Nuclear Factor-κB: A Holy Grail in Cancer Prevention and Therapy

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Abstract: Nuclear factor-κB (NF-κB) is a transcription factor that is activated in response to various inflammatory stimuli such as cytokines, growth factors, hormones, mitogens, carcinogens, chemotherapeutic agents, viral products, eukaryotic parasites, endotoxin, fatty acids, metals, radiation, hypoxia, and psychological, physical, oxidative, and chemical stresses. In addition, constitutively active NF-κB is frequently encountered in a wide variety of tumors, including breast, ovarian, colon, pancreatic, thyroid, prostate, lung, head and neck, bladder, and skin cancers; B-cell lymphoma; Hodgkin's disease; T-cell lymphoma; adult T-cell leukemia; acute lymphoblastic leukemia; multiple myeloma; chronic lymphocytic leukemia; and acute myelogenous leukemia. Furthermore, NF-κB activation has been shown to regulate the expression of over 400 genes involved in cellular transformation, proliferation, inflammation, viral replication, antiapoptosis, angiogenesis, invasion and metastasis, oxidative stress, and osteoclastogenesis. Therefore, because of the critical role NF-κB plays in the pathogenesis of cancer, specific inhibitors of this factor are being sought. Agents that prevent cancer or inflammation have been found to suppress NF-κB activation. Although IκBα kinase is the major kinase, over 30 different protein kinases have been linked to the activation of NF-κB by different stimuli. The development of a drug that can specifically suppress NF-κB activation requires a full understanding of the mechanism by which NF-κB is activated in response to these various stimuli.

Key Words: NF-κB, IKK, antitumor, TNF, inflammation.

1. INTRODUCTION

Nuclear factor-κB (NF-κB), a nuclear transcription factor, was first identified in 1986 by Sen and Baltimore [1]. As its name implies, it is a nuclear factor bound to an enhancer element of the immunoglobulin kappa light chain gene in B cells [1]. First considered a B-cell transcription factor, NF-κB is now known to comprise a family of ubiquitous proteins. NF-κB proteins contain a Rel homology domain (DNA-binding domain/dimerization domain) with a nuclear localization sequence; such sequences are conserved from *Drosophila* to man. Class I proteins include p50, p52, p100, and p105. Multiple copies of ankyrin repeats are present in p100 and p105; proteolytic cleavage of p100 forms p52 and that of p105 forms p50. These protein, in turn, form dimers with class II proteins (c-Rel, RelB, and RelA/p65), which exclusively contain C-terminal activation domains. Whereas RelB forms only heterodimers, all the other proteins can form both homo- and heterodimers. NF-κB is the most common heterodimer formed between Rel A and p50. Dimeric NF-κB transcription factors bind to the 10-base-pair consensus site GGGPuNNPyPyCC, where Pu is purine, Py is pyrimidine, and N is any base. The individual dimers have distinct DNA-binding specificities for a collection of related κB sites [2, 3].

The various inhibitors of NF-κB include IκBα, IκBβ, IκBγ (derived from the C-terminal of p100), IκBε, Bcl-3, pp40 (a chicken homologue), cactus (a *Drosophila* homologue), and avian swine fever virus protein p28.2. p105 and p100 can also function to retain NF-κB subunits in the cytoplasm. All of these proteins are characterized by the presence of multiple ankyrin repeats. Perhaps the most common and best-understood form of NF-κB consists of p50, p65, and IκBα. IκBα mediates transient gene expression, whereas IκBβ mediates persistent response.

The IκB proteins are expressed in a tissue-specific manner and have distinct affinities for individual Rel/NF-κB complexes. IκBs contain six or more ankyrin repeats, an N-terminal regulatory domain, and a C-terminal domain that contains a proline-glutamic acid-serine-threonine motif. IκBs bind to NF-κB dimers and sterically block the function of their nuclear localization sequences, thereby causing their cytoplasmic retention. Most agents that activate NF-κB mediate the phosphorylation-induced degradation of IκB. On receipt of a signal, phosphorylation of IκBα takes place on two conserved serine residues (S32 and S36) in the N-terminal regulatory domain. However, another member of the IκB family, Bcl-3, stimulates transcription after interacting with p50 and p52 subunits of NF-κB.

Several of the IκB kinases (IKKs) have been characterized, namely, IKKα, IKKβ, and IKKγ. Mutation analysis revealed that IKKα and not IKKβ mediates proinflammatory signals. Once phosphorylated, the IκBs, which are still bound to NF-κB, almost immediately undergo a second posttranslational modification known as polyubiquitination. The major ubiquitin acceptor sites in human IκBα are lysines 21 and 22. Protein ubiquitination occurs through the E1 ubiquitin-activating enzyme, the E2 ubiquitin-conjugating enzyme, and the E3 ubiquitin protein ligases. After ubiquitination, IκBs are degraded in 26S proteasomes, leading to the release of NF-κB dimers, which translocate into the nucleus [3, 4]. In contrast, the activation of NF-κB in response to ultraviolet (UV) radiation is accompanied by IκBα degradation but not phosphorylation on the N-terminus of IκBα [5]. Hypoxia or

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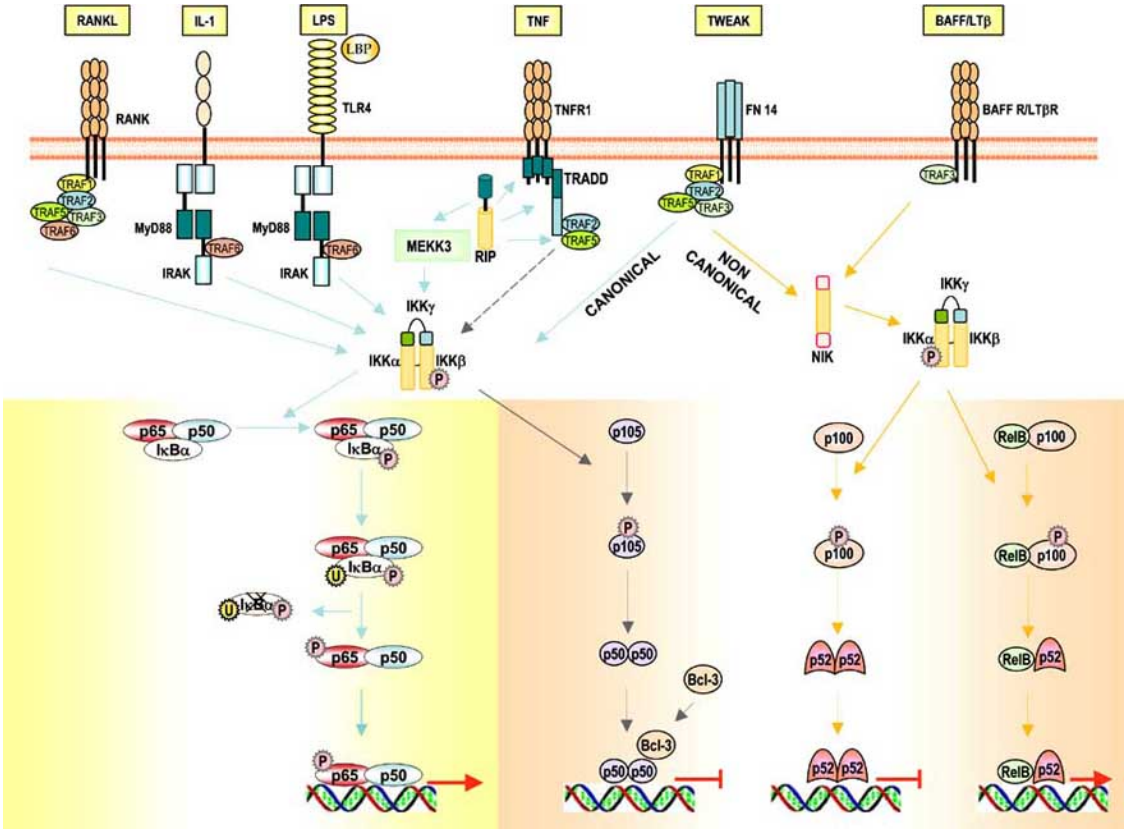

Fig. (1). Signaling pathway of NF-κB.

pervanadate treatment stimulates the phosphorylation of IκBα at tyrosine 42, but other IκBs do not have a tyrosine at this position [6]. Phosphorylation on Ser-276 by the catalytic subunit of protein kinase A contributes to the intrinsic transcriptional capacity of the p65 subunit of NF-κB. The catalytic subunit of protein kinase A was also found to be associated with NF-κB and IκB in the cytoplasm and was able to phosphorylate p65 only after IκB degradation [7]. In addition, a site-directed mutant of p65 (Ser-276 to Ala) is phosphorylated at Ser 529 in response to tumor necrosis factor (TNF), suggesting that multiple physiologic stimuli modulate p65 through distinct phosphorylation sites to control transcriptional activity. RelA (C-terminus) has been shown to interact with basal transcriptional apparatus proteins such as TATA-binding protein (TBP), transcription factor (TF) IIB and TBP-associated factor (TAF) 105 and with coactivators such as cAMP responsive element binding protein (CBP) and p300, although the actual role of these interactions is not clear [7]. This pathway is well conserved, both in structure and function, from *Drosophila* to humans.

NF-κB is activated by many divergent stimuli, including proinflammatory cytokines (e.g., TNF-α, interleukin-1 [IL-1]), T- and B-cell mitogens, bacteria, lipopolysaccharide (LPS), viruses, viral proteins, double-stranded RNA, and physical and chemical stresses [8] (Table **1**). Cellular stresses, including ionizing radiation and chemotherapeutic agents, also activate NF-κB [9].

Although much has been learned since the discovery of NF-κB, the precise mechanism of its activation is still not fully understood. Depending on the stimulus, this mechanism involves overlapping and nonoverlapping steps. For example, TNF, one of the most potent activators of NF-κB, interacts with the TNF receptor (TNFR) and then recruits a protein called TNFR-associated death domain. This protein binds to TNFR-associated factor (TRAF) 2, which recruits NF-κB–inducing kinase (NIK), which in turn activates IKK. IKK phosphorylates IκBα at serines 32 and 36, which leads to ubiquitination at lysines 21 and 22, and this leads to the degradation of IκBα by the 26S proteasome. This degradation results in translocation of NF-κB to the nucleus, where it binds to its consensus sequence (5'-GGGACTTTC-3') and activates gene expression. Thus, NF-κB can be monitored by the IκBα degradation seen on Western blotting, by the NF-κB binding to DNA seen on electrophoretic mobility shift assay, or by the NF-κB–dependent reporter gene expression seen on transient transfection.

Besides the previously described canonical NF-κB activation pathway, a noncanonical NF-κB activation pathway is activated by CD40L, lymphotoxin (LT)-β, receptor activator of NF-κB ligand (RANKL), and B-cell-activating factor of the TNF family (BAFF), all members of the TNF family (Table **1**) [10, 11]. This pathway does not involve IκBα but instead involves direct phosphorylation and ubiquitin-dependent degradation of p100. Current research

Table 1. A List of Inducers of NF-κB Activation

Cytokines	All-trans retinoic acid	*Borrelia burgdorferi*	Pneumocystis
TNFα[10,11]	RET/PTC3 fusion oncoprotein	*Chlamydia pneumoniae*	*Theileria parva*
TNFβ	S100B	*Cryptococcus neoformans*	*Trypanoplasma borreli*
RANKL[10,11,154]	Serum	*Ehrlichia chaffeensis*	
IL-1	Sulphatide	Enteropathogenic *Escherichia coli*	**Physiologic (stress) conditions**
TRAIL	TGF-α	*Fusobacterium nucleatum*	Asthma
CD40 ligand[10,11]	TGF-β	*Gardnerella vaginalis*	Rheumatoid arthritis
CD30 ligand[10,11]		*Helicobacter pylori*	Crohn's disease/ulcerative colitis
TWEAK[10,11]	**Viruses**	Lactobacilli	Corneal epidemic
THANK	Adenovirus	*Listeria monocytogenes*	keratoconjunctivitis
BAFF[10,11]	Cytomegalovirus	*Mycoplasma fermentans*	Coronary artery bypass
LTβ[10,11]	EBV	*Mycobacteria tuberculosis*	Hypercholesterolemia
IL-2	Hammarskjöld and hepatitis B virus	*Neisseria gonorrhoeae*	Hyperglycemia
IL-4	HVS	*Neisseria meningitidis*	Hyperhomocysteinemia
IL-8	Human herpes virus 6	*Porphyromonas gingivalis*	Hyperosmotic shock
IL-12	HIV-1	*Prevotella intermedia*	Hypoxia[90]
IL-15	Herpes simplex virus-1	*Pseudomonas aerogenosa*	Acute lung injury
IL-17	HTLV-1	*Rhodococcus equi*	Acute respiratory distress syndrome
IL-18	Influenza virus	*Rickettsia rickettsii*	Adhesion
CD11b/CD18 ligand	Measles virus	*Salmonella dublin*	Angina pectoris
CD28 ligand	Moloney murine leukemia virus	*Salmonella typhimurium*	Antiphospholipid antibodies
CD2 ligand	Newcastle disease virus	*Shigella flexneri*	Appendicitis
CD35 ligand	Respiratory syncytial virus	*Staphyllococcus aureus*	Depolarization
CD3 ligand	Rhinovirus	Streptococcus	Hemorrhage
CD43 ligand	SARS coronavirus	*Streptomyces californicus*	Ischemia (transient, focal)
CD4 ligand	Sendai paramyxovirus	*Trichomonas vaginalis Ureaplasma*	Ischemic preconditioning
CD66a ligand	Sindbis virus	*urealyticum*	Liver regeneration
LIF	Vaccinia virus akara	*Yersinia enterocolitica*	Human labor
S100B	West Nile flavavirus		Biotin deficiency
Agonist antibodies to AT1 receptor		**Bacterial or fungal products**	Butter and walnut diet
IgM ligand	**Viral products**	LPS	Cecal ligatin and puncture
β-D-glucan ligand	Adenovirus 5: E1A	Wogonin	Mechanical ventilation
IgG2a	Adenovirus: E3/19K	Enterotoxin	Memory retrieval
Flt-1 ligand	African swine fever virus: IAP	Glycosylphosphatidylinositols	Middle cerebral artery occlusion
Ly6A/E ligand	Antibody to dengue virus	Fumonisin B1	Muscle disuse
	nonstructural protein 1	Staphylococcus enterotoxins A and	Muscular dystrophy
Growth factors and hormones	CMV: ie1	B	Neuronal firing
GMCSF	EBNA-2	Apicularen A	Overventilation pancreatitis
M-CSF	EBV: LMP1	CpG	Proteinuria
NGF	HBV: HBx	Cytolysin	Reoxygenation
EGF	HBV: LHBs	Diphosphoryl lipid A	Senescence
Insulin-like growth factor 1	HBV: MHBst	Exotoxin B Lipoprotein LpK	Shear stress
Insulin	HCV: Core protein	Lipoprotein LpK	Neuroonal trimethyltin injury
Hepatocyte growth factor	HVS13	Lipoteichoic acid	Uniaxial cyclic cell stretching
Gastrin	HVS: StpC	Membrane lipoproteins	T-cell selection
PEDF	HIV-1: gp160	(*Mycoplasma fermentans, Mycoplasma penetrans*)	**Physical stress**
Platelet-derived growth factor	HIV-1: Nef	Mannoproteins	UV A, B, and C radiation[106]
Bone morphogenic protein 2	HIV-1: p9 (9 aa peptide)	Muramyl peptides	Bile duct ligation
Bone morphogenic protein 4	HIV-1: Tat	*Mycobacterium* species	Cyclic mechanical muscle strain
Cortical releasing hormone EGF	HTLV-I: Tax	lipoarabinomannan	Exercise γ radiation
Follicle-stimulating hormone	HTLV-II: Tax	PlcA	Heavy ion irradiation
Mullerian inhibiting substance	Influenza virus: hemagglutinin	PlcB	High-fat diet
PAF	KSHV: K1	Porins	Laminar shear stress
Plant steroids (diosgenin,	Parvovirus B19: NSI	Porin 1B	Dietary lipid hydroperoxide
hecogenin, tigogenin)	Respiratory syncytial virus: M2-1	Toxic shock syndrome toxin 1	PPME
Prostratin	SV40		photosensitization
	Bacteria and fungi	**Eukaryotic parasites**	Mechanical lung ventilation
Oncogenes	*Anaplasma phagocytophilum*	*Eimeria tenella* and *Eimeria*	Obesity
Ras[115]			

(Table 1. Contd….)

Myc[117]	*Angiostrongylus cantonensis*	*necatrix*	Wounding combined with
bcr-ab1[116]	*Bacteroides forsythus*	Leishmania	TUDCA
HeNe irradiation	*Bartonella henselae*	Phospholipomannan	Tuberous sclerosis complex
Wounding combined with thermal	*Bordetella pertussis*		Uremic toxins VLDL
irradiation	Cycloprodigiosin	**Physiological Mediators**	
	Dacarbazine	Alloxan	**Chemical Agents**
Oxidative stress	Orengedeokuto	Angiotensin II	L-NMA
Glutathione	Diazoxide	Bradykinin	Ethanol
Hydrogen peroxide[224]	5,6-dimethylxanthenone-4-acetic	Fibrinogen	Monensin
Catalase	acid	Hemoglobin	Nicotine
Nitric oxide	Flavone-8-acetic acid	Hyaluronan	Adriamycin
Ozone	Haloperidol	Thioredoxin	2-Deoxyglucose
Peroxynitrite	Kunbi-Boshin-Hangam-Tang	Kainic acid	Anisomycin
Butyl peroxide	Methamphetamine	L-Glutamate	Benzyl isothiocyanate
Cerulein	Norepinephrine	Melanin	Bisperoxovanadium phosphotyrosyl
Pervanadate	Oltipraz	Violacein	Brefeldin A
Reoxygenation	Phenobarbital	Vitamin D_3	Cadmium
	Protocatechuic acid	Adenosine	Colchicine
Environmental hazards		Activin A	Calcium Ionophore plus phorbol
Cigarette smoke[87-88]	**Fatty acids**	Albumin	ester
PH[91]	Palmitate	Allergin	Calyculin A
Arsenic	Leukotriene B4	Acrp30/adiponectin	Ceramide-β-galactose
Polychlorinated biophenyl-77	Free fatty acids	Adrenomedullin	2-chloroethyl ethyl sulfide
Benzo[a]pyrene diol epoxide[87]	Linoleic acid	*Aeginetia indica* L (AIL)b-A	Con A
Crocidolite asbestos fibers	Oleic acid	Bronchoalveolar lavage fluid	Cycloheximide
Dicamba	Saturated fatty acids	Brusatol	Cyclopiazonic acid
Diesel exhaust particles	Lysophosphatidic acid	Homocysteine	Diquat
Gliadin		Cryptdins	2,4-Dinitrofluorobenzene
House dust mite	**Modified Proteins**	Deoxycholic acid	Ferrocene
Dust particles	LDL	12(R)-hydroxyeicosatrienoic acid	Forskolin
Fear-potentiated startle response	AGEs	6-Hydroxydopamine	Gadolinium chloride
Lead	Amyloid protein fragment	Human peptide transporter 1	Glass fibers
Lead chromate	Glycosylated oxyhemaglobin	LTP	Sodium butyrate trichostatin A
Noise	Maleylated bovine serum Albumin	LysoPC	Malondialdehyde
Oily fly ash	Neurotrophin receptor proteolytic	Neuormelanin	MDMA
Silica particles	fragments	NS-398	MEN 17055[113]
Wood smoke	Nonamyloid beta component of	Osteopontin	N-methyl-D-aspartate
Zymosan	Alzheimer's disease	PAF	Mycophenolic acid
	ATM[108]	PCSC	Nafenopin
Chemotherapeutic drugs		Prolactin	N-Nitrosomorphine
Cisplatin	**Heat shock protein**	PIF	Nocodazol
Paclitaxel[103]	HSP60	12-Lipoxygenase	Okadaic acid[89]
Doxorubicin[100]	HSPb1	Thrombin	Peplomycin
Tamoxifen		Streptozotocin	Phytohemagglutinin
Vinblastine	**Apoptotic Mediators**	Substance P	Phorbol ester[87]
Vincristine[222]	Anti-Fas/Apo-1	Potassium	Podophyllotoxin
Etoposide	PARP	Burn patient serum	Polychlorinated biophenyl-77
ara-C		Anaphylatoxin C3a and C5a	Prostratin
Bryostatin-1	**Metals**	Angiotensin II type 1	Pyrogallol
Camptothecin[101]	Lithium	receptor–activating antibodies	Staurosporine
Daunomycin	Titanium and copper implants	Antiphospholipid antibodies	Transglutaminase 2
Daunorubicin	Nickel sulfate	Basic calcium phosphate crystals	Tunicamycin
Mitoxantrone	Iron	C2-ceramide cerulein	WF10
Gemcitabine[102]	Manganese	Chelidonium majus extract	WY-14 643
	Nickel	Collagen lattice	CFTR
Therapeutically used drugs	Chromium	Collagen type I	GPR56
Celecoxib	Cobalt	Cysteinyl leukotrines	Hematopoietic progenitor kinase 1
AZT	Aluminum	Des-Arg10-kallidin	Ig heavy chain
Bleomycin		Mixed meal ingestion Monosodium	MHC Class I

(Table 1. Contd…)

WR1065	**Natural products**	urate crystals	
ABR-25757 SN38	Areca nut extract	Neutrophil elastase	
SA 981	Baicalin	Phellinus linteus proteoglycan	
AdAMP	β-carotene	Platelet type arachidonate	
Anthralin	Quercetin	Polysaccharides of Poria cocos	
Ciprofibrate	Quinolinic acid	Regulatory RNA	
	Safflower polysaccharides	S100B	
	Sanglifehrin A	Sleep deprivation	

TNF, tumor necrosis factor; RANKL, receptor activator of nuclear factor-κB ligand; IL, interleukin; TRAIL, TNF-related apoptosis-inducing ligand; TWEAK, TNF-like weak inducer of apoptosis; THANK, TNF homologue that activates apoptosis, nuclear factor-κB, and c-Jun NH2 terminal kinase; BAFF, B cell-activating factor; LT, lymphotoxin; LIF, leukemia inhibitory factor; AT, ataxia telangiectasia; Ig, immunoglobulin; GMCSF, granulocyte macrophage colony-stimulating factor; M-CSF, macrophage colony-stimulating factor; NGF, nerve growth factor; EGF, epidermal growth factor; PEDF, pigment epithelium-derived growth factor; PAF, platelet-activating factor; TGF. transforming growth factor; EBV, Epstein-Barr virus; HVS, herpesvirus saimiri; HTLV-1, human T-cell leukemia virus type 1; SARS, severe acute respiratory syndrome; IAP, inhibitor of apoptosis protein; CMV, cytomegalovirus; EBNA, EBV nuclear antigen; LMP1, latent membrane protein 1; HBx, hepatitis B virus X; LHBs, large hepatitis B virus surface protein; MHBst, C-terminally truncated middle surface protein; HCV, hepatitis C virus; KSHV, Kaposi sarcoma herpesvirus; L-NMA, Nomega-methyl-L-arginine; NSI, non-specific lymphadenitis; SV40, simian virus 40; LPS, lipopolysaccharide; PPME, Pyropheophorbide-a methyl ester; LpK. L pyruvate kinase; PIc, poly(rI:rC); UV, ultraviolet; ara-C, 1-β-D-arabinofuranosylcytosine; AZT, azidothymidine; AdAMP, 2-1-adamantylamino)-6-methylpyridine; LDL, low-density lipoprotein; AGEs, advanced glycated end products;, HSP, heat shock protein; PARP, poly(ADP)ribose polymerase; Acrp30, adipocyte complement-related protein of 30 kDa; LTP, long-term potentiation, LysoPC, lysophosphatidylcholine; PCSC, polysaccharide from Poria cocos; PIF, proteolysis-inducing factor; TUDCA, taurosodeoxycholic acid; VLDL, very LDL; MDMA, 3,4-methylenedioxymethamphetamine.

Source: http://www.ncbi.nih.gov/entrez/query.fcgi, http://people.bu.edu/gilmore/nf-kb/.

indicates that NF-κB activation is highly complex and may involve dozens of different protein kinases [12-53] (Table **2**). Besides NIK, IKK-α, and IKK-β, NF-κB activation may also require the involvement of other kinases, such as atypical protein kinase C, protein kinase C-z, pp90rsk, double-stranded RNA-dependent protein kinase, cot kinase (also called TPL2), mitogen-activated protein kinase kinase kinase 1, 2, and 3), phosphatidylinositol 3 protein kinase, Akt, mixed lineage kinase 3, hematopoietic progenitor kinase-1, transforming growth factor β–activated kinase 1, and c-raf kinase. These kinases may form a cascade, and different cascades may form depending on the NF-κB activator. For instance, IKK can be phosphorylated by NIK, mitogen-activated protein kinase kinase kinase, or Akt. Although IKK is required for NF-κB activation by most agents, a few (such as human epithelil receptor type 2, H_2O_2, pervanadate, x-rays, and γ-radiation) activate NF-κB through IKK-independent pathways [54]. Although several signaling proteins and protein kinases have been identified recently that mediate NF-κB activation, more kinases and protein phosphatases remain to be identified. Besides the ubiquitin-dependent 26S proteasome, which has a role in IκBα degradation [55], other proteases have also been implicated in NF-κB activation [56].

The genetic deletions of different NF-κB proteins produce numerous phenotypic changes [57]. For instance, deletion of the *rel* a gene induced embryonic lethality in mice, probably due to massive apoptosis in the liver [58]. In addition, the mouse embryo fibroblasts (MEFs) from rel a-deletion mice were found to be hypersensitive to TNF-induced apoptosis. These results indicate a negative role for NF-κB in TNF-induced apoptosis. Furthermore, mice lacking the RelA subunit were brought to term only in aTNFR1-deficient background [59]. These mice lacked lymph nodes, Peyer's patches, and an organized splenic microarchitecture, and they had a profound defect in their T-cell–dependent antigen responses. Analyses of TNFR1/RelA-deficient embryonic tissues and of radiation chimeras suggest that the dependence on RelA is manifested not in hematopoietic cells but rather in radioresistant stromal cells, which are needed for the development of secondary lymphoid organs. In contrast to the deletion of Rel A, the deletion of the IκBα gene leads to early neonatal lethality caused by inflammatory dermatitis and granulocytosis [60] that are most likely induced by constitutive activation of NF-κB, leading to expression of the granulocyte colony-stimulating factor.

NF-κB activation has been implicated in a wide variety of diseases, including cancers, diabetes mellitus, cardio-vascular diseases, autoimmune diseases, viral replication, septic shock, neurodegenerative disorders, ataxia telangiect-asia (AT), arthritis, asthma, inflammatory bowel disease, and several other inflammatory conditions (Table **3**). For example, activation of NF-κB by LPS may contribute to the development of septic shock because NF-κB activates transcription of the inducible nitric oxide synthase (iNOS) genes known to be involved in septic shock [61]. Similarly, autoimmune diseases such as systemic lupus erythematosus may also involve activation of NF-κB. Additionally, in chronic Alzheimer's disease, the amyloid β peptide causes production of reactive oxygen intermediates and indirectly activates gene expression through κB sites [62]. The influenza virus protein hemagglutinin also activates NF-κB, and this activation may contribute to viral induction of cytokines and to some of the symptoms associated with influenza [63]. Furthermore, the oxidized lipids from the low density lipoproteins associated with atherosclerosis activate NF-κB, which then activates other genes [64], and mice that are susceptible to atherosclerosis exhibit NF-κB activation when fed an atherogenic diet [65]. Another important contributor to atherosclerosis is thrombin, which stimulates the proliferation of vascular smooth muscle cells through the activation of NF-κB [66]. Finally, a truncated form of IκBα was shown to protect AT cells, which express constitutive levels of an NF-κB–like activity, from ionizing radiation [67]. In light of all these findings, the abnormal activation or expression of NF-κB is clearly associated with a wide variety of pathologic conditions. How NF-κB activation mediates tumorigenesis is the focus of the current review.

Table 2. A List of Protein Kinases Implicated in NF-κB Activation

Kinase	Ligand
IKKα	Proinflammatory cytokines (eg TNF, IL-1, IL6), LPS[20-22]
IKKβ	Proinflammatory cytokines (eg TNF, IL-1, IL6), LPS[20-22]
IKKγ	Proinflammatory cytokines (eg TNF, IL-1, IL6), LPS[14,15]
IKKi/IKKε	TNF, PMA[16-18]
NIK	TNF, CD95, and IL-1[19]
NAK	Phorbol esters, growth factors[38-40]
PI3K	IL-1[41]
Akt1	TNF, TCR/CD28[31,42]
HPK-1	Unknown[44]
PKC-α	TPA, TNF, Bimp1, Bcl 10/MALT1[26-28]
PKC-β	IgM receptor, Bimp1, Bcl 10/MALT1[27,29]
PKC-τ	CD3-CD28 (TCR/CD28), Bimp 1, Bcl 10/MALT1[27,30,31]
PKC-δ	TNFα[32]
PKC-ε	Phorbol ester, Bimp1, Bcl 10/ MALT1[27,33]
aPKC	TNF, ras p21[32,23]
MEKK1/2	Proinflammatory cytokines[22-36]
MEKK3	TNF, other proinflammatory cytokines[22,37]
TPL-2/Cot kinase	TNF [34]
pp90rsk	*Pseudomonas aeruginosa*[54]
Raf-1 kinase	Serum growth factors, phorbol ester, and PTK oncogenes[45]
PKR	dsRNA[33]
TAK1/MEKK	XIAP, TGF-β[44]
MLK3	CD3/CD28[42]
PAK1	p21, LPS[46]
BTK	B-cell antigen receptor[47]
JAK2	Erythropoietin[12]
PKA	IL-1, LPS, many others[48,49]
IRAK-1	IL-1[50]
IRAK-2	IL-1, LPS[52]
IRAK-M	IL-1, LPS[52]
P56 lck	Ceramide[53]
Syk	TNF, H_2O_2[224]

IKK, IκB kinase; TNF, tumor necrosis factor; IL, interleukin; LPS, lipopolysaccharide; PMA, phorbol 12-myristate 13-acetate; NIK, NF-κB–inducing kinase; NAK, NF-κB–activating kinase (also called T2K-, TBK1-, or TRAF2-interacting kinase, PI3K, phosphatidylinositol 3 kinase; Akt, protein kinase Bα; TRC, T-cell receptor; HPK, hematopoietic protein kinase; PKC, protein kinase C; TPA, 12-O-tetradecanoylphorbol-13-acetat; MALT, mucosa-associated lymphoid tissue; IgM, immunoglobulin M; aPKC, atypical PKC; MEKK, mitogen-activated protein kinase kinase kinase; pp90rsk, ribosomal protein S6 kinase; PKR, RNA-dependent protein kinase; dsRNA, double-stranded RNA; XIAP, X-linked inhibitor of apoptosis; TGF, transforming growth factor; MLK, mixed lineage kinase; PAK; p21 activated; BTK, Bruton's tyrosine kinase; JAK, Janus kinase; PKA, protein kinase A; IRAK, IL-1 receptor-associated kinase; Syk, spleen tyrosine kinase.

2. NF-κB MEDIATES CARCINOGENESIS

NF-κB has been implicated in carcinogenesis because it plays a critical role in cell survival, cell adhesion, inflammation, differentiation, and cell growth. Cancer is a hyperproliferative disorder that results from tumor initiation and tumor promotion and ultimately produces tumor metastasis. Notably, several genes involved in cellular transformation, proliferation, invasion, and angiogenesis are regulated by NF-κB.

2.1. NF-κB Genes are Proto-Oncogenes

NF-κB genes are members of a proto-oncogene family, and many functions of their encoded proteins have obvious implications for the development of cancer and its therapy. Retroviruses encoding *v-rel* were shown to be oncogenic in avian species, and *rel* genes to be prone to rearrangements and translocations [68]. For example, tumors with *rel* amplification had an increased frequency of chromosomal aberrations previously associated with tumor progression,

Table 3. Association of NF-κB Activation with Major Diseases

Cancer	**Inflammatory diseases**
B-cell lymphoma	Alzheimer's disease
Breast cancer	Arthritis
Hodgkin's disease	Asthma
Liver cancer	Bone resorption
Mantle cell lymphoma	Chronic obstructive pulmonary disease
Multiple myeloma	Crohn's disease
Non-Hodgkin's lymphoma	Huntington's disease
Ovarian cancer	Inflammatory bowel disease
Prostate cancer	Inflammatory response syndrome
T-cell lymphoma	Multiple sclerosis
Thyroid cancer	Ocular allergy
Bladder cancer	Parkinson's disease
Colon cancer	
Esophageal cancer	**Other diseases**
Head and neck squamous cell carcinoma	Aging
Laryngeal cancer	AIDS
Lung cancer	Catabolic disorders
Pancreatic cancer	Ectodermal dysplasia
Pharyngeal cancer	Gut diseases
Renal carcinoma	Headache
Acute lymphoblastic leukemia	*Helicobacter pylori*-associated gastritis
Adult T-cell leukemia	Inontinentia pigmenti
Cervical cancer	Ischemia/reperfusion injury
Nasopharyngeal carcinoma	Systemic lupus erythematosus
Melanoma	Muscular dystrophy
	Neuropathologic diseases
Cardiovascular diseases	Psychosocial stress diseases
Atherosclerosis	Renal diseases
Cardiac hypertrophy	Sepsis
Heart failure	Skin diseases
Hypercholesterolemia	Sleep apnea
	Viral infection
Diabetes mellitus	
Type I	
Type II	

Source: http://www.ncbi.nih.gov/entrez/query.fcgi, http://people.bu.edu/gilmore/nf-kb/.

thus suggesting an oncogenic effect of amplified *rel* in B-lymphoid cells already containing a transforming genetic lesion. According to one study's findings, *rel* amplification is a frequent event in diffuse lymphoma with a large-cell component and probably constitutes a progression-associated marker of primary extranodal lymphomas [69]. The *rel* proto-oncogene has been mapped to chromosome region 2p11.2-14, a site associated with nonrandom rearrangements in non-Hodgkin's lymphoma. Lu *et al*. [28] characterized an abnormal rel mRNA from a cell line derived from a diffuse large cell lymphoma in which the evolutionarily conserved N-terminal half of the *rel* coding region was fused with the C-terminal coding region of an unrelated gene. In addition, the rearrangement or amplification of the *rel* locus was found in the lymphomatous tissue of two follicular and one diffuse large cell lymphoma [28]. These findings suggest the involvement of rel in the pathogenesis of large-cell lymphoma [70].

Recent studies have also implicated the p50 subunit of the NF-κB transcription factor complex in tumorigenesis. Mukhopadhyay *et al*. [29] investigated the expression of this complex in paired normal and non-small cell lung carcinoma (NSCLC) tissues and found that 81.8% of fresh NSCLC tissues expressed from two- to 20-fold the levels of the p50 subunit compared with normal lung tissue. Thirteen NSCLC cell lines also exhibited high levels of p50. Such alterations in the normal NF-κB/rel pathway of regulation may therefore play a role in the genesis of NSCLC [71]. Alterations in the Rel A protein were also implicated in squamous head and neck carcinoma [72], adenocarcinomas of the breast and stomach [73], thyroid carcinoma [74], and multiple myeloma [75]. In addition, the overexpression of the p50 subunit of NF-κB has been detected in cell lines derived from cancers of the lung, prostate, breast, bone, and brain [71].

In certain lymphomas, the *bcl-3* and *nf-κb2* genes are known to be translocated [76]. Furthermore, mutations in the *iκbα* gene were observed in Hodgkin's lymphoma [77], and blocking of *iκbα* by antisense mechanisms appears to induce oncogenic transformation [78]. Additionally, antisense to *relA* blocks tumorigenesis induced by Tax, a protein derived from human T-cell leukemia virus type 1 [79]. A recent study similarly showed that human breast tumors accumulate activated NF-κB complexes consisting of p50, p52, and Bcl-3 rather than p65 [7].

In 1992, Liptay *et al*. [38] discovered that the NF-κB subunits p49/p100 and p105 map to regions associated with certain types of acute lymphoblastic leukemia. During molecular characterization of translocations associated with some human T-cell acute lymphocytic leukemias, the LYL1 gene was first identified. In adult tissues, LYL1 expression was restricted to hematopoietic cells, with the notable exception of cells of the T-cell lineage. LYL1 encodes a basic helix-loop-helix protein that is highly related to TAL-1, whose activation is also associated with a high proportion of human T-cell acute lymphocytic leukemias. Liptay *et al*. found that p105, the precursor of NF-κB1 p50, was the major LYL1-interacting protein in this system. Ectopic expression of LYL1 in a human T-cell line caused a significant decrease in NF-κB–dependent transcription and a reduced level of NF-κB1 proteins [80]. It was also reported that p105/p50 is altered in cancers of the bone, colon, prostate, breast, and brain [81].

Alterations in p100/p52 implicated in chronic lymphocytic leukemia, multiple myeloma, and cutaneous T-cell lymphomas [82]. In addition, NF-κB2/p52 (lyt-10) was shown to be involved in the breakpoint of at (10;14)(q24;q32) chromosomal translocation in a case of B-cell lymphoma. Fracchiolla *et al*. [42] have demonstrated that lyt-10 gene

rearrangements are recurrent lesions that may be involved in the pathogenesis of both B-cell and T-cell malignancies and suggested that the truncation of the ankyrin domain is a common mechanism leading to abnormal lyt-10 activation in lymphoid neoplasia. These authors also showed in an MDA–MB-435 breast cancer model that most p65 protein is complexed with p100 in these cells but predominantly with IκBα in cell lines expressing less p100. Based on these observations, Dejardin *et al*. [43] hypothesized that NF-κB is involved in carcinogenesis and suggested that the p100/p52 NF-κB subunit plays a role in the development of human breast cancers, possibly by sequestering other NF-κB–related proteins in the cytoplasm.

2.2. Constitutively Active NF-κB is Common in Tumor Cells

Constitutive expression of NF-κB has been shown in cell lines derived from breast, ovarian, colon, pancreatic, thyroid, prostate, lung, head and neck, bladder, and skin tumors [83]. This has also been seen for B-cell lymphoma, Hodgkin's disease, T-cell lymphoma, adult T-cell leukemia, acute lymphoblastic leukemia, multiple myeloma, chronic lymphocytic leukemia, and acute myelogenous leukemia.

Besides these cell lines, activated NF-κB has also been noted in tissue samples from patients with cancer. For instance, NF-κB is constitutively activated in high-grade squamous intraepithelial lesions and squamous cell carcinomas of the human uterine cervix [84]. NF-κB was also implicated in an aggressive phenotype of renal cell carcinoma [47]. Of 45 cases of renal cell carcinoma, 33% showed an NF-κB activity that was greater than 200% higher than that of normal renal tissue. In locally advanced cases, 64% showed an increased activity, and tissue from their metastases showed an even greater increase. In addition, the elevation of patients' serum C-reactive protein levels correlated with increases in the NF-κB activation in their tumors; therefore, NF-κB may be a cause of the inflammatory paraneoplastic syndrome [85]. Along similar lines, Yu *et al*. [48] reported that increased expression of NF-κB in colorectal tumorigenesis plays an important role in the pathogenesis of colon cancer in humans by mediating the transition from colorectal adenoma with low-grade dysplasia to adenocarcinoma.

Why tumor cells express constitutively active NF-κB, however, is not fully understood. The aberrant IKK activity and shorter half-life of IκBα in B-cell lymphoma, the mutated IκBα in Hodgkins' lymphoma, the IL-1β production in acute myelogenous leukemia, and the TNF production in cutaneous T-cell lymphoma and Burkitt's cell lymphoma are some of the reasons that have been postulated for constitutive NF-κB activation [57].

2.3. Carcinogens, Inflammatory Agents, Pro-Oxidants, and Stress and Tumor Promoters Activate NF-κB

During the last few years, various carcinogens have been shown to activate NF-κB, including 7,12-dimethylbenz(*a*) anthracene (DMBA) [86], benzo(*a*)pyrene diol epoxide, 4-(methylnitrosamino)-1-(3-pyridyl)-1-butanone, and nicotine (for a review, see reference[87]) (Table **1**). For example, DMBA-induced NF-κB activation was shown to occur not only *in vitro* but also when DMBA was administered *in vivo* to animals [49]. Furthermore, our laboratory showed that cigarette smoke condensate is a potent activator of NF-κB in different cell types [88]. Unlike TNF-induced NF-κB activation, the NF-κB activation induced by cigarette smoke condensate was persistent [87]. Besides tumor initiators, tumor promoters such as phorbol ester and okadaic acid have also been shown to activate NF-κB [89]. Additionally, NF-κB is activated by hypoxia [90] and an acidic pH [91], both characteristics of the tumor microenvironment.

TNF is one of the most potent activators of NF-κB. Although initially identified as an anticancer agent [92], TNF has since been shown to be involved in cellular transformation [93], tumor promotion [94], and induction of metastasis [95]. In agreement with these observations, mice deficient in TNF were shown to be resistant to skin carcinogenesis [96]. For several tumors, TNF has even been shown to be a growth factor [97]. Like phorbol ester, TNF mediates these effects in part by activating a protein kinase C pathway [98]. Other inflammatory cytokines have also been implicated in tumorigenesis [99].

2.4. Chemotherapeutic Agents and γ-Radiation Induce NF-κB Activation

Besides inducing apoptosis, almost all chemotherapeutic agents also activate NF-κB (Table **1**). These include DNA-damaging agents such as doxorubicin (DoxR) [100], camptothecin [101], gemcitabine [102], and cisplatin; microtubule depolymerizing agents such as taxol [103]; alkylating agents such as melphalan [104]; and the glutathione reductase inhibitor 1,3-bis(2-chloroethyl)-1-nitrosourea [105]. Similarly, γ-radiation, x-rays, and UV radiation have also been shown to activate NF-κB [106]. Egan *et al*. [107] found that IκB-kinase-β-dependent NF-κB activation provides radioprotection to the intestinal epithelium, and Panta *et al*. [108] found that the ataxia telangiectasia mutated (ATM) gene/protein and the catalytic subunit of DNA-dependent protein kinase activate NF-κB through a common mitogen-activated protein kinase kinase/extracellular signal-regulated kinase/p90(rsk) signaling pathway in response to distinct forms of DNA damage. Why these proapoptotic agents activate NF-κB, however, is not clear.

Although in most instances NF-κB mediates chemoresistance [109] or radioresistance [110], in other cases it may mediate apoptosis [111]. Additionally, chemotherapeutic agents activate NF-κB by mechanisms that appear to differ from that of TNF. The antiapoptotic activity of NF-κB in tumors contributes to their acquired resistance to chemotherapy.

Degradation of IκB is a seminal step in the activation of NF-κB. The IκB kinases, IKKα and IKKβ, have been implicated in both IκB degradation and subsequent modification of NF-κB. Using MEFs devoid of both IKKα and IKKβ genes (IKKα/β(-/-), Tergaonkar *et al*. [112] showed that the novel IκB degradation mechanism induced by the chemotherapeutic agent DoxR) does not require the classic serine 32 and 36 phosphorylation or the proline-glutamic acid-serine-threonine domain of IκBα. The degradation of IκBα was also shown to be partially blocked by the phosphatidylinositol 3-kinase inhibitor LY294002 and to be

mediated by the proteasome. The free NF-κB generated by the DoxR-induced IκB degradation in IKKα/β(-/-) cells activated the chromatin-based NF-κB reporter gene and the expression of the endogenous target gene, IκBα [112]. These results also imply that modification of NF-κB by IKKα or IKKβ, either prior or subsequent to its release from IκB, is not essential for NF-κB–mediated gene expression, at least in response to DNA damage. In addition, DoxR-induced cell death in IKKα/β(-/-) MEFs was enhanced by the simultaneous inhibition of NF-κB activation by the blockage of protea-somal activity [112] . These findings show an additional pathway for activating NF-κB during anticancer therapy and provide a mechanistic basis for the observation that proteasome inhibitors can be used as adjuvant agents in chemotherapy. Thus, DoxR-induced NF-κB activation is an IKK-independent IκBα degradation pathway.

The new disaccharide anthracycline MEN 10755 induces activation of both NF-κB and p53 transcription factors in A2780 cells [113]. However, the pharmacologic inhibition of NF-κB activation does not modify the sensitivity of A2780 cells to MEN 10755 treatment. To better characterize the role of NF-κB in MEN 10755-induced cytotoxicity, Camarda *et al*. [113] analyzed the expression of a number of genes known to be regulated by NF-κB and found that none of them was modified by MEN 10755. On the contrary, as our results also suggest, the p53 DNA damage-responsive pathway is fully activated in A2780 cells, with several genes controlled by p53 being either up- or downregulated, according to the described action of p53 on their promoters [113]. Thus, in the A2780 cell line, the role of p53 in transducing the DNA-damage signal appears to be relevant, whereas NF-κB, although activated, appears to be non-functional. Other human carcinoma cell lines also activate NF-κB DNA binding in response to MEN 10755, but again this binding does not always lead to target gene activation. These results suggest that other tumor type-specific factors different from those that merely activate influence NF-κB transcriptional activity. Therefore, care should be taken when considering the pharmacologic inhibition of NF-κB as a means to improve anticancer therapy.

Chemotherapeutic agents simultaneously induce transcription factors p53 and NF-κB. p53 induction can activate an apoptotic program, and resistance to chemotherapy correlates with the loss of a functional p53 pathway. By contrast, NF-κB prevents apoptosis in response to chemotherapeutic agents. Tergaonkar *et al*. [114] analyzed the p53 response in IKKα/β(-/-) MEFs, which lack detectable NF-κB activity. Compared with wild-type fibroblasts, IKKα/β(-/-) fibroblasts showed increased cell death and p53 induction in response to the chemotherapeutic agent DoxR [114]. Reconstitution of IKKβ, but not IKKα, increased murine double minute 2 (Mdm2) levels and decreased DoxR-induced p53 stabilization and cell death. IKKβ-mediated effects required its kinase function and were abrogated by coexpression of the dominant negative mutant IκBα (IκBαM), thus suggesting a role for NF-κB in the blocking of chemotherapy-induced p53 and cell death.

2.5. Oncogenes Activate NF-κB

Several oncogene products that can activate NF-κB have been identified, including *ras* [115], *bcr-abl* [116], and *myc* [117]. Oncogenic Ha-Ras–induced signaling also activates NF-κB transcriptional activity, which is required for cellular transformation [118]. How these genes induce NF-κB activation, however, is poorly understood. Oncogenic Ras enhances NF-κB transcriptional activity through Raf-dependent and Raf-independent mitogen-activated protein kinase signaling pathways [119]. NF-κB activation was also found to be required for apoptosis induced by oncogenic Ras [120]. As many as 50% of all cancers (mostly solid tumors) express activated ras [121], which can lead to NF-κB activation. Bcr-abl, which has been shown to cause chronic myelogenous leukemia, can elicit survival signals through induction of NF-κB [122]. NF-κB activation is also required in Bcr-abl–mediated transformation. Finally, the oncogene c-myc, which can mediate tumor cell survival, has also been shown to be regulated by NF-κB [117].

2.6. NF-κB Activation is Required for Cell Proliferation

Several genes that mediate cell proliferation are also regulated by NF-κB, including the growth factors TNF, IL-1β, and IL-6 [123]. For instance, TNF was shown to be a growth factor for glioblastoma cells [124] and cutaneous T-cell lymphomas [97], IL-1β for acute myelogenous leukemia [125], and IL-6 for multiple myeloma [126] and head and neck squamous cell carcinoma [127]. Suppression of NF-κB in these tumors downregulates the cytokine expression and inhibits tumor cell proliferation. Besides growth factors, certain cell cycle regulatory proteins such as cyclin D1, which is required for transition of cells from the G1 to S phase, are also regulated by NF-κB [128]. Additionally, in some cells, prostaglandin E_2 (PGE_2) was shown to induce proliferation of tumor cells, and the synthesis of cyclooxygenase-2, which controls PGE_2 production, was also shown to be regulated by NF-κB activation [129].

The constitutive activation of NF-κB also appears to have a role in cell proliferation. Bargou *et al*. [130] showed that proliferation of Hodgkin/Reed-Sternberg cells depended on activation of NF-κB. Furthermore, constitutive NF-κB prevented Hodgkin's lymphoma cells from undergoing apoptosis under stress conditions [130].

It was further shown that growth factors such as epithelial growth factor (EGF) and platelet-derived growth factor induce proliferation of tumor cells through activation of NF-κB [131]. The EGF receptor was found to engage a receptor-interacting protein and NIK to activate NF-κB, thus identifying a novel receptor-tyrosine kinase signalosome [132]. NF-κB signaling was also shown to promote both cell survival and the formation of neurite processes in nerve growth factor–stimulated PC12 cells [133].

2.7. Activation of NF-κB Inhibits Apoptosis

Both the prosurvival and antiapoptotic functions of NF-κB were recently reviewed [134]. Several gene products that negatively regulate apoptosis in tumor cells, including inhibitor of apoptosis proteins (IAPs) 1 and 2, X-linked IAP, cellular Fas-associated death domain-like interleukin-1β-converting enzyme (FLICE)-like inhibitory protein (cFLIP), were shown to be controlled by NF-κB activation (Table **4**) [134]. For example, Bcl-x_L suppressed cytochrome C release from the mitochondria, the IAPs inhibited caspase-3 and caspase-9 [135], and FLIP inhibited caspase-8 [136]. A n

Table 4. A List of Genes Regulated by NF-κB

Cytokines	**Receptors**	TIEG	**Cell proliferation**	**P450**	**Miscellaneous**
TNFα[123]	β2 microglobulin		COX-2[129]	CYP2C11	α-1 Acid glycoprotein
TNFβ	μ-opioid receptor	**Transcription factors**	c-myc	CYP2E1	a1-Antitrypsin,
IL-1α[123,125]	2A	NF-κB1			a2(1)-Collagen
IFN-α	A1 Adenosine receptor	NF-κB2	**Cell cycle**	**Oxidative stress**	α-Fetprotein
IFN-γ	Amiloride-sensitive sodium channel	c-Rel	Cyclin D1[128]	XOD	AMH
IL-1β[123,126]	Androgen receptor	p53	p21	GST	Apolipoprotein C III
TRAIL	B 7.1	RelB	Cyclin D2	Mn SOD	β-Amyloid
M-CSF	Bradykinin B-1 receptor	IκBα	Cyclin D3	iNOS	Biglycan
RANTES	BRL-1	c-fos	GADD 45β		Caveolin-1
FAS ligand	CCR 5	c-myb		**Enzymes**	Claudin-2
Lymphotoxin α	CCR7	JunB	**Viruses**	γ-GCS	Clone 156
Lymphotoxin β	CD 137	E2F3a	HIV-1	11bGSD2	Clone 330
G-CSF	CD 154	Elf 3	Epstein-Barr virus (Wp promoter)	12-LOX	Clone 68
GM-CSF	CD 23	ELYS	Hepatitis B virus (pregenomic promoter)	5-LOX	Connexin 32
IL-2	CD 40	ETR 1001	HSV	ABC transporters	Epsilon-globin
IL-6[86]	CD 48	IRF-1	SV-40	ADH	Factor VIII
IL-8[111]	CD 69	IRF-2	Adenovirus (E3 region)	ARFRP 1	Gadd45β
IL-9	CD 83	IRF-4	Avian leukosis virus	Aromatase	Galectin 3
IL-10	CD 95	IRF-7	Bovine leukemia virus	BACE	GIF
IL-11	EGFR[132]	Mail	Cytomegalovirus	Catepsin B and ceramide glycosyl transferase	GS 3686
IL-12	Gal 1 receptor	Nurr1	JC virus	Collagenase 1	HMG-14
IL-13	GFBP-2	Stat5a	Human papillomavirus type 16	CRAD1 and 2	K15 keratin
IL-15	Glucocorticoid receptor	WT1	SIV	Dihydrodiol dehydrogenase	K3 keratin
NK4	IGFBP-1			DT-diaphorase	K3 keratin
GCP-2 (CXCL6)	IL-1 receptor antagonist	**Metastasis**	**Acute phase response proteins**	Gelatinase B	K6 keratin
Gro α	IL-2 receptor α-chain	ICAM-1	Angiotensinogen	GSTP1-1	Laminin B2 chain
Gro γ	Immunoglobulin γ4	VCAM-1	β-defensin-2	Guanylyl cyclase α	mCGM3
Gro-1	Immunoglobulin Cγ1	ELAM-1	C4b binding protein	H^+-K^+-ATPase a2	Mts1
ICOS	Immunoglobulin E heavy chain	E-selectin	Complement factor B	Heparanase	MUC-2
IL-1 receptor antagonist	Immunoglobulin κ light chain	Endoglin	Complement factor C4	HO-1	Mx 1
IP-10	Invariant chain II	Fibronectin	C-reactive protein	Hyaluronan synthase	Neutrophil gelatinase-assosiated lipocalin
KC	Lox-1	MadCAM-1	LPS binding protein	Iodothyronine deiodinase	NLF 1
BMP-2	Mdr 1	P-selectin	Pentraxin PTX3	Lysozyme	P11
CCL15	MHC class (HLA-B7)	Tenascin-C	SAA1 and SAA2	MKP-1	PAI-1
CCL22	MHC class I (H-2Kb)	DC-SIGN	Tissue factor-1	N-Acethylglucosaminyltransferase 1	Pax 8
CCL28	Neuropeptode YY1-receptor	MMP-3	Urokinase-type plasminogen activator	NGAL	PCBD
CCL5	NMDA receptor subunint NR-1	MMP-9[139]		NQO1	Perforin
CD40 ligand	NMDA receptor subunit	CXCR4[151]	**Proteins involved in antigen presentation**	PDE 7A1	PGK
CINC-1		KAI1/CD82[137]	Tapasin	PGES\L-PGDS	PSA
CXCL 11		uPA[142]		Phospholipase A2	RICK
ENA-78 (CXCL5)					S100A6 (calcyclin)
Eotaxin		**Antiapoptosis**			Spergen-1
Erythropoietin		Bcl-2[134]			Syndecan-4
MCP-1/JE		Bcl-x_L[134]			TFPI-2
		Bfl1/A1[134]			

(Table 4. Contd….)

MIP-1α,β	Nod 2	TRAF-1[134]	LMP 2	Phospholipase C δ1	Transferrin
MIP-2	Polymeric Ig receptor	TRAF-2[134]	TAP 1	PIM-1	Urokin 16
MIP-3α	RAF receptor 1	TRAF6	Complement B	PTGIS	UBE 2M
Mob-1	RAGE	c-FLIP[134]	Complement receptor 2	RACK 1	UCP-2
Neutrophil-activating peptide 78	T-cell receptor β chain	IAPs[134]	Complement component 3	REV 3	Vimentin
NK-1R	T-cell receptor/ CD3γ	Survivin [135]		Seprin 2A	Wilms' tumor suppressor gene
Stem cell factor	TNF-Receptor, p75/80	A20	**Kinases**	SNARK	
Angiotensinogen	DR4[167]	Bax	PI3K[143]	TERT	
TCA3	DR5[167]	Caspase-11	PlK3	Transglutaminase	
TFF3		FAP-1	PKCδ		
TSP-1	**Early response genes**	Fas-ligand	MAP 4K1		
TSP-2	p62	IEX-1L			
	p22/RPG1	Nr13			
	B 94				
	Egr-1	**Angiogenesis**			
		VEGF[144,145]			

TNF, tumor necrosis factor; IL, interleukin, IFN, interferon; TRAIL, tumor necrosis factor–related apoptosis-inducing ligand; M-CSF, macrophage colony-stimulating factor; RANTES, regulated upon activation, normal T-cell expressed and secreted; G-CSF, granulocyte colony-stimulating factor; GM-CSF, granulocyte-macrophage colony-stimulating factor; NK, neurokinin; GCP, granulocyte chemotactic protein; CXCL, CXC chemokine ligand; Gro, growth-related oncogene; ICOS, inducible costimulator; IP-10, IFN-γ–inducible protein 10; KC, Kupffer cells; BMP, bone morphogenic protein; CCL, chemokine ligand; CINC, cytokine-induced neutrophil chemoattractant; CXCL, CXC ligand, ENA-78, epithelial cell-derived neutrophil-activating protein 78; MCP-1, monocyte chemoattractant protein-1; MIP, macrophage inflammatory protein; TCA, T-cell activation; TFF3, trefoil factor 3; Tsp, thrombospondin; BRL, ; EGFR, epidermal growth factor receptor; GFBP, growth factor binding protein; IGFBP, insulin-like GFBP; Lox-1, lectin-like oxidized low-density lipoprotein receptor; Mdr, Mdr1, multidrug resistance mediator 1; MHC, major histocompatibility complex; NMDA, N-methyl-D-aspartate; Ig, immunoglobulin; RAGE, receptor of advanced glycation end products; DR, death receptor; Egr-1, early growth response 1; TIEG, TGFβ-inducible early gene; NF-κB, nuclear factor κB; IRF, interferon regulatory factor; ICAM-1, intercellular adhesion molecule-1; VCAM-1, vascular cell adhesion molecule 1; ELAM, endothelial leukocyte adhersion molecule; MadCAM-1, mucosal addressin cell adhesion molecule; DC-SIGN, dendritic cell surface C-type lectin; MMP, matrix metalloproteinase; CXCR, CXC chemokine receptor; TRAF, TNF-receptor associated factor; c-FLIP, c-FLICE inhibitory protein; IAPs, inhibitors of apoptosis; IEX-1L, immediate early response factor 1; VEGF; vascular endothelial growth factor; COX-2, cyclooxygenase-2; GADD, growth arrest and DNA damage–inducible; HSV, herpes simplex virus; SV-40, simian virus 40; SIV, simian immunodeficiency virus; SAA, serum amyloid A proteins; LMP, latent membrane protein; Pl3K, phosphatidylinositol 3 kinase; PIK3, polo-like kinase 3; MAP, mitogen-activated protein; CYP; cytochrome p450; XOD, xanthine oxidase; GST, glutathione S-transferase; Mn SOD, superoxide dismutase; iNOS, inducible nitric oxide synthase; GCS, glutamylcysteine synthetase; LOX, lipoxygenase; ADH, alcohol dehydrogenase; ARFRP 1, ARF-related protein 1; CRAD, conditionally replicating adenoviruses; NQO1, NAD(P)H quinone oxidoreductase 1; TERT, telomerase catalytic subunit; AMH, anti-mullerian hormone; HMG-14, high mobility group 14; Nod 2, Nucleotide oligomerization domain 2; TAP1, Human transporter associated with antigen processing 1; BACE, β-secretase, GSTP1, glutathione S-transferase P1; MKP-1, MAP kinase phosphatase-1; NGAL, Neutrophil gelatinase-associated lipocalin; NQO, NAD(P)H:quinone oxidoreductase; PGE, PGE synthase, L-PGDS, lipocalin-type PGD synthase, PIM, phosphatidylinositol; PTGIS, prostacyclin synthase; Rack1, Receptor for activated C kinase 1; SNARK, SNF1/AMP kinase-related kinase; GIF, growth inhibitory factor; PSA, prostate-specific antigen; RICK, Rip-like interacting caspase-like apoptosis-regulatory protein kinase; TFPI, tissue factor pathway inhibitor; UCP, uncoupling protein. Source: http://www.ncbi.nih.gov/entrez/query.fcgi, http://people.bu.edu/gilmore/nf-kb/.

antiapoptotic role of NF-κB has been alleged in T-cell lymphoma, osteoclasts, melanoma, pancreatic cancer, bladder cancer, and breast cancer. Cell types that display an antiapoptotic role for NF-κB include B cells, T cells, granulocytes, macrophages, neuronal cells, and smooth muscle cells [57].

Although rare, there are systems in which NF-κB plays a proapoptotic role in addition to its more common antiapoptotic role. This is seen, for example, in B-cells, T-cells, neuronal cells, and endothelial cells. These opposing effects of NF-κB are thought to be cell-type specific and/or dependent on the inducing signal (e.g., IL-1, TNF, UV radiation). These outwardly paradoxical effects of NF-κB on apoptosis may result from the different activation pathways of NF-κB that cause the expression of proteins that promote (e.g., Fas, c-myc, p53, and IB) or inhibit (e.g., TRAF2, IAP proteins, and Bcl-2-like proteins) apoptosis. In addition, NF-κB activation variably regulates cell cycle proteins (e.g., cyclin D1 and CDK2 kinase) and their interaction with various cellular components (e.g., p300 and p53) that promote or induce apoptosis [134]. Along similar lines, Shinohara *et al*. [137] showed that NF-κB activation induces the expression of the metastasis suppressor gene KAI1/CD82 in lung cancer cell lines expressing mutant p53. Mucosa-associated lymphoid tissue (MALT) lymphoma, the most common extranodal lymphoid cell neoplasia that frequently follows chronic bacteria-induced inflammation in various tissues, is also related to NF-κB inhibition of apoptosis. MALT lymphomas are characterized genetically by the t(11;18)(q21;q21) translocation, which yields chimeric transcripts encoding structurally distinct API2/MALT1 fusion proteins. Stoffel *et al*. [138] provided functional evidence for the contribution of API2/MALT1 fusion proteins to the transformation of cells in culture by activating the NF-κB pathway through a RelB/p50 dimer. They also provided evidence of the emerging role of the NF-κB signaling pathway in the inhibition of apoptosis. Thus,

activation of NF-κB and inhibition of p53-mediated apoptosis by API2/MALT 1 fusions promote oncogenesis.

2.8. NF-κB Mediates the Invasion of Tumor Cells

When cancer treatment is ineffective, the tumor cells remaining after treatment inevitably infiltrate the surrounding normal tissue, which leads to tumor recurrence. According to recent studies, the ability of tumor cells to digest the extracellular matrix (ECM) by secreting proteolytic enzymes correlates well with their tissue invasiveness. [139-141] For most primary human tumors, invasion is thought to be accomplished, at least in part, by proteases — serine, cysteine, and metalloproteinases — that penetrate connective tissue barriers, induce vascular remodeling, and destroy normal tissue. Several proteases (e.g., matrix metalloproteinases [MMPs] and the serine protease urokinase-type plasminogen activator [uPA]) that influence the invasive characteristics of tumors are regulated by NF-κB [139-141].

MMPs are a pivotal family of zinc enzymes responsible for the degradation of ECM components, including basement membrane collagen, interstitial collagen, fibronectin, and various proteoglycans. MMPs promote the growth of cancer cells through the interaction of ECM molecules and integrins, thereby cleaving insulin-like growth factors and shedding transmembrane precursors of growth factors, including TGF-β. MMPs promote angiogenesis by increasing the bioavailability of proangiogenic growth factors. MMPs also regulate invasion and migration by degrading structural ECM components — in particular, by cleaving laminin-5. Notably, it has been shown that MMP-9 expression is regulated transcriptionally through NF-κB elements within the MMP-9 gene [139]. Bond *et al*. [141], showed by using an adenovirus that overexpressed the inhibitory subunit IκBα that NF-κB activation was an absolute requirement in the upregulation of MMP-9.

uPA is another critical protease involved in tumor invasion and metastasis. Novak *et al*. [140] reported that the transcriptional activation of the uPA gene by phorbol myristate, IL-1, and TNFα requires the induction of NF-κB activity and the decay of its short-lived repressor protein IκBα. Wang *et al*. [142] also reported that uPA was overexpressed in pancreatic tumor cells and that its overexpression was induced by constitutive RelA activity. The uPA promoter contains an NF-κB binding site that directly mediates the induction of uPA expression by RelA. Treating pancreatic tumor cell lines with the NF-κB inhibitors dexamethasone and N-tosyl-L-phenylalanine chloromethyl ketone abolished constitutive RelA activity and uPA overexpression [142]. These results showed that uPA is one of the downstream target genes induced by constitutively activated RelA in human pancreatic tumor cells and suggested that constitutive RelA activity plays a critical role in tumor invasion and metastasis.

It was further shown that constitutively active phosphatidylinositol 3′-kinase (PI3k) controls cell motility by regulating the expression of uPA through the activation of NF-κB [143]. Mahabeleshwar *et al*. [107] demonstrated that the activation of Syk, a protein-tyrosine kinase, suppressed cell motility and NF-κB–mediated secretion of uPA by inhibiting PI3k activity in breast cancer cells. Thus, one of the ways to block the invasion of tumor cells is to target NF-κB and thus prevent its activation of the genes involved in cancer progression.

2.9. NF-κB Activation is Needed for Angiogenesis

It is now well recognized that the induction of the tumor vasculature (i.e., angiogenesis) is critical for the progression of tumors. Tumor vascularization has been shown to be dependent on chemokines (e.g., monocyte chemoattractant protein-1, IL-8), and growth factors (e.g., TNF, vascular EGF [VEGF]) produced by macrophages, neutrophils, and other inflammatory cells [144]. The production of these angiogeneic factors has been shown to be regulated by NF-κB activation [145].

NF-κB has been shown to mediate the upregulation of IL-8 and VEGF expression in bombesin-stimulated PC-3 cells [146]. Yu *et al*. [147] demonstrated that NF-κB expression was associated with VEGF expression and microvessel density in human colorectal cancer. Specifically, they detected the immunohistochemical expression of NF-κB, VEGF, and CD34 in 10 paraffin-embedded specimens of normal colorectal mucosa and 52 paraffin-embedded colorectal adenocarcinomas obtained by surgery or endoscopy; NF-κB and VEGF were significantly overexpressed and associated with increased microvessel density in the colorectal cancer specimens. These findings suggest that increased expression of NF-κB contributes to tumor angiogenesis in colorectal cancer and that VEGF plays an important role in mediating the NF-κB angiogenic pathway.

Highly metastatic melanoma cells were found to express high levels of constitutive NF-κB activity that was suppressed by transfection with IκBαM, and suppression of constitutive NF-κB activity inhibited tumor growth, prevented lung metastasis, and decreased microvessel density (angiogenesis), which correlated with a decrease in the level of IL-8 expression [148]. In another study, Pollet *et al*. (112) demonstrated that LPS directly stimulated endothelial sprouting *in vitro*, which is mediated through TRAF6. Inhibition of NF-κB activity, downstream of TRAF6, was sufficient to inhibit LPS-induced endothelial sprouting [149]. Also, inhibition of NF-κB activity blocked basic fibroblast growth factor-induced angiogenesis. These findings further underscore the role of NF-κB activation in mediating angiogenesis.

2.10. NF-κB is Involved in Metastasis of Tumor Cells

The metastasis of cancer requires the migration of cancerous cells both into and out of the vessel walls and their transport to other parts of the body. The ability of tumor cells to penetrate vessel walls is mediated by specific molecules that are expressed on the endothelial cells of the blood vessels in response to a number of signals from inflammatory cells, tumor cells, and so on. Among those special molecules are intercellular adhesion molecule-1, endothelial leukocyte adhesion molecule-1, and vascular cell adhesion molecule-1, all of which are expressed in response to NF-κB activation [150] (Table **4**).

Helbig *et al*. [151] demonstrated that NF-κB regulates the motility of breast cancer cells by directly upregulating the expression of CXCR4. They further showed that the NF-

κB subunits p65 and p50 bind directly to sequences within the –66 to +7 region of the CXCR4 promoter and activate transcription. They also showed that the cell surface expression of CXCR4 and that stromal derived factor-1α–mediated migration are enhanced in breast cancer cells isolated from mammary fat pad xenografts compared with parental cells grown in culture. A further increase in CXCR4 cell surface expression and stromal-1α–mediated migration was observed for cancer cells that metastasized to the lungs. These results implicate NF-κB in the migration and organ-specific homing of metastatic breast cancer cells. In addition, Fujioka *et al*. [152] showed that inhibiting constitutive NF-κB activity by expressing IκBαM suppressed liver metastasis, but not tumorigenesis, in the metastatic human pancreatic tumor cell line AsPc-1 in an orthotopic nude mouse model. These findings demonstrate the importance of the suppression of NF-κB activation in reducing the metastasis of cancer cells to other sites. Recently, Loercher *et al*. [153] further showed that NF-κB is an important modulator of the altered gene expression profile and malignant phenotype in squamous cell carcinoma.

2.11. NF-κB Activation Mediates Bone Loss

The adult skeleton is in a dynamic state, undergoing turnover by the coordinated actions of osteoclasts and osteoblasts. The focal net loss of bone at sites of inflammation in conditions such as arthritis and osteoporosis is due to an imbalance in favor of bone resorption. The loss of bone is also common to certain type of cancers, such as breast cancer, melanoma, and prostate cancer (Table **3**). Binding of RANKL, a member of the TNF superfamily, to its receptor RANK leads to the activation of NF-κB [154]. Furthermore, osteoclast differentiation and activation are mediated by RANKL, which is induced by osteoblasts or bone-lining cells in response to stimuli by the parathyroid hormone, IL-1, or PGE_2. Double-knockout mice for both NF-κB1 and NF-κB2 developed osteopetrosis because of a defect in osteoclast differentiation [155] and could not generate mature osteoclasts and B cells [156]. These findings establish the criticality of NF-κB in bone loss and development.

NF-κB proteins transmit growth factor signals between the ectoderm and the underlying mesenchyme during embryonic limb formation. In one study, the interruption of NF-κB activity through the virus-mediated delivery of an inhibitor resulted in a highly dysmorphic apical ectodermal ridge, a reduction in overall limb size, a loss of distal elements, and a reversal in the direction of limb outgrowth [157]. Yao *et al*. [158] showed that ethyl alcohol induced NF-κB nuclear translocation through p56lck in human osteoblast-like cells. This activation of NF-κB thus may contribute to bone loss through activation of signal transduction that results in production of the osteoclastogenic cytokine IL-6 in osteoblasts [158].

Inhibition of NF-κB signaling activity in Saos-2 cells resulted in a marked decrease in cellular proliferation accompanied by the induction of bone morphogenic proteins 4 and 7 and of the osteoblast-specific transcription factor Cbfa1, which heralds osteoblast differentiation, the induction of alkaline phosphatase, osteopontin and osteocalcin production, and an attendant increase in matrix deposition and mineralization *in vitro*. These results point to the negative regulation of osteoblast differentiation by NF-κB, with implications for the pathogenesis and progression of osteosarcomas [159]. They also suggest the critical involvement of NF-κB proteins in regulating the dynamics of bone development, thus raising new possibilities for the treatment of bone disorders.

3. SUPPRESSION OF NF-κB INHIBITS TUMORIGENESIS

The evidence presented in the previous sections suggests that activation of NF-κB can lead to tumor cell proliferation, invasion, angiogenesis, and metastasis. Therefore, the suppression of NF-κB in cancer cells may provide a target for treatment that prevents cancer.

3.1. Tumor-Suppressor Genes Inhibit NF-κB Activation

Whereas oncogenes can stimulate NF-κB activation, several tumor-suppressor genes, including the phosphatase and tensin gene (PTEN) [160], CYLD [161-163], and p53 [164], inactivate NF-κB. (Table **5**). The mutated in multiple advanced cancers (MMAC)/PTEN homologue is a natural antagonist of PI3k activity. PTEN is a lipid phosphatase responsible for downregulating the PI3k product phosphatidylinositol 3,4,5-triphosphate. In a glioma cell line that was stably transfected with MMAC/PTEN, the IL-1–induced DNA binding and transcriptional activity of NF-κB were both inhibited [160]. The ability of IL-1 to induce IκBα degradation or the nuclear translocation of NF-κB was, however, unaffected by MMAC/PTEN expression. Moreover, the IL-1–induced phosphorylation of p50 NF-κB was potently inhibited in MMAC/PTEN-expressing cells. However, the IL-1–induced interaction between the PI 3-k target Akt kinase and the IKK complex was antagonized by MMAC/PTEN.

Mayo *et al*. [128] found that PTEN inhibits TNF-stimulated NF-κB transcriptional activity. Specifically, PTEN did not block TNF-induced IKK activation, IκBα degradation, p105 processing, p65 (RelA) nuclear translocation, or DNA binding of NF-κB [165]. However, PTEN did inhibit NF-κB–dependent transcription by blocking the ability of TNF to stimulate the transactivation domain of the p65 subunit. Thus, maintenance of the PTEN tumor-suppressor protein is required to modulate Akt activity and to concomitantly control the transcriptional activity of NF-κB as an antiapoptotic transcription factor.

Familial cylindromatosis is caused by mutations in a gene encoding CYLD, a tumor-suppressor gene with deubiquitinating enzymatic activity that negatively regulates the activation of NF-κB. A loss of the deubiquitinating activity of CYLD correlates with tumorigenesis. CYLD inhibits activation of NF-κB by the TNFR family members CD40, X-linkedectodertmal dysplasoa receptor (XEDAR), and ectodysplason-A receptor (EDAR) in a manner that depends on the deubiquitinating activity of CYLD. Downregulation of CYLD by RNA-mediated interference augments both basal and CD40-mediated activation of NF-κB. The inhibition of NF-κB activation by CYLD is mediated, at least in part, by the deubiquitination and

Table 5. A List of Agents that Inhibit NF-κB Activation

Anti-inflammatory agents	**Antioxidants**	**HIV-1 protease inhibitors**	**Others**
Aspirin[57]	ADT	Nelfinavir	β-Amyloid protein
Indomethacin[57]	Apocynin	Ritonavir	β-Catenin
Diclofenac[57]	Benidipine	Saquinavir	γ-Glutamylcysteine synthetase[234]
Naproxen[57]	BHA		
Dexamethasone[57]	Carvedilol	**ACE inhibitor**	1,2,3,4,6-Penta-O-galloyl-β-D-glucose
Phenylbutazone[57]	Catechol derivatives	Quinadril	
Tamoxifen[57]	DHEAS		1,2,4-Thiadiazolidine derivatives
Sulfasalazine	DMDTC	**COX-2 inhibitor**	
Sulindac	DMSO	Celecoxib[216,218]	15-Deoxy-prostaglandin J(2)
Ibuprofen[57]	Disulfiram		17-Allylamino-17-demethoxygeldanamycin
Leflunomide[173]	Ebselen	**Interferon inducible protein**	
Trichodion	EPC-K1 21	p202a	2-Acetylaminofluorene
Acetaminophen	Ethyl pyruvate		3,4,5-Trimethoxy-4'-fluorochalcone
Gabexate mesilate	Ethylene glycol tetraacetic acid	**Methanesulfonilide antiarthritis inhibitor**	
Glucosamine sulfate	Iron tetrakis	T-614	4-Hydroxy-2-nonenal[183]
Petrosaspongiolide M	Mn-SOD		5′-Methylthioadenosine
Dexanabinol	Melatonin		Acrolein
BSASM	NAL	**PTK inhibitor**	Adenosine[226]
Cloricromene	NDGA	Herbimycin A[244]	AMP-activated protein kinase
Rolipram	Orthophenanthroline		Amrinone
Pranlukast	Hydroquinone and tert-butyl hydroquinone	**Farnesyl protein transferase inhibitor**	Anandamide
		SCH66336[220]	Angiopoietin-1
Immunosupressive agents	Ref-1		ANP
Cyclosporin	Tempol	**Protease inhibitor**	Antithrombin III
LF15-0195	Tepoxaline	Nafamostat mesilate	APC0576
15-Deoxyspergualin	2-Amino-3-cyano-4-aryl-6-(2-hydroxy-phenyl)		Arsenite[243]
6(5H)-Phenanthridinone and benzamide	N-Acetylcysteine	**PPARγ ligand**	Auranofin
		Pioglitazone	AvrA protein
			AZT
Chemopreventive agents	**Reactive oxygen species**	**Heat shock proteins**	Bcl-Abl[228]
Carboplatin	Nitric oxide	HSP70	Benfotiamine
Vesnarinone[232]		HSP72	Biliverdin
Cycloprodigiosin	**Antibacterial agents**		Biotinylated isopanepoxydone
(-)-Cycloepoxydon	Fosfomycin	**Statins**	
Campthothecin	5-Aminosalicylic acid	Atorvastatin	Bisphenol A
Gemcitabine	Clarithromycin	Erbstatin	BMT
Levamisole	Rifampicin	Fluvastatin	Bovine serum albumin
Epoxomicin	Roxithromycin	Lovastatin	BZLF1
			C5a
Angiogenesis	**Antidiabetes agents**	**Metals**	Carbon monoxide
2-Methoxyestradiol	Adiponectin	Chromium	Cyclopentones
TNP-470	Glimepiride	Cadmium	CYL-19s and CYL-26z
		Gold	CYLD[161-163]
Cell proliferation	**Tyrosine phosphatase inhibitors**	Lead	Desloratadine
6-Aminoquinazoline derivatives	Diamides[242]	Mercury	Disulfiram
Raxofelast	Pervanadate[242]	Zinc	Dobutamine
	Phenylarsine oxide[242]	Arsenic	DQ 65-79
Virus		Titanium	DTD
K1L	**Serine protease inhibitor**		E3330
HIV-1 Vpu protein	Pefabloc	**IκB-like protein**	E-73
Canine distemper virus		Encoded by ASFV	Ecabet sodium
Hypochlorite	**Phosphathidylcholine-phospholipase C inhibitor**		Hydroquinone
Inhaled isobutyl nitrite		**Acetylation inhibitor**	RKIP
Jesterone dimer	D609	HDAC[187]	Rocaglamide
KT-90 APC	Hyperosmolarity	Nucling	Saline
Losartin	Hypothermia	ox-LDL and HNE	Selenomethionine
Calcitriol	IMD-0354	OXPAPC	Siah2

(Table 5. Contd….)

CaMKK	Intravenous immunoglobulin	p8	SiRNA[191]
Campthothecin	Jesterone	p53[164]	SLPI
Caprofin	JSH-21	PACAP	SOCS1
Decoy oligonucleotides	JSH-23	PAN1	Sodium salicylate
Dehydroxymethylepoxyquinomicin	Kaposi's sarcoma-associated	Panepoxidone	SUN C8079
Diethyldithiocarbamic acid	herpesvirus K1 protein	PBN	Surfactant protein A
Diethylmaleate	Ketamine	PDTC	Survanta
Diltiazem	KL-1156	PEDF	TAT-SR-IκBα
DMF	LMB	Pentoxifylline	Tetrathiomolybdate
Epoxyquinol	Macrolide antibiotics	Photo oxidative stress [236]	Thalidomide[229]
Erythromycin	MAST205	PIAS	THI 52
Estrogen (E2)	MEB	Pirfenidone	Thiopental
Ethacrynic acid	Melatonin	PPM-18	TNAP
Ethyl 2-[(3-methyl-2,5-dioxo(3-pyrrolinyl))	Mercaptopyrazine	Probiotics	Tom1
	Mesalamine	Probiotics	Tranilast
Ethyl pyruvate	Methotrexate[230]	Prostaglandin 15-deoxy-Delta(12,14)-PGJ(2)	Triflusal
Evans blue	Monochloramine		Triglyceride-rich lipoproteins
Fibrates	Monomethylfumarate	15-deoxy-delta 12,14-prostaglandin J2	Troglitazone
Fish oil feeding	MTA	Prostaglandin A1	Tyrphostin
Flunixin meglumine	Murr1	Prostaglandin E2	Ursodeoxycholic acid
Flurbiprofen	MX781	Prostaglandin E2	Uteroglobin
Fox1j	Myxoma Virus MNF	PSK	Vasoactive intestinal peptide
G-120 3032	N-(p-coumaroyl) serotonin	Psychosine	vIRF3
Gangliosides	NEM	PTEN [160]	YopJ
Gemfibrozil	Neurofibromatosis-2 protein	PTX-B	ZAS3 protein
Ghrelin	nicotinamide, 3-aminobenzamide	Pyrithione	ZUD protein
GILZ	Nilvadipine	Pyrrolidinedithiocarbamic -acid	
Glucocorticoids	Nitrosoglutathione	Quinazolines	
Hydroquinone	Nitrosylcobalamin mild hypothermia	Rapamycin	
HSCO	N-Octylcaffeate	Rebamipide	
Human breast milk	N-Oleoyldopamine	RelA peptides	
Hycrochloride	NPM-ALK oncoprotein	Retinoic acid receptor-related orphan receptor-alpha	
	NRF	Ribavirin	
		Rifamides	
		Ritonavir	

ADT, anethole dithiolthione; BHA, butylated hydroxyanisole; DHEAS, dehydroepiandrosterone sulfate; DMDTC, dimethyldithiocarbamates; DMSO, dimethylsulfoxide; EPC-K1, phosphodiester compound of vitamin E and vitamin C; EGTA, ethylene glycol-bis(2-aminoethylether)-N,N,N',N'-tetraacetic acid; Mn-SOD, manganese superoxide dismutase; NAC, N-acetylcysteine; NAL, nacyselyn; NDGA, nordihydroguaiaritic acid phenolic antioxidants; Ref-1, redox factor 1; ACE, angiotensin-converting enzyme; COX-2, cyclooxygenase-2; PTK, protein tyrosine kinase; PPAR, peroxisome proliferator-activated receptor; HSP, heat shock protein; ASFV, African swine fever virus; HDAC, histone deacetylase; ANP, atrial natriuretic peptide; APC, activated protein C; BMT, o,o′-bismyristoyl thiamine disulfide; ALLnL, N-acetyl-leucinyl-leucynil-norleucynal; APNE, N-acetyl-DL-phenylalanine-b-naphthylester; BTEE, N-benzoyl L-tyrosine-ethylester; DCIC, 3,4-dichloroisocoumarin; LLM, N-acetyl-leucinyl-leucynil-methional; TLCK, N-a-tosyl-L-lysine chloromethyl ketone; TPCK , N-a-tosyl-L-phenylalanine chloromethyl ketone; Z-LLL, carbobenzoxyl-leucinyl-leucynil-leucynal; Z-LlnV, carbobenzoxyl-leucinyl-leucynil-norvalinal; a-MSH, alpha-melanocyte-stimulating hormone; ox-LDL, oxidized low density lipoprotein; HNE, Hydroxynonenal; HB-EGF, Heparin-binding epidermal growth factor-like growth factor; PACAP, Pituitary adenylate cyclase-activating polypeptide; THI 52; 1-naphthylethyl-6,7-dihydroxy-1,2,3,4- tetrahydroisoquinoline; PDTC, Pyrrolinedithiocarbamate; DMF, Dimethylfumarate; DTD, 4,10-dichloropyrido[5,6:4,5]thieno[3,2-d':3,2- d]-1, 2, 3-ditriazine; Tranilast; N-(3,4-dimethoxycinnamoyl)anthranilic acid, MEB; 2-(4-morpholynl) ethyl butyrate hydrochloride; FLIP, FLICE-Like Inhibitory Protein; PTX-B, pertussis toxin binding protein; RKIP; Raf Kinase Inhibitor Protein; GILZ, Glucorticoid-induced leucine zipper protein; LMB, Leptomycin B; MTA, Mevinolin; 5'-methylthioadenosine; NEM, N-ethyl-maleimide; NRF, NF-kB repression factor; OXPAPC, oxidized 1-palmitoyl-2-arachidonoyl-sn-glycero-3-phosphorylcholine; PEDF, pigment epithelium derived factor; PTX, Pentoxifylline; PBN, Phenyl-N-tert-butylnitrone; PIAS1, protein inhibitor of activatated STAT1; PSK, Protein-bound polysaccharide; VEGF; Vascular endothelial growth factor; Avr, avirulence; AZT, 3'-azido-3'-deoxythymidine; CaMKK, Calcium/calmodulin-dependent protein kinase kinase; DMF, dimethylfumarate; HSCO, Hepatoma Subtracted-cDNA library Clone One; MEB, 2-(4-morpholynl) ethyl butyrate hydrochloride; NPM, nucleophosmin; ALK, anaplastic lymphoma kinase;PAN, PYRIN and NACHT domain; Ppm-18, a-benzoylamino-1,4-naphthoquinon; Siah, seven in absentia homolog; SLPI, secretory leukocyte protease inhibitor; SOCS, Suppressor of cytokine signalin;THI52, 1-naphthylethyl-6,7-dihydroxy-1,2,3,4-tetrahydroisoquinoline; TNAP, TRAFs and NIK-associated protein; vIRF, human herpesvirus 8-encoded interferon regulatory factor; YopJ, Yersinia outer protein; ZUD, ZU 5 and death domain-containing protein.

Source: http://www.ncbi.nih.gov/entrez/query.fcgi, http://people.bu.edu/gilmore/nf-kb/.

inactivation of TRAF2. CYLD also binds to the NF-κB essential modulator (also known as IKKγ) component of the IKK complex and appears to regulate its activity through deubiquitination of TRAF2, since TRAF2 ubiquitination can be modulated by CYLD [161-163]**.** These results indicate that CYLD is a negative regulator of the cytokine-mediated activation of NF-κB that is required for appropriate cellular homeostasis of skin appendages. In addition, the inhibition of CYLD increases resistance to apoptosis, suggesting a mechanism through which the loss of CYLD contributes to oncogenesis. This effect can be reversed by aspirin derivatives that inhibit NF-κB activity, which suggests a therapeutic intervention strategy to restore growth control in patients with familial cylindromatosis.

Whereas NF-κB provides a proliferative signal, p53 is a mediator of antiproliferation. A mechanism by which p53 regulates NF-κB function and cell cycle progression has been proposed: p53 was shown to downregulate the expression of cyclin D1 by inhibiting the expression of the Bcl-3 protein [166], a member of the IκB family that functions as a transcriptional coactivator for p52 NF-κB; p53 also reduced p52/Bcl-3 complex levels. At the same time, p53 induced a significant increase in the association of p52 and histone deacetylase 1 (HDAC1). Importantly, p53-mediated suppression of the cyclin D1 promoter was reversed by coexpression of Bcl-3 and inhibition of p52 or deacetylase activity. p53 therefore induces a transcriptional switch by which p52/Bcl-3 activator complexes are replaced by p52/HDAC1 repressor complexes, resulting in active repression of cyclin D1 transcription.

The alternative reading frame (ARF) tumor suppressor is a central component of the cellular defense against oncogene activation. In addition to activating p53 by binding to Mdm2, ARF can repress the transcriptional activity of the antiapoptotic RelA(p65) NF-κB subunit. Rocha *et al*. [166] demonstrated that ARF induces the ataxia-telangiectasia mutated and Rad3-related (ATR) and checkpoint kinase 1 (Chk1)-dependent phosphorylation of the RelA transactivation domain at threonine 505, a site required for ARF-dependent repression of RelA transcriptional activity [166]. Consistent with this finding, ATR and Chk1 are required for ARF-induced sensitivity to TNF-induced cell death. Significantly, ATR activity was also required for ARF-induced p53 activity and inhibition of proliferation; ARF achieved these effects by activating ATR and Chk1. These results reveal novel functions for ARF, ATR, and Chk1, together with a new pathway regulating the RelA NF-κB function. Moreover, this pathway provides a mechanism through which ARF can remodel the cellular response to an oncogenic challenge and execute its function as a tumor suppressor.

3.2. Inhibition of NF-κB Inhibits Tumorigenesis

That NF-κB is critical for tumorigenesis is also indicated by studies showing that suppression of NF-κB blocks tumorigenesis. For instance, fibrosarcoma and colore tumors grown in nude mice were induced to undergo apoptosis after infection with an adenovirus expressing ctal a modified form of IκBα and the systemic delivery of campothecin-11 chemotherapy [7]. In addition, HT1080 fibrosarcoma cells exposed to ionizing radiation, TNF-α, or daunorubicin exhibited enhanced activation of NF-κB, and inhibition of NF-κB dramatically enhanced apoptosis in response to radiation or daunorubicin [9]. There are, however, studies suggesting that suppression of NF-κB has no effect on apoptosis. For example, the stable transfection of *iκbα* did not result in enhanced cytotoxicity in response to chemotherapeutic agents, despite the ability of these agents to activate NF-κB [167].

3.3. Chemopreventive Agents Inhibit NF-κB Activation

Because of the critical role of NF-κB in proliferation, invasion, angiogenesis, and metastasis of tumors, there has been great interest in modulators of the NF-κB signaling pathway. Several agents that have been described as natural chemopreventive agents have also been found to be potent inhibitors of NF-κB activation (Table **6**). How these agents suppress NF-κB activation is becoming increasingly apparent. For example, these inhibitors may block any one or more steps in the NF-κB signaling pathway, such as the incoming signals that activate the NF-κB signaling cascade, the translocation of NF-κB into the nucleus, DNA binding of the dimers, and interactions with the basal transcriptional machinery. For instance, whereas curcumin blocks IKK activation [126], resveratrol suppresses p65 translocation to the nucleus [86] and caffeic acid phenethyl ester (CAPE) suppresses the binding of the p50-p65 complex directly to the DNA [168].

Other chemopreventive agents that have been shown to suppress NF-κB activation include betulinic acid, ursolic acid, piceatannol, green tea polyphenols, oleandrin, and anethole [89, 169-172]. Similarly leflunomide, a drug approved for the treatment of rheumatoid arthritis, was also shown to suppress NF-κB activation [173].

3.4. Some Cytokines Block NF-κB Activation

Several cytokines have also been found to suppress NF-κB activation. These include cytokines produced by Th2 cells, such as IL-4, IL-11, IL-13, and IL-10 [174], and hormones of the endocrine system, such as human chorionic gonadotropin [175], melanocyte-stimulating hormone (MSH) [176], and growth hormone [177] (Table **7**). Besides these cytokines, IFN-α was also found to be a potent suppressor of NF-κB activation [178]. How these agents suppress NF-κB activation varies. For instance, the nuclear translocation of the RelA subunit and the degradation of IκBα induced by TNF were prevented by IL-13 and human chorionic gonadotropin in one study [175]. Both IL-10 and IL-13 suppressed nuclear localization of NF-κB and increased IκBα mRNA expression in another study [179].

4. NF-κB IS A POTENTIAL TARGET FOR DRUG DEVELOPMENT

NF-κB is an ideal target for anticancer drugs. Given the hyperproliferative nature of cancer, which involves transformation, initiation, promotion, angiogenesis, invasion, and metastasis, and the diversity of its clinical presentation, aggressiveness, and current treatment strategies, the implication is that an equally diverse number of potential targets exist in the molecular pathways leading to its formation. In this regard, several strategies have been used to block the activation of NF-κB, and a wide range of compounds, such as IKK inhibitors, inhibitory peptides, antisense RNA, and proteasome inhibitors, have been found to block various steps leading to NF-κB activation (Table **8**).

4.1. Inhibitors of Proteasomes Block NF-κB Activation

Proteasome inhibitors block the 26S proteasome, which is necessary to degrade the IκBα inhibitory subunit after its phosphorylation and ubiquitination in the cytoplasm, and thus its subunit release from the NF-κB complex [55]. Some of the well-known proteasome inhibitors are peptide aldehydes, such as ALLnL, LLM, Z-LLnV, and Z-LLL; lactacystine; PS-341; N-cbz-Leu-Leu-leucinal (MG132); MG115; and ubiquitin ligase inhibitors (for a review, see reference[57]) (Table **8**).

Table 6. A List of Natural Products that Suppress NF-κB Activation

α-Lipoic acid	Cheongyeolsaseuptang	Isomallotochromanol and	Tetrandine
α-Pinene	Chromene derivatives	isomallotochromene	Theaflavin
α-Tocopherol	Chicory root	Isorhapontigenin	Trilinolein
α-Torphryl acetate	Chitosan	Kahweol	Triptolide
α-Torphryl succinate	Curcumin[219,247-266]	Quinic acid	Ursolic acid [89]
β-Lapachone[235]	Cyclolinteinone	Kamebakaurin	Vitamin C
1′-Acetoxychavicole	Danshenshu	Lacidipine	Wedelolactone
acetate[213]	DDC	L-Ascorbic acid	Withaferin A
20(S)-Protopanaxatriol	Dehydroascorbic acid	Lazaroids	Wogonin
23-Hydroxyursolic acid	Diarylheptanoid	Lupeol	Wortmannin[238]
7-Amino-4-	Dibenzylbutyrolactone lignans	Luteolin	Yakuchinone A and B
methylcoumarin	Diferoxamine	Magnolol	Zerumbone[211]
Acetyl-boswellic acids	Digitoxin	γ-Mangostin	
Allyl-cysteine	Dihydroisoeugenol	Manassantins	**Others**
Amentoflavone	Dihydrolipoic acid	Mangiferin	1,2,4-Thiadiazolidine derivatives
Andalusol	DIM/I3C[212]	Nicotine	2-Amino-3-cyano-4-aryl-6-(2-hydroxy-phenyl)
Andrographolide	Ebselen	Oleandrin[171]	Aged garlic extract
Anethole[170]	EGCG	Onconase	Apple juice
Apigenin	Emodin[238]	Panduratin A	*Artemisia capillaris* Thunb extract
Artemisia sylvatica	ent-Kaurane diterpenoids	Palthenolide	*Artemisia iwayomogi* extract
Artemisinin	Ergothioneine	PC-SPES	Blueberry and berry mix
Astaxanthin	Evodiamine[214]	Phenethylisothiocyanate	Black raspberry extracts
Astragaloside IV	Epoxyquinone A	Phomol	Cat's claw bark
Aucubin	Falcarindol	Phytic acid	Extract of the stem bark of *Mangifera indica* L.
Avicin	Flavonoids	Phallacidin	Extracts of *Ochna macrocalyx*
Baicalein	Flavopiridol[223]	Piceatannol[227]	Fomes fomentarius methanol extracts
Bambara groundnut	Fungal gliotoxin	Piperine	Fructus benincasae recens extract
Bee venom	*Ganoderma lucidum* polysaccharides	Qingkailing,	*Ginkgo biloba* extract
Benzyl isothiocyanate	Garcinol	Shuanghuanglian	*Harpagophytum procumbens* extracts
Betulinic acid[172]	Genistein	Quercetin	*Kochia scoparia* fruit
Bis-eugenol	Geldanamycin	Resiniferatoxin	Omega 3 fatty acids
Bupleurum fruticosum	Genipin	Resveratrol[225,231]	*Phyllanthus amarus* extracts
phenylpropanoids	Ginkgolide B	Rhein	*Platycodi radix* extract
Cacospongionolide B	Glabridin	Rocaglamide	Red wine
Calagualine[210]	Glitoxin	Rotenone	*Sophorae radix* extract
Cancer bush	*Glossogyne tenuifolia*	Sanggenon C	Stinging nettle plant extracts
CAPE	Glycyrrhizin	Sanguinarine	*Tanacetum larvatum* extract
Capsaicin[240]	Guggulsterone[215]	Saucerneol	*Uncaria tomentosum* plant extract
Capsiate	Helenarin	Sauchinone	*Trichomomas vaginalis*
Carnosol	Hematein	Sesamin	*Xanthium strumarium* L.
Catalposide	Herbal compound 861	Sesquiterpene lactones	*Yucca schidigera* extract
Cinnamaldehyde/2-	Hydroxyethyl starch	Silymarin[209]	
methoxycinnamaldehyde	Hypericin	Sinomenine	
Cycloepoxydon	10Z-hymenialdisine	*Solana nigrum* L.	
Celastrol	Hypoestoxide	Sphondin	
Cepharantin	Indirubin-3'-oxime	Staurosporine	
Cytochalasin D	IRFI 042	Tansinones	
Conophylline			

SAC, garlic compound, DDC; Diethyldithiocarbamate, EGCG; Epigallocatechin-3-gallate (green tea polyphenols), Flavonoids (Crategus), Garcinol from extract of Garcinia indica fruit rind, IRFI 042; Vitamin E-like compound, 20(S)-Protopanaxatriol; ginsenoside metabolite, Apigenin; plant flavinoid, Calagualine ; fern derivative, Capsaicin; 8-methyl-N-vanillyl-6-nonenamide, Cyclolinteinone ; sponge sesterterpene, Emodin; 3-methyl-1,6,8-trihydroxyanthraquinone, *Kochia scoparia* fruit; methanol extract, K1L ; Vaccinia virus protein, Panduratin A from *Kaempferia pandurata*, Zingiberaceae, Phytic acid ; inositol hexakisphosphate, Rocaglamides ; Aglaia derivatives , Sanguinarine ; pseudochelerythrine, 13-methyl-[1,3]-benzodioxolo-[5,6-c]-1,3-dioxolo-4,5 phenanthridinium, Sesquiterpene lactones ; parthenolide; ergolide; guaianolides, Apigenin ;4',5,7-trihydroxyflavone, Baicalein; 5,6,7-trihydroxyflavone, Bambara groundnut ;Vignea subterranean, Bee venom ;melittin, Blueberry and berry mix; Optiberry, Cancer bush ; Sutherlandia frutescenss, Onconase;Ranpirnase, Catalposide; stem bark, Cat's claw bark ; *Uncaria tomentosa*; Rubiaceae, Chicory root ;guaianolide 8-deoxylactucin, Diarylheptanoid (7-(4'-hydroxy-3'-methoxyphenyl)-1-phenylhept-4-en-3-one), ent-kaurane diterpenoids ; Croton tonkinensis leaves, Harpagophytum procumbens , Devil's Claw extracts, PC-SPES ; 8 herb mixture, Qingkailing and Shuanghuanglian; Chinese medicinal preparations, *Solana nigrum* L. 150 kDa glycoprotein, Sphondin ; furanocoumarin derivative from *Heracleum laciniatum*, Tansinones ; *Salvia miltiorrhiza* Bunge, Labiatae roots, Sesamin from sesame oil, Triptolide ; PG490, extract of Chinese herb, Wogonin ;5,7-dihydroxy-8-methoxyflavone, Wortmannin ;fungal metabolite, Aged garlic extract ;allicin, DIM, 3,3'-Diindolylmethane. Source: http://www.ncbi.nih.gov/entrez/query.fcgi, http://people.bu.edu/gilmore/nf-kb/.

Table 7. A List of Proteins and Peptides that Inhibit NF-κB Activation

Interleukin	**Peptides**
IL-4[237]	TRAF6 inhibitory peptide
IL-11	NEMO binding peptide
IL-10[179]	p65 binding pepride[198]
IL-13[179, 239]	Boronic acid peptide
	NLS cell permeable peptides[195]
Interferon	Peptide nucleic acid-DNA decoys
IFN-α[147]	Peptide YY
	TIRAP inhibitor peptide
Growth factors and hormones	
VEGF	
Hepatocyte growth factor	
MSH[176]	
HB-EGF	
Chorionic gonadotropin[175,221]	

IL, Interleukin; IFN, interferon; VEGF, vascular epidermal growth factor; MSH; melanocyte stimulating hormone TIRAP, toll IL1 receptor domain-containing adapter protein; HB-EGF, Heparin-binding epidermal growth factor-like growth factor; TRAF, TNF receptor-associated factor; NEMO, NF-κB essential modulator; NLS, nuclear localization sequence. Source: http://www.ncbi.nih.gov/entrez/query.fcgi, http://people.bu.edu/gilmore/nf-kb/.

PS-341 blocks TNF-α-induced NF-κB activation in multiple myeloma cells through the inhibition of IκBα phosphorylation and the degradation of IκBα [180]. Proteasome inhibitors also inhibit the chymotrypsin-like activity of the proteasome complex. Fiedler *et al*. [181] have demonstrated that the pretreatment of cells with the proteasome inhibitor N-cbz-Leu-Leu-leucinal (MG-132) reverses TNFα-induced NF-κB activation by inhibiting proteasome-mediated IκBα degradation and NF-κB-mediated gene transcription [181].

Calpain inhibitor I is a cysteine protease inhibitor that is less potent than MG132 and MG115. Molecules such as lactacystine irreversibly block proteasome activity by acylating a threonine residue in the active site of the subunit X of the mammalian proteasome. Some serine protease inhibitors also act as proteasome inhibitors and block the phosphorylation and degradation of IκBα [182]. Retaining the NF-κB dimers in the cytoplasm by preventing the phosphorylation and degradation of IκBα is also an effective way to inhibit NF-κB, and this can be achieved by several signaling molecules such as nitric oxide, estrogen, oxidized low-density lipoproteins, 4-hydroxynonenal, and prostaglandin A.

4.2. Inhibitors of IKK Block NF-κB Activation

IκBα phosphorylation is a critical step in NF-κB activation, and compounds that block this phosphorylation also prevent IκBα's ubiquitination and further degradation. Recently, 4-hydroxy-2-nonenal, a lipid peroxidation product, has been shown to block this phosphorylation by directly inhibiting of IKK [183]. Also, 5-bromo-6-methoxy-β-carboline, a natural product derivative, is a nonspecific IKK inhibitor that inhibits the phosphorylation of IκBα and the subsequent activation of NF-κB; this compound has a 50% inhibitory concentration (IC_{50}) in the nanomolar range [184]. SC-514 is another novel inhibitor of IKK, specifically inhibiting IKKβ, which has an IC_{50} of appproximately 10 μM; it does not inhibit other IKK isoforms or other serine-

Table 8. Synthetic Compounds That Inhibit NF-κB Activation

IKK inhibitors	SN-50	**P42 MAPK inhibitor**
BAY-11-7082	FK506	AG 126
BAY-11-7085	Ro106-9920	
BMS-345541[186]	PS5190	**P38 MAPK inhibitor**
SPC839	TLCK ,TPCK	SB203580
AS602868	Cyclosporin A	
PS1145	DCIC	**PI3K-kinase inhibitor**
MNL120	Deoxyspergualin	LY294002
SC-514[185]	DFP	
	LLM	**PKC inhibitor**
Polyubiquitinqtion inhibitor	BTEE	RO31-8220
RO106-9920	MG115[57]	
	MG120	**MEK1/2 inhibitor**
Proteosome inhibitors	APNE	U0126
ALLnL [57]	Ubiquitin Ligase Inhibitors	
PS-341[57,218]	Z-LLL	**Protein kinase inhibitor**
Lactacystine, β-lactone [57]	Z-LLnV	A77 1726

IKK, IκB kinase; PI3K, phosphotidylinositole 3; PKC, protein kinase, protein kinase C, MEK, mitogen-activated protein kinase (MAPK) and extracellular signal-regulated kinase kinase. Source: http://www.ncbi.nih.gov/entrez/query.fcgi, http://people.bu.edu/gilmore/nf-kb/.

threonine and tyrosine kinases. SC-514 inhibits the native IKK complex and the recombinant human IKK-α/IKK-β heterodimer and IKK-β homodimer in a similar manner [185] (Table **8**).

BMS-345541 (4(2′-aminoethyl) amino-1,8-dimethylimidazo(1,2-α)quinoxaline) was identified as a selective inhibitor of the catalytic subunits of IKK with an IC_{50} of 4 μM for IKKα and 0.3 μM for IKKβ [186] (Table **8**). Burke *et al*. [186] proposed a binding model in which BMS-345541 binds to similar allosteric sites on IKKα and IKKβ and then affects the active sites of the subunits differently. BMS-345541 was also shown to have excellent pharmacokinetics in mice, to dose dependently inhibit the production of serum TNFα after intraperitoneal challenge with LPS in mice, and to represent an important tool for investigating the role of IKK in disease models [186].

4.3. Acetylation Inhibitors can Block NF-κB Activation

Histone acetylation modulates gene expression, cellular differentiation, and survival and is regulated by the opposing activities of histone acetyltransferases and HDACs (Table **5**). HDAC3 acts directly on nuclear Rel A, enabling its association with IκBα and its subsequent export from the nucleus, and is another potential target for gene transfer. In one study, the expression of HDAC3 in TNFα-stimulated HeLa cells repressed both NF-κB DNA binding and the levels of Rel A, with a corresponding increase in the level of inactive cytoplasmic IκBα–NF-κB complexes [187]. This repressive mechanism was shown to control the duration of NF-κB activation and thus may be a potential weapon against constitutive NF-κB activation.

4.4. Gene Transfer of Inhibitory Proteins can Block NF-κB Activation

The transfer of genes that encode inhibitory proteins is another strategy used to block the activation of NF-κB. The most direct target is the IκB gene. This transfer entails the modification of IκB at its specific phosphorylation (ser 32 and 36 switched with ala) and ubiquitination (lys 21 and 22 switched with arg) sites to prevent its degradation. In several studies, the superrepressor of NF-κB activity was a mutated, nondegradable IκBα resistant to phosphorylation and degradation that could be delivered into intestinal epithelial cells by using an adenoviral vector (Ad5 IκB). There was a strong inhibition of NF-κB activity because the superrepressor kept the NF-κB complex in the cytoplasm indefinitely [167]. These studies suggest an exciting approach for *in vivo* intestinal gene therapy and illustrate a key role for NF-κB in transcriptional regulation of the inflammatory phenotype of intestinal epithelial cells. Along similar lines, a nonphosphorylatable form of IκBα was recently shown to inhibit osteoclastogenesis and block bone resorption when injected into bone marrow macrophages [188].

4.5. Antisense and Small Interfering RNA can Block NF-κB Activation

Antisense agents that inhibit the expression of a target gene in a sequence-specific manner may be used against NF-κB for therapeutic purposes. Three types of anti-mRNA strategies can be distinguished: 1) single-stranded antisense oligonucleotides, 2) RNA cleavage triggered by catalytically active oligonucleotides, referred to as ribozymes, and 3) RNA interference induced by small interfering RNA (siRNA) molecules.

In one study, phosphorothioate antisense oligonucleotides to p65 inhibited *in vitro* growth, reduced soft-agar colony formation, and eliminated the ability of cells to adhere to an ECM in diverse transformed cell lines [189]. In addition, stable transfectants of a fibrosarcoma cell line expressing dexamethasone-inducible antisense RNA to p65 showed inhibition of *in vitro* growth and *in vivo* tumor development. Furthermore, in response to inducible expression of antisense RNA, a pronounced tumor regression was seen in nude mice. The administration of antisense but not sense p65 oligonucleotides also caused a pronounced inhibition of tumorigenicity in nude mice injected with diverse tumor-derived cell lines [189].

RNA interference is the mechanism of sequence-specific, posttranscriptional gene silencing initiated by double-stranded RNA homologous to the gene being suppressed and has the potential to be a new and powerful tool in cancer therapeutics [190]. Surabhi and Gaynor [191] showed the potential of siRNAs in decreasing the level of expression of p65 protein [191]. Also, when siRNAs were directed against IKKα, IKKβ, and the upstream regulatory kinase transforming growth factor β-activated kinase-1, both IKKα and IKKβ were found to be important in activating the NF-κB pathway [192]. Another study showed that the blocking of c-Rel in primary macrophages with siRNA selectively blocked IL-12 production and normalized the minimal, residual IL-12 levels in a lupus erythematosus-prone and diabetes mellitus-prone mouse strain with aberrant IL-12 production [193].

4.6. Peptides can Cross the Cell Membrane and Block NF-κB Activation

Another approach to inhibiting NF-κB activation is to use peptides that cross the cell membrane and block the nuclear localization of the NF-κB complex (Table **8**). For example, SN-50 and o,o′-bismyristoyl thiamine disulfide [194] work by mimicking the sequence of p50 responsible for transporting the NF-κB complex from the cytoplasm to the nucleus to block the normal import machinery. A dual nuclear localization signal (NLS) peptide has been shown to block the karyopherin–NF-κB interaction, which is required for the translocation of NF-κB into the nucleus [195]. Also, a novel peptide that selectively blocks the association of IKKγ with the rest of the IKK complex has been shown to inhibit NF-κB activation in response to proinflammatory cytokines in mice and at the same time to preserve basal NF-κB activity [196]. P1, another novel hybrid peptide derived from cecropin-A and magainin-2, reduced osteoclast differentiation in various osteoclast culture systems by inhibiting the NF-κB activation induced by RANKL [197].

NF-κB activation is known to require p65 phosphorylation at serine residues 276, 529, and 536 before it undergoes nuclear translocation. Small protein domains, termed protein transduction domains (PTDs), that are able to penetrate cell membranes can be used to transport other

proteins across the cell membrane. Our laboratory has identified two peptides from the p65 subunit of NF-κB (P1 and P6 from amino acid residues 271-282 and 525-537, respectively) that, when linked with a PTD derived from the third helix sequence of antennapedia, inhibited TNF-induced NF-κB activation *in vivo* [198]. Linkage to the PTD was not, however, required to suppress the binding of the p50-p65 heterodimer to the DNA *in vitro*, and PTD-p65-P1 had no effect on TNF-induced AP-1 activation. On the other hand, PTD-p65-P1 did suppress NF-κB activation induced by LPS, IL-1, okadaic acid, phorbol 12-myristate 13-acetate, H_2O_2, cigarette smoke condensate, and TNF. Although PTD-p65-P1 had no effect on the phosphorylation and degradation of the TNF-induced inhibitory subunit of NF-κB (IκBα) or on IκBα kinase activation, it blocked TNF-induced p65 phosphorylation and nuclear translocation. These peptides also suppressed the NF-κB–regulated reporter gene expression induced by TNF, TNFR1, the TNFR-associated death domain, TNFR-associated factor-2, NF-κB–inducing kinase, IκBα kinase, and p65. Notably, the suppression of NF-κB by PTD-p65-P1 enhanced the apoptosis induced by TNF and chemotherapeutic agents.

4.7. Antiinflammatory Agents Block NF-κB Activation

Additionally, several anti-inflammatory agents have been identified that suppress NF-κB activation. Examples include aspirin, ibuprofen, indomethacin, tamoxifen, dexamethasone, and sulindac [57] (Table **5**). However, the exact mechanism of their action is not fully understood. Kopp and Ghosh [190] demonstrated that the anti-inflammatory drugs sodium salicylate and aspirin inhibited the activation of NF-κB by preventing the degradation of the NF-κB inhibitor, IκB, so that NF-κB was retained in the cytosol.

Our laboratory has investigated the effect of almost a dozen different commonly used nonsteroidal anti-inflammatory drugs and dexamethasone on TNF-induced NF-κB activation and NF-κB–regulated gene products and cell proliferation [199]. We included dexamethasone, an anti-inflammatory steroid, in this analysis for comparison with NSAIDs. As indicated by DNA binding, none of the drugs alone activated NF-κB. In fact, all of the compounds inhibited TNF-induced NF-κB activation, but with highly variable efficacy. Specifically, the IC_{50}s required were 5.67 (aspirin), 3.49 (ibuprofen), 3.03 (sulindac), 1.25 (phenylbutazone), 0.94 (naproxen), 0.60 (indomethacin), 0.38 (diclofenac), 0.084 (resveratrol), 0.043 (curcumin), 0.027 (dexamethasone), 0.024 (celecoxib), and 0.010 (tamoxifen) mM. All drugs inhibited IκBα kinase and suppressed IκBα degradation and NF-κB–regulated reporter gene expression. They also suppressed NF-κB–regulated cyclooxygenase-2 and cyclin D1 protein expression in a dose-dependent manner. Furthermore, all compounds inhibited the proliferation of tumor cells, with IC_{50}s of 6.09 (aspirin), 1.12 (ibuprofen), 0.65 (sulindac), 0.49 (phenylbutazone), 1.01 (naproxen), 0.19 (indomethacin), 0.36 (diclofenac), 0.012 (resveratrol), 0.016 (curcumin), 0.047 (dexamethasone), 0.013 (celecoxib), and 0.008 (tamoxifen) mM. Overall, we found that aspirin and ibuprofen are the least potent and resveratrol, curcumin, celecoxib, and tamoxifen are the most potent anti-inflammatory and antiproliferative agents of those studied.

The inhibition of NF-κB with these drugs represents a possible approach to solving the more complicated issue of creating drug therapies that are effective in preventing or attenuating tumorigenesis. Indeed, aspirin has been shown to be an effective therapeutic agent against cylindromatosis, colorectal cancers, and human hepatoma. Our understanding of the precise mechanisms of action, specificity, and even toxicity of these drugs with respect to NF-κB is still incomplete. However, targeting NF-κB seems central to designing an effective therapy for cancer.

4.8. NF-κB can be Suppressed by Chemical Modifications

Various proteins that constitute the NF-κB machinery contain a highly reactive cysteine residue on their surfaces (Table **9**). Because modifications of this residue can also suppress NF-κB activation, various agents have been tested for their ability to modify these cysteine residues. Some found to be effective in this regard include N-tosyl-L-phenylalanine chloromethyl ketone, caffeic acid phenethyl ester (CAPE), herbimycin, arsenite, sesquiterpene lactone, ethyl pyruvate, ethacrynic acid, and kaurane diterpene (kamebakaurin) (Table **6**).

Table 9. Inhibition of NF-κB Through the Critical Cystein Residue

IKKα/β	Cys 179[242]
IKKβ	Cys 59[243]
IKKγ	Cys 417[244]
p65	Cys 38[245]
p50	Cys 62[246]

5. NF-κB ACTIVATION IS A DOUBLE-EDGED SWORD

Although NF-κB activation in most cases is harmful [200], there is evidence that NF-κB activation also has beneficial effects. As previously shown, NF-κB activation regulates the expression of both antiapoptotic and proapoptotic genes. However, the downstream target genes regulated by NF-κB in the initiation of proapoptotic signaling are unclear. Perfettini *et al*. [201] found that NF-κB and p53 are the dominant apoptosis-inducing transcription factors elicited by the HIV-1 envelope. Additionally, Ravi *et al*. [202] found that NF-κB induces expression of death receptor 4 (DR4) and DR5 and that, conversely, a transdominant mutant of the inhibitory protein IκBα or a transactivation-deficient mutant of c-Rel reduces expression of either DR. Whereas NF-κB promotes DR expression, cytokine-mediated activation of the RelA subunit of NF-κB also increases expression of the apoptosis inhibitor Bcl-x_L and protects cells from the TNF-related apoptosis-inducing ligand (TRAIL). Inhibition of NF-κB by blocking the activation of the IKK complex reduces Bcl-x_L expression and sensitizes tumor cells to TRAIL-induced apoptosis. The ability to induce DRs or Bcl-x_L may explain the dual role of NF-κB as a mediator and inhibitor of cell death during immune and stress responses. Fujioka *et al*. [203] showed that NF-κB mediates proapoptotic effects by activating the p53-signaling pathway in response to doxycycline-induced

superoxide. Additionally, Li *et al*. [204] demonstrated that the ability of doxycycline or superoxide to induce expression of polo-like kinase 3 (Plk3) depends on NF-κB activity. They identified a κB binding site in the promoter of Plk3 and showed that this site is directly involved in Plk3 induction by the RelA–NF-κB complex [204]. Specifically, Plk3 formed a complex with p53 and was involved in the phosphorylation of p53 on Ser-20 in response to superoxide. The inhibition of Plk3 expression by Plk3 siRNA suppressed the doxycycline- and superoxide-mediated apoptosis. In addition, the overexpression of wild-type Plk3 in HCT116 p53+/+ cells induced rapid apoptosis. However, the overexpression of wild-type Plk3 in HCT116 p53-/- cells and of the kinase-defective mutant Plk3(K91R) in p53+/+ cells induced a delayed onset of apoptosis. Furthermore, the mutagenesis of Plk3 showed that the N-terminal domain (amino acids 1–26) is essential for the induction of delayed onset of apoptosis. These findings show that Plk3 is a RelA-NF-κB–regulated gene that induces apoptosis in both p53-dependent and -independent signaling pathways, thus suggesting a possible mechanism for RelA-NF-κB–regulated proapoptotic responses.

DR5 (TRAIL-R2) is a proapoptotic protein considered a potential target for cancer therapy; its expression is mediated by NF-κB. Shetty *et al*. [205] found that NF-κB differentially regulates DR5 expression that involves HDAC1. They specifically showed that etoposide-induced DR5 expression requires the first intronic region of the DR5 gene [205]. The mutation of a putative NF-κB binding site in this intron eliminates the DR5 promoter activity, as do mutations in the p53 binding site in this region. The reduction in p53 expression also blocked p65 binding to the intronic region of the DR5 gene, indicating cooperation between p53 and p65 in DR5 expression. They further found that HDAC inhibitors activate NF-κB and p53 and upregulate DR5 expression. The blockage of DR5 activation decreased HDAC inhibitor-induced apoptosis, and a combination of HDAC inhibitors and TRAIL increased apoptosis [205]. This provides a mechanism for regulating NF-κB–mediated DR5 expression and could explain the differential roles NF-κB plays in regulating apoptosis.

It is known that inactivation of apoptotic pathways is a common event in cancer and that p53 and NF-κB are transcription factors that regulate apoptosis during tumorigenesis. Although NF-κB is generally considered a suppressor of cell death, Ryan *et al*. [206] showed that NF-κB can contribute to p53-induced death. Specifically, they found that whereas the loss of NF-κB is tumor promoting, it does not substitute for the loss of p53. They also showed that the loss of p65, a critical subunit of NF-κB, can cause resistance to different agents that signal death through p53. The loss of p65 also enhances the tumorigenesis induced by E1a and Ras. Unlike the loss of p53, however, the loss of p65 does not cause anchorage-independent growth or enable tumor development after the expression of a single oncogene. These findings reaffirm the role of NF-κB in p53-induced death and also show that its loss does not substitute for the loss of p53 in tumor development. These findings further indicate that loss of the ability to induce programmed cell death, even if this ability is central to p53's function, does not completely inactivate p53's tumor-suppressive effects. Interestingly, Bohuslav *et al*. [207] found that p53 induces NF-κB activation by an IκB kinase-independent mechanism involving the phosphorylation of p65 by ribosomal S6 kinase 1.

The role of the NF-κB family of transcription factors as tumor promoters is firmly established. However, some data suggest that NF-κB can also inhibit tumor growth. Moreover, NF-κB activity is modulated by tumor suppressors such as p53 and ARF whereby NF-κB subunits repress, rather than activate, the expression of tumor-promoting genes. This suggests a dual function of NF-κB during tumor progression: in the early stages, NF-κB inhibits tumor growth, but as further mutations lead to a loss of tumor-suppressor expression, the oncogenic functions of NF-κB become unleashed, allowing it to actively contribute to tumorigenesis. Perkins reviewed the implications of NF-κB function and how it might influence the use of NF-κB–based anticancer therapies [208].

CONCLUSIONS

Overall, the cited studies have clearly shown that NF-κB plays a critical role in tumorigenesis. Similarly, the suppression of NF-κB can inhibit various steps in the tumorigenic process. Thus, the design of NF-κB inhibitors that are pharmacologically safe will be critical for the treatment of cancer. Because of the multiple agents that activate NF-κB through diverse pathways, it is unlikely that any single inhibitor of NF-κB could be effective against all tumors. Our efforts, therefore, should be directed toward developing NF-κB inhibitors that work best against individual cancers. Bortezomib (Velcade), also called PS341, is one such inhibitor that has already been approved by the US Food and Drug Administration for treatment of multiple myeloma.

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ABBREVIATIONS

NF-κB	=	Nuclear factor-κB
IKK	=	IκB kianses
TNF	=	Tumor necrosis factor
IL-1	=	Interleukin-1
LPS	=	Lipopolysaccharide
TNFR	=	TNF receptor
TRAF	=	TNF receptor-associated factor
NIK	=	NF-κB inducing kinase
NSCLC	=	Non-small cell lung carcinoma

MEF = Mouse embryo fibroblats

PGE_2 = Prostaglandin E_2

EGF = Epithelial growth factor

MALT = Mucosa-associated lymphoid tissue

MMP = Matrix metalloproteinase

uPA = Urokinase-type plasminogen activator

ECM = Extracellular matrix

PI3k = Phosphatidylinositol 3'-kinase

IκBαM = Mutant IκBα

HDAC = Histone deacetylase

PlK3 = Polo-like kinase 3

siRNA = Small interfering RNA

DR = Death receptor

PTD = Protein transduction domain

REFERENCES

[1] Sen R, Baltimore D. Inducibility of kappa immunoglobulin enhancer-binding protein Nf-kappa B by a posttranslational mechanism. Cell 1986; 47: 921-8.

[2] Gilmore TD. The Rel/NF-kappaB signal transduction pathway: introduction. Oncogene 1999; 18: 6842-4.

[3] Ghosh S, May MJ, Kopp EB. NF-kappa B and Rel proteins: evolutionarily conserved mediators of immune responses. Annu Rev Immunol 1998; 16: 225-60.

[4] Karin M. How NF-kappaB is activated: the role of the IkappaB kinase (IKK) complex. Oncogene 1999; 18: 6867-74.

[5] Li N, Karin M. Ionizing radiation and short wavelength UV activate NF-kappaB through two distinct mechanisms. Proc Natl Acad Sci U S A 1998; 95: 13012-7.

[6] Singh S, Darnay BG, Aggarwal BB. Site-specific tyrosine phosphorylation of IkappaBalpha negatively regulates its inducible phosphorylation and degradation. J Biol Chem 1996; 271: 31049-54.

[7] Mayo MW, Baldwin AS. The transcription factor NF-kappaB: control of oncogenesis and cancer therapy resistance. Biochim Biophys Acta 2000; 1470: M55-62.

[8] Pahl HL. Activators and target genes of Rel/NF-kappaB transcription factors. Oncogene 1999; 18: 6853-66.

[9] Wang CY, Mayo MW, Baldwin AS Jr. TNF- and cancer therapy-induced apoptosis: potentiation by inhibition of NF-kappaB. Science 1996; 274: 784-7.

[10] Dejardin E, Droin NM, Delhase M, *et al*. The lymphotoxin-beta receptor induces different patterns of gene expression *via* two NF-kappaB pathways. Immunity 2002; 17: 525-35.

[11] Cao Y, Bonizzi G, Seagroves TN, *et al*. IKKalpha provides an essential link between RANK signaling and cyclin D1 expression during mammary gland development. Cell 2001; 107: 763-75.

[12] Digicaylioglu M, Lipton SA. Erythropoietin-mediated neuroprotection involves cross-talk between Jak2 and NF-kappaB signalling cascades. Nature 2001; 412: 641-7.

[13] Li J, Peet GW, Balzarano D, *et al*. Novel NEMO/IkappaB kinase and NF-kappa B target genes at the pre-B to immature B cell transition. J Biol Chem 2001; 276: 18579-90.

[14] Rothwarf DM, Zandi E, Natoli G, *et al*. IKK-gamma is an essential regulatory subunit of the IkappaB kinase complex. Nature 1998; 395: 297-300.

[15] Shimada T, Kawai T, Takeda K, *et al*. IKK-i, a novel lipopolysaccharide-inducible kinase that is related to IkappaB kinases. Int Immunol 1999; 11: 1357-62.

[16] Peters RT, Liao SM, Maniatis T. IKKepsilon is part of a novel PMA-inducible IkappaB kinase complex. Mol Cell 2000; 5: 513-22.

[17] Nomura F, Kawai T, Nakanishi K, *et al*. NF-kappaB activation through IKK-i-dependent I-TRAF/TANK phosphorylation. Genes Cells 2000; 5: 191-202.

[18] Malinin NL, Boldin MP, Kovalenko AV, *et al*. MAP3K-related kinase involved in NF-kappaB induction by TNF, CD95 and IL-1. Nature 1997; 385: 540-4.

[19] DiDonato JA, Hayakawa M, Rothwarf DM, *et al*. A cytokine-responsive IkappaB kinase that activates the transcription factor NF-kappaB. Nature 1997; 388: 548-54.

[20] Mercurio F, Zhu H, Murray BW, *et al*. IKK-1 and IKK-2: cytokine-activated IkappaB kinases essential for NF-kappaB activation. Science 1997; 278: 860-6.

[21] Zandi E, Rothwarf DM, Delhase M, *et al*. The IkappaB kinase complex (IKK) contains two kinase subunits, IKKalpha and IKKbeta, necessary for IkappaB phosphorylation and NF-kappaB activation. Cell 1997; 91: 243-52.

[22] Zandi E, Chen Y, Karin M. Direct phosphorylation of IkappaB by IKKalpha and IKKbeta: discrimination between free and NF-kappaB-bound substrate. Science 1998; 281: 1360-3.

[23] Sanz L, Sanchez P, Lallena MJ, *et al*. The interaction of p62 with RIP links the atypical PKCs to NF-kappaB activation. EMBO J 1999; 18: 3044-53.

[24] Diaz-Meco MT, Berra E, Municio MM, *et al*. A dominant negative protein kinase C zeta subspecies blocks NF-kappa B activation. Mol Cell Biol 1993; 13: 4770-5.

[25] Chen CC, Sun YT, Chen JJ, *et al*. TNF-alpha-induced cyclooxygenase-2 expression in human lung epithelial cells: involvement of the phospholipase C-gamma 2, protein kinase C-alpha, tyrosine kinase, NF-kappa B-inducing kinase, and I-kappa B kinase 1/2 pathway. J Immunol 2000; 165: 2719-28.

[26] Vertegaal AC, Kuiperij HB, Yamaoka S, *et al*. Protein kinase C-alpha is an upstream activator of the IkappaB kinase complex in the TPA signal transduction pathway to NF-kappaB in U2OS cells. Cell Signal 2000; 12: 759-68.

[27] McAllister-Lucas LM, Inohara N, Lucas PC, *et al*. Bimp1, a MAGUK family member linking protein kinase C activation to Bcl10-mediated NF-kappaB induction. J Biol Chem 2001; 276: 30589-97.

[28] Leitges M, Schmedt C, Guinamard R, *et al*. Immunodeficiency in protein kinase cbeta-deficient mice. Science 1996; 273: 788-91.

[29] Lin X, O'Mahony A, Mu Y, *et al*. Protein kinase C-theta participates in NF-kappaB activation induced by CD3-CD28 costimulation through selective activation of IkappaB kinase beta. Mol Cell Biol 2000; 20: 2933-40.

[30] Bauer B, Krumbock N, Fresser F, *et al*. Complex formation and cooperation of protein kinase C theta and Akt1/protein kinase B alpha in the NF-kappa B transactivation cascade in Jurkat T cells. J Biol Chem 2001; 276: 31627-34.

[31] Vancurova I, Miskolci V, Davidson D. NF-kappa B activation in tumor necrosis factor alpha-stimulated neutrophils is mediated by protein kinase Cdelta. Correlation to nuclear Ikappa Balpha. J Biol Chem 2001; 276: 19746-52.

[32] Diaz-Guerra MJ, Bodelon OG, Velasco M, *et al*. Up-regulation of protein kinase C-epsilon promotes the expression of cytokine-inducible nitric oxide synthase in RAW 264.7 cells. J Biol Chem 1996; 271: 32028-33.

[33] Kumar A, Haque J, Lacoste J, *et al*. Double-stranded RNA-dependent protein kinase activates transcription factor NF-kappa B by phosphorylating I kappa B. Proc Natl Acad Sci USA 1994; 91: 6288-92.

[34] Belich MP, Salmeron A, Johnston LH, *et al*. TPL-2 kinase regulates the proteolysis of the NF-kappaB-inhibitory protein NF-kappaB1 p105. Nature 1999; 397: 363-8.

[35] Zhao Q, Lee FS. Mitogen-activated protein kinase/ERK kinase kinases 2 and 3 activate nuclear factor-kappaB through IkappaB kinase-alpha and IkappaB kinase-beta. J Biol Chem 1999; 274: 8355-8.

[36] Yang J, Lin Y, Guo Z, *et al*. The essential role of MEKK3 in TNF-induced NF-kappaB activation. Nat Immunol 2001; 2: 620-4.

[37] Tojima Y, Fujimoto A, Delhase M, *et al*. NAK is an IkappaB kinase-activating kinase. Nature 2000; 404: 778-82.

[38] Pomerantz JL, Baltimore D. NF-kappaB activation by a signaling complex containing TRAF2, TANK and TBK1, a novel IKK-related kinase. EMBO J 1999; 18: 6694-704.

[39] Bonnard M, Mirtsos C, Suzuki S, *et al*. Deficiency of T2K leads to apoptotic liver degeneration and impaired NF-kappaB-dependent gene transcription. EMBO J 2000; 19: 4976-85.

[40] Reddy SA, Huang JH, Liao WS. Phosphatidylinositol 3-kinase in interleukin 1 signaling. Physical interaction with the interleukin 1 receptor and requirement in NFkappaB and AP-1 activation. J Biol Chem 1997; 272: 29167-73.

[41] Ozes ON, Mayo LD, Gustin JA, *et al*. NF-kappaB activation by tumour necrosis factor requires the Akt serine-threonine kinase. Nature 1999; 401: 82-5.

[42] Hehner SP, Hofmann TG, Ushmorov A, *et al*. Mixed-lineage kinase 3 delivers CD3/CD28-derived signals into the IkappaB kinase complex. Mol Cell Biol 2000; 20: 2556-68.

[43] Hu MC, Wang Y, Qiu WR, *et al*. Hematopoietic progenitor kinase-1 (HPK1) stress response signaling pathway activates IkappaB kinases (IKK-alpha/beta) and IKK-beta is a developmentally regulated protein kinase. Oncogene 1999; 18: 5514-24.

[44] Wang W, Zhou G, Hu MC, *et al*. Activation of the hematopoietic progenitor kinase-1 (HPK1)-dependent, stress-activated c-Jun N-terminal kinase (JNK) pathway by transforming growth factor beta (TGF-beta)-activated kinase (TAK1), a kinase mediator of TGF beta signal transduction. J Biol Chem 1997; 272: 22771-5.

[45] Troppmair J, Hartkamp J, Rapp UR. Activation of NF-kappa B by oncogenic Raf in HEK 293 cells occurs through autocrine recruitment of the stress kinase cascade. Oncogene 1998; 17: 685-90.

[46] Frost JA, Swantek JL, Stippec S, *et al*. Stimulation of NFkappa B activity by multiple signaling pathways requires PAK1. J Biol Chem 2000; 275: 19693-9.

[47] Petro JB, Rahman SM, Ballard DW, *et al*. Bruton's tyrosine kinase is required for activation of IkappaB kinase and nuclear factor kappaB in response to B cell receptor engagement. J Exp Med 2000; 191: 1745-54.

[48] Zhong H, Voll RE, Ghosh S. Phosphorylation of NF-kappa B p65 by PKA stimulates transcriptional activity by promoting a novel bivalent interaction with the coactivator CBP/p300. Mol Cell 1998; 1: 661-71.

[49] Zhong H, SuYang H, Erdjument-Bromage H, *et al*. The transcriptional activity of NF-kappaB is regulated by the IkappaB-associated PKAc subunit through a cyclic AMP-independent mechanism. Cell 1997; 89: 413-24.

[50] Li YL, Guo FK, Wu SG. Effects of antisense IRAK-2 oligonucleotides on PGI2 release induced by IL-1 and TNF. Acta Pharmacol Sin 2000; 21: 646-8.

[51] Jefferies C, Bowie A, Brady G, *et al*. Transactivation by the p65 subunit of NF-kappaB in response to interleukin-1 (IL-1) involves MyD88, IL-1 receptor-associated kinase 1, TRAF-6, and Rac1. Mol Cell Biol 2001; 21: 4544-52.

[52] Wesche H, Gao X, Li X, *et al*. IRAK-M is a novel member of the Pelle/interleukin-1 receptor-associated kinase (IRAK) family. J Biol Chem 1999; 274: 19403-10.

[53] Manna SK, Sah NK, Aggarwal BB. Protein tyrosine kinase p56lck is required for ceramide-induced but not tumor necrosis factor-induced activation of NF-kappa B, AP-1, JNK, and apoptosis. J Biol Chem 2000; 275: 13297-306.

[54] Fan C, Li Q, Ross D, *et al*. Tyrosine phosphorylation of I kappa B alpha activates NF kappa B through a redox-regulated and c-Src-dependent mechanism following hypoxia/reoxygenation. J Biol Chem 2003; 278: 2072-80.

[55] Palombella VJ, Rando OJ, Goldberg AL, *et al*. The ubiquitin-proteasome pathway is required for processing the NF-kappa B1 precursor protein and the activation of NF-kappa B. Cell 1994; 78: 773-85.

[56] Parcellier A, Schmitt E, Gurbuxani S, *et al*. HSP27 is a ubiquitin-binding protein involved in I-kappaBalpha proteasomal degradation. Mol Cell Biol 2003; 23: 5790-802.

[57] Garg A, Aggarwal BB. Nuclear transcription factor-kappaB as a target for cancer drug development. Leukemia 2002; 16: 1053-68.

[58] Rosenfeld ME, Prichard L, Shiojiri N, *et al*. Prevention of hepatic apoptosis and embryonic lethality in RelA/TNFR-1 double knockout mice. Am J Pathol 2000; 156: 997-1007.

[59] Alcamo E, Hacohen N, Schulte LC, *et al*. Requirement for the NF-kappaB family member RelA in the development of secondary lymphoid organs. J Exp Med 2002; 195: 233-44.

[60] Beg AA, Sha WC, Bronson RT, *et al*. Embryonic lethality and liver degeneration in mice lacking the RelA component of NF-kappa B. Nature 1995; 376: 167-70.

[61] MacMicking JD, Nathan C, Hom G, *et al*. Altered responses to bacterial infection and endotoxic shock in mice lacking inducible nitric oxide synthase. Cell 1995; 81: 641-50.

[62] Behl PN. Vitiligo: treatment by dermabrasion and epithelial sheet grafting. J Am Acad Dermatol 1994; 30: 1044-5.

[63] Pahl HL, Baeuerle PA. A novel signal transduction pathway from the endoplasmic reticulum to the nucleus is mediated by transcription factor NF-kappa B. EMBO J 1995; 14: 2580-8.

[64] Berliner JA, Navab M, Fogelman AM, *et al*. Atherosclerosis: basic mechanisms. Oxidation, inflammation, and genetics. Circulation 1995; 91: 2488-96.

[65] Liao F, Andalibi A, Qiao JH, *et al*. Genetic evidence for a common pathway mediating oxidative stress, inflammatory gene induction, and aortic fatty streak formation in mice. J Clin Invest 1994; 94: 877-84.

[66] Nakajima T, Kitajima I, Shin H, *et al*. Involvement of NF-kappa B activation in thrombin-induced human vascular smooth muscle cell proliferation. Biochem Biophys Res Commun 1994; 204: 950-8.

[67] Jung M, Zhang Y, Lee S, *et al*. Correction of radiation sensitivity in ataxia telangiectasia cells by a truncated I kappa B-alpha. Science 1995; 268: 1619-21.

[68] Luque I, Gelinas C. Rel/NF-kappa B and I kappa B factors in oncogenesis. Semin Cancer Biol 1997; 8: 103-11.

[69] Houldsworth J, Mathew S, Rao PH, *et al*. REL proto-oncogene is frequently amplified in extranodal diffuse large cell lymphoma. Blood 1996; 87: 25-9.

[70] Lu D, Thompson JD, Gorski GK, *et al*. Alterations at the rel locus in human lymphoma. Oncogene 1991; 6: 1235-41.

[71] Mukhopadhyay T, Roth JA, Maxwell SA. Altered expression of the p50 subunit of the NF-kappa B transcription factor complex in non-small cell lung carcinoma. Oncogene 1995; 11: 999-1003.

[72] Mathew S, Murty VV, Dalla-Favera R, *et al*. Chromosomal localization of genes encoding the transcription factors, c-rel, NF-kappa Bp50, NF-kappa Bp65, and lyt-10 by fluorescence *in situ* hybridization. Oncogene 1993; 8: 191-3.

[73] Wang CY, Guttridge DC, Mayo MW, *et al*. NF-kappaB induces expression of the Bcl-2 homologue A1/Bfl-1 to preferentially suppress chemotherapy-induced apoptosis. Mol Cell Biol 1999; 19: 5923-9.

[74] Visconti R, Cerutti J, Battista S, *et al*. Expression of the neoplastic phenotype by human thyroid carcinoma cell lines requires NFkappaB p65 protein expression. Oncogene 1997; 15: 1987-94.

[75] Trecca D, Guerrini L, Facchiolla NS, *et al*. Identification of a tumor-associated mutant form of the NF-kappaB RelA gene with reduced DNA-binding and transactivating activities. Oncogene 1997; 14: 791-9.

[76] Baldwin AS Jr. The NF-kappa B and I kappa B proteins: new discoveries and insights. Annu Rev Immunol 1996; 14: 649-83.

[77] Cabannes E, Khan G, Aillet F, *et al*. Mutations in the IkBa gene in Hodgkin's disease suggest a tumour suppressor role for IkappaBalpha. Oncogene 1999; 18: 3063-70.

[78] Beauparlant P, Kwan I, Bitar R, *et al*. Disruption of I kappa B alpha regulation by antisense RNA expression leads to malignant transformation. Oncogene 1994; 9: 3189-97.

[79] Kitajima I, Shinohara T, Bilakovics J, *et al*. Ablation of transplanted HTLV-I Tax-transformed tumors in mice by antisense inhibition of NF-kappa B. Science 1992; 258: 1792-5.

[80] Ferrier R, Nougarede R, Doucet S, *et al*. Physical interaction of the bHLH LYL1 protein and NF-kappaB1 p105. Oncogene 1999; 18: 995-1005.

[81] Motokura T, Arnold A. PRAD1/cyclin D1 proto-oncogene: genomic organization, 5' DNA sequence, and sequence of a tumor-specific rearrangement breakpoint. Genes Chromosomes Cancer 1993; 7: 89-95.

[82] Nakshatri H, Bhat-Nakshatri P, Martin DA, *et al*. Constitutive activation of NF-kappaB during progression of breast cancer to hormone-independent growth. Mol Cell Biol 1997; 17: 3629-39.

[83] Rayet B, Gelinas C. Aberrant rel/nfkb genes and activity in human cancer. Oncogene 1999; 18: 6938-47.

[84] Nair A, Venkatraman M, Maliekal TT, *et al*. NF-kappaB is constitutively activated in high-grade squamous intraepithelial

lesions and squamous cell carcinomas of the human uterine cervix. Oncogene 2003; 22: 50-8.

[85] Oya M, Takayanagi A, Horiguchi A, *et al*. Increased nuclear factor-kappa B activation is related to the tumor development of renal cell carcinoma. Carcinogenesis 2003; 24: 377-84.

[86] Banerjee S, Bueso-Ramos C, Aggarwal BB. Suppression of 7,12-dimethylbenz(a)anthracene-induced mammary carcinogenesis in rats by resveratrol: role of nuclear factor-kappaB, cyclooxygenase 2, and matrix metalloprotease 9. Cancer Res 2002; 62: 4945-54.

[87] Shishodia S, Potdar P, Gairola CG, *et al*. Curcumin (diferuloylmethane) down-regulates cigarette smoke-induced NF-kappaB activation through inhibition of IkappaBalpha kinase in human lung epithelial cells: correlation with suppression of COX-2, MMP-9 and cyclin D1. Carcinogenesis 2003; 24: 1269-79.

[88] Anto RJ, Mukhopadhyay A, Shishodia S, *et al*. Cigarette smoke condensate activates nuclear transcription factor-kappaB through phosphorylation and degradation of IkappaB(alpha): correlation with induction of cyclooxygenase-2. Carcinogenesis 2002; 23: 1511-8.

[89] Shishodia S, Majumdar S, Banerjee S, *et al*. Ursolic acid inhibits nuclear factor-kappaB activation induced by carcinogenic agents through suppression of IkappaBalpha kinase and p65 phosphorylation: correlation with down-regulation of cyclooxygenase 2, matrix metalloproteinase 9, and cyclin D1. Cancer Res 2003; 63: 4375-83.

[90] Koong AC, Chen EY, Giaccia AJ. Hypoxia causes the activation of nuclear factor kappa B through the phosphorylation of I kappa B alpha on tyrosine residues. Cancer Res 1994; 54: 1425-30.

[91] Schutze S, Potthoff K, Machleidt T, *et al*. TNF activates NF-kappa B by phosphatidylcholine-specific phospholipase C-induced "acidic" sphingomyelin breakdown. Cell 1992; 71: 765-76.

[92] Aggarwal BB. Tumour necrosis factors receptor associated signalling molecules and their role in activation of apoptosis, JNK and NF-kappaB. Ann Rheum Dis 2000; 59 (Suppl. 1): i6-16.

[93] Komori A, Yatsunami J, Suganuma M, *et al*. Tumor necrosis factor acts as a tumor promoter in BALB/3T3 cell transformation. Cancer Res 1993; 53: 1982-5.

[94] Suganuma M, Okabe S, Marino MW, *et al*. Essential role of tumor necrosis factor alpha (TNF-alpha) in tumor promotion as revealed by TNF-alpha-deficient mice. Cancer Res 1999; 59: 4516-8.

[95] Hafner M, Orosz P, Kruger A, *et al*. TNF promotes metastasis by impairing natural killer cell activity. Int J Cancer 1996; 66: 388-92.

[96] Moore RJ, Owens DM, Stamp G, *et al*. Mice deficient in tumor necrosis factor-alpha are resistant to skin carcinogenesis. Nat Med 1999; 5: 828-31.

[97] Giri DK, Aggarwal BB. Constitutive activation of NF-kappaB causes resistance to apoptosis in human cutaneous T cell lymphoma HuT-78 cells. Autocrine role of tumor necrosis factor and reactive oxygen intermediates. J Biol Chem 1998; 273: 14008-14.

[98] Arnott CH, Scott KA, Moore RJ, *et al*. Tumour necrosis factor-alpha mediates tumour promotion *via* a PKC alpha- and AP-1-dependent pathway. Oncogene 2002; 21: 4728-38.

[99] Hehlgans T, Stoelcker B, Stopfer P, *et al*. Lymphotoxin-beta receptor immune interaction promotes tumor growth by inducing angiogenesis. Cancer Res 2002; 62: 4034-40.

[100] Ashikawa K, Shishodia S, Fokt I, *et al*. Evidence that activation of nuclear factor-kappaB is essential for the cytotoxic effects of doxorubicin and its analogues. Biochem Pharmacol 2004; 67: 353-64.

[101] Singh S, Raju U, Mendoza J, *et al*. Acquisition of cellular resistance to 9-nitro-camptothecin correlates with suppression of transcription factor NF-kappa B activation and potentiation of cytotoxicity by tumor necrosis factor in human histiocytic lymphoma U-937 cells. Anticancer Drugs 1998; 9: 703-14.

[102] Arlt A, Gehrz A, Muerkoster S, *et al*. Role of NF-kappaB and Akt/PI3K in the resistance of pancreatic carcinoma cell lines against gemcitabine-induced cell death. Oncogene 2003; 22: 3243-51.

[103] Hwang S, Ding A. Activation of NF-kappa B in murine macrophages by taxol. Cancer Biochem Biophys 1995; 14: 265-72.

[104] Donepudi M, Raychaudhuri P, Bluestone JA, *et al*. Mechanism of melphalan-induced B7-1 gene expression in P815 tumor cells. J Immunol 2001; 166: 6491-9.

[105] Galter D, Mihm S, Droge W. Distinct effects of glutathione disulphide on the nuclear transcription factor kappa B and the activator protein-1. Eur J Biochem 1994; 221: 639-48.

[106] Brach MA, Hass R, Sherman ML, *et al*. Ionizing radiation induces expression and binding activity of the nuclear factor kappa B. J Clin Invest 1991; 88: 691-5.

[107] Egan LJ, Eckmann L, Greten FR, *et al*. IkappaB-kinasebeta-dependent NF-kappaB activation provides radioprotection to the intestinal epithelium. Proc Natl Acad Sci USA 2004; 101: 2452-7.

[108] Panta GR, Kaur S, Cavin LG, *et al*. ATM and the catalytic subunit of DNA-dependent protein kinase activate NF-kappaB through a common MEK/extracellular signal-regulated kinase/p90(rsk) signaling pathway in response to distinct forms of DNA damage. Mol Cell Biol 2004; 24: 1823-35.

[109] Wang CY, Cusack JC, Jr., Liu R, *et al*. Control of inducible chemoresistance: enhanced anti-tumor therapy through increased apoptosis by inhibition of NF-kappaB. Nat Med 1999; 5: 412-7.

[110] Chen X, Shen B, Xia L, *et al*. Activation of nuclear factor kappaB in radioresistance of TP53-inactive human keratinocytes. Cancer Res 2002; 62: 1213-21.

[111] Zhang Q, Siebert R, Yan M, *et al*. Inactivating mutations and overexpression of BCL10, a caspase recruitment domain-containing gene, in MALT lymphoma with t(1;14)(p22;q32). Nat Genet 1999; 22: 63-8.

[112] Tergaonkar V, Bottero V, Ikawa M, *et al*. IkappaB kinase-independent IkappaBalpha degradation pathway: functional NF-kappaB activity and implications for cancer therapy. Mol Cell Biol 2003; 23: 8070-83.

[113] Camarda G, Binaschi M, Maggi CA, *et al*. Nuclear factor-kappaB, induced in human carcinoma cell line A2780 by the new anthracycline men 10755, is devoid of transcriptional activity. Int J Cancer 2002; 102: 476-82.

[114] Tergaonkar V, Pando M, Vafa O, *et al*. p53 stabilization is decreased upon NFkappaB activation: a role for NFkappaB in acquisition of resistance to chemotherapy. Cancer Cell 2002; 1: 493-503.

[115] Finco TS, Baldwin AS Jr. Kappa B site-dependent induction of gene expression by diverse inducers of nuclear factor kappa B requires Raf-1. J Biol Chem 1993; 268: 17676-9.

[116] Hamdane M, David-Cordonnier MH, D'Halluin JC. Activation of p65 NF-kappaB protein by p210BCR-ABL in a myeloid cell line (P210BCR-ABL activates p65 NF-kappaB). Oncogene 1997; 15: 2267-75.

[117] Duyao MP, Kessler DJ, Spicer DB, *et al*. Transactivation of the c-myc promoter by human T cell leukemia virus type 1 tax is mediated by NF kappa B. J Biol Chem 1992; 267: 16288-91.

[118] Finco TS, Westwick JK, Norris JL, *et al*. Oncogenic Ha-Ras-induced signaling activates NF-kappaB transcriptional activity, which is required for cellular transformation. J Biol Chem 1997; 272: 24113-6.

[119] Norris JL, Baldwin AS Jr. Oncogenic Ras enhances NF-kappaB transcriptional activity through Raf-dependent and Raf-independent mitogen-activated protein kinase signaling pathways. J Biol Chem 1999; 274: 13841-6.

[120] Mayo MW, Norris JL, Baldwin AS. Ras regulation of NF-kappa B and apoptosis. Methods Enzymol 2001; 333: 73-87.

[121] Balmain A, Pragnell IB. Mouse skin carcinomas induced *in vivo* by chemical carcinogens have a transforming Harvey-ras oncogene. Nature 1983; 303: 72-4.

[122] Lu Y, Jamieson L, Brasier AR, *et al*. NF-kappaB/RelA transactivation is required for atypical protein kinase C iota-mediated cell survival. Oncogene 2001; 20: 4777-92.

[123] Libermann TA, Baltimore D. Activation of interleukin-6 gene expression through the NF-kappa B transcription factor. Mol Cell Biol 1990; 10: 2327-34.

[124] Aggarwal BB, Schwarz L, Hogan ME, *et al*. Triple helix-forming oligodeoxyribonucleotides targeted to the human tumor necrosis factor (TNF) gene inhibit TNF production and block the TNF-dependent growth of human glioblastoma tumor cells. Cancer Res 1996; 56: 5156-64.

[125] Estrov Z, Thall PF, Talpaz M, *et al*. Caspase 2 and caspase 3 protein levels as predictors of survival in acute myelogenous leukemia. Blood 1998; 92: 3090-7.

[126] Bharti AC, Donato N, Singh S, *et al*. Curcumin (diferuloylmethane) down-regulates the constitutive activation of nuclear factor-kappa B and IkappaBalpha kinase in human multiple myeloma cells, leading to suppression of proliferation and induction of apoptosis. Blood 2003; 101: 1053-62.

[127] Kato T, Duffey DC, Ondrey FG, *et al*. Cisplatin and radiation sensitivity in human head and neck squamous carcinomas are independently modulated by glutathione and transcription factor NF-kappaB. Head Neck 2000; 22: 748-59.

[128] Mukhopadhyay A, Banerjee S, Stafford LJ, *et al*. Curcumin-induced suppression of cell proliferation correlates with down-regulation of cyclin D1 expression and CDK4-mediated retino-blastoma protein phosphorylation. Oncogene 2002; 21: 8852-61.

[129] Yamamoto K, Arakawa T, Ueda N, *et al*. Transcriptional roles of nuclear factor kappa B and nuclear factor-interleukin-6 in the tumor necrosis factor alpha-dependent induction of cyclooxygenase-2 in MC3T3-E1 cells. J Biol Chem 1995; 270: 31315-20.

[130] Bargou RC, Emmerich F, Krappmann D, *et al*. Constitutive nuclear factor-kappaB-RelA activation is required for proliferation and survival of Hodgkin's disease tumor cells. J Clin Invest 1997; 100: 2961-9.

[131] Romashkova JA, Makarov SS. NF-kappaB is a target of AKT in anti-apoptotic PDGF signalling. Nature 1999; 401: 86-90.

[132] Habib AA, Chatterjee S, Park SK, *et al*. The epidermal growth factor receptor engages receptor interacting protein and nuclear factor-kappa B (NF-kappa B)-inducing kinase to activate NF-kappa B. Identification of a novel receptor-tyrosine kinase signalosome. J Biol Chem 2001; 276: 8865-74.

[133] Foehr ED, Lin X, O'Mahony A, *et al*. NF-kappa B signaling promotes both cell survival and neurite process formation in nerve growth factor-stimulated PC12 cells. J Neurosci 2000; 20: 7556-63.

[134] Shishodia SA, ggarwal BB. Nuclear factor-kappa B activation: A question of life and death. J Biochem Mol Biol 2002; 35: 28-40.

[135] Kawamura K, Sato N, Fukuda J, *et al*. Survivin acts as an antiapoptotic factor during the development of mouse preimplantation embryos. Dev Biol 2003; 256: 331-41.

[136] Matta H, Eby MT, Gazdar AF, *et al*. Role of MRIT/cFLIP in protection against chemotherapy-induced apoptosis. Cancer Biol Ther 2002; 1: 652-60.

[137] Shinohara T, Miki T, Nishimura N, *et al*. Nuclear factor-kappaB-dependent expression of metastasis suppressor KAI1/CD82 gene in lung cancer cell lines expressing mutant p53. Cancer Res 2001; 61: 673-8.

[138] Stoffel A, Levine AJ. Actvation of NF-kappaB by the API2/ MALT1 fusions inhibits p53 dependant but not FAS induced apoptosis: a directional link between NF-kappaB and p53. Cell Cycle 2004; 3: 1017-20.

[139] Farina AR, Tacconelli A, Vacca A, *et al*. Transcriptional up-regulation of matrix metalloproteinase-9 expression during spontaneous epithelial to neuroblast phenotype conversion by SK-N-SH neuroblastoma cells, involved in enhanced invasivity, depends upon GT-box and nuclear factor kappaB elements. Cell Growth Differ 1999; 10: 353-67.

[140] Novak U, Cocks BG, Hamilton JA. A labile repressor acts through the NFkB-like binding sites of the human urokinase gene. Nucleic Acids Res 1991; 19: 3389-93.

[141] Bond M, Fabunmi RP, Baker AH, *et al*. Synergistic upregulation of metalloproteinase-9 by growth factors and inflammatory cytokines: an absolute requirement for transcription factor NF-kappa B. FEBS Lett 1998; 435: 29-34.

[142] Wang W, Abbruzzese JL, Evans DB, *et al*. Overexpression of urokinase-type plasminogen activator in pancreatic adenocarcinoma is regulated by constitutively activated RelA. Oncogene 1999; 18: 4554-63.

[143] Sliva D, English D, Lyons D, *et al*. Protein kinase C induces motility of breast cancers by upregulating secretion of urokinase-type plasminogen activator through activation of AP-1 and NF-kappaB. Biochem Biophys Res Commun 2002; 290: 552-7.

[144] Loch T, Michalski B, Mazurek U, *et al*. [Vascular endothelial growth factor (VEGF) and its role in neoplastic processes]. Postepy Hig Med Dosw 2001; 55: 257-74.

[145] Chilov D, Kukk E, Taira S, *et al*. Genomic organization of human and mouse genes for vascular endothelial growth factor C. J Biol Chem 1997; 272: 25176-83.

[146] Levine L, Lucci JA, 3rd, Pazdrak B, *et al*. Bombesin stimulates nuclear factor kappa B activation and expression of proangiogenic factors in prostate cancer cells. Cancer Res 2003; 63: 3495-502.

[147] Yu HG, Yu LL, Yang Y, *et al*. Increased expression of RelA/nuclear factor-kappa B protein correlates with colorectal tumorigenesis. Oncology 2003; 65: 37-45.

[148] Huang S, DeGuzman A, Bucana CD, *et al*. Nuclear factor-kappaB activity correlates with growth, angiogenesis, and metastasis of human melanoma cells in nude mice. Clin Cancer Res 2000; 6: 2573-81.

[149] Pollet I, Opina CJ, Zimmerman C, *et al*. Bacterial lipopolysaccharide directly induces angiogenesis through TRAF6-mediated activation of NF-kappaB and c-Jun N-terminal kinase. Blood 2003; 102: 1740-2.

[150] van de Stolpe A, Caldenhoven E, Stade BG, *et al*. 12-O-tetradecanoylphorbol-13-acetate- and tumor necrosis factor alpha-mediated induction of intercellular adhesion molecule-1 is inhibited by dexamethasone. Functional analysis of the human intercellular adhesion molecular-1 promoter. J Biol Chem 1994; 269: 6185-92.

[151] Helbig G, Christopherson KW, 2nd, Bhat-Nakshatri P, *et al*. NF-kappaB promotes breast cancer cell migration and metastasis by inducing the expression of the chemokine receptor CXCR4. J Biol Chem 2003; 278: 21631-8.

[152] Fujioka S, Sclabas GM, Schmidt C, *et al*. Function of nuclear factor kappaB in pancreatic cancer metastasis. Clin Cancer Res 2003; 9: 346-54.

[153] Loercher A, Lee TL, Ricker JL, *et al*. Nuclear factor-kappaB is an important modulator of the altered gene expression profile and malignant phenotype in squamous cell carcinoma. Cancer Res 2004; 64: 6511-23.

[154] Darnay BG, Haridas V, Ni J, *et al*. Characterization of the intracellular domain of receptor activator of NF-kappaB (RANK). Interaction with tumor necrosis factor receptor-associated factors and activation of NF-kappab and c-Jun N-terminal kinase. J Biol Chem 1998; 273: 20551-5.

[155] Iotsova V, Caamano J, Loy J, *et al*. Osteopetrosis in mice lacking NF-kappaB1 and NF-kappaB2. Nat Med 1997; 3: 1285-9.

[156] Franzoso G, Carlson L, Xing L, *et al*. Requirement for NF-kappaB in osteoclast and B-cell development. Genes Dev 1997; 11: 3482-96.

[157] Bushdid PB, Brantley DM, Yull FE, *et al*. Inhibition of NF-kappaB activity results in disruption of the apical ectodermal ridge and aberrant limb morphogenesis. Nature 1998; 392: 615-8.

[158] Yao Z, Zhang J, Dai J, *et al*. Ethanol activates NFkappaB DNA binding and p56lck protein tyrosine kinase in human osteoblast-like cells. Bone 2001; 28: 167-73.

[159] Andela VB, Sheu TJ, Puzas EJ, *et al*. Malignant reversion of a human osteosarcoma cell line, Saos-2, by inhibition of NFkappaB. Biochem Biophys Res Commun 2002; 297: 237-41.

[160] Koul D, Yao Y, Abbruzzese JL, *et al*. Tumor suppressor MMAC/PTEN inhibits cytokine-induced NFkappaB activation without interfering with the IkappaB degradation pathway. J Biol Chem 2001; 276: 11402-8.

[161] Trompouki E, Hatzivassiliou E, Tsichritzis T, *et al*. CYLD is a deubiquitinating enzyme that negatively regulates NF-kappaB activation by TNFR family members. Nature 2003; 424: 793-6.

[162] Brummelkamp TR, Nijman SM, Dirac AM, *et al*. Loss of the cylindromatosis tumour suppressor inhibits apoptosis by activating NF-kappaB. Nature 2003; 424: 797-801.

[163] Kovalenko A, Chable-Bessia C, Cantarella G, *et al*. The tumour suppressor CYLD negatively regulates NF-kappaB signalling by deubiquitination. Nature 2003; 424: 801-5.

[164] Rocha S, Martin AM, Meek DW, *et al*. p53 represses cyclin D1 transcription through down regulation of Bcl-3 and inducing increased association of the p52 NF-kappaB subunit with histone deacetylase 1. Mol Cell Biol 2003; 23: 4713-27.

[165] Mayo MW, Madrid LV, Westerheide SD, *et al*. PTEN blocks tumor necrosis factor-induced NF-kappa B-dependent transcription by inhibiting the transactivation potential of the p65 subunit. J Biol Chem 2002; 277: 11116-25.

[166] Rocha SPerkins ND. ARF the integrator: linking NF-kappaB, p53 and checkpoint kinases. Cell Cycle 2005; 4: 756-9.

[167] Bentires-Alj M, Hellin AC, Ameyar M, *et al*. Stable inhibition of nuclear factor kappaB in cancer cells does not increase sensitivity to cytotoxic drugs. Cancer Res 1999; 59: 811-5.

[168] Natarajan K, Singh S, Burke TR Jr., *et al*. Caffeic acid phenethyl ester is a potent and specific inhibitor of activation of nuclear transcription factor NF-kappa B. Proc Natl Acad Sci USA 1996; 93: 9090-5.

[169] Singh R, Ahmed S, Islam N, *et al*. Epigallocatechin-3-gallate inhibits interleukin-1beta-induced expression of nitric oxide

synthase and production of nitric oxide in human chondrocytes: suppression of nuclear factor kappaB activation by degradation of the inhibitor of nuclear factor kappaB. Arthritis Rheum 2002; 46: 2079-86.

[170] Chainy GB, Manna SK, Chaturvedi MM, *et al*. Anethole blocks both early and late cellular responses transduced by tumor necrosis factor: effect on NF-kappaB, AP-1, JNK, MAPKK and apoptosis. Oncogene 2000; 19: 2943-50.

[171] Manna SK, Sah NK, Newman RA, *et al*. Oleandrin suppresses activation of nuclear transcription factor-kappaB, activator protein-1, and c-Jun NH2-terminal kinase. Cancer Res 2000; 60: 3838-47.

[172] Takada Y, Aggarwal BB. Betulinic acid suppresses carcinogen-induced NF-kappa B activation through inhibition of I kappa B alpha kinase and p65 phosphorylation: abrogation of cyclooxygenase-2 and matrix metalloprotease-9. J Immunol 2003; 171: 3278-86.

[173] Manna SK, Mukhopadhyay A, Aggarwal BB. Leflunomide suppresses TNF-induced cellular responses: effects on NF-kappa B, activator protein-1, c-Jun N-terminal protein kinase, and apoptosis. J Immunol 2000; 165: 5962-9.

[174] Wang P, Wu P, Siegel MI, *et al*. Interleukin (IL)-10 inhibits nuclear factor kappa B (NF kappa B) activation in human monocytes. IL-10 and IL-4 suppress cytokine synthesis by different mechanisms. J Biol Chem 1995; 270: 9558-63.

[175] Manna SK, Mukhopadhyay A, Aggarwal BB. Human chorionic gonadotropin suppresses activation of nuclear transcription factor-kappa B and activator protein-1 induced by tumor necrosis factor. J Biol Chem 2000; 275: 13307-14.

[176] Manna SK, Aggarwal BB. Alpha-melanocyte-stimulating hormone inhibits the nuclear transcription factor NF-kappa B activation induced by various inflammatory agents. J Immunol 1998; 161: 2873-80.

[177] Haeffner A, Thieblemont N, Deas O, *et al*. Inhibitory effect of growth hormone on TNF-alpha secretion and nuclear factor-kappaB translocation in lipopolysaccharide-stimulated human monocytes. J Immunol 1997; 158: 1310-4.

[178] Manna SK, Mukhopadhyay A, Aggarwal BB. IFN-alpha suppresses activation of nuclear transcription factors NF-kappa B and activator protein 1 and potentiates TNF-induced apoptosis. J Immunol 2000; 165: 4927-34.

[179] Ehrlich LC, Hu S, Sheng WS, *et al*. Cytokine regulation of human microglial cell IL-8 production. J Immunol 1998; 160: 1944-8.

[180] Hideshima T, Chauhan D, Schlossman R, *et al*. The role of tumor necrosis factor alpha in the pathophysiology of human multiple myeloma: therapeutic applications. Oncogene 2001; 20: 4519-27.

[181] Fiedler MA, Wernke-Dollries K, Stark JM. Inhibition of TNF-alpha-induced NF-kappaB activation and IL-8 release in A549 cells with the proteasome inhibitor MG-132. Am J Respir Cell Mol Biol 1998; 19: 259-68.

[182] Page S, Fischer C, Baumgartner B, *et al*. 4-Hydroxynonenal prevents NF-kappaB activation and tumor necrosis factor expression by inhibiting IkappaB phosphorylation and subsequent proteolysis. J Biol Chem 1999; 274: 11611-8.

[183] Ji C, Kozak KR, Marnett LJ. IkappaB kinase, a molecular target for inhibition by 4-hydroxy-2-nonenal. J Biol Chem 2001; 276: 18223-8.

[184] Castro AC, Dang LC, Soucy F, *et al*. Novel IKK inhibitors: beta-carbolines. Bioorg Med Chem Lett 2003; 13: 2419-22.

[185] Kishore N, Sommers C, Mathialagan S, *et al*. A selective IKK-2 inhibitor blocks NF-kappa B-dependent gene expression in interleukin-1 beta-stimulated synovial fibroblasts. J Biol Chem 2003; 278: 32861-71.

[186] Burke JR, Pattoli MA, Gregor KR, *et al*. BMS-345541 is a highly selective inhibitor of I kappa B kinase that binds at an allosteric site of the enzyme and blocks NF-kappa B-dependent transcription in mice. J Biol Chem 2003; 278: 1450-6.

[187] Chen L, Fischle W, Verdin E, *et al*. Duration of nuclear NF-kappaB action regulated by reversible acetylation. Science 2001; 293: 1653-7.

[188] Abu-Amer Y, Dowdy SF, Ross FP, *et al*. TAT fusion proteins containing tyrosine 42-deleted IkappaBalpha arrest osteoclasto-genesis. J Biol Chem 2001; 276: 30499-503.

[189] Higgins KA, Perez JR, Coleman TA, *et al*. Antisense inhibition of the p65 subunit of NF-kappa B tumorigenicity and causes tumor regression. Proc Natl Acad Sci USA 1993; 90: 9901-5.

[190] Kopp E, Ghosh S. Inhibition of NF-kappa B by sodium salicylate and aspirin. Science 1994; 265: 956-9.

[191] Surabhi RM, Gaynor RB. RNA interference directed against viral and cellular targets inhibits human immunodeficiency Virus Type 1 replication. J Virol 2002; 76: 12963-73.

[192] Takaesu G, Surabhi RM, Park KJ, *et al*. TAK1 is critical for IkappaB kinase-mediated activation of the NF-kappaB pathway. J Mol Biol 2003; 326: 105-15.

[193] Liu J, Beller DI. Distinct pathways for NF-kappa B regulation are associated with aberrant macrophage IL-12 production in lupus- and diabetes-prone mouse strains. J Immunol 2003; 170: 4489-96.

[194] Pieper GM, Riaz ul H. Activation of nuclear factor-kappaB in cultured endothelial cells by increased glucose concentration: prevention by calphostin C. J Cardiovasc Pharmacol 1997; 30: 528-32.

[195] Cunningham MD, Cleaveland J, Nadler SG. An intracellular targeted NLS peptide inhibitor of karyopherin alpha:NF-kappa B interactions. Biochem Biophys Res Commun 2003; 300: 403-7.

[196] May MJ, D'Acquisto F, Madge LA, *et al*. Selective inhibition of NF-kappaB activation by a peptide that blocks the interaction of NEMO with the IkappaB kinase complex. Science 2000; 289: 1550-4.

[197] Kwak HB, Lee SW, Lee DG, *et al*. A hybrid peptide derived from cecropin-A and magainin-2 inhibits osteoclast differentiation. Life Sci 2003; 73: 993-1005.

[198] Takada Y, Singh S, Aggarwal BB. Identification of a p65 peptide that selectively inhibits NF-kappa B activation induced by various inflammatory stimuli and its role in down-regulation of NF-kappaB-mediated gene expression and up-regulation of apoptosis. J Biol Chem 2004; 279: 15096-104.

[199] Takada Y, Bhardwaj A, Potdar P, *et al*. Nonsteroidal anti-inflammatory agents differ in their ability to suppress NF-kappaB activation, inhibition of expression of cyclooxygenase-2 and cyclin D1, and abrogation of tumor cell proliferation. Oncogene 2004; 23: 9247-58.

[200] Aggarwal BB. Nuclear factor-kappaB: the enemy within. Cancer Cell 2004; 6: 203-8.

[201] Perfettini JL, Roumier T, Castedo M, *et al*. NF-kappaB and p53 are the dominant apoptosis-inducing transcription factors elicited by the HIV-1 envelope. J Exp Med 2004; 199: 629-40.

[202] Ravi R, Bedi GC, Engstrom LW, *et al*. Regulation of death receptor expression and TRAIL/Apo2L-induced apoptosis by NF-kappaB. Nat Cell Biol 2001; 3: 409-16.

[203] Fujioka S, Schmidt C, Sclabas GM, *et al*. Stabilization of p53 is a novel mechanism for proapoptotic function of NF-kappaB. J Biol Chem 2004; 279: 27549-59.

[204] Li Z, Niu J, Uwagawa T, *et al*. Function of polo-like kinase 3 in NF-kappaB-mediated proapoptotic response. J Biol Chem 2005; 280: 16843-50.

[205] Shetty S, Graham BA, Brown JG, *et al*. Transcription factor NF-kappaB differentially regulates death receptor 5 expression involving histone deacetylase 1. Mol Cell Biol 2005; 25: 5404-16.

[206] Ryan KM, O'Prey J, Vousden KH Loss of nuclear factor-kappaB is tumor promoting but does not substitute for loss of p53. Cancer Res 2004; 64: 4415-8.

[207] Bohuslav J, Chen LF, Kwon H, *et al*. p53 induces NF-kappaB activation by an IkappaB kinase-independent mechanism involving phosphorylation of p65 by ribosomal S6 kinase 1. J Biol Chem 2004; 279: 26115-25.

[208] Perkins ND. NF-kappaB: tumor promoter or suppressor? Trends Cell Biol 2004; 14: 64-9.

[209] Manna SK, Mukhopadhyay A, Van NT, *et al*. Silymarin suppresses TNF-induced activation of NF-kappa B, c-Jun N-terminal kinase, and apoptosis. J Immunol 1999; 163: 6800-9.

[210] Manna SK, Bueso-Ramos C, Alvarado F, *et al*. Calagualine inhibits nuclear transcription factors-kappaB activated by various inflammatory and tumor promoting agents. Cancer Lett 2003; 190: 171-82.

[211] Takada Y, Murakami A, Aggarwal BB. Zerumbone abolishes NF-kappaB and IkappaBalpha kinase activation leading to suppression of antiapoptotic and metastatic gene expression, upregulation of apoptosis, and downregulation of invasion. Oncogene 2005.

[212] Takada Y, Andreeff M, Aggarwal BB. Indole-3-carbinol suppresses NF-kappaB and IkappaBalpha kinase activation, causing inhibition of expression of NF-kappaB-regulated antiapoptotic and metastatic

gene products and enhancement of apoptosis in myeloid and leukemia cells. Blood 2005; 106: 641-9.

[213] Ichikawa H, Takada Y, Murakami A, *et al*. Identification of a novel blocker of I kappa B alpha kinase that enhances cellular apoptosis and inhibits cellular invasion through suppression of NF-kappa B-regulated gene products. J Immunol 2005; 174: 7383-92.

[214] Takada Y, Kobayashi Y, Aggarwal BB. Evodiamine abolishes constitutive and inducible NF-kappaB activation by inhibiting IkappaBalpha kinase activation, thereby suppressing NF-kappaB-regulated antiapoptotic and metastatic gene expression, up-regulating apoptosis, and inhibiting invasion. J Biol Chem 2005; 280: 17203-12.

[215] Shishodia S, Aggarwal BB. Guggulsterone inhibits NF-kappaB and IkappaBalpha kinase activation, suppresses expression of anti-apoptotic gene products, and enhances apoptosis. J Biol Chem 2004; 279: 47148-58.

[216] Shishodia S, Koul D, Aggarwal BB. Cyclooxygenase (COX)-2 inhibitor celecoxib abrogates TNF-induced NF-kappa B activation through inhibition of activation of I kappa B alpha kinase and Akt in human non-small cell lung carcinoma: correlation with suppression of COX-2 synthesis. J Immunol 2004; 173: 2011-22.

[217] Shishodia S, Aggarwal BB. Cyclooxygenase (COX)-2 inhibitor celecoxib abrogates activation of cigarette smoke-induced nuclear factor (NF)-kappaB by suppressing activation of IkappaBalpha kinase in human non-small cell lung carcinoma: correlation with suppression of cyclin D1, COX-2, and matrix metalloproteinase-9. Cancer Res 2004; 64: 5004-12.

[218] Zheng B, Georgakis GV, Li Y, *et al*. Induction of cell cycle arrest and apoptosis by the proteasome inhibitor PS-341 in Hodgkin disease cell lines is independent of inhibitor of nuclear factor-kappaB mutations or activation of the CD30, CD40, and RANK receptors. Clin Cancer Res 2004; 10: 3207-15.

[219] Bharti AC, Takada Y, Aggarwal BB. Curcumin (diferuloylmethane) inhibits receptor activator of NF-kappa B ligand-induced NF-kappa B activation in osteoclast precursors and suppresses osteoclastogenesis. J Immunol 2004; 172: 5940-7.

[220] Takada Y, Khuri FR, Aggarwal BB. Protein farnesyltransferase inhibitor (SCH 66336) abolishes NF-kappaB activation induced by various carcinogens and inflammatory stimuli leading to suppression of NF-kappaB-regulated gene expression and up-regulation of apoptosis. J Biol Chem 2004; 279: 26287-99.

[221] Rao Ch V, Li X, Manna SK, *et al*. Human chorionic gonadotropin decreases proliferation and invasion of breast cancer MCF-7 cells by inhibiting NF-kappaB and AP-1 activation. J Biol Chem 2004; 279: 25503-10.

[222] Giri DK, Pantazis P, Aggarwal BB. Cellular resistance to vincristine suppresses NF-kappa B activation and apoptosis but enhances c-Jun-NH2-terminal protein kinase activation by tumor necrosis. Apoptosis 1999; 4: 291-301.

[223] Takada Y, Aggarwal BB. Flavopiridol inhibits NF-kappaB activation induced by various carcinogens and inflammatory agents through inhibition of IkappaBalpha kinase and p65 phosphorylation: abrogation of cyclin D1, cyclooxygenase-2, and matrix metalloprotease-9. J Biol Chem 2004; 279: 4750-9.

[224] Takada Y, Mukhopadhyay A, Kundu GC, *et al*. Hydrogen peroxide activates NF-kappa B through tyrosine phosphorylation of I kappa B alpha and serine phosphorylation of p65: evidence for the involvement of I kappa B alpha kinase and Syk protein-tyrosine kinase. J Biol Chem 2003; 278: 24233-41.

[225] Estrov Z, Shishodia S, Faderl S, *et al*. Resveratrol blocks interleukin-1beta-induced activation of the nuclear transcription factor NF-kappaB, inhibits proliferation, causes S-phase arrest, and induces apoptosis of acute myeloid leukemia cells. Blood 2003; 102: 987-95.

[226] Majumdar S, Aggarwal BB. Adenosine suppresses activation of nuclear factor-kappaB selectively induced by tumor necrosis factor in different cell types. Oncogene 2003; 22: 1206-18.

[227] Ashikawa K, Majumdar S, Banerjee S, *et al*. Piceatannol inhibits TNF-induced NF-kappaB activation and NF-kappaB-mediated gene expression through suppression of IkappaBalpha kinase and p65 phosphorylation. J Immunol 2002; 169: 6490-7.

[228] Mukhopadhyay A, Shishodia S, Suttles J, *et al*. Ectopic expression of protein-tyrosine kinase Bcr-Abl suppresses tumor necrosis factor (TNF)-induced NF-kappa B activation and IkappaBalpha phosphorylation. Relationship with down-regulation of TNF receptors. J Biol Chem 2002; 277: 30622-8.

[229] Majumdar S, Lamothe B, Aggarwal BB. Thalidomide suppresses NF-kappa B activation induced by TNF and H2O2, but not that activated by ceramide, lipopolysaccharides, or phorbol ester. J Immunol 2002; 168: 2644-51.

[230] Majumdar S, Aggarwal BB. Methotrexate suppresses NF-kappaB activation through inhibition of IkappaBalpha phosphorylation and degradation. J Immunol 2001; 167: 2911-20.

[231] Manna SK, Mukhopadhyay A, Aggarwal BB. Resveratrol suppresses TNF-induced activation of nuclear transcription factors NF-kappa B, activator protein-1, and apoptosis: potential role of reactive oxygen intermediates and lipid peroxidation. J Immunol 2000; 164: 6509-19.

[232] Manna SK, Aggarwal BB. Vesnarinone suppresses TNF-induced activation of NF-kappa B, c-Jun kinase, and apoptosis. J Immunol 2000; 164: 5815-25.

[233] Manna SK, Aggarwal BB. Wortmannin inhibits activation of nuclear transcription factors NF-kappaB and activated protein-1 induced by lipopolysaccharide and phorbol ester. FEBS Lett 2000; 473: 113-8.

[234] Manna SK, Kuo MT, Aggarwal BB. Overexpression of gamma-glutamylcysteine synthetase suppresses tumor necrosis factor-induced apoptosis and activation of nuclear transcription factor-kappa B and activator protein-1. Oncogene 1999; 18: 4371-82.

[235] Manna SK, Gad YP, Mukhopadhyay A, *et al*. Suppression of tumor necrosis factor-activated nuclear transcription factor-kappaB, activator protein-1, c-Jun N-terminal kinase, and apoptosis by beta-lapachone. Biochem Pharmacol 1999; 57: 763-74.

[236] Krishnamoorthy RR, Crawford MJ, Chaturvedi MM, *et al*. Photo-oxidative stress down-modulates the activity of nuclear factor-kappaB *via* involvement of caspase-1, leading to apoptosis of photoreceptor cells. J Biol Chem 1999; 274: 3734-43.

[237] Manna SK, Aggarwal BB. Interleukin-4 down-regulates both forms of tumor necrosis factor receptor and receptor-mediated apoptosis, NF-kappaB, AP-1, and c-Jun N-terminal kinase. Comparison with interleukin-13. J Biol Chem 1998; 273: 33333-41.

[238] Kumar A, Dhawan S, Aggarwal BB. Emodin (3-methyl-1,6,8-trihydroxyanthraquinone) inhibits TNF-induced NF-kappaB activation, IkappaB degradation, and expression of cell surface adhesion proteins in human vascular endothelial cells. Oncogene 1998; 17: 913-8.

[239] Manna SK, Aggarwal BB. IL-13 suppresses TNF-induced activation of nuclear factor-kappa B, activation protein-1, and apoptosis. J Immunol 1998; 161: 2863-72.

[240] Singh S, Natarajan K, Aggarwal BB. Capsaicin (8-methyl-N-vanillyl-6-nonenamide) is a potent inhibitor of nuclear transcription factor-kappa B activation by diverse agents. J Immunol 1996; 157: 4412-20.

[241] Singh S, Aggarwal BB. Protein-tyrosine phosphatase inhibitors block tumor necrosis factor-dependent activation of the nuclear transcription factor NF-kappa B. J Biol Chem 1995; 270: 10631-9.

[242] Kapahi P, Takahashi T, Natoli G, *et al*. Inhibition of NF-kappa B activation by arsenite through reaction with a critical cysteine in the activation loop of Ikappa B kinase. J Biol Chem 2000; 275: 36062-6.

[243] Ogino S, Tsuruma K, Uehara T, *et al*. Herbimycin A abrogates nuclear factor-kappaB activation by interacting preferentially with the IkappaB kinase beta subunit. Mol Pharmacol 2004; 65: 1344-51.

[244] Yang F, Yamashita J, Tang E, *et al*. The zinc finger mutation C417R of I-kappa B kinase gamma impairs lipopolysaccharide- and TNF-mediated NF-kappa B activation through inhibiting phosphorylation of the I-kappa B kinase beta activation loop. J Immunol 2004; 172: 2446-52.

[245] Garcia-Pineres AJ, Lindenmeyer MT, Merfort I. Role of cysteine residues of p65/NF-kappaB on the inhibition by the sesquiterpene lactone parthenolide and N-ethyl maleimide, and on its transactivating potential. Life Sci 2004; 75: 841-56.

[246] Nishi T, Shimizu N, Hiramoto M, *et al*. Spatial redox regulation of a critical cysteine residue of NF-kappa B *in vivo*. J Biol Chem 2002; 277: 44548-56.

[247] Singh S, Aggarwal BB. Activation of transcription factor NF-kappa B is suppressed by curcumin (diferuloylmethane). J Biol Chem 1995; 270: 24995-5000.

[248] Han SS, Chung ST, Robertson DA, *et al*. Curcumin causes the growth arrest and apoptosis of B cell lymphoma by downregulation of egr-1, c-myc, bcl-XL, NF-kappa B, and p53. Clin Immunol 1999; 93: 152-61.

[249] Natarajan C, Bright JJ. Curcumin inhibits experimental allergic encephalomyelitis by blocking IL-12 signaling through Janus kinase-STAT pathway in T lymphocytes. J Immunol 2002; 168: 6506-13.

[250] Bharti AC, Shishodia S, Reuben JM, *et al*. Nuclear factor-kappaB and STAT3 are constitutively active in CD138+ cells derived from multiple myeloma patients, and suppression of these transcription factors leads to apoptosis. Blood 2004; 103: 3175-84.

[251] Anto RJ, Mukhopadhyay A, Denning K, *et al*. Curcumin (diferuloylmethane) induces apoptosis through activation of caspase-8, BID cleavage and cytochrome c release: its suppression by ectopic expression of Bcl-2 and Bcl-xl. Carcinogenesis 2002; 23: 143-50.

[252] Shishodia S, Amin HM, Lai R, *et al*. Curcumin (diferuloylmethane) inhibits constitutive NF-kappaB activation, induces G1/S arrest, suppresses proliferation, and induces apoptosis in mantle cell lymphoma. Biochem Pharmacol 2005; 70: 700-13.

[253] Siwak DR, Shishodia S, Aggarwal BB, *et al*. Curcumin-induced antiproliferative and proapoptotic effects in melanoma cells are associated with suppression of IkappaB kinase and nuclear factor kappaB activity and are independent of the B-Raf/mitogen-activated/extracellular signal-regulated protein kinase pathway and the Akt pathway. Cancer 2005; 104: 879-90.

[254] Bharti AC, Takada Y, Aggarwal BB. PARP cleavage and caspase activity to assess chemosensitivity. Methods Mol Med 2005; 111: 69-78.

[255] Dorai T, Aggarwal BB. Role of chemopreventive agents in cancer therapy. Cancer Lett 2004; 215: 129-40.

[256] Li L, Aggarwal BB, Shishodia S, *et al*. Nuclear factor-kappaB and IkappaB kinase are constitutively active in human pancreatic cells, and their down-regulation by curcumin (diferuloylmethane) is associated with the suppression of proliferation and the induction of apoptosis. Cancer 2004; 101: 2351-62.

[257] Aggarwal BB, Takada Y, Oommen OV. From chemoprevention to chemotherapy: common targets and common goals. Expert Opin Investig Drugs 2004; 13: 1327-38.

[258] Aggarwal S, Takada Y, Singh S, *et al*. Inhibition of growth and survival of human head and neck squamous cell carcinoma cells by curcumin *via* modulation of nuclear factor-kappaB signaling. Int J Cancer 2004; 111: 679-92.

[259] Mukhopadhyay A, Bueso-Ramos C, Chatterjee D, *et al*. Curcumin downregulates cell survival mechanisms in human prostate cancer cell lines. Oncogene 2001; 20: 7597-609.

[260] Aggarwal BB, Kumar A, Bharti AC. Anticancer potential of curcumin: preclinical and clinical studies. Anticancer Res 2003; 23: 363-98.

[261] Aggarwal BB, Shishodia S. Suppression of the nuclear factor-kappaB activation pathway by spice-derived phytochemicals: reasoning for seasoning. Ann N Y Acad Sci 2004; 1030: 434-41.

[262] Bharti AC, Donato N, Aggarwal BB. Curcumin (diferuloylmethane) inhibits constitutive and IL-6-inducible STAT3 phosphorylation in human multiple myeloma cells. J Immunol 2003; 171: 3863-71.

[263] Yan C, Jamaluddin MS, Aggarwal B, *et al*. Gene expression profiling identifies activating transcription factor 3 as a novel contributor to the proapoptotic effect of curcumin. Mol Cancer Ther 2005; 4: 233-41.

[264] Shishodia S, Gethi G, Aggarwal BB. Curcumin: Getting back to the roots., Vol. In press: The annals of the New York Academy of Sciences, 2005.

[265] Aggarwal BB, Kumer S, Aggarwal S, *et al*. Curcumin derived from turmeric (Curcuma longa): A spice for all seasons,, p. 349-87: CRC press, 2005.

[266] Aggarwal BB, Kumar A, Bharti AC. Therapeutic potential of curcumin derived from turmeric (Curcuma longa). p. 781-812. New York: Marcel Dekker, 2004.

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