The Immune System, Our Personal Bodyguard

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The central role of the human immune system

The complexity of our immune system evolved over millions of years to minimize the threat by pathogens and neoplasms. Although we normally are not aware of its subtle functions as long as we enjoy our health, an early inflammatory reaction clearly denotes the beginning of the fight of our immune cells against invaders, such as viruses, bacteria, fungi, and even parasites. Close collaboration between innate and specific immunity ensures the elimination of the infectious agent by cellular and/or humoral immune responses. In some instances, long-lived immunity is generated. The aim of the present article is to briefly outline important mechanisms of immune reactions against infectious microorganisms. The molecular details of these interactions are beyond the scope of this article, but they can easily be found in the reviews cited.

Structural organization of the human immune system

Whereas innate immune responses are immediately available on contact with pathogens, the activation of specific immunity takes longer. The T and B cells, with their highly diverse antigen receptors, play a central role in this activation.

All immune cells originate from hematopoietic stem cells in the bone marrow. Under the influence of numerous cytokines and growth factors, the so-called pluripotent stem cells differentiate in a multistep process into several types of granulocytes (i.e., neutrophils, eosinophils, and basophils), each of which has specialized functions; monocytes (which differentiate to the tissue macrophages when settling in different organs); natural killer (NK) cells; and B and T cells.

The lymph nodes are localized as a large network throughout the human body to sample antigens from the tissues via the lymph vessels. Lymph nodes are usually the site of sensitization of T cells by antigen-presenting cells (APCs). The mucosa-associated lymphoid tissue (MALT) includes the Peyer's patches along the gastrointestinal tract, the tonsils, and the nasal- and bronchus-associated lymphoid tissue. All of these tissues form highly organized structures supporting the interaction of antigens with the few available antigen-specific lymphocytes circulating in the blood or the lymph. The MALT is essential as a protective barrier at the highly vulnerable mucosal surfaces.[1-3]

Innate immunity: a powerful first-line defense

The first defense against infectious agents starts when the invader is recognized by phagocytes that nonspecifically engulf and digest pathogens. Even this most primitive defense function is a highly complex cellular process.[4,5] Two different types of phagocytosis exist: the removal of pathogens and the elimination of apoptotic tissue cells (apoptosis means programmed cell death). The former causes an inflammatory alarm response, whereas the latter (which is, for example, necessary during embyrogenesis) prevents inflammation. Moreover, phagocytosis bridges innate and adaptive immunity, through antigen presentation.

The engulfment of pathogens by neutrophils and macrophages discriminates between diverse particles through an array of receptors expressed on their surface. Among these receptors are several for complement proteins, combinations of scavenger receptors, and numerous integrins, described extensively by Stuart and Ezekowitz in 2008.[5] Most of these receptors are able to recognize both pathogens and altered-self ligands, such as apoptotic cells.

After receptor ligation by the particle, a "phagosome" is formed within the phagocyte. This phagosome then fuses with a lysosome, generating the "phagolysosome." In the latter, the final destruction of pathogens occurs by an arsenal of degrading enzymes, oxygen radicals, bactericides, etc. Proteomic analysis has revealed that phagosomes contain more than 600 different types of

proteins. A key role of phagolysosomes is to provide, by only partial degradation, the antigenic ligands for the stimulation of the T and B cells (which are further described later in the article).

Role of toll-like receptors in antimicrobial immunity

The family of receptors called toll-like receptors (TLRs) is essential for the discrimination between self and nonself structures, a central requirement for the immune system. This topic was extensively reviewed by Akira and Takeda[6] and Iwasaki and Medzhitov.[7]

The TLRs sense microbial infections as a "general danger" to the body, recognizing conserved molecular structures unique to the microbial world and widely invariant among the single classes of pathogens. Each of these pathogen-associated molecular patterns (PAMPs) is detected by a different TLR subtype (e.g., TLR4 recognizes lipopolysaccharides). The PAMPs are among the strongest stimuli for immune cells. The signal transduction pathways that TLRs activate in different immune cell subtypes result in a multitude of antimicrobial and inflammatory responses, which usually lead to the elimination of the pathogen. The TLRs also do the following:

1. help recruit cells to infected sites by triggering the release of chemotactic mediators (chemokines)
2. help make functionally mature APCs
3. contribute to antiviral immunity[8]

Therefore, PAMPs very efficiently link innate and adaptive immune mechanisms, thus potentiating defense efficacy.

The neutrophil: a prototypic cell type of antibacterial defense

Neutrophil granulocytes are the most abundant cells of the immune system. Beyond being pure "eaters and killers," they are recognized as major contributors to the overall regulation of immune responses. Neutrophils also contribute to the recruitment, activation, and programming of APCs by producing an array of cytokines, chemokines, lipid mediators, and, last but definitely not least, an arsenal of cytolytic agents for killing ingested pathogens, as described by Nathan.[9] Among the latter are bactericidal peptides (defensins), oxygen radicals produced by myeloperoxidase, and others. Lactoferrin, or lipocalin, can slow down bacterial growth by depleting iron at the site of infection. In addition, neutrophils secrete factors that assist B-cell maturation and proliferation and can also function as prominent suppressors of T-cell function (e.g., by secreting prostaglandins).

The role of complement proteins in immunity

The complement system deserves attention in that this proteolytic machinery, resembling in its cascade-like mode of action the coagulation cascade, effectively interlinks innate and specific immune mechanisms. First described as a heat-sensitive factor in fresh serum that is able to "complement" the effects of specific antibodies in the lysis of bacteria, the complement system now represents a system of more than 30 serum proteins and cell surface receptors. An excellent review on complement concerning numerous immunoregulatory roles beyond the killing of bacteria has been published by Carroll.[10]

Messaging between cells of the innate immune system

To accomplish the antibacterial defense during innate immune reactions, the phagocytosis of microbial pathogens is accompanied by the release of several messenger molecules, such as arachidonic acid metabolites (prostaglandins and leukotrienes), chemokines, cytokines, and proteases. Only a fine-tuned release of these hundreds of mediators coordinates the activities of different immune cells sufficiently to successfully clear the tissues of almost all infectious microorganisms before they can create problems.

Initiation of an antigen-specific immune response against infection

In many cases, the first line of defense established by the phagocytes is not enough, especially when microbial and viral pathogens evolved to exhibit sophisticated survival strategies. In such cases, antigen-specific immune responses are initiated. Even these begin with phagocytosis, although, as reviewed recently by Finlay and McFadden,[11] some pathogens may resist phagocytosis and others interfere with antigen presentation. Those pathogens that resist digestion and multiply within the phagocytes constitute a tremendous threat to the body (e.g., mycobacteria and parasitic helminth worms). Despite their subversive activities, these pathogens can be destroyed by more specific cellular immune mechanisms. There is antibody-dependent cytotoxicity and enforced cellular immunity; the latter results in a profound activation of macrophages, boosting them to destroy even microorganisms as resistant as mycobacteria. Such reenforcement usually involves the cells from the antigen-specific part of the immune system, the T and B cells.

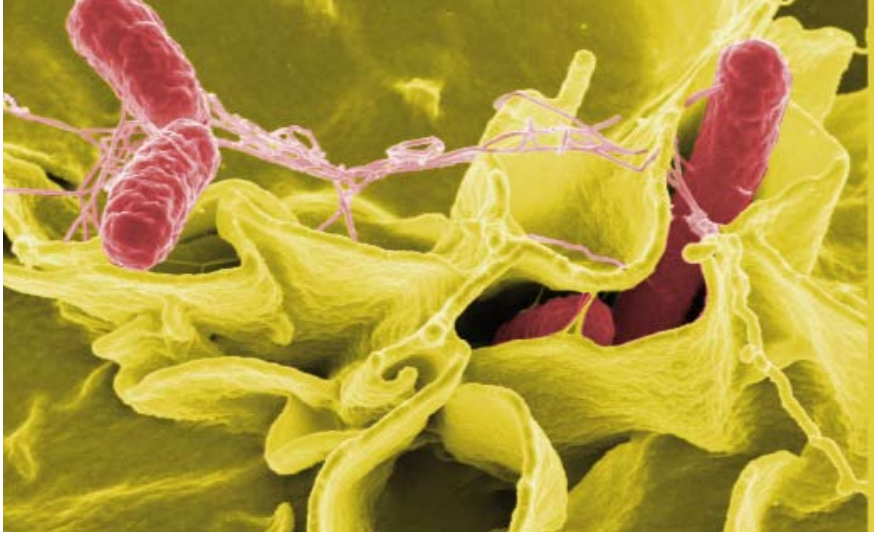


Photo by Rocky Mountain Laboratories, NIAID, NIH, http://en.wikipedia.org/wiki/Image:SalmonellaNIAID.jpg

Bacteria display a wide diversity of shapes and sizes. Here: Salmonella typhimurium (red) invading cultured human cells.

T and B lymphocytes are responsible for generating antigen-specific immune responses

True antigen specificity resides in the T and B lymphocytes, which are able to recognize antigens through highly specific membrane-bound receptors. Each cell has peculiar receptors that recognize only one antigen. Yet, hypothetically, all B and T lymphocytes together are able to respond to virtually any antigen in the world. The antigen size may range from small chemical structures to highly complex molecules. The receptors of both cell types recognize only a small part of large antigens, referred to as the epitope. Complex antigens usually consist of more than one epitope. This tremendous variability in T- and B-cell specificities is achieved by DNA rearrangement.[12,13]

Antigens are internalized and processed to smaller fragments, which are then presented at the surface of APCs to "naïve" T cells, teaching the latter about the current antigen load. Compared with T cells, B cells do respond to nondigested epitopes. The surface structures to which the antigens or the fragments are attached are the proteins of the major histocompatibility complex (MHC), of which 2 classes are highly important for immune and tissue cells: class I (MHC-I) and class II (MHC-II).

Antigen presentation by MHCs

Recognition of antigens in the binding grooves of MHC molecules by specific T-cell receptors (TCRs) is the central event to T-cell activation. MHC-I, found on all cells of the human body, was originally described as transplantation antigen(s), being the cause for organ rejection. The natural function of MHC-I is to sample antigens from within the cells (e.g., during infection [viruses or intracellular bacteria] or tumorigenesis).[14] The recognition of antigens presented by MHC-I molecules leads to the activation of cytotoxic T cells (CTLs) bearing the CD8 surface marker (CD8+ CTLs).

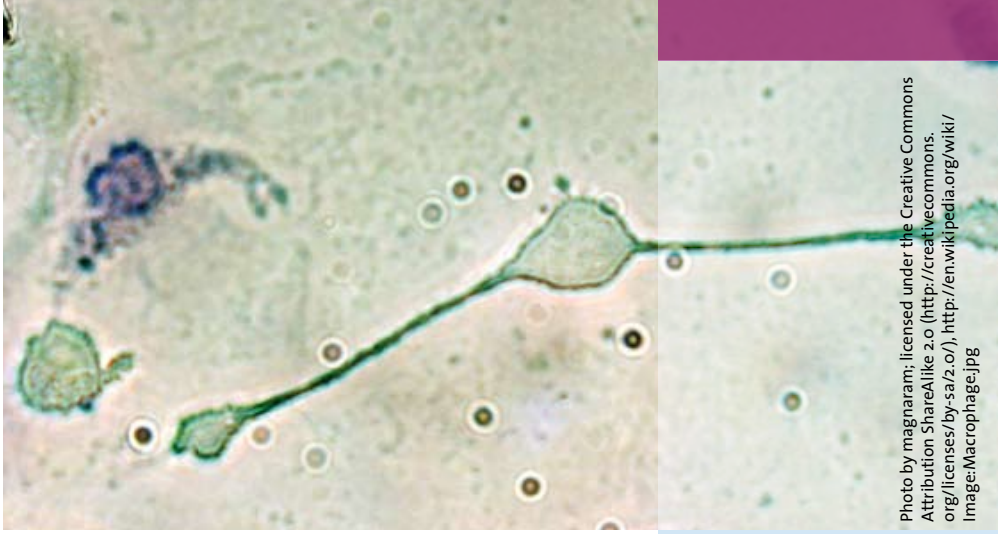
The MHC-II proteins are found exclusively on cells of the immune system (e.g., macrophages, B cells, and dendritic cells [DCs]). The DCs are recognized as the most efficient APCs to stimulate naïve T cells. The DCs seem to decide which type of T-cell response is induced by different antigens (Reis e Sousa[15] and Shortman and Naik[16] provide reviews). In contrast to MHC-I molecules, MHC-II molecules sample antigens from the extracellular space to activate CD4-positive (CD4+) T-helper (Th) cells.

Effector/inflammatory CD4+ Th cells and cytotoxic CD8 T cells

Viruses are crucial pathogens because they hide and multiply inside susceptible tissue cells. Antibodies neutralize viruses only outside cells (i.e., before they enter target cells or when they are released by these cells after replication). The elimination of virus-infected host cells is, therefore, a real challenge for the immune system.

Evolution enabled infectious pathogens (i.e., viruses, bacteria, and parasites) to develop improved strategies to override the immune defense, which, in turn, improved its effector mechanisms to destroy even these pathogens. This is the reason why we have specialized populations of T cells, such as CTLs and various Th cells.

Basically, CD4+ T-cell activation is initiated by the interaction of the antigen receptor (TCR) with antigen/MHC-II complexes on APC surfaces. Antigen/MHC-II complexes trigger a complex concert of intracellular signals, activating a whole series of genes that control the proliferation, differentiation, and effector functions of T cells. Antigen-specific T-cell activation is initiated only when these signals are strong enough.[17] When a T cell is activated, it proliferates to give a clone, with each clone cell having the same TCR specificity as the parent cell. Notably, proliferation needs several growth factors (e.g., the very well-known interleukin [IL] 2). IL-2 is the prototype of a T-cell growth factor and acts to promote proliferation and differentiation of antigen-activated T cells.[18,19]



A macrophage forming two processes to phagocytize two smaller particles.

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CD4+ T cells activate cellular immunity

A core function of CD4+ T cells in antibacterial defense is the re-enforcement of tissue macrophages to better kill intracellular parasites and bacteria that otherwise may survive phagocytosis and use these cells as incubators. Macrophage activation by T cells is essential to cellular immunity against pathogens, such as leishmania and mycobacteria. This activation of macrophages depends on cytokines from activated CD4 Th cells, most importantly interferon (IFN) γ, which is also provided by NK cells. Other cytokines supporting cell-mediated immunity are mediators, such as IL-12 and IL-18, which are produced by activated APCs in a positive feedback loop. Macrophages activated in this manner express a higher ability to present antigens, provide stronger costimulation, and secrete more activating cytokines (e.g., IL-1, IL-6, and IL-10) or tumor necrosis factor α. Moreover, the CD4+ T lymphocytes are important helper cells for antiviral CTLs. CD4+ T cells are not only crucial for macrophage activation but also provide help to B cells by secreting growth factors favoring antibody production.

Cytotoxic CD8+ T cells and NK cells kill virus-infected and tumor cells

Virus-specific CD8+ CTLs are the major effector cells for eliminating established viral infections. NK cells also lyse virus-infected cells and tumor cells. Therefore, both cell types are often summarized as cytotoxic lymphocytes. It seems that both cell types share common mechanisms for antiviral and antitumor defense.[20-22] For example, both cell types secrete the cytotoxic protein perforin, along with granzymes. Together, they kill infected cells and tumor cells on cell-to-cell contact. This is a thoroughly controlled process to kill only the diseased target cell (not healthy neighbor cells).

The most important difference between the CTL and NK cells is that NK cells do not have a TCR; NK cells recognize virally infected cells by their ability to recognize and lyse virally infected cells by other receptors showing a more general specificity for pathogen-induced changes in tissue cells (e.g., intracellular infection or neoplasia). Other NK cell receptors possess inhibitory activity, enabling a close control of cell killing.[23]

The major advantage that NK cells have over antigen-specific CD8+ T cells is that they kill target cells without the need for clonal expansion (i.e., without a "lag" phase). Therefore, NK cells effectively limit the early spread of infection.

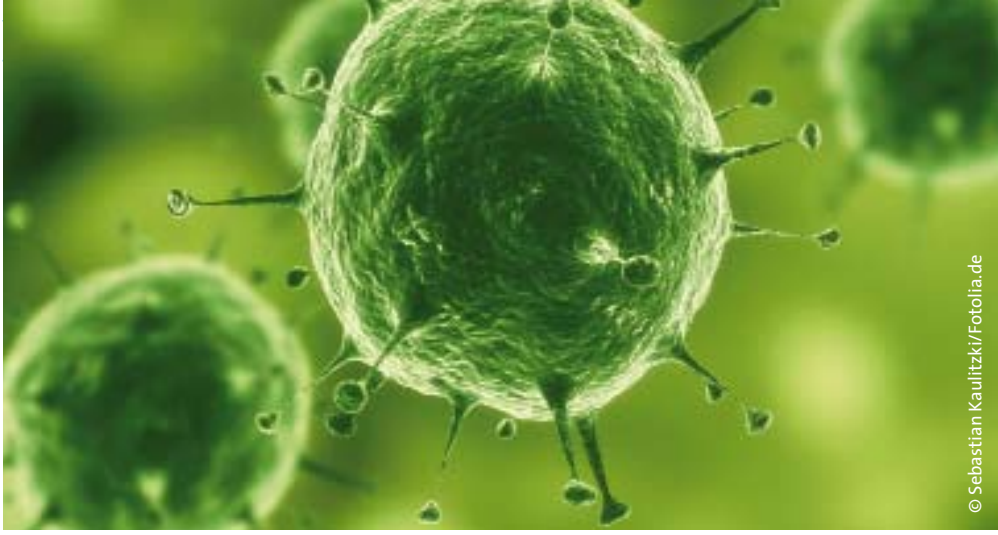
CD8+ CTLs recognize antigens by their TCR in association with MHC-I molecules. In addition, similar to CD4+ T cells, CD8+ T cells need clonal expansion to establish full effector functions.

Role of IFN synthesis in antiviral immunity

In addition to cell contact-dependent killing mechanisms, soluble mediators released during viral infection of cells directly stimulate the production of IFNs, of which type 1 IFNs (α and β) possess the strongest direct antiviral activity. Type 1 IFNs are produced by many cell types and cause an "antiviral state" in the infected cells; this state is characterized by inhibition of viral replication and cell proliferation. Type 1 IFNs also enhance NK cell activity to lyse target cells and improve antigen presentation in APCs.

Multiple ways to control T-cell activation

T-cell responses do not only consume lots of energy by clonal expansion but are also highly powerful when it comes to destroying tissue cells. Taken together, the costs of false alarms are high; therefore, such a process needs to be controlled strictly.



The elimination of viruses is a real challenge for the immune system.

The ability of T cells to become activated primarily depends on the signal strength received by the TCR; therefore, only those T cells showing the best binding fit to the antigen will become fully activated. Another potent means to effectively control T-cell activation is by "costimulation." This is accomplished by a series of costimulating counterreceptors on the APC surface binding to ligands on T cells. These add positively and negatively to the regulation of the proliferation and differentiation of a given T-cell clone. One of the best characterized costimulation signals is induced by the CD28/CD80 receptor pair.[24]

Finally, much progress has been made to characterize the functional diversity of T cells, leading to the current description of subpopulations such as the Th cells (Th1, Th2, and Th3) and the regulatory T cells (which have been extensively reviewed by Kalinski and Moser[25] and Belkaid[26]). Briefly, each of these subtypes of T cells expresses its own spectrum of activities and soluble mediators that it secretes. The Th1 cells are involved in cell-mediated immunity, the Th2 cells support antibody production and participate in the induction of hypersensitivity, and the Th3 and Treg cells can generally be seen as the protectors ("down-regulators") against reactions that are too strong, outdated, or undesired. Interestingly, the decision as to which type of T cell is generated is probably met largely by DCs, which are able to polarize nondifferentiated T cells toward these functional subtypes. This concept is a subject of continuing debate.[25]

Humoral immunity: the generation of antibodies

B cells are the only cells that produce antibodies (immunoglobulins). As with T cells, each B cell is specific for a particular epitope on an antigen (e.g., protein or carbohydrate). Antigens are specifically recognized by surface-anchored antibodies on these cells. By this B-cell receptor, antigens can be internalized. They are then broken down into fragments and displayed at the B-cell surface together with MHC-II to CD4+ Th cells, which subsequently trigger the activation of the presenting B cell. Activated B cells develop into plasma cells, producing huge amounts of antibodies of the same specificity as the B-cell receptor on their surface originally encoded. In the further course of the immune response, the interaction with T cells causes the B cells to switch their production from immunoglobulin M (IgM), which is always the first to be secreted, to the more versatile IgG.

Effector functions of different antibody classes

In humans, 5 different classes of immunoglobulins, called isotypes, are known (i.e., IgM, IgG, IgA, IgE, and IgD). These immunoglobulins all have different structures and activities. On primary activation, B cells first always synthesize IgM, peaking about 7 to 10 days after initial exposure. Because of its pentameric structure, representing 10 binding sites for antigen per IgM, IgM is particularly potent for agglutinating antigens, enhancing phagocytosis, and activating complement.

At some point during B-cell activation, these lymphocytes can switch from IgM to a different class of antibodies. The most prominent of these antibodies is IgG. Its capacity to "coat" bacteria to improve phagocytosis is called opsonization. IgG also neutralizes microbial toxins, blocks viral adherence to target cells, activates complement, and is the main antibody found on repeated antigen contact.

IgA, on the other hand, is the antibody found primarily in mucus, colostrum, and milk. It protects against respiratory and gastrointestinal tract infectious agents.

Finally, IgE is produced in response to parasites and is also a characteristic mediator of type 1 allergy. In both of these instances, IgE collaborates closely with Th2 cells to shape this particular type of immune response.[27]

Antibodies usually neutralize viruses through binding to their surface, blocking the virus from entering the host cell. In addition, some viral infections lead to the expression of viral proteins on the surface of infected cells. These may bind virus-specific antibodies, leading to complement-mediated lysis, or activate a subset of NK cells to lyse infected cells through antibody-dependent cellular cytotoxicity.

Immune cell memory

Adaptive immune responses lead to a state of long-lived immunity, which is established by the generation of memory cells in the T- and B-cell lineage, exhibiting the same antigen specificity as their parent cells. By contrast, innate defense does not create memory. The advantage of memory cells is that they can be activated upon any repeated contact with their specific antigen much more rapidly than on first contact, which helps to keep reinfection down efficiently.

Intercellular communication during infection

The communication between different immune cells to establish a well-coordinated response during antimicrobial defense, as previously described, would be impossible without the help of the vast array of soluble mediators that evolution elaborated to fine-tune immune responses. They comprise a large number of chemokines, cytokines, and growth factors; there are also whole series of lipid-derived mediators, proteases, antiproteases, coagulation cascade-derived mediators, kinins, and even neurotransmitters. All of these bind to receptors on the cells of the immune system and modify their reactions in a highly controlled manner. Most of these mediators form positive- and negative-feedback signaling loops that timely adjust the general type and the extent of response to the current needs, which in fact differ substantially between the different types and phases of a defense reaction.

Concluding remarks

Immunological knowledge is growing fast. The recent discovery of the TLRs and their functions and of functionally different DC types, the ever-growing list of lymphocyte subpopulations displaying different functions, and the enormous amount of newly discovered mediators have contributed tremendously to our understanding of antimicrobial immunity. In addition, these discoveries are helping us understand the switch from well-regulated immune responses to detrimental conditions such as chronic inflammation. Readers are encouraged to consult one or more of the articles cited herein, which will provide a deeper guide into the complex and highly fascinating world of the immune system, our personal bodyguard.

References

1. Coombes JL, Powrie F. Dendritic cells in intestinal immune regulation. *Nat Rev Immunol.* 2008;8(6):435-446.
2. Sansonetti PJ. War and peace at mucosal surfaces. *Nat Rev Immunol.* 2004;4(12):953-964.
3. Holt PG, Strickland DH, Wikström ME, Jahnsen FL. Regulation of immunological homeostasis in the respiratory tract. *Nat Rev Immunol.* 2008;8(2):142-152.
4. Stuart LM, Ezekowitz RA. Phagocytosis: elegant complexity. *Immunity.* 2005;22(5):539-550.
5. Stuart LM, Ezekowitz RA. Phagocytosis and comparative innate immunity: learning on the fly. *Nat Rev Immunol.* 2008;8(2):131-141.
6. Akira S, Takeda K. Toll-like receptor signalling. *Nat Rev Immunol.* 2004;4(7):499-511.
7. Iwasaki A, Medzhitov R. Toll-like receptor control of the adaptive immune responses. *Nat Immunol.* 2004;5(10):987-995.
8. Kawai T, Akira S. Innate immune recognition of viral infection. *Nat Immunol.* 2006;7(2):131-137.
9. Nathan C. Neutrophils and immunity: challenges and opportunities. *Nat Rev Immunol.* 2006;6(3):173-182.
10. Carroll MC. The complement system in regulation of adaptive immunity. *Nat Immunol.* 2004;5(10):981-986.
11. Finlay BB, McFadden G. Anti-immunology: evasion of the host immune system by bacterial and viral pathogens. *Cell.* 2006;124(4):767-782.
12. Chaudhuri J, Alt FW. Class-switch recombination: interplay of transcription, DNA deamination and DNA repair. *Nat Rev Immunol.* 2004;4(7):541-552.
13. Schlissel MS. Regulating antigen-receptor gene assembly. *Nat Rev Immunol.* 2003;3(11):890-899.
14. Vyas JM, Van der Veen AG, Ploegh HL. The known unknowns of antigen processing and presentation. *Nat Rev Immunol.* 2008;8(8):607-618.
15. Reis e Sousa C. Dendritic cells in a mature age. *Nat Rev Immunol.* 2006;6(6):476-483.
16. Shortman K, Naik SH. Steady-state and inflammatory dendritic-cell development. *Nat Rev Immunol.* 2007;7(1):19-30.
17. Acuto O, Bartolo VD, Michel F. Tailoring T-cell receptor signals by proximal negative feedback mechanisms. *Nat Rev Immunol.* 2008;8(9):699-712.
18. Waldmann TA. The biology of interleukin-2 and interleukin-15: implications for cancer therapy and vaccine design. *Nat Rev Immunol.* 2006;6(8):595-601.
19. Taniguchi T, Minami Y. The IL-2/IL-2 receptor system: a current overview. *Cell.* 1993;73(1):5-8.
20. Voskoboinik I, Smyth MJ, Trapani JA. Perforin-mediated target-cell death and immune homeostasis. *Nat Rev Immunol.* 2006;6(12):940-952.
21. Trapani JA, Smyth MF. Functional significance of the perforin/granzyme cell death pathway. *Nat Rev Immunol.* 2002;2(10):735-747.
22. Orange JS. Formation and function of the lytic NK-cell immunological synapse. *Nat Rev Immunol.* 2008;8(9):713-725.
23. Kumar V, McNerney ME. A new self: MHC-class-I-independent natural-killer-cell self-tolerance. *Nat Rev Immunol.* 2005;5(5):363-374.
24. Acuto O, Michel F. CD28-mediated co-stimulation: a quantitative support for TCR signalling. *Nat Rev Immunol.* 2003;3(12):939-951.
25. Kalinski P, Moser M. Consensual immunity: success-driven development of T-helper-1 and T-helper-2 responses. *Nat Rev Immunol.* 2005;5(3):251-260.
26. Belkaid Y. Regulatory T cells and infection: a dangerous necessity. *Nat Rev Immunol.* 2007;7(11):875-888.
27. Anthony RM, Rutitzky LI, Urban JF Jr, Stadecker MJ, Gause WC. Protective immune mechanisms in helminth infection. *Nat Rev Immunol.* 2007;7(12):975-987.