Severity of Meningococcal Disease Associated with Genomic Bacterial Load

Tom Darton, Malcolm Guiver, Simone Naylor, Dominic L. Jack, Edward B. Kaczmarski, Raymond Borrow, and Robert C. Read

Department of Infection and Immunity, University of Sheffield, and Department of Infection and Tropical Medicine, Royal Hallamshire Hospital, Sheffield Teaching Hospitals National Health Service Foundation Trust, Sheffield, and Meningococcal Reference Unit for England and Wales, Health Protection Agency, Manchester, United Kingdom

(See the editorial commentary by Brouwer and van de Beek on pages 595–7)

**Background.** Diagnostic polymerase chain reaction (PCR) detection of *Neisseria meningitidis* has enabled accurate quantification of the bacterial load in patients with meningococcal disease.

**Methods.** Quantification of the *N. meningitidis* DNA level by real time-PCR was conducted on whole-blood samples obtained from patients presenting with meningococcal disease to hospitals throughout England and Wales over a 3-year period. Levels were correlated with clinical outcome, infecting serogroup, and host factors including, interleukin-1 genotype (*IL-1*).

**Results.** Bacterial loads were available for 1045 patients and were not associated with the age of the patient, delay in sample submission, or administration of antibiotics prior to admission. The median log bacterial load was higher in 95 patients who died (5.29 log_{10} copies/mL; interquartile range, 4.41–6.30 log_{10} copies/mL) than in 950 patients who survived (3.79 log_{10} copies/mL; interquartile range, 2.87–4.71 log_{10} copies/mL). Logistic regression revealed that age (odds ratio, 1.04 per 1-year increase in age) and bacterial load (odds ratio, 2.04 per log_{10}-copies/mL increase) had a statistically significant effect on the risk of death. Infection with *N. meningitidis* serogroup C was associated with increased risk of death and an increased bacterial load. Also associated with a higher bacterial load were prolonged hospitalization (duration, >10 days); digit, limb, or soft-tissue loss; and requirement of hemodialysis. Carriage of *IL-1RN*(+2018) was associated with increased mortality (odds ratio, 2.14; *P* < .07) but not with a higher bacterial load.

**Conclusions.** In meningococcal disease, bacterial load is associated with likelihood of death, development of permanent disease sequelae, and prolonged hospitalization. The bacterial load was relatively higher in patients infected with *N. meningitidis* serogroup C than in those infected with other serogroups. The effects of age and *IL-1* genotype on mortality are independent of a high genomic bacterial load.

*Neisseria meningitidis* causes a spectrum of illness ranging from asymptomatic carriage to fulminant meningococcemia and meningitis. In one large European epidemiological survey, the mortality rate of *N. meningitidis* infection was 8% [1].

Non--culture-based diagnostic PCR techniques were introduced as a national service in Great Britain in 1996 [2–7], and this has allowed the accurate and rapid quantification of bacterial load (consisting of both viable and nonviable bacteria). It is likely that all routine laboratories with molecular facilities will be capable of providing this service in the future [8, 9].

Previous studies, mainly involving small pediatric samples, have demonstrated a correlation between bacterial load and death [10–12]. The relationship between meningococcal bacterial load and other clinical outcomes, such as the duration of hospitalization or the development of permanent sequelae of disease, has not been investigated. Certain factors have been shown to increase likelihood of death due to meningococcal disease, including age, infection with *N. meningitidis* serogroup C, and host polymorphisms of the *IL-1* and *IL-1RN* receptor antagonist genes (*IL-1* and *IL-1RN*, respectively) [13], but it is not known whether these factors are also associated with a high bacterial load. Such
information would improve our understanding of the mechanisms that lead to death.

In this study we analyzed the quantitative bacterial load of a national cohort of patients presenting to hospitals with meningococcal disease. These data were correlated with clinical and microbiological data, outcomes of disease, and IL-1 and IL-1RN genotypes to determine (1) the relationship between bacterial load and important sequelae of disease and length of hospital stay, and (2) whether the mortality-associated factors of age, infecting serogroup, and IL-1/IL-1RN genotype exert their effects via increased susceptibility to high bacterial load.

METHODS

Patients and samples. Patient and specimen data were included in this analysis if an EDTA-treated blood sample had been submitted to the Meningococcal Reference Unit for England and Wales (Manchester, England) for the purpose of non-culture-based diagnosis and if the patient was subsequently proven to have meningococcal disease on the basis of culture or PCR of material from any invasive site. From this group, we identified persons who were found to have a positive PCR signal for meningococcal DNA in peripheral blood specimens and those for whom a quantitative bacterial load measurement had been determined. All samples submitted to the Meningococcal Reference Unit were accompanied by simple clinical, demographic, and outcome (death vs. survival) data relating to the patient. A subset of this sample was included in a previous study in which patients’ DNA had been genotyped at markers within the IL-1 locus [13]. In the case of this subset, a research nurse (S.N.) contacted the patients or their families and used a structured questionnaire to obtain additional details about the clinical episode and outcome. Patients or their families were interviewed 1–3 years after the clinical episode. Specifically, data were recorded on administration of antibiotics prior to hospitalization, time spent in high-dependency areas (high-dependency units or the intensive treatment unit), use of hemodialysis, and duration of hospitalization. Occurrence of clinical sequelae and outcome were also recorded: patients were asked whether they had experienced sustained hearing loss, visual problems (specifically, diplopia and loss of visual acuity), or digit or limb loss and a requirement for skin grafting. All data were made anonymous after collection; samples were assigned a unique identifier, which was used for the remainder of the study. Ethics approval for the study was granted by the Public Health Laboratory Service (now the Health Protection Agency; Colindale, London, UK) and the ethics committee of the Sheffield Teaching Hospitals NHS Foundation Trust (Sheffield, England).

Isolate confirmation and serogroup characterization. Isolate confirmation was performed as described elsewhere [14]. In addition, a non–culture-based serogrouping technique (i.e., PCR) was performed with the use of siaD assays to confirm serogroups of the sialic acid–containing capsules for serogroups B and C [15, 16] and serogroups Y and W135 [16, 17]. Serogroup A was confirmed using the mynA PCR assay from the year 2000 onwards [18].

Quantification of the N. meningitidis load. Quantification of the N. meningitidis bacterial load was performed at the Meningococcal Reference Unit, as previously described elsewhere [14]. This assay targets the capsular transfer gene (ctrA), which has been shown to be specific for N. meningitidis, and provides accurate quantification in the range of 10^3–10^9 copies/mL [6]. Because ctrA is a single-copy gene, the number of copies measured is equivalent to the bacterial load number.

Genetic analysis. Patient DNA was extracted from the whole-blood EDTA samples using standard methods and was probed for single-nucleotide polymorphisms at the IL-1B(-511) and IL-1RN(+2018) positions, as described elsewhere [19] (rs16944 and rs315952, respectively). The nomenclature used to describe genotype is as follows: 1, frequent allele; 2, rare allele; +, allele carriage; −, lack of allele carriage. For statistical analysis, individuals were classified on the basis of carriage of the rare alleles (IL-1 2+ and IL-1RN 2+) or according to IL-1 and IL-1RN composite genotypes.

Statistical analysis. Bacterial load data were log transformed to assume a normal distribution. To select variables for inclusion in regression analysis, preliminary unadjusted univariate tests (the Mann-Whitney U test for skewed data, unpaired Student’s t test for approximately normally distributed data, and the χ² test for categorical data) were performed. Variables that were found to be statistically significant (P<.05) were subsequently included in the primary regression analysis. Secondary analyses to examine the effects of variables on clinical sequelae and duration of hospital stay were then performed, with significance assumed at P<.01. Database formation and statistical analyses were performed using Excel, version 11.5 (Microsoft), and SPSS software, version 16 (SPSS). All bacterial load data are presented as the number of log₁₀ DNA copies per milliliter of blood.

RESULTS

Patients and Samples Available for Analysis

During the period January 1999 through December 2001, a total of 1910 EDTA-treated blood samples were received by the Meningococcal Reference Unit from patients who fulfilled diagnostic criteria (table 1). Of these patients, 1719 (90%) survived, and 191 (10%) died. The mean age (±SE) of patients who died (21.8 ± 1.8 years; median, 16 years; range, 0–82 years) was significantly higher than that of those who survived (12.3 ± 0.4 years; median, 5 years; range, 0–88 years; P<
Table 1. Population characteristics for the entire study population and for those for whom bacterial load data were available.

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Survivors All (n = 1719)</th>
<th>Survivors Bacterial load data available (n = 950)</th>
<th>Nonsurvivors All (n = 191)</th>
<th>Nonsurvivors Bacterial load data available (n = 95)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mortality rate, %</td>
<td>...</td>
<td>10.0</td>
<td>9.1</td>
<td></td>
</tr>
<tr>
<td>Male sex</td>
<td>869 (50.6)</td>
<td>490 (51.6)</td>
<td>87 (51.2)</td>
<td>55 (57.9)</td>
</tr>
<tr>
<td>Age</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mean years ± SD</td>
<td>12 ± 16</td>
<td>11 ± 16</td>
<td>22 ± 23</td>
<td>19 ± 23</td>
</tr>
<tr>
<td>Range</td>
<td>0–88</td>
<td>0–88</td>
<td>0–82</td>
<td>0–90</td>
</tr>
<tr>
<td>Age group</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>&lt;1 year</td>
<td>437 (25.4)</td>
<td>272 (28.6)</td>
<td>45 (26.5)</td>
<td>29 (30.5)</td>
</tr>
<tr>
<td>1–3 years</td>
<td>382 (22.2)</td>
<td>249 (26.2)</td>
<td>15 (8.8)</td>
<td>13 (13.7)</td>
</tr>
<tr>
<td>4–11 years</td>
<td>260 (15.1)</td>
<td>144 (15.2)</td>
<td>12 (7.1)</td>
<td>5 (5.3)</td>
</tr>
<tr>
<td>12–17 years</td>
<td>282 (16.4)</td>
<td>123 (12.9)</td>
<td>33 (19.4)</td>
<td>16 (16.8)</td>
</tr>
<tr>
<td>18–20 years</td>
<td>83 (4.8)</td>
<td>33 (3.5)</td>
<td>7 (4.1)</td>
<td>2 (2.1)</td>
</tr>
<tr>
<td>21–60 years</td>
<td>234 (13.6)</td>
<td>112 (11.8)</td>
<td>40 (23.5)</td>
<td>23 (24.2)</td>
</tr>
<tr>
<td>&gt;60 years</td>
<td>41 (2.4)</td>
<td>17 (1.8)</td>
<td>18 (10.6)</td>
<td>7 (7.4)</td>
</tr>
<tr>
<td>Serogroup</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>A</td>
<td>1 (0.1)</td>
<td>0 (0)</td>
<td>0 (0)</td>
<td>0 (0)</td>
</tr>
<tr>
<td>B</td>
<td>948 (55.8)</td>
<td>555 (59.0)</td>
<td>62 (36.9)</td>
<td>32 (34)</td>
</tr>
<tr>
<td>C</td>
<td>522 (30.7)</td>
<td>261 (27.8)</td>
<td>94 (56.0)</td>
<td>56 (59.6)</td>
</tr>
<tr>
<td>Y</td>
<td>8 (0.5)</td>
<td>4 (0.4)</td>
<td>0 (0)</td>
<td>0 (0)</td>
</tr>
<tr>
<td>W135</td>
<td>29 (1.7)</td>
<td>15 (1.6)</td>
<td>2 (1.2)</td>
<td>1 (1.1)</td>
</tr>
<tr>
<td>Ungrouped</td>
<td>192 (11.3)</td>
<td>105 (11.2)</td>
<td>10 (6)</td>
<td>5 (5.3)</td>
</tr>
<tr>
<td>Antibiotics received</td>
<td>273 (99.3)</td>
<td>162 (99.4)</td>
<td>2 (0.7)</td>
<td>1 (0.6)</td>
</tr>
</tbody>
</table>

**NOTE.** Data are no. (%) of patients, unless otherwise indicated.

.001, by Mann-Whitney U test). Receipt of antibiotics before hospital admission (data were available for 15 patients who died) did not have a statistically significant effect on the risk of death (P = .165, by Fisher’s exact test).

**Method of Diagnosis Confirmation**

The diagnosis of meningococcal disease from all sites was made by PCR in 984 cases (52%), by culture in 251 cases (13%), and by both PCR and culture in 661 cases (35%). Serogroup determination was possible for 1683 isolates (88%) (table 1). Of the remaining isolates, 203 (11%) were ungrouped—that is, bacterial DNA was detected by ctrA PCR, but it was not possible to determine the serogroups by either siaD or mynA assay. Patients with serogroup B infection (1010 patients) were one-half as likely to die as those with non-serogroup B infection (i.e., all other serogroups combined; 858 patients; OR, 0.46; 95% CI, 0.33–0.64; P < .001, by χ² test). Patients infected with serogroup C (616 patients) had an almost 3-fold risk of death, compared with patients with non-serogroup C infection (1252 patients; OR, 2.87; 95% CI, 2.08–3.95; P < .001, by χ² test).

**Bacterial Load**

Of 1645 cases diagnosed by techniques that included a PCR assay, a measurement of bacterial load in EDTA-treated blood specimens was available for 1045 patients (table 1). Of the remaining patients, 248 (19%) had a nonquantifiable bacterial load, and information on the remaining 352 was not available. Patients with nonquantifiable bacterial loads tended to be younger (mean age, 13 years) and to have serogroup B disease (58.3%), although these characteristics were not statistically significant (as determined by χ² test). This group also had better outcomes: they had a lower mortality rate (2.4% vs. 9.1%; P < .001, by χ² test) and lower rates of prolonged hospitalization and permanent sequelae (data not shown).

The median bacterial load for all 1045 patients was 3.90 log₁₀ copies/mL (interquartile range [IQR], 2.94–4.91 log₁₀ copies/mL; mean ± SD, 4.12 ± 1.29 log₁₀ copies/mL) and ranged between the upper and lower limit of the PCR assay capability (3–8 log₁₀ copies/mL; M. Guiver, personal communication). Patients whose bacterial loads were determined within 5 days after hospital admission (n = 695) had a median bacterial load
Factors Affecting the Bacterial Load

Timing of sample submission. Data relating to timing of submission of the sample to the Meningococcal Reference Unit was available for 706 samples. The majority (60%) were submitted within the first 24 h after admission to the hospital (range, 0–12 days). In a comparison of samples submitted on days 0, 1, 2, 3, 4, and 5 after hospital admission (after this point, there was an insufficient number samples available for analysis; these samples accounted for 1.56% of the total), there was a significant but negligible decrease in the measured bacterial load ($r = -0.092$; $P = .016$, by Pearson’s correlation) (figure 1). Additional comparison was performed by grouping samples according to the day of submission, but this comparison confirmed no significant difference in median bacterial load ($P = .258$, by Kruskal Wallis test). The median bacterial load for samples sent on the day of hospital admission was 4.10 log$_{10}$ copies/mL; those sent on day 5 had a median bacterial load of 3.85 log$_{10}$ copies/mL.

Pre–hospital admission administration of antibiotics.

The median bacterial load was not significantly different for 165 patients who did versus 314 patients who did not receive antibiotics before hospital admission (3.82 log$_{10}$ copies/mL [IQR, 3.16–4.97 log$_{10}$ copies/mL] vs. 3.77 log$_{10}$ copies/mL [IQR, 2.90–4.86 log$_{10}$ copies/mL]; $P = .455$, by Mann-Whitney $U$ test). No difference was seen if only samples sent within 24 h after hospital admission to the hospital were compared (among 76 patients who had received antibiotics, the median bacterial load was 4.05 log$_{10}$ copies/mL [IQR, 3.88–5.38 log$_{10}$ copies/mL]; among 132 patients who had not received antibiotics, it was 3.99 log$_{10}$ copies/mL [IQR, 3.10–5.13 log$_{10}$ copies/mL; $P = .299$, by Mann-Whitney $U$ test).

As a result of these analyses, all samples, regardless of time lapsed prior to receipt and regardless of whether antibiotics had been administered prior to hospital admission, were included in further regression modeling.

Age and serogroup. Patient age had no significant effect on the bacterial load, whether assessed as a scale ($r = -0.014$; $P = not significant, by Pearson’s correlation) (figure 2) or as a grouped variable ($P = .256$, by Kruskal Wallis test). In contrast, serogroup had a statistically significant influence on the circulating bacterial load. The median bacterial loads, by serogroup, were as follows: serogroup B, 3.74 log$_{10}$ copies/mL (IQR, 2.70–4.72 log$_{10}$ copies/mL); serogroup C, 4.39 log$_{10}$ copies/mL (IQR, 3.44–5.35 log$_{10}$ copies/mL); serogroup Y, 2.70 log$_{10}$ copies/mL (IQR, 2.70–4.30 log$_{10}$ copies/mL); and serogroup W135, 3.22 log$_{10}$ copies/mL (IQR, 2.74–3.66 log$_{10}$ copies/mL). Compared with non–serogroup B infection, disease caused by $N$. meningitidis serogroup B was associated with a statistically significantly lower bacterial load ($P < .001$, by
Mann-Whitney U test). Conversely, disease due to serogroup C was associated with a significantly higher bacterial load (P < .001, by Mann-Whitney U test).

**Combination of factors affecting the bacterial load.** To assess the effect of the combination of these factors on the measured bacterial load, a multiple regression model was created. After adjustment and removal of nonsignificant variables (i.e., those with a P value > .05), only serogroup C disease was associated with a statistically significant effect. The median difference in the bacterial load between patients with serogroup C disease and those with non-serogroup C disease was +0.55 log_{10} copies/mL (95% CI, +0.38 to +0.71 log_{10} copies/mL; P < .001).

**Effect of Bacterial Load on Outcome**

**Bacterial load and death.** Multivariate logistic regression analysis of complete data sets (n = 507) was performed, including only bacterial loads for samples obtained ≤48 h after hospital admission (to exclude any possible impact of hospital treatment) (table 2). Significant factors in the final model are shown below

- **Bacterial load.** Increased bacterial was statistically significantly associated with an increased risk of death. In patients with consistent age and IL-1 genotype, there was a 2-fold risk of death for every 1–log_{10} copy/mL increment in the bacterial load.

- **Age.** There was a significant and independent association between increasing age and increased risk of death. For each additional year of life, the risk of death increased 1.04-fold. Therefore, bacterial load and carriage of the IL-1RN(+) genotype, serogroup B, and serogroup C. Only statistically significant variables are shown.

- **IL-1RN(+) genotype.** Carriage of the rare allele at IL-1RN(+) indicated a tendency toward an increased risk of death. The effect of genotype on death was independent of the bacterial load and age, as seen by the effect on the univariate OR of addition to the regression model. Patients with the IL-1RN(+) genotype (n = 192) had a 2.04-fold increased risk of death, compared with a 30-year-old patient (OR = 1.04^30/OR of 3.2 [i.e., 1.04^30]). Age, however, had effect on bacterial load.

- **IL-1RN(+) genotype.** Carriage of the rare allele at IL-1RN(+) indicated a tendency toward an increased risk of death. The effect of genotype on death was independent of the bacterial load and age, as seen by the effect on the univariate OR of addition to the regression model. Patients with the IL-1RN(+) genotype (n = 192) had a 2.04-fold increased risk of death, compared with a 30-year-old patient (OR = 1.04^30/OR of 3.2 [i.e., 1.04^30]). Age, however, had effect on bacterial load.

**Combination of factors affecting hospital stay and clinical sequelae.** Bacterial load was found to be a significant variable affecting the duration of hospital stay, the need for hemodialysis, and limb or digit loss or a requirement for skin grafting (table 4). Age was also a significant factor that affected the duration of hospital stay, a need for hemodialysis, and the subsequent likely reporting of visual impairment after the clinical episode. Carriage of the rare allele at IL-1RN(+) was found to be protective against the development of auditory problems, although this variable failed to reach the level of statistical significance set for secondary analysis (i.e., P > .01).

**DISCUSSION**

This study confirms the association between a high quantitative bacterial genomic load of *N. meningitidis* in blood samples and increased mortality. Among patients who survive, a high bacterial load is also associated with loss of limbs, digits, or skin/soft tissue; with renal failure requiring hemodialysis; and with prolonged hospitalization. It also reveals that the lethality of serogroup C–expressing *N. meningitidis* strains during the period 1999–2001 was likely related to high bacterial load, but that the influence of host factors (e.g., age and IL-1 genotype) on mortality is independent of an effect upon the intensity of bacteremia. Although this is a retrospective analysis, it has the advantage of a large national cohort of samples that were all processed at the same reference laboratory. In patients with meningococcemia, proliferation of bacteria correlates with production of lipopolysaccharide [12] [20], complement activation [21, 22], and cytokine or chemokine disruption [23–27], all of which affect severity of disease and likely clinical outcome [28].

The observation that bacterial load measurements are unaffected by delay in sample submission or by administration of antibiotics before hospital admission is relevant to the interpreting clinician. The implication of these findings is that a sample obtained for bacterial load measurement will provide valuable prognostic information. Our observations also reveal some insight into mechanisms that underlie risk factors associated with death in patients with meningococcal disease. Thus, increasing age is a direct and independent risk factor for death.
and the development of several clinical sequelae (visual deficit, longer hospital stay, and need for hemodialysis).

The work of Brandtzaeg et al. [28] revealed a clear relationship between lipopolysaccharide concentrations in blood and CSF specimens and the severity of meningococcal septicemia and meningitis and also with levels of plasma proinflammatory markers, such as TNF and IL-1. Plasma lipopolysaccharide concentrations correlate with bacterial load [12]. It can be concluded that a primary determinant for septic injury in patients with meningococcal disease is failure of the hosts' innate immunity to limit bacterial replication in sterile sites.

It has been shown by a number of investigators, including ourselves, that polymorphisms of the IL-1/IL-1RN complex are associated with death in persons with meningococcal disease [9, 28, 29]. The rare allele of the single-nucleotide polymorphism at IL-1RN(+2018) is found in 28% of the white population [30] and is associated with enhanced IL-1β availability in vitro [31]. This study allowed us to show that carriage of rare alleles within IL-1B and IL-1RN likely do not mediate their effects on disease mortality via failure to control bacterial replication. Therefore, we conclude that the effect of IL-1/IL1-RN genotype on likelihood of death is related to perturbation of the metabolic/cytokine profile. An interesting observation in this study is that carriers of IL-1RN(+2018) are less likely to experience auditory loss. This could suggest that a proinflammatory profile protects against meningitis in individuals with meningococcal bacteremia.

Infection with serogroup C has been shown by several authors to be associated with higher morbidity and mortality rates, particularly in the noninfant population [32, 33]. Our results indicate that infection with serogroup C–expressing strains prevalent in the period 1999–2001 was associated with an increased risk of death because of a higher bacterial load. The majority of serogroup C isolates of N. meningitidis during the period commencing in 1995 were of sequence type (ST) 11 [34]. In Europe, ST11 has an OR for mortality of 2.19 (95% confidence interval).
CI, 1.53–3.15) [1], and in South Africa (where the serogroup was determined to be W135), it caused a recent lethal outbreak of meningococcal disease [35]. The mechanism whereby the clonal complex might be associated with a higher bacterial load is obscure but is presumably related to virulence determinants capable of allowing the organism to evade innate immunity.

The relatively long persistence of DNA in these EDTA-treated whole-blood samples (compared with plasma specimens [12]) is consistent with the findings of Hackett et al. [10] and is probably related to sequestration of meningococci in peripheral blood leukocytes. We found that EDTA-treated whole-blood specimens are more sensitive than plasma for PCR detection (M. Guiver, personal communication).

We attach some reservations to our conclusions. The database analyzed broadly reflects the distribution of serotypes seen nationally in the United Kingdom during the same time period, as shown by serogroup rate comparison with averaged national data (table 1) (national data were as follows: serogroup A, 0.04%; serogroup B, 52.0%; serogroup C, 33.8%; serogroup W135, 2.2%; serogroup Y, 0.08%; and ungrouped, 11.9%) [14]. Non-B and non-C serogroups are uncommon in the United Kingdom, so our findings may not apply in populations with higher prevalence of other serogroups. The database did demonstrate an underrepresentation of culture-only diagnostic methods; national/study rates of diagnostic methods were 36.8%/52%, 41.3%/13% and 22.3%/35% for PCR only, culture only, and PCR and culture methods combined, respectively [14]. An additional potential source of error was the requirement that samples be transported to a central laboratory, although we feel that this is unlikely to have caused significant DNA degradation or any repeat freeze/thaw events [6]. The clinical outcomes used were all self-reported by the patient or the patient’s next-of-kin and, thus, are subject to recall bias. However, in a subset of 20 patients, there was 100% fidelity of recall in the structured interview when tested against details recorded in the clinical charts (data not shown).

In summary, the intensity of bacteremia in patients with meningococcal disease is associated with poor outcome, and is influenced by the serogroup of the infecting strain. The effects of age and IL-1 genotype on likelihood of death are independent of a high genomic bacterial load.

Acknowledgments

We thank all of the Consultants in Communicable Disease Control, the patients, and their families for assistance in the construction of the original database. We also thank the Statistical Services Unit of The University of Sheffield and Dr. James Wing for assistance with the statistical analysis.

Financial support. Meningitis Research Foundation (award 4/00).

Potential conflicts of interest. R.C.R. is a stockholder in Interleukin Genetics. All other authors: no conflicts.

References


Table 4. Variables affecting duration of hospital stay and development of clinical sequelae

<table>
<thead>
<tr>
<th>Outcome, variable</th>
<th>Multivariatea OR (95% CI)</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Duration of hospitalization &gt;10 days</td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Neisseria meningitidis</em> bacterial load (per log10 copy/mL increase)</td>
<td>1.36 (1.12–1.65)</td>
<td>.002</td>
</tr>
<tr>
<td>Age (per 1-year increase)</td>
<td></td>
<td>.003</td>
</tr>
<tr>
<td>Auditory deficit: IL-1R+M+2018+2</td>
<td>0.52 (0.28–0.97)</td>
<td>.038</td>
</tr>
<tr>
<td>Visual deficit: age (per 1-year increase)</td>
<td>1.04 (1.02–1.05)</td>
<td>&lt;.001</td>
</tr>
<tr>
<td>Need for hemodialysis</td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Neisseria meningitidis</em> bacterial load (per log10 copy/mL increase)</td>
<td>1.64 (1.21–2.22)</td>
<td>.001</td>
</tr>
<tr>
<td>Age (per 1-year increase)</td>
<td>1.03 (1.01–1.06)</td>
<td>.012</td>
</tr>
<tr>
<td>Digit or limb loss: <em>Neisseria meningitidis</em> bacterial load (per log10 copy/mL increase)</td>
<td>1.54 (1.08–2.20)</td>
<td>.017</td>
</tr>
</tbody>
</table>

NOTE. The variables included for each model were bacterial load, age, serogroup B, serogroup C, IL-1R+M+2018+2 carriage, and IL-1R+M+2018+2 carriage. Only statistically significant variables are shown.

a Multivariate logistic regression model.


