ISSN 1990-7508, Biochemistry (Moscow), Supplement Series B: Biomedical Chemistry, 2019, Vol. 13, No. 2, pp. 97–112. © Pleiades Publishing, Ltd., 2019.
Russian Text © The Author(s), 2019, published in Biomeditsinskaya Khimiya.

The Role of the Urokinase-Type Plasminogen Activator System in Tumor Progression

E. V. Kugaevskaya[a], T. A. Gureeva[a], O. S. Timoshenko[a], and N. I. Solovyeva[a, *]

[a] *Institute of Biomedical Chemistry, ul. Pogodinskaya 10, Moscow, 119121 Russia*

**e-mail: nina.solovyeva@ibmc.msk.ru*

Received July 18, 2018; revised December 5, 2018; accepted December 5, 2018

Abstract—In the multistage process of carcinogenesis, invasion of malignant cells into normal tissue and their distribution as well as the degree of tissue destruction are the key links in tumor growth and progression. The urokinase-type plasminogen activator system (uPA system) plays the most important role in the development of these processes. The uPA system consists of several components: serine proteinase—uPA, its receptor—uPAR and its two endogenous inhibitors – PAI-1 and PAI-2. The role of uPA, a highly specific protease, consists in triggering the processes performed by the uPA system, which result in destruction of connective tissue matrix (CTM) and basal membranes as well as activation of numerous extracellular and intracellular signaling pathways. uPA converts plasminogen into plasmin and in addition to the regulation of fibrinolysis, it can hydrolyze a number of CTM components and activate zymogens of secreted matrix metalloproteinases (MMPs)—pro-MMPs. MMPs can hydrolyze all the main CTM components and thus play a key role in the development of invasive processes, as well as to perform regulatory functions by activating and releasing from CTM a number of biologically active molecules involved in the regulation of the main processes of carcinogenesis. uPA, PAI-1 and PAI-2 not only regulate uPA/uPAR activity; they are also involved in proliferation, apoptosis, chemotaxis, adhesion, migration, and activation of epithelial-mesenchymal transition pathways. All the above mentioned processes are aimed at regulating invasion, metastasis and angiogenesis. The components of the uPA system are used as prognostic and diagnostic markers of many cancers, as well as serve as targets for anticancer therapy. Selective uPA inhibitors, uPAR peptide antagonists, monoclonal antibodies that can prevent uPA binding to uPAR, as well as antisense oligonucleotides directed against uPA and uPAR are intensively studied in this context as putative and actual pharmacological agents.

Keywords: urokinase-type plasminogen activator (uPA), uPAR receptor, PAI-1 and PAI-2 inhibitors, tumor progression

DOI: 10.1134/S1990750819020069

INTRODUCTION

Invasion of tumor cells into normal tissue is a key process in growth and progression of malignant tumors. This multistep process occurs due to destruction of the connective tissue matrix (CTM) [1–5]. Various proteolytic enzymes are actively involved in degradation of CTM and the basal cell membrane; however, the urokinase-type plasminogen activator system (uPA system) is unquestionably crucial. The uPA system includes uPA, a specific polyfunctional serine proteinase (EC 3.4.21.31), its receptor, uPAR, and two endogenous inhibitors, PAI-1 and PAI-2 [6, 7] (Fig. 1). This system is involved in the modulation of the main processes of tumor growth and development: angiogenesis, invasion, and metastasis [6, 8–10]. Its components—uPA, uPAR, as well as PAI-1 and PAI-2 inhibitors—are involved in cell proliferation, apoptosis, chemotaxis, adhesion, and migration, as well as activation of epithelial-mesenchymal transition (EMI) pathways and signal transduction pathways that are directly associated with tumor progression [6, 11–22] (Figs. 1, 2).

The multifunctional uPA system performs both proteolytic and regulatory functions. The proteolytic functions of the uPA system are aimed at conversion of inactive plasminogen to plasmin (EC 3.4.21.7), a broad-spectrum serine protease; its main function consists in cleavage of thrombin. Besides involvement in fibrinolysis, plasmin hydrolyzes various extracellular components and activates secreted metalloproteinases (MMPs), which together can hydrolyze all major CTM components and activate a number of biologically active molecules, thus promoting development of the malignant process [5–7] (Fig. 1). The regulatory functions of the uPA system are realized through uPAR, which is currently considered as a signal receptor involved in activation of a number of kinases and interacting with such transmembrane proteins as integrins and growth factor receptors; this results in activation of certain signaling pathways that promote tumor

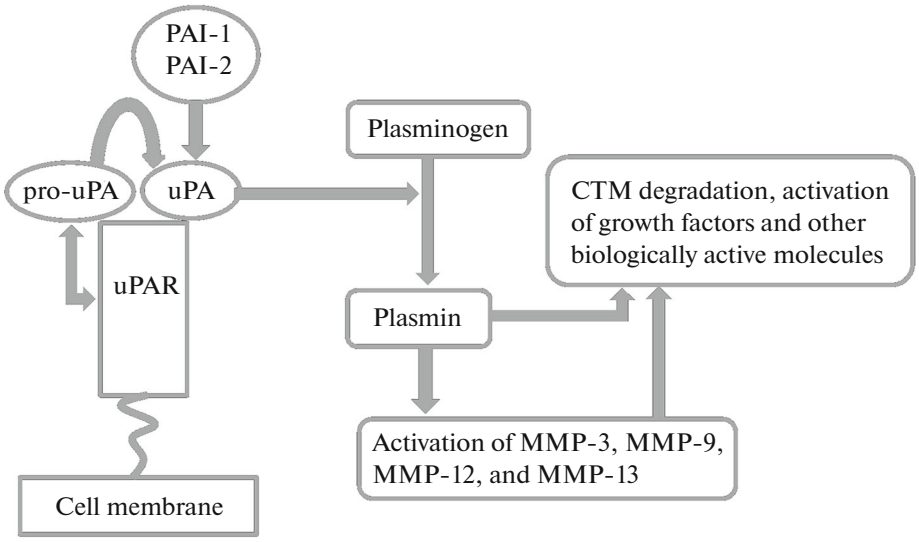

Fig. 1. The uPA system and its proteolytic and regulatory functions. uPA—urokinase type plasminogen activator; pro-uPA—uPA proenzyme; uPAR—uPA receptor; uPA inhibitors—PAI-1 and PAI-2; MMP—matrix metalloproteinases. The interaction of pro-uPA with uPAR results in effective activation of pro-uPA and formation of active uPA, which converts plasminogen to plasmin that performs proteolytic functions by participating in CTM degradation and activation of MMP; MMP can degrade all the main CTM components and release and activate growth factors and other biologically active molecules and thus participate in regulatory processes. Endogenous inhibitors PAI-1 and PAI-2 inhibit uPA activity and plasmin formation.

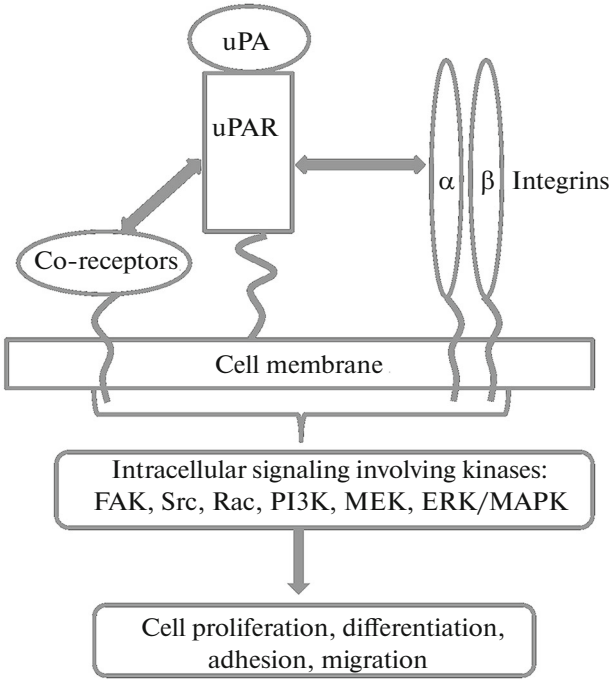

Fig. 2. Functions of uPAR as a signal receptor. uPA—urokinase type plasminogen activator; pro-uPA—uPA proenzyme; uPAR—uPA receptor; α- and β-integrins; co-receptors are receptors for growth factors and chemokines. uPAR interacts with uPA, integrins and co-receptors (receptors of growth factors and chemokines) and this triggers intracellular signaling pathways through activation of signal kinases, such as MAPK, FAK, Src and Rac, JAK, STAT, PI3-K.

progression [6, 23, 24] (Fig. 2). Cancer cells exhibit higher expression of the components of the uPA system than normal cells. In normal cells expression of the *PLAU* gene encoding uPA is at a minimum level, while in tumor cells it is several times higher; this leads to an increase in uPA concentrations. *PLAU* expression is induced by various factors (cytokines, hormones, growth factors, T-cell factors, etc.), which are overexpressed in cancer [6, 25]. uPAR is encoded by the *PLAUR* gene; its transcription is regulated by both different transcription factors [6, 25, 26] and the epidermal growth factor 2 receptor (EGFR 2) [27]. The uPAR expression is increased in tumors, but not in surrounding normal tissue. It serves as a marker for the aggressive course of cancer. Currently, uPAR is considered as a promising therapeutic target for creation of anticancer drugs, but the signaling pathways activated by uPAR in the tumor contribute to the reduction or interruption of the effects of pharmacological agents [17]. The soluble form of uPAR, suPAR, interacts with the same extracellular components as membrane-bound uPAR [28–30]. Although contradictory results have been reported on suPAR involvement in carcinogenesis, most researchers indicate the anticarcinogenic effect of suPAR [31, 32]. The action of PAI-1 and PAI-2 is aimed at suppressing uPA activity, and PAI-1 is more effective than PAI-2. PAI-1 demonstrates anticarcinogenic properties, as it inhibits its activity of the uPA/uPAR complex. In addition, PAI-1 can interact with the region of the amino acid sequence of integrin responsible for binding to vitronectin, which leads to inhibition of cell migration [6, 33]. However, PAI-1 binding to uPA during

uPA/PAI-1 complex formation reduces PAI-1 affinity to vitronectin [33]. Although PAI-1 is an effective uPA inhibitor, its overexpression promotes tumor growth and correlates with a poor prognosis [34–36]. This suggests that PAI-1 has ligands promoting tumor growth and these ligands differ from the uPA system [37]. In contrast to PAI-1, a high level of PAI-2 in cancer is associated with a decrease in tumor growth and metastasis [34].

Thus, overexpression of uPA, uPAR and PAI-1 is a sign of malignancy and correlates with the progression of tumors, invasion, and metastasis, and also serves as an unfavorable prognostic factor. In contrast, inhibition of expression of these components leads to a decrease in the invasive and metastatic ability of many tumors.

1. CHARACTERISTICS OF COMPONENTS OF THE uPA SYSTEM: uPA, uPAR, PAI-1, PAI-2

1.1. Urokinase-Type Plasminogen Activator—uPA

uPA is a serine proteinase of the trypsin family; it is characterized by extremely narrow substrate specificity. The main hydrolytic function of this enzyme consists in plasminogen transformation into plasmin, a multifunctional proteinase with broad substrate specificity (Fig. 1). In mammals, a tissue-type plasminogen activator, tPA, has been also identified; it plays a crucial role in vascular fibrinolysis (EC 3.4.21.68). In a plasminogen molecule consisting of 791 residues, uPA and tPA hydrolyze the same peptide bond located between Arg561–Val562 [38]; at the same time plasminogen is converted into plasmin. uPA and tPA are synthesized as precursors, and plasmin is their main activator [6, 10]. The uPA molecule is synthesized as a precursor, pro-uPA; in the human body it is encoded by the *PLAU* gene located on chromosome 10q24. Pro-uPA is secreted as a single-chain protein consisting of 411 residues with molecular mass of 53 kDa [39, 40]. Activation of pro-uPA occurs when it binds to its own receptor—uPAR; this is accompanied by cleavage of one peptide bond located between Lys158–Ile159. This process involves membrane-bound plasmin (Fig. 1) or some other proteinases, such as cathepsins B and L, thermolysin, trypsin, kallikrein. The activation leads to the formation of the active form of uPA, consisting of two polypeptide chains, A and B, containing 158 and 253 residues, respectively, which are linked together by a disulfide bond Cys148–Cys270 [19, 41, 42]. Activated uPA triggers a proteolytic cascade, including plasminogen activation followed by its transformation into plasmin, which serves as the main activator of matrix metalloproteinase precursors (pro-MMPs) responsible for CTM destruction and participating in activation of a number of regulatory molecules that are involved in both external and in the internal signaling (Fig. 1) [6, 8, 10]. The catalytically active uPA (consisting of two polypeptide chains), which has a very narrow specific activity, converts plasminogen into plasmin, the main pro-uPA activator. The resultant uPA activates plasminogen. Thus, reactivation, reproduction, and accumulation of uPA and plasmin occur in the pericellular space (Fig. 1). This process is a fast and effective source of uPA, plasmin, MMPs and regulatory factors that play a crucial role in the processes of adhesion, migration and invasion of cells in normal physiological and pathological conditions.

1.2. uPA Receptors: Membrane Bound uPAR and Soluble suPAR

The uPAR receptor belongs to the family of receptors associated with the cell membrane via glycosylphosphatidyl-inositol (GPI-anchored receptors). uPAR lacks a transmembrane region and this determines its mobility on the cell surface (Fig. 2). The human uPAR gene *PLAUR* is located on chromosome 19q13 and encodes a protein consisting of 335 residues [40]. uPAR serves as a receptor for both uPA and pro-uPA. uPAR binding to uPA and to pro-uPA stimulates conversion of single-chain pro-uPA into active double-chain uPA, which contributes to the manifestation of its proteolytic functions and first of all conversion of plasminogen to plasmin [43]. uPAR is a signaling receptor that binds CTM adhesive proteins, vitronectin and fibronectin; different receptor sites are involved in interaction with vitronectin and uPA, which makes it possible to y bind simultaneousl both ligands and affect its adhesive and proteolytic properties. It has been found that uPAR can interact with transmembrane proteins, such as integrins and growth factor receptors coupled to G-proteins (GPCR); this results in activation of intracellular signaling through activation of various kinases, such as MAPK, FAK, Src, Rac, Jak and others (Fig. 2) [6, 7, 44, 45]. uPAR interaction with transmembrane ligands activates ion proliferation, differentiation, adhesion, and cell migration. These processes occur independently of the proteolytic activity of uPA.

uPAR exists not only in membrane-bound, but also in soluble form (suPAR), which is released from the plasma membrane cleavage of the GPI-anchor catalyzed by specific phospholipases C and D [32]. suPAR is found in biological body fluids, endothelial cell lines, and also in various tumors [46, 47]. It is suggested that formation of suPAR is a regulatory mechanism aimed at reducing the amount of membrane-bound uPAR on the cell surface; it includes competitive displacement of the uPA/uPAR complex followed by termination of its signaling. It has been found that suPAR can directly (and independently of uPA) block the uPAR signaling; suPAR binds the same extracellular ligands that interact with uPAR, for example, to uPA and vitronectin; this prevents interaction with uPAR, and thus reduces proteolytic and adhesive activities of the cell [28, 29]. At the same time, suPAR

interaction of with integrins can induce adhesion [30]. The role of suPAR in the development of different types of tumors is ambiguous. An elevated level of suPAR was detected in various carcinomas [48]; it also served as a negative prognostic factor in multiple myeloma [49], colorectal [50], breast and lung [51] carcinomas. At the same time, at high levels of suPAR (which could result in increased binding of suPAR to uPA) there was a decrease in the growth and development of ovarian and mammary gland carcinomas [52, 53]. Animal studies have shown that suPAR cleavage by proteinases neutralized its inhibitory effect on tumor development. It is suggested that the proteolytic cleavage of suPAR represents the mechanism of neutralization of the anticarcinogenic activity of suPAR [31].

1.3. Endogenous Inhibitors of uPA: PAI-1 and PAI-2

PAI-1 and PAI-2 are specific endogenous uPA inhibitors (Fig. 1); they belong to the family of serine proteinase inhibitors, serpins. Two other members of the serpin family, nexin-1 and protein C inhibitor (PAI-3), can also interact with uPA; however, their effect on the uPA activity is very small.

PAI-1 (serpin-1) is the main uPA inhibitor, which is widely distributed in cells, organs and tissues of the human body. It has been found in endothelial cells, platelets, fibroblasts, hepatocytes, macrophages, as well as in plasma, placenta, vascular smooth muscle, and adipose tissue stromal cells [54]. PAI-1 is localized in the extracellular space [55, 56]. PAI-1 is encoded by the *serpin1* gene located on chromosome 7q21.3-q22. Gene transcription is controlled by various regulatory factors, characterized by altered expression in carcinogenesis [40, 57]. The PAI-1 molecule is a single polypeptide chain with molecular mass of 45 kDa, which consists of 379 or 381 residues. The binding site of uPAR with uPA is located at the C-terminal part of the molecule and the interaction occurs via the peptide bond Arg346–Met347 [54]. The stable PAI-1/uPA complex with a 1 : 1 stoichiometry is formed very quickly. PAI-1 can interact with vitronectin, affect cell adhesion and migration, as well as inhibit apoptosis regardless of its interaction with uPA [42]. The data obtained indicate that PAI-1 blocks the activity of the uPA system and the uPa/uPAR complex, and thus exhibit an anticarcinogenic effect. However, PAI-1 overexpression promotes tumor growth, invasion, as well as angiogenesis and correlates with poor prognosis [6, 55, 56]. It is suggested that there are sites on the surface of PAI-1 that differ from the binding sites with the uPA system and these sites promote tumor growth [6, 55, 56, 58].

PAI-2 is encoded by the *serpin2* gene located on chromosome 18q21.3. As in the case of PAI-I, gene transcription is regulated by various factors. PAI-2 belongs to the group of structurally conservative ovalbumin-like serpins [19]. PAI-2 exists in 2 forms; the first is an intracellular protein of 415 residues and molecular mass of 47 kDa. The second form is a secreted extracellular protein from molecular mass of 60 kDa. The inhibitor reactive center includes the Arg380–Thr381 peptide bond, which participates in interaction with uPA. The major proportion of PAI-2 is a protein that remains inside the cell and whereas only a small part of PAI-2 is secreted. The secreted PAI-2 protein is an effective uPA inhibitor, although it acts slower than PAI-1 [59, 60]. Intracellular PAI-2 has been shown to play a role in the control of apoptosis [61, 62]. Good evidence exists that PAI-2 is involved in regulation of collagen remodeling in the stroma, thus affecting tumor growth and invasion [12]. High expression of PAI-2 has been found to correlate with increased lifespan of patients, a decrease in tumor growth and metastasis, as well as a decrease in the tumor growth rate in various types of cancer [55, 56, 63, 64].

2. PHYSIOLOGICAL FUNCTIONS OF THE uPA SYSTEM

The components of the uPA system play an important role in various normal physiological and pathological processes (Fig. 3). In normal cells, expression of the *PLAU* and *PLAUR* genes encoding uPA and uPAR, respectively, is at a minimally low level, which increases dramatically under pathological conditions especially pronounced in carcinogenesis and inflammation [40]. The uPA system plays a key role in the fibrinolytic system, which is responsible for conversion of inactive plasminogen to plasmin. The main physiological function of plasmin consists in cleavage of fibrin. Under physiological conditions, fibrin formation induces plasminogen activation by its activators (uPA and tPA), which convert plasminogen into plasmin on the surface of fibrin clots and resultant plasmin degrades these clots to soluble fragments [65]. Plasmin can cleave fibrinogen and a number of blood coagulation factors, as well as activate inactive forms of MMPs (pro-MMPs) responsible for the CTM degradation. These processes are involved in the normal physiological homeostatic mechanism of wound healing [66].

The components of the uPA system and plasmin may be involved in the regulation of hematopoiesis. They activate cytokines such as fibroblast growth factor (FGF), transforming growth factor β (TGF-β), interleukin-1β (IL-1β). The study of the effect of these cytokines on the regulation of plasminogen activators (uPA and tPA) and their inhibitors PAI-1 and PAI-2 in human bone marrow stromal fibroblasts has shown that all three cytokines stimulated secretion and expression of uPA and tPA (from 10 to 300 times). PAI-1 and PAI-2 are also regulated by these cytokines. IL-1β insignificantly reduced the PAI-1 level but caused a 6-fold increase in the PAI-2 level. FGF and

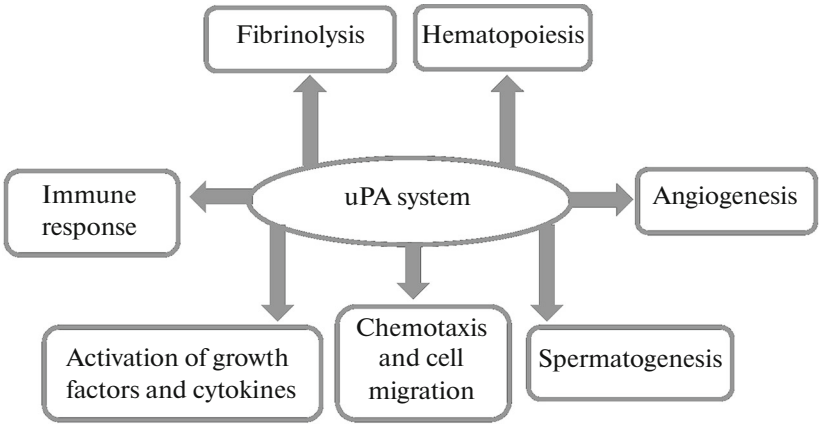

Fig. 3. Physiological functions of the uPA system. Under normal physiological conditions, the uPA system is involved in various biological processes, such as fibrinolysis, wound healing, hematopoiesis, innate and acquired immunity, angiogenesis, activation of growth factors and cytokines, chemotaxis, cell migration, spermatogenesis.

TGF-β had no effect on the PAI-2 level. Thus, these three cytokines are involved in the regulation of uPA and tPA levels and their inhibitors in the bone marrow stromal cells. Formation of uPA and tPA and plasmin in the bone marrow can be one of the factors regulating hematopoiesis [67, 68].

The uPA system is involved in both normal physiological and pathological angiogenesis. Angiogenesis is a multistep process that begins with activation of endothelial cell induced by proangiogenic factors and, first of all, VEGF and its receptors, the main regulators of angiogenesis in both the embryonic and postnatal development of the organism [69]. VEGF is released from the CTM via the proteolytic cascade triggered by the uPA/uPAR complex. uPA is the key regulator of this process [6, 10]. The proteolytic cascade results in formation of plasmin, activation of MMPs and a number of physiologically active factors required for the development of angiogenesis. Physiological angiogenesis occurs during fetal development and growth; it develops evenly and orderly, with moderate intensity, and leads to formation of normal vessels. Pathological angiogenesis occurs during regeneration of damaged tissues, carcinogenesis, chronic inflammatory diseases, myocardial infarction, etc.; it leads to formation of abnormal, heterogeneous vessels with irregular branching and high permeability for plasma proteins [69].

The uPA system is involved in both innate and acquired immunity. It has been found that expression of components of the uPA system occurs in various types of hematopoietic cells responsible for immunity [70]. Their expression levels change dramatically in infections thus implying involvement of the uPA system in the immune response. Bacterial infection is characterized by activation of proinflammatory cytokines, such as tumor necrosis factor (TGF-α), interleukin-1α and interleukin-1β (IL-1α, IL-1β), interferon γ; this results in an increase in the expression and secretion of uPA by various types of monocytes, neutrophils, parts of lymphocytes, epithelial and endothelial cells involved in innate immunity [71]. The uPA system is also involved in acquired immunity; uPA and uPAR expression was significantly increased during T cell activation as compared to their expression level in resting cells [72]. In uPA-deficient mice, T-helpers 1 and 2 were shown to lose their ability to react on pathogen action [73, 74]. uPAR blockade limited migration of leukocytes in vitro and prevented T cell concentrating in vivo [75].

The uPA system plays a key role in chemotaxis. The chemotactic activity of uPA is determined by its N-terminal domain, which lacks a catalytic center and does not possess proteolytic activity. Chemotaxis depends on the interaction of uPA with uPAR. The presence of uPAR on the surface of leukocytes provides uPA concentrating on them [68]. It is suggested that formation of suPAR causes a decrease in the uPAR content on the cell surface [31]. In non-migrating resting cells, uPAR is located on their apical side, and during migration it moves to the leading edge of cells [76]. In addition, cell migration contributes to CTM destruction by products of the uPA system, such as MMPs and plasmin, and the components of the uPA system provide activation, modification and release of biologically active molecules from CTM, which activate a number of extracellular and intracellular signaling pathways [5–7, 23, 24].

The uPA system is involved in the regulation of male reproductive function; uPA is found in semen, in the testice and its appendages, as well as in the prostate. uPA improves sperm motility, induces acrosomal reaction and increases the sperm ability to fertilize eggs [77]. These data suggest the development of a new approach to male contraception [78].

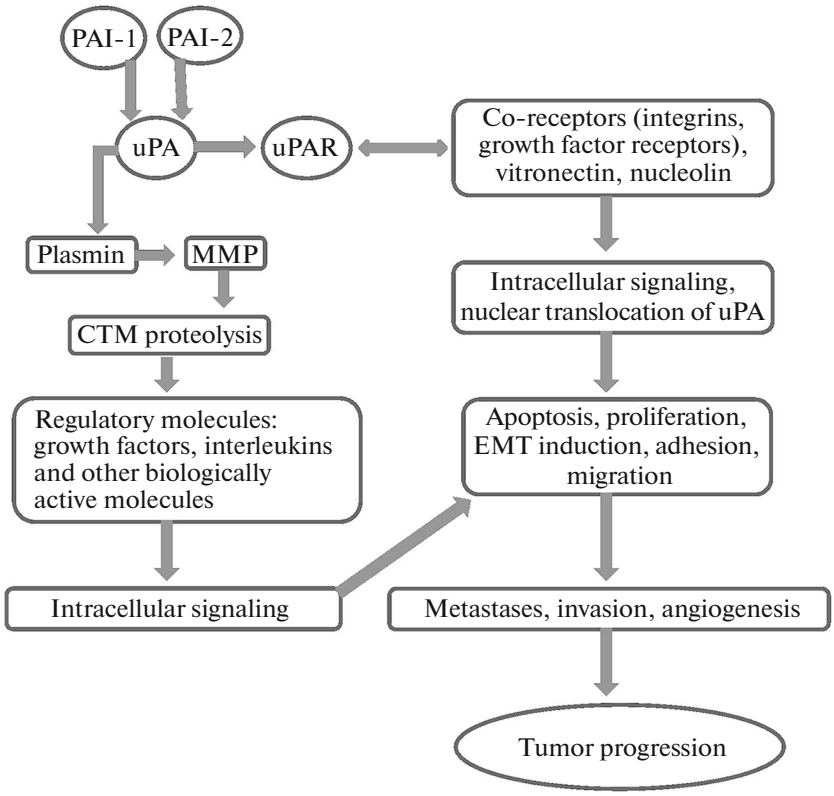

Fig. 4. The role of the uPA system in tumor progression. uPA—urokinase type plasminogen activator; uPAR—uPA receptor; uPA inhibitors—PAI-1 and PAI-2. uPA binding to uPAR activates uPA and initiates the further proteolytic cascade, including plasmin formation and MMP activation, leading to degradation of extracellular matrix proteins and release of regulatory molecules, such as growth factors, cytokines, and others. uPA binding to vitronectin and co-receptors activates intracellular signaling, which may be involved in uPA nuclear translocation and gene expression. Both pathways affect cell migration, adhesion, proliferation, apoptosis and EMT induction, thus playing a key role in the main processes of tumor progression: invasion, metastasis and angiogenesis.

3. FUNCTIONS OF THE uPA SYSTEM IN THE REGULATION OF DESTRUCTIVE PROCESSES IN CARCINOGENESIS

The components of the uPA system are involved in the development of the main processes of tumor growth and development: invasion, metastasis, and angiogenesis (Fig. 4). The functions of the uPA system are divided into two types: dependent and independent of its proteolytic action.

3.1. Proteolytic Functions of the uPA System

The first type of the uPA system functions includes the proteolytic cascade, which is initiated by uPA after its interaction with uPAR; the cascade includes activation of pro-uPA and formation of plasmin caused by uPA. Plasmin can independently degrade many CTM proteins, including fibrin, laminin, fibronectin and perlecan (heparan sulfate proteoglycan), as well as activate latent secreted MMPs, such as MMP-1, MMP-3, MMP-8, MMP-9, MMP-12, and MMP-13 [6, 7, 16]. MMPs exhibit relative substrate specificity, but collectively they are able to hydrolyze all the major CTM components: MMPs-1-8-13 cleave the fibrillar collagen types I, II, III, V, VII, X, XI, fibronectin, laminin, vitronectin, aggrecan, entactin and others; MMPs-2-9-3-12 cleave type IV collagen (the main component of the basement membranes), as well as collagen types III, V, VII, X, XI, elastin, fibronectin, laminin, vitronectin, aggrecan, plasminogen, etc.; MMPs-3-10-12-13 cleave elastins, proteoglycans, collagen types III, IV, V, VII, IX, X, XI, fibronectin, vitronectin, laminin, etc., as well as other substrates unrelated to CTM [1, 2, 5]. The action of MMPs leads to destruction of CTM and basement membranes, which promoting development of invasive and metastatic processes. In addition to destructive functions MMPs play a regulatory role by activating, inactivating and modifying properties of a number of biologically active molecules (Figs. 1, 4). It has been shown that proteolysis of CTM components such as laminin and fibronectin leads to formation of fragments of these proteins exhibiting their own biological activity: they can stimulate motility and migration of cancer cells [3–5, 42]. Degradation of CTM leads to release of STM-associated mitogenic and angiogenic growth factors such as bFGF (basic fibroblast growth factor),

VEGF (vascular endothelial growth factor), HGF (hepatocyte growth factor), IGF (insulin-like growth factor), EGF (epidermal growth factor), and TGF-β (transforming growth factor β), which are able to stimulate growth and proliferation of cancer cells and angiogenesis [6, 7]. This proteolytic cascade depends on the type of tumors and is especially important in the early stages of tumor progression, when degradation and remodeling of the tumor-surrounding tissue is important [1, 7, 10, 19].

3.2. Functions of the uPA System Independent of Its Proteolytic Functions

The second type of uPA system functions include functions that are not related to its proteolytic role but may also contribute to tumor progression. During the last decade, experimental evidence has been obtained indicating that uPAR functions not only as a proteinase receptor, but also as a signaling receptor that have a significant impact on migration, adhesion, differentiation, and cell proliferation via intracellular signaling pathways (Figs. 2, 4). The uPAR functioning as a signal receptor does not depend on the uPA proteolytic activity [6, 7, 15]. uPAR is able to interact with a number of plasma membrane proteins, such as some integrins, transmembrane growth factor receptors (EGFR) and chemokine receptors coupled to G-proteins [16, 79, 80]. uPAR binding to uPA and co-receptors can initiate various signaling pathways either through activation of MAPK, FAK, Src and Rac kinases, or through activation of JAK, STAT, PI3-K kinases [6, 7, 10, 79].

Integrins are the most studied and important co-receptors of uPAR. They are associated with various signaling molecules, for example, kinases from the FAK and Src families, which activate signaling pathways stimulating tumor progression [6, 7, 43, 81]. uPAR interacts with β1, β2 and αV integrins [6, 7, 43] as it has been demonstrated on various types of cancer cells including: lung cancer, breast cancer, ovarian cancer, melanoma, prostate cancer and head and neck carcinoma [80]. It is suggested that activation of a specific uPAR mediated signaling pathway depends on its interaction with certain integrins. For example, uPAR interactions with β1-integrins are associated with activation of FAK and ERK signaling, while uPAR interaction with β3 integrins is associated with activation of Rac signaling [79]. It has been shown that some uPAR co-receptors, such as the proteins caveolin and tetraspanin (CD82/KAI1, tumor metastasis suppressor) [82], can also form complexes with integrins, and thus affect their interaction with uPAR and thus influence tumor progression.

It should be emphasized that during tumor development increased expression of uPAR is observed only in the tumor tissue, but not in the normal tissue surrounding the tumor. Therefore, uPAR can serve as a marker for the tumor destructive potential [83].

The soluble receptor suPAR competes with uPAR for binding to uPA and a number of extracellular regulatory molecules, thus suggesting its anticarcinogenic effect. However, existing data on the suPAR involvement in the process of tumor development are contradictory and suggest that an increase in the suPAR expression can serve as a negative prognostic factor [49–51, 84] and can also reduce the development of tumor growth and development processes [52, 53]. Experiments on cell lines have shown that suPAR can directly and independently of uPA block uPAR signaling pathways. This process depended on the cell type and signaling pathways induced by the uPA/uPAR system in these cells. The inhibitory effect of suPAR signaling on cell growth and invasion processes was established. The proteolytic cleavage of suPAR neutralized its anticarcinogenic action [31].

There is another pathway by which uPA can affect intracellular processes, regardless of proteolytic activity and enzyme degradation, as well as uPAR binding (Fig. 4). It has been shown that uPA interaction with the nucleocytoplasmic shuttle protein, nucleolin, leads to rapid uPA translocation into the nucleus, where it can participate in regulation of gene transcription, particularly, to induce expression of smooth muscle α-actin [16, 85].

3.3. The Role of the uPA System in Induction of Proliferation, Adhesion, Migration and Apoptosis of Tumor Cells

The ratio of the processes of increased proliferation to inhibition of apoptosis is an important link in the development of tumor progression. The ability of the uPA system to induce proliferation of human cancer cells has been demonstrated in a number of in vitro and in vivo studies [16, 85]. Molecular mechanisms determining involvement of the uPA system in the proliferation process include both proteolytic and regulatory functions. The proteolytic cascade, initiated by uPA interaction with uPAR, provides CTM destruction and activation/release of growth factors (VEGF, FGF, IGF, etc.), that stimulate proliferation and tumor growth [6, 8, 10]. In addition, proliferation of cancer cells depends on the interaction of the uPA/uPAR complex with integrins; this leads to activation of proliferation through the p38MAPK signaling system [16]. uPA stimulates proliferation regardless of its proteolytic activity and interaction with uPAR. It is suggested that this process is regulated via the ERK/MARK signaling pathway triggered by integrins [16, 86]. The uPAR interaction with integrins of the β1 family (α5β1 and α3β1) and with EGFR is responsible for its participation in proliferation. Thus, binding of uPAR to α5β1 integrin induces activation of the ERK/MAPK signaling pathway that controls proliferation, cell motility, and apoptosis [6, 7, 44, 45]. The ERK/MAPK signaling pathway is also activated when uPAR interacts with EGFR [44]; the latter

is involved in regulation of cell proliferation, survival, and tumor growth [6, 7, 87]. It is suggested that the important role of EGFR in tumor progression is directly related to uPAR signaling, because both in vitro and in vivo experiments have shown that expression of uPAR is a necessary precondition for activation and manifestation of the EGFR mitogenic activity [10]. It has been shown that in serum of prostate cancer patients, the levels of EGFR and uPAR are positively correlated with each other [88]. Using the COS-7 cell line (green African monkey kidney fibroblasts), it has been demonstrated that EGFR inhibitors are able to block signal transduction pathways initiated by the uPA-uPAR interaction, and therefore have a negative impact on proliferation [31].

Involvement of the uPA-system in adhesion and migration of cell in cancer is currently well studied [6, 7, 9, 10, 42]. Cell migration is of great importance for development of metastasis and angiogenesis; it depends on particular components that regulate cell adhesion and their release from CTM. Regulation of these processes occurs in several pathways. As in the case of proliferation, the influence of the uPA system on adhesion and migration is determined by its proteolytic and regulatory functions. First, CTM degradation performed by products of the uPA system—plasmin and MMPs—results in destruction and remodeling of tumor-surrounding tissues; this promotes cell migration and modification of a number of adhesive molecules (fibrin, laminin, vitronectin, integrins, interleukins, etc.), as well as activation of physiologically active molecules, such as growth factors, cytokines, etc. These processes stimulate migration and affect the adhesive properties of the cells. Secondly, uPAR interaction with uPA and co-receptors leads to activation of intracellular signaling pathways, which affect cell adhesion, proliferation, differentiation, and migration regardless of the proteolytic function of the uPA system [1, 6, 7]. It has been shown that the integrin-dependent activation of the ERK/MAPK signaling pathway, which induces cell proliferation, survival, and cell motility, is essential for stimulation of cell migration, [6, 7, 42].

However, uPAR interaction of with various integrins and adhesive molecules leads to different effects. Direct uPAR binding to vitronectin in the extracellular matrix facilitates cell adhesion, migration, and invasion [89–91]. This interaction is stimulated by the uPAR binding to uPA and is inhibited by PAI-1 (uPAR and PAI-1 interact with the same vitronectin site) [91].

The uPA system is involved in inhibition of apoptosis. It has been shown that cells of a number of malignant tumors, such as breast, rectum and lung cancers, are resistant to apoptosis [6, 7, 9, 10]. There is a correlation between expression of uPA and uPAR and cell sensitivity to apoptosis. In epithelial cell lines the uPAR expression level positively correlated with cell resistance to apoptosis, while uPA or uPAR blockade with specific antibodies led to increased apoptosis in tumor cells [16, 92]. PAI-1 can be involved in both increasing and inhibiting apoptosis. For example, in prostate cancer, PAI-1 expression correlated with an increase in apoptosis in tumor endothelial cells [93]. However, in some cases, PAI-1 reduced apoptosis through inhibition of caspase-3, involved in regulation of this process [94, 95].

3.4. The uPA System in the Epithelial-Mesenchymal Transition

Cancer cells that have separated from the tumor and migrated into the surrounding tissue can change their morphological properties and acquire the phenotype characteristic of mesenchymal cells. These cells are characterized by higher invasiveness, migrating capacities, as well as increased resistance to apoptosis than epithelial cells [96, 97]. This process is known as the epithelial-mesenchymal transition (EMT). It represents an important step leading to invasion and metastasis, in which the uPA system plays a significant role [98]. Mesenchymal type invasion was observed during development of various malignant tumors, for example, fibrosarcoma, glioblastoma, melanoma, etc. [99–102]. One of the mechanisms responsible for EMT induction is hypoxia, which is observed when cancer cells are separated from a tumor and located at a distance of more than 180 μm from blood vessels [20]. Using the epithelial cell lines CHO and 293 (hamster ovary and human embryonic kidney, respectively), it was demonstrated that phenotypic changes similar to the changes occurring in EMT were observed in cells expressing uPAR and depended on direct interaction of uPAR with vitronectin [103, 104]. In the human breast cancer cell line MDA-MB-468 characterized by an epithelial cell phenotype, hypoxia was found to induce EMT by increasing uPAR expression and activating uPAR-dependent cellular signaling. uPA binding to uPAR activates ERK1/2- and PI3-K-mediated signaling pathways, and direct interaction of uPAR with vitronectin activates Rac1 signaling, which promotes cell migration [20, 105]. EMT induced by uPAR is a reversible phenomenon that can be canceled by reoxygenation, which prevents uPA interaction with uPAR and inhibits PI3K, Src and ERK-mediated signaling. The study of the MDA-MB-231 breast cancer cell line, which has a mesenchymal phenotype, has shown that a high level of uPAR expression in cells is necessary maintenance of mesenchymal morphology by the cells, while uPAR gene knockdown changed their phenotype to the epithelial one [20]. It is suggested that EMT represents the main route for metastasis of tumor cells [7].

3.5. uPA System in Angiogenesis

Angiogenesis, the process of formation of new blood vessels, depends on activation, proliferation, adhesion, migration, and maturity of endothelial cells. It is one of the most important mechanisms of tumor growth. Increased intensity of angiogenesis is one of the main causes of the rapid growth of malignant tumors, as well as one of the mechanisms for its metastasis. Angiogenesis begins with activation of endothelial cells by pro-angiogenic factors, such as VEGF and interleukins [6, 106, 107]. One of the main stimulators of angiogenesis is hypoxia, in which the activator of the transcription of angiogenesis factors, factor 1 (HF1), induces expression of many angiogenic factors, and, first of all, the main inducer of angiogenesis—VEGF and its receptors [16]. The uPA system increases tumor angiogenesis induced by VEGF via activation of pericellular proteolysis, increased vascular permeability and by maintaining proliferation and migration of endothelial cells [108, 109]. The main mechanism by which the uPA system stimulates angiogenesis is the proteolytic cascade triggered on the cell surface by the uPA/uPAR complex, which involves formation of plasmin, activation of MMPs, CTM degradation, release of CTM-associated growth factors, such as VEGF, VEGF-E, bFGF, EGF, HGF, and cytokines [10, 19, 108, 110]. In stomach cancer there was a significant correlation between the expression levels of uPA, uPAR, PAI-1 and VEGF, and clinical and pathological factors. Evaluation of the number of microvessels in endothelial cells showed that their number was significantly higher in patients with increased expression of uPA, uPAR or VEGF [111]. Ex vivo experiments have demonstrated that single-chain uPA increased expression of the VEGF receptors (VEGFR-1 and VEGFR-2) on the endothelial cell surface and promoted cell migration [112]. Increase uPA expression in endothelial cells resulted in an increase not only in cell proliferation, but also in cell migration and formation of capillary structures, while uPA binding to uPAR and activation of not only pro-uPA, but also the MAP kinase signaling pathway led to the migration of endothelial cells and angiogenesis [16]. In addition, it was shown that suPAR could promote angiogenesis regardless of its participation in the proteolytic cascade [113]. The role of PAI-1 in the regulation of angiogenesis is multifunctional; low expression of PAI-1 has a stimulating effect on angiogenesis, while increased expression inhibits it [114]. However, studies have shown that PAI-1 expression by host cells but not by the tumor cells is the determining factor for development of the vascularization process. In the in vivo system, high PAI-1 concentrations produced by tumor cells did not compensate the lack of PAI-1 in host cells and did not inhibit tumor angiogenesis [11, 115, 116].

4. CLINICAL IMPORTANCE OF DETERMINATION OF THE uPA SYSTEM COMPONENTS IN CANCER

The knowledge on the important role of the uPA system components in the progression of malignant tumors is employed in clinical practice, where they are used as diagnostic, prognostic and therapeutic targets, because in malignant tumors their expression is usually higher than in normal tissues (Table 1).

The overexpression of uPA is an indicator of low overall and relapse-free survival. The higher uPA expression was found in actively invasive and metastatic tumors as compared with the primary tumor [107, 111] (Table 1). uPAR, as well as uPA, is expressed by tumor cells and its level is associated with tumor aggressiveness. Since increased uPAR expression is observed in the tumor tissue, but not in surrounding normal tissues, it is proposed to use uPAR as a therapeutic target [8, 10, 15, 117, 118]. The uPA inhibitors, PAI-1 and PAI-2, are present in the tumor tissue and plasma. The role of PAI-1 consists not only in inhibition of invasion and metastasis (through inhibition of plasminogen activation and the subsequent proteolytic cascade), but also in tumor protection against proteolysis [36, 119–121]. This may be one of the plausible explanations that a high level of PAI-1 is often an unfavorable prognostic factor. The increased PAI-1 level is associated with increased risk of metastasis and tumor recurrence, while the increased PAI-2 level in the tissue correlates with a favorable prognosis [6, 34]. In various human malignant tumors, increased expression of uPA, uPAR, and PAI-1, as well as positive correlation between their levels, is a poor prognosis [6, 9, 10, 121, 122].

Data of Table 1 on the role of various components of the uPA system in most common malignant tumors of various organs, indicate that as a rule, the components of the uPA system are good prognostic markers for overall and relapse-free survival, as well as for the results of cancer treatment, and the level of their expression correlates with invasion and metastasis. For example, an increased level of uPA in tumor tissue has a prognostic value in breast, stomach, esophagus, ovarian, prostate, lung, liver, and other cancers; high levels of uPAR are associated with poor prognosis for breast, endometrial, prostate, lung, stomach, liver and colon cancers; an increased level of PAI-1 correlates with reduction in overall and/or relapse-free survival in cancer of the kidney, ovary, breast, colon, liver and other cancers (Table 1).

Data presented in this review show that changes in the expression level of components of the uPA system, particularly increased concentrations of uPA, uPAR and PAI-1, are observed in almost all malignant tumors of various localization. Data available in the literature indicate that the uPA/uPAR complex may serve as the best prognostic indicator, and changes in the ratio of uPA to its endogenous inhibitors PAI-1

Table 1. The role of the uPA system in malignant tumor development in various organs

Cancer localization	Components	The role of the uPA system in tumor development	References
Breast	uPA, PAI-1, and uPAR	Prognostic markers for relapse-free and overall survival and also for results of cancer treatment; correlate with tumor invasion and metastasis; high expression is associated with poor prognosis	123 124 125 126
	PAI-2	High level is associated with favorable prognosis	127
Ovarian carcinoma	uPA and PAI-1	The prognostic value for overall survival during tumor progression, where expression of these components has been increased	128 129 130 131
	suPAR (soluble uPAR)	It is associated with poor survival of patients with a high level of expression before surgery; it is important for predicting and evaluating chemotherapy effectiveness	132
Cervical carcinoma	uPA	Increased expression in the tumor. Assessment of localization in tissues may predict lymph node metastasis	133
	uPA and PAI-1		134
	uPA and PAI-1 and PAI-2	Increased expression in invasive cervical carcinoma	13 120
Endometrium	uPAR, suPAR	Increased expression in plasma and tissues correlated with tumor progression	135 136
	PAI-1	High levels correlated with shorter relapse-free and overall survival and advanced cancer stages	137 138
Prostate	uPA, and uPAR	High expression level correlated with aggressiveness, progression after surgery, invasion, metastases, and also with overall and relapse-free survival	139
	suPAR	Low overall survival at increased expression	140
	PAI-1	Increased expression was associated with tumor development and survival of patients	141 142
Colon/rectum	uPA, PAI-1 and uPAR	Prognostic markers for tumor progression, survival of patients after surgery and therapy	143 144
	uPAR, PAI-1 and PAI-2	Overexpression is associated with poor response to therapy.	145
	suPAR	High level before surgery is associated with poor prognosis for survival	146
Lung	uPA, uPAR, PAI-1 and PAI-2	Expression increases in tumor tissues of non-small cell lung cancer	147 148
	suPAR	Increased expression is associated with short overall survival in small cell lung cancer	149
Pancreas	uPA	Increased expression is associated with low survival of patients after pancreatic resection	150
	PAI-2	The gene encoding PAI-2, often eliminates with pancreatic adenocarcinoma and this leads to increased metastasis	151
Stomach	uPA and uPAR	Increased expression correlates with low survival rate and can be used as a prognostic marker (although some studies question this viewpoint)	152 153
	uPA, uPAR	Low expression correlates with better survival rate of patients	8
	uPA and PAI-1	Prognostic factors for relapse-free survival	154
Esophagus	uPA, PAI-1	The ratio uPA/PAI-1 correlates with invasion and survival rate of patients	9
	uPA	Increased expression is associated with low overall survival	155
	PAI-2	Protects against local invasion	8
Liver	uPA, uPAR, PAI-1	High expression in tumor tissues promotes increased invasion and metastases	8
	uPAR, PAI-1	Expression level correlates with poor prognosis	156
Head and neck	uPA, PAI-1 and suPAR	Increased expression correlates with invasion and metastasis and has a prognostic value	157 158 159

and PAI-2 are the better survival parameter than determination of only the levels of uPA/uPAR or PAI- 1 and PAI-2 [6, 7, 9]. Inhibition of expression of uPA and uPAR or blockade of their interaction suppresses progression of various types of cancer [11, 19, 107, 160]. It should be noted that besides determination of the components of the uPA system, products of uPA-mediated activation of pro-MMPs by plasmin can be used as direct markers of tumor potential, as evidenced by numerous data on the role of MMPs in invasion and metastasis, including our results of the study of the role of MMP-1, -2, -9 in squamous carcinoma of the cervix [3–5, 133, 134].

5. DIAGNOSTIC METHODS FOR ESTIMATION OF EXPRESSION OF THE uPA SYSTEM COMPONENTS IN MALIGNANT TUMORS

Numerous data on the altered expression of the uPA system components in malignant tumors suggest their use as informative diagnostic, prognostic, and therapeutic targets [6–8, 106, 161]. For this purpose several methods are used. Determination of the content of particular components of the uPA system is performed by an enzyme immunoassay (ELISA) method, which requires the use of fresh or freshly frozen tissue samples [162]. The other method employs analysis of uPA and PAI-1 expression by the level of their mRNA [163–165]. In contrast to the ELISA method, the advantage of the latter method consists in the use of formalin-fixed tissue samples. A comparative analysis of changes in the methylation degree of the promoter region of genes encoding components of the uPA system in tumor and normal cells is also used in diagnostics of cancer. The change (most often DNA hypermethylation) is found in tissues of various types of tumors. The advantage of this method is that DNA is more stable and can be isolated from paraffinized tissue samples prefixed in-formalin [166].

Various approaches aimed at creating drugs that interact with components of the uPA system have been used for the development of therapeutic agents with anticancer activity. Since the components of the uPA system are located outside the cell, they can serve as targets for various peptides and antibodies. Peptides or peptidomimetics as well as antibodies were used as uPA inhibitors [6–8, 11, 167]; low molecular weight antagonists of PAI-1 and antibodies were used to suppress PAI-1; in the case of uPAR low molecular weight peptides, monoclonal antibodies aimed at blocking the interaction of uPAR with uPA, as well as antisense RNA and target toxins were used. The data on the crystal structures of uPA, uPAR and PAI-1 can be used for development of drugs based on the structure of the inhibitors of these proteins.

CONCLUSIONS

The polyfunctional uPA system is essential for the tumor progression; it plays the key role in invasion, metastasis, and angiogenesis. Involvement of the uPA system in these processes is due to its proteolytic and regulatory functions. Proteolytic functions are mediated by the proteolytic cascade, which is triggered by a highly specific protease uPA and leads to formation of a polyfunctional protease, plasmin, and then MMPs, thus forming the proteolytic potential for destruction of CTM and basement membranes, triggering regulatory functions through activation and release from CTM a number of biologically active molecules involved in the regulation of the main processes of carcinogenesis. The uPA system, regardless of its proteolytic functions, contributes to tumor progression through uPAR, PAI-1 and PAI-2, which are involved not only in regulating the uPA/uPAR activity, but also in proliferation, apoptosis, chemotaxis, adhesion, migration, and activation of the pathways of the epithelial-mesenchymal transition considered as one of the main pathways of the metastatic process. All the above processes are aimed at the regulation of invasion, metastasis, and angiogenesis. Data on altered expression of the uPA system components in malignant tumors suggest their use as informative diagnostic, prognostic and therapeutic targets. Using the uPA system components as diagnostic markers, it is necessary to take into consideration that at certain concentrations, PAI-1 can act both as an inhibitor and as an activator of tumor progression.

FUNDING

This work was performed within the framework of the Program for Basic Research of State Academies of Sciences for 2013–2020.

COMPLIANCE WITH ETHICAL STANDARDS

The authors declare that they have no conflict of interest. This article does not contain any studies involving animals or human participants performed by any of the authors.

REFERENCES

1. Theocharis, A.D., Skandalis, S.S., Gialeli, C., and Karamanos, N.K., *Adv. Drug Delivery Revs.*, 2016, vol. 97, pp. 4–27. doi 10.1016/j.addr.2015.11.001
2. Benjamin, M.M. and Khalil, R.A., *EXS*, 2012, vol. 103, pp. 209–279. doi 10.1007/978-3-0348-0364-9_7
3. Shay, G., Lynch, C.C., and Fingleton, B., *Matrix Biol.*, 2015, vols. 44–46, pp. 200–206. doi 10.1016/j.matbio.2015.01.019
4. Amar, S., Smith, L., and Fields, G.B., *Biochim. Biophys. Acta*, 2017, vol. 1864 (11PtA), pp. 1940–1951. doi 10.1016/j.bbamcr.2017.04.2015

5. Cui, N., Hu, M., and Khalil, R.A., *Prog. Mol. Biol. Transl. Sci.*, 2017, vol. 147, pp. 1–73. doi 10.1016/bs.pmbts.2017.02.005
6. Mahmood, N., Mihalcioiu, C., and Rabbani, S.A., *Front. Oncol.*, 2018, vol. 8, 24. doi 10.3389/fonc.2018.00024
7. Jaiswal, R.K., Varshney, A.K., and Yadava, P.K., *Biomed. Pharmacother.*, 2018, vol. 98, pp. 886–898. doi 10.1016/j.biopha.2018.01.029
8. Brungs, D., Chen, J., Aghmesheh, M., Vine, K.L., Becker, T.M., Carolan, M.G., and Ranson, M., *Oncotarget*, 2017, vol. 8, pp. 23099–23109. doi 10.18632/oncotarget.15485
9. McMahon, B.J. and Kwaan, H.C., *Adv. Exp. Med. Biol.*, 2015, vol. 867, pp. 145–156. doi 10.1007/978-94-017-7215-0_10
10. Mekkawy, A.H., Pourgholami, M.H., and Morris, D.L., *Med. Res. Rev.*, 2014, vol. 34, pp. 918–956. doi 10.1002/med.21308
11. Eatemadi, A., Aiyelabegan, H.T., Negahdari, B., Mazlomi, M.A., Daraee, H., Daraee, N., Eatemadi, R., and Sadroddiny, E., *Biomed. Pharmacother.*, 2017, vol. 86, pp. 221–231. doi 10.1016/j.biopha.2016.12.021
12. Harris, N., Vennin, C., Conway, J., Vine, K.L., Pinese, M., and Cowley, M., *Oncogene*, 2017, vol. 36, no. 30, pp. 4288–4298. doi 10.1038/onc.2017.63
13. Sato, M., Kawana, K., Adachi, K., Fujimoto, A., Yoshida, M., Nakamura, H., Nishida, H., Inoue, T., Taguchi, A., Takahashi, J., Kojima, S., Yamashita, A., Tomio, K., Nagamatsu, T., Wada-Hiraike, O., Oda, K., Osuga, Y., and Fujii, T., *Int. J. Oncol.*, 2016, vol. 48, no. 2, pp. 829–835. doi 10.3892/ijo.2015.3283
14. Ellis, V., in *Handbook of Proteolytic Enzymes,* Rawlings, N.D. and Salvesen, G., Eds., Academic Press, 2013, vol. 3, pp. 2938–2945.
15. Noh, H., Hong, S., and Huang, S., *Theranostics*, 2013, vol. 3, pp. 487–495. doi 10.7150/thno.4218
16. Tkachuk, V.A., Plakhanova, O.S., Beloglazova, I.B., and Parfenova, E.V., *Ukr. Biokhim. Zh.*, 2013, vol. 85, pp. 18–45.
17. Gonias, S.L. and Hu, J., *Front. Pharmacol.*, 2015, vol. 6, 154. doi 10.3389/fphar.2015.00154
18. Santibanez, J.F., Obradović, H., Kukolj, T., and Krstić, J., *Dev. Dyn.*, 2018, vol. 247, no. 3, pp. 382–395. doi 10.1002/dvdy.24554
19. Ulisse, S., Baldini, E., Sorrenti, S., and D'Armiento, M., *Curr. Cancer Drug Targets*, 2009, vol. 9, pp. 32–71. doi 10.2174/156800909787314002
20. Jo, M., Lester, R.D., Montel, V., Eastman, B., Takimoto, S., and Gonias, S.L., *J. Biol. Chem.*, 2009, vol. 284, pp. 22825–22833. doi 10.1074/jbc.M109.023960
21. Gilder, A.S., Natali, L., Van Dyk, D.M., Zalfa, C., Banki, M.A., Pizzo, D.P., Wang, H., Klemke, R.L., Mantuano, E., and Gonias, S.L., *Sci. Rep.*, 2018, vol. 8, 2982. doi 10.1038/s41598-018-21358-1
22. Vandooren, J., Opdenakker, G., Loadman, P.M., and Edwards, D.R., *Adv. Drug. Deliv. Rev.*, 2016, vol. 97, pp. 144–155. doi 10.1016/j.addr.2015.12.020
23. D'Alessio, S. and Blasi, F., *Front. Biosci.* (Landmark Ed.), 2009, vol. 14, pp. 4575–4587.
24. Ferraris, G.M. and Sidenius, N., *Semin. Thromb. Hemost.*, 2013, vol. 39, no. 4, pp. 347–455. doi 10.1055/s-0033-1334485
25. Minchenko, A.G., Khar'kovskaya, A.P., Kubaichuk, K.I., Minchenko, D.O., Glushchak, N.A., and Kovalevskaya, O.V., *Ukr. Biokhim. Zh.*, 2014, vol. 86, no. 4, pp. 79–89.
26. Miles, L.A. and Parmer, R.J., *Semin. Thromb. Hemost.*, 2013, vol. 39, no. 4, pp. 329–337. doi 10.1055/s-0033-1334483
27. Chandran, V.I., Eppenberger-Castori, S., Venkatesh, T., Vine, K.L., and Ranson, M., *Oncoscience*, 2015, vol. 2, no. 3, pp. 207–224. doi 10.18632/ oncoscience
28. Høyer-Hansen, G., Behrendt, N., Ploug, M., Danø, K., and Preissner, K.T., *FEBS Letts.*, 1997, vol. 420, pp. 79–85. doi 10.1016/S0014-5793(97)01491-9
29. Higazi, A., Cohen, R.L., Henkin, J., Kniss, D., Schwartz, B.S., and Cines, D.B., *J. Biol. Chem.*, 1995, vol. 270, pp. 17375–17380. doi 10.1074/jbc.270.29.17375
30. Wei, Y., Lukashev, M., Simon, D.I., Bodary, S.C., Rosenberg, S., Doyle, M.V., and Chapman, H.A., *Science*, 1996, vol. 273, pp. 1551–1555. doi 10.1126/science.273.5281.1551
31. Jo, M., Thomas, K.S., O'Donnell, D.M., and Gonias, S.L., *J. Biol. Chem.*, 2003, vol. 278, no. 3, pp. 1642–1646. doi 10.1074/jbc.M210877200
32. Montuori, N., Visconte, V., Rossi, G., and Ragno, P., *Thromb. Haemost.*, 2005, vol. 93, no. 2, pp. 192–198. doi 10.1160/TH04-09-0580
33. Stefansson, S. and Lawrence, D.A., *Nature*, 1996, vol. 383, no. 6599, pp. 441–443. doi .10.1038/383441a0
34. Croucher, D.R., Saunders, D.N., Lobov, S., and Ranson, M., *Nat. Rev. Cancer*, 2008, vol. 8, no. 7, pp. 535–545. doi 10.1038/nrc2400
35. Harbeck, N., Schmitt, M., Meisner, C., Friedel, C., Untch, M., Schmidt, M., Sweep, C.G., Lisboa, B.W., Lux, M.P., Beck, T., Hasmüller, S., Kiechle, M., Jänicke, F., and Thomssen, C., *Eur. J. Cancer*, 2013, vol. 49, no. 8, pp. 1825–1835. doi 10.1016/j.ejca.2013.01.007
36. Halamkova, J., Kiss, I., Pavlovsky, Z., Jarkovsky, J., Tomasek, J., Tucek, S., Hanakova, L., Moulis, M., Cech, Z., Zavrelova, J., and Penka, M., *Hepatogastroenterology*, 2011, vol. 58, no. 112, pp. 1918–1925. doi 10.5754/hge10232
37. Lee, C.C. and Huang, T.S., *J. Cancer. Mol.*, 2005, vol. 1, no. 1, pp. 25–36. doi 10.29685/JCM.200510.0003
38. Binder, B.R., *Fibrinolysis*, 1995, vol. 9, suppl. 1, pp. 3–8. doi 10.1016/S0268-9499(05)80002-5
39. Crippa, M.P., *Int. J. Biochem. Cell Biol.*, 2007, vol. 39, no. 4, pp. 690–694. doi 10.1016/j.biocel.2006.10.008
40. Nagamine, Y., Medcalf, R.L., and Muñoz-Cánoves, P., *Thromb. Haemost.*, 2005, vol. 93, no. 4, pp. 661–675. doi 10.1160/TH04-12-0814

41. Behrens, M.A., Botkjaer, K.A., Goswami, S., Oliveira, C.L., Jensen, J.K., Schar, C.R., Declerck, P.J., Peterson, C.B., Andreasen, P.A., and Pedersen, J.S., *J. Mol. Biol.*, 2011, vol. 411, no. 2, pp. 417–429. doi 10.1016/j.jmb.2011.05.026

42. Duffy, M.J., *Curr. Pharm. Des.*, 2004, vol. 10, pp. 39–49. doi 10.2174/1381612043453559

43. Ashour, A.A., Gurbuz, N., Alpay, S.N., Abdel-Aziz, A.A., Mansour, A.M., Huo, L., and Ozpolat, B., *J. Cell. Mol. Med.*, 2014, vol. 18, no. 11, pp. 2235–2251. doi 10.1111/jcmm.12361

44. Eden, G., Archinti, M., Arnaudova, R., Andreotti, G., Motta, A., Furlan, F., Citro, V., Cubellis, M.V., and Degryse, B., *Cell. Mol. Life Sci.*, 2018, vol. 75, no. 10, pp. 1889–1907. doi 10.1007/s00018-017-2718-3

45. Liu, D., Aguirre Ghiso, J., Estrada, Y., and Ossowski, L., *Cancer Cell*, 2002, vol. 1, no. 5, pp. 445–457. doi 10.1016/S1535-6108(02)00072-7

46. Ploug, M., Eriksen, J., Plesner, T., Hansen, N.E., and Danø, K., *Eur. J. Biochem.*, 1992, vol. 208, pp. 397–404. doi 10.1111/j.1432-1033.1992.tb17200.x

47. Pedersen, N., Schmitt, M., Rønne, E., Nicoletti, M.I., Høyer-Hansen, G., Conese, M., Giavazzi, R., Dano, K., Kuhn, W., and Jänicke, F., *J. Clin. Invest.*, 1993, vol. 92, pp. 2160–2167. doi 10.1172/JCI116817

48. Brünner, N., Nielsen, H.J., Hamers, M., Christensen, I.J., Thorlacius-Ussing, O., and Stephens, R.W., *APMIS*, 1999, vol. 107, pp. 160–167. doi 10.1111/j.1699-0463.1999.tb01539.x

49. Rigolin, G.M., Tieghi, A., Ciccone, M., Bragotti, L.Z., Cavazzini, F., Della Porta, M., Castagnari, B., Carroccia, R., Guerra, G., Cuneo, A., and Castoldi, G., *Br. J. Haematol.*, 2003, vol. 120, pp. 953–959. doi 10.1046/j.1365-2141.2003.04176.x

50. Stephens, R.W., Nielsen, H.J., Christensen, I.J., Thorlacius-Ussing, O., Sørensen, S., Danø, K., and Brünner, N., *J. Natl. Cancer Inst.*, 1999, vol. 91, pp. 869–874.

51. Pappot, H., Høyer-Hansen, G., Rønne, E., Hansen, H.H., Brünner, N., Danø, K., and Grøndahl-Hansen, J., *Eur. J. Cancer*, 1997, vol. 33, pp. 867–872. doi 10.1016/S0959-8049(96)00523-0

52. Krüger, A., Soeltl, R., Lutz, V., Wilhelm, O.G., Magdolen, V., Rojo, E.E., Hantzopoulos, P.A., Graeff, H., Gänsbacher, B., and Schmitt, M., *Cancer Gene Ther.*, 2000, vol. 7, pp. 292–299. doi 10.1038/sj.cgt.7700144

53. Lutz, V., Reuning, U., Krüger, A., Luther, T., von Steinburg, S.P., Graeff, H., Schmitt, M., Wilhelm, O.G., and Magdolen, V., *Biol. Chem.*, 2001, vol. 382, pp. 789–798. doi 10.1515/BC.2001.095

54. Lijnen, H.R., *J. Thromb. Haemost.*, 2005, vol. 3, no. 1, pp. 35–45. doi 10.1111/j.1538-7836.2004.00827.x

55. Croucher, D.R., Saunders, D.N., Lobov, S., and Ranson, M., *Nat. Rev. Cancer*, 2008, vol. 8, no. 7, p. 535. doi 10.1038/nrc2400

56. Halamkova, J., Kiss, I., Pavlovsky, Z., Jarkovsky, J., Tomasek, J., Tucek, S., Hanakova, L., Moulis, M., Cech, Z., Zavrelova, J., and Penka, M., *Hepatogastroenterology*, 2011, vol. 58, no. 112, pp. 1918–1925. doi 10.5754/hge10232

57. Su, S.C., Lin, C.W., Yang, W.E., Fan, W.L., and Yang, S.F., *Expert Opin. Ther. Targets*, 2016, vol. 20, no. 5, pp. 551–566. doi 10.1517/14728222.2016.1113260

58. Placencio, V.R. and DeClerck, Y.A., *Cancer Res.*, 2015, vol. 75, pp. 2969–2974. doi 10.1158/0008-5472.CAN-15-0876

59. Kruithof, E.K., Baker, M.S., and Bunn, C.L., *Blood*, 1995, vol. 86, no. 11, pp. 4007–4024.

60. Ellis, V., Wun, T.C., Behrendt, N., Ronne, E., and Danø, K., *J. Biol. Chem.*, 1990, vol. 265, no. 17, pp. 9904–9908.

61. Kumar, S. and Baglioni, C., *J. Biol. Chem.*, 1991, vol. 266, no. 31, pp. 20960–20964.

62. Dickinson, J.L., Bates, E.J., Ferrante, A., and Antalis, T.M., *J. Biol. Chem.*, 1995, vol. 270, no. 46, pp. 27894–27904. doi 10.1074/jbc.270.46.27894

63. Cerami, E., Gao, J., Dogrusoz, U., Gross, B.E., Sumer, S.O., Aksoy, B.A., Jacobsen, A., Byrne, C.J., Heuer, M.L., Larsson, E., Antipin, Y., Reva, B., Goldberg, A.P., Sander, C., and Schultz, N., *Cancer Discov.*, 2012, vol. 2, pp. 401–404. doi 10.1158/2159-8290.CD-12-0095

64. Gao, J., Aksoy, B.A., Dogrusoz, U., Dresdner, G., Gross, B., Sumer, S.O., Sun, Y., Jacobsen, A., Sinha, R., Larsson, E., Cerami, E., Sander, C., and Schultz, N., *Sci. Signal.*, 2013, vol. 6, no. 269, pl1. doi 10.1126/scisignal.2004088

65. Aisina, R.B. and Mukhametova, L.I., *Bioorgan. Khim.*, 2014, vol. 40, pp. 642–657. doi 10.7868/S0132342314060025

66. Ogiwara, K., Nogami, K., Nishiya, K., and Shima, M., *Blood Coagul. Fibrinolysis*, 2010, vol. 21, pp. 568–576. doi 10.1097/MBC.0b013e32833c9a9f

67. Hannocks, M.J., Oliver, L., Gabrilove, J.L., and Wilson, E.L., *Blood*, 1992, vol. 79, no. 5, pp. 1178–1184.

68. Mukhina, S., Stepanova, V., Traktouev, D., Poliakov, A., Beabealashvilly, R., Gursky, Y., Minashkin, M., Shevelev, A., and Tkachuk, V., *J. Biol. Chem.*, 2000, vol. 275, no. 22, pp. 16450–16458. doi 10.1074/jbc.M909080199

69. Dvorak, H.F., *J. Thromb. Haemost.*, 2005, vol. 3, pp. 1835–1842. doi 10.1111/j.1538-7836.2005.01361.x

70. Plesner, T., Behrendt, N., and Ploug, M., *Stem Cells*, 1997, vol. 15, no. 6, pp. 398–408. doi 10.1002/stem.150398

71. Blasi, F., *Bioessays*, 1993, vol. 15, no. 2, pp. 105–111. doi 10.1002/bies.950150206

72. Nykjaer, A., Møller, B., Todd, R., Christensen, T., Andreasen, P.A., Gliemann, J., and Petersen, C.M., *J. Immunol.*, 1994, vol. 152, no. 2, pp. 505–516.

73. Gyetko, M.R., Sud, S., and Chensue, S.W., *Infect. Immun.*, 2004, vol. 72, no. 1, pp. 461–467. doi 10.1128/IAI.72.1.461-467.2004

74. Gyetko, M.R., Sud, S., Chen, G.H., Fuller, J.A., Chensue, S.W., and Toews, G.B., *J. Immunol.*, 2002, vol. 168, no. 2, pp. 801–809. doi 10.4049/jimmunol.168.2.801

75. Gyetko, M.R., Todd, R.F., 3rd, Wilkinson, C.C., and Sitrin, R.G., *J. Clin. Invest.*, 1994, vol. 93. no. 4, p. 1380. doi 10.1172/JCI117114

76. Estreicher, A., Mühlhauser, J., Carpentier, J.L., Orci, L., and Vassalli, J.D., *J. Cell Biol.*, 1990, vol. 111, no. 2, pp. 783–792. doi 10.1083/jcb.111.2.783

77. Zheng, P., Zou, R., and Liu, Y., *Sheng Li Xue Bao.*, 2001, vol. 53, no. 1, pp. 45–50.

78. Qin, Y., Han, Y., Xiong, C.L., Li, H.G., Hu, L., and Zhang, L., *Asian J. Androl.*, 2015, vol. 17, no. 2, pp. 269–273. doi 10.4103/1008-682X.143316

79. Smith, H.W., and Marshall, C.J., *Nat. Rev. Mol. Cell Biol.*, 2010, vol. 11, pp. 23–36. doi 10.1038/nrm2821

80. Tang, C.H. and Wei, Y., *Cell. Mol. Life Sci.*, 2008, vol. 65, no. 12, pp. 1916–1932. doi 10.1007/s00018-008-7573-9

81. Montuori, N., Pesapane, A., Rossi, F.W., Giudice, V., De Paulis, A., Selleri, C., and Ragno, P., *Transl. Med. UniSa.*, 2016, vol. 15, pp. 15–21.

82. Bass, R., Werner, F., Odintsova, E., Sugiura, T., Berditchevski, F., and Ellis, V., *J. Biol. Chem.*, 2005, vol. 280, pp. 14811–14818. doi 10.1074/jbc.M414189200

83. Mazar, A.P., *Clin. Cancer. Res.*, 2008, vol. 14, no. 18, pp. 5649–5455. doi 10.1158/1078-0432.CCR-07-4863

84. Riisbro, R., Christensen, I.J., Piironen, T., Greenall, M., Larsen, B., Stephens, R.W., Han, C., Høyer-Hansen, G., Smith, K., Brünner, N., and Harris, A.L., *Clin. Cancer Res.*, 2002, vol. 8, pp. 1132–1141.

85. Stepanova, V., Lebedeva, T., Kuo, A., Yarovoi, S., Tkachuk, S., Zaitsev, S., Bdeir, K., Dumler, I., Marks, M.S., Parfyonova, Y., Tkachuk, V.A., Higazi, A.A., and Cines, D.B., *Blood*, 2008, vol. 112, no. 1, pp. 100–110. doi 10.1182/blood-2007-07-104455

86. Gandhari, M., Arens, N., Majety, M., Dorn-Beineke, A., and Hildenbrand, R., *Int. J. Oncol.*, 2006, vol. 28, no. 6, pp. 1463–1470. doi 10.3892/ijo.28.6.1463

87. Jo, M., Thomas, K.S., Takimoto, S., Gaultier, A., Hsieh, E.H., Lester, R.D., and Gonias, S.L., *Oncogene*, 2007, vol. 26, no. 18, pp. 2585–2594. doi 10.1038/sj.onc.1210066

88. Milanese, G., Dellabella, M., Fazioli, F., Pierpaoli, E., Polito, M., Siednius, N., Montironi, R., Blasi, F., and Muzzonigro, G., *J. Urol.*, 2009, vol. 181, no. 3, pp. 1393–1400. doi 10.1016/j.juro.2008.10.147

89. Chabot, V., Dromard, C., Rico, A., Langonné, A., Gaillard, J., Guilloton, F., Casteilla, L., and Sensebé, L., *Stem Cell Res. Ther.*, 2015, vol. 6, p. 188. doi 10.1186/s13287-015-0163-5

90. De Lorenzi, V., Sarra Ferraris, G.M., Madsen, J.B., Lupia, M., Andreasen, P.A., and Sidenius, N., *EMBO Rep.*, 2016, vol. 17, no. 7, pp. 982–998. doi 10.15252/embr.201541681

91. Ahn, S.B., Mohamedali, A., Anand, S., Cheruku, H.R., Birch, D., Sowmya, G., Cantor, D., Ranganathan, S., Inglis, D.W., Frank, R., Agrez, M., Nice, E.C., and Baker, M.S., *J. Proteome Res.*, 2014, vol. 13, no. 12, pp. 5956–5964. doi 10.1021/pr500849x

92. Kenny, H.A., Leonhardt, P., Ladanyi, A., Yamada, S.D., Montag, A., Im, H.K., Jagadeeswaran, S., Shaw, D.E., Mazar, A.P., and Lengyel, E., *Clin. Cancer Res.*, 2011, vol. 17, no. 3, pp. 459–471. doi 10.1158/1078-0432.CCR-10-2258

93. Chen, S.C., Henry, D.O., Reczek, P.R., and Wong, M.K., *Mol. Cancer. Ther.*, 2008, vol. 7, no. 5, pp. 1227–1236. doi 10.1158/1535-7163.MCT-08-0051

94. Balsara, R.D. and Ploplis, V.A., *Thromb. Haemost.*, 2008, vol. 100, no. 6, pp. 1029–1036.

95. Chen, Y., Kelm, R.J., Budd, R.C., Sobel, B.E., and Schneider, D.J., *J. Cell. Biochem.*, 2004, vol. 92, no. 1, pp. 178–188. doi 10.1002/jcb.20058

96. Kalluri, R. and Weinberg, R.A., *J. Clin. Invest.*, 2009, vol. 119, no. 6, pp. 1420–1428. doi 10.1172/JCI39104

97. Kalluri, R. and Neilson, E.G., *J. Clin. Invest.*, 2003, vol. 112, no. 12, pp. 1776–1784. doi 10.1172/JCI200320530

98. Thompson, E.W., Newgreen, D.F., and Tarin, D., *Cancer Res.*, 2005, vol. 65, pp. 5991–5995.

99. van Zijl, F., Krupitza, G., and Mikulits, W., *Mutat. Res.*, 2011, vol. 728, nos. 1–2, pp. 23–34. doi 10.1016/j.mrrev.2011.05.002

100. Sanz-Moreno, V., Gadea, G., Ahn, J., Paterson, H., Marra, P., Pinner, S., Sahai, E., and Marshall, C.J., *Cell*, 2008, vol. 135, no. 3, pp. 510–523. doi 10.1016/j.cell.2008.09.043

101. Carragher, N.O., Walker, S.M., Scott Carragher, L.A., Harris, F., Sawyer, T.K., Brunton, V.G., Ozanne, B.W., and Frame, M.C., *Oncogene*, 2006, vol. 25, no. 42, pp. 5726–5740. doi 10.1038/sj.onc.1209582

102. Yamazaki, D., Kurisu, S., and Takenawa, T., *Oncogene*, 2009, vol. 28, no. 13, pp. 1570–1583. doi 10.1038/onc.2009.2

103. Madsen, C.D., Ferraris, G.M., Andolfo, A., Cunningham, O., and Sidenius, N., *J. Cell Biol.*, 2007, vol. 177, no. 5, pp. 927–939. doi 10.1083/jcb.200612058

104. Zhang, F., Tom, C.C., Kugler, M.C., Ching, T.T., Kreidberg, J.A., Wei, Y., and Chapman, H.A., *J. Cell Biol.*, 2003, vol. 163, pp. 177–188. doi 10.1083/jcb.200304065

105. Lester, R.D., Jo, M., Montel, V., Takimoto, S., and Gonias, S.L., *J. Cell Biol.*, 2007, vol. 178, pp. 425–436. doi 10.1083/jcb.200701092

106. Nyberg, P., Salo, T., and Kalluri R., *Front. Biosci.*, 2008, vol. 13, pp. 6537–6553.

107. Su, S.C., Lin, C.W., Yang, W.E., Fan, W.L., and Yang, S.F., *Expert Opin. Thera. Targets*, 2016, vol. 20, pp. 551–566. doi 10.1517/14728222.2016.1113260

108. Herkenne, S., Paques, C., Nivelles, O., Lion, M., Bajou, K., Pollenus, T., Fontaine, M., Carmeliet, P., Martial, J.A., Nguyen, N.Q., and Struman, I., *Sci. Signal.*, 2015, vol. 8, no. 403, ra117. doi 10.1126/scisignal.aaa2403

109. Breuss, J.M. and Uhrin, P., *Cell. Adh. Migr.,* 2012, vol. 6, no. 6, pp. 535–615. doi 10.4161/cam.22243

110. Poettler, M., Unseld, M., Mihaly-Bison, J., Uhrin, P., Koban, F., Binder, B.R., Zielinski, C.C., and Prager, G.W., *Thromb. Haemost.*, 2012, vol. 108, no. 2, pp. 357–366. doi 10.1160/TH11-12-0868

111. Kaneko, T., Konno, H., Baba, M., Tanaka, T., and Nakamura, S., *Cancer Sci.*, 2003, vol. 94, no. 1, pp. 43–49. doi 10.1111/j.1349-7006.2003.tb01350.x

112. Stepanova, V., Jayaraman, P.S., Zaitsev, S.V., Lebedeva, T., Bdeir, K., Kershaw, R., Holman, K.R.,

Parfyonova, Y.V., Semina, E.V., Beloglazova, I.B., Tkachuk, V.A., and Cines, D.B., *J. Biol. Chem*., 2016, vol. 291, no. 29, pp. 15029–15045. doi 10.1074/jbc.M115.678490

113. Bifulco, K., Longanesi-Cattani, I., Gala, M., Masucci, M.T., Pavone, V., Lista, L., Arra, C., Stoppelli, M.P., and Carriero, M.V., *J. Thromb. Haemost.*, 2010, vol. 8, no. 12, pp. 2789–2799. doi 10.1111/j.1538-7836.2010.04075.x
114. McMahon, G.A., Petitclerc, E., Stefansson, S., Smith, E., Wong, M.K., Westrick, R.J., Ginsburg, D., Brooks, P.C., and Lawrence, D.A., *J. Biol. Chem.*, 2001, vol. 276, no. 36, pp. 33964–33968. doi 10.1074/jbc.M105980200
115. Bajou, K., Noel, A., Gerard, R.D., Masson, V., Brunner, N., Holst-Hansen, C., Skobe, M., Fusenig, N.E., Carmeliet, P., Collen, D., and Foidart, J.M., *Nat. Med.*, 1998, vol. 4, no. 8, pp. 923–928. doi 10.1038/nm0898-923
116. Bajou, K., Maillard, C., Jost, M., Lijnen, R.H., Gils, A., Declerck, P., Carmeliet, P., Foidart, J.M., and Noel, A., *Oncogene*, 2004, vol. 23, no. 41, pp. 6986–6990. doi 10.1038/sj.onc.1207859
117. Mauro, C.D., Pesapane, A., Formisano, L., Rosa, R., D'Amato, V., Ciciola, P., Servetto, A., Marciano, R., Orsini, R.C., Monteleone, F., Zambrano, N., Fontanini, G., Servadio, A., Pignataro, G., Grumetto, L., Lavecchia, A., Bruzzese, D., Iaccarino, A., Troncone, G., Veneziani, B.M., Montuori, N., Placido, S., and Bianco, R., *Sci. Rep.*, 2017, vol. 7, no. 1, 9388. doi 10.1038/s41598-017-10062-1
118. Santi, A.L., Gorrasi, A., Alfieri, M., Montuori, N., and Ragno, P., *Oncotarget*, 2018, vol. 9, no. 45, pp. 27823–27834. doi 10.18632/oncotarget.25597
119. Duffy, M.J., McGowan, P.M., Harbeck, N., Thomssen, C., and Schmitt, M., *Breast Cancer Res.*, 2014, vol. 16, no. 4, p. 428. doi 10.1186/s13058-014-0428-4
120. Zhang, W., Ling, D., Tan, J., Zhang, J., and Li, L., *Oncol. Rep.*, 2013, vol. 29, no. 2, pp. 637–645. doi 10.3892/or.2012.2148
121. Rabi, Z.A., Todorović-Raković, N., Vujasinović, T., Milovanović, J., and Nikolić-Vukosavljević, D., *Cancer Biomark.*, 2015, vol. 15, no. 6, pp. 745–754. doi 10.3233/CBM-150516
122. Li, S., Wei, X., He, J., Tian, X., Yuan, S., and Sun, L., *Biomed. Pharmacother.*, 2018, vol. 105, pp. 83–94. doi 10.1016/j.biopha.2018.05.119
123. Kwaan, H.C., Mazar, A.P., and McMahon, B., *J. Semin. Thromb. Hemost.*, 2013, vol. 39, no. 4, pp. 382–391. doi 10.1055/s-0033-1338127
124. Viala, M., Alexandre, M., Thezenas, S., Lamy, P.J., Maran-Gonzalez, A., Gutowski, M., Colombo, P.E., Romieu, G., Jacot, W., and Guiu, S., *Breast Cancer Res. Treat.*, 2017, vol. 165, no. 3, pp. 611–621. doi 10.1007/s10549-017-4373-7
125. Duffy, M.J., Walsh, S., McDermott, E.W., and Crown, J., *Adv. Clin. Chem.*, 2015, vol. 71, pp. 1–23. doi 10.1016/bs.acc.2015.05.001
126. Duffy, M.J., O'Donovan, N., McDermott, E., and Crown, J., *Breast,* 2016, vol. 29, pp. 192–201. doi 10.1016/j.breast.2016.07.009
127. Duffy, M.J., Harbeck, N., Nap, M., Molina, R., Nicolini, A., Senkus, E., and Cardoso, F., *Eur. J. Cancer*, 2017, vol. 75, pp. 284–298. doi 10.1016/j.ejca.2017.01.017
128. van Tilborg, A.A., Sweep, F.C., Geurts-Moespot, A.J., Wetzels, A.M., de Waal, R.M., Westphal, J.R., and Massuger, L.F., *J. Oncol.*, 2014, vol. 44, no. 4, pp. 1394–1400. doi 10.3892/ijo.2014.2303
129. van Dam, P.A., Coelho, A., and Rolfo, C., *Eur. J. Surg. Oncol.*, 2016, vol. 43, no. 2, pp. 252–257. doi 10.1016/j.ejso.2016.06.002
130. Dorn, J., Harbeck, N., Kates, R., Gkazepis, A., Scorilas, A., Soosaipillai, A., Diamandis, E., Kiechle, M., Schmalfeldt, B., and Schmitt, M., *Ann. Oncol.*, 2011, vol. 22, no. 4, pp. 877–883. doi 10.1093/annonc/mdq462
131. Hoffmann, G., Pollow, K., Weikel, W., Strittmatter, H.-J., Bach, J., Schaffrath, M., Knapstein, P., Melchert, F., and Pollow, B., *Clin. Chem. Lab. Med.*, 1999, vol. 37, no. 1, pp. 47–54. doi 10.1515/CCLM.1999.007
132. Ljuca, D., Fatusić, Z., Iljazović, E., and Ahmetović, B., *Bosn. J. Basic. Med. Sci.*, 2007, vol. 7, no. 2, pp. 111–116. doi 10.17305/bjbms.2007.3063
133. Timoshenko, O.S., Kugaevskaya, E.V., Gureeva, T.A., Zavalishina, L.E., Andreeva, Yu.Yu., and Solovyeva, N.I., *Arkhiv Patologii*, 2015, vol. 77, no. 5, pp. 31–35.
134. Solovyeva, N.I., Timoshenko, O.S., Gureeva, T.A., and Kugaevskaya, E.V., *Biomed. Khim.*, 2015, vol. 61, no. 6, pp. 694–704. doi 10.18097/PBMC20156106694
135. Gershtein, E.S., Gritsaenko, E.V., and Tereshkina, I.V., *Molekulyarnaya Meditsina*, 2013, vol. 6, pp. 27–32.
136. Memarzadeh, S., Kozak, K.R., Chang, L., Natarajan, S., Shintaku, P., Reddy, S.T., and Farias-Eisner, R., *Proc. Natl. Acad. Sci. USA*, 2002, vol. 99, no. 16, pp. 10647–10652. doi 10.1073/pnas.152127499
137. Steiner, E., Pollow, K., Hasenclever, D., Schormann, W., Hermes, M., Schmidt, M., Puhl, A., Brulport, M., Bauer, A., Petry, I.B., Koelbl, H., and Hengstler, J.G., *Gynecol. Oncol.*, 2008, vol. 108, no. 3, pp. 569–576. doi 10.1016/j.ygyno.2007.11.025
138. Dariusz, S., Agnieszka, M., Elzbieta, R., Danuta, O.-N., Maciej, Z., Piotr, D., and Nowicki, M., *Eur. J. Obstet. Gynecol. Reprod. Biol.*, 2012, vol. 163, no. 2, pp. 193–199. doi 10.1016/j.ejogrb.2012.03.031
139. Shariat, S.F., Roehrborn, C.G., McConnell, J.D., Park, S., Alam, N., Wheeler, T.M., and Slawin, KM., *J. Clin. Oncol.*, 2007, vol. 25, no. 4, pp. 349–355. doi 10.1200/JCO.2006.05.6853
140. Kumano, M., Miyake, H., Muramaki, M., Furukawa, J., Takenaka, A., and Fujisawa, M., *Urol. Oncol.*, 2009, vol. 27, no. 2, pp. 180–186. doi 10.1016/j.urolonc.2008.01.012
141. Kjellman, A., Akre, O., Gustafsson, O., Høyer-Hansen, G., Lilja, H., Norming, U., Piironen, T., and Törnblom, M., *J. Intern. Med.*, 2011, vol. 269, no. 3, pp. 299–305. doi 10.1111/j.1365-2796.2010.02284.x
142. Serafin, A.M., Akudugu, J.M., and Bohm, L., *Ann. Clin. Biochem.*, 2015, vol. 52, pt. 3, pp. 387–394. doi 10.1177/0004563214559546

143. Yang, J.L., Seetoo, D.Q., Wang, Y., Ranson, M., Berney, C.R., Ham, J.M., Russell, P.J., and Crowe, P.J., *Int. J. Cancer*, 2000, vol. 89, no. 5, pp. 431–439. doi 10.1002/1097-0215(20000920)89:5<431::AID-IJC6>3.0.CO;2-V

144. Halamkova, J., Kiss, I., Pavlovsky, Z., Tomasek, J., Jarkovsky, J., Cech, Z., Tucek, S., Hanakova, L., Moulis, M., Zavrelova, J., Man, M., Benda, P., Robek, O., Kala, Z., and Penka, M., *Neoplasma*, 2011, vol. 58, no. 5, pp. 377–385. doi 10.4149/neo_2013_020

145. Riisbro, R., Christensen, I.J., Nielsen, H.J., Brünner, N., Nilbert, M., and Fernebro, E., *Int. J. Biol. Markers*, 2005, vol. 20, no. 2, pp. 93–102. doi 10.5301/JBM.2008.5473

146. Gershtein, E.S., Korotkova, E.A., Kushlinskii, N.E., and Prorokov, V.V., *Klin. Lab. Diagn.*, 2013, no. 10, pp. 5–10.

147. Salden, M., Splinter, T., Peters, H., Look, M., Timmermans, M., van Meerbeeck, J., and Foekens, J.A., *Ann. Oncol.*, 2000, vol. 11, no. 3, pp. 327–332.

148. Su, C.Y., Liu, Y.P., Yang, C.J., Lin, Y.F., Chiou, J., Chi, L.H., Lee, J.J., Wu, A.T., Lu, P.J., Huang, M.S., and Hsiao, M., *PLoS One*, 2015, vol. 10, no. 7, e0133411. doi 10.1371/journal.pone.0133411

149. Langkilde, A., Hansen, T.W., Ladelund, S., Linneberg, A., Andersen, O., Haugaard, S.B., Jeppesen, J., and Eugen-Olsen, J., *Cancer Epidemiol. Biomarkers Prev.*, 2011, vol. 20, pp. 609–618. doi 10.1158/1055-9965.EPI-10-1009

150. Harris, N., Vennin, C., Conway, J., Vine, K.L., Pinese, M., Cowley, M., Shearer, R.F. Lucas, M.C., Herrmann, D., Allam, A.H., Pajic, M., Morton, J.P., Biankin, A.V., Ranson, M., Timpson, P., and Saunders, D.N., *Oncogene*, 2017, vol. 36, no. 30, pp. 4288–4298. doi 10.1038/onc.2017.63

151. Asuthkar, S., Stepanova, V., Lebedeva, T., Holterman, A.L., Estes, N., Cines, D.B., Rao, J.S., and Gondi, C.S., *Mol. Biol. Cell.*, 2013, vol. 24, no. 17, pp. 2620–2632. doi 10.1091/mbc.E12-04-0306

152. Beyer, B., Heiss, M.M., Simon, E.H., Gruetzner, K.U., Babic, R., Jauch, K.W., Schildberg, F.W., and Allgayer, H., *Cancer*, 2006, vol. 106, no. 5, pp. 1026–1035. doi 10.1002/cncr.21682

153. Luebke, T., Baldus, S., Spieker, D., Grass, G., Bollschweiler, E., Schneider, P., Thiele, J., Dienes, H.P., Hoelscher, A.H., and Moenig, S.P., *Int. J. Biol. Markers*, 2006, vol. 21, no. 3, pp. 162–169.

154. Ding, Y., Zhang, H., Lu, A., Zhou, Z., Zhong, M., Shen, D., Wang, X., and Zhu, Z., *Oncol. Letts.*, 2016, vol. 11, no. 6, pp. 4208–4216. doi 10.3892/ol.2016.4498

155. Kit, O.I., Frantsiyants, E.M., Kozlova, L.S., Kolesnikov, E.N., Pogorelova, Yu.A., Cheryarina, N.D., and Tavaryan, I.S., *Eksper. Klin. Gastroenterol.*, 2016, vol. 129, no. 5, pp. 16–21.

156. Illemann, M., Bird, N., Majeed, A., Laerum, O.D., Lund, L.R., Danø, K., and Nielsen, B.S., *Int. J. Cancer*, 2009, vol. 124, no. 8, pp. 1860–1870. doi 10.1002/ijc.24166

157. Pavón, M.A., Arroyo-Solera, I., Céspedes, M.V., Casanova, I., León, X., and Mangues, R., *Oncotarget*, 2016, vol. 7, no. 35, pp. 57351–57366. doi 10.18632/oncotarget.10344

158. Lee, Y.C., Yu, C.C., Lan, C., Lee, C.H., Lee, H.T., Kuo, Y.L., Wang, P.H., and Chang, W.W., *Head Neck*, 2016, vol. 38, no. 1, pp. 895–904. doi 10.1002/hed.24124

159. Magnussen, S.N., Hadler-Olsen, E., Costea, D.E., Berg, E., Jacobsen, C.C., Mortensen, B., Salo, T., Martinez-Zubiaurre, I., Winberg, J.O., Uhlin-Hansen, L., and Svineng, G., *BMC Cancer*, 2017, vol. 17, no. 1, p. 350. doi 10.1186/s12885-017-3349-7

160. Carriero, M.V. and Stoppelli, M.P., *Curr. Pharm. Des.*, 2011, vol. 17, pp. 1944–1961. doi 10.2174/138161211796718143

161. Lu, J.J., Guo, H., Gao, B., Zhang, Y., Lin, Q.L., Shi, J., Liu, J.J., and Liu, J., *Mol. Clin. Oncol.*, 2018, vol. 8, no. 1, pp. 127–132. doi 10.3892/mco.2017.1484

162. Nicolini, A., Ferrari, P., and Duffy, M.J., *Semin. Cancer Biol.*, 2017. pii: S1044-579X(17)30052-30054. doi 10.1016/j.semcancer.2017.08.010

163. Paik, S., Shak, S., Tang, G., Kim, C., Baker, J., Cronin, M., Baehner, F.L., Walker, M.G., Watson, D., Park, T., Hiller, W., Fisher, E.R., Wickerham, D.L., Bryant, J., and Wolmark, N., *N. Engl. J. Med.*, 2004, vol. 351, no. 27, pp. 2817–2826. doi 10.1056/NEJMoa041588

164. Wang, X., Jiang, Z., An, J., Mao, X., Lin, F., and Sun, P., *Mol. Med. Rep.*, 2018, vol. 17, no. 3, pp. 4273–4280. doi 10.3892/mmr.2018.8414

165. Al-Janabi, O., Taubert, H., Lohse-Fischer, A., Fröhner, M., Wach, S., Stöhr, R., Keck, B., Burger, M., Wieland, W., Erdmann, K., Wirth, M.P., Wullich, B., Baretton, G., Magdolen, V., Kotzsch, M., and Füssel, S., *Biomed. Res. Int.*, 2014, vol. 2014, 972587. doi 10.1155/2014/972587

166. Schmitt, M., Mengele, K., Gkazepis, A., Napieralski, R., Magdolen, V., Reuning, U., and Harbeck, N., *Breast Care*, 2008, vol. 3, pp. 3–10. doi 10.1159/000151737

167. Xu, X., Cai, Y., Wei, Y., Donate, F., Juarez, J., Parry, G., Chen, L., Meehan, E.J., Ahn, R.W., Ugolkov, A., Dubrovskyi, O., O'Halloran, T.V., Huang, M., and Mazar, A.P., *PLoS One*, 2014, vol. 9, no. 1, e85349. doi 10.1371/journal.pone.0085349

Translated by A. Medvedev