

## EXTENDED REPORT

# Fcγ receptor IIa, IIIa, and IIIb polymorphisms in German patients with systemic lupus erythematosus: association with clinical symptoms

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**Background:** Receptors for IgG play an important part in immune complex clearance. Several studies have identified polymorphisms of receptors for the Fc fragment of IgG (FcγR) as genetic factors influencing susceptibility to disease or disease course of systemic lupus erythematosus (SLE).

**Objective:** To examine these possibilities by evaluating a panel of clinical parameters in a cohort of 140 German patients with SLE for correlations with the FcγRIIa, IIIa, and IIIb polymorphisms in an explorative study.

**Methods:** 140 German patients with SLE according to American College of Rheumatology (ACR) criteria and 187 German controls were genotyped for the FcγRIIa, IIIa, and IIIb polymorphisms. Associations between FcγR genotypes, combined genotypes and clinical as well as laboratory features were analysed.

**Results:** No significant skewing of any of the three FcγR polymorphisms was seen in the German SLE cohort studied. Various clinical and serological parameters were found more frequently and at younger age in homozygous patients with the genotypes IIA-R/R131 or IIIA-F/F158 than in patients with IIA-H/H131 or IIIA-V/V158. These effects were even more pronounced in patients with the low binding combined phenotypes of the FcγRIIa, IIIa (double negative phenotypes) and FcγRIIa, IIIa, and IIIb (triple negative phenotypes). In patients with the double negative IIA and IIIA genotypes significantly higher frequencies of nephritis (63% v 33%) and proteinuria according to ACR criteria (58% v 11%), anaemia (84% v 55%), and anticardiolipin antibodies (63% v 22%) were found than in patients with the double positive genotypes. Patients with the IIA-R/R131 genotype and the double negative homozygous genotype had an earlier incidence of clinical symptoms, haematological and immunological abnormalities. Accordingly, SLE is diagnosed earlier in these patients, the difference reaching statistical significance only in the double negative v the double positive genotype (26.3 v 39.5 years) and the IIIA-F/F158 genotype v the rest (26.7 v 32.0 years). Most relevant is the fact that a higher median disease activity (ECLAM score) was demonstrated, both in the IIA-R/R131 homozygous (3.3 v 2.7) and the double negative (3.4 v 2.3) patients, reaching statistical significance in the first group.

**Conclusion:** The results of this explorative study support the view that the FcγRIIa/IIIa and IIIb polymorphisms constitute factors influencing clinical manifestations and the disease course of SLE but do not represent genetic risk factors for the occurrence of SLE. Higher frequencies of clinical symptoms, haematological and immunological abnormalities as well as an earlier onset of clinical symptoms, haematological and immunological markers of active disease were found in patients with the IIA-R/R131 genotype and the double negative and triple negative genotypes.

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Systemic lupus erythematosus (SLE) is a prototypic immune complex mediated disease with a broad spectrum of autoimmune phenomena, clinical symptoms, and a still unknown cause. Evidence for a hereditary predisposition to SLE comes from family studies demonstrating a high concordance among homozygotic twins, and a high prevalence of disease among relatives of patients with SLE, but a uniform HLA association has not been found. HLA studies have shown the importance of HLA-DR2 and 3, as well as their linked HLA-DQ specificities DQ1 and DQ2 and the C4A null allele. In particular, HLA data from multiplex families with SLE led to a multigene theory to explain SLE.<sup>1</sup>

In vivo and in vitro abnormalities in both complement mediated and FcγR mediated immune complex clearance have been described in SLE.<sup>2–4</sup> The major site of immune complex clearance is the mononuclear phagocyte system of the liver and spleen. Immune complexes are primarily removed through FcγR dependent mechanisms by splenic phagocytes. Clearance studies with IgG opsonised erythrocytes provide

strong evidence for impaired Fc receptor function in SLE.<sup>2</sup> The underlying hypothesis includes anti-FcγR autoantibodies,<sup>5</sup> defects of intrinsic phagocytic cell functions, and high levels of immune complexes or immunoglobulins.<sup>4,6</sup> More and more susceptibility genes for SLE can be located on the long arm of chromosome 1 in the region 1(q23–24), where FcγRII and FcγRIII genes are located, and in the region 1(q41–44)<sup>7</sup> and 1p36.<sup>8</sup> In addition, it has been suggested that FcγR gene polymorphisms have an important role in determining pathogenesis and disease course.<sup>9</sup>

**Abbreviations:** ACR, American College of Rheumatology; CRP, C reactive protein; ECLAM, European Consensus Lupus Activity Measurement; OR, odds ratio; PCR, polymerase chain reaction; RR, rate ratio; SDI, SLICC/ACR damage index; SLE, systemic lupus erythematosus; SLICC, Systemic Lupus International Collaborating Clinics

**Table 1** Characteristics of the study group. Results are shown as median (range)

	Controls (n)	All patients with SLE (n)	Age at diagnosis (y)	Patient age (y)	ECLAM score	SDI, year 5
All	187*	140*	30.1 (12.4–74.7)	40.0 (18.0–77.3)	2.9 (0.4–6.2)	2.0 (0–10)
FcγRIIA:						
R/R131	50	39	28.0 (12.4–74.7)	36.5 (18–76.8)	<b>3.3</b> (0.5–5.8)	3.0 (0–7)
H/H131	53	46	36.2 (15.1–73.7)	47.1 (20.3–80.3)	<b>2.7</b> (0.5–4.7)	2.0 (0–8)
FcγRIIIA:						
F/F158	62	55	<b>26.7</b> (13.7–74.7)	36.6 (18–77.3)	3.1 (0.5–4.8)	3.0 (0–10)
V/V158	50	21	<b>31.1</b> (15.4–72.0)	41.6 (21.3–78.2)	2.7 (1.4–5.1)	2.5 (0–8)
FcγRIIB:						
NA2/NA2	80	40	29.4 (12.4–62.7)	38.8 (18–69.7)	3.0 (0.5–6)	3.0 (0–8)
NA1/NA1	20	13	33.2 (17.8–57.8)	46.2 (23.8–62.9)	3.0 (0.9–3.8)	2.0 (0–10)
FcγRII/IIIa:						
R/R131, F/F158	18	19	<b>26.3</b> (13.7–74.7)	36.0 (18–77.3)	3.4 (0.5–4.3)	3.0 (0–7)
H/H131, V/V158	16	9	<b>39.5</b> (21.7–51.9)	52.0 (24.3–80.3)	2.3 (1.4–3.4)	3.0 (2–8)

ECLAM, European Consensus Lupus Activity Measurement; SDI, ACR damage index; SLICC, Systemic Lupus International Collaborating Clinics; significant differences are shown in bold.

\*The maximum number of controls and patients with SLE was 187 and 140 respectively.

Human leucocyte FcγR belong to the immunoglobulin supergene family, and three classes of FcγR (FcγRI, FcγRII, FcγRIII) are currently distinguished.<sup>10</sup> FcγRII (CD32) represents the most widely expressed class of FcγR, encoded by three genes (FcγRIIA, IIB, IIC) located on the long arm of chromosome 1, specifying six isoforms (FcγRIIa1, IIA2, IIB1, IIB2, IIB3, IIC). FcγRIIA is expressed on most myeloid cells and engagement of this receptor avidly triggers immune complex uptake. The FcγRIIA molecule occurs in two co-dominantly expressed allelic forms, IIA-R131 and H131; the IIA-H131 genotype exhibits a higher ability to bind complexed IgG2 and IgG3.<sup>11–12</sup> Data on African American and Dutch patients with SLE show that the homozygous IIA-R/R131 genotype is more common in patients with SLE with nephritis.<sup>13–14</sup> A phenylalanine (FcγRIIIA-F158) to valine (FcγRIIIA-V158) substitution at amino acid position 158 of FcγRIIIa, expressed on macrophages and NK cells, affects binding of IgG1, IgG3, and IgG4 subclasses, which are all more avidly bound by FcγRIIIa-V158.<sup>15–16</sup> The neutrophil FcγRIIb bears the neutrophil antigen polymorphism which is caused by four amino acid substitutions and is involved in both alloimmune and autoimmune neutropenias. Functionally, FcγRIIb-NA1 is more efficient in binding immune complexes containing IgG1 and IgG3 than FcγRIIb-NA2.<sup>17–18</sup> These in vitro findings suggest that the efficiency of FcγR mediated immunological functions differs between subjects. Interestingly, a large number of studies report skewed distributions of FcγRIIA, IIIa, and IIB alleles in patients with autoimmune diseases.<sup>13–14–19–22</sup>

Therefore we wondered whether such a skewed distribution is also present in German patients with SLE. We also evaluated whether the course of disease depends on FcγRIIA, IIIa, and IIB polymorphisms.

## PATIENTS AND METHODS

### Study group

One hundred and forty unrelated German patients with SLE, followed up at the Department of Medicine III, University Erlangen-Nuremberg, were randomly selected irrespective of their disease severity or stage of disease. In addition, a total of 187 healthy unrelated blood donors served as control group. All patients fulfilled the 1982 and 1997 revised criteria of the American college of Rheumatology (ACR) for the diagnosis of

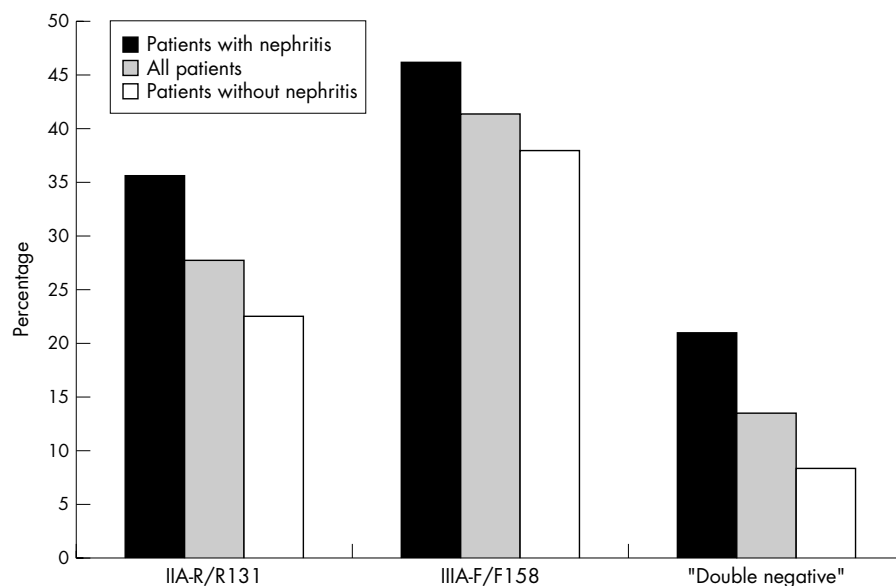
SLE.<sup>23–24</sup> Skin manifestations, including malar rash, discoid rash, photosensitivity, and oral ulcers, were present in 81%, 44%, 86%, 33% of our SLE cohort; arthritis was seen in 79%, serositis in 36%, nephritis in 41%, neurological disorder in 20%, haematological and immunological abnormalities in 97% and 96% each, and antinuclear antibodies in 99% of our patients. The major organ involvements were defined according to ACR criteria,<sup>24</sup> which implies that each patient with a renal disorder has nephritis. All abnormalities associated with SLE were recorded at the time of the first appearance either under our care or from a well documented history and for the whole follow up period.

### FcγRIIA, IIIA, and IIB genotyping

FcγR genotyping was performed for all patients and 187 controls, by polymerase chain reaction (PCR) based allotyping methods with allele-specific primers. For FcγRIIA genotyping, a 1000 bp PCR product containing the polymorphic FcγRIIA site was generated with FcγRIIA specific primers. This template was then used to amplify a 278 bp fragment with allele-specific primers.<sup>25</sup> PCR based genotyping methods were also used for the IIIA<sup>26</sup> and IIB allotyping.<sup>27</sup>

### Demographic, clinical, and laboratory characteristics

Demographic data for each patient were obtained retrospectively from the official medical record at the time of the first visit until September 1999 and included sex, age, and age at the time of diagnosis. Common clinical manifestations (according to Hahn<sup>28</sup>), haematological and immunological parameters, including C reactive protein (CRP), antinuclear antibodies, and dsDNA, Ro, La, nRNP, Sm, and cardiolipin antibodies, lupus anticoagulant, cryoglobulins, low C3 and C4 as well as an activity score (European Consensus Lupus Activity Measurement (ECLAM))<sup>29</sup> and a damage score (Systemic Lupus International Collaborating Clinics/ACR damage index for systemic lupus erythematosus (SLICC))<sup>30</sup> were recorded at each visit, with a maximum of three visits/year. Because the distribution of the FcγRIIA, IIIA, and IIB genotypic forms and genotype combinations is known to be different between ethnic groups,<sup>31</sup> we limited our study to white German subjects.



**Figure 1** Overrepresentation of the homozygous low binding genotypes FcγRIIA-R/R131, FcγRIIAA-F/F158, and FcγRIIA/IIIA-R/R131, F/F158 (double negative) in patients with SLE and nephritis. Differences are not significant. n=57 for patients with nephritis, n=140 for all patients, n=83 for patients without nephritis.

### Statistical analyses

Data analysis was done with the statistic package SPSS 10.0 for Windows. To test the null hypothesis that categorical variables such as clinical and serological parameters are equally distributed between patients with the low binding and high binding homozygous genotypes of the FcγRIIA, IIIA, and IIIB, two tailed Pearson's  $\chi^2$  tests were applied. The strength of the association of FcγRIIA, IIIA, and IIIB genotypes as genetic factor with the frequency of a certain SLE symptom was estimated by the calculation of the odds ratios (OR; OR>1: positive association; OR=1: no influence; OR<1: protective).<sup>32</sup> Median and range were calculated for age at diagnosis, ECLAM and SLICC scores because no parametric distribution can be assumed for these data. Accordingly, Mann-Whitney tests were used to test for the null hypothesis that these parameters did not differ between patients with the low binding and high binding groups of the FcγRIIA, IIIA, and IIIB. A p value <0.05 was considered significant. The correlation between age at the onset of symptoms and FcγRIIA, IIIA, and IIIB genotypes and genotype combinations was estimated according to the method of Kaplan and Meier,<sup>33,34</sup> and differences were assessed using log rank tests.<sup>35</sup> Finally, multiple logistic and Cox regression models were performed<sup>36</sup> to define the possible role of FcγRIIA, IIIA, and IIIB genotypes and genotype combinations as risk factors for outcome criteria like—for example, nephritis or a high damage score, accumulated since disease onset and calculated by the SLICC/ACR damage index (SDI). All analyses were adjusted for sex and age at disease onset by defining categorical variables. In these models, a p value of 0.1 was considered significant.

## RESULTS

### Distribution of FcγRIIA, IIIA, and IIIB genotypes and genotype combinations

FcγRIIA, IIIA, and IIIB genotypes were determined by genotyping 140 patients with SLE and 187 German controls using PCR based methods. The prevalence of IIA-R/R131, IIIA-F/F158, and IIIB-NA2/NA2 among patients with SLE was 28% (n=39), 41.3% (n=55), and 39.6% (n=40), comparable with that of the control group (27%, 35%, 43%) (table 1). No skewing of the FcγRIIA, IIIA, and IIIB polymorphisms was seen in this German SLE cohort. The double negative (IIA-R/R131, IIIA-F/F158) and triple negative (IIA-R/R131, IIIA-F/F158,

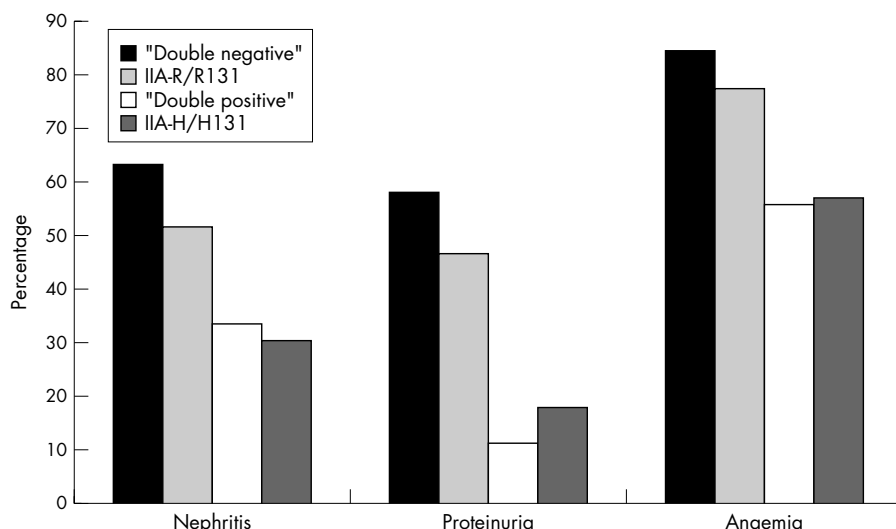
and IIIB-NA2/NA2) homozygous genotypes were detected in 14% (n=19) (table 1) and 8% (n=8) of patients and 11% and 5% of controls; again no significant skewing, but an overrepresentation of the double negative and triple negative genotypes was found in the SLE cohort. Figure 1 shows the distribution of the homozygous, low binding FcγRIIA/IIIA and B genotypes in 57 patients with SLE and nephritis. There was an overrepresentation of the genotype FcγRIIA-R/R131 (36%) compared with all patients with SLE (28%), patients without nephritis (23%), and controls (27%). The FcγRIIAA-F/F158 genotype was present in 46% compared with 38% in patients without nephritis, and the double negative genotype in 21% compared with 8% and 14% in patients without nephritis and all patients; the differences did not reach statistical significance (fig 1).

### Characteristics of the study group

The demographic data did not show significant differences between the three FcγR genotypes (table 1). Notably, the median age at diagnosis was 8.2, 4.4, and 3.8 years earlier in patients with SLE with the IIA-R/R131, IIIA-F/F158, and IIIB-NA2/NA2 genotypes than in patients with the IIA-H/H131, IIIA-V/V158, and IIIB-NA1/NA1 genotypes, reaching significance (p<0.05) only for IIIA. Interestingly, this difference was even more pronounced in the group of patients with the double negative homozygous genotype, who were 13.2 years younger at disease onset than patients with the double positive genotype (p<0.05). When the ECLAM score was tested as a measure for overall disease activity, a higher score for patients with the IIA-R/R131, IIIA-F/F158, and the double negative genotype was found compared with patients with the IIA-H/H131, IIIA-V/V158, and double positive genotype (3.3, 3.1, 3.4 v 2.7, 2.7, 2.3), reaching significance only for the FcγRIIA (p<0.05). The same trend was seen for the SDI (3.0, 3.0, 3.0 v 2.0, 2.5, 3.0) without reaching statistical significance.

### Clinical presentation

When the distribution of clinical symptoms and different genetically defined subgroups was analysed, a strong protective effect of the IIA-H/H131 genotype in comparison with the IIA-R/R131 genotype was found for all symptoms except pericarditis and malar rash (NS). In patients with the IIA-R/R131 genotype the incidence of nephritis (51% v 30% for the IIA-H/



**Figure 2** Prevalence of nephritis, proteinuria, and anaemia in patients with SLE with the homozygous genotypes FcγRIIA/IIIA-R/R131, F/F158 (double negative), FcγRIIA-R/R131, FcγRIIA/IIIA-H/H131, V/V158 (double positive), and FcγRIIA-H/H131. Statistical differences for the frequency of each symptom were calculated with a  $\chi^2$  test. The differences between columns 1 and 3 and columns 2 and 4 were significant for nephritis, proteinuria, and anaemia ( $p < 0.05$ ).

H131 genotype,  $p < 0.05$ , OR=1.5) and of proteinuria (46% v 17%,  $p < 0.01$ , OR=2.0) (according to ACR criteria) was significantly higher (fig 2). The prevalence of nephritis and proteinuria was even higher in the double and triple negative patients as compared with the double positive patients with a prevalence of nephritis of 63% ( $p < 0.05$ ) and 75% (NS) v 33% and 39% and a prevalence of proteinuria of 58% ( $p < 0.01$ ) and 62% (NS) v 11% and 29% (fig 2). Also the amount of maximum proteinuria was significantly higher in the double negative group as compared with the double positive group (2921 mg/d v 44 mg/d,  $p = 0.01$ ). Onset of nephritis, proteinuria (fig 3), and all clinical symptoms was earlier in double negative patients. In multivariate Cox regression models including age, sex, and FcγR genotypes and genotype combinations we identified FcγRIIA-R/R131 as the most important FcγR genotype for the occurrence of nephritis ( $p = 0.01$ , rate ratio (RR) = 1.4) (table 2).

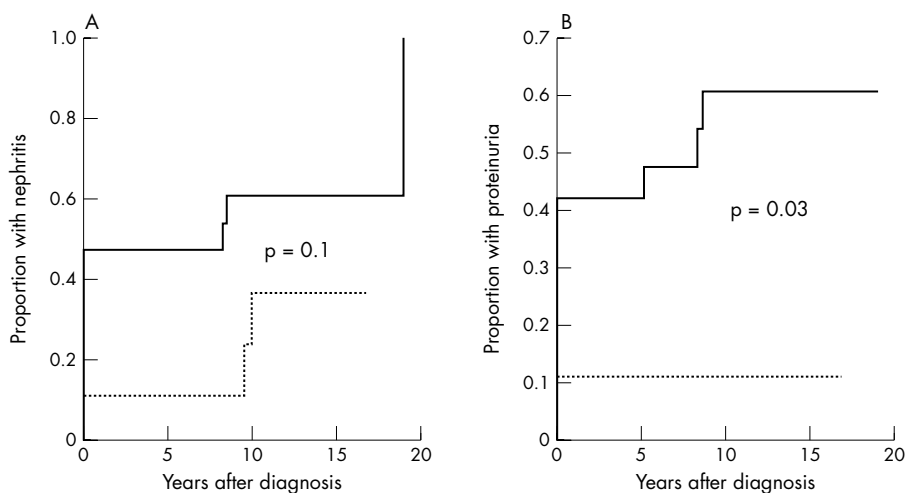
**Haematological characteristics**

When the genetically defined subgroups with special haematological characteristics are analysed it can be seen that cyto-

penias occurred earlier and more often in patients with the IIA-R/R131 or IIIA-F/F158 genotype (data not shown), reaching statistical significance for anaemia ( $p < 0.05$ ) in both subgroups. Again, an even higher prevalence of anaemia ( $p < 0.05$ ) was found for patients with the double (fig 2) and triple negative genotypes (prevalence 84%).

**Immunological characteristics**

Patients with the IIA-R/R131 genotype compared with the H/H131 genotype more often had antibodies against nRNP (64% v 30%, OR=2.0,  $p < 0.01$ ) as well as antibodies against Sm (44% v 24%, OR=1.6,  $p < 0.05$ ) and a low C4 (85% v 63%, OR=1.8,  $p < 0.05$ ). All autoimmune phenomena occurred more frequently and earlier in patients with the IIA-R/R131 genotype than in patients with the H/H131 genotype. There was the same trend in IIIA-F/F158 positive patients. In patients with the double negative genotype all tested parameters occurred more frequently, reaching statistical significance for cardiolipin antibodies (63% v 22%, OR= 2.4,  $p < 0.05$ ) and significantly earlier except for lupus anticoagulant and cryoglobulins.



**Figure 3** Kaplan Meier analysis of onset of (A) nephritis and (B) proteinuria according to the double negative (dashed line) versus double positive (solid line) homozygous FcγRIIA/IIIA genotypes. The difference between haplotypes was estimated with a log rank test; the p value indicates the level of significance.



**Table 2** Homozygous FcγR genotype in SLE as prognostic marker for the risk of nephritis

Risk factors	All nephritis		RR (95%CI)	p	RR (95%CI)
	No (%)	p			
FcγRIIA-R/R131	39 (51.3)	<b>0.01</b>	<b>1.4 (1.1 to 1.9)</b>	<b>0.01</b>	<b>1.4 (1.1 to 1.9)</b>
FcγRIIIA-F/F158	55 (45.4)	NS	NS	0.1	1.2 (0.9 to 1.6)
FcγRII/IIIA-R/R131, F/F158	19 (63.3)	NS	NS	0.08	1.6 (1.1 to 2.2)
FcγRIIA/IIA/IIIB-R/R131, F/F158, NA2/NA2	8 (75.0)	NS	NS	<b>0.04</b>	<b>1.6 (1.0 to 2.5)</b>
All patients with SLE	133 (40.6)	Multivariate analysis		Univariate analysis	

Multiple Cox regression analyses. After adjustment for age and sex the genotype FcγRIIA-R/R131 is still a risk factor (p=0.01). Significantly different values are shown in bold. RR, rate ratio.

No unidirectional trend was found in the FcγRIIIB genotypes.

## DISCUSSION

The present data document an association between the tested FcγRIIA, IIIa, and IIIB polymorphisms, the onset of disease, the onset, frequency, and quality of different clinical symptoms and disease-specific markers in German patients with SLE. To our knowledge, this is the first report providing information on all known functional FcγR polymorphisms and on FcγR genotype combinations and the clinical impact of these genotype combinations on the course of SLE.

We, as well as others, found no significant differences in the distribution of genotypes<sup>14–37–43</sup> or genotype combinations between patients with SLE and controls of an ethnically homogenous group, thus excluding the FcγRIIA, IIIA, and IIIB polymorphisms as susceptibility factors for SLE as a whole in the Erlangen cohort. Only Hatta *et al* detected significant difference in the frequencies of FcγRIIIB genotypes between SLE and healthy subjects.<sup>44</sup> The odds ratio of the FcγRIIIB-NA2/NA2 homozygotes for the development of SLE was 2.5. We found a unidirectional trend for an earlier occurrence (p<0.05 in 48% of the tested variables) and a higher incidence (p<0.05 in 15% of the tested variables) of all clinical features (except pericarditis and malar rash) and serological markers of SLE in patients with the homozygous genotype FcγRIIA-R/R131; this genotype has a lower ability to bind IgG2 and IgG3, and may therefore negatively influence the outcome in individual patients. When we evaluated associations between FcγRIIA polymorphism and the onset of disease, we found an 8.2 years' delay in disease onset (median) in the homozygous group with the IIA-H/H131 genotype. A delayed disease onset has been reported to be associated with a less severe course of disease.<sup>45</sup> Yun *et al* described an earlier occurrence of photosensitivity and oral ulcers in Korean patients with the IIA-R/R131 genotype, which is in accordance with our findings.<sup>46</sup> This trend was also found for patients with the genotype FcγRIIIA-F/F158 (earlier occurrence of symptoms: p<0.05 in 22% of the tested variables; higher frequency: p<0.05 in 12.5% of the tested variables) and FcγRIIIB-NA2/2 (p<0.05 in 9% and 5% respectively). In the double negative genotypes we observed an accumulation of effects with even higher incidence and earlier onset of various symptoms (earlier occurrence of symptoms: p<0.05% in 42% of the tested variables; higher incidence: p<0.05 in 14%). In a Dutch cohort of patients with Wegener's disease, the relapse rate was increased in patients with the double negative genotype.<sup>47</sup>

Our results support the hypothesis, that FcγRIIA/IIIa and IIIB polymorphisms influence the onset and the course of disease and therefore directly influence human biology, as concluded also for a Malay-Chinese cohort.<sup>39</sup> However, we did exclude an influence of these polymorphisms on the risk of developing SLE, at least in this German group. Going into detail, the outcome of SLE is negatively influenced by nephritis or anaemia, as we<sup>48</sup> and others have shown.<sup>45–49</sup> For both

features, this study demonstrated a higher incidence and an earlier onset for the group with the homozygous FcγRIIA-R/R131, the double and triple negative genotypes. Song *et al*, like us (data not shown), found a higher proteinuria in FcγRIIA-R/R131 positive patients with lupus nephritis.<sup>30</sup>

Most interesting is the predominance of IgG2 and IgG3 in glomerular immune complexes of 74 patients with lupus nephritis, as analysed by Zuniga *et al*.<sup>51</sup> They found an enrichment of the IIA-R/R131 genotype (47%), particularly in patients with immune complexes containing IgG2.<sup>51</sup> In multivariate Cox regression analyses we showed that FcγRIIA-R/R131 is an independent risk factor for nephritis. In 57 patients with lupus nephritis from the Erlangen cohort there is an overrepresentation of patients with the homozygous genotypes FcγRIIA-R/R131 (36% v 23%), FcγRIIIA-F/F158 (46% v 38%), and double negative genotype (21% v 9%). These differences between the groups were not significant here and in other studies,<sup>14–21–22–37–38–42</sup> as described for FcγRIIA and FcγRIIIA for white subjects<sup>13–55</sup> and other ethnic groups,<sup>14–15–21–30–52–54</sup> thus emphasising the need for larger studies with ethnically homogenous populations or meta-analyses. These results suggest that ethnic variation is critical in defining the specific genetic factors underlying the pathogenesis of SLE, and they have important prognostic and therapeutic implications also. As we demonstrated a strong influence of the FcγRIIA, IIIA, and IIIB polymorphisms on the course of disease our conclusion is different from that of Northworthy *et al*, which limited the influence of FcγR on the prevalence of lupus nephritis to patients with C1q antibodies of the subclass IgG2.<sup>20</sup>

Also for anaemia and cytopenias in general we found a higher incidence and earlier onset in patients with the low binding phenotypes; again, there was an accumulation of effects in patients with the double negative and triple negative genotypes. This is interesting because FcγRIIA and FcγRIIIB are important for the clearance of erythrocytes,<sup>18</sup> as confirmed by in vivo data of Dijkstra *et al* which showed a poorer clearance of IgG coated erythrocytes in IIA-R/R131 patients.<sup>56</sup>

As previously reported,<sup>38</sup> Sm and nRNP antibodies are more frequent and occur earlier in IIA-R/R131 positive patients, which fits well with the overrepresentation of the IgG2 subclass in these autoantibodies.<sup>57</sup>

FcγRII and III genes are mapped in a 500 kB region of 1q(23–24). As previously discussed,<sup>38</sup> the interpretation of association studies is complicated by the presence of immunologically relevant genes—for example, CRP, SAP, and interleukin 6 receptor located on the long arm of chromosome 1 in the region 1q(23–24) and linked to the FcγRII and FcγRIII genes. The possibility of linkage disequilibrium with any of these genes might also influence the course of disease.<sup>31</sup> A non-random distribution can be assumed for FcγR genotype combinations, as described by van der Pol *et al* for Dutch white subjects<sup>31</sup> and could be shown in our SLE cohort for the double negative and triple negative genotypes.

In conclusion, our results support the hypothesis, that the lower ability to clear immune complexes—depending on FcγRIIA, IIIA, or IIIB genotype—influences the prognosis and

outcome in patients with SLE. Ethnic differences are large and have to be kept in mind when the results of different studies are compared. The results on the non-random distribution of FcγRIIA, IIIA, and IIIB genotype combinations show the importance of looking for genotype combinations in association studies.

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