

THE ROLE OF FOCAL-ADHESION KINASE IN CANCER — A NEW THERAPEUTIC OPPORTUNITY

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Abstract | Focal-adhesion kinase (FAK) is an important mediator of growth-factor signalling, cell proliferation, cell survival and cell migration. Given that the development of malignancy is often associated with perturbations in these processes, it is not surprising that FAK activity is altered in cancer cells. Mouse models have shown that FAK is involved in tumour formation and progression, and other studies showing that FAK expression is increased in human tumours make FAK a potentially important new therapeutic target.

Focal-adhesion kinase (FAK) resides at sites of integrin clustering — the so-called focal adhesions — that are prominent in cells that are grown in tissue culture. FAK carries out protein–protein-interaction adaptor functions at sites of cell attachment to the extracellular matrix (ECM), thereby contributing to focal-adhesion ‘scaffolding’, and also transmits adhesion-dependent and growth-factor-dependent signals into the cell interior (FIG. 1). In the cancer context, the synergistic signalling between growth-factor receptors and FAK might be particularly relevant as both are often upregulated in tumour cells. Together, the action of FAK and signalling from growth-factor receptors might control the altered growth of tumour cells as well as their responses to autocrine or paracrine factors. In addition, FAK influences the dynamic regulation of integrin-associated adhesions, and the actin cytoskeleton that is tethered there, through diverse molecular interactions. This, in turn, regulates cell migration by controlling the focal-complex assembly/disassembly cycle at the leading lamellipodia of migrating cells, while also controlling adhesion disassembly at the trailing edge. As these processes are crucial components of cell migration, and therefore also of invasion by cancer cells, FAK might well be involved in the spread of cancer cells. Focal adhesions are places from which adhesion and actin dynamics are coordinately regulated with survival

and growth signalling, at least in part, through FAK-dependent functions¹. Therefore, it is timely to review whether — and, if so, how — FAK might contribute to the development of malignancy.

Several reports have linked FAK expression with cancer. FAK mRNA was found to be increased in 49 human tissue samples, including a wide range of paired normal and neoplastic tissue samples. In this survey, increased levels of FAK were found in 1 of 8 adenomatous tissues, in 17 of 20 invasive tumours, and in all 15 metastatic tumours of different origins. However, no FAK mRNA was detected in 6 normal tissue samples², although sub-detectable levels might have been present. Another study showed that FAK protein levels were increased in 100% of colon and 88% of breast tumour samples, with FAK protein expression often being associated with advanced disease³.

Since these studies were published, there have been many reports documenting similar findings in a wide range of human malignancies. These studies, which are based mainly on immunohistochemical and immunoblotting analysis, have shown increased FAK levels in cancers of the **thyroid, prostate, cervix, colon, rectum**, oral epithelium and **ovary**^{4–12}. Increased FAK expression and activity are frequently correlated with malignant or metastatic disease and poor patient prognosis^{3,10,13,14}. Interestingly, a recent report has also

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Summary

- Focal-adhesion kinase (FAK) is a non-receptor tyrosine kinase that provides signalling and scaffolding functions at sites of integrin adhesion. It is involved in the regulation of turnover of these adhesion sites, a process that is crucial in the control of cell migration.
- FAK is linked to the protection of cells from anoikis (suspension-induced cell death). This anti-apoptotic function is potentially linked to the ability of FAK to sequester receptor-interacting protein (RIP) from the death-receptor machinery.
- Substantial circumstantial evidence has accumulated linking overexpression of FAK to a wide range of human epithelial cancers. Levels of FAK expression correlate with the invasive potential of tumours.
- Using a mouse model of skin carcinogenesis, a direct requirement for FAK has now been shown during tumour progression *in vivo*. These observations are probably linked to the ability of FAK to protect cells from apoptosis.
- Inhibition of FAK function might provide an attractive anticancer target, however it is not yet clear what the most effective strategy would be. Potential intervention routes are inhibition of the kinase activity of FAK or disruption of crucial protein–protein interactions.

highlighted a possible correlation between FAK expression and clinical outcome. In this ovarian cancer study, FAK overexpression in primary tumour biopsy material was correlated with metastasis to lymph nodes and distant organs, as well as with reduced survival times¹⁵. These findings indicate that FAK protein levels might be an appropriate prognostic indicator. By contrast, there is also one report of reduced FAK expression in

metastatic liver tumours, compared with their matched primary human colorectal adenocarcinoma samples¹⁶. So FAK might have different roles in different tumours, or during different stages of tumour progression.

Although the mechanisms that underlie the increased expression of FAK in tumour cells are not fully understood, amplification of the *FAK* gene has been reported in a few cancer cell lines, and gains in gene copy number are found in cells derived from **head and neck cancer**¹⁷.

Despite the numerous impressive correlates, experimental proof for a causative role for FAK in cancer has been lacking. However, recent evidence implies that FAK promotes tumorigenesis in an animal model. In this review, we consider key cellular properties regulated by FAK that could mediate its tumorigenic activity. This is probably linked to its well-documented ability to control cell adhesion and migration, as well as to influence cell-survival pathways. FAK is itself regulated by a range of mechanisms, including tyrosine phosphorylation, serine/threonine phosphorylation and many protein-binding interactions (these have been reviewed in detail recently¹⁴).

Regulation of FAK in cancer cells

Given the evidence that high levels of FAK are associated with human cancer, it is surprising that the *FAK* gene promoter has not been more extensively studied. However, a recent report identified binding sites for the **p53** tumour suppressor in the *FAK* promoter. These studies showed that p53 binding to this site was able to suppress expression of FAK¹⁸. This observation raises the intriguing possibility that transcriptional silencing by p53 might be involved in the normal control of FAK expression. However, it remains untested whether — and, if so, how — p53 loss or mutation contributes to altered FAK expression.

Another mode of FAK regulation that is well understood is phosphorylation, particularly tyrosine phosphorylation¹⁴. This has been reviewed thoroughly in other articles, (for example, see REF. 19). Autophosphorylation of FAK on a particular tyrosine (Y) residue, Y397, occurs in response to many stimuli, including integrin engagement, and this creates a high-affinity binding site for the SRC homology 2 (SH2) domain of several proteins including the upstream **SRC** kinase itself^{20,21}. The association of SRC with FAK leads to a conformational change and activation of the kinase activity of SRC. The ensuing phosphorylation of FAK by SRC on Y576 and Y577 within the FAK catalytic domain is required for the full enzymatic activity of FAK²². SRC can further phosphorylate FAK on Y407, Y861 and Y925 with phosphorylated Y925 acting as a docking site for growth-factor-receptor-bound protein 2 (**GRB2**; REFS 22,23), which permits signalling to the RAS–MAPK (mitogen-activated protein kinase) cascade²⁴. The FAK–SRC signalling complex acts to recruit and/or phosphorylate a number of signalling proteins and is involved in adhesion regulation and the motile and invasive phenotype, as well as in growth and survival signalling (FIG. 1).

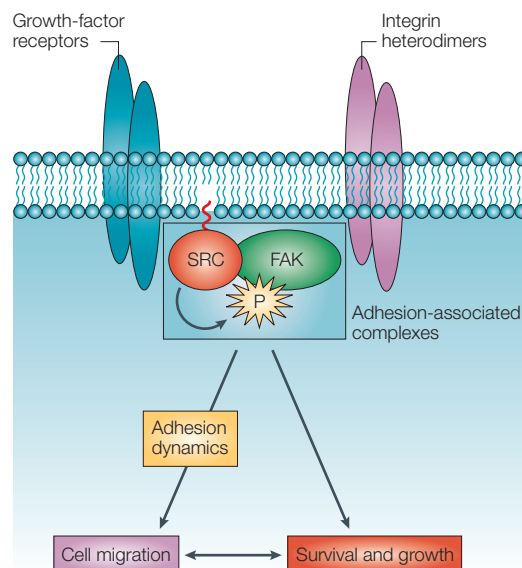


Figure 1 | Focal-adhesion kinase as a signal integrator. Focal-adhesion kinase (FAK) acts to integrate signals from extracellular cues, such as growth-factor receptors and integrins, and from the upstream SRC-family kinases, to control and coordinate adhesion dynamics/cell migration with survival signalling. SRC binds to growth-factor receptors and FAK to integrins (although, this has not been shown *in vivo*), and they bind to each other. SRC binding to growth-factor receptors is widely believed to be important, as is FAK signalling from integrins, regardless of whether the interaction between FAK and these transmembrane receptors is direct.

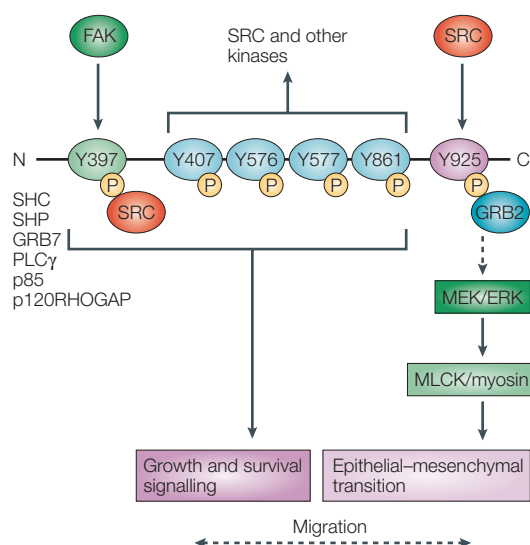


Figure 2 | Focal-adhesion kinase tyrosine phosphorylation regulates downstream signalling events. Phosphorylation of tyrosine (Y)397 is mainly due to autophosphorylation, although transphosphorylation by growth-factor receptors might also occur. This creates a high-affinity binding site for the SRC homology 2 (SH2) domain of SRC (and other SH2-domain-containing proteins), which can then phosphorylate focal-adhesion kinase (FAK) on additional tyrosine residues, of which Y925, in particular, appears to be a SRC-specific site. How signalling through increased phospho-FAK contributes to the behaviour of cancer cells is beginning to be understood. Phosphorylation of Y397 or Y925 might cause increased complex formation between FAK and its SH2-containing proteins. For example, SRC, SHC, p85 (a phosphatidylinositol 3-kinase regulatory subunit), phospholipase C γ (PLC γ), growth factor receptor-bound protein 7 (GRB7), GRB2, p120RHOGAP and others. The lower bracket indicates phosphorylation sites that are likely to mediate growth and survival signalling, probably by creating binding sites for partner proteins. In the case of the SRC-specific FAK-Y925 phospho-acceptor site, phosphorylation at this site can cause FAK exclusion from focal adhesions¹¹⁸. This site is also proposed to link FAK to the RAS–MAPK (mitogen-activated protein kinase) pathway, which is associated with SRC-induced adhesion changes that cause an epithelial–mesenchymal transition¹¹⁹ (FIG. 6). In this way, altered tyrosine phosphorylation of FAK in tumour cells could control subcellular localization, adhesion type predominance, growth and survival signalling, and cancer-cell behaviour. ERK, extracellular signal-regulated kinase; MEK, MAPK/ERK kinase; MLCK, myosin light-chain kinase.

FAK phosphorylation in cancer

Phosphorylation of FAK at specific sites has been reported to be associated with different tumour types. In ovarian tissue for example, phospho-FAK-Y397 (phosphorylation of FAK at Y397 in the FAK sequence) was found in invasive tumours, but not in normal epithelium²⁵. Other studies have also shown increases in phospho-FAK-Y397 in different tumour types^{13,26,27}, although phosphorylation of FAK-Y397 might not necessarily reflect FAK kinase activity^{28,29}. In colon cancer, phosphorylation of FAK has been linked to expression of gastrin-releasing peptide and tumour differentiation³⁰. There is also evidence that FAK

phosphorylation could regulate tumour-cell adhesion. In colon cancer cells, phosphorylation of FAK occurs on multiple sites (FIG. 2), although phosphorylation at FAK-Y925 is the major SRC-specific phosphorylation event that is associated with integrin adhesion dynamics and E-cadherin deregulation during SRC-induced epithelial–mesenchymal transition^{28,31}. This indicates that FAK-Y925 could be important in some aspects of the cancer phenotype (FIG. 2). In addition, phosphorylation of FAK-Y861 promotes association of FAK with the $\alpha_v\beta_5$ integrin following vascular endothelial growth factor stimulation³², and this could influence the tumour vasculature. Differential phosphorylation of individual FAK tyrosine residues after receipt of mitogenic or oncogenic stimuli^{29,32–35}, combined with the likelihood of kinases other than SRC can phosphorylate FAK^{28,29}, imply that more complexities will be unravelled.

FAK mechanisms of action

Cell migration. FAK is an important regulator of cell migration¹⁴ — a function required for the invasion and metastasis of cancer cells. The latter requires individual cells, or probably small groups of cells, to initially move through three-dimensional (3D) ECM around the region of the primary tumour. This often, although not always, requires the combined action of matrix metalloproteinases (MMPs), to degrade ECM barriers. However, recent evidence implies that there are also proteolysis-independent mechanisms of invasion, and it is possible that FAK might be differentially required for distinct modes of tumour-cell invasion.

FAK controls the dynamic regulation of integrin-linked adhesions (or focal adhesions), cadherin-dependent cell–cell adhesions and peripheral actin structures, and so contributes to cell migration and invasion^{14,36,37}. Its mechanism of action is complex and probably involves a web of downstream signalling connections. Recent advances in molecular intervention, combined with real-time fluorescence imaging of adhesion dynamics and actin remodelling, are beginning to imply mechanisms by which FAK can promote, and at times also suppress, cell migration.

FAK signalling might mediate the initial assembly of integrin-associated focal adhesions, which promote cell adhesion to the ECM³⁸, although maintenance of focal adhesions does not generally require FAK^{38,39}. Migration also requires the regulated turnover, or disassembly, of these adhesions and FAK also controls this process, specifically by releasing points of attachment between the cell and the surrounding ECM^{39–42}. Moreover, as the cycle of assembly and disassembly of these adhesions — that is, the ability to adhere and let go — controls the rate of cell movement, or indeed whether a cell can move or not, FAK is implicated as a key regulator of cell movement. In this regard, FAK-deficient cells exhibit larger focal adhesions at the cell periphery and migrate poorly in comparison to normal fibroblasts⁴¹.

The recent development of live-cell imaging techniques used with fluorescently labelled focal-adhesion components confirms a role for FAK in promoting

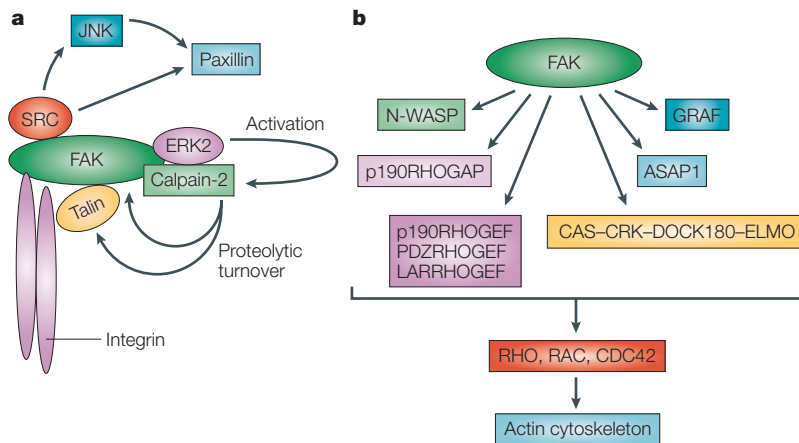


Figure 3 | Focal-adhesion kinase influences cell migration by molecular signalling pathways. **a** | During focal-adhesion turnover, integrin-mediated phosphorylation of focal-adhesion kinase (FAK) recruits a number of signalling and structural proteins, promoting the initial assembly of focal adhesions. Once phosphorylated by SRC, FAK functions as an efficient adaptor molecule that recruits both extracellular signal-regulated kinase 2 (ERK2) and calpain-2 to focal-adhesion sites. This facilitates ERK2-induced activation of calpain-2 and calpain-2-mediated cleavage of focal-adhesion components (for example, talin and FAK), resulting in focal-adhesion disassembly^{40,47}. FAK also recruits JUN N-terminal kinase (JNK) to focal-adhesion sites, and JNK-mediated phosphorylation of paxillin on a specific serine residue promotes focal-adhesion remodelling and cell motility⁵⁶. **b** | During actin remodelling, the RHO family of small GTPases — RHO, RAC and CDC42 — drive cell migration across two-dimensional substrates by controlling actin polymerization/de-polymerization and the actin-stress-fibre assembly/disassembly cycle. FAK both positively and negatively regulates the RHO-family GTPases by modulating various upstream regulators. FAK-mediated phosphorylation of the GTPase exchange factors RHO guanine nucleotide exchange factor of 190kDa (p190RHOGEF), PDZ-domain-containing RHOGEF (PDZRHOGEF) and LAR tyrosine phosphatase RHOGEF correlates with enhanced RHOA activity^{14,64,120}. FAK also modifies several GTPase-activating proteins including, ARF-GTPase-activating protein 1 (ASAP1), GTPase regulator associated with FAK (GRAF) and RHO-GTPase-activating protein of 190kDa (p190RHOGAP), which regulate activity of the RHO family^{65,66}. In addition, FAK participates in the assembly of a complex consisting of CRK-associated substrate (CAS, also known as p130cas), CRK, dedicator of cytokinesis of 180kDa (DOCK180) and engulfment and cell motility protein (ELMO), where the association of DOCK180 with ELMO might locally stimulate RAC1 activity⁶⁸. FAK-mediated phosphorylation of neuronal Wiscott–Aldrich syndrome protein (N-WASP) influences its subcellular localization and cell motility⁶⁹, probably by modifying the ability of N-WASP to promote actin polymerization through affecting the Arp2/3 complex.

focal-adhesion disassembly^{39,42}. Quantitative measurements of rate constants for focal-adhesion turnover show that FAK signalling activates the MAPK signalling pathway, as well as the protease calpain-2, which promotes focal-adhesion turnover^{39,43} (FIG. 3a). FAK therefore integrates several signalling responses that control focal-adhesion dynamics.

FAK expression, but not its kinase activity, is required for platelet-derived growth factor (PDGF)- and epidermal growth factor (EGF)-stimulated cell motility, indicating that FAK acts as an adaptor, or scaffold, to recruit molecules that promote adhesion turnover²⁹. Such a role is supported by findings that FAK adaptor function promotes the assembly of a functional complex consisting of calpain-2, FAK, SRC and ERK2¹ (FIG. 3a). Calpain-2 is a ubiquitously expressed member of the calpain family of intracellular cysteine proteases, which were first implicated in cell migration by the use of pharmacological inhibitors. These inhibitors impair the release of cell–substrate interactions at the rear of migrating cells and thereby suppress cell movement⁴⁴.

Moreover, calpain-2-mediated cleavage of focal-adhesion components, such as talin, paxillin or FAK, often occurs in parallel with focal-adhesion turnover and reduced cell–matrix adhesiveness^{45,46}. Indeed, recent work has shown that calpain-mediated cleavage of talin is a rate-limiting step in focal-adhesion turnover that is needed for cell migration⁴⁷. Also, calpain-2 is directly phosphorylated and activated by EGF-induced MAPK, and EGF-induced de-adhesion and cell migration requires MAPK-induced activation of the calpain-2 isoform at the membrane⁴⁸. Recent work (reviewed in REF. 1) has elucidated how the adaptor function of FAK promotes the motile response through MAPK and calpain-2. Such signalling events might have importance in invasion and metastasis, as increased expression and/or activity of EGFR (or its close relatives), SRC-family kinases, FAK, calpain-2 and MAPK have all been linked in some way to tumour invasion *in vivo*^{49–54}. FAK expression can also promote recruitment of JUN N-terminal kinase (JNK) to focal-contact sites after integrin stimulation⁵⁵, and JNK-mediated phosphorylation of paxillin-S178 promotes cell migration⁵⁶ (FIG. 3a). This highlights the multiple ways in which FAK can induce focal-adhesion turnover and cell migration, and indeed these might not yet all have been uncovered. In this regard, the kinase activity of FAK could also influence focal-adhesion turnover by tyrosine phosphorylation of other focal-adhesion substrates, such as CRK-associated substrate (CAS, also known as p130cas) and paxillin^{39,57–60}. Additionally, focal-adhesion dynamics are clearly also under the control of the RHO family of small GTPases (see below), which themselves are perturbed, and involved, in cancer.

So there is no doubt that FAK occupies an important position within the migration-regulatory signalling network, although it remains to be established whether the kinase activity of FAK is required in all situations, and whether FAK-dependent cell migration (as currently understood for cells in culture) is important for epithelial cancer cell migration *in vivo* (discussed in more detail below).

In addition to tumour-cell migration, FAK has been found to be expressed in angiogenic blood vessels of malignant astrocytomas, where it might contribute to angiogenesis by enabling HAPTOTACTIC MIGRATION towards ECM proteins⁶¹. Additionally, it is now known that FAK deficiency in endothelial cells (which causes vascular defects and lethality in null mice at day 8.5) impairs the normal organization of fibronectin, an important ECM component⁶². These data imply that FAK has important roles in the behaviour of endothelial cells, which, in turn, might affect tumour development.

Remodelling the actin cytoskeleton. Actin remodelling is another crucial element of the cell-motility process (key effectors of FAK in actin remodelling are depicted in FIG. 3b). Adhesion dynamics are tightly linked to control of actin assembly and disassembly and FAK contributes to both, the latter usually by influencing RHO-GTPase pathways. The contribution of RHO GTPases to actin remodelling is described in BOX 1. The fact that impaired

HAPTOTACTIC MIGRATION
Migration of cells towards fixed attractants to which the cells bind. Often used to describe movement of cells towards extracellular-matrix components mediated by binding to specific integrins.

Box 1 | The contribution of RHO GTPases to actin remodelling

The initial stages of cell adhesion and focal-complex formation are associated with activation of RAC1 and CDC42, which stimulate lamellipodia and filopodia, respectively. These processes enable membrane protrusion and cell polarization in the direction of forward movement, whereas subsequent assembly of tension-inducing actin stress fibres and mature focal adhesions are controlled by RHOA and activation of downstream effectors such as DIA1/2 or RHO-associated kinase (ROCK). The activity of the RHO-family GTPases themselves is positively regulated by guanine nucleotide exchange factors (GEFs) and negatively by GTPase-activating proteins (GAPs). The contribution of focal-adhesion kinase to actin remodelling that is needed for cell migration is mediated through binding to RHO protein effectors and subsequent effects on RHO GTPases.

motility of FAK-deficient cells could be rescued by an inhibitor of ROCK, a downstream effector of RHOA, implies that FAK works, at least in part, through the modulation of RHO proteins⁶³. Furthermore, the intrinsic activity of RHOA is increased in FAK-deficient cells⁶³, although re-expression of FAK in *Fak*^{-/-} fibroblasts decreases RHOA activity^{37,42}. These events promote actin remodelling and adhesion reorganization. However, in other cell types FAK-induced phosphorylation and activation of p190RHOGEF (guanine nucleotide exchange factor) is linked to RHOA activation^{14,64}, indicating that FAK-mediated control of RHO-GTPase regulators is complex, probably cell and context dependent, and perhaps spatially regulated. There are other examples of how FAK can influence actin remodelling. For example, FAK also interacts with ARF GTPase-activating protein 1 (ASAP1), which possesses GTPase-activating protein (GAP) activity for the ARF family of GTPases, and GTPase regulator associated with FAK (GRAF), which possesses GAP activity for RHOA and CDC42 (REFS 65,66) (FIG. 3b).

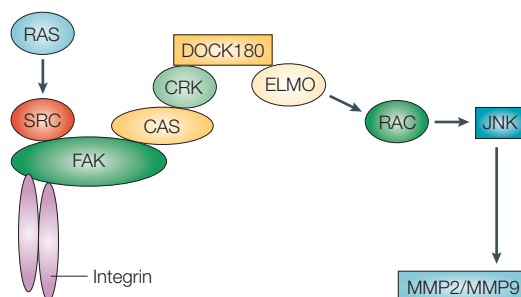


Figure 4 | Focal-adhesion kinase influences cell migration through additional molecular signalling pathways.

Additional focal-adhesion kinase (FAK)-mediated signalling events induce the expression of genes encoding matrix metalloproteinases (MMPs)¹²¹. Once MMPs are secreted, they mediate the breakdown of surrounding extracellular-matrix substrates and promote cell invasion. Recent studies on RAS-transformed cells indicate that tyrosine phosphorylation of FAK on tyrosine (Y)861 promotes association with CRK-associated substrate (CAS, also known as p130cas) and recruitment of a CRK–DOCK180–ELMO complex (where DOCK180 is a 180kDa protein that is a regulator of cytokinesis and cell motility protein) that activates RAC1, subsequently activating JUN N-terminal kinase (JNK) that promotes MMP expression and cell invasion³⁵.

Another FAK function that is important for cell migration is the assembly of a complex between FAK, CAS and the proto-oncoprotein CRK, which serves to recruit the RAC1 GEF, dedicator of cytokinesis of 180kDa (DOCK180; reviewed in REF. 67). DOCK180, when associated with its binding partner engulphment and cell motility protein (ELMO), stimulates RAC1 activity⁶⁸. So, FAK controls recruitment and activation of the CAS–CRK–DOCK180–ELMO complex, stimulating localized activation of RAC1, peripheral actin assembly and stabilization of focal complexes (FIG. 4 and reviewed in REF. 67). Moreover, as discussed below, this pathway contributes to the invasive process.

FAK probably also influences actin polymerization more directly by binding and inducing phosphorylation of the CDC42 effector protein neuronal Wiskott–Aldrich syndrome protein (N-WASP)⁶⁹. N-WASP promotes actin polymerization by activation of the Arp2/3 complex. FAK-mediated phosphorylation of N-WASP at Y256 appears to influence its intracellular localization, but it is not yet clear whether, and if so how, FAK influences N-WASP-mediated Arp2/3 activity and actin assembly, or whether this occurs in cancer cells⁶⁹ (FIG. 3b).

Invasion. FAK regulates the invasive activity of both normal and SRC-transformed fibroblasts through reconstituted 3D MATRIX^{37,70}. By contrast, a negative role for FAK during EGF-induced invasion of A431 epidermal carcinoma cells has been reported⁷¹. Although integrin-linked adhesion complexes still form in cells cultured within a 3D matrix (referred to as ‘3D-matrix adhesions’), these are structurally distinct from the large mature focal adhesions formed by cells on two-dimensional (2D) substrates⁷² (reviewed in REF. 73). In addition, phosphorylation of FAK on Y397 in cells that are growing in 3D gels is substantially reduced when compared with cells on 2D substrates⁷². In addition, phosphorylation of FAK on Y397 contributes to CYTOTROPHOBLAST migration and invasion⁷⁴. These findings indicate that FAK might be differently regulated, or have distinct roles, depending on whether cells are moving within a 3D matrix, or across a 2D planar substrate.

Although v-Src can promote transformation in *FAK*^{-/-} cells⁷⁵, FAK kinase activity is required for the efficient invasion of v-Src-transformed fibroblasts through a 3D matrix *in vitro*³⁷. Importantly, whereas FAK-deficient v-Src-transformed fibroblasts are not impaired in their ability to migrate across 2D substrates, they are defective in invading 3D matrices *in vitro*³⁷. Moreover, dominant-negative forms of FAK suppress matrix metalloproteinase 2 (MMP2) expression and activity^{70,76}, whereas anti-sense-mediated reduction in FAK also decreases expression of MMPs in carcinoma cells⁷⁷, implying a general requirement for FAK for the production and activity of MMPs that have cancer-associated matrix-degrading activities. The FAK-promoted assembly of a SRC–CAS–CRK–DOCK180 complex in

3D MATRIX

Reconstituted cell growth matrix such as Matrigel or fibrillar collagen, which is designed to mimic the *in vivo* environment encountered by tumour cells and so provide a surrogate when they are invading *in vitro*. This allows monitoring of cancer cells in culture migrating through a 3D matrix environment.

CYTOTROPHOBLAST

Part of the mammalian placenta; that is, the inner cellular layer of the trophoblast (trophoblast), between the syncytiotrophoblast and chorionic villus capillaries.

v-Src-transformed fibroblasts, results in activation of RAC1 and JNK, and consequent increased MMP2 and MMP9 expression³⁷ (FIG. 4). So, FAK regulates cell motility and invasion by distinct pathways; that is, by promoting the dynamic regulation of focal adhesions and peripheral actin structures during migration, as well as by MMP-mediated matrix degradation (reviewed in REF. 14).

Interestingly, it is now recognized that tumour cells can invade a 3D matrix by both MMP-dependent and MMP-independent mechanisms — referred to as mesenchymal-like and amoeboid-like mechanisms, respectively^{78,79} (reviewed in REF. 80). The precise roles of FAK during mesenchymal-like and amoeboid-like mechanisms of invasion by tumour cells remain to be established, although, as mentioned above, FAK has been implicated in MMP expression, indicating that FAK is probably required for mesenchymal-like invasion. By contrast, amoeboid-like invasion might be independent of this FAK function, as this mode of invasion through a 3D matrix is less dependent on integrin-mediated adhesions within the matrix⁸¹. Further studies are needed to address whether tumour cells that invade by distinct mechanisms are indeed differently dependent on FAK. But if this is the case, and tumour cells can switch between these types of invasive mechanisms, this could go some way to explaining the apparently contradictory FAK requirement for invasion by different cell types^{37,71}.

FAK in crosstalk between integrin- and cadherin-mediated adhesion. Integrin-dependent cell–matrix adhesions and cadherin-mediated cell–cell contacts can communicate with each other. Such crosstalk contributes to an altered balance between the two adhesion types, and is likely to contribute to cancer progression. Usually, cells that assemble dynamic cell–matrix contacts are more migratory and have less robust cell–cell adhesions, whereas cells that predominantly form cadherin-mediated cell–cell contacts maintain polarized epithelial-cell morphology, and typically have less robust focal-adhesion complexes. FAK has emerged as a mediator of crosstalk between integrin-mediated focal adhesions and intercellular junctions. Inhibiting signalling through FAK, or decreasing FAK expression, can either promote assembly or disassembly of cadherin-mediated cell–cell adhesions, depending on cell context and cadherin type^{31,82}. In one case, treatment of HeLa cells with short interfering RNA (siRNA) for FAK, led to aberrant membrane protrusion and deregulation of N-cadherin-mediated intercellular adhesions, both of which correlated with increased peripheral RAC1 activity⁸².

The positive effect of FAK on intercellular adhesion might be through regulation of expression, or localization, of the cadherins themselves. For example, transforming growth factor- β (TGF β) suppresses the malignant phenotype of TGF β -responsive human colon adenocarcinoma cells (Moser cells) by inducing E-cadherin expression that parallels FAK activation⁸³.

However, in endothelial cells, FAK activation is required for proper localization of E-cadherin to the cell periphery and consequent strengthening of the barrier of endothelial cells⁸⁴. So, in some cell types, FAK positively regulates cadherin-mediated cell–cell contact and maintenance of a non-migratory epithelial phenotype; however, in other cell types, as is the case with KM12C colon cancer cells that have retained E-cadherin, FAK can have a negative influence on cadherin-mediated intercellular adhesion³¹. This apparent discrepancy could be due to some cell- or context-dependent signalling from FAK to RAC1, depending perhaps on other upstream signalling inputs, such as SRC activity.

A role for FAK in the survival of tumour cells. Evidence linking FAK to cell survival was first reported by Frisch *et al.*, who showed that FAK was able to suppress suspension-induced cell death (known as ‘anoikis’) in kidney epithelial (MDCK) cells⁸⁵. The ability of FAK to do this depends on both FAK-Y397 phosphorylation and kinase activity. Overexpression of FAK also protects MDCK cells from ultraviolet-light-induced cell death, and this is linked to its association with CAS⁸⁶. Increased FAK expression in the HL60 leukaemia cell line has also been linked with suppression of apoptosis. This study showed that phosphorylation of FAK at Y397 and Y925, as well as the kinase activity of FAK, are required for the observed effects on cell survival^{87,88}. In addition, attenuation of FAK activity by either antibody injection^{89,90}, antisense oligonucleotides⁹¹ or expression of the isolated focal-adhesion targeting domain all lead to induction of apoptosis^{92–94}.

FAK inhibition has been shown to have synergistic effects with inhibition of EGF-receptor signalling, leading to apoptosis in breast cancer cells⁹⁵. Also, simultaneous inhibition of SRC and FAK robustly induces apoptosis in some colon cancer cell lines, whereas inhibition of FAK function alone more modestly induces cell death⁹⁶. The ability of FAK to protect cells from death also varies between cancer cell lines. In one breast cancer cell line, inhibition of FAK alone caused apoptosis⁹⁶, whereas two other studies have shown that expression of the FAK amino-terminus, which causes dephosphorylation of FAK-Y397 and activation of caspase-3, leads to apoptosis. However, expression of this portion of FAK in normal breast cells had no effect on cell survival^{97,98}, raising the intriguing possibility that suppressing FAK function blocks survival pathways only in malignant cells.

So, overexpression of FAK in human tumour cells might contribute to malignancy by promoting survival under conditions that would normally lead to cell death. Some recent work has elucidated a mechanism by which FAK might achieve this. Kurenova *et al.* showed that the ability of FAK to suppress apoptosis is mediated by binding to receptor-interacting protein (RIP)⁹⁹, a major component of the DEATH-RECEPTOR COMPLEX, which has been shown to interact with both FAS and tumour-necrosis factor. FAK could sequester RIP from the death-receptor

DEATH-RECEPTOR COMPLEX
A multiprotein complex involved in the cellular response to pro-apoptotic stimuli. It links cell-surface receptors to the intracellular signalling cascade that accompanies programmed cell death.

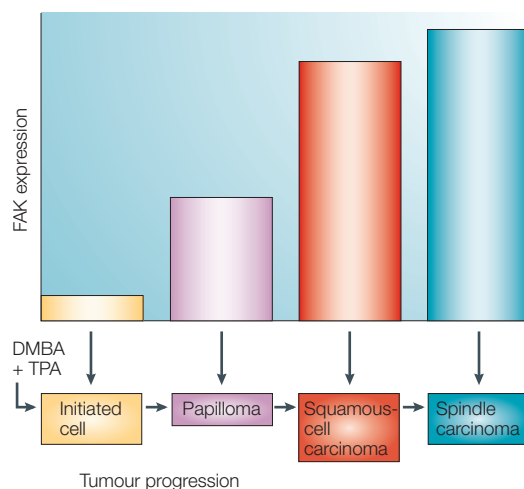


Figure 5 | Expression of focal-adhesion kinase during tumour progression in the mouse skin carcinogenesis model. Diagrammatic representation of the various stages in tumour progression that result from chemical initiation with 7,12-dimethylbenz[*a*]anthracene (DMBA) and promotion with 12-*O*-tetradecanoylphorbol-13-acetate (TPA). The relative focal-adhesion kinase (FAK) expression levels from cell lines generated from the various defined stages in tumour progression in the skin carcinogenesis model are depicted.

complex, suppressing apoptosis⁹⁹. If this proves to be a common mechanism, disrupting the interaction between FAK and RIP might be a useful intervention strategy.

The importance of loss of FAK-mediated survival to the behaviour of tumour cells is primarily two-fold. First, adhesion-dependent survival, which is often lost in tumour cells, is mediated by FAK^{85,91}. So, increased FAK expression might promote matrix-independent survival of tumour cells, which is required for invasion and metastasis. Support for this comes from studies in which FAK protein levels have been suppressed by RNA interference (RNAi), resulting in anoikis¹⁰⁰. Second, the modulation of FAK protein levels influences the sensitivity of tumour cells to various chemotherapeutic agents^{13,87,101} and ultraviolet irradiation⁸⁶. Again, this could be linked to the ability of FAK to protect against anoikis. In support of this, signalling from $\beta 1$ -integrins is involved in matrix-dependent protection from drug-induced apoptosis in small-cell lung cancer cells¹⁰². In acute myeloid leukaemia cells, however, the increased sensitization of cells to daunorubicin following decreased FAK expression is not dependent on the matrix¹³. So, although the exact mechanisms by which FAK contributes to aberrant survival of tumour cells have yet to be worked out, the level of FAK protein in tumour cells might contribute to clinical outcome, particularly the response to therapeutic regimens¹³.

A role for FAK in the growth of tumour cells. Several studies have indicated that FAK has a direct role in tumour growth. For example, inhibition of FAK

signalling downstream of the urokinase receptor, by use of the well-characterized dominant-negative FAK variant, FAK-related non-kinase, suppresses proliferation of Hep3 cells *in vivo*¹⁰³. Induction of dormancy correlates with reduced signalling through FAK, RAS and extracellular signal-regulated kinase (ERK)–MAPK, and dormancy can be reversed by expression of an active mutant of MEK1 (MAPK/ERK kinase 1). These findings indicate that FAK signalling through the ERK–MAPK pathway is required to maintain the growth of at least some tumour cells¹⁰³. Interestingly, FAK signalling through SHC to the RAS and ERK–MAPK pathway has also been shown in anaplastic astrocytoma samples, and this correlates with proliferation of tumour cells¹⁰⁴, although in U251MG astrocytoma cells in culture, FAK forms a complex with p120RASGAP, leading to increased RAS activity associated with proliferation of tumour cells¹⁰⁵.

FAK levels affect carcinogenesis

Most of the evidence linking FAK to tumorigenesis has been circumstantial, positioning FAK ‘at the scene of the crime’. However, there is now evidence that FAK mediates both tumour formation and malignant progression. In a study using the two stage 7,12-dimethylbenz[*a*]anthracene/12-*O*-tetradecanoylphorbol-13-acetate (DMBA/TPA) MOUSE CARCINOGENESIS MODEL (described in REF. 106), FAK expression is increased in a stepwise fashion in cell lines derived from various stages of the mouse skin carcinogenesis model (FIG. 5). Furthermore, phosphorylation of FAK at Y925, which has been identified as an important site for phosphorylation by SRC, is increased in several malignant tumours that were examined from this model (G.M., unpublished observations).

When *Fak*^{+/-} mice are analysed in the same skin carcinogenesis model, the reduced FAK expression is sufficient to impair papilloma formation, compared with wild-type controls¹⁰⁷. However, although this work demonstrated a clear link between the level of FAK protein expression and the propensity to form benign tumours, it was not possible to assess the role of FAK in the malignant progression of papillomas to squamous-cell carcinomas, because the levels of FAK protein were increased in papillomas from both *Fak*^{+/-} and wild-type mice to a similar extent¹⁰⁷. As a result, there was no visible difference in the conversion rates between *Fak*^{+/-} and wild-type mice¹⁰⁷. These findings indicated that there is a selection pressure that raises FAK expression during tumour formation.

Using an INDUCIBLE Cre-LOX SYSTEM to delete FAK expression specifically in epidermal cells and in the hair follicles^{108,109}, papilloma formation is significantly reduced. When papillomas did form in the FAK-deleted skin, conversion to squamous-cell carcinomas was also substantially reduced¹⁰⁹. These findings provide compelling evidence that FAK contributes to both tumour formation, and the acquisition of malignancy, at least in this model¹⁰⁹.

DMBA/TPA MOUSE SKIN CARCINOGENESIS MODEL
Two-stage chemical carcinogenesis model that progresses from normal skin to benign papillomas to invasive tumours through several well-characterized stages. An initial treatment with DMBA serves as the tumour initiator followed by treatment with TPA as the promoter during tumour formation.

INDUCIBLE Cre-LOX SYSTEM
Method for the introduction of genetic modifications into specific genes by homologous recombination using Cre, a site-specific bacteriophage-P1-derived recombinase. The Cre recombinase cuts at the LOXP-tagged genes.

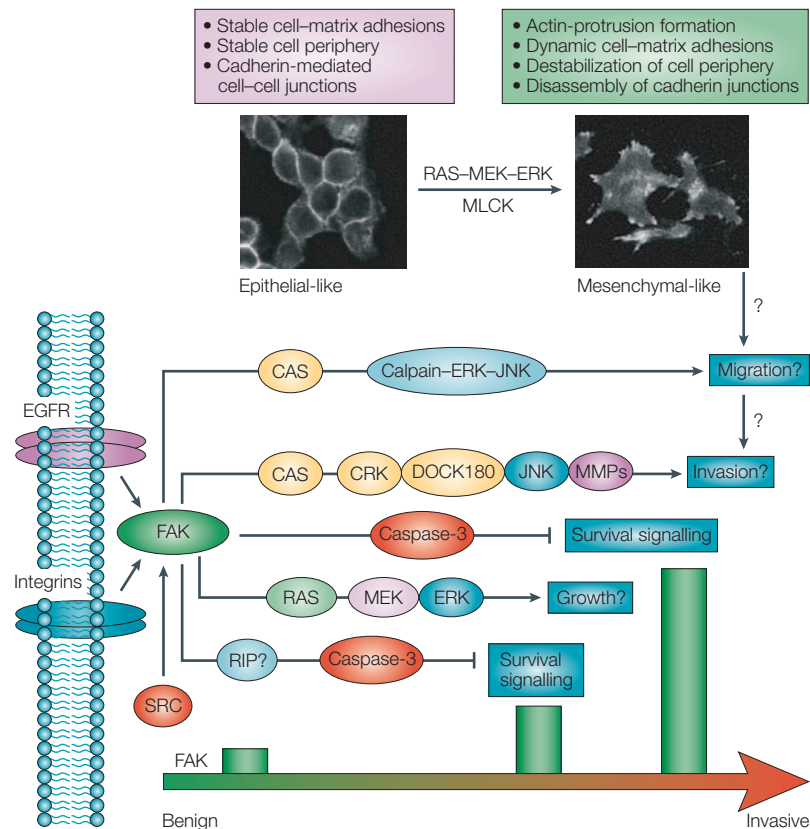


Figure 6 | Model of the possible contributions of focal-adhesion kinase in cancer development. Phosphorylation of focal-adhesion kinase (FAK) by SRC not only regulates its kinase activity and localization, but also the formation of phosphorylation-dependent protein complexes, integrin- and E-cadherin-mediated adhesions, and cellular motility and invasion. FAK-Y925 phosphorylation is particularly associated with SRC-induced adhesion changes associated with the epithelial-mesenchymal transition (FIG. 1). KM12C colon cancer cells stained with anti-E-cadherin (left panel) or anti-paxillin (right panel) indicate the morphological differences associated with phosphorylation of FAK¹¹⁹. These effects cause an epithelial-mesenchymal transition and could contribute to migration of cancer cells, perhaps at the tumour edges, although this has not been shown. Depicted is FAK-mediated induction of the invasive pathway involving signalling to RAC1 and JUN N-terminal kinase (JNK) and matrix metalloproteinases (MMPs). Shown also is the clear role of FAK in preventing apoptosis downstream of integrin or growth-factor-receptor signalling, as judged by its requirement to keep caspase-3 inactive, the latter associated with tumour formation and progression in a mouse model¹⁰⁹. The role proposed for FAK in contributing to growth through the RAS-MAPK (mitogen-activated protein kinase) pathway is also shown. ERK, extracellular signal-regulated kinase; EGFR, epidermal growth factor receptor; DOCK180, dedicator of cytokinesis of 180kDa; MEK, MAPK/ERK kinase; MLCK, myosin light-chain kinase; RIP, receptor-interacting protein.

The role of FAK in skin carcinogenesis model

The role of FAK as a regulator of migration and cell survival prompted testing of which, if either, of these activities is associated with tumorigenesis in the skin carcinogenesis model¹⁰⁹. These activities were investigated *in vitro* (using an inducible model of expression in mouse keratinocytes) and *in vivo* by examining mouse skin. *Fak*-null keratinocytes are unable to repopulate a disrupted monolayer like normal keratinocytes. This is consistent with reduced cell migration, and the tracking of individual *Fak*-deficient keratinocytes by time-lapse microscopy confirmed reduced rates of migration (by about 50%), although *Fak*-null cells were still visibly motile¹⁰⁹. However, fewer *Fak*-null cells also remained in the monolayer, consistent with detachment and/or

cell death. Collection of adherent and detached keratinocytes revealed that FAK loss of expression leads to aberrant cell-cycle profiles and cell death¹⁰⁹.

In mice, *FAK*-deficient keratinocytes are capable of re-forming the epithelium of wounded skin¹⁰⁹. This observation is surprising, as it seems at odds with the extensive literature from *in vitro* experiments describing a key role for FAK in cell migration (reviewed in REFS 14,110) and, indeed, evidence from the same study that *FAK*-deficient keratinocytes display impaired migration *in vitro*¹⁰⁹. These apparently paradoxical findings probably reflect a difference between cells migrating as isolated cells with no cell-cell contact, and the forward movement of epithelial sheets to repair wounded skin *in vivo*. In this regard, it is known that FAK can promote N-cadherin-mediated intercellular adhesion, and suppress migration of epithelial sheets, by downregulating peripheral RAC1 activity⁸².

Although there was no obvious effect of *FAK* deletion on the epithelial repair of wounds in mouse skin, we cannot rule out that *FAK*-dependent influences on some types of tumour-cell migration — perhaps of isolated cells or small groups of cells that have undergone a mesenchymal transition at the leading edges of migrating sheets, or at the edges of tumours — might have a role in invasion of malignant cells. However, these findings highlight the importance of developing physiologically relevant 3D *in vitro* culture systems, at least for the movement of epithelial sheets, and for testing the effects of molecular or pharmacological intervention in modulating the migration of cells *in vivo*.

In contrast to the disparity between the *in vitro* and *in vivo* migration results, *FAK*-deficient keratinocytes did have increased rates of cell death both *in vitro* and *in vivo*. Increased levels of activated caspase-3 staining, a reliable marker of apoptosis in tissue sections^{111,112}, are present in both skin and papillomas from *FAK*-deficient mice when compared to normal controls¹⁰⁹. Interestingly, in skin, caspase-3 staining is strongest in cells of the hair follicles, where most target cells for DMBA-induced carcinogenesis probably reside¹¹³. Taken together, these data are consistent with *FAK*-dependent survival signalling being required for tumour formation and progression in the DMBA/TPA mouse skin carcinogenesis model¹⁰⁹.

Future directions

FAK has been causally implicated in tumour development *in vivo*, albeit in a single mouse carcinogenesis model (FIG. 5). Much more work needs to be done to fully determine the role of FAK in tumorigenesis, which might vary in different tumour types. For example, as discussed in this article, contributing effects could come from cell-cycle and survival signalling, effects on cadherins or other cell-adhesion molecules, on migration and invasion of mesenchymal-like cells, on MMP production, and on other cancer-associated processes (FIG. 6). The causal role of FAK in tumour development, combined with reports that increased FAK expression is associated with poor clinical outcome^{13,114}, indicate that FAK might be a useful therapeutic target³⁴.

There is considerable interest and effort now in the development of tyrosine-kinase inhibitors as cancer treatments. However, in the case of FAK, the requirement for kinase activity in the development of cancer has not been proven — therefore, it is not clear whether targeting this domain will have any therapeutic benefit. Indeed, studies in cell lines have demonstrated that the ability of FAK to regulate cell motility and invasion cannot be wholly attributed its kinase activity^{29,71,115}. By contrast, the kinase activity of FAK does seem to be required for adhesion-regulated survival⁸⁵. It is important now to determine whether this is required for the contribution of FAK to tumour development. Although no inhibitors of FAK are currently being evaluated in clinical trials, patents have been taken out on several FAK-inhibitory compounds; however, no animal studies using these compounds have been reported so far. These compounds are based largely on pyrimidine derivatives that are ATP competitors. It is also not clear whether continued FAK expression and activity is required for maintenance of the tumour phenotype — this is likely to vary between tumour types, and is an important factor in determining the clinical use of FAK kinase inhibitors.

In addition to its kinase activity, FAK is also an important molecular adaptor that regulates the dynamics of multiprotein complexes. So it might be possible to specifically block the interaction between FAK and its key binding partners as a therapeutic strategy. For example, the interaction between signalling effectors and important FAK phosphotyrosine residues, such as Y397 and Y925, or with the SH3-binding proline-rich sequences of FAK, could be targeted. Such approaches have been used for other signalling proteins, including SRC and GRB2. Inhibitors of the SRC SH2 and SH3 domains have been based on modifications of cognate peptide sequences, or have been identified by screening of non-peptide compounds based on the preferred binding sequences of the SRC SH2 and SH3 domains¹¹⁶.

However, this approach has recognized problems, due to relatively low-affinity interactions, promiscuity, and problems with delivery and recognized redundancy in achieving signalling end points. For these reasons, such strategies do not yet have a high success rate, but the possibility of achieving specificity with limited toxicity means that these strategies should continue to be pursued. There have been some promising results with inhibitors of GRB2 protein–protein interactions. For example, SH2 inhibitors that suppress GRB2 binding to the EGF receptor prevent downstream activation of RAS signalling and growth of tumour cells¹¹⁷. By

analogy, specifically targeting particular adaptor functions of FAK might be an interesting approach.

In the case of FAK, there is some evidence that RNAi might be a valuable therapeutic tool. Systemic administration of FAK siRNA to mice results in inhibition of metastasis in a model of pancreatic cancer¹⁰⁰. Furthermore, FAK siRNA potentiates gemcitabine-induced cytotoxicity *in vivo*¹⁰¹, raising the possibility that these therapeutic approaches could be potentially useful in combination with conventional cytotoxic agents.

Although current data implicate the survival function of FAK as an important contributor to tumour formation and progression in a mouse skin carcinogenesis model¹⁰⁹, there remains considerable circumstantial evidence implicating FAK as a regulator of cancer-cell motility and invasion. Indeed, it is likely that the kinase and/or adaptor functions of FAK contribute to both positive and negative outcomes regarding cancer-cell motility and invasion that are context dependent. These outcomes are perhaps controlled by tumour cell type or the localized production of motogenic growth factors, as well as production of the ECM substrates and expression and activities of the integrin receptors involved.

Further studies in additional tumour models are required to determine the range of physiological functions of FAK in different tumour types, and particularly to assess whether FAK has a role in metastasis. Increasing our knowledge of where, and when, the diverse functions of FAK are required during cancer progression, and pursuing the development of clinically relevant biomarkers of the biochemical and biological activities of FAK, will aid the design and future evaluation of therapeutic strategies that target FAK function.

Note added in proof

A very recently published paper by Van de Water and colleagues has demonstrated that FAK is required for metastasis in a syngeneic rat model¹²². Specifically, inducible expression of an inhibitory FAK protein, FAK-related non-kinase (FRNK), in MTLn3 mammary adenocarcinoma cells, suppressed the growth of primary tumours and blocked metastasis formation in the lungs. Importantly, in this case, FRNK expression was linked to adhesion and migration defects, rather than induction of apoptosis. This work implies that impaired signalling through FAK can eliminate the targeting of tumour cells to the lungs - most likely by blocking the adhesion and migration of breast cancer cells, processes that are probably required for establishment of distant metastasis - and confirms that FAK can contribute to tumour development in diverse ways.

1. Carragher, N. O. & Frame, M. C. Focal adhesion and actin dynamics: a place where kinases and proteases meet to promote invasion. *Trends Cell Biol.* **14**, 241–249 (2004).
2. Weiner, T. M., Liu, E. T., Craven, R. J. & Cance, W. G. Expression of focal adhesion kinase gene and invasive cancer. *Lancet* **342**, 1024–1025 (1993).
3. Owens, L. V. *et al.* Overexpression of the focal adhesion kinase (p125^{FAK}) in invasive human tumours. *Cancer Res.* **55**, 2752–2755 (1995).

4. Owens, L. V. *et al.* Focal adhesion kinase as a marker of invasive potential in differentiated human thyroid cancer. *Ann. Surg. Oncol.* **3**, 100–105 (1996).
5. Tremblay, L. *et al.* Focal adhesion kinase (pp125^{FAK}) expression, activation and association with paxillin and p50^{CRK} in human metastatic prostate carcinoma. *Int. J. Cancer* **68**, 164–171 (1996).
6. McCormack, S. J., Brazinski, S. E., Moore, J. L. Jr, Werness, B. A. & Goldstein, D. J. Activation of the focal

- adhesion kinase signal transduction pathway in cervical carcinoma cell lines and human genital epithelial cells immortalized with human papillomavirus type 18. *Oncogene* **15**, 265–274 (1997).
7. Kornberg, L. J. Focal adhesion kinase expression in oral cancers. *Head Neck* **20**, 634–639 (1998).
8. Kornberg, L. J. Focal adhesion kinase and its potential involvement in tumour invasion and metastasis. *Head Neck* **20**, 745–752 (1998).

9. Judson, P. L., He, X., Cance, W. G. & Van Le, L. Overexpression of focal adhesion kinase, a protein tyrosine kinase, in ovarian carcinoma. *Cancer* **86**, 1551–1556 (1999).
10. Cance, W. G. *et al.* Immunohistochemical analyses of focal adhesion kinase expression in benign and malignant human breast and colon tissues: correlation with preinvasive and invasive phenotypes. *Clin. Cancer Res.* **6**, 2417–2423 (2000).
11. Lark, A. L. *et al.* Overexpression of focal adhesion kinase in primary colorectal carcinomas and colorectal liver metastases: immunohistochemistry and real-time PCR analyses. *Clin. Cancer Res.* **9**, 215–222 (2003).
12. Gabriel, B. *et al.* Focal adhesion kinase interacts with the transcriptional co-activator FHL2 and both are overexpressed in epithelial ovarian cancer. *Anticancer Res.* **24**, 921–927 (2004).
13. Recher, C. *et al.* Expression of focal adhesion kinase in acute myeloid leukemia is associated with enhanced blast migration, increased cellularity, and poor prognosis. *Cancer Res.* **64**, 3191–3197 (2004).
14. Schlaepfer, D. D., Mitra, S. K. & Ilic, D. Control of motile and invasive cell phenotypes by focal adhesion kinase. *Biochim. Biophys. Acta* **1692**, 77–102 (2004).
15. Sood, A. K. *et al.* Biological significance of focal adhesion kinase in ovarian cancer: role in migration and invasion. *Am. J. Pathol.* **165**, 1087–1095 (2004).
16. Ayaki, M. *et al.* Reduced expression of focal adhesion kinase in liver metastases compared with matched primary human colorectal adenocarcinomas. *Clin. Cancer Res.* **7**, 3106–3112 (2001).
17. Agochiya, M. *et al.* Increased dosage and amplification of the focal adhesion kinase gene in human cancer cells. *Oncogene* **18**, 5646–5653 (1999).
18. Golubovskaya, V., Kaur, A. & Cance, W. Cloning and characterization of the promoter region of human focal adhesion kinase gene: nuclear factor κ B and p53 binding sites. *Biochim. Biophys. Acta* **1678**, 111–125 (2004).
19. Mitra, S. K., Hanson, D. A. & Schlaepfer, D. D. Focal adhesion kinase: in command and control of cell motility. *Nature Rev. Mol. Cell Biol.* **6**, 56–68 (2005).
20. Schlaepfer, D. D., Hanks, S. K., Hunter, T. & van der Geer, P. Integrin-mediated signal transduction linked to Ras pathway by GRB2 binding to focal adhesion kinase. *Nature* **372**, 786–791 (1994).
21. Schaller, M. D. *et al.* Autophosphorylation of the focal adhesion kinase, pp125FAK, directs SH2-dependent binding of pp60src. *Mol. Cell. Biol.* **14**, 1680–1688 (1994).
22. Calalb, M. B., Polte, T. R. & Hanks, S. K. Tyrosine phosphorylation of focal adhesion kinase at sites in the catalytic domain regulates kinase activity: a role for Src family kinases. *Mol. Cell. Biol.* **15**, 954–963 (1995).
23. Calalb, M. B., Zhang, X., Polte, T. R. & Hanks, S. K. Focal adhesion kinase tyrosine-861 is a major site of phosphorylation by Src. *Biochem. Biophys. Res. Commun.* **228**, 662–668 (1996).
24. Schlaepfer, D. D. & Hunter, T. Evidence for *in vivo* phosphorylation of the Grb2 SH2-domain binding site on focal adhesion kinase by Src-family protein-tyrosine kinases. *Mol. Cell. Biol.* **16**, 5623–5633 (1996).
25. Grisar-Granovsky, S. *et al.* Differential expression of Protease activated receptor 1 (Par1) and pY397FAK in benign and malignant human ovarian tissue samples. *Int. J. Cancer* **113**, 372–378 (2004).
26. Aronson, M. S., Brown, H. M., Hauptman, G. & Kornberg, L. J. Expression of focal adhesion kinase and phosphorylated focal adhesion kinase in squamous cell carcinoma of the larynx. *Laryngoscope* **113**, 1944–1948 (2003).
27. Moon, H. S., Park, W. I., Choi, E. A., Chung, H. W. & Kim, S. C. The expression and tyrosine phosphorylation of E-cadherin/catenin adhesion complex, and focal adhesion kinase in invasive cervical carcinomas. *Int. J. Gynecol. Cancer* **13**, 640–646 (2003).
28. Brunton, V. G. *et al.* Identification of Src-specific phosphorylation site on FAK: dissection of the role of Src SH2 and catalytic functions and their consequences for tumour cell behaviour. *Cancer Res.* **65**, 1335–1342 (2005).
29. Sieg, D. J. *et al.* FAK integrates growth-factor and integrin signals to promote cell migration. *Nature Cell Biol.* **2**, 249–256 (2000).
30. Matkowskyj, K. A. *et al.* Expression of GRP and its receptor in well-differentiated colon cancer cells correlates with the presence of focal adhesion kinase phosphorylated at tyrosines 397 and 407. *J. Histochem. Cytochem.* **51**, 1041–1048 (2003).
31. Avizienyte, E. *et al.* Src-induced de-regulation of E-cadherin in colon cancer cells requires integrin signalling. *Nature Cell Biol.* **4**, 632–638 (2002).
32. **Demonstrated a requirement for SRC-mediated phosphorylation of FAK in the deregulation of E-cadherin-based cell-cell junctions in colon cancer cells. This paper linked integrin-induced signals to adhesion changes at cell junctions and provides evidence that FAK signalling could be important for the disassembly of cell-cell junctions that is associated with the epithelial-mesenchymal transition, and therefore perhaps invasion.**
33. Eliceiri, B. P. *et al.* Src-mediated coupling of focal adhesion kinase to integrin α v β 5 in vascular endothelial growth factor signalling. *J. Cell Biol.* **157**, 149–160 (2002).
34. Chen, H. C., Chan, P. C., Tang, M. J., Cheng, C. H. & Chang, T. J. Tyrosine phosphorylation of focal adhesion kinase stimulated by hepatocyte growth factor leads to mitogen-activated protein kinase activation. *J. Biol. Chem.* **273**, 25777–25782 (1998).
35. Nakamura, K., Yano, H., Schaefer, E. & Sabe, H. Different modes and qualities of tyrosine phosphorylation of FAK and Pyk2 during epithelial-mesenchymal transdifferentiation and cell migration: analysis of specific phosphorylation events using site-directed antibodies. *Oncogene* **20**, 2626–2635 (2001).
36. Lim, Y. *et al.* Phosphorylation of focal adhesion kinase at tyrosine 861 is crucial for Ras transformation of fibroblasts. *J. Biol. Chem.* **279**, 29060–29065 (2004).
37. Parsons, J. T., Martin, K. H., Slack, J. K., Taylor, J. M. & Weed, S. A. Focal adhesion kinase: a regulator of focal adhesion dynamics and cell movement. *Oncogene* **19**, 5606–5613 (2000).
38. Hsia, D. A. *et al.* Differential regulation of cell motility and invasion by FAK. *J. Cell Biol.* **160**, 753–767 (2003).
39. **Demonstrates that FAK regulates both cell motility and invasion by distinct signalling pathways. Whereas v-Src expression in FAK^{-/-} fibroblasts was sufficient to restore migration defects, FAK re-expression and the formation of a FAK–SRC–CAS–DOCK180 complex was necessary to promote invasion.**
40. Richardson, A. & Parsons, T. A mechanism for regulation of the adhesion-associated protein tyrosine kinase pp125^{FAK}. *Nature* **380**, 538–540 (1996).
41. Webb, D. J. *et al.* FAK–Src signalling through paxillin, ERK and MLCK regulates adhesion disassembly. *Nature Cell Biol.* **6**, 154–161 (2004).
42. Carragher, N. O., Westhoff, M. A., Fincham, V. J., Schaller, M. D. & Frame, M. C. A novel role for FAK as a protease-targeting adaptor protein: regulation by p42 ERK and Src. *Curr. Biol.* **13**, 1442–1450 (2003).
43. Ilic, D. *et al.* Reduced cell motility and enhanced focal adhesion contact formation in cells from FAK-deficient mice. *Nature* **377**, 539–544 (1995).
44. Ren, X. D. *et al.* Focal adhesion kinase suppresses Rho activity to promote focal adhesion turnover. *J. Cell Sci.* **113**, 3673–3678 (2000).
45. Franco, S., Perrin, B. & Huttenlocher, A. Isoform specific function of calpain 2 in regulating membrane protrusion. *Exp. Cell Res.* **299**, 179–187 (2004).
46. Huttenlocher, A. *et al.* Regulation of cell migration by the calcium-dependent protease calpain. *J. Biol. Chem.* **272**, 32719–32722 (1997).
47. Carragher, N. O., Levkau, B., Ross, R. & Raines, E. W. Degraded collagen fragments promote rapid disassembly of smooth muscle focal adhesions that correlates with cleavage of pp125^{FAK}, paxillin, and talin. *J. Cell Biol.* **147**, 619–630 (1999).
48. Carragher, N. O., Fincham, V. J., Riley, D. & Frame, M. C. Cleavage of focal adhesion kinase by different proteases during SRC-regulated transformation and apoptosis. Distinct roles for calpain and caspases. *J. Biol. Chem.* **276**, 4270–4275 (2001).
49. Franco, S. J. *et al.* Calpain-mediated proteolysis of talin regulates adhesion dynamics. *Nature Cell Biol.* **6**, 977–983 (2004).
50. Glading, A. *et al.* Epidermal growth factor activates m-calpain (calpain II), at least in part, by extracellular signal-regulated kinase-mediated phosphorylation. *Mol. Cell. Biol.* **24**, 2499–2512 (2004).
51. Westhoff, M. A., Serrels, B., Fincham, V. J., Frame, M. C. & Carragher, N. O. SRC-mediated phosphorylation of focal adhesion kinase couples actin and adhesion dynamics to survival signalling. *Mol. Cell. Biol.* **24**, 8113–8133 (2004).
52. Sebolt-Leopold, J. S. Development of anticancer drugs targeting the MAP kinase pathway. *Oncogene* **19**, 6594–6599 (2000).
53. Mamouni, A. *et al.* DU145 human prostate carcinoma invasiveness is modulated by urokinase receptor (uPAR) downstream of epidermal growth factor receptor (EGFR) signalling. *Exp. Cell Res.* **299**, 91–100 (2004).
54. Braun, C. *et al.* Expression of calpain I messenger RNA in human renal cell carcinoma: correlation with lymph node metastasis and histological type. *Int. J. Cancer* **84**, 6–9 (1999).
55. Frame, M. C. Src in cancer: deregulation and consequences for cell behaviour. *Biochim. Biophys. Acta* **1602**, 114–130 (2002).
56. McLean, G. W., Avizienyte, E. & Frame, M. C. Focal adhesion kinase as a potential target in oncology. *Expert Opin. Pharmacother.* **4**, 227–234 (2003).
57. Almeida, E. A. *et al.* Matrix survival signalling: from fibronectin via focal adhesion kinase to c-Jun NH₂-terminal kinase. *J. Cell Biol.* **149**, 741–754 (2000).
58. Huang, C., Rajfur, Z., Borchers, C., Schaller, M. D. & Jacobson, K. JNK phosphorylates paxillin and regulates cell migration. *Nature* **424**, 219–223 (2003).
59. Subauste, M. C. *et al.* Vinculin modulation of paxillin–FAK interactions regulates ERK to control survival and motility. *J. Cell Biol.* **165**, 371–381 (2004).
60. Schaller, M. D. Biochemical signals and biological responses elicited by the focal adhesion kinase. *Biochim. Biophys. Acta* **1540**, 1–21 (2001).
61. Cary, L. A., Han, D. C., Polte, T. R., Hanks, S. K. & Guan, J. L. Identification of p130^{cas} as a mediator of focal adhesion kinase-promoted cell migration. *J. Cell Biol.* **140**, 211–221 (1998).
62. Sieg, D. J., Hauck, C. R. & Schlaepfer, D. D. Required role of focal adhesion kinase (FAK) for integrin-stimulated cell migration. *J. Cell Sci.* **112**, 2677–2691 (1999).
63. Haskell, H. *et al.* Focal adhesion kinase is expressed in the angiogenic blood vessels of malignant astrocytic tumours *in vivo* and promotes capillary tube formation of brain microvascular endothelial cells. *Clin. Cancer Res.* **9**, 2157–2165 (2003).
64. Ilic, D. *et al.* FAK promotes organization of fibronectin matrix and fibrillar adhesions. *J. Cell Sci.* **117**, 177–187 (2004).
65. Chen, B. H., Tzen, J. T., Bresnick, A. R. & Chen, H. C. Roles of Rho-associated kinase and myosin light chain kinase in morphological and migratory defects of focal adhesion kinase-null cells. *J. Biol. Chem.* **277**, 33857–33863 (2002).
66. Zhai, J. *et al.* Direct interaction of focal adhesion kinase with p190RhoGEF. *J. Biol. Chem.* **278**, 24865–24873 (2003).
67. Liu, Y., Lojens, J. C., Martin, K. H., Karginov, A. V. & Parsons, J. T. The association of ASAP1, an ADP ribosylation factor-GTPase activating protein, with focal adhesion kinase contributes to the process of focal adhesion assembly. *Mol. Biol. Cell* **13**, 2147–2156 (2002).
68. Hildebrand, J. D., Taylor, J. M. & Parsons, J. T. An SH3 domain-containing GTPase-activating protein for Rho and Cdc42 associates with focal adhesion kinase. *Mol. Cell. Biol.* **16**, 3169–3178 (1996).
69. Parsons, J. T. Focal adhesion kinase: the first ten years. *J. Cell Sci.* **116**, 1409–1416 (2003).
70. Brugnera, E. *et al.* Unconventional Rac-GEF activity is mediated through the Dock180–ELMO complex. *Nature Cell Biol.* **4**, 574–582 (2002).
71. Wu, X., Suetsugu, S., Cooper, L. A., Takenawa, T. & Guan, J. L. Focal adhesion kinase regulation of N-WASP subcellular localization and function. *J. Biol. Chem.* **279**, 9565–9576 (2004).
72. Hauck, C. R., Hsia, D. A., Ilic, D. & Schlaepfer, D. D. v-Src SH3-enhanced interaction with focal adhesion kinase at β 1 integrin-containing invadopodia promotes cell invasion. *J. Biol. Chem.* **277**, 12487–12490 (2002).
73. Lu, Z., Jiang, G., Blume-Jensen, P. & Hunter, T. Epidermal growth factor-induced tumour cell invasion and metastasis initiated by dephosphorylation and downregulation of focal adhesion kinase. *Mol. Cell. Biol.* **21**, 4016–4031 (2001).
74. Cukierman, E., Pankov, R., Stevens, D. R. & Yamada, K. M. Taking cell–matrix adhesions to the third dimension. *Science* **294**, 1708–1712 (2001).
75. Yamada, K. M., Pankov, R. & Cukierman, E. Dimensions and dynamics in integrin function. *Braz. J. Med. Biol. Res.* **36**, 959–966 (2003).
76. Ilic, D. *et al.* Plasma membrane-associated pY397FAK is a marker of cytotrophoblast invasion *in vivo* and *in vitro*. *Am. J. Pathol.* **159**, 93–108 (2001).
77. **Showed that FAK was phosphorylated on Y397 during cytotrophoblast invasion of surrounding ECM. Knocking down FAK expression suppressed**

- cytotrophoblast invasion, showing that FAK signalling, and autophosphorylation in particular, is also associated with normal invasive processes.**
75. Moissoglio, K. & Gelman, I. H. v-Src rescues actin-based cytoskeletal architecture and cell motility and induces enhanced anchorage independence during oncogenic transformation of focal adhesion kinase-null fibroblasts. *J. Biol. Chem.* **278**, 47946–47959 (2003).
 76. Zhang, Y. *et al.* A role for focal adhesion kinase in hyaluronan-dependent MMP-2 secretion in a human small-cell lung carcinoma cell line, QG90. *Biochem. Biophys. Res. Commun.* **290**, 1123–1127 (2002).
 77. Shibata, K. *et al.* Both focal adhesion kinase and c-Ras are required for the enhanced matrix metalloproteinase 9 secretion by fibronectin in ovarian cancer cells. *Cancer Res.* **58**, 900–903 (1998).
 78. Wolf, K. *et al.* Compensation mechanism in tumour cell migration: mesenchymal-amoeboid transition after blocking of pericellular proteolysis. *J. Cell Biol.* **160**, 267–277 (2003).
 79. Sahai, E. & Marshall, C. J. Differing modes of tumour cell invasion have distinct requirements for Rho/ROCK signalling and extracellular proteolysis. *Nature Cell Biol.* **5**, 711–719 (2003).
 80. Friedl, P. & Wolf, K. Tumour-cell invasion and migration: diversity and escape mechanisms. *Nature Rev. Cancer* **3**, 362–374 (2003).
 81. Friedl, P. Prespecification and plasticity: shifting mechanisms of cell migration. *Curr. Opin. Cell Biol.* **16**, 14–23 (2004).
 82. Yano, H. *et al.* Roles played by a subset of integrin signalling molecules in cadherin-based cell–cell adhesion. *J. Cell Biol.* **166**, 283–295 (2004).
 83. Wang, H., Radjendirane, V., Wary, K. K. & Chakrabarty, S. Transforming growth factor β regulates cell–cell adhesion through extracellular matrix remodeling and activation of focal adhesion kinase in human colon carcinoma Moser cells. *Oncogene* **23**, 5558–5561 (2004).
 84. Quadri, S. K., Bhattacharjee, M., Parthasarathi, K., Tanita, T. & Bhattacharya, J. Endothelial barrier strengthening by activation of focal adhesion kinase. *J. Biol. Chem.* **278**, 13342–13349 (2003).
 85. Frisch, S. M., Vuori, K., Ruoslahti, E. & Chan-Hui, P. Y. Control of adhesion-dependent cell survival by focal adhesion kinase. *J. Cell Biol.* **134**, 793–799 (1996).
 86. Chan, P. C. *et al.* Suppression of ultraviolet irradiation-induced apoptosis by overexpression of focal adhesion kinase in Madin–Darby canine kidney cells. *J. Biol. Chem.* **274**, 26901–26906 (1999).
 87. Sonoda, Y. *et al.* Anti-apoptotic role of focal adhesion kinase (FAK). Induction of inhibitor-of-apoptosis proteins and apoptosis suppression by the overexpression of FAK in a human leukemic cell line, HL-60. *J. Biol. Chem.* **275**, 16309–16315 (2000).
 88. Sakurai, S. *et al.* Mutated focal adhesion kinase induces apoptosis in a human glioma cell line, T98G. *Biochem. Biophys. Res. Commun.* **293**, 174–181 (2002).
 89. Liu, X. J. *et al.* Apoptosis of rat hepatic stellate cells induced by anti-focal adhesion kinase antibody. *World J. Gastroenterol.* **8**, 734–738 (2002).
 90. Hungerford, J. E., Compton, M. T., Matter, M. L., Hoffstrom, B. G. & Otey, C. A. Inhibition of pp125FAK in cultured fibroblasts results in apoptosis. *J. Cell Biol.* **135**, 1383–1390 (1996).
 91. Xu, L. H. *et al.* Attenuation of the expression of the focal adhesion kinase induces apoptosis in tumour cells. *Cell Growth Differ.* **7**, 413–418 (1996).
 92. van de Water, B., Houtepen, F., Huigsloot, M. & Tijdens, I. B. Suppression of chemically induced apoptosis but not necrosis of renal proximal tubular epithelial (LLC-PK1) cells by focal adhesion kinase (FAK). Role of FAK in maintaining focal adhesion organization after acute renal cell injury. *J. Biol. Chem.* **276**, 36183–36193 (2001).
 93. Jones, G., Machado, J. Jr, Tolnay, M. & Merlo, A. PTEN-independent induction of caspase-mediated cell death and reduced invasion by the focal adhesion targeting domain (FAT) in human astrocytic brain tumours which highly express focal adhesion kinase (FAK). *Cancer Res.* **61**, 5688–5691 (2001).
 94. Ilic, D. *et al.* Extracellular matrix survival signals transduced by focal adhesion kinase suppress p53-mediated apoptosis. *J. Cell Biol.* **143**, 547–560 (1998).
 95. Golubovskaya, V. *et al.* Dual inhibition of focal adhesion kinase and epidermal growth factor receptor pathways cooperatively induces death receptor-mediated apoptosis in human breast cancer cells. *J. Biol. Chem.* **277**, 38978–38987 (2002).
 96. Golubovskaya, V. M. *et al.* Simultaneous inhibition of focal adhesion kinase and SRC enhances detachment and apoptosis in colon cancer cell lines. *Mol. Cancer Res.* **1**, 755–764 (2003).
 97. Xu, L. H. *et al.* The focal adhesion kinase suppresses transformation-associated, anchorage-independent apoptosis in human breast cancer cells. Involvement of death receptor-related signalling pathways. *J. Biol. Chem.* **275**, 30597–30604 (2000).
 98. Bevilacqua, L. *et al.* Focal adhesion kinase N-terminus in breast carcinoma cells induces rounding, detachment and apoptosis. *Biochem. J.* **373**, 201–210 (2003).
 99. Kurenova, E. *et al.* Focal adhesion kinase suppresses apoptosis by binding to the death domain of receptor-interacting protein. *Mol. Cell Biol.* **24**, 4361–4371 (2004).
- Provides new mechanistic insight into the role of FAK as a suppressor of apoptosis by providing evidence of a link between FAK and the death-receptor complex. FAK is shown to bind to RIP (a pro-apoptotic member of the death-receptor complex) and inhibit its function, so providing a possible link between the overexpression of FAK found in many tumours and aberrantly regulated survival signalling.**
100. Duxbury, M. S., Ito, H., Zinner, M. J., Ashley, S. W. & Whang, E. E. Focal adhesion kinase gene silencing promotes anoikis and suppresses metastasis of human pancreatic adenocarcinoma cells. *Surgery* **135**, 555–562 (2004).
- Highlights the possible therapeutic benefit of FAK gene silencing. In pancreatic cell lines FAK RNAi resulted in death of anoikis-resistant resistant cell lines, and in suppression of metastasis.**
101. Duxbury, M. S. *et al.* RNA interference targeting focal adhesion kinase enhances pancreatic adenocarcinoma gemcitabine chemosensitivity. *Biochem. Biophys. Res. Commun.* **311**, 786–792 (2003).
 102. Sethi, T. *et al.* Extracellular matrix proteins protect small cell lung cancer cells against apoptosis: a mechanism for small cell lung cancer growth and drug resistance *in vivo*. *Nature Med.* **5**, 662–668 (1999).
 103. Aguirre Ghiso, J. A. Inhibition of FAK signalling activated by urokinase receptor induces dormancy in human carcinoma cells *in vivo*. *Oncogene* **21**, 2513–2524 (2002).
 104. Hecker, T. P., Grammer, J. R., Gillespie, G. Y., Stewart, J. Jr & Gladson, C. L. Focal adhesion kinase enhances signalling through the Shc/extracellular signal-regulated kinase pathway in anaplastic astrocytoma tumour biopsy samples. *Cancer Res.* **62**, 2699–2707 (2002).
 105. Hecker, T. P., Ding, Q., Rege, T. A., Hanks, S. K. & Gladson, C. L. Overexpression of FAK promotes Ras activity through the formation of a FAK/p120RasGAP complex in malignant astrocytoma cells. *Oncogene* **23**, 3962–3971 (2004).
 106. Quintanilla, M., Brown, K., Ramsden, M. & Balmain, A. Carcinogen-specific mutation and amplification of Ha-ras during mouse skin carcinogenesis. *Nature* **322**, 78–80 (1986).
 107. McLean, G. W. *et al.* Decreased focal adhesion kinase suppresses papilloma formation during experimental mouse skin carcinogenesis. *Cancer Res.* **61**, 8385–8389 (2001).
 108. Indra, A. K. *et al.* Temporally-controlled site-specific mutagenesis in the basal layer of the epidermis: comparison of the recombinase activity of the tamoxifen-inducible Cre-ER(T) and Cre-ER(T2) recombinases. *Nucleic Acids Res.* **27**, 4324–4327 (1999).
 109. McLean, G. W. *et al.* Specific deletion of focal adhesion kinase suppresses tumour formation and blocks malignant progression. *Genes Dev.* **18**, 2998–3003 (2004).
- Using a mouse skin carcinogenesis model and skin-specific conditional FAK deletion, this report provides the first *in vivo* evidence for a causative role for FAK during tumour progression. Cre-LOX driven FAK ablation both reduced benign papilloma formation and blocked progression to malignant squamous-cell carcinoma, an effect that was shown to be linked to the anti-apoptotic function of FAK.**
110. Hanks, S. K., Ryzhova, L., Shin, N. Y. & Brabek, J. Focal adhesion kinase signalling activities and their implications in the control of cell survival and motility. *Front Biosci.* **8**, 982–996 (2003).
 111. Duan, W. R. *et al.* Comparison of immunohistochemistry for activated caspase-3 and cleaved cytokeratin 18 with the TUNEL method for quantification of apoptosis in histological sections of PC-3 subcutaneous xenografts. *J. Pathol.* **199**, 221–228 (2003).
 112. Marshman, E., Ottewill, P. D., Potten, C. S. & Watson, A. J. Caspase activation during spontaneous and radiation-induced apoptosis in the murine intestine. *J. Pathol.* **195**, 285–292 (2001).
 113. Argyris, T. S. Tumour promotion by abrasion induced epidermal hyperplasia in the skin of mice. *J. Invest. Dermatol.* **75**, 360–362 (1980).
 114. Miyazaki, T. *et al.* FAK overexpression is correlated with tumour invasiveness and lymph node metastasis in oesophageal squamous cell carcinoma. *Br. J. Cancer* **89**, 140–145 (2003).
 115. Cary, L. A., Chang, J. F. & Guan, J. L. Stimulation of cell migration by overexpression of focal adhesion kinase and its association with Src and Fyn. *J. Cell Sci.* **109**, 1787–1794 (1996).
 116. Sawyer, T., Boyce, B., Dalgarno, D. & Iulucci, J. Src inhibitors: genomics to therapeutics. *Expert Opin. Investig. Drugs* **10**, 1327–1344 (2001).
 117. Gay, B. *et al.* Selective GRB2 SH2 inhibitors as anti-Ras therapy. *Int. J. Cancer* **83**, 235–241 (1999).
 118. Katz, B. Z. *et al.* Targeting membrane-localized focal adhesion kinase to focal adhesions: roles of tyrosine phosphorylation and SRC family kinases. *J. Biol. Chem.* **278**, 29115–29120 (2003).
 119. Avizienyte, E., Fincham, V. J., Brunton, V. G. & Frame, M. C. Src SH3/2 domain-mediated peripheral accumulation of Src and phospho-myosin is linked to deregulation of E-cadherin and the epithelial–mesenchymal transition. *Mol. Biol. Cell* **15**, 2794–2803 (2004).
 120. Chikumi, H., Fukuhara, S. & Gutkind, J. S. Regulation of G protein-linked guanine nucleotide exchange factors for Rho, PDZ-RhoGEF, and LARG by tyrosine phosphorylation: evidence of a role for focal adhesion kinase. *J. Biol. Chem.* **277**, 12463–12473 (2002).
 121. Hauck, C. R., Hsia, D. A., Puente, X. S., Cheresch, D. A. & Schlaepfer, D. D. FRNK blocks v-Src-stimulated invasion and experimental metastases without effects on cell motility or growth. *EMBO J.* **21**, 6289–6302 (2002).
 122. van Nimwegen, M. J., Verkoelen, S., van Buren, L., Burg, D. & van de Water, B. Requirement for focal adhesion kinase in the early phase of mammary adenocarcinoma lung metastasis formation. *Cancer Res.* **65**, 4698–4706 (2005).

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Competing interests statement

The authors declare no competing financial interests.

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