Signaling network in focus
The endothelin axis in cancer
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Abstract
The endothelin axis, comprising endothelins and their receptors, has recently emerged as relevant player in tumor growth and metastasis by regulating mitogenesis, cell survival, angiogenesis, bone remodeling, stimulation of nociceptor receptor, tumor-infiltrating immune cells, epithelial-to-mesenchymal transition, invasion and metastatic dissemination. Endothelin-1 participates in the growth and progression of a variety of tumors such as prostatic, ovarian, renal, pulmonary, colorectal, cervical, breast, bladder, endometrial carcinomas, Kaposi’s sarcoma, brain tumors, melanoma, and bone metastases. This review highlights key signaling pathways activated by endothelin-1 axis in cancer, since the understanding the full spectrum activated by endothelin-1 is critical for the optimal design of targeted therapies. Preliminary experimental and clinical data demonstrate that interfering with endothelin receptor by using endothelin-1 receptor antagonists alone and in combination with cytotoxic drugs or molecular inhibitors could represent a new mechanism-based antitumor strategy.
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Keywords: Endothelin; Endothelin receptor antagonist; G-protein-coupled receptor; Antitumor therapy

1. Introduction
Endothelins (ETs) are a family of three 21-aa peptides ET-1, ET-2 and ET-3. ETs mediate their action by activating two G-protein-coupled receptor (GPCR) subtypes, ETA receptor (ETAR) and ETB receptor (ETBR). In addition to its role as a potent endogenous vasoconstrictor and mediator of cardiovascular and renal disorders, the endothelin axis has a relevant role in various cancer cells and stromal cells leading to autocrine/paracrine loops that activate aberrant proliferation, escape from apoptosis, new vessels formation, immune modulation, abnormal osteogenesis, alteration of nociceptive stimuli, invasion and metastatic dissemination (Fig. 1) (Nelson, Bagnato, Battistini, & Nisen, 2003).

2. Synthesis
ET-1, ET-2 and ET-3 are characterized by a single α-helix and two disulfide bridges. ET-1 and ET-2 have similar structures, whereas ET-3 differs in structure at 6 of 21 aa. The three peptides are encoded by distinct genes and are regulated at the level of mRNA transcription. The primary translation product of the ET-1 gene is the 212-aa prepro-ET-1, which is cleaved by an endothelin converting enzyme (ECE-1) to form the 38-aa big-ET-1, and then to the biologically active peptide and a C-terminal fragment. The half-life of ET-1 in circulation is 1 min. Two pathways for endothelin clearance have been described: ETBR-mediated uptake followed by...
Signaling network facts

- Endothelin axis, by triggering multiple signaling pathways contributes to tumor growth and progression, inducing cell proliferation, survival, angiogenesis and metastatic spread, indicating that endothelin-1 receptor blockade might improve cancer treatment.
- Endothelin-1, acting primarily through the endothelin A receptor, triggers a highly interconnected signaling network that is further amplified by cross-talk with receptor tyrosine kinase such as the epidermal growth factor receptor.
- In clinical studies, endothelin A receptor antagonists can be administered orally and have a favorable tolerability profile.
- Preliminary data from clinical studies indicate that this class of drugs is promising for targeted therapy. Future clinical trials will evaluate the use of endothelin A receptor antagonist in combination with current chemotherapy or with other molecular therapy.

Endosomes and catabolism by extracellular neutral endopeptidase 24.11 (NEP, neprilysin). ET-1 production is stimulated by a variety of cytokines and growth factors, hypoxia, and shear stress. Inhibitory factors include nitric oxide, prostacyclin and atrial natriuretic peptide (Nelson et al., 2003).

3. Endothelin receptors and G-protein signaling

ETs exert their effects by binding to two distinct cell surface ET receptors, ETAR and ETBR. ETBR has equal affinities for either ETs, whereas ETAR exhibits an affinity for ET-3 that is two orders of magnitude lower than that for ET-1 and ET-2. Most important, ligand activation of ETBR leads to induction of intracellular pathways being counter-regulatory to ETAR signaling. The receptors belong to the superfamily of GPCR and contain seven hydrophobic transmembrane domains, an intracytoplasmatic C terminus and an extracellular N terminus. Due to their differences in C terminus sequences, which are pivotal for coupling of G proteins, either receptor induces divergent intracellular effects. GPCR interact with heterotrimeric G proteins composed of α, β and γ subunits. The α subunits of G proteins are divided into four subfamilies: Gαs, Gαi, Gαq and Gα12. Each G-protein activates several downstream effectors. Typically, Gαq,s coupling results into activation of adenyl cyclase. Gαi coupling leads to an inhibition of adenyl cyclase and activation of Ca2+ channels. Coupling of Gα12 entails activation of phospholipase C (PLC) (Masaki, 2000) which cleaves phosphatidylinositol bisphosphate (PIP2) into diacylglycerol and inositol triphosphate (IP3). Gαq and Gα12 can also control the activity of key intracellular signal transducing molecules, including mitogen activated protein kinase (MAPK) as well as induction of various immediate early genes (Rozengurt, 2007).

3.1. Signaling of ET-1 in cell proliferation

The binding of endothelin to its cognate GPCR triggers the activation of a network of a multiple signaling pathways rather than a linear sequence of intracellular signaling cascades, including PLC activity, increases in intracellular Ca2+ levels, activation of protein kinase C, and MAPK, that act in a synergistic and combinatorial fashion to relay the mitogenic signal to the nucleus and promote cell proliferation. The epidermal growth factor receptor (EGFR) transactivation has emerged as a transducer in the signaling of endothelin receptor. Moreover, further analysis of the signaling pathway showed that ET-1 stimulated phosphatidylinositol 3-kinase (PI3K)-mediated AKT activation, indicating that endothelin axis activates a complex signaling network that is finely tuned during cell growth. ET-1 stimulates DNA synthesis and cell proliferation in various tumor cells, including prostate, cervical, ovary cancer cells. In these carcinoma cell lines, spontaneous growth was significantly inhibited in the presence of ETAR and not ETBR antagonist, demonstrating that endogenous ET-1 acts as an autocrine modulator of cell proliferation only through ETAR. The mitogenic activity of ET-1 can be amplified by synergistic interactions with other growth factors including EGF, basic fibroblast growth factor (bFGF), insulin, insulin-like growth factor (IGF), platelet-derived growth factor (PDGF), transforming growth factor (TGF), and interleukin-6 (IL-6) (Nelson et al., 2003).

3.2. Cross-signaling between ETAR and EGFR pathways

Cross-talk between cell surface receptors represents the main mechanism to expand the cellular communication signaling network. A major pathway utilized by many GPCR agonists is the transactivation of EGFR
Fig. 1. By activating ET\(_{AR}\), ET-1 is involved in facilitating several aspects of cancer growth and progression, including cell proliferation, escape from programmed cell death, new vessel formation, invasion and metastatic spread. In some tumors, such as prostate and breast cancer, ET-1 stimulates osteoblast proliferation, decreasing both osteoclastic bone resorption and motility, resulting in osteoblastic lesions associated with osteoblastic bone metastases, alteration of nociceptive stimuli and malignant bone pain. ETs modulate also trafficking, differentiation and activation of tumor-infiltrating immune cells.

that leads to RAS-dependent MAPK activation. GPCR-induced EGFR transactivation is mediated by release of precursor forms of EGFR ligands generated by activation of matrix-metalloproteinases (MMPs). EGFR is also phosphorylated directly by c-Src. For example in colon cancer cells, EGFR transactivation appears to proceed through an intracellular pathway involving GPCR, \(\beta\)-arrestin and c-Src (Rozengurt, 2007). In this context, in ovarian cancer cells ET-1 causes EGFR transactivation, that is in part responsible for MAPK activation, by c-Src. This event leads through the formation of Shc/Grb-2 complexes to activation of the RAS/MAPK pathway as well as of AKT activation (Bagnato, Spinella, & Rosanò, 2005; Vacca, Bagnato, Catt, & Tecce, 2000). The cross-signaling between the EGFR/ET\(_{AR}\) pathways provides a rationale to combine EGFR inhibitors with ET\(_{AR}\) antagonists. ZD4054, a specific ET\(_{AR}\) antagonist, was able to reduce the ET-1-induced EGFR transactivation. The EGFR inhibitor gefitinib significantly inhibited EGFR- and ET-1-induced EGFR phosphorylation, but incompletely reduced the ET-1-induced activation of downstream targets, indicating that dual alternative pathways, one EGFR-dependent, and the other independent, converge on MAPK and AKT activation. ZD4054 plus gefitinib resulted in a greater inhibition of EGFR, MAPK, and AKT phosphorylation, indicating the critical role of these interconnected signaling proteins. The drug combination resulted in a significant decrease in cell proliferation, invasion, and vascular endothelial growth factor (VEGF) production, accompanied by a two fold increase in apoptosis, suggesting that this drug combination by simultaneous disabling multiple signaling circuits activates by EGFR and ET\(_{AR}\) may be potentially advantageous in cancer therapy (Rosanò et al., 2007).

3.3. Signaling of ET-1 in tumor neovascularization

ETs, which are mitogens for endothelial cells, vascular smooth muscle cells, fibroblasts, and pericytes, are also angiogenic factors. Endothelial cell mitogenesis is mediated by ET\(_{BR}\), whilst vascular smooth muscle cells and pericycle mitogenesis is mediated by ET\(_{AR}\). ET-1 modulates various stages of neovascularization, including endothelial cell proliferation, migration, invasion, protease production, tube formation and stimulates neovascularization in vivo. Elevated expression of ET-1 and its cognate receptor is significantly associated with microvessel density (MVD) and VEGF expression in tumor cells. Thus, ET-1 increases VEGF mRNA
expression and induces VEGF levels in a time- and dose-dependent fashion, and does so to a greater extent under hypoxia.

The transcriptional upregulation of VEGF has been linked to the hypoxic inducible factor-1α (HIF-1α), the critical transcriptional factor that conveys signaling elicited by hypoxia and growth factors. Degradation of HIF-1α was reduced in ET-1-treated tumor cells under both hypoxic and normoxic conditions. Following ET-1 stimulation, HIF-1α protein levels increase, the HIF-1 transcription complex is formed which binds to the HRE binding site. Thus, under hypoxic conditions, ET-1 potentiates hypoxia stimulus by amplifying HIF-1α stability and VEGF production (Bagnato et al., 2005). Addition of a specific ETAR antagonist blocked the ET-1-induced upregulation of VEGF expression and secretion as well as activation of HIF-1 transcription complex. There is a reciprocal relationship between ET-1 expression and HIF-1α activity: not only ET-1 stabilizes HIF-1α during normoxia and to a greater extent in hypoxia leading to HIF-1α-mediated transcription of angiogenic genes, but HIF-1α mediates transcription of ET-1 in different cell types. Therefore expression of ETs is controlled by the tumor microenvironment, whilst the endothelins themselves modify that environment via HIF-1α (Grimshaw, 2007).

In normoxic conditions, ET-1 significantly increases the expression of cyclooxygenase (COX)-1 and -2, at mRNA and protein level, the COX-2 promoter activity and prostaglandin E2 (PGE2) production, and do so to a greater extent under hypoxia. COX-2 and -1 inhibitors blocked ET-1-induced PGE2 and VEGF release, MMP activation and cell invasion, demonstrating that both enzymes function as downstream mediators of ET-induced angiogenic and invasive properties. The ET-1 receptor specific antagonist or transfection with a dominant negative ILK mutant effectively reverses ET-1-mediated transcription of angiogenic genes, but HIF-1α mediates transcription of ET-1 in different cell types. Therefore expression of ETs is controlled by the tumor microenvironment, whilst the endothelins themselves modify that environment via HIF-1α (Grimshaw, 2007).

Defects in intercellular communication, including reduced or inappropriate expression of connexin (Cx), predominantly Cx43, has emerged as key factors in tumor progression. In tumor cells, ET-1 axis induces a transient and a dose-dependent reduction of gap junction intercellular communications (GJIC) and tyrosine phosphorylates Cx43 by c-Src (Bagnato, Rosanò, Spinella, Di Castro, Tecce, & Natali, 2004; Bagnato et al., 2005). In this context, ET-1 enhances the expression of α2β1 and α3β1 integrins. Integrin-linked kinase (ILK) activity increases as ovarian cancer cells adhere to type I collagen through a β1 integrin signaling, even more upon ET-1 stimulation. ET-1 increases ILK expression and activity and an ILK inhibitor KP-392 or transfection with a dominant negative ILK mutant effectively blocks the phosphorylation of the downstream signals, activation. In different tumor cells, the addition of ET-1 markedly inhibited paclitaxel-induced apoptosis in a concentration-dependent manner, as result of Bcl-2 phosphorylation. Interestingly, the addition of a specific ETAR antagonist blocked this effect, indicating that ET-1 contributes to trigger resistance to paclitaxel through ETAR binding via activation of anti-apoptotic signaling pathways such as Akt (Del Bufalo et al., 2002).

Specific ETAR antagonists may therefore provide an additional approach to the treatment of carcinoma in which ETAR blockade could result in the tumor growth inhibition by reducing tumor growth as well as by inducing apoptosis. Furthermore, when combined with the conventional chemotherapy the ETAR antagonists would more effectively induce apoptosis by contributing to the reversal of paclitaxel resistance, as observed in ovarian, prostate, cervical, and nasopharyngeal carcinoma (Akhavan et al., 2006; Bagnato et al., 2002; Rosanò et al., 2007).

3.5. Signaling of ET-1 in tumor invasion

ET-1 acting through the ETAR consistently induced the activity of two families of metastasis-related proteinases, the MMPs and the urokinase type plasminogen activator (uPA) system at several levels: mRNA transcription, zymogen secretion and pro-enzymes activation, resulting into the highest invasive potential of tumor cells. Furthermore in these cells, ET-1 stimulated focal adhesion kinase (FAK) and paxillin phosphorylation suggesting that targeting ET-1 axis can inhibit cell migration and possibly other FAK-associated processes which also contributes to invasion and metastasis. Moreover, EGFR transactivation by ETAR is in part responsible for PGE2 and VEGF secretion, MMP activity and tumor cell invasion (Bagnato et al., 2005).

3.4. Signaling of ET-1 in cell survival

ET-1 is an anti-apoptotic factor in different cell types, indicating that the peptide may also modulate cell survival pathways, such as PI3-K-dependent AKT pathways such as Akt (Del Bufalo et al., 2002).
AKT and glycogen synthase kinase-3β (GSK-3β). The blockade of ET-1/ETAR-induced ILK activity results into an inhibition of MMP activation as well as of cell motility and invasiveness in a PI3-K-dependent manner, indicating that ILK functions as a downstream mediator of ET-1/ETAR axis to potentiate aggressive cellular behaviour. One hallmark of epithelial cancer progression is epithelial-to-mesenchymal transition (EMT), in which tumor cells undergo loss of polarity and cell–cell junctions, acquire a mesenchymal phenotype, the ability to invade the extracellular matrix, and to migrate to distant sites. In cancer cells, such as melanoma or ovarian carcinoma cells, activation of ET-1 axis contributes to disruption of normal host–tumor interactions by downregulating the expression of E-cadherin and associated β-catenin and concomitant upregulation of the mesenchymal N-cadherin. In ovarian carcinoma cells, activation of ETAR pathway by ET-1 drives inhibition of GSK-3β by a PI3-K-dependent ILK-mediated signaling pathway to stabilize snail and β-catenin proteins in a coordinate fashion so as to cooperatively engage transcriptional programs that control repression of E-cadherin leading to EMT (Bagnato et al., 2004; Rosanò et al., 2005). Interestingly, in colon cancer cells, ET-1 is a downstream target of β-catenin, and can rescue cells from apoptosis after β-catenin inhibition. Moreover in both normal and malignant prostate cells, β-catenin transcriptionally activates ET-1 expression. Meanwhile, ET-1 stimulates β-catenin signaling via a PI3K-dependent pathway. The positive inter-regulation between β-catenin and ET-1 signaling plays an important role in promoting proliferation and survival of cancer cells, thereby representing a novel mechanism that contributes to cancer progression (Sun, Xiong, Kim, Ren, & Zhang, 2006).

3.6. Signaling of ET-1 axis in osteogenesis

Several observations implicate ET-1 in the osteoblastic response. Osteoblasts display a high density of ETAR, and respond to ET-1, driving osteoblastic proliferation and new bone formation through this receptor (Guise et al., 2006). Thus, ET-1 stimulates mitogenesis in osteoblasts and decreases osteoclastic bone resorption and osteoclast motility. Accumulating data have shown that ET-1 stimulates bone formation associated with metastatic breast and prostate tumors. Experimental models of osteoblast proliferation and bone metastasis are inhibited by ETAR antagonists in vivo, suggesting that the ETAR are attractive targets for the management of tumors that metastasize to the bone (Nelson et al., 2003).

3.7. Signaling of ET-1 axis in pain

ET-1 is described as a new pain mediator implicated in the pathogenesis of refractory pain of certain metastatic cancers. Local cutaneous injection of ET-1 causes pain and excitation of nociceptors through ETAR and concurrently produces analgesia through ETBR by inducing the release of β-endorphin and the activation of opioid pool. Thus, antagonist of ETAR has been shown to ameliorate pain. This knowledge may lead to improved, targeted analgesia in patients with advanced cancer (Davar, 2001).

3.8. Signaling of ET-1 axis in immune modulation

ETs modulate trafficking, differentiation and activation of tumor-infiltrating immune cells. ETs have a role in recruiting tumor-associated macrophages (TAMs): macrophages express both endothelin receptors and chemotaxis towards ETs via ETβR and a MAPK-mediated signaling pathway. Macrophages not only react to ETs but also produce ETs themselves; in contrast, no immunoreactive ET can be detected in cell extracts from human neutrophils and lymphocytes (Grimshaw, 2007).

4. Targeting endothelin receptor as novel approach in cancer treatment

The demonstration that ET-1 actions are concordant with many of the “hallmarks of cancer” indicates a critical role for ET-1 in the initiation or progression of many tumors clearly identifying the ET axis as a potential therapeutic target. This has propelled the development of several approaches targeting ET-1 axis in cancer therapy including ECE inhibitors or NEP transfection, which efficiently degrades ET-1, or selective or non-selective antagonists for ETAR and ETBR (Smollich & Wulfing, 2007). To date, endothelin receptor blocker represents the most promising approach in controlling the pleiotropic activities of ET-1, which are all pivotal in the gain of malignant phenotype. In particular the development of small molecules acting as selective ETAR antagonists has contributed to understanding of the physiopathologic relevance of the ET-1 axis and its signaling circuitry in tumor progression and metastasis providing a rationale for the translation of this information into clinical trials (Fig. 2). Among various ETAR antagonists, ABT-627 (atrasentan) and ZD4054 are orally bioavailable ETAR antagonists that potently and specifically bind to the ETAR, blocking signal transduction pathways implicated in cancer cell proliferation and other host-dependent processes pro-
Fig. 2. ETAR-induced signal transduction pathways in cancer cells. Binding of ET-1 to ETAR triggers signaling pathways through a pertussis-insensitive Gq subunit that is coupled to the ETAR intracellular domain. This led to the activation of a signaling network that includes PLC with subsequent parallel mobilization of intracellular Ca^{2+} and PKC activation, and PTKs (such as FAK and paxillin) that ultimately results in the activation of the RAF/MEK/MAPK pathway. ET-1 also activates PI3-K, which results in the stimulation of the Akt survival pathway and increased protein translation by the activation of mTOR signaling pathway, and parallel ILK-dependent GSK-3β phosphorylation stabilizing snail and β-catenin that translocate to the nucleus to engage transcriptional programs leading to EMT. ET-1 also causes c-Src-mediated EGFR transactivation that is in part responsible for MAPK activation. Moreover, ET-1 promotes COX-1 and -2 expression, PGE2 and VEGF production. The activation of multiple and coordinated signaling pathways, regulates pleiotropic functions, including tumor growth, survival, angiogenesis and, as recently emerged, EMT and invasion.

moting cancer growth (Nelson et al., 2003; Rosanò et al., 2007).

4.1. Ovarian carcinoma

ET-1/ETAR autocrine pathway is overexpressed in primary and metastatic ovarian carcinomas. Levels of ET-1 are markedly elevated in the ascites of patients with epithelial ovarian cancer (Bagnato et al., 2005) and, together with the ETAR, are overexpressed and activated in 85% of ovarian tumors, correlating with advanced stages. Recent analysis of gene expression profile of late-stage ovarian cancer identified ETAR as a metastasis-associated gene. Moreover, ETAR has been also recognized as one of the genes highly expressed in post-chemotherapy samples compared with untreated primary tumors.

Treatment with ZD4054 or ABT-627 produced tumor growth inhibition in well established HEY xenografts with no detectable signs of acute or delayed toxicity. More marked or complete tumor growth inhibition was obtained by combined treatment of ETAR antagonist with cytotoxic drug, such as paclitaxel, or with molecular inhibitor, such as gefitinib (Rosanò et al., 2003a, 2007).

4.2. Prostate carcinoma

The ET axis has recently been identified as contributing to the pathophysiology of prostate cancer. In the normal prostate gland, ET-1 is produced by epithelial cells; the highest concentrations of ET-1 are found in seminal fluid. In prostate cancer, key components of the ET-1 clearance pathway, ETBR and NEP, are diminished, resulting in an increase in local ET-1 concentrations. Increased ETAR expression is also seen with advancing tumor stage and grade in both primary and metastatic prostate cancer. By activating ETAR, ET-1 is pathogenically involved in facilitating several aspects of prostate cancer progression, including proliferation, escape from apoptosis, new bone formation or altering the equilibrium in
Table 1
Role of ET receptors in different tumors

<table>
<thead>
<tr>
<th>Tumor Type</th>
<th>Endothelin receptors</th>
<th>Receptor antagonists and their effects</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>Prostate cancer</td>
<td>Expression of the ETAR increases with tumor stage and grade, and is associated with decreased ETBR expression</td>
<td>ETAR antagonist has demonstrated benefit in PSA progression, markers of bone turnover and pain</td>
<td>Carducci and Jimeno (2006)</td>
</tr>
<tr>
<td>Ovarian cancer</td>
<td>ETAR mRNA is present in 85% of primary and metastatic cancers, and correlates with tumor grade. ETAR mediates all ET-1-induced tumor promoting effects</td>
<td>In preclinical studies ETAR antagonists display antitumor effects and additive effects in combination with taxanes or gefitinib</td>
<td>Bagnato et al. (2005); Rosanò et al. (2007)</td>
</tr>
<tr>
<td>Bladder cancer</td>
<td>Both ETAR and ETBR are expressed in bladder tumors. ETAR is overexpressed and correlates with tumor progression. ETBR mediates all ET-1-induced tumor promoting effects</td>
<td>ETAR antagonist decreases lung metastases. ETBR antagonists inhibit melanoma cell growth in vitro and in vivo. The dual ETA,BR antagonist Bosentan has benefit in disease stabilization in metastatic melanoma patients</td>
<td>Herrmann et al. (2007)</td>
</tr>
<tr>
<td>Melanoma</td>
<td>ETBR is overexpressed and correlates with tumor progression. ETAR mediates all ET-1-induced tumor promoting effects</td>
<td>ETAR antagonist inhibits tumor growth in vitro and in vivo. The dual ETA,BR antagonist Bosentan has benefit in disease stabilization in metastatic melanoma patients</td>
<td>Bagnato et al. (2004); Kefford et al. (2007)</td>
</tr>
<tr>
<td>Bone malignancies</td>
<td>ETAR is expressed in osteoblasts</td>
<td>ETAR antagonist significantly reduced osteoblastic bone metastases and tumor burden in bone</td>
<td>Guise et al. (2006)</td>
</tr>
<tr>
<td>Breast cancer</td>
<td>ETAR expression correlates with several clinicopathological parameters of aggressive carcinoma</td>
<td>In preclinical studies ETAR antagonist inhibits tumor growth</td>
<td>Smolich and Wulfing (2007)</td>
</tr>
<tr>
<td>Renal cancer</td>
<td>ETAR is expressed in different cell lines</td>
<td>ETAR antagonist inhibits tumor growth and metastasis and shows synergistic effects in combination with cytotoxic drugs</td>
<td>Pflug et al. (2007)</td>
</tr>
<tr>
<td>Nasopharyngeal carcinoma</td>
<td>ETAR is overexpressed in 74% of tumors</td>
<td>ETAR antagonist inhibits tumor growth and metastasis and shows synergistic effects in combination with cytotoxic drugs</td>
<td>Mai et al. (2006)</td>
</tr>
<tr>
<td>Colon cancer</td>
<td>Increased expression of ETAR and ETBR</td>
<td>Dual ETAR antagonist Bosentan induces apoptosis or sensitises cells to Fas-induced apoptosis</td>
<td>Peduto Eberl et al. (2003)</td>
</tr>
<tr>
<td>Cervical cancer</td>
<td>HPV-positive cervical carcinoma cells predominantly express functional ETAR</td>
<td>ETAR antagonist inhibits tumor growth in monotherapy as well as in association with taxane</td>
<td>Bagnato et al. (2002)</td>
</tr>
<tr>
<td>Kaposi’s sarcoma</td>
<td>Both ETAR and ETBR are expressed</td>
<td>Dual ETAR antagonist inhibits tumor growth in nude mice</td>
<td>Rosanò et al. (2003b)</td>
</tr>
<tr>
<td>Glioblastoma</td>
<td>ETBR is expressed in cancer cells</td>
<td>Dual ETAR antagonist induced apoptosis</td>
<td>Egidy et al. (2000)</td>
</tr>
</tbody>
</table>
pain modulation. Biological activity of atrasentan in patients with prostate cancer has demonstrated benefit in biological markers of prostate cancer progression, markers in bone turnover and pain (Carducci & Jimeno, 2006). Recent results from a Phase II clinical trial indicate that ZD4054 could offer a promising improvement in overall survival in men with metastatic hormone refractory prostate cancer HRPC (James et al., 2007).

4.3. Other cancers

ET-1 has been studied in several other cancers, including those arising in colon, lung, breast, cervix, bladder, nasopharynx, as well as Kaposi’s sarcoma, melanoma, neuroblastoma, and glioblastoma. In some of these models, ET\textsubscript{A}R or ET\textsubscript{B}R or dual ET\textsubscript{A}\textsubscript{B}R blockers have been tested as antitumor agents in mono and combination therapy with chemotherapeutics. The role of the ET axis and the therapeutic relevance of ET-1 receptor antagonists in a range of malignancies requires future investigation that may lead to a new generation of molecularly targeted therapies for cancer (Table 1).

5. Conclusion

The endothelin axis has a crucial role in stromal-cancer cell interactions that promote tumor initiation and progression representing a new and mostly unexplored target for cancer therapy. ET-1 receptor antagonism remains a promising therapeutic approach. However in some tumors it is still unclear when to use selective ET\textsubscript{A}R antagonists and when to use mixed ET\textsubscript{A}\textsubscript{B}R blockers. To this end, further information regarding the expression of endothelin receptors in different neoplasia is required.

Experimental and preclinical evidence have shown that ET-1 receptor antagonism potentiates the therapeutic efficacy of conventional cytotoxic drugs, offering a rationale for its clinical evaluation. A novel promising approach to cancer therapy emerges by the treatment with multiple selective inhibitors of different growth factor receptors or to key post-receptor signaling pathways. In this context, the improved knowledge of the interconnected molecular mechanism promoted by ET-1 axis in cancer will certainly fuel the interest of basic and translational scientists to evaluate new treatment strategy that incorporate ET\textsubscript{A}R blockade in combination with other molecularly targeted drug in order to overcome compensatory mechanism of escape that can be eliminated therapeutically.

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