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Forum Review

Redox Regulation of Fcγ Receptor-Mediated Phagocytosis: Implications for Host Defense and Tissue Injury

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ABSTRACT

Recent advances in our understanding of the mechanisms that regulate acute and chronic inflammatory responses have revealed a key role for reactive oxygen intermediates in modulating the activation of neutrophils. Opsonized microbes and immune complexes initiate the oxidative burst by the engagement of receptors for immunoglobulin G, termed Fcγ receptors. The regulation of phagocytic cell function by oxidant-sensitive signaling pathways optimizes host defense capabilities, but it also amplifies tissue damage. This review will focus on the cross-talk between Fcγ receptors and reactive oxygen intermediates at sites of inflammation and its role in microbial immunity. Antioxid. Redox Signal. 4, 85–95.

INTRODUCTION

IN MANY ACUTE AND CHRONIC INFLAMMATORY DISORDERS, important components of pathological processes are linked to the ability of phagocytes, in particular polymorphonuclear leukocytes (PMN), to produce reactive oxygen intermediates (ROI). The oxidative burst in PMN is initiated by the interaction of cell-surface receptors with specific ligands found on microbial targets or in the inflammatory milieu that elicit intracellular signaling pathways leading to biological responses. Fcγ receptors (FcγR) are receptors expressed on the surface of neutrophils that recognize the constant region of immunoglobulin G (IgG). Engagement of FcγR by opsonized microbes or immune complexes stimulates phagocytosis and generation of ROI. Although ROI have the potential to kill invading microbes, they also initiate a number of physiological responses, including cell activation, proliferation, and migration. A new mechanism to regulate microbicidal responses to optimize host defense is the amplification of FcγR signaling and function by ROI. The same effectors activated during antimicrobial responses may lead to inflammatory tissue damage in autoimmune disorders. By modulating the effector potential of FcγR on PMN, ROI regulate a broad program of cell functions relevant to host defense against microbes, inflammation, and autoimmunity.

ROI ENHANCE FcγR-MEDIATED FUNCTION IN PMN

The NADPH oxidase is a membrane-associated enzyme that generates a family of ROI (33). The NADPH oxidase is inactive in unstimulated PMN. PMN triggered via FcγR

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rapidly activate the enzyme system resulting in the generation of superoxide anion (O_2^-). Other ROI are generated by subsequent catalyzed or spontaneous reactions. For example, hydrogen peroxide (H_2O_2) forms from the spontaneous dismutation of O_2^-, but superoxide dismutase will accelerate this process (52).

ROI include highly reactive, diffusible molecules. Whereas at high concentrations ROI are toxic to cells, at lower concentrations ROI can serve as intra- or extracellular second messengers. Several lines of evidence support a role for oxidants in the amplification of neutrophil FcγR phagocytic function. Phorbol esters stimulate FcγR-mediated phagocytosis, an effect that is blocked by superoxide dismutase and catalase (23). Patients with chronic granulomatous disease (CGD), characterized by genetic defects in the NADPH oxidase system that result in markedly diminished generation of ROI, have served as clinical paradigms that establish the importance of oxidants in phagocyte function (52). PMN from such patients show impaired phorbol myristate acetate and cytokine-dependent amplification of FcγR-mediated internalization, emphasizing the possibility that increased FcγR responsiveness is mediated by products of the respiratory burst (23).

CROSS-LINKING FcγRIIIb AMPLIFIES FcγRIIA FUNCTION IN AN OXIDANT-DEPENDENT MANNER

Human PMN constitutively express two structurally distinct activating FcγR, FcγRIIa and FcγRIIIb (44) (Fig. 1). They can also be induced to express FcγRI by interferon-γ (IFN-γ) (15, 36). FcγRIIa, a transmembrane receptor, is the predominant phagocytic receptor in neutrophils. Allelic variants of human FcγRIIa profoundly influence phagocyte biologic activity. A histidine to arginine substitution at amino acid position 131 in the extracellular domain of FcγRIIa changes the ability to bind IgG2 and C reactive protein (CRP), and thereby alters effector responses to these ligands (7, 53, 58) (Fig. 1).

The second activating FcγR isoform, FcγRIIIb, is anchored to the plasma membrane via a C-terminus-linked glycosylphosphatidylinositol (GPI) moiety. With 10-fold greater expression than FcγRIIa, it may play a predominant role in PMN binding of immune complexes.

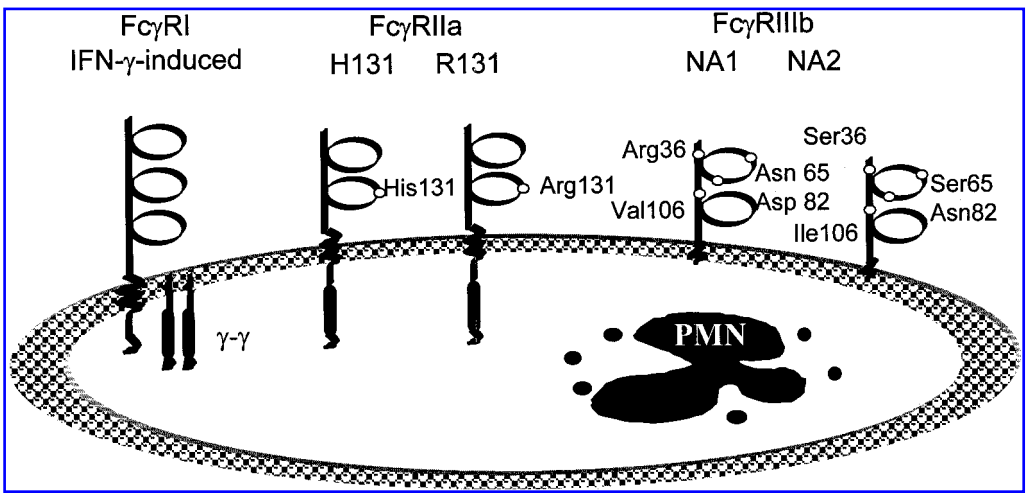

FIG. 1. Schematic representation of the human FcγR family members expressed in PMN. FcγRI is a multichain receptor, induced by IFN-γ, which associates with immunoreceptor tyrosine activation motif (ITAM; black cylinders)-bearing γ-chain dimers to mediate positive signaling. FcγRIIa is a single-chain receptor containing two ITAMs in its cytoplasmic tail. The two allelic variants of FcγRIIa are a consequence of a histidine-to-arginine (H131 to R131) substitution at position 131 in the extracellular domain. FcγRIIIb is a GPI-anchored receptor. The neutrophil antigen (NA) 1 and NA2 polymorphism of FcγRIIIb reflects four amino acid substitutions resulting in quantitative differences in oxidative burst and phagocytic function.

Because both FcγR isoforms are likely to be engaged by immune complexes, the questions of whether and how the GPI-anchored receptor may interact with FcγRIIa have been subject to debate. Although one view is that FcγRIIIb serves merely to enhance immune complex binding for presentation to FcγRIIa, clear evidence supports an active role for the GPI-anchored isoform in signaling and PMN activation. In the absence of FcγRIIa ligation, FcγRIIIb cross-linking induces a rise in the intracellular free Ca^{2+} concentration ($[Ca^{2+}]_i$) and triggers degranulation and the respiratory burst (25). FcγRIIIb-triggered $[Ca^{2+}]_i$ transients and O_2^- production are also inhibited by high concentrations of D-mannose or *N*-acetyl-D-glucosamine, each part of the conserved core structure of GPI anchors (50). It has been suggested that the signaling capacity of GPI-anchored proteins may derive from their ability to induce the formation of microdomains of defined composition within the plasma membrane (6).

Although FcγRIIIb is much less efficient than FcγRIIa in initiating phagocytosis, it can interact synergistically to amplify FcγRIIa-specific function (12). Cross-linking of FcγRIIIb increases the phagocytic activity of FcγRIIa that has been engaged independently and leads to FcγRIIa activation, in a similar manner to that induced by phorbol esters (23). Activation of FcγRIIa by FcγRIIIb is transferable by supernatants from activated cells and is blocked by inhibitors of reactive oxygen species and the H_2O_2–myeloperoxidase–chloride system. The increase in FcγRIIa-specific internalization induced by oxidants reflects both an increase in ligand binding by FcγRIIa and an increase in internalization efficiency of targets bound. Taken together, these studies show that cross-linking of FcγRIIIb, which leads to the generation of ROI, alters FcγRIIa avidity and efficiency (46).

ENHANCEMENT OF FcγRIIA FUNCTION IS SENSITIVE TO ALLELES OF FcγRIIIB

The capacity of FcγRIIIb to augment FcγRIIa function varies according to differences in the primary structure of FcγRIIIb. Two common allelic variants of FcγRIIIb have been characterized, and they differentially modulate PMN function (45). The allotypes, known as neutrophil antigen 1 (NA1) and NA2, differ by five nucleotides that result in substitutions of four amino acids in the first extracellular domain (34, 35) (Fig. 1). PMN from NA1 homozygous donors have a more robust FcγR-mediated phagocytic response than cells from NA2 donors, despite equivalent receptor density (3, 45). Because FcγRIIIb mediates phagocytosis poorly (1, 12), we predicted that it would influence internalization by modulating FcγRIIa function in an allele-sensitive fashion. Indeed, donors homozygous for the NA1 allele of FcγRIIIb showed greater activation of FcγRIIa following FcγRIIIb cross-linking than donors homozygous for the NA2 allele of FcγRIIIb (46). This altered phagocytic capacity appears to be due, at least in part, to the ability of the NA1 allele to elicit a quantitatively larger oxidative burst and degranulation response compared with the NA2 allele. These oxidant-mediated changes in FcγRIIa function provide another mechanism for receptors to collaborate in both an autocrine and paracrine fashion.

FcγR triggering also induces secretion of the contents of granules/vesicles. The contents of specific granules can be delivered to the extracellular environment through secretion or to the phagosome containing an ingested particle. Proteolysis by serine proteases leads to enhanced ligand binding to FcγRIIa, but the structural basis for this effect in unknown (11). Nonetheless, to amplify the effects of proteases on FcγRIIa, PMN use the H_2O_2–myeloperoxidase–chloride system to generate chlorinated oxidants, such as HOCl. Chlorinated oxidants activate protease zymogens and inactivate protease inhibitors, and therefore may represent a mechanism to enhance FcγRIIa function. Upon cross-linking FcγRIIIb, PMN bearing NA1 alleles have more potent generation of ROI and secretion of serine proteases, which may account for the greater amplification of FcγRIIa function by NA1 than NA2 alleles (46). This modulation of FcγR function by reactive oxidants and proteases occurs very rapidly, over a time frame of minutes, in contrast to cytokine-induced

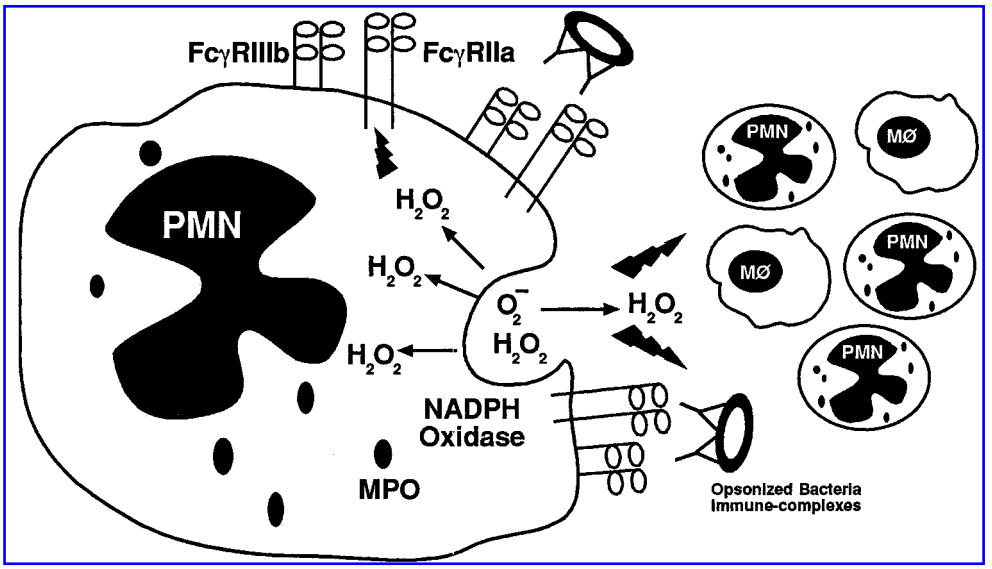

FIG. 2. Autocrine and paracrine effects of ROI generated by human PMN following FcγR triggering. At sites of inflammation, FcγRIIa and FcγRIIIb on PMN are co-clustered by opsonized bacteria or immune complexes, leading to the rapid activation of the NADPH oxidase system and generation of O_2^- and H_2O_2. Myeloperoxidase (MPO), an enzyme contained in neutrophil granules, amplifies the oxidative potential of H_2O_2 by generating cytotoxic chlorinated oxidants and other radical species. These highly diffusible molecules serve as intra- or extracellular second messengers that alter the signal transduction and amplify the effector potential of FcγR on both the same and adjacent phagocytic cells. MØ, macrophages.

changes in receptor expression that occur over hours or days. Through coordinated degranulation and generation of ROI, FcγR triggering can act in an autocrine or paracrine manner to rapidly activate other receptors (Fig. 2). FcγR isoforms cooperate through this oxidant-dependent mechanism to produce more efficient effector cell function.

MODULATION OF FcγR SIGNAL TRANSDUCTION: ROI ENHANCE TYROSINE PHOSPHORYLATION OF FcγRIIA AND SYK

PMN activation is initiated when FcγR are clustered at the cell surface by multivalent antigen–antibody complexes; monovalent ligand binding is insufficient to generate a signal. Stimulatory FcγR have no intrinsic enzymatic activity, but are associated with membrane anchored Src family kinases. Tyrosine phosphorylation is essential for FcγR-mediated responses. Endogenous generation of ROI or exposure to exogeneous H_2O_2 has been shown to induce phosphorylation of several intracellular proteins in human PMN (4, 16). Extracellular or intracellular ROI easily penetrate the plasma membrane and function as second messengers modulating signal transduction pathways. The deficiency in the capacity to generate ROI, such as that found in the PMN from patients with CGD, which results in limited accumulation in phosphoprotein after stimulation, underscores the role of ROI in signaling (18).

The importance of tyrosine phosphorylation in FcγR-mediated internalization, taken together with observations that oxidants influence phosphotyrosine accumulation, led us to examine systematically the influence of endogenously generated and exogenously added oxidants on the proximal events of FcγRIIa signaling. Upon receptor cross-linking, FcγRIIa immunoreceptor tyrosine-based activation motif (ITAM) motifs are phosphorylated on tyrosines by Src family protein kinases, and the phosphorylated ITAM functions as a scaffold to recruit and organize effector molecules (Fig. 3). In the presence of H_2O_2, there is accelerated and increased phosphorylation of the ITAM of FcγRIIa following

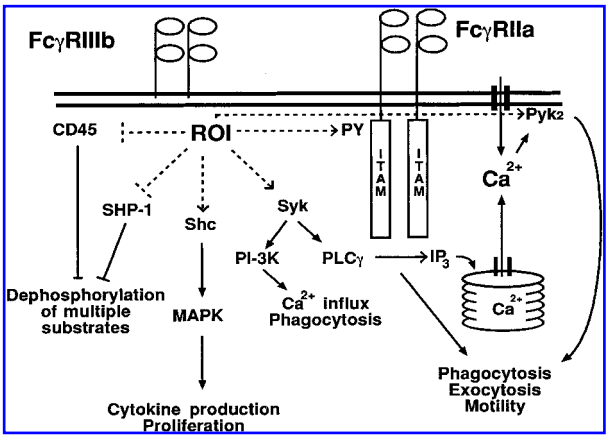

FIG. 3. ROI amplifies FcγR-mediated signaling pathways. FcγRIIa triggering in the presence of ROI results in enhanced phosphorylation (P) of the tyrosines (Y) in the ITAM motif of FcγRIIa, as well as increased phosphorylation and activation of the SH2-domain tyrosine kinase Syk, which activates phosphatidylinositol 3-kinase (PI-3K) and phospholipase Cγ (PLCγ). This amplifies a series of downstream events that lead to greater influx of Ca^{2+} from intra- and extracellular sources, increased activation of adapter proteins (Shc), and focal adhesion family members (Pyk2) and ultimately to cytoskeletal changes and transcriptional activation of cytokine genes. ROI inactivate CD45 and the SH2 domain-containing phosphatase SHP-1 phosphatase activity and thereby prevent the dephosphorylation and inactivation of many intermediates of this signaling cascade. MAPK, mitogen-activated protein kinase.

receptor triggering in PMN (41, 62). Endogenous generation of ROI initiated by FcγRIIIb triggering induced an even greater enhancement of FcγRIIa phosphorylation in PMN, which may be related to higher and more sustained intracellular oxidant levels (41). This enhanced phosphorylation of FcγRIIa ITAM may be a mechanism by which ROI enhance FcγRIIa-mediated phagocytosis.

Phosphorylation of FcγRIIa leads to the recruitment, phosphorylation, and activation of Syk, which then phosphorylates downstream signaling targets. Syk phosphorylation and activity correlate with the magnitude of FcγRIIa-mediated effector function (32, 49). We have shown that ROI increase the rate and magnitude of FcγRIIa-triggered phosphorylation of this critical kinase in PMN (41). Both exogenous H_2O_2 and endogenously generated oxidants amplify tyrosine phosphorylation of Syk. Inhibition of FcγRIIIb-stimulated Syk hyperphosphorylation in PMN in the presence of catalase emphasizes the role for oxidants as intracellular second messengers with the potential to modulate effector function. ROI also contribute to lectin-induced phosphorylation of Syk (43). Syk has been shown to be required for FcγR-mediated phagocytosis (9). Indeed, transfected cells expressing an FcγRIII-Syk chimera internalize particles that cross-link FcγRIII, indicating that Syk kinase is sufficient for initiating cytoskeletal coupling and phagocytosis (22), and alterations in Syk expression modify efficiency of phagocytosis (26, 32). These studies, taken together with our evidence for ROI-induced enhanced Syk phosphorylation, reveal a mechanism by which oxidants mediate synergism of FcγRIIa and FcγRIIIb in PMN.

ROI AMPLIFY AND ACCELERATE FcγRIIA-TRIGGERED TYROSINE PHOSPHORYLATION OF Shc AND Pyk2

Tyrosine kinases phosphorylate many intracellular substrates, including phospholipid kinases, phospholipases, adapter molecules, and cytoskeletal proteins (Fig. 3). Activation of phosphatidylinositol 3-kinase and phospholipase Cγ leads to the production of phosphoinositol messengers and a sustained increase in cytoplasmic Ca^{2+} (31). The activation of phospholipase Cγ by oxidative radical stress elevates $[Ca^{2+}]_i$ levels by influx from extracellular and intracellular sources (Fig. 3). Ca^{2+} signals are also directly initiated by H_2O_2 (48, 49). The transient rise in Ca^{2+} induced by oxidants may cause preactivation of intracellular signaling elements and allow for a more sustained Ca^{2+} flux following specific FcγR triggering, thereby contributing to the amplification of phagocytic function (13).

The adapter protein Shc is phosphorylated upon triggering through FcγRIIa (51). Shc provides a link between the membrane-localized receptor and downstream signaling pathways, such as Ras/Raf/mitogen-activated protein kinase, that lead to activation of transcription factors and induction of gene expression, and the family of focal adhesion kinases that are involved in the modulation of the cytoskeleton. We found that in the presence of exogenous H_2O_2 there is amplified and accelerated Shc

phosphorylation induced after FcγRIIa clustering. When PMN were stimulated to generate ROI through FcγRIIIb, the FcγRIIa-induced phosphotyrosine acumulation on Shc is also markedly increased (Fig. 4). This enhanced phosphorylation has the potential to amplify effector signaling or alter the threshold for effector function.

Cross-linking of FcγR leads to rapid and transient phosphorylation of focal adhesion kinase, a protein tyrosine kinase localized to focal adhesions, and paxillin, a cytoskeleton-associated substrate for tyrosine kinases. The proline-rich tyrosine kinase (Pyk2), another member of the focal adhesion kinase family, is also tyrosine-phosphorylated and activated following FcγR triggering in human PMN. Pyk2 provides the link between the cell-surface signals and the cytoskeleton, which is the essential framework for phagocytosis and cell migration (21). It is likely that Pyk2 phosphorylation and activation modulate phagocytic functions of cells, because Pyk2 constitutively binds to and phosphorylates paxillin. We have recent evidence that oxidants amplify Pyk2 phosphorylation (J. Gokhale and L. Pricop, unpublished results). PMN exposed to exogenous H_2O_2 or triggered through FcγRIIIb to generate endogenous ROI show an increase in the rate and magnitude of Pyk2 phosphorylation (Fig. 5). This pathway provides another means by which oxidants can increase phagocytic function. In addition, because Pyk2 co-localizes with vinculin and paxillin in podosomes and is crucial for the cytoskeletal reorganization required for cell motility, it is likely to influence the efficiency

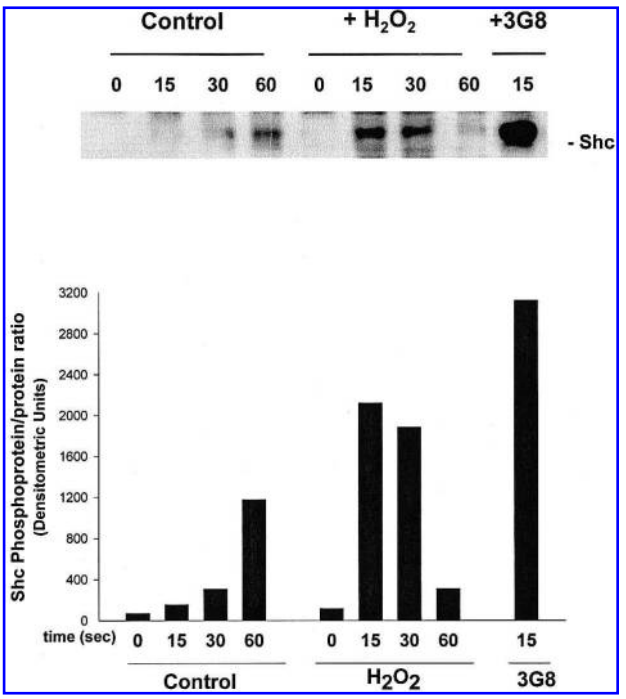

FIG. 4. Exogenous and endogenously generated oxidants accelerate and amplify FcγRIIa-stimulated phosphorylation of the adapter protein Shc in PMN. Freshly isolated PMN (2×10^7/lane) were pretreated with medium (lanes 1–4), H_2O_2 (500 μ*M*) (lanes 5–8), or F(ab′)$_2$ fragments of anti-FcγRIIIb monoclonal antibody (mAb; clone 3G8) (5 μg/ml) (lane 9) for 10 min at room temperature. Subsequently, cells were opsonized with Fab fragments of FcγRIIa mAb (clone IV.3) (5 μg/ml) and stimulated with goat anti-mouse F(ab′)$_2$ (30 μg/ml) for 15, 30, or 60 s at 37°C. Cells were lysed and proteins were immunoprecipitated with anti-Shc antibody, run on 10% sodium dodecyl sulfate–polyacrylamide gels, and immunoblotted with anti-phosphotyrosine mAb clone 4G10 (**top panel**). Phosphoprotein-to-protein ratios were determined by densitometric measurements and are expressed as arbitrary units (**bottom panel**).

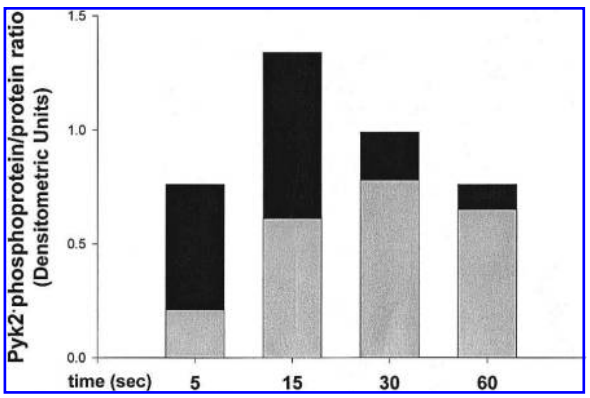

FIG. 5. Enhanced phosphorylation of Pyk2 in PMN treated with ROI. Freshly isolated PMN (2×10^7/lane) were pretreated with medium or H_2O_2 (500 μ*M*) for 10 min at room temperature. Subsequently, cells were treated with Fab fragments of FcγRIIa mAb (clone IV.3) (5 μg/ml) and stimulated with goat anti-mouse F(ab′)$_2$ (30 μg/ml) for 5, 15, 30, or 60 s at 37°C. Cells were lysed and proteins were immunoprecipitated with rabbit polyclonal anti-Pyk2 antibody (obtained from Dr. Jerome Groopman, Harvard Medical School), run on 10% sodium dodecyl sulfate–polyacrylamide gels, and immunoblotted with anti-phosphotyrosine mAb (clone 4G10) (top panel). Phosphoprotein-to-protein ratios were determined by densitometric measurements. Overlaid bars represent the intensity of Pyk2 phosphorylation (phosphoprotein-to-protein ratio) in the absence (gray bars) or in the presence of exogenous oxidants (black bars).

of recruitment of neutrophils to sites of inflammation.

POTENTIAL MECHANISMS OF ROI-INDUCED AMPLIFICATION OF FcγRIIa-SIGNAL TRANSDUCTION

The mechanism by which ROI increase total phosphotyrosine is not clear. It has been suggested that endogenous or exogenous oxidants can promote tyrosine phosphorylation by combined activation of kinases and inhibition of phosphatases (4, 16, 18, 63). The balance between protein tyrosine kinase and tyrosine phosphatase activity determines the magnitude and kinetics of phosphorylation of signaling elements, and thereby regulates effector cell activation. Tyrosine phosphatases may be inactivated by oxidants that target critical cysteine residues in their catalytic domains (19, 54, 57). As a consequence, constitutive autophosphorylation and stimulation of kinases, which are no longer offset by phosphatase activity, result in accumulation of phosphotyrosine. Indeed, CD45, a known inhibitor of FcγRIIa signaling in PMN, is susceptible to inactivation by oxidants (17, 18). The fact that CGD neutrophils have diminished inhibition of CD45 tyrosine phosphatase activity in response to activation of NADPH oxidase and show impaired PMA-induced amplification of FcγR function provides indirect support for this mechanism of oxidant-induced modulation of FcγR signaling (18, 23).

Neutrophil function is also regulated by intracellular src homology domain 2 (SH2)-containing protein tyrosine phosphatases 1 (SHP-1) and src homology inositol polyphosphate 5′-phosphatase (SHIP) (5, 24). Activation of SHP-1 in PMN has been shown to be inhibited by treatment with H_2O_2 (10). Given that SHP-1 deficiency results in abnormalities in neutrophil function, oxidative-induced inactivation of SHP-1 may be an important mechanism to regulate neutrophil activation (28, 61).

Another negative regulator of phagocyte activation is FcγRIIb, an inhibitory FcγR isoform. We have recently shown that FcγRIIb is expressed in human monocytes and neutrophils and that it is an important negative regulator of phagocyte activation (42). FcγRIIb is a single-chain low-affinity receptor with extracellular domains highly homologous to FcγRIIa and cytoplasmic domains containing an immunoreceptor tyrosine-based inhibitory motif (ITIM). Like FcγRIIa, the FcγRIIb intracellular tyrosine motif is phosphorylated by protein tyrosine kinases and is therefore subject to oxidant-induced modulation. The ITIMs recruit SH2-containing phosphatases upon phosphorylation. Although the protein tyrosine phosphatases SHP-1 and SHP-2 bind to FcγRIIb-phosphorylated ITIM motifs, the inositol polyphosphate 5′-phosphatase SHIP has been shown to be preferentially recruited to FcγRIIb and appears to play the predominant role in FcγRIIb-mediated inhibition. In contrast to SHP-1, a direct role for oxidants in the regulation of SHIP phosphatase activity has not been reported. However, recent reports suggest that conditions that favor hypo- or hyperphosphorylation of FcγRIIb might interfere with phosphatase recruitment (30). Although speculative, oxidant-mediated alteration of FcγRIIb phosphorylation has the potential to influence the responsiveness of neutrophils to immune complex-mediated inflammation.

ALLELIC VARIANTS OF FcγR: IMPLICATIONS FOR HOST DEFENSE

Allelic variants identified in two of the FcγR expressed on PMN, FcγRIIa and FcγRIIIb, profoundly influence phagocyte biologic activity. Single amino acid substitutions within the extracellular domains of stimulatory FcγR alter the ability of the receptor to bind IgG and have been associated with risk for and phenotype of autoimmune and infectious disease (Fig. 1).

The alleles of FcγRIIa, H131 and R131, differ substantially in their ability to bind human IgG2 (7, 58). H131 is the high-binding allele, R131 is low binding, whereas heterozygotes have intermediate function. Because IgG2 is a poor activator of the classical com-

plement pathway, FcγRIIa-H131 is essential for handling IgG2 immune complexes. The two common allelic variants of FcγRIIIb, NA1 and NA2, are associated with distinct neutrophil phenotypes. PMN from NA1 homozygous donors have more robust FcγR-mediated phagocytosis than those from NA2 donors, which is not due to a difference in FcγR binding, but likely to be related to the larger oxidative burst and degranulation mediated by NA1 alleles, resulting in increased FcγRIIa function (3, 45, 46).

FcγRIIa has substantial clinical importance for host defense against infection with encapsulated bacteria known to elicit IgG2 responses, such as *Neisseria meningitidis*, *Hemophilus influenzae,* and *Streptococcus pneumoniae* (2, 40, 47). There is an increased frequency of homozygosity of FcγRIIa-R131 among otherwise healthy children who suffer from recurrent respiratory tract infections or fulminant meningococcal sepsis. FcγRIIa-R131 has also been shown to be a risk factor for invasive pneumococcal infection in patients with systemic lupus erythematosus (60). Like IgG2, CRP binds to several encapsulated bacteria. Evidence for a reciprocal relationship between the binding affinities of IgG2 and CRP for FcγRIIa suggests a mechanism for partial protection from invasive infection in individuals homozygous for FcγRIIa-R131 (53).

Functional differences between the NA1 and NA2 alleles also appear to have clinical significance. Homozygous NA1 individuals are more resistant to bacterial infection, especially when FcγRIIa cannot be effectively engaged, as suggested by the finding of increased *Neisseria meningitidis* infection among hosts with complement component 6 or 8 deficiency who are homozygous for FcγRIIIb-NA2 and FcγRIIa-R131 (39). Alternatively, increased ROI generated by the NA1-FcγRIIIb allele may enhance function of PMN bearing FcγRIIa-R131 and lead to more efficient defense against encapsulated microbes. In Wegener's granulomatosis, a systemic vasculitis characterized by anti-neutrophil cytoplasmic antibodies (ANCA) that activate PMN and lead to inflammation and damage of blood vessels, both FcγRIIa and FcγRIIIb are engaged by ANCA on neutrophils to trigger cell activation (27). It has been suggested that alleles with increased binding capacity predispose to more severe tissue injury (14). The importance of the interplay between ROI and FcγR in host defense is underscored by the recent report that the risk for immune-mediated complications of CGD is associated with FcγR allelic polymorphisms (20).

FcγR AND ROI: IMPLICATIONS FOR HOST DEFENSE AND TISSUE INJURY

Our observations and those of others provide the basis for a better understanding of the regulation of FcγR at sites of inflammation. Perhaps more importantly, the data presented in this review indicate a mechanism for priming phagocytes for enhanced responses to receptor-driven effects. ROI generated in an inflammatory milieu act in an autocrine and paracrine manner to rapidly amplify the effector potential of FcγR on quiescent phagocytes by altering signal transduction. Indeed, for FcγRIIa, exposure to oxidants enables uptake of an IgG2-opsonized particle by FcγRIIa-R131 homozygotes, albeit to a lesser extent than that of other FcγRIIa genotypes, and thus allows removal of IgG2-opsonized microbes and immune complexes despite relatively low binding capacity. Hence, for antimicrobial defense, ROI-initiated increases in phagocytosis are protective.

In contrast, at sites of immune complex deposition, such as the kidney in systemic lupus erythematosus, amplification of FcγRIIa-triggered release of inflammatory mediators may promote tissue injury. Of note, in the absence of PMN influx, renal injury is attenuated in murine models of autoimmune glomerulonephritis (56). Alternatively, FcγR-driven phagocyte-derived ROI may act as second messengers to increase platelet aggregation, vascular smooth muscle cell proliferation, and mesangial cell proliferation (29, 38, 55, 59), all characteristic findings in diffuse proliferative glomerulonephritis. Our experiments showing that oxidants from activated PMN augment "bystander" monocyte FcγRIIa function underscore the importance of this paracrine mechanism (41). These find-

ings, along with evidence that FcγR-deficient mice are protected from autoimmune glomerulonephritis (8, 37), highlight the importance of the identifying the factors, which modulate the efficiency of FcγR function. Definition of the role of oxidants as amplifiers of FcγR signaling provides a novel target for therapeutic intervention in immune complex-mediated tissue injury.

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ABBREVIATIONS

ANCA, anti-neutrophil cytoplasmic antibodies; $[Ca^{2+}]_i$, intracellular free Ca^{2+} concentration; CGD, chronic granulomatous disease; CRP, C reactive protein; FcγR, receptors for Fc portion of IgG; FcγRI, type I high-affinity receptor for IgG; FcγRIIa, type IIa low-affinity receptor for IgG; FcγRIIIb, type IIIb receptor for IgG; GPI, glycosylphosphatidylinositol; H_2O_2, hydrogen peroxide; IFN-γ, interferon-γ; IgG, immunoglobulin G; ITAM, immunoreceptor tyrosine-based activation motif; ITIM, immunoreceptor tyrosine-based inhibitory motif; mAb; monoclonal antibody; NA, neutrophil antigen; NADPH oxidase, nicotinamide adenine dinucleotide phosphate H; O_2^-, superoxide anion; PMN, polymorphonuclear neutrophils; ROI, reactive oxygen intermediates; SH2, src homology domain 2; SHIP, src homology inositol polyphosphate 5′-phosphatase; SHP-1, SH2 domain-containing protein tyrosine phosphatase-1.

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