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REVIEW ARTICLE

Cell signaling in cancer

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Summary

The pathways that control cell differentiation and growth are almost always compromised in cancer. Although in the last years there have been major advances in understanding these changes and how they contribute to tumor initiation and growth, the task is far from complete. In this review we discuss some of the factors that are found in major key nodes of the signaling pathways. Included among them are the nuclear factor κB (NF-κB) that has a quite central role in inflammation, and the mammalian target of rapamycin (mTOR) kinase. Also an eminent role is played by the EGF (epidermal growth factor) and the Snail/Slug family of repressors. Since the expansion of tumor cells depends heavily on nutrient and oxygen supply, it requires the growth of new blood vasculature which is directed by the hypoxia inducible factor (HIF).

Key words: cancer, cell, signaling

Inflammation and NF-κB

The fact that chronic inflammation can lead to the development of malignancy was suggested as early as the 19th century [1]. Epidemiological studies have linked chronic inflammation as a major risk factor for various cancers [2,3], like hepatocellular carcinoma or gastric cancers [4], chronic inflammatory bowel diseases that significantly increase the risk for colorectal cancer [5], or respiratory system irritation due to various causes that increase the risk for lung cancer [6].

To find the link between chronic inflammation and the development of malignancy, the cellular processes that lead to neoplasia must be taken into account. These are, according to Hanahan and Weinberg [7], evasion of apoptosis, self-sufficiency in growth signals, insensitivity to growth-inhibition signals, limitless replication, tissue invasion and metastasis and sustained angiogenesis.

It is generally accepted that the NF-κB group of transcription factors play a central role in inflammation [8]. These factors are assembled by dimerization of 5 subunits: RelA (p65), c-Rel, RelB, p50/NF-κB1 and p52/NF-κB2 [9]. c-Rel is the homologue of the v-rel oncogene. All these protein subunits have the same DNA-binding domain [10]. When unstimulated, the dimers are localized in the cytoplasm bound to specific inhibitors (inhibitors of NF-κB-IκB). When the cells are stimulated, a specific kinase phosphorylates these inhibitors, targeting them for ubiquitination and degradation [11]. The free dimers translocate to the nucleus where they transcriptionally activate a multitude of other genes [12-14].

Another pathway in NF-κB signaling is the activation of p52: RelB heterodimers [15]. Activation of a specific kinase results in the degradation of the precursor of p52/NF-κB2 (which keeps itself and RelB in the cytoplasm), thereby allowing the p52: RelB heterodimers to enter the nucleus. A third pathway involves the activation of casein kinase 2, which causes the degradation of IκBs (inhibitors of NF-κBs) [16].

NF-κB can either promote or inhibit carcinogenesis, depending on the cell type it acts on. Inflammation can enhance tumor development by influencing factors that are involved either in malignant progression or

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repair mechanisms. Organisms that cause infection and inflammation affect the host through pattern-recognition receptors (PRR), like the Toll-like receptors (TLR) [17]. PRRs can activate many signal-transduction pathways that control the transcription of genes for chemokines, cytokines and other enzymes [18,19]. Some of these proteins activate other receptors that amplify the inflammation. Although there are many molecules that play important roles in the activation of signal-transduction pathways, it is accepted that the NF-κB group of transcription factors play a central role in inflammation [8] and in tumor promotion, since it is an important activator for the expression of many anti-apoptotic genes [20-23].

NF-κB in cancer

Colitis-associated cancer is a disease that occurs in patients suffering from ulcerative colitis. When mice are injected with the procarcinogen azoxymethane this cancer develops with low incidence which can be augmented by exposure to dextran-sulfate sodium salt, an irritant that causes colitis (colonic inflammation) by eroding the intestinal mucosa [24]. Inactivation of the *Ikbkb* (IκB kinase β-subunit, IKKβ) gene resulted in 80% decrease in colitis-associated cancer incidence [25] without a corresponding decrease in tumor size. This fact implies that IKKβ-dependent NF-κB in enterocytes affects tumor initiation but not tumor growth.

Another model of inflammation-caused cancer in mice is the hepatocellular carcinoma in multidrug-resistance 2 (*Mdr2*) knockout mice. The absence of the MDR2 transporter causes accumulation of bile acids and phospholipids in hepatocytes, which results in liver inflammation and eventually to hepatocellular carcinoma [26]. Inhibition of NF-κB by expression of the IκB-superrepressor (an inhibitor of NF-κB that is resistant to degradation) blocks tumor development [27].

Another model of NF-κB-associated cancer is a skin carcinogenesis model caused by exposure to 7,12-dimethylbenzanthracene (DMBA) as tumor initiator plus TPA as tumor promoter. When NF-κB in keratinocytes was inhibited, the multiplicity of squamous cell carcinomas caused by the exposure to the above agents increased [28].

mTOR in cancer

mTOR is a conserved serine/threonine kinase that receives both mitogenic and nutrient signals. It controls cell growth in response to growth factors and environmental inputs, like the availability of glucose, oxygen and aminoacids. Very important upstream activators of mTOR are the Ras and phosphatidylinositol-3-OH kinase (PI3K) proteins that both converge to mTOR [29] and stimulate cell growth. There are two separate TOR complexes, mTORC1 and mTORC2. mTORC1 is involved in transcription and translation and is strongly inhibited by rapamycin. mTORC2 is involved in the control of cell growth [30]. Before we further analyze the role of mTOR in cancer we should elaborate on the role of some of its effectors, like Ras, PI3K and AKT.

A direct effector of Ras is the Raf-1 serine/threonine kinase [29] which is an extracellular signal regulated kinase (ERK) that is linked to the mitogen-activated protein kinase (MAPK) pathway.

The PI3K pathway is believed to be responsible for about 30% of all human cancers [31]. The PI3K pathway is activated by receptor tyrosine-kinases (RTK) and Ras, which in turn activates other signaling pathways using the second messenger phosphatidylinositol-3,4,5-triphosphate [$PI(3,4,5)P_3$]. The AKT protein (or protein kinase B, PKB) is an important target of $PI(3,4,5)P_3$ in human cancers.

AKT regulates cell survival, cell-cycle progression, cell metabolism and growth by phosphorylating and controlling various substrates, like the glycogen synthase kinase-3, that is inhibited by AKT phosphorylation [32]. AKT also stimulates glycolysis by phosphorylating phosphofructokinase-2 (PFK2) and the synthesis of fatty acids by phosphorylation of ATP citrate lyase [33]. Another substrate of AKT is MDM2, that degrades the p53 tumor suppressor [34]. AKT also inactivates the pro-apoptotic protein BAD [35,36] and activates the IKK pathway [37].

An important target of AKT that is involved in the regulation of mTOR is the TSC2 tumor suppressor protein tuberin [38], which negatively regulates mTOR by acting as a GTPase-activating protein for the Rheb GTPase [30]. Tuberin is also a direct substrate of ERK [39,40] as well as the ribosomal protein S6 kinase (RSK) [41].

A case in which abnormal regulation of mTOR can lead to cancer is in cells that lack LKB1, a kinase that phosphorylates and activates the AMP-activated protein kinase (AMPK) [42,43]. When AMP levels rise and ATP levels drop (e.g. in hypoxia or glucose starvation), AMP binds to AMPK causing its phosphorylation by LKB1 [44], which causes the inhibition of mTOR. In cells that lack LKB1, the AMPK cannot be phosphorylated and therefore mTOR remains active [45]. This can lead to the development of various types of cancers.

EGF in cancer

EGF is an extracellular signaling molecule, important for cell survival [46] which binds to 4 related tyrosine kinase receptors, ErbB1/EGFR, ErbB2/Her2/Neu, ErbB3/HER3 and ErbB4/HER4 [47], with subsequent dimerization and activation of the receptors [48], which leads to their phosphorylation in the C-terminal end by the tyrosine kinase domain. This phosphorylation is followed by the binding of proteins that have the SH2 (Src homology 2) domain. When these proteins bind to the receptors they associate with various other proteins resulting in the activation of other serine-threonine kinases that can in turn phosphorylate other proteins kinases and transcription factors, leading to a kinase cascade [49]. This cascade amplifies various signaling pathways, thereby changing the function of a diverse range of proteins and activating transcription, thus interacting with the apoptotic pathways and preventing cell death.

Since a necessary step towards cancer development is the prevention of apoptosis, the EGF, which is a strong survival signal, is exploited by tumor cells [50]. The ErbB receptors are implicated in breast, head and neck, lung, glioma and renal carcinomas [51]. In about 25% of breast cancers ErbB2 expression is increased, leading to increased aggressiveness of the tumor and poor prognosis [47]. In gliomas the ErbB1 gene is often mutated causing activation of the tyrosine kinase without the presence of EGF, which also leads to more aggressive tumor forms [52].

Apart from alterations in the EGF receptors and their ligands, EGF is indirectly implicated in many other forms of cancer through the compromise of intracellular signaling pathways that are regulated by EGF. In some breast and lung cancers the RAS/ERK pathway is deregulated by mutations in Ras that increase the signal of ERK [53]. In other cancers, like breast and glioblastomas, mutations are found in the tumor suppressor PTEN that dephosphorylates $PI(3,4,5)P_3$ [54].

The Snail/Slug family of repressors

Members of the Snail/Slug family play an important role in the development and cancer [55], acting as transcriptional repressors. Fibroblast growth factor (FGF) is necessary for the expression of mSnail in mice, thereby controlling the expression of E-cadherin. The Snail family has an anti-apoptotic role [56]. However, there are many other observations that point to the Snail family as positive regulators for apoptosis. In chicken limb development cSnail is induced in interdigital areas in limb outgrowth. In an NBT-II rat carcinoma epithelial cell culture, when the cells were transfected with antisense Slug cDNA they became resistant to epithelial mesenchymal induction by FGF [57]. In the same cells, the EGF activated Src and Ras. When A431 epidermoid carcinoma cells were treated with EGF the Snail expression levels were elevated [58].

Hypoxia in cancer

When cells are grown *in vitro*, the supply of nutrients is usually uninterrupted. On the other hand, when cells grow *in vivo*, they must constantly monitor the environment for availability of nutrients and adjust their metabolic balance to compensate for any shortage. When tumors grow at a fast rate, this rapid growth results in tumor cells being deprived of oxygen and nutrients since they are far away from the existing vasculature. The tumor cells send signals that can induce the formation of new blood vessels, a process called angiogenesis. Angiogenesis is found in every tissue, but in solid tumors it is often greatly enhanced. The resulting tumor vessels are not normal, as in angiogenesis in normal tissues, but they are malformed as a result of the fast growth pace. Sensing of the levels of oxygen is critical in the control of vasculogenesis [59].

At the center of this regulatory system is HIF which controls the expression of vascular endothelial growth factor-A (VEFG-A) [60] and angiopoietin-2 [61]. Although the former two products are the most important ones that HIF controls, they are not the only ones. HIF induces many other factors that control metabolism, survival, vascularization and intracellular pH. It is established that HIF promotes tumor growth strongly [62]. Apart from oxygen homeostasis, HIF can induce pro-apoptotic genes in hypoxic tissues.

HIF is a heterodimer of a stable HIF-1β subunit that is expressed regardless of the oxygen levels and one of 3 oxygen-regulated subunits (HIF-1α, 2α or 3α). Although HIF is the primary mediator in hypoxia, it is not the director sensor of oxygen levels. The primary oxygen sensors is a class of iron-dependent non-haem dioxygenases, whose activity depends directly on the cellular oxygen tension [63]. There are 2 types of these oxygen sensors that control the action of HIF. The first are enzymes that hydroxylate 2 prolyl residues in the HIF-1α subunit. This hydroxylation causes the ubiquitination of HIF-1α by the E3 ubiquitin ligase complex [64] that drives HIF-1 to destruction by the proteasome. When cells are well oxygenated the half-life of HIF-1α in less than 5 min [65]. The second class of oxygen sensors is an asparaginyl hydroxylase that is referred to as Factor Inhibiting HIF-1 (FIH-1) [66].

It hydroxylates an asparagine residue of HIF-1α in the carboxy-terminal transcriptional activation domain (C-TAD), which in turn inhibits the interaction of C-TAD with transcriptional co-activators. Under hypoxic conditions the HIF subunits and consequently HIF are stabilized and bind to hypoxia-response elements (HRE) in the nucleus with the consensus sequence RCGTG that are near the genes responsible for the response to hypoxia (more that 100 genes have been characterized so far).

The expression of VEGF-A is controlled at transcription, mRNA stability and translation [67] (Figure 1). The HIF-1 heterodimer binds to and enhances the transcription of the VEGF-A mRNA. However, HIF is not the only activator of VEGF-A transcription. There are two other important transcriptional controls through the Ras-ERK and the PI(3)K-AKT pathways [68,69].

Another important angiogenic factor that is induced by HIF is angiopoietin-2. Under normal conditions the capillaries are in a dormant state by recruiting pericytes (PC) through the platelet-derived growth factor-B (PDGF-B) and the signaling of the Tie-2 receptor that is activated by the binding of angiopoietin-1. Angiopoietin-2 is induced under hypoxic conditions and it is an antagonist for the Tie-2 receptor (Figure 2). When signaling by Tie-2 is blocked, the VEGF-A can induce cell migration and division with consequent sprouting angiogenesis [67].

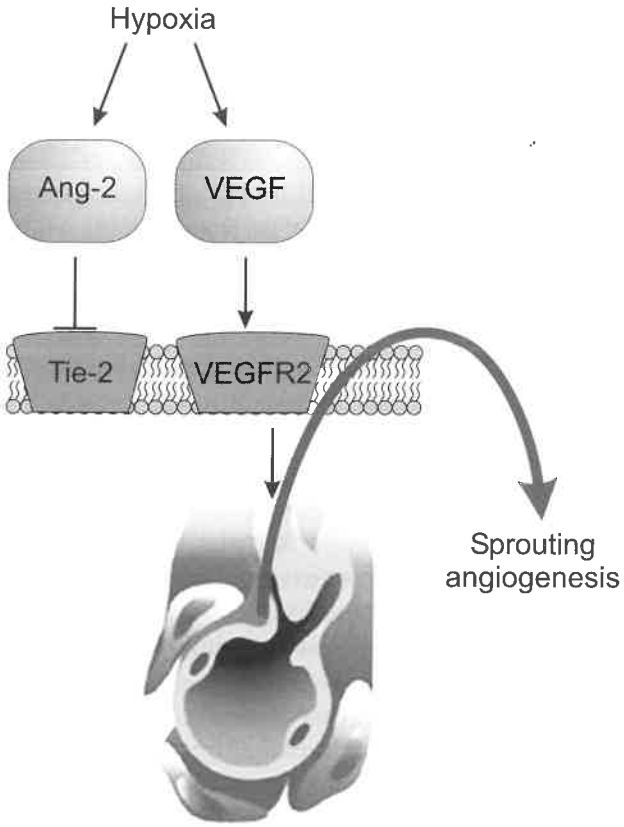


Figure 2. Induction of angiogenesis by angiopoietin-2 and VEGF-A [67]. Ang-2: angiopoietin-2, VEGF: vascular endothelial growth factor, VEGFR2: VEGF receptor-2, Tie-2: endothelial receptor Tie-2.

Conclusions

Although our understanding of the cell signaling pathways has improved greatly over the last years, there still remain significant challenges. In order to perturb a signaling pathway so as to use it for therapeutic intervention against cancer we must understand this pathway in full. Since the signaling pathways are built with high redundancy, it is unlikely that the interference in a single point of a single pathway will be effective against cancer. Instead, a combination of drugs will be required.

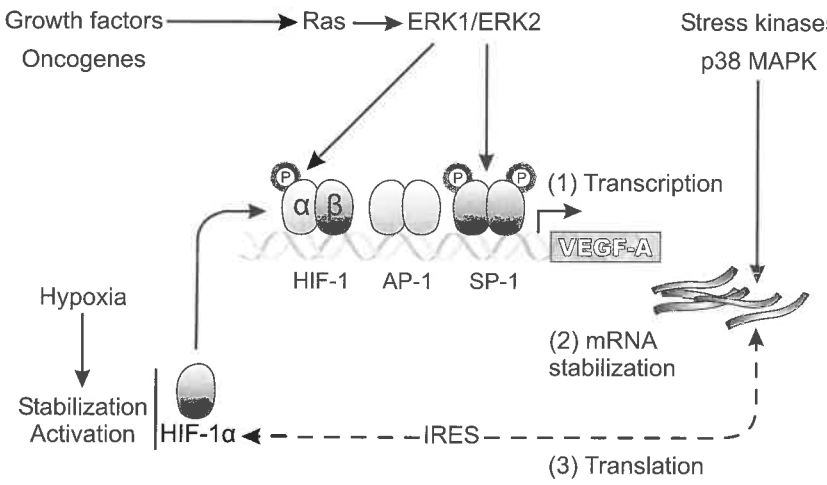


Figure 1. Induction of VEGF-A by HIF [67]. ERK: extracellular signal regulated kinase, MAPK: mitogen-activated protein kinase, HIF: hypoxia inducible factor, AP-1: transcription factor activator protein-1, SP-1: transcription factor Sp1, IRES: internal ribosome entry site.

As more of these drugs that target specific points of the signal transduction pathways become available, the selection of the correct combination of these drugs will be crucial. This requires a deeper understanding of these pathways as well as the development of new diagnostic technologies that will make the detection of the underlying molecular defect or defects in each individual tumor feasible.

References

1. Balkwill F, Mantovani A. Inflammation and cancer: back to Virchow? Lancet 2001; 357: 539-545.
2. Coussens L, Werb Z. Inflammation and cancer. Nature 2002; 420: 860-867.
3. Kuper H, Adami H, Trichopoulos D. Infections as a major preventable cause of human cancer. J Intern Med 2000; 248: 171-183.
4. Roder D. The epidemiology of gastric cancer. Gastric Cancer 2002; 5 (Suppl 1): 5-11.
5. Ekbom A. Risk of cancer in ulcerative colitis. J Gastrointest Surg 1998; 2: 312-313.
6. Dhala A, Pinser K, Prezant D. Respiratory health consequences of environmental tobacco smoke. Med Clin North Am 2004; 88: 1535-1552.
7. Hanahan D, Weinberg R. The hallmarks of cancer. Cell 2000; 100: 57-70.
8. Li Q, Verma I. NF-κB regulation in the immune system. Nature Rev Immunol 2002; 2: 725-734.
9. Ghosh S, Karin M. Missing pieces in the NF-κB puzzle. Cell 2002; 109 (Suppl): S81-S96.
10. Gilmore T. The Rel/NF-κB signal transduction pathway: introduction. Oncogene 1999; 18: 6842-6844.
11. Karin M, Ben-Neriah Y. Phosphorylation meets ubiquitination: the control of NF-κB activity. Annu Rev Immunol 2000; 18: 621-663.
12. Werner S, Barken D, Hoffmann A. Stimulus specificity of gene expression programs determined by temporal control of IKK activity. Science 2005; 309: 1857-1861.
13. Park J, Greten F, Wong A et al. Signaling pathways and genes that inhibit pathogen-induced macrophage apoptosis - CREB and NF-κB as key regulators. Immunity 2005; 23: 319-329.
14. Covert M, Leung TH, Gaston JE, Baltimore D. Achieving stability of lipopolysaccharide-induced NF-κB activation. Science 2005; 309: 1854-1857.
15. Bonizzi G, Karin M. The two NF-κB activation pathways and their role in innate and adaptive immunity. Trends Immunol 2004; 25: 280-288.
16. Kato TJ, Delhase M, Hoffmann A, Karin M. CK2 is a c-terminal IκB kinase responsible for NF-κB activation during the UV response. Mol Cell 2003; 12: 829-839.
17. Medzhitov R. Toll-like receptors and innate immunity. Nature Rev Immunol 2001; 1: 135-145.
18. Akira S, Uematsu S, Takeuchi O. Pathogen recognition and innate immunity. Cell 2006; 124: 783-801.
19. Nizet V, Lawrence T, Karin M. Innate immunity gone awry: mechanisms linking microbial infections to chronic inflammatory disorders and cancer. Cell 2006; 124: 823-835.
20. Liu Z-G, Hu H, Goeddel D, Karin M. Dissection of TNF receptor 1 effector functions: JNK activation is not linked to apoptosis, while NF-κB activation prevents cell death. Cell 1996; 67: 565-576.
21. Beg A, Baltimore D. An essential role for NF-κB in preventing TNF-α induced cell death. Science 1996; 274: 782-784.
22. Van Antwerp D, Martin S, Kafri T, Green D, Verma I. Suppression of TNFα-induced apoptosis by NF-κB. Science 1996; 274: 787-789.
23. Wang C-Y, Mayo M, Baldwin AJ. TNF-α and cancer therapy-induced apoptosis: potentiation by inhibition of NF-κB. Science 1996; 274: 784-787.
24. Okayasu I, Ohkusa T, Kajiura K, Kanno J, Sakamoto S. Promotion of colorectal neoplasia in experimental murine ulcerative colitis. Gut 1996; 39: 87-92.
25. Greten F, Eckmann L, Greten T et al. IKKβ links inflammation and tumorigenesis in a mouse model of colitis associated cancer. Cell 2004; 118: 285-296.
26. Mauad T, van Nieuwkerk C, Dingemans K et al. Mice with homozygous disruption of the mdr2 P-glycoprotein gene. A novel animal model for studies of nonsuppurative inflammatory cholangitis and hepatocarcinogenesis. Am J Pathol 1994; 145: 1237-1245.
27. Pikarsky E, Porat R, Stein I et al. NF-κB functions as a tumour promoter in inflammation-associated cancer. Nature 2004; 431: 461-466.
28. van Hogerlinden M, Rozell BL, Ahrlund-Richter L, Toftgard R. Squamous cell carcinomas and increased apoptosis in skin with inhibited Rel/nuclear factor-κB signaling. Cancer Res 1999; 59: 3299-3303.
29. McCormick F. Signalling networks that cause cancer. Trends Cell Biol 1999; 9: M53-M56.
30. Wullschleger S, Loewith R, Hall M. TOR signaling in growth and metabolism. Cell 2006; 124: 471-484.
31. Luo J, Manning B, Cantley L. Targeting the PI3K–Akt pathway in human cancer: rationale and promise. Cancer Cell 2003; 4: 257-262.
32. Frame S, Cohen P. GSK3 takes centre stage more than 20 years after its discovery. Biochem J 2001; 359: 1-16.
33. Hammerman P, Fox C, Thompson C. Beginnings of a signal-transduction pathway for bioenergetic control of cell survival. Trends Biochem Sci 2004; 29: 586-592.
34. Mayo L, Donner D. The PTEN, Mdm2, p53 tumor suppressor–oncoprotein network. Trends Biochem Sci 2002; 27: 462-467.
35. Datta S, Dudek H, Tao X et al. Akt phosphorylation of BAD couples survival signals to the cell-intrinsic death machinery. Cell 1997; 91: 231-241.
36. del Peso L, Gonzalez-Garcia M, Page C, Herrera R, Nunez G. Interleukin-3-induced phosphorylation of BAD through the protein kinase Akt. Science 1997; 278: 687-689.
37. Karin M. Nuclear factor-κB in cancer development and progression. Nature 2006; 44: 431-436.
38. Manning B, Cantley L. United at last: the tuberous sclerosis complex gene products connect the phosphoinositide 3-kinase/Akt pathway to mammalian target of rapamycin (mTOR) signalling. Biochem Soc Trans 2003; 31: 573-578.
39. Ma L, Chen Z, Erdjument-Bromage H, Tempst P, Pandolfi P. Phosphorylation and functional inactivation of TSC2 by Erk implications for tuberous sclerosis and cancer pathogenesis. Cell 2005; 121: 179-193.
40. Ballif B, Roux P, Gerber S, MacKeigan J, Blenis J, Gygi S. Quantitative phosphorylation profiling of the ERK/p90

ribosomal S6 kinase-signaling cassette and its targets, the tuberous sclerosis tumor suppressors. Proc Natl Acad Sci USA 2005; 102: 667-672.
41. Roux P, Ballif B, Anjum R, Gygi S, Blenis J. Tumor-promoting phorbol esters and activated Ras inactivate the tuberous sclerosis tumor suppressor complex via p90 ribosomal S6 kinase. Proc Natl Acad Sci USA 2004; 101: 13489-13494.
42. Woods A, Johnstone S, Dickerson K et al. LKB1 is the upstream kinase in the AMP-activated protein kinase cascade. Curr Biol 2003; 13: 2004-2008.
43. Shaw R, Kosmatka M, Bardeesy N et al. The tumor suppressor LKB1 kinase directly activates AMP-activated kinase and regulates apoptosis in response to energy stress. Proc Natl Acad Sci USA 2004; 101: 3329-3335.
44. Hardie DG. New roles for the LKB1–AMPK pathway. Curr Opin Cell Biol 2005; 17: 167-173.
45. Shaw R, Bardeesy N, Manning B et al. The LKB1 tumor suppressor negatively regulates mTOR signaling. Cancer Cell 2004; 6: 91-99.
46. Jost M KC, Rodeck U. The EGF receptor - an essential regulator of multiple epidermal function. Eur J Dermatol 2000; 10: 505-510.
47. Normanno N, Strizzi L, Mancino M et al. The ErbB receptors and their ligands in cancer: an overview. Curr Drug Targets 2005; 6: 243-257.
48. Muthuswamy S, Gilman M, Brugge J. Controlled dimerization of ErbB receptors provides evidence for differential signaling by homo- and heterodimers. Mol Cell Biol 1999; 19: 6845-6857.
49. Olayioye MA, Neve RM, Lane HA, NE H. The ErbB signaling network: receptor heterodimerization in development and cancer. EMBO J 2000; 19: 3159-3167.
50. Kabore A, Johnston J, Gibson S. Changes in the apoptotic and survival signaling in cancer cells and their potential therapeutic implications. Curr Cancer Drug Targets 2004; 4: 147-163.
51. Janmaat ML, Giaccone G. The epidermal growth factor receptor pathway and its inhibition as anticancer therapy. Drugs Today (Barc) 2003; 39 (Suppl C): 61-80.
52. Pedersen M, Meltorn M, Damstrup L, Poulsen H. The type III epidermal growth factor receptor mutation. Biological significance and potential target for anti-cancer therapy. Ann Oncol 2001; 12: 745-760.
53. Duursma A, Agami R. Ras interference as cancer therapy. Semin Cancer Biol 2003; 13: 267-273.
54. Sansal I, Sellers W. The biology and clinical relevance of the PTEN tumor suppressor pathway. J Clin Oncol 2004; 22: 2954-2963.
55. Hemavathy K, Ashraf S, Ip Y. Snail/slug family of repressors: slowly going into the fast lane of development and cancer. Gene 2000; 257: 1-12.
56. Inukai T, Inoue A, Kurosawa H et al. SLUG, a ces-1-related zinc finger transcription factor gene with antiapoptotic activity, is a downstream target of the E2A-HLF oncoprotein. Mol Cell 1999; 4: 343-352.
57. Savagner P, Yamada K, Thiery J. The zinc-finger protein slug causes desmosome dissociation, an initial and necessary step for growth factor-induced epithelial-mesenchymal transition. J Cell Biol 1997; 137: 1403-1419.
58. Lu Z, Ghosh S, Wang Z, Hunter T. Downregulation of caveolin-1 function by EGF leads to the loss of E-cadherin, increased transcriptional activity of beta-catenin, and enhanced tumor cell invasion. Cancer Cell 2003; 4: 499-515.
59. Pugh C, Ratcliffe PJ. Regulation of angiogenesis by hypoxia: role of the HIF system. Nature Med 2003; 9: 677-684.
60. Ikeda E, Achen M, Breier G, Risau W. Hypoxia-induced transcriptional activation and increased mRNA stability of vascular endothelial growth factor in C6 glioma cells. J Biol Chem 1995; 270: 19761-19766.
61. Mandriota S, Pepper M. Regulation of angiopoietin-2 mRNA levels in bovine microvascular endothelial cells by cytokines and hypoxia. Circ Res 1998; 83: 852-859.
62. Semenza G. Targeting HIF-1 for cancer therapy. Nature Rev Cancer 2003; 3: 721-732.
63. Berra E, Ginouves A, Pouyssegur J. The hypoxia-inducible-factor hydroxylases bring fresh air into hypoxia signalling. EMBO Rep 2006; 7: 41-45.
64. Maxwell P, Pugh C, Ratcliffe P. Activation of the HIF pathway in cancer. Curr Opin Genet Dev 2001; 11: 239-299.
65. Berra E, Richard D, Gothie E, Pouyssegur J. HIF-1-dependent transcriptional activity is required for oxygen-mediated HIF-1α degradation. FEBS Lett 2001; 491: 85-90.
66. Lando D, Peet D, Gorman J, Whelan D, Whitelaw M, Bruick R. FIH-1 is an asparaginyl hydroxylase enzyme that regulates the transcriptional activity of hypoxia-inducible factor. Genes Dev 2002; 16: 1466-1471.
67. Pouyssegur J, Dayan F, Mazure N. Hypoxia signaling in cancer and approaches to enforce tumor regression. Nature 2006; 441: 437-443.
68. Rak J, Mitsuhashi Y, Sheehan C et al. Oncogenes and tumor angiogenesis: differential modes of vascular endothelial growth factor up-regulation in ras-transformed epithelial cells and fibroblasts. Cancer Res 2000; 60: 490-498.
69. Pages G, Pouyssegur J. Transcriptional regulation of the vascular endothelial growth factor gene: a concert of activating factors. Cardiovasc Res 2005; 65: 564-573.