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Repurposing the anti-parasitic agent pentamidine for cancer therapy; a novel approach with promising anti-tumor properties

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Abstract

Pentamidine (PTM) is an aromatic diamidine administered for infectious diseases, e.g. sleeping sickness, malaria, and *Pneumocystis jirovecii* pneumonia. Due to similarities of cellular mechanisms between human cells and such infections, PTM has also been proposed for repurposing in non-infectious diseases such as cancer. Indeed, by modulating different signaling pathways such as PI3K/AKT, MAPK/ERK, p53, PD-1/PD-L1, etc., PTM has been shown to inhibit different properties of cancer, including proliferation, invasion, migration, hypoxia, and angiogenesis, while inducing anti-tumor immune responses and apoptosis. Given the promising implications of PTM for cancer treatment, however, the clinical translation of PTM in cancer is not without certain challenges. In fact, clinical trials have shown that systemic administration of PTM can be concurrent with serious adverse effects, e.g. hypoglycemia. Therefore, to reduce the administered doses of PTM, lower the risk of adverse effects, and prevent any potential drug resistance, while maintaining the anti-tumor efficacy, two main strategies have been suggested. One is combination therapy that employs PTM in conjunction with other anti-cancer modalities, such as chemotherapy and radiotherapy, and attacks tumor cells with significant additive or synergistic anti-tumor effects. The other is developing PTM-loaded nanocarrier drug delivery systems e.g. pegylated liposomes, chitosan-coated niosomes, squalene-based nanoparticles, hyaluronated lipid-polymer hybrid nanoparticles, etc., that offer enhanced pharmacokinetic characteristics, including increased bioavailability, sit-targeting, and controlled/sustained drug release. This review highlights the anti-tumor properties of PTM that favor its repurposing for cancer treatment, as well as, PTM-based combination therapies and nanocarrier delivery systems which can enhance therapeutic efficacy and simultaneously reduce toxicity.

Keywords Pentamidine, Cancer, Cancer therapy, Drug repurposing, Combination therapy, Nanocarrier delivery systems

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Introduction

Being one of the leading causes of death, cancer has persistently been posing a great threat to human health and, subsequently, inflicting a significant financial burden on health organizations worldwide [1]. While there is currently a wide range of procedures and drugs adopted against this malignancy, including chemotherapy, surgery, radiotherapy, immunotherapy, hormone therapy, etc., unfortunately, in many cases, none could resolve the menace of cancer for good which might even claim the patients' lives, eventually [2]. On top of that, new findings are indicative of an alarming increase in cancer prevalence as statistics in 2022 show nearly 20 million newly diagnosed cases, as well as 9.7 million cancer-associated deaths worldwide [3]. For this reason, many attempts have been made to find and develop novel approaches for overcoming cancer with better efficacy and safety. About 90% of candidate pharmaceuticals fail to reach clinical trials owing to inadequate safety, efficacy, or both; therefore, in order to cut back on eye-watering financial expenses and time-consuming studies, as much as is feasible, one initiative approach that has emerged as a promising alternative is repurposing non-oncology drugs, such as diabetes medications, antihypertensives, antibiotics, antifibrotic agents, etc., for their off-label indications in the treatment of cancer [4, 5]. Indeed, with well-studied safety and side effect profiles and their unique mechanisms of action such medications may undergo a relatively rapid approval process and clear the way for a potentially successful course of therapy in cancer patients [6].

Given that humans and many parasite cells share great similarities in cellular mechanisms, for example, DNA replication, protein biosynthesis, mitochondrial function, and cellular metabolism, and that cancer cells heavily rely on these mechanisms for their rapid proliferation, many FDA-approved anti-parasitics such as niclosamide, ivermectin, albendazole, and pentamidine, among others, have been proposed for repurposing against cancer [7]. Indeed, due to having overlapping targets with antitumor agents, including microtubules, kinases, folate pathway, DNA topoisomerases, etc., anti-parasitics have shown promising anti-tumor properties induction of apoptosis, immune modulation, reduction of inflammation, disruption of cancer metabolism, overcoming drug resistance, and so on, and therefore, secured a place in the field of cancer research [8].

Aromatic diamidines are a group of compounds encompassing two amidine (-C(=NH)-NH2) functional groups attached to an aromatic ring, as shown in Fig. 1. Owing to their ability to bind to AT-rich regions of DNA, aromatic diamidines have shown significant

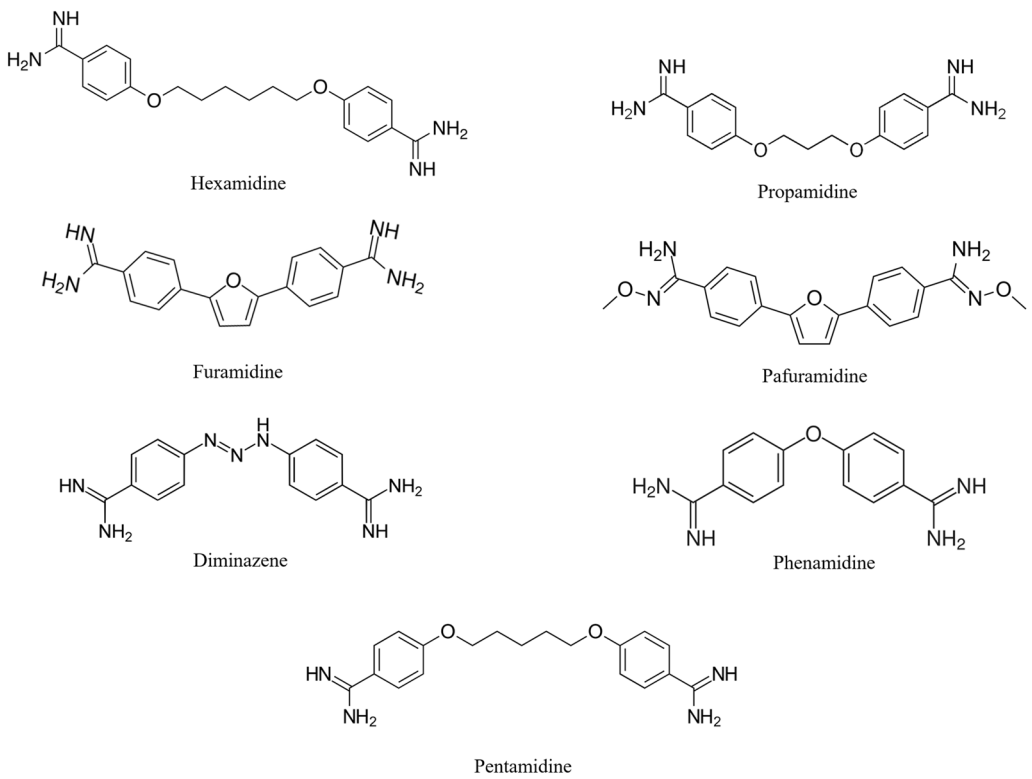

Fig. 1 The chemical structures of aromatic diamidines, a group of compounds with indications in antimicrobial and anti-parasitic treatments

antimicrobial and anti-parasitic effects, some of which, including propamidine, hexamidine, furamidine, pafuramidine, diminazene, phenamidine, and pentamidine are administered for treatment of some infections [9, 10]. Propamidine and hexamidine are antiseptic agents that in the form of eye drops are indicated for the treatment of some eye infections such as *Acanthamoeba* keratitis [11]. Pafuramidine is the O-methyl amidoxime prodrug of furamidine (DB75); it is an experimental compound, which was primarily granted orphan drug status by the FDA for the treatment of *Pneumocystis jiroveci* pneumonia in patients with HIV/AIDS [12]. Later, it also entered clinical trials for the treatment of first-stage human African trypanosomiasis but later failed due to late-onset toxicity most notably glomerulonephritis and nephropathy [13]. Diminazene and phenamidine are aromatic diamidines commonly used in veterinary for the treatment of infections such as cytauxzoonosis, trypanosomiasis, and babesiosis [14, 15].

Pentamidine (PTM) is another member of aromatic diamidines FDA-approved for the treatment of *Pneumocystis jiroveci* pneumonia in patients with HIV/AIDS which has not only been used against various other infections such as leishmaniasis and human African trypanosomiasis but also holds great promise in other contexts, especially in cancer [16]. Indeed, emerging evidence proposes that PTM has considerable anti-tumor effects on different types of cancer by blocking some signaling pathways, such as PI3K/AKT, mitochondrial targeting, DNA damage response inhibition, increasing reactive oxygen species (ROS) production, calcium homeostasis disruption, and gene expression modulation, which in turn inhibit cancer proliferation, stemness, invasion, migration, and angiogenesis while inducing apoptosis and anti-tumor immune responses [17–20]. In addition, some studies have reported that PTM displays synergistic effects in combination with other modalities such as chemotherapy and radiotherapy which might lead to the establishment of novel drug regimens for cancer therapy [21–23]. Furthermore, owing to potential side effects of its conventional administration such as nephrotoxicity, thrombocytopenia, leucopenia, hypoglycemia, etc., which could undesirably decrease compliance and success of therapy in clinical scenarios [16]. Nanocarrier-based drug delivery has been offered as a promising delivery tool that not only reduces the possible side effects but also improves the pharmacokinetic properties of PTM, including pharmacological half-life (t½), and bioavailability, among others [24].

Herein, we initially introduce PTM, its chemical structure, pharmacology, and both approved and off-lab indications. Importantly, to better appreciate the gravity of repurposing PTM in cancer therapy, we discuss the latest knowledge on the different anti-tumor properties of PTM and its combined effects with other anti-tumor agents. Lastly, we briefly take a peek at the nanomedicine-based strategies that have so far been employed to administer PTM against cancer with the goal of better therapeutic efficacy and lower risks of side effects.

Pentamidine; structure, pharmacology, and indications

PTM, chemically 4,4′-(Pentane-1,5-diylbis(oxy))dibenzimidamide, is a symmetric aromatic diamidine with a molecular formula of $C_{19}H_{24}N_4O_2$ that comprises a pentane-1,5-diol where both hydroxyl hydrogens are swapped for 4-amidinophenyl groups. It was first synthesized in the late 1930s by Yorke et al. PTM [25, 26]. Approved by the FDA, aerosolized PTM is used for the treatment and prevention of *Pneumocystis jirovecii* pneumonia -an opportunistic fungal infection- in high-risk HIV-infected patients. In addition, infectious diseases, including leishmaniasis, human African trypanosomiasis, and malarial infections are some of the well-known non-FDA-approved indications of PTM [16, 27]. The exact pharmacology of PTM, from both pharmacodynamic and pharmacokinetic perspectives, is not fully understood. Nonetheless, the findings suggest that PTM interferes with nuclear metabolism; it binds to transfer RNA, inhibits polyamine synthesis and RNA polymerase activity, and prevents the production of proteins, phospholipids, nucleic acids, and folate [3, 28]. The terminal polar amidine functional groups give PTM a significant topological polar surface area (TPSA) of 118 $Å^2$ which results in its poor gastrointestinal absorption and blood–brain barrier (BBB) penetration [24, 29]. PTM has been commercially available in the form of two salts, mesylate and isethionate. Besides, due to its poor absorption through the gastrointestinal tract, PTM is usually administered in the form of intravenous (IV) and intramuscular (IM) injections, as well as inhalation [24].

Pentamidine repurposing

With the concept of drug repurposing growing, attention has also been drawn to redirecting less frequently used medicines from the treatment of original diseases to other conditions [5]. Similarly, as illustrated in Fig. 2, recent studies have introduced PTM as a potential candidate for the treatment of non-parasitic diseases [16]. For example, studies have introduced PTM as a promising agent against Alzheimer's and Parkinson's diseases as, by inhibiting the N-methyl-D-aspartate (NMDA) receptor, PTM could show neuroprotective effects [30]. Recent findings suggest that PTM could block the SARS-CoV-2 3a channel, which makes it a potential candidate against COVID-19 [31]. Also, PTM has been found to reverse the

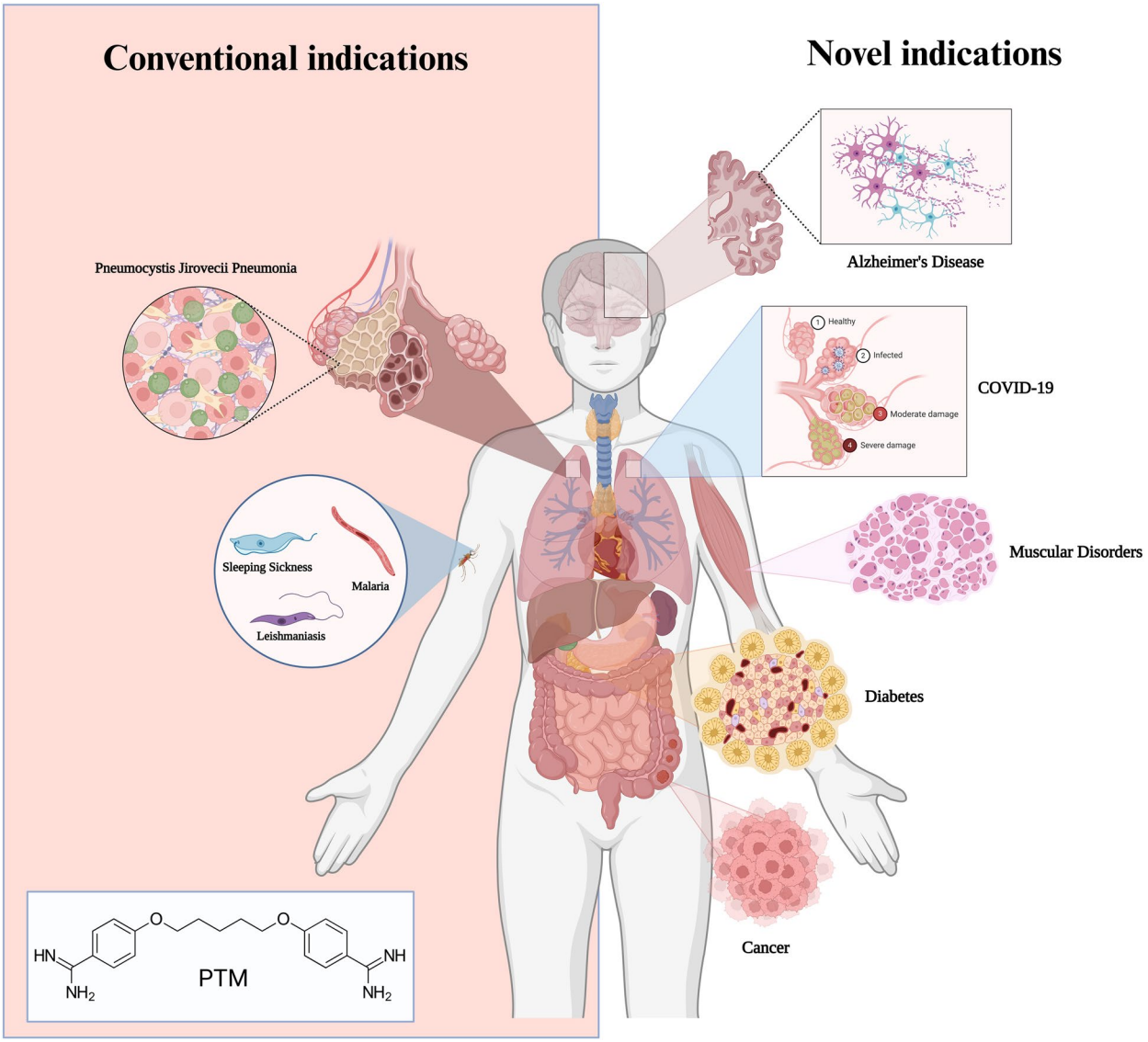

Fig. 2 Redirecting PTM administration from conventional applications to novel ones. Recent findings suggest that not only can PTM act as an anti-microbial agent but also exhibit other pharmacological characteristics that may be harnessed in other therapy fields such as the treatment of Alzheimer's disease, diabetes, muscular disorders, COVID-19, and cancer

splicing defects correlated with myotonic dystrophy type 1, a genetic disorder of autosomal dominant inheritance, which stems from the expansion of a CTG trinucleotide repeats in the noncoding region of the DMPK gene [32]. Due to its hypoglycemic effects, PTM can potentially be employed for the treatment of diabetes mellitus although some studies have demonstrated that such initial hypoglycemia might even lead to hyperglycemia as a consequence [33]. Moreover, a considerable body of evidence implies appealing effects of PTM against different types of cancer both in the form of monotherapy and combination therapy with other anti-cancer drugs [16, 24]. As such, PTM emerges as a promising agent that may be further investigated with the view to being translated into the clinics for the fulfillment of the aforementioned purposes.

Implications of PTM for cancer therapy

PTM has long been administered as antibiotic prophylaxis against *Pneumocystis jirovecii* pneumonia in cancer patients receiving immunosuppressive chemotherapy and/or radiotherapy [34]. Furthermore, a growing body of research, as shown in Fig. 3, has recently found PTM with appealing anti-tumor capacity with or without

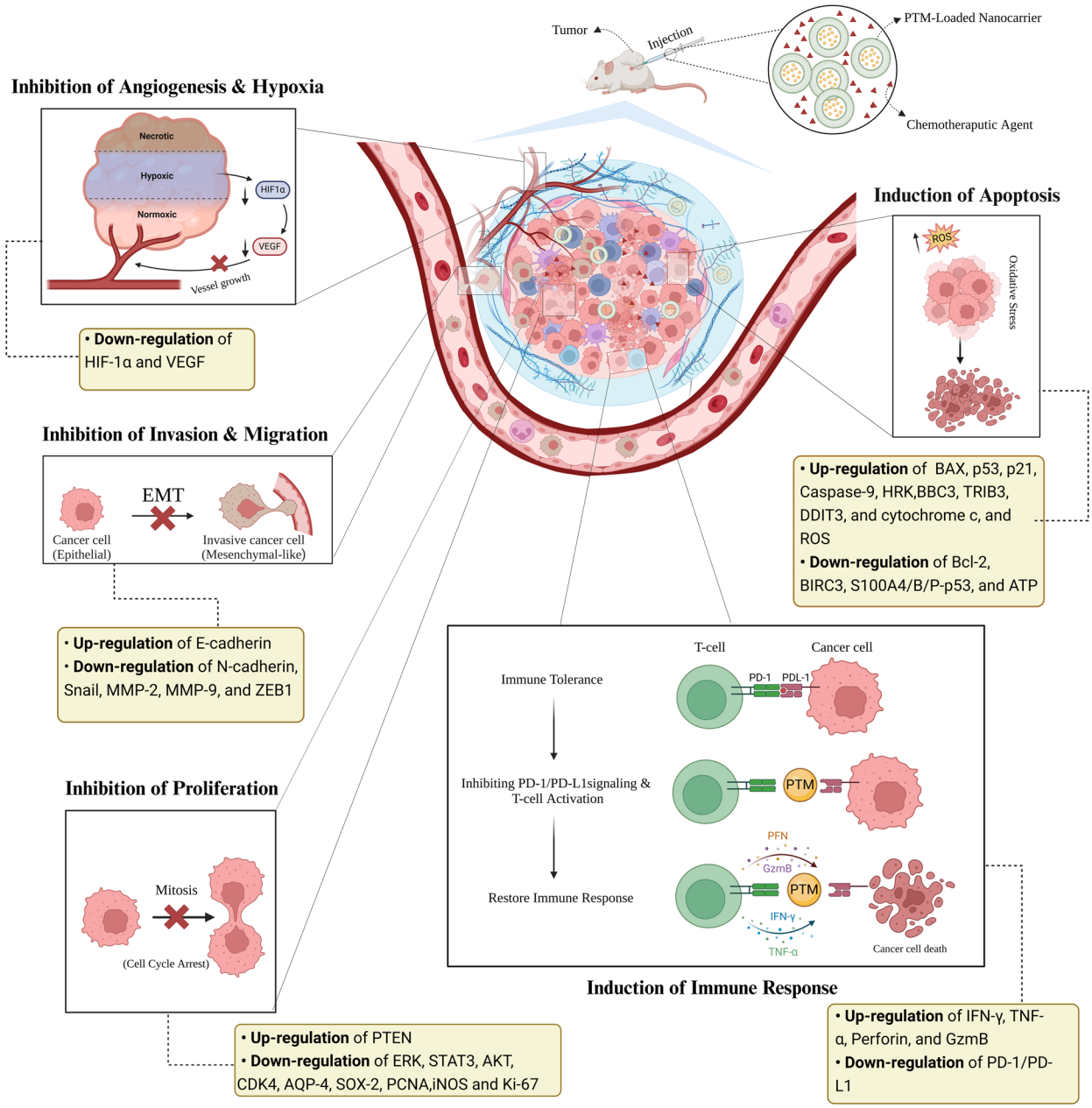

Fig. 3 Implications of PTM for Cancer Therapy. Growing evidence indicates that, by modulation of various pathways, PTM can show its anti-tumor effects through inhibition of cancer proliferation, invasion, migration, angiogenesis, and hypoxia while inducing apoptosis and restoring anti-tumor immune responses

other modalities in different types of cancer, including pancreatic [18], ovarian [35–37], prostate [38–40], lung [17, 22, 35, 41], breast [17, 19, 21, 38, 40–42], endometrial [43], and colon [17, 22, 35, 44, 45] cancers, as well as melanoma [17] and glioma [20, 40, 46]. Mechanistically, it inhibits signaling pathways such as PI3K/AKT [43], JAK/STAT [47], MAPK/ERK [45, 46], NF-κB [45], HIF-1α [40], Wnt/β-catenin [23], PD-1/PD-L1 [17] pathways while inducing p53 pathway[19, 20, 39, 42, 45, 46]. Indeed, PTM has been reported to interfere with cellular processes such as DNA and RNA synthesis, protein translation, mitosis, and calcium homeostasis in cancer cells which are critical to cancer progression [48]. The gravity of recent findings is to such an extent that several clinical trials have resultantly been conducted to investigate the eligibility of PTM for the treatment of various

types of cancer such as metastatic colon cancer [49, 50], stage IV non-small cell lung cancer [51], etc.; however, the results of them are still undisclosed. In the following, we scrutinize the latest knowledge regarding the anti-tumor properties of PTM that have made it a promising candidate in the field of cancer.

Proliferation and apoptosis

Deregulated cell proliferation and evasion of apoptosis are some of the well-known characteristics of tumors [52, 53]. Indeed, cancer cells may embrace specific strategies such as the upregulation of antiapoptotic factors and inhibition of apoptotic proteins to circumvent the programmed cell death apoptosis, which grants tumor cells an irregularly extended lifespan resulting in augmentation of tumorigenic mutations, and, subsequently, deregulated cell proliferation and perturbed differentiation [54]. For that reason, inhibition of proliferation and/or induction of apoptotic cell death has long been a chief goal in the development of anti-cancer therapeutics [55]. Remarkably, PTM has been found to inhibit cancer proliferation and tumor growth in a dose-dependent manner (mostly in the range of 5–20 μM in vitro) among various types of cancer [16]. For example, findings by Lin et al. demonstrated that PTM was able to inhibit the proliferation of endometrial carcinoma HEC-1A and Ishikawa cells dose-dependently, with the strongest inhibitory effect at 15 μM [43]. Indeed, PTM causes chromosomal segregation defects resulting in abnormal or inhibited mitosis in cancer cells [22]. Additionally, by inhibiting endo-exonucleases, PTM inhibits the DNA double-stranded break repair in cancer cells [56]. A study by Wu et al. has reported that 20 μM PTM in ovarian cancer cells can preserve the stability of the tumor suppressor PTEN, loss of or mutation in which is substantially correlated with cancer progression, through the ubiquitin/proteasome pathway, hence inhibiting the cancer proliferation [37]. Moreover, blockage of G1/S progression in cancer cells is another pronounced mechanism whereby PTM can suppress proliferation [57]. Indeed, by down-regulating phosphorylation levels of AKT protein, PTM at concentrations higher than 10 μM may increase the percentage of G1 phase cells while reducing the number of cells arrested in the S phase, and, subsequently, inhibit proliferation and growth in tumors [37, 43]. In vivo and in vitro (1 μM and 5 μM PTM) findings on glioma have identified PTM as a promising candidate against proliferation that plays an inhibitory role through a dose-dependent reduction in the expression of CDK4, STAT3, SOX-2, and Ki-67, as well as ERK activation [46]. Induction of apoptosis is another strategy, by which PTM may offer interesting inhibitory effects on proliferation [58]. Indeed, findings show that PTM up-regulates the expression levels of pro-apoptotic genes, such as BAX, p53, p21, BBC3, TRIB3 DDIT3, cleaved caspase-9, and HRK while down-regulating anti-apoptotic genes such as Bcl-2 and BIRC3 [19, 20, 39, 42, 46]. Katte and colleagues found that PTM up-regulated the expression of p53 protein in ZR-75-1 human breast cancer cells in a dose-dependent manner with effect at 20 μM [19]. The results of a study by Capoccia et al. indicated that 5 μM PTM (5 μM > 0.5 μM > 0.05 μM) had the strongest effects on the proliferation of C6 glioma cells, up-regulation of p53 and BAX, and down-regulation of Bcl-2 and AQP-4, highlighting a dose-dependent PTM cytotoxicity [20]. Moreover, PTM inhibits proliferation through the induction of mitochondrial DNA depletion and dysfunction [59]. Similarly, in the context of cancer, PTM has been found to suppress the growth of xenograft tumors of the prostate following morphological and functional impairment in mitochondria, ATP level reduction, increasing the expression levels of cytochrome c, down-regulation of the transcription levels of mitochondria-encoded genes, and, ultimately, inducing ROS production and apoptosis [39]. Another route, whereby PTM can inhibit proliferation and induce apoptosis, is targeting the interactions between the S100 family and other proteins [60, 61]. S100 proteins are a prominent family of widely expressed calcium-binding proteins, that play a crucial role in an ample range of cellular processes, including Ca^{2+} homeostasis, apoptosis, proliferation, differentiation, etc., and overexpression thereof has been reported in some types of cancer [62]. S100 proteins destabilize the wild-type p53 and form a complex of S100-p53 thus maintaining proliferation [63]. Interestingly, a growing body of evidence suggests that some of the S100 proteins, such as S100A4 [42], S100B [20], and S100P [19] share the same binding site for p53 and PTM. As a result, PTM can disrupt the interactions between these proteins and p53 which, in turn, reactivates the p53–p21 pathway and, ultimately, inhibits cell proliferation in cancer [64]. Indeed, a study on human colon cancer biopsies has revealed that not only does PTM disrupt S100B-activated RAGE/phosphor-p38 MAPK/NF-κB pathway and the S100B-p53 interaction, thereby restoring the p53-mediated apoptotic pathway, but also reduces the protein expression levels of PCNA, AQP4, IL-6, and inducible nitric oxide synthase (iNOS), as well as relative NO accumulation, which, collectively, lead to higher rates of apoptotic cell death and inhibition of proliferation [45]. Other findings also suggest that interactions between S100A1 and V domains of RAGE, a member of the immunoglobulin superfamily, can lead to RAGE dimerization, which has been found to highly contribute to cell proliferation and tumor growth [44].

Invasion and migration

Metastasis is reckoned the leading cause of death from cancer and frequently co-occurs with cancer recurrence [65]. Unfortunately, despite recent breakthroughs in cancer therapy, no efficient approach is yet available for patients with metastatic cancer. Thus, the research around this formidable issue has persistently been ongoing to hopefully devise novel anti-metastatic strategies [66, 67]. Relevantly, growing findings, both in vitro and in vivo, have introduced PTM as a possible agent against invasive properties in several types of cancer such as endometrial [43], ovarian [37], and prostate [39] cancer, as well as glioma [20]. Indeed, wound-healing assays and transwell experiments indicate that PTM can exhibit inhibitory effects on the migration ability of cancer cells in a dose- and time-dependently manner [43]. Besides, PTM has been shown to down-regulate the expression levels of MMP-2 [20, 43] and MMP-9 [43] -two prominent type IV collagenases- that play an important role in extracellular matrix (ECM) remodeling and facilitate the migration of cancer cells [68]. In addition, PTM could inhibit guanidinobenzoatase, a cell surface proteolytic enzyme, which, by degrading fibronectin throughout ECM, promotes cancer migration [69].

Glioblastoma (GBM) is the most aggressive and malignant type of glioma. Due to its intense invasiveness, GBM can diffuse into the healthy brain tissue and as a result make the surgical removal of tumors a very challenging task [70, 71]. Thus, the safest course may leave a portion of tumor cells intact and increase the risk of cancer recurrence [72]. Therefore, efforts have been made to develop new strategies to diminish the spread of these invasive glioma cells across the brain. Interestingly, a study by Capoccia et al. investigated the potential effect of PTM on fusiform C6 rat glioma cells that mimics human GBM behaviors following injection in brains of neonatal rats [20, 73]. Interestingly, they showed that 0.05, 0.5, and 5 μM PTM substantially impaired the migration of C6 rat glioma cells in vitro; in contrast, the untreated cells were able to invade and grow again in the scratched region [20]. Besides, PTM was able to down-regulate the expression levels of MMP-2, the overexpression of which is correlated with GBM invasion and poor prognosis [20, 74, 75]. Likewise, a study by Tamai et al. reported that PTM was able to reduce pseudopodia formation in both human GBM cell lines and human patient-derived cells, a morphological change that is correlated with cell mobility. In a similar manner, the results of the scratch assay showed the anti-migration effects of PTM on these cells [46, 76]. Moreover, they found that IM injection of PTM (150 μg PTM dissolved in 100 μL PBS with 5% DMSO every 2 days) was able to reduce the number of migrated cells and proliferation index in the xenograft mouse model of GBM [46]. Taken altogether, these findings implicate the potential of PTM as a promising candidate against invasion and migration of GBM cells which indeed calls for further studies in the future.

Epithelial-mesenchymal transition (EMT) is a crucial step in the initiation of metastasis, which naturally occurs during early embryogenesis and wound healing; however, it can also be appropriated by epithelial cancer cells, which in turn endows cells with invasive and migratory properties, as well as gene expression profile and morphology similar to mesenchymal cells [77]. Chemotherapy- and radiotherapy-mediated EMT has been recently shown to be a daunting challenge whereby the mesenchymal-like cells put up significant resistance to cytotoxic agents and highly correlate to poor prognosis in patients [78]. Interestingly, PTM has been found to notably reverse this transition as it modulates the expression levels of EMT markers. Indeed, gene expression studies show reduced expression levels of N-cadherin, snail, and ZEB1, along with increased levels of E-cadherin following treatment with PTM in clear cell renal cell carcinoma and ovarian cancer [23, 37]. Collectively, findings imply that PTM may be adopted against metastatic cancer cells in conjunction with other modalities, especially with those, that potentially induce EMT in malignant cells such as paclitaxel [79], cisplatin [80], fluorouracil [81], radiotherapy [82], etc.

Hypoxia and angiogenesis

Hypoxia is one of the hallmarks among many solid tumors that arises due to the rapid proliferation of cancer cells and insufficient performance of existing blood capillaries for supplying oxygen and nutrients, and it is highly associated with poor prognosis in patients [83]. Hypoxic tumors exhibit an extracellular pH significantly lower compared to their normal counterparts, which mostly stems from the reshaped metabolism of glucose and leads to activation of the alpha/beta subunit of a heterodimeric transcription factor hypoxia-inducible factor-1 (HIF-1) [84, 85]. Active HIF-1 modulates the expression levels of an assortment of genes associated with proliferation, apoptosis, metabolism, pH regulation, angiogenesis, migration, and metastasis [86]. Thus far, several agents, such as small molecules (FM19G11, IDF-11774, and KC7F2), monoclonal antibodies (mAbs) (cetuximab and trastuzumab), siRNAs, and histone deacetylase inhibitors (apigenin, trichostatin A, and suberoylanilide hydroxamic acid), and nanobodies, have been suggested to target this protein [87–89].

Interestingly, PTM has also been found to inhibit the expression of HIF-1α, an important subunit of HIF-1, in prostate and breast cancer, as well as glioma [40]. In fact, findings propose that PTM, in a time- and

dose-dependent manner, has been able to reduce HIF-1α protein expression, likely through decreasing HIF-1α protein stability and inhibiting protein translation. Such reduction, mediated by PTM, could reduce hypoxia-induced promoter activity of iNOS—a calcium-dependent isoform of NOS enzyme- and down-regulate the expression levels of glucose transporter-1 (GLUT-1), which plays an important role in glucose metabolism via facilitating the entry of glucose into tumor cells [40, 45]. Moreover, as mentioned above, angiogenesis has been described as an indispensable part of cancer progression proceeding from tumor hypoxia and stimulated by several angiogenic factors such as VEGF, PDGF, FGF, and IGF [90]. Having an outstanding contribution to both cancer growth and metastasis, tumor angiogenesis acts as a limiting factor and is regarded as a promising target for cancer therapy, hence the development of angiogenesis inhibitors, for example, bevacizumab, cabozantinib, and sunitinib, among others [91]. Similarly, PTM also shows anti-angiogenic implications in cancer; indeed, it has been found that PTM dose-dependently reduced hypoxia-induced production of VEGF and capillary tube formation in breast, colon, and prostate cancer [40, 45]. Taken altogether, PTM might be a promising candidate against different hypoxia-mediated tumorigenic effects, especially angiogenesis; however, to reach a better understating of underlying mechanisms for this indication of PTM and possible adverse effects thereof, further investigations are absolutely necessary in view of any prospective translation into the clinics.

Immune evasion

The immune system plays an indispensable part in checking tumor progression; once compromised, it will profoundly lead to a highly increased risk of cancer [92]. Several lineages of immune cells contribute to anti-tumor responses, including natural killer (NK) cells, cytotoxic T lymphocytes (CTLs; CD8+), B lymphocytes, and macrophages, among others [93, 94]. Despite the elaborate immune surveillance, there have been found a handful of escape mechanisms whereby tumors may circumvent the hostile immune actions including the PD-1/PD-L1 pathway, down-regulation of MHC class I, overexpression of anti-apoptotic factors, for example, Bcl-2 proteins, secretion of cytokines such as IL-10 and TGF-β, as well as immunosuppressive exosomes, infiltration of regulatory T cells (Tregs) and myeloid-derived suppressor cells (MDSCs) within the tumor microenvironment (TME) [95, 96]. Clinically, mAbs, checkpoint inhibitors, vaccines such as T-VEC, CAR T-cell therapy, and cytokines such as aldesleukin and interferon are common modalities adopted to restore anti-tumor immunity and elicit stronger immune responses [97].

Several non-oncology compounds, including melatonin, metformin, tetracycline, amphotericin B, etc., have recently been found to aid the immune system in recognizing and attacking cancer cells, which can be co-administered with well-established immunotherapy drugs to boost the immune responses against tumor progression more effectively [98, 99]. Similarly, it has been found that PTM could manifest interesting immunomodulatory effects [100]. Indeed, PTM has been able to up-regulate the protein levels of cytokines IFN-γ, TNF-α, perforin, and Granzyme B, the production of which are vital for the maintenance of $CD8^+$ T cell cytotoxicity against cancer cells [17]. Pharmacological findings also suggest that PTM can restore anti-tumor response from the immune system via blocking PD-L1 and, subsequently, restricting PD-1 /PD-L1 interaction, which plays an important role in tumor-mediated immunosuppression and development of self-tolerance via exhausting T-cells, induction of apoptosis among antigen-specific T cells, and inhibition of apoptosis in Tregs [101]. Moreover, following intraperitoneal (IP) administration of PTM, a PD-L1 humanized syngeneic mouse model of breast cancer has been found with an increased number of tumor-infiltrating lymphocytes and $CD8^+/FoxP3^+$ ratio in T cells, which, indeed, reflects the reinvigoration of tumor-infiltrating lymphocyte-mediated immune responses [17, 102]. Although limited, these findings provide an uncharted perspective on the immunomodulatory capacity of PTM for cancer therapy which undoubtedly calls for further research to better delineate its impact on tumor progression most notably in combination with other immunotherapy drugs.

Clinical trials investigating PTM repurposing in cancer therapy

Clinical trials are a crucial step for clinical translation that certify the safety and efficacy of new treatment options, such as drug repurposing, in human subjects before being widely used in the clinics. Like many other repurposed non-oncology drugs, PTM has entered several clinical trials for evaluation of its eligibility in cancer therapy chiefly for refractory tumors where available modalities usually fail (Table 1). For example, a phase I/II clinical study (NCT00809796) in 2008 was conducted to evaluate the safety of PTM in patients with metastatic colon cancer receiving standard Folinic Acid, 5-Fluorouracil (5-FU), or capecitabine and oxaliplatin Chemotherapy as second-line and/or third-line treatment [49]. Likewise, a clinical trial was held among patients with metastatic pancreatic cancer to evaluate the safety and potential efficacy of PTM administration in conjunction with standard therapy [103]. In 2011, a phase II clinical

Table 1 The list of clinical trials investigating the anti-cancer properties of PTM

Cancer type	ClinicalTrials.gov ID	Intervention	Outcome measures	Study period	Phase	Reference
Relapsed/Refractory Classical Hodgkin Lymphoma	NCT03730363	PTM; administered as an IV infusion on treatment day 1–3 of a 21-day cycle 3 cycles using 2, 3, and 4 mg/kg dose escalation schedules. Ifosfamide; 5000 mg/m2. Carboplatin; 5 area under curve (AUC). Etoposide; 100 mg/mg^2 by IV infusion	Maximum tolerated dose (MTD), best overall response, defining the duration of response, identifying immunohistochemistry biomarkers, PCR analysis of biomarkers, identifying phosphorylation biomarkers, identifying soluble CD30 (sCD30) and TARC biomarkers, and measuring circulating-free (cf)mRNA and cfDNA biomarkers	2018–2019	Phase I	[104]
Hepatocellular Carcinoma	NCT02210182	PTM; orally given at 300 mg, 600 mg, 900 mg or 1200 mg QD × 3 consecutive days	Pharmacokinetics of PTM including liver and plasma concentrations of PTM after oral administration, adverse events, the tissue biomarker of endoexonuclease, and the levels of plasma pharmacodynamic markers of efficacy, namely ALT and AST	2014–2015	Phase I	[105]

Table 1 (continued)

Cancer type	ClinicalTrials.gov ID	Intervention	Outcome measures	Study period	Phase	Reference
Stage IV Non-Small Cell Lung Cancer	NCT01844791	PTM in combination with chemotherapy, including platinum and gemcitabine, each cycle (dosings not mentioned)	Overall response	2012–2014	Phase II	[51]
Unresectable and Locally Recurrent or Metastatic Colorectal Cancer	NCT01378143	PTM in combination with mFOLFOX6 or FOLFIRI as standard of care (dosings not mentioned)	Tumor size, progression free survival (PFS), overall survival (OS), peak plasma concentration (Cmax) of PTM, number of participants with adverse events as a measure of safety and tolerability, objective response (OR), duration of response (DR)	2011–2014	Phase II	[50]
Locally advanced or metastatic pancreatic cancer	NCT00810953	PTM; two dose of 6 mg/kg with or without standard chemotherapy	Any severe events, tumor marker CA19-9, and tumor size	2009–2011	Phase I/II	[103]
Metastatic Colon Cancer	NCT00809796	PTM; one dose of 4 mg/kg to two dose of 4 mg/kg with possibility of escalated to two dose of 6 mg/kg with or without standard chemotherapy (folinic acid, 5-fluorouracil) or else (capecitabine and oxaliplatin)	Any severe events, tumor marker CEA, and tumor size	2008–2011	Phase I/II	[49]

Table 1 (continued)

Cancer type		ClinicalTrials.gov ID	Intervention	Outcome measures	Study period	Phase	Reference
Relapsed or Refractory Melanoma	NCT00729807		PTM; IV over 2 h 5 days a week for 2 weeks	Number of participants with both p21 and S100B expression in accessible tumor biopsies pre and post pentamidine exposure in cycle 1, expression of S100B before and during pentamidine exposure, number of participants with serious and non-serious adverse events, time to progression	2008–2012	Phase II	[106]

study (NCT01378143) started to investigate the safety and efficacy of the PTM administration in combination with standard chemotherapy regimen, either folinic acid, fluorouracil, and irinotecan hydrochloride (FOLFIRI) or 5-FU, leucovorin, and oxaliplatin (mFOLFOX6) as a second line treatment in patients diagnosed with unresectable and locally recurrent or metastatic colorectal cancer [50]. A single-arm, open-label investigation (NCT01844791), in 2013, was performed to evaluate the safety and efficacy of PTM combined with gemcitabine and platinum-based first-line therapy in patients with the late-stage non-small cell lung cancer (NSCLC) [51]. A Phase I clinical trial of PTM combined with salvage chemotherapy, including ifosfamide, carboplatin, and etoposide (ICE), for recurrent or resistant Hodgkin's lymphoma [104]. Another phase I study (NCT02210182) investigated the pharmacokinetic profile of PTM, such as hepatic uptake and PTM serum levels, as well as the safety and tolerance of a new oral formulation of PTM in patients with hepatocellular carcinoma who underwent thermal ablation [105]. Unfortunately, for some reason, none of the abovementioned clinical trials have so far disclosed the results thereof. However, a recent clinical study (NCT00729807) has yielded considerable outcomes. Indeed, this investigation was to explore the response rate in individuals diagnosed with relapsed/refractory melanoma, which is known for the up-regulated levels of the S100B-p53 complex, following slow infusion of PTM isethionate (4 mg/kg/day) with each therapy cycle including 2 weeks of treatment (5 days/week) and, then, 2 weeks of monitoring. The results clearly showed that PTM can lead to a partial response and reduction in the sum of the longest diameter of target lesions and lower serum levels of S100B, which is reportedly associated with a better prognosis. However, there were serious adverse effects observed following PTM administration, most notably, infection and hypoglycemia, for which patients were hospitalized and patients' course of therapy was subsequently halted. The severity of these events was to such an extent that resulted in the termination of this study at the suggestion of the Data and Safety Monitoring Board (DSMB) prior to obtaining target enrollment [106].

Altogether, findings indicate that while PTM might efficiently reduce tumor progression it could also lead to the emergence of daunting challenges such as adverse effects, as described above, comprise patient compliance and, eventually, reduce the likelihood of PTM clinical translation at the current form, which indeed calls for further initiatives regarding this matter as discussed in the following sections.

Innovative approaches for PTM administration

As discussed earlier, repurposing PTM for cancer therapy may create a remarkable avenue for expanding its indications beyond infections [16]. However, like many other clinical translations, certain challenges may emerge on this path that need to be resolved. As the few clinical trials, using PTM in cancer therapy reflect, conventional PTM administration has de facto been proposed as salvage therapy in patients with metastatic cancers and, obviously, very poor prognosis. Indeed, systemic administration of PTM could be accompanied by some serious adverse effects, most notably, nephrotoxicity and hypoglycemia, and thus, should be administered with extra caution [24]. Besides, by adopting different strategies, such as inhibition of apoptosis, enhanced drug efflux activity, and accelerated DNA repair, among others, there is always the possibility that tumor cells secure resistance to PTM, which is one of the daunting hurdles in the course of cancer therapy [107, 108]; as cytotoxic drugs are usually wanting in a broad therapeutic index, administration of higher than recommended doses thereof may inflict unrecoverable damage and even death in individuals [109]. Besides, although some studies suggest that GBM can disrupt BBB integrity, due to its poor BBB penetration and low delivery into the brain, PTM administration may face considerable challenges in the treatment of different types of brain tumors [24]. Therefore, in order to reduce the administered doses of PTM, lower the risk of adverse effects, overcome resistance, maintain the therapeutic efficacy, improve the BBB penetration (in case of brain tumors), and, overall, improve the clinical significance of PTM for cancer therapy, even for patients with low-grade cancer, several approaches such as combination therapy and nanocarrier drug delivery systems have been proposed. In the following subsections, we bring outstanding findings regarding PTM-based combination therapy and nanocarrier systems, as well as, consideration for improving BBB penetration of PTM which may be translated into cancer research.

PTM-based combination therapy

Espoused by oncologists, combination therapy is one of the possible courses of action that involves the concurrent adoption of multiple modalities, including chemotherapy, hormone therapy, radiation therapy, etc., and improves efficacy, prevents resistance, and lowers the risk of toxicity [110]. As its name suggests, by combining different anti-cancer therapeutics, combination therapy targets tumor cells with significant additive or synergistic anti-tumor effects via a wide range of mechanisms [111]. Moreover, combination therapy may adopt a unique strategy known as synthetic lethality which occurs when

the concurrent disruption of two or more genes causes cell death, whereas the deficiency of only one gene is usually unable to do so [112, 113]. As a case in point, BRCA1 and BRCA2 are two renowned proteins that play a great role in homologous recombination (HR) in the repair of DNA double-stranded breaks (DSBs); once mutated, as typically observed in ovarian and breast cancers, cancer cells become highly dependent on the enzyme poly(ADP-ribose) polymerase (PARP) that can mediate alternative DNA repair mechanisms in cancer cells [114]. Therefore, in the case of BRCA-mutant cancers, PARP inhibitors, such as talazoparib, olaparib, niraparib, etc., can block these alternative pathways and lead to synthetic lethality, which can even lead to synergistic effects once combined with DNA-damaging agents such as cisplatin, doxorubicin, and temozolomide, among others [115, 116].

With the concept of drug repurposing garnering ever-increasing attention for cancer therapy, many efforts have also been dedicated to investigating the efficacy of these medications, such as metformin, sildenafil, chlorpromazine, and aspirin, in conjunction with the aforementioned anti-cancer modalities in preclinical and/or clinical models [117, 118]. Notably, some studies have indicated the considerable potential of PTM for combination therapy against cancer cells (Table 2) [18, 21–23, 43]. It has been reported that a combination of PTM and PEG-stabilized gold nanoparticles (PEG-AuNPs) can increase the sensitivity of triple-negative breast cancer (TNBC) cells to radiotherapy. Interestingly, the adsorption of PTM onto the surfaces of PEG-AuNPs could enhance the cellular uptake of gold compared to the nanoparticle alone, which in turn leads to a significantly greater number of residual DNA double-strand breaks [21]. PTM also synergizes with the antipsychotic chlorpromazine against cancer proliferation; mechanistically, PTM causes chromosomal segregation defects, and chlorpromazine inhibits mitotic kinesin KSP/Eg5 and accumulates monopolar spindles, which collectively leads to dual inhibition of mitosis and, consequently, synergistic antiproliferative effects against cancer cells. Of note, this aforementioned combination has also exhibited considerable synergism with microtubule-binding agents, such as vinorelbine and paclitaxel, as evidenced by lower tumor volumes in xenograft models of cancer [22]. Likewise, once combined with non-oncology drugs such as oligomycin and amitriptyline, PTM has been found to synergistically induce apoptosis in renal cell carcinoma [23]. Other findings have demonstrated that PTM can improve the inhibitory effects of LY294002—a potent chemical inhibitor of PI3Ks—on cancer proliferation. Indeed, a combination of PTM and LY294002 led to increased repression of the PI3K/AKT pathway and, eventually, inhibition of proliferation in endometrial cancer [43]. Moreover, a recent study on animal models of pancreatic cancer suggests that pharmacological inhibition of the polyamine biosynthesis enzyme SAT1 by PTM can synergize the anti-tumor efficacy of FOLFIRINOX regimen -including folinic acid, fluorouracil, irinotecan hydrochloride, and oxaliplatin- which is the current standard of care therapy for patients with pancreatic cancer [18, 119]. Furthermore, PTM has found its way into some clinical trials with the intention of PTM-based combination therapy; however, in most cases, no study results are still provided by the responsible party. For example, a single-arm, open-label study investigated both the safety and efficacy of co-administering PTM and platinum-gemcitabine-based doublet chemotherapy in patients with stage IV non-small cell lung cancer [51]. In addition, a phase I/II clinical trial was conducted to study the safety of PTM in subjects with metastatic colon cancer receiving standard leucovorin, fluorouracil, or capecitabine and oxaliplatin chemotherapy as second- and/or third-line treatment [49]. Although, in the case of PTM, the concept of synthetic lethality has never been put to the test explicitly, there

Table 2 The list of studies employing PTM with other anti-cancer modalities in the form of combination therapy

Combination	Cancer type	Subject	Advantage	Reference
Radiotherapy + gold nanoparticles + PTM	Triple Negative Breast Cancer	MDA-MB-231 and MDA-MB-436 cells	Increase in cytotoxicity, radiosensitization, and number of residual DNA double-strand breaks compared to radiation alone	[21]
Chlorpromazine + PTM	lung cancer and colon cancer	Severe combined immunodeficient Hsd:ICR mice bearing A549 or HCT116 cells	Inhibiting tumor growth more effectively than either PTM or chlorpromazine alone	[22]
LY294002 + PTM	Endometrial cancer	Ishikawa cells	Increased inhibition of cell viability and the protein expression of p-AKT	[43]
FOLFIRINOX + PTM	Pancreatic cancer	Athymic nude mice bearing S2-013 and HPAF-II cells	Improved efficacy of FOLFIRINOX, a significant decrease in the tumor growth kinetics, reduced tumor volume and weight parameters	[18]

are interesting, yet limited, implications that may support its potential for this matter. For example, findings in parasites have shown that PTM can exhibit inhibitory effects on topoisomerase II [120], an enzyme, that plays an important role in the HR repair pathway in BRCA-deficient cells [121], which highlights the hypothetical synthetic lethality with PTM most notably in combination with PARP inhibitors and DNA-damaging agents, as mentioned above. By and large, PTM has come into view as a compelling candidate for combination therapy, but any possible clinical translation of PTM for combination therapy indeed requires further investigation regarding its safety and efficacy.

Nanocarrier PTM delivery systems

Nanocarrier drug delivery systems such as liposomes, noisomes, dendrimers, micelles, etc., have gained growing popularity for cancer treatment. With enhanced pharmaceutical characteristics over conventional chemotherapy, including the capacity to encapsulate both hydrophobic and hydrophilic agents, increased solubility and bioavailability, site-targeting, and controlled and/or sustained release, among others, nanocarrier drug delivery systems offer lower risks of adverse effects, higher therapeutic efficacy, reduced dosage frequency, and, eventually, improves patient compliance [122, 123]. Due to poor oral bioavailability, commercially available formulations of PTM have been limited to lyophilized powders for IM and/or IV injection (Pentacarinat® and Pentam®), as well as, inhalable powders (Nebupent®) and aerosols (Pneumopent®) [24]. However, following systemic PTM delivery with these approved formulations, the emergence of adverse effects is mostly inevitable, and, co-administration of PTM and chemotherapeutic agents with formidable cytotoxicity in cancer patients may add insult to injury, worsening the prognosis [124]. To tackle this alarming issue, nanocarrier drug delivery systems have been suggested as a promising approach for PTM administration, using different preparation techniques. In the following, we discuss the very latest nanotechnological advances for PTM administration against cancer (Table 3) as Andreana et al. specifically covered this area in a previous review article [24].

Liposomes are small vesicles widely used for novel drug delivery systems. They have one or more concentric phospholipid bilayers surrounding an aqueous solution core [125]. Findings on tumor-bearing mice demonstrate that PTM-loaded liposomes, prepared by the thin-film hydration method, enhance biodistribution and tumor accumulation, and limit kidney drug levels, hence the lower risk of nephrotoxicity compared to the free drug [35]. Noisomes are another type of drug delivery system that consist of non-ionic surfactant vesicles and have several advantages compared to liposomes, for example, higher physical stability of formulation and pronounced cost-effectiveness [126]. As a result, they have recently been adopted in many cancer studies. Seguella et al. coated PTM-loaded niosomes with chitosan in order to improve mucoadhesion with mucosal tissues and accelerate interpenetration. These chitosan-coated niosomes of PTM showed substantially increased drug permeabilization in human biopsies of colon cancer [45]. Andreana and colleagues developed squalene-based nanoparticles of PTM (SQ-COOH/PTM-B) using ion-pairing at physiological pH (7.4) between the negatively charged squalene derivative (1,1′,2-tris-norsqualenoic acid) and the positively charged base free form of PTM, squalene-based nanoparticles of PTM (SQ-COOH/PTM-B), which required no surfactant for carrier stabilization. Given the high affinity of squalene for low-density lipoproteins (LDL) and preferential concentration in cells with LDL receptor (LDLR) overexpression, findings interestingly demonstrated that SQ-nanoparticles could deliver a higher intracellular amount of PTM-B via an LDLR-mediated approach. Indeed, the produced nanoparticles of PTM showed interestingly superior cytotoxicity compared to PTM-B in cancer cells with LDLR overexpression such as HepG2 (hepatocellular carcinoma), PC-3 (grade IV prostatic adenocarcinoma), MDA-MB-231 (TNBC) cells, whereas such a difference was not found between the two in cancer cells with undetectable or low LDLR levels such as MCF7 and LNCap cells [38, 127]. Poly(lactic-co-glycolic acid) (PLGA) nanoparticles are another commonly used type of drug delivery system. PLGA, a copolymer of lactic acid and glycolic acid, breaks down into safe by-products, namely lactic acid and glycolic acid, which are easily metabolized and removed from the body [128]. Stella et al., synthesized PTM-loaded nanoparticles, following ionic interactions between PTM and PLGA, which could notably release PTM in a pH-dependent manner. Remarkably, the PTM-loaded PLGA nanoparticles showed higher cytotoxicity against cancer cells compared to liposomal PTM, which was proposedly caused by the kinetics of matrix degradation that could affect the drug release, considering that PTM is dispersed within the polymer matrix of PLGA nanoparticles, as for liposomal PTM, it is located in the central aqueous core [36]. An in vitro study by Carton et al. reported that the interaction between the cationic PTM and the anionic hyaluronic acid (HA) led to the formation of polyelectrolyte complexes (PECs). Furthermore, to formulate nanoparticles, they applied polyarginine (PArg), a biocompatible cationic poly(aminoacid), which crosslinked HA, stabilized PTM-HA PECs, augmented the concentration of loaded PTM, and enhanced intracellular delivery. The produced HA-PArg NPs

Table 3 The description of PTM-loaded nanocarriers against cancer

Formulation	Preparation technique	Cancer type	Subject	Advantage	Reference
Squalene-based nanoparticles of PTM	Nanoprecipitation	Breast, prostate, and liver cancer	MCF7, MDA-MB-231, LNCap, PC-3, and HepG2 cells	A superior cytotoxicity compared to free PTM-B and preferential accumulation in cells overexpressing the LDL receptor	[38]
PTM-hyaluronic acid polyelectrolyte complexes	Polyelectrolyte complexation	Lung and breast cancer	A549 and MDA-MB-231 cells	More cytotoxicity in comparison to the free drug and enhanced internalization of encapsulated drugs by cancer cells	[41]
Pegylated liposomal PTM	Thin film hydration	Colorectal, ovarian, lung, and breast cancer	Female SCID mice bearing subcutaneous HT29, A549, SKOV3, or orthotopic MDA-MB-231 tumors	Improved biodistribution and tumor accumulation, as well as decreased kidney drug levels compared to the free drug	[35]
Chitosan coated niosome of PTM	Thin film hydration	Colon cancer	Human biopsies of colon cancer	Maximized drug permeabilization in the tissue	[45]
PTM-loaded poly(lactide-co-glycolide) nanoparticles & Liposomal PTM	Nanoprecipitation & Thin film hydration	Ovarian cancer	A2780 cells	Higher cytotoxicity of PLGA nanoparticles compared to liposomes	[36]
PTM-loaded PLGA/HA-DPPE nanoparticles	Nanoprecipitation	Breast cancer	MCF-7 and MDA-MB-231 cells	Higher cytotoxicity of PTM-B-loaded PLGA/HA-DPPE nanoparticles compared to the free PTM-B and PTM-B-loaded PLGA nanoparticles in the CD44-expressing cells	[129]

loaded with PTM showed stronger cytotoxicity against human lung and breast cancer cells compared to that of the free PTM [41]. Moreover, another recent study by Andreana et al. took the initiative and synthesized a hybrid of lipid and polymer-based nanosystems. To this end, HA-phospholipid conjugate (HA-DPPE) was added during nanoprecipitation of PLGA nanoparticles where, by ionic interactions, PTM was encapsulated. With HA presented at the surface, the hybrid nanosystem, preferentially targeted cancer cells with overexpression of CD44, an important cell surface receptor of HA. In fact, the aforementioned nanoparticles showed greater cytotoxic effects on C44-overexpressing MDA-MB-231 cells in comparison to the low-C44-expressing MCF7 cells due to endocytosis-triggered mechanism by CD44 [129, 130].

Overall, the aforementioned PTM nanoparticles have exhibited improved tumor targeting, intracellular delivery, and cytotoxicity in cancer cells and, therefore, administering them can increase the therapeutic efficacy of PTM, minimize the toxicity in comparison to its free form, and, eventually bring interesting aspects to future clinical trials investigation PTM in cancer therapy.

Considerations for improving BBB penetration of pentamidine

As mentioned earlier, PTM has shown interesting inhibitory effects on glioma cells both in vitro and in vivo. Some findings suggest that in both primary and metastatic brain tumors, BBB integrity may be disrupted and become more permeable which may further offset poor and slow BBB penetration of PTM [131, 132]. In support of this event, a study by Tamai et al. showed that IM injection of PTM (150 µg PTM dissolved in 100 µL PBS with 5% DMSO every 2 days) was able to significantly reduce the proliferation and migration of glioma cells in a xenograft mouse model of GBM [46]. On the other hand, growing evidence indicates that in many cases of GBM, BBB indeed remains intact [133]. Therefore, developing novel approaches to increase PTM delivery towards brain tumors is of great importance. To date, several approaches have been proposed in this regard yet none has been evaluated for BBB penetration in the field of cancer. Implementing these approaches, including synthesizing lipophilic PTM prodrugs (such as diacetyldiamidoximeester derivative [134]) and bioconjugates (hyaluronic acid-PTM and PLGA-PEG copolymer-PTM [135]), nanocarriers (e.g. polycaprolactone nanoparticles, phosphatidylcholine liposomes [136], and chitosan glutamate-coated niosomes [137]), sucrose-corrected distribution of PTM with or without adenosine perfusion, coadministration of PTM with the P-glycoprotein and/or MRP efflux pump inhibitors such as indomethacin [138], and intranasal delivery [139], might circumvent BBB and deliver higher doses of PTM into the brain. Indeed, in the field of cancer, these are only hypothetical approaches that warrant future studies in this matter in the hope of modifying PTM's pharmacokinetic properties for higher BBB penetration and attacking brain tumors.

Conclusion and future perspectives

Having the promising anti-tumor capacity, PTM may be harnessed by innovative approaches, such as combination therapy and nanodrug delivery systems, to reach better efficacy and lower toxicity in cancer therapy. Nevertheless, PTM administration for the treatment of cancer still harbors uncertainty over some areas that clearly need to be resolved for any clinical translation in the future. For example, further research needs to delineate the exact mechanisms whereby PTM manifests the aforementioned anti-tumor properties, which may even advance its utility in combination therapies. Moreover, future investigations focusing on novel PTM-based combinations with other chemotherapeutic agents and anti-cancer modalities may lead to the establishment of treatment approaches with higher therapeutic efficacy. Developing state-of-the-art nanocarrier systems with the goal of optimizing PTM's pharmacokinetic properties while minimizing systemic toxicity is of the essence. In addition, PTM repurposing for the treatment of brain tumors may face pharmacokinetic challenges most notably poor BBB penetration which needs to be addressed in future studies. In the end, PTM remains an intriguing drug with great anti-tumor potential that may open up a whole new avenue for cancer therapy in the near future.

Abbreviations

AT	Adenine and thymine
PTM	Pentamidine
t½	Pharmacological half-life
IV	Intravenous
IM	Intramuscular
TPSA	Topological polar surface area
BBB	Blood–brain barrier
NMDA	N-methyl-D-aspartate
DMPK	DM1 protein kinase
PTEN	Phosphatase and tensin homolog
AKT	Protein kinase B
CDK4	Cyclin-dependent kinase 4
STAT3	Signal transducer and activator of transcription 3
SOX-2	SRY-box transcription factor 2
AQP-4	Aquaporin-4
ERK	Extracellular signal-regulated kinase
BAX	BCL2 associated X
BBC3	BCL2 binding component 3
TRIB3	Tribbles pseudokinase 3
DDIT3	DNA damage inducible transcript 3
HRK	Harakiri, BCL2 interacting protein
BIRC3	Baculoviral IAP repeat containing 3
RAGE	Receptor for advanced glycation end products
ROS	Reactive oxygen species
MAPK	Mitogen-activated protein kinase
PCNA	Proliferating cell nuclear antigen
IL-6	Interleukin 6

ECM	Extracellular matrix
GBM	Glioblastoma
PBS	Phosphate-buffered saline
DMSO	Dimethyl sulfoxide
EMT	Epithelial-mesenchymal transition
MMP	Matrix metalloproteinase
ZEB1	Zinc finger E-box binding homeobox 1
HIF-1	Hypoxia-inducible factor-1
MAbs	Monoclonal antibodies
iNOS	Inducible nitric oxide synthase
GLUT-1	Glucose transporter-1
VEGF	Vascular endothelial growth factor
PDGF	Platelet-derived growth factor
FGF	Fibroblast growth factor
IGF	Insulin-like growth factor
CTLs	Cytotoxic T lymphocytes
Tregs	Regulatory T cells
NK	Natural killer
MDSCs	Myeloid-derived suppressor cells
PD-1	Programmed cell death protein 1
PD-L1	Programmed death-ligand 1
MHC	Major histocompatibility complex
TGF-β	Transforming growth factor beta
TME	Tumor microenvironment
T-VEC	Talimogene laherparepvec
IFN-γ	Interferon gamma
TNF-α	Tumor Necrosis Factor Alpha
FOXP3	Forkhead box P3
IP	Intraperitoneal
5-FU	5-Fluorouracil
FOLFIRI	Folinic acid, fluorouracil, and irinotecan hydrochloride
mFOLFOX6	5-FU, leucovorin, and oxaliplatin
NSCLC	Non-small cell lung cancer
ICE	Ifosfamide, carboplatin, and etoposide
DSMB	Data and Safety Monitoring Board
MTD	Maximum tolerated dose
sCD30	Soluble CD30
cfmRNA	Circulating-free mRNA
TARC	Thymus and activation regulated chemokine
ALT	Alanine aminotransferase
AST	Aspartate aminotransferase
PFS	Progression free survival
OS	Overall survival
Cmax	Peak plasma concentration
OR	Objective response
DR	Duration of response
CEA	Carcinoembryonic antigen
AUC	Area under curve
QD	Once daily
HR	Homologous recombination
DSBs	DNA double-stranded breaks
PARP	Enzyme poly(ADP-ribose) polymerase
PEG-AuNPs	PEG-stabilized gold nanoparticles
TNBC	Triple-negative breast cancer
PI3K	Phosphoinositide 3-kinases
SAT1	Spermidine/spermine N1-acetyltransferase 1
SQ-COOH/PTM-B	Squalene-based nanoparticles of PTM
LDL	Low-density lipoproteins
LDLR	Low-density lipoproteins receptor
PLGA	Poly(lactide-co-glycolide)
HA	Hyaluronic acid
PECs	Polyelectrolyte complexes
PEG	Polyethylene glycol
MRP	Multidrug resistance-associated protein

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