International Immunopharmacology 17 (2013) 638–650

Contents lists available at ScienceDirect

International Immunopharmacology

journal homepage: www.elsevier.com/locate/intimp

Neutrophil cell surface receptors and their intracellular signal transduction pathways☆

Krisztina Futosi [a], Szabina Fodor [b], Attila Mócsai [a,*]

[a] Department of Physiology, Semmelweis University School of Medicine, 1094 Budapest, Hungary
[b] Department of Computer Science, Corvinus University of Budapest, 1093 Budapest, Hungary

ARTICLE INFO

Article history:
Received 16 October 2012
Received in revised form 7 December 2012
Accepted 9 June 2013
Available online 30 August 2013

Keywords:
Neutrophils
Signaling
Receptors
Kinases
Inflammation

ABSTRACT

Neutrophils play a critical role in the host defense against bacterial and fungal infections, but their inappropriate activation also contributes to tissue damage during autoimmune and inflammatory diseases. Neutrophils express a large number of cell surface receptors for the recognition of pathogen invasion and the inflammatory environment. Those include G-protein-coupled chemokine and chemoattractant receptors, Fc-receptors, adhesion receptors such as selectins/selectin ligands and integrins, various cytokine receptors, as well as innate immune receptors such as Toll-like receptors and C-type lectins. The various cell surface receptors trigger very diverse signal transduction pathways including activation of heterotrimeric and monomeric G-proteins, receptor-induced and store-operated Ca^{2+} signals, protein and lipid kinases, adapter proteins and cytoskeletal rearrangement. Here we provide an overview of the receptors involved in neutrophil activation and the intracellular signal transduction processes they trigger. This knowledge is crucial for understanding how neutrophils participate in antimicrobial host defense and inflammatory tissue damage and may also point to possible future targets of the pharmacological therapy of neutrophil-mediated autoimmune or inflammatory diseases.

1. Introduction

Neutrophils are the most abundant circulating leukocytes in the human blood. They develop in the bone marrow from the myeloid hematopoietic system and share a number of characteristic features with other myeloid cells such as monocytes/macrophages and mast cells [1,2]. Neutrophils are short-lived, terminally differentiated cells that, unless activated by a microbial or inflammatory stimulus, only

Abbreviations: Abl, Abelson leukemia proto-oncogene; ADAP, adhesion and degranulation promoting adapter protein (Fyb, SLAP-130); Asc, apoptosis-associated speck-like protein containing a CARD; BCR, B-cell receptor; C3G, Crk SH3 domain-binding guanine nucleotide exchange factor (RapGEF1); CALDAG-GEFI, calcium and DAG-regulated guanine nucleotide exchange factor I; CARD, caspase activation and recruitment domain; CEACAM3, carcinoembryonic antigen-related cell adhesion molecule 3 (CD66b); CHO, Chinese hamster ovary cells; cIAP, cellular inhibitor of apoptosis; CLEC, C-type lectin; DAG, diacyl-glycerol; DAP12, DNAX activating protein 12; DISC, death-inducing signaling complex; Epac1, exchange protein activated by cyclic AMP 1; ERK, extracellular signal-regulated kinase; ERM, ezrin-radixin-moesin; ESL-1, E-selectin ligand 1; FADD, Fas-associated protein with death domain; FcR, Fc-receptor; FcRγ, Fc-receptor γ-chain; Fgr, Gardner–Rasheed feline sarcoma proto-oncogene; fMLP, formly-Met-Leu-Phe; GAP, GTPase activating protein; G-CSF, granulocyte colony-stimulating factor; GM-CSF, granulocyte/monocyte colony-stimulating factor; GPCR, G protein-coupled receptor; GPI, glycosylphosphatidylinositol anchor; GRK, GPCR kinase; Hck, hematopoietic cell kinase; ICAM-1, intercellular adhesion molecule 1; IFN, interferon; IKK, IκB kinase; IL, interleukin; IP_3, inositol-tris-phosphate; IRAK, IL-1 receptor-associated kinase; IRF, IFN regulatory factor; ITAM, immunoreceptor tyrosine-based activation motif; IκB, inhibitor of NF-κB; JAK, Janus kinase; JNK, c-Jun N-terminal kinase; LAD, leukocyte adhesion deficiency; LFA-1, lymphocyte function-associated receptor 1 ($\alpha_L\beta_2$ integrin); LTβ, lymphotoxin β; LTB_4, leukotriene B_4; Mac-1, macrophage antigen 1 ($\alpha_M\beta_2$ integrin); MAP kinase, mitogen-activated protein kinase; MAPKAP-kinase, MAP kinase-associated protein kinase; Mcl, macrophage C-type lectin; MDA5, melanoma differentiation-associated protein 5; MDL-1, myeloid DAP12-associating lectin 1; MIP, macrophage inflammatory protein; MKK, MAP kinase kinase; MyD88, myeloid differentiation protein 88; NF-κB, nuclear factor κB; NLRP3, NOD-like receptor family, pyrin domain containing 3; NOD, nucleotide-binding oligomerization domain containing protein; OSCAR, osteoclast-associated receptor; PAF, platelet activating factor; PAK, p21-activated kinase; PI3K, phoshoinositide-3-kinase; PIP_3, phosphatidylinositol-3-phosphate; PIR, paired immunoglobulin-like receptor; PKB, protein kinase B; PKC, protein kinase C; PLC, phospholipase C; PSGL-1, P-selectin glycoprotein ligand; Rac, Ras-related C3 botulinum toxin substrate; RANK, receptor activator of NF-κB; Rap, Ras-related protein; RIG, retinoic acid-inducible gene; RIP3, receptor-interacting serine-threonine protein kinase 3; ROS, reactive oxygen species; SAP130, Sin3A-associated protein of 130 kDa; SH2, Src-homology 2 domain; SHP-1, SH2 domain-containing protein tyrosine phosphatase 1; SLP-76, SH2 domain-containing leukocyte protein of 76 kDa; SOCS, suppressor of cytokine signaling; Src, Rous sarcoma virus proto-oncogene; STAT, signal transducer and activator of transcription; Syk, spleen tyrosine kinase; TAK, TGFβ-activated kinase 1; TCR, T-cell receptor; TGFβ, transforming growth factor β; TLR, Toll-like receptor; TNF, tumor necrosis factor; TRADD, TNFR1-associated death domain protein; TRAF, TNF receptor-associated factor; TRAIL, TNF-related apoptosis-inducing ligand; TREM, triggering receptor expressed on myeloid cells; Tyk2, tyrosine protein kinase 2; VASP, vasodilator-stimulated phosphoprotein; VCAM-1, vascular cell adhesion molecule 1; VLA-4, very late antigen 4 ($\alpha_4\beta_1$ integrin); ZAP-70, ζ-chain-associated protein of 70 kDa.

* Corresponding author at: Department of Physiology, Semmelweis University School of Medicine, Tűzoltó utca 37–47, 1094 Budapest, Hungary. Tel.: +36 1 459 1500x60 409; fax: +36 1 266 7480.
E-mail address: mocsai.attila@med.semmelweis-univ.hu (A. Mócsai).

1567-5769/$ – see front matter © 2013 The Authors. Published by Elsevier B.V. All rights reserved.
http://dx.doi.org/10.1016/j.intimp.2013.06.034

survive for a short time in the bloodstream and die by a spontaneous apoptotic program, followed by removal of dead neutrophils by macrophages. Neutrophils show a condensed and multilobed nuclear morphology (likely reflecting the limited transcriptional activity of the cells) and contain a large number of intracellular granules and vesicles with no prominent staining characteristics [3]. Those features explain the alternative designation of the cells as polymorphonuclear cells or neutrophilic granulocytes.

The primary role of neutrophils is host defense against bacterial and fungal pathogens, providing the first line of defense against invading microorganisms. Neutrophils express a large number of cell surface receptors for the recognition of microbial invasion. Some of those receptors are capable of innate recognition of microbial structures while others (such as Fc-receptors) are linked to the activation of the adaptive immune response, and yet other receptors recognize the inflammatory environment.

The antimicrobial activity of neutrophils relies on the effective recognition and elimination of microbial pathogens, as well as complex intracellular signal transduction pathways linking those processes to each other. Additional signal transduction processes are not directly involved in microbial recognition and elimination but inform the cells of their environment (such as an inflammatory interstitium) or promote additional processes (such as chemotaxis) indirectly required for the elimination of pathogens. Taken together, intracellular signal transduction processes need to convey a large amount of complex information to support an efficient antimicrobial immune response.

There are several classes of receptors expressed on the surface of neutrophils, including G-protein-coupled seven-transmembrane receptors, Fc-receptors, adhesion molecules like selectins/selectin ligands and integrins, various cytokine receptors, as well as innate immune receptors including Toll-like receptors and C-type lectins (Table 1). Activation of those receptors leads to complex cellular activation and elimination processes such as phagocytosis, exocytosis of intracellular granules, production of reactive oxygen species, release of neutrophil extracellular traps, as well as additional responses like chemotactic migration or chemokine and cytokine release.

The aim of this review is to provide an overview of neutrophil cell surface receptors and their intracellular signal transduction processes. Given the very large amount of information available on that subject, only a small portion of the available data will be discussed, focusing on pathways where genetic data from primary mammalian neutrophils are available and where results may have implications in the understanding, diagnosis and therapy of autoimmune and inflammatory diseases.

2. Signaling by G-protein-coupled receptors

2.1. G-protein-coupled receptors on neutrophils

Neutrophils express a large number of G-protein-coupled receptors (GPCRs) that participate in host defense and inflammation (Table 1). Those include formyl-peptide receptors [4–6] that sense bacterial products and tissue injury (through recognition of release of mitochondrially synthesized proteins), receptors for a diverse set of "classical chemoattractants" such as leukotriene B_4 (LTB_4), platelet activating factor (PAF) and complement fragment C5a [6–9], as well as CXC (CXCR1, CXCR2) and, to a lesser extent, CC (CCR1, CCR2) chemokine receptors [10–13].

A common feature of the above G-protein-coupled receptors is that they strongly activate the chemotactic migration of neutrophils; therefore their agonists are conventionally termed "chemoattractants". It should nevertheless be stated that most of those ligands (especially formyl-peptides, lipid mediators and C5a) also trigger neutrophil responses other than chemotaxis, including ROS production and exocytosis of intracellular granules and vesicles, and they are also able to augment the responses of neutrophils to subsequent stimulation by other agonists ("priming" effect).

2.2. GPCR signal transduction

All of the above GPCR agonists signal through pertussis toxin-sensitive heterotrimeric G-proteins of the $G_{i/o}$ family. Activation of those receptors triggers the dissociation of the GPCR-specific Gα subunit from the shared Gβγ dimer and concomitant activation of various signal transduction pathways by both G-protein fragments (Fig. 1). The $G\alpha_i$ subunit inhibits adenylyl cyclase activity and therefore reduces cytoplasmic cAMP levels. However, it is unclear whether that inhibition plays any major role in GPCR signaling in neutrophils. Instead, our current understanding is that the majority of GPCR signal transduction in neutrophils occurs through the Gβγ subunit [14–16].

One of the classical signals triggered by GPCRs in neutrophils is a prominent biphasic Ca^{2+}-signal. The first phase of this signal is likely mediated by phospholipase Cβ (PLCβ) enzymes leading to the generation of IP_3 and concomitant release of Ca^{2+} from intracellular stores. Indeed, the combined genetic deficiency of PLCβ2 and PLCβ3 completely abrogated fMLP-induced IP_3 production, the increase of cytoplasmic Ca^{2+}-concentration, the activation of conventional PKC isoforms and

Table 1
The most important neutrophil receptors. See the text for further details.

G-protein-coupled receptors	Fc-receptors	Adhesion receptors	Cytokine receptors	Innate immune receptors
Formyl-peptide receptors	*Fcγ-receptors*	*Selectins and selectin ligands*	*Type I cytokine receptors*	*Toll-like receptors*
• FPR1 (FPR)	• FcγRI	• L-selectin	• IL-4R	• TLR1
• FPR2 (FPRL1)	• FcγRIIA (human)	• PSGL-1	• IL-6R	• TLR2
• FPR3 (FPRL2)	• FcγRIIB (inhibitory)	*Integrins*	• IL-12R	• TLR4
Classical chemoattractant receptors	• FcγRIII (mouse)	• LFA-1 ($\alpha_L\beta_2$)	• IL-15R	• TLR5
• BLT1 (LTB_4-rec.)	• FcγRIIIB (human)	• Mac-1 ($\alpha_M\beta_2$)	• G-CSFR	• TLR6
• BLT2 (LTB_4-rec.)	• FcγRIV (mouse)	• VLA-4 ($\alpha_4\beta_1$)	• GM-CSFR	• TLR7 (?)
• PAFR	*Fcα-receptors*		*Type II cytokine receptors*	• TLR8
• C5aR	• FcαRI (human)		• IFNAR (IFNα/β-rec.)	• TLR9
Chemokine receptors	*Fcε-receptors*		• IFNGR	*C-type lectins*
• CXCR1 (human)	• FcεRI		• IL-10R	• Dectin-1
• CXCR2	• FcεRII		*IL-1R family*	• Mincle
• CCR1			• IL-1RI	• MDL-1
• CCR2			• IL1RII (decoy)	• Mcl
			• IL-18R	• CLEC-2
			TNFR family	*NOD-like receptors*
			• TNFR1 (p55)	• NOD2
			• TNFR2 (p75)	• NLRP3
			• Fas	*RIG-like receptors*
			• LTβR	• RIG-I
			• RANK	• MDA5
			• TRAIL-R2	
			• TRAIL-R3	