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Original Article

Mammalian target of rapamycin (mTOR) inhibitors and combined chemotherapy in breast cancer: a meta-analysis of randomized controlled trials

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Abstract: The mammalian target of rapamycin (mTOR) inhibitor, in combination with other chemotherapeutic drugs, has been used for treatment of breast cancer that develops resistance to endocrine therapy. However, the efficacy and safety need further evaluation. Here, we report a meta-analysis of randomized controlled trials (RCT) in breast cancer patients undergoing chemotherapy using steroid (exemestane) or nonsteroid (letrozole) aromatase inhibitors with or without mTOR inhibitors (everolimus). The overall response rate (ORR), progression-free survival (PFS), clinical benefit rate with 95% confidence interval (CI), and the major toxicities/adverse effects were analyzed. Data were extracted from twelve studies that meet the selection criteria. Among these, six studies that enrolled 3693 women received treatment of everolimus plus exemestane, or placebo with exemestane. The results showed that everolimus plus exemestane significantly increased the ORR relative risk (relative risk = 9.18, 95% CI = 5.21-16.15), PFS hazard ratio (hazard ratio = 0.44, 95% CI = 0.41-0.48), and clinical benefit rate (relative risk = 1.92, 95% CI 1.69-2.17) compared to placebo control, while the risks of stomatitis, rash, hyperglycemia, diarrhea, fatigue, anorexia and pneumonitis also increased. Three studies that enrolled 715 women who received everolimus as neoadjuvant therapy were analyzed. Compared to chemotherapy with placebo, chemotherapy plus everolimus did not increase the ORR relative risk (relative risk = 0.90, 95% CI = 0.77-1.05). Meanwhile, two other studies that enrolled 2104 women examined the efficacy of temsirolimus (or placebo control) plus letrozole. The results indicated that emsirolimus plus letrozole did not increase the ORR relative risk and clinical benefit rate ($p > 0.05$). Together, these data suggest that the combined mTOR inhibitor (everolimus) plus endocrine therapy (exemestane) is superior to endocrine therapy alone. As a neoadjuvant, everolimus did not increase the ORR, while temsirolimus plus letrozole treatment has limited effect on the ORR and the CBR of breast cancer patients.

Keywords: mTOR inhibitors, breast cancer, meta-analysis, aromatase inhibitors, chemotherapy

Introduction

Mammalian target of rapamycin (mTOR) is a serine/threonine kinase that integrates multiple signals from growth factors and hormones. mTOR pathway is known to play essential roles in regulating cell growth and proliferation [1, 2]. Correspondingly, mTOR inhibitors have been extensively studied and used for treatment of cancer, including breast cancer [3, 4]. Breast cancer is one of the most common cancers with greater than 1,300,000 cases and greater than 450,000 deaths each year worldwide [5]. Clinically, target therapies in patients with early-stage breast cancer using estrogen receptor (ER) and human epidermal growth factor receptor antagonists have markedly reduced tumor recurrence and death [6-8]. However, a subset of hormone receptor-positive breast cancers do not benefit from above endocrine therapies and almost all hormone receptor-positive metastatic breast cancer patients ultimately develop resistance to hormonal therapies [9]. This largely hindered the efficacy of endocrine therapy treatment on breast cancer.

Although not very clear, one potential mechanism of the treatment resistance in breast cancer is attributed to the phosphatidyl inositol 3-kinase (PI3K) pathway, which is the most fre-

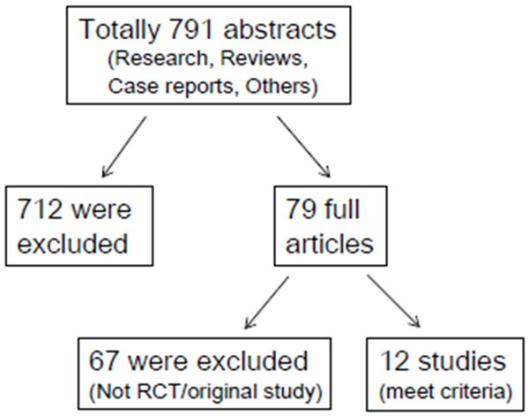

Figure 1. Illustrated is an outline of the search-flow diagram. Among the 79 full-length research articles, 12 studies meet the selection criteria and were subjected to analysis.

quently altered pathway in breast cancer [10]. In addition, PI3K activation has been associated with hormone therapy resistance in preclinical studies. This raises a possibility of targeting p13K pathway so as to reduce resistance. Notably, mTOR is a signal transduction kinase in the PI3K pathway containing two complexes mTORC1 and mTORC2. Both mTORC1 and mTORC2 are associated with AKT, a downstream target of PI3K that enhances cell proliferation, survival, and angiogenesis [10]. Therefore, inhibition of mTOR pathway (such as using the selective mTOR inhibitor, Everolimus) in breast cancer may restore its sensitivity to endocrine therapy [11, 12]. Rapalogs, agents that primarily inhibit mTORC1, have been used in combination with endocrine therapy to overcome endocrine resistance [4]. mTOR has been demonstrated as a crucial mediator of tumor progression and a promising target in a significant proportion of breast cancer patients [13]. Specifically, everolimus and temsrrolimus have been the major mTOR inhibitors that in combination with aromatase inhibitors as the therapeutic agents for breast cancer patients [13], while some clinical trials used steroidal aromatase inhibitors (such as exemestane) or nonsteroidal aromatase inhibitors (such as letrozole), instead.

To evaluate the clinical effectiveness of mTOR inhibitors in breast cancer therapeutics, we performed a meta-analysis to determine the efficacy and safety of mTOR inhibitors combined chemotherapy (with steroidal or nonsteroidal aromatase inhibitors) in treatment of breast cancer, such that more reliable and evidence-based medicine to guide clinical practice may be achieved.

Methods

Search strategy

Eligible articles were identified by searching the MEDLINE bibliographical database for the period up to the end of 2013. The search strategy included the following keywords: (breast AND (neoplasms OR neoplasm OR cancer OR cancers OR carcinoma OR carcinomas) AND (mTOR AND inhibitor) OR BEZ235 OR NVP-BEZ235 OR everolimus OR RAD001 OR rapamycin OR sirolimus OR PI-103 OR temsirolimus OR torisel OR AZD8055 OR Ku-0063794 OR PF-04691502 OR CH5132799 OR GDC-0980 OR RG7422 OR WAY-600 OR WYE-125132 OR WYE-687 OR GSK2126458 OR PKI-587 OR PP-121 OR OSI-027 OR “palomid 529” OR P529 OR PP242 OR XL765 OR GSK1059615 OR WYE-354 OR deforolimus OR ridaforolimus) [13]. To obtain more information that may be missed by above methods, we performed Google academic search on the internet to track relevant references.

Inclusion criteria

Any randomized clinical trials evaluating the therapeutic efficacy of the mTOR inhibitors everolimus, temsrrolimus, or sirolimus for the treatment of breast cancer were included. All these clinical trials include placebo control treatment. Conference abstracts were included if they provided sufficient data to assess the quality of the studies or the results were described in detail. Non-randomized trials, pharmacokinetic studies, animal studies, and laboratory studies were excluded from this study. Only studies with institutional review board approval and appropriate consent were included [14].

Data extraction

Data extraction was conducted using a standardized form. Two reviewers independently examined the titles and abstracts of the studies identified by the searching criteria and then performed initial screening of the abstracts to exclude obviously ineligible studies. Full texts of all relevant and eligible studies were reviewed [14]. Disagreements were resolved by discussion with an independent expert. The following information was extracted from each paper

Table 1. Summary of everolimus plus endocrine therapy in HR^+, $HER2^-$ advanced breast cancer (6 studies)

Author/phase	Patients	N	Chemotherapy regimens	Efficacy
Mario Campone et al., 2013/BOLERO-2	with HR^+, $HER2^-$ visceral metastases	271	Everolimus + exemestane	PFS: 6.8 vs 2.8 months HR: 0.47; 95% CI 0.37-0.60
		135	Placebo + exemestane	CBR: 44.6% vs 22.2%
	without visceral metastases	214	Everolimus + exemestane	PFS: 9.9 vs 4.2 months; HR: 0.41; 95% CI 0.31-0.55;
		104	Placebo + exemestane	CBR: 59.8% vs 31.7%
José Baselga, M.D et al., 2012/BOLERO-2	Postmenopausal advanced BC	485	Exemestane + everolimus	PFS: 6.9 vs 2.8 months HR: 0.43; 95% CI: 0.35-0.54
		239	Exemestane + placebo	ORR: 9.5% vs 0.4%
G. N. Hortobagyi et al., 2011/BOLERO-2	Postmenopausal advanced BC	485	Exemestane + everolimus	PFS: 7.4 vs 3.2 months HR: 0.44; 95% CI: 0.36-0.53
		239	Exemestane + placebo	ORR: 12.0% vs 1.3% CBR: 50.5% vs 25.5%
Shinzaburo Noguchi et al., 2013/BOLERO-2	metastatic Asian	98	Exemestane+everolimus	PFS: 8.48 vs 4.14 months HR: 0.62; 95% CI 0.41-0.94 CBR: 58.2 vs 28.9% ORR: 19.4% vs 0
		45	Exemestane + placebo	
	Non-Asian	387	Exemestane + everolimus	PFS: 7.33 vs 2.83 months HR: 0.41; 95% CI, 0.33-0.50
		194	Exemestane + placebo	CBR: 49.6% vs 25.8% ORR: 10.9% vs 2.1%
Novartis Pharmaceuticals Corporation/BOLERO-2	HR^+, $HER2^-$ metastatic	485	Exemestane+everolimus	PFS: 7.8 vs 3.2 months HR: 0.45; ORR: 12.6% vs 1.7%
		239	Exemestane + placebo	
Thomas Bachelot et al., 2012/Phase II	HR^+, $HER2^-$ metastatic	54	Tamoxifen + everolimus	PFS: 8.6 vs 4.5 months HR: 0.54; 95% CI, 0.36-0.81 CBR: 61% vs 42% ORR: 14% vs 13%
		57	Tamoxifen	

(although data from some of the papers may not be intact): the trial's name, first author, year of publication, journal, number of patients in both groups, age, hazard ratios (HR) for PFS and their 95% confidence intervals (CI), overall response rate, and clinical benefit rate. Data on adverse effects/toxicities, including stomatitis, rash, hyperglycemia, diarrhea, fatigue, anorexia and pneumonitis were also collected.

Quality assessment

An open assessment of the trials was performed using the methods reported by Jadad and his colleagues [15]. The assessment recorded and scored the trials based on answers to following three questions: 1) whether the trial reported an appropriate randomization method (score 0-2); 2) whether the trials reported an appropriate blind method (score 0-2); and 3) whether the trial reported withdrawals or dropouts (score 0-1). The quality scale ranged from 0 to 5 points. A score equal or less than 2 indicates low quality, while a score equal or more than 3 indicates high quality [16].

Statistical analysis

Hazard ratios (HR) for PFS and OS, relative risk (RR) for overall response to treatment, clinical benefit rate (CBR) and different types of toxicity

Table 2. Summary of everolimus plus endocrine therapy in HR^+, $HER2^-$ advanced breast cancer (6 studies)

Author/phase	Patients	Chemotherapy regimens	Most common toxic effects
Mario Campone et al 2013/BOLERO-2	Postmenopausal women with HR^+, $HER2^-$ visceral metastases	Everolimus + exemestane	Stomatitis (59%) Rash (40%) Nausea (33%) Pneumonitis (14%) Diarrhea (34%) Fatigue (40%)
		Placebo + exemestane	Stomatitis (13%) Rash (4%) Nausea (29%) Pneumonitis (0%) Diarrhea (16%) Fatigue (29%)
	without visceral metastases	Everolimus + exemestane	Stomatitis (59%) Rash (39%) Nausea (27%) Pneumonitis (19%) Diarrhea (34%) Fatigue (36%)
		Placebo + exemestane	Stomatitis (10%) Rash (10%) Nausea (28%) Pneumonitis (0%) Diarrhea (22%) Fatigue (25%)
José Baselga, M.D et al 2012/BOLERO-2	Postmenopausal ER^+ advanced BC	Exemestane + everolimus	Stomatitis (56%) Rash (36%) Nausea (27%) Pneumonitis (12%) Diarrhea (30%) Fatigue (33%)
		Exemestane + placebo	Stomatitis (11%) Rash (6%) Nausea (27%) Pneumonitis (0%) Diarrhea (16%)
G. N. Hortobagyi et al 2011/BOLERO-2	Postmenopausal ER^+ advanced BC	Exemestane + everolimus	Stomatitis (59%) Rash (39%) Nausea (27%) Interstitial lung disease (15%) Diarrhea (33%)
		Exemestane + placebo	Stomatitis (11%) Rash (6%) Nausea (28%) Diarrhea (19%) Interstitial lung disease (0%)
Shinzaburo Noguchi et al 2013/BOLERO-2	metastatic advanced Asian	Exemestane + everolimus	Stomatitis (80%) Rash (40%) Nausea (22%) Interstitial lung disease (23%) Diarrhea (23%)
		Exemestane + placebo	Stomatitis (15%) Rash (9%) Nausea (24%) Decreased appetite (8%) Diarrhea (13%)
	Non-Asian	Exemestane + everolimus	Stomatitis (54%) Rash (37%) Nausea (32%) Interstitial lung disease (14%) Diarrhea (37%)
		Exemestane + placebo	Stomatitis (11%) Rash (6%) Nausea (30%) Decreased appetite (14%) Diarrhea (20%)
Novartis Pharmaceuticals Corporation/BOLERO-2	HR^+, $HER2^-$, metastatic advanced	Exemestane + everolimus	Stomatitis (67%) Rash (39%) Nausea (29%) Interstitial lung disease (19%) Diarrhea (33%)
		Exemestane + placebo	Stomatitis (11%) Rash (6%) Nausea (28%) Interstitial lung disease (0.4%) Diarrhea (18%)
Thomas Bachelot et al 2012/Phase II	HR^+, $HER2^-$, metastatic advanced	Tamoxifen + everolimus	Stomatitis (56%) Rash (44%) Nausea (35%) Interstitial lung disease (17%) Diarrhea (39%)
		Tamoxifen	Stomatitis (7%) Rash (7%) Nausea (35%) Decreased appetite (30%)

were calculated and generally compared between two groups: chemotherapy plus either mTOR inhibitor (s) or placebo controls. A statistical test with a p value < 0.05 was considered to be significant. The values of HR, OR, and RR > 1 reflect more progression or deaths, more overall response, and more toxicities in the chemotherapy plus mTOR inhibitors group respectively. To investigate statistical heterogeneity among the different trials, the standard chi-squared (χ^2 Q) test was applied ($p < 0.10$ indicated meaningful differences between studies). The results were generated using a fixed-effect model. A random-effect model was employed when there was evidence of statistically significant heterogeneity, which generates a more conservative estimate. All CI had two-sided probability coverage of 95%. An estimate of potential publication bias was carried out using the funnel plot. An asymmetric plot suggested a possible publication bias. We used a forest plot to analyze and to display the results. All calculations were accomplished using the Review Manager 5 software.

Results

Selection of the twelve clinical trial studies

Using above searching strategy, we retrieved 791 articles which include 761 articles from

Table 3. Summary of combined everolimus with neoadjuvant chemotherapy in breast cancer (3 studies)

Author/phase	N	Chemotherapy regimens	RR	Grades 3 to 4
Baselga et al 2009/Phase II	138	Everolimus + Letrozole	36.2%	Stomatitis (2.2%) Rash (0.7%) Asthenia (0%) Hypercholesterolemia (0.7%) Fatigue (1.5%) Anorexia (0%) Hyperglycemia (5.1%)
	132	Letrozole	39.4%	Stomatitis (0%) Rash (0%) Asthenia (0.8%) Hypercholesterolemia (0%) Fatigue (0%) Anorexia (0%) Hyperglycemia (0%)
Jens Huober et al 2013/Phase II	197	Paclitaxel + everolimus	52.2%	Anaemia (1%) Leukopaenia (12.3%) Neutropenia (17.8%) Nausea (0%) Diarrhoea (1%)
	198	Paclitaxel	61.7%	Anaemia (1.5%) Leukopaenia (7.7%) Neutropenia (9.5%) Nausea (0%) Diarrhoea (0.5%)
Gonzalez et al 2011/Phase II	23	Paclitaxel + everolimus-5FU + epirubicin + cyclophosphamide	47.8%	Anemia (13%) Leukopenia (17%) Rash/Desquamation (9%) Vomiting (13%)
	27	Paclitaxel-5FU + epirubicin + cyclophosphamide	29.6%	Anemia (4%) Leukopenia (11%) Rash/Desquamation (7%) Vomiting (4%)

Table 4. summary of temsirolimus plus letrozole in postmenopausal breast cancer (2 studies)

Author/phase	N	Chemotherapy regimens	Efficacy	Gradt 3 and 4 toxicities
Antonio C. Wolff et al., 2013/Phase III	556	Letrozole + Temsirolimus	PFS: 8.9 vs 9.0 months HR: 0.90	Asthenia (3%) Diarrhea (2%) Rash (1%) Fever (1%) Pruritus (1%) Stomatitis (1%) Nausea (1%) Anorexia (1%) Hyperlipemia (2%) Anemia (1%) Dyspnea (3%)
	556	Letrozole + Placebo		Asthenia (2%) Diarrhea (1%) Rash (< 0.5%) Fever (1%) Pruritus (0) Stomatitis (< 0.5%) Nausea (1%) Anorexia (1%) Hyperlipemia (< 0.5%) Anemia (1%) Dyspnea (3%)
Chow, et al., 2006/Phase III	493	Letrozole + Temsirolimus	PFS: 9.2 vs 9.2 months ORR: 24% vs 24% CBR: 40% vs 43%	grade 3-5 AEs neutropenia (3%) hyperglycemia (4%) dyspnea (3%),
	499	Letrozole + Placebo		dyspnea (3%) asthenia (1%)

MEDLINE bibliographical database and 30 articles from Google academic. 712 papers were excluded as they were neither RCTs, nor original studies. Studies that involved neither of our target drugs were also excluded. The remaining 79 articles were further reviewed and only 12 articles met our inclusion criteria. The searching and selection process is outlined in **Figure 1**. Among these 12 articles, 6 studies evaluated everolimus plus endocrine therapy [17-21, 31], including 5 studies that described the results of phase III trials, while the remaining one study described the results of phase II trials. All these studies were conducted on postmenopausal women with advanced breast cancer who are hormone receptor (HR) positive and human epidermal growth factor receptor-2 (HER2) negative. 3 other studies evaluated everolimus in combination with neoadjuvant chemotherapy [22, 23, 32]. There were 2 studies that evaluated temsirolimus plus letrozole [24, 25], while the last one was a phase II study about sirolimus that were conducted in patients with metastatic breast cancer [26]. Detailed

Table 5. Quality of reports of 12 clinical trials using the Jadad assessment scale

Trials	Randomization (range: 0-2)	Blindness (range: 0-2)	Withdrawals/dropouts (range: 0-1)	Total score (range: 0-5)
Campone 2013	1	2	1	4
Baselga 2012	1	2	0	3
Hortobagyi 2011	1	0	0	1
Noguchi 2013	1	2	1	4
Novartis 2012	1	2	0	3
Bachelot 2012	1	0	1	2
Huober 2013	1	0	1	2
Gonzalez 2011	1	0	0	1
Baselga 2009	1	2	1	4
Antonio 2013	1	2	1	4
Chow 2006	1	0	0	1
Bhattacharyya 2011	1	0	0	1

information about these studies is provided in **Tables 1-4**. The quality of the methods used in these studies were also assessed by the Jaded score system (**Table 5**).

Efficacy and safety comparison between endocrine therapy with everolimus and with placebo

To determine the efficacy and safety of endocrine therapy (exemestane) in combination with mTOR inhibitor (everolimus), we measured the progression-free survival (PFS), clinical benefit rate (CBR), and overall response (OR) and compared to the same endocrine therapy with placebo. The results showed that Everolimus plus exemestane significantly increased PFS (pooled HR for PFS, HR 0.44, 95% CI 0.41-0.48, **Figure 2A**), CBR (pooled RR for CBR, RR 1.92, 95% CI 1.69-2.17, **Figure 2B**), and ORR (pooled RR for ORR, RR 9.18, 95% CI 5.21-16.15, **Figure 2C**). There was no significant heterogeneity both for PFS (p = 0.71) and for CBR (p = 0.8). The heterogeneity of ORR was significant (p = 0.009) using the STATA version 11.0 sensitivity analysis (**Figure 2D**). However, when one of the study, which is out of range [21], was removed from the analysis, the heterogeneity showed no significance (p = 0.699). All the pooled HR for PFS, the pooled RR for CBR, and the pooled RR for overall response were performed using the fixed-effect model.

Adverse events

The meta-analyses of any adverse events showed that the risks of stomatitis (5.44, 95% CI, 4.63-6.38), rash (6.30, 95% CI, 5.04-7.86), hyperglycemia (6.68, 95% CI, 4.48-9.96), diarrhea (1.85, 95% CI, 1.62-2.11), fatigue (1.34, 95% CI, 1.21-1.49), anorexia (2.47, 95% CI, 2.11-2.89) and pneumonitis (47.36, 95% CI, 17.74-126.39) were higher in patients treated with everolimus plus exemestane than in those treated with placebo plus endocrine therapy. The risk of nausea was comparable between two groups (OR = 1.05, 95% CI = 0.94-1.16, **Table 6**). We also analyzed the Grade 3 or 4 adverse events and the result showed that the risks of stomatitis (9.28, 95% CI, 4.77-18.08), rash (6.07, 95% CI, 1.65-22.39), hyperglycemia (8.38, 95% CI, 3.82-18.39), diarrhea (3.34, 95% CI, 1.63-6.86), nausea (2.43, 95% CI, 1.26-4.71), fatigue (4.03, 95% CI, 2.13-7.62), and pneumonia (13.34, 95% CI, 3.79-46.91) were higher in patients receiving everolimus plus exemestane than in those receiving placebo plus exemestane. The risk of anorexia were comparable between two groups (1.91 95% CI, 0.89-4.09, **Table 7**).

The efficacy of everolimus and other mTOR inhibitors

We have analyzed the efficacy of everolimus in combination with neoadjuvant chemotherapy. The result showed that the pooled RR for ORR of everolimus in combination with neoadjuvant chemotherapy significantly improved the ORR (RR = 0.9, 95% CI = 0.77-1.05; **Figure 3A**), while there was no significant heterogeneity (p = 0.22). We also analyzed the efficacy of endocrine therapy by adding temsirolimus to letro-

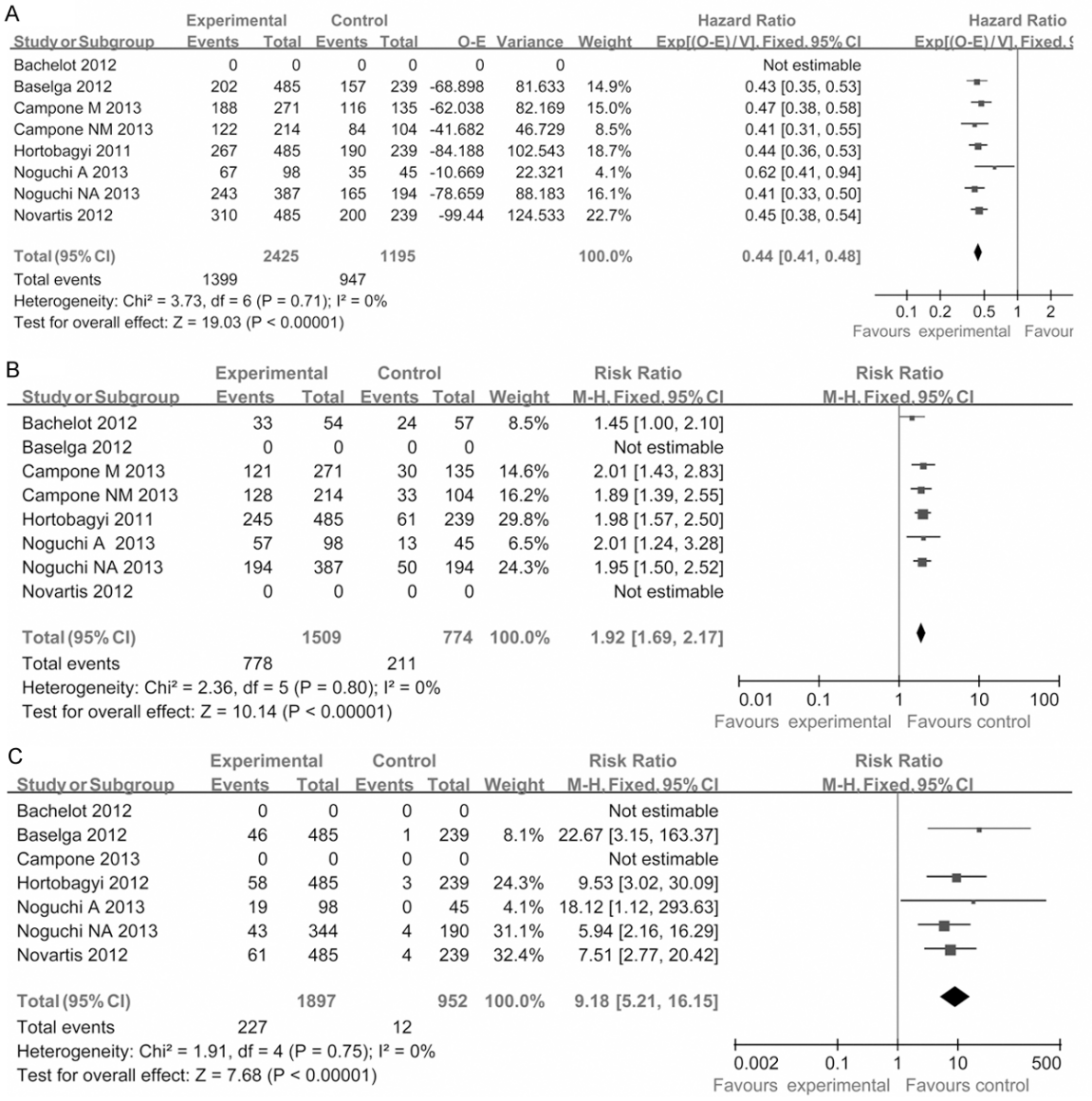

Figure 2. Forest plot of 6 studies comparing everolimus plus endocrine therapy and placebo (control) plus endocrine therapy. Everolimus plus exemestane significantly increased A: PFS (pooled HR for PFS, HR 0.44, 95% CI 0.41-0.48, p < 0.00001); B: CBR (pooled RR for CBR, RR 1.92, 95% CI 1.69-2.17, p < 0.00001), and C: ORR (pooled RR for ORR, RR 9.18, 95% CI 5.21-16.15, p < 0.00001). There was no significant heterogeneity for PFS (p = 0.71) and for CBR (p = 0.8), and RR (p = 0.75). C: The heterogeneity of ORR was significant (p = 0.009) using the STATA version 11.0 sensitivity analysis.

zole in postmenopausal breast cancer. The result suggested that the pooled RR for ORR of temsirolimus plus letrozole did not improve the ORR (RR = 1.0, 95% CI = 0.86-1.15; **Figure 3B**).

The heterogeneity did not show significant difference either (p = 0.97). Meanwhile, analysis of the pooled RR for CBR showed that temsirolimus plus letrozole did not improved the CBR

Table 6. Meta-analysis of any adverse events

	Heterogeneity I^2 (%)	analysis p	Effects model	RR (95%CI)	Z	P
stomatitis	0	0.93	Fixed	5.44 [4.63,6.38]	20.7	P < 0.00001
rash	0	0.81	Fixed	6.30 [5.04,7.86]	16.22	P < 0.00001
hyperglycemia	0	0.84	Fixed	6.68 [4.48, 9.96]	9.31	P < 0.00001
diarrhea	0	0.77	Fixed	1.85 [1.62, 2.11]	9.15	P < 0.00001
nausea	0	0.99	Fixed	1.05 [0.94, 1.16]	0.82	P = 0.41
fatigue	0	1.00	Fixed	1.34 [1.21, 1.49]	5.64	P < 0.00001
anorexia	0	1.00	Fixed	2.47 [2.11, 2.89]	11.17	P < 0.00001
pneumonitis	0	1.00	Fixed	47.36 [17.74,126.39]	7.7	P < 0.00001

Table 7. Meta-analysis of grade 3 or 4 adverse events

	Heterogeneity I^2 (%)	analysis p	Effects model	RR (95% CI)	Z	P
stomatitis	0	1.00	Fixed	9.28 [4.77, 18.08]	6.55	P < 0.00001
rash	0	1.00	Fixed	6.07 [1.65, 22.39]	2.71	P = 0.007
hyperglycemia	0	0.55	Fixed	8.38 [3.82, 18.39]	5.30	P < 0.00001
diarrhea	0	1.00	Fixed	3.34 [1.63, 6.86]	3.29	P = 0.001
nausea	0	0.53	Fixed	2.43 [1.26, 4.71]	2.64	P = 0.008
Fatigue e	0	0.85	Fixed	4.03 [2.13, 7.62]	4.29	P < 0.0001
anorexia	0	0.86	Fixed	1.91 [0.89, 4.09]	1.66	P = 0.1
pneumonitis	0	0.85	Fixed	13.34 [3.79, 46.91]	4.04	P < 0.0001

(RR = 0.94, 95% CI = 0.86-1.04; **Figure 3C**), while there was no significant heterogeneity (p = 0.75). All the pooled RR for ORR and CBR were performed using the fixed-effect model.

Discussion

Inconsistence of combined endocrine therapy in breast cancer

Given the essential function of mTOR signaling pathway in cell growth and survival, mTOR inhibitors have recently drawn attention from clinical oncologists in the field of cancer treatment. As to breast cancer, endocrine therapy has been widely used and is generally effective for its treatment. However, patients usually develop resistance over a long period of hormonal therapy, which may be overcome by alternative therapeutic strategy, or by addition of other therapeutic drugs, such as mTOR inhibitors [4]. In recent years, multiple phase III clinical trials using endocrine therapy plus mTOR inhibitors have been conducted in breast cancer patients and the efficacy compared to endocrine therapy alone. Notably, the results were very inconsistent. Dr. Baselga and his colleagues have shown that the median PFS difference is 4.1 months with 6.9 months in the everolimus plus exemestane group versus 2.8 months in the placebo plus exemestane group [17]. Similar results were obtained in another study conducted by the same group: the difference of PFS is 4 months with 6.8 months and 2.8 months in the everolimus plus exemestane and the placebo plus exemestane groups respectively [18]. Surprisingly, a phase III trial using a different combination of hormone/kinase inhibitors obtained a quiet different result: the median PFS difference is only 0.1 month with 8.9 months in the Letrozole plus Temsirolimus group versus 9.0 months in Letrozole plus Placebo group [25]. It has also been reported that the response rate by clinical palpation in the everolimus arm was higher than that with letrozole alone (68.1% v 59.1%) [22]. These results demonstrated the inconsistency of these clinical trials using combined endocrine therapy with mTOR inhibitors and motivated current meta-analysis.

Everolimus in treatment of breast cancer

Compared to placebo plus endocrine therapy, everolimus plus exemestane, a steroidal aromatase inhibitor, significantly increased the ORR [relative risk (RR) 9.18], PFS [hazard ratio (HR) 0.44], CBR [relative risk (RR) 1.92], and prolonged PFS by 4 months in breast cancer. The quality of life as measured by time to 5%

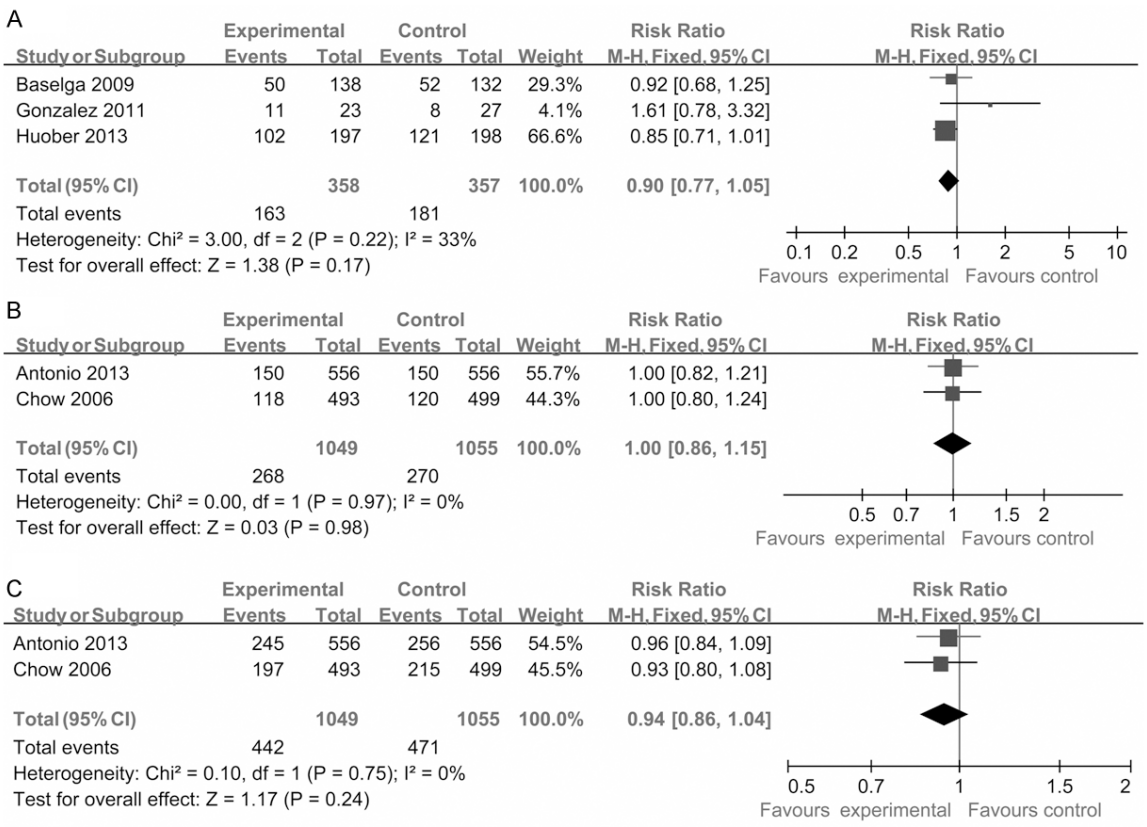

Figure 3. A: Forest plot of everolimus in combination with neoadjuvant chemotherapy for ORR; the pooled RR significantly improved the ORR (RR = 0.9, 95% CI = 0.77-1.05, Right); B: Forest plot of ORR of temsirolimus plus letrozole did not improve the ORR (RR = 1.0, 95% CI = 0.86-1.15, p =); C: Forest plot of CBR of temsirolimus plus letrozole. The heterogeneity did not show significant difference either (p = 0.97).

deterioration in global health status was also maintained in patients receiving everolimus [17]. Although the risks of stomatitis, rash, hyperglycemia, diarrhea, fatigue, and pneumonitis were higher in patients receiving everolimus plus endocrine therapy compared to placebo plus endocrine therapy, the everolimus combination still provides an alternative treatment with less toxicity compared to most of the cytotoxic chemotherapy [17]. The above toxic effects did influence the practical use of this class of drugs and compliance [27]. However, selected adverse effects associated with the use of everolimus for the treatment of metastatic renal cell carcinoma (mRCC) have been well-discussed and managed by a group of expert physicians and nurses [28]. This will provide a reference for the use of everolimus in treatment of breast cancer.

Everolimus and neoadjuvant chemotherapy

Neoadjuvant chemotherapy plus everolimus did not increase the ORR of breast cancer patients compared to neoadjuvant chemotherapy plus placebo, (RR 0.9, 95% CI 0.77-1.05). mTOR inhibition is known to cause cell cycle arrest, therefore, everolimus treatment may reduce the efectiveness of chemotherapy [23]. Compared to placebo plus letrozole, a nonsteroidal aromatase inhibitor, temsirolimus plus letrozole did not improve the ORR (RR 1.0, 95% CI 0.86-1.15) and CBR (RR 0.94, 95% CI 0.86-1.04). The discrepancy with above everolimus treatment is partially due to the population difference, as the temsirolimus trial included only endocrine treatment-naive patients, while the everolimus population composed of patients refractory to a previous treatment with aromatase inhibitor. The difference may also be attributed to the use of steroidal or nonsteroidal aromatase inhibitors, which may show alteration in terms of chemical structure and effect when they are combined with mTOR inhibitors. Finally, due to its high rate of toxic effects (37% versus 11% in the temsirolimus and everolimus groups), temsirolimus may be biologically inactive for the studies analyzed [27].

In summary, we have systematically reviewed all the available randomized studies. These studies were randomized controlled trials and were designed strictly with good homogeneity, and therefore, the data of this meta-analysis is convincing, although the number of studies included was relatively small for a statistics power [29]. Based on the results of our meta-analysis, mTOR inhibitiors in combination with endocrine therapy is likely to be considered as a new therapeutic strategy for women with advanced breast cancer that were previously treated with aromatose inhibitor [27]. Notably, mTOR inhibition can block the negative feedback on IGF-1R signaling interfering on AkT/PI3K signaling. While increasing of Akt phosphorylation, protein kinase activity, and downstream signaling may potentially counteract the inhibition of mTOR [30]. A better understanding is needed regarding which patients will most likely benefit from these therapies and have limited potential to develop resistance to mTOR agents.

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Disclosure of conflict of interest

All authors have no conflict of interest.

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