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# Review on flavonoids targeting STAT pathway involved in breast cancer

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**Abstract:**

Breast cancer is a heterogeneous disease classified into several subtypes, including Luminal A, Luminal B, HER2-positive, and basal-like breast cancer. The STAT signaling pathway plays a crucial role in breast cancer progression, proliferation, apoptosis, metastasis, and chemoresistance. Recent studies have highlighted the potential of flavonoids, a class of plant-derived compounds, in targeting the STAT pathway in breast cancer cells.

This review provides an overview of the classification of breast cancer subtypes, the mechanism of action of the STAT signaling pathway, and the advances in the study of STAT3 signaling pathways in breast cancer. We also discuss the anti-cancer properties of flavonoids, their subclassification, and their anti-proliferative activities in cancer. Furthermore, we focus on the specific flavonoids that have been shown to target the STAT pathway in breast cancer cells, including quercetin.

Preclinical studies have demonstrated that flavonoids can inhibit STAT protein activation, dimerization, and nuclear translocation, leading to the suppression of downstream effectors involved in breast cancer progression. Co-administration of flavonoids with paclitaxel has also shown promising results in enhancing anti-cancer efficacy.

This review aims to provide a comprehensive understanding of the potential of flavonoids in targeting the STAT signaling pathway in breast cancer, highlighting their mechanism of action, preclinical studies, and clinical implications.

**Keywords**: Anticancer activity, progression, proliferation, apoptosis, metastasis, chemoresistance.

**Introduction:**

Breast cancer is defined as a malignant tumor that starts in the cells of the breast. The type of breast cancer is determined by which cells in the breast become cancerous. There are numerous locations in the breast where breast cancer can begin. Breasts primarily consist of lobules, ducts, and connective tissue. The milk travels through the ducts, which are tubes, from the breast to the nipple. Connective tissue, composed of fibrous and fatty tissue, holds everything together. Usually, ducts or lobules are places where breast cancer develops. Blood and lymphatic vessels are two ways that breast cancer can spread to other body areas. Metastasis refers to the spread of breast cancer to other bodily regions. Cancer is a disorder wherein some body cells proliferate out of control and spread to other body regions. Cancer can start almost anywhere in any of the billions of cells that make up the human body. With more than 10 million deaths from cancer in the previous year, it is the leading cause of mortality worldwide.[1]

Cancer is a condition in which some cells in the body grow out of control and spread to other parts of the body. Cancer can begin anywhere in the trillions of cells that make up the human body. Various cancers, such as lung, stomach, esophageal, and breast cancer, exist all over the body without our knowledge. These cancers initially disappear due to the body's immune response, but if not treated, the tumor grows and causes damage to the body and develops into cancer. These cancers have the following symptoms: weight fluctuations, such as

unexpected weight loss or increase; yellowing, darkening, or redness of the skin; unhealed wounds, alterations to existing moles; and coughing that will not go away or breathing problems.[2]

Among cancers that are a global issue, breast cancer is the most common cancer in the world. Breast cancer is a frequent condition among women in their forties and fifties. It has a 20% mortality rate and a 30% morbidity rate. Furthermore, the cancer incidence rate is increasing daily due to the development of human physiological instability and current nutritional eating habits. Premature menstruation and delayed menopause, for example, increase the risk of breast cancer. In addition, many side effects often exist even after overcoming cancer. The main side effects of breast cancer are fatigue, loss of appetite, nausea, pain, and weight loss.[2]

Breast cancer is the most common cancer and also the primary cause of mortality due to cancer in females around the World. About 1.38 million new breast cancer cases were diagnosed in 2008, with almost 50% of all breast cancer patients and approximately 60% of deaths occurring in developing countries. There is a huge difference in breast cancer survival rates worldwide, with an estimated 5-year survival of 80% in developed countries to below 40% in developing countries.[3]

Developing countries face resource and infrastructure constraints that challenge the objective of improving breast cancer outcomes by timely recognition, diagnosis, and management. In developed countries like the United States, about 232,340 females will be diagnosed and 39,620 females will be due to breast cancer in 2013. The lifetime risk of developing breast cancer in an American female is 12.38%. The significant decline in mortality due to breast cancer in the United States from 1975 to 2000 is attributed to constant enhancement in both screening mammography and management. According to the World Health Organization (WHO), enhancing breast cancer outcome and survival by early detection remains the foundation of breast cancer regulations. Different modern medicines are prescribed to treat breast cancer. Medical therapy of breast cancer with antiestrogens such as raloxifene or tamoxifen might prevent breast cancer in individuals who are at increased risk of developing it. Surgery of both breasts is an added preventative measure in the increased probability of developing cancer in females. In patients who have been identified with breast tumors, different strategies of management are used, such as targeted therapy, hormonal therapy, radiation therapy, surgery, and chemotherapy. In individuals with distant metastasis, managements are typically aimed at enhancing life quality and survival rate. The unpleasant side effects of breast cancer treatment are one of the most motivating factors to find some alternative methods. The use of herbs for treating patients having breast cancer is considered a natural alternative, because some plants may contain properties that can naturally treat breast cancer.[3]

**Subclassification of luminal-like breast cancer**

**Luminal-A**

The luminal-A is the most common subtype and represents 50%-60% of all breast cancers. These tumors frequently have a low histological grade, low degree of nuclear pleomorphism, and low mitotic activity, and include special histological types (i.e., tubular, invasive cribriform, mucinous, and lobular) with good

prognosis. Luminal A is characterized by higher levels of ER and lower levels of proliferation-related genes. It is characterized by the expression of luminal epithelial cytokeratin's (CK) 8 and 18, other luminal-associated markers including ER1, genes associated with ER function such as LIV1 (zinc transporter ZIP6 or SLC39A6; solute carrier family 39 zinc transporter, member 6), hepatocyte nuclear factor 3 alpha (FOXA1), X-box binding protein 1 (XBP1), GATA binding protein 3 (GATA3), B cell lymphoma 2 (BCL2), erbB3 and erbB4. Luminal-A subtype is defined as ER-positive and/or PR-positive tumors with negative HER2 and low Ki67 (proliferating cell nuclear antigen) index by immunohistochemistry. Patients with luminal-A breast cancer have a good prognosis; the relapse rate is significantly lower than the other subtypes. Recurrence is common in bone, whereas liver, lung, and central nervous system metastases occur in less than 10% of patients, and treatment is mainly based on hormonal therapy.

**Luminal-B**

Luminal B tumors are of higher grade and worse prognosis compared to Luminal A. They are ER-positive and can be PR negative and have a high expression of Ki67 (greater than 20%). They are generally of intermediate/high histologic grade. These tumors may benefit from hormonal therapy along with chemotherapy. The elevated Ki67 makes them grow faster than luminal A and worse prognosis. It constitutes 10–20% of luminal tumors. It has a moderately low expression of estrogen receptors and increased expression of proliferation and cell cycle genes. It represents the group of luminal tumors with the worst prognosis. They benefit from hormone therapy and in a higher percentage from chemotherapy compared to the previous group. Although bone recurrence is frequent, they have a higher rate of visceral recurrence, and survival from diagnosis to relapse is lower. [4]

**Human epidermal receptor 2 (HER2)-positive breast cancer**

Human epidermal receptor 2 (HER2)-positive breast cancer accounts for approximately 15% of all breast cancer diagnoses. However, the HER2-positive subtype is not a uniform entity. A first distinction can be made according to the expression of hormone receptors, which allows one to distinguish between hormone receptor-negative and hormone receptor-positive tumors, representing around one-third and two-thirds of all HER2-positive tumors, respectively. Nonetheless, hormone receptor status does not fully recapitulate the heterogeneity of the disease. By gene expression profiling, four molecular intrinsic subtypes can be identified: HER2-enriched (the most represented subtype, accounting for approximately 47% of HER2-positive tumors), luminal A and B tumors (accounting for approximately 24% and 20% of cases, respectively), and basal-like (around 9%). The distribution of these molecular subtypes is different according to hormone receptor status, with a higher prevalence of HER2-enriched subtype (up to 80%–90%) in tumors without expression of hormone receptors, and a relatively higher representation of luminal intrinsic subtypes (up to 60%–70%) in triple-positive tumors.[5][6]

**Basal-like breast cancer**

In contrast to the controversy regarding the definition of basal-like breast cancers, there is uniform agreement that triple-negative cancers are defined as tumors that lack ER, PR, and HER2 expression. These tumors account for 10–17% of all breast carcinomas,1, depending on the thresholds used to define ER and PR positivity and the methods used for HER2 assessment. Future studies are likely to produce slightly different prevalence rates for triple-negative breast cancers given the change in the definition of HER2 and hormone receptor positivity according to the ASCO/CAP guidelines. Despite these definitional issues, the clinical interest in these tumors stems from the lack of tailored therapies for this group of breast cancer patients and the overlap with the profiles of basal-like cancers. The main characteristics of triple-negative cancers that have emerged from the literature illustrate their similarities to basal-like cancers, including the fact that they more frequently affect younger patients (50 years), are more prevalent in African-American women, often present as interval cancers, and are significantly more aggressive than tumors of other molecular subtypes. This aggressiveness is best exemplified by the fact that the peak risk of recurrence is between the first and third years and the majority of deaths occur in the first 5 years following therapy. Patients with triple-negative cancers, similar to those with basal-like cancers, have a significantly shorter survival following the first metastatic event when compared with those with non-basal-like/non-triple-negative controls.[7]

**Mechanism action of STAT signaling pathway**

The classical IL-6/STAT3 signaling pathway in cancer cells. IL-6 binds to the membrane-bound IL-6 receptor α (IL-6R) and IL-6 receptor β (gp130). The IL-6/IL-6R/gp130 complex activates the phosphorylation of JAKs, followed by STAT3 phosphorylation and activation. Growth factors, such as FGF, IGF, and EGF, can also phosphorylate STAT3 by binding to their cognate membrane receptors. Then, phosphorylated STAT3 forms a homodimer and translocates into the nucleus to bind to the promotor region of target genes and activate target gene transcription

**Advances in the study of STAT3 signaling pathways in breast cancer**

**The role of STAT3 in breast cancer progression**

Advances of the STAT3 signaling pathways involved in breast cancer progression. Interleukins, including IL-6, IL-8, and IL-35, can bind to their receptors and activate the phosphorylation of JAKs and STAT3, OSM can increase IL-6-mediated activation, and IL-17 binding to its receptor leads to inhibition of STAT3 phosphorylation. PTPN2 can inhibit STAT3 phosphorylation by EGF. COX2 and prostaglandin E2 upregulated by HDAC6 can activate STAT3 phosphorylation, and SMYD2 has a similar effect. Additionally, STAT3 and NEAT1 can form a loop to activate the phosphorylation of STAT3, which is inhibited by miR-124. The activated and phosphorylated STAT3 dimers translocate into the nucleus and activate the transcription of target genes involved in breast cancer progression.

**The role of STAT3 in breast cancer proliferation and apoptosis**

Advances of the STAT3 signaling pathways involving breast cancer proliferation and apoptosis. Classical IL-6 /JAK/STAT3 pathways can activate the transcription of cyclin D-1, c-myc, bcl-2, and Bax to promote the

proliferation and inhibit the apoptosis of breast cancer. miR-125a, miR-25-3p, and p16 can promote the binding of IL-6 to its receptors, whereas Wwox has the opposite effect. CCL-18 binding to its receptor can activate the phosphorylation of STAT3, which can be inhibited by IL-32θ. The circuit loop of phosphorylated STAT3, TMEM16A, and EGF leads to continuous activation of STAT3. miR-93-5p, SMYD2, TRIM14 and PKT-M2 induce the activation of STAT3, whereas miR-124 and miR-9 inhibit the activation of STAT3 and breast cancer proliferation. Let-7a-5p, hnRN-A, and phosphorylated STAT3 dimers form a circuit loop to upregulate PKM2 and promote the proliferation and inhibit the apoptosis of breast cancer cells. DPF3 suppressed by phosphorylated STAT3 can promote breast cancer proliferation. Additionally, transcription factor EB (TFEB) can combine with phosphorylated STAT3 dimers to promote the transcription of target genes involved in breast cancer proliferation.

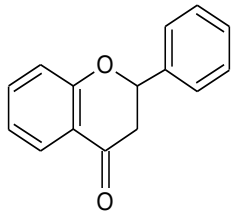
**The role of STAT3 in breast cancer metastasis**

Advances of the STAT3 signaling pathways involving breast cancer metastasis. Classical IL-6/JAK/STAT3 pathways activate the transcription of MMP2, MMP9, Twist, Snail, Slug, and vimentin to promote breast cancer metastasis, which can be suppressed by MEST and activated by GRAMD1B. Wwox can inhibit the binding of IL-6 and IL-6R/gp130. IL-11 and KLF-11 can also activate STAT3 to promote breast cancer metastasis by binding to their receptors. ARHGAP24, MUC1-C, NPRA, and OSM-mediated SMAD3 function to upregulate the phosphorylation of STAT3. Estrogen-related receptor alpha (ERR-α) can be transcriptionally activated by STAT3 and promote breast cancer metastasis. Phosphorylated STAT3 induces the activation of VASP to inhibit the metastasis of breast cancer, whereas PIM1 induced by phosphorylated STAT3 may have the opposite effect. The combination of phosphorylated STAT3 and RhoU inhibits breast cancer metastasis. Additionally, TFEB can activate the phosphorylation of STAT3 and AKT to promote breast cancer metastasis.

**The role of STAT3 in breast cancer chemoresistance**

Advances of the STAT3 signaling pathways involving breast cancer chemoresistance. Classical IL-6/JAK/STAT3 pathways can induce chemoresistance in breast cancer, while miR-4532 may attenuate this effect by inhibiting HIC-1 and IL-6/STAT3 pathways. Leukemia inhibitory factor (LIF) binding to its receptor LIFR can increase the activation of STAT3. STAT3 and pSTAT3 levels are regulated by GRP78. Then, phosphorylated STAT3 activates cellular molecules including FAO, CPT1B, and MAPK/AKT to induce the chemoresistance of breast cancer. Oct-4 and c-Myc form a signaling loop to promote STAT3/NF-κB activation and chemoresistance in breast cancer. Additionally, miR-124 can inhibit HIF 1 and promote breast cancer chemoresistance.[8]

**Flavonoids and Their Anticancer Properties**



Structure of Flavonoids

Flavonoids are bioactive compounds belonging to an important class of low molecular weight plant secondary metabolites having a polyphenolic structure. Flavonoids are abundant in fruits, vegetables, herbs, beverages, spices, and oils. Hence, they are also known as dietary flavonoids. Following terpenoids (30,000) and alkaloids (12,000), the third-largest group of natural products is represented by flavonoids, comprising nearly 10,000 compounds. All flavonoids contain 15 carbon atoms in their basic skeleton, which is composed of two six-membered rings and one three-carbon unit linked to them as C6-C3-C6. The 3-carbon unit bridging the phenyl groups usually cyclizes with oxygen to form a third ring. This core structure is called 2-phenylbenzopyranone. Flavonoids are most often associated with sugar in the conjugated form, O-glycosides or C-glycosides. They can also exist as aglycones. The glycosides are normally attached to position 3 or 7, with the most common carbohydrates occupying those positions being D-glucose, L-rhamnose, glucorhamnose, galactose, or arabinose. The other factors about the varied chemical nature of the flavonoids include patterns of hydroxylation, conjugation between aromatic rings, methoxy groups, and other substituents such as sulfates and prenyl groups. Flavonoids have been known to exhibit a broad spectrum of pharmacological and biochemical reactions associated with health-promoting effects.[9]

**Overview of flavonoids and their subclasses**

Flavonoids are mainly found in plant cell vacuoles in the form of C-glycosides or O-glycosides. The basic molecular structure of flavonoids depends upon their basic C6–C3–C6 skeleton. Flavonoids are classified into seven subclasses based on modifications to their basic skeletons; these subclasses include flavones, flavanones, isoflavones, flavonols, chalcones, flavonols, and anthocyanins.[10]

| Classification of flavonoids | Example |
|---|---|
| Flavanol | Catechin<br>Epicatechin<br>Gallocatechin |
| Flavanone | Naringenin<br>Hesperin<br>Naringenin<br>Prunin |
| Flavonol | Quercetin<br>Rutin<br>Myricetin |
| Isoflavone | Formononetin<br>Genistein<br>Biochanin A |
| Flavone | Apigenin<br>Luteolin<br>Baicalein<br>Diometin |

**Table No. 1: Classification of flavonoids**

**Table No.2: A bird-eye view of various anti-proliferative activities of flavonoids in cancer**

| Sr No | Flavonoids | Type of cancer | Cell model | Mechanistic insight |
|---|---|---|---|---|
| 1. | Quercetin | Breast cancer | MCF-7 | ↓Bcl-2, ↓ RIPK1, ↓ RIPK3 |
| 2. | Luteolin | Breast Cancer | MCF7-TamR | ↓PI3K/AKT/mTOR |
| 3. | Silibinin | Breast cancer | MCF-7 | ↓ERα, ↑ERβ, ↑apoptosis |
| 4. | Cyanidin-3-glucoside | Breast cancer | MDA-MB-453 | ↑caspase-3, ↑DNA fragmentation, ↓Bcl-2, ↑ Bax |

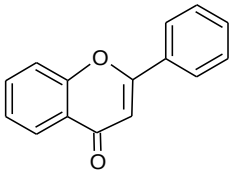
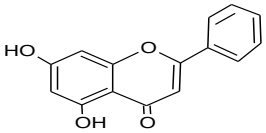
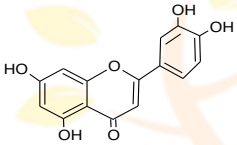
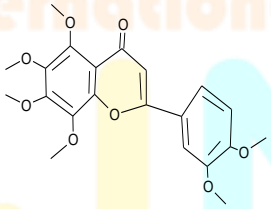
| Anticancer Potential of Flavonoids: Biological Activities |
|---|
| Inhibition of cell growth |
| Inhibition of tumor cell invasion |
| Induction of apoptosis |
| Antiangiogenic properties |
| Inhibition of the STAT signaling pathway |

**Table no.3: Anticancer Potential of flavonoids in biological activities**

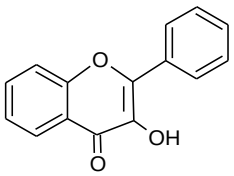
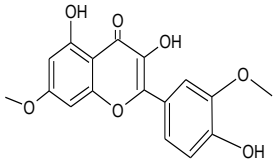
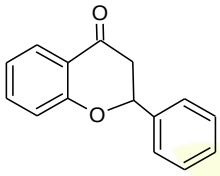
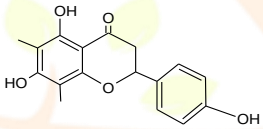
**Table No. 4: Antiangiogenic molecular mechanisms of different flavonoids.[12]**

**Mechanisms of flavonoid-induced anti-cancer effects, including modulation of signaling pathways**

Female breast cancer accounts for 11.7% of all cases and 6.9% of cancer deaths globally. There are several studies on kaempferol's effects and possible mechanisms on breast cancer. In 2004, Hung published an article that showed the significant efficacy of kaempferol on the impairment of the estrogen receptor-α and blocking the estradiol-induced cell proliferation, while estrogen receptor (ER)-negative breast cancer cells didn't show growth resistance toward the kaempferol. In some in vitro and in vivo studies, kaempferol is a phytoestrogen that inhibits triclosan and exhibits estrogen's effects on the growth of breast cancer cells. Cyclin D1, cyclin E, and cathepsin D expressions were upregulated in cells treated with triclosan, whereas p21 and Bax expressions

| Type | Compounds | Mechanism |
|---|---|---|
| Flavone  | Chrysin  | Regulating PI3K/Akt signaling; downregulating JAK1/STAT3 pathway and VEGF/VEGFR2 expression |
| | Luteolin  | Downregulating AEG-1, MMP-2, MMP-9, HIF-1α, and STAT3; stimulating immune response; inhibiting AKT/ERK/mTOR/ P70S6K/MMPspathway or PI3K/Akt/mTOR pathway; elevating JNK phosphorylation; inhibiting NF-κB-DNA binding activity; modulating IL-6/STAT3 pathway |
| | Nobiletin  | Inhibiting VEGF-and bFGF-induced signaling; activating caspase pathway; inhibiting Akt phosphorylation; mediating Src/FAK/STAT3signaling |

were downregulated, and these gene expressions were blocked by kaempferol. This compound also inhibited 17β-estradiol and triclosan-induced expression of pIRS 1, pAkt, and pMEK1/2 [44]. Similar results to in vitro studies on tumor growth inhibition were observed in the in vivo xenografted mouse model. In addition to ER agonist, this phytoconstituent was known as AHR antagonist (aryl hydrocarbon receptor), which was related to the inhibition activity on transcription of AHR in ER-α negative breast cancer cell lines independent of ER-α expression. Generally, the biphasic responses (via ER-dependent and ER-independent pathways) of kaempferol on. The other factors influencing the kaempferol mechanism of action in breast cancer are its pharmaceutical formulation and particle size, considered in two previous studies. The gold nanoparticles

| | | |
|---|---|---|
| Flavonol  | Rhamnazin  | Regulating VEGF and PEDF; downregulating the VEGFR2/STAT3/MAPK/Ak pathway |
| Flavanones  | Farrerol  | Downregulating Akt/mTOR, ErkandJak2/Stat3signalings |

(AuNPs) and nanostructured lipid carriers (NLCs) of kaempferol are suggested as powerful drug delivery techniques in the management of breast cancer cells through the antioxidant, anti-proliferative, and antiangiogenic properties.[13]

**Flavonoids Targeting STAT Pathway in Breast Cancer**

In interconnected networks in a cell, the JAK/STAT (Janus kinase/signal transducers and activators of transcription) signaling pathway detects stimulus signals from outside the cell transmits its message to the cell nucleus, and activates several transcription factors. The JAK/STAT pathway is involved in regulating processes such as immune maintenance, cell division and growth, cell death, and tumor formation. JAK/STAT pathway components are regulated by other paths such as ERK MAPK and PI3K. Skin, immune system, and cancer disorders are caused by JAK/STAT pathway disruptions. In line with this, Qin et al. examined the effect of quercetin on leptin and its receptor in MGC-803 cells via the JAK/STAT signaling pathway. They found that quercetin caused the arrest of cells in the G2/M stage of the cell cycle through the p-STAT3 pathway and caused apoptosis and necrosis. On the other hand, the flavonoid compound reduces the expression of leptin and its receptor. It has been shown that quercetin can inhibit IL-6-induced glioblastoma cell growth and migration by regulating the STAT3 signaling pathway, which affects the expression of this protein in glioblastoma cells. In T98Gand U-87 cell lines, quercetin blocked the IL-6-induced STAT3 pathway, reducing the expression of GP130, JAK1, and STAT3. As a result, there is a reduction in the proliferation and migration of cancer cells.

The effects of quercetin and epigalactochin-3-gallate (EGCG) on cholangiocarcinoma cells were investigated by Senggunprai et al. IL-6 and IFN-gamma were found to regulate JAK/STAT (STAT1/3 phosphorylation) pathways in cholangiocarcinoma cells, and the results suggested that these two chemicals can be employed as chemopreventive agents against these cells. Lastly, these compounds were able to suppress KKU100 cancer cell proliferation and migration. Quercetin suppresses clonogenic survival in BT-474 cells, triggers apoptosis via caspase 3, 8, and PARP cleavage, and causes cell cycle arrest in the sub-G0/G1 phase, according to a study by Seo et al., which also found that quercetin reduces p-JAK1 and p-STAT3 expression and inhibits MMP-9 secretion. When it comes to HER-2-ex pressing breast cancer, this flavonoid can both prevent and treat the disease. Luo et al. examined the processes of apoptosis, autophagy, and quercetin proliferation in cervical cancer cells. Using quercetin-conjugated gold nanoparticles, they found that the complex shows a similar role in suppressing JAK2, a protein expressed in cervical cancer cells, which suppresses proliferation, invasion, and migration processes. They also found that apoptosis and autophagy processes occurred through caspase-3 in cancer cells inhibited by JAK2, and that cyclin D1 and mTOR were suppressed by the STAT3/5 and PI3K/AKT signaling pathways.[14]

**Specific flavonoids that have been shown to target the STAT pathway in breast cancer cells**

| Signaling pathways | Subfamily involved in the signaling pathway | Cancer types | Quercetin IC50 | Target genes | Cell line (s)/in vitro model | Possible mechanisms |
|---|---|---|---|---|---|---|
| JAK/STAT (family) | JAK1/STAT3 | Breast cancer | 0–100 µM | HER-2, MMP-9 | BT474 | ↓ growth and ↓clono genic↑apoptosis ↑STAT3 |

**Table No. 5: The role of quercetin in various cancers mediated by signalling pathways—evidence from preclinical studies**

| flavonoid (cancer type) | Study design | Flavonoid dosage | Paclitaxel Dosage | Duration of study | Mechanism of action |
|---|---|---|---|---|---|
| Tangeretin (Breast cancer) | in vitro: MX-1 and taxol-resistant MX-1/T cells; MDR1–MDCKII cells for modeling epithelial cells | in vitro: 2 folds of IC50 (IC50: 25.3 µM) | In vitro: 75 µM | In vitro: 4 h | toxicity and decrease the cell viability. Inhibitory effects on P-glycoprotein |
| Sciadopitysin (Breast cancer) | In vitro: MX-1 and taxol-resistant MX-1/T cells; MDR1–MDCKII cells for modeling epithelial cells | In vitro:: 2 folds of IC50 (IC50: 106.8 µM) | In vitro: 75 µM | In vitro: 4 h | Increase Taxol cytotoxicity and decrease the cell viability. Inhibitory effects on P-glycoprotein |
| Silibinin (Breast cancer) | In vitro: MCF-7 | In vitro: 1-400 µM | In vitro: 1-200 nM | In vitro: 24 h | Decreasing in anti-apoptotic Bcl-2 level Increasing in pro-apoptotic Bax, P53, BRCA1, and ATM mRNA levels |

**Table No.6: Flavonoids and paclitaxel Co-administration results [15]**

**Mechanisms of action, including inhibition of STAT protein activation, dimerization, and nuclear translocation**

Upon activation of cytokine receptors or growth-factor receptors, STATs are recruited via their SH2 domains and phosphorylated on a tyrosine residue adjacent to the SH2 domain by receptor-associated tyrosine kinases or the intrinsic kinase activity of growth-factor receptors. STATs can also be phosphorylated by constitutively active nonreceptor tyrosine kinases (e.g., v-Src). Tyrosine-phosphorylated STATs form homo- or heterodimers via reciprocal phosphotyrosine (pTyr) SH2 interactions and translocate to the nucleus, where they bind to their respective DNA-binding motifs within the promoter elements of target genes and induce transcription. Since the SH2 domain is required for both tyrosine-phosphorylation and dimerization of STATs, the most logical approach toward inhibition of any STAT, including STAT3, would impair the function of its SH2 domain. This should not only inhibit STAT3 activation but also prevent dimerization of any STAT3 molecules that escape inhibition of activation (Figure 1). Direct inhibition of STAT3 itself is also less likely to result in unintentional inhibition of additional signaling pathways than the targeting of upstream molecules. While the principal feasibility to inhibit STAT3 in cells by a ligand for its SH2 domain has been demonstrated with fusion peptides

carrying hydrophobic or basic peptide sequences to achieve cell permeability and peptide-derived molecule, then a tour of the reagents required their use at relatively high concentrations in tissue culture, and their conversion to nonpeptidic molecules is likely to be difficult.[16]

**Upstream regulators and downstream effectors of the STAT pathway in breast cancer**

STAT3 has received particular attention among the seven members of the STAT family since it is considered an antidote target for the treatment of human tumors. It is constitutively activated by aberrant upstream tyrosine kinase activities in a broad spectrum of cancer cell lines and human tumors. Inhibition of STAT3 signaling by a dominant-negative mutant, anti-sense approaches, decoy oligonucleotides, siRNAs, or G-quartet oligonucleotides has been demonstrated to suppress tumor growth and to induce apoptosis in cancer cells. Numerous small molecules have been reported to inhibit STAT3 signaling; the vast majority of them act on targets other than STAT3.

STAT3 signaling inhibitors with unknown targets include members of the cucurbitacin family of natural products. A recent study suggests that the JAK/STAT3 pathway inhibitor cucurbita cin I also affected the actin cytoskeleton in nontumor cells. Curcumin, another indirect natural product in the inhibition of STAT3 signaling, has also been identified as an inhibitor of numerous additional signaling pathways. Similarly, magnolol was shown to inhibit signaling via STAT3 and other pathways. In addition to these inhibitors with unknown targets, several natural products have been shown to inhibit kinases up to STAT3.

They include Indirubin, a constituent of a Chinese herbal prescription used for the treatment of chronic myelogenous leukemia, and a known inhibitor of a cyclin-dependent kinase, which was shown to inhibit STAT3 signaling in breast cancer cells by inhibiting upstream kinase activity, presumably that of c-Src. A similar mechanism of action was suggested for the at-ura product Resveratrol. Flavopiridol, an acytotoxic compound with several identified targets, was shown to inhibit STAT3 signaling, presumably by intercalation into DNA. Among the small molecules that are thought to bind to STAT3 directly, galiellalactone, a natural product with weak activity against the de Novo synthesis of a-amylases, proteases, and tases in embryoless halves of wheat seeds, was reported to inhibit interleukin-6 (IL-6)-mediated STAT3 signaling. The compound was assumed to bind to the DNA-binding domain of dimeric STAT3, possibly by covalently modifying a cysteine residue in the STAT3 DNA-binding domain. A similar mechanism of action was discussed for a platinum complex that was reported to bind to STAT3 and inhibit STAT3 signaling. Other platinum complexes were also reported to inhibit STAT3 signaling.

**Preclinical and Clinical Evidence**

**Overview of preclinical studies demonstrating the anti-cancer effects of flavonoids in breast cancer models**

| Flavonoid | Type of cancer | Inhibits or decrease the expression of metastatic protein molecules |
|---|---|---|
| Quercetin | Breast | VEGF, MMP9, Nf-κB |
| Cardamonin | Breast | GSK3β, VEGF |
| Baicalein | Breast | GSK3β |
| Luteolin | Breast | MMP9/2,β-catenin |
| Chalcone | Colon | STAT3 |

**Table No.7: Shows different flavonoids downregulating/inhibiting the expression of metastatic protein molecules in various cancers**.

| Flavonoid | Cancer Type | Participant Count | FDA certification Status | Trial Stage |
|---|---|---|---|---|
| Hesperidin | Breast Cancer | 40 | N/A | Finished |
| Catechins | Breast Cancer | 1075 | PhaseII | Finished |
| | | | | |

**Table No.8: Flavonoids in anticancer clinical studies.**[17]

**Conclusion:**

Flavonoids, a class of plant-derived compounds, have shown promising anti-cancer properties, particularly in targeting the STAT signaling pathway involved in breast cancer. This review highlights the potential of flavonoids in inhibiting STAT signaling, leading to suppressed breast cancer cell growth, proliferation, and survival.

Flavonoids inhibit STAT activation: Flavonoids, such as quercetin, kaempferol, and apigenin, have been shown to inhibit STAT activation, thereby suppressing the expression of downstream target genes involved in breast cancer progression. Anti-proliferative and pro-apoptotic effects: Flavonoids have been found to exhibit anti-proliferative and pro-apoptotic effects in breast cancer cells, leading to reduced tumor growth and increased cancer cell death. Synergistic effects with conventional therapies: Flavonoids have been shown to

enhance the efficacy of conventional breast cancer therapies, such as chemotherapy and hormone therapy, by targeting complementary signaling pathways.

Flavonoids have shown promising anti-cancer properties by targeting the STAT signaling pathway involved in breast cancer. Further research is needed to fully elucidate the therapeutic potential of flavonoids in breast cancer treatment. However, the existing evidence suggests that flavonoids may be a valuable adjunct to conventional therapies, offering a novel and targeted approach to breast cancer treatment.

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