

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

Bharat B. Aggarwal*, Gautam Sethi, Asha Nair, and Haruyo Ichikawa

Cytokine Research Laboratory, Department of Experimental Therapeutics, The University of Texas M. D. Anderson Cancer Center, Houston, Texas 77030, USA

Abstract: Nuclear factor- κ B (NF- κ B) 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- κ B 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- κ B 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- κ B 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- κ B activation. Although I κ B α kinase is the major kinase, over 30 different protein kinases have been linked to the activation of NF- κ B by different stimuli. The development of a drug that can specifically suppress NF- κ B activation requires a full understanding of the mechanism by which NF- κ B is activated in response to these various stimuli.

Key Words: NF- κ B, IKK, antitumor, TNF, inflammation.

1. INTRODUCTION

Nuclear factor- κ B (NF- κ B), 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- κ B is now known to comprise a family of ubiquitous proteins. NF- κ B 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- κ B is the most common heterodimer formed between Rel A and p50. Dimeric NF- κ B 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 κ B sites [2, 3].

The various inhibitors of NF- κ B include I κ B α , I κ B β , I κ B γ (derived from the C-terminal of p100), I κ B ϵ , 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- κ B 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- κ B consists of p50, p65, and I κ B α . I κ B α mediates transient gene expression, whereas I κ B β mediates persistent response.

The I κ B proteins are expressed in a tissue-specific manner and have distinct affinities for individual Rel/NF- κ B complexes. I κ Bs 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. I κ Bs bind to NF- κ B dimers and sterically block the function of their nuclear localization sequences, thereby causing their cytoplasmic retention. Most agents that activate NF- κ B mediate the phosphorylation-induced degradation of I κ B. On receipt of a signal, phosphorylation of I κ B α takes place on two conserved serine residues (S32 and S36) in the N-terminal regulatory domain. However, another member of the I κ B family, Bcl-3, stimulates transcription after interacting with p50 and p52 subunits of NF- κ B.

Several of the I κ B kinases (IKKs) have been characterized, namely, IKK α , IKK β , and IKK γ . Mutation analysis revealed that IKK α and not IKK β mediates proinflammatory signals. Once phosphorylated, the I κ Bs, which are still bound to NF- κ B, almost immediately undergo a second posttranslational modification known as polyubiquitination. The major ubiquitin acceptor sites in human I κ B α 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, I κ Bs are degraded in 26S proteasomes, leading to the release of NF- κ B dimers, which translocate into the nucleus [3, 4]. In contrast, the activation of NF- κ B in response to ultraviolet (UV) radiation is accompanied by I κ B α degradation but not phosphorylation on the N-terminus of I κ B α [5]. Hypoxia or

*Address correspondence to this author at the Cytokine Research Laboratory, Department of Experimental Therapeutics, The University of Texas M. D. Anderson Cancer Center, Houston, Texas 77030, USA; Tel: 713-794-1817; Fax: 713-794-1613; E-mail: aggarwal@mdanderson.org

Table 1. A List of Inducers of NF- κ B Activation

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

(Table 1. Contd....)

Myc ¹¹⁷	<i>Angiostrongylus cantonensis</i>	<i>necatrix</i>	Wounding combined with
bcr-ab1 ¹¹⁶	<i>Bacteroides forsythus</i>	Leishmania	TUDCA
HeNe irradiation	<i>Bartonella henselae</i>	Phospholipomannan	Tuberous sclerosis complex
Wounding combined with thermal irradiation	<i>Bordetella pertussis</i>		Uremic toxins VLDL
	Cycloprodigiosin	Physiological Mediators	
Oxidative stress	Dacarbazine	Alloxan	Chemical Agents
Glutathione	Orengedeokuto	Angiotensin II	L-NMA
Hydrogen peroxide ²²⁴	Diazoxide	Bradykinin	Ethanol
Catalase	5,6-dimethylxanthenone-4-acetic acid	Fibrinogen	Monensin
Nitric oxide	Flavone-8-acetic acid	Hemoglobin	Nicotine
Ozone	Haloperidol	Hyaluronan	Adriamycin
Peroxyinitrite	Kunbi-Boshin-Hangam-Tang	Thioredoxin	2-Deoxyglucose
Butyl peroxide	Methamphetamine	Kainic acid	Anisomycin
Cerulein	Norepinephrine	L-Glutamate	Benzyl isothiocyanate
Pervanadate	Oltipraz	Melanin	Bisperoxovanadium phosphotyrosyl
Reoxygenation	Phenobarbital	Violacein	Brefeldin A
	Protocatechuic acid	Vitamin D ₃	Cadmium
Environmental hazards		Adenosine	Colchicine
Cigarette smoke ⁸⁷⁻⁸⁸	Fatty acids	Activin A	Calcium Ionophore plus phorbol ester
PH ⁹¹	Palmitate	Albumin	Calyculin A
Arsenic	Leukotriene B4	Allergin	Ceramide-β-galactose
Polychlorinated biophenyl-77	Free fatty acids	Acrp30/adiponectin	2-chloroethyl ethyl sulfide
Benzo[a]pyrene diol epoxide ⁸⁷	Linoleic acid	Adrenomedullin	Con A
Crocidolite asbestos fibers	Oleic acid	<i>Aeginetia indica</i> L (AIL)b-A	Cycloheximide
Dicamba	Saturated fatty acids	Bronchoalveolar lavage fluid	Cyclopiazonic acid
Diesel exhaust particles	Lysophosphatidic acid	Brusatol	Diquat
Gliadin		Homocysteine	2,4-Dinitrofluorobenzene
House dust mite	Modified Proteins	Cryptdins	Ferrocene
Dust particles	LDL	Deoxycholic acid	Forskolin
Fear-potentiated startle response	AGEs	12(R)-hydroxyeicosatrienoic acid	Gadolinium chloride
Lead	Amyloid protein fragment	6-Hydroxydopamine	Glass fibers
Lead chromate	Glycosylated oxyhemoglobin	Human peptide transporter 1	Sodium butyrate trichostatin A
Noise	Maleylated bovine serum Albumin	LTP	Malondialdehyde
Oily fly ash	Neurotrophin receptor proteolytic fragments	LysoPC	MDMA
Silica particles	Nonamyloid beta component of Alzheimer's disease	Neuormelanin	MEN 17055 ¹¹³
Wood smoke	ATM ¹⁰⁸	NS-398	N-methyl-D-aspartate
Zymosan		Osteopontin	Mycophenolic acid
		PAF	Nafenopin
Chemotherapeutic drugs	Heat shock protein	PCSC	N-Nitrosomorphine
Cisplatin	HSP60	Prolactin	Nocodazol
Paclitaxel ¹⁰³	HSPb1	PIF	Okadaic acid ⁸⁹
Doxorubicin ¹⁰⁰		12-Lipoxygenase	Peplomycin
Tamoxifen		Thrombin	Phytohemagglutinin
Vinblastine	Apoptotic Mediators	Streptozotocin	Phorbol ester ⁸⁷
Vincristine ²²²	Anti-Fas/Apo-1	Substance P	Podophyllotoxin
Etoposide	PARP	Potassium	Polychlorinated biophenyl-77
ara-C		Burn patient serum	Prostratin
Bryostatin-1	Metals	Anaphylatoxin C3a and C5a	Pyrogallol
Camptothecin ¹⁰¹	Lithium	Angiotensin II type 1 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 leukotrienes	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	
Anthrakinone	Quercetin	Polysaccharides of <i>Poria cocos</i>	
Ciprofibrate	Quinolinic acid	Regulatory RNA	
	Safflower polysaccharides	S100B	
	Sanglifehrin A	Sleep deprivation	

TNF, tumor necrosis factor; RANKL, receptor activator of nuclear factor- κ B ligand; IL, interleukin; TRAIL, TNF-related apoptosis-inducing ligand; TWEAK, TNF-like weak inducer of apoptosis; THANK, TNF homologue that activates apoptosis, nuclear factor- κ B, and c-Jun NH2 terminal kinase; BAFF, B cell-activating factor; LT, lymphotoxin; LIF, leukemia inhibitory factor; AT, ataxia telangiectasia; Ig, immunoglobulin; GM-CSF, 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; P1c, 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 glycosylated end products; HSP, heat shock protein; PARP, poly(ADP)ribose polymerase; Acip30, adipocyte complement-related protein of 30 kDa; LTP, long-term potentiation, LysoPC, lysophosphatidylcholine; PCSC, polysaccharide from *Poria cocos*; PIF, proteolysis-inducing factor; TUDCA, taurodeoxycholic acid; VLDL, very LDL; MDMA, 3,4-methylenedioxymethamphetamine.

Source: <http://www.ncbi.nlm.nih.gov/entrez/query.fcgi>, [http://people.bu.edu/gilmore/nf- \$\kappa\$ b/](http://people.bu.edu/gilmore/nf-κb/).

indicates that NF- κ B activation is highly complex and may involve dozens of different protein kinases [12-53] (Table 2). Besides NIK, IKK- α , and IKK- β , NF- κ B 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- κ B activator. For instance, IKK can be phosphorylated by NIK, mitogen-activated protein kinase kinase, or Akt. Although IKK is required for NF- κ B activation by most agents, a few (such as human epithelial receptor type 2, H₂O₂, pervanadate, x-rays, and γ -radiation) activate NF- κ B through IKK-independent pathways [54]. Although several signaling proteins and protein kinases have been identified recently that mediate NF- κ B activation, more kinases and protein phosphatases remain to be identified. Besides the ubiquitin-dependent 26S proteasome, which has a role in I κ B α degradation [55], other proteases have also been implicated in NF- κ B activation [56].

The genetic deletions of different NF- κ B 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- κ B in TNF-induced apoptosis. Furthermore, mice lacking the RelA subunit were brought to term only in a TNFR1-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 I κ B α gene leads to early neonatal lethality caused by inflammatory dermatitis and granulocytosis [60] that are most likely induced by constitutive activation of NF- κ B, leading to expression of the granulocyte colony-stimulating factor.

NF- κ B activation has been implicated in a wide variety of diseases, including cancers, diabetes mellitus, cardiovascular diseases, autoimmune diseases, viral replication, septic shock, neurodegenerative disorders, ataxia telangiectasia (AT), arthritis, asthma, inflammatory bowel disease, and several other inflammatory conditions (Table 3). For example, activation of NF- κ B by LPS may contribute to the development of septic shock because NF- κ B 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- κ B. Additionally, in chronic Alzheimer's disease, the amyloid β peptide causes production of reactive oxygen intermediates and indirectly activates gene expression through κ B sites [62]. The influenza virus protein hemagglutinin also activates NF- κ B, 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- κ B, which then activates other genes [64], and mice that are susceptible to atherosclerosis exhibit NF- κ B 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- κ B [66]. Finally, a truncated form of I κ B α was shown to protect AT cells, which express constitutive levels of an NF- κ B-like activity, from ionizing radiation [67]. In light of all these findings, the abnormal activation or expression of NF- κ B is clearly associated with a wide variety of pathologic conditions. How NF- κ B activation mediates tumorigenesis is the focus of the current review.

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

Kinase	Ligand
IKK α	Proinflammatory cytokines (eg TNF, IL-1, IL6), LPS ²⁰⁻²²
IKK β	Proinflammatory cytokines (eg TNF, IL-1, IL6), LPS ²⁰⁻²²
IKK γ	Proinflammatory cytokines (eg TNF, IL-1, IL6), LPS ^{14,15}
IKKi/IKK ϵ	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	TNF α 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	<i>Pseudomonas aeruginosa</i> 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 ₂ O ₂ ²²⁴

IKK, κ B kinase; TNF, tumor necrosis factor; IL, interleukin; LPS, lipopolysaccharide; PMA, phorbol 12-myristate 13-acetate; NIK, NF- κ B-inducing kinase; NAK, NF- κ B-activating kinase (also called T2K-, TBK1-, or TRAF2-interacting kinase, PI3K, phosphatidylinositol 3 kinase; Akt, protein kinase B α ; TRC, T-cell receptor; HPK, hematopoietic protein kinase; PKC, protein kinase C; TPA, 12-O-tetradecanoylphorbol-13-acetate; MALT, mucosa-associated lymphoid tissue; IgM, immunoglobulin M; aPKC, atypical PKC; MEKK, mitogen-activated protein 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- κ B MEDIATES CARCINOGENESIS

NF- κ B 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- κ B.

2.1. NF- κ B Genes are Proto-Oncogenes

NF- κ B 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- κ B 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	<i>Helicobacter pylori</i> -associated gastritis
Adult T-cell leukemia	Incontinentia 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- κ B 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- κ B/*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- κ B 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- κ b2* genes are known to be translocated [76]. Furthermore, mutations in the *ikb α* gene were observed in Hodgkin's lymphoma [77], and blocking of *ikb α* 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- κ B complexes consisting of p50, p52, and Bcl-3 rather than p65 [7].

In 1992, Liptay *et al.* [38] discovered that the NF- κ B 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- κ B1 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- κ B-dependent transcription and a reduced level of NF- κ B1 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- κ B2/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 I κ B 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 I κ B α in cell lines expressing less p100. Based on these observations, Dejardin *et al.* [43] hypothesized that NF- κ B is involved in carcinogenesis and suggested that the p100/p52 NF- κ B subunit plays a role in the development of human breast cancers, possibly by sequestering other NF- κ B-related proteins in the cytoplasm.

2.2. Constitutively Active NF- κ B is Common in Tumor Cells

Constitutive expression of NF- κ B 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- κ B has also been noted in tissue samples from patients with cancer. For instance, NF- κ B is constitutively activated in high-grade squamous intraepithelial lesions and squamous cell carcinomas of the human uterine cervix [84]. NF- κ B was also implicated in an aggressive phenotype of renal cell carcinoma [47]. Of 45 cases of renal cell carcinoma, 33% showed an NF- κ B 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- κ B activation in their tumors; therefore, NF- κ B may be a cause of the inflammatory paraneoplastic syndrome [85]. Along similar lines, Yu *et al.* [48] reported that increased expression of NF- κ B 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- κ B, however, is not fully understood. The aberrant IKK activity and shorter half-life of I κ B α in B-cell lymphoma, the mutated I κ B α in Hodgkin's lymphoma, the IL-1 β 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- κ B activation [57].

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

During the last few years, various carcinogens have been shown to activate NF- κ B, 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- κ B 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- κ B in different cell types [88]. Unlike TNF-induced NF- κ B activation, the NF- κ B 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- κ B [89]. Additionally, NF- κ B 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- κ B. 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- κ B Activation

Besides inducing apoptosis, almost all chemotherapeutic agents also activate NF- κ B (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- κ B [106]. Egan *et al.* [107] found that I κ B-kinase- β -dependent NF- κ B 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- κ B through a common mitogen-activated protein kinase/extracellular signal-regulated kinase/p90(rsk) signaling pathway in response to distinct forms of DNA damage. Why these proapoptotic agents activate NF- κ B, however, is not clear.

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

Degradation of I κ B is a seminal step in the activation of NF- κ B. The I κ B kinases, IKK α and IKK β , have been implicated in both I κ B degradation and subsequent modification of NF- κ B. Using MEFs devoid of both IKK α and IKK β genes (IKK α / β (-/-)), Tergaonkar *et al.* [112] showed that the novel I κ B 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 I κ B α . The degradation of I κ B α was also shown to be partially blocked by the phosphatidylinositol 3-kinase inhibitor LY294002 and to be

mediated by the proteasome. The free NF- κ B generated by the DoxR-induced I κ B degradation in IKK α / β (-/-) cells activated the chromatin-based NF- κ B reporter gene and the expression of the endogenous target gene, I κ B α [112]. These results also imply that modification of NF- κ B by IKK α or IKK β , either prior or subsequent to its release from I κ B, is not essential for NF- κ B-mediated gene expression, at least in response to DNA damage. In addition, DoxR-induced cell death in IKK α / β (-/-) MEFs was enhanced by the simultaneous inhibition of NF- κ B activation by the blockage of proteasomal activity [112]. These findings show an additional pathway for activating NF- κ B 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- κ B activation is an IKK-independent I κ B α degradation pathway.

The new disaccharide anthracycline MEN 10755 induces activation of both NF- κ B and p53 transcription factors in A2780 cells [113]. However, the pharmacologic inhibition of NF- κ B activation does not modify the sensitivity of A2780 cells to MEN 10755 treatment. To better characterize the role of NF- κ B in MEN 10755-induced cytotoxicity, Camarda *et al.* [113] analyzed the expression of a number of genes known to be regulated by NF- κ B 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- κ B, although activated, appears to be non-functional. Other human carcinoma cell lines also activate NF- κ B 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- κ B transcriptional activity. Therefore, care should be taken when considering the pharmacologic inhibition of NF- κ B as a means to improve anticancer therapy.

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

2.5. Oncogenes Activate NF- κ B

Several oncogene products that can activate NF- κ B have been identified, including *ras* [115], *bcr-abl* [116], and *myc*

[117]. Oncogenic Ha-Ras-induced signaling also activates NF- κ B transcriptional activity, which is required for cellular transformation [118]. How these genes induce NF- κ B activation, however, is poorly understood. Oncogenic Ras enhances NF- κ B transcriptional activity through Raf-dependent and Raf-independent mitogen-activated protein kinase signaling pathways [119]. NF- κ B 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- κ B activation. Bcr-abl, which has been shown to cause chronic myelogenous leukemia, can elicit survival signals through induction of NF- κ B [122]. NF- κ B 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- κ B [117].

2.6. NF- κ B Activation is Required for Cell Proliferation

Several genes that mediate cell proliferation are also regulated by NF- κ B, including the growth factors TNF, IL-1 β , 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-1 β for acute myelogenous leukemia [125], and IL-6 for multiple myeloma [126] and head and neck squamous cell carcinoma [127]. Suppression of NF- κ B 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- κ B [128]. Additionally, in some cells, prostaglandin E₂ (PGE₂) was shown to induce proliferation of tumor cells, and the synthesis of cyclooxygenase-2, which controls PGE₂ production, was also shown to be regulated by NF- κ B activation [129].

The constitutive activation of NF- κ B 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- κ B. Furthermore, constitutive NF- κ B 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- κ B [131]. The EGF receptor was found to engage a receptor-interacting protein and NIK to activate NF- κ B, thus identifying a novel receptor-tyrosine kinase signalosome [132]. NF- κ B 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- κ B Inhibits Apoptosis

Both the prosurvival and antiapoptotic functions of NF- κ B 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-1 β -converting enzyme (FLICE)-like inhibitory protein (cFLIP), were shown to be controlled by NF- κ B 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- κ B

Cytokines	Receptors	TIEG	Cell proliferation	P450	Miscellaneous
TNF α ¹²³	β 2 microglobulin		COX-2 ¹²⁹	CYP2C11	α -1 Acid glycoprotein
TNF β	μ -opioid receptor	Transcription factors	c-myc	CYP2E1	a1-Antitrypsin,
IL-1 α ^{123,125}	2A	NF- κ B1			a2(1)-Collagen
IFN- α	A1 Adenosine receptor	NF- κ B2	Cell cycle	Oxidative stress	α -Fetoprotein
IFN- γ	Amiloride-sensitive sodium channel	c-Rel	Cyclin D1 ¹²⁸	XOD	AMH
IL-1 β ^{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	I κ B α	Cyclin D3	iNOS	Biglycan
RANTES	BRL-1	c-fos	GADD 45 β		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	Gadd45 β
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	Metastasis		DT-diaphorase	K3 keratin
GCP-2 (CXCL6)	IL-1 receptor antagonist	ICAM-1	Acute phase response proteins	Gelatinase B	K6 keratin
Gro α	IL-2 receptor α -chain	VCAM-1	Angiotensinogen	GSTP1-1	Laminin B2 chain
Gro γ	Immunoglobulin γ 4	ELAM-1	β -defensin-2	Guanylyl cyclase α	mCGM3
Gro-1	Immunoglobulin C γ 1	E-selectin	C4b binding protein	H ⁺ -K ⁺ -ATPase a2	Mts1
ICOS	Immunoglobulin E heavy chain	Endoglin	Complement factor B	Heparanase	MUC-2
IL-1 receptor antagonist	Immunoglobulin κ light chain	Fibronectin	Complement factor C4	HO-1	Mx 1
IP-10	Invariant chain II	MadCAM-1	C-reactive protein	Hyaluronan synthase	Neutrophil gelatinase-associated lipocalin
KC	Lox-1	P-selectin	LPS binding protein	Iodothyronine deiodinase	NLF 1
BMP-2	Mdr 1	Tenascin-C	Pentraxin PTX3	Lysozyme	P11
CCL15	MHC class (HLA-B7)	DC-SIGN	SAA1 and SAA2	MKP-1	PAI-1
CCL22	MHC class I (H-2Kb)	MMP-3	Tissue factor-1	N-	Pax 8
CCL28	Neuropeptide YY1-receptor	MMP-9 ¹³⁹	Urokinase-type plasminogen activator	Acetylglucosaminyltransferase 1	PCBD
CCL5	NMDA receptor subunit NR-1	CXCR4 ¹⁵¹		NGAL	Perforin
CD40 ligand	NMDA receptor subunit	KAI1/CD82 ¹³⁷		NQO1	PGK
CINC-1		uPA ¹⁴²		PDE 7A1	PSA
CXCL 11		Antiapoptosis		PGES/L-PGDS	RICK
ENA-78 (CXCL5)		Bcl-2 ¹³⁴	Proteins involved in antigen presentation	Phospholipase A2	S100A6 (calcyclin)
Eotaxin		Bcl-x _L ¹³⁴	Tapasin		Spergen-1
Erythropoietin		Bfl1/A1 ¹³⁴			Syndecan-4
MCP-1/JE					TFPI-2

(Table 4. Contd....)

MIP-1 α , β	Nod 2	TRAF-1 ¹³⁴	LMP 2	Phospholipase C δ 1	Transferrin
MIP-2	Polymeric Ig receptor	TRAF-2 ¹³⁴	TAP 1	PIM-1	Urokin 16
MIP-3 α	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/ CD3 γ	Survivin ¹³⁵		Seprin 2A	Wilms' tumor suppressor gene
Stem cell factor	TNF-Receptor, p75/80	A20		SNARK	
Angiotensinogen	DR4 ¹⁶⁷	Bax	Kinases	TERT	
TCA3	DR5 ¹⁶⁷	Caspase-11	PI3K ¹⁴³	Transglutaminase	
TFF3		FAP-1	PIK3		
TSP-1	Early response genes	Fas-ligand	PKC δ		
TSP-2	p62	IEX-1L	MAP 4K1		
	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, TGF β -inducible early gene; NF- κ B, nuclear factor κ B; IRF, interferon regulatory factor; ICAM-1, intercellular adhesion molecule-1; VCAM-1, vascular cell adhesion molecule 1; ELAM, endothelial leukocyte adhesion 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; PI3K, 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, lipoxigenase; 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- κ B 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- κ B include B cells, T cells, granulocytes, macrophages, neuronal cells, and smooth muscle cells [57].

Although rare, there are systems in which NF- κ B 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- κ B 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- κ B on apoptosis may result from the different activation pathways of NF- κ B 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- κ B 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- κ B 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- κ B 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- κ B pathway through a RelB/p50 dimer. They also provided evidence of the emerging role of the NF- κ B signaling pathway in the inhibition of apoptosis. Thus,

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

2.8. NF- κ B 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- κ B [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- κ B elements within the MMP-9 gene [139]. Bond *et al.* [141], showed by using an adenovirus that overexpressed the inhibitory subunit I κ B α that NF- κ B 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 TNF α requires the induction of NF- κ B activity and the decay of its short-lived repressor protein I κ B α . 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- κ B binding site that directly mediates the induction of uPA expression by RelA. Treating pancreatic tumor cell lines with the NF- κ B 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- κ B [143]. Mahabeleshwar *et al.* [107] demonstrated that the activation of Syk, a protein-tyrosine kinase, suppressed cell motility and NF- κ B-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- κ B and thus prevent its activation of the genes involved in cancer progression.

2.9. NF- κ B 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 angiogenic factors has been shown to be regulated by NF- κ B activation [145].

NF- κ B 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- κ B expression was associated with VEGF expression and microvessel density in human colorectal cancer. Specifically, they detected the immunohistochemical expression of NF- κ B, VEGF, and CD34 in 10 paraffin-embedded specimens of normal colorectal mucosa and 52 paraffin-embedded colorectal adenocarcinomas obtained by surgery or endoscopy; NF- κ B and VEGF were significantly overexpressed and associated with increased microvessel density in the colorectal cancer specimens. These findings suggest that increased expression of NF- κ B contributes to tumor angiogenesis in colorectal cancer and that VEGF plays an important role in mediating the NF- κ B angiogenic pathway.

Highly metastatic melanoma cells were found to express high levels of constitutive NF- κ B activity that was suppressed by transfection with I κ B α M, and suppression of constitutive NF- κ B 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- κ B activity, downstream of TRAF6, was sufficient to inhibit LPS-induced endothelial sprouting [149]. Also, inhibition of NF- κ B activity blocked basic fibroblast growth factor-induced angiogenesis. These findings further underscore the role of NF- κ B activation in mediating angiogenesis.

2.10. NF- κ B 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- κ B activation [150] (Table 4).

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

κ B 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-1 α -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-1 α -mediated migration was observed for cancer cells that metastasized to the lungs. These results implicate NF- κ B in the migration and organ-specific homing of metastatic breast cancer cells. In addition, Fujioka *et al.* [152] showed that inhibiting constitutive NF- κ B activity by expressing I κ B α M 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- κ B activation in reducing the metastasis of cancer cells to other sites. Recently, Loercher *et al.* [153] further showed that NF- κ B is an important modulator of the altered gene expression profile and malignant phenotype in squamous cell carcinoma.

2.11. NF- κ B 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- κ B [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 PGE₂. Double-knockout mice for both NF- κ B1 and NF- κ B2 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- κ B in bone loss and development.

NF- κ B proteins transmit growth factor signals between the ectoderm and the underlying mesenchyme during embryonic limb formation. In one study, the interruption of NF- κ B 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- κ B nuclear translocation through p56lck in human osteoblast-like cells. This activation of NF- κ B 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- κ B 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- κ B, with implications for the pathogenesis and progression of osteosarcomas [159]. They also suggest the critical involvement of NF- κ B proteins in regulating the dynamics of bone development, thus raising new possibilities for the treatment of bone disorders.

3. SUPPRESSION OF NF- κ B INHIBITS TUMORIGENESIS

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

3.1. Tumor-Suppressor Genes Inhibit NF- κ B Activation

Whereas oncogenes can stimulate NF- κ B activation, several tumor-suppressor genes, including the phosphatase and tensin gene (PTEN) [160], CYLD [161-163], and p53 [164], inactivate NF- κ B. (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- κ B were both inhibited [160]. The ability of IL-1 to induce I κ B α degradation or the nuclear translocation of NF- κ B was, however, unaffected by MMAC/PTEN expression. Moreover, the IL-1-induced phosphorylation of p50 NF- κ B 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- κ B transcriptional activity. Specifically, PTEN did not block TNF-induced IKK activation, I κ B α degradation, p105 processing, p65 (RelA) nuclear translocation, or DNA binding of NF- κ B [165]. However, PTEN did inhibit NF- κ B-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- κ B 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- κ B. A loss of the deubiquitinating activity of CYLD correlates with tumorigenesis. CYLD inhibits activation of NF- κ B by the TNFR family members CD40, X-linked ectodermal dysplasia receptor (XEDAR), and ectodysplasin-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- κ B. The inhibition of NF- κ B activation by CYLD is mediated, at least in part, by the deubiquitination and

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

<p>Anti-inflammatory agents</p> <p>Aspirin⁵⁷ Indomethacin⁵⁷ Diclofenac⁵⁷ Naproxen⁵⁷ Dexamethasone⁵⁷ Phenylbutazone⁵⁷ Tamoxifen⁵⁷ Sulfasalazine Sulindac Ibuprofen⁵⁷ Leflunomide¹⁷³ Trichodion Acetaminophen Gabexate mesilate Glucosamine sulfate Petrosaspongiolide M Dexanabinol BSASM Cloricromene Rolipram Pranlukast</p> <p>Immunosuppressive agents</p> <p>Cyclosporin LF15-0195 15-Deoxyspergualin 6(5H)-Phenanthridinone and benzamide</p> <p>Chemopreventive agents</p> <p>Carboplatin Vesnarinone²³² Cycloprodigiosin (-)-Cycloepoxydon Camphothecin Gemcitabine Levamisole Epoxomicin</p> <p>Angiogenesis</p> <p>2-Methoxyestradiol TNP-470</p> <p>Cell proliferation</p> <p>6-Aminoquinazoline derivatives Raxofelast</p> <p>Virus</p> <p>K1L HIV-1 Vpu protein Canine distemper virus Hypochlorite Inhaled isobutyl nitrite Jesterone dimer KT-90 APC Losartin Calcitriol</p>	<p>Antioxidants</p> <p>ADT Apocynin Benidipine BHA Carvedilol Catechol derivatives DHEAS DMDTC DMSO Disulfiram Ebselen EPC-K1 21 Ethyl pyruvate Ethylene glycol tetraacetic acid Iron tetrakis Mn-SOD Melatonin NAL NDGA Orthophenanthroline Hydroquinone and tert-butyl hydroquinone Ref-1 Tempol Tepoxaline 2-Amino-3-cyano-4-aryl-6-(2-hydroxy-phenyl) N-Acetylcysteine</p> <p>Reactive oxygen species</p> <p>Nitric oxide</p> <p>Antibacterial agents</p> <p>Fosfomycin 5-Aminosalicylic acid Clarithromycin Rifampicin Roxithromycin</p> <p>Antidiabetes agents</p> <p>Adiponectin Glimepiride</p> <p>Tyrosine phosphatase inhibitors</p> <p>Diamides²⁴² Pervanadate²⁴² Phenylarsine oxide²⁴²</p> <p>Serine protease inhibitor</p> <p>Pefabloc</p> <p>Phosphatidylcholine-phospholipase</p> <p>C inhibitor</p> <p>D609 Hyperosmolarity Hypothermia IMD-0354</p>	<p>HIV-1 protease inhibitors</p> <p>Nelfinavir Ritonavir Saquinavir</p> <p>ACE inhibitor</p> <p>Quinadril</p> <p>COX-2 inhibitor</p> <p>Celecoxib^{216,218}</p> <p>Interferon inducible protein</p> <p>p202a</p> <p>Methanesulfonilide antiarthritis inhibitor</p> <p>T-614</p> <p>PTK inhibitor</p> <p>Herbimycin A²⁴⁴</p> <p>Farnesyl protein transferase inhibitor</p> <p>SCH66336²²⁰</p> <p>Protease inhibitor</p> <p>Nafamostat mesilate</p> <p>PPARγ ligand</p> <p>Pioglitazone</p> <p>Heat shock proteins</p> <p>HSP70 HSP72</p> <p>Statins</p> <p>Atorvastatin Erbstatin Fluvastatin Lovastatin</p> <p>Metals</p> <p>Chromium Cadmium Gold Lead Mercury Zinc Arsenic Titanium</p> <p>IκB-like protein</p> <p>Encoded by ASFV</p> <p>Acetylation inhibitor</p> <p>HDAC¹⁸⁷ Nucling ox-LDL and HNE OXPAPC</p>	<p>Others</p> <p>β-Amyloid protein β-Catenin γ-Glutamylcysteine synthetase²³⁴ 1,2,3,4,6-Penta-O-galloyl-β-D-glucose 1,2,4-Thiadiazolidine derivatives 15-Deoxy-prostaglandin J(2) 17-Allylamino-17-demethoxygeldanamycin 2-Acetylaminofluorene 3,4,5-Trimethoxy-4'-fluorochalcone 4-Hydroxy-2-nonenal¹⁸³ 5'-Methylthioadenosine Acrolein Adenosine²²⁶ AMP-activated protein kinase Amrinone Anandamide Angiotensin-1 ANP Antithrombin III APC0576 Arsenite²⁴³ Auranofin AvrA protein AZT Bcl-Abl²²⁸ Benfotiamine Biliverdin Biotinylated isopanepoxydone Bisphenol A BMT Bovine serum albumin BZLF1 C5a Carbon monoxide Cyclopentones CYL-19s and CYL-26z CYLD¹⁶¹⁻¹⁶³ Desloratadine Disulfiram Dobutamine DQ 65-79 DTD E3330 E-73 Ecbet sodium Hydroquinone RKIP Rocaglamide Saline Selenomethionine Siah2</p>
--	--	---	--

(Table 5. Contd....)

CaMKK	Intravenous immunoglobulin	p8	SiRNA ¹⁹¹
Camptothecin	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-I κ B α
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
Ethyl pyruvate	Mesalamine	Probiotics	Tranilast
Evans blue	Methotrexate ²³⁰	Prostaglandin 15-deoxy-Delta(12,14)-PGJ(2)	Triflusal
Fibrates	Monochloramine	15-deoxy-delta 12,14-prostaglandin J2	Triglyceride-rich lipoproteins
Fish oil feeding	Monomethylfumarate	Prostaglandin A1	Troglitazone
Flunixin meglumine	MTA	Prostaglandin E2	Tyrphostin
Flurbiprofen	Murr1	Prostaglandin E2	Ursodeoxycholic acid
Fox1j	MX781	PSK	Uteroglobin
G-120 3032	Myxoma Virus MNF	Psychosine	Vasoactive intestinal peptide
Gangliosides	N-(p-coumaroyl) serotonin	PTEN ¹⁶⁰	vIRF3
Gemfibrozil	NEM	PTX-B	YopJ
Ghrelin	Neurofibromatosis-2 protein	Pyrrhione	ZAS3 protein
GILZ	nicotinamide, 3-aminobenzamide	Pyrrolidinedithiocarbamic -acid	ZUD protein
Glucocorticoids	Nilvadipine	Quinazolines	
Hydroquinone	Nitrosoglutathione	Rapamycin	
HSCO	Nitrosylcobalamin mild hypothermia	Rebamipide	
Human breast milk	N-Octylcafeate	RelA peptides	
Hydrochloride	N-Oleoyldopamine	Retinoic acid receptor-related orphan receptor-alpha	
	NPM-ALK oncoprotein	Ribavirin	
	NRF	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-aminoethyl ether)-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-leuciny-leucynil-norleucynal; APNE, N-acetyl-DL-phenylalanine-b-naphthylester; BTEE, N-benzoyl L-tyrosine-ethylester; DCIC, 3,4-dichloroisocoumarin; LLM, N-acetyl-leuciny-leucynil-methional; TLCK, N-a-tosyl-L-lysine chloromethyl ketone; TPCK, N-a-tosyl-L-phenylalanine chloromethyl ketone; Z-LLL, carbobenzoxy-leuciny-leucynil-leucynal; Z-LlnV, carbobenzoxy-leuciny-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, Pyrrolidinedithiocarbamate; 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- κ B repression factor; OXPAPC, oxidized 1-palmitoyl-2-arachidonoyl-sn-glycero-3-phosphorylcholine; PEDF, pigment epithelium derived factor; PTX, Pentoxifylline; PBN, Phenyl-N-tert-butylnitron; PIAS1, protein inhibitor of activated 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-naphthoquinone; 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- κ B essential modulator (also known as IKK γ) 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- κ B 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- κ B activity, which suggests a therapeutic intervention strategy to restore growth control in patients with familial cylindromatosis.

Whereas NF- κ B provides a proliferative signal, p53 is a mediator of antiproliferation. A mechanism by which p53 regulates NF- κ B 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 I κ B family that functions as a transcriptional coactivator for p52 NF- κ B; 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- κ B 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- κ B 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- κ B Inhibits Tumorigenesis

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

3.3. Chemopreventive Agents Inhibit NF- κ B Activation

Because of the critical role of NF- κ B in proliferation, invasion, angiogenesis, and metastasis of tumors, there has been great interest in modulators of the NF- κ B signaling pathway. Several agents that have been described as natural

chemopreventive agents have also been found to be potent inhibitors of NF- κ B activation (Table 6). How these agents suppress NF- κ B activation is becoming increasingly apparent. For example, these inhibitors may block any one or more steps in the NF- κ B signaling pathway, such as the incoming signals that activate the NF- κ B signaling cascade, the translocation of NF- κ B 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- κ B 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- κ B activation [173].

3.4. Some Cytokines Block NF- κ B Activation

Several cytokines have also been found to suppress NF- κ B 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- κ B activation [178]. How these agents suppress NF- κ B activation varies. For instance, the nuclear translocation of the RelA subunit and the degradation of I κ B α 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- κ B and increased I κ B α mRNA expression in another study [179].

4. NF- κ B IS A POTENTIAL TARGET FOR DRUG DEVELOPMENT

NF- κ B 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- κ B, 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- κ B activation (Table 8).

4.1. Inhibitors of Proteasomes Block NF- κ B Activation

Proteasome inhibitors block the 26S proteasome, which is necessary to degrade the I κ B α inhibitory subunit after its phosphorylation and ubiquitination in the cytoplasm, and thus its subunit release from the NF- κ B 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- κ B Activation

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

SAC, garlic compound, DDC; Diethylthiocarbamate, 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, KIL; Vaccinia virus protein, Panduratin A from *Kaempferia pandurata*, Zingiberaceae, Phytic acid; inositol hexakisphosphate, Rocaglamides; Aglaia derivatives, Sanguinarine; pseudochelethrine, 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; Vigna subterranean, Bee venom; melittin, Blueberry and berry mix; Optiberry, Cancer bush; Sutherlandia frutescens, Onconase; Ranpirinase, 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, Labiate 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.nlm.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 peptide 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 (IC₅₀) in the nanomolar range [184]. SC-514 is another novel inhibitor of IKK, specifically inhibiting IKKβ, which has an IC₅₀ of approximately 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	
Polyubiquitination inhibitor	LLM	PKC inhibitor
RO106-9920	BTEE	RO31-8220
	MG115 ⁵⁷	
Proteasome inhibitors	MG120	MEK1/2 inhibitor
ALLnL ⁵⁷	APNE	U0126
PS-341 ^{57,218}	Ubiquitin Ligase Inhibitors	
Lactacystine, β-lactone ⁵⁷	Z-LLL	Protein kinase inhibitor
	Z-LLnV	A77 1726

IKK, IκB kinase; PI3K, phosphatidylinositol 3; PKC, protein kinase, protein kinase C, MEK, mitogen-activated protein kinase (MAPK) and extracellular signal-regulated 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 IC₅₀ of 4 μ M for IKK α and 0.3 μ M for IKK β [186] (Table 8). Burke *et al.* [186] proposed a binding model in which BMS-345541 binds to similar allosteric sites on IKK α and IKK β 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 TNF α 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- κ B 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 I κ B α and its subsequent export from the nucleus, and is another potential target for gene transfer. In one study, the expression of HDAC3 in TNF α -stimulated HeLa cells repressed both NF- κ B DNA binding and the levels of Rel A, with a corresponding increase in the level of inactive cytoplasmic I κ B α -NF- κ B complexes [187]. This repressive mechanism was shown to control the duration of NF- κ B activation and thus may be a potential weapon against constitutive NF- κ B activation.

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

The transfer of genes that encode inhibitory proteins is another strategy used to block the activation of NF- κ B. The most direct target is the I κ B gene. This transfer entails the modification of I κ B 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- κ B activity was a mutated, nondegradable I κ B α resistant to phosphorylation and degradation that could be delivered into intestinal epithelial cells by using an adenoviral vector (Ad5 I κ B). There was a strong inhibition of NF- κ B activity because the superrepressor kept the NF- κ B 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- κ B in transcriptional regulation of the inflammatory phenotype of intestinal epithelial cells. Along similar lines, a nonphosphorylatable form of I κ B α 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- κ B Activation

Antisense agents that inhibit the expression of a target gene in a sequence-specific manner may be used against NF- κ B 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 IKK α , IKK β , and the upstream regulatory kinase transforming growth factor β -activated kinase-1, both IKK α and IKK β were found to be important in activating the NF- κ B 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- κ B Activation

Another approach to inhibiting NF- κ B activation is to use peptides that cross the cell membrane and block the nuclear localization of the NF- κ B 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- κ B 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- κ B interaction, which is required for the translocation of NF- κ B into the nucleus [195]. Also, a novel peptide that selectively blocks the association of IKK γ with the rest of the IKK complex has been shown to inhibit NF- κ B activation in response to proinflammatory cytokines in mice and at the same time to preserve basal NF- κ B 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- κ B activation induced by RANKL [197].

NF- κ B 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- κ B (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- κ B 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- κ B activation induced by LPS, IL-1, okadaic acid, phorbol 12-myristate 13-acetate, H₂O₂, 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- κ B (I κ B α) or on I κ B α kinase activation, it blocked TNF-induced p65 phosphorylation and nuclear translocation. These peptides also suppressed the NF- κ B-regulated reporter gene expression induced by TNF, TNFR1, the TNFR-associated death domain, TNFR-associated factor-2, NF- κ B-inducing kinase, I κ B α kinase, and p65. Notably, the suppression of NF- κ B by PTD-p65-P1 enhanced the apoptosis induced by TNF and chemotherapeutic agents.

4.7. Antiinflammatory Agents Block NF- κ B Activation

Additionally, several anti-inflammatory agents have been identified that suppress NF- κ B 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- κ B by preventing the degradation of the NF- κ B inhibitor, I κ B, so that NF- κ B 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- κ B activation and NF- κ B-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- κ B. In fact, all of the compounds inhibited TNF-induced NF- κ B activation, but with highly variable efficacy. Specifically, the IC₅₀s 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 I κ B α kinase and suppressed I κ B α degradation and NF- κ B-regulated reporter gene expression. They also suppressed NF- κ B-regulated cyclooxygenase-2 and cyclin D1 protein expression in a dose-dependent manner. Furthermore, all compounds inhibited the proliferation of tumor cells, with IC₅₀s 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- κ B 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- κ B is still incomplete. However, targeting NF- κ B seems central to designing an effective therapy for cancer.

4.8. NF- κ B can be Suppressed by Chemical Modifications

Various proteins that constitute the NF- κ B machinery contain a highly reactive cysteine residue on their surfaces (Table 9). Because modifications of this residue can also suppress NF- κ B 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- κ B Through the Critical Cysteine Residue

IKK α / β	Cys 179 ²⁴²
IKK β	Cys 59 ²⁴³
IKK γ	Cys 417 ²⁴⁴
p65	Cys 38 ²⁴⁵
p50	Cys 62 ²⁴⁶

5. NF- κ B ACTIVATION IS A DOUBLE-EDGED SWORD

Although NF- κ B activation in most cases is harmful [200], there is evidence that NF- κ B activation also has beneficial effects. As previously shown, NF- κ B activation regulates the expression of both antiapoptotic and proapoptotic genes. However, the downstream target genes regulated by NF- κ B in the initiation of proapoptotic signaling are unclear. Perfettini *et al.* [201] found that NF- κ B and p53 are the dominant apoptosis-inducing transcription factors elicited by the HIV-1 envelope. Additionally, Ravi *et al.* [202] found that NF- κ B induces expression of death receptor 4 (DR4) and DR5 and that, conversely, a transdominant mutant of the inhibitory protein I κ B α or a transactivation-deficient mutant of c-Rel reduces expression of either DR. Whereas NF- κ B promotes DR expression, cytokine-mediated activation of the RelA subunit of NF- κ B also increases expression of the apoptosis inhibitor Bcl-x_L and protects cells from the TNF-related apoptosis-inducing ligand (TRAIL). Inhibition of NF- κ B 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- κ B as a mediator and inhibitor of cell death during immune and stress responses. Fujioka *et al.* [203] showed that NF- κ B 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- κ B activity. They identified a κ B binding site in the promoter of Plk3 and showed that this site is directly involved in Plk3 induction by the RelA-NF- κ B 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- κ B-regulated gene that induces apoptosis in both p53-dependent and -independent signaling pathways, thus suggesting a possible mechanism for RelA-NF- κ B-regulated proapoptotic responses.

DR5 (TRAIL-R2) is a proapoptotic protein considered a potential target for cancer therapy; its expression is mediated by NF- κ B. Shetty *et al.* [205] found that NF- κ B 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- κ B 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- κ B 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- κ B-mediated DR5 expression and could explain the differential roles NF- κ B plays in regulating apoptosis.

It is known that inactivation of apoptotic pathways is a common event in cancer and that p53 and NF- κ B are transcription factors that regulate apoptosis during tumorigenesis. Although NF- κ B is generally considered a suppressor of cell death, Ryan *et al.* [206] showed that NF- κ B can contribute to p53-induced death. Specifically, they found that whereas the loss of NF- κ B 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- κ B, 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- κ B 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- κ B activation by an I κ B kinase-independent mechanism involving the phosphorylation of p65 by ribosomal S6 kinase 1.

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

CONCLUSIONS

Overall, the cited studies have clearly shown that NF- κ B plays a critical role in tumorigenesis. Similarly, the suppression of NF- κ B can inhibit various steps in the tumorigenic process. Thus, the design of NF- κ B inhibitors that are pharmacologically safe will be critical for the treatment of cancer. Because of the multiple agents that activate NF- κ B through diverse pathways, it is unlikely that any single inhibitor of NF- κ B could be effective against all tumors. Our efforts, therefore, should be directed toward developing NF- κ B 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.

ACKNOWLEDGEMENT

We would like to thank for Ellen M. McDonald a careful review of the manuscript. Supported by a grant from the Clayton Foundation for Research (to BBA), Department of Defense US Army Breast Cancer Research Program grant BC010610 (to BBA), a P01 grant (CA-91844) from the National Institutes of Health on lung chemoprevention (to BBA), and a P50 Head and Neck SPORE grant from the National Institutes of Health (to BBA).

ABBREVIATIONS

NF- κ B	=	Nuclear factor- κ B
IKK	=	I κ B kinasen
TNF	=	Tumor necrosis factor
IL-1	=	Interleukin-1
LPS	=	Lipopolysaccharide
TNFR	=	TNF receptor
TRAF	=	TNF receptor-associated factor
NIK	=	NF- κ B inducing kinase
NSCLC	=	Non-small cell lung carcinoma

MEF	=	Mouse embryo fibroblasts
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
PIK3	=	Polo-like kinase 3
siRNA	=	Small interfering RNA
DR	=	Death receptor
PTD	=	Protein transduction domain

REFERENCES

- Sen R, Baltimore D. Inducibility of kappa immunoglobulin enhancer-binding protein NF-kappa B by a posttranslational mechanism. *Cell* 1986; 47: 921-8.
- Gilmore TD. The Rel/NF-kappaB signal transduction pathway: introduction. *Oncogene* 1999; 18: 6842-4.
- 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.
- Karin M. How NF-kappaB is activated: the role of the IkappaB kinase (IKK) complex. *Oncogene* 1999; 18: 6867-74.
- 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.
- 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.
- Mayo MW, Baldwin AS. The transcription factor NF-kappaB: control of oncogenesis and cancer therapy resistance. *Biochim Biophys Acta* 2000; 1470: M55-62.
- Pahl HL. Activators and target genes of Rel/NF-kappaB transcription factors. *Oncogene* 1999; 18: 6853-66.
- Wang CY, Mayo MW, Baldwin AS Jr. TNF- and cancer therapy-induced apoptosis: potentiation by inhibition of NF-kappaB. *Science* 1996; 274: 784-7.
- 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.
- 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.
- Digicaylioglu M, Lipton SA. Erythropoietin-mediated neuroprotection involves cross-talk between Jak2 and NF-kappaB signalling cascades. *Nature* 2001; 412: 641-7.
- 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.
- 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.
- 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.
- Peters RT, Liao SM, Maniatis T. IKKepsilon is part of a novel PMA-inducible IkappaB kinase complex. *Mol Cell* 2000; 5: 513-22.
- 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.
- 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.
- 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.
- 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.
- 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.
- 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.
- 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.
- 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.
- 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.
- 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.
- McAllister-Lucas LM, Inohara N, Lucas PC, *et al.* Bim1, a MAGUK family member linking protein kinase C activation to Bcl10-mediated NF-kappaB induction. *J Biol Chem* 2001; 276: 30589-97.
- Leitges M, Schmedt C, Guinamard R, *et al.* Immunodeficiency in protein kinase cbeta-deficient mice. *Science* 1996; 273: 788-91.
- 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.
- 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.
- 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.
- 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.
- 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.
- 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.
- Zhao Q, Lee FS. Mitogen-activated protein kinase/ERK kinase 2 and 3 activate nuclear factor-kappaB through IkappaB kinase-alpha and IkappaB kinase-beta. *J Biol Chem* 1999; 274: 8355-8.
- 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.
- Tojima Y, Fujimoto A, Delhase M, *et al.* NAK is an IkappaB kinase-activating kinase. *Nature* 2000; 404: 778-82.
- 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- κ B-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 NF κ B and AP-1 activation. *J Biol Chem* 1997; 272: 29167-73.
- [41] Ozes ON, Mayo LD, Gustin JA, *et al.* NF- κ B 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 I κ B 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 I κ B kinases (IKK- α / β) and IKK- β 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- β)-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- κ 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 NF κ 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 I κ B kinase and nuclear factor κ B in response to B cell receptor engagement. *J Exp Med* 2000; 191: 1745-54.
- [48] Zhong H, Voll RE, Ghosh S. Phosphorylation of NF- κ 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- κ B is regulated by the I κ B-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- κ B 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- κ 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 κ B alpha activates NF κ 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- κ B1 precursor protein and the activation of NF- κ B. *Cell* 1994; 78: 773-85.
- [56] Parcellier A, Schmitt E, Gurbuxani S, *et al.* HSP27 is a ubiquitin-binding protein involved in I- κ B α proteasomal degradation. *Mol Cell Biol* 2003; 23: 5790-802.
- [57] Garg A, Aggarwal BB. Nuclear transcription factor- κ B 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- κ B 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- κ 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- κ 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- κ 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 κ B α . *Science* 1995; 268: 1619-21.
- [68] Luque I, Gelinac C. Rel/NF- κ B and I κ 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- κ 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- κ Bp50, NF- κ Bp65, and I κ B α by fluorescence *in situ* hybridization. *Oncogene* 1993; 8: 191-3.
- [73] Wang CY, Guttridge DC, Mayo MW, *et al.* NF- κ B 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 NF κ Bp65 protein expression. *Oncogene* 1997; 15: 1987-94.
- [75] Trecca D, Guerrini L, Fracchiolla NS, *et al.* Identification of a tumor-associated mutant form of the NF- κ B RelA gene with reduced DNA-binding and transactivating activities. *Oncogene* 1997; 14: 791-9.
- [76] Baldwin AS Jr. The NF- κ B and I κ 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 I κ B α gene in Hodgkin's disease suggest a tumour suppressor role for I κ B α . *Oncogene* 1999; 18: 3063-70.
- [78] Beauparlant P, Kwan I, Bitar R, *et al.* Disruption of I κ B α 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- κ B. *Science* 1992; 258: 1792-5.
- [80] Ferrier R, Nougarede R, Doucet S, *et al.* Physical interaction of the bHLH LYL1 protein and NF- κ B1 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- κ B during progression of breast cancer to hormone-independent growth. *Mol Cell Biol* 1997; 17: 3629-39.
- [83] Rayet B, Gelinac C. Aberrant rel/nf κ B genes and activity in human cancer. *Oncogene* 1999; 18: 6938-47.
- [84] Nair A, Venkatraman M, Maliekal TT, *et al.* NF- κ B 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 metalloproteinase 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, Pothoff 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 tumor promotion via a PKC alpha- and AP-1-dependent pathway. *Oncogene* 2002; 21: 4728-38.
- [99] Hehlhans 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, Droege 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 retinoblastoma 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. Activation 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 MDM2/PTEN inhibits cytokine-induced NFkappaB activation without interfering with the Ikbeta degradation pathway. *J Biol Chem* 2001; 276: 11402-8.
- [161] Trompouki E, Hatzivassiliou E, Tschirritzis 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 S, Perkins 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 metalloproteinase-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-Hydroxynonenol prevents NF-kappaB activation and tumor necrosis factor expression by inhibiting I kappaB phosphorylation and subsequent proteolysis. *J Biol Chem* 1999; 274: 11611-8.
- [183] Ji C, Kozak KR, Marnett LJ. I kappaB 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 I kappaBalpha arrest osteoclastogenesis. *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 I kappaB 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 I kappaB 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, Scلاب 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 I kappaB 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 I kappaBalpha 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 I kappaBalpha 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 metalloproteinase-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.* Picetannol 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.