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THE ROLE OF BLOOD PLATELETS IN IMMUNE SYSTEM

Blood platelets are anucleate cells derived from the megakaryocyte series, and have long been considered only as cells responsible for coagulation and the fibrinolysis process. However, recently more data shows that they are also effector cells in the inflammatory response and important elements of the immunological response. Platelets store and release many biologically active substances, including growth factors, cytokines and chemokines, which actively affect i.a. elements of the immune system, and thus become regulators of immunity and mediators of inflammatory response. Their impact on the immune system cells is also associated with the induction of leucocytes and progenitor cells to the site of pathogen permeation or vascular injury inflow, as well as endothelial cells. Interacting with neutrophils, monocytes and lymphocytes, they not only activate them, but also form platelet-leukocyte aggregates that immobilise pathogens and prevent their spreading. Furthermore, platelets are capable of absorbing pathogens, affecting anti-infection immunity of the system. It is also assumed that the presence of receptors on their surface, such as Toll-like receptors (TLRs), affects their initiation and activity of the immunological response.

Key words: *blood platelets, substances of platelets, receptors of platelets, immune system*

Characteristics of platelets. Platelets are anucleate cells derived from the megakaryocyte series, characterised by high morphological variation. Under resting conditions platelets are oval, without cell processes and surrounded with shapeless glycocalyx which prevents their sticking and adhesion e.g. to the endothelium [18, 19]. Their membrane is connected to an intercellular open canalicular system (OCS) which is necessary for their granule content exocytosis which takes part not only in coagulation and homeostasis, but also in inflammation and also activates the immune system [9,14,17,21,23]. The function of platelets during the immunological response is strictly connected with activation of their surface receptors, including the aforementioned TLR markers and the Fc receptor which recognises immunoglobulins G, E and A as well as selectin P and receptor CD40 [1,3,8,9,11,17,21].

The most important receptors of platelets, in relation to immunological response,

are TLRs, which play a major role in the activation of immune system cells, determining the non-specific response, including elements of natural immunity and, indirectly, also the specific response, including acquired (adaptive) immunity components [1,17,21]. The receptors recognize exogenous and endogenous ligands and bind many various agonists and antagonists, such as antigens of many bacteria, viruses and auto-antigens of vertebrates and invertebrates [1,17,21]. The occurrence of TLRs on the surface of platelets is evidence of the fact that the cells evolve not only as hemostatic cells, but also as immune system cells, including cells recognizing antigens, such as bacterial lipopolysaccharide (LPS). It was evidenced that there are platelets with TLR 7 1,2,4,5,6 and 9 receptors in humans [1,14,16,17,21]. Among them, the role of TLR 4 and 2 was very well recognised and described, as they are also present in mice platelets [1,14,21]. *In vitro* studies revealed that, in response to LPS, TLR 4 affects the release of TNFα and induces an interaction between the activated platelets and PMN cells [21]. Furthermore, it was evidenced that TLR4 induces activation of platelets manifested by increasing their adhesion to PMN cell capacity as a result of LPS action. This leads to robust neutrophil activation and formation of neutrophil extracellular traps (NETs) which also prevents bacterial spreading and is very important in severe sepsis [6, 21]. Also, *in vivo* studies revealed that the expression of TLR 4 and TLR2 on platelets is changed in patients suffering from diseases related to inflammation of the organism, which evidences their role in immunity. This role is also confirmed by TLR9, present on the platelets, which recognizes bacterial DNA as well as other pathogen ligands, and its expression is increased after activation of platelets with thrombin. The presence of TLR 1, 5 and 6 on the platelets was only evidenced on the level of their expression, yet their role in the activation of platelets is still unknown. Another group of platelet receptors are Fc markers and related to them receptors, including FcγRIIA, Glycoprotein VI (GPVI) and C-type lectin receptor (CLEC-2), activation of which occurs by phosphorylation of immunoreceptor tyrosine-based activation motifs (ITAM), present in the cytoplasmic part of these receptors, or by binding to subunit FcRγ [5, 21].

The role of platelets in immunity. Platelets are important in the "formation" of immunity as the first cells occurring at the site of damage, pathogen permeation, as well as at the inflammation site [18, 21]. On the surface, of platelets, there is increased expression of receptors specific to antagonists and ligands binding platelets with activated endothelial cells or the stroma of vessels uncovered after infection or trauma, which results in their activation [18]. The process causes a shape change of such ceils from smooth and oval to ameboid and also a change in their function, as they begin to secrete biologically active substances; this leads to the final and irreversible phase of activation, including aggregation [17, 18, 19]. Cytokines released by platelets, including growth factors and chemokines, regulate immunological response by mobilising and directing leucocytes to the site of the inflammation, and affect the activity of other immune system cells outside the circulatory system [6,14,18,21]. The role of platelets in the modulation of immunological responses has also been noted, pointing i.a. to the action of CD40T as a bridge between innate and adaptive immunity, as it was proven that this molecule induces maturation of dendritic cells (DC) a base for development of adaptive immunity in response to infections [8,16,17,21]. It was also observed that platelets activated in response to viral infections can affect CD40T-depenclent differentiation of B ceils and switching of immunoglobulin classes, as well as the response of active CD8+ T cells by enhancing their cytotoxic properties against viral antigens [8,16,17,21]. It was hypothesized that platelets affect signal transmission to the interior of various immune system cells as it was evidenced that CD40L causes an increase in IgG secretion even

when the number of helper T cells with receptor CD4+ is reduced. Hence the presumption [7,8] in physiological conditions where the number of B and T cells and their antigen-specific response is weakened, platelets take over the functions of *Y* cells and cause signal transmission to B cells, in the direction of stimulating specific humoral response. During the inflammation, platelets also affect leucocytes, which leads to the formation of aggregates with neutrophils, monocytes and lymphocytes [16, 21]. This is possible owing to P-seleetin present on the membrane of the activated platelets, which binds to P-selectin glycoprotein ligand-1 (PSGL-1), present on the surface of leucocytes. Owing to this, the leucocyte CDl1b/CD18-receptoi (Mac-1) is activated and, together with platelet glycoprotein Ilb/IIIa, binds to fibrinogen, causing strengthening of the bond and formation of platelet-leucocyte aggregates [16,18,21]. P-selectin presented on the surface of activated platelets not only allows their binding to leucocytes, but can also affect activation of leucocytes themselves. It was evidenced that P-selectin, as mentioned earlier, apart from the production of superoxide anion radical in neutrophils and monocytes, together with PAF and RANTES, can stimulate monocytes to synthesis of IL-8, MCP-1, ¡ MIPl-α and TNF α [21,23]. Another mechanism by which platelets can impact the activity of immune system cells is their interaction with the triggering receptor expressed on myeloid cells 1 (TREM-1) present mainly on neutrophils and monocytes proved that ligand for this receptor is largely expressed in human platelets [10,17]. They also evidenced that interaction between platelets and neutrophils in the presence of LPS increases TREM-1-dependent neutrophil activation, which results in increased production of reactive oxygen species and release of IL-8 [10]. It was also evidenced that neutrophil-platelet complexes carry out phagocytosis, cytotoxicity or cytolysis more actively than single circulating neutrophils. It was also observed that platelets affect PMN cells causing formation of NETs, mainly in bacterial infections e.g. in severe sepsis [17,21]. *In vitro* studies of this process indicate the major role of platelet TLR4, which is responsible for binding bacterial LPS [6]. It was also evidenced that interaction of platelets and monocytes induces translocation of nuclear factor kappa-light-chain-enhancer of activated B cells (NF-kB), and expression of NF-kB-dependent genes and proteins-in monocytes, involved in the regulation of the inflammation, as a result of action of platelet-derived RANTES, IL-ip and PAF, which increase the secretion of e.g. TNFα, IL-8 and MCP-1 in monocytes [18,21]. *In vitro* studies indicate that platelets also contribute to acceleration of chemokine synthesis in monocytes, and that they can provide the “signal" which triggers monocyte differentiation into DC cells rather than into settled macrophages [21].

The least data refer to the interaction between platelets and lymphocytes; it is only known that activated platelets secrete substances acting as chemoattractants and have an activating effect on helper T cells (Th), cytotoxic T cells (Tc), and NK cells, as well as B cells [13]. As mentioned earlier, T cells can contribute to platelet secretion of RANTES chemokine, as a result of a mechanism dependent on CD40L-CD40, which increases the inflow of lymphocytes to the inflammation site. Also, platelets facilitate lymphocyte aggregation in lymph nodes, which contributes to the formation of immunological memory [21]. Moreover, platelets activate Tc cells in response to viral infections, e.g. during hepatitis B virus infection, as they affect the accumulation of such lymphocytes in the liver [17]. It was evidenced that the role of platelets in anti-infection immunity of the, system is related to the fact that they are capable of destroying pathogens both indirectly – by involving other cells of the immune system, as well as directly, by secretion of substances destroying and damaging bacteria, such as PMP or reactive forms of oxygen [14,16,21]. Furthermore, their effect on pathogens is not only limited to the release of

active substances from granules, but they are also capable of absorbing small extraneous molecules via an open canalicular system, without activation or changing shape [19]. They detect bacteria in the circulatory system and surround them by adhesion, and hence it is assumed that their activity can be helpful in "clearing" microorganisms from the blood, which prevents their spreading across the system, even despite the fact that they are not professional phagocytes [18,19].

Conclusion. Previous studies on platelets, which once were regarded only as megakaryocyte cytoplasm fragments, indicate that they constitute an important element of the immune system, including, natural immunity, and also of acquired immunity by their interaction with B cells. This is due to their unique properties, such as the capacity of expressing receptors and secretion of pro- and anti-inflammatory substances, as well as their capacity of interaction with granulocytes, monocytes and lymphocytes. The data indicate the important role in platelet function as ar essential element in maintaining the antigen integrity of the entire organism.

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