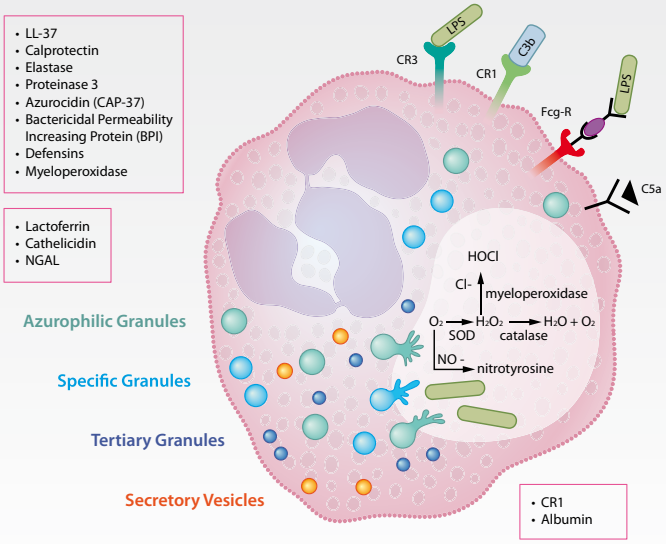
Hycult Scope

Neutrophil proteins in innate host defense

Neutrophils represent roughly 50-70 % of the total white blood cell population in human. Together with basophils and eosinophils, they compose the polymorphonuclear cell family (PMNs). Structural hallmarks of a neutrophil are its multilobed nucleus and the abundant storage granules in the cytoplasm. Neutrophils undergo a process called chemotaxis, which allows them to migrate toward sites of infection or inflammation. They are attracted by cytokines expressed by activated endothelium, mast cells, and macrophages. Cell surface receptors are able to detect chemical gradients of molecules such as interleukin-8 (IL-8), interferon gamma (IFN-gamma), C3a/b and C5a, which these cells use to direct the path of their migration. Upon arrival, complement receptors (for instance CR1, CR3; Figure 1) and LPS-recognition-pathways are activated, resulting in phagocytosis, destruction of infectious agents or initiation of an adaptive immune response. Activation leads to an oxidative response, which consists of the production of radical oxygen species involving myeloperoxidase, hypochlorous acid (HOCl), chloramines and nitrotyrosine. Furthermore, a non-oxygen dependent pathway is triggered and consists of the release of preformed proteins stored in granules in the phagolysosome. Calprotectin, MPO, NGAL, bactericidal and cytotoxic proteins (defensins, BPI, lactoferrin), proteolytic enzymes (elastase, proteinase 3, lysozyme, arginase I) and surface receptors are all proteins stored in granules in the neutrophil cytoplasm. The process of neutrophil activation has to be regulated tightly, since over-activation of neutrophils can mediate tissue damage.

Immunological relevance

- Calprotectin, lactoferrin, MPO, Elastase and Elafin levels are useful markers in inflammatory bowel diseases (IBD)
- NGAL is a marker for acute renal injury after cardiac surgery
- First line of host defense by releasing antimicrobial peptides
- People suffering from neutropenia (low neutrophil counts) predispose heavily for infection
- Production of cytokines upon neutrophil activation enhances the immune response



LEUKOCYTES

Assays leukocytes and neutrophils (2x96 det.)

Specifity	Cat. #	
Alpha-Defensin 1-3, Human, ELISA	HK317	*Unique*
Arginase-I, Human, ELISA	HK322	
BPI, Human, ELISA	HK314	*Unique*
Calprotectin, Human, ELISA	HK325	
Elastase, Human, ELISA	HK319	
Lactoferrin, Human, ELISA	HK329	
LL-37, Human, ELISA	HK321	
MPO, Human, ELISA	HK324	
MPO, Mouse, ELISA	HK210	*Unique*
MPO, Rat, ELISA	HK105	*Unique*
NGAL, Human, ELISA	HK330	
Nitrotyrosine, ELISA	HK501	*Unique*

In this Issue:

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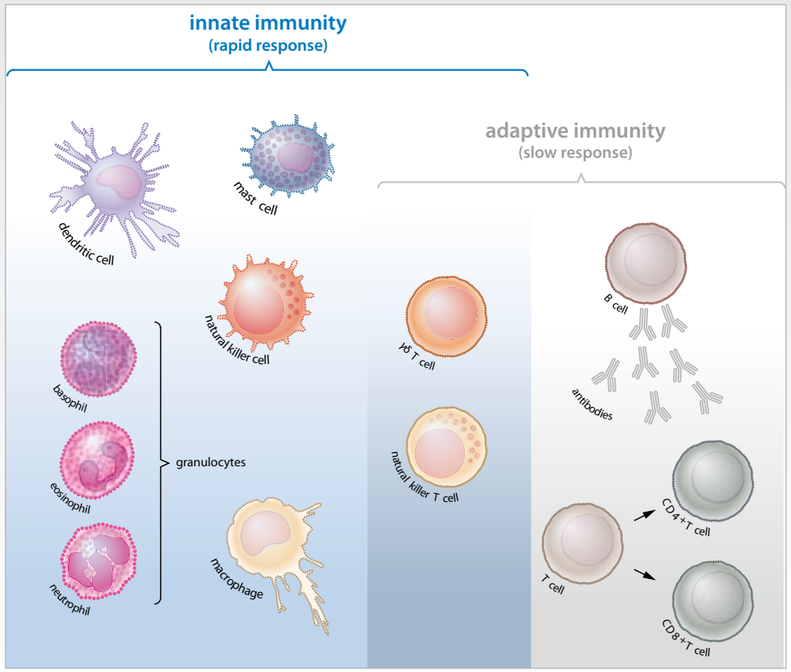
Eosinophils, basophils and mast cells

Eosinophils, basophils and mast cells are white blood cells involved in the innate immune responses especially associated with allergic reactions, asthma and parasite infections. They are key effector cells in IgE-associated immune response and participate in the innate immune responses to many pathogens, including bacteria and viruses. These cells contain small granules within the cellular cytoplasm, which upon stimulation release their contents (degranulation), predominantly mediators and cytokines. Basophils (< 1 % of white blood cells) and eosinophils (~1.6 %) are granulocytes that develop during hematopoiesis in the bone marrow before migrating into blood. Mast cells originate also from hematopoietic stem cells in the bone marrow, but they complete their differentiation in the peripheral tissues.

Immunological relevance:

- Mast cells have been implicated in rheumatoid arthritis, multiple sclerosis, atherosclerosis, inflammatory bowel disease and angiogenesis
- Activation of mast cells initiates allergic inflammatory reactions in response to antigen
- Mast cells can function both as negative as well as positive regulator of immunity
- Basophils enhance immunological memory responses
- Basophils are required for adoption of T-helper 2 fate in response to allergens with protease activity
- Eosinophils have recently been implicated in antigen presentation to T cells
- Eosinophils are also involved in many other biological processes, including postpuberal mammary gland development, estrus cycling, allograft rejection and neoplasia

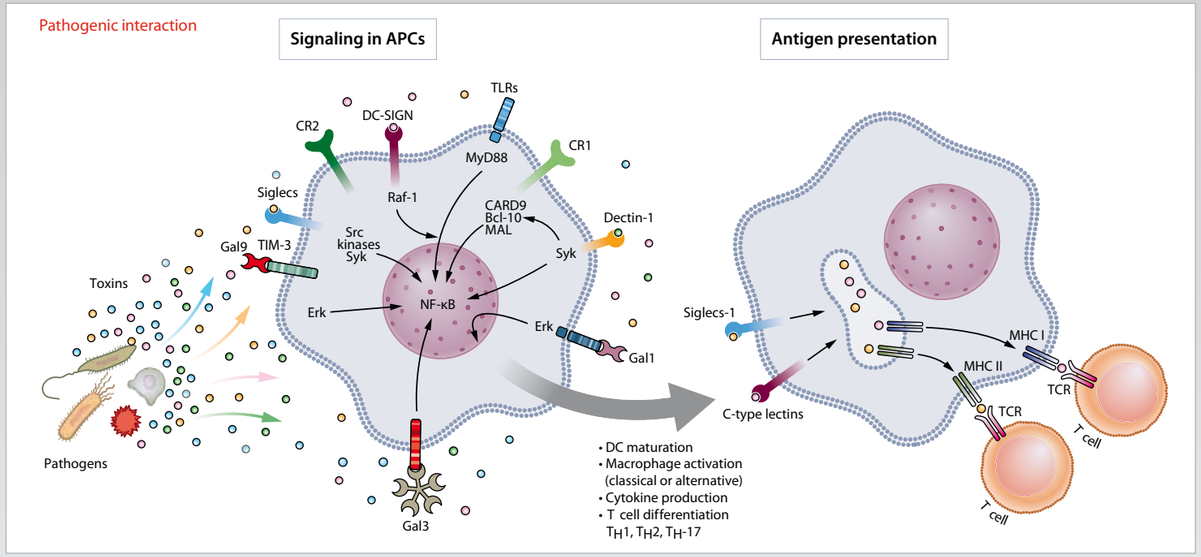
Product	Applications	Cat. #
Monoclonal antibody against Mouse CD200R3, clone Ba103	W IP FC FS	HM1103
Monoclonal antibody against Mouse CD200R3, clone Ba91	W IP FC FS	HM1104



In the heat of bleach

The fearsome bacteria-fighting power of bleach lies in its ability to unfold proteins, a process that resembles the effects of boiling an egg: proteins are damaged and begin to unfold, exposing once hidden 'sticky' amino acids that can interact with other damaged proteins, eventually forming intractable, nonfunctional aggregates. Bleach is a solution of sodium hypochlorite, which is rapidly dissociated into the highly reactive hypochlorous acid. Neutrophils produce this acid to kill bacteria. In that view, bleach and the immune system fight bacteria using a same mechanism.

Winter, J et al; Bleach activates a redox-regulated chaperone by oxidative protein unfolding. Cell 2008, 135: 691



Dendritic cells

Dendritic cells (DC) are the professional antigen presenting cells (APC) of the immune system. They induce adaptive immune responses by activation of naive T cells. DCs also have important effector functions during the innate immune response, such as pathogen recognition and cytokine production. In fact, DC represent the crucial link between innate and adaptive immune responses. Immature DCs reside in the tissue (for example, in the skin, lungs and gastrointestinal tract) and mature following exposure to pathogens. DC activation by TLR ligands induces the formation of endolysosomal tubules, which contain several proteins. Subsequently, these proteins are delivered to the cell surface, where they are available to CD4+ T cells for activation.

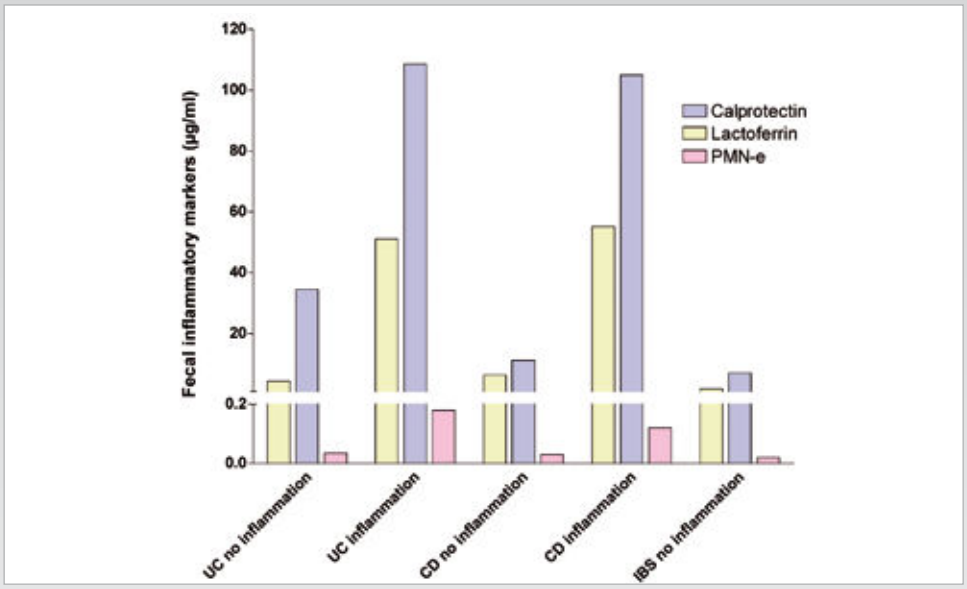
Immunological relevance:

- DCs are crucial in the balance between tolerance and immune responses
- So-called killer DCs mediate another important innate function: the cell-mediated cytotoxicity
- DCs play a role in protection against Plasmodium infection via cross-presentation
- Minor imbalances in the feedback control of DCs are associated with autoimmunity and allergic reactions in genetically prone individuals
- DC mediated therapy can be used for the immunoregulation of type 1 diabetes mellitus, allergic disease, cancer, and autoimmune diseases

B: Inhibition of biological activity
F: Frozen sections
FC: Flow cytometry
FS: Functional studies
IA: Immuno assays
IF: Immuno fluorescence
IP: Immuno precipitation
P: Paraffin sections
W: Western blot

Product	Applications	Cat. #
Dectin-1, Mouse, mAb 2A11	B FC IP	HM1067
MHC Class II, Mouse, mAb ER-TR3	F FC P	HM1087
MHC Class I, Mouse, mAb ER-HR52	F FC	HM1090
MHC Class I, Mouse, mAb ER-MP42	F FC	HM1091
Siglec-H, Mouse, mAb 440c	B F FC	HM1075
DC-SIGN (CD209), Human, mAb DCN47.5	FC FS	HM2209
CR1 (CD35), Human, mAb 31R	F FC IA IP P W	HM2107
CR2 (CD21), Human, 21B9	FC W	HM2139
Galectin-3, Human, mAb B2C10	F FC FS IA P W	HM2186
TLR2 (CD282), Mouse, mAb 6C2	FC IP IF	HM1047
TLR2 (CD282), Mouse, mAb mT2.7	F FC IP	HM1058
TLR2 (CD282), Mouse, mAb T2.5	B F FC IP	HM1054
TLR4/MD2, Mouse, mAb MTS510	B F FC IP	HM1029
TLR9 (CD289), Mouse, mAb 5G5	F FC IA W	HM1042
TLR2 (CD282), Human, mAb TL2.1	B F FC IA IP P W	HM2064
TLR4 (CD284), Human, mAb HTA125	B F FC IP	HM2068
TLR9 (CD289), Human, Mouse, mAb 5G5	F FC IA W	HM2087

Noninvasive assessment of inflammationin inflammatory bowel diseases (IBD) by measuring neutrophil proteins



Product	Applications	Cat. #
CD200R3, Mouse clone Ba103	W IP FC FS	HM1103
CD200R3, Mouse clone Ba91	W IP FC FS	HM1104

Figure 4: Noninvasive assessment of inflammation in inflammatory bowel diseases (IBD). Median values of fecal calprotectin, lactoferrin and polymorphonuclear neutrophil elastase (PMN-e) for endoscopy-based classification of inflammation for ulcerative colitis (UC), Crohn's disease (CD) and inflammatory bowel syndrome (IBS). Inflammation was defined as endoscopic score of 1; no inflammation was defined as endoscopic score = 0. Data are shown as µg/ml.

(Adapted from: Langhorst J, et al; Noninvasive markers in the assessment of intestinal inflammation in inflammatory bowel diseases: performance of fecal lactoferrin, calprotectin, and PMN-elastase, CRP, and clinical indices. Am J Gastroenterol 2008, 103: 162)

Unique antibodies for functional studies on mouse basophils and mast cells

Special features

- mAb Ba103 is useful for depletion of basophils but not mast cells
- mAb Ba91 and mAb Ba103 added to basophils or mast cells induce degranulation of the cells in vitro
- bulk quantity available of 1 mg (Cat.# HM1103b; HM1104b)
- CD200R3 functions as an activating receptor on mast cells and basophils
- CD200R3 regulates IgE-independent immune responses in cooperation with an inhibitory receptor CD200R
- Basophils are candidate effector cells for the immune response against parasites
- Basophils are a major source of the typical Th2 cytokine IL-4 during parasite infections

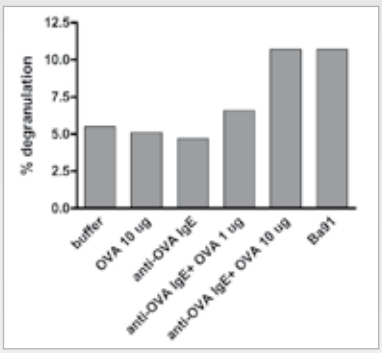


Figure 5: mAb Ba91 induced degranualtion of MC/9 cells. MC/9 cells were incubated with the indicated reagents. Degranulation was calculated by measuring ß-hexosaminidase release (Cat. # HM1104).

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