Human Fc receptor polymorphisms in relation to bacterial infection

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ABSTRACT

Human IgG receptors are very heterogeneous. Currently there are three Fc gamma receptor classes specifying at least 12 receptor isoforms to be distinguished. On top of this complexity, Fc gamma receptors (FcγRs) differ between different individuals. Polymorphisms have been identified for two FcγR classes, representing allelic variation of the FcγRIIa (CD32), FcγRIIIa and FcγRIIIb (CD16). The FcγRIIa polymorphisms are now considered to be a heritable risk factor for infectious diseases and some manifestations of autoimmune disorders. A relevant role of the IIIb polymorphism in infectious disease has been suggested, though less convincingly. Detailed analysis of the exact contribution of each of these polymorphisms in relation to previously implicated risk factors for infectious or immunological disease should unravel the pathophysiological contribution of FcγR polymorphisms in the wide variety of factors now being investigated. The information obtained about the multiple genes that impact inflammatory responses of the host – and most likely in reaction to exogenous triggers and pathogens – is becoming overwhelming. The rapid development of molecular techniques makes it possible to determine the incidence of all these individual genetic polymorphisms. Although its relevance will be more difficult to ascertain, the information on all the different genes and their allelic variations involved in the host immune response will be very important for our understanding of infectious disease in modern times.

INTRODUCTION

Human genomics and the rapidly growing insight into the host inflammatory response explain the increasing interest in infectious disease genetics over the last five to ten years. Host genetic factors are major determinants of susceptibility to infectious diseases in humans. Candidate gene studies by association and human genome-wide analysis have been used to identify susceptibility and resistance genes in infectious diseases. However, a single gene defect has rarely been directly related to a devastating diseases, such as interferon-gamma receptor (IFNγR) or interleukin-12 receptor (IL-12R) mutations leading to severe or fatal infections with mycobacterial strains.
In clinical terms, gene polymorphisms of the host immune defence factors appear to have a much broader role and contribution to health and disease. These genetic variants, which modify the regulation or function of the mediators, have been associated with susceptibility to and/or outcome of severe sepsis and septic shock. All steps of the host response to bacteria may be affected by genetic factors. For example, Fcγ receptors (FcγRs), Toll-like receptors (TLRs), CD14 and mannose-binding lectin (MBL) mutations have all been shown to modify the host detection of pathogens leading to pneumococcal infections, Gram-negative bacteria septic shock, and meningococcal disease. Polymorphisms of cytokine genes (TNF-α, IL-1 receptor antagonist (IL-1RA), IL-10, etc.) have been reported to influence the level of secreted mediators and to unbalance the inflammatory cascade. Coagulation response to sepsis may also be affected by gene variants such as the plasminogen activator inhibitor 1 (PAI-1) common functional polymorphism that increases the risk of death from meningococcal infection.
In this paper we will largely focus on the contribution of FcγRs in infectious disease.

PHAGOCYTES

The expression of FcγRs is limited to cells of the haematopoietic lineage (*table 1*). The consequence of IgG binding

Table 1
Opsonin receptors on leucocytes

RECEPTOR	CD	GENE	EXPRESSION
Fc receptors			
FcγR-I	CD64	*FCGRIA*	Monocytes, macrophages, IFN-γ- or G-CSF-stimulated neutrophils
		FCGRIB	-
		FCGRIC	-
FcγR-II	CD32	*FCGRIIA*	Neutrophils, eosinophils, basophils, Monocytes, macrophages Platelets
		FCGRIIB	B lymphocytes Macrophages (neutrophils?)
		FCGIIC	NK cells
FcγR-III	CD16	*FCGRIIIA*	Macrophages, NK cells
		FCGRIIIB	Neutrophils
FcRn		*FCRN*	Syncytiotrophoblasts, intestinal epithelial cells
FcαR	CD89	*FCAR*	Neutrophils, monocytes, mesangium cells
Complement receptors			
CR1	CD35	*CR1*	Erythrocytes, granulocytes, monocytes
CR2	CD21	*CR2*	B lymphocytes
CR3	CD11b/CD18	*CD11B and INTG2*	Neutrophils, eosinophils, basophils, monocytes, macrophages, NK cells
CR4	CD11c/CD18	*CD11C and INTG2*	Monocytes, macrophages, neutrophils, eosinophils, basophils
C1qR?	CD91?		Monocytes, neutrophils, eosinophils, endothelial cells?

to these receptors varies among the cells in relation to their primary function. In the case of infectious disease, it is clear that the phagocytes have the greatest impact on clearance and elimination of bacterial and fungal pathogens, both from tissues and the circulation. The professional phagocytes to be distinguished are mainly formed of neutrophilic granulocytes, monocytes and monocyte-derived tissue macrophages. Neutrophils constitute the major type of leucocytes in peripheral blood, with counts ranging from 40 to 70% under normal conditions. Neutrophils contain numerous proteases and other proteins in their granules and can produce – when activated – a large quantity of toxic oxygen metabolites through the NADPH oxidase activity to kill tissue-invading pathogens.[1,2] Neutrophils protect our bodies against so-called extracellular bacterial and fungal micro-organisms, whereas the macrophages are particularly involved in and equipped for the killing of intracellular pathogens. Neutrophils have a high turnover rate. Once these cells have egressed from the bone marrow, their half-life in the peripheral circulation is about six to eight hours before they leave rather randomly into the extravascular tissue where the neutrophils roam for another 24 hours. In the case of inflammation, circulating neutrophils sense the site of infection, rapidly extravasate and crawl in large quantities towards the invading micro-organisms to ingest and kill them.[2-4] During an infection monocytes follow only later to perform the role of scavenging cell, to engulf the remaining apoptotic neutrophils, and to prepare the local environment for recovery from the tissue damage inflicted during the first wave of neutrophil-mediated inflammation. Macrophages are tissue-dwelling cells derived from monocytes with an estimated half-life of three to four months. Macrophages can be found in any organ or tissue, though in different forms and endowed with different functions. These cells are well situated and actively involved in liver and spleen clearing the blood compartment from aged cells, oxidatively damaged material, as well as filtering out any bacterial pathogen that escaped the defence system of skin or mucosal lining, including the destructive power of tissue-infiltrated neutrophils. Thus, for proper functioning of this first line of defence by phagocytes, a number of prerequisites have to be fulfilled.

PHAGOCYTOSIS AND MICROBICIDAL ACTIVITY

Phagocytes operate in concert with antibodies and complement factors, so-called opsonins. Micro-organisms covered with these opsonins are bound through specific receptors for these opsonins on the plasma membrane (*table 1*). Antibodies bind with their Fab regions to microbial antigens. In this way, the Fc regions of these antibodies are closely packed together. This spatial arrangement enhances complement activation, thus leading to binding of C3b and C3bi to the micro-organisms and subsequent binding of the microbes to the complement receptor type 1 (CR1)

and complement receptor type 3 (CR3) respectively. On the other hand, the proximity of the antibody Fc regions also promotes direct binding of the opsonised micro-organisms to the Fc receptors on the phagocytes.[5]

Two types of Fcγ receptors are present on neutrophils: FcγRIIa and FcγRIIIb. Only after long-term activation of neutrophils by interferons or growth factors is a third Fcγ receptor expressed, i.e. the FcγRI, which binds monomeric IgG with high affinity (*table 2*). In contrast, the constitutively expressed FcγRIIa and FcγRIIIb bind monomeric IgG only with low avidity, but can efficiently bind immune complexes containing multiple IgG molecules. A polymorphism in FcRγRIIa defines the intrinsic ability to recognise all four IgG subclasses or not. IgG2 antibodies, often formed as the most predominant IgG subclass of antibodies against the microbial carbohydrate structures, only react with the so-called L131 type of FcγRIIa (with a leucine at amino acid position 131). As will be discussed later, it is generally assumed that individuals with this isotype are better protected against infections with certain micro-organisms than individuals with the R131 type of FcγRIIa (arginine at position 131).
FcγRIII has two variants, a transmembrane form (FcγRIIIa) expressed on macrophages and natural killer cells, and a neutrophil-specific form (FcγRIIIb) linked to the plasma membrane by a lipid anchor, which allows very rapid redistribution and early localisation of opsonised material. Although there is a polymorphic site in the FcγRIIIb on neutrophils (NA1/NA2 depending on differences in glycosylation), its effect on clinical outcome is as yet not as clear as in the case of the FcγRIIa. In contrast to all other transmembrane FcγR members, the FcγRIIIb on neutrophils can be rapidly shed from the membrane during functional activation or during the process of programmed cell death (apoptosis) in the tissues. The presence of soluble FcγRIIIb plasma in combination with its relatively long half-life renders this molecule a prime candidate for the estimation of total neutrophil mass.[6]
Monocytes express the FcγRI and FcγRIIa whereas macrophages show FcγRI, reduced FcγRIIa and a high level of FcγRIIIa. FcγRIIIa has two polymorphic sites of which the FcγRIIIa-158V/F allotypes show some functional influence on the avidity of IgG subclass binding, of IgG4 in particular.[7] The FcγRIIIa-V/F158 allelic variation possibly influences autoimmune disease in its presentation, course and outcome. Even in one type of disease, the published data of groups of patients with various racial backgrounds are contradictory or at best inconclusive.[8,9]
Studies on the inhibitory action of FcγRIIb were largely limited to B-cell functions. With the current insight from experimental FcγRIIb-knockout models,[10,11] its expression on macrophages (and other phagocytes) and – as a consequence – its impact on the outcome in inflammatory responses as well as on the efficacy of intravenous immunoglobulin infusions in an experimental model for immune thrombocytopenia[12] has been demonstrated. The precise working mechanism of the disease-mitigating role of FcγRIIb should be elucidated in the near future, also in human disease.

MICROBICIDAL ACTIVITY

Binding of opsonised material to surface receptors leads to concentration of such receptors around the area of contact. Subsequently, the cell extends pseudopods that engulf the particle. By consecutive receptor binding, these pseudopods fit tightly around the particle and finally fuse with each other to form a closed membrane vesicle (phagosome) around the particle, within the phagocyte. Neutrophils may overeat themselves in infected areas and die of congestion. Macroscopically, this is manifested as pus formation. Apart from phagocytosis, receptor binding may also start two other processes, i.e. the generation of reactive oxygen compounds and the release of granule

Table 2
Function of Fcγ and complement receptors on phagocytes

			EFFECT		
NOMENCLATURE	EXPRESSION	LIGAND	PHAGOCYTOSIS	MEDIATOR RELEASE	CYTOTOXICITY
FcγRI (CD64)	Constitutive*/long-term activation**	IgG	+	+	+
FcγRII (CD32)	Constitutive, stable expression	IgG	+	+	+
FcγRIII (CD16)	Shed upon activation	IgG	-	+	+
FcγR (CD89)	Constitutive, stable expression	IgA	+	?	?
CR1 (CD35)	Upregulated upon activation	C4b/C3b	+	+	+
CR3 (CD11b/18)	Upregulated upon activation	C3bi	+	+	+
CR4 (CD11c/18)	Upregulated upon activation	C3bi/C3dg	+	+	?

** Constitutive expression on monocytes and macrophages, ** activation-dependent expression on neutrophils.*

contents. Both reactions are localised events in that they are restricted to the release of microbicidal products into the phagosome. However, the secretion of these products begins before the phagosome is closed, and some of the oxygen compounds and granule enzymes may thus escape into the extracellular environment of the neutrophils. Moreover, neutrophils adhering to opsonised material that is too large to be ingested (e.g. immune complexes deposited along basement membranes) may secrete these products in large quantities into the extracellular space. Under such conditions, macrophages can fuse with each other forming large tissue multinuclear histiocytes as an important cell type, as is often present in granulomata.

Degranulation does not occur in resting neutrophils, monocytes, or macrophages. During phagocytosis or adherence of neutrophils to large substrates, intracellular signalling events induce the fusion of granules with the plasma membrane. Neutrophils contain at least two different types of granule; most likely the same holds true for monocytes. In macrophages, the granular content is less apparent. The azurophil granules resemble the lysosomes in other cell types in that they contain acid hydrolases, with a low pH optimum. Moreover, these granules also contain myeloperoxidase (MPO) and a number of serine proteinases (elastase, cathepsin G, proteinase 3). Further, the azurophil granules also contain large numbers of defensins, small peptides with a broad range of bactericidal activity, and bactericidal permeability-increasing protein (BPI), a very potent antibiotic against Gram-negative bacteria. Lysozyme, an enzyme that hydrolyses certain peptidoglycans of Gram-positive bacteria, is present in the azurophil as well as in the specific granules of the phagocytes. Proteins specifically found in the specific granules comprise lactoferrin, an iron-binding and therefore bacteriostatic protein, vitamin B_{12}-binding protein, and the metallo-proteinases collagenase and gelatinase. Neutrophils possess far more granules than monocytes or macrophages. Finally, neutrophils also contain so-called secretory vesicles, which actively exchange their membrane-bound receptors and enzymes with the plasma membrane. In contrast, macrophages have a vesicular system of endosomal trafficking – although not as elaborate as the dendritic cells – for exchange and presentation of antigen as an antigen-presenting cell, which is absent in neutrophils.

Simultaneous with degranulation, a membrane-bound oxidase enzyme complex located in the membrane of secretory vesicles and specific granules can be activated to generate reactive oxygen compounds needed in the killing process. This NADPH oxidase complex is composed of several subunits in the plasma membrane (cytochrome b_{558} α and β subunit: p22-*phox* and gp91-*phox*) and a number of activity-regulating proteins in the cytoplasm (e.g. p47-*phox*, p67-*phox*). Phagocytes at rest do not generate superoxide. Only after opsonin/ligand binding to cell surface receptors is the active NADPH oxidase assembled to generate superoxide in phagocytes. Superoxide spontaneously dismutates into hydrogen peroxide that may then react with chloride ions to form hypochlorous acid (HOCl). This reaction is catalysed by MPO, when released into the phagosome or the extracellular space. HOCl is very toxic for a broad range of micro-organisms but is rather short-lived. However, it can react with primary and secondary amines, and thus give rise to *N*-chloramines, some of which are very stable microbicidal agents. Under normal phagocytosing conditions, neutrophils convert more than 75% of their superoxide into hypochlorous acid and *N*-chloramines, and thus create a highly toxic environment within the phagosomes and in the cell surroundings.

MEANING OF FCγR POLYMORPHISMS IN INFLECTION

Since the FcγRIIa (CD32) is the sole IgG FcR capable of interacting with human IgG2, the main IgG subclass of bacterial capsular polysaccharides, most studies on infections with encapsulated bacteria have focused on the FcγR allotypes: i.e. FcγRIIa-R131 and IIa-H131.
The retrospective study by Bredius *et al*. first showed in a small cohort of 25 children with prior fulminant meningococcal septic shock that almost half of the children were homozygous for FcγRIIA-R/R131, the poor IgG2-binding allotype. This allele frequency was significantly different from its frequency in a healthy white population (44% *vs* 23%, p=0.03). The relevance of this finding was further supported by the fact that neutrophils with the FcγRIIa-R/R131 allotype phagocytised *N. meningitidis* opsonised with polyclonal IgG2 antibodies less effectively than did IIa-H/H131 neutrophils.[13] Another Dutch group had determined the FcγRIIa and FcγRIIIb phenotypes of 48 children with recurrent bacterial respiratory tract infections. FcγRIIa-H/H131 was less than half that observed in 123 healthy adults (p=0.01). IgG2 responses were low in 25 out of 48 patients after immunisation with pneumococcal vaccine. The authors' conclusion was that FcγRIIa polymorphisms contribute to increased susceptibility to infections with encapsulated bacteria in a childhood population with low IgG2 anti-carbohydrate antibodies.[14]

Similar variation in allele frequency was studied in small groups of patients with additional opsonisation defects in the complement system. The distribution of FcγRIIa and IIIb allotypes in 15 individuals with a deficiency in one of the late complement components (LCCD) and in 15 properdin-deficient patients with/without previous meningococcal disease was analysed. The combination of FcγRIIa-R/R131

with FcγRIIIb-NA2/NA2 allotypes was associated with previous meningococcal disease (OR=13.9, p=0.036). No such relation was observed in the properdin-deficient patients.[15] Another study in LCCD patients had previously shown that the distribution of FcγRIIa genotypes and disease demonstrated an apparently clear age effect. The R/R131, R/H131, and H/H131 genotype distribution was 0.14:0.29:0.57 for patients with their first disease episode <10 years of age, as compared with the distribution of 0.21:0.64:0.14 for those with their first episode >10 years (OR=8.0, p<0.05). Meningococcal disease had a more severe course in four out of 31 episodes in patients with the R/R131 or R/H131 allotypes, in contrast to one out of 18 in patients with H/H131 (OR=14.2, p<0.01). Thus, CD32-mediated phagocytosis may restrict the severity of meningococcal disease in LCCD patients with the H/H131 genotype.[16]

In a rather heterogeneous cohort of paediatric and adult HIV-positive individuals, skewing in FcγRIIa R/H131 allele frequency was not observed in relation to pneumococcal invasive disease.[17] In 60 black children with sickle-cell anaemia (SCA) and well-documented bacterial infections, the R/H131 genotype distribution in the 51 individuals with a history of *Streptococcus pneumoniae* infection was also not statistically significantly different from that of the control population. In contrast, however, the H/H131 genotype was unexpectedly overrepresented in the 11 individuals with a well-documented history *of Haemophilus influenzae* type-b infection (64% H/H131, 27% H/R131, 9% R/R131, p=0.002) when compared with ethnically matched controls (14% H/H131, 60% H/R131, 26% R/R131).[18]
A case-control study on the risk and outcome of meningococcal disease in 130 patients with microbiologically proven meningococcal disease and 260 asymptomatic sex-matched controls indicated a lack of skewing in allele distributions of FcγRIIA allotypes.[19] In comparison with meningococcal meningitis, however, both the fulminant meningococcal disease (OR=3.9, 95% CI 1.0 to 16, p=0.04) and sepsis without meningitis (OR=3.0, 95% CI 1.4 to 7.8, p=0.004) were associated more commonly with the FcγRIIA-R/R131 allotype than to those with meningococcal meningitis. Of the 42 patients with the R/R131 allotype, 31 (74%) had an adverse prognostic score, compared with 7% (4 of 59) of those with the R/H131 allotype and 3% (1 of 29) of those with the H/H131 allotype (p<0.0001).

Thus, the FcγRIIA-R/R131 allotype may not act as a strict susceptibility gene but – instead – a severity marker, associated with the more severe forms of meningococcal disease. This remains to be confirmed for other encapsulated pathogens.

CONCLUDING REMARKS

The meaning of polymorphic variations in the genes for the FcγRIIa, FcγRIIIa and FcγRIIIb is far from clear at the moment. Most studies describe small and heterogeneous groups, and may at best tell us something about differences within a certain cohort. Then, the polymorphisms indeed show their most characteristic features, such as an age effect, a difference in outcome and symptomatology, and – presumably – a different treatment effect. The role of these polymorphisms seen as a separate issue in the host defence response indicates a subtle impact, more interesting from the perspective of pathophysiology than for the therapeutic options to date. This may change rapidly as soon as monoclonal antibodies enter the clinics for the reasons outlined above.

In the first phase of inflammatory responses or other forms of stress, chemotaxins such as C5a, lipid mediators, and so-called chemokines are produced. The secretion of chemokines can be firmly induced by the proinflammatory cytokines derived from tissue macrophage and T cells such as TNF-α, IL-1, and IFN-γ, or by bacterial products such as lipopolysaccharide (LPS) from Gram-negative bacteria, peptidoglycans from Gram-positive bacteria, or lipoarabinomannans from mycobacteria.
The process of recruitment is complex and still incompletely understood. Once recruited, the phagocytes – and neutrophils foremost – have a wide range of toxic mechanisms to fight any invading micro-organism, as described. These mechanisms are strongly regulated and delicately controlled because an over-excessive or premature induction of the toxic activities may result in the inactivation of protease inhibitors and activation of several cascades of activating substances (e.g. coagulation and fibrinolysis, the complement system). As a consequence, bacteraemia may progress to septic shock and disseminated intravascular coagulation, a community-acquired pneumonia may develop into acute or adult respiratory distress syndrome, and hypoxia/reperfusion injury can lead to fatal circulatory collapse.

In many of these early steps of inflammation, molecules are involved in which allelic variation has been characterised, often in the coding or promotor regions, as well as in the nontranscribed intronic sequences or 3'-regions. Their meaning in biological and – as a consequence – in clinical terms remains to be clarified. The FcγR allotypic variation is only a minute factor within the complex sequence of reactions in the host immune reaction to an invading pathogen.

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Discussion following lecture by T.W. Kuijpers

De Groot: I appreciate your remark about susceptibility and severity at the end of your presentation, which brings me to my question. It also relates to the first speaker, Dr Turner, who said, as I understand, that in the study on *Mycoplasma pneumoniae* infection there was a question of susceptibility. I fully agree with your statement that these molecules of innate immunity are more likely related to the severity of infectious diseases than to susceptibility. But if that is the case, could you comment on the type of studies that we have done so far, which are skewed, because most of them deal with retrospectively acquired material in highly selected populations from university settings and they are not prospectively done, i.e. they are not population-based. Can you give us your perspective on this? Maybe Dr Turner has a response to this question?

Kuijpers: First of all, we are now trying to do all these studies in a larger cohort of about 200 patients with recurrent respiratory tract infections more or less similar to the study already mentioned by Summerfield in which Dr Turner cooperated for his study of mannose-binding lectin (MBL). Once again, by checking all the known genotypic or allelic variation in these children, there is no clear-cut skewing in any of the polymorphisms presently known, not even for the MBL mutations. You can count the number of infections that these 200 children have, but indeed once again this is an anamnestic response by parents, and thus prone to recall bias. The only way to find out the truth is in a proper prospective study, preferably not in a tertiary centre, in order to have the broadest population possible. But what surprised us was that even the MBL mutations were definitely not skewed to a particular mutation as compared to controls.

Turner: I would not disagree with that. I think most of the studies on MBL today should have been done in centres where there have been tertiary referral hospitals collecting the samples. That is one of the reasons why we are doing a large study in the West of England – it is called ALSPAC, the Avon Longitudinal Study of Pregnancy And

Childhood – which Professor Jean Golding set up in the early 1990s to follow 14,000 pregnancies, and we are just completing a study of 8000 children to see what correlates we can find for disease associations in those children. That is a true prospective study. These sorts of studies are desperately needed for these innate immune markers.

Van der Meer: Just to get back to the point of the *Mycoplasma* infection as a complication in hypogammaglobulinaemic patients, the question there as well is whether it is a severity or a susceptibility marker.

Turner: You are right, it was mainly infection due to *M. hominis*, and the patient cohort was a mixture of common variable immunodeficiency and also some X-linked agammaglobulinaemia patients who had started off as a study of common variable immunodeficiency – the *Mycoplasma* association was discovered as an incidental part of that. So we then enlarged the cohort for *Mycoplasma*.

Kuijpers: I want to point out the study already mentioned by Peter Garred in cystic fibrosis. Considering a drop of eight years in lifespan, it is crucial when you are also MBL-deficient. Then you may think about prospective treatment options being available nowadays for those CF patients who are MBL-deficient. But that can only be done in a proper setting with a multicentre study. Without such collaboration we probably remain stuck with the question, is it susceptibility or severity? A severity marker is already important, but only in certain cohorts.

Appelmelk: Dr Kuijpers, first can you explain to me, when you have an allele in a low-affinity receptor causing inactivity, why doesn't the high-affinity receptor take over? It sounds as though there is some redundancy in the system. So how does that work?

Kuijpers: I think you are completely right in mentioning this. Of course, all comes down to the initial thought and idea that these polymorphic sites in case of IgG2 opsonising mechanisms were of importance because there was a strict difference in whether you had the one or the other genotype, FcγRIIa. Of course, the other studies were a kind of follow-up to that, first of all trying to confirm their ideas, and then suddenly these genotypes for the IIIa were showing different expression patterns and especially so in the autoimmune diseases. If a macrophage expresses so many Fcγ-receptors, having the opportunity to bind IgG-laden material, it is indeed questionable why it would not be taken over by other Fcγ receptors. I think a lot of the work in autoimmune diseases in fact confirms the genotype skewing. There are more molecules involved in immune reactivity as such. Think about the IL-6 or proteins related to apoptosis. Nobody knows in fact. This might simply be a way to look at something different in that particular genetic locus, not meaning that it is that particular receptor as such.

McAdam: Are these same receptors expressed on a placenta to facilitate the passage of immunoglobulin across the placenta? Would it be useful to look at babies with neonatal infections?

Kuijpers: The only receptor believed to be important in the transport of IgG from the mother to the child is thought to be the FcRN, so the neonatal receptor, which shows homology to the HLA class-I molecules. This is not even related to the FcγRI, II or III. So it is a very different mechanism. There is, as far as I know, no information about allelic variation in that receptor. Of course, I just mentioned the polymorphic sites in the FcγRIIb: they might be another thing to study, and especially that it is not in the extracellular part, where it may have an effect on the function of the other receptors. That is of course something that may or may not affect the outcome of an inflammatory reaction. But once again, I wonder whether that would tell us very much about susceptibility as such.

Van Agtmael: I would like to ask you a question from a clinical point of view. In March 2001 there was a study in Cochrane Review on the use of intravenous immunoglobulins (IVIG) for sepsis and septic shock and it showed a moderate effect.[1] It would be interesting to look at this group of patients to find factors in those patients who would benefit from the intravenous immunoglobulins. Do you think that it would be useful to look at the Fc polymorphisms in this broad group of patients with sepsis and to identify those who might benefit from the IVIG?

Kuijpers: Well, it would not be the first molecule I would look at, although potentially IVIG may be beneficial. I think it will be very hard to see in which patients they are effective. Of course, I am talking very much as a paediatrician seeing a lot of meningococcal infections – that is a very homogeneous population. But when you are practising internal medicine, there is a huge variation. There are so many parameters that you have to exclude or include to come up with a firm idea about which group is definitely helped by IVIG. The follow-up of those patients studied is difficult. Did they show a normal humoral response etc? All that crucial information is completely lacking in these cohorts. So I don't know.

De Groot: A small remark on the question concerning the neonatal infection, which brought me back to the presentation by Dr Turner in the beginning, where he said that surfactants may play a role as part of innate immunity. There is data to show that mutations in the genes for

surfactants A and D are associated with an increased risk of chronic recurrent lower respiratory tract infection. It is all very preliminary, and that actually brings me to my question. I will turn the gun onto myself immediately after I have said this because so far all these studies – if you have read Kimman's nice book – have been done on associations between single-gene polymorphisms and diseases. You have already said so, but knowing the literature for instance on leucocytes or chronic granulomatous disease (CGD), even in a clear-cut serious mutation disease as CGD there is a tremendous variability in the expression of the disease. This varies from patients who do not become ill at all to those who are very seriously ill with many in between. The way we study these diseases as clinicians is at this point in time very simplistic. We try to take one gene mutation and apply it to a population and look for an association, whereas it might very well be possible that there are a hundred genes involved and that the effects of twenty genes are upregulating severity and there are thirty down-regulating it.

Verbrugh: I have to intervene as Chairman, because we have a time problem here. Could you answer that briefly, because we would like to have two more comments from speakers before we go on to the next topic?

Kuijpers: This is one of the main points set out by the Dutch Society for Science, that you have to enter this field by doing gene arrays and snip arrays, and that may not even bring a clear answer to your question, because if you have twenty different genes or snips being characterised in a certain cohort of patients you come up with the same question. You have then characterised only a limited number of these snips. You may be more susceptible, but then also of course it is a matter of meeting the agent that can potentially affect you.

Kimman: Are Fcγ-receptors involved in the response to vaccinations, and has anybody looked for associations between response to vaccination and Fcγ-receptor polymorphisms?

Kuijpers: I would say yes, most likely, but whether somebody has studied that, I am not aware of that.

Van Furth: Just in emulation of the discussion with Van Agtmael and you, Dr Kuijpers, is there a difference between persons regarding the turnover of IgG, given intravenously or just autologous? Have these studies been done?

Kuijpers: I don't think anybody has done that in a detailed fashion, and the only thing I could say is that of course the turnover of IgG very much relates to the FcRN effect. So I think that a different receptor is involved in rescuing IgG from being cleared and broken down.

REFERENCE

1. Alejandria MM, Lansang MA, Dans LF, Mautaring JB. Intravenous immunoglobulin for treating sepsis and septic shock. Cochrane Database Syst Rev 2001;2:CD001090.