EXPRESSION OF FILARIAL-SPECIFIC IgG SUBCLASSES UNDER DIFFERENT TRANSMISSION INTENSITIES IN A REGION ENDEMIC FOR LOIASIS

J. P. AKUE, E. DEVANEY, G. WAHL, AND H. MOUKANA
Department of Medical Parasitology, Centre International de Recherches Médicales de Franceville, Gabon; Department of Veterinary Parasitology, Veterinary School, University of Glasgow, Glasgow, United Kingdom

Abstract. Specific IgG subclasses were investigated in two villages (Okoumbi and Ndjokaye) in southeast Gabon with different Loa loa transmission intensities of ~9,000 and 1,300 infective larvae (L3) per person per year, respectively. IgG subclasses were measured by an enzyme-linked immunosorbent assay (ELISA) using extracts of L. loa L3, microfilariae (MF), or adult worms. Levels of L3-specific IgG3 were significantly higher in the village with low transmission (Nnjokaye) (P = 0.006). In contrast, MF-specific IgG2 was significantly higher in Okoumbi than in Ndjokaye (P = 0.0009). In the high-transmission village (Okoumbi), levels of both MF- and adult-specific IgG4 were significantly increased in MF carriers compared with microfilaricaric subjects (P = 0.0015 and P = 0.0003, respectively), while levels of L3- and MF-specific IgG1 were significantly higher in microfilaricaric individuals compared with MF carriers (P = 0.04 and P = 0.03, respectively). Furthermore, among microfilaricaric individuals, the level of the specific IgG1 subclass was much lower in Okoumbi than in Ndjokaye (P = 0.036). These results suggest that the expression of antigen-specific IgG3 and IgG2 is more likely to vary with transmission intensity, whereas antigen-specific IgG4 and IgG1 varies with adult worm and MF burden.

INTRODUCTION

Loa loa is a human filarial parasite that is endemic to the West African tropical forest region, with an estimated 3 million people infected.1 Symptoms include allergic signs, hydrocele, inflammation of the conjunctiva, cardiopathy, and encephalitis.2 While in the absence of an effective therapy for infection with adult worms, any alternative means of control requires an improved knowledge of the disease. We have previously reported that occult infection, or the microfilaremic state, was the most common presentation of infection (70% of the endemic population in the region studied).3 Previous studies have shown that levels of IgG4 antibodies were increased in both microfilaricaric and microfilaricaric individuals,5 whereas levels of IgG1 were increased only in microfilaricaric subjects,6 suggesting a relationship between parasitic status and the immune response. However, the immunologic factors implicated in the spectrum of infection remain ill-defined. In other filarial infections, humoral immune responses vary with transmission levels,7 while seasonal changes in microfilariae (MF) density are reported to influence cellular immune responses (interferon-γ [IFN-γ] production) to Brugia malayi.8 In the absence of studies on the relationship between the intensity of transmission and loiasis, it is not known whether variation in transmission rates may affect the immune response in this infection. In this study, we have compared the levels of different IgG subclasses among residents of two villages in Gabon that differed in the number of infective larvae (L3) potentially infecting an individual per year.

MATERIALS AND METHODS

Entomologic studies. Two villages in southeast Gabon were designated for entomologic follow-up: Okoumbi and Ndjokaye, which are situated in the forest and savannah area, respectively. Chrysops were captured in these villages twice a week for more than a year from 7:00 AM to 6:00 PM. Vectors were identified and brought to the laboratory for microscopic dissection to examine the presence of L3 and the maturity of the females (gonadotrophic status). The daily biting rate (DBR) was defined as the number of bites by Chrysops per day. The daily transmission potential (DTP) was calculated as the DBR × the percentage of infected Chrysops × the mean number of L3 per infected Chrysops. The monthly transmission potential (MTP) is equal to the DTP × 30 days. Therefore, the annual transmission potential (ATP) was calculated as the MTP × 12 months. The ATP was defined as the number of L3 potentially infecting an individual per year (virtual ATP). The actual ATP (or corrected ATP) was defined as the transmission potential taking into account the actual exposure. To determine the actual exposure, four individuals were followed up for four days during their activities in the village and in their plantation. One individual served as a standard and any Chrysops approaching this individual were captured. The other three individuals were observed to 1) determine their natural behavior towards Chrysops, 2) estimate the number of Chrysops that landed on their bodies but could not bite for some reason (e.g., chased away, killed before biting), and 3) measure those that landed and bit these individuals. The mean number of Chrysops recorded in each of these scenarios for these three individuals over the four days of follow-up were determined. The percentage of Chrysops that bit human subjects and the ratio of the DBR for three individuals to the DBR of the standard individual were taken into account when estimating the corrected ATP. Vectorial capacity was also determined during this study as total number of L3 found in all dissected Chrysops × the percentage of infected Chrysops.

Study population. The populations of these two villages donated blood after informed consent was obtained from all individual participants. The project was approved by the Ethical Committee of the Centre International de Recherches Médicales de Franceville. Blood was examined for the presence of circulating MF using a modified Knott’s technique4, and plasma was collected and stored at −20°C for further analysis of IgG subclasses. To avoid any potential age bias, only individuals more than 18 years of age were selected (Okoumbi, n = 68; Ndjokaye, n = 44). Subsequently, the population was divided into four groups: high microfilaricaric individuals (Okoumbi, n = 12; Ndjokaye, n = 6; MF ≥ 100/ml); low microfilaricaric individuals (Okoumbi, n = 6; Ndjokaye,
n = 6; MF < 100/ml); amicrofilaremic individuals with the ocular passage of an adult worm, calabar edema, or positive IgG4 serology indicative of active infection (Okoumbi, n = 46; Ndjokaye, n = 29); and endemic controls with no parasitologic or clinical sign of filarial infection (Okoumbi, n = 4; Ndjokaye, n = 3). Another species of filarial parasite, *M. perstans*, was prevalent in both villages and was distributed equally among *L. loa*-infected groups. All parasitized individuals were later treated by a general practitioner (G.P.). Serum samples were scored as positive for IgG4 on the basis of an optical density (OD) value greater than the mean + 1 SD of 11 serum samples from Gambians infected with *M. perstans* but not *L. loa*. Endemic controls were residents of the village who did not have *L. loa* or *M. perstans*. Caucasians (n = 4) visiting Africa for the first time also served as non-endemic controls.

**Parasite materials.** Adult worms of *L. loa* were removed by an ophthalmologist during ocular passage. The MF were obtained from heavily infected patients and purified on a Percoll gradient, as previously described. The L3 were obtained by growing extract of adult, MF, or L3 of *M. perstans* was prevalent in both villages and was distributed equally among *L. loa*-infected groups. All parasitized individuals were later treated by a general practitioner (G.P.). Serum samples were scored as positive for IgG4 on the basis of an optical density (OD) value greater than the mean + 1 SD of 11 serum samples from Gambians infected with *M. perstans* but not *L. loa*. Endemic controls were residents of the village who did not have *L. loa* or *M. perstans*. Caucasians (n = 4) visiting Africa for the first time also served as non-endemic controls.

**Antigen preparations.** Adult worms or L3 were washed and then homogenized in 10 mM Tris-HCl, pH 8.3, containing protease inhibitors. The MF were disrupted by sonication in the same buffer. Antigens were extracted with 1% sodium deoxycholate for 1 hr at 4°C. The extracts were centrifuged at 10,000 × g for 10 min, and the soluble supernatant was aliquoted and stored at −70°C. The protein content was measured using the Bio-Rad (Richmond, CA) method.

**Enzyme-linked immunosorbent assay.** Detergent-soluble extracts of adult, MF, or L3 of *L. loa* were diluted in carbonate-bicarbonate buffer, pH 9.6, and 100-μl volumes were distributed into the wells of microtiter plates (Immunolon II; Dynatech Laboratories, Chantilly, VA) at a concentration of 10 μg/ml. After an overnight incubation at 4°C, the plates were washed three times (10 min/wash) at room temperature with 50 mM Tris-HCl (pH 7.4), 200 mM NaCl, and 0.05% Tween 20 (TBST). The plates were then blocked for 1 hr with TBST-5% bovine serum albumin (BSA), and the washing step was repeated. Human serum diluted 1:200 in TBST-1% BSA was added for 1 hr, and the washing step was repeated and followed by incubation with mouse monoclonal antihuman subclass reagents (Oxoid Unipath, Hampshire, United Kingdom) at the following dilutions: IgG1, 1:2,000 (HP6012); IgG2, 1:2,000 (HP6014); IgG3, 1:1,000 (HP 6050); and IgG4, 1:30,000 (HP 6011). Optimal dilutions of each subclass-specific reagent detected the appropriate subclass as the correct proportion of total IgG, while antigen and serum dilutions were optimized by checkerboard titration. After three washes in TBST, the plates were incubated with a 1:1,000 dilution of anti-mouse IgG Fc-specific alkaline phosphatase conjugate (Sigma, St. Louis, MO) for 1 hr. The reaction was revealed with p-nitrophenyl phosphate (Sigma) diluted in diethanolamine buffer, pH 9.8, and the OD was read at 405 nm.

**Statistical analysis.** The Mann-Whitney U test was used to assess the significance of the differences in median values between IgG subclasses. The chi-square test was used for comparisons between the sexes and for the distribution of vectors. *P* values < 0.05 were considered significant.

**RESULTS**

**Entomologic characteristic of the villages.** The ATP for *L. loa* was estimated for both villages as described in the Materials and Methods. This analysis demonstrated that the ATP in Okoumbi was approximately seven times higher than that in Ndjokaye. Therefore, Okoumbi was designated as a high transmission area with an actual ATP of ∼9,000 L3 per person per year, while Ndjokaye was designated as a low transmission area with ∼1,300 L3 per person per year (Table 1). Two vectors (*Chrysops silacea* and *C. dimidiata*) were found to be prevalent in both villages. *C. silacea* had the highest vectorial capacity and was the predominant vector in both Okoumbi and Ndjokaye, accounting for 91% and 79% of *Chrysops* caught in the respective villages (Table 2). No statistically significant difference in the distribution of *C. silacea* was found between the two villages (χ² = 0.878, degrees of freedom [df] = 1, *P* = 0.349).

**Study population and parasitologic examination.** Sixty-eight adults from Okoumbi and 44 adults from Ndjokaye participated in the study. There was no significant difference in sex ratios in these villages (χ² = 1.067; df = 1, *P* = 0.302 and χ² = 0.045, df = 1, *P* = 0.831 for Okoumbi and Ndjokaye, respectively), or in age of the study populations (*P* = 0.15). In Okoumbi, 18 subjects had circulating MF in their blood (mean = 8,930 MF/ml) and in Ndjokaye, 12 individuals had circulating MF (mean = 204 MF/ml). Although the mean MF density in Okoumbi was much higher than that in Ndjokaye (Table 3), neither the difference in MF density (*P* = 0.065) nor prevalence reached statistical significance in the two villages, despite the large difference in transmission intensity. Another filarial parasite, *M. perstans*, was present in both villages and was distributed equally among the infected groups.

**Analysis of IgG subclass responses in the two villages.** The level of each IgG subclass was determined in participating individuals more than 18 years of age in the two villages. There were no significant differences in any subclass response to adult antigen between overall population of the two villages.

---

**Table 1**

<table>
<thead>
<tr>
<th>Vectorial capacity in each village</th>
<th>Okoumbi</th>
<th>Ndjokaye</th>
</tr>
</thead>
<tbody>
<tr>
<td>Capture (number of days)</td>
<td>86</td>
<td>64</td>
</tr>
<tr>
<td>Daily Biting Rate (DBR)</td>
<td>93</td>
<td>15</td>
</tr>
<tr>
<td>Daily Transmission Potential (DTP)</td>
<td>184</td>
<td>26</td>
</tr>
<tr>
<td>Monthly Transmission Potential (MTP)</td>
<td>5520</td>
<td>780</td>
</tr>
<tr>
<td>Annual Transmission Potential (ATP)</td>
<td>66240</td>
<td>9360</td>
</tr>
<tr>
<td>Corrected ATP</td>
<td>9273</td>
<td>1310</td>
</tr>
</tbody>
</table>

**Table 2**

<table>
<thead>
<tr>
<th>Vector</th>
<th>Total</th>
<th>% Infected</th>
<th>L3/Chrysops</th>
<th>Vectorial capacity</th>
<th>Distribution</th>
</tr>
</thead>
<tbody>
<tr>
<td>C. silacea</td>
<td>3,103</td>
<td>1.7</td>
<td>152</td>
<td>2.6</td>
<td>91/79</td>
</tr>
<tr>
<td>C. dimidiata</td>
<td>1,542</td>
<td>0.6</td>
<td>36</td>
<td>0.2</td>
<td>9/21</td>
</tr>
</tbody>
</table>

*Total = Total number of flies of either species collected; L3/Chrysops = mean number of infective L3 per fly; Vectorial capacity = total number of L3 in all dissected Chrysops x % infected Chrysops. Distribution = % of species in Okoumbi/Ndjokaye.*
lages, or in IgG1 or IgG4 levels to MF or L3 antigen. How-
however, levels of MF-specific IgG2 were significantly increased
in Okoumbi, the high transmission village ($P = 0.0009$), while
levels of L3-specific IgG3 were significantly higher in Nd-
jokaye, the low transmission village ($P = 0.006$). These re-
sults are summarized in Table 4.

To investigate whether the differences observed in IgG sub-
classes in the two villages were related to the presence or
absence of MF, rather than to transmission intensity, IgG subclass levels were measured in microfilaremic versus am-
icrofilaremic individuals in the two villages. When L3 antigen
was used, the level of IgG1 was significantly higher in am-
crofilaremic individuals compared with MF+ individuals, but
only in the high transmission village of Okoumbi (mean ± SD
= 0.509 ± 0.439 versus 0.285 ± 0.227; $P = 0.04$). No other
difference reached statistical significance either between
groups or between the two villages. When adult antigen was
used, the only difference to reach significance was the level of
IgG4, which was significantly higher in MF+ individuals in
Okoumbi than in microfilaremic subjects in either Okoumbi
(1.886 ± 0.889 versus 1.071 ± 0.991; $P = 0.003$) or Ndjokaye
(1.886 ± 0.889 versus 0.879 ± 0.843; $P = 0.0006$). When MF
antigen was used, a similar pattern of IgG4 reactivity was
observed with IgG4 levels being significantly higher in MF+
individuals from Okoumbi compared with amicrofilaremic in-
dividuals in the same village (1.027 ± 0.853 versus 0.471 ±
0.529; $P = 0.0015$) or to amicrofilaremic individuals from the
low transmission village (1.027 ± 0.853 versus 0.378 ± 0.311; $P
= 0.0011$). Conversely, in the high transmission village, the
level of specific IgG1 was significantly lower in MF+ subjects
than in amicrofilaremic subjects (0.627 ± 0.395 versus 0.364 ±
0.316; $P = 0.04$). When MF+ subjects were compared be-
tween the two villages, levels of IgG1 were significantly increased in
the low transmission village compared with the high transmis-
sion village (0.627 ± 0.395 versus 0.364 ± 0.316; $P = 0.036$),
while amicrofilaremic individuals in both villages had similar
levels of IgG1 ($P = 0.445$). A similar trend was observed with
IgG1 responses to adult antigen; MF+ subjects from Nd-
jokaye had higher IgG1 levels (mean ± SD = 1.59 ± 0.738)
compared with those from Okoumbi (0.970 ± 0.644), but the
difference did not reach statistical significance ($P = 0.054$).
Thus, the analysis of IgG subclasses in MF+ individuals versus
amicrofilaremic individuals highlighted significant differences in
IgG1 and IgG4 levels, and these differences were much more
pronounced under conditions of high transmission.

To investigate whether the observed differences in IgG levels in
the two villages were a function of transmission intensity or
were related to microfilarial density, the population was then
further divided into four different groups: endemic controls, amicrofilaremic individuals, high microfilaric in-
dividuals ($\geq 100$ MF/ml), and low microfilaric individuals
($< 100$ MF/ml). Comparisons were made both between dif-
cerent clinical groups in each village and between the same
groups in the different villages. When L3 antigen was used in

### Table 3

<table>
<thead>
<tr>
<th>Village</th>
<th>Number (Male/Female)</th>
<th>Mean age (years)</th>
<th>Microfilaremics (prevalence)</th>
<th>MF density (MF/mL)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Okoumbi</td>
<td>68 (28/40)</td>
<td>48.5</td>
<td>18 (28%)</td>
<td>8930 ± 21270</td>
</tr>
<tr>
<td>Ndjokaye</td>
<td>44 (21/23)</td>
<td>54.7</td>
<td>12 (27%)</td>
<td>204 ± 287</td>
</tr>
</tbody>
</table>

### Table 4

<table>
<thead>
<tr>
<th>Group status</th>
<th>IgG1 (mean ± SD)</th>
<th>IgG2 (mean ± SD)</th>
<th>IgG3 (mean ± SD)</th>
<th>IgG4 (mean ± SD)</th>
</tr>
</thead>
<tbody>
<tr>
<td>L3 Ag</td>
<td>0.417 ± 0.387</td>
<td>0.187 ± 0.141</td>
<td>0.119 ± 0.086</td>
<td>0.434 ± 0.582</td>
</tr>
<tr>
<td>MF Ag</td>
<td>0.536 ± 0.447</td>
<td>0.191 ± 0.114†</td>
<td>0.214 ± 0.098</td>
<td>0.434 ± 0.582</td>
</tr>
<tr>
<td>Adult Ag</td>
<td>1.047 ± 0.762</td>
<td>0.390 ± 0.341†</td>
<td>0.174 ± 0.305</td>
<td>1.276 ± 1.032</td>
</tr>
</tbody>
</table>

* $P = 0.006$ L3-specific IgG1 low transmission versus high transmission village.
† $P = 0.0009$ MF-specific IgG2 high transmission versus low transmission.
the ELISA (Table 5), the only result to reach statistical significance was the level of IgG3 in high microfilaremic individuals, which was significantly greater in Ndjokaye compared with high microfilaremic individuals \( (P = 0.039) \) or to low microfilaremic individuals \( (P = 0.036) \) in Okoumbi. However, the levels of IgG3 did not vary significantly between high and low microfilaremic individuals or amicrofilaremic individuals in each village.

In the high transmission village, IgG4 levels to adult antigen were increased in the high MF group compared with the low MF group \( (P = 0.013) \) or the amicrofilaremic group \( (P = 0.0009) \) (Table 6). When villages were compared, there was no significant difference in IgG4 levels between the high MF group of either village \( (P = 0.246) \), although IgG4 was significantly increased in the high MF group in Okoumbi compared with the low MF group \( (P = 0.022) \), or the amicrofilaremic group \( (P = 0.0002) \) in Ndjokaye. When MF antigen was used (Table 7), the level of IgG1 was significantly increased in amicrofilaremic individuals from the high transmission village compared with the low MF and high MF groups from the same village \( (P = 0.0209 \text{ and } P = 0.042, \text{ respectively}) \). However, no differences between the high and low microfilaremic groups of the same village were detected for any IgG subclass \( (P > 0.05) \). The level of MF-specific IgG4 in the high MF group in the high transmission village was significantly greater than in the low MF group from the low transmission village \( (P = 0.044) \).

**DISCUSSION**

To date, there have been no investigations on the possible effects of transmission intensity on the immune response in loiasis, although previous studies have shown differential immune responses between parasitologically defined groups.\(^6,11\) Two villages, each with well characterized entomologic data on transmission, were used in this study to determine whether transmission intensity had an influence on the expression of different IgG subclasses. *Chrysops silacea*, the vector with the highest vectorial capacity, was the most prevalent in both villages; therefore, differences between the villages could not be attributed to variation in the efficiency of the vector. The ATP for Okoumbi was approximately seven-fold greater than for Ndjokaye, an observation consistent with the generally higher levels of infection in the former village. Data were analyzed in four steps: first, a comparison of the prevalence of each IgG subclass was carried out between villages. Second, the mean level of subclasses between the study populations of both villages was compared. Third, differences due to the presence or absence of MF were assessed by comparing IgG levels in MF+ and amicrofilaremic subjects. Fourth, differences due to MF density rather than transmission intensity were examined by comparing high microfilaremic subjects, low microfilaremic subjects, and amicrofilaremic individuals in both villages.

No statistically significant differences in MF density or microfilarial prevalence were seen between villages, although MF levels tended to be higher under conditions of intense transmission. These observations are in agreement with a previous report showing the spatial stability of MF prevalence.\(^4\) Such stability has not been observed with immunologic markers. In other filarial infections, increased levels of IgG have been shown to be associated with high transmission intensity.\(^7\)
### Table 6
Mean levels (OD ± SD) of IgG subclasses using the adult antigen

<table>
<thead>
<tr>
<th>Group status</th>
<th>High transmission village</th>
<th>Low transmission village</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>IgG1</td>
<td>IgG2</td>
</tr>
<tr>
<td>Amicrofilaremics</td>
<td>1.134 ± 0.800</td>
<td>0.454 ± 0.367</td>
</tr>
<tr>
<td>High MF+</td>
<td>1.048 ± 0.690</td>
<td>0.251 ± 0.218</td>
</tr>
<tr>
<td>Low MF+</td>
<td>0.767 ± 0.600</td>
<td>0.320 ± 0.309</td>
</tr>
<tr>
<td>Endemic control</td>
<td>0.322 ± 0.367</td>
<td>0.114 ± 0.054</td>
</tr>
</tbody>
</table>

* Level of IgG4 in high MF+ significantly elevated over low MF+ (P = 0.013) and amicrofilaremic (P = 0.0009) from high transmission village and significantly higher than low MF+ and amf from Ndjokaye (P = 0.022, P = 0.0002, respectively).

### Table 7
Mean levels (OD ± SD) of IgG subclasses using MF antigen

<table>
<thead>
<tr>
<th>Group status</th>
<th>High transmission village</th>
<th>Low transmission village</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>IgG1</td>
<td>IgG2</td>
</tr>
<tr>
<td>Amicrofilaremics</td>
<td>0.645 ± 0.5*</td>
<td>0.206 ± 0.122</td>
</tr>
<tr>
<td>High MF+</td>
<td>0.413 ± 0.370</td>
<td>0.172 ± 0.090</td>
</tr>
<tr>
<td>Low MF+</td>
<td>0.240 ± 0.155</td>
<td>0.188 ± 0.069</td>
</tr>
<tr>
<td>Endemic control</td>
<td>0.182 ± 0.153</td>
<td>0.108 ± 0.031</td>
</tr>
</tbody>
</table>

* IgG1 significantly elevated in amicrofilaraemics versus low MF+ (P = 0.029) and high mf (P = 0.042) within the high transmission village.
† IgG4 significantly elevated in high MF+ versus low MF+ of low transmission village (P = 0.044).
In the present study, levels of L3-specific IgG3 and MF-specific IgG2 were significantly different between the two villages. In the absence of any statistical differences in MF density, age or sex between the villages, it is likely that the difference in transmission intensity causes these differences in sub-class responses. Furthermore, when the population was split into different groups, no association was found between the presence or the density of MF and the level of specific IgG3 and IgG2, further supporting the hypothesis that both subclasses were influenced by transmission intensity. L3-specific IgG3 was highest in the low transmission area, while MF-specific IgG2 was higher in the high transmission village. Whether these differences relate to exposure to L3 or are modulated by other life cycle stages remains to be determined.

Previous work on well-defined groups of microfilaricarriers of *L. loa* demonstrated that the level of antigen-specific IgG4 was similar between groups. However, the present analysis shows that levels of adult antigen-specific IgG4 are higher in microfilarie as opposed to amicrofilaricarriers of *Loa loa* and *M Mansonella perstans*, but that these differences are only apparent under conditions of high transmission intensity. In contrast, levels of IgG1 to both L3 and MF antigen were significantly higher in amicrofilaricarriers than in MF+ subjects. Differences were also apparent between the two villages in MF+ subjects, with IgG1 levels being significantly higher in the low transmission village than in the high transmission village. These data confirm a negative association between the presence of MF and levels of IgG1. In this case, it is likely that the presence of MF may actively down-regulate IgG1 levels. Interferon-γ is known to be selectively induced by MF in a mouse model of *Brugia* infection, and IFN-γ can also down-regulate the synthesis of the IgG1 subclass in humans. Alternatively, specific IgG1 may be associated with resistance to infection, as previously suggested, since an inverse correlation was reported between MF density and IgG1. Similarly, in this study it was found that the village with the lowest MF density had increased IgG1 levels, whereas the other village had a higher MF density (although not statistically significant) and lower IgG1 levels. The fact that the immune response to extracts of different parasite stages gave characteristic profiles indicates that there is an element of stage-specificity to the humoral immune response.

The present study demonstrates a dissociation in the response of IgG4 and IgG1 depending on whether an individual was microfilaricar, or from one particular village. This suggests that IgG4 and IgG1 are not regulated by the same factors, although it is generally accepted that the humoral immune response is under Th2 control. It has been proposed that IgG4 may block IgE and IgG3 and thus prevent pathologic responses. Thus, the pressure on the immune system induced by high levels of transmission may result in a shift in expression of IgG subclasses to avoid pathology. Alternatively, the new incoming parasite stage may also modulate this shift for its own survival. In conclusion, we have shown that IgG4 and IgG1 levels vary with the presence, absence, or density of MF, while levels of L3-specific IgG3 decrease and MF-specific IgG2 increase with transmission intensity.

Acknowledgments: We thank Drs. Jean Wickings, P. Deloron, and Nicholas Anthony for reviewing the manuscript.

Financial support: This study was partially supported by Wellcome Trust grant 047101/2/96/077/CSD/JL/CG. The Centre International de Recherches Médicales de Franceville is supported by Elf-Gabon, State of Gabon, and The Ministère Français de la Cooperation.

Authors’ addresses: J. P. Akue, G. Wahl, and H. Moukana, Centre International de Recherches Médicales de Franceville, BP 769, Franceville, Gabon. E. Devaney, Department of Veterinary Parasitology, Veterinary School, University of Glasgow, Bearsden Road, Glasgow G61 1QH, United Kingdom.

Reprint requests: J. P. Akue, Centre International de Recherches Médicales de Franceville, BP 769, Franceville, Gabon.

REFERENCES


