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SUPPLEMENT ARTICLE

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Emerging data and future directions for CDK4/6 inhibitor treatment of patients with hormone receptor positive HER2-non-amplified metastatic breast cancer

Elgene Lim[1,2] | Jane Beith[3,4] | Frances Boyle[3,5] | Richard de Boer[6] | Rina Hui[3,7] | Nicole McCarthy[8,9] | Andrew Redfern[10] | Theresa Wade[11] | Natasha Woodward[9,12]

[1]St.Vincent's Clinical School, University of New South Wales, Darlinghurst, NSW 2010, Australia

[2]Garvan Institute of Medical Research, Darlinghurst, NSW 2010, Australia

[3]University of Sydney, Camperdown, NSW 2006, Australia

[4]Chris O'Brien Lifehouse, Camperdown, NSW 2050, Australia

[5]Mater Hospital, North Sydney, NSW 2060, Australia

[6]Peter MacCallum Cancer Centre, Melbourne, VIC 3000, Australia

[7]Westmead Hospital, Hawkesbury Road and Darcy Road, Westmead, NSW 2145, Australia

[8]ICON Cancer Care Wesley, Auchenflower, QLD 4066

[9]University of Queensland, St Lucia, QLD 4072, Australia

[10]Fiona Stanley Hospital, Murdoch, Perth, WA 6150

[11]WriteSource Medical Pty Ltd, Lane Cove, NSW 1595, Australia

[12]Mater Misericordiae Ltd and Mater Research Institute Raymond Terrace, South Brisbane, QLD 4101, Australia

Correspondence
Elgene Lim, The Kinghorn Cancer Centre, 370 Victoria St, Darlinghurst, NSW 2010, Australia.
Email: e.lim@garvan.org.au

Abstract

Cyclin-dependent kinase (CDK4/6) inhibitors in combination with endocrine therapy are currently the optimal first line treatment for hormone receptor (HR) positive, human epidermal growth factor receptor 2 (HER2) non-amplified metastatic breast cancer (MBC). However, not all patients benefit from this treatment and all patients will inevitably progress. Identifying therapeutic strategies in this setting is therefore of immediate clinical importance. We present an overview of the mechanisms of resistance to CDK4/6 inhibitors and review potential biomarkers that may guide therapy selection. We also discuss the use of CDK4/6 inhibitors in the context of non-HR-positive/HER2-non-amplified breast cancer and in combination with therapies other than endocrine therapy.

KEYWORDS

breast cancer, cyclin-dependent kinase inhibitor, endocrine therapy, treatment resistance

1 | INTRODUCTION

Cyclin-dependent kinase 4 and 6 (CDK4/6) inhibitors in combination with endocrine therapy have been shown to improve progression free survival (PFS) in hormone receptor (HR) positive, human epidermal growth factor receptor 2 (HER2) non-amplified metastatic breast cancer (MBC) compared with endocrine therapy alone in both the first and second line treatment settings. This combination represents the current standard of care in an increasing number of countries, and has Food and Drug Administration (FDA) approval in the United States (US) and Pharmaceutical Benefits Scheme (PBS) approval in Australia.

Currently estrogen receptor (ER) positivity and HER2 negativity are the only biomarkers used to select patients with MBC for combination endocrine therapy and CDK4/6 inhibitor treatment. However, clinical trials suggest that a proportion of patients have de novo resistance to this treatment combination, and the vast majority of patients will inevitably develop resistance. The strategies to combat resistance to these combination treatments are not yet defined and represent the next major clinical challenge in ER-positive breast cancer. Further research is needed to identify predictive biomarkers of response and new therapeutic strategies for patients who progress on this combination therapy. In this review, we explore the mechanisms of resistance

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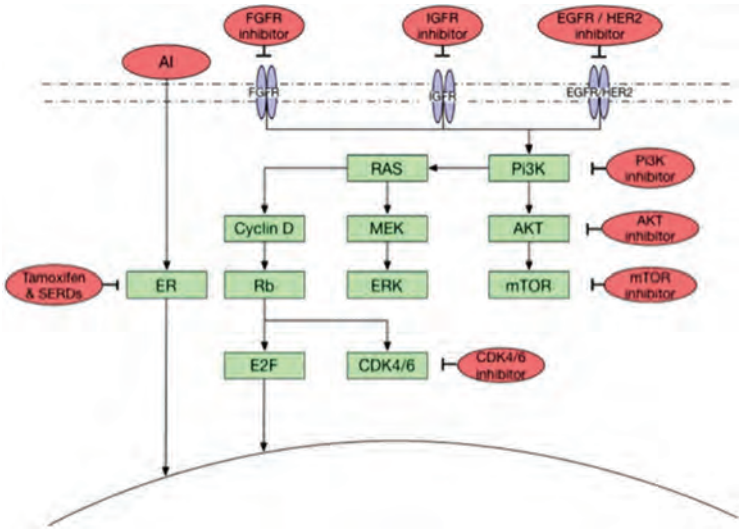

FIGURE 1 Signaling pathways in ER-positive breast cancer. AI, aromatase inhibitor; AKT, serine/threonine protein kinase; CDK4/6, cyclin-dependent kinase 4/6; E2F, E2F transcription factors; EGFR, epidermal growth factor receptor; ER, estrogen receptor; ERK, extra-cellular signal-related kinase; FGFR, fibroblast growth factor receptor; IGFR, insulin growth factor receptor; HER2, human epidermal growth factor receptor 2; MEK, mitogen-activated protein kinase/extra-cellular signal-regulated kinase; mTOR, mammalian target of rapamycin; Pi3K, phosphatidylinositol-3 kinase; RAS, RAS gene; Rb, retinoblastoma; SERDs, selective estrogen receptor degraders

to CDK4/6 inhibitors and review potential biomarkers that may guide therapy selection. We also consider treatment options for patients who progress following treatment with a CDK4/6 inhibitor, the use of CDK4/6 inhibitors in the context of non-HR-positive/HER2-non-amplified breast cancer, and combinations with therapies other than endocrine therapy.

2 | BIOMARKERS OF RESPONSE TO CDK4/6 INHIBITORS

CDK4/6 inhibitors are effective in both endocrine-sensitive and endocrine-resistant breast cancer. This has been demonstrated in pre-clinical studies, and then subsequently in the seminal clinical trials, which have included treatment-naïve patients as well as patients who had relapsed or progressed on endocrine therapy.[1–8] A core reason underlying the efficacy of CDK4/6 inhibitors is their ability to overcome some of the mechanisms that mediate endocrine-resistance, although the eventual progression of disease in the majority of patients treated points to the existence or development of overlapping resistance mechanisms.

Potential predictive biomarkers of response have been evaluated for CDK4/6 inhibitors. Other than ER and retinoblastoma (Rb) positivity, the results have been disappointing in general with none having been clinically validated in patients with HR-positive, HER2-non-amplified breast cancer to date. Dysregulation of the cyclin-CDK-Rb axis by upregulation of cyclin-CDK activity and/or abrogation of suppressors are features of many tumor types, including ER-positive breast cancer. In this cancer, growth is driven by estradiol, which consequently drives cellular proliferation, partly through the increase in levels of cyclin D1 and CDK4/6 activity.[9] Figure 1 shows the interacting signaling pathways in ER-positive, HER2-non-amplified breast cancer and therapeutic targets.

Biomarker studies performed in the PALOMA-2 trial included central blinded analysis on the immunohistochemistry for ER, phosphorylated Rb (pRb), p16, cyclin D1 and Ki-67. A PFS improvement in the palbociclib arm was observed in patients with tumors in all ER quartiles, in patients that were Rb-positive (> 90% of intent-to-treat population (ITP); 24.2 vs. 13.7 months; HR, 0.53; $P < 0.0001$), and in p16-positive patients (85% ITP; 24.8 vs. 13.8 months; HR, 0.52; $P < 0.0001$). The vast majority of patients were found to be cyclin D1-positive (97%), and the benefit of adding palbociclib did not vary with H-score.[10] Similarly, higher expression of genes in the cyclin D-CDK4/6-Rb pathway did not correlate with greater benefit from palbociclib.[11]

One study in patients with HR-positive MBC evaluated the association between response to CDK4/6 inhibitors in second line use and beyond and *PIK3CA* and *TP53* mutations in metastatic tumor DNA.[12] No significant difference in PFS was seen among patients with *PIK3CA* mutations (7.1 vs. 9.7 months; $P = 0.28$) or with any alteration in the PI3K/Akt/mTOR pathway (8.2 vs. 9.3 months; $P = 0.40$). Patients who had *TP53* mutations or complex tumor genomics had a trend toward shorter PFS, however statistical significance was not reached possibly due to the small number of patients in this subgroup.

A study of baseline tumor tissue samples from patients in the PALOMA-3 trial found that patients with low tumor cyclin E expression experienced a greater benefit from palbociclib compared to patients with high expression (median PFS 14.1 vs. 7.6 months, $P = 0.0024$). However, other cell cycle genes did not show any interaction as a biomarker, with all subgroups deriving similar benefit from palbociclib compared to placebo.[13]

Studies of baseline circulating tumor DNA (ctDNA) and tumor tissues from the PALOMA trials found no link between response to palbociclib and *PIK3CA* mutational status.[3,10] In contrast, when sequential plasma samples at baseline and day 15 were studied in a subset of patients in the PALOMA-3 study, a relative decrease in *PIK3CA* ctDNA strongly predicted for a PFS improvement with palbociclib plus fulvestrant (HR 3.94; 95% CI 1.61–9.64; $P = 0.0013$).[14] In this study, *ESR1* mutations (a mutation in gene coding for the ERα) were not predictive of long term PFS improvement. Similar correlative studies performed in the MONALEESA-2 clinical trial demonstrated that the addition of ribociclib to letrozole resulted in an improved PFS irrespective of the status of baseline ctDNA biomarkers (*RTK* or *ZNF703/FGFR1* alterations).[15]

In a small study, the longitudinal mutational landscape was evaluated in the ctDNA of 15 patients who progressed during treatment with palbociclib plus endocrine therapy. There was stable and persistent incidence of *PIK3CA*, *TP53* and *ESR1* mutations before treatment and at progression. In addition, there was also a low but measurable incidence of new mutations found in other genes, and an increased rate of *MYC* gene amplification that may be related to drivers of CDK4/6 inhibitor-resistance.[16]

3 | NOVEL THERAPEUTIC COMBINATIONS WITH CDK4/6 INHIBITORS IN ENDOCRINE-RESISTANT BREAST CANCER

While CDK4/6 inhibitors have been developed for clinical use primarily in the context of combination with endocrine therapy, a number of rational combinations with other classes of drugs can also be considered in the setting of progression on endocrine-based therapy based upon their ability to overcome endocrine-resistance. These preclinical studies provide the rationale for the further clinical development of novel therapeutic combinations with CDK4/6 inhibitors. Details of ongoing clinical trials of novel combinations with CDK4/6 inhibitors are listed in Table 1.

First, there is a growing number of endocrine therapies being developed that are potentially more effective than tamoxifen, aromatase inhibitors and fulvestrant. These new endocrine therapies would make logical partners with CDK4/6 inhibitors. Next generation selective ER degraders (SERDs) currently in clinical development have been shown to be more potent than their prototype fulvestrant with activity in endocrine-resistance and *ESR1* mutations.[17] Furthermore, many of these are orally bioavailable in contrast to fulvestrant, which is administered as an injection, increasing patient convenience. One such example is elacestrant (RAD1901), which has enhanced the efficacy of both palbociclib and abemaciclib *in vitro*.[18] Other SERDs that are currently being evaluated in clinical trials in combination with CDK4/6 inhibitors include LSZ102 in combination with ribociclib (NCT02734615; Table 1), and GDC 9545 in combination with palbociclib (NCT03332797; Table 1).

Other potent ER-directed agents in development include the ER coregulatory binding modulator EtiraRx-11 (ERX-11), which has demonstrated efficacy in blocking ER and ER-coregulator mediated oncogenic signaling, and inhibited proliferation of both endocrine-sensitive and -resistant breast cancer cells. Co-treatment of ERX-11 with palbociclib showed synergistic activity preclinically in *in vitro* and *in vivo* models.[19] Another group of compounds with a unique ER antagonist activity in clinical development are the selective estrogen receptor covalent antagonists (SERCAs), which include H3B-5942. H3B-5942 covalently inactivates both wild-type and mutant ERα by targeting Cys530 and enforcing a unique antagonist conformation resulting in antitumor activity *in vitro* and *in vivo*. Additionally, the efficacy of H3B-5942 was further improved in combination with palbociclib.[20]

The PI3K signaling pathway is a major alternative signaling pathway that is upregulated in endocrine-resistance (Figure 1). Everolimus, a mammalian target of rapamycin (mTOR) inhibitor, in combination with exemestane, has demonstrated prolonged median PFS compared with exemestane alone in patients who had progressed on nonsteroidal aromatase inhibitors (NSAIs) in the BOLERO-2 trial (7.8 vs. 3.2 months; HR 0.45; 95% CI 0.38–0.54)[21] and has been FDA and PBS approved for this indication. Similarly, everolimus extended PFS when added to fulvestrant (12.2 vs. 7.6 months; HR 0.64; 95% CI 0.45–0.91).[22] Preclinical studies with triplet combinations of PI3K/mTOR inhibitors, CDK4/6 inhibitors and endocrine therapies have been promising,[23] forming the basis of a number of phase I clinical studies currently underway (Table 1).[23–27] Initial results suggest that these combinations are feasible with manageable toxicity and showed encouraging signs of clinical activity including in some patients with prior exposure to PI3K/AKT/mTOR or CDK4/6 inhibitors.[25–27] One study found that duration of treatment was longer in patients with *cyclin D* amplification, possibly due to the inclusion of a CDK4/6 inhibitor. These results suggest that triplet therapy might be beneficial for patients who progress on doublet therapy or who have *cyclin D* amplification.[26]

Entinostat is an oral, class 1, selective histone deacetylase (HDAC) inhibitor, which has been shown to inhibit ER-positive breast cancer cells and restore endocrine sensitivity as a result of the downregulation of estrogen-independent growth factor signaling pathways.[28] The phase II trial of exemestane with or without entinostat demonstrated an improvement in median PFS (4.3 vs. 2.3 months; HR 0.73, 95% CI 0.50–1.07) and median overall survival (OS) (28.1 vs. 19.8 months; HR 0.59, 95% CI 0.36–0.97).[29] Consequently, a phase 3 trial of this combination is currently underway in patients with advanced ER-positive breast cancer (NCT02115282). The combination of entinostat and palbociclib has demonstrated synergistic activity *in vitro* and thus also warrants further exploration.[30]

Other signaling pathways that are upregulated in the context of endocrine-resistance include the fibroblast growth factor receptor 1 (FGFR1) and insulin-like growth factor 1(IGF-1) signaling pathway. *FGFR1* amplification occurs in approximately 10% of ER-positive human breast cancers, and is associated with early recurrence after adjuvant endocrine therapy.[31] The ER signaling pathway remains active in ER-positive, *FGFR1*-amplified breast cancer models in estrogen-deprived conditions.[32] These findings have led to the initiation of a phase 2 trial of lucitanib, a vascular endothelial growth factor (VEGF), platelet-derived growth factor receptor (PDGFR) and FGFR inhibitor in patients with ER-positive metastatic breast cancer (NCT02053636). IGF ligand-dependent signaling via the IGF receptor results in upregulation of cyclin D1, and subsequent progression through the cell cycle,[33] and the combination of palbociclib with IGF-1 receptor (IGF-1R) inhibitory antibody has demonstrated promising *in vitro* activity in preclinical ER-positive breast cancer cells.[34] A phase I clinical trial (NCT03099174; Table 1) of an IGF-1 ligand-neutralizing antibody, xentuzumab, in combination with abemaciclib, with and without endocrine therapy, is currently underway.

Finally, CDK4/6 inhibitors have also been shown to increase tumor immunogenicity through the activation of tumor cell expression of endogenous retroviral elements and enhancement of tumor antigen presentation, as well as by the suppression of regulatory T cell (Treg) proliferation.[35] This seminal finding has provided a rationale for new combination regimens comprising CDK4/6 inhibitors and

TABLE 1 Current accruing clinical trials for the treatment of hormone receptor positive, HER2-non-amplified metastatic breast cancer

Study Title	Agents	Therapy class	Study status	Estimated completion date	Clinicaltrials.gov identifier	Ref.
Novel therapeutic combinations with CDK4/6 inhibitors						
A phase I/Ib study of LSZ102 (LSZ102+ ribociclib or LSZ102 + alpelisib) in patients with advanced ER+ breast cancer who have progressed after endocrine therapy	Ribociclib Alpelisib LSZ102	CDK4/6i Pi3K/mTORi SERD	Recruiting	February 2018	NCT02734615	74
A phase Ia/Ib, study of GDC 9545 alone or in combination with Palbociclib and/or LHRH Agonist in patients with advanced ER+ breast cancer	Palbociclib GDC 9545 LHRH agonist	CDK4/6i SERD LHRH agonist	Recruiting	July 2021	NCT03332797	N/A
A phase Ib study of Gedatolisib in combination with Palbociclib and either Letrozole or Fulvestrant in women with advanced ER + breast cancer	Palbociclib Gedatolisib Fulvestrant or Letrozole	CDK4/6i Pi3K/mTORi SERD NSAI	Recruiting	November 2019	NCT02684032	25
A phase Ib study of Ribociclib in combination with Everolimus and Exemestane in postmenopausal women with advanced ER+ breast cancer	Ribociclib Everolimus Exemestane	CDK4/6i Pi3K/mTORi SAI	Active, not recruiting	September 2018	NCT01857193	26
A phase Ib/II, study of the combination of Ribociclib and Alpelisib with letrozole in patients with advanced ER+ breast cancer	Ribociclib Alpelisib Letrozole	CDK4/6i Pi3K/mTORi NSAI	Recruiting	March 2019	NCT01872260	27
A phase Ib study of Xentuzumab and Abemaciclib in patients with advanced solid tumors and in combination with endocrine therapy in patients with advanced ER+ breast cancer	Abemaciclib Xentuzumab Letrozole Anastrozole Fulvestrant	CDK4/6i IGF antibody NSAI NSAI SERD	Recruiting	December 2018	NCT03099174	33
A phase Ib study of Abemaciclib in combination with Pembrolizumab for patients with advanced non-small cell lung cancer or ER+ breast cancer	Abemaciclib Pembrolizumab	CDK4/6i PD-L1 antibody	Recruiting	May 2019	NCT02779751	36
A phase II study of the combination of Pembrolizumab, Letrozole and Palbociclib in postmenopausal patients with newly diagnosed advanced ER+ breast cancer	Palbociclib Pembrolizumab Letrozole	CDK4/6i PD-L1 antibody NSAI	Recruiting	September 2018	NCT02778685	37
Potential treatment strategies to overcome resistance to CDK4/6 inhibitors						
A phase Ib trial of Fulvestrant, Palbociclib, and Erdafitinib in advanced ER+/HER2-/FGFR-amplified breast cancer	Palbociclib Erdafitinib Fulvestrant	CDK4/6i FGFR inhibitor SERD	Recruiting	September 2020	NCT03238196	49
Treatment of HR-positive HER2-non-amplified MBC following progression with a CDK4/6 inhibitor						
A phase II study of Fulvestrant, Palbociclib and Avelumab for endocrine pre-treated ER+ advanced breast cancer	Palbociclib Fulvestrant Avelumab	CDK4/6 SERD PD-L1 antibody	Recruiting	December 2020	NCT03147287	57
A phase II, study of Alpelisib Plus Fulvestrant or Letrozole in patients with PIK3CA mutant, ER+ advanced breast cancer, who have progressed on or after CDK4/6i treatment	Alpelisib Fulvestrant or Letrozole	Pi3K/mTORi SERD NSAI	Recruiting	February 2020	NCT03056755	58
A phase II study of Fulvestrant with or without Ribociclib after progression on anti-estrogen therapy plus CDK4/6i in patients with advanced ER+ breast cancer	Ribociclib Fulvestrant	CDK4/6i SERD	Recruiting	May 2019	NCT02632045	59
A phase I/II, study of Ribociclib in combination with Everolimus + Exemestane in the treatment of men and postmenopausal women with ER+ advanced breast cancer following progression on a CDK 4/6i	Ribociclib Everolimus Exemestane	CDK4/6i PI3K/mTORi SAI	Recruiting	May 2018	NCT02732119	60
Patients with CNS metastases						
A phase II study of Abemaciclib in patients with brain metastases secondary to ER+ breast cancer, non-small cell lung cancer, or melanoma	Abemaciclib	CDK4/6i	Recruiting	November 2018	NCT02308020	64
A phase II study of Palbociclib in patients with metastatic HER2-positive breast cancer with brain metastasis	Palbociclib	CDK4/6i	Recruiting	November 2019	NCT02774681	65

(Continues)

TABLE 1 (Continued)

Study Title	Agents	Therapy class	Study status	Estimated completion date	Clinicaltrials.gov identifier	Ref.
HR-positive, HER2-amplified MBC						
A phase II, study of Abemaciclib plus Trastuzumab with or without Fulvestrant vs chemotherapy of physician's choice Plus trastuzumab in patients with HR+, HER2+ advanced breast cancer	Abemaciclib Trastuzumab Fulvestrant	CDK4/6i HER2 antibody SERD	Active, not recruiting	August 2018	NCT02675231	69
A phase Ib/II study of Ribociclib, in combination with Trastuzumab or T-DM1 for advanced HER2+ breast cancer	Ribociclib Trastuzumab T-DM1 Fulvestrant	CDK4/6i HER2 antibody HER2 antibody-chemotherapy conjugate SERD	Recruiting	February 2019	NCT02657343	70
A phase Ib/II study Tucatinib in combination with Palbociclib and Letrozole in subjects with advanced ER+HER2+ breast cancer	Palbociclib Tucatinib Letrozole	CDK4/6i TKI NSAI	Recruiting	October 2018	NCT03054363	71
A phase III study of Palbociclib + Anti-HER2 therapy + endocrine therapy vs. Anti-HER2 therapy + endocrine therapy after induction treatment for advanced ER+HER2+ breast cancer	Palbociclib Trastuzumab Pertuzumab Letrozole or Anastrozole or Exemestane or Fulvestrant	CDK4/6i HER2 antibody NSAI NSAI SAI SERD	Recruiting	October 2020	NCT02947685	72
A phase III study of chemo vs endocrine therapy in combination with dual HER2-targeted therapy in patients with advanced ER+HER2+ breast cancer	Ribociclib Trastuzumab Pertuzumab Letrozole or Anastrozole or Exemestane or Fulvestrant or Tamoxifen Paclitaxel or Vinorelbine or Docataxel	CDK4/6i HER2 antibody NSAI NSAI SAI SERD SERM chemo	Recruiting	September 2021	NCT02344472	73

CDK4/6i, cyclin-dependent kinase 4/6 inhibitor; HR, hormone receptor; HER2, human epidermal growth factor receptor 2; CNS, central nervous system; Pi3K/mTORi, phosphatidylinositol-3 kinase/mammalian target of rapamycin inhibitor; SERD, selective estrogen receptor degrader; LHRH agonist, luteinizing hormone-releasing hormone agonist; NSAI, non-steroidal aromatase inhibitor; SAI, steroidal aromatase inhibitor; IGF, insulin growth factor; PD-L1, programmed death ligand 1; FGFR, fibroblast growth factor receptor; TKI, tyrosine kinase inhibitor; SERM, selective estrogen receptor modulator.

immunotherapies. Pembrolizumab, an anti-PD-1 antibody, is currently being evaluated in combination with abemaciclib in a phase II clinical trial (NCT02779751; Table 1).[36] Pembrolizumab is also being evaluated in a phase II study (NCT02778685; Table 1) in postmenopausal patients who have achieved stable disease during treatment with letrozole and palbociclib.[37]

4 | RESISTANCE TO CDK4/6 INHIBITORS

Many of the preclinical studies to date have evaluated single agent resistance to CDK4/6 inhibitors, in contrast to the clinical context in which resistance occurs in combination with endocrine therapy. An important consideration is how the ER and CDK4/6 directed therapy resistance mechanisms overlap. CDK4/6 inhibition acts directly downstream of endocrine therapy and it is therefore inevitable that some mechanisms of resistance will be common to both types of treatments.

Mechanisms of endocrine-resistance have been well described, including upregulation of alternate signaling pathways, genomic and epigenetic aberrations.[38–40] Rb is maintained in the vast majority of ER-positive breast cancer, and in most cases functional Rb protein is retained through the development of endocrine-resistance,[41] rendering these tumors amenable to CDK4/6 inhibition. In addition, the incidence of *Rb* gene deletion/mutation is very rare (3.9%) in ER-positive breast cancer.[42] While there have been preclinical studies demonstrating that Rb loss is a mechanism for resistance to CDK4/6 inhibition,[43,44] the jury is still out as to whether Rb is a predictive biomarker for CDK4/6 inhibitor response as there are studies in other cancer types to suggest that functional Rb is not an absolute requirement for CDK4/6 inhibitors to be effective.[45]

Other proposed mechanisms of resistance to CDK4/6 inhibitors include the upregulation of CDK4 or CDK6. In one study, CDK4 T172-phosphorylation was found to correlate with sensitivity to palbociclb, and an 11-gene expression signature that predicted CDK4 modification profiles of breast tumors and cell lines correlated with palbociclib response.[46] Low levels of CDK6 expression have been shown to be positively correlated with the efficacy of ribociclib in endocrine-resistant breast cancer cell lines.[47] CDK6 has also been shown to play a role in mediating endocrine-resistance, highlighting the potential overlap in mechanisms of resistance to endocrine therapy and CDK4/6 inhibitors. Increased expression of CDK6 have been described in fulvestrant-resistant breast cancer cell lines.[48] Furthermore, high CDK6 levels in

metastatic tumor tissue from two independent cohorts of breast cancer patients treated with fulvestrant ($n = 45$ and 46) correlated significantly with shorter PFS in patients treated with fulvestrant (median time to progression 2.5 vs. 8.2 months, $P = 0.0006$ and 3.4 vs. 8.9 months, $P = 0.018$). Treatment with palbociclib resulted in alteration of Rb protein phosphorylation and re-sensitization of the cells to fulvestrant in preclinical models.[48]

5 | POTENTIAL TREATMENT STRATEGIES TO OVERCOME RESISTANCE TO CDK4/6 INHIBITORS

In addition to playing a role in endocrine-resistance, *FGFR1* amplification has also been reported to induce resistance to fulvestrant and CDK4/6 inhibitor combinations in gain-of-function kinase library screens in cell lines.[49] The resistance phenotype was abrogated with the FGFR tyrosine kinase inhibitor lucitanib. Furthermore, in the MONALEESA-2 study, *FGFR1* amplification in ctDNA was shown to correlate with early progression on ribociclib.[15] The data suggest that FGFR signaling is a mechanism of resistance to endocrine therapies with or without CDK4/6 inhibitors, with overexpression of cyclin D1 induced by both FGFR signaling and ER transcription.[49] Additionally, two activating *FGFR2* mutations (*M538I* and *N550K*) have been identified in patients with endocrine-resistant ER-positive MBC, which are associated with resistance to fulvestrant, palbociclib and the combination of the two agents.[50] Building on these findings, a phase Ib study has been initiated to evaluate fulvestrant, palbociclib and erdafitinib (an FGFR inhibitor) in patients with endocrine-resistant ER-positive, HER2-non-amplified and *FGFR1-4* amplified MBC (NCT03238196; Table 1).

In another study using ribociclib-resistant breast cancer cells, dinaciclib, a pan-CDK inhibitor and GSK2334470, a 3-phoshoinositide dependent protein kinase 1 (PDK1) inhibitor was found to re-sensitize the resistant cells to CDK4/6 inhibitors.[51] Ribociclib plus GSK2334470 completely abrogated components of the cell cycle, including pRb, pS6, pRSK2, pCDK2, cyclin A and cyclin E, making this combination another potential strategy to overcome CDK4/6 inhibitor resistance.

Finally, higher activator protein 1 (AP-1) transcriptional activity has been demonstrated in dually tamoxifen and palbociclib-resistant cell lines.[52] The combination of palbociclib and AP-1 blockade was effective in inhibiting cell growth and reducing pRb and CDK2 levels compared to either agent alone.

6 | TREATMENT OF HR-POSITIVE, HER2-NON-AMPLIFIED MBC FOLLOWING PROGRESSION ON A CDK4/6 INHIBITOR

This clinical scenario will increase in frequency as CDK4/6 inhibitors become established as the standard of care for patients with advanced ER-positive breast cancer in combination with endocrine therapy. There are currently no treatment guidelines as to how to manage patients who progress on CDK4/6 inhibitors, even though CDK4/6 inhibitor use is most often confined to the first and second line settings. Several studies have investigated the follow-up treatment of patients after participation in clinical trials with a CDK4/6 inhibitor. In patients studied for 9 months immediately after participation in the PALOMA-3 study, the median time to subsequent chemotherapy was significantly longer for the palbociclib arm compared to the placebo arm (252 vs. 132 days), and fewer patients in the palbociclib arm discontinued next-line treatment (33% vs. 46%), suggesting that palbociclib did not adversely affect the efficacy of subsequent treatments.[53] Similar trends were observed for post-progression treatment of patients receiving the next line of therapy immediately following participation in the PALOMA-1 trial.[54] In the MONALEESA-2 study, ribociclib plus letrozole also prolonged the time to next treatment compared to letrozole alone (24.2 vs. 16.7 months).[55] Fewer patients treated with the combination received subsequent chemotherapy compared with letrozole alone (15.8% vs. 22.4%).

In contrast, there are case studies of rapid secondary disease progression during subsequent lines of therapy following cessation of CDK4/6 inhibitors.[56] This observation has not been reported in larger CDK4/6 inhibitor clinical trials.

Table 1 outlines several phase II clinical trials that are ongoing to investigate the treatment of HR-positive, HER2-non-amplified MBC with endocrine therapy, CDK4/6 inhibitors or PI3K/mTOR inhibitors following progression on a CDK4/6 inhibitor.[57–60]

7 | OTHER CLINICAL SCENARIOS IN WHICH CDK4/6 INHIBITORS MAY PLAY A ROLE IN BREAST CANCER

7.1 | Patients with central nervous system (CNS) metastases

A lower percentage of patients with HR-positive, HER2-non-amplified MBC develop brain metastases when compared with patients with triple negative breast cancer or HER2-amplified MBC.[61] However, owing to the high incidence of the HR-positive, HER2-non-amplified breast cancer sub-type, cases are still relatively common. There are no specific regulatory approved systemic agents for the treatment of HR-positive breast cancer brain metastases, and so this remains an unmet medical need. Standard local treatment options include surgery, stereotactic radiosurgery (SRS) and/or whole brain radiation therapy (WBRT).[61]

There is preliminary evidence and preclinical data that ribociclib, abemaciclib and palbociclib cross the brain–blood barrier,[62,63] with data suggesting that albemaciclib may be more efficient in crossing the blood–brain barrier compared to palbociclb.[62] Several clinical trials are ongoing to evaluate CDK4/6 inhibitors in patients with brain metastases (Table 1).[64,65]

7.2 | HR-positive, HER2-amplified MBC

There have been results from three phase II clinical studies investigating the treatment of HER2-amplified MBC with palbociclib. PATRICIA

assessed the efficacy of palbociclib and trastuzumab in advanced HER2-amplified breast cancer (patients had received two to four prior lines of anti-HER2 based therapies). Patients with luminal disease showed a statistically significantly longer median PFS than patients with non-luminal disease (10.37 vs. 3.53 months, $P = 0.023$) and it was concluded that patients with non-luminal disease might not derive a large benefit from this treatment strategy.[66] Another clinical trial investigated the addition of palbociclib with or without trastuzumab in patients with Rb-positive advanced cancer, including a sub-cohort of patients with HER2-amplified disease. Of the 10 patients with HER2-amplified disease receiving palbociclib plus trastuzumab, a response rate of 30% and a PFS of 6.7 months (95% CI 1.6–17.8) was observed. It was concluded that further investigation into this combination was warranted.[67] Neoadjuvant triplet therapy with trastuzumab, pertuzumab and palbociclib for women with non-metastatic HER2-amplified breast cancer was also investigated (NCT02530424). The primary endpoint was characterization of Ki67 changes from baseline before therapy, at 2 weeks and at surgery. This triplet targeting resulted in a significant and rapid decrease of Ki67 that was of larger magnitude after 2 weeks than at surgery irrespective of the recorded objective clinical response. It was suggested that tolerability and clinical response merit further clinical testing and additional molecular characterization of this chemotherapy-free approach.[68]

There are several ongoing clinical trials investigating CDK4/6 inhibitor combinations in patients with advanced HR-positive, HER2-amplified breast cancer (Table 1). There are two studies evaluating the combination of a CDK4/6 inhibitor with trastuzumab with or without fulvestrant.[69,70] A phase Ib/II study will test the combination of the novel oral HER2 small molecule inhibitor tucatinib with palbociclib and letrozole as a potential non-chemotherapy based regimen.[71] PATINA (NCT02947685; Table 1) is a phase III registration trial investigating the addition of palbociclib to dual HER2-directed (pertuzumab and trastuzumab) and endocrine therapy to determine if the addition of a CDK4/6 inhibitor will prolong PFS compared to dual HER2-directed and endocrine therapy.[72] Similarly, the DETECT V study (NCT02344472; Table 1) is comparing the safety and efficacy of dual HER2-directed (pertuzumab and trastuzumab) and endocrine therapy with ribociclib.[73]

8 | SUMMARY AND CONCLUSIONS

ER-positivity and HER2-non-amplification are currently the only biomarkers used to select breast cancer patients for treatment with CDK4/6 inhibitors. Further analysis from PALOMA and MONALEESA clinical trials showed response to CDK4/6 inhibitors were not linked to a range of biomarkers from baseline samples.[3,10,11,13,15] However sequential plasma samples analyzed from the PALOMA-3 trial did show a decrease in *PIK3CA* ctDNA after 2 weeks strongly predicted for prolonged PFS with palbociclib compared to placebo.[14] There clearly needs to be further work in this area, as identifying patients who do really well with or without the addition of a CDK4/6 inhibitor is critical to allow best use of these expensive drugs.

Several clinical trials are investigating novel therapeutic combinations with CDK4/6 inhibitors in endocrine-resistant breast cancer. These include the combination of CDK4/6 inhibitors with next generation SERDs and new classes of endocrine therapies, HDAC inhibitors, IGFR and FGFR inhibitors and immunotherapies.[29,33,74] Triplet combinations of CDK4/6 inhibitors, PI3K/mTOR inhibitors and endocrine therapies, and CDK4/6 inhibitors, HER2-directed antibodies and endocrine therapies are also being explored.[23–27,37] There are also concerted efforts to identify potential treatment strategies to overcome CDK4/6 inhibitor resistance, including the use of FGFR, PDK1 and AP-1 inhibitors.[49,51,52]

There are several ongoing clinical trials to investigate treatment of patients with HR-positive, HER2-non-amplified MBC following progression on a CDK4/6 inhibitor and endocrine therapy, a major emerging clinical challenge where there is currently little data and no guidelines.

Given the observed benefit of CDK4/6 inhibitors in combination with endocrine therapy for patients with HR-positive, HER2-non-amplified MBC, large adjuvant studies are now underway to investigate the combination in HR-positive, HER2-non-amplified early breast cancer.[75–77]

CDK4/6 inhibitors in combination with endocrine therapy represent a significant development in the treatment of HR-positive, HER2-non-amplified MBC. However, identifying therapeutic strategies to follow this treatment is of immediate clinical importance since the vast majority of patients will inevitably progress on this combination.

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DISCLOSURES

F.B. has served as an investigator on CDK inhibitor trials, and on advisory boards for Pfizer, Novartis, Eli Lilly, Astra Zeneca and Roche. N.W. has been an investigator on CDK inhibitor trials, owns stock in CSL, has participated in advisory boards for Roche and Novartis and has provided editorial support to Pfizer. R.H. has participated in advisory boards for Novartis, Merck Sharp and Dohme, AstraZeneca, Roche, Bristol Myer Squibb and Pfizer and has received speaker honoraria from Novartis, Merck Sharp and Dohme, AstraZeneca, Roche, Bristol Myer Squibb and Boehringer Ingelheim. J.B. has participated in advisory boards for Pfizer, Novartis, Eli Lilly and Roche. R.D.B. served as an investigator on CDK inhibitor trials, is a member of advisory boards for Novartis and Roche and has received speaker honoraria from Novartis, Roche, Amgen and Genomic Health. A.R. has been an investigator on CDK inhibitor trials, and participated on advisory boards for Pfizer, Novartis, Roche and Eisai. E.L. has received research funding from Novartis and Bayer Pharmaceuticals, and has participated in advisory boards for Pfizer, Novartis, Roche, Eisai and Lilly. N.M.C. has been an investigator on CDK inhibitor trials and participated in advisory boards for Pfizer, Novartis, Roche and Astra Zeneca.

REFERENCES

1. Finn RS, Crown JP, Lang I, et al. The cyclin-dependent kinase 4/6 inhibitor palbociclib in combination with letrozole versus letrozole alone as first-line treatment of oestrogen receptor-positive, HER2-negative, advanced breast cancer (PALOMA-1/TRIO-18): a randomised phase 2 study. *Lancet Oncol*. 2015;16:25–35.
2. Finn RS, Martin M, Rugo HS, et al. Palbociclib and Letrozole in Advanced Breast Cancer. *N Engl J Med*. 2016;375:1925–1936.
3. Cristofanilli M, Turner NC, Bondarenko I, et al. Fulvestrant plus palbociclib versus fulvestrant plus placebo for treatment of hormone-receptor-positive, HER2-negative metastatic breast cancer that progressed on previous endocrine therapy (PALOMA-3): final analysis of the multicentre, double-blind, phase 3 randomised controlled trial. *Lancet Oncol*. 2016;17:425–439.
4. Hortobagyi GN, Stemmer SM, Burris HA, et al. Ribociclib as first-line therapy for HR-positive, advanced breast cancer. *N Engl J Med*. 2016;375:1738–1748.
5. Hortobagyi GN, Stemmer SM, Burris HA, et al. Updated results from MONALEESA-2, a phase III trial of first-line ribociclib plus letrozole versus placebo plus letrozole in hormone receptor-positive, HER2-negative advanced breast cancer. *Ann Oncol*. 2018;29:1541–1547.
6. Slamon DJ, Neven P, Chia S, et al. Phase III randomized study of ribociclib and fulvestrant in hormone receptor-positive, human epidermal growth factor receptor 2-negative advanced breast cancer: mONALEESA-3. *J Clin Oncol*. 2018:JCO2018789909.
7. Sledge GW, Jr, Toi M, Neven P, et al. MONARCH 2: abemaciclib in combination with fulvestrant in women with HR+/HER2– advanced breast cancer who had progressed while receiving endocrine therapy. *J Clin Oncol*. 2017;35:2875–2884.
8. Goetz MP, Toi M, Campone M, et al. MONARCH 3: abemaciclib as initial therapy for advanced breast cancer. *J Clin Oncol*. 2017;35:3638–3646.
9. Filmus J, Robles AI, Shi W, Wong MJ, Colombo LL, Conti CJ. Induction of cyclin D1 overexpression by activated ras. *Oncogene*. 1994;9:3627–3633.
10. Finn R, Jiang Y, Rugo H, et al. Biomarker analyses from the phase 3 PALOMA-2 trial of palbociclib (P) with letrozole (L) compared with placebo (PLB) plus L in postmenopausal women with ER+/HER2– advanced breast cancer (ABC). *Ann Oncol*. 2016;27:LBA15.
11. Finn RS, Liu Y, Martin M, et al. Abstract P2-09-10: comprehensive gene expression biomarker analysis of CDK 4/6 and endocrine pathways from the PALOMA-2 study. *Cancer Research*. 2018;78:P2-09-10.
12. Spring L, Niemierko A, Juric D, et al. Tumor genomics and response to CDK 4/6 inhibitors for patients with hormone receptor-positive (HR+) metastatic breast cancer (MBC). *J Clin Oncol*. 2017;35:1046.
13. Turner NC, Liu Y, Zhu Z, et al. Abstract CT039: cyclin E1 (*CCNE1*) expression associates with benefit from palbociclib in metastatic breast cancer (MBC) in the PALOMA3 trial. *Cancer Res*. 2018;78:CT039.
14. O'Leary B, Hrebien S, Morden JP, et al. Early circulating tumor DNA dynamics and clonal selection with palbociclib and fulvestrant for breast cancer. *Nat Commun*. 2018;9:896.
15. Hortobagyi GN, Stemmer S, Campone M, et al. Abstract PD4-06: first-line ribociclib + letrozole in hormone receptor-positive, HER2-negative advanced breast cancer: efficacy by baseline circulating tumor DNA alterations in MONALEESA-2. *Cancer Res*. 2018;78:PD4-06.
16. Cruz MR, Limentani K, Taxter T, et al. Abstract PD4-05: patterns of genomic alterations in ER-positive advanced breast cancer patients treated with CDK4/6 inhibitors. *Cancer Res*. 2018;78:PD4-05.
17. McDonnell DP, Wardell SE, Norris JD. Oral selective estrogen receptor downregulators (SERDs), a breakthrough endocrine therapy for breast cancer. *J Med Chem*. 2015;58:4883–4887.
18. Martin LA, Pancholi S, Simigdala N, et al. Abstract P4-04-09: new oral SERD elacestrant (RAD1901) shows efficacy in breast cancer models harbouring ESR1 mutations and enhances the antiproliferative activity of mTORC1 and CDK4/6 inhibitors. *Cancer Res*. 2018;78:P4-04-09.
19. Viswanadhapalli S, Sareddy GR, Zhou M, et al. Abstract P1-09-06: blocking ER coregulator signaling enhances CDK4/6 inhibitor palbociclib therapy in ER-positive advanced breast cancer. *Cancer Res*. 2018;78:P1-09-06.
20. Puyang X, Furman C, Zheng GZ, et al. Discovery of selective estrogen receptor covalent antagonists (SERCAs) for the treatment of ERa(WT) and ERa(MUT) breast cancer. *Cancer Discov*. 2018.
21. Yardley DA, Noguchi S, Pritchard KI, et al. Everolimus plus exemestane in postmenopausal patients with HR(+) breast cancer: bOLERO-2 final progression-free survival analysis. *Adv Ther*. 2013;30:870–884.
22. Schmid P, Zaiss M, Harper-Wynne C, et al. Abstract GS2-07: mANTA - a randomized phase II study of fulvestrant in combination with the dual mTOR inhibitor AZD2014 or everolimus or fulvestrant alone in estrogen receptor-positive advanced or metastatic breast cancer. *Cancer Res*. 2018;78:GS2-07.
23. Oelmann E, Michaloglou C, Crafter C, et al. Abstract PD4-04: combined inhibition of mTOR and CDK4/6 is required for optimal blockade of E2F function and long term growth inhibition in estrogen receptor positive breast cancer. *Cancer Res*. 2018;78:PD4-04.
24. Cortes J, Im SA, Holgado E, Perez-Garcia JM, Schmid P, Chavez-MacGregor M. The next era of treatment for hormone receptor-positive, HER2-negative advanced breast cancer: triplet combination-based endocrine therapies. *Cancer Treat Rev*. 2017;61:53–60.
25. Forero A, Han HS, Dees EC, et al. Abstract OT2-07-06: phase Ib study to assess the safety, tolerability, and clinical activity of gedatolisib in combination with palbociclib and either letrozole or fulvestrant in women with metastatic or locally advanced/recurrent breast cancer (B2151009). *Cancer Res*. 2018;78:OT2-07-06.
26. Bardia A, Modi S, Oliveira M, et al. Abstract P6-13-01: triplet therapy with ribociclib, everolimus, and exemestane in women with HR+/HER2– advanced breast cancer. *Cancer Res*. 2016;76:P6-13-01.
27. Juric D, Ismail-Khan R, Campone M, et al. Abstract P3-14-01: phase Ib/II study of ribociclib and alpelisib and letrozole in ER+, HER2– breast cancer: safety, preliminary efficacy and molecular analysis. *Cancer Res*. 2016;76:P3-14-01.
28. Sabnis GJ, Goloubeva O, Chumsri S, Nguyen N, Sukumar S, Brodie AM. Functional activation of the estrogen receptor-alpha and aromatase by the HDAC inhibitor entinostat sensitizes ER-negative tumors to letrozole. *Cancer Res*. 2011;71:1893–1903.
29. Yardley DA, Ismail-Khan RR, Melichar B, et al. Randomized phase II, double-blind, placebo-controlled study of exemestane with or without entinostat in postmenopausal women with locally recurrent or metastatic estrogen receptor-positive breast cancer progressing on treatment with a nonsteroidal aromatase inhibitor. *J Clin Oncol*. 2013;31:2128–2135.
30. Lee J, Lim B, Pearson T, Tripathy D, Ordentlich P, Ueno NT. Abstract P5-21-15: the synergistic antitumor activity of entinostat (MS-275) in combination with palbociclib (PD 0332991) in estrogen receptor-positive and triple-negative breast cancer. *Cancer Res*. 2018;78:P5–21–15.
31. Turner N, Pearson A, Sharpe R, et al. FGFR1 amplification drives endocrine therapy resistance and is a therapeutic target in breast cancer. *Cancer Res*. 2010;70:2085–2094.

32. Formisano L, Stauffer KM, Young CD, et al. Association of FGFR1 with ERalpha maintains ligand-independent ER transcription and mediates resistance to estrogen deprivation in ER(+) breast cancer. *Clin Cancer Res*. 2017;23:6138–6150.
33. Yee D, Prat A, Sablin MP, et al. 90TiPA phase Ib trial of xentuzumab and abemaciclib in patients with locally advanced or metastatic solid tumors, hormone receptor-positive (HR+), HER2-negative (HER2-) breast cancer (BC; +/- endocrine therapy), or non-small-cell lung cancer (NSCLC). *Ann Oncol*. 2017;28. mdx656.001.
34. Sachdev D, Hoff K. Abstract PD4-03: cDK4/6 inhibition blocks effects of IGFs and insulin in estrogen receptor positive and triple negative breast cancers: implications for cotargeting IGF1R/IR and CDKs. *Cancer Res*. 2018;78:PD4-03.
35. Goel S, DeCristo MJ, Watt AC, et al. CDK4/6 inhibition triggers anti-tumour immunity. *Nature*. 2017;548:471–475.
36. Rugo H, Tolaney S, Dickler M, et al. Abstract OT2-01-07: a phase 2 study of abemaciclib plus pembrolizumab for patients with hormone receptor positive (HR+), HER2 negative (HER2-) metastatic breast cancer (MBC). *Cancer Res*. 2017;77:OT2-01-07.
37. Yuan Y, Frankel P, Synold T, et al. Abstract OT2-01-03: phase II trial of the addition of pembrolizumab to letrozole and palbociclib in patients with metastatic estrogen receptor positive breast cancer who have stable disease on letrozole and palbociclib. *Cancer Res*. 2017;77:OT2-01-03.
38. Musgrove EA, Sutherland RL. Biological determinants of endocrine resistance in breast cancer. *Nat Rev Cancer*. 2009;9:631–643.
39. Jeselsohn R, Yelensky R, Buchwalter G, et al. Emergence of constitutively active estrogen receptor-alpha mutations in pretreated advanced estrogen receptor-positive breast cancer. *Clin Cancer Res*. 2014;20:1757–1767.
40. Stone A, Zotenko E, Locke WJ, et al. DNA methylation of oestrogen-regulated enhancers defines endocrine sensitivity in breast cancer. *Nat Commun*. 2015;6:7758.
41. Musgrove EA, Caldon CE, Barraclough J, Stone A, Sutherland RL. Cyclin D as a therapeutic target in cancer. *Nat Rev Cancer*. 2011;11:558–572.
42. Ciriello G, Gatza ML, Beck AH, et al. Comprehensive molecular portraits of invasive lobular breast cancer. *Cell*. 2015;163:506–519.
43. Dean JL, McClendon AK, Hickey TE, et al. Therapeutic response to CDK4/6 inhibition in breast cancer defined by ex vivo analyses of human tumors. *Cell Cycle*. 2012;11:2756–2761.
44. Chou A, Froio D, Nagrial AM, et al. Tailored first-line and second-line CDK4-targeting treatment combinations in mouse models of pancreatic cancer. *Gut*. 2017. https://doi.org/10.1136/gutjnl-2017-315144.
45. Castellano D, Rubio C, López-Calderón F, et al. Cdk4/6 inhibitor activity in metastatic bladder cancer cell lines is independently of RB1 status. *Ann Oncol*. 2016;27:ix1–ix8.
46. Raspe ES, Coulonval K, Pita J, et al. Abstract P6-07-02: cDK4 phosphorylation status and corresponding gene expression profile predict sensitivity to Palbociclib. *Cancer Res*. 2017;77:P6-07-02.
47. Iida M, Nakamura M, Tokuda E, Niwa T, Ishida T, Hayashi SI. Abstract P6-04-02: cDK6 might be a key factor for efficacy of CDK4/6 inhibitor and the hormone sensitivity following acquired resistance. *Cancer Res*. 2018;78:P6-04-02.
48. Alves CL, Elias D, Lyng M, et al. High CDK6 protects cells from fulvestrant-mediated apoptosis and is a predictor of resistance to fulvestrant in estrogen receptor-positive metastatic breast cancer. *Clin Cancer Res*. 2016;22:5514–5526.
49. Formisano L, Lu Y, Jansen VM, et al. Abstract GS6-05: gain-of-function kinase library screen identifies FGFR1 amplification as a mechanism of resistance to antiestrogens and CDK4/6 inhibitors in ER+ breast cancer. *Cancer Research*. 2018;78:GS6–05.
50. Mao P, Kusiel J, Cohen O, Wagle N. Abstract PD4-01: the role of FGF/FGFR axis in resistance to SERDs and CDK4/6 inhibitors in ER+ breast cancer. *Cancer Res*. 2018;78:PD4-01.
51. Jansen VM, Formisano L, Witkiewicz A, et al. Abstract P3-03-05: pI3K/PDK1 mediates resistance to CDK4/6 inhibitors through dysregulation of S-phase cyclins/cyclin dependent kinases (CDKs). *Cancer Research*. 2017;77:P3-03-05.
52. De Angelis C, Nardone A, Cataldo ML, et al. Abstract P4-03-05: aP-1 as a potential mediator of resistance to the cyclin-dependent kinase (CDK) 4/6-inhibitor palbociclib in ER-positive endocrine-resistant breast cancer. *Cancer Res*. 2018;78:P4-03-05.
53. Turner NC, André F, Cristofanilli M, et al. Abstract P4-22-06: treatment postprogression in women with endocrine-resistant HR+/HER2- advanced breast cancer who received palbociclib plus fulvestrant in PALOMA-3. *Cancer Res*. 2017;77:P4-22-06.
54. Finn RS, Crown JP, Ettl J, et al. Abstract P4-13-02: treatment patterns of post-disease progression in the PALOMA-1/TRIO-18 trial. *Cancer Res*. 2016;76:P4-13-02.
55. Blackwell KL, Paluch-Shimon S, Campone M, et al. Abstract P5-21-18: subsequent treatment for postmenopausal women with hormone receptor-positive, HER2-negative advanced breast cancer who received ribociclib + letrozole vs placebo + letrozole in the phase III MONALEESA-2 study. *Cancer Res*. 2018;78:P5-21-18.
56. Bashour SI, Doostan I, Keyomarsi K, et al. Rapid breast cancer disease progression following cyclin dependent kinase 4 and 6 inhibitor discontinuation. *J Cancer*. 2017;8:2004–2009.
57. Mayer EL, Wander SA, Regan MM, et al. Abstract OT3-05-11: palbociclib after CDK inhibitor and endocrine therapy (PACE): a randomized phase II study of fulvestrant versus palbociclib plus fulvestrant, with and without avelumab, for CDK inhibitor pre-treated HR+/HER2- metastatic breast cancer. *Cancer Research*. 2018;78:OT3-05-11.
58. Rugo HS, Turner N, Chia S, et al. Abstract OT3-05-02: bYLieve: a phase 2 study of alpelisib with fulvestrant or letrozole for treatment of *PIK3CA* mutant, hormone receptor-positive (HR+), human epidermal growth factor receptor 2-negative (HER2-) advanced breast cancer (aBC) progressing on/after cyclin-dependent kinase (CDK)4/6 inhibitor therapy. *Cancer Res*. 2018;78:OT3–05–02.
59. Kalinsky K, Mundi PS, Chiuzan C, et al. Abstract OT3-05-09: a randomized phase II trial of fulvestrant with or without ribociclib after progression on aromatase inhibition plus cyclin-dependent kinase 4/6 inhibition in patients with unresectable or metastatic hormone receptor positive, HER2 negative breast cancer (Maintain trial). *Cancer Res*. 2018;78:OT3–05–09.
60. Bardia A, Hurvitz S, Yardley DA, et al. Abstract OT2-01-05: tRINITI-1: ribociclib + everolimus (EVE) + exemestane (EXE) triplet combination in men or postmenopausal women with HR+, HER2– advanced breast cancer (ABC) following progression on a cyclin-dependent kinase (CDK) 4/6 inhibitor. *Cancer Research*. 2017;77:OT2–01–05.
61. Liu MC, Cortes J, O'Shaughnessy J. Challenges in the treatment of hormone receptor-positive, HER2-negative metastatic breast cancer with brain metastases. *Cancer Metastasis Rev*. 2016;35:323–332.
62. Raub TJ, Wishart GN, Kulanthaivel P, et al. Brain exposure of two selective dual CDK4 and CDK6 inhibitors and the antitumor activity of CDK4 and CDK6 inhibition in combination with temozolomide in an intracranial glioblastoma xenograft. *Drug Metab Dispos*. 2015;43:1360–1371.

63. Patel Y. PDTB-12. CNS penetration of the CDK4/6 inhibitor Ribociclib (LEE011) in non-tumor bearing mice and mice bearing orthotopic pediatric brain tumors. *Neuro-Oncology*. 2016;18:vi152.
64. Tolaney SM, Lin NU, Thornton D, et al. Abemaciclib for the treatment of brain metastases (BM) secondary to hormone receptor positive (HR+), HER2 negative breast cancer. *J Clin Oncol*. 2017;35:1019.
65. Santa-Maria CA, Kumthekar P, Rademaker A, et al. A pilot study of palbociclib in patients with HER2-positive breast cancer with brain metastasis. *J Clin Oncol*. 2017;35:TPS1110-TPS1110.
66. Ciruelos E, Villagrasa P, Paré L, et al. Abstract P5-20-19: pAM50 intrinsic subtype predicts survival outcome in HER2-positive/hormone receptor-positive metastatic breast cancer treated with palbociclib and trastuzumab: a correlative analysis of the PATRICIA (SOLTI 13-03) trial. *Cancer Res*. 2018;78:P5–20–19.
67. Clark AS, Wang X, Troxel A, et al. Abstract P4-22-14: single agent palbociclib with or without trastuzumab for the treatment of Rb+ advanced breast cancer. *Cancer Res*. 2017;77:P4–22–14.
68. Gianni L, Bisagni G, Colleoni M, et al. Abstract P4-21-39: neo-adjuvant treatment with trastuzumab and pertuzumab associated with palbociclib and fulvestrant in HER2-positive and ER-positive breast cancer: effect on Ki67 during and after treatment. A phase II Michelangelo study. *Cancer Res*. 2017;77:P4–21–39.
69. Tolaney SM, Bourayou N, Goel S, Forrester T, André F. monarcHER: a phase 2 randomized open-label study of abemaciclib plus trastuzumab (T) with or without fulvestrant (F) compared to standard-of-care chemotherapy of physician's choice plus T in women with HR +, HER2+ advanced breast cancer. *Ann Oncol*. 2016;27:314TiP.
70. Spring L, Goel S, Juric D, et al. Trastuzumab emtansine (T-DM1) and ribociclib, an oral inhibitor of cyclin dependent kinase 4 and 6 (CDK 4/6), for patients with metastatic HER2-positive breast cancer: phase 1b clinical trial. *J Clin Oncol*. 2017;35:TPS1106-TPS1106.
71. Shagisultanova E, Diamond J, Stopeck A, et al. Abstract OT1-03-06: phase IB/II clinical trial to evaluate safety and efficacy of tucatinib in combination with palbociclib and letrozole in patients with hormone receptor positive and HER2-positive metastatic breast cancer. *Cancer Res*. 2018;78:OT1–03–06.
72. Metzger-Filho O, Mandrekar S, Loibl S, et al. Abstract OT3-05-07: pATINA: a randomized open label phase III trial to evaluate the efficacy and safety of palbociclib + anti HER2 therapy + endocrine therapy vs anti HER2 therapy + endocrine therapy after induction treatment for hormone receptor positive, HER2 positive metastatic breast cancer. *Cancer Res*. 2018;78:OT3–05–07.
73. Romashova T, Polasik A, Friedl TWP, et al. Abstract OT1-03-05: the DETECT V-Study – comparison of dual HER2-targeted therapy with trastuzumab plus pertuzumab in combination with chemo- or endocrine therapy in addition with CDK4/6 inhibition in patients with HER2-positive and hormone-receptor positive metastatic breast cancer. *Cancer Res*. 2018;78:OT1–03–05.
74. Juric D, Curigliano G, Cresta S, et al. Abstract P5-21-04: phase I/Ib study of the SERD LSZ102 alone or in combination with ribociclib in ER+ breast cancer. *Cancer Res*. 2018;78:P5–21–04.
75. Mayer EL, DeMichele AM, Guo H, et al. Abstract PD5-06: adjuvant palbociclib plus endocrine therapy for hormone receptor positive/HER2 negative breast cancer: a phase II feasibility study. *Cancer Research*. 2018;78:PD5–06.
76. Martin M, Hurvitz SA, Chan D, et al. Abstract PD5-01: final results of NeoMONARCH: a phase 2 neoadjuvant study of abemaciclib in postmenopausal women with hormone receptor positive (HR+), HER2 negative breast cancer (BC). *Cancer Res*. 2018;78:PD5–01.
77. Mayer EL, Demichele AM, Pfeiler G, et al. PALLAS: pALbociclib CoLlaborative adjuvant study: a randomized phase 3 trial of palbociclib with standard adjuvant endocrine therapy versus standard adjuvant endocrine therapy alone for HR1/HER2-early breast cancer. *Annals of Oncology*. 2017;28:v66.

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