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ONCOGENE-DRIVEN HEMOSTATIC CHANGES IN CANCER

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ABSTRACT.

A common feature in progression of multiple human malignancies is the protracted deregulation of the coagulation system, often referred to as cancer coagulopathy. The genesis of this syndrome can be traced to changes in the tumour vascular interface, formed through vascular invasion, angiogenesis and metastasis. The resulting contact between cancer cells and the circulating components of the coagulation system compromise the regulatory barriers that normally control physiological haemostasis. In addition, cancer cells and their stroma often exhibit procoagulant properties. While these changes have long been thought to be unspecific in nature evidence now exists to suggest that cancer coagulopathy and the related Trousseau syndrome are a function of the genetic tumor progression. Indeed, the expression of several effector molecules of the coagulation and fibrinolytic systems, including: tissue factor (TF), plasminogen activator inhibitor 1 (PAI-1), cyclooxygenase 2 (COX-2) or urokinase (uPA) are often *direct* regulatory targets of oncogenes (K-ras, EGFR, HER-2, c-MET) and tumour suppressors (p53, PTEN). Moreover, oncogenic alterations act on coagulation *indirectly* by driving formation of leaky vessels, metastasis or inflammation. These procoagulant influences of oncogenic pathways are modulated by hypoxia, stress responses and cellular differentiation, the latter involving formation of cancer stem cells and their niches to which coagulation factors may contribute. It is possible that targeting cancer-related coagulopathy may require more cancer-specific measures, to achieve improved thromboprophylaxis and impact patients survival.

INTRODUCTION - HEMOSTATIC PERTURBATIONS IN CANCER

Persistent activation of the coagulation system is amongst the most consistent correlates of advanced malignancy, and its thrombotic consequences remain the second leading cause of cancer related deaths (recently reviewed in [1-4]). In spite of the fact that these changes have long been recognized clinically, and studied experimentally, their nature is still poorly understood and rather complex [1;3], hence the descriptive term of cancer coagulopathy is frequently used to describe them collectively [1;3]. In memory of Armand Trousseau who firmly introduced the notion of cancer-related thrombosis to the literature [5] (although preceding reports do exist [1]), this condition is also sometimes referred to as Trousseau (Trousseau's) syndrome, a term which has historically been reserved to a rather specific set of circumstances, where unexplained thrombotic events preceded the diagnosis of previously unrecognized malignancy [6]. It is argued that the term Trousseau syndrome perhaps should continue to be defined in these strict clinical terms, namely as a cluster of abnormalities that usually include: migratory thrombophlebitis, chronic disseminated intravascular coagulopathy, verrucous endocarditis, microangiopathic anaemia and arterial microemboli, all emerging on the background of occult visceral tumour [6]. At the same time it is recognized that the traditional definition of this important syndrome is essentially retrospective in nature, and thereby of uncertain practical (diagnostic) utility, and also that it may divide what is ultimately a continuum of biological changes (however diverse) [6].

Indeed, not entirely dissimilar procoagulant events may occur both before and after cancer diagnosis, and cancer patients often develop venous thromboembolism (VTE), including deep vein thrombosis (DVT), pulmonary embolism (PE)[7], or syndromes that resemble low grade disseminated intravascular coagulation (DIC) [27, 28], or else asymptomatic alterations in laboratory

tests indicative of the activated coagulation [1;2;4]. Notably, up to 90% of patients with metastatic cancer are affected by some form of coagulopathy, which is also a cause of considerable morbidity and mortality [3;8;9].

It is noteworthy that owing to the progress in imaging technology, screening programs and medical education of the public cancer diagnosis is made in many instances at much earlier stage of the disease than would have been the case even a decade ago. Therefore, the contribution of various factors that are thought to be responsible for cancer coagulopathy may undergo some rearrangement. These factors include external influences such as immobility and iatrogenic effects of surgery, central lines or anticancer medication, including novel targeted agents, but is perhaps most strongly defined by the continuous and progressive nature of the disease itself, in its many local, regional and systemic manifestations [10]. While we are awaiting a consensus as to maintaining or broadening the usage of different terms to describe cancer coagulopathy, its nature remains amongst the most fascinating and daunting medical challenges. This article will touch on but one aspect of this rapidly evolving field.

IS CANCER COAGULOPATHY CANCER-SPECIFIC?

A sound argument could be made that coagulation perturbations and thrombosis in cancer patients could be induced by factors similar to those operative during tissue injury, vascular damage, inflammation, infection, intake of certain types of medication and immobility imposed for various reasons (all present in cancer patients), i.e. cancer coagulopathy could be cancer-unspecific in nature. In the other words, the release of procoagulant tissue material into the circulation (e.g. mucins), abnormal vascular barrier, stasis, hypoxia and production of cytokines

could occur in conjunction of cancer-unrelated injury, surgery, inflammatory bowel disease, still causing an increased risk of thrombosis [6].

While the contribution of these 'unspecific' triggers to the cancer-related coagulopathy is undeniable, there are several reasons to think that the malignant process adds to these changes another novel, quantitative and qualitative dimension. First, the risk of thrombosis is generally 6-7 fold greater in cancer patients than in patients with a similar configuration of other risk factors [11]. Second, this risk differs in different types of malignancies, even with seemingly similar constellations of 'unspecific' characteristics. For instance, several studies [11-13] show that the risk of thrombosis is markedly greater in ovarian, pancreatic or brain tumours (glioblastoma) than in head and neck cancer (as reviewed extensively [1;2;4]). Also normal haemostasis has recently been shown to be regulated in an organ-specific manner, notably as a result of differences in involvement of tissue factor (TF) [14]. Therefore, the site-specific differences cancer-related risks of thrombosis are inconsistent with the idea of some generic, or external, 'unspecific' mechanism being involved in their causation. Third, as mentioned earlier, the risk of coagulation perturbations increases with cancer progression [1;15], often without a particular, discrete change in the configuration of 'unspecific' factors. Fourth, these perturbations are often exacerbated [1;16], or else sometimes alleviated by cancer-specific medication, e.g. by chemotherapy in breast cancer [17], or in multiple myeloma (MM), especially in conjunction with thalidomide [1], or by all-trans retinoic acid (ATRA) therapy in acute promyelocytic leukemia (APL), respectively. Fifth, certain relevant elements of cancer pathogenesis have no parallel in other chronic conditions that might be associated with thrombosis, especially as it relates to processes of metastasis, tumour angiogenesis and the underlying mutational changes at the cellular level. These factors are

especially worthy of closer analysis, as they may illustrate the various levels at which cancer progression affects the cogulation system [10].

WHAT ARE THE CANCER-SPECIFIC TRIGGERS OF HYPERCOAGULABILITY?

Malignancies are associated with a host of complex and heterogeneous, localized and systemic changes. These processes are thought to involve a succession of premalignant alterations, followed by cellular diversification and emergence of cancer stem cells (CSCs), also known as tumour initiating cells (TICs)[18]. It is thought that in a conducive tissue context, known as cancer stem cell niche, dormant CSCs cells can be triggered to multiply and produce their mitogenically active cancer cell progeny, the main constituent of the tumour mass [18]. Further tumour growth also depends on generation of the vascular stroma, influx of inflammatory cells, macrophages and well as endothelial and hematopoietic progenitors [19;20]. These local changes in the tissue composition and architecture are associated with a multitude of systemic effects, including activation of bone marrow and formation of premetastatic niches in distant organ sites, eventually followed by overt tumour dissemination [21]. Arguably, at the heart of all these complex changes are the abnormal regulatory networks triggered within cancer cells by mutational (or epigenetic) events that ultimately underlie the activation of oncogenes and inactivation of tumour (and metastasis) suppressors [22;23].

It is now well established that oncogenic alterations in cancer cells impact various facets of the vascular system. For instance, oncogenic transformation triggers expression of vascular endothelial growth factor (VEGF) and several other cytokines and chemokines [24;25], whereby cancer cells become capable of attracting blood vessels and recruitment of inflammatory and

bone marrow-derived cell of endothelial and hematopoietic origin [26]. These various cells are known to participate in formation of the 'private' tumour microcirculation, namely through processes of tumour angiogenesis, vasculogenesis, blood vessel invasion, vessel cooption and vasculogenic mimicry [10;26;27].

However, the resulting vascular structures are vastly different from those that emerge during normal development or wound healing, including their architectural [28], cellular [29] and molecular [30] properties. Notably, tumour blood vessels are heterogeneously hyperpermeable, and leaky to plasma-derived macromolecules, including coagulation factors, which thereby come into direct contact with the procoagulant extravascular microenvironment of cancer cells and tumour stroma [4;10]. In addition, the actions of VEGF [25;31], inflammatory cytokines (e.g. tumour necrosis factor alpha) and other stimuli [32] may turn on the expression of tissue factor (TF) in normally anticoagulant endothelial cells, thereby further compromising their barrier function [33;34].

Procoagulant tumour cells not only interact with extravascular clotting factors, but also physically enter the vascular compartment through processes of blood vessel invasion, microscpoic intravasation, or as circulating metastatic deposits [35]. In addition tumor-related proteins, including TF, may enter the blood stream as cargo of procoagulant microvesicles/microparticles [7;36;37]. The sum of these emerging new points of abnormal contact between the tumour mass and the vascular system (tumour-vascular interface), is likely to strongly influence cancer coagulopathy, their nature is, for the most part, unique to cancer [38] and, we postulate that their causative stimulus lies to a large extent with cancer causing oncogenic mutations [10].

ONCOGENE-DEPENDENT ALTERATIONS AFFECTING THE COAGULATION SYSTEM

The impact of the oncogenic pathways on procoagulant events in cancer has only recently been recognized [39], and can be viewed as either *indirect* or *direct* in nature. In the first instance, it is reasoned that the wide spread changes in gene expression profiles induced by the malignant transformation also include the various modifiers of the tumor-vascular interface, resulting in changes impacting the haemostatic system. Paradigmatic in this regard is the influence exerted by the loss of the p53 tumor suppressor gene. Seminal studies of Bouck and collaborators established for the first time that intrinsic changes in cancer cells, and not necessarily the microenvironment, are primary inducers of the angiogenic phenotype, here epitomized by deregulation of the antiangiogenic protein thrombospondin 1 (TSP-1)[40-42]. Subsequent studies revealed the key role of *ras* oncogene in regulation of both VEGF, the central inducer of angiogenesis and vasculogenesis, and TSP-1 its antithetical angiogenesis inhibitor [24;25;43]. Collectively, these and numerous follow up studies involving over 20 different oncogenic lesions and several of their downstream pro- and anti-angiogenic growth factors, cytokines and polypeptides (cancer cell *angiome*) provided ample demonstration that the consequences of cancer-causing genetic events transcend cellular boundaries, and through paracrine influences lead to formation of abnormal, static, leaky, TF expressing and sometimes occlusive intratumoral blood vessels. This, in turn, may be one of the oncogene-dependent sources of pro-coagulant events associated with cancer [10].

There is also mounting evidence that intracellular signalling cascades triggered downstream of mutant oncogenes and tumor suppressors directly control the ability of cancer cells to express (sometimes ectopically) several genes related to the coagulation or fibrinolytic systems, i.e. the

cancer cell *coagulome* [10]. What are the most important activities in this regard remains to be established, but Table 1 summarizes the presently available, albeit still limited, literature. Thus, evidence exists that oncogenic lesions impact the expression of professional elements of the coagulation (e.g. TF) and fibrinolytic pathways (urokinmase/uPA), which are otherwise normally expressed in various tissues. However, cancer cells also acquire the ability to express coagulation factors ectopically. For instance, factor VII was recently found to be expressed by several different liver-unrelated tumour cell lines [44]. This is interesting as activation of this factor and its binding to TF on the same cells could potentiate their procoagulant properties and, at the same time contribute to the autocrine (e.g. PAR-2-mediated) TF signalling [44]. Similarly, thrombin-like protein was found to be produced by a human breast cancer cell line [45]. If confirmed, this finding could suggest the existence of TF-unrelated mechanisms of procoagulant conversion of these cancer cells, and potentially, once again, their autocrine stimulation *via* PAR-1 [44]. In both of these cases the ectopic production of coagulation factors appears to be transformation-specific, as such production is largely restricted to cancer cells. If so, this would implicitly suggest an involvement of some, presently unknown, oncogenic events. The third category of cancer- and oncogene-dependent procoagulant factors are ones in which this putative activity has no known role in normal haemostasis. The case in point are cancer-related mucins, some which are targets of oncogenes and have been previously implicated in the activation of the coagulation system [6;46]. Fourth, class of factors is composed of the protease known as cancer procoagulant (CP). There is presently no data as to the regulation of CP by oncogenic events, and the discussion of the mechanisms leading to the release of this interesting factor would be more informative once its origin and identity are understood in more detail. Fifth category of procoagulant activities released by cancer cells under the influence of oncogenic events are TF-containing membrane-

related tumour microvesicles (Table 1). Recent studies revealed that oncogenic events, such as mutations of p53, or activation of epidermal growth factor receptor (EGFR) and other changes may drive the release of microvesicular TF into the circulation [37], a process that involves both stimulation of the cellular vesiculation process [47] and well as oncogene-driven overexprssion of TF itself [37;48]. The latter process is worth considering in more detail as TF represents an interesting paradigm for the control of cancer coagulopathy, due to its pivotal role in the coagulation cascade, cellular association and well-recognized biological and signalling activity [44].

REGULATION OF TISSUE FACTOR BY ONCOGENIC PATHWAYS

Tissue factor (TF) is a 47 kDa transmembrane cellular receptor for the blood borne coagulation protease, factor VIIa (FVIIa) [49;50]. As TF is normally absent in the vascular lumen this interaction can only occur upon vascular injury, abnormal TF expression on endothelial cells (e.g during angiogenesis), or after entry into the blood stream of a large number of TF expressing material, such as TF expressing cells and microvesicles [50-53]. Formation of the TF/VIIa complex[50;52] triggers proteolytic conversion of circulating factor X (FX) to an active form (FXa) (along with activation of FIX). FXa activates small amounts of prothrombin (FII) to thrombin (FIIa)[54]. A subsequent burst of thrombin activity is facilitated by platelets, and factors Va, VIIIa and IXa leading to conversion of soluble fibrinogen into insoluble fibrin [54], activation of protease-activated (G protein-coupled) receptors (PARs), followed by further recruitment of platelets and rapid clot formation [55]. These processes are kept in check by several opposing influences [52;54], including: tissue factor pathway inhibitor (TFPI), protein C, antithrombin [52] and the fibrinolytic system [54;56].

TF expression is not restricted to cancer and can be detected on extravascular smooth muscle and connective tissue cells surrounding blood vessels (hemostatic envelope), and in association with inflammatory cells, especially upon activation [32]. However, it is also true that TF activity on the surface of cancer cells can be up to 1000 fold greater than that associated with their normal counterparts [34;59]. Indeed, TF is overexpressed in many cancers, especially at their late stages of progression [34;60-64], where the degree of TF-positivity often correlates with poor prognosis [65], metastasis [66], high microvascular density (MVD), and expression of VEGF [67].

While it has been postulated that TF may be expressed as a result of tumour hypoxia or stimulation with inflammatory cytokines [4;68], may types of cancer cells express this receptor constitutively, including in cell culture, i.e. in the absence of hypoxia or inflammation, and often as a function of their malignant transformation [10;37]. This led to the suggestion that oncogenic lesions in the cancer cell genome may contribute to their TF overexpression and procoagulant conversion, in very much the same way as these lesions control the tumor-related angiogenic phenotype and neovascularization [39]. Indeed, studies involving human colorectal cancer cells (CRC) first revealed that the succession of oncogenic mutations affecting K-*ras* proto-oncogene and *p53* tumor suppressor (two pivotal events during CRC progression) trigger the corresponding, stepwise increases in TF gene expression, protein production, procoagulant activity and shedding of TF-containing tumour-cell derived microvesicles [37]. Interestingly, when two isogenic and tumourigenic CRC cell lines with different p53 status were compared in an *in vivo* xenograft assay, it was found at the same tumour size that the cells that have lost *p53* expression were more proficient in shedding microvesicular TF into the circulation of the tumour

bearing mice than their *p53* proficient counterparts [37]. As K-*ras* mutations were present in both tumour types, it is unclear whether this effect of *p53* is independent, or requires the concomitant activation of the Ras pathway.

In the absence of *ras* gene mutations Ras pathway may become activated *via* upstream stimulation of protooncogenic receptor tyrosine kinases (RTKs), such as members of the ErbB family, including EGFR and or ErbB2/HER-2. These RTKs are frequently involved (activated) in epidermal, epithelial, brain and other human malignancies [69] and, as recently demonstrated, their activation robustly triggers overexpression of TF [48;70;71]. For instance, copious amounts of TF are produced by the human, epithelial, squamous cell carcinoma cell line, A431, which also contains an amplified wild type *EGFR* gene and overexpress the oncogenic EGFR protein. Moreover, treatment of these cells with EGFR agonists, such as transforming growth factor alpha (TGF α), leads to further dramatic increases in TF gene expression, protein production and procoagulant activity, while several different pharmacological inhibitors of EGFR (C225, AG1478, CI-1033) induce the opposite effect ([48] and Milsom & Rak – unpublished data).

Oncogenic activity of EGFR in A431 cells not only drives the cell-associated ovrerexpression of TF, but also the release of this receptor into the cellular micromilieu, as membrane microvesicles [72]. The significance of TF shedding is in its independent procoagulant actions [53] but also in the uptake of the TF containing microvesicles by other cells and platelets [7;73]. In cancer, such mechanism could lead to propagation of the procoagulant potential, both within the tumour and systemically. Moreover, we postulated earlier that the intercellular transfer of TF as microvesicle cargo may represent an interesting paradigm for ‘sharing’ this and other cellular receptors,

between the cells, a process that could include also the oncogenic RTKs (Yu and Rak – unpublished). Indeed, when microvesicles derived from A431 cells were incubated with TF-negative mouse endothelial cells this led to a transfer of both fluorescently labelled tumour cell membrane material and the associated human TF activity (Yu, Milsom & Rak unpublished). Interestingly, our subsequent studies based on this paradigm revealed that indeed, other receptors could be transferred between the cells in a similar manner. For instance, oncogenic EGFR was found to be incorporated into the cargo of tumour related exosomes [74] and microvesicles, and in that form could traffic between the adjacent cells [47]. The consequences of this latter process for cancer related coagulopathy remain presently unclear. However, blood borne TF-containing microvesicles are increasingly viewed as a possible source of prognostic information regarding the risk of thrombosis in cancer patients. This notion is consistent with the suggestion that cancer progression and the related oncogenic events influence the risk of thrombosis by impacting TF expression [37] , cellular vesiculation [37;47] and possibly the intercellular microvesicle transfer [47].

TF [75] and EGFR [76] are highly expressed in human glioblastoma multiforme (GBM), and are both of potential prognostic significance. In particular, the presence of the ligand-independent EGFR mutant (EGFRvIII) signifies the transition from lower grade disease to a high-grade aggressive GBM [76], of which vascular proliferation, necrosis and thrombosis are important hallmarks [77]. To explore the possible linkage between EGFRvIII and the prothromotic characteristics of GBM cells we employed an indolent glioma cell line (U373), which expresses low levels of TF. When these cells were transfected with EGFRvIII cDNA we observed a dramatic increase in TF expression and procoagulant activity, properties that were readily reversible upon exposure to the several EGFR inhibitors (Milsom, Yu, Magnus & Rak unpublished).

Although EGFR may play a significant role in TF expression in primary GBM, other genetic events are clearly operative in this disease, and could contribute to its hypercoagulable nature [77]. Pioneering work in this regard was recently published by Brat, Rong and colleagues who documented the impact associated with the loss of the *PTEN* tumor suppressor on hypoxia-regulated levels of TF in astrocytic and GBM cells [78]. PTEN is a lipid phosphatise and tensin homologue localized on chromosome ten, which is mutated, lost or silenced in up to 70% of all GBM. PTEN plays a crucial role in down modulating the excessive activity of the PI3K/Akt cascade, downstream of several growth and survival regulating receptors, including in cells harbouring EGFR and EGFRvIII oncogenes [69;76;79]. In their study Rong et al demonstrated that loss of PTEN and hypoxia cooperate in upregulation of TF in astrocytic cells, likely in a manner that involves Akt and hypoxia responsive transcription factor Egr-1 [78]. As loss of PTEN often overlaps in a prognostically significant manner with the activation of the EGFR/EGFRvIII pathway [80], it is of interest whether both of these lesions also interact in regulating TF expression and coagulopathy of GBM cells and are linked to thrombotic events in the respective GBM patients. It is also of great interest whether TF regulation by these oncogenic pathways influences in a more direct manner the biology of the disease.

COMPLEXITY OF PROCOAGULANT CHANGES REGULATED BY ONCOGENES

While TF serves as an instructive paradigm to consider the role of oncogenic pathways in the regulation of the cancer coagulome, the scope of these latter changes is likely much wider. For instance, while mutant *ras* upregulates TF in various settings, it also regulates the expression of fibrinolytic activities (uPA), a property that may explain why massive vascular occlusion and

thrombosis is relatively infrequent, even in highly TF-positive tumors [10]. However, there are instances, at least in experimental settings, where a single oncogenic lesion may provoke an overt and extremely severe thrombohaemorrhagic syndrome. The case in point is the recent elegant study by Boccaccio et al, in which c-MET oncogene was introduced into the mouse liver by a lentiviral vector, and the resulting neoplastic growth was found to precipitate a life threatening thrombosis, ostensibly due to upregulation of PAI-1 and COX-2 [81]. While this may represent an extreme case, with few direct parallels in the clinical context, somewhat milder thrombotic complications do emerge as a result of oncogenic activity in patients with certain types of malignant diseases. For instance, in acute promyelocytic leukemia (APL), the *PML-RARa* oncogene plays a pivotal transforming role [82;83], which can be partially blocked by treatment with all-trans retinoid acid (ATRA) [82]. Interestingly, this treatment also leads to the resolution of the APL-associated coagulopathy, possibly due to downregulation of TF on leukemic cells, along with other changes [82]. These examples merely scratch the surface of what is likely to be a much larger network of molecular interactions that influence haemostatic parameters in cancer patients. Until more data is available, in can only be speculated that in addition to TF, also other professional (ectopic FVII) and opportunistic (mucins, adhesion molecules) procoagulant molecules could be regulated by the oncogenic transformation and contribute to hypercoagulability.

CONCLUSIONS AND IMPLICATIONS

The ‘oncogene hypothesis’ provides a workable platform to understand the cancer-specific alterations in the haemostatic system, however it is hardly the ultimate answer to the puzzle of cancer coagulopathy. As in the case of other cancer related processes, oncogenic pathways that

control expression of TF and other coagulation-related effectors intersect at many levels with pathways of stress response, hypoxia [68] and cellular differentiation [84]. The latter regulation is of particular interest as it controls how oncogene-harbouring cells acquire properties of cancer stem cells (CSCs/TICs) or become their limited proegeny[18]. In this regard it was recently postulated that the procoagulant microenvironment may serve as a provisional CSC niche and thereby promote disease progression [84].

Thus, it is increasingly appreciated that cancer coagulopathy may contribute not only to thrombotic side effects but also to disease progression and aggressiveness as such [8-10;85;86]. This is highlighted by improved survival of subsets of patients included in several recent clinical trials involving administration of low molecular weight heparin [1;87-89]. While this is encouraging, it is also possible that the pathogenesis of cancer-related coagulopathy is sufficiently dissimilar from conditions found in cardiovascular, orthopaedic and other contexts, that it may benefit from including additional cancer-specific measures, e.g. targeted agents to obliterate oncogene-driven expression of TF, and/or TF antagonists [90]. It is likely that a better understanding of cancer coagulopathy may provide additional insights into the pathogenesis of cancer [10].

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Table 2: Linkage between oncogenic events and procoagulant phenotype of cancer cells

Genetic Influence		Consequence of Deregulation	References
Oncogenes	K-ras	Upregulation of TF in human CRC cells	[37]
	H-ras	Downregulation of TFPI 2	[91]
	H-ras	Upregulation of uPA	[92;93]
	H-ras	Downregulation of uPA in invasive cells	[94]
	H-ras	Upregulation of mucins	[95]
	src	Upregulation of TF	[96]
	EGFR	Upregulation of TF	[48]
	EGFRvIII	Upregulation of TF	Milsom, Yu, Magnus & Rak unpublished
	RGFRvIII	Increase in vesiculation (may contribute to release of procoagulant activity)	[47]
	HER-2	Increase in TF expression	(Yu & Rak unpublished)
	HER-2	Upregulation of mucins	[95]
	Unknown	Ectopic expression of in cancer FVII	[97]
	Unknown	Ectopic expression of thrombin-like protein	[45]
	PML-RARa	TF-dependent coagulopathy	[82]
	c-MET	Deregulation of PAI-1 and COX-2	[81]
Tumour Suppressors	P53	Upregulation of TF in human CRC cells	[37]
	P53	Increase in release TF containing microvesicles into the circulation	[37]
	PTEN	Upregulation of TF	[78]

TF, tissue factor; PAI -1, plasminogen activator inhibitor type 1; COX 2, cyclooxygenase; CRC – colorectal carcimoma; TFPI 2 – tissue factor pathway inhibitor; FVII – coagulation factor VII; uPA – urokinase plasminogen activator

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FIGURES

Figure 1.