Biomedicine & Pharmacotherapy 86 (2017) 221–231

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Review

Role of protease and protease inhibitors in cancer pathogenesis and treatment

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ARTICLE INFO

Article history:
Received 25 August 2016
Received in revised form 26 November 2016
Accepted 5 December 2016

Keywords:
Protease
Protease inhibitor
cancer

ABSTRACT

Cancer is the second cause of death in 2015, and it has been estimated to surpass heart diseases as the leading cause of death in the next few years. Several mechanisms are involved in cancer pathogenesis. Studies have indicated that proteases are also implicated in tumor growth and progression which is highly dependent on nutrient and oxygen supply. On the other hand, protease inhibitors could be considered as a potent strategy in cancer therapy. On the basis of the type of the key amino acid in the active site of the protease and the mechanism of peptide bond cleavage, proteases can be classified into six groups: cysteine, serine, threonine, glutamic acid, aspartate proteases, as well as matrix metalloproteases. In this review, we focus on the role of different types of proteases and protease inhibitors in cancer pathogenesis.

Contents

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http://dx.doi.org/10.1016/j.biopha.2016.12.021
0753-3322/

1. Proteases

One of the most important biological catalytic reactions is proteolysis and this is known as proteolytic activity, which has been attributed to a class of enzymes called proteases. Proteolysis is the hydrolysis of peptide bond by attacking the carbonyl group of the peptide. Proteases are of broad enzymes distribution. In human, there are about 990 known protease genes. In addition, about 1605 known protease inhibitor genes have been reported in human [1].

On the basis of the nature of the key amino acid in the active site of the protease and the mechanism of peptide bond cleavage, proteases can be classified into six groups: cysteine, serine, threonine, glutamic acid,aspartate proteases, as well as matrix metalloproteases [2–4].

The cleaving mechanism of a peptide bond with a protease usually occur in the presence of water molecule (in aspartate, metallo- and glutamic acid proteases) or a cysteine, serine, or threonine residue (typically a histidine residue activation) as the nucleophile in the active site [5] (Fig. 1).

The association between stromal and tumor cells modulates two protease systems that are involve in proteolysis outside the cell, these are the MMPs and urokinase plasminogen activator (uPA)/uPA receptor (uPAR)/plasminogen network. Stromal MMP-2 and uPA are synthesized as inactive precursors and then stimulated on the tumor cells surface (Fig. 1), thereby causing malignant cells to rupture the basement membranes. These enzymes also promotes blood vessels sprouting to feed the growing cancer. Antitumor therapies targeting these stromal contributions to metastasis, invasion, and angiogenesis, attack a genetically constant cell population, so they may not attack the resistance related to the use of traditional chemotherapeutic drugs.

2. The role of protease in cancer development

Proteases in normal cells are very essential in carrying out imperative biological processes, and can regulate a diversity of different cellular processes such as gene expression, differentiation, and cell death [6]. However, recent studies have indicated that

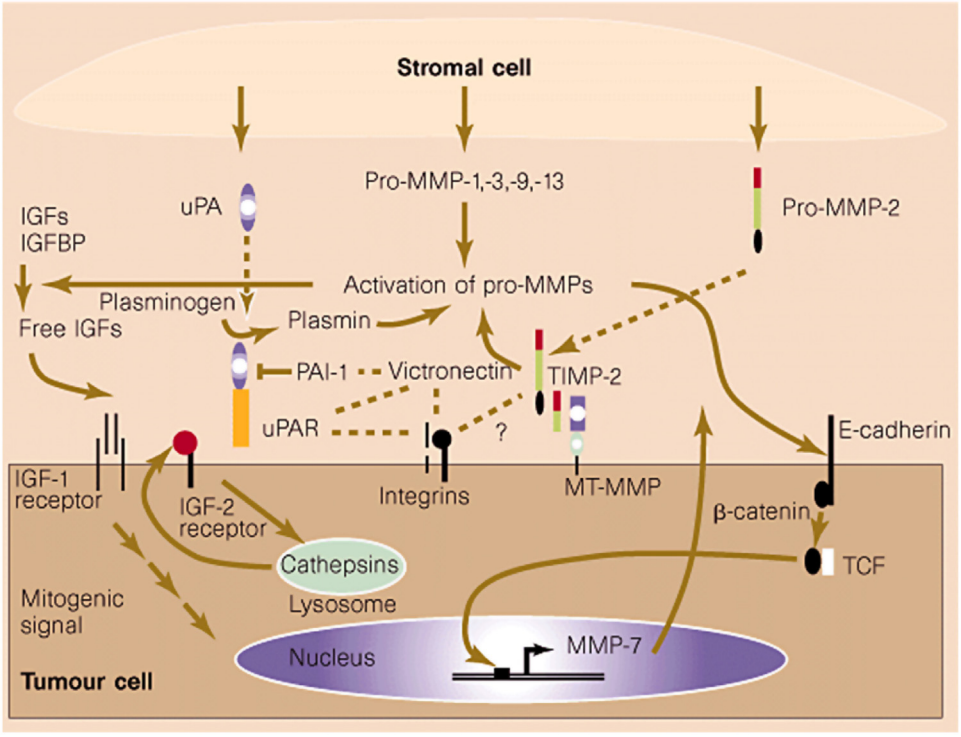

Fig. 1. General cascade of protease inhibitor mechanism of action on tumor cells.

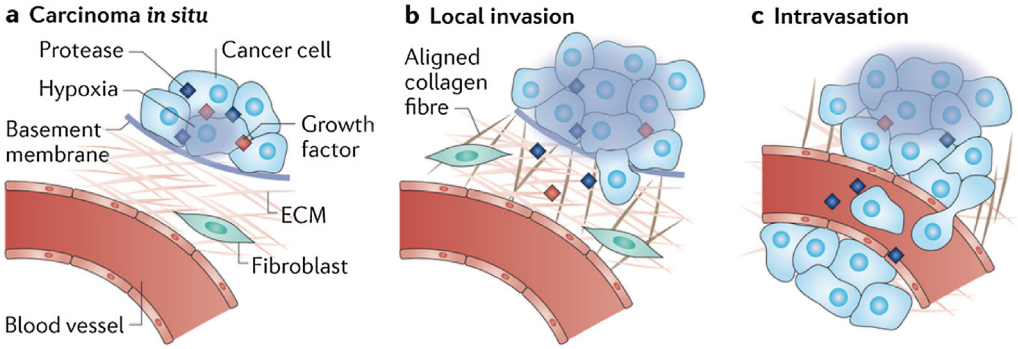

Fig. 2. Epithelial to mesenchymal transition in metastasis.

proteases are also implicated in tumor growth and progression, both at primary and metastatic sites [3].

It has been shown, that tumor cells stimulate the expression of proteolytic enzymes in non-neoplastic neighboring cells, hijacking their activity to favor tumor expansion [7,8].

Metastases and tumor progression are highly dependent on nutrient and oxygen supply, which are motivated by various proteases in the tumor and/or surrounding tissues and organs [3]. Several tumors have been indicated to have increased the levels of proteases at an early stage and these proteases are now indicated to be involved in many aspects of cancer, such as proliferation, immune responses, inflammatory cell recruitment, tumor invasion, angiogenesis, metastasis, apoptosis, epithelial to mesenchymal transition (EMT) as depicted in Fig. 2, the mobilization of normal cells from their tissue compartments to assist in metastasis (Fig. 3), as well as response to therapy such as chemoresistance to drugs [9,10]. In the subsequent section of this review, we have gathered evidence emphasizing on the role of each protease classes on different types of cancers. Table 1 shows different proteases and types of cancer cell where their function is elucidated.

2.1. Cysteine proteases

Cysteine proteases (calpains) are localized in the cytosol, or the lysosome (cathepsins B, L, H and S) and are secreted in some cell types under pathological conditions. This class of protease mediate general functions such as catabolism of intracellular protein and specialized functions such as selective activation of extracellular protein degradation, macrophage function, bone resorption or signaling molecules (e.g. interleukin, protein kinase C, encephalin) [8]. The family of cathepsin cysteine proteases can degrade both intracellular and extracellular matrix (ECM) proteins [11,12]. Cathepsins have been shown to function extracellularly as well as intracellularly. Cathepsins are predicted to be a potential targets for anti-cancer therapy [12,13], as the extracellular activity of cathepsins promotes cancer cells progression to nearby tissues, blood and lymph vessels and metastasize to outlying tissues [14,15]. Cathepsin B has been actually implicated in the remodeling and dissolution of basement membrane and connective tissue in the processes of tumor growth, invasion, and metastasis [16], which may results in podosome-mediated ECM degradation and invasion via secreted lysosomes [17]. Cathepsins are also considered as useful prognosis markers for recognizing patients who are suffering from colorectal cancer [18], breast cancer [19], pancreatic cancer [20] and tongue carcinoma [21]. Increased levels of other lysosomal proteases, such as cathepsins D, H, or L, have also been reported in several cancer types [22]. Cathepsin L2 (CTSL2) has been shown to be upregulated in a different malignancies like lung, breast, gastric, colon, head and neck carcinomas, gliomas, melanomas [23] as well as endometrial cancer [24].

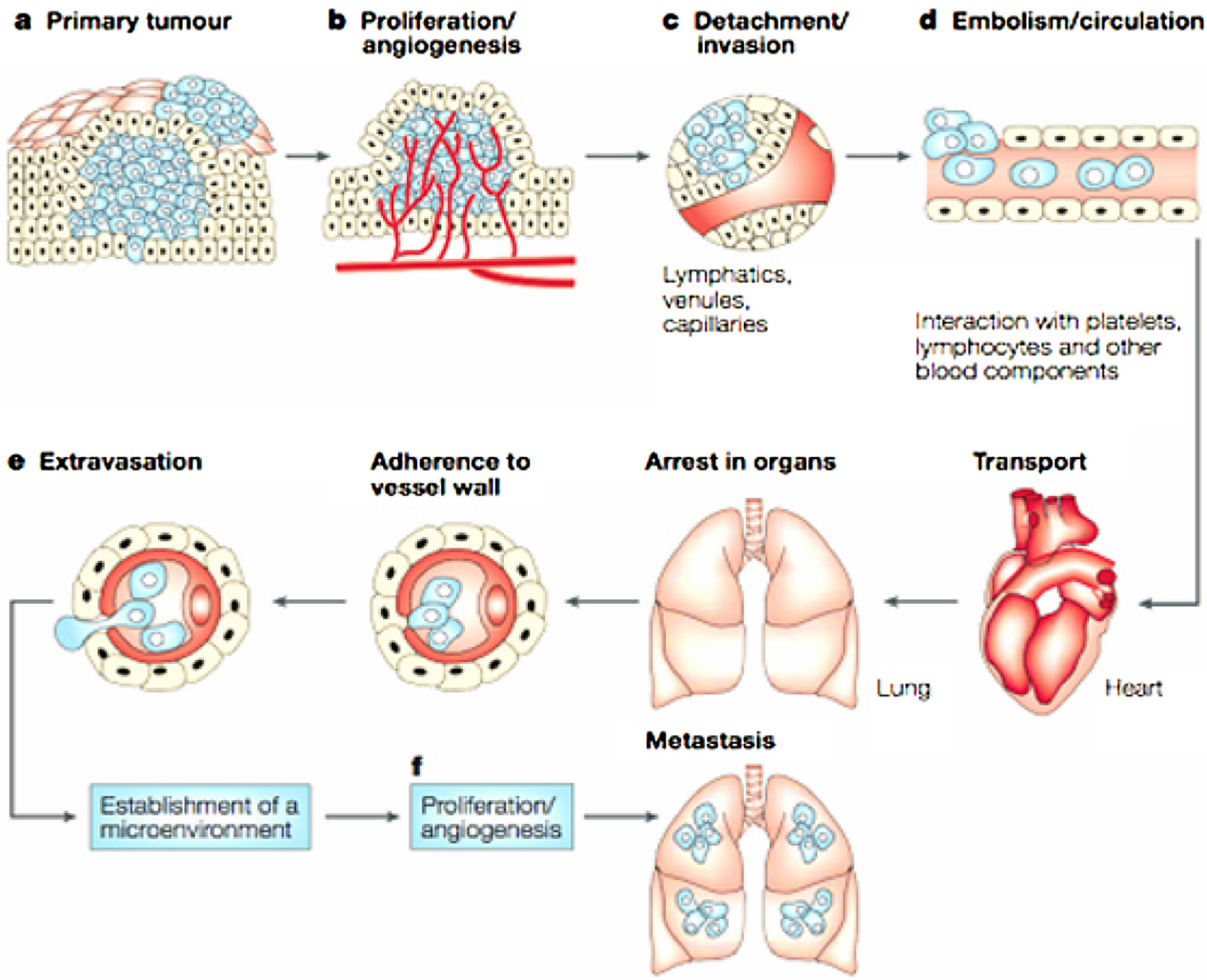

Fig. 3. Principal stages in the formation of metastasis.

Table 1
All classes of protease and types of cancer where they are localize.

Family	Protease	Location	Cancer	Ref.
Cysteine Cathepsins	General	Intracellular, lysosome	Most	[1,2]
	Cathepsin K	Extracellular, bone	Breast	[3]
	Cathepsin B	Extracellular and pericellular under pathological conditions	Breast, cervix, colon, colorectal, gastric, head and neck, liver, lung, melanoma, ovarian, pancreatic, prostate, thyroid	[4–6]
	Cathepsin L		Breast, colorectal	[7]
Aspartatic Cathepsins	Cathepsin E	Endosomal structures, ER, Golgi	Cervical, gastric, lung, pancreas adenocarcinomas	[8]
	Cathepsin D	Lysosome	Breast, colorectal, ovarian	[8]
Kallikreins (hK)	General	Intracellular, secreted	Most	[9,10]
	Hk1	Tissue	Lungs adenocarcinoma	[11]
	PSA (Hk3)		Prostate, ovarian	[2]
	Hk10		Colon, ovarian, pancreatic, head and neck	[12]
	Hk15		Ovarian, prostate	[13]
Serine Proteases	Upa, uPAR	Membrane, pericellular	Cervical, colorectal, gastric, prostate	[14]
Caspases		Intracellular		
MMPs	General	Extracellular	Most	[15]
	MMP-1, −8,−13		Breast	[15]
	MMP-2, −9		Breast, Colorectal, lung, malignant gliomas, ovarian	[16,17]
	MMP −14	Membrane	Breast	[18]
ADAM		Extracellular		

2.2. Serine proteases

Normal regulation of serine proteases activity is critical for physiological activities of the cell and tissue. But abnormal regulation of these proteases activity can lead to pathological conditions such as cancer [25]. Just about one third of all proteases can be classified as serine proteases, which are known for the existence of the nucleophilic Ser residue at the active site [26]. Serine proteases was initially well-known by the existence of the Asp-His-Ser “charge relay” system or “catalytic triad” [27]. The Asp-His-Ser triad can be establish in at least four diverse structural contexts; subtilisin, chymotrypsin, carboxypeptidase Y, and Clp protease (MEROPS) nomenclature [28]; signifying that this catalytic machinery have developed on at least four separate occasions [27].

There are another serine proteases with different catalytic triads and dyads, comprising Ser-His-Glu, Ser-Lys/His, His-Ser-His, and N-terminal Ser [27].

One of the well characterized serine proteases is trypsin. These proteases play a critical roles in a wide spectrum of pathological processes like, inflammation, atherosclerosis and cancer [25].

2.3. Aspartate proteases

Cathepsin-D (Cath-D) is an aspartic endo-protease that is universally found in lysosomes [29]. Cath-D had two main roles: precursor’s activation of biologically active proteins in pre-lysosomal compartments of specialized cells, and as a major protein-degrading enzyme in lysosomes and phagosomes [30]. The aspartic protease Cath-D is overexpressed and secreted at high levels by human epithelial breast cancer cells [31–34], thus, it is a marker of poor prognosis in breast cancer [35]. Cath-D motivates fibroblast outgrowth, cancer cell proliferation, angiogenesis and metastasis [36–38]. Cath-D, like other aspartate proteases such as renin, pepsinogen, chymosin, has a bilobed structure [39].

2.4. Threonine proteases

Threonine proteases or proteasomes partake in removing cellular proteins, which is tagged for degradation through a complex modification known as poly-ubiquitination; a procedure of adding a series of ubiquitin molecules to another protein, targeted for degradation [40]. The expression of the majority proteins is controlled by the proximal activity of the ubiquitin proteasome system through the activity of specific proteins and enzyme complexes, including the 76-amino-acid protein upstream ubiquitin, the E1, E2, and E3 ubiquitin ligase machinery, and deubiquitinating enzymes [41]. Aberrant proteasome-dependent proteolysis appears to be associated with the pathophysiology of some malignancies such as cancer [42].

2.5. Matrix metalloproteases

Matrix metalloproteases (MMPs) are class of nine or more highly homologous Zn^{2+} endopeptidase that together cleaves most of the constituents of the ECM. Increased level of MMPs expression has been reported in multiple tumor types [43,44]. MMPs are responsible for the remodeling and turnover of ECM proteins [7] and the increased expression associated with the pathophysiology of cancer

3. Role of protease inhibitors in cancer treatment

While protease are involve in pathogenesis of cancer, protease inhibitors have been noted for their role in cancer therapy. However, protease inhibitor therapy design is complicated since

Table 2
Selected cysteine cancer protease inhibitors.

Inhibitors	Cancer type	Description	Ref(s)
Stefin A	breast cancer	Inhibits distant metastasis	[59]
Stefin A	human prostate cancer	Ratio of cathepsin B to stefin A identifies heterogeneity within Gleason histologic scores	[60]
Stefin A	in laryngeal cancer	Expression and clinical significance of cathepsin B	[61]
Stefin A	breast cancer	Co-expression of cathepsin B and its inhibitor Stefin A in breast cancer metastasis to lung and bone	[62]
Stefin B	breast cancer model	Stefin B deficiency reduces tumor growth via sensitization of tumor cells to oxidative stress	[57]
Stefin B	Meningiomas	Lower protein and message levels	[63]
Stefin B	Melanoma	down-regulated gene	[64]
Stefin B	in human esophageal carcinoma	downregulated and this change was related to lymph node-metastasis	[65]
Cystatin C	glioma tumor	levels were decreased in high grade glioma tumor masses	[14]
Cathepsin-cystatin C	colorectal cancers	decrease in cathepsin-cystatin C complexes with an increase in stage of colorectal cancers	[40]
Cystatin F or leukocystatin or CMAP	certain lymphoid cell types	shows a rather restricted expression pattern, being confined to certain lymphoid cell types	[43]
Cystatin F or leukocystatin or CMAP	several murine cancer types	This gene encodes a glycosylated cysteine protease inhibitor with a putative role in immune regulation through inhibition of a unique target in the hematopoietic system	[44]
Cystatin F or leukocystatin or CMAP	the primary tumor	Little or no expression of cystatin F was found in the primary tumor	[45]
Cystatin E/M	breast cancers	expressed in normal tissues, but expression is lost in most late stage/metastatic breast cancers	[46]
Cystatin M	endothelial cells	increased cystatin M expression was shown to decrease tumor cell proliferation, invasion, and adhesion to endothelial cells	[47]
Cystatin D	human colon cancer cells	Tumor suppressor gene induced by vitamin D	[66]
Cystatin SN	colorectal cancer	Tumor marker	[67]

different types of cancers use different proteases at the fluctuating stages of cancer development and no single inhibitor can be used on all classes of proteases [45,46].

3.1. Cysteine cancer protease inhibitors

Cathepsins (lysosomal cysteine proteases) can be controlled by the endogenous cysteine protease inhibitor, known as cystatin, in normal tissues and cells. The cystatin superfamily are a group of reversible, tight-binding competitive inhibitors for cysteine peptidases such as cathepsins B, H, and L that inhibits the proteolytic activity of cysteine proteases [46].

It was proposed that cysteine cancer protease inhibitors plays an important role in cancer, which are related with alterations of the proteolytic system [47]. Recent studies have showed cystatins to block metastasis or invasion of diverse cancers in experimental systems [48].

The prime role of cystatins as protease inhibitors is to limit extra cysteine protease activity released from lysosomes or generated during inflammation. As cysteine proteases play important roles in tumor growth, development, and metastasis, the cystatins should mediate the control these processes.

There are 3 types of cystatin; type I and II cystatins being the most studied in cancer. Main species of type I cystatins are cystatins A and B, which are commonly referred to as Stefins A and B. Type 2 includes cystatins C, D, E/M, F, G (CRES), S, SN and SA [48,49].

Decreased level of Stefin A expression have been found in some epithelial-type cancers which correlates with lower patient survival [50]. Furthermore, evidence shows that Stefin A levels could be reduced at the protein and transcriptional levels during tumor progression [51]. Studies have also show a decreases in Stefins A and B in breast cancer cell lines in increasing invasiveness [52]. Immunostaining Stefin A existing in benign but not malignant meningiomas have been shown [53,54].

In glioblastoma, the invasive capability of tumors can be confirmed by cystatin markers [55]. Immunologic staining of Stefin A was also markedly decreased in pituitary adenomas while cathepsin levels were amplified [54]. Poor patient survival in head and neck cancer can be correlated with low Stefin A levels [56].

Altogether, lower expression of stefins A and B has been shown in aggressive tumor types and increase in primary tumor Stefin levels appear to correlate with a more favorable prognosis [48,49,57]. Patients with small cell lung cancer (SCLC) demonstrated a higher levels of Stefin A and B in tumor than in normal tissue [58]. Some of cysteine cancer protease inhibitors are summarized in Table 2. Fig. 4 depict the 3-D structure of several protease inhibitors discussed in Tables 2 and 3.

3.2. Serine cancer protease inhibitors

A type II transmembrane serine protease known as matriptase, is involved in the degradation of ECM, angiogenesis, and in the development of some epithelial cancers [68]. But in normal cells and tissues, it is repressed by hepatocyte growth factor activator inhibitor-1(HAI-1). For the period of the development of human prostate cancer (CaP), there is loss of HAI-1 and expression of matriptase. It has been proposed that the ratio of these two proteins may serve as a favorable biomarker for human prostate cancer progression and a probable marker for establishing the efficacy of chemopreventive and therapeutic interventions [69].

Serpins (serine protease inhibitors) are a group of proteins with similar structures that were first recognized as a set of proteins able to inhibit serine proteases [70,71]. Important serpins in cancer are summarized in Table 3.

3.3. Aspartate cancer protease inhibitors

There has been several studies in the improvement of aspartic proteinase inhibitors as therapeutic agents for the treatment of

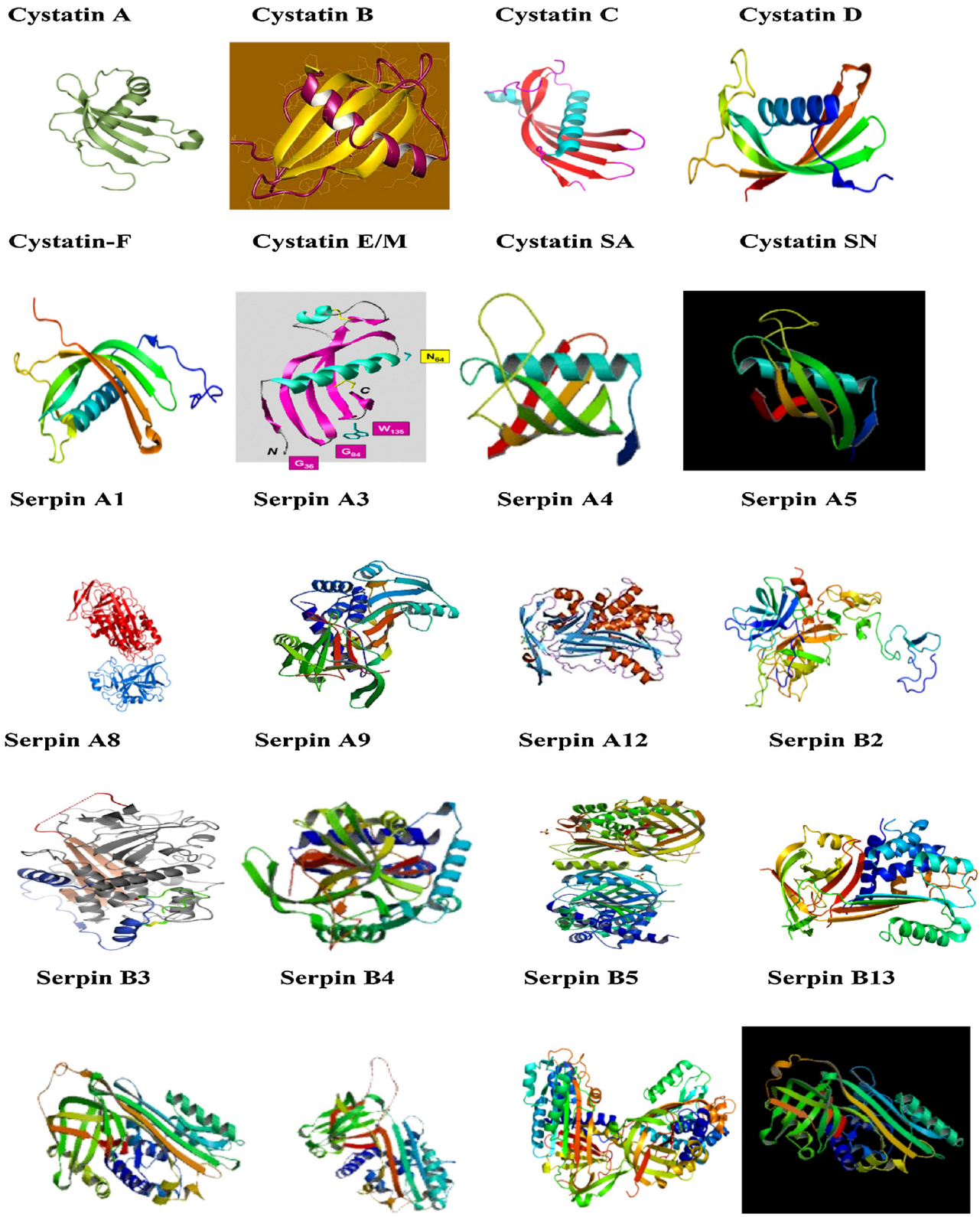

Fig. 4. Collections of several protease inhibitors, showing their 3-D structure (Adapted from PDB).

AIDS, hypertension, gastric diseases, parasitic infections, amyloid diseases, and cancer. The cathepsin D (an aspartic proteinase) is hyper secreted and overexpressed by cells of epithelial breast cancer [39]. Augmented levels of the aspartic proteinase cathepsin D were first reported in several human neoplastic, and cancer tissues in the mid-1980s [94]. These results generated focus on a promising role for cathepsin D in cancer and neoplastic processes. The findings showed a strong risk factor for measurement of

Table 3
Selected serine cancer protease inhibitors.

Serpin types	Commonly name	Cancer type	Description	Ref(s)
SERPINA1	Alpha1 antitrypsin	Lung cancer, colorectal cancer	Serum biomarkers for the diagnosis of lung cancer and colorectal cancer	[72,73]
SERPINA3	Alpha1 antichymotrypsin	prostate cancer	As an indicator of prostate cancer, predicting bone metastases of prostate cancer,	[74,75]
SERPINA4	Kallistatin	Lung cancer	Inhibits angiogenesis and tumor growth,	[76]
SERPINA5	Protein C inhibitor	prostate cancer	As a biomarker for prostate cancer	[77]
SERPINA8	Angiotensinogen	Lung Cancer	Polymorphisms in Lung Cancer	[78]
SERPINA9	Centerin (Gcet1)	B-cell lymphomas	Extracellular; inhibitory, maintenance of naive B cells	[79,80]
SERPINA12	Vaspin	endometrial cancer	Low vaspin are associated with an increased risk of developing endometrial cancer	[81,82]
SERPINB2	Plasminogen activator inhibitor-2	head and neck cancer	Down-regulation contributes to chemoresistance in head and neck cancer	[83]
SERPINB3	Squamous cell carcinoma antigen-1 (SCCA-1)	breast cancer, cervical cancer treatment, tongue cancer,	Prognostic tool	[84–87]
SERPINB4	Squamous cell carcinoma antigen-2 (SCCA-2)	cervical cancer	Prognostic tool	[87]
SERPINB5	Maspin	Human mammary epithelial cells, lung cancer	Proposed to function as a tumor suppressor, Biomarker in lung cancer	[88,89]
SERPINI2	Pancpin	pancreatic cancer	inhibition of pancreatic cancer metastasis has been suggested	[93]
SERPINB13	Hurpin/Headpin	Brain and ovarian cancer	Single nucleotide polymorphisms and treatment of cancer	[90–92]

cathepsin D concentrations in breast cancer as well as many other tumor types. Level of cathepsin D is correlated with the frequency of clinical metastasis and is therefore a promising marker of poor prognosis in breast cancer and other cancers. Cathepsin D is a functional factor in a varieties of tissues during their regression or remodeling, and overexpression of the enzyme promotes metastasis and tumorigenesis. The enzyme has also been implicated in apoptosis and aids tumor angiogenesis [39]. In the mammary gland, cathepsin D function appears to be related to the processing of the prolactin (a peptide hormone). The cathepsin D synthesis is regulated by steroid hormones. As an example, in breast cancer cell lines, expression of cathepsin D is controlled by estrogen hormones. Studies has shown the amplified levels of cathepsin D (both at the mRNA and protein levels) in several human neoplastic tissues [95]. The design of antagonists to block the interaction of the protein with its receptor may be a promising tool in the treatment of breast cancer and other neoplastic diseases [39].

In contrast to other tissue proteases (e.g. metalloproteinases and serine proteases), no endogeneous cath-D tissue proteases inhibitor is known in mammals. Pepstatin, a natural aspartic proteases inhibitor has been isolated from various species of actinomycetes [96], and often used not only to study its function in some in vitro systems, but also in affinity chromatography purification of cath-D.

3.4. Glutamic acid cancer protease inhibitors

This is a new class of proteases derived from pepstatin-insensitive carboxyl protease. Glutamic acid proteases have been isolated from *Aspergillus niger var. macrosporus and Stylidium lignicola*. The mechanism of action is based on the two enzymes from the scytalidoglutamic and aspergilloglutamic proteases. The active site of diad glutamic acid and glutamine is implicated in catalysis and substrate binding. These amino acids association with water molecules behaves as a nucleophiles to possess an acid-base mechanism different from that of the aspartic proteases. The glutamic acid behaves as a general acid in the catalysis first phase, donating a proton to the carbonyl oxygen of the scissile peptide bond of the substrate. Concurrently, an OH— is donated by water in the active site of the enzyme to the carbonyl oxygen of the peptide bond substrate. In some cases, two water molecules plays a role in the reaction. The substrate transition state is thought to be maintained by hydrogen bonding with the two catalytic residues. Thereafter, glutamic acid donates a proton to the amide nitrogen atom of the scissile peptide bond stimulating the breakdown of the tetrahedral intermediate, and thus leading to peptide bond cleavage. The glutamine residue is then charged for reversing the original state of the glutamic acid residue [19].

3.5. Threonine cancer protease inhibitors

Proteasome, a threonine protease unlike other proteases contains an active site N-terminal threonine residue, residing in the MB1 subunits of its b-rings that can be aimed by pharmacophores linked to short peptides [97,98]. Proteasome Inhibitor (PSI) is an inhibitor of chymotrypsin-like activity of 20S proteasome. Recently, PSI has been demonstrated by Oyajobi and Mundy [99], to be capable of reducing the tumor accumulation in mice injected with murine 5TGM1 plasmacytoma cells and inhibits the growth of osteolytic lesions. However, the good pharmacodynamics of PSI are outweighed by its poor pharmaceutical properties.

Lactacystin being the first of such specific PSI is a Streptomyces metabolite that inhibit the 20S subunit, and its peptidase enzyme activities include trypsin, chymotrypsin and peptidylglutamyl-peptide-like hydrolyzing activities, the first two bind irreversibly, but they all bind at different rates to lactacystin [98]. In spite of the selective effects of lactacystin in tumor cells, it stands as a tool for proteasomal process studies only in *in vitro* systems, due to its poor metabolic stability and binding irreversibly to proteasomal subunit [100].

Several pharmaceutical inhibitors have been developed to this end; Epoxomicin is an actinomycetes-derived PSI which binds irreversibly and possesses *in vivo* and *in vitro* activities [101]. Epoxomicin like lactacystin binds solely to the proteasome 20S catalytic subunits leading to the inhibition of the chymotrypsin-like function. It acts more faster with greater specificity when compared to lactacystin, unlike other reversible inhibitors [101]. Nevertheless, it use in human is restricted for similar reason raised by lactacystin. Thus, PSI which binds irreversibly doesn't have

desirable pharmacological characteristics and are therefore presently no longer investigated for cancer drug development.

Dipeptide boronic acid (BA) analogues, compared to peptide aldehyde agents, are selective, potent and are PSI which bind reversibly [101–103]. BA block the activity of threonine protease by inhibiting chymotryptic activity of proteasome leading to the weakening of the degradation of cell cycle regulatory proteins, e.g., p53 or p27, I-jB and cyclin E [102,104,105]. While affecting multiple proteins, BA treatment leads to the inhibition of cancer cell growth and initiation of apoptosis[102]. Bortezomib (Velcadee) being the first in the BA group is generally active in tumor cells, possessing little toxic effects on normal cells and demonstrated as a single active agent in preclinical human cancer xenografts models and also in primary cultures of hematological and solid tumor types like prostate, neck and head cancers, multiple myeloma and lymphoma [40,104].

Ritonavir; another PSI has been shown to reversibly interact with the active site of the subunit responsible for chymotrypsin-like activity in isolated 20S proteasomes [106]. Gaedicke et al. [107] showed that ritonavir, like other PSIs' has antitumor activities, it strongly reduce the degree of proliferation of various tumor cell lines and induced their apoptosis *in vitro*. Chymotrypsin-like function of isolated 26S proteasomes is activated by ritonavir compared to its action on isolated 20S proteasomes. The final outcome of low micromolar concentrations of ritonavir on the chymotrypsin-like activity in cells and cell lysates was a weak inhibition, acid-soluble proteolytic peptide levels marginal alterations, consistent marginal alterations of polyubiquitinated proteins and a little accumulation of cancer suppressor protein p53 in cells that are treated with ritonavir. Accumulation of p21 in the presence of this PSI is associated with the hindrance of proteolytic degradation and this shows selective proteasomes inhibition in line with abnormal degradation of p21, which doesn't need to be ubiquitinated. This discovery proposed that the selective perturbation of proteasomal protein degradation can partake in ritonavir antitumor activity.

3.6. Matrix cancer protease inhibitors

Zinc dependent MMPs are the most abundant group of non-serine proteases existing in invasive and metastatic cancer, it breakdown structural proteins comprising the ECM, like elastin, collagen, fibronectin, laminin, fibrinogen and vitronectin as an important step in the complicated process of hematogenous metastasis. MMPs have long been related with tumor-cell metastasis and invasion, and this contributed to the rationale of MMPs inhibitors for clinical trials [6,108,109]. The center of attraction of cancer research is mutations in cancer cells, and this result in either loss-of-function in tumor-suppressor genes or gain-of-function in oncogenes [110].

Tissue inhibitors of metalloproteases (TIMPs) are groups of small extracellular proteins inhibiting MMPs. Direct proof for the role of MMPs in tumor progression comes from xenograft experiments using cancer cells with decreased and increased expression levels of TIMPs or MMPs, so also as carcinogenesis experiments using mice lacking a specific TIMP-1 or MMP or those having organ-specific TIMP-1 or MMP overexpression. There are four members of the TIMP family, TIMP-1, −2, −3 and −4, each inhibiting the activities of different MMPs with varying efficiency [6,111]. TIMPs are crucial in determining the influence of the ECM of cell adhesion molecules, chemokines, several cytokines and growth factors on the phenotype of cells in various pathological and physiological conditions [112].

MMPs pharmaceutical inhibitors such as batimastat and its chemical analog, marimastat, have been synthesized and administered to decrease the cancer metastasis in patient, mimicking the cleavage sites of MMP substrates, but, the clinical trials outcome with these drugs failed [113]. Krüger et al. [108] mentioned that batimastat treatment have been said to give rise to liver metastasis of human breast carcinoma cells in nude mice and also a rise in liver metastasis of murine T-cell lymphoma in syngeneic mice, overexpression of liver specific MMP2, MMP9, mRNA angiogenesis up-regulation and caspase1 even in cancer free animals have been observed. They mentioned that in improving this drug further, induction of organ-specific side effect should be controlled. Batimastat can't be administered orally and it's no more tested for tumor treatment in human [113].

Marimastat have gone through various Phase III clinical trials [113], advanced pancreatic cancer trial was aimed at detecting differences in survival comparing patients treated with several doses of this drug and conventional chemotherapy. Although it was unsuccessful to identify increased survival for groups treated with marimastat, however, the highest dose of marimastat was as effective as the conventional therapy [114]. Bramhall et al. [98] administered marimastat to some patient in other to prolong survival in patients suffering from gastro-oesophageal adenocarcinoma, this experiment is one of the first to show the therapeutic use of MMP inhibitor in tumor patients, it was observed that patient who have previously received a chemotherapy treatment before this drug has a longer span than those who receive the placebo after chemotherapy. Certainly, an achievement in both academia and pharmaceutical industry previously aimed at the inhibition of main class of proteases in tumor. The failure of MMP inhibitors (MMPIs) during its clinical trial in the late 1990s led to the abortion of several drug programs and miserably had a negative effect on other protease inhibitor class development. In analyzing why MMPIs reached phase III clinical trials only to be unsuccessful, various points that might be important for consideration of targeting other classes of proteases have been raised [111,115,116].

4. Conclusion

In this review, we provided an insight into the role of the protease and protease inhibitors in cancer. Proteases, are functionally involved in many processes of cancer progression, from benign to malignancy [4]. In cancer conditions, proteases were initially considered to stimulate cancer cell escape through tissue barriers. However, process of proteolysis is complicated in many aspects of cancer involving inflammatory cell recruitment, immune responses, proliferation, and apoptosis [10]. Activity of proteases is regulated by interactions with endogenous inhibitors of the protease (TIMPs) targeting metalloproteases, serpins being effective against serine proteases, and cystatins predominantly inhibiting cysteine proteases. The studies of the role of protease and protease inhibitors in cancer progression extend the therapeutic window for cancer treatment. Collectively, further perceptive of the roles of proteases in tumor progression, metastasis and cancer development will guide the development of novel therapeutic strategies against cancer.

Conflict of interest

The authors declare no conflict of interest.

Acknowledgement

The authors thank Department of Medical Biotechnology, School of Advanced Technologies in Medicine, Tehran University of Medical Sciences, Tehran, Iran.

References

[1] A.J. Rawlings, Using the MEROPS database for proteolytic enzymes and their inhibitors and substrates, Curr. Protoc. Bioinf. (2014) 1.25.1–1.25.33.
[2] B. Turk, D. Turk, V. Turk, Protease signalling: the cutting edge, EMBO J. 31 (7) (2012) 1630–1643.
[3] Y. Yang, H. Hong, Y. Zhang, W. Cai, Molecular imaging of proteases in cancer, Cancer Growth and Metastasis 2 (2009) 13.
[4] U. Verbovšek, C.J. Van Noorden, T.T. Lah, Complexity of cancer protease biology: cathepsin K expression and function in cancer progression, Semin. Cancer Biol. (2015) Elsevier.
[5] R.G. Boy, M. Knapp, U. Eisenhut, W. Mier, Enzymes/transporters, Molecular Imaging II, Springer, 2008, pp. 131–143.
[6] M. Egeblad, Z. Werb, New functions for the matrix metalloproteinases in cancer progression, Nat. Rev. Cancer 2 (3) (2002) 161–174.
[7] S. Rakashanda, F. Rana, S. Rafiq, A. Masood, S. Amin, Role of proteases in cancer: a review, Biotechnol. Mol. Biol. Rev. 7 (4) (2012) 90–101.
[8] S. Zucker, J. Cao, W.-T. Chen, Critical appraisal of the use of matrix metalloproteinase inhibitors in cancer treatment, Oncogene 19 (56) (2000) 6642–6650.
[9] J. Kim, W. Yu, K. Kovalski, L. Ossowski, Requirement for specific proteases in cancer cell intravasation as revealed by a novel semiquantitative PCR-based assay, Cell 94 (3) (1998) 353–362.
[10] S.D. Mason, J.A. Joyce, Proteolytic networks in cancer, Trends Cell Biol. 21 (4) (2011) 228–237.
[11] M.M. Mohamed, B.F. Sloane, Cysteine cathepsins: multifunctional enzymes in cancer, Nat. Rev. Cancer 6 (10) (2006) 764–775.
[12] V. Gocheva, J.A. Joyce, Cysteine cathepsins and the cutting edge of cancer invasion, ABBV Cell Cycle 6 (1) (2007) 60–64.
[13] C. Jedeszko, B.F. Sloane, Cysteine cathepsins in human cancer, Biol. Chem. 385 (11) (2004) 1017–1027.
[14] O. Vasiljeva, B. Turk, Dual contrasting roles of cysteine cathepsins in cancer progression: apoptosis versus tumor invasion, Biochimie 90 (2) (2008) 380–386.
[15] P. Matarrese, B. Ascione, L. Ciarlo, R. Vona, C. Leonetti, M. Scarsella, A.M. Mileo, C. Catricalà, M.G. Paggi, W. Malorni, Cathepsin B inhibition interferes with metastatic potential of human melanoma: an in vitro and in vivo study, Mol. Cancer 9 (2) (2010) 207.
[16] J.A. Joyce, D. Hanahan, Multiple roles for cysteine cathepsins in cancer, ABBV Cell Cycle 3 (12) (2004) 1516–1519.
[17] C. Tu, C.F. Ortega-Cava, G. Chen, N.D. Fernandes, D. Cavallo-Medved, B.F. Sloane, V. Band, H. Band, Lysosomal cathepsin B participates in the podosome-mediated extracellular matrix degradation and invasion via secreted lysosomes in v-Src fibroblasts, Cancer Res. 68 (22) (2008) 9147–9156.
[18] K. Hirai, M. Yokoyama, G. Asano, S. Tanaka, Expression of cathepsin B and cystatin C in human colorectal cancer, Hum. Pathol. 30 (6) (1999) 680–686.
[19] P.L. Kandalaft, K.L. Chang, C.W. Ahn, S.T. Traweek, P. Mehta, H. Battifora, Prognostic significance of immunohistochemical analysis of cathepsin D in low-stage breast cancer, Cancer 71 (9) (1993) 2756–2763.
[20] P. Michl, Targeting cathepsins: a new glimmer of hope for pancreatic cancer therapy? Gut 61 (6) (2012) 790–791.
[21] Y. Saleh, J. Wnukiewicz, R. Andrzejak, T. Trziszka, M. Siewinski, P. Ziolkowski, W. Kopec, Cathepsin B and cysteine protease inhibitors in human tongue cancer: correlation with tumor staging and in vitro inhibition of cathepsin B by chicken cystatin, J. Cancer Mol. 2 (2006) 67–72.
[22] N. Fujise, A. Nanashima, Y. Taniguchi, S. Matsuo, K. Hatano, Y. Matsumoto, Y. Tagawa, H. Ayabe, Prognostic impact of cathepsin B and matrix metalloproteinase-9 in pulmonary adenocarcinomas by immunohistochemical study, Lung Cancer 27 (1) (2000) 19–26.
[23] J.M. Lankelma, D.M. Voorend, T. Barwari, J. Koetsveld, A.H. Van der Spek, A.P. De Porto, G. Van Rooijen, C.J. Van Noorden, Cathepsin L, target in cancer treatment? Life Sci. 86 (7) (2010) 225–233.
[24] M. Skrzypczak, A. Springwald, C. Lattrich, J. Häring, S. Schüler, O. Ortmann, O. Treeck, Expression of cysteine protease cathepsin L is increased in endometrial cancer and correlates with expression of growth regulatory genes, Cancer Invest. 30 (5) (2012) 398–403.
[25] Y.A. DeClerck, A.M. Mercurio, M.S. Stack, H.A. Chapman, M.M. Zutter, R.J. Muschel, A. Raz, L.M. Matrisian, B.F. Sloane, A. Noel, Proteases, extracellular matrix, and cancer: a workshop of the path B study section, Am. J. Pathol. 164 (4) (2004) 1131–1139.
[26] L. Hedstrom, Serine protease mechanism and specificity, Chem. Rev. 102 (12) (2002) 4501–4524.
[27] G. Dodson, A. Wlodawer, Catalytic triads and their relatives, Trends Biochem. Sci. 23 (9) (1998) 347–352.
[28] N.D. Rawlings, A.J. Barrett, MEROPS: the peptidase database, Nucleic Acids Res. 28 (1) (2000) 323–325.
[29] A. Barrett, D. Cathepsin, Purification of isoenzymes from human and chicken liver, Biochem. J. 117 (1970) 601–607.
[30] S. Diment, K. Martin, P. Stahl, Cleavage of parathyroid hormone in macrophage endosomes illustrates a novel pathway for intracellular processing of proteins, J. Biol. Chem. 264 (23) (1989) 13403–13406.
[31] A. Vashishta, S.S. Ohri, V. Vetvicka, Pleiotropic effects of cathepsin D, Endocrine, Endocr. Metab. Immune Disord. Drug Targets 9 (4) (2009) 385–391 (Formerly Current Drug Targets-Immune, Endocrine & Metabolic Disorders).
[32] G. Nicotra, R. Castino, C. Follo, C. Peracchio, G. Valente, C. Isidoro, The dilemma: does tissue expression of cathepsin D reflect tumor malignancy? The question: does the assay truly mirror cathepsin D mis-function in the tumor?, Cancer Biomark. Sect. A Dis. Mark. 7 (1) (2009) 47–64.
[33] E.S. Radisky, D.C. Radisky, Matrix metalloproteinase-induced epithelial-mesenchymal transition in breast cancer, J. Mammary Gland Biol. Neoplasia 15 (2) (2010) 201–212.
[34] O. Masson, C. Prébois, D. Derocq, A. Meulle, C. Dray, D. Daviaud, D. Quilliot, P. Valet, C. Muller, E. Liaudet-Coopman, Cathepsin-D, key protease in breast cancer, is up-regulated in obese mouse and human adipose tissue, and controls adipogenesis, PLoS One 6 (2) (2011) e16452.
[35] J. Rodriguez, J. Vazquez, M. Corte, M. Lamelas, M. Bongera, M. Corte, A. Alvarez, M. Allende, L. Gonzalez, M. Sanchez, Clinical significance of cathepsin D concentration in tumor cytosol of primary breast cancer, Int. J. Biol. Mark. 20 (2) (2004) 103–111.
[36] A. Vashishta, S.S. Ohri, M. Proctor, M. Fusek, V. Vetvicka, Ribozyme-targeting procathepsin D and its effect on invasion and growth of breast cancer cells: an implication in breast cancer therapy, Int. J. Oncol. 30 (5) (2007) 1223–1230.
[37] H. Huang, E. Pierstorff, E. Osawa, D. Ho, Protein-mediated assembly of nanodiamond hydrogels into a biocompatible and biofunctional multilayer nanofilm, ACS Nano 2 (2) (2008) 203–212.
[38] S.S. Ohri, A. Vashishta, M. Proctor, M. Fusek, V. Vetvicka, The propeptide of cathepsin D increases proliferation, invasion and metastasis of breast cancer cells, Int. J. Oncol. 32 (2) (2008) 491–498.
[39] E. Liaudet-Coopman, M. Beaujouin, D. Derocq, M. Garcia, M. Glondu-Lassis, V. Laurent-Matha, C. Prébois, H. Rochefort, F. Vignon, D. Cathepsin, newly discovered functions of a long-standing aspartic protease in cancer and apoptosis, Cancer Lett. 237 (2) (2006) 167–179.
[40] B.S. Mitchell, The proteasome-an emerging therapeutic target in cancer, New Engl. J. Med. 348 (26) (2003) 2597–2598.
[41] N.M. Weathington, R.K. Mallampalli, Emerging therapies targeting the ubiquitin proteasome system in cancer, J. Clin. Invest. 124 (1) (2014) 6–12.
[42] D. Chen, M. Frezza, S. Schmitt, J. Kanwar, Q. Dou, Bortezomib as the first proteasome inhibitor anticancer drug: current status and future perspectives, Curr. Cancer Drug Targets 11 (3) (2011) 239.
[43] A. Anastasov, P. Vihinen, J. Nikkola, S. Pyrhonen, T. Vlaykova, Matrix metalloproteinses in development and progression of skin malignant melanoma, Med. (Baltimore) 1 (1) (2011).
[44] B. Evrosimovska, B. Velickovski, C. Dimova, D. Veleska-Stefkovska, Matrix metalloproteinases (with accent to collagenases), J. Cell Anim. Biol. 5 (7) (2011) 113–120.
[45] G. Fear, S. Komarnytsky, I. Raskin, Protease inhibitors and their peptidomimetic derivatives as potential drugs, Pharmacol. Ther. 113 (2) (2007) 354–368.
[46] A.R. Folgueras, A.M. Pendás, L.M. Sánchez, C. López-Otín, Matrix metalloproteinases in cancer: from new functions to improved inhibition strategies, Int. J. Dev. Biol. 48 (2004) 411–424.
[47] B. Turk, V. Turk, D. Turk, Structural and functional aspects of papain-like cysteine proteinases and their protein inhibitors, Biol. Chem. 378 (3–4) (1996) 141–150.
[48] J.L. Cox, Cystatins and cancer, Front. Biosci. 14 (2009) 463–474.
[49] G. Dubin, Proteinaceous cysteine protease inhibitors, Cell. Mol. Life Sci. 62 (6) (2005) 653–669.
[50] T.T. Lah, M. Čerček, A. Blejec, J. Kos, E. Gorodetsky, R. Somers, I. Daskal, B. Cathepsin, a prognostic indicator in lymph node-negative breast carcinoma patients: comparison with cathepsin D, cathepsin L, and other clinical indicators, Clin. Cancer Res. 6 (2) (2000) 578–584.
[51] P. Hawley-Nelson, D.R. Roop, C.K. Cheng, T.M. Krieg, S.H. Yuspa, Molecular cloning of mouse epidermal cystatin A and detection of regulated expression in differentiation and tumorigenesis, Mol. Carcinog. 1 (3) (1988) 202–211.
[52] I. Zajc, N. Sever, A. Bervar, T.T. Lah, Expression of cysteine peptidase cathepsin L and its inhibitors stefins A and B in relation to tumorigenicity of breast cancer cell lines, Cancer Lett. 187 (1) (2002) 185–190.
[53] T. Strojnik, J. Kos, T.T. Lah, Cathepsins B and L are markers for clinically invasive types of meningiomas, Neurosurgery 48 (3) (2001) 598–605.
[54] T. Strojnik, T.T. LAH, B. Židanik, Immunohistochemical staining of cathepsins B, L and stefin A in human hypophysis and pituitary adenomas, Anticancer Res. 25 (1B) (2005) 587–594.
[55] N. Levičar, T. Strojnik, J. Kos, R.A. Dewey, G.J. Pilkington, T.T. Lah, Lysosomal enzymes, cathepsins in brain tumor invasion, J. Neurooncol. 58 (1) (2002) 21–32.
[56] P. Strojan, B. Svetic, L. Šmid, J. Kos, Serum cystatin C in patients with head and neck carcinoma, Clin. Chim. Acta 344 (1) (2004) 155–161.
[57] M. Butinar, M.T. Prebanda, J. Rajković, B. Jerič, V. Stoka, C. Peters, T. Reinheckel, A. Krüger, V. Turk, B. Turk, Stefin B deficiency reduces tumor growth via sensitization of tumor cells to oxidative stress in a breast cancer model, Oncogene 33 (26) (2014) 3392–3400.
[58] H.-H. Heidtmann, U. Salge, M. Abrahamson, M. Bencina, L. Kastelic, N. Kopitar-Jerala, V. Turk, T.T. Lah, Cathepsin B and cysteine proteinase inhibitors in human lung cancer cell lines, Clin. Exp. Metastasis 15 (4) (1997) 368–381.
[59] B. Parker, D. Ciocca, B. Bidwell, F. Gago, M. Fanelli, J. George, J. Slavin, A. Möller, R. Steel, N. Pouliot, Primary tumor expression of the cysteine cathepsin

inhibitor Stefin A inhibits distant metastasis in breast cancer, J. Pathol. 214 (3) (2008) 337–346.
[60] A.A. Sinha, B.J. Quast, M.J. Wilson, E.T. Fernandes, P.K. Reddy, S.L. Ewing, B.F. Sloane, D.F. Gleason, Ratio of cathepsin B to stefin A identifies heterogeneity within Gleason histologic scores for human prostate cancer, The Prostate 48 (4) (2001) 274–284.
[61] M.S. Khan, H. Devaraj, N. Devaraj, Chrysin abrogates early hepatocarcinogenesis and induces apoptosis in N-nitrosodiethylamine-induced preneoplastic nodules in rats, Toxicol. Appl. Pharmacol. 251 (1) (2011) 85–94.
[62] B. Parker, B. Bidwell, J. Slavin, R. Steel, B. Eckhardt, M. Henderson, R. Anderson, Co-expression of cathepsin B and its inhibitor Stefin A in breast cancer metastasis to lung and bone, Cancer Res. 67 (Suppl. 9) (2007) 3095.
[63] M. Trinkaus, A. Vranic, V.V. Dolenc, T. Lah, Cathepsins B and L and their inhibitors stefin B and cystatin C as markers for malignant progression of benign meningiomas, Int. J. Biol. Mark. 20 (1) (2004) 50–59.
[64] S.L. McDonald, H.D. Edington, J.M. Kirkwood, D. Becker, Expression analysis of genes identified by molecular profiling of VGP melanomas and MGP melanoma-positive lymph nodes, Cancer. Biol. Ther. 3 (1) (2004) 110–120.
[65] T. Shiraishi, M. Mori, S. Tanaka, K. Sugimachi, T. Akiyoshi, Identification of cystatin B in human esophageal carcinoma, using differential displays in which the gene expression is related to lymph-node metastasis, Int. J. Cancer 79 (2) (1998) 175–178.
[66] N. Valle, J.M. García, C. Peña, J.M. Freije, V. Quesada, A. Astudillo, F. Bonilla, C. López-Otín, A. Muñoz, Cystatin D is a candidate tumor suppressor gene induced by vitamin D in human colon cancer cells, J. Clin. Invest. 119 (8) (2009) 2343.
[67] K. Yoneda, H. Iida, H. Endo, K. Hosono, T. Akiyama, H. Takahashi, M. Inamori, Y. Abe, M. Yoneda, K. Fujita, Identification of Cystatin SN as a novel tumor marker for colorectal cancer, Int. J. Oncol. 35 (1) (2009) 33–40.
[68] K. Nakamura, A. Hongo, J. Kodama, F. Abarzua, Y. Nasu, H. Kumon, Y. Hiramatsu, Expression of matriptase and clinical outcome of human endometrial cancer, Anticancer Res. 29 (5) (2009) 1685–1690.
[69] M. Saleem, V.M. Adhami, W. Zhong, B.J. Longley, C.-Y. Lin, R.B. Dickson, S. Reagan-Shaw, D.F. Jarrard, H. Mukhtar, A novel biomarker for staging human prostate adenocarcinoma: overexpression of matriptase with concomitant loss of its inhibitor, hepatocyte growth factor activator inhibitor-1, Cancer Epidemiol. Biomark. Prev. 15 (2) (2006) 217–227.
[70] J.C. Whisstock, G.A. Silverman, P.I. Bird, S.P. Bottomley, D. Kaiserman, C.J. Luke, S.C. Pak, J.-M. Reichhart, J.A. Huntington, Serpins flex their muscle II. Structural insights into target peptidase recognition, polymerization, and transport functions, J. Biol. Chem. 285 (32) (2010) 24307–24312.
[71] G.A. Silverman, J.C. Whisstock, S.P. Bottomley, J.A. Huntington, D. Kaiserman, C.J. Luke, S.C. Pak, J.-M. Reichhart, P.I. Bird, Serpins flex their muscle I. Putting the clamps on proteolysis in diverse biological systems, J. Biol. Chem. 285 (32) (2010) 24299–24305.
[72] Y. Liang, T. Ma, A. Thakur, H. Yu, L. Gao, P. Shi, X. Li, H. Ren, L. Jia, S. Zhang, Differentially expressed glycosylated patterns of alpha-1-antitrypsin as serum biomarkers for the diagnosis of lung cancer, Glycobiology (2014) cwu115.
[73] S. Pérez-Holanda, I. Blanco, M. Menéndez, L. Rodrigo, Serum concentration of alpha-1 antitrypsin is significantly higher in colorectal cancer patients than in healthy controls, BMC Cancer 14 (1) (2014) 355.
[74] A. Christensson, T. Björk, O. Nilsson, U. Dahlén, M. Matikainen, A. Cockett, P. Abrahamsson, H. Lilja, Serum prostate specific antigen complexed to alpha 1-antichymotrypsin as an indicator of prostate cancer, J. Urol. 150 (1) (1993) 100–105.
[75] E. Kikuchi, J. Nakashima, M. Ishibashi, T. Ohigashi, M. Oya, K. Nakagawa, A. Miyajima, M. Murai, Usefulness of alpha 1-antichymotrypsin-PSA complex for predicting bone metastases of prostate cancer, Urology 68 (2) (2006) 371–375.
[76] N. Wang, J. Zou, Y. Diao, [Plasmid-mediated expression of kallistatin and its biological activity in lung cancer related cells], Yao xue bao = Acta Pharmaceutica Sinica 48 (3) (2013) 359–365.
[77] Z. Zhang D.W. Chan C.N. Rosenzwelg Protein C inhibitor as a biomarker for prostate cancer Google Patents 2013.
[78] H. Wang, K. Zhang, H. Qin, L. Yang, L. Zhang, Y. Cao, Genetic association between angiotensinogen polymorphisms and lung cancer risk, Med. (Baltimore) 94 (37) (2015) e1250.
[79] S. Montes-Moreno, G. Roncador, L. Maestre, N. Martinez, L. Sanchez-Verde, F. I. Camacho, J. Cannata, J.L. Martinez-Torrecuadrada, Y. Shen, W.C. Chan, Gcet1 (centerin), a highly restricted marker for a subset of germinal center-derived lymphomas, Blood 111 (1) (2008) 351–358.
[80] M.A. Paterson, P.S. Hosking, P.B. Coughlin, Expression of the serpin centerin defines a germinal center phenotype in B-cell lymphomas, Am. J. Clin. Pathol. 30 (1) (2008) 117–126.
[81] S. Erdogan, S. Sezer, E. Baser, O. Gun-Eryilmaz, T. Gungor, S. Uysal, F.M. Yilmaz, Evaluating vaspin and adiponectin in postmenopausal women with endometrial cancer, Endocr. Relat. Cancer 20 (5) (2013) 669–675.
[82] S. Erdogan, S. Sezer, E. Baser, O. Eryilmaz, T. Gungor, S. Uysal, F. Yilmaz, Low serum adiponectin and vaspin levels as novel risk factors for postmenopausal endometrial cancer, Int. J. Gynecol. Cancer (2013) Lippincott Williams & Wilkins 530 Walnut St, Philadelphia, Pa 19106-3621 USA.
[83] Z. Huang, H. Li, Q. Huang, D. Chen, J. Han, L. Wang, C. Pan, W. Chen, M.G. House, K.P. Nephew, SERPINB2 down-regulation contributes to chemoresistance in head and neck cancer, Mol. Carcinog. 53 (10) (2014) 777–786.
[84] E.S. Collie-Duguid, K. Sweeney, K.N. Stewart, I.D. Miller, E. Smyth, S.D. Heys, SerpinB3, a new prognostic tool in breast cancer patients treated with neoadjuvant chemotherapy, Breast Cancer Res. Treat. 132 (3) (2012) 807–818.
[85] E. Pras, P.H. Willemse, A.A. Canrinus, H.W. de Bruijn, W.J. Sluiter, K.A. ten hoor, J.G., aalders, B.G., szabo, E.G. de vries, serum squamous cell carcinoma antigen and CYFRA 21-1 in cervical cancer treatment, Int. J. Radiat. Oncolo. Biol. Phys. 52 (1) (2002) 23–32.
[86] X. Huang, Y. Wei, L. Li, Y. Wen, J. Yang, B. Liu, X. Song, J. Zhao, Serum proteomics study of the squamous cell carcinoma antigen 1 in tongue cancer, Oral Oncol. 2 (1) (2006) 25–30.
[87] K.F. Hsu, S.C. Huang, A.L. Shiau, Y.M. Cheng, M.R. Shen, Y.F. Chen, C.Y. Lin, B.H. Lee, C.Y. Chou, Increased expression level of squamous cell carcinoma antigen 2 and 1 ratio is associated with poor prognosis in early-stage uterine cervical cancer, Int. J. Gynecol. Cancer 17 (1) (2007) 174–181.
[88] Z. Zou, A. Anisowicz, M. Hendrix, A. Thor, M. Neveu, S. Sheng, K. Rafidi, E. Seftor, R. Sager, Maspin, a serpin with tumor-suppressing activity in human mammary epithelial cells, Science 263 (5146) (1994) 526–529.
[89] R. Berardi, F. Morgese, A. Savini, A. Onofri, S. Cascinu, Maspin staining and its use as biomarker in lung cancer, Biomark. Cancer (2015) 345–358.
[90] W.-S. Chang, C.-W. Wu, Diagnosing brain and ovarian cancer via presence of allelic variance in gene associated with serine protease inhibitors (SERPINB13); restriction fragment length polymorphisms, Google Patents, 2003.
[91] W.-s.W. Chang, C.-w. Wu, SERPINB13 single nucleotide polymorphisms and treatment of cancer, US Patent 20,040,005,617, 2004.
[92] M.-T. Wu, S.-C. Lin, C.-Y. Chi, C.-F. Hsiao, J.-T. Ho, H.-C. Lai, L., Chang, W.-S.W. Chang, SERPINB13 SNPS AND THE RISK OF GENETIC CANCER, Program & Abstracts of the 4th Asian-Pacific Organization for Cell Biology Congress Cell Biology in the Post-Genomic Era, 2002.
[93] K. Ozaki, M. Nagata, M. Suzuki, T. Fujiwara, Y. Miyoshi, O. Ishikawa, H. Ohigashi, S. Imaoka, E.i. Takahashi, Y. Nakamura, Isolation and characterization of a novel human pancreas-specific gene, pancpin, that is down-regulated in pancreatic cancer cells, Genes Chromosomes and Cancer 22 (3) (1998) 179–185.
[94] V. Vetvicka, J. Vetvickova, M. Fusek, Anti-human procathepsin D activation peptide antibodies inhibit breast cancer development, Breast Cancer Res. Treat. 57 (3) (1999) 261–269.
[95] M. Fusek, V. Vetvicka, Mitogenic function of human procathepsin D: the role of the propeptide, Biochem. J. 303 (1994) 775–780.
[96] H. Umezawa, T. Aoyagi, H. Morishima, M. Matsuzaki, M. Hamada, T. Takeuchi, Pepstatin, a new pepsin inhibitor produced by agtinomygetes, J. Antibiot. 23 (5) (1970) 259–262.
[97] M.H. Glickman, A. Ciechanover, The ubiquitin-proteasome proteolytic pathway: destruction for the sake of construction, Physiol. Rev. 82 (2) (2002) 373–428.
[98] G. Fenteany, R.F. Standaert, W.S. Lane, S. Choi, E. Corey, S.L. Schreiber, Inhibition of proteasome activities and subunit-specific amino-terminal threonine modification by lactacystin, Science 268 (5211) (1995) 726–731.
[99] B.O. Oyajobi, G.R. Mundy, Receptor activator of NF-κB ligand, macrophage inflammatory protein-1α, and the proteasome, Cancer 97 (S3) (2003) 813–817.
[100] J. Adams, The proteasome: structure, function, and role in the cell, Cancer Treat. Rev. 29 (2003) 3–9.
[101] L. Meng, R. Mohan, B.H. Kwok, M. Elofsson, N. Sin, C.M. Crews, Epoxomicin, a potent and selective proteasome inhibitor, exhibits in vivo antiinflammatory activity, Proc. Natl. Acad. Sci. 96 (18) (1999) 10403–10408.
[102] J. Adams, V.J. Palombella, E.A. Sausville, J. Johnson, A. Destree, D.D. Lazarus, J. Maas, C.S. Pien, S. Prakash, P.J. Elliott, Proteasome inhibitors: a novel class of potent and effective antitumor agents, Cancer Res. 59 (11) (1999) 2615–2622.
[103] P.M. Voorhees, E.C. Dees, B. O'Neil, R.Z. Orlowski, The proteasome as a target for cancer therapy, Clin. Cancer Res. 9 (17) (2003) 6316–6325.
[104] J. Adams, Development of the proteasome inhibitor PS-341, Oncologist 7 (1) (2002) 9–16.
[105] E. Seemuller, A. Lupas, D. Stock, J. Lowe, Proteasome from Thermoplasma acidophilum: a threonine protease, Science 268 (5210) (1995) 579.
[106] G. Schmidtke, H.-G. Holzhütter, M. Bogyo, N. Kairies, M. Groll, R. de Giuli, S. Emch, M. Groettrup, How an inhibitor of the HIV-I protease modulates proteasome activity, J. Biol. Chem. 274 (50) (1999) 35734–35740.
[107] S. Gaedicke, E. Firat-Geier, O. Constantiniu, M. Lucchiari-Hartz, M. Freudenberg, C. Galanos, G. Niedermann, Antitumor effect of the human immunodeficiency virus protease inhibitor ritonavir induction of tumor-cell apoptosis associated with perturbation of proteasomal proteolysis, Cancer Res. 62 (23) (2002) 6901–6908.
[108] A. Krüger, R. Soeltl, I. Sopov, C. Kopitz, M. Arlt, V. Magdolen, N. Harbeck, B. Gänsbacher, M. Schmitt, Hydroxamate-type matrix metalloproteinase inhibitor batimastat promotes liver metastasis, Cancer Res. 61 (4) (2001) 1272–1275.
[109] M.D. Sternlicht, Z. Werb, How matrix metalloproteinases regulate cell behavior, Annu. Rev. Cell Dev. Biol. 17 (2001) 463.
[110] D. Hanahan, R.A. Weinberg, The hallmarks of cancer, Cell 100 (1) (2000) 57–70.

[111] K. Brew, H. Nagase, The tissue inhibitors of metalloproteinases (TIMPs): an ancient family with structural and functional diversity, Biochim. et Biophys. Acta (BBA)-Mol. Cell Res. 1803 (1) (2010) 55–71.
[112] G. Murphy, Tissue inhibitors of metalloproteinases, Genome Biol. 12 (11) (2011) 233.
[113] H.S. Rasmussen, P.P. McCann, Matrix metalloproteinase inhibition as a novel anticancer strategy: a review with special focus on batimastat and marimastat, Pharmacol. Ther. 75 (1) (1997) 69–75.
[114] S. Bramhall, A. Rosemurgy, P. Brown, C. Bowry, J. Buckels, M.P.C.S. Group, Marimastat as first-line therapy for patients with unresectable pancreatic cancer: a randomized trial, J. Clin. Oncol. 19 (15) (2001) 3447–3455.
[115] C.M.O.O. Kleifeld, Validating matrix metalloproteinases as drug targets and anti-targets for cancer therapy, Nat. Rev. Cancer 6 (2006) 227.
[116] B. Turk, Targeting proteases: successes, failures and future prospects, Nat. Rev. Drug Discov. 5 (9) (2006) 785–799.