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Inhibition of the Cyclin-Dependent Kinases (CDK) 4/6 as Therapy for Estrogen Receptor Positive Breast Cancer

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Abstract

Endocrine therapy (ET) is the usual first-line therapy for patients with hormone receptor-positive metastatic breast cancer (HR+MBC). However, resistance to ET frequently occurs during the course of treatment. Cellular pathways involved in cell proliferation are targets for new drugs that interfere with development of resistance to ET. Cyclin-dependent kinases (CDKs) are a subgroup of serine/threonine kinases that play a key role in regulating cell cycle progression. In this review, we discuss the currently approved and under investigation CDK 4/6 inhibitors, in addition to their preclinical data and clinical trials that demonstrated their benefit in the treatment of HR+ breast cancer.

Keywords: Breast Cancer; CDK 4/6 Inhibitors

Introduction

Given their proven efficacy and generally favorable toxicity profile, with the exception of patients with advanced visceral disease, most patients will receive endocrine therapies (ET) in the treatment of HR+MBC. Unfortunately, not all patients respond to first-line ET due to intrinsic resistance, while others may initially respond but eventually progress with secondary acquired resistance leading to disease progression and endocrine resistance [1]. Mechanisms of resistance to anti-estrogen therapy include among others, estrogen receptor (ER) loss over time in the tumor which occurs in about 20% of patients treated with ET, acquired mutations in ERα (ESR1) and constitutive activation cyclin-dependent kinases (CDK) 4 and 6 [2].

Cell cycle regulation is identified as an attractive target for targeted drug therapy. Cyclin D1, a cell cycle proto-oncogene, and its binding to CDK 4/6, promotes G1-to-S phase transition by phosphorylating the retinoblastoma protein (Rb), which releases the E2F transcription factor and activates downstream target genes [3]. Given their kinase activity, the cyclin dependent kinases have been pursued as drug targets [4] CDK 4/6 inhibitors prevent the cyclin D-CDK4/6 complex phosphorylation of Rb required for the commitment to S-phase and ultimately, cellular mitosis. Selective inhibition of CDK 6 has been reported to have a role in anti-angiogenesis as well [5]. An additional suggested mechanism of action for the novel CDK 4/6 inhibitor, palbociclib, is decreasing the expression of cyclooxygenase-II (COX-II), an enzyme associated with the epithelial-mesenchymal transition (EMT) in metastasis [6]. CDK 4/6 inhibition also results in un-phosphorylation of Forkhead Box M1 (FOXM1), a transcription factor involved in the expression of genes that upregulate proliferative capacity [7]. Preclinical data in melanoma cell lines, for example, have shown that pharmacological CDK 4/6 inhibition led to degradation of FOXM1 transcription factor and resulted in subsequent phenotypic expression of cellular senescence [8].

Growth-promoting agents such as estrogen up-regulate cyclin D gene expression [9]. Furthermore, CDK4/6 is particularly activated in ER+ breast cancer via the ER, along with other oncogenic signaling pathways [10]. After disappointing results of first and second generation CDK inhibitors, mainly due to low single agent efficacy and increased toxicity [11], the development of specific CDK 4/6 inhibitors has produced results never before seen in the treatment of breast cancer (BC) [12] **(Table 1).**

Agents Approved and Under Investigation

CDK 4/6 inhibitors, including palbociclib, abemaciclib, and ribociclib, are being investigated in both the preclinical and clinical settings; some are becoming available for treatment of HR+MBC. These potent, ATP-competitive CDK 4/6 selective inhibitors are orally administered and have little to no function on other CDK enzymes, even at clinical doses. Selectivity for the CDK 4/6 proteins has been shown to be important in mitigating cytotoxic effects that were highlighted in the pan-CDK inhibitor predecessors. CDK1 inhibition, for example, causes arrest in M-phase, leading to toxicities such as myelosuppression [7]. Preclinical data for the novel selective inhibitors demonstrated over 1000-fold less potency for CDK1 vs. CDK4/6 in palbociclib and ribociclib, and over 160-fold less potency in abemaciclib [7,13,14]. Furthermore, Rb-depleted ER+ human BC cell lines displayed no anti-growth effects for

palbociclib; loss of Rb expression is suggested as a mechanism for CDK 4/6 inhibitor resistance development [14]. Gelbert et al. also confirmed this property being a requirement for abemaciclib when evaluating abemaciclib effects on Rb-proficient and Rb-deficient BC cell lines [15].

Previous *in vitro* studies of palbociclib confirmed growth inhibition sensitivity of human luminal ER+ and HER2-amplified breast cancer lines to the agent. Finn et al. evaluated forty-seven lines of the aforementioned BC subgroups and demonstrated G0/G1 phase arrest via cell cycle analysis and inhibited Rb phosphorylation in sensitive subtypes using Western blot analysis. Elevated Rb was found in this cell line subtype. When combined with tamoxifen in ER+ lines, synergistic growth inhibition was observed for the three line subtypes that were evaluated (CIm < 1). In three additional HER2-amplified BC lines targeted in the study, palbociclib with trastuzumab proved to act synergistically as well (CIm < 1). Tamoxifen-resistant subgroups were examined for response to palbociclib too. The MCF7 resistant line was sensitive to palbociclib monotherapy and demonstrated increased sensitivity to tamoxifen-palbociclib combination, albeit not at the level of parental non-resistant lines to tamoxifen alone [16].

Palbociclib was the first of the novel selective CDK 4/6 inhibitors to gain FDA approval for use in HR+MBC; it did so by demonstrating activity in the phase II open label randomized PALOMA 1 clinical trial. In this study, 165 postmenopausal women with advanced ER+ and HER2 negative BC, who had not received any systemic treatment for their advanced disease were randomly assigned in a 1:1 to receive continuous letrozole or letrozole daily plus oral palbociclib 125 mg, given once daily for 3 weeks followed by 1 week off over 28-day cycles. The major efficacy outcome measure was investigator-assessed progression free survival (PFS) of 10.2 months for the letrozole group and 20.2 months for the palbociclib plus letrozole group (p=0·0004) [17]. These results were confirmed in the larger phase III PALOMA-2 clinical trial. This study randomized 666 patients 2:1 to receive the same dose and frequency of letrozole or letrozole plus palbociclib. Patients in the palbociclib containing arm experienced a PFS of 24.8 months in comparison to the control arm of 14.5 months (p<0.000001). Objective response rate (ORR) was also higher with palbociclib with 55.3% of patients who had measurable disease experiencing a reduction in size vs 44% (P=0.013). In terms of side effects, neutropenia (79.5 vs 6.3%), fatigue (37.4 vs. 27.5%), and nausea (35.1% vs 26.1%) were more noticeable in the investigational arm; neutropenic fever was only seen in 2.5% of the patients [18].

Similarly, in the second line setting, the PALOMA-3 double-blind phase III clinical trial randomized 427 patients whose disease had progressed within 12 months of adjuvant therapy or within one month of ET for HR+/HER2 negative MBC to receive palbociclib plus fulvestrant versus fulvestrant plus placebo. In conjunction with study treatment, premenopausal and perimenopausal women were required to take goserelin. The study met the primary endpoint, PFS which was 9.2 months in the palbociclib and fulvestrant arm versus 3.8 months in the fulvestrant and placebo arm (p< 0.001) [19].

In the neoadjuvant setting, a phase II clinical trial evaluated the early introduction of CDK 4/6 inhibitors in the treatment of BC. This is based on the observation that decreasing Ki 67 by complete cell cycle arrest, could have a positive effect on long term outcomes [20]. The study included patients with clinical stage II/III HR+, HER2 negative breast cancer. Patients received palbociclib and anastrozole for a four month period and a proportion of patients was kept on palbociclib up until surgery; serial biopsies were performed. The primary end point was complete cell-cycle arrest, which was defined as a proportion of tumor cells positive for Ki 67 ≤ 2.7% on cycle 1, day 15 after 2 weeks of treatment with both drugs. Of the 45 evaluable patients 87% experienced complete cell-cycle arrest at cycle 1, day 15. Clinical responses were observed in 67%. Patients tended to have a rebound in Ki67 level in the washout period; however, this increase was not observed in patients who continued palbociclib [21].

The results from another of these drugs, ribociclib were recently reported in the MONALEESA-2 Phase III randomized, double blind, placebo controlled, and multicenter global registration trial. The study randomized 668 postmenopausal women with HR+/HER2 negative advanced BC in a 1:1 stratified by the presence of liver and/or lung metastases. Patients received ribociclib 600 mg/daily (three weeks on and one week off), or placebo, in combination with letrozole 2.5 mg/daily. The first interim analysis showed a 44% improvement in median PFS and has not been yet reached at the data cut-off over 14.7 months seen in the placebo arm (p = 0.00000329). As seen with other CDK4/6 inhibitors there is a significantly higher objective response rate when combined with aromatase-inhibitors (AIs) (53% vs. 37%; p=0.00028). Overall survival results are currently pending. This agent is also associated with neutropenia, which occurred in 59% of patients in the ribociclib arm compared to 1% of the placebo arm; leukopenia occurred in 21% vs 1% while increased alanine aminotransferase and aspartate aminotransferase were associated with combined therapy as well (9% vs. 1% and 5.7% vs. 1.2%, respectively) Despite the increased adverse event frequency, only 7.5% of patients in the investigational arm required permanent discontinuation of combined therapy [22]. The MONALEESA-3 trial is evaluating ribociclib in combination with fulvestrant compared to fulvestrant alone in men and postmenopausal women with HR+/HER2 negative MBC in the second line ET setting [23].

The third CDK 4/6 inhibitor in development Abemaciclib has demonstrated single agent activity as reported in the MONARCH 1 phase II single arm study where 132 patients received abemaciclib monotherapy 200 mg every 12 hours until progression of disease. Patients had a median of 3 lines of prior therapy for advanced disease, including a median of 2 lines of chemotherapy, 90.2% had visceral disease. At the 8 month interim the confirmed ORR (per RECIST v1.1) was 17.4%, the clinical benefit rate (CBR) defined as objective response or stable disease for ≥24 weeks was 42.4%, and median PFS was 5.7 months [24]. MONARCH 3 is a Phase III

trial of abemaciclib in combination with anastrozole in patients with HR+/HER2 negative locoregionally recurrent or MBC [25].

The neoMONARCH phase II trial evaluated abemaciclib in combination with anastrozole in the neoadjuvant setting. In this study 173 women were randomized to receive abemaciclib plus anastrozole (n = 56), abemaciclib monotherapy (n = 58) or anastrozole monotherapy (n = 59) for the first two weeks. At the conclusion of that regimen, all patients underwent a second core biopsy and then subsequently received the abemaciclib–anastrozole combination for 14 weeks. Abemaciclib was administered in 150 mg oral doses every 12 hours, and anastrozole was administered in 1 mg oral doses daily. Patients also received loperamide as primary prophylaxis with each abemaciclib dose. The percentage of Ki67 responders, defined as patients with Ki67 levels < 2.7% at week 2, was higher among those assigned the combination (69.6%) and abemaciclib monotherapy (68.4%) than anastrozole alone (22.7%), radiographic response rate (RR) was 54.7%. [26]. Additionally, the monarcHER phase II trial is evaluating abemaciclib plus trastuzumab (with or without fulvestrant) in women with HR+/HER2 positive locally advanced or MBC [27].

Table 1 Selected clinical trials of CDK 4/6 inhibitors.

Study/reference	Design	Treatment	N	Results
Paloma 1 [8]	Phase II MBC 1st line	letrozole-palbociclib vs. letrozole	165	Improvement in PFS from 10.2 to 20.2 m
Paloma 2 [9]	Phase III MBC 1st line	letrozole-palbociclib vs. letrozole	666	Improvement in PFS from 14.5 to 24.8 m
Paloma 3 [10]	Phase III MBC pretreated	Fulvestrant palbociclib vs. fulvestrant	427	Improvement in PFS from 3.8 to 9.2 m
Ma et al. [12]	Phase II Neoadjuvant	anastrozole-palbociclib	45	87% complete cell-cycle arrest at cycle 1, day 15. Clinical RR 67%
MONALEESA-2 [13]	Phase III MBC 1st line	letrozole-ribociclib vs. letrozole	668	44% improvement in PFS 14.7 months .vs NR for ribociclib
MONARCH-1 [15]	Phase II MBC (heavily-pretreated)	abemaciclib	132	ORR 17.4%, CB 42.4%, PFS 5.7 m
neoMONARCH [17]	Phase II Neoadjuvant	abemaciclib + anastrozole vs. abemaciclib vs. anastrozole	173	Ki67 < 2.7% at week 2. combination (69.6%); abemaciclib (68.4%); anastrozole (22.7%). RR 54.7%
Abbreviations: m: Months; PFS: Progression Free Survival; TTP: Time to Progression; ORR: Objective Response Rate; RR: Response Rate; CB: Clinical Benefit				

Conclusion

ER-targeted therapy is important for many women with breast cancer, but resistance to therapy inevitably occurs. The ER signaling pathway is a complex network of extensive crosstalk with growth-factor signaling pathways, cell cycle control pathways, and protein degradation pathways. These pathways provide many alternative targets for agents that may be useful in combination with ET to decrease resistance to treatment and to extend benefit to patients who do not achieve optimal benefit from ET alone.

With the development of CDK 4/6 inhibitors, significant improvement in PFS has been documented in each MBC clinical trial where these agents have been combined with antiestrogen therapies. However, it is unknown if there is benefit to the continuation of CDK 4/6 inhibitors following progression. Recent neoadjuvant studies have also demonstrated clinical activity when introducing these agents earlier. Clinical comparison of CDK 4/6 inhibitors have not been studied for palbociclib vs. ribociclib as well.

The future looks very promising for the ET of patients with MBC, with unprecedented PFS findings on recent trials; it is likely that the overall survival of patients will continue to improve overtime. Part of the success of these agents is in overcoming intrinsic resistance of cancer and preventing acquired resistance over time. However, the question remains on which patients are these drug combinations needed, as adding these agents to endocrine therapy increases toxicity. Biomarkers that predict the benefit of these agents are greatly needed.

References

1. Lumachi F, Luisetto G, Basso SM (2011) Endocrine therapy of breast cancer. Curr Med Chem 18: 513-522.
2. Osborne CK, Schiff R (2011) Mechanisms of endocrine resistance in breast cancer. Annu Rev Med 62: 233-247.

3. Miller TW, Balko JM, Fox EM (2011) ERα-dependent E2F transcription can mediate resistance to estrogen deprivation in human breast cancer. Cancer Discov 1: 338-351.

4. Shapiro GI (2006) Cyclin-dependent kinase pathways as targets for cancer treatment. J Clin Oncol 24: 1770–1783.

5. Ehab M, Elbaz M (2016) Profile of palbociclib in the treatment of metastatic breast cancer. Breast cancer (Dove Medical Press) 8: 83-91.

6. Qin G, Xu F, Qin T (2015) Palbociclib inhibits epithelial-mesenchymal transition and metastasis in breast cancer via c-Jun/COX-2 signaling pathway. Oncotarget 6: 41794-41808.

7. VanArsdale T, Boshoff C, Arndt KT, Abraham RT (2015) Molecular pathways: Targeting the cyclin D-CDK4/6 axis for cancer treatment. Clinical cancer research: an official journal of the American Association for Cancer Research 21: 2905-2910.

8. Anders L, Ke N, Hydbring P (2011) A systematic screen for CDK4/6 substrates links FOXM1 phosphorylation to senescence suppression in cancer cells. Cancer cell 20: 620-634.

9. Klein EA, Assoian RK (2008) Transcriptional regulation of the cyclin D1 gene at a glance. Journal of Cell Science 121: 3853-3857.

10. Dean JL, Thangavel C, McClendon AK (2010) Therapeutic CDK4/6 inhibition in breast cancer: key mechanisms of response and failure. Oncogene 29: 4018–4032.

11. Santo L, Siu KT, Raje N (2015) Targeting cyclin-dependent kinases and cell cycle progression in human cancers. Semin Oncol 42: 788-800.

12. Turner NC, Ro J, André F (2015) Palbociclib in hormone-receptor-positive advanced breast cancer. N Engl J Med 373: 209–219.

13. Toogood PL, Harvey PJ, Repine JT (2005) Discovery of a potent and selective inhibitor of cyclin-dependent kinase 4/6. Journal of medicinal chemistry 48: 2388-2406.

14. Fry DW, Harvey PJ, Keller PR (2004) Specific inhibition of cyclin-dependent kinase 4/6 by PD 0332991 and associated antitumor activity in human tumor xenografts. Molecular cancer therapeutics 3: 1427-1438.

15. Gelbert LM, Cai S, Lin X (2014) Preclinical characterization of the CDK4/6 inhibitor LY2835219: in-vivo cell cycle-dependent/independent anti-tumor activities alone/in combination with gemcitabine. Investigational new drugs 32: 825-837.

16. Finn RS, Dering J, Conklin D (2009) PD 0332991, a selective cyclin D kinase 4/6 inhibitor, preferentially inhibits proliferation of luminal estrogen receptor-positive human breast cancer cell lines in vitro. Breast cancer research: BCR 11: 77.

17. Finn RS, Crown JP, Lang I (2015) 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 16: 25–35.

18. Finn RS, Martin M, Rugo HS (2016) PALOMA-2: Primary results from a phase III trial of palbociclib (P) with letrozole (L) compared with letrozole alone in postmenopausal women with ER+/HER2– advanced breast cancer. J Clin Oncol 34.

19. Turner NC, Ro J, Andre F (2015) PALOMA3: A double-blind, phase III trial of fulvestrant with or without palbociclib in pre- and post-menopausal women with hormone receptor-positive, HER2-negative metastatic breast cancer that progressed on prior endocrine therapy. Abstract J Clin Oncol 33.

20. Montagna E, Bagnardi V, Viale G (2015) Changes in PgR and Ki-67 in residual tumour and outcome of breast cancer patients treated with neoadjuvant chemotherapy. Ann Oncol 26: 307-313.

21. Ma CX, Gao F, Northfelt D (2015) A phase II trial of neoadjuvant palbociclib, a cyclin-dependent kinase 4/6 inhibitor, in combination with anastrozole for clinical stage 2 or 3 estrogen receptor positive HER2 negative breast cancer. SABCS S6-05.

22. Hortobagyi GN, Stemmer SM, Burris HA (2016) First-line ribociclib + letrozole for postmenopausal women with hormone receptor-positive (HR+), HER2-negative (HER2–), advanced breast cancer (ABC). ESMO.

23. MONALEESA-3 (2016) Study of efficacy and safety of LEE011 in men and postmenopausal women with advanced breast cancer. (MONALEESA-3) NCT02422615.

24. Tolaney SM, Rosen LS, Beeram M (2015) Clinical activity of abemaciclib, an oral cell cycle inhibitor, in metastatic breast cancer. Cancer Res 75: 19-13.

25. MONARCH3 (2016) A Study of Nonsteroidal Aromatase Inhibitors Plus Abemaciclib (LY2835219) in Postmenopausal Women With Breast Cancer (MONARCH3) NCT02246621.

26. Hurvitz S, Abad MF, Rostorfer R (2016) Interim results from neoMONARCH: A neoadjuvant phase II study of abemaciclib in postmenopausal women with HR + /HER2- breast cancer (BC). Ann Oncol 27.

27. MONARCHER (2016) A Study of Abemaciclib (LY2835219) in Women With HR+, HER2+ Locally Advanced or Metastatic Breast Cancer. (MONARCHER) NCT02675231.