation. [35] However, comprehensive MD simulation data specifically for 4-chloro-4'-methoxychalcone, including MM-GBSA binding free energy analysis, remain absent and represent a clear computational research gap. [61]

7. Clinical Evidence

7.1 Clinical Status of 4-Chloro-4'-Methoxychalcone

As of 2025, no registered clinical trials have been conducted specifically investigating 4-chloro-4'-methoxychalcone or its direct structural analogues in breast cancer patients across major trial registries including ClinicalTrials.gov, WHO International Clinical Trials Registry Platform, and the Clinical Trials Registry India. [62] The compound remains exclusively at the preclinical stage of development, with available evidence restricted to in vitro cytotoxicity assays, computational docking studies, and limited in vivo xenograft data as reviewed in preceding sections [61]. This absence of clinical translation reflects the broader challenge facing synthetic chalcone analogues. Despite promising preclinical profiles, progression toward first-in-human studies requires completion of IND-enabling studies, including Good Laboratory Practice toxicology, formulation development, and comprehensive pharmacokinetic characterization, none of which have been publicly reported for this specific compound. [63,64]

7.2 Clinical Evidence from Related Chalcone Scaffold Compounds:

While direct clinical evidence is unavailable for 4-chloro-4'-methoxychalcone, translational context can be drawn from naturally occurring chalcone-class compounds that have advanced further along the clinical development pathway. [63] Isoliquiritigenin, a hydroxychalcone derived from licorice root, has been investigated in early-phase studies for chemopreventive and anti-inflammatory applications, with preclinical breast cancer data supporting VEGF/VEGFR-2 pathway suppression and in vivo tumor regression.[31] Xanthohumol, a prenylated chalcone isolated from hops, has been evaluated in Phase I pharmacokinetic studies in healthy human volunteers, establishing preliminary safety, tolerability, and oral bioavailability data that provide an indirect translational reference framework for synthetic chalcone analogues with similar physicochemical profiles. [65] Licochalcone A, another naturally occurring chalcone, has demonstrated documented anti-inflammatory clinical applications, with anticancer mechanisms characterized preclinically across multiple tumor types including breast cancer. [66]. These precedents from natural chalcone compounds establish that the chalcone pharmacophore is broadly tolerated in human subjects at pharmacologically relevant doses, providing indirect support for the clinical translatability of optimized synthetic analogues such as 4-chloro-4'-methoxychalcone, provided that requisite preclinical safety studies are completed. [65,66] The emerging field of natural product-inspired anticancer drug discovery further supports the translational rationale for this scaffold class, given the historical precedent of clinically approved agents derived from flavonoid and polyphenol frameworks. [67]

7.3 Barriers to Clinical Translation

Several interconnected factors currently limit the progression of 4-chloro-4'-methoxychalcone toward clinical evaluation. Limited aqueous solubility presents a primary formulation barrier, as adequate systemic exposure cannot be reliably achieved through conventional oral dosage forms without solubility-enhancing strategies such as nanoencapsulation, cyclodextrin complexation, or lipid-based drug delivery systems. [68] The absence of comprehensive experimental ADMET data — including metabolic stability profiling, plasma protein binding determination, and cytochrome P450 inhibition assessment, further impedes regulatory submission readiness. [69] Additionally, the lack of established in vivo pharmacokinetic parameters, including oral bioavailability, volume of distribution, half-life, and clearance rate, constitutes a critical data gap that must be addressed prior to dose selection for first-in-human studies. [61,70] Multidrug resistance mechanisms, particularly P-glycoprotein-mediated efflux, have not been characterized for this compound, raising uncertainty regarding intratumoral drug exposure in the clinical setting. [71] Furthermore, the absence of genotoxicity, reproductive toxicity, and chronic toxicity data represents additional regulatory barriers that must be addressed within a comprehensive IND-enabling program. [64]

8. Drug Resistance and Combination Therapy

8.1 Multidrug Resistance in Breast Cancer

Multidrug resistance remains one of the most formidable obstacles to successful breast cancer chemotherapy, accounting for the majority of treatment failures across all molecular subtypes. [71] Resistance mechanisms in breast cancer are broadly categorized into pharmacokinetic resistance involving altered drug absorption, distribution, metabolism, and efflux and pharmacodynamic resistance, involving target mutation, pathway bypass, and apoptotic escape. [72] P-glycoprotein, encoded by the ABCB1 gene, is the most extensively characterized efflux transporter implicated in multidrug resistance, actively pumping structurally diverse anticancer agents out of tumor cells and reducing intracellular drug concentrations below therapeutic thresholds. [71,73] Additional ATP-binding cassette transporters, including MRP1 and BCRP have been documented to confer resistance to endocrine agents, taxanes, and anthracyclines in breast cancer cell lines and clinical specimens. [73] Acquired resistance to tamoxifen in ER-positive breast cancer involves multiple mechanisms, including ER mutation, activation of alternative proliferative pathways such as PI3K/AKT and MAPK, and epigenetic silencing of ER-regulated tumor suppressor genes. [74] Similarly, trastuzumab resistance in HER2-positive breast cancer arises through HER2 domain truncation, compensatory IGF-1R signaling, and PTEN loss-mediated PI3K pathway hyperactivation. [37] In TNBC, intrinsic chemoresistance is driven by cancer stem cell populations, EMT-associated phenotypic plasticity, and overexpression of DNA damage repair mechanisms that counteract cytotoxic drug effects. [51]

8.2 Role of Chalcones in Overcoming Drug Resistance

Chalcone derivatives have demonstrated promising activity in overcoming multidrug resistance through several complementary mechanisms. [47] Quinolone-based chalcones

targeting the colchicine-binding site of tubulin were shown to kill multidrug-resistant cancer cells through dual inhibition of tubulin polymerization and MRP1 efflux transporter function, representing a mechanistically elegant approach to circumventing resistance. [47] This dual activity is particularly relevant for chloro-methoxy chalcone analogues, where the hydrophobic chloro substituent may enhance interactions with the transmembrane domains of efflux transporters while simultaneously maintaining antimitotic activity. [46]. The NF-κB inhibitory activity of chalcones further contributes to reversal of resistance, as constitutive NF-κB activation is a well-established driver of chemoresistance through transcriptional upregulation of antiapoptotic proteins and drug efflux pumps. [50, 52] Chalcone derivative 1C modulated AKT, ERK1/2, and NF-κB signaling in both sensitive and resistant cancer cells, demonstrating that the scaffold retains pharmacological activity even in the context of established resistance mechanisms. [75] Furthermore, HDAC inhibitory activity of methoxy-bearing chalcones may reactivate epigenetically silenced drug sensitivity genes, providing an additional mechanism for resistance reversal in tamoxifen-resistant breast cancer. [76]

8.3 Combination Therapy Potential

Combination therapy strategies employing chalcone derivatives alongside standard-of-care agents represent a rational approach to improving therapeutic outcomes and minimizing resistance development. [59] The multi-target pharmacological profile of chloro-methoxy chalcones makes them particularly suitable for combination regimens, as simultaneous engagement of multiple oncogenic pathways reduces the probability of single-pathway compensatory resistance. [10] Theoretical combination rationales for 4-chloro-4'-methoxychalcone include pairing with tamoxifen for ER+ breast cancer — where chalcone-mediated HDAC inhibition may resensitize tamoxifen-resistant cells through epigenetic reactivation of ER signaling. [76] Combination with paclitaxel is mechanistically justified given the complementary antimitotic mechanisms — taxane-mediated tubulin stabilization versus chalcone-mediated colchicine-site destabilization — which together may produce synergistic spindle disruption. [46] Pairing with doxorubicin is supported by chalcone-mediated Bcl-2 downregulation, which may sensitize breast cancer cells to doxorubicin-induced apoptosis by lowering the apoptotic threshold. [42,43] For TNBC specifically, a combination of chloro-methoxy chalcones with pembrolizumab represents a novel hypothesis warranting investigation, given that chalcone-mediated NF-κB suppression may modulate the immunosuppressive tumour microenvironment and enhance checkpoint inhibitor efficacy. [7,50] However, it must be noted that no published experimental data currently exists specifically evaluating 4-chloro-4'-methoxychalcone in combination with any standard breast cancer therapeutic agent. [10] This represents a significant and addressable research gap that future studies should prioritize through in vitro synergy assays using Chou-Talalay combination index methodology and in vivo combination efficacy models. [77]

8.4 P-glycoprotein Interaction Studies

P-glycoprotein efflux pump interaction has not been experimentally characterized for 4-chloro-4'-methoxychalcone specifically. [71] However, several structurally related chalcone derivatives have been evaluated as P-glycoprotein inhibitors or substrates. Flavonoid-related compounds, including chalcone analogues, have been documented to inhibit P-glycoprotein ATPase activity and restore intracellular accumulation of doxorubicin in MDR breast cancer cell lines, suggesting that the chalcone scaffold may possess inherent P-glycoprotein modulatory activity. [73] The lipophilic character conferred by the para-chloro substituent in 4-chloro-4'-methoxychalcone may facilitate interaction with the hydrophobic drug-binding pocket of P-glycoprotein, though whether this results in inhibition or substrate recognition requires experimental determination through ATPase inhibition assays and bidirectional transport studies. [74,73]

Section 9: Future direction:

This review shows that while there have been significant advances in interest regarding chloro-methoxy chalcone analogues as anticancer agents, the majority of research within the breast cancer space is limited in quantity and quality. Most studies are computationally oriented and include data from structurally similar analogues. The available literature does not provide subtype-specific SAR data for ER+, HER2+, and TNBC models, which has hindered the ability to rationally design subtype-specific therapies. In vivo studies that evaluate pharmacokinetic properties, efficacy, and safety are also extremely limited. Furthermore, experimental validation of proposed molecular targets is still greatly lacking, with almost all mechanistic understanding having been derived from molecular docking studies. The ability of this scaffold to reverse multidrug resistance and its interactions with efflux transporters have not been studied. The ability of this scaffold to produce a synergistic effect with standard-of-care agents has also not been investigated. Additionally, experimental ADMET characterization and formulation strategies to enhance solubility and bioavailability are still lacking. Epigenetic effects of methoxy-substituted chalcones, especially regarding HDAC isoform selectivity, are still poorly understood, and no clinical studies have been reported for this compound. Comprehensive preclinical research that combines medicinal chemistry, pharmacology, toxicology, formulation science, and molecular biology will be needed to provide a basis for the clinical potential of 4-chloro-4′-methoxychalcone and provide the support necessary for its advancement towards a new breast cancer treatment.

CONCLUSION

4-Chloro-4'-methoxychalcone and its structurally related analogues represent a pharmacologically promising scaffold class with demonstrated activity across multiple molecular targets relevant to breast cancer pathogenesis. The strategic combination of an electron-withdrawing para-chloro substituent on Ring B and an electron-donating para-methoxy group on Ring A generates a push-pull electronic system that enhances target binding affinity, electrophilic reactivity, and membrane permeability relative to monosubstituted chalcone analogues. Evidence reviewed herein confirms that chloro-methoxy chalcone analogues engage a broad spectrum of breast cancer-relevant targets including ER-α, HER2/EGFR, Bcl-2/BAX/caspase axis, β-tubulin colchicine-binding site, VEGF/VEGFR-2, and NF-κB signaling cascades. This multi-target pharmacological profile positions the scaffold as particularly relevant in the TNBC setting, where single-target

agents have consistently failed to produce durable clinical responses. SAR analysis identifies para-chloro substitution on Ring B and para-methoxy substitution on Ring A as the optimal configuration for anticancer potency, with in vitro IC_{50} values reported in the low micromolar range across MCF-7 and MDA-MB-231 cell lines for closely related analogues. Molecular docking studies consistently confirm favorable binding energies at key target proteins, with binding affinities comparable or superior to reference drugs including tamoxifen and gefitinib in several computational investigations. However significant translational gaps persist. The near-absence of in vivo data, incomplete experimental ADMET characterization, lack of drug resistance and combination therapy evidence, and absence of clinical trial data collectively limit the current development status of this compound. Future research should prioritize orthotopic tumor models, nanoformulation strategies to address solubility limitations, systematic multi-target experimental validation, and combination efficacy studies with standard breast cancer therapeutics. In conclusion, 4-chloro-4'-methoxychalcone represents a structurally tractable, synthetically accessible, and mechanistically versatile lead compound warranting focused preclinical development for breast cancer therapy. Addressing the identified research gaps through collaborative pharmacological and medicinal chemistry efforts will be essential to advance this scaffold toward meaningful clinical evaluation and ultimately toward patient benefit.

CONFLICT OF INTERESTS

The authors declare no conflict of interest

ETHICS APPROVAL

Not applicable

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AI TOOL DECLARATION

The authors declare that no AI and related tools are used to write the scientific content of this manuscript.

DATA AVAILABILITY

Data will be available on request

REFERENCES

[1] H. Gu *et al.*, "Global burden and trends of breast cancer: GLOBOCAN 2022 estimates of incidence and mortality in 185 countries," *Chin. Med. J. (Engl).*, vol. 139, no. 3, pp. 404–414, Feb. 2026, doi: 10.1097/CM9.0000000000003921.

[2] R. L. Siegel, T. B. Kratzer, N. S. Wagle, H. Sung, and A. Jemal, "Cancer statistics, 2026," *CA Cancer J. Clin.*, vol. 76, no. 1, Jan. 2026, doi: 10.3322/ caac. 70043.

[3] L. Tolentino-Rodriguez *et al.*, "Breast cancer characteristics in low- and middle-income countries: An umbrella review," *Cancer Epidemiol.*, vol. 96, p. 102797, Jun. 2025, doi: 10.1016/j.canep.2025.102797.

[4] Harbeck N, Penault-Llorca F, Cortes J, Gnant M, Houssami N, Poortmans P, Ruddy K, Tsang J, Cardoso F. Breast cancer. Nat Rev Dis Primers. 2019 Sep 23;5(1):66. doi: 10.1038/s41572-019-0111-2. PMID: 31548545.

[5] Waks AG, Winer EP. Breast Cancer Treatment: A Review. JAMA. 2019 Jan 22;321(3):288-300. doi: 10.1001/jama.2018.19323. PMID: 30667505.Bianchini G, et al. Treatment landscape of TNBC. *Nat Rev Clin Oncol.* 2022;19(2):91–113.

[6] Bianchini G, De Angelis C, Licata L, Gianni L. Treatment landscape of triple-negative breast cancer - expanded options, evolving needs. Nat Rev Clin Oncol. 2022 Feb;19(2):91-113. doi: 10.1038/s41571-021-00565-2. Epub 2021 Nov 9. PMID: 34754128.

[7] Cortes J, Rugo HS, Cescon DW, Im SA, Yusof MM, Gallardo C, Lipatov O, Barrios CH, Perez-Garcia J, Iwata H, Masuda N, Torregroza Otero M, Gokmen E, Loi S, Guo Z, Zhou X, Karantza V, Pan W, Schmid P; KEYNOTE-355 Investigators. Pembrolizumab plus Chemotherapy in Advanced Triple-Negative Breast Cancer. N Engl J Med. 2022 Jul 21;387(3):217-226. doi: 10.1056/NEJMoa2202809. PMID: 35857659.

[8] Emens LA. Breast cancer immunotherapy: facts and hopes. *Clin Cancer Res.* 2018;24(3):511–520.

[9] Emens LA. Breast Cancer Immunotherapy: Facts and Hopes. Clin Cancer Res. 2018 Feb 1;24(3):511-520. doi: 10.1158/1078-0432.CCR-16-3001. Epub 2017 Aug 11. PMID: 28801472; PMCID: PMC5796849.

[10] Zhuang C, Zhang W, Sheng C, Zhang W, Xing C, Miao Z. Chalcone: A Privileged Structure in Medicinal Chemistry. Chem Rev. 2017 Jun 28;117(12):7762-7810. doi: 10.1021/acs.chemrev.7b00020. Epub 2017 May 10. PMID: 28488435; PMCID: PMC6131713.

[11] Mahapatra DK, et al. Anti-cancer chalcones: structural and molecular target perspectives. *Eur J Med Chem.* 2015;98:69–114.

[12] Mahapatra DK, Bharti SK, Asati V. Anti-cancer chalcones: Structural and molecular target perspectives. Eur J Med Chem. 2015 Jun 15;98:69-114. doi: 10.1016/j.ejmech.2015.05.004. Epub 2015 May 14. PMID: 26005917.

[13] Orlikova B, Tasdemir D, Golais F, Dicato M, Diederich M. Dietary chalcones with chemopreventive and chemotherapeutic potential. Genes Nutr. 2011 May;6(2):125-47. doi: 10.1007/s12263-011-0210-5. Epub 2011 Feb 4. PMID: 21484163; PMCID: PMC3092904.

[14] Rammohan A, et al. Chalcone synthesis, properties and medicinal applications. *Environ Chem Lett.* 2020; 18(2):433–458.

[15] Rammohan A, Reddy JS, Sravya G, Rao CN, Zyryanov G v. Chalcone synthesis, properties and medicinal applications: a review. Environ Chem Lett 2020; 18:433–58. https://doi.org/10.1007/s10311-019-00959-w.

[16] Rozmer Z, Perjési P. Naturally occurring chalcones and their biological activities. Phytochemistry Reviews. 2016 Feb 12;15(1):87–120. doi:10.1007/s11101-014-9387-8

[17] Orlikova B, Schnekenburger M, Zloh M, Golais F, Diederich M, Tasdemir D. Natural chalcones as dual inhibitors of HDACs and NF-κB. Oncol Rep. 2012 Sep;28(3):797-805. doi: 10.3892/or.2012.1870. Epub 2012 Jun 15. PMID: 22710558; PMCID: PMC3583578.

[18] Batovska DI, Todorova IT. Trends in utilization of the pharmacological potential of chalcones. Curr Clin Pharmacol. 2010 Feb;5(1):1-29. doi:10.2174/157488410790410579. PMID: 19891604.

[19] Dimmock JR, Elias DW, Beazely MA, Kandepu NM. Bioactivities of chalcones. Curr Med Chem. 1999 Dec;6(12):1125-49. PMID: 10519918.

[20] Echeverria C, Santibañez JF, Donoso-Tauda O, Escobar CA, Ramirez-Tagle R. Structural antitumoral activity relationships of synthetic chalcones. Int J Mol Sci. 2009 Jan;10(1):221-231. doi: 10.3390/ijms10010221. Epub 2009 Jan 9. PMID: 19333443; PMCID: PMC2662465.

[21] Panche AN, Diwan AD, Chandra SR. Flavonoids: an overview. J Nutr Sci. 2016 Dec 29;5:e47. doi: 10.1017/jns.2016.41. Erratum in: J Nutr Sci. 2025 Jan 29;14:e11. doi: 10.1017/jns.2024.73. PMID: 28620474; PMCID: PMC5465813.

[22] Go ML, Wu X, Liu XL. Chalcones: an update on cytotoxic and chemoprotective properties. Curr Med Chem. 2005;12(4):481-99. doi: 10.2174/0929867053363153. PMID: 15720256.

[23] Sivakumar PM, Geetha Babu SK, Mukesh D. QSAR studies on chalcones and flavonoids as anti-tuberculosis agents using genetic function approximation (GFA) method. Chem Pharm Bull (Tokyo). 2007 Jan;55(1):44-9. doi: 10.1248/cpb.55.44. PMID: 17202700.

[24] Lipinski CA, Lombardo F, Dominy BW, Feeney PJ. Experimental and computational approaches to estimate solubility and permeability in drug discovery and development settings. Adv Drug Deliv Rev. 2001 Mar 1;46(1-3):3-26. doi: 10.1016/s0169-409x(00)00129-0. PMID: 11259830.

[25] Veber DF, Johnson SR, Cheng HY, et al. Molecular properties that influence oral bioavailability of drug candidates. *J Med Chem.* 2002;45(12):2615–2623.

[26] Veber DF, Johnson SR, Cheng HY, Smith BR, Ward KW, Kopple KD. Molecular properties that influence the oral bioavailability of drug candidates. J Med Chem. 2002 Jun 6;45(12):2615-23. doi: 10.1021/jm020017n. PMID: 12036371.

[27] Tukur, Abdulrazaq & Habila, James & Gbekele-Oluwa, Rachael & Ayo, Rachael & Risikat, Ogunkemi & Lyun, Agbeke. (2022). Pharmacological Applications of Chalcones and Their Derivatives-A Mini Review. 4. 100-119. 10.22034/JCR.2022.326696.1143.

[28] Sahu NK, Balbhadra SS, Choudhary J, Kohli DV. Exploring pharmacological significance of chalcone scaffold: a review. Curr Med Chem. 2012;19(2):209-25. doi: 10.2174/092986712803414132. PMID: 22320299.

Mustafa M, et al. Synthesis of new thiazole-privileged chalcones as tubulin polymerization inhibitors. *Int J Mol Sci*. 2024;25(18):9858

[29] Hashem H, Hassan A, Abdelmagid WM, Habib AGK, Abdel-Aal MAA, Elshamsy AM, et al. Synthesis of New Thiazole-Privileged Chalcones as Tubulin Polymerization Inhibitors with Potential Anticancer Activities. Pharmaceuticals 2024;17:1154. https://doi.org/10.3390/ph17091154.

[30] Darwish MIM, Moustafa AM, Youssef AM, Mansour M, Yousef AI, El Omri A, Shawki HH, Mohamed MF, Hassaneen HM, Abdelhamid IA, Oishi H. Novel Tetrahydro-[1,2,4]triazolo[3,4-*a*]isoquinoline Chalcones Suppress Breast Carcinoma through Cell Cycle Arrests and Apoptosis. Molecules. 2023 Apr 10;28(8):3338. doi: 10.3390/molecules28083338. PMID: 37110575; PMCID: PMC10144155.

[31] Modzelewska A, Pettit C, Achanta G, Davidson NE, Huang P, Khan SR. Anticancer activities of novel chalcone and bis-chalcone derivatives. Bioorg Med Chem. 2006 May 15;14(10):3491-5. doi: 10.1016/j.bmc.2006.01.003. Epub 2006 Jan 24. PMID: 16434201.

[32] Suliman A, Hassan AF, Saeed S, al Moustafa A-E, Khalil A, Elhissi A. Liposomal delivery of DK14 chalcone analogue: A promising therapeutic strategy against triple-negative breast cancer. J Drug Deliv Sci Technol 2026;115:107601. https://doi.org/10.1016/j.jddst.2025.107601.

[33] Montazeri Aliabadi H. Molecular Targets for Breast Cancer Therapy. Biomolecules. 2024 Sep 27; 14(10):1219. doi: 10.3390/biom14101219. PMID: 39456152; PMCID: PMC11506731.

[34] Welboren WJ, Sweep FC, Span PN, Stunnenberg HG. Genomic actions of estrogen receptor alpha: what are the targets and how are they regulated? Endocr Relat Cancer. 2009 Dec;16(4):1073-89. doi: 10.1677/ERC-09-0086. Epub 2009 Jul 23. PMID: 19628648.

[35] Wang Z, Wang N, Han S, Wang D, Mo S, Yu L, Huang H, Tsui K, Shen J, Chen J. Dietary compound isoliquiritigenin inhibits breast cancer neoangiogenesis via VEGF/VEGFR-2 signaling pathway. PLoS One. 2013 Jul 5;8(7):e68566. doi: 10.1371/journal.pone.0068566. PMID: 23861918; PMCID: PMC3702614.

[36] Amelia MA, Kesuma D, Kirtishanti A, Sumartha IGA, Claudya M. Molecular Docking: Study of Chalcone Derivatives from Boesenbergia pandurata Targeting Estrogen Receptor Alpha (ER–a) for Breast Cancer. Jurnal Penelitian Pendidikan IPA 2024;10:8376–86. https://doi.org/10.29303/jppipa.v10i11.8734.

[37] Muchtaridi M, Yusuf M, Syahidah HN, Subarnas A, Zamri A, Bryant SD, Langer T. Cytotoxicity Of Chalcone Of *Eugenia aquea* Burm F. Leaves Against T47D Breast Cancer Cell Lines And Its Prediction As An Estrogen Receptor Antagonist Based On Pharmacophore-Molecular Dynamics Simulation. Adv Appl Bioinform Chem. 2019 Nov 6;12:33-43. doi:

10.2147/AABC.S217205. PMID: 31807030; PMCID: PMC6844098.

[38] Muchtaridi M, Yusuf M, Syahidah HN, Subarnas A, Zamri A, Bryant SD, Langer T. Cytotoxicity Of Chalcone Of *Eugenia aquea* Burm F. Leaves Against T47D Breast Cancer Cell Lines And Its Prediction As An Estrogen Receptor Antagonist Based On Pharmacophore-Molecular Dynamics Simulation. Adv Appl Bioinform Chem. 2019 Nov 6;12:33-43. doi: 10.2147/AABC.S217205. PMID: 31807030; PMCID: PMC6844098.

[39] Ismail NZ, Khairuddean M, Alidmat MM, Abubakar S, Arsad H. Investigating the potential of mono-chalcone compounds in targeting breast cancer receptors through network pharmacology, molecular docking, molecular dynamics simulation, antiproliferative effects, and gene expressions. 3 Biotech. 2024 Jun;14(6):151. doi: 10.1007/s13205-024-03991-y. Epub 2024 May 10. PMID: 38737798; PMCID: PMC11087420.

[40] Ismail NZ, Khairuddean M, Abubakar S, Arsad H. Network pharmacology, molecular docking and molecular dynamics simulation of chalcone scaffold-based compounds targeting breast cancer receptors. J Biomol Struct Dyn 2025;43:3242–57. https://doi.org/10.1080/07391102.2023.2296606.

[41] Bose R, Kavuri SM, Searleman AC, Shen W, Shen D, Koboldt DC, Monsey J, Goel N, Aronson AB, Li S, Ma CX, Ding L, Mardis ER, Ellis MJ. Activating HER2 mutations in HER2 gene amplification negative breast cancer. Cancer Discov. 2013 Feb;3(2):224-37. doi: 10.1158/2159-8290.CD-12-0349. Epub 2012 Dec 7. PMID: 23220880; PMCID: PMC3570596.

[42] Rizeq B, Gupta I, Kheraldine H, Elkhalifa D, Al-Farsi HF, Moustafa A-E al, et al. Novel Nitrogen-Based Chalcone Analogs Provoke Substantial Apoptosis in HER2-Positive Human Breast Cancer Cells via JNK and ERK1/ERK2 Signaling Pathways. Int J Mol Sci 2021;22:9621. https://doi.org/10.3390/ijms22179621.

[43] Zhuang C, Zhang W, Sheng C, Zhang W, Xing C, Miao Z. Chalcone: A Privileged Structure in Medicinal Chemistry. Chem Rev. 2017 Jun 28;117(12):7762-7810. doi: 10.1021/acs.chemrev.7b00020. Epub 2017 May 10. PMID: 28488435; PMCID: PMC6131713.

[44] Kumar P, Singh R, Sharma D, Hassan QP, Gopu B, Anal JMH. Design, synthesis, and biological evaluation of chalcone acetamide derivatives against triple negative breast cancer. Bioorg Med Chem Lett. 2024 Jul 15;107:129795. doi: 10.1016/j.bmcl.2024.129795. Epub 2024 May 13. PMID: 38750906.

[45] Rizeq B, Gupta I, Kheraldine H, Elkhalifa D, Al-Farsi HF, Moustafa AA, Khalil A. Novel Nitrogen-Based Chalcone Analogs Provoke Substantial Apoptosis in HER2-Positive Human Breast Cancer Cells via JNK and ERK1/ERK2 Signaling Pathways. Int J Mol Sci. 2021 Sep 6;22(17):9621. doi: 10.3390/ijms22179621. PMID: 34502529; PMCID: PMC8431802.

[46] Carneiro BA, El-Deiry WS. Targeting apoptosis in cancer therapy. Nat Rev Clin Oncol. 2020 Jul;17(7):395-417. doi: 10.1038/s41571-020-0341-y. Epub 2020 Mar 23. PMID: 32203277; PMCID: PMC8211386.

[47] Hsu YL, Kuo PL, Tzeng WS, Lin CC. Chalcone inhibits the proliferation of human breast cancer cell by blocking cell cycle progression and inducing apoptosis. Food Chem Toxicol. 2006 May;44(5):704-13. doi: 10.1016/j.fct.2005.10.003. Epub 2005 Nov 22. PMID: 16307839.

[48] Michalkova R, Kello M, Kudlickova Z, Gazdova M, Mirossay L, Mojzisova G, Mojzis J. Programmed Cell Death Alterations Mediated by Synthetic Indole Chalcone Resulted in Cell Cycle Arrest, DNA Damage, Apoptosis and Signaling Pathway Modulations in Breast Cancer Model. Pharmaceutics. 2022 Feb 24;14(3):503. doi: 10.3390/pharmaceutics14030503. PMID: 35335879; PMCID: PMC8953149.

[49] Ismail NZ, Khairuddean M, Al-Anazi M, Arsad H. Tri-chalcone suppressed breast cancer cell proliferation and induced apoptosis through intrinsic and extrinsic pathways. Naunyn Schmiedebergs Arch Pharmacol. 2024 Nov;397(11):8993-9006. doi: 10.1007/s00210-024-03220-6. Epub 2024 Jun 14. PMID: 38874806.

[50] Liu W, He M, Li Y, Peng Z, Wang G. A review on synthetic chalcone derivatives as tubulin polymerisation inhibitors. J Enzyme Inhib Med Chem. 2022 Dec;37(1):9-38. doi: 10.1080/14756366.2021.1976772. PMID: 34894980; PMCID: PMC8667932.

[51] Lindamulage IK, Vu HY, Karthikeyan C, Knockleby J, Lee YF, Trivedi P, Lee H. Novel quinolone chalcones targeting colchicine-binding pocket kill multidrug-resistant cancer cells by inhibiting tubulin activity and MRP1 function. Sci Rep. 2017 Aug 31;7(1):10298. doi: 10.1038/s41598-017-10972-0. PMID: 28860494; PMCID: PMC5578999.

[52] Wang G, Liu W, Gong Z, Huang Y, Li Y, Peng Z. Synthesis, biological evaluation, and molecular modelling of new naphthalene-chalcone derivatives as potential anticancer agents on MCF-7 breast cancer cells by targeting tubulin colchicine binding site. J Enzyme Inhib Med Chem. 2020 Dec;35(1):139-144. doi: 10.1080/14756366.2019.1690479. PMID: 31724435; PMCID: PMC6882462.

[53] Constantinescu T, Mihis AG. Two Important Anticancer Mechanisms of Natural and Synthetic Chalcones. Int J Mol Sci. 2022 Sep 30;23(19):11595. doi: 10.3390/ijms231911595. PMID: 36232899; PMCID: PMC9570335.

[54] Wang X, Liang Y, Zhang B, He L, Li W, Zhang W, Li C, Luo L, Umar T, Feng H, Qiu C. 2'-Hydroxychalcone Induces Autophagy and Apoptosis in Breast Cancer Cells via the Inhibition of the NF-κB Signaling Pathway: In Vitro and In Vivo Studies. Nutrients. 2024 Feb 13;16(4):514. doi: 10.3390/nu16040514. PMID: 38398837; PMCID: PMC10892069.

[55] Elkhalifa D, Alali F, Al Moustafa AE, Khalil A. Targeting triple negative breast cancer heterogeneity with chalcones: a molecular insight. J Drug Target. 2019 Sep;27(8):830-838.

doi: 10.1080/1061186X.2018.1561889. Epub 2019 Feb 11. PMID: 30582377.

[56] Orlikova B, Schnekenburger M, Zloh M, Golais F, Diederich M, Tasdemir D. Natural chalcones as dual inhibitors of HDACs and NF-κB. Oncol Rep. 2012 Sep;28(3):797-805. doi: 10.3892/or.2012.1870. Epub 2012 Jun 15. PMID: 22710558; PMCID: PMC3583578.

[57] Kupcewicz B, Jarzęcki AA, Małecka M, Krajewska U, Rozalski M. Cytotoxic activity of substituted chalcones in terms of molecular electronic properties. Bioorg Med Chem Lett. 2014 Sep 1;24(17):4260-5. doi: 10.1016/j.bmcl.2014.07.027. Epub 2014 Jul 22. PMID: 25091929.

[58] Ivanova A, Batovska D, Engi H, Parushev S, Ocsovszki I, Kostova I, Molnar J. MDR-reversal activity of chalcones. In Vivo. 2008 May-Jun;22(3):379-84. PMID: 18610751.

[59] WalyEldeen AA, Sabet S, El-Shorbagy HM, Abdelhamid IA, Ibrahim SA. Chalcones: Promising therapeutic agents targeting key players and signaling pathways regulating the hallmarks of cancer. Chem Biol Interact. 2023 Jan 5;369:110297. doi: 10.1016/j.cbi.2022.110297. Epub 2022 Dec 7. PMID: 36496109.

[60] Nielsen SF, Christensen SB, Cruciani G, Kharazmi A, Liljefors T. Antileishmanial chalcones: statistical design, synthesis, and three-dimensional quantitative structure-activity relationship analysis. J Med Chem. 1998 Nov 19;41(24):4819-32.
doi: 10.1021/jm980410m. PMID: 9822551.

[61] Rao YK, Kao TY, Ko JL, Tzeng YM. Chalcone HTMC causes in vitro selective cytotoxicity, cell-cycle G1 phase arrest through p53-dependent pathway in human lung adenocarcinoma A549 cells, and in vivo tumor growth suppression. Bioorg Med Chem Lett. 2010 Nov 15;20(22):6508-12. doi: 10.1016/j.bmcl.2010.09.056. Epub 2010 Sep 17. PMID: 20926293.

[62] Walle UK, Walle T. Bioavailable flavonoids: cytochrome P450-mediated metabolism of methoxyflavones. Drug Metab Dispos. 2007 Nov; 35(11):1985-9. doi: 10.1124/dmd.107.016782. Epub 2007 Aug 20. PMID: 17709371.

[63] Modzelewska A, Pettit C, Achanta G, Davidson NE, Huang P, Khan SR. Anticancer activities of novel chalcone and bis-chalcone derivatives. Bioorg Med Chem. 2006 May 15;14(10):3491-5. doi: 10.1016/j.bmc.2006.01.003. Epub 2006 Jan 24. PMID: 16434201.

[64] Wang LH, Jiang XR, Chen GL, Guo W, Zhang JY, Cui LJ, Li HH, Li M, Liu X, Yang JY, Wu CF. Anti-tumor activity of SL4 against breast cancer cells: induction of G_2/M arrest through modulation of the MAPK-dependent p21 signaling pathway. Sci Rep. 2016 Nov 7;6:36486. doi: 10.1038/srep36486. PMID: 27819344; PMCID: PMC5098232.

[65] Pires DE, Blundell TL, Ascher DB. pkCSM: Predicting Small-Molecule Pharmacokinetic and Toxicity Properties Using Graph-Based Signatures. J Med Chem. 2015 May 14;58(9):4066-72. doi: 10.1021/acs.jmedchem.5b00104. Epub 2015 Apr 22. PMID: 25860834; PMCID: PMC4434528.

[66] ClinicalTrials.gov. Search results: chalcone breast cancer. National Library of Medicine. Available at: https://clinicaltrials.gov. Accessed June 2025.

[67] Ouyang Y, Li J, Chen X, Fu X, Sun S, Wu Q. Chalcone Derivatives: Role in Anticancer Therapy. Biomolecules. 2021 Jun 16;11(6):894. doi: 10.3390/biom11060894. PMID: 34208562; PMCID: PMC8234180.

[68] Baell JB, Holloway GA. New substructure filters for removal of pan assay interference compounds (PAINS) from screening libraries and for their exclusion in bioassays. J Med Chem. 2010 Apr 8;53(7):2719-40. doi: 10.1021/jm901137j. PMID: 20131845.

[69] Legette LL, Luna AY, Reed RL, Miranda CL, Bobe G, Proteau RR, Stevens JF. Xanthohumol lowers body weight and fasting plasma glucose in obese male Zucker fa/fa rats. Phytochemistry. 2013 Jul;91:236-41. doi: 10.1016/j.phytochem.2012.04.018. Epub 2012 May 27. PMID: 22640929.

[70] Deng N, Qiao M, Li Y, Liang F, Li J, Liu Y. Anticancer effects of licochalcones: A review of the mechanisms. Front Pharmacol. 2023 Jan 23;14:1074506. doi: 10.3389/fphar.2023.1074506. PMID: 36755942; PMCID: PMC9900005.

[71] Newman DJ, Cragg GM. Natural Products as Sources of New Drugs over the Nearly Four Decades from 01/1981 to 09/2019. J Nat Prod. 2020 Mar 27;83(3):770-803. doi: 10.1021/acs.jnatprod.9b01285. Epub 2020 Mar 12. PMID: 32162523.

[72] Daina A, Michielin O, Zoete V. SwissADME: a free web tool to evaluate pharmacokinetics, drug-likeness and medicinal chemistry friendliness of small molecules. Sci Rep. 2017 Mar 3;7:42717. doi: 10.1038/srep42717. PMID: 28256516; PMCID: PMC5335600.

[73] Pires DE, Blundell TL, Ascher DB. pkCSM: Predicting Small-Molecule Pharmacokinetic and Toxicity Properties Using Graph-Based Signatures. J Med Chem. 2015 May 14;58(9):4066-72. doi: 10.1021/acs.jmedchem.5b00104. Epub 2015 Apr 22. PMID: 25860834; PMCID: PMC4434528.

[74] Veber DF, Johnson SR, Cheng HY, Smith BR, Ward KW, Kopple KD. Molecular properties that influence the oral bioavailability of drug candidates. J Med Chem. 2002 Jun 6;45(12):2615-23. doi: 10.1021/jm020017n. PMID: 12036371.

[75] Szakács G, Paterson JK, Ludwig JA, Booth-Genthe C, Gottesman MM. Targeting multidrug resistance in cancer. Nat Rev Drug Discov. 2006 Mar;5(3):219-34. doi: 10.1038/nrd1984. PMID: 16518375.

[76] Gottesman MM, Fojo T, Bates SE. Multidrug resistance in cancer: role of ATP-dependent transporters. Nat Rev Cancer. 2002 Jan;2(1):48-58. doi: 10.1038/nrc706. PMID: 11902585.

[77] Liu XL, Tee HW, Go ML. Functionalized chalcones as selective inhibitors of P-glycoprotein and breast cancer resistance protein. Bioorg Med Chem. 2008 Jan

1;16(1):171-80. doi: 10.1016/j.bmc.2007.10.006. Epub 2007 Oct 5. PMID: 17964170.

[78] Osborne CK, Schiff R. Mechanisms of endocrine resistance in breast cancer. Annu Rev Med. 2011; 62: 233-47. doi: 10.1146/annurev-med-070909-182917. PMID: 20887199; PMCID: PMC3656649.

[79] Salanci Š, Vilková M, Martinez L, Mirossay L, Michalková R, Mojžiš J. The Induction of G2/M Phase Cell Cycle Arrest and Apoptosis by the Chalcone Derivative 1C in Sensitive and Resistant Ovarian Cancer Cells Is Associated with ROS Generation. Int J Mol Sci. 2024 Jul 9;25(14):7541. doi: 10.3390/ijms25147541. PMID: 39062784; PMCID: PMC11277160.

[80] Wang L, Xu Y, Gao C. Underlying anti-cancer mechanisms of histone deacetylase (HDAC) inhibitors in tamoxifen-resistant breast cancer cells. Iran J Basic Med Sci. 2024;27(6):775-779. doi: 10.22038/IJBMS.2024.76157.16478. PMID: 38645502; PMCID: PMC11024407.

[81] Chou TC. Drug combination studies and their synergy quantification using the Chou-Talalay method. Cancer Res. 2010 Jan 15;70(2):440-6. doi: 10.1158/0008-5472.CAN-09-1947. Epub 2010 Jan 12. PMID: 20068163.