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Review article

Recent advancement in developing small molecular inhibitors targeting key kinase pathways against triple-negative breast cancer

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ABSTRACT

Triple-negative breast cancer (TNBC) stands out as the most formidable variant of breast cancer, predominantly affecting younger women and characterized by a bleak outlook and a high likelihood of spreading. The absence of safe and effective targeted treatments leaves standard cytotoxic chemotherapy as the primary option. The role of protein kinases, frequently altered in many cancers, is significant in the advancement and drug resistance of TNBC, making them a logical target for creating new, potent therapies against TNBC. Recently, an array of promising small molecules aimed at various kinases have been developed specifically for TNBC, with combination studies showing a synergistic improvement in combatting this condition. This review underscores the effectiveness of small molecule kinase inhibitors in battling the most lethal form of breast cancer and sheds light on prospective pathways for crafting novel treatments.

1. Introduction

Triple-negative breast cancer (TNBC), a particularly lethal form of breast cancer, predominantly affects younger women with a poor prognosis and the rate of metastasis is also high.[1] Due to the absence of specific, effective targeted therapies, cytotoxic chemotherapy remains the primary treatment, despite common occurrences of chemoresistance.[2,3] The ineffectiveness of endocrine or HER2-targeted therapies in TNBC patients stems from the lack or diminished presence of estrogen receptor (ER), progesterone receptor (PR), and human epidermal growth factor receptor 2 (HER2).[2]

Protein kinases (PKs) are a group of enzymes responsible for catalyzing a reaction known as protein phosphorylation.[4] Kinases are crucial for signaling pathways that regulate cellular processes such as cell growth, replication, metabolism, and regulation of apoptosis. Aberrant activation of PKs led to many diseases, particularly cancer.[5–7] The mitogen-activated protein (MAPK) kinase pathways are crucial in the oncogenesis of various cancers, including TNBC, with elevated MAPK expressions in TNBC tissues being linked to poorer prognoses and reduced survival rates. A significant elevated expression of MAPKs was identified in TNBC tissues compared to the para cancerous tissues by Jiang et al. [8] Wang et al reported that NNMT promoted metastasis of TNBC by targeting PP2A/MEK/ERK/c-Jun/ABCA1 pathway.[9] Additionally, MAPK4 has been identified as a promoter of TNBC cell

Abbreviations: 4E-BP1, eukaryotic translation initiation factor 4E binding protein 1; AKT, protein kinase B; BAD, Bcl2 associated agonist of cell death; Bax, Bcl-2-associated X protein; Bcl-2, B-cell lymphoma 2; CCNB1, cyclin B1; CDKs, cyclin dependent kinases; DYRK1, dual-specificity tyrosine phosphorylation-regulated kinase; eEF-2K, eukaryotic elongation factor 2 kinase; EGFR, epidermal growth factor receptor; EMT, epithelial-mesenchymal transition; ERK, extracellular signal-regulated kinase; FAK, Focal adhesion kinase; FGFR, fibroblast growth factor receptor; IC50, half-maximal inhibitory concentration chain 3B; JNKs, c-Jun *N*-terminal kinases; LC3, microtubule-associated proteins 1A/1B light; MAPK, mitogen-activated protein kinase; MEK, MAP kinase kinase; MMP-2, matrix metalloproteinase-2; MMP-9, matrix metalloproteinase-9; MPS1, monopolar spindle 1; mTOR, mammalian target of rapamycin; NF-kB, nuclear factor k-light-chain-enhancer of activated B cells; PAK4, p21 activated kinase 4; PARP, Poly (ADP ribose) polymerase; PI3K, phosphatidylinositol 3-kinases; PIK3CA, phosphatidylinositol-4,5-bisphosphate 3-kinase catalytic subunit alpha; PIM, Proviral Integration site for Moloney murine leukemia virus kinase; PTEN, Phosphatase and tensin homologue; RB, the retinoblastoma protein; ROS, reactive oxygen species; SRc, Proto-oncogene tyrosine-protein kinase; STAT3, Signal transducer and activator of transcription 3; TNBC, Triple-negative breast cancer; TNK2, Tyrosine kinase non receptor2; VEGF, vascular endothelial growth factor XBP1, X-box binding protein 1; XIAP, X-linked inhibitor of apoptosis protein.

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proliferation, concurrently diminishing tumor responsiveness to phosphatidylinositol 3-kinases (PI3K) inhibition.[10] PI3K/Akt/mTOR signaling pathway has been found to be aberrantly activated nearly all kinds of cancers[11] and their overactivation also common in TNBC.[12] Mutations in PIK3CA, protein kinase B1 (Akt1), and Phosphatase and tensin homologue (PTEN) or PTEN loss are commonly observed in TNBCs, leading to the aberrant activation of the PI3K/Akt/mTOR signaling pathway. This activation enhances cell growth, cancer advancement, and resistance to chemotherapy by suppressing programmed cell death.[13,14]

Cyclin-dependent kinases (CDKs) are a group of serine/threonine protein kinase, regulated by subunits known as cyclins, play a crucial role in controlling cell cycle progression and transcriptional events that drive cell proliferation. Due to the fact that uncontrolled cell growth is a defining feature of cancer, targeting CDKs has also emerged as a promising approach in the creation of novel anti-cancer medications. Specifically, CDK4/6, CDK7, CDK12/13 and CDK16 have been identified as potential therapeutic targets in the development drugs for treating TNBC.[15] Additionally, elevated expression of PIM1 confirmed in TNBCs was associated with apoptosis inhibition via Bcl-2, thereby driving the progression of TNBC.[16,17] On the contrary, tyrosine kinases have demonstrated a link to the progression of TNBC, with high levels of expression observed in TNBC cases.[18–20] Therefore, targeting kinases presents a viable approach for developing therapies for TNBC. Hence, this review is intended to offer a comprehensive summary of the latest insights into TNBC effects of pivotal kinase inhibitors and their prospective roles in TNBC therapy. It encompasses significant and innovative advancement in pre-clinical research and evaluates the effectiveness of selected combination studies involving kinase inhibitors and other agents. This approach represents a novel strategy for developing potent anti-TNBC treatments and combating chemoresistance. Additionally, the review assesses the safety and effectiveness of various clinical trials involving kinase inhibitors.

It's important to note that the role of kinases in TNBC tumorigenesis and kinase inhibitors have been extensively reviewed by numerous authors in recent years, including the works by Li et al.,[11] Jiawei et al.,[21] Costa et al.,[12] Khan et al.,[22] Wang and Micky,[23] Zhao et al.,[24] Gerosa et al.,[25] and Mehlich and Marusiak.[26] Yet, these reviews primarily concentrate on clinical and biological aspects, with only a partial examination of kinase inhibitors, and tend to overlook pre-clinical small molecule inhibitors. Consequently, our review presents a comprehensive view of small molecule kinase inhibitors, including detailed mechanisms of action against TNBC. In summary, this review sheds light on the potential of kinase inhibitors in the treatment of TNBC. Fig. 1 depicted a simplified overview of kinases in TNBC.

2. Mitogen activated protein (MAPK) kinase pathways inhibitors/modulators in preclinical TNBCs

Ahn et al demonstrated that compounds **1** and **2,** as well as microtubule acetylation-specific inhibitors, had anti-TNBC response and induced cell cycle arrest via activating JNK/AP-1 pathway.[27] Lee et al reported that anti-cancer and anti- metastasis activity of MEK1 inhibitor with compound **3**, not only in vitro, it prompted G1 cell cycle interruption and programmed cell death, while also impeding the progression of triple-negative breast cancer (TNBC) xenograft tumors and hindering metastasis to the lungs in a murine model by diminishing extracellular signal-regulated kinase (ERK) phosphorylation.[28] Furthermore, Chiu et al showed that compound **4**, a derivative of curcumin, can also reduce the invasion of TNBC MDA-MB-231 cells by targeting MAPK/ERK/Akt signaling pathway.[29] Earlier research corroborated the effectiveness against TNBC by inhibiting MAPK pathway. For example, compound **5,** a MEK inhibitor has been shown to possess anti-TNBC activity by reducing lung metastasis in a xenograft model.[30] Importantly, Ebelt and colleague identified compound **6** a JNK inhibitor which demonstrated efficacious response in TNBC in vivo and also sensitizes TNBC cells to lapatinib.[31] Furthermore, compounds **7**,[32] investigated by Chen et al. displayed anti-TNBC activity both in vitro and in vivo while produced synergistic apoptosis inducing effects in TNBC cells with a HSP90 inhibitors via potently inhibited Raf/ERK pathway. Similarly, compounds **8**,[33] and **9**[34] have also presented effectiveness triggering apoptosis against TNBC via modulating MAPK signaling pathways. Fig. 2 depicts the structures of the compounds **(1–9)** targeting MAPK kinase pathway in TNBC.

3. PI3K/Akt/mTOR pathway inhibitors in preclinical TNBCs

Xu et al investigated compound **10,** a dual inhibitor of PI3Kα and

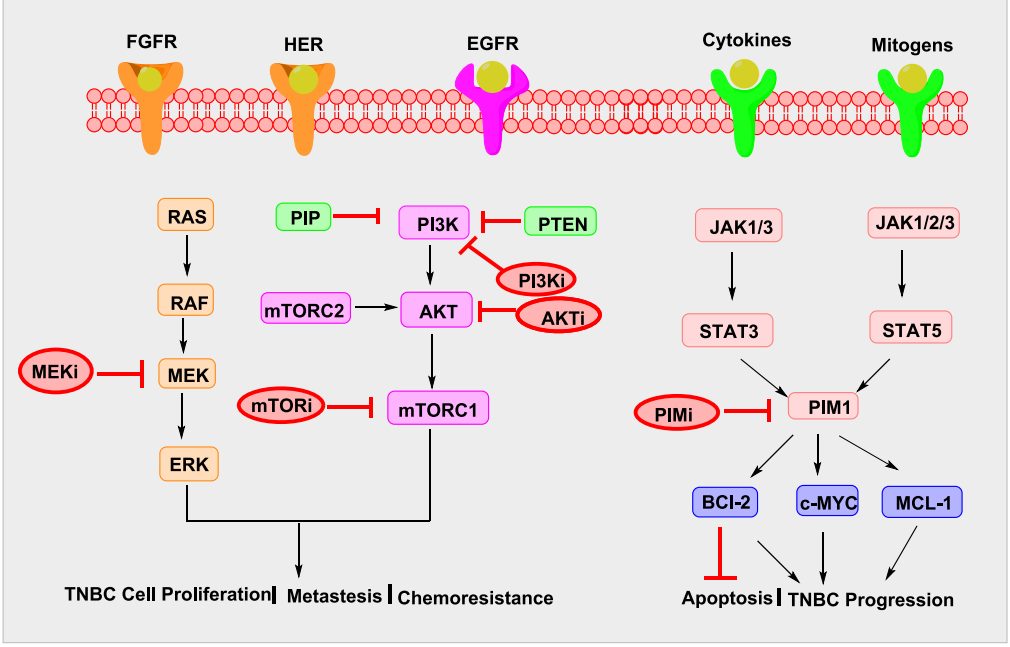

Fig. 1. A simplified overview of RAS/MEK/ERK, PI3K/Akt/mTOR and JAK/PIM pathways in TNBC.

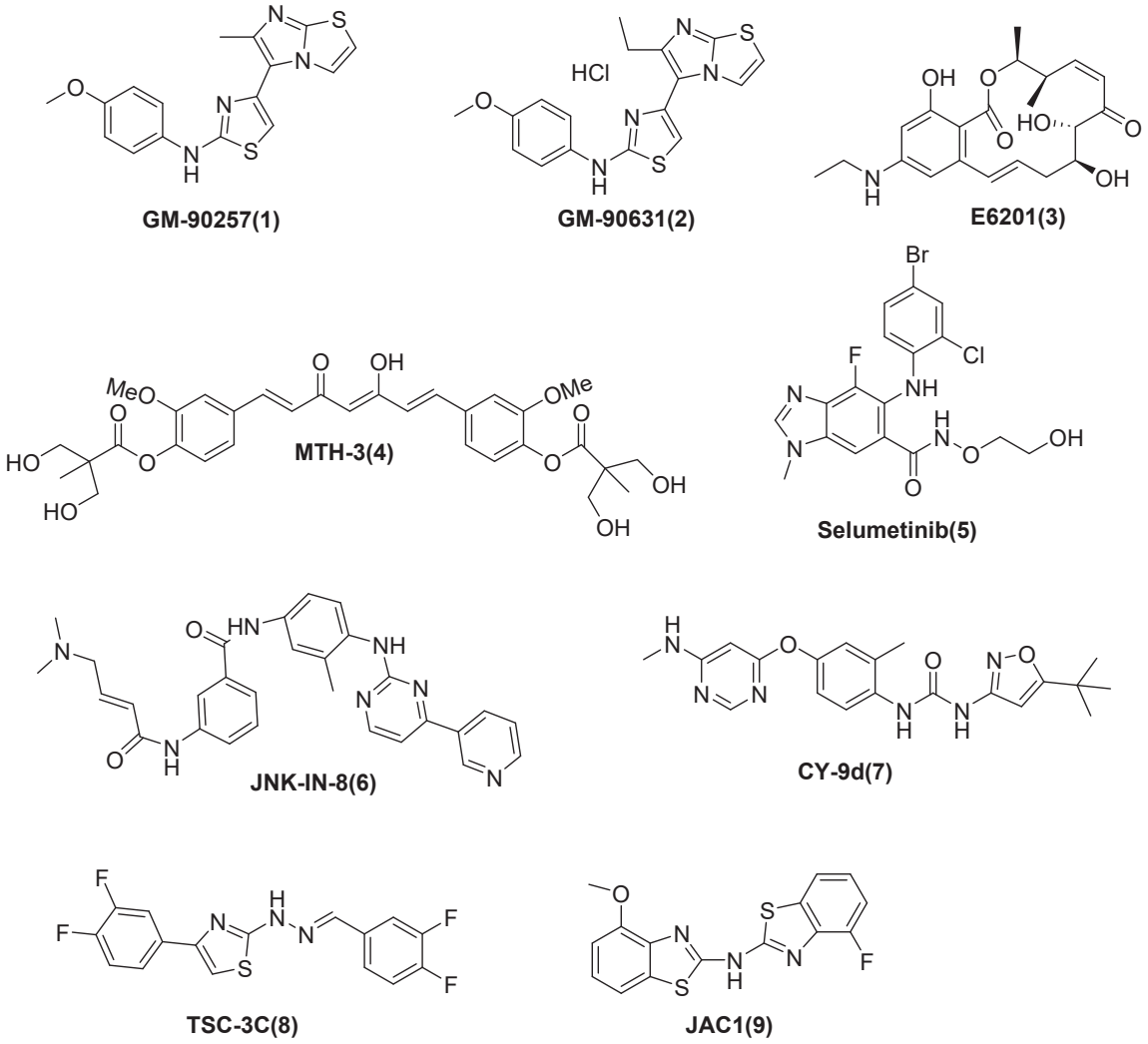

Fig. 2. Chemical structure of the compounds **(1–9)** targeting MAPK kinase pathway in TNBCs.

mTOR (IC_{50} values of 291 and 60.8 nM, respectively). The compound exhibited promising anti-breast cancer activity in both in vitro and in vivo models, demonstrating particular efficacy against TNBC by suppressing cell invasion and migration in vitro. The molecule exhibited inhibition activity against several cancer cell lines including MCF-7 and MDA-MB-231 cell lines with IC_{50} values between 1.07 to 0.002 μM. Furthermore, in vivo studies using BALB/c nude mice harboring MCF-7 xenografts demonstrated significant tumor suppression (57 % inhibition). The treatment mechanism involved activating caspase-3 and promoting Bax expression while decreasing Bcl-2, alongside inhibiting the expression of PI3K, Akt, mTOR (mammalian target of rapamycin), and S6K1 in vivo. Additionally, it suppressed the epidermal growth factor (EGF) induced expression of *N*-cadherin and vimentin while enhancing E-cadherin expression, thereby reducing epithelial-mesenchymal-transition (EMT) in the MCF-7 and MDA-MB-231 cell lines in vitro.[35] Conversely, Cordover et al discovered that the PAK4 and NAMPT inhibitor, known as KPT-9274 **(11)** exhibited anti-TNBC activity via mTORC2 signaling modulation.[36] Moreover, compound **12** validated its anti-TNBC activity via attenuating CDK4, Cyclin D1 and Cyclin B1 expression and promoted p21 and p27 expression. The treatment markedly reduced Akt phosphorylation activity. In addition, it reduced the growth of HCC1806 tumor in Balb/c nude mice at the dose of 20 mg/kg.[37] Another in vitro experiment found Compound **13** to effectively induce apoptosis against MDA-MB-231 and T47D cells lines that inhibited the migration of MDA-MB-231 cell line. It was found that the molecule **(13)** could suppress PI3K/Akt/mTOR and hedgehog (Hh) signaling pathways.[38]

Similarly, a shikonin derivative compound **14,** exhibited potential efficacy against TNBC cell line. Treatment with compound **14** triggered apoptosis and arrested the cell cycle in the G2/M phase, in addition to inhibiting migration of MDA-MB-231 cells. It also increased the levels of cleaved PARP, caspase-3 and Cyt c, while decreasing Bcl-2 expression. Further investigation suggests that the anti-TNBC efficacy of compound **14** is associated with disruptions in the PI3K/Akt/mTOR and Wnt/β-catenin signaling pathways.[39] Shawish and colleagues recently synthesized a novel series of pyrazolyl s-triazine derivatives. Within this series, compounds **15** and **16** were ascertained as having the most potent anti-cancer effects, exhibiting significant activity against various cancer cell lines, notably including TNBC MDA-MB-231 cells. It induced apoptosis via promoting p53, Bax, caspase-3.-8 and-9, and reducing PI3K, Akt, mTOR and Bcl-2 mRNA expression.[40] Similarly, P.M and colleagues synthesized a group of compounds in which compound **17** appeared to have the highest anti-cancer property with a GI_{50} values of 2.96 μM against TNBC cell. It induced apoptosis and inhibited p-ERK and p-Akt (Thr308) expression in MDA-MB-468 cells.[41] Xu et al. synthesized compound **18,** which demonstrated superior inhibitory effects on MDA-MB-231 and MDA-MB-468 cells line through the induction of apoptosis and autophagic cell death. The treatment with compound **18** led to a decrease in Bcl-2 and an increase in Bax expression. It also reduced the phosphorylation of mTOR (Ser2448), p-p70S6K (Thr389), p-Akt

(Ser473), p-4EBP1 (Thr37/46) while enhancing the expression of Beclin1 and LC3-II expression in both cells lines during in vitro studies.[42] Similarly, Yao and co-workers developed molecule **19,** which exhibited anti-tumor activity against TNBC cells lines by inducing apoptosis, evidenced by lowered Bcl-2 levels and heightened expression of Bax, cleaved caspase-3 and-8. Molecule **19** also promoted autophagy, as indicated by increased levels of Beclin-1 and LC3-II expression in MDA-MB-231 cells in vitro. Additionally, inhibited histone deacetylases (HDAC) and p-p70S6K (ser371) and MMP-2 expression while enhancing E-cadherin levels.[43] Previous studies have also identified several active molecules for the treatment of TNBC, such as compound **20**. It is a selective, irreversible mTOR inhibitor, that was found to be effective against TNBC cell growth in vitro. Apart from that, **20** suppressed the growth of MDA-MB-231 xenograft tumor and lung metastasis at the dose of 10 mg/kg.[44] Jose et al reported that serotonin derivatives compounds **21** and **22** also exhibited antiproliferative activity and induced apoptosis in TNBC cells by modulating the Akt/mTOR pathway.[45] Interestingly, compound **23**, a selective mTOR inhibitor, displayed excellent pharmacokinetic property and metabolic stability in in vivo experiments.[46] Likewise, compound **24,** suppressed viability and metastasis progression of TNBC cell line via induction of autophagy which was obtained by inhibiting Akt/mTOR pathway.[47]

Another compound, UNBS5162 **(25)** has significantly attenuated cell proliferation, migration, and invasion of MDA-MB-231 cells in vitro. It promoted active caspase-3 and Bax expression while downregulated Bcl-2 expression. Additionally, decreased Akt, mTOR, P70S6 kinase and 4 EB1 phosphorylation.[48] Chopra et al found that Torin2 **(26)** diminished proliferation of TNBC cells by blocking the activity of mTOR and other PI3K-like kinases (PIKKs).[49] Importantly, Hossan and others have demonstrated that Cu (II)–cardamonin complex **(27)** induced anticancer activity against MDA-MB-468 and PANC-1 cancer cells. Further studies disclosed that it induced DNA damage and accumulated ROS, activated caspase -3/7, PARP cleavage, and decreased Mcl-1, p-Akt and p-4EBP1 expression.[50] In a similar manner, compound **(28)** decreased cell growth and induced apoptosis in MDA-MB-231 cells in vitro as well as remarkably suppressed the growth of TNBC xenograft tumor in mice. Mechanistically, inhibited PI3K/Akt pathway via reducing the DNA methylation level of PTEN.[51] It was reported that M2698 **(29)** suppressed growth of tumor in in vivo model of TNBC and HER-2 cells mouse xenograft in a dose-dependent manner while potently inhibited p70S6K and Akt and thus suppressed the PI3K/Akt/mTOR pathway.[52] Likewise, LG25 **(30)** demonstrated cell growth reduction and induced apoptosis and cell cycle arrest at G2/M phase in MDA-MB-231 cells. Also remarkably suppressed the growth of TNBC xenograft tumor in mice, promoted Bax expression, down regulated Bcl-2 and inhibited Akt/mTOR/NF-κB signaling pathway.[53]

Another compound, I194496 **(31)** displayed cell growth attenuation and reduced the metastasis of human TNBC cells in vitro as well as exhibited strong TNBC xenograft tumor growth inhibition in nude mice. Treatment with **31** downregulated PI3K/Akt and Ras/Raf/ERK pathway. It also suppressed Anxa2/ STAT3 and VEGF/FAK/Paxillin signaling pathways.[54] Liu and co-authors have found that Lapatinib **(32)** diminished cell growth and induced apoptosis in TNBC cells via suppressing CIP2A/PP2A/p-Akt signaling cascade.[55] Similarly, compounds **(33/34)** attenuated growth of TNBC cells and induced G0/G1 phase cycle arrest primarily targeting PI3K/Akt pathway.[56] Finally, NVP-BEZ235 **(35)** abates the growth of MDAMB-231 and MDA-MB-468 cells line by triggering mutant p53 degradation and autophagy. Mechanism studies revealed that treatment with **35** significantly reduced the expression of p-Akt at ser473, p-mTOR at ser2448 and p-P70 at Thr389. Additionally, promoted LC3 protein expression and AMPK phosphorylation.[57] Fig. 3 depicts the structures of the compounds **(10–35)** targeting PI3K/Akt/mTOR in TNBC.

4. Cyclin dependant kinases (CDKs) inhibitors in preclinical TNBCs

Umed et al. identified compound **36** from a newly synthesized series of 3-pyrimidinylazaindoles as the most effective inhibitor of CDK2/9. This compound exhibited a favourable pharmacokinetic profile and significantly inhibited growth of 4T1 TNBC tumor at a dosage of 15 mg/kg.[58] Quereda et al. described compound **37**, a selective dual inhibitor of CDK12 & CDK13, which exhibited effectiveness against TNBC in preclinical trial models. Moreover, compound **37** demonstrated a response combating TNBC when used combination with the other agents such as PARP inhibitors.[59] A series of compounds were synthesised by Jing and co-workers, in which, compounds **38** and **39** displayed significant cell growth reduction activity against the MDA-MB-231 cells by inhibiting the CDK2/CDK9.[60] Similarly, dinaciclib (compound **40**), a novel CDK9 inhibitor was found to suppressed TNBC cells via inducing apoptosis and G2/M phase cell cycle arrest in vitro studies, thus effectively inhibiting tumor growth in TNBC PDX model. In in vitro studies, it was observed to supress cyclin B1, cyclin T1, MYC, pRb, and pCDK1. As for in vivo studies, it significantly inhibiting cyclin B1, cyclin T1, MYC, and CDK9.[61] Moreover, compound **41**, a pan-CDK inhibitor exerted anti-TNBC activity within a single treatment while exhibited enhanced effectivity in combination with doxorubicin in p53-mutated.[62]. Likewise, another novel CDK2/9 inhibitor compound **42**, lead to TNBC growth inhibition both in vitro and in vivo tumor xenografts. Mechanistically, it inhibited p-Smad3 at T179 and c-MYC expression along with increasing p15 and sub-G1 phase cell distribution in MDA-MB-231 cells. Furthermore, the combination of compound **42** with eribulin resulted in a heightened anti-TNBC response, surpassing the efficacy observed with either treatment alone.[63] A recent study showed that compound **43** suppressed bone metastasis in ER+ and TNBC breast cancer in animal models.[64] Liang et al developed compound **44**, a derivative of 2-aminopurine, which was effective in inhibiting the growth of MDA-MB-231 cells. It induced a G2/M phase cell arrest by effectively suppressing CDK2 kinase activity.[65] Fig. 4 depicts the structures of the compounds (**36–44**) targeting CDKs in TNBC.

5. Proviral integration site for Moloney murine leukemia virus kinase (PIM) inhibitors in preclinical TNBCs

Several studies have found that inhibition of PIM1 and PIM2 kinase showed anti-TNBC response. In one study, the researchers developed a chromones based compound, called compound **45** which inhibited in vitro TNBC cell proliferation and induced apoptosis. Further investigation of this compound revealed that it was a multi-kinase inhibitor with maximum inhibition against PIM1 and PM2 kinase. Furthermore, molecular docking suggested a favorable binding interaction between both the kinases.[66] Katsuta et al demonstrated that compound **46** significantly inhibited tumor growth both in vitro and in animal models against several cancer cells including TNBC MDA-MB-231 cell line.[67] Likewise, compound **47** that is a potent PIM1 kinase inhibitor, inhibited MCF-7 breast cancer cell growth activity while showed a weak inhibition against MDA-MB-231 cell line.[68] In addition, PIM2 inhibitor, compound **48** developed by Zhao et al demonstrated effectiveness against TNBC tumor. It induced apoptosis and autophagic cell death which are evidenced by the reduced expression of Bcl-2, upregulation of Bax and LC3-II expression. Moreover, treatment also inhibited growth of MDA-MB-231 xenograft tumor in mice at the dose of 40 mg/kg.[69] Kennedy and colleague demonstrated compound **49**, a PIM kinase inhibitor, as the anti-breast cancer agent which potently inhibited many different breast cancer cells including TNBC cells. Also, attenuated growth of BT-474 and HCC-1954 tumor xenograft in mice. It inhibited PIM/PI3K/mTOR pathway, leading to an anti-cancer effects.[70] Conversely, Horiuchi et al examined the effects of PIM1 kinase inhibitors, specifically compounds **50** and **51**, and observed their ability to suppress tumor growth in human TNBC tumors within a PDX model that

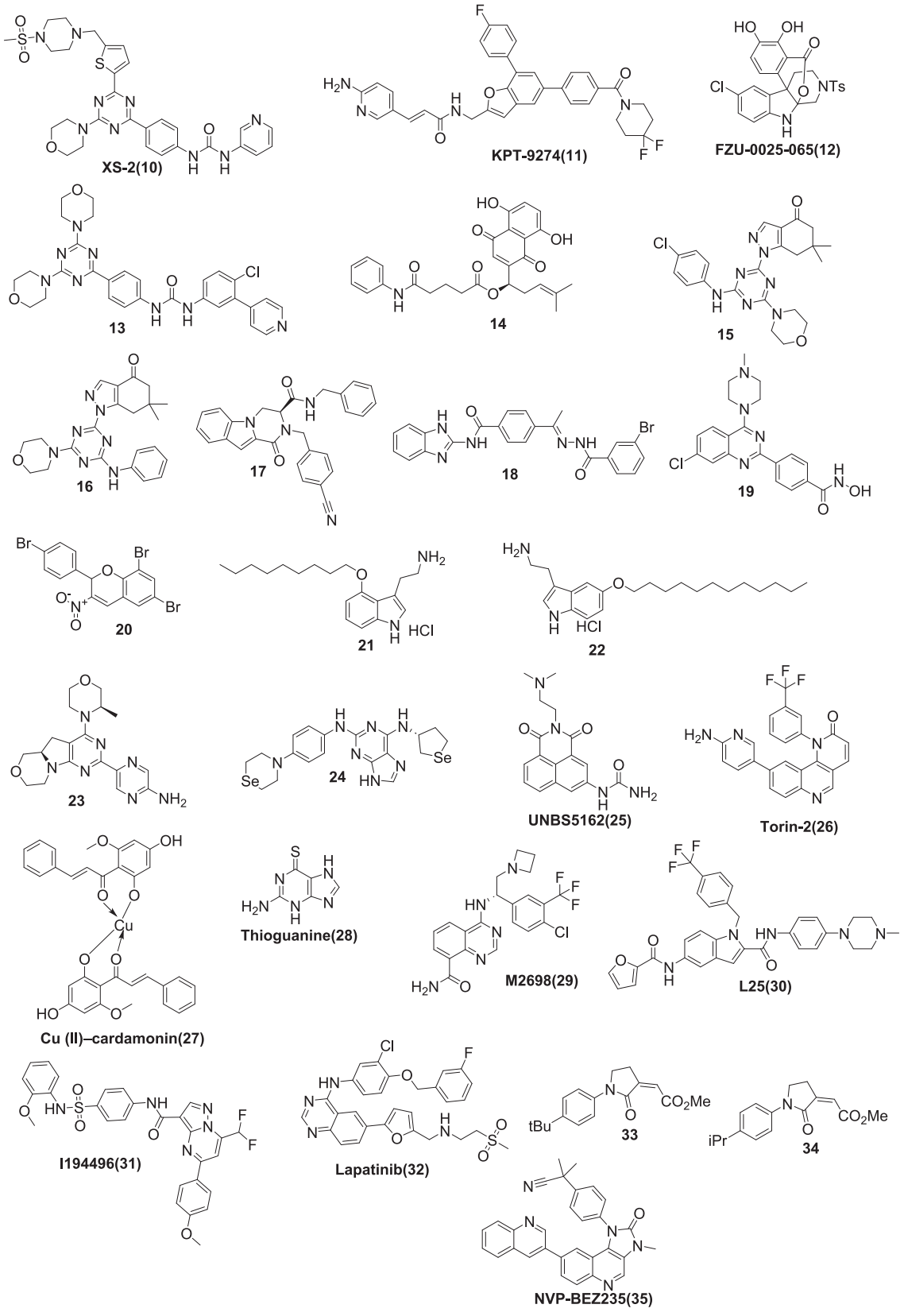

Fig. 3. Chemical structures of the compounds **(10–35)** targeting PI3K/Akt/mTOR pathway in TNBC.

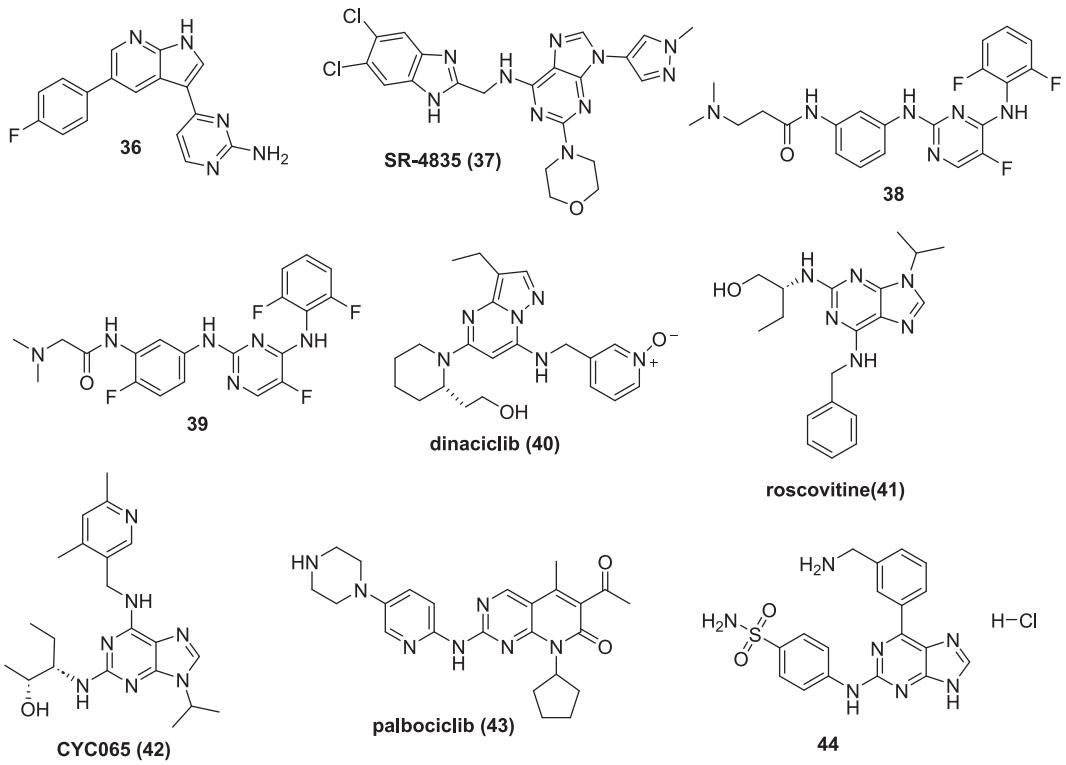

Fig. 4. Chemical structures of the compounds **(36–44)** targeting CDKs in TNBC.

exhibited high MYC expression [97]. Additionally, the PIM1 inhibitor, known as compound **52**,[71] along with the dual CK2/PIM1 inhibitor, referred as compound **53**,[72] have also been identified as effective in combating TNBCs. Fig. 5 depicts the chemical structure of PIM kinase inhibitors **(45–53)** in TNBCs.

6. Receptor and non-receptor tyrosine kinase inhibitors in preclinical TNBC

Compound **54**, a tyrosine kinase inhibitor was found as the potent anti-TNBC agent by Dong et al. which could inhibit AR+ TNBC cells within a single course of treatment in vitro. It significantly suppressed the RTK/ACK/AR and Akt/mTOR signaling pathway in AR+ TNBC MDA-MB 453 cell. Moreover, at the dose of 25 mg/kg, compound **54** suppressed tumor growth of MDA-MB-231 xenograft in mouse model and inhibited the growth of TU-BcX-4EALNb PDX tumor. In addition, in combination with paclitaxel (10 mg/kg), compound **54** synergistically inhibited the tumor growth of MDA-MB-231 xenograft and lung metastases in vivo. The combination treatment also remarkably inhibited the growth of TU-BcX-4EALNb PDX tumor and inhibited p-FAK and p-YB-1 expression.[73] Another tyrosine kinases inhibitor compound **55** which can inhibit ABL1 and other kinases including FLT3, TIE2, SRC, PDGFRα, FYN, AXL and MET, has demonstrated inhibitory activity against TNBC cancer stem cells (CSCs) via suppressing AXL and the transcription factor KLF5 expression.[74] The synthetic compound **56** has exhibited a potent inhibition against Src and KDR kinase with IC_{50} values of 0.003 and 0.032 μM, respectively. However, it was also found to inhibit several other kinases involved in the MAPK signal transduction. This compound also exhibited a notable response in vitro and significantly suppressed the tumor growth of MDA-MB-231 and the MDA-MB-435 cells xenograft at the dose of 20 and 40 mg/kg. Moreover, it also displayed an excellent pharmacokinetic profile. Besides, treatment considerably decreased p-Src, p-FAK, p-MEK and p-ERK expression.[75] Similarly, Zhang and co-author developed the anti-TNBC compound **57** which showed a potent inhibitory activity against Src with an IC_{50} vales of 0.0009 μM. However, it also inhibited other kinases such as B-RAF and C-RAF. It not only inhibited cell growth, but induced apoptosis and G0/G1 cell cycle arrest of MDA-MB-231 cell line in vitro as well as inhibited tumor growth in mice bearing MDA-MB-231 tumor xenograft. Additionally, it remarkably inhibited p-Src, p-FAK, p-MEK and p-ERK expression in vitro.[76] Fig. 6 depicts the structures of the compounds (**54–65**) targeting tyrosine kinase in TNBC. Table 1 **(58–65)** contains a summary of tyrosine kinase inhibitors. Apart from the compounds already described several others miscellaneous kinase inhibitors **(1–8)** have also been reported and their activity summarized in Supplementary Table T1 while the structure can be found in Supplementary Fig. S1.

7. Naturally occurring compounds and kinases inhibition in preclinical TNBCs

7.1. Natural products targeting MAPK kinase pathways in preclinical TNBCs

Gao et al exhibited that compound **66** induced inhibition of TNBC cell growth, migration, and invasion in a dose-dependent manner. He went on to discover that it induced apoptosis in Hs 578 T and MDA-MB-468 cells in vitro via reactive oxygen species (ROS) accumulation. In addition, when compound **66** used in the treatment process, it suppressed the cellular inhibitor of apoptosis 2 (cIAP-2) expression and c-Jun *N*-terminal kinases (JNK/c-Jun) signaling pathway.[85] A different study pinpointed compound **67** as a highly effective anti-TNBC agent that curbs cell proliferation and triggers both apoptosis and cell cycle arrest in MDA-MB-231 and mouse TNBC cell line 4 T1, with its impact intensifying in proportion to the dose administered. Mechanistic studies revealed that the treatment suppressed CDK1, cyclin B1, CDK2, and cyclin A2 phosphorylation. In fact, it also promoted the expression of E-cadherin, Bax and cleaved PARP and attenuated vimentin expression. However, it was observed to also increase p-p38, p-JNK, and p-ERK1/2 expression.[86] Whereas Huang et al showed that compound **68**,

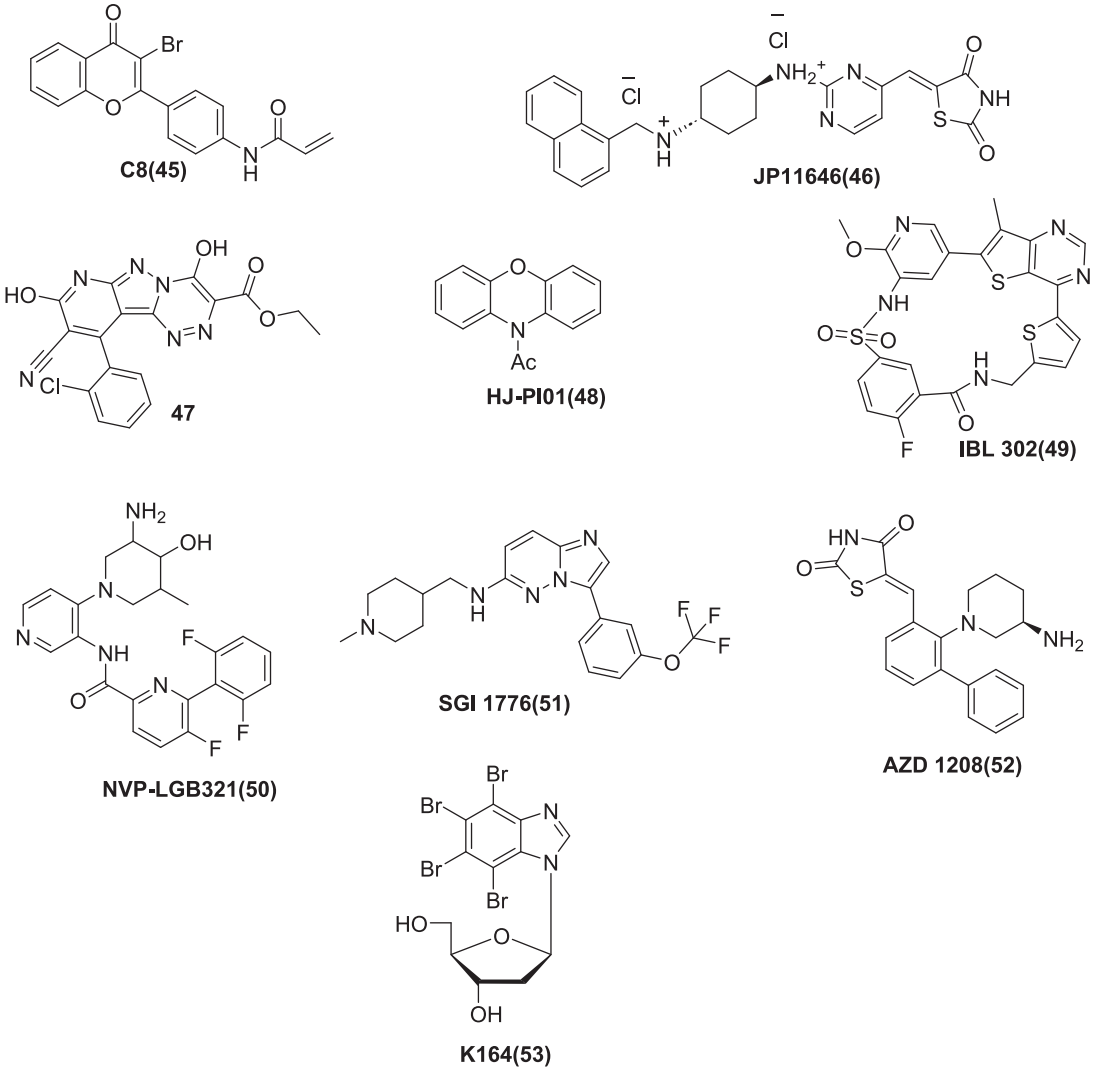

Fig. 5. Chemical structure of PIM kinase inhibitors **(45–53)** in TNBCs.

attenuated proliferation of MDA-MB-468 and MDA-MB-231cells in vitro as well as suppressed MDA-MB-231 xenograft tumor in mice targeting DUSP6/MAPK signaling.[87] Moreover, the compound **69** had exhibited anti-cancer activity across multiple TNBC cell lines in vitro and suppressed growth of MDA-MB-231 tumor on chick chorioallantoic membranes. It was found to accumulate ROS, activate ERK1/2, inhibit p-Akt and induce apoptosis. It was also established to induce autophagy through empirical evidence of the expression of microtubule-associated proteins 1A/1B light-II (LC3-II) and *P*62 protein in MDA-MB-231 cells.[88] Finally, compound **70** demonstrated effectiveness against TNBC by inducing cell cycle arrest and blocking MAPK and Akt signaling pathways.[89] Besides, Compound **71** exhibited TNBC growth inhibition via modulating MAPK signaling pathways.[90] Fig. 7 depicts the structures of the compounds **(66–71)** targeting MAPK kinase pathways in TNBC.

7.2. Natural products targeting PI3K/Akt/mTOR pathway in preclinical TNBCs

In contrast, compound **72** showed in vitro anti-tumor activity via inducing apoptosis and cell cycle arrest at G2/M phase in MDA-MB-231 and MDA-MB-468 cells in a time and dose dependent manner. The molecular mechanism revealed that treatment with compound **72** significantly suppressed cyclin D1, cyclin B1, CDK6 expression as well as Akt/mTOR and Signal transducer and activator of transcription 3 (STAT3) signaling pathways. Furthermore, it significantly reduced the growth of MDA-MB-231 xenograft tumor the doses of 25 and 50 mg/kg.[91] Li et al reported that molecule **73** induced non-protective autophagy and apoptosis via blocking Akt/mTOR signaling cascade. Compound **73** induced anti-proliferative activity, apoptosis, and inhibited migration of SUM-159-PT and MDA-MB-231 cells in vitro. Mechanistic study revealed that the treatment significantly enhanced the expression of LC3-II protein, concurrently decreasing the levels of Akt and mTOR in both in SUM-159-PT and MDA-MB-231 cell lines during in vitro studies. Additionally, the treatment inhibited the growth of MDA-MB-231 xenograft tumors in mice by inducing autophagy and apoptosis. The effect was confirmed by the reduced expression of B-cell lymphoma 2 (Bcl-2), Ki67, p62, alongside a marked increase in Bax and LC3B expression within the tumor tissues.[92] Xu and co-authors reported that compound **74** inserted anti-cancer activities against MDA-MB-231 and EFM-192A cells lines with IC_{50} values of 70.96 and 78.58 nM, respectively. Moreover, it induced apoptosis and G2/M phase cell cycle arrest. Further investigation unveiled that the molecule suppressed the expression of CDK1 and Cyclin B1, Bcl-2, p-PI3K, and p-Akt while enhanced the expression of Cytosine c, PARP, Bax, active Caspase-3, and -9 as well as p21 and p27 in vitro.[93] On the contrary, molecule **75** was reported to have anti-TNBC activity via autophagy induction by inhibiting PI3K/Akt/mTOR cascade.[94] Kumar et al exhibited that compound **76** suppressed in vitro cell viability, migration and angiogenesis and

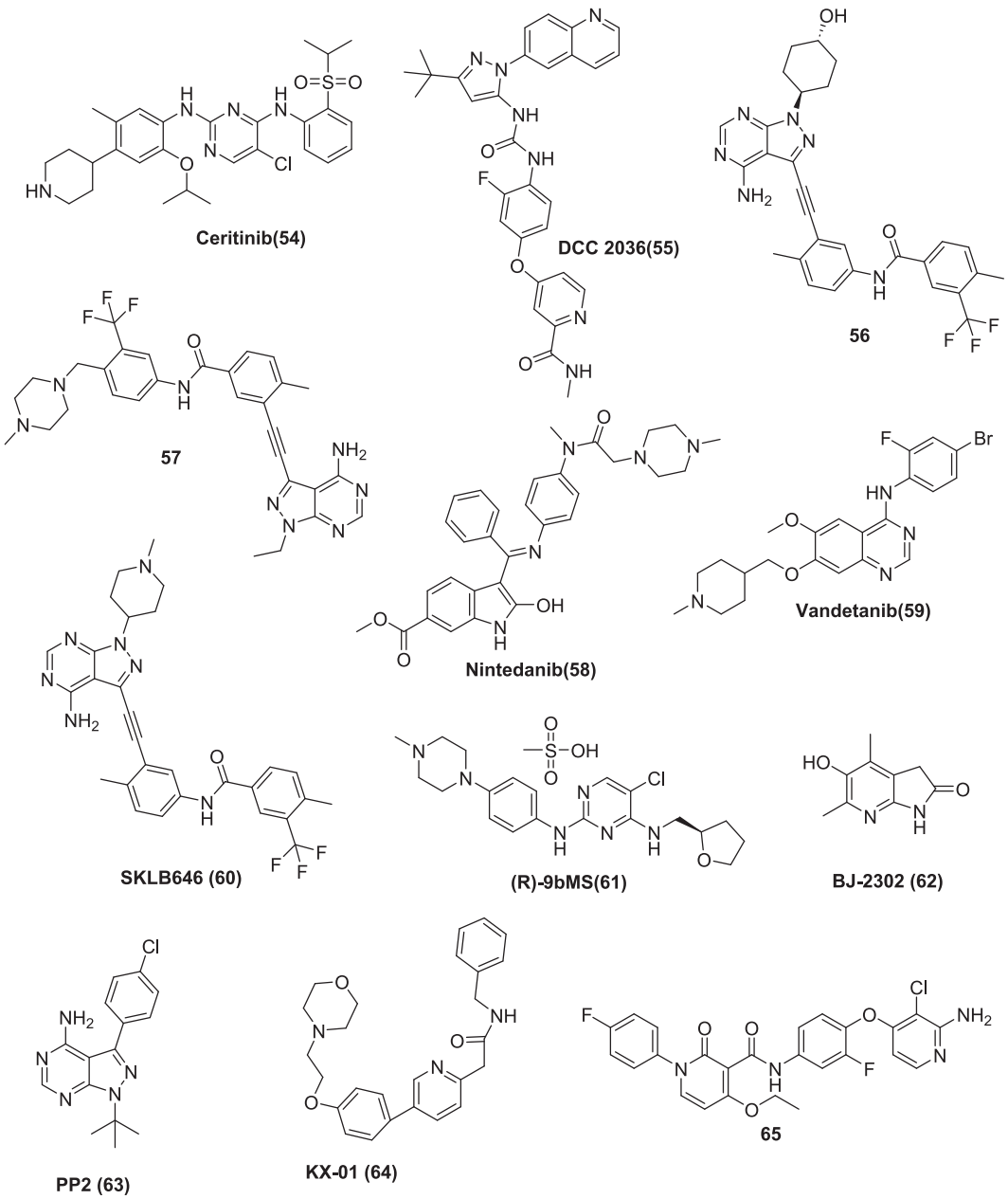

Fig. 6. Chemical structure of Src and other tyrosine kinase inhibitors **(54–65)** in TNBC.

induced apoptosis in MDA-MB-231 and ER+ MCF-7 breast cancer cell lines. Moreover, it significantly decreased the growth of MDAMB-231-Luc cell line in NOD/SCID mice at the dose of 25 mg/kg and 100 mg/kg. The treatment enhanced the expression of Bax, Bad, cleaved caspase-3, -9, and PARP, while suppressing Bcl-2 expression in MDA-MB-231 cells, observed both in vitro and in vivo studies.[95] In a similar manner, compound **77** was found to suppress proliferation and induce apoptosis, necrosis, and autophagy in MDA-MB-231 cell line in a time and dose dependent manner in vitro studies. Thereby, significantly reducing the growth of MDA-MB-231 cell xenograft tumor in nude mice at the dose of 80 mg/kg which was similar to paclitaxel at 10 mg/kg in the same TNBC tumor. Mechanistic study revealed that it activated cleaved caspase-3, and promoted Bax, LC3-II, p-RIP3 expression while reduced expression of p-Akt-473 and Bcl-2 in a dose dependent manner in vitro. Additionally, it also promoted RIP3 and MLKL expression both in vitro and in vivo.[96] Zhao et al demonstrated the anti-TNBC response of compound **78**, which was obtained mainly via the alteration of Akt/p38 MAPK signaling was linked to reactive oxygen species (ROS) accumulation too.[97] A study disclosed the anti-TNBC effects of compound **79** by inducing apoptosis which are evidenced by DNA fragmentation, caspase-3 activation and PARP cleavage, reduced expression of Bcl-2 and promote expression of Bax protein. It also inhibited PI3K 110α/85α, p-Akt (ser473), p70S6K1 p- 4E-BP1 in vitro.[98] The activities of compounds **(80–92)** are summarized in Table 2. Fig. 8 depicts the structures of the compounds **(72–92)** targeting PI3K/Akt/mTOR in TNBC.

7.3. Natural compounds targeting miscellaneous kinases in preclinical TNBCs

Zhao and co-author found that compound **93** suppressed cell proliferation and migration, and induced apoptosis in MDA-MB-231 cells via blocking PIM1 kinase and suppressing ROCK2/STAT3 signaling.[112] Another compound, ziyuglycoside II **(94)** inhibited TNBC cell proliferation in vitro and downregulated ITGB4/FAK and Akt as well as p38 MAPK signalling pathways.[113] It is noteworthy that Thymoquinone **(95)**

Table 1
Summary of small molecules **(58–65)** targeting tyrosine kinases in preclinical TNBCs.

Compound	Target kinase	Biological response	Mechanism of Action	Reference
Nintedanib (58)	TKs	Induced apoptosis and inhibited tumor growth in vivo in TNBC cells.	Suppressed the SHP-1/p-STAT3 pathway	[77]
Vandetanib (59)	TKs	Induces tumor regression in both HER2+ and TNBC PDX xenografts with high expression of RET or EGFR	Suppressed RET/ EGFR phosphorylation in PDX tumors.	[78]
SKLB646 (60)	Src/Raf/ VEGFR2	Significantly inhibited cell growth and migration of TNBC cells as well as induced apoptosis and cell cycle arrest in vitro. Also, attenuated the tumor growth of TNBC xenograft in a dose dependent manner	It inhibited the Src. and VEGFR2 with IC_{50} values of 0.002 mmol/L and 0.012 mmol/L, respectively. It potently inhibited the expression of p-Src in TNBC cells and tumors. Also, inhibitedp-FAK (Tyr925) , p-MEK(Ser217/ 221),p-ERK1/2 (Thr202/tyr204) expression in vitro.	[79]
(R)-9bMS (61)	TNK2	Significantly inhibited the viability of TNBC cell lines in vitro.	Remarkably inhibited phosphorylated TNK2 expression.	[80]
BJ-2302 (62)	Src	Inhibited MDA-MB-231 cell viability in vitro. Also, repressed the tumor growth and metastases in the xenograft mouse model	Treatment downregulated both lysosomal enzyme cathepsin S(CTSS) and MMP-9 and effectively inhibited p-Src and suppressed PI3K/ Akt and Ras/Raf/ ERK pathways	[81]
PP2(63)	Src	Inhibited cell growth and migration of SUM1315MO2 and MDA-MB-231 cells. Also, remarkably inhibited tumor growth of a SUM1315MO2 xenograft on female BALB/c nude mice	Effectively inhibited c-Src phosphorylation and reduced Vimentin expression while promoting E-cadherin. Also decreased p-Akt, CDK4 and cyclin E1 and D1 expression in TNBC cells in vitro.	[82]
KX-01(64)	Src/ Pretubulin	Suppressed the cell growth and migration of TNBC cells and induced cell cycle arrest at G2/M phase in vitro and MDA-MB-231 xenograft in animal model	Effectively inhibited p-Src and p-FAK (Y397) in TNBC cell lines and MDA-MB-231 tumor. Also suppressed the expression of p-ERK, p-Akt, p-STAT3 in vitro.	[83]
BMS-777607 (65)	TKs	Inhibited TNBC cell proliferation in vitro and significantly inhibited MDA-MB-231 xenograft tumor in mice.	Inhibited p-MET and p-RON expression, also suppressed p-Akt and p-ERK1/2 expression	[84]

has also significantly inhibited proliferation, migration, and invasion of TNBC cells. Also, remarkably suppressed the growth of MDA-MB-231 tumor in mice. Mechanistically, it suppresses the expression of eEF-2K both in vitro and in vivo. It also reduced Src/FAK and Akt phosphorylation.[114] Li et al showed that Rubioncolin C **(96)** has anti-TNBC activity via the modulation of multiple pathways. Compound **96** reduced cell growth of TNBCs via inducing apoptosis and autophagy. Also suppressed tumor growth in animal models. It activated MAPK and inhibited mTOR/Akt/p70S6K and NF-κB pathways.[115] Interestingly, Liu and colleague reported that DATS **(97)** suppress migration and invasion of MDA-MB-231 and HS 578T TNBC cells by inhibiting MMP2/9 expression and deactivating NF-κB and ERK/MAPK pathways.[116] Fig. 9 depicts the chemical structures of the compounds **(93–97)** targeting various kinase in TNBC.

8. Preclinical combination studies of kinases with kinase inhibitors and other agents in TNBCs

A significant amount of research has been conducted on the combined efficacy of two or more kinase inhibitors, or kinase inhibitors paired with other agents, against TNBCs. studies combining different kinase inhibitors have shown promise, as many combinations exhibit synergistic anti-TNBC effects, justifying further investigation in clinical trials. The majority of the studies described under this section were conducted combining clinical phase compounds and preclinical active molecules against TNBC to examine their further potentiality. For instance, Coussy et al examined the effects of a clinical phase PI3KCa inhibitor, (compound **121**), both alone and combined with a MEK inhibitor (compound **5**) in metastatic PDX TNBC model. The combination treatment exhibited remarkable improvement in anti-tumor activity in PDXs harboring of PIK3CA, Akt1, BRAF, and FGFR4 genomic alterations compared to a single treatment, such as, in the HBCx-60 model tumor carrying FGFR4 amplification, PIK3CA mutation, and Akt1 amplification, combination treatment induced tumor growth inhibition with a TGI of 94 %, which was much superior to monotherapy. Furthermore, two additional models, HBCx-165and HBCx-178, which are resistant to anthracycline, also demonstrated increased anti-tumor activity. Importantly, all three PDX models displayed some complete and durable responses via the inhibition of PI3K/Akt/mTOR and RTK/MAPK signaling pathways.[117] Likewise, results from another study suggested that the combination between MEK inhibitor (compound **98**) and Pan-HER inhibitor (compound **99**) together with clinical phase mTOR inhibitor (compound **124**) and pan-HER inhibitor (compound **99**) significantly increased cancer growth inhibition in both HER2+ and TNBC breast cancer models.[118] Interestingly, Haga et al showed that by blocking the Akt/mTOR pathway with compound **120** (a clinical phase Akt inhibitor) and compound **100** (mTOR inhibitor) inhibited intrinsic resistance to Src inhibitor, dasatinib (compound **101**) in TNBC.[119] Johnson and

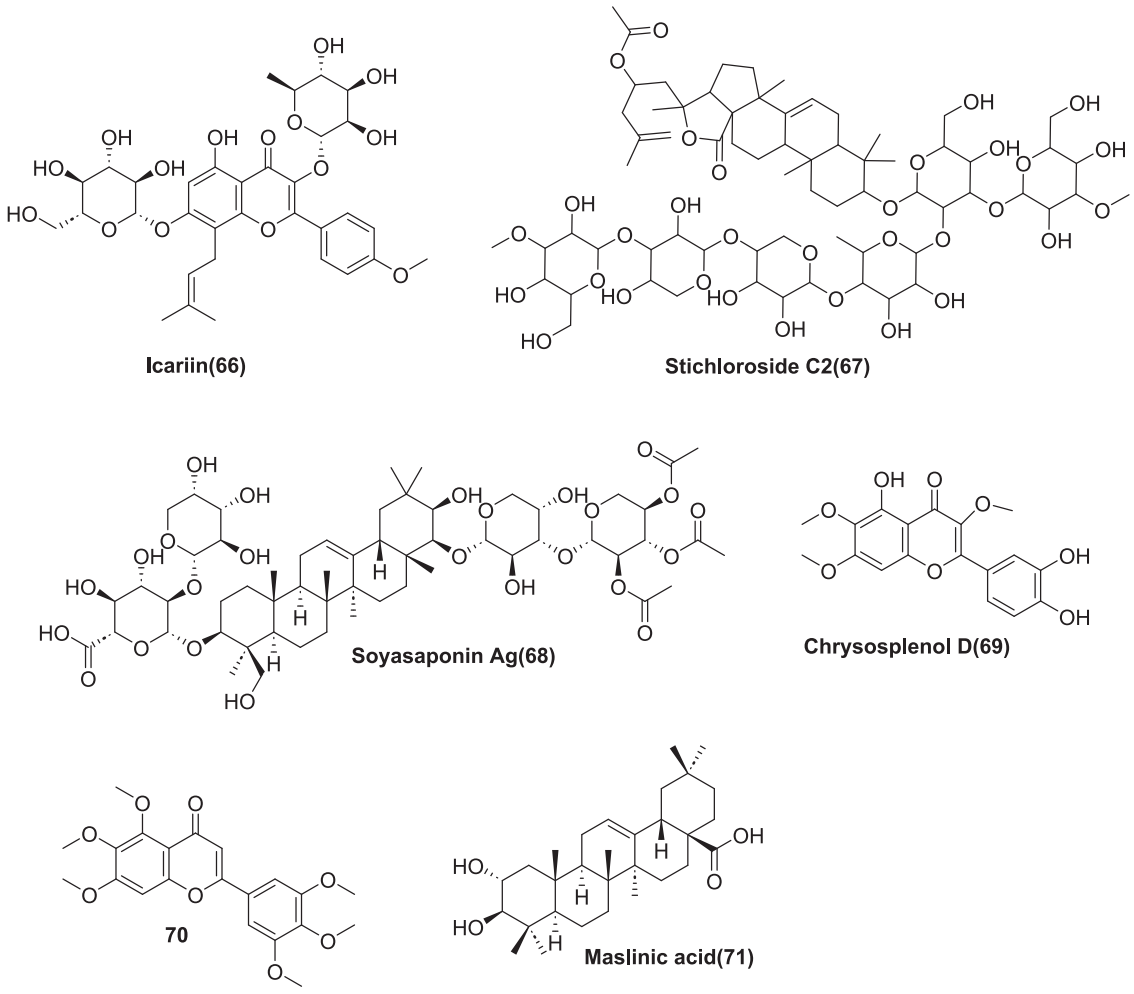

Fig. 7. Chemical structures of the natural compounds **(66–71)** targeting MAPK kinase pathways in TNBC.

colleagues reported that blocking of PI3K and AMPK signaling in fact enhanced the radiosensitivity of TNBC.[120] Conversely, Zhu et al showed that a concomitant treatment of PARP inhibitor (compound **102**) and CDK4/6 inhibitor (compound **43**) demonstrated in vitro and in vivo synergistic anti-cancer response in BRCAmut TNBC cells.[121] Similarly, co-administration of CDK4/6 inhibitor (compound **103**) and an AR inhibitor was found synergistic in AR+ TNBC cells.[122] Additionally, another similar study have reported synergistic activity of CDK4/6 inhibitor (compound **104**) together with and AR inhibitor (compound**105**) against AR+ TNBC model.[123] Microtubule-targeting agent (compound **106**) and a clinical phase mTOR inhibitor (compound **124**) might have exhibited good synergistic anti-TNBC activity via the inhibition of pAkt/pS6K/pS6 signaling pathway.[124] Cocco et al reported that resistance of PI3K/Akt inhibitors such as compound **107/122** (clinical phase Akt inhibitor) in TNBC can be prevented via the inhibition of autophagy by compound **108** also, the triple combination of **107 or 122** and **108** and paclitaxel sensitize to TNBC cells.[125] Reports also showed that concomitant inhibition of PI3K/Akt and MEK5/ERK5 pathways can be more beneficial than treatment alone in reducing TNBC cell proliferation. For instance, co-administrations of Akt inhibitor (compound **122**) and MEK5 inhibitor (compound **109)** and ERK5 inhibitor (compound **110)** were investigated and found that both of the combination significantly enhanced the anti-cancer activity than single treatment.[126] On the contrary, the combination of compound **111** and **100** as well as its analogs was found to effectively inhibits p-mTOR and p-4EBP1, as well as suppressed Akt and ERK signaling, and thus sensitizing rapamycin and apagoges resistance TNBC cells that offers a potential therapeutic strategy to inhibit mTOR-targeted therapy resistance in TNBC cells.[127] Fig. 10 depicts the structures of the compounds **(98–118)**. Synergistic action of kinase inhibitors **(112–118)** and other compounds in combination against TNBCs have been summaries in Supplementary Table T2.

9. Clinical studies of kinase inhibitors in TNBC: Outcome of mono and combination therapy

9.1. Buparlisib

A phase II single clinical trial of a Pan-PI3K inhibitor buparlisib **(119)** in 50 TNBC patients revealed inadequate response. The trial exhibited no conformed CR nor PR, but the disease was stabilized in 12 % of the study participants (6 patients, all SD ≥4 months). The study had a median progression-free survival (PFS) of 1.8 months (95 %, CI:1.6–2.3) and the median overall survival (OS) was 11.2 months (95 %, CI 6.2–25). Among the major toxicities, fatigue appeared in 58 % of the participants, followed by nausea and hyperglycemia (34 %), and 30 % of the participants were anorexic. In addition, while 12 % of the participants were depressed, another 10 % experienced anxiety (10 %).[128]

9.2. MK-2206

Similarly, another phase II clinical trial of Akt inhibitor MK-2206 **(120)**, as monotherapy was also carried out in 27 participants with advanced breast cancer (HR+, TNBC and HER+) harboring either

Table 2
Summary of the naturally occurring compounds **(80–92)** targeting PI3K/Akt/mTOR pathway in preclinical TNBCs.

Molecules	Targets	Biological Activities	Mechanism of Action	Reference
Luteolin (80)	Akt/mTOR	Suppressed proliferation and metastasis of AR-positive TNBC cells	Treatment attenuated the expression of MMP9 and Akt/mTOR protein	[99]
Gomisin G (81)	Akt	Reduced proliferation of MDA-MB-231 and MDA-MB-468 cells	Downregulated Cyclin D1expression and Akt Phosphorylation	[100]
Strictinin(82)	PI3K/Akt/GSK3ß	Reduced cell survival and migration of TNBC cells and induced apoptosis	Inhibited ROR1, and p-Akt (Ser-473), p-BAD (ser-136), and p- GSK3 (ser-9)	[101]
Gallic acid (83)	PI3K/Akt	Suppressed cell proliferation and induced apoptosis in HCC1806 cell line	Treatment up regulated the expression of Bax, cleaved-caspase-3, and -9, P53, P-ERK1/2, P-JNK, and P-P38 proteins. On contrary it down regulated the expression of Bcl-2, P-PI3K, P-Akt, and P-EGFR proteins in vitro	[102]
Fisetin (84)	PTEN/Akt/GSK3β	Inhibited cell growth, migration, and invasion in TNBC cell lines MDA-MB-231 and BT549 cells in a dose-dependent manner	Inhibited PTEN/Akt/GSK3β Signal Pathway	[103]
Berbamine (85)	PI3K/Akt/mTOR	Reduced the viability of MDA-MB-231 and MCF-7 breast cancer cell line.	Treatment reduces the expression of PI3K, mTOR, Akt phosphorylation, as well as COX-2, LOX, and MDM2 expression in dose dependent manner. At the same time, promoted p53 expression.	[104]
Lupiwighteone (86)	PI3K/Akt/mTOR	Inhibited proliferation of MDA-MB-231 and MCF-7 breast cancer cells by inducing apoptosis	Treatment promoted the expression of cleaved caspase-3, -7, -8, -9, PARP, and Bax while reduced the Bcl-2 expression. Also inhibited PI3K/Akt/mTOR signaling cascade.	[105]
Arnicolide D (87)	Akt/mTOR/STAT3	Significantly attenuated cell growth and induced apoptosis and G2/M phase cell cycle arrest in human TNBC cells. Also suppressed MDA-MB-231 xenograft tumor growth in female BALB/c nude mice	Treatments inhibited Akt/mTOR and STAT3 signaling Pathways	[106]
Shikonin (88)	miR-17-5p/PTEN/Akt	Reduced the migration and invasion of MDA-MB-231 and BT549 cells	Inhibited EMT such as suppressed *N*-cadherin and promoted Vimentin expression and inhibited miR-17-5p/PTEN/Akt pathway	[107]
4-HW (89)	Akt	Inhibited growth of MDA-MB-231 cells and induced G1 phase arrest.	Suppressed PI3K/Akt pathway, also inhibited the expression of survivin and promoted p21 and p27 expression	[108]
Quercetin (90)	EGFR/PI3K/Akt	Gold nanoparticles-conjugated quercetin induced apoptosis in MCF-7 and TNBC MDA-MB-231 cell lines	Suppressed EGFR/PI3K/Akt pathway	[109]
Quercetin-3-ME (91)	PI3K/Akt	Attenuated cells proliferation, migration, and invasion and induce cell cycle arrest at the G2/M phase and apoptosis in TNBC and ER/PR positive, HER-2 negative cells	Downregulated EMT expression by promoting E-cadherin and decreasing MMP-2 expression, also reduced p-PI3K, p-Akt at Ser473 and p-mTOR levels.	[110]
Casticin (92)	PI3K/Akt	Significantly suppressed migration and invasion of MDA-MB-231 and 4T1cells line, also attenuated lung metastasis of 4T1 cells in mice.	Treatment reduced MMP-9 mRNA and protein expression, c-Jun and c-Fos as well as inhibited Akt phosphorylation, and PI3K expression.	[111]

PIK3CA or Akt and PTEN mutations or PTEN loss. Among the administered 33 % (9 participants) TNBC, 22 % were either PIK3CA or Akt mutated and 56 % were PTEN mutations/PTEN loss. However, results exhibited limited clinical response in all types of breast cancer which suggests that Akt inhibitor **120** alone is insufficient to prevent advanced breast cancer.[129]

9.3. Alpelisib

Similar to this, a phase II clinical trial was conducted to assess the effects of alpelisib **(121),** a PI3K inhibitor, as a monotherapy in participants with advanced breast cancer (ER+/HER2-, n = 33 and TNBC, n = 10), that had a mutation in the PI3K pathway. While the trial showed a

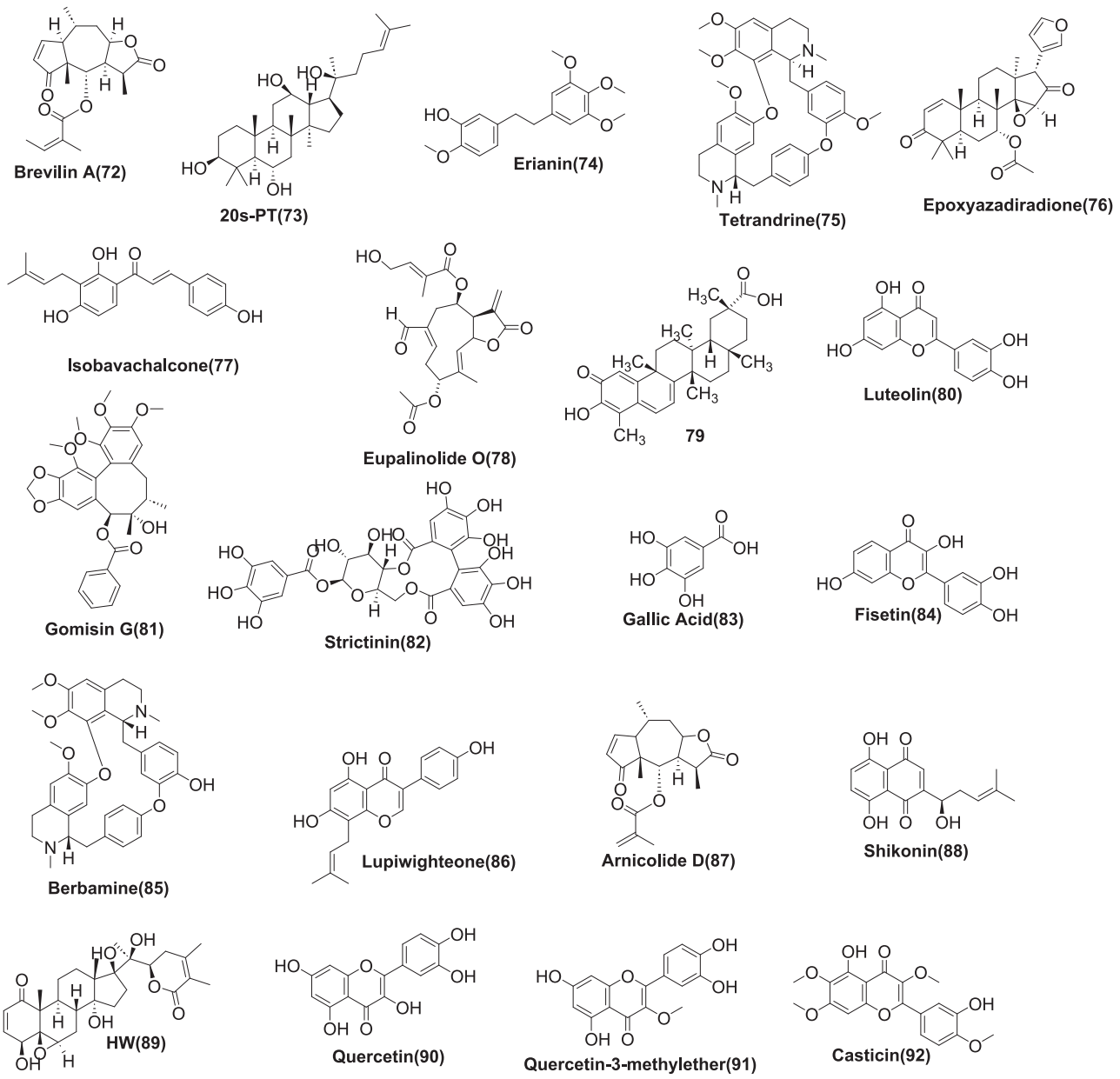

Fig. 8. Chemical structures of the natural compounds **(72–92)** targeting PI3K/Akt/mTOR in TNBC.

relatively encouraging outcome with an overall response rate of 30 % and a clinical benefit rate of 36 % for PI3K mutated ER+ breast cancer, the results against TNBC were unsatisfactory. The majority of participants in this trial reported grade 3 and 4 adverse effects, with hyperglycemia accounting for 32.6 % of cases, maculopapular rash for 25.6 %, colitis and diarrhea for 7.0 % respectively.[130]

9.4. Ipatasertib and paclitaxel

In contrast, compared to the paclitaxel + placebo group, the ipatasertib (**122**) and paclitaxel combination phase 2 trial has shown some encouraging results in the treatment of TNBC. With ipatasertib intention-to-treat population (n = 62) and PTEN-low tumors (n = 48), the median progressive-free survival was 6.2 months. Whereas the paclitaxel pls placebo treated groups looked to be 4.9 months and 3.7 months, respectively. But no discernable changes was found in this investigation.[131]

9.5. Alpelisib and Olaparib

In a similar vein, a phase 1b clinical combination trial of alpelisib **(121)** and olaparib (PARP inhibitor) was conducted in 17 pre-treated advanced TNBC participants with median age of 51 years and there were 18 % participants having BRCA1/2 mutation. The study's findings showed that 41 % of participants had SD and 18 % (95 % CI, 3.8–43.4) of participants had a PR. The median response period was 7.4 months, and the median PFS and OS were, respectively 3.6 months (95 % CI, 1.8–9.2) and 11.8 months (95 % CI, 4.2–19.6), respectively. Treatment-related fatigue (71 %), anorexia (59 %), hyperglycemia (59 %), and nausea (53 %) were most common all-grade toxicities.[132]

9.6. Gedatolisib and cofetuzumab pelidotin

Eighteen participants were included in a second phase I trial examining gedatolisib **(123)** and cofetuzumab pelidotin. A confirmed PR of 3/18 (ORR 16.7 %) and stable disease was obtained in this trial. In

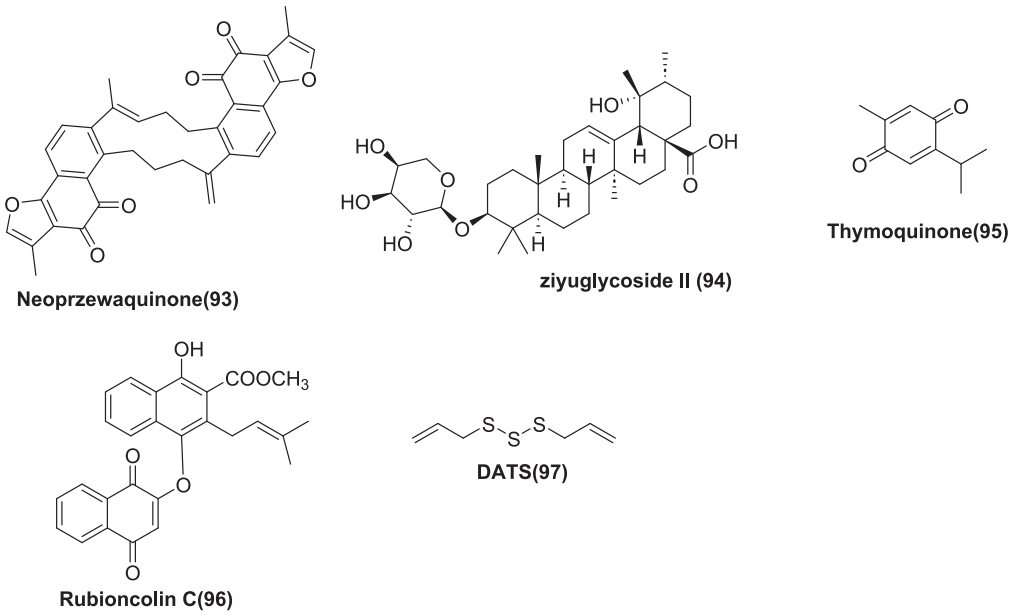

Fig. 9. Chemical structures of the natural compounds **(93–97)** targeting various kinase in TNBCs.

addition, at 18 weeks, the trial's clinical benefit rate (CB18) was 27.8 % (5/18). The meridian OS was 9.4 months (95 % CI:4.3–15.8), and median PFS was 2.0 months (95 % CI:1.2–6.2) overall. Nonetheless, there were a few typical side effects, including mucositis, nausea, anorexia, constipation and exhaustion.[133]

9.7. Everolimus and eribulin

The phase 1 clinical trial of eribulin with everolimus (**124**) involved 27 patients in total, with a median age of 55 years, to investigate the safety, efficacy, and toxicity of this combination research in participants with metastatic TNBC. The highest dose of everolimus that was shown to be safe was 5 mg per day when combined with an eribulin dose of 1.1 mg/m2 on days 1 and 8 of every three weeks (RP2D). Of the 25 participants who received treatment, 9 participants (36 %) showed a partial response (PR), and the remaining 9 patients (36 %) showed stable disease. Conversely, 7 participants (28 %) showed signs of illness progression. The median time to progression in this trial was found to be 2.6 months (95 %, CI: 2.1, 4.0), and the OS was 8.3 months (95 % CI [5.5, undefined]). Regretfully, participants with TNBC co-administration of eribulin and everolimus does not result in any CR response. In this investigation, the most frequent toxicities discovered were hyperglycemia, stomatitis, and neutropenia.[28] In an analogous vein, everolimus was also the subject of an additional phase 1 clinical trial combined with gemcitabine/cisplatin chemotherapy. Unfortunately, the study failed to yield synergistic anti-TNBC activity in the participants.[134]

9.8. Capivasertib and chemo-drugs

Finally, 140 patients participated in a phase II trial comparing capivasertib **(125)** and paclitaxel (Cap-P) against placebo plus paclitaxel (PP). The median PFS and OS in the Cap-p treated group were 5.9 months and 19.1 months, respectively, but the median PFS and OS in the PP treated group were lower at 4.2 months and 12.6 months, respectively. Interestingly, PFS rose considerably with Cap-P treatment in participants with PIK3CA/Akt1/PTEN-altered malignancies. Akt inhibitors in combination may be a better therapeutic option for this type of mutant TNBC, as evidenced by the study's documentation of a median PFS of 9.3 months versus 3.7 months in PIK3CA/Akt1/PTEN-altered TNBC.[135] The molecular structure of kinase inhibitors **(119–129)** under TNBC clinical trials was shown in Fig. 11. Table 3 provides an overview of the ongoing clinical trial.

9.8.1. Perspective and future directions

Recent progress in the discovery of new targets biomolecules across the TNBC molecular subtypes open a new window to treat this complex heterogeneous disease.[26] Kinases, along with other targets might offer a promising therapeutic option in the cure and preventing TNBC, particularly its potentiality for the subtypes specific targeted therapy development. Kinases such as PI3K, Akt, and mTOR inhibitors have exhibited success in numerous preclinical in vitro and in vivo trials, also currently under clinical trials which are discussed before. It worth mentioning that clinical trial results of single kinase inhibitors against TNBCs are disappointing, however, a combination strategy displayed improved activity. It is also a challenge to find the right combination regime for the right kinds of patients. Next, to reduce side effects and minimize toxicities appeared to be another challenge. Similarly, overcoming drug resistance in TNBCs is another concern. On the other hand, many of the existing molecules are not potent enough to reach in clinical trials, besides, selectivity is also an issue to be considered. In addition, maximum pre-clinical studies discussed before, are lack of pharmacokinetics investigations. This insufficient pharmacokinetic data may interrupt further development. Therefore, it is crucial to develop more novel and potent kinase selective molecules. In synthetic approach, presently available SAR can provide an important starting point, for example 1) there are certain functional groups such as fluoro (–F), chloro (–Cl), bromo (–Br), hydroxy (–OH), methoxy (–OCH_3), and morpholine, but not limited to this, are appeared to be abundant throughout the molecular structure of kinase inhibitors presented in this paper, also some scaffold for instance,1,3,5-trazine, chromone, and pyrrolo[2,3-*b*]pyridine could also be taken into consideration in designing new molecules. 2) Computer aided drug design is another popular current trend which may also help to design and synthesis new kinase inhibitors.[136] Besides, 3) structural optimization also led to new potent and selective anti-TNBC kinase inhibitors which are cited in this article. 4) Natural products can also serve as a promising source of small molecular kinase inhibitors, since a noteworthy number of Phyto-molecules have been summarized in this paper which induced potent TNBC growth suppression primarily via kinase inhibitions.

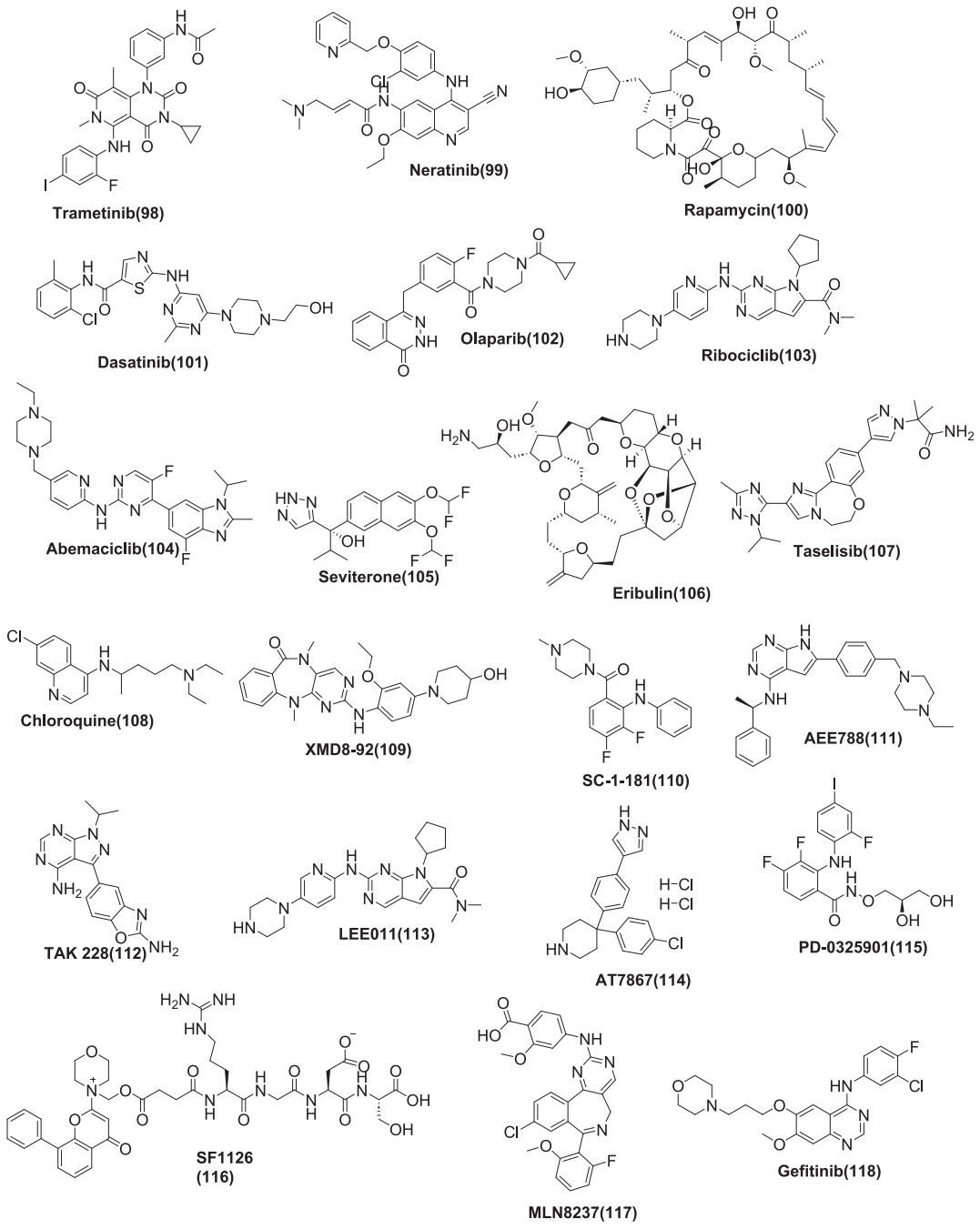

Fig. 10. Chemical structure of kinase inhibitors and other compounds **(98–118)** acting synergistically in preclinical TNBC studies.

10. Conclusion

Kinase inhibitors have shown considerable promise in pre-clinical trials for treating advanced metastatic breast cancers, with many advancing to clinical trials. However, due to the heterogeneous nature of TNBC, targeting a single kinase may not adequately halt TNBC progression. Conversely, studies combining various kinase inhibitors with other agents have not only synergistically improved anti-TNBC responses but also facilitated chemo sensitization, suggesting a more effective strategy for developing targeted therapies in the future. It remains imperative to innovate and evaluate novel kinase inhibitors, understanding their structure–activity relationships (SAR) to uncover how individual functional groups contribute to their anti-cancer effects by targeting different kinases. Furthermore, recent progress in kinase targeting presents an optimistic perspective for the creation of new anti-TNBC treatments.

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Declaration of Generative AI and AI-assisted technologies in the writing process

During the preparation of this work the author(s) used no generative AI and AI-assisted technologies.

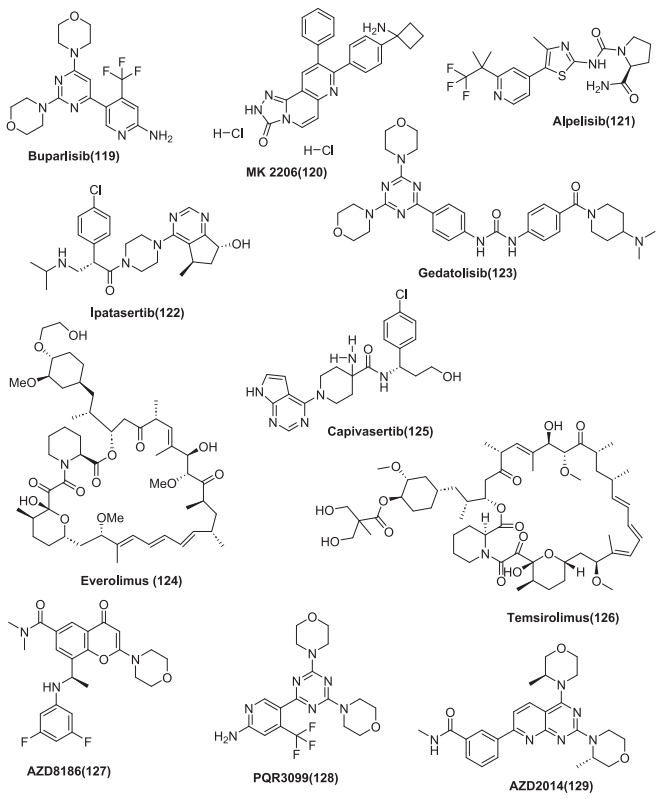

Fig. 11. Chemical structure of kinases inhibitors **(119–129)** under TNBC clinical trials (Clinicaltrial.gov).

Table 3
Single and combination clinical studies of small molecular kinase inhibitors in TNBC (Clinicaltrial.gov).

Treatments	Trial Phase	NCT identifier
CUDC-907(dual Akt/HDACi)	Phase I	NCT02307240
AZD8186(mTORi)	Phase I	NCT01884285
Everolimus (mTORi)	Phase II	NCT01931163
BKM120(PI3Ki)	Phase II	NCT01790932
AZD2014 (mTORC1/2i) + AZD8186	Phase I	NCT01884285
Gedatolisib (PI3K/mTOR) + PTK7-ADC	Phase I	NCT03243331
BKM120 (PI3Ki) + Olaparib	Phase I	NCT01623349
BYL719 (PI3Ki) + Olaparib	Phase I	NCT01623349
PQR309 (dual PI3K/mTORi) + eribulin	Phase I, II	NCT02723877
Temsirolimus (mTORi) + Neratinib	Phase I, II	NCT01111825
Vistusertib (AZD2014) (mTORC1/2i) + Olaparib	Phase I, II	NCT02208375
Capivasertib(AZD5363)(Akti) + Olaparib	Phase I, II	NCT02208375
Gedatolisib (PI3K/mTORi) + Talazoparib	Phase I, II	NCT03911973
Ipatasertib(Akti) + Atezolizumab Cyclophosphamide + Paclitaxel + Doxorubicin	Phase II	NCT05498896
Ipatasertib (Akti) + Paclitaxel	Phase II	NCT02301988
Ipatasertib + Capecitabine + Eribulin + Carboplatin + Gemcitabine	Phase II	NCT04464174
Everolimus(mTORi) + nab-Paclitaxel	Phase II	NCT04395989
Capivasertib + Olaparib	Phase II	NCT03801369
Everolimus + Carboplatin	Phase II	NCT02531932
Everolimus + Doxorubicin + Bevacizumab	Phase II	NCT02456857
GSK2141795(Akti) + Trametinib	Phase II	NCT01964924
Alpelisib (PI3Ki) + nab-Paclitaxel	Phase III	NCT04251533
Capivasertib + Paclitaxe	Phase III	NCT03997123

CRediT authorship contribution statement

Rajibul Islam: Writing – review & editing, Writing – original draft, Conceptualization. **Khor Poh Yen:** Writing – review & editing, Writing – original draft. **Nur Najihah 'Izzati Mat Rani:** Writing – review & editing, Visualization, Software. **Md. Selim Hossain:** Review and editing.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.bmc.2024.117877.

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