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Review Article

FER as a novel target for cancer therapy

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CONTENTS

Abstract

FER is an intracellular tyrosine kinase which resides in both the cytoplasm and the nucleus of mammalian cells. Although FER is present in a variety of tissues and cells, mice devoid of active FER develop normally and the proliferation of fibroblasts derived from these mice is not impaired *in vitro*. Thus, FER does not appear to exert an essential function in the proliferation of normal cells. However, several lines of evidence suggest a regulatory role of FER in the progression and growth of malignant tumors. FER is highly expressed in numerous malignant cell lines and the levels of FER in malignant prostate tumors are significantly higher than those detected in benign prostate tumors. Furthermore, downregulation of FER impairs the proliferation of prostate carcinoma cells and attenuates the development of prostate cancer. Recent findings also suggest the involvement of FER in the progression of breast cancer and it has been shown to play a unique regulatory role during cell cycle progression in breast cancer cells. In the current review, we discuss the molecular mechanisms which underlie the pivotal role of FER in the proliferation of malignant cells. The validity of FER as a novel intervening target for cancer therapy is discussed as well.

Introduction

FER is an intracellular tyrosine kinase which resides in both the cytoplasm and the nucleus of mammalian cells (1-3). Together with FES, FER constitutes a distinct subfamily of intracellular tyrosine kinases which share a unique structure. Both kinases bear an extended *N*-terminal tail containing an FPS/FES/FER/CIP4 homology domain (FCH), followed by three coiled-coil-forming regions (Fig. 1). While the FCH domain is thought to mediate the association of FER and FES with microtubular structures, the coiled-coil domains have been shown to direct the oligomerization of these kinases. The kinase domain of the two enzymes is located at their carboxy terminal part (Fig. 1) and is 70% identical between the two proteins (4).

FER is activated by growth factors such as epidermal growth factor (EGF) and platelet-derived growth factor (PDGF) in fibroblast cells (5), and by occupation of the Fcγ receptor in mast cells (6). Activation of FER in these systems could be linked to the modulation of cell-cell and cell-substrate interactions, since FER has been shown to associate with and to phosphorylate adhesion molecules (4, 5, 7-11). FER has also been shown to associate with key cellular regulatory proteins, including phosphatidylinositol 3-kinase (PI3K) (12), signal transducer and activator of transcription 3 (STAT3) (13) and the cytoskeletal linker protein plectin (14).

Although FER is present in a variety of tissues and cells, mice devoid of active FER develop normally and the proliferation of fibroblasts derived from these mice is not impaired *in vitro* (15). Similarly, mice that lack functional FES are viable and only exhibit slight alterations in myelopoiesis (4, 16). However, several lines of evidence suggest a regulatory role of FER and FES in the progression and growth of malignant tumors. Surprisingly, while FES was originally identified as a proto-oncogene (17), it has recently been shown to exert tumor-suppressive activity in both breast and colon carcinomas (18, 19). On the other hand, various observations suggest the involvement of FER in the promotion of malignant processes. FER is highly expressed in numerous malignant cell lines (2, 20) and the levels of FER in malignant prostate tumors are significantly higher than those detected in benign prostate tumors (21). Furthermore, downregulation of FER impairs the proliferation of prostate carcinoma cells and abolishes their ability to form colonies in soft agar (21). Recent findings also suggest the involvement of FER in the progression of breast cancer and it has been shown to play a unique regulatory role during cell cycle progression in breast cancer cells (22). These findings suggest that, unlike FES, FER supports the development

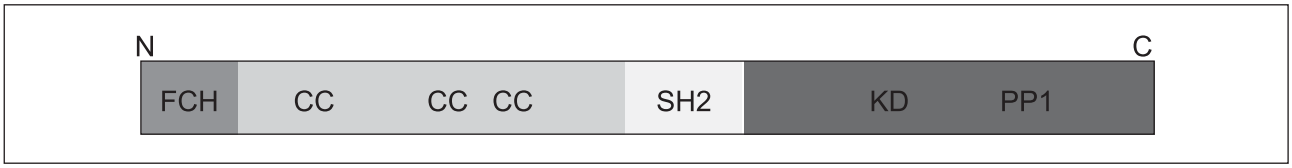

Fig. 1. Schematic description of the FER protein. The different subdomains in FER are depicted. FCH, FES/FER/CIP4 homology domain; CC, coiled-coil sequences; SH2, SH2 domain; KD, kinase domain; PP1, PP1 binding sites.

and progression of specific tumors. Thus, while FER and FES belong to the same tyrosine kinase subgroup, they seem to exert opposite effects on tumor progression. In this review, the molecular mechanisms through which FER supports tumor progression will be described and the possible exploitation of FER as a novel target for cancer therapy will be discussed.

FER and proto-oncogenes

FER and STAT3

One of the cellular activities of FER that could hint at its involvement in malignant processes is its ability to activate the pro-oncogenic transcription factor STAT3 (signal transducer and activator of transcription 3). Like FES, FER associates with STAT3 in various cell types (13), including malignant cells. Furthermore, the association of FER with STAT3 has been shown to be regulated by interferon gamma (IFN-γ), which causes cell cycle arrest in colon carcinoma cells. Treatment of colon cancer cells with IFN-γ leads to downregulation of FER and its association with inactive STAT3 (20). However, there is no evidence for the direct activation of STAT3 by FER in malignant cells. Moreover, activation of STAT3 in cells stimulated with PDGF is not affected by the absence of functional FER. Detailed studies aimed at the identification of growth factors that could elicit the FER-dependent activation of STAT3 revealed the ability of insulin to induce the activation of STAT3 in myogenic cells via a pathway involving PI3K (23). Knockdown of FER severely impairs the insulin-driven activation of STAT3 in these myogenic cells (23). Further studies should reveal whether FER mediates the insulin-stimulated activation of STAT3 in other cell types. However, FER was found to be required for the proliferation of prostate carcinoma PC-3 cells, which do not harbor active STAT3 (21). Thus, to date, there is no direct evidence for the involvement of STAT3 in the pro-oncogenic role of FER in malignant cells.

FER and β-catenin

β-Catenin was originally identified as a key effector of Wnt signaling (24). Later, it was suggested to be a component of cell adhesion complexes where it associates with cadherins (25). The function of classic type 1 cadherins depends on their association with the actin cytoskeleton, an interaction mediated by α- and β-catenin (25). When the level of free cytoplasmic β-catenin rises following Wnt signaling, or after being released from adhesion complexes, β-catenin enters the nucleus and binds the transcription factor T-cell factor (TCF) to initiate transcription of Wnt-responsive genes (26). These include pro-oncogenic genes such as cyclin D1 (27-29). Thus, β-catenin itself functions as a potent proto-oncogene, and indeed, the protein has been found to be deregulated and constitutively active in a wide range of tumor types (29). β-Catenin association with E-cadherin and α-catenin recruits β-catenin to cell adhesion complexes and excludes it from the nucleus.

The involvement of FER in the establishment of cell adhesion complexes has led to its identification as a regulator of β-catenin accumulation in these complexes. This would also affect the accumulation of β-catenin in other cellular compartments and would consequently alter the availability of β-catenin to nuclear transcription complexes. However, there are contradictory reports concerning the effect of FER on the presence of β-catenin in nuclear pools.

Plakoglobin is a protein closely related to β-catenin that links desmosomal cadherins to intermediate filaments. Plakoglobin can also substitute for β-catenin in adherens junctions, providing a connection between E-cadherin and α-catenin. β-Catenin association with E-cadherin and α-catenin is regulated by phosphorylation of specific tyrosine residues; modification of β-catenin Tyr654 and Tyr142 decreases binding to E-cadherin and α-catenin, respectively. Plakoglobin can also be phosphorylated on tyrosine residues, but unlike β-catenin, this modification is not always associated with a disrupted association with junctional components.

Protein tyrosine kinases can phosphorylate different tyrosine residues on β-catenin and plakoglobin, and tyrosine phosphorylation of β-catenin and plakoglobin can have different effects on their interactions with components of desmosomes or adherens junctions. The tyrosine kinase activity of FER, which modifies β-catenin Tyr142 and thereby weakens its association with α-catenin, phosphorylates plakoglobin on Tyr549 and produces the opposite effect by strengthening the binding of plakoglobin to α-catenin. It has been suggested that phosphorylation of Tyr549 and increased binding of plakoglobin to components of adherens junctions may contribute to the release of β-catenin from adhesion complexes and may lead to upregulation of the transcriptional activity of the β-catenin-TCF complex observed in many epithelial tumor cells (9). However, later studies indicated an opposite role of FER in modulating the partitioning of β-catenin between adhesion complexes and the cell nucleus.

As described above, the phosphorylation state of β-catenin controls its association with cadherins and thus the association of cadherin with the cellular cytoskeleton. Xu *et al.* reported that the phosphorylation of β-catenin is regulated by the combined activities of the tyrosine kinase FER and the tyrosine phosphatase PTP1B. FER was shown to phosphorylate PTP1B at Tyr152, regulating its binding to cadherin and the continuous dephosphorylation of β-catenin at Tyr654. Furthermore, FER was found to interact with cadherin indirectly, through p120ctn. The interaction domains of FER and p120ctn have been mapped and competing peptides corresponding to these sequences release FER from p120ctn *in vitro* and in live cells. This results in a loss of cadherin-associated PTP1B, an increase in the pool of tyrosine-phosphorylated β-catenin and a loss of cadherin adhesion function. The effect of the peptides is lost when a β-catenin mutant with a substitution at Tyr654 is introduced into cells. Thus, FER phosphorylates PTP1B at Tyr152, enabling it to bind to the cytoplasmic domain of cadherin, where it maintains β-catenin in a dephosphorylated state. Cultured fibroblasts from mouse embryos targeted with a kinase-inactivating ferD743R mutation lose cadherin-associated PTP1B and β-catenin, as well as localization of cadherin and β-catenin in areas of cell-cell contacts (30). Hence, this study implicates FER in recruiting β-catenin to adhesion complexes rather than increasing its nuclear pool. In accordance with this notion, knockdown of FER in prostate and breast carcinoma cells does not cause any change in the cyclin D1 gene expression level (22), indicating that β-catenin is not involved in the pro-oncogenic activity of FER in carcinoma cells.

The findings summarized above imply that although FER supports the proliferation of malignant cells, a STAT-dependent signaling pathway has not been shown to be involved and it is also not clear if β-catenin mediates this tumor-promoting activity of FER.

FER and tumor suppressors

While there is currently no experimental evidence for a functional link between FER and known oncogenes in the context of malignant cell proliferation, several findings indicate a regulatory impact of FER on the functioning of tumor suppressor proteins.

FER and pRb1

Orlovsky *et al.* and Allard *et al.* were the first to show that downregulation of FER leads to cell cycle arrest in malignant cells. While Allard *et al.* used an antisense approach in prostate carcinoma PC-3 cells (31), Orlovsky *et al.* overexpressed a dominant negative FER mutant, which led to the downregulation of the endogenous FER in HeLa cells and to the arrest of the treated cells at the G1-S transition point (32). These findings were further corroborated by a later study, which demonstrated that the knockdown of FER using a short interfering RNA (siRNA) approach also arrested PC-3 cells at the G1-S transition stage (22). Both HeLa and PC-3 cells harbor a functional retinoblastoma protein pRb1 (p105), which serves as a key regulator of G1-S transition in mammalian cells (33, 34). This suggested the possibility that FER may be involved in the regulation of pRb1 function in malignant cells.

pRb1 plays a restrictive role in eukaryotic cell cycle progression when cells exit G0 or G1 and enter the S phase, thereby acting as a crucial negative regulator of cellular proliferation and neoplasia (35). In quiescent or early G1-phase cells, pRb1 is hypophosphorylated and associates with specific members of the E2F transcription factor family, converting them to active transcriptional repressors (36, 37). Gene repression is also mediated by pRb1 and pRb2 (p130) recruitment of histone deacetylase (HDAC) to promote the formation of inhibitory nucleosomes (38-40). The many proteins found in association with pRb1 suggest that additional regulatory mechanisms may also be involved (41), although the physiological role of most of these interactions remains unproven. Cell cycle progression from the G to the S phase occurs when complexes of D-type cyclins/CDK4/CDK6 phosphorylate pRb1, thereby derepressing E2Fs to direct transcription of the DNA replication machinery and nucleotide biosynthesis genes (42). Mutational inactivation of pRb1 causes the pediatric cancer retinoblastoma, while deregulation of the pathway in which pRb1 functions is common in most types of human cancers (43).

Knockdown of FER in prostate and breast cancer cells leads to dephosphorylation and activation of the suppressive activity of pRb1. This most probably results from the G0/G1 arrest in the treated malignant cells. While in FER-depleted breast carcinoma cells pRb1 became dephosphorylated only in several of its 16 serine/threonine phosphorylation sites, in prostate carcinoma cells pRb1 became hypophosphorylated on all sites analyzed (22).

The intense dephosphorylation of pRb1 following the downregulation of a single kinase is unusual and even surprising in light of recent information on the regulation of G1-S transition in mammalian cells. G1 progression is governed by a large number of control steps, most of which are deregulated in malignant cells (44). However, recent findings suggested a high functional redundancy of the G1-S regulatory components, such as CDK2 and CDK4 (45-49). In accordance with this redundancy, knockdown of either CDK4 or CDK2 in prostate carcinoma cells does not lead to hypophosphorylation of pRb1 (22). Hence, the profound effect of FER on the phosphorylation state of pRb1 is unique. Intriguingly, no effect of FER depletion on the cellular activity or cellular levels of known CDKs, CDK activators or CDK inhibitors has been observed (22).

Thus, the potential regulatory link between FER and other modulators of pRb1 functions requires further investigation.

FER and PP1

Whereas G1/S CDKs phosphorylate pRb1 on defined sites (50-52), the protein phosphatase 1 (PP1) isoforms

(53) dephosphorylate these sites in pRb1 (54). Thus, unlike CDKs, PP1 phosphatases enhance rather than counteract the tumor-suppressive activity of pRb1. The balance between these two opposing regulatory pathways determines the net phosphorylation and activation state of pRb1. PP1 is therefore another regulator of pRb1 that might be controlled by FER and may thereby mediate the effect of FER on pRb1.

Phosphorylation of Thr320 in PP1-α suppresses the phosphatase activity of PP1-α, while increased phosphorylation of Thr320 is inversely correlated with the phosphatase activity of PP1-α, *in vivo* (55, 56, 57). Concomitantly, dephosphorylation of the Thr320 site induces the phosphatase activity of PP1-α toward pRb1 (58). Knockdown of FER does not affect the cellular levels of PP1-α, but leads to a profound decrease in the phosphorylation of PP1-α on Thr320 (22). Hence, downregulation of FER results in the induction of PP1-α phosphatase activity, a process which is expected to lead to the dephosphorylation and activation of pRb1. Conversely, overexpression of FER decreases the enzymatic activity of PP1, as determined in both *in vivo* and *in vitro* assays (22).

How could a tyrosine kinase like FER modulate the enzymatic activity of a serine/threonine phosphatase like PP1α Amino acid sequence analysis has revealed two PP1-binding motifs in the kinase domain of FER, and the association of FER and PP1-α has been verified using a co-immunoprecipitation assay. The binding of FER to PP1 depends on the presence of intact PP1-binding motifs and does not require FER kinase activity (22). It should be noted that tyrosine-phosphorylated PP1-α has not been detected in FER-expressing cells. This corroborates the notion that the regulatory effect of FER on PP1-α is kinase activity-independent and may result from the physical interaction between these two proteins. Similarly, FER binds ERK1/2 and maintains the phosphorylation states of these kinases, independently of its own tyrosine kinase activity (59). The association of FER with PP1-α might enhance the CDK-dependent phosphorylation of PP1-α on Thr320. Alternatively, FER could attenuate the auto- or transdephosphorylation of PP1-α on Thr320 (55). FER mutated in one of its PP1-binding motifs (FerF606A) has dominant negative activity, causing dephosphorylation of PP1-α and cell cycle arrest in breast carcinoma cells (22). These findings strongly suggest that PP1 is one of the key mediators of the oncogenic role of FER in malignant cells.

The function of the PP1 phosphatases is regulated by a diverse set of proteins in eukaryotic cells (60, 61). However, FER is the first intracellular tyrosine kinase shown to directly associate with PP1 and to modulate its activity. The ability of FER kinase to bind and modulate PP1 seems to be harnessed by malignant cells for manipulating the balance between "signaling" and "antisignaling" cascades in which PP1 phosphatases are involved (62).

Interestingly, unlike its role in prostate and breast carcinoma cells, FER is not essential for the proliferation of primary fibroblasts (7). Moreover, no interaction between FER and PP1 could be detected in primary mice embryonic fibroblasts (Orel *et al.,* data not shown). This suggests that the regulatory role of FER in cell cycle progression might differ between normal and malignant cells. Hence, one of the key roles of FER in malignant cells is to restrain phosphatase activities in growth-promoting pathways (62) and thereby negate the activity of the tumor suppressor pRb1 (Fig. 2).

FER and TMF/ARA160

TMF/ARA160 is a Golgi-resident protein (63) which consists of 1,093 amino acids and has an apparent molecular mass of 160 kDa (64). This protein was initially identified as a DNA-binding factor that preferentially binds to the TATA element in the HIV-1 long terminal repeat (LTR) and modulates its function (65). It was therefore termed "TATA element modulatory factor" (TMF) (65). TMF was later shown to be a potential co-activator of the androgen receptor (AR), thus endowing it with the name AR-co-activator 160 kDa (ARA160) (64). The central and *C*-terminal parts of TMF/ARA160 contain coiled-coil-forming domains that could mediate the interaction of TMF with other cellular factors. Several cellular proteins besides nuclear receptors were found to interact with TMF/ARA160 in various protein-protein binding assays. These include the Rab6 GTP-binding protein, which appears to anchor TMF/ARA160 to Golgi membranes (66), the ATPase subunits of the human chromatin remodeling complex SNF/SWI-hbrm/hSNF2-α and BRG-1/hSNF2-β (63). Furthermore, FER has been shown to cause tyrosine phosphorylation of TMF/ARA160 in *in vitro* phosphorylation assays (67). Interestingly, the Rab6-, AR- and FER-binding domains of TMF/ARA160 overlap and include coiled-coil-forming sequences (64, 67). However, these findings did not conclusively reveal the function of TMF/ARA160 *in vivo*, and its cellular role remained elusive. In a recent study, TMF/ARA160 was shown to bear a "BC-box" element (68), which exists in elongin C-binding proteins (69). This group of proteins also includes E3 ubiquitin ligases such as the VHL protein (70, 71). TMF/ARA160 was further found to recruit, under defined stress conditions, the proto-oncogene STAT3 to ubiquitination and subsequent proteasomal degradation (68). Since STAT3 is constitutively activated in various types of cancer, these findings suggest that TMF/ARA160 may have potential tumor-suppressor functions. Accordingly, TMF/ARA160 has been found to be downregulated in malignant brain tumors (68). Further work should reveal whether FER inhibits the activity of TMF/ARA160 and whether this contributes to the promalignant function of FER.

Role of FER in angiogenesis

As solid tumors expand, they can rapidly outgrow the carrying capacity of the local vasculature. Thus, tumors are often riddled with areas of lowered oxygen content;

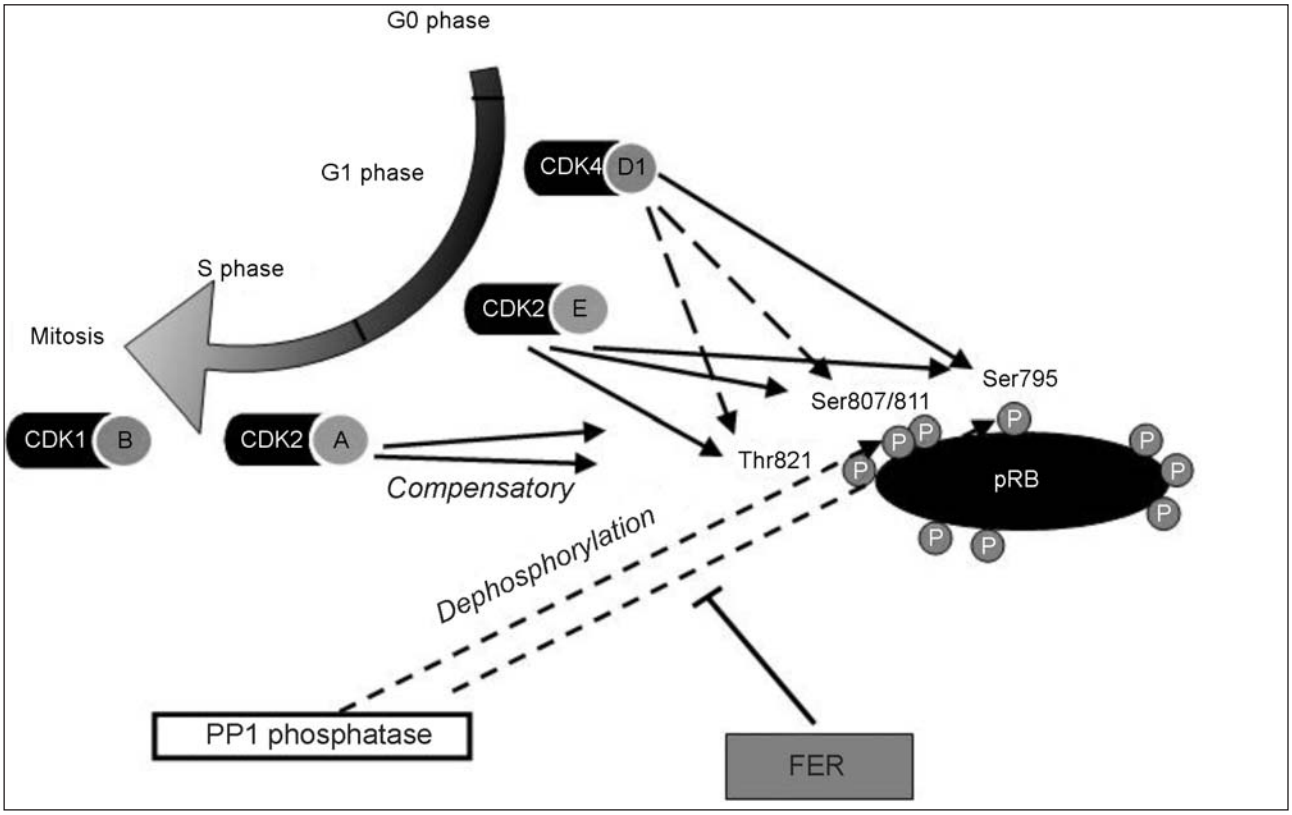

Fig. 2. Roles of FER in modulating the activities of PP1 and pRb1 in malignant cells. CDKs phosphorylate pRb1 and FER restrains the dephosphorylation activity of PP1.

this hypoxia has many therapeutic ramifications (72). Hypoxia induces the release of angiogenic factors such as vascular endothelial growth factor (VEGF), and thereby contributes to tumor vascularization and progression (73). The expression of VEGF is regulated at the transcriptional, post-transcriptional and translational levels (74). At the transcriptional level, the *vegf* gene is activated by the hypoxia-inducible transcription complex (HIF) (75). The oxygen-regulated component of this complex is named HIF-1α (76, 77). One of the regulators of HIF-1α is ERK1, which phosphorylates HIF-1α and activates its transcription-inducing activity (78-81).

FER has been shown to sustain the activation state of several mitogen-activated protein kinase (MAPK) members. These include the p38 kinase, upon activation of the Kit or FcεRI receptors in mast cells (7), and the ERK1/2 kinases in hypoxic cells (59). Furthermore, FER has been shown to associate with ERK1/2 in hypoxic cells in a manner that does not depend on the kinase activity of FER (59). Knockdown of FER induces the deactivation of ERK1/2 in hypoxic cells and a concomitant decrease in the production of VEGF by the treated cells. The dephosphorylation of ERK1/2 in FER-depleted hypoxic cells does not result from an impaired upstream signaling, but rather from direct deactivation of the ERK1/2 kinases (59). Hence, in this system as well, FER appears to modulate the activity of serine/threonine phosphatase(s) which can dephosphorylate and deactivate the ERK1/2 kinases (82). One of the ERK1/2-specific phosphatases, MKP-1, is upregulated in hypoxic cells (83-85) and the potential interaction of FER with this phosphatase should be further investigated. Interestingly, the knockdown of FER not only led to the deactivation of ERK1/2 and to the consequent deactivation of HIF-1α, but it also resulted in a significant decrease in the cellular level of the HIF-1α transcription factor (59). However, the molecular mechanism that links FER to the accumulation of HIF-1α has not been revealed. Further studies are therefore required to elucidate the molecular role of FER in VEGF production and angiogenesis. These findings implicate FER not only in the proliferation of malignant cells, but also in processes supportive of tumor growth, such as angiogenesis, which are required for tumor progression *in vivo*.

It is interesting that the two best-characterized pro-oncogenic activities of FER, namely its interactions with PP1-α and ERK, do not involve its own kinase activity. A similar kinase-independent pro-oncogenic activity has been manifested in the Abl tyrosine kinases by their kinase-independent ability to stabilize the ERK5 protein and thereby efficiently transform Rat-1 fibroblasts (86).

FER as a potential target for cancer intervention

From our current knowledge of the molecular pathways in which FER is involved in malignant cells, we can

postulate a unique regulatory role for this tyrosine kinase in cancer cells. FER appears to restrain the activity of serine/threonine phosphatases like PP1, which exert both proliferation-restricting and -promoting functions in eukaryotic cells (87-89). The dual role of PP1 precludes the ability of malignant cells to gain a growth advantage by elimination or mutation of the *pp1* genes. FER therefore provides cancer cells with a novel tool for the modulation of PP1 activity.

The involvement of PP1 phosphatases in the regulation of several transition points in the course of cell cycle progression (88, 89) also suggests a role for FER in the control of several key regulatory junctions during cell cycle progression in cancer cells. This is corroborated by

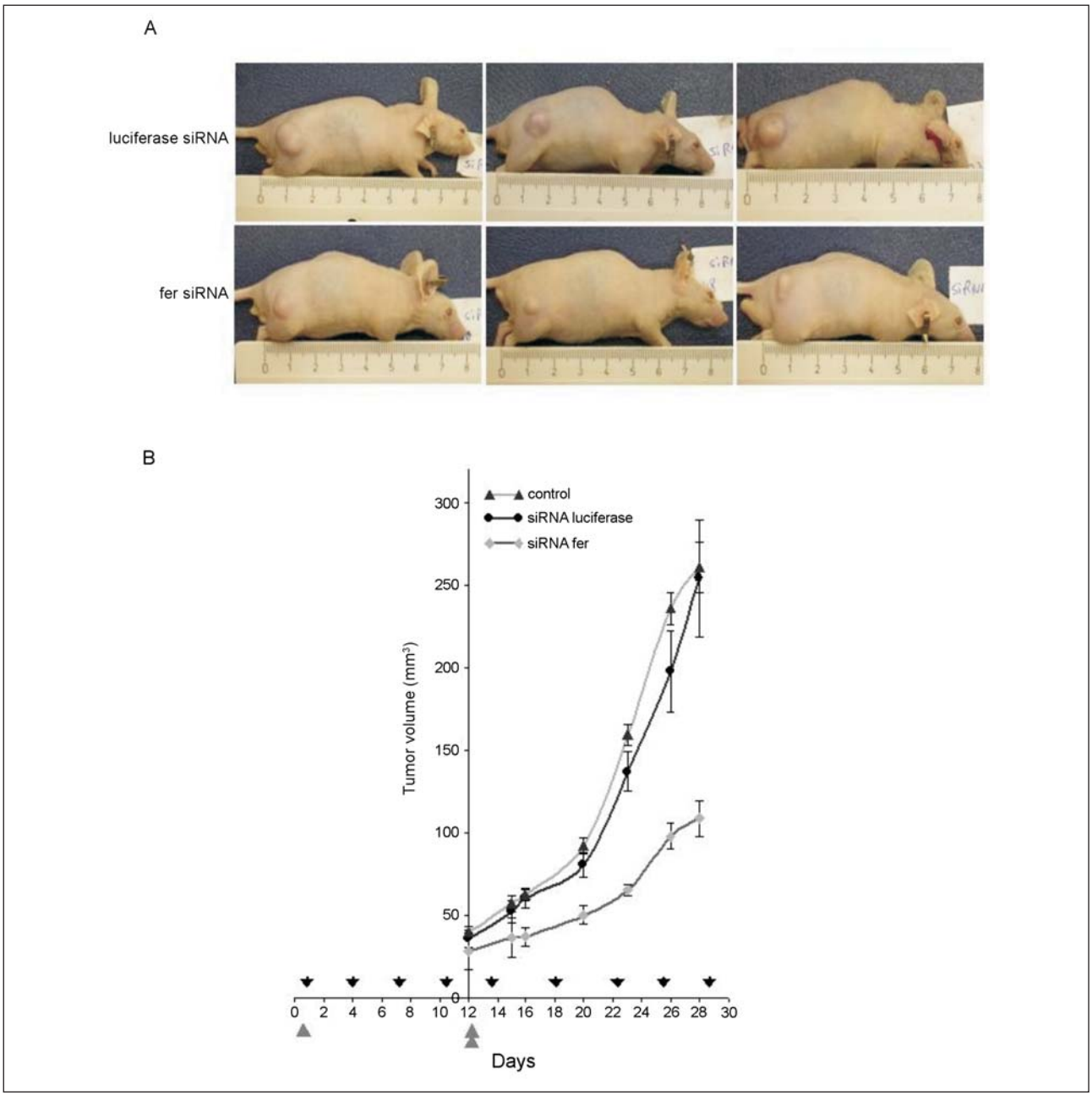

Fig. 3. fer siRNA attenuates the progression of PC-3 tumors in athymic nude mice. **A.** 10^6 PC-3 cells were subcutaneously injected into 21 athymic male mice. The animals were injected immediately thereafter with luciferase (n=7) or fer siRNA (n=7), and from then on were injected repeatedly with the siRNA samples twice a week. A control group of 7 animals were not treated with siRNA. **B.** Tumors were measured repeatedly starting 12 days after the injection of cells, using a caliber at three planes. Downward-oriented dark arrowheads indicate days of siRNA injection. The single upward-oriented grey arrowhead indicates the day of injection of the cells. Double upward-oriented grey arrowheads indicate the starting day of tumor measurement. Average values were plotted with a significance of $p < 0.01$ (Student's t-test). Bars represent standard errors (n=7).

the fact that FER is bound to chromosomes (90), implying an additional regulatory role for this kinase in the S phase progression and/or the G2-M transition. Thus, FER is engaged in the regulation and maintenance of a signaling balance at multiple regulatory stages during cell cycle progression, which could facilitate the deregulated proliferation of cancer cells. The targeting of FER should therefore interfere with the progression of tumors which exhibit deregulated G1-S and/or G2-M transitions points.

To examine whether FER can indeed serve as a therapeutic target *in vivo*, a prostate tumor xenograft model was adopted by our group (Pasder and Nir, personal communication). Prostate carcinoma PC-3 tumors were induced in athymic (nude) mice by injecting cells subcutaneously into the animals, and the xenografts were then repeatedly treated with either luciferase or fer siRNAs. The progression of xenografts subjected to the two treatments was followed by determining tumor volumes as a function of time. The growth profiles of the PC-3 tumor xenografts differed significantly between the luciferase siRNA- and fer siRNA-treated animals. Luciferase siRNA-treated tumors developed with a slope of 5.6 ($y = 5.6 X –31.4$, $r^2 = 0.9951$) between days 12 and 20 and with a slope of 21.4 ($y = 21.4 X –351.1$, $r^2 = 0.993$) between days 20 and 28 of siRNA injection. The average volume of these tumors was 254 mm^3 28 days after injection of the PC-3 cells. However, tumors injected with the fer siRNA developed with a slope of 2.7 ($y = 2.7 X –4.7$, $r^2 = 0.9818$) between days 12 and 20 and with a slope of 7.7 ($y = 7.7 X –106.7$, $r^2 = 0.9768$) between days 20 and 28 of the siRNA injection. The average volume of tumors exposed to the fer siRNA was 108 mm^3 (Fig. 3A, B). Thus, downregulation of FER significantly decreased the progression rate of the PC-3 xenografts and consequently affected their final size.

These results corroborate the potential of FER as a novel target for prostate cancer intervention. This is in accordance with the observed overexpression of FER in prostate carcinoma tumors (21). Thorough analysis of the FER expression profiles in various normal tissues and malignant tumors derived from them should further reveal in which other tumor types FER is overexpressed.

Future prospects

One strategy for translating the targeting of FER into a therapeutic approach is to identify low-molecular-weight compounds which would target FER and impair its pro-oncogenic activities. These agents could inhibit the kinase activity of FER and/or interfere with its interaction with other cellular proteins. Since some of the cellular regulatory roles of FER are kinase activity-independent, a high-throughput screening assay would need to select for low-molecular-weight compounds which both inhibit the kinase activity of FER and interfere with its interaction with other downstream effectors. The biotech start-up company UriFer is currently developing accurate and reproducible high-throughput screening assays to enable the identification of low-molecular-weight compounds that affect the functioning of FER at multiple levels in malignant cells. This should lay the ground for the development of new anticancer drugs.

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