REVIEW

Clinical evidence of the efficacy of everolimus and its potential in the treatment of breast cancer

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Abstract: The PI3K/Akt/mTOR (phosphatidylinositol 3-kinase/protein kinase B/mammalian target of rapamycin) pathway regulates several key cellular functions and its dysregulation creates an environment that promotes tumorigenesis as well as resistance to therapy. The mTOR inhibitor everolimus has emerged as a promising agent in the treatment of breast cancer and was recently approved in combination with exemestane for advanced hormone receptor–positive disease after progression on a nonsteroidal aromatase inhibitor. Everolimus may also be effective in combination with cytotoxic and human epidermal growth factor receptor-2-directed therapies for the treatment of other subtypes of breast cancer. This paper highlights preclinical and clinical data that have emerged on the role of mTOR inhibition in breast cancer. Although generally well tolerated, everolimus carries a unique side effect profile of which both patients and providers should be made aware. Recommendations related to the administration of everolimus in the clinical setting are also discussed.

Keywords: everolimus, breast cancer, mTOR inhibition

Introduction

It is estimated that in 2012 there were approximately 226,870 new cases of invasive breast cancer among women in the United States. An estimated 39,920 breast cancer deaths were expected, making it the second leading cause of cancer death in women. Since 1990, death rates for breast cancer have steadily decreased, reflecting progress in earlier detection and improved treatment.[1]

Treatment strategies for breast cancer vary depending on stage, hormone receptor status, human epidermal growth factor receptor-2 (HER2) status, and other tumor- and patient-specific characteristics. However, one of the major challenges of treatment is overcoming intrinsic and/or acquired drug resistance. Multiple mechanisms of resistance exist, including reduced accumulation of chemotherapy in cancer cells, alterations in drug targets, activation of detoxifying mechanisms, increased repair of drug-induced cellular damage, and alterations in cell signaling, cell cycle control, and apoptosis signaling.[2] Each one of these mechanisms represents a potential opportunity to develop therapeutic agents in an effort to circumvent resistance.

The field of oncology has witnessed a paradigm shift from a "one size fits all" approach to treatment to a more individualized approach in which a deeper understanding of target oncogenic proteins and pathways has allowed us to develop more rational therapies, resulting in increased efficacy and decreased toxicity.

This paper discusses our understanding of the phosphatidylinositol 3-kinase/protein kinase B/mammalian target of rapamycin (PI3K/Akt/mTOR) pathway in

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the pathogenesis of breast cancer as well as the successful development of an inhibitor of this pathway, everolimus, and its role in the treatment of breast cancer.

PI3K/Akt/mTOR pathway

The PI3K/Akt/mTOR pathway regulates several normal cellular functions, including cellular growth, proliferation, survival, and differentiation.[3] Dysregulation of this pathway creates an environment that promotes tumorigenesis, and activating mutations are frequently reported in human cancers.[4]

Activation of this pathway can occur through activation or mutation of either PI3K or Akt, overexpression of growth factor receptors such as HER2 or insulin-like growth factor receptor, or through loss of tumor suppressor phosphatase and tensin homolog deleted from chromosome 10 (PTEN) activity.[5–7]

Activated PI3K leads to downstream phosphorylation of Akt, a serine/threonine kinase, which in turn activates other key downstream effectors such as mTOR. mTOR is a serine/threonine kinase that is a key mediator of cellular proliferation, apoptosis, angiogenesis, and cellular metabolism.

The effects of mTOR are mediated by two distinct multiprotein complexes, mTORC1, which is sensitive to inhibition by rapamycin, and mTORC2, which is not.[8] mTORC1 exerts its effects via phosphorylation of the eukaryotic initiation factor 4E-binding protein 1 (4E-BP1) and the p70 ribosomal S6 kinase 1 (S6K1), leading to enhanced mRNA translation, cell proliferation, growth, and survival. The function of mTORC2 is incompletely understood but is thought to be related to cytoskeleton organization as well as cell proliferation, survival, and metabolism.[8,9]

Mutations in the PI3K catalytic alpha subunit (*PIK3CA*) gene and the loss of PTEN, which inhibits the mTOR pathway, are frequently observed in breast cancer.[10] Several studies have shown that alterations of the *PIK3CA* gene are observed in approximately 10%–40% of breast cancers.[11] Activation of the PI3K/Akt signaling pathway in breast cancer cells leads to the development of resistance to therapy, and high Akt and mTOR activity are especially associated with the development of resistance to endocrine therapy.[12–14] Similar associations have been made between mTOR activation and trastuzumab resistance.[15–17] These findings provide the rationale for the addition of mTOR inhibition to chemotherapy, endocrine therapy, anti-HER2 therapy, or a combination of these, in an effort to delay or reverse resistance.

mTOR inhibitors

Rapamycin is a naturally occurring fungicide produced by the bacterium *Streptomyces hygroscopicus*. It forms a complex with the cytosolic protein FK-binding protein 12 (FKBP12) which blocks the actions of 4E-BP1 and p70 ribosomal S6K, resulting in cell cycle arrest.[18] Historically, rapamycin has been used as an immunosuppressant in solid organ transplant recipients. In recent years, the use of synthetic rapamycin analogs, or "rapalogs," with more favorable pharmacokinetic properties, has extended to the field of oncology.

Temsirolimus (Torisel®, Pfizer, Inc, New York, NY, USA) is US Food and Drug Administration (FDA)-approved for the treatment of renal cell carcinoma; everolimus (Afinitor®, Novartis Pharmaceuticals Corporation, East Hanover, NJ, USA) is FDA-approved for the treatment of renal cell carcinoma, pancreatic neuroendocrine tumor, angiomyolipoma, advanced estrogen receptor (ER)+/HER2 breast cancer, and subependymal giant cell astrocytoma in individuals with tuberous sclerosis complex; ridaforolimus is currently in clinical development.

Preclinical data

Several preclinical studies have demonstrated the efficacy of mTOR inhibition in breast cancer. One of the drivers of the PI3K pathway is ER activation, and activation of both PI3K and Akt may be invoked by crosstalk between epidermal growth factor receptor and ER. Such crosstalk has been associated with estrogen-independent transcriptional activity and estrogen resistance.[19–21] DeGraffenried et al demonstrated that breast cancer cells with constitutively active Akt proliferate in the absence of exogenous estrogen and develop resistance to the growth inhibitory and proapoptotic effects of tamoxifen both in vitro and in vivo. Cotreatment of these cells with rapamycin derivatives restores the apoptotic responses to tamoxifen.[20]

When everolimus is combined with letrozole or 4-hydroxytamoxifen (an active metabolite of tamoxifen), there is synergistic inhibition of the proliferation of estrogen-dependent breast cancer cells and increased apoptosis.[22,23] In another study, coadministration of everolimus with letrozole and fulvestrant reversed Akt-mediated resistance to endocrine therapy and restored responsiveness to endocrine therapy in breast cancer cell lines.[24] Thus, ample preclinical data are available supporting the potential role of mTOR inhibitors in the treatment of breast cancer, particularly in endocrine-resistant tumors.

Temsirolimus

A randomized three-arm Phase II study evaluated temsirolimus, also known as CCI-779 or Torisel, in combination with

letrozole in postmenopausal women with heavily pretreated ER+ metastatic breast cancer (MBC). Patients received letrozole 2.5 mg/day alone or with temsirolimus given orally on a daily or intermittent schedule. The combination showed tolerability and there was a suggestion that progression-free survival (PFS) was longer with the addition of temsirolimus.[25]

A subsequent Phase III study evaluated first-line letrozole 2.5 mg/day plus temsirolimus 30 mg/day (5 days every 2 weeks) versus letrozole plus placebo in 1112 patients with aromatase inhibitor (AI)-naïve, hormone receptor–positive advanced breast cancer. However, the study was terminated prematurely after a planned interim analysis showed that the combination was unlikely to achieve the expected level of efficacy. There was no improvement in the primary endpoint of PFS (median, 9 months; hazard ratio (HR), 0.90; 95% CI, 0.76–1.07; $P = 0.25$).[26] Possible reasons for the failure of the study to meet its primary endpoint include patient selection and/or suboptimal dosing of the drug.

Everolimus

The rapamycin analog everolimus, also known as RAD 001 or Afinitor, is a highly specific mTOR inhibitor that also carries anti-angiogenic properties. After oral administration, everolimus is absorbed rapidly, with peak concentrations occurring at 1.3–1.8 hours after a single dose. After multiple doses, steady-state concentrations are achieved in approximately 7 days. It has a half-life of 18–35 hours. Everolimus has a predominantly hepatic clearance, and dose adjustment is not needed for renal insufficiency as only 5% of the drug is excreted in the urine.[27]

Currently, everolimus is FDA-approved for use in advanced renal cell cancer, pancreatic neuroendocrine cancers, and subependymal giant cell astrocytomas. Its most recent FDA approval was granted in July 2012 for treatment of postmenopausal women with advanced hormone receptor–positive, HER2-negative breast cancer in combination with exemestane. This last indication was granted on the basis of study results that will be discussed below.

Everolimus in the treatment of hormone receptor–positive breast cancer

On the basis of the preclinical data discussed above, a number of studies were conducted evaluating the role of everolimus in the clinical setting.

A Phase I, dose-escalating study evaluated everolimus plus letrozole in 18 postmenopausal patients with stable MBC or progression after at least 4 months of first- or second-line therapy with letrozole alone.[28] Six patients received

everolimus 5 mg/day, and 12 patients received 10 mg/day. Among these patients, one had a complete response (CR) lasting more than 22 months, and another experienced a 28% reduction in liver metastases. Both had received everolimus 10 mg/day. There was one dose-limiting toxicity, grade 3 thrombocytopenia, which occurred in a patient assigned to the higher dose. Based on the results of this study, everolimus at a daily dose of 10 mg/day was recommended for subsequent studies.

A Phase II study randomized 270 postmenopausal women with operable ER+ breast cancer to receive 4 months of neoadjuvant treatment with letrozole 2.5 mg/day plus either everolimus 10 mg/day or placebo. The primary endpoint was clinical response by palpation. The response rate (RR) in the everolimus arm was higher than that with letrozole alone (68.1% versus 59.1%). An antiproliferative response, defined by a reduction in Ki67 expression at day 15 occurred in 52 of 91 (57%) patients in the everolimus arm and in 25 of 82 (30%) patients in the placebo arm ($P < 0.01$). The authors concluded that everolimus significantly increased letrozole efficacy in the neoadjuvant treatment of ER+ breast cancer.[29]

Sabine et al characterized the effects of preoperative everolimus in primary breast cancer patients through gene expression profiling. Twenty-seven patients with ER+ breast cancer completed 11–14 days of neoadjuvant everolimus 5 mg/day. Patients whose tumors responded with significant reductions in proliferation also had significant decreases in the expression of genes involved in cell cycle and p53 signaling pathways. Overall, everolimus was noted to decrease proliferation, increase apoptosis, and reduce Akt/mTOR signaling in tumors.[30]

The Tamoxifen-RAD001 (TAMRAD) study was a Phase II study of 111 patients with ER+, HER2-negative MBC treated previously with AI therapy.[31] Patients were randomized to either tamoxifen alone or tamoxifen with everolimus 10 mg/day. The primary outcome was clinical benefit rate (CBR), defined as CR + partial response (PR) + stable disease (SD) at 6 months. Earlier AI therapy had been administered in either the adjuvant (31%) or metastatic setting (60%). Patients were stratified according to primary (49%) or secondary (51%) hormone resistance. Primary resistance was defined as disease relapse during or within 6 months of stopping adjuvant AI therapy, or disease progression within 6 months of starting AI therapy in the metastatic setting. Secondary resistance was defined as disease relapse greater than 6 months after stopping adjuvant AI therapy or progression after 6 months of AI therapy in the metastatic setting.

The CBR was 42% in the tamoxifen-alone group, compared with 61% in the everolimus plus tamoxifen group (exploratory $P = 0.045$). The time to progression (TTP) was significantly longer in the combination group (8.6 months versus 4.5 months, HR, 0.54; 95% CI, 0.36–0.81). At the time of the last update, there were 31 deaths in the tamoxifen-alone group, compared with 16 deaths in the combination group. Median overall survival (OS) had not been reached in the combination group; OS was 32.9 months in the tamoxifen group, yielding a HR for survival of 0.45 (95% CI, 0.24–0.81, exploratory $P = 0.007$), favoring the addition of everolimus.

Results from exploratory subgroup analyses suggested that the benefit from the addition of everolimus to tamoxifen may have been restricted to patients with secondary hormone resistance. The CBR among patients with secondary resistance was 74% with the combination, compared with 48% with tamoxifen alone; whereas the CBR in patients with primary resistance was only slightly better with the addition of everolimus (46%) compared with tamoxifen alone (36%). Similarly, the magnitude of improvement in TTP with the combination was greater in patients with secondary resistance (HR, 0.46; 95% CI, 0.26–0.83) than that observed in patients with primary resistance (HR, 0.70; 95% CI, 0.40–1.21). The main toxicities associated with tamoxifen plus everolimus were fatigue (72% versus 53% with tamoxifen alone), stomatitis (56% versus 7%), rash (44% versus 7%), anorexia (43% versus 18%), and diarrhea (39% versus 11%).

BOLERO-2 was a randomized Phase III trial that enrolled 724 postmenopausal women with ER+/HER2 locally advanced cancer or MBC that was refractory to letrozole or anastrozole.[32] Patients were randomized in a 2:1 ratio to receive exemestane 25 mg/day plus everolimus 10 mg/day versus exemestane plus placebo. The primary endpoint was PFS. At the time of interim analysis, the group assigned to everolimus plus exemestane demonstrated a significant improvement in PFS (6.9 months) compared with exemestane alone (2.8 months) (HR, 0.43; 95% CI, 0.35–0.54; $P < 0.001$). The benefit appeared consistent across all subgroups. Overall survival analysis is not yet mature. There were more grade 3 and 4 toxicities observed with the addition of everolimus, the most common being stomatitis (8% versus 1%), anemia (6% versus < 1%), dyspnea (4% versus 1%), hyperglycemia (4% versus < 1%), fatigue (4% versus 1%) and pneumonitis (3% versus 0%). Based on results from this study, in July 2012, the FDA approved the use of everolimus in combination with exemestane for hormone receptor–positive/HER2 advanced breast cancer in postmenopausal women after progression on a nonsteroidal AI.

Everolimus in the treatment of HER2-overexpressing breast cancer

HER2+ breast cancers, which account for 25%–30% of breast cancers, are biologically aggressive and are associated with altered response to therapy and poor clinical outcomes.[33] The development of trastuzumab, a humanized monoclonal antibody against HER2, represents a major advance in the treatment of both early-stage and advanced HER2+ breast cancer.[34–37] Nevertheless, many tumors do not respond to trastuzumab-based therapy, and even among those that do, resistance often develops.

HER2 stimulates cell proliferation and survival through the PI3K/Akt/mTOR pathway,[38] and activation of this pathway is associated with trastuzumab resistance.[15–17] Preclinical data have shown that everolimus has synergistic activity with trastuzumab and may overcome trastuzumab resistance.[39]

The efficacy of everolimus in women with HER2+ MBC has been evaluated in several Phase I–II studies. In a Phase Ib study, 33 patients who were pretreated with trastuzumab received everolimus 5 mg daily, 10 mg daily, or 30 mg weekly in combination with weekly trastuzumab and paclitaxel on days 1, 8, and 15 of a 28-day cycle.[40] Among the 27 patients with measurable disease, two patients had a CR and 10 patients had a PR, for an overall response rate (ORR) of 44%. An additional 13 patients (48%) had SD. The median PFS was 34 weeks.

In a subsequent Phase II follow-up trial, everolimus 10 mg/day in combination with paclitaxel and trastuzumab in doses similar to the Phase Ib study described above was evaluated in patients who were resistant to taxanes and trastuzumab. Among 48 patients, 19% had a PR and 62% had SD. Median PFS was 26 weeks.[41]

In another Phase Ib trial, everolimus was evaluated in patients with HER2+ MBC who had progressed after receiving trastuzumab.[42] Most patients had received taxanes and anthracyclines previously, and 24% had been pretreated with lapatinib. Patients received everolimus 5 mg/day, 20 mg/week, or 30 mg/week in combination with weekly trastuzumab and vinorelbine on days 1 and 8 of a 21-day cycle. Among 47 patients in all dosage groups, the ORR was 19% and the CBR (CR + PR + SD ≥ 24 weeks) was 54%. Median PFS was 30.7 weeks for the overall population. In the extension phase of the trial, patients were allowed to continue everolimus, and vinorelbine could be discontinued at the investigator's discretion. Two additional patients

achieved CR, one achieved PR, and the overall median PFS was 41 weeks.

A post hoc analysis was performed that pooled data from the two previously discussed Phase I[40,42] and Phase II[41] studies to evaluate the efficacy of everolimus in patients pretreated with lapatinib.[43] Among 101 patients, the ORR was 21% for those patients treated with lapatinib compared with 29% for those not treated with lapatinib. The disease control rate was 88% and 81%, respectively; overall mean PFS was 29.0 and 36.1 weeks, respectively. These data suggest that everolimus, in combination with trastuzumab and chemotherapy, has antitumor efficacy in patients with HER2+ MBC, regardless of whether they were pretreated with lapatinib.

Patients with HER2+ MBC with progressive disease after previous HER2-targeted therapy were evaluated in two Phase I–II trials; data were combined for analysis.[44] Everolimus was administered at 5 or 10 mg/day plus a trastuzumab 8 mg/kg loading dose and then 6 mg/kg every 3 weeks. Among 47 patients, 15% had PR and 19% had SD. The median PFS was 4.1 months. Results from this study suggest that everolimus may have promising activity in the absence of cytotoxic chemotherapy.

The abovementioned trials show encouraging results for everolimus use in patients with HER2+ MBC. Two ongoing Phase III studies, BOLERO-1 and BOLERO-3, will help to further define the role of everolimus in this population.[45,46] The BOLERO-1 trial is evaluating everolimus in combination with paclitaxel and trastuzumab as first-line therapy, and BOLERO-3 is evaluating everolimus in combination with vinorelbine and trastuzumab in patients with previous taxane therapy and trastuzumab resistance.

Everolimus in the treatment of triple-negative breast cancer (TNBC)

TNBCs lack the expression of ER, progesterone receptor and HER2 and comprise about 15% of all breast cancers.[47] Such tumors are associated with aggressive behavior and worse survival compared with other subtypes of breast cancer.[48] Treatment of TNBC remains a challenge, and cytotoxic chemotherapy remains the standard.

A Phase II study was conducted to evaluate the role of mTOR inhibition in combination with chemotherapy in this population.[49] Fifty patients with early-stage or locally advanced TNBC were randomized to receive neoadjuvant weekly paclitaxel 80 mg/m^2 either alone or in combination with everolimus 30 mg/week for 12 weeks followed by 5-fluorouracil, epirubicin, and cyclophosphamide every 3 weeks for four cycles. The 12-week RR by ultrasonography

was 48% when everolimus was administered in combination with paclitaxel compared with 30% when paclitaxel was used alone ($P = 0.15$). Pathologic CR rate was not significantly different between the two groups.

Another Phase II study evaluated the combination of everolimus at a dose of 5 mg daily plus carboplatin (area under the curve [AUC] = 6) given every 3 weeks in patients with metastatic TNBC.[50] The primary objective was CBR. At the time of reporting, 18 out of a planned total of 25 patients had been enrolled. One patient achieved a CR; four achieved a PR, and two SD lasting greater than 6 months. One of the patients who achieved SD had progressed on single-agent carboplatin at the time of study entry. Dose-limiting thrombocytopenia was an unexpected toxicity requiring amendment of carboplatin dosing to AUC = 4.

Due to the small sample sizes of the above studies, no clear conclusions can be drawn at this time. More studies are needed in this population to determine whether any subset exists that may benefit from the addition of everolimus.

Everolimus – toxicities and management

Safety data from the pivotal BOLERO-2 study showed that the most common adverse events (AEs) with an incidence of 30% or greater were stomatitis, infections, rash, fatigue, diarrhea, and decreased appetite. Grade 3/4 AEs with an incidence of 2% or greater included stomatitis, infections, hyperglycemia, fatigue, dyspnea, pneumonitis, and diarrhea. All these AEs occurred with greater frequency in the combination arm.[32]

Noninfectious pneumonitis is a rare but serious AE known to occur with mTOR inhibitors.[52] It should be considered in patients presenting with cough, dyspnea, hypoxia, or pleural effusions when other causes have been excluded. On computed tomography scans, pneumonitis can present as ground glass or patchy opacities or infiltrates. It appears to be immunologically mediated, with biopsies showing organizing pneumonia, granulomatous inflammation, and lymphocytic infiltration or vasculitis. Management of symptomatic pneumonitis involves cessation of therapy and the use of corticosteroids if severe. Most cases have been reversible upon drug discontinuation. A pooled analysis of five studies evaluating everolimus in breast cancer patients reported variability in the incidence of noninfectious pneumonitis. In four of the studies, the incidence was approximately 3%; however, the incidence was 35% in the fifth study.[51]

The immunosuppressive properties of everolimus may predispose patients to opportunistic infections and reactivation of previous infections including hepatitis B. Pneumonia,

Table 1 Ongoing studies with everolimus

Study	Phase	Treatment
BOLERO-1: everolimus in combination with trastuzumab and paclitaxel, as first line therapy in women with HER2+ locally advanced or metastatic breast cancer	III	Experimental arm: everolimus 10 mg daily + paclitaxel 80 mg/m^2 weekly on days 1, 8, and 15 + trastuzumab 2 mg/kg weekly on days 1, 8, 15, and 22 Control arm: placebo + paclitaxel 80 mg/m^2 weekly on days 1, 8, and 15 + trastuzumab 2 mg/kg weekly on days 1, 8, 15, and 22
BOLERO-3: daily everolimus in combination with trastuzumab and vinorelbine, in pretreated women with HER2+ locally advanced or metastatic breast cancer	III	Experimental arm: everolimus + vinorelbine + trastuzumab Control arm: placebo + vinorelbine + trastuzumab
Randomized, placebo-controlled clinical trial evaluating the use of adjuvant endocrine therapy ± 1 year of everolimus in patients with high-risk, hormone receptor–positive and HER2/neu negative breast cancer	III	Experimental arm: adjuvant endocrine therapy × 2–5 years + everolimus daily × 1 year Control arm: adjuvant endocrine therapy × 2–5 years + placebo × 1 year
GeparQuinto – integration of bevacizumab, everolimus, and lapatinib into current neoadjuvant chemotherapy regimens for primary breast cancer	III	Arm 1: epirubicin – cyclophosphamide/docetaxel Arm 2: epirubicin – cyclophosphamide/docetaxel + bevacizumab Arm 3: paclitaxel Arm 4: paclitaxel + everolimus Arm 5: epirubicin – cyclophosphamide/docetaxel + trastuzumab Arm 6: epirubicin – cyclophosphamide/docetaxel + lapatinib
Lapatinib in combination with everolimus in triple negative metastatic or locally advanced breast cancer	II	Lapatinib 1250 mg daily + everolimus 5 mg daily
Randomized study to compare vinorelbine in combination with everolimus vs vinorelbine monotherapy for second-line treatment in advanced breast cancer (VicTORia)	II	Experimental arm: vinorelbine 25 mg/m^2 days 1, 8, and 15 + everolimus 5 mg daily Control arm: vinorelbine 25 mg/m^2 days 1, 8, and 15
Trastuzumab or everolimus in hormone-refractory metastatic breast cancer	II	Arm A: trastuzumab every 3 weeks + endocrine therapy (everolimus added at progression) Arm B: everolimus 10 mg daily + endocrine therapy (trastuzumab added at progression)
Combined fulvestrant (Faslodex) and RAD001 in advanced/metastatic breast cancer after aromatase inhibitor failure	II	Everolimus 10 mg daily + fulvestrant 500 mg day 1, then 250 mg days 14 and 28, then 250 mg every 4 weeks
Value of adding RAD001 to trastuzumab as preoperative therapy of HER2+ primary breast cancer amenable to surgery	II	Experimental arm: weekly trastuzumab + everolimus daily × 6 weeks Control arm: weekly trastuzumab × 6 weeks
Weekly abraxane and RAD001 in women with locally advanced or metastatic breast cancer	I/II	Everolimus daily + paclitaxel albumin-stabilized nanoparticle formulation days 1, 8, and 15
Neo-adjuvant study of cisplatin, paclitaxel with or without RAD001 in patients with triple-negative locally advanced breast cancer	II	Experimental arm: cisplatin 25 mg/m^2 weekly + everolimus 5 mg daily × 1 week followed by cisplatin 25 mg/m^2 + paclitaxel 80 mg/m^2 weekly + everolimus 5 mg daily × 11 weeks Control arm: cisplatin 25 mg/m^2 IV weekly + placebo daily × 1 week followed by cisplatin 25 mg/m^2 + paclitaxel 80 mg/m^2 IV weekly + placebo daily × 11 weeks
The efficacy and tolerability of everolimus in combination with trastuzumab and vinorelbine in the treatment of progressive HER2+ breast cancer brain metastases	II	Everolimus 5 mg daily + plus vinorelbine 25 mg/m^2 days 1, 8, and 15 + weekly trastuzumab
Lapatinib and RAD001 for HER2+ metastatic breast cancer	II	Lapatinib 1250 mg daily + everolimus 5 mg daily
GCC 0901 – letrozole in combination with lapatinib followed by an addition of everolimus in postmenopausal women with advanced endocrine resistant breast cancer	II	Letrozole 2.5 mg daily + lapatinib 1500 mg daily Everolimus 5 mg daily added upon progression (with reduction of lapatinib to 1250 mg daily)
Everolimus in invasive breast cancer patients after preoperative use of anthracycline and/or taxane-based chemotherapy	II	Experimental arm: everolimus 10 mg daily × 3 weeks 1 week after completion of neoadjuvant treatment and before surgery Control arm: no intervention

Abbreviations: HER2, human epidermal growth factor receptor-2; IV, intravenously.

mycobacterial infections, other bacterial infections, and invasive fungal infections have been noted in those treated with everolimus.[52]

Everolimus is also associated with metabolic derangements, notably hyperglycemia, hypercholesterolemia, and hypertriglyceridemia.[52] Optimal glycemic and lipid control should be achieved before starting everolimus, and serum glucose and lipid levels should be checked at baseline and periodically during treatment. These abnormalities can be treated using standard guidelines for diabetes and hyperlipidemia.

Everolimus – drug interactions

Everolimus is a substrate of CYP3A4 and P-glycoprotein. Coadministration with ketoconazole, a strong CYP3A4 inhibitor, results in increased everolimus blood concentrations, with resultant maximum concentration (C_{max}) and AUC 3.9-fold and 15-fold higher, respectively. It is recommended that concurrent administration of strong CYP3A4 inhibitors be avoided. If coadministration with a moderate inhibitor is necessary, it is suggested that the dose of everolimus be reduced to avoid excess toxicity. Conversely, coadministration with the strong CYP3A4 inducer rifampin decreases C_{max} and AUC of everolimus by 58% and 63%, respectively. If concurrent use of a strong inducer is necessary, it is suggested that the dose of everolimus be increased.[52]

Predictive biomarkers

In this era of targeted therapy it is crucial to be able to identify subsets of patients who are either likely or unlikely to respond to a given drug. Such identification would maximize efficacy and minimize unnecessary toxicity. Currently, no reliable biomarkers exist to predict response to treatment with everolimus, although some molecular patterns are emerging as potential predictors of sensitivity and/or resistance.

Di Nicolantonio et al performed a series of elegant experiments to assess the sensitivity to everolimus of various cancer cell lines harboring mutations in *PIK3CA* or *PTEN*.[53] They found that the tumors could be divided into two groups based on response to everolimus: those resistant to treatment harbored both a mutation in the PI3K pathway as well as a mutation in either *KRAS* or *BRAF*. On the other hand, those that were sensitive to everolimus had only a mutation in the PI3K pathway. These results suggest that treatment with an mTOR inhibitor may be ineffective against *KRAS* or *BRAF* mutant tumors despite the presence of PI3K/AKT/mTOR pathway activation.

This hypothesis was tested by Janku et al in a study evaluating response in 25 patients with breast or gynecologic tumors harboring the *PIK3CA* mutation, 23 of whom were treated on a protocol that included a PI3K/AKT/mTOR pathway inhibitor.[54] Two (9%) of the 23 patients had SD for more than 6 months, and seven patients (30%) had a PR. Seven patients with *PIK3CA* mutations had coexisting mutations in *KRAS*, *NRAS*, or *BRAF*, and two of these patients (ovarian cancer) achieved a response. Although the numbers are small, these findings suggest that not all patients with simultaneous mutations in both pathways demonstrate resistance.

Chen et al found that expression of the cell cycle regulator p27 correlated with the anticancer activity of rapamycin and temsirolimus in breast cancer cells in vitro and in vivo. Cells expressing high levels of p27 were sensitive to treatment, whereas those with low expression demonstrated resistance. Moreover, they observed consistently that downregulation of p27 by silencing RNA rendered cells with normally high levels of expression resistant to treatment. They propose that p27 expression levels might serve as a predictive biomarker for patient selection for rapalog-based therapy.[55]

Conclusion

The mTOR pathway is pivotal to the pathogenesis of many cancers, including breast cancer. Dysregulation of this pathway is associated with resistance both to endocrine and to HER2-directed therapies. The mTOR inhibitor everolimus has emerged as a promising agent in the treatment of several cancers and is now approved in combination with exemestane in postmenopausal hormone receptor–positive advanced breast cancer.

Everolimus carries a unique side-effect profile, which clinicians and patients should be aware of. With proper pretreatment planning and careful monitoring, treatment-related toxicities are generally manageable and the drug well tolerated.

Studies are ongoing to further define the role of everolimus in various subtypes and stages of breast cancer (see Table 1). Identification of biomarkers capable of predicting sensitivity or resistance to mTOR inhibition in breast cancer remains an unmet need. As our understanding of the molecular profiles of tumors improves, we will be able to develop increasingly refined targeted agents as well as appropriate selection criteria that will lead to improved outcomes with minimal toxicities.

Disclosure

The authors disclose no potential conflicts of interest.

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