REVIEW

Functional Role of Fertility α_2-Microglobulin

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Fertility α_2-microglobulin is one of the main proteins expressed between the late lutein phase of the menstrual cycle and the first gestation trimester. It is produced by endometrial secretory glandular epithelium and decidual membrane. It is believed to be involved in the preparation to gestation, conception, normal development of the feto-placental system, and initiation of labor. The immunomodulating effect of fertility α_2-microglobulin and its possible involvement in the regulation of fertilization by blocking the spermatozoon reaction with the ovocyte lucid membrane were demonstrated *in vitro*. The data of structural analysis (appurtenance to lipocalines and unique pattern of N-glycosylation) and analysis of the spatial and temporal parameters of the expression in connection with other events in the organism within the same system of coordinates prompted us to investigate the probability of realization of other, so far unknown functions of α_2-microglobulin. The probable mechanisms of realization of the immunomodulating function are analyzed.

Key Words: *fertility α_2-microglobulin; lipocalines; N-glycanes; immunity; angiogenesis; endometrium; decidual membrane; trophoblast; normal killer cells; lectins*

Fundamental theoretical and experimental research has yielded new data on the structure and function of proteins. The progress in this field became possible due to coordinated efforts of molecular and cellular biologists, biochemists, and specialists in x-ray structural analysis. Each protein is characterized by a precisely determined structure and exhibits unique properties providing the realization of strictly determined functions; at the same time, it is an integral component of a universal controlling system which supports the viability of an organism. Each protein possesses individual functions, while the principles regulating the molecular mechanisms underlying their effects are common. The key aspects of protein molecule organization are the recognition of a certain ligand, ability to form complexes in which the specific features of reacting molecules are manifested, combined function of highly organized ensembles of functional groups, arrangement of the microenvironment affecting the nature of chemical transformations, transfer of effects in a molecule or a supramolecular complex, etc. [4].

Some protein molecules are polyfunctional despite their strictly individual structure which is probably due to the presence of spatially discrete recognition sites for several ligands and (for proteins transmitting biological information) by a cellular receptor system, and the differences in cellular response to transmitted signals regulated by programmed support in specialized cells.

Proteins often act as "functional duplicators". This occurs under conditions of an emergency increase in the level of macromolecular partners of a protein when the incorporation of more precision counter ligands for these partners lags behind.

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The ability to duplicate functions saves a biological system in case of a counter ligand defect or deficiency. For instance, gene "knockout" for a cytokine binding to type 1 cytokine receptors carrying a common signal-transferring gp130 chain does not disturb considerably the vital functions of an organism. A T cell in the immune system receives a highly specialized signal via the TCR/CD3 antigen-specific receptor complex and needs an additional costrimulatory signal for an adequate response. The sources of these second signal are different and are largely determined by the microenvironment in which the cell functions.

At present the bank of information on molecular organization of proteins and their structural genes is rapidly increasing. The information about protein isoforms and their allele variants, carbohydrate and lipid components of glyco- and lipoproteins has been accumulated. The use of monoclonal antibodies as probes for the investigation of protein functions and methods of molecular biology ("knockout" and gene transfection, directed site-specific mutagenesis, and recombinant proteins) will in future reveal the functional significance of these molecules.

On the other hand, the gap between the progress in studies of structural organization of protein macromolecules and our knowledge of the functions of detected proteins is increasing. This concerns human proteins, such as fertility α_2-microglobulin (FAMG).

The aim of our review is to validate the strategy of investigation of the biological function of a protein of a known structure, as exemplified by FAMG. The following approaches to the evaluation of the functional role of FAMG are proposed:

1) Structural analysis based on appurtenance to a family of molecules with known functions;

2) Analysis of spatial and temporal parameters of protein expression in connection with other events realized in the given system of coordinates.

Let us begin with the history of FAMG discovery and our knowledge of this protein. An antigenic component with electrophoretic mobility of α_2-globulins was detected in water extracts of human placenta by D. D. Petrunin *et al.* in 1976 [5]. Since this protein was identified in menstrual blood and in the endometrium at the secretory phase of the cycle and in the semen and seminal vesicles, it was termed fertility α_2-microglobulin [1,2,6]. Then a protein with similar characteristics was described as uterine α-globulin, progesterone-dependent endometrial protein, placental protein 14, and endometrial protein 15 [7,10,27,54]. Later it was shown that these proteins are identical. The term FAMG has been used in Russian publications [3,6]. FAMG is secreted by secretory decidual endometrium [28,30], fallopian tube epithelium [32], and seminal epithelium [31], early erythroid precursors and megakaryocytes in the bone marrow, and peripheral blood platelets [33,41]. Recently, FAMG has been referred to as glycodelin, to emphasize the unique nature of the oligosaccharide component of this glycoprotein [18].

The term FAMG defines a family of glycoproteins recognized by anti-pp14 antibodies. These glycoproteins possess a unique DNA sequence, but are polymorphous inside the family [30]. The differences between the family members are due to posttranslation modifications [18,40]. Molecular weight of native FAMG is about 50 kD; it is a dimer.

The time and spatial expression of FAMG in female reproductive organs are strictly regulated [6,46]. The corpus luteus hormone progesterone, whose effect is mediated by unknown mechanisms, is believed to play the key role in this regulation. FAMG is not expressed in proliferating endometrium, during the cycle appears in secretory glandular endometrium; its level gradually increases from the 4th day after ovulation and reaches the peak by the 12th day [6]. Thus, its expression during the periovulatory period of the cycle is minimal. During the embryo implantation, the production of FAMG in decidual tissue amounts for 4-10% of the total secreted protein [29]. FAMG is secreted into the amniotic cavity, and its content in the amniotic fluid reaches the maximum between weeks 10-14 of gestation. Its serum content in nonpregnant and pregnant women is much lower than in the amniotic fluid and decidual tissue [46]. Changes in serum content of chorionic gonadotropin and FAMG during pregnancy strongly correlate with each other. Intramuscular injection of chorionic gonadotropin to sterile women induced the production of FAMG in the endometrium. Together with progesterone, gonadotropin may indirectly affect the production of FAMG in the endometrium. When the chorionic villi invade uterine mucosa, their cells start producing chorionic gonadotropin which triggers the synthesis of FAMG in the adjacent decidual tissue [6].

FAMG belongs to lipocalines [20]. Crystallographic analysis of some proteins of this family has shown that a lipocaline molecule has an 8-stripe antiparallel barrel-like β structure with an internal ligand-binding site open from one side. Individual stripes of β plicated layer are bound to each other by β folds (L2-L7), typical β "pins", while L1 is a large W-loop forming the "roof" of the barrel, crooked towards the internal ligand-binding site.

This loop conformationally varies in different lipocalines but retains the common shape, size, and position toward the molecule. A short 3/10 spiral immediately preceding stripe A covers the "bottom" of the barrel. α-Spiral, curved parallel to stripes G and H, is outside the last stripe of β barrel.

The members of the lipocaline family are minor secretory proteins characterized by some common properties: the ability to bind minor hydrophobic molecules and specific receptors of cell surface and to form complexes with soluble molecules. The members of the family initially characterized as a group of transport proteins perform a variety of functions. Lipocalines participate in the retinol transport, cryptic pigmentation in invertebrates, the sense of smell, pheromone transport, and prostaglandin production. They are involved in the cellular homeostasis regulation, modulation of immune response, and, as protein carriers, in total clearance of endo- and exogenous compounds. Structural organization of lipocalines, namely, a large cup-like cavity in the β barrel and a loop hanging over the entry into this cavity, makes these molecules ideally adapted for binding various ligands [20]. The set of amino acid residues forming the walls of the ligand-binding channel and the loop-like lid of β barrel determine sufficient variety within the family; therefore, the ligand-binding sites vary within a wide range.

Some lipocalines bind hydrophobic ligands deeply submerged in the intramolecular channel, while others interact with large and relatively hydrophobic ligands through a site exposed with a solvent in the lid of β barrel. Among the known lipocaline ligands are molecules with important biological functions: retinoids, arachidonic acid, and steroids. Experimental validation of the binding of minor hydrophobic molecules by a certain lipocaline does not allow us to conclude that the transfer of these ligands is an evidence of transport or other specialized function of the lipocaline.

Some lipocalines bind to specific receptors on the cell surface and are internalized by receptor-mediated endocytosis. Presumably, three conservative motives determining the molecule appurtenance to lipocalines, being spatially approximated, participate in the formation of a common binding site for cellular receptors on the bottom of the lipocaline barrel.

The ability to form complexes with soluble molecules is another important characteristic of lipocalines. In some cases the binding is noncovalent, as with retinol-binding protein forming complexes with thyroxin-transferring protein. Another lipocaline, apolipoprotein D, also forms disulfide-bound complexes with apoA-II in high-density lipoproteins and with apoB 100 in low- and very low-density lipoproteins. Neutrophilic lipocaline (NLC) covalently binds type IV collagenase through intermolecular disulfide bonds. Lipocaline C8-γ, a subunit of the C8 complement component, covalently binds to C8-α through a disulfide bridge. C8-γ forms a site of reception with C8-binding protein (a homologous restriction factor expressed on erythrocytes and other cells) [19,22].

The ability of some lipocalines to form complexes simultaneously with several unrelated macromolecules is noteworthy. α_1-Macroglobulin forms covalent complexes with human immunoglobulin A and fibronectin and with rat α_1-inhibitor III. In the latter case the formation of α_1-macroglobulin-α_1-inhibitor III complex cancels the inhibitor function, presumably by preventing its cross-binding to proteinases. α_1-Macroglobulin, NLC, and C8-γ are believed to decrease the activities of plasma proteins via complex formation. These intermolecular reactions are mediated by the loop hanging over the open end of a lipocaline molecule. The variability of the length, conformation, and primary structure of the loop explain the ability of lipocalines to form high-affinity and selective complexes.

The structural organization of lipocalines allows an *a priori* assumption of a polyfunctional nature of at least some representatives of this family. Lipocalines in general are classified as extracellular transport proteins. A classical prototype retinol-binding protein is the only retinol vector in the plasma binding transretinoic acid as a physiological ligand. Retinol-binding protein is believed to facilitate the transport of insoluble retinol deposited by the liver to the peripheral tissues and protects it from oxidation. In addition, the production of this protein regulates the release of retinol by the liver and guarantees the specificity of its capture by target cells. The hypothesis on analogous transfer of retinoids by FAMG was not confirmed [21]. It was hypothesized that FAMG is involved in directed transport of prostaglandins E (PGE). Pheromones, pigments, and proteins are lipocalines involved in olfactory processes and taste sensations, production of prostaglandin D, regulation of cell growth, and immunomodulation. Changed level of many plasma proteins during the acute-phase response and complex physiological reaction to stress and inflammation plays an important role in many diseases.

The acute-phase proteins α_1-acid glycoprotein (AGP), NLC, α_1-macroglobulins, and probably FAMG, belong to lipocalines. These proteins are characterized by anti-inflammatory properties, prevent tissue destruction, and ensure transport of biologically significant factors. The only known function of FAMG is its involvement in the immune

system regulation [12,48]. FAMG suppresses proliferation of T cells in response to phytohemagglutinin [49], reaction of mixed lymphocyte culture [11], and suppresses the activity of natural killer cell cells. Interleukin-2 (IL-2) does not stimulate mitogen-induced T cell proliferation in the presence of FAMG. Antibodies to FAMG abolish the immunosuppressor effect of decidual tissue extracts. Decidual extracts containing FAMG suppress the production of IL-1 and IL-2-receptors in a population of cells stimulated by mitogen [47]. Purified FAMG suppresses the lysis of K562 target cells by human natural killer cells [45]. In addition, FAMG stimulates the production of IL-6 anti-inflammatory cytokine by endometrial epithelial cells [36].

Are there common or overlapping approaches to realization of the immunomodulating effects of the lipocalines FAMG, NLC, α_1-macroglobulin, and AGP? α_1-Macroglobulin expressed in the liver suppresses antigen-induced polyclonal proliferation of lymphocytes and spontaneous migration of neutrophils *in vitro* and inhibits granulocyte chemotaxis in a concentrated gradient of cytokines released by activated macrophages and T and B cells. Its immunosuppressor activity is probably mediated by the carbohydrate component constituting 22% of the total molecular weight. As a product of neutrophils and liver cells, NLC possesses anti-inflammatory and immunosuppressor properties by regulating the gelatinase function in neutrophils and by binding and probably neutralizing some products of bacterial origin, specifically, lipopolysaccharide (inflammatory cytokine inductor) and N-formyl-methionyl-leucyl-phenyl alanine, a potent chemoattractant. In mice the NLC analog 24p3 is produced in the uterus; its level reaches the maximum during labor. Lipocaline or α-acid glycoprotein (AGP or orosomucoid) is a quantitatively predominating liver protein, that accumulates in the foci of inflammation. It inhibits platelet aggregation, participates in wound healing, and suppresses neutrophil activation and phagocytosis. The acute-phase response is associated with characteristic changes in orosomucoid glycosylation. It acquires pronounced immunomodulating properties. The content of an orosomucoid with the carbohydrate component, sialyl-Lewis X, is increased during acute inflammation. AGP inhibits E-selectin-mediated granulocyte invasion into inflamed epithelium. Thus, structural organization of lipocalines allows some of them to perform multiple functions due to the presence of several dissociated sites for the binding of different ligands. High-level glycosylation is significant for some immunomodulating lipocalines; in many cases the immunomodulating effect is realized through carbohydrate components.

Philogenetically, among the earliest acquisitions are the reactions between the proteins expressed on the cell surface and their carbohydrate counterligands represented on the cellular and extracellular matrix. By reacting with glycoconjugates (glycoproteins and glycolipids), carbohydrate-binding proteins glycosyl transferases and lectin-like molecules ensure mutual recognition of cells, which may permit the transfer of positional information and trigger certain cell functions. The protein-carbohydrate reactions play an important role in realization of a variety of functions in an adult organism. Fixation and permanent modification of these early philogenetic acquisitions is an important characteristic of biological systems.

Structural analysis of the carbohydrate component of FAMG molecule revealed the unique nature of FAMG oligosaccharides. FAMG is glycosylated by two sites: Asn-28 and Asn-63. Site Asn-28 carries a mannose-rich hybrid and complex structures, while the other site is occupied solely by complex glycanes [18]. The major nonrecovering epitopes of complex glycanes are Gal-β_{1-4} GlcNAc (lacNAc), GalNAc-β_{1-4} GlcNAc (lacdiNAc), NeuAc-α_{2-6} GalNAc-β_{1-4} GlcNAc (sialylated lacNAc), NeuAc-α_{2-6} GalNAc-β_{1-4} GlcNAc (sialated lacdiNAc), Gal-β_{1-4} (Fuc-α_{1-3}) GlcNAc (Lewis X), and GalNAc-β_{1-4} (Fuc-α_{1-3}) GlcNAc (lacdiNAc analog of Lewis X).

The majority of N-bound FAMG oligosaccharides are not typical of mammalian glycoproteins. Nonsulfated lacdiNac structures in FAMG were previously detected in three categories of mammalian glycoproteins. The first category includes lactotransferrin [16], high-density lipoprotein receptor CD36 [43], and butyrophyllin [53]. The second category includes some serine proteases, such as tissue inactivator of Bowes melanoma plasminogen [13] and urokinase plasminogen activator [8], which convert plasminogen into plasmin, and the urine callidinokinase [55], which cleaves kininogens with a release of the vasoactive peptide lysylbradykinin. Recombinant human protein C expressed by renal cell strain 293 belongs to the third category [59].

Unique glycosylation of FAMG suggests that this lipocaline can act as a productive glycoconjugate counterligand for certain cell surface lectin-like molecules. FAMG isolated from the amniotic fluid (glycodelin A) effectively suppresses spermatozoon binding to the oocyte lucid membrane. Its testicular analog glycodelin S carries other glycoforms of N-glycanes and does not prevent spermatozoon binding to the oocyte [40].

The important role of oligosaccharides in cell-mediated adhesion in inflammatory and immune response led to the hypothesis that some FAMG N-

glycanes react with bioactive receptor proteins of the immune system [18], for example, CD22, a molecule associated with the β cell receptor complex which participates in cell-to-cell adhesion and activation. CD22 binds the NeuAc-α_{2-6} Gal-β_{1-4} GlcNAc sequence [51] and NeuAc-α_{2-6} GalNAc disaccharide [50] with similar affinity. Presumably, FAMG reacts with CD22 through antigens containing these carbohydrate chains by inhibiting specific immunoadhesion and activation. Impaired proliferative response of T lymphocytes during the blocking of the reaction between CD22 on B cells and its ligand on T cells by monoclonal antibodies confirms this hypothesis. As orosomucoids, FAMG may block the E-selectin-mediated adhesion of neutrophils to the endothelium. FAMG from the amniotic fluid (glycodelin A) blocks spermatozoon binding to the oocyte lucid membrane, while spermal glycodelin S differing from it by the composition of the carbohydrate component does not prevent this reaction.

Thus, on the basis of experimental data it can be concluded that FAMG supports local immunosuppression in the fetoplacental system and regulates the process of fertilization. Let us formulate some hypotheses on the functional significance of FAMG, based on the correlation between the spatial and temporal parameters of its expression and the events occurring in an organism.

The FAMG production markedly increases on days 6-7 after fertilization and reaches the plateau on days 20-21. On day, 9 lacunae appear in the trophoblastic testa, into which maternal blood flows as a result of small vessel and capillary erosion. Fetoplacental vessels start to form in a 10-day-old embryo. Vascular rudiments capable of independent growth (angioblastic structures) are detected on days 13-15 after fertilization. Vascularization of chorionic villi is completed on weeks 8-10, i.e., simultaneously with a drastic rise of FAMG content in the amniotic fluid. The coincidence of the terms of FAMG expression and formation of vascular connection between the mother and fetus suggests the participation of FAMG in angiogenesis.

Angiogenesis is a multi-step mechanism involving through the growth factors activation of endothelial cells, cell migration and proliferation, formation of the vascular tube and external layer from smooth muscle cells. Growth factors secreted by macrophages and fibroblasts or released after capillary injury activate the endothelium of adjacent capillaries and induce the production of proteases and antiproteases, paving the way for vessels and remodeling the forming extracellular matrix. Biantennal N-bound oligosaccharides carrying GalNAc-β_{1-4} (Fuc-α_{1-3}) Glc Nac inhibit the binding of blood cells (neutrophils, normal killer cells, and monocytes) to E-selectin on the endothelial cell surface, thus creating the primary barrier for adhesion and subsequent release of cells from blood vessels. Since free E-selectin (extracellular domains) is a chemoattractant for endotheliocytes, it is involved in angiogenesis [35]. FAMG probably binds free E-selectin, which prevents endotheliocyte chemotaxis, thus modulating the formation of vascular connection between the mother and fetus.

During the menstrual cycle the expression of FAMG and matrix metalloproteinases (matrixins) indirectly depends on progesterone and is almost synchronous. Together with other data, this fact suggests that FAMG inhibits or modulates the activities of matrixins that play an important role in the extracellular matrix remodeling. Neutrophilic lipocaline NLC structurally similar to FAMG forms a covalent complex with gelatinase through a free loop (lid of the β barrel). Metalloproteinase-I tissue inhibitor possesses a similar free loop participating in its reaction with a relevant enzyme [9]. During pregnancy, metalloproteinase inhibitors, as well as FAMG, are present in the amniotic fluid in rather high concentrations. The production of FAMG, matrixins, and their inhibitors is conjugated in time [39,42].

The expression of FAMG matrixins can be blocked through competitive inhibition of the serine protease functions. For example, urokinase/plasminogen activator is activated after its binding to a specific receptor on activated monocytes/macrophages. Plasminogen is converted into plasmin, the main activator of matrixins [42].

FAMG probably competes with plasminogen activator for receptor through a common complex N-glycanes. As a result, matrixin expression is canceled. Here we make an allowance, because the carbohydrate component of plasminogen activator is localized outside its receptor-binding site. Detection of two isoforms of FAMG (pp14.1 and pp14.2) in platelets may indicate their probable role in the functional relationship between the coagulation and immune systems [41]. Plasminogen and plasmin are the components of the coagulation cascade; therefore, FAMG may act as a mediator in the realization of this integrative function. As FAMG, protein C participating in fibrinolysis carries nonsulfated structures of the lacdiNac type. In addition to the regulatory function in the blood clotting system (inhibition of thrombin formation by proteolytically degrading factors Va and VIIIa), activated protein C exerts a potent anti-inflammatory effect by binding to a macrophage specific receptor [23,25]. It protects higher animals from lethal endotoxemia and

suppresses lipopolysaccharide-induced production of tumor necrosis factor-α. Activated protein C inhibits macrophage-dependent production of cytokines and proliferation of T lymphocytes and blocks E-selectin-dependent adhesion of leukocytes on endotheliocytes. FAMG and activated protein C may react with cells through common receptor structures if their common carbohydrate components are involved. By binding to the macrophage receptor for activated protein C, FAMG may mediate the negative signal, thus preventing the macrophage-dependent proliferative response of T cells. FAMG may block the production of IL-1 by macrophages by this mechanism. To what extent the results of FAMG-mediated immunosuppression *in vitro* agree with the presumable role of this glycoprotein as a natural immunosuppressant in the context of reproductive immunology? Expression of HLA molecules presenting a processed antigenic peptide to maternal T lymphocytes is the key event in the recognition of fetal antigen by maternal cells.

During the first trimester of gestation, the majority of cells of the nonvillous trophoblast express HLA-I, HLA-C, and HLC-G associated with β_2-microglobulin. In addition, placental macrophages (Hoffbauer cells) express HLA-I from weeks 8-11, and HLA-II starting from the end of the second trimester. However, these cells do not directly react with maternal leukocytes, because they are separated from them by a layer of syncytiotrophoblast which does not express HLA molecules [37]. T and B cells of the decidual membrane come in contact with fetal tissue extremely rarely. B lymphocytes are scarce in the basal endometrial layer. In normal gestation they are not present in the implantation zone, thus ruling out the probability of local production of antibodies to the trophoblast antigens. T cells are present in the uterine mucosa of the endometrium in nonpregnant women. The count of these cells virtually does not change over the menstrual cycle or pregnancy. The CD4/CD8 ratio changes during the menstrual cycle. CD8 lymphocytes predominate during the late secretory phase, menses, and in chronic miscarriage. In normal pregnancy, type 2 T helper cytokines are produced predominantly in the fetoplacental system of mice.

In vitro stimulation of peripheral blood mononuclear cells in women with chronic miscarriages of unknown etiology by trophoblast extracts results in the production of predominantly type I T helper cytokines, while in parous women with intact reproductive function type 2 T helper cytokines are detected. Type 2 T helper cytokines (anti-inflammatory cytokines) are locally expressed in the uteroplacental and other reproductive tissues in normal gestation [26]. Local deficiency of IL-2 creates conditions for the development of relative T cellular anergy in these tissues [58]. Such anergy can be provided via the immunosuppressive effect of the IL-3 production inhibitor FAMG. The immunosuppressive effect of FAMG on T cell functions has been little studied.

We do not know whether FAMG reacts with T lymphocytes directly or via the antigen-presenting cells (B lymphocytes, macrophages, and dendritic cells). If FAMG is a potential vector of minor lipophilic molecules, this effect is mediated by prostaglandins E transported by it, specifically, by PGE_2. The range of FAMG concentrations suppressing T cell proliferation is such that if the potential sites for PGE binding are saturated, any amount of the bound lipophilic ligand is sufficient for PGE-induced suppression. The intensity of PGE production in fetoplacental tissue during pregnancy suggests that FAMG is responsible for targeted delivery of this immunomodulating factor to T cells. The solution of this problem requires the evidence that PGE binds to FAMG. Lack of immunosuppressive effect of lipid-free FAMG products and recovery of immunomodulating function of FAMG in repeated loading with PGE might prove the rightfulness of such a hypothesis. PGE_2-induced suppression of the response manifests it self in a rapid short term rise of cAMP level in T cells. Similar early changes in the intracellular cAMP concentration induced by FAMG also prove the involvement of PGE in the realization of the suppressor effect. PGE_2 affects the type 1/type 2 T helper ratio, determining the choice in favor of cellular or humoral immunity; therefore, it is desirable to screen the cytokine profiles of FAMG-treated T lymphocytes. Although PGE receptors on lymphocytes have been partially characterized, a detailed description of the presumable effect of FAMG-PGE complexes on T cells is hardly possible at present.

Another mechanism of realization of the immunosuppressor function can be realized through the unique FAMG N-glycanes. This permits a tentative conclusion concerning a potential FAMG counterligand expressed on the target cells: it possesses lectin-like properties. Selectins (molecules responsible for the initial stage of leukocyte adhesion to vascular endothelium), the B cell markers CD22 and CD23, lectin-like receptors of normal killer cells (NK), and some other receptors on the cell surface, whose functional significance is not yet known, possess such properties.

The important role of the natural resistance cells and NK in reproduction has been recognized. Large granular lymphocytes expressing the NK

marker CD56 are present in the endometrium, and their count rises at the beginning of the secretory phase of the menstrual cycle. During gestation, NK constitute up to 30% of stromal cells in the parietal and basal decidual tissue. Large accumulations of these cells are observed in the zones of extensive invasion of interstitial trophoblast at the site of implantation. NK directly contact with the invading trophoblasts, and the increase in their pool in the decidual membrane is due to the inflow of precursor cells from peripheral blood and intense proliferation *in utero*. Decidual NK differ from peripheral blood NK by a higher level of CD56 expression, size, and greater content of granules. These cells retain their cytolytic potential *in vitro* and produce a wide spectrum of cytokines, including colony-stimulating factors, tumor necrosis factor-α, and leukemia-inhibiting factor [37]. Uterine NK are believed to participate in the maintenance of normal pregnancy. Their role may consist in protection of the decidual membrane from excessive penetration of trophoblasts, regulation of the epitheliocyte growth, and maintenance of adequate epithelial differentiation [37]. Cytokines may be involved in the information exchange between decidual NK and trophoblasts.

During the first trimester of gestation, villous and nonvillous trophoblasts express the receptors for many cytokines produced by NK. A nonvillous trophoblast, whose vascular adaptation occurrs according to the same principle as the invasion of malignant cells, directly contacts with maternal blood. Its resistance to NK-mediated cytolysis is determined by the expression of HLC-C and HLC-G on cell surface. The cytolytic program of NK is not realized when they contact with potential target cells carrying HLC-I (the "avoid your kin" principle) [34]. During the reaction with HLC-C or HLC-G, inhibitory receptors on NK surface on the nonvillous trophoblast cells transduce the signal blocking the lythic program of effectors [57]. The expression of HLC-G molecules on nonvillous trophoblast coincides in time with a sharp rise in FAMG level in the amnion, which is associated with anatomical restructuring of the fetoplacental system (celom obliteration with the amnion-chorion contact) in the presence of progressive production of FAMG by decidual tissue [14,37].

FAMG suppressing the lythic activity of NK may duplicate this protective function by simulating or modulating the signal delivered to the effector from class I HLA molecules. Lectin-like HLA-I-recognizing inhibitory or activating molecules containing C-type carbohydrate-specific domains may act as potential NK receptors for FAMG. Specifically, this may be the heterodimer receptor CD94 expressed on the majority of decidual NK, and CD69 and NK G2 receptors [15,17,52]. NK are highly adaptive to cellular environment; the ensembles of the receptors of individual NK are rearranged depending on the density of the class I HLA expression within the framework of certain tissue compartments. FAMG may play an important role in the precision adaptogenic set up of NK in the fetoplacental system.

We have evaluated the significance of biological functions of FAMG and possible mechanisms of their realization by analyzing the structure of this glycoprotein and the events associated in time with its expression in the body. Our conclusions are as follows:

- ❖ The appurtenance to lipocalines suggests with a high degree of probability that FAMG are polyfunctional.
- ❖ The unique composition of FAMG N-glycanes allows us to regard this glycoprotein as a potential counterligand for certain cellular receptors of the lectin series.
- ❖ Spatial and temporal coordinates of FAMG expression and its molecular structure suggest the participation of this lipocaline in the restructuring of immunity, hemostasis, and angiogenesis in gestation.

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