ELSEVIER

CrossMark

Experimental Hematology

Experimental Hematology 2015;43:158–167

REVIEW

Role of toll-like receptors in multiple myeloma and recent advances

Krishan K. Thakur[a], Nityanand B. Bolshette[a], Cristiana Trandafir[b], Vinayak S. Jamdade[c], Alexandru Istrate[b], Ranadeep Gogoi[a], and Andrei Cucuianu[d]

[a]*Laboratory of Biotechnology, Department of Biotechnology, National Institute of Pharmaceutical Education and Research, Gauhati Medical College, Assam, India;* [b]*Faculty of Medicine, University of Medicine and Pharmacy Iuliu Haţieganu, Cluj-Napoca, Romania;* [c]*Laboratory of Molecular Pharmacology and Toxicology, Department of Pharmacology and Toxicology, National Institute of Pharmaceutical Education and Research, Gauhati Medical College, Assam, India;* [d]*Department of Hematology, Ion Chiricuta Cancer Institute, Cluj-Napoca, Romania*

(Received 20 June 2014; revised 2 November 2014; accepted 10 November 2014)

Multiple myeloma (MM) is a hematologic malignancy characterized as an abnormal proliferation and invasion of plasma cells into the bone marrow. Toll-like receptors (TLRs) connect the innate and adaptive immune responses and represent a significant and potentially linking element between inflammation and cancer. When TLRs bind to their ligands, they trigger two major signaling pathways such that both share overlapping downstream signals: one is a myeloid differentiation primary response 88 (MyD88)-dependent production and activation of nuclear factor-κB, whereas the other is a MyD88-independent production of type-I interferon. Whereas the MyD88 pathway results in proinflammatory cytokine production, the other pathway stimulates cell proliferation. Dysregulations of these pathways may eventually lead to abnormal cell proliferation and MM. Despite recent biomedical advances, MM continues to be an incurable disease. There are an increasing number of TLR-based therapeutic approaches currently being tested in a number of preclinical and clinical studies. We here attempt to outline in detail the currently available information on TLRs in various types of cancer.

Multiple myeloma (MM) is currently the second most common hematologic malignancy after chronic lymphocytic leukemia, and the incidence of MM has been increasing lately [1]. The so-called CRAB criteria (elevated calcium level, renal failure, anemia, and osteolytic bone lesions) [2–4] clinically define this pathology. In MM, monoclonal plasma cells multiply and secrete large quantities of monoclonal immunoglobulins. Usually, normal immunoglobulins in the bloodstream decrease in number and leave MM patients susceptible to infection or inflammation [5]. This renders therapeutic approaches difficult; MM is practically incurable, since the currently available treatments only lead to transient responses [3,6]. Moreover, chronic infection and inflammation increase the risk of developing MM and thus might be involved in its pathogenesis and progression [7,8]. However, until now, the fundamental molecular mechanisms of these processes have not been clearly decoded [2,4,9].

Toll-like receptors (TLRs) are noncatalytic, transmembrane, single receptors that link at a molecular level infection, tissue injury, and inflammation [10,11]. They stimulate the organism to respond with both innate and adaptive immune responses and increase tumor resistance and invasiveness. Multiple myeloma cells express TLRs, and this shows that the bone marrow environment responds to tumor-induced signals with inflammation [2,3]. However, most studies that have so far found a connection between TLRs and cancer have been restricted to mRNA studies: the mRNA level differences explain the variability of TLRs expressed in various cells and cellular responses

KKT and NBB contributed equally to this article.

Offprint requests to: Nityanand B. Bolshette, Department of Biotechnology, National Institute of Pharmaceutical Education and Research (NIPER), Gauhati Medical College, Guwahati, Assam 781032, India; E-mail: nbolshette@gmail.com

0301-472X/Copyright © 2015 ISEH - International Society for Experimental Hematology. Published by Elsevier Inc.
http://dx.doi.org/10.1016/j.exphem.2014.11.003

[12]. Also, the expression patterns of the functional forms of these proteins are insufficiently known. Since TLRs modulate adaptive immune responses, current research is focused on TLR-based therapeutic approaches that enhance the efficiency of anticancer immunotherapies [13]. Toll-like receptors and their distinct signaling pathways in myeloma cells might be potential therapeutic targets for tumor progression inhibitors [8]. In this review, we summarize the main advances in the mechanisms of TLR involvement in homeostasis. The review supports the link between TLR activation and cell proliferation and indicates that TLR overexpression might be involved in MM development. We focused on the role of TLRs in inflammatory processes present in MM and to emphasized their relation to tumor progression.

Summary of Toll-like receptors

Toll-like receptors are members of the pattern recognition receptor family, a group of proteins that facilitate the accurate identification of preserved pathogen-associated molecular patterns (PAMPs) [14–17]. They are type-I transmembrane glycoproteins expressed preferentially in the cells of the innate immune system, but also in platelets and B and T lymphocytes [18]. Toll-like receptors are expressed in myeloma cells and nonhematopoietic cells, where they may influence tumor growth and host immune responses [19].

At present, 13 TLRs have been discovered, of which the first 10 in humans and the remaining ones in mice [20]. All of them consist of three major domains: extracellular, transmembrane, and cytoplasmic. The extracellular domain harbors a leucine-rich repeat consisting of 19–25 tandem copies of the "xLxxLxLxx" motif [21], which detects and binds bacterial cell-wall components, viral single- or double-stranded RNAs, and small molecules of antiviral compounds. The transmembrane domain is embedded in the cell membrane or the endosomal membrane. Lastly, the cytoplasmic domain, or Toll–interleukin (IL) 1 receptor domain (TIR domain), binds to the adaptors eloid differentiation primary response 88 (MyD88), TIRAP/Mal, TIR-domain-containing adapter-inducing interferon-β (TRIF), and TRIF-related adaptor molecule (TRAM), and it initiates the downstream signaling pathway [22–26].

Toll-like receptors play vital roles in the structural recognition of the innate immunity [27,28]: bacterial membrane-associated PAMPs activate TLR1, -2, -4, -5, and -6 on the plasma membrane [5], whereas nucleic-acid-based PAMPs from bacteria and viruses activate TLR3, -4, -7 and -9 expressed only on endosomes [28] (Table 1). Toll-like receptor activation triggers the adaptive immune response. Here, we present the roles of individual TLRs: TLR1 and TLR6 are active only after they form heterodimers with TLR2. The TLR1/2 heterodimer is a receptor for lipopeptides from mycobacteria and meningococci, and the TLR2/6 heterodimer is a receptor for mycoplasma lipoproteins and peptidoglycans [27,29]. Overexpression of TLR1 and TLR2 on MM cells enhances their adhesion to bone marrow stromal cells and their sensitivity to bortezomib and neutralizes the protective effect of bone marrow stromal cells over MM cells [30]. Toll-like receptor 2 forms heterodimers with either TLR1 or TLR6. These heterodimers respond to triacylated or diacylated bacterial lipopeptides. The TLR3 homodimers directly or indirectly

Table 1. Overview of TLRs with their respective functions

TLRs	Species	Location	PAMPs recognized by TLRs	Function
TLR1/2	H/M	Cell surface	Ac3LP, glycolipids, triacyllipopeptides	receptors for lipopeptides
TLR2/6	H/M	Cell surface	Ac2LP, LTA, zymosan, peptidoglycan, ciacyllipopeptides	receptors for lipopeptides
TLR3	H/M	Endosome	polyI:C, dsRNA(reovirus), RSV, MCMV	stimulates IFN-α/β and IL-12p70 production
TLR4	H/M	Cell surface	LPS, taxol, heparan, hyaluronate, F-prost, VSV, envprost,	stimulates rapid proliferation and survival of CD4+ T-cells
TLR5	H/M	Cell surface	Flagellin	modulates the transcription of TNFα in rheumatoid arthritis
TLR7	H/M	Endosome	ssRNA, imiquimod, loxoribine, and guanine analogs such as loxoribine	activates NF-κB, increases antibody secretion and cytokine production in B cells
TLR8	H/M	Endosome	ssRNA, IAQ (R848)	activates NF-κB, mediates the inflammation of the nervous tissue after an ischemic stroke attack
TLR9	H/M	Endosome	dsDNA viruses (HSV, MCMV), CpG motifs from bacteria and viruses, hemozoin (plasmodium)	increases proliferation, survival and differentiation, as well as secretion of IL-6, IL-10, immunoglobulins, autoimmune disease prevention
TLR10	H	Cell surface	Unknown	modulates cellular responses to gram positive bacterial peptidoglycan
TLR11	M	Cell surface	Profiling	expressed and completely functional in mice
TLR12	M	Cell surface	Unknown	expressed and completely functional in mice
TLR13	M	Endosome (Probably)	Unknown	expressed and completely functional in mice

H = Human; *LTA* = lipoteichoic acid; *LPS* = lipopolysaccharide; *M* = mouse; *MCMV* = murine cytomegalovirus; *R848* = resiquimod; *RSV* = respiratory syncytial virus; *ssRNA* = single-stranded RNA; *VSV* = vesicular stomatitis virus.