Activation of NF-κB Is Involved in the Survival of Osteoclasts Promoted by Interleukin-1*

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We previously reported that interleukin-1 (IL-1) promoted the survival of murine osteoclast-like cells (OCLs) formed in vitro and activated a transcription factor, NF-κB, of OCLs. The present study examined whether the activation of NF-κB is directly involved in the survival of OCLs promoted by IL-1. The expression of IL-1 type I receptor mRNA in OCLs was detected by the polymerase chain reaction amplification of reverse-transcribed mRNA. An electrophoretic mobility shift assay showed that IL-1 transiently activated NF-κB in the nuclei of the OCLs, and the maximal activation occurred at 30 min. The degradation of IκBα coincided with the activation of NF-κB in the OCLs. The immunocytochemical study revealed that p65, a subunit of NF-κB, was translocated from the cytoplasm into almost all of the nuclei of the OCLs within 30 min after IL-1 stimulation. The purified OCLs spontaneously died via apoptosis, and IL-1 promoted the survival of OCLs by preventing their apoptosis. The pretreatment of purified OCLs with proteasome inhibitors suppressed the IL-1-induced activation of NF-κB and prevented the survival of OCLs supported by IL-1. The pretreatment of OCLs with proteasome inhibitors or antisense oligodeoxynucleotides to p65 and p50 of NF-κB prevented the IL-1-induced survival of OCLs supported by IL-1. These results indicate that IL-1 promotes the survival of osteoclasts through the activation of NF-κB.

Osteoclasts are multinucleated giant cells responsible for bone resorption (1–3). Osteoclastic bone resorption consists of several important processes: the development of osteoclasts from hematopoietic progenitor cells, the attachment of osteoclasts to the bone surface, the formation of a ruffled border and clear zone, and the secretion of acids and lysosomal enzymes into the space beneath the ruffled border (2, 4). Osteoclasts are terminally differentiated cells with a limited life span. Although osteoclasts are believed to die soon after they play their role in bone (bone resorption), recent findings suggest that the survival of osteoclasts is tightly regulated by several factors (5–9). Two cytokines, macrophage colony stimulating factor (M-CSF), also called CSF-1 (5, 6) and interleukin 1 (IL-1) (6), and a calcium-regulating hormone, calcitonin (7), stimulate the survival of osteoclasts. In contrast, estrogen (8) and transforming growth factor-β (TGF-β) (9) induce the apoptosis of osteoclasts. However, it is not yet known how the apoptosis and survival of osteoclasts is regulated by those factors.

Nuclear factor κB (NF-κB) is a ubiquitous transcription factor that regulates the expression of many genes involved in immune and inflammatory responses (10, 11). Conventional NF-κB is a heterodimer that consists of p50 and p55 subunits. The amino acid sequences of both subunits of NF-κB show a high homology to the Rel family, which includes c-Rel, Rel B, and p52, and they are now categorized as the NF-κB/Rel family (10, 11). The activity of NF-κB is strictly regulated by an inhibitor, IκBα, that forms a complex with NF-κB and keeps NF-κB in the cytoplasm (10, 11). When cells receive signals that activate NF-κB, IκBαs are phosphorylated and degraded through a ubiquitin/proteasome pathway. Multiple ubiquitin molecules attach to the phosphorylated IκBαs, and then ubiquitinated IκBαs are degraded by 26 S proteasome, an organella of intracellular protease complexes (11–14). The degradation of IκBαs triggers the translocation of NF-κB from the cytoplasm into the nucleus. Thus, proteasome is believed to be a key enzyme which is involved in NF-κB activation (11–14).

Recently several lines of evidence have reported that activation of NF-κB is involved in cell survival (15–19) besides in immune responses and inflammation. We have also reported that IL-1 stimulates the survival of osteoclasts (6) and activates NF-κB in osteoclasts (20), separately. Therefore, in this report, we examined whether the effect of IL-1 on osteoclast survival is mediated by the activation of NF-κB. IL-1 transiently induced an activation of NF-κB in osteoclast-like cells (OCLs), which was concomitant with the degradation of IκBα. The OCLs spontaneously died via apoptosis, which was markedly blocked by the addition of IL-1. The pretreatment of OCLs with either proteasome inhibitors or antisense oligodeoxynucleotides to p65 and p50 prevented the IL-1-induced survival of OCLs. These results indicate that IL-1 promotes the survival of osteoclasts by preventing apoptosis through the activation of NF-κB.

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1 The abbreviations used are: M-CSF, macrophage colony stimulating factor; TRAP, tartrate-resistant acid phosphatase; NF-κB, nuclear factor κB; IL, interleukin; ALLN, aldehydes N-acetyl-leucinyl-leucinyl-norleucinal-H; ALLM, N-acetyl-leucinyl-leucinyl-methional; ZLLal, carbobenzoxyl-leucinyl-leucinyl-leucinal-H; ZLLal, carbobenzoxyl-leucinyl-leucinal-H; OCL, osteoclast-like cell; α-MEM, α-minimal essential medium; FBS, fetal bovine serum; RT-PCR, reverse-transcribed polymerase chain reaction; PBS, phosphate-buffered saline; CTR, calcitonin receptor; GAPDH, glyceraldehyde-3-phosphate dehydrogenase; FITC, fluorescein isothiocyanate; S-ODN, synthetic phosphorothioate oligodeoxynucleotide; DOTAP, N-[1-(2,3-dioleolxyloxy)propyl]-N,N,N-trimethyl-ammonium salts; EMSA, electrophoretic mobility shift assay; TNFα, tumor necrosis factor α; NIK, NF-κB-inducing kinase.

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Experimental Procedures

Antibodies and Chemicals—Anti-human p65 (sc-109) and anti-human IκBα rabbit polyclonal antibodies were purchased from Santa Cruz Biotechnology (Santa Cruz, CA) and New England BioLabs (Lake Placid, NY), respectively. The peptide aldehydes (Boc-Lys(Acm)-Nle-Nle-Acm-OH) and (Boc-Lys(Acm)-cyclo(Acm)-Nle-OH) and (Boc-Lys(Acm)-cyclo(Acm)-Nle-OH) were synthesized by Sigma. Recombinant human interleukin 1 (rhuIL-1) and murine IL-1 receptor antagonist (IL-1ra) were obtained from R&D Systems (Minneapolis, MN).

Culture of Osteoclast-like Cells—Osteoblasts obtained from the calvaria of newborn mice and bone marrow cells obtained from the tibiae of newborn mice were co-cultured in α-minimal essential medium (α-MEM) (Life Technologies, Inc., Grand Island, NY) containing 10% fetal bovine serum (FBS), 1×25-hydroxyvitamin D₃ (1α,25(OH)₂D₃), 10⁻⁶ M prostaglandin E₂ (PGE₂) in 100-mm-diameter dishes coated with collagen gels (Nitta Gelatin Co., Osaka, Japan). OCLs were formed within 6 days in culture and were removed from the dishes by treatment with 0.2% collagenase (Wako Pure Chemical Co.). The purity of OCLs in this fraction (crude OCL preparation) was about 5%. To further purify the OCLs, the crude OCL preparation was replated on culture dishes. After an 8-h culture, osteoblasts were removed with phosphate-buffered saline (PBS) containing 0.001% Pronase E (Calbiochem, La Jolla, CA) and 0.02% EDTA for 10 min at 37 °C according to the method described previously (20).

Polymerase Chain Reaction Amplification of Reverse-transcribed mRNA—Total RNA from purified OCLs, crude OCLs and primary osteoblasts was extracted twice with phenol/chloroform/isoamyl alcohol and once with chloroform and precipitated in ethanol with 150 mM CH₃COONa, pH 5.2. The DNA was then dissolved in TE buffer (10 mM Tris-HCl, 1 mM EDTA, pH 8.0) and treated with 20 μg/ml RNase A. The procedures for DNA extraction and precipitation were repeated. Two μg of DNA was separated by electrophoresis on a 1% agarose gel and visualized by ethidium bromide staining with ultraviolet light illumination.

Antisense Oligodeoxynucleotides and Cell Cultures—Synthetic phosphorothioate oligodeoxynucleotides (S-ODN) that include the ATG initiation codon of the cDNA for mouse p50 or p65 were used for the sense and antisense experiments (see Table II). Crude OCLs were incubated in the presence of 5 μM antisense or sense S-ODN/cationic liposomes (DOTAP, Boehringer Mannheim, Mannheim, Germany) as the S-ODN carrier in α-MEM. They were then cultured for 2 h, 5% FBS was added, and the OCLs were further cultured for 8 h. After FBS were added, they were incubated in the presence of the antisense or sense S-ODN/DOTAP in α-MEM. They were then cultured for 2 h, and 5% FBS was added. After incubation for 6 h, OCLs were treated with IL-1, and the incubation was continued for an additional 15 h. The expression of p50 or p65 mRNA was detected by RT-PCR using gene-specific PCR primers specific for IL-1RI, CTR (a marker of osteoclasts), and osteocalcin. The p50 primers (1) 5'-TCGAGACTGAGCCTGTGG-3' and (2) 5'-CCCTGCGTTGGATTTCGTGACT-3' define an amplicon of 604 base pairs. The p65 primers (1) 5'-GGGAAGGAGGAGCTGAGCACGCA-3' and (2) 5'-GATTAGTGGTCGAGGAGATGACT-3' define an amplicon of 604 base pairs. The p65 primers (1) 5'-GGGAAGGAGGAGCTGAGCACGCA-3' and (2) 5'-GATTAGTGGTCGAGGAGATGACT-3' define an amplicon of 604 base pairs.

Survival of Osteoclasts by NF-κB Activation—The survival rate was measured as reported previously (6, 25). After OCLs were purified, some of the cultures were subjected to tartrate-resistant acid phosphatase (TRAP) staining. TRAP-positive multinucleated cells were counted as living OCLs. Other cultures were further incubated for indicated times in the presence or absence of IL-1. After incubation, the remaining OCLs were counted.

Results

Expression of IL-1 Type I Receptor mRNA by OCLs—We first examined whether OCLs express IL-1RI mRNA. Total RNA isolated from the cell preparations of purified OCLs, crude OCLs, and primary osteoblasts was analyzed by RT-PCR using primers specific for IL-1RI, CTR (a marker of osteoclasts), and osteocalcin (a marker of osteoblasts) (Table I). CTR mRNA was detected in the purified OCL and crude OCL preparations (Fig. IA, 1st middle panel, lanes 1–4), whereas osteocalcin mRNA was detected in the crude OCL and osteoblast preparations.
IL-1 activates NF-κB of OCLs induced by IL-1 (Fig. 1). This result suggests that IL-1 activates NF-κB of OCLs through IL-1RI expressed by OCLs—thereby activating NF-κB in OCLs within 30 min (Fig. 1B). The gene encoding IκBα is one of the target genes of NF-κB (10, 11). When purified OCLs were treated with actinomycin D (an inhibitor of mRNA synthesis) and incubated with IL-1, the activation of NF-κB was first detected within 5 min, attained a maximal level at 30 min, and declined thereafter. IκBα rapidly disappeared upon IL-1 stimulation and reappeared after 30 min (Fig. 2A). The gene encoding IκBα is one of the target genes of NF-κB (10, 11). When purified OCLs were treated with actinomycin D (an inhibitor of mRNA synthesis) and incubated with IL-1, the activation of NF-κB was first detected within 5 min, attained a maximal level at 30 min, and declined thereafter. IκBα rapidly disappeared upon IL-1 stimulation and reappeared after 30 min (Fig. 2A). The gene encoding IκBα is one of the target genes of NF-κB (10, 11). When purified OCLs were treated with actinomycin D (an inhibitor of mRNA synthesis) and incubated with IL-1, the activation of NF-κB was first detected within 5 min, attained a maximal level at 30 min, and declined thereafter. IκBα rapidly disappeared upon IL-1 stimulation and reappeared after 30 min (Fig. 2A).

To clarify whether IL-1 activates NF-κB of OCLs through their own IL-1RI, the effect of murine IL-1 receptor antagonist (IL-1ra) on the IL-1-induced activation of NF-κB was examined in the purified OCL preparations. The addition of IL-1 markedly activated NF-κB in OCLs within 30 min (Fig. 1B). The pretreatment of OCLs with IL-1ra suppressed NF-κB activity of OCLs induced by IL-1 (Fig. 1B). This result suggests that IL-1 activates NF-κB of OCLs through IL-1RI expressed by OCLs.
IL-1 stimulation, p65 was distributed throughout the cytoplasm (Fig. 3A) and especially around the nuclei of OCLs, which were detected by staining with Hoechst 33342 (Fig. 3B). When purified OCLs were treated with IL-1 for 15 min (Fig. 3, C and D), p65 was translocated into some nuclei in OCLs, but the remaining nuclei of OCLs showed no accumulation of p65 at this time point. However, p65 was detected in most of the nuclei of OCLs after IL-1 stimulation for 30 min (Fig. 3, E and F). The immunoreactivity of p65 completely disappeared from the nuclei of the OCLs after stimulation for 90 min (Fig. 3, G and H). When nonimmune immunoglobulins were used as the first antibody, no specific immunolabeling was detected in the OCLs (data not shown). Thus, the immunocytochemical findings were well consistent with the findings by electrophoretic mobility shift assay (EMSA) for NF-κB in OCLs treated with IL-1.

Proteasome Inhibitors Block the Activation of NF-κB and the Survival of OCLs Promoted by IL-1—Proteasome inhibitors were used to explore whether the activation of NF-κB is directly involved in the IL-1-induced survival of OCLs. The peptide aldehydes ALLN (calpain inhibitor I) and ZLLLal inhibit the proteolytic activity of proteasome (12). These inhibitors also suppress the protease activity of cathepsin B and calpain. Therefore, the structurally related compounds, ALLM (calpain inhibitor II) and ZLLLal, which inhibit cathepsin B and calpain but not proteasome (12), were used as the controls. The pretreatment of OCLs with ALLN or ZLLLal prior to IL-1 addition markedly decreased the NF-κB activity induced by IL-1, whereas the pretreatment with ALLM or ZLLLal did not (Fig. 4A). Neither ALLN nor ZLLLal affected the DNA-binding activity of another transcription factor, Oct-1, of the OCLs (Fig. 4A).

As reported previously (6), when purified OCLs were cultured for longer than 24 h, most died spontaneously, and the addition of IL-1 markedly supported the survival of the OCLs (Fig. 4B). The pretreatment of purified OCLs with ZLLLal, a proteasome inhibitor, completely prevented the IL-1-promoted survival of OCLs, but pretreatment with the control peptide, ZLLal, did not (Fig. 4B). Fragmentation of DNA was detected in the purified OCLs after culturing for 18 h (Fig. 4C, lane 2), suggesting that the death of OCLs was due to apoptosis. The treatment of OCLs with IL-1 inhibited ladder formation of DNA (Fig. 4C, lane 3). DNA fragmentation was also observed even in the presence of IL-1 in OCLs pretreated with ZLLLal (Fig. 4C, lane 5) but not with ZLLal (Fig. 4C, lane 4). These findings suggest that the activation of NF-κB by IL-1 contributes greatly to the prevention of the apoptosis of OCLs.

Antisense Oligodeoxynucleotides to NF-κB Prevent the IL-1-Induced Survival of OCLs—S-ODNs to p50 and p65 were also used to confirm the notion that the activation of NF-κB is involved in the survival of OCLs promoted by IL-1. When purified OCLs were treated with the antisense S-ODNs to p50 or p65, expression of the respective mRNA by OCLs was suppressed (Fig. 5A). Treatment of OCLs with the sense S-ODNs showed no effect on the expression of p50 and p65 mRNA (Fig. 5A). The viability of OCLs treated with sense S-ODNs to p50 and p65 went down slightly when compared with that treated with IL-1 alone (p < 0.05, Fig. 5B). This indicates that the transfection of S-ODNs partially affects the viability of OCLs at a basal level. The survival of OCLs was markedly reduced even in the presence of IL-1 by pretreatment with antisense S-ODNs to p50 and p65, compared with sense S-ODNs to p50 and p65 (Fig. 5, B and C).

DISCUSSION

The present study clearly indicates that the activation of NF-κB is involved in the survival of OCLs promoted by IL-1. The OCLs formed in our co-cultures expressed IL-1 type I receptors, and IL-1 directly activated NF-κB of OCLs through the binding to the IL-1 type I receptors. The degradation of IκBα appeared to trigger the activation of NF-κB in the OCLs because the level of IκBα in OCLs varied inversely with the activation of NF-κB. The immunocytochemical study also showed that the nuclear translocation of p65 was well correlated with the activation of NF-κB detected by the EMSA. Almost all of the nuclei of OCLs accumulated NF-κB within 30 min in response to IL-1 and discharged NF-κB after the stimulation for 90 min. This suggests that all of the nuclei of OCLs are functionally active and also that the function of multiple nuclei in osteoclasts is harmoniously regulated by stimuli from outside of the cell. The experiments using proteasome inhibitors and antisense S-ODNs to NF-κB showed that the IL-1-promoted survival of OCLs was mediated by the activation of...
NF-κB. These results indicate that activation of NF-κB directly prevents the spontaneously occurring apoptosis of OCLs.

Although IL-1 is a potent bone-resorbing factor in vivo (24) and in vitro (25), it is still not fully understood how IL-1 regulates osteoclast development and function. We previously reported that IL-1 stimulated OCL formation in murine bone marrow cultures by a mechanism involving prostaglandin E2 synthesis (26). Osteoblasts or marrow stromal cells have been proposed to play an essential role in OCL formation induced by osteotropic factors including IL-1 (3). Thomson et al. (27) showed that IL-1 stimulates the pit-forming activity of isolated rat osteoclasts through soluble factors secreted by osteoblasts. However, neither contaminating osteoblasts nor nonspecific esterase-positive macrophages were detected in our purified OCL preparation (20). Using an in situ hybridization technique, Xu et al. (28) showed the expression of mRNAs to IL-1 types I and II receptors by osteoclasts in normal bone tissues in mice and rats and inflammatory bone tissues in rats. Yu and Ferrier (29) also obtained evidence that osteoclasts are one of the target cells of IL-1. These findings together with the present report strongly suggest that IL-1 also acts on osteoclasts directly through their IL-1 receptors and regulates their functions without the help of other stromal cells.

M-CSF (5, 6, 23) and calcitonin (7) have been shown to prevent the apoptosis of osteoclasts. We also reported that like IL-1, M-CSF strongly supported the survival of OCLs (6, 23). It was shown that the tyrosine phosphorylation of IκBα represented a proteolysis-independent mechanism of NF-κB activation that directly coupled NF-κB to cellular tyrosine kinase (30). M-CSF, the receptors of which possess tyrosine kinase in the cytoplasmic domain, activated NF-κB of liver macrophages (Kupffer cells) of rats (31). However, M-CSF did not activate NF-κB of OCLs, and antisense S-ODNs to NF-κB failed to block the M-CSF-supported survival of OCLs in our culture system (data not shown). Furthermore, the pretreatment of actinomycin D suppressed the reappearance of IκBα protein in the purified OCLs treated with IL-1. These results indicated that the IL-1-induced NF-κB activation was mediated by a proteolysis-independent mechanism in OCLs. NF-κB of OCLs was not activated by the addition of calcitonin (20). In our preliminary experiments, IL-1 as well as M-CSF reduced the caspase activity in purified OCLs. These results suggest that signals other than the activation of NF-κB are involved in the survival of OCLs promoted by M-CSF and calcitonin.

Among the several factors examined, tumor necrosis factor α (TNFα, 10 ng/ml) also activated NF-κB of OCLs and prevented their spontaneous apoptosis (data not shown). It was recently reported that a novel serine/threonine protein kinase, NF-κB-inducing kinase (NIK), which binds to TRAF2 (TNF receptor associated factor 2) and activates NF-κB, is a necessary component of an NF-κB-activating cascade common to TNFα and IL-1 signalings (32). More recently, IκB kinase was identified which binds to NIK (33, 34). These findings suggest that OCLs also express functionally active TNFα receptors, and that serine/threonine protein kinases such as NIK and IκB kinase are commonly involved in the activation of NF-κB in OCLs treated with IL-1 and TNFα.

On the contrary to the present report, it has been shown that the activation of NF-κB induces the apoptosis in certain cells (22, 35). Abbadie et al. (35) reported that the overexpression of c-Rel induced apoptosis of avian bone marrow cells. Bessho et al. (22) also showed that the treatment of human leukemia cells and thymocytes with a protease inhibitor, pyrrolinedithiocarbamate (PDTC), which inhibited the activation of NF-κB, prevented their apoptosis. In addition, radiation or agents such as lipopolysaccharides and TNFα triggered the hydrolysis of membrane phospholipids to produce ceramide, which in turn activated NF-κB and induced apoptosis (11). It is not known at present if NF-κB is involved in promotion of apoptosis in certain cells and if also responsible for inhibition of apoptosis in other cells.

A novel function of NF-κB in apoptosis was recently found in...
Survival of Osteoclasts by NF-κB Activation

Fig. 5. Effects of antisense oligodeoxynucleotides to p50 and p65 subunits of NF-κB on the survival of OCLs. The nucleotide sequences of S-ODNs to murine p50 or p65 used in this study are shown in Table II. Crude OCL preparations plated on culture dishes were incubated for 10 h in the presence or absence of antisense S-ODNs (p50AS and p65AS) or sense S-ODNs (p50S and p65S) to p50 and p65. The cultures were then treated with Pronase to purify the OCLs. The purified OCLs were further incubated with or without the antisense or sense S-ODNs for 8 h. Total RNA was then isolated from some cultures to determine the expression of mRNAs to p50 and p65 (A). The expression of GAPDH mRNA was used as the control. The other cultures were further incubated with IL-1α (10 ng/ml) for an additional 15 h and then stained for TRAP. TRAP-positive multinucleated cells were counted as OCLs. Similar results were obtained from three independent sets of experiments. *, significantly different from the sense group; p < 0.01 (B). Panel C shows typical TRAP stainings of purified OCLs from the experiment described in panel B. Bar = 100 μm.

Rel A (p65) knockout mice (15, 37). Beg et al. (36) showed that a considerable apoptosis occurred in hepatocytes in Rel A-deficient embryos. They also reported that TNFα induced the apoptosis of fibroblasts and macrophages derived from Rel A knockout mice, and transfection of Rel A into the cells rescued them from TNFα-induced apoptosis (15). Wang et al. (16) and Antwerp et al. (17) independently demonstrated the anti-apoptotic role of NF-κB using cell lines stably transfected with the dominant negative IkBa. Liu et al. (18) also showed that three different responses to TNFα were mediated by TNFα receptor complex: the activation of Jun N-terminal kinase (JNK), the activation of NF-κB, and the induction of apoptosis. The activation of NF-κB protected the cells against TNFα-induced apoptosis. Ozaki et al. (19) also reported that NF-κB inhibitors pyrrolidine dithiocarbamate and chromomethyl ketone stimulated apoptosis of rabbit mature osteoclasts, which resulted in the inhibition of bone resorption. These findings together with the present report indicate that the NF-κB signaling cascade is closely involved in the prevention of apoptosis of cells. Cell death therefore appears to be tightly regulated through a balance between apoptosis-inducing and apoptosis-preventing signals. The signaling cascade mediated by NF-κB may be important in the survival of osteoclasts.

Osteoclasts reported to have a short half-life undergo apoptosis within a few days (3, 5, 6, 23). Studying the control mechanism of limited life span of osteoclasts may be important for understanding the regulation of bone remodeling not only in vitro but also in vivo. Treatment of OCLs with IL-1 for 1 h could support their survival, which was evaluated 20 h later (data not shown). This indicates that NF-κB stimulates gene expression of long term effective proteins on the survival of OCLs. Identification of such proteins in the OCLs must be important in future studies. IL-1 has been implicated in increased bone loss in pathological conditions such as rheumatoid arthritis, lymphoma, and osteoporosis (37). The IL-1-induced survival of osteoclasts may also play an important role in the bone resorption stimulated by these pathological conditions. Further studies are required to determine a more detailed mechanism of involvement of NF-κB in the survival of osteoclasts and the gene expression of proteins that exert anti-apoptotic effects promoted by IL-1.

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REFERENCES
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