Review Article

A Cross-TALK OF TFPI, Protein S, Protein C, Antithrombin III Genes And Their Expression And Markers OF Thrombogenesis In Breast Cancer: A Review

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Abstract

Cancer-associated thrombosis is a major cause of mortality in cancer patients. Several risk factors for developing thrombogenesis also coexist with cancer patients, such as chemotherapy and immobilization, contributing to the increased risk of cancer patients developing thrombosis compared with non-cancer patients. Cancer cells are capable of activating the coagulation cascade and other prothrombotic properties of host cells, and many anticancer treatments themselves are being described as additional mechanisms for promoting thrombosis. There is a scarcity of literature which explores the pathogenesis of thrombogenesis and its association with the cancer progression in breast cancer patients to the best of our knowledge.

Overall aim is to discuss the cross-talk between TFPI, protein S, Protein C, anti-thrombin III genes and their expression in patients with breastcancer. We also aim to link the process of thrombogenesis and fibrinolysis activation in patients with breast cancer. As thrombogenesis and fibrinolysis in systemic circulation are interrelated with inflammation and endothelial function we also aim to measure markers of inflammation and endothelial activation. Ultimate goal is to find out whether the above-mentioned association could be an early predictor of the progression of breast malignancy.

Keywords: breast cancer, thrombosis, coagulation factors

INTRODUCTION

Cancer-associated thrombosis is a major cause of mortality in cancer patients. Several risk factors for developing thrombogenesis also coexist with cancer patients, such as chemotherapy and immobilization, contributing to the increased risk of cancer patients developing thrombosis compared with non-cancer patients. Cancer cells are capable of activating the coagulation cascade and other prothrombotic properties of host cells, and many anticancer treatments themselves are being described as additional mechanisms for promoting thrombosis.There is a scarcity of literature which explores the pathogenesis of thrombogenesis and its association with the cancer progression in breast cancer patients to the best of our knowledge.The cross-talk between TFPI, Proteins C and S and anti-thrombin III as well as their association with thrombogenesis profile may be early predictors of disease progression.

Thrombosis is a prevalent consequence for cancer patients, according to numerous studies, and it contributes to the second-leading cause of cancer-related death [1,2]. Multiple clinical variables, as well as biological procoagulant pathways generated by cancer tissues, all contribute to the activation of blood coagulation and, as a result, to the patients' total thrombotic risk [3,4]. Clinical risk factors include general and biological characteristics that are common to both cancer and non-cancer patients, but there are also a number of disease-specific clinical and biological factors that distinguish the pathogenesis of cancer-associated thrombosis in patients with malignancy.

These factors, taken together, support a shift in the hemostatic balance toward a prothrombotic state, as evidenced by the presence of subclinical coagulation abnormalities in almost all cancer patients, who have high levels of circulating hypercoagulability biomarkers. Even without obvious thrombosis, patients with solid tumors commonly present with subclinical prothrombotic or hypercoagulable conditions [5,6].

Arterial or venous thromboembolism to disseminated intravascular coagulation are all examples of thrombotic consequences in cancer [7,8]. Despite the well-established link between cancer and thromboembolic disorder, the mechanisms that enhance thromboembolic events in cancer patients are unknown and appear to be complex [9]. Patients with cancer are commonly hypercoagulable or prothrombotic, as they have anomalies in each component of Virchow's

triad, which contribute to thrombosis. Blood flow stasis, endothelial damage, and hypercoagulability are the three components, with the latter involving anomalies in the coagulation and fibrinolytic pathways, as well as platelet activation. The specific processes causing Virchow's triad anomalies in cancer patients, notably the effect on the host hemostatic system to increase the prothrombotic condition, remain unknown.

Laboratory studies that reveal abnormalities in the hemostatic system are common in cancer patients [10]. In cancer patients, activation of blood coagulation caused by cancer-dependent stimuli increases the risk of thromboembolic consequences [11,12]. The most frequent cancers in humans are colon and breast cancers. Hypercoagulability in cancer patients may be caused, at least in part, by coagulation inhibitors with insufficient efficacy.

Tissue factor pathway inhibitor (TFPI), a member of the Kunitz-type serine protease inhibitor superfamily, is one of the most important inhibitors of blood coagulation. Endothelial cells (ECs) of tiny blood arteries produce TFPI [13,14]. TFPI mRNA was found in the placenta, lung, liver, heart, kidney, skeletal muscles, pancreas, and brain, among other places [13,15].

Activated macrophages, monocytes, and fibroblasts produce small amounts of TFPI [14]. In humans, the majority of TFPI is anchored to the EC membrane by a glycosylphosphatidylinositol (GPI)-anchored protein, rather than by glycosaminoglycans (GAGs) [16,17]. It binds to plasma lipoproteins in a lesser proportion. It's also found in blood platelets and circulates freely in plasma [15,17]. Both coagulation factor Xa and the tissue factor/factor VIIa (TF/VIIa) complex are inhibited by TFPI [18].

TFPI regulates TF activity by a two-step feedback mechanism that starts with the creation of a bimolecular FXa/TFPI complex, which then interacts with TF/FVIIa, resulting in an inactive quaternary complex and the end of TF/FVIIa-catalyzed FX activation[19,20]. Because of an intact C-terminal basic tail that interacts with anionic membrane surfaces, full-length TFPI is most effective in this reaction[21,22]. The development of a binary TFPI-FXa complex is required for the suppression of TF/FVIIa by TFPI in this negative feedback pathway[19].

Patients with various neoplasms have higher plasma concentrations of TFPI than healthy participants [23-25], especially at an advanced stage of the disease. Heparin treatment causes a larger release of TFPI in cancer patients than in non-cancer participants [23]. Furthermore, in compared to healthy people, patients with solid tumours had a higher amount of factor Xa/TFPI complex in their plasma [24]. A high plasma TFPI concentration could indicate a host compensating mechanism in cancer patients with a hypercoagulable state. The normalisation of TFPI plasma levels is typical after anticancer treatment (surgery, radiation, or chemotherapy) [25,26].

Blood coagulation is activated not only intravascularly, but also extra-vascularly at the tumor site in cancer patients [27]. In colon and breast cancer tissues, components of the TF-dependent blood coagulation pathway (e.g., TF, factor VII, factor X) were found [28]. The aforementioned factors' activity is thought to play a role in the control of apoptosis, cell migration, angiogenesis, and metastasis development [29,30]. Furthermore, activation of blood coagulation results in the production of thrombin and fibrin, both of which aid tumor growth through a variety of interconnected processes [27,31].The data on TFPI expression in tumor tissue in loco is inconsistent.

Werling et al. found no TFPI in colon, breast, and renal cancer cells, non-small cell lung cancer tissue, or lymphoma tissue [32]. The authors, however, failed to mention a number of cases in which the study was conducted [32]. TFPI mRNA and protein, on the other hand, were found in colon, breast, and pancreatic cancer cell lines [33].

Low TFPI concentration, as well as high TF mRNA, protein expression, and activity, were found in colon cancer cell lines with a high potential for metastatic propagation to the liver in in vitro investigations [34]. However, in a metastatic colon cancer cell line, increased TF expression was observed alongside normal TFPI expression [34].

Since its presence was confirmed in 70% of colon cancer cases and 100% of breast cancer instances, it is possible that TFPI activity may, at least in part, offset TF and Xa biological activities. TFPI has little effect on cancer cell proliferation [35], but it's plausible that the inhibitor stimulates cancer growth locally. The findings of a study indicating ECM-bound TFPI via an interaction with the TF/VIIa complex found on cancer cells increased tumor cell adhesion and migration are particularly intriguing [36].

The procoagulant function of TF is regulated by TFPI in a highly aggressive melanoma, and this activity is required for perfusion of vasculogenic mimicry channels generated by TF-expressing melanoma cells, which play a key role in providing blood to the growing tumor [37].

TFPI was also found in the ECs of tiny blood arteries feeding neoplasms, as well as in TAMs. This type of protein localization has been documented before [32] and could be a counterbalance mechanism in response to the activation of coagulation in malignant tissue [27]. Furthermore, the presence of TFPI in ECs is unsurprising, given that ECs of tiny blood arteries are the primary source of TFPI production [14].

It's worth noting that good tumor growth requires robust angiogenesis, and newly formed blood vessels only have EC linings [38]. As a result, TFPI may either reduce angiogenesis indirectly by inhibiting TF proangiogenic activity or directly by inhibiting EC motility and creation of capillary-like structures by the ECs [39]. TFPI production has also been observed in activated macrophages [32].

Given that TAMs provide all of the components needed to build a full coagulation pathway, the presence of TFPI could indicate a reaction to activated blood coagulation in the proximity of TAMs. Plasma TFPI, interestingly, has been shown to have an antimetastatic impact in investigations [35,40]. When cancer cells are circulating in the bloodstream during the early stages of metastatic dissemination, TFPI has the most powerful effect. TFPI may help prevent TF-induced metastasis [41]. Through multiple methods, tumor cell TF-mediated thrombin production and tumor cell-induced platelet activation and aggregation (TCIPA) contribute to metastatic dissemination [42,43]. The neutralization of TF/VIIa or TF/VIIa/Xa complex activity is one of the mechanisms of TFPI anti-metastatic action [36].

Protein S is a vitamin K-dependent plasma protein (Mr75 kDa) that is synthesized in the liver and in endothelial cells and which circulates in plasma both in a free form (150 nmol/L) and in complex with C4b-binding protein (200 nmol/L;). It is well known that protein S acts as a cofactor of activated protein C (APC) in the proteolytic inactivation of blood coagulation factors Va and VIIIa, thus providing a negative feedback on coagulation and making the APC/protein S pathway essential for normal hemostasis[44,45].Homozygous deficiencies in either protein C or protein S result in severe neonatal procoagulant phenotypes, whereas heterozygous deficiencies are associated with a 10-fold increased risk of venous thrombosis[46].

It was reported that protein S acts as cofactor of TFPI11It was reported that C-terminally truncated TFPI was a weak inhibitor of factor Xa. In addition, truncated TFPI (1–161) was not stimulated by protein S [47]. This strongly suggests that the interaction between protein S and TFPI is mediated through TFPI Kunitz-3 and the C-terminal tail. For both protein S and full-length TFPI activity it is crucial that they can bind to negatively charged phospholipids through their Gla domain and C-terminal tail, respectively [21,48,49]. In addition, FXa protein S interactions have been reported [50,51]. Together with the observation that the C terminus of TFPI directly interacts with the Gla domain of FXa [52]. This implies that during inhibition of FXa by TFPI/protein S at a certain moment a trimolecular complex between FXa, TFPI, and protein S on phospholipids will exist.

Protein S (PS; encoded by the PROS1 gene), a key vitamin K-dependent anticoagulant protein, is emerging as a key structural and functional protein that is overexpressed in various malignancies, but how PROS1 gene and its cross talk with TFPI, Protein C and antithrombin III to promote breast cancer progression is unclear.

Activated protein C (APC) is a serine protease that inhibits blood coagulation by degrading the procoagulant cofactors Va and VIIIa [53]. After thrombin forms a compound with cell surface bound thrombomodulin, APC is formed by proteolytic cleavage of zymogen protein C. (TM). Protein C activation is enhanced by the endothelial cell protein C receptor, which sequesters it from circulation and presents it to the thrombin–TM complex. The importance of APC as a natural anticoagulant is demonstrated by the fact that heterozygous protein C deficiency increases the risk of thrombophlebitis, deep vein thrombosis, and pulmonary embolism [54-56], whereas homozygous protein C deficiency causes fatal massive disseminated intravascular coagulation (DIC) or purpurafulminans [57-60] unless treated with protein C replacement therapy. Because anticoagulant therapy slows cancer advancement, it's possible that the natural anticoagulant protein C pathway has a protective function in cancer progression as well.

Future research perspectives

Research may be planned to explore the cross-talk between TFPI, protein S, Protein C, anti-thrombin III genes and their expression in patients with breastcancer. We also aim to characterize the process of thrombogenesis and fibrinolysis activation in patients with breast cancer. As thrombogenesis and fibrinolysis in systemic circulation are interrelated with inflammation and endothelial function we also aim to measure markers of inflammation and endothelial activation. Ultimate goal must be to find out whether the above-mentioned association could be an early predictor of the progression of breast malignancy.

Expected outcome

- Such study may reveal the role of Cross-talk of TFPI, Proteins C and S and antithrombin III genes and expression in thrombogenesis in breast cancer.
- The study might reveal that laboratory profiling of thrombogenesis may be predictors of progression of breast cancer.
- Early prediction of cancer spread may be useful in planning therapeutic strategy and hence improving the quality of life of patients with breast cancer.
- The TFPI, Proteins C and S and antithrombin III genes may be therapeutic targets of metastatic breast cancer.

Conclusion

If the intended goal of establishing the association between the cross-talk between TFPI, Proteins C, S ,anti-thrombin III genes and their expressions with the markers of thrombogenesis, this may be an early predictor of progression of cancer. The study may be beneficial to the breast cancer patients in early identification of cancer spread and hence therapeutic strategy planned accordingly may improve the quality of life of the patients.

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