Anti-cancer Role of Phosphodiesterase-5 Inhibitors

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1 Introduction

Phosphodiesterase-5 (PDE5) is an enzyme that hydrolyzes the 3’, 5’-phosphodiester bond in the second messenger molecule cGMP to 5’-GMP, which is biologically inactive. The PDE family includes 11 members based on tissue localization, substrate specificity, inhibitor sensitivity and regulatory properties. However, over 100 distinct PDE isoforms or splice variants may be expressed in humans based on alternative mRNA splicing, of multiple genes expression, and post-translational protein modifications (Beavo, 1995; Lugnier, 2006). It is known that certain PDEs are highly specific for hydrolysis of cAMP (PDEs 4, 7, and 8) or cGMP (PDEs 5, 6 and 9), and others hydrolyze both cAMP and cGMP (PDEs 1, 2, 3, 10, and 11) (Bender et al., 2006). PDE5 was originally identified in platelets (Hamet et al., 1978), and was subsequently shown to specifically hydrolyze cGMP and to have high affinity binding sites for cGMP(Coquil et al., 1980). Only one gene for PDE5 has been discovered, PDE5A, although it has three variants (PDE5A1-PDE5A3) (Lin et al., 2000a, 2000b). The PDE5 protein is a homodimer and the subunits are ~100 kDa and each one contains an N-terminal regulatory domain and a conserved C-terminal catalytic domain. Within the regulatory domain, there are two tandem GAF (cGMP-binding PDEs, Adenylyl cyclase, Fh1A) subdomains (Thomas et al., 1990a). The binding of cGMP to the GAF subdomain stimulates phosphorylation of the enzyme by cGMP-dependent protein kinase (PKG), leading to an increase in catalytic activity (Okada et al., 2002; Thomas et al., 1990b). Key aspects of cellular activity and gene expression may be regulated by cGMP-induced PKG activation, in tandem with cGMP binding PDEs and cyclic nucleotide gated ion channels, resulting in cyclic nucleotide hydrolysis, protein phosphorylation, or ionic fluxes (Lincoln et al., 1993). It has been reported that besides its prominent expression in the corpus cavernosum, PDE5 is also expressed in smooth muscles of the systemic vasculature, prostate, heart, brain, lungs and platelets (Sandner et al., 2007). These tissues contain significant amounts of cGMP signaling entities such as guanylylcyclase (GC), nitric oxide (NO) synthase, atrial natriuretic peptide (ANF) receptors, PKG and specific cGMP-gated ion channels. It has been shown that PDE5 inhibition increases intracellular cGMP levels and this activates the cGMP cascade reactions. For example, cGMP produces PKG activation and decreases intracellular calcium levels, thereby promoting relaxation of smooth muscle cells, as well as affecting many other calcium-dependent processes in these tissues (Lucas et al., 2000). Currently, there are a number of PDE5 inhibitors that are clinically used to treat erectile dysfunction, such as sildenafil, vardenafil and tadalafil. Table 1 summarizes the PDE5 inhibitors that have been approved for clinical use, as well as those that are undergoing clinical trials(Palit et al., 2010). A number of PDE5 inhibitors are being tested for other medical conditions such as pulmonary arterial hypertension (PAH), heart failure, Raynaud's disease, stroke, benign prostate hyperplasia (BPH) among others (Sandner, et al., 2007). The wide and safe use of PDE5 inhibitors, together with an increasing understanding of physiological role of PDE5 in various tissues, have triggered a number of attempts to find novel functions and applications for these drugs. Recently, it has been reported that PDE5 expression is increased in various types of cancers compared to normal or surrounding non-cancerous specimens. In addition, several groups have reported that PDE5 inhibitors can enhance the sensitivity of certain cancer cells to standard chemotherapeutic drugs (Zhu et al., 2007). The aim of this chapter is to review and discuss recent findings in this field and highlight possible future therapeutic uses of PDE5 inhibitors in cancer chemotherapy.
2  Anti-cancer Effects of PDE5 Inhibitors

Accumulating evidence suggests that modulating the intracellular secondary messengers, cAMP and cGMP, may be a promising approach in the field of anti-cancer therapy. It is well known that certain hormones and neurotransmitters can activate GPCR that are positively coupled to the enzyme adenylyl cyclase, which catalyzes the conversion of ATP (in the presence of Mg$^{2+}$) to cAMP. Cyclic AMP can produce its effects primarily via activation of protein kinase A (PKA), cAMP-activated exchange factors (EPAC) and cAMP-gated ion channels. The differential regulation of cAMP, PKA and EPAC has been linked to certain PDEs (1, 2, 3, 4, 7, 8 and 10) inhibitors. Whereas cyclic GMP can produce its effects through PKG and its regulation has been linked PDEs (1, 2, 3, 5, 6, 9 and 11). The inhibitors of PDEs 1-11 have been shown to modulate the process mediated by these important cyclic nucleotides. For the purpose of this chapter, the following section will discuss the possible mechanisms by which PDE5 inhibitors could produce anti-cancer effects. This includes regulation of cGMP-PKG levels, PDE5 expression and modulation of host immunity by PDE5 inhibitors (see Figure 1).

2.1  Regulating cGMP and PKG Levels

Natriuretic peptides (NP) and NO stimulate soluble guanylylcyclase (sGC) to generate cGMP. Subsequently, cGMP can bind to and activate GMP-gated ion channels and cGMP-dependent PKG. PKG is normally involved in activating specific proteins that modulate the contractile activity of smooth muscle cells. Interestingly, accumulating evidence suggests that cancer cells express lower levels of PKG compared to normal tissue (Hou et al., 2006; Yamanaka et al., 2002). In addition, xenograft models in nude mice have shown an increased expression of PKG in human colon cancer cells can lead to decreased invasiveness and tumor growth (Yamanaka, et al., 2002). Similarly, cGMP levels are significantly reduced in the cancer cells and tissues compared with their normal counterparts (Thompson et al., 2000). The biochemical basis for this effect could be due to decreased levels of NO synthase, NP, sGC or particulate guanylatecyclase or possibly an increase in cGMP metabolism by PDEs. The increase in cGMP levels either by NP, 1-benzyl-3-(5'-hydroxymethyl-2'-furylindazole (YC-1), an NO donor, or the GC-activator sNOR-3, uroguanylin/guanylin, have been shown to induce apoptosis, inhibit migration and decrease cell growth in colon cancer cells (Thompson, et al., 2000). A recent study showed that increased activation of PKG by cGMP produces apoptosis and growth arrest in breast cancer MCF-7 and MDA-MB-468 cells (Fallahian et al., 2011). PDE inhibitors such as sulindac sulfide, trequinsin and MY5445, which inhibit PDE5, inhibit the growth of breast cancer cells (Tinsley et al., 2009). Sulindac sulfide specifically inhibited cGMP hydrolysis and activated PKG to produce apoptosis in breast cancer cells, but had no effect on normal human mammary epithelial cells (Tinsley, et al., 2009). Furthermore, it has been recently reported that the inhibition of cGMP-induced PKG activation by specific PKG inhibitors such as KT5823 and Rp-8-pCPT-cGMP also inhibited the growth of breast cancer cells via induction of apoptosis regardless of their estrogenstatus (Fallahian, et al., 2011). Since PDE5 inhibitors increase cGMP by preventing their metabolism by PDEs, it is possible that the anti-cancer action of these drugs may be partly attributed to cGMP regulated downstream pathways. The induction of apoptosis by the PDE5 inhibitors is a complicated process. For example, cGMP activates c-Jun NH2-terminal kinase (JNK) (Soh et al., 2000), inhibits extracellular-signal regulated kinases 1/2 (ERK1/2) (Rice et al., 2004) and regulates p42/p44 mitogen activated-protein kinase (MAPK) and p21. As hypothesized in Figure 1, cGMP activated PKG could ac-
Figure 1: Proposed mechanisms for the anti-cancer role of PDE5 inhibitors - A schematic model. PDE5 inhibitors may produce anti-cancer activity by 1) blocking the substrate efflux activity of multidrug resistant transporters such as ABCC4, ABCC5, ABCC10, ABCB1 and ABCG2 and/or 2) increasing cGMP levels via their inhibition of PDE5 activity, ABCC4- and ABCC5-mediated efflux of cGMP, thereby increasing the likelihood of cGMP-induced activation of PKG, which produces growth suppression or apoptosis. In addition, PDE5 inhibition also leads to apoptosis through increased phosphorylation of β-catenin and/or MEKK1/SEK1/JNK1 signaling pathways by increased PKG activation through cGMP induction. ANP/BNP, Atrial natriuretic peptides/brain natriuretic peptides; cGMP, cyclic guanosine monophosphate; ERK1/2, extra-cellular regulated kinases ½; JNK1, c-Jun N-terminal kinase 1; MAPK, mitogen-activated protein kinase; MEKK1, MAP kinase/ERK kinase kinase; PDE5, Phosphodiesterase type 5; PKG, Protein kinase G; SEK1, stress-activated protein kinase/extracellular signal-regulated kinase kinase.
tivate the JNK1 pathways via phosphorylation of mitogen-activated protein kinase kinasekinase 1 (MEKK1) (Soh et al., 2001), decrease the expression of Wnt/β-catenin (Tinsley et al., 2011) and down-regulate cyclin D1 (Li et al., 2002), and all these processes produce caspase-dependent apoptosis and cell growth arrest. On the contrary, increased levels of cGMP were observed in human oral squamous cell carcinoma (OSCC) with and without lymph node metastases (Spoto et al., 2003). The exact role of cGMP in tumorigenesis may be dependent on tissue contexts.

<table>
<thead>
<tr>
<th>Inhibitors</th>
<th>Status</th>
<th>IC$_{50}$ for PDE5, nmol/l</th>
<th>Interaction with other PDE</th>
<th>T$_{max}$, h</th>
<th>C$_{max}$, ng/ml</th>
<th>T$_{1/2}$, h</th>
<th>Structure</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sildenafil</td>
<td>Approved for ED, PAH</td>
<td>3.7</td>
<td>Low activity against PDE6, very low activity against PDE1</td>
<td>0.8</td>
<td>450</td>
<td>3-5</td>
<td><img src="image" alt="Sildenafil Structure" /></td>
</tr>
<tr>
<td>Tadalafil</td>
<td>Approved for ED, BPH</td>
<td>1.8</td>
<td>Low activity against PDE11</td>
<td>2</td>
<td>378</td>
<td>17.5</td>
<td><img src="image" alt="Tadalafil Structure" /></td>
</tr>
<tr>
<td>Vardenafil</td>
<td>Approved for ED</td>
<td>0.091</td>
<td>Low activity against PDE6, very low activity against PDE1</td>
<td>0.7-0.9</td>
<td>20.9</td>
<td>4-5</td>
<td><img src="image" alt="Vardenafil Structure" /></td>
</tr>
<tr>
<td>Udenafil</td>
<td>Clinical Trial</td>
<td>8.2</td>
<td>Low activity against PDE3, 6</td>
<td>1.0-1.5</td>
<td>416.2</td>
<td>11-13</td>
<td><img src="image" alt="Udenafil Structure" /></td>
</tr>
<tr>
<td>Microdenafil</td>
<td>Clinical Trial</td>
<td>0.33</td>
<td>Similar to sildenafil</td>
<td>1.25</td>
<td>No data</td>
<td>2.5</td>
<td><img src="image" alt="Microdenafil Structure" /></td>
</tr>
<tr>
<td>Lodenafil</td>
<td>Clinical Trial</td>
<td>No data</td>
<td>No data</td>
<td>1.2</td>
<td>157</td>
<td>2.4</td>
<td><img src="image" alt="Lodenafil Structure" /></td>
</tr>
<tr>
<td>Avanafil</td>
<td>Approved for ED</td>
<td>1</td>
<td>“Highly” Selective</td>
<td>0.5-1.5</td>
<td>No data</td>
<td>&lt;1.5</td>
<td><img src="image" alt="Avanafil Structure" /></td>
</tr>
</tbody>
</table>

Table 1: Summary of approved and emerging PDE5 inhibitors
2.2 Targeting PDE5 Expression

Increased PDE5 expression occurs in various human carcinomas, including urinary bladder cancers (Piazza et al., 2001), metastatic breast cancers (Pusztai et al., 2003) and non-small cell lung cancers (Whitehead et al., 2003). Increased PDE5 expression has been associated with the development of human OSCC (Spoto, et al., 2003). An increase in the expression of cGMP-PDEs has been confirmed in various cell lines originating from breast cancer (MCF-7, HTB-26, MDA-MB-468), prostate cancer (LNCAP, PC3), colonic adenocarcinomas (HT29, HCT-116, SW480, T84), bladder cancer (HTB-76, HT1376) and some chronic lymphocytic leukemia cells (CLL) (Zhu et al., 2007). These findings suggest that PDE5 may play a role in tumorigenesis and that inhibiting PDE5 activity may produce antineoplastic action. Several groups have evaluated the anti-cancer effect of sildenafil and other PDE5 inhibitors in multiple carcinomas and cancer cell lines. For example, sildenafil and vardenafil suppress tumor cell growth and induce caspase-dependent apoptosis of B-cell chronic lymphocytic leukemia cells in vitro (Sarfati et al., 2003). The non-specific PDE5 inhibitor exisulind and its analogues selectively induce the apoptosis of various human prostate, colon and breast cancer cells. This effect is most likely due to the inhibition of PDE5 expression, thereby decreasing the cGMP-induced activation of PKG. Furthermore, the effects of exisulind and its analogues were not related to their increase in prostaglandin levels, cell cycle arrest, p53 or cyclooxygenase inducing actions (Abadi et al., 2010; Mohamed et al., 2011; Piazza, et al., 2001; Thompson, et al., 2000; Tinsley, et al., 2011; Tinsley, et al., 2009; Tinsley et al., 2010; Whitehead, et al., 2003; Whitt et al., 2012; Zhu et al., 2005).

2.3 Modulating Host Immunity

It is well known that immune system plays a key role in limiting tumorigenesis in the early stages (Zou, 2005). However, the tumor cells ability to effectively change their microenvironment impairs the tumor-specific T cells. Emerging reports in mouse tumor models suggest PDE5 inhibitors may reverse tumor progression by generating a substantial tumor immune response by enhancing the intratumoral T cell adaptation and activity (Bronte et al., 2003). Serafini et al. reported that sildenafil inhibits tumor progression by significantly down-regulating NO synthase-2 (NOS-2) and arginase-1 (ARG1), the key enzymes in L-arginine catabolism, thereby disrupting the machinery of myeloid-derived suppressor cells (MDSCs) (Bronte, et al., 2003). MDSCs induces apoptosis of beneficial CD8+ T cells through NO and super-oxide radicals. The inhibition or differentiation of MDSCs by PDE5 inhibition leads to the restoration and infiltration of CD8+ T cell response, in addition to its tumoricidal activity (Terabe et al., 2003). The down-regulation of MDSCs by PDE5 inhibitors has also been reported in patients with multiple myeloma and head and neck cancers (Serafini et al., 2006). These data suggests that PDE5 inhibitors may be used as an adjunct to maximally enhance antitumor immunity as they have the potential to permit the tumor-specific T cells to alter the tumor microenvironment.

3 PDE5 Inhibitors as Chemo-Sensitizers

Recently, PDE5 inhibitors have been reported to have efficacy as chemo-sensitizers. The ABC transporters (such as ABCB1, ABCC4, ABCC5, ABCC10 and ABCG2), whose overexpression is a main cause of multi-drug resistance (MDR) in cancer cells, were found to be the new targets of PDE5 inhibitors, and PDE5 inhibitors sildenafil, vardenafil and tadalafil were shown to enhance the action of various antineo-
plastic drugs (Chen et al., 2012; Chen et al., 2001; Z. S. Chen et al., 2011; Ding et al., 2011; Jedlitschky et al., 2000; Shi, Tiwari, Patel, et al., 2011; Shi, Tiwari, Shukla, et al., 2011; Sun et al., 2012). Importantly, most of these PDE5 inhibitors are in clinical use and have acceptable tolerability profiles. This section will focus on the chemosensitizing effect of PDE5 inhibitors, which is most likely mediated by their inhibition of the efflux activity of ABC transporters (Figure 1 and Table 2) in addition to its PDE5 inhibitory effect.

<table>
<thead>
<tr>
<th>PDE5 inhibitors</th>
<th>ABC transporters</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>ABCB1</td>
</tr>
<tr>
<td>Sildenafil</td>
<td>++</td>
</tr>
<tr>
<td>Vardenafil</td>
<td>++</td>
</tr>
<tr>
<td>Tadalafil</td>
<td>+</td>
</tr>
</tbody>
</table>

Table 2: Summary of inhibitory effect of PDE5 inhibitors on ABC transporters

### 3.1 Inhibiting ABC Transporters Function

The ABC transporters belong to a superfamily of transmembrane proteins that transport a wide variety of substrates across extra- and intracellular membranes, ranging from ions, sugars, amino acids, vitamins, lipids and drugs to larger molecules such as oligosaccharides, oligopeptides and even higher molecular weight proteins (Dean et al., 2001). Several ABC transporters have shown to transport antineoplastic drugs from cancer cells, including taxanes, anthracyclines, vinca alkaloids, and topoisomerase inhibitors (Dean, et al., 2001). These transporters were discovered in in vitro models of drug resistance and have been postulated to produce multidrug resistance (MDR) in patients. Currently, the major members of the ABC transporters linked to MDR in cancer cells include ABCB1 (P-glycoprotein, ABCB1/MDR1), ABCCs (MRPs) and ABCG2 (BCRP/MXR/ABCP) (Szakacs et al., 2006). These proteins share the ability to transport a large number of structurally diverse, mainly hydrophobic compounds from cells, but each transporter has their own specific substrates (Szakacs, et al., 2006).

It has been shown that cGMP and cAMP are substrates with low micromolar affinity for ABCC4 and ABCC5 transporter and the PDE5 inhibitor sildenafil significantly inhibited the efflux activity of ABCC4 and ABCC5 (Z. S. Chen, et al., 2001; Jedlitschky, et al., 2000). These findings prompted us to determine if sildenafil, as well as other PDE5 inhibitors had the similar effect on other ABC transporters. Our data showed that sildenafil inhibited the activity of ABCB1, ABCC10, ABCG2 and reverse MDR to anti-cancer drugs in cancer cells mediated by these transporters (Chen, et al., 2012; Shi, et al., 2011). In ABCB1-overexpressing cells, non-toxic doses of sildenafil inhibited resistance and increased the effective intracellular concentration of ABCB1 substrate drugs such as paclitaxel. Similarly, in ABCG2-overexpressing cells, sildenafil inhibited the efflux of the ABCG2 substrate anti-cancer drug mitoxantrone as well as the fluorescent compound BODIPY-prazosin. Sildenafil also inhibited the transport of E217βG and methotrexate by the ABCG2 transporter. Mechanistic experiments indicated that sildenafil stimulated ABCB1 ATPase activity and inhibited the photolabeling of ABCB1 with [125I]-IAAP, whereas it also stimulated ABCG2 ATPase activity and inhibited photolabeling of ABCG2 with [125I]-IAAP. These aforementioned effects of sildenafil were specific as it did not alter the sensitivity of parental ABCB1-
or ABCG2-overexpressing cells to non-ABCB1 and non-ABCG2 substrate drugs and sildenafil did not significantly affect the function of ABCC1. Homology modeling predicted that the binding conformation of sildenafil occurred within the large cavity of the transmembrane region of ABCB1 (Shi, et al., 2011). Additionally, sildenafil also produced a concentration-dependent increase in the sensitivity of ABCC10-transfected HEK293 cells to paclitaxel, docetaxel and vinblastine, and enhanced the intracellular accumulation of [3H]-paclitaxel by inhibiting the efflux of [3H]-paclitaxel in HEK293/ABCC10 cells. (Chen, et al., 2012) Further, other PDE5 inhibitors, vardenafil and tadalafil, were also examined for their effect on ABC transporter-mediated MDR in cancer cells (Chen, et al., 2012; Ding, et al., 2011). The results showed that vardenafil when used in combination with anti-cancer substrates of ABCB1, significantly potentiated their cytotoxicity in ABCB1 overexpressing cells in a concentration-dependent manner, and this effect was significantly greater than that of tadalafil. The sensitivity of the parenteral cell lines to cyto-toxic anti-cancer drugs was not significantly altered by vardenafil. The differential effects of vardenafil and tadalafil appear to be specific for the ABCB1 transporter, as both vardenafil and tadalafil had no significant effect on the reversal of drug resistance conferred by ABCC1 and ABCG2 transporters. Vardenafil significantly increased the intracellular accumulation of [3H]-paclitaxel in the ABCB1-overexpressing KB-C2 cells. In addition, vardenafil significantly stimulated the ATPase activity of ABCB1 and inhibited the photolabeling of ABCB1 with [125I]-IAAP. Furthermore, Western blot analysis indicated the incubation of cells with either vardenafil or tadalafil for 72 h did not alter ABCB1 protein expression (Ding, et al., 2011). Additionally, vardenafil and tadalafil produced a concentration-dependent increase in the sensitivity of ABCC10-transfected HEK293 cells to paclitaxel, docetaxel and vinblastine. The accumulation and efflux experiments demonstrated that vardenafil and tadalafil increased the intracellular accumulation of [3H]-paclitaxel by inhibiting the efflux of [3H]-paclitaxel in HEK293/ABCC10 cells. (Chen, et al., 2012). Summary of inhibitory effect of PDE5 inhibitors on ABC transporters is shown in Table 2. The reason why certain PDE5 inhibitors can affect certain ABC transporters and not others might be due to their structural–activity relationship or their ability to inhibit ABC transporters. The molecular configuration of tadalafil departs entirely from that of both sildenafil and vardenafil, whereas sildenafil and vardenafil differ only in particular by their nitrogen atoms in the heterocyclic ring system.

### 3.2 Blocking PDE5 Activity

The apoptotic activity of exisulind was shown to be beneficial when combined with docetaxel in non-small cell lung cancer orthotopic lung tumor model (Whitehead, et al., 2003) or in combination with capecitabine in Phase I and II metastatic breast cancer (Pusztai, et al., 2003). In a rat brain tumor model, the PDE5 inhibitors sildenafil and vardenafil increased the transport of doxorubicin across blood-brain tumor barrier and enhanced the efficacy of chemotherapy (Black et al., 2008). These two PDE5 inhibitors also selectively increased transport of herceptin across brain tumor capillaries and significantly enhanced the antitumor effect of herceptin in mouse models of metastatic HER2/neu-positive brain tumors (Hu et al., 2010). Moreover, sildenafil was reported to enhance the sensitivity of breast cancer cells to doxorubicin without exacerbating its toxic effects on either bone marrow cells or macrophages (Di et al., 2010). Sildenafil also increased the chemotherapeutic efficacy of doxorubicin in prostate cancer in vivo and ameliorated cardiac dysfunction (Das et al., 2010). Sildenafil produced penile vasodilation and reduced tumor hypoxia in squamous carcinoma of penis, when it was used as radio-sensitizers (Sun, et al., 2012). PDE5 inhibitor’s radio-sensitization mechanism may be due to increased perfusion of blood in the penile
structure, leading to cGMP-induced relaxation in the corpus cavernosum, which reduces hypoxia and thus increase radio-sensitivity.

4 Implications

Based on the studies discussed in this chapter, it is reasonable to hypothesize that PDE5 inhibitors may enhance anti-cancer drugs sensitivity and potentially improve the chemotherapeutic outcome in cancer patients due to its inhibitory effect on PDE5 and specific ABC transporters. PDE5 inhibitors alone produced transient and mild to moderate adverse effects, which usually dissipates on continued treatment. In addition, favorable safety and efficacy profiles are reported for all the clinically used PDE5 inhibitors, where vardenafil and tadalafil are slightly more selective than sildenafil based on their efficacy profile towards PDE5. Moreover, one may rationalize tadalafil to offer more flexible dosage regimen as it acts for 36 hour duration in patients compared to around 4 hour action with vardenafil and sildenafil. Future studies examining the combined use of PDE5 inhibitors with anti-cancer drugs need to address several issues. First, the pharmacokinetic profile of PDE5 inhibitors and anti-cancer drugs may be affected by each other, potentially resulting in an increased therapeutic response, but also in adverse effects. This is possible as some ABC transporters are highly expressed in normal tissues (Szakacs, et al., 2006), where the concentration and distribution of PDE5 inhibitors and anti-cancer drugs may be altered if used in combination. Second, most PDE5 inhibitors, including sildenafil, vardenafil and tadalafil are primarily metabolized by the cytochrome P450 (CYP) isoenzyme CYP3A4 (Warrington et al., 2000) and the substrates for CYP3A4 overlap considerably with those of ABCB1 transporters (Zhou, 2008). Consequently, the metabolism and elimination of these PDE5 inhibitors, as well as the anti-cancer drugs, some of which are substrates of both CYP3A4 and ABCB1 (Zhou, 2008), may be affected when these drugs are used in combination. Finally, determining the concentrations that would be effective in vivo would definitely improve the outcome of the combined use of PDE5 inhibitors with anti-cancer drugs. For example, the maximum observed plasma concentration (Cmax) of a single oral dose of 25-200 mg of sildenafil in healthy male subjects is 127-1150 ng/ml (0.2-2 µM) (Nichols et al., 2002), which is slightly lower than the concentration that was effective for MDR reversal (Shi, et al., 2011). Therefore, concentrations of sildenafil exceeding those required for PDE5 inhibition seem to be required to enhance the effects of chemotherapeutic drugs. Third, it is important to determine the affinity of PDE5 inhibitors towards the variable expression levels of PDE5 and some ABC transporters in cancer tissues. In addition to the overexpression of ABC transporters, other drug resistance determinants in cancer cells include changes in metabolizing and detoxifying systems, such as DNA repair and the cytochrome P450 oxidases and drug-induced alterations in apoptosis (Szakacs, et al., 2006). Hence, the expression levels of PDE5 inhibitors target proteins such as PDE5 and some ABC transporters would significantly determine the efficiency of PDE5 inhibitors.

5 Summary

Greater benefits are seen with multi-targeted agents than those observed with single-targeted compounds, as they are usually active against a broader range of tumor types. As our knowledge and understanding of
physiological role of PDE5 in various tissues is broadening, novel functions and applications of these drugs are apparent. An increasing number of studies indicate an essential role of PDE5 expression in regulating apoptosis and growth of cancer cells. The functional role of PDE5 in controlling cGMP-mediated downstream signaling suggests potential anti-cancer targets that could be used for the development of novel anti-cancer agents. In Figure 1, a comprehensive understanding of both direct and indirect anti-cancer activity of PDE5 inhibitors is shown. In summary, the PDE5 inhibitors may have anti-cancer activity by: 1) blocking the substrate efflux function of multidrug resistant transporters such as ABCC4 (Chen, et al., 2001), ABCC5 (Jedlitschky, et al., 2000), ABCC10 (Chen, et al., 2012), ABCB1 and ABCG2 (Ding, et al., 2011; Shi, et al., 2011); 2) increasing cGMP levels via its inhibition of PDE5, ABCC4- and ABCC5-mediated efflux of cGMP, which may activate PKG-mediated signaling, thus producing growth suppression or apoptosis (Zhu, et al., 2007); 3) some unknown pathways, yet not clear. For these reasons, analogues of tadalafil, have been developed to be tested specifically for blocking tumor growth (Abadi et al., 2009; Abadi, et al., 2010). It is believed that additional PDE5 inhibitors will be undergoing experimental trials as anti-cancer drugs or adjuvants to cancer chemotherapy in the future.

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Reference


