Editorial

# CDK inhibitors as potential breast cancer therapeutics: new evidence for enhanced efficacy in ER+ disease

Robert L Sutherland[1,2] and Elizabeth A Musgrove[1,2]

[1]Cancer Research Program, Garvan Institute of Medical Research, 384 Victoria Street, Darlinghurst, Sydney, NSW 2010, Australia
[2]St Vincent's Clinical School, Faculty of Medicine, University of New South Wales, Sydney, NSW 2052, Australia

Corresponding author: Robert L Sutherland, r.sutherland@garvan.org.au

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## Abstract

Loss of cell cycle control is a hallmark of cancer, and aberrations in the cyclin-CDK-RB (cyclin-dependent kinase-retinoblastoma protein) pathway are common in breast cancer. Consequently, inhibition of this pathway is an attractive therapeutic strategy, but results from clinical trials of CDK inhibitors in breast cancer have been disappointing. A recent study now shows that in cell culture a selective CDK4/6 inhibitor is preferentially effective in estrogen receptor-positive (ER+) disease and apparently acts synergistically with tamoxifen or trastuzumab. These exciting new preclinical data set the scene for a more targeted approach to further clinical evaluation wherein this class of drugs is targeted to subgroups of ER+ patients, including those with resistance to endocrine therapy, alone or in combination with current standard therapies.

## Introduction

Almost as soon as it became clear that cyclin activation of cyclin-dependent kinases (CDKs) is pivotal to the control of cell cycle progression, investigations began into the likely role of cyclins and CDKs in cancer, both in the development and progression of the disease and as therapeutic targets. The article by Finn and colleagues [1] in the previous issue of *Breast Cancer Research* provides the most recent insight into targeting this basic cell cycle regulatory mechanism in the context of breast cancer therapy.

Cyclin D1 was the first mammalian $G_1$ cyclin identified and is now firmly established as a mammary oncogene [2]. Amplification at 11q13, the locus of the *CCND1* gene encoding cyclin D1, occurs in 15% to 20% of breast cancers, and cyclin D1 overexpression is even more common (up to 50% of breast cancers) [2,3]. Accumulating evidence that inhibiting the activity of CDKs may be an effective therapy in cancers, including breast cancer, led to the development of small molecules that specifically target subgroups of CDKs, including CDK4 and CDK6, the kinases activated by cyclin D1 [4].

Though well tolerated, CDK inhibitors that have entered clinical trials have been of limited efficacy except in haematological malignancies [5]. One reason for this disappointing outcome is that early CDK inhibitors frequently targeted CDK2 and many (though not all) cancer cells are refractory to CDK2 inhibition [6,7]. However, cells that continue to proliferate despite CDK2 inhibition are arrested by CDK4 inhibition *in vitro* [6]. Thus, more selective CDK inhibitors, and identification of cancer subtypes that are likely to be susceptible to CDK inhibition, are needed to clarify the degree to which CDK inhibition, alone or in combination with other therapeutic approaches, may be useful clinically. The publication by Finn and colleagues [1] is pivotal in this context as it addresses this need by investigating predictors of response to the CDK4/6-specific inhibitor PD 0332991 in a panel of 41 immortalised breast epithelial and breast cancer cell lines representative of the major subtypes of breast cancer.

## Determinants of response to CDK4/6 inhibition

PD 0332991 is highly selective for inhibition of CDK4 and CDK6 [8] and is being tested in ongoing clinical trials in myeloma and breast cancer [5]. It causes a specific cell cycle arrest in $G_1$ phase and inhibits proliferation in cultured and xenografted leukaemia, myeloma, breast cancer, colon cancer, and lung cancer cells [8,9]. This is accompanied by decreased phosphorylation of the CDK4/6 substrate retinoblastoma protein (RB), and cell lines lacking RB, and therefore not dependent on cyclin D1-CDK4/6 for proliferation, are resistant to PD 0332991 treatment [8,9]. Thus, CDK4/6 inhibition appears to be the primary mechanism for PD 0332991 inhibition of proliferation.

CDK = cyclin-dependent kinase; ER = estrogen receptor; $IC_{50}$ = half inhibitory concentration; RB = retinoblastoma protein.

Finn and colleagues [1] compared baseline gene expression profiles from 21 cell lines highly sensitive to PD 0332991 (half inhibitory concentration [$IC_{50}$] <150 nM) and 12 resistant cell lines ($IC_{50}$ >1 μM) and identified 450 differentially expressed genes. Estrogen receptor-positive ($ER^+$) cell lines, including those with *HER2* amplification, were the most sensitive, and there was significant overlap between the gene set associated with PD 0332991 sensitivity and that which distinguishes breast cancer subtypes [1]. However, the PD 0332991 sensitivity signature also included genes that are not part of the subtype signature but are components of the cyclin D1-RB pathway (for example, *RB1*, *CCND1*, and *CDKN2A*, which encodes $p16^{INK4A}$, an endogenous inhibitor of CDK4 and CDK6). Interestingly, some cell lines were insensitive despite the presence of RB and failed to downregulate RB phosphorylation following PD 0332991 treatment. Comparison of measures of RB pathway activity (for example, expression of the RB-responsive gene set [10]) with response to PD 0332991 may give some insight into whether this insensitivity arises through functional inactivation of RB and consequent loss of dependence on cyclin D1-CDK4/6.

Although to date CDK inhibitors have not been particularly effective in the clinic as single agents, combination studies have yielded more promising results [5]. Consequently, Finn and colleagues tested whether there was any interaction between PD 0332991 and therapies commonly used in the subtypes of breast cancer most sensitive to CDK4/6 inhibition: tamoxifen and trastuzumab. In both cases, the combination synergistically inhibited proliferation [1], and so the results of an ongoing clinical trial of breast cancer treated with PD 0332991 in combination with letrozole [5], an aromatase inhibitor that, like tamoxifen, targets the ER signalling pathway, will be of particular interest. Synergy between PD 0332991 and tamoxifen was also apparent in a tamoxifen-insensitive cell line [1], raising the possibility that CDK4/6 inhibitors like PD 0332991 may be useful in endocrine-resistant breast cancers, which are not well served by current targeted therapies [11]. This latter result is somewhat unexpected given that cyclin D1-CDK4 is a major target of tamoxifen-mediated growth arrest [12] and that RB inactivation causes tamoxifen resistance [10]. Thus, the mechanistic basis for these apparent synergies requires further detailed investigation.

## Conclusions

The study of Finn and colleagues has important implications for the further clinical evaluation of PD 0332991 and other specific CDK4/6 inhibitors in breast cancer. First, it identifies a subgroup of patients most likely to benefit: the $ER^+$ luminal subtype. This is perhaps counterintuitive in that these are not the highly proliferative phenotype but are the subgroup enriched for cyclin D1 overexpression [2,3]. Second, it identifies potential synergy with standard best practice therapies (that is, tamoxifen and trastuzumab), which raises the questions of potential interactions with aromatase inhibitors and the efficacy of CDK4/6 inhibitors in endocrine- and trastuzumab-resistant $ER^+$ disease. Finally, this study emphasises the importance of strong preclinical data and the identification of potential therapeutic response parameters (in this case, ER, cyclin D1, and a gene expression signature) in targeting therapeutic trials to the most appropriate patient subgroups. Further preclinical and clinical data on this class of agents in breast cancer are awaited with interest.

## Competing interests

The authors declare that they have no competing interests.

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## References

1. Finn RS, Dering J, Conklin D, Kalous O, Cohen DJ, Desai A, Ginther C, Atefi M, Chen I, Fowst C, Los G, Slamon DJ: **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 Res* 2009, **11:**R77.
2. Arnold A, Papanikolaou A: **Cyclin D1 in breast cancer pathogenesis.** *J Clin Oncol* 2005, **23:**4215-4224.
3. Sutherland RL, Musgrove EA: **Cyclins and breast cancer.** *J Mammary Gland Biol Neoplasia* 2004, **9:**95-104.
4. Lee YM, Sicinski P: **Targeting cyclins and cyclin-dependent kinases in cancer: lessons from mice, hopes for therapeutic applications in human.** *Cell Cycle* 2006, **5:**2110-2114.
5. Dickson MA, Schwartz GK: **Development of cell-cycle inhibitors for cancer therapy.** *Curr Oncol* 2009, **16:**36-43.
6. Tetsu O, McCormick F: **Proliferation of cancer cells despite CDK2 inhibition.** *Cancer Cell* 2003, **3:**233-245.
7. Du J, Widlund HR, Horstmann MA, Ramaswamy S, Ross K, Huber WE, Nishimura EK, Golub TR, Fisher DE: **Critical role of CDK2 for melanoma growth linked to its melanocyte-specific transcriptional regulation by MITF.** *Cancer Cell* 2004, **6:**565-576.
8. Fry DW, Harvey PJ, Keller PR, Elliott WL, Meade M, Trachet E, Albassam M, Zheng X, Leopold WR, Pryer NK, Toogood PL: **Specific inhibition of cyclin-dependent kinase 4/6 by PD 0332991 and associated antitumor activity in human tumor xenografts.** *Mol Cancer Ther* 2004, **3:**1427-1438.
9. Baughn LB, Di Liberto M, Wu K, Toogood PL, Louie T, Gottschalk R, Niesvizky R, Cho H, Ely S, Moore MA, Chen-Kiang S: **A novel orally active small molecule potently induces $G_1$ arrest in primary myeloma cells and prevents tumor growth by specific inhibition of cyclin-dependent kinase 4/6.** *Cancer Res* 2006, **66:**7661-7667.
10. Bosco EE, Wang Y, Xu H, Zilfou JT, Knudsen KE, Aronow BJ, Lowe SW, Knudsen ES: **The retinoblastoma tumor suppressor modifies the therapeutic response of breast cancer.** *J Clin Invest* 2007, **117:**218-228.
11. Musgrove EA, Sutherland RL: **Biological determinants of endocrine resistance in breast cancer.** *Nat Rev Cancer* 2009, **9:**631-643.
12. Watts CK, Brady A, Sarcevic B, deFazio A, Musgrove EA, Sutherland RL: **Antiestrogen inhibition of cell cycle progression in breast cancer cells in associated with inhibition of cyclin-dependent kinase activity and decreased retinoblastoma protein phosphorylation.** *Mol Endocrinol* 1995, **9:**1804-1813.