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Human water contacts patterns in *Schistosoma mansoni* epidemic foci in northern Senegal change according to age, sex and place of residence, but are not related to intensity of infection

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Summary In an epidemic focus in northern Senegal, adults had lower intensities of infection than adolescents, a phenomenon that could not be attributed to immunity acquired over the previous 10–15 years of exposure to the parasite because all age groups had had the same number of years' experience of the worm. This article considers whether this pattern could have been because of higher levels of exposure to the parasite in younger age groups. Personal contact with infected water was recorded using a questionnaire in *Schistosoma mansoni* foci not more than 3 years old and in another, 10-year-old focus. Many aspects of contact (e.g. frequency, duration or time of day of contact) may contribute to the number of encounters with infective cercariae (true exposure), so various assumptions regarding the relationship between water contact and true exposure were tested resulting in a range of exposure indices. People reported a mean of 4.4 separate contacts, and spent a median of 57 min per day in water. Patterns of water contact differed depending on the exposure index used, e.g. considering duration, males spent a longer time in water than females ($P < 0.001$). But using frequency, females had more contacts with water than males in most villages ($P < 0.001$). Generally, exposure levels dropped as people become aged ($P < 0.001$) and residents of the older focus were more exposed than residents of other foci ($P < 0.002$). Intensity of (re)infection was not related to exposure either alone or in models incorporating age, sex and/or village irrespective of the index used. There is therefore evidence that age, sex and place of residence determine exposure but none to suggest that exposure had an influence on the relationship between these factors and intensity of infection. We propose therefore that in this population other factors have principal importance in determining intensity of infection.

keywords *Schistosoma mansoni*, human, Senegal, water contact

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Introduction

Prevalence and intensity of *Schistosoma mansoni* infection in humans characteristically increases during early childhood, peaks during adolescence and drops to much lower levels thereafter. In a young epidemic focus in northern Senegal, adults had lower intensities of infection than adolescents (Gryseels *et al*. 1994). This difference between adults and adolescents could not be as a result of immunity acquired over the previous 10–15 years of exposure to the parasite because, in this recent focus, the adults had had the same number of years' exposure of schistosomiasis as children, defined in this context as 'the number of years in daily contact with infected water'. The observations in the epidemic focus raised the possibility that host maturation in itself could be one factor in resistance to *S. mansoni* (Gryseels 1994) although the most obvious explanation of differing prevalence and infection intensity in study populations is the contribution of individual levels of exposure to infected water. We therefore investigated whether individual heterogeneity in intensity of infection, in residents of five recently infected villages in northern Senegal, could be affected by water contact behaviour.

Changing levels of exposure have been shown to influence intensity of infection in some foci (Wilkins *et al.* 1987a); but Butterworth *et al.* (1992) reviewed evidence from several studies and concluded that exposure alone does not explain the drop in infection that occurs with age. Intrinsic to this conclusion was evidence from The Gambia and Kenya. Adult Gambian women were as heavily exposed as children despite having significantly lower intensities of re-infection with *S. haematobium* (Wilkins *et al.* 1987b; Hagan 1992). In Kenya, the level of re-infection with *S. mansoni* was highest in children aged between 8 and 12 years, whereas greatest exposure to infected water did not occur until the ages of 16–24 years. Even when water contact rates were taken into account, they did not explain the large differences in infection intensities between the different age groups (Butterworth *et al.* 1988). This study differs from exposure studies of endemic populations in that subjects in any one village had been exposed to the parasite for the same number of years.

The type of water contact activity, the age groups in which it is important and the extent of the relationship with intensity or prevalence of infection vary between studies. Chandiwana and Woolhouse (1991) observed a correlation between individual contact rates and intensity of infection (*S. haematobium*), although it accounted for less than 3% of the variation in intensity of infection. In Egypt, 12–13-year-old boys had both the highest geometrical mean (GM) intensity of infection (*S. haematobium*), and the highest levels of exposure (Kloos *et al.* 1983). In Brazil, frequency of fishing and washing parts of the body were correlated with GM intensity of infection (*S. mansoni*), although the predictive value of the model was limited (Kloos *et al.* 1998). Factors associated with intensity of infection (*S. mansoni*) in rural Kenya differed according to area and age group (Kloos *et al.* 1997), although the relationship between age and intensity of infection remained the same even in villages with very different patterns of water contact behaviour (Kabatereine *et al.* 1999). These differences highlight the importance of addressing the issue of exposure afresh in each new situation.

Water contact is not in itself a measure of exposure. Many aspects of contact (such as the frequency or total duration of contact, how much of the body is exposed and when) may contribute to the likelihood of encountering infective cercariae. These and other factors have been used by other authors to approximate exposure from water contact information (Wilkins *et al.* 1987a; Chandiwana & Woolhouse 1991; Demeure *et al.* 1993; Etard *et al.* 1995; Fulford *et al.* 1996; Kloos *et al.* 1998; Gazzinelli *et al.* 2001). However, it is far from clear which of the many available exposure indices most accurately reflects true exposure. Our approach was therefore to test a range of assumptions rather than to assume that one index is better than another. The analysis focuses on differences in exposure index in terms of age, sex, number of years' exposure and intensity of infection.

Materials and methods

Study sample

This study took place in five villages. Ndigue, Maraye, Ravette and Rone are clustered together to the north-east of the district of Saint Louis, Senegal, in an *S. mansoni* focus not more than 3 years old. Ten per cent of the population of the villages were surveyed in 1994 and found to be negative for *S. mansoni* (Ernould 1996). In 1997, the prevalence of *S. mansoni* infection was measured in all compliant, permanently resident volunteers (77% of a total population of 1095). It was 73% in Ndigue [95% confidence interval (CI): 65–80], 62% in Maraye (95% CI: 55–68), 84% in Ravette (95% CI: 75–87) and 86% in Rone (95% CI: 73–95). A fifth village, Buntabat, just to the south of Richard Toll, had had cases of *S. mansoni* infection since the beginning of the outbreak in northern Senegal in 1989. In 1997 the prevalence of infection was 70% (95% CI: 63–76).

Residents of the villages had no access to piped water and used principally canal or river water for domestic and agricultural purposes. Economic activities include making straw mats, fishing and growing rice, all of which entail spending long periods of time in infected water. Water contact sites are within a 5-min walk from all houses in Rone, Ravette and Buntabat, whereas in Maraye and Ndigue, water collection occurred at a site, a 10–15-min walk from the village.

All permanent residents were invited to participate in the study. People who frequently travelled out of the area were excluded, including absences for schooling, temporary work (migrant fishermen) and travelling trade. Some chose not to participate in the study at all. Reasons for this included personal dislike at giving stool and urine samples or lack of permission to participate. For example, men who were working away at the time of recruitment headed some households, and therefore could not give their permission. This excluded 238 people (22%). No permanent residents reported having moved into the area from somewhere else, although there was some movement between Ndigue, Maraye, Ravette and Rone. A further 231 were excluded because they were under 8 years old. Young children were excluded because the older focus was approximately 8-year-old and we wanted all subjects in that focus to have had the same number of years' exposure.

In the remaining population who decided to take part (626 people) compliance was very good. Complete samples for diagnosis of *S. mansoni* were received from 98% before treatment. Of the remaining 615 people, 239 returned water contact questionnaires in all three seasons. The main reason for this low follow-up for all three questionnaires was lack of manpower: two interviewers had to apply all questionnaires. Of the participating population ($n = 626$), treatment was not verified in 10, these were therefore excluded from the analysis 1 year after treatment. Compliance 1 year later was lower than pre-treatment levels: 54% of the possible 616 did not give all samples necessary to ascertain intensity of *S. mansoni* infection. Of these 334 people, water contact questionnaires were completed in all three seasons for 147 people. There was no significant difference in intensity of infection between those who gave all three questionnaires and those who did not before or after treatment (pre-treatment: $P < 0.3$, 1 year post-treatment: $P < 0.4$: measured using a negative binomial regression).

Determination of intensity of infection

Infection was measured by egg counts in 2 × 25 mg kato slides from each of two stools taken on different days using the Kato-Katz method (Katz *et al.* 1972). Sample collection pots were labelled with the name of the subject, personal identification number and whether it was 'pot 1' or 'pot 2'. Pots were labelled in both Roman and Arabic script because adults read only Arabic script. First samples were collected the following morning after distribution of pots and the second sample pots immediately distributed. Those who had not given a sample were reminded and their samples collected the next day. Kato-Katz slides were prepared immediately on return to the laboratory, never more than 5 h after collection. Slides were stored at 4 °C for 48 h and read by two experienced microscopists, who each read one slide per stool sample. Ten per cent of slides were re-read on the same day by a third microscopist to ensure continued quality of diagnosis. A range of quality control checks maximized the quality of the data. These included double checks on the person identification numbers on sample pots and on slides, the dates of arrival of stool samples compared with the dates, results were given and a double-entry data check protocol.

Treatment

All subjects were treated using a standard course of praziquantel (40 mg/kg) and followed at 6 weeks to assess the efficacy of chemotherapy. Individuals who remained positive at 6 weeks were retreated. Re-infection was measured 1 year post-treatment.

Measurement of water contact activities

Personal water use was recorded using a questionnaire which asked about water contact activity 'yesterday', and also 'usual' water contact. Each subject was asked to return three questionnaires, one in February, one in July and one in October corresponding to the cold dry, hot dry and hot wet seasons. Residents were asked about their contact with water bodies confirmed as habitats for the intermediate snail host *Biomphalaria* spp. (usually *Biomphalaria pfeifferi*). The design of the questionnaire was performed in collaboration with focus groups, then the design was adjusted and the interview approach practised until water contact activities observed directly were actually found reported in questionnaires the next day. Two adult male members of the field team who were resident in the villages at the time carried out direct observations. They noted water contacts as they went about their normal routine in the village, in order to minimize the possibility of changing behaviour. Before commencing the water contact study, the aim of the questionnaire and the study was fully explained to residents of the study villages. Questionnaires were administered by two trained interviewers (members of the field team) who had previously worked in the village for over a year, had been actively involved in the design and testing of the questionnaire and quality control protocols. They were resident in the village for the duration of the water contact study.

Data analysis

Several exposure indices based on commonly used indices from the literature were calculated to approximate exposure in different ways: frequency of contact with water, total duration of contact with water, duration × body surface (BS) exposed, duration × BS exposed × time of day (assuming that 0.1% of cercariae were viable after 24 h) and duration × BS exposed × time of day (assuming that 33% of cercariae were still viable after 24 h) (Tchuem Tchuente *et al.* 1999). Body surface area was calculated from individual weight and height measurements (Mosteller 1987). The percentage of the body exposed was calculated from charts used to assess the severity of burns (Lund & Browder 1944). Exposure indices were calculated either including or not including domestic water, i.e. water collected from infected sites, but used in the home. Two assumptions regarding seasonality of transmission were tested: (1) assuming that transmission was equal in all seasons; (2) the season was weighed according to the mean prevalence of infection in snails in the area in that season.

Time of day correction

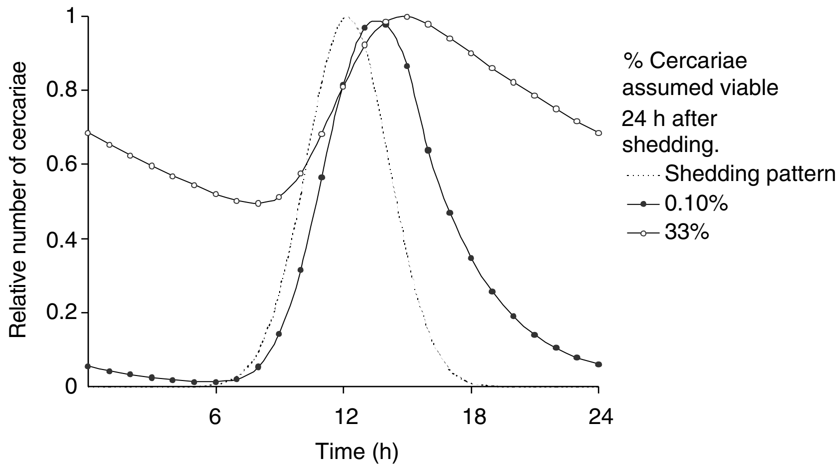
The relative number of cercariae shed at different times of day was assumed to follow a bell curve peaking just after midday – as previously used by Chandiwana and Woolhouse (1991). This shape approximates the shedding patterns of *S. mansoni* cercariae from *B. pfeifferi* snails from northern Senegal (Tchuem Tchuente *et al.* 1999). Chandiwana and Woolhouse (1991) assumed that cercariae were immediately swept away from water contact sites after shedding. However, in the current study villages, discrete water contact sites were rare; rather, people used water all along a stretch of river or canal. Cercariae swept away from one site were therefore assumed to be replaced from further upstream. The rate at which infective cercariae left water via death, predation, or entering a host was assumed to occur at an exponential rate. One exposure index, 'fast', assumed 0.1% of cercariae were viable after 24 h (Prentice & Ouma 1999). Tchuent *et al.* (1999) observed that 33% of cercariae from northern Senegal can remain viable in laboratory tests after 24 h. Another version of this exposure index, 'slow', therefore made this assumption.

Assuming that 0.1% of cercariae were viable 24 h after shedding did not alter the shape of the bell curve markedly from the assumption that cercariae had negligible life expectancy. However, if 33% of cercariae remain viable 24 h after shedding, the relative number of cercariae in water should be higher at all times. This scenario implies that early morning and evening water contact carries a greater risk of encountering viable cercariae compared with the alternative hypothesis (Figure 1).

It was not possible to ascertain the time of water contact using the questionnaires, as people could not recall the exact time they entered water. However, they were able to report in what part of day the activity occurred. Time of day was split into four, following the natural pattern of daily life and prayer times: morning (6–10 am), middle of the day (10 am–2 pm), afternoon (2–5 pm) and evening/night: (5 pm–6 am). Activities reported were corrected for the mean relative number of cercariae assumed to be in the water over these hours. Assuming that 0.1% of cercariae were viable after 24 h, total duration was multiplied by 0.109 for activities between 6 and 10 am, 0.833 between 10 am and 2 pm, 0.659 between 2 and 5 pm and 0.104 between 5 pm and 6 am. The equivalent figures assuming that 33% of cercariae were viable after 24 h were: 0.508 (6–10 am), 0.749 (10 am–2 pm), 0.977 (2–5 pm) and 0.727 (5 pm–6 am).

Seasonality assumptions

Water is present in all seasons, although water contact is at its peak in the hot dry season in these villages. In the wet season, vegetation grows rapidly and restricts access to water. Mosquitoes are also much more prevalent in the wet season, further reducing water contact. Such human behaviour is accounted for by the seasonal data available from questionnaires. A previous study over 3 years in a



$$N(T) = \int_0^T e^{-b[\mathrm{mod}(t,24) - T_c]^2 - k(T-t)} dt$$

Figure 1 A comparison of correction factors for time of day assuming that either 0.1% or 33% of cercariae remain viable 24 h after shedding The dotted line represents the assumed diurnal cycle of cercariae emerging from snails. N(T) = number of cercariae at time T, b (a constant = 0.369. $T_c = 7.32$, mod(t,24) = t modulo 24, k is the constant of exponential decay function: $k = 0.077$ assuming that 0.1% of cercariae are viable after 24 h, and $k = 0.046$ assuming that 33% of cercariae are still viable after 24 h.

similar village indicated that the number of *Biomphalaria* spp. snails and the number of positive snails differed markedly between months. However, the seasonal patterns were consistent over 3 years (Stelma 1997). Seasonality was incorporated into the index using the average proportion of positive snails found in each season over 3 years. The total duration of activities reported in the cold dry season was therefore multiplied by 0.24, by 0.69 in the hot dry season and by 0.08 in the hot wet season.

Statistical analysis

The relationship between exposure (expressed as eggs per gram, i.e. a count variable), age, sex, place of residence and intensity of (re)infection was analysed by means of Poisson regression, the standard generalized linear model for this type of variable (Lindsey 1997). When the data did not conform to the assumption of Poisson regression (variance equals mean) a negative binomial model was used (Lindsey 1997). This model allows for heterogeneity within the different groups by allowing the residuals to vary according to a probability distribution instead of varying randomly around a probability constant, as is the case with Poisson regression. A negative binomial model was necessary in cases where data were over-dispersed (the histogram was extremely right-skewed), as is common in distributions of parasite populations. Analyses were carried out in Stata 7.0 (Stata Corporation 2001) and hypothesis testing was done using a likelihood ratio test.

Unless otherwise stated the *P*-values quoted in the text are derived from the models described in Table 1 or Figure 2. They relate to the difference between the stated group and the base group. In one instance the base group was altered. To illustrate that there was no difference in the frequency of contact between males and females in Buntabat, the base group was altered from 8-year-old boys in the younger focus to 8-year-old boys in the older focus. This facilitated a comparison of males with females in the older focus, as opposed to females in the older focus with males in the younger focus as in the standard model. In this case incidence rate ratios, 95% CI and probability values that are exposed by this arrangement are given. Other parts of the model are not affected, except that if the comparison is old compared with young focus instead of young compared with old, the IRRs (incidence rate ratio) are inverted.

Ethical permission

Ethical permission for this study was obtained from the ethical committee of the Prince Leopold Institute of Tropical Medicine, Antwerp, Belgium and the Région Médiciale, Saint Louis, Sénégal, in accordance with the principles and practice of the Helsinki Declaration.

Results

All of the exposure indices are significantly correlated with one another, and none of them were significantly related to intensity of infection or re-infection 1 year after treatment, either as the sole factor in a negative binomial model, or in models incorporating any combination of age, sex and village of residence. Results are therefore described here for the simplest two exposure indices – ‘frequency’ (number of contacts per day) and duration (minutes per day) and one of the most complex: ‘fast’: total duration corrected for body surface exposed, time of day (assuming that 0.1% of cercariae remain viable 24 h after shedding), and weighed for seasonal differences in infectivity of water. Two assumptions with regard to domestic water contact were tested. First, that domestic water was of negligible risk, and secondly that contact with domestic water held equal risk of encountering an infective cercaria compared with non-domestic water. The conclusions were the same regardless

Table 1 Distribution of (A) intensity of infection with *S. mansoni* before treatment and (B) intensity of infection 1 year after treatment, with regard to water contact measured by either ‘frequency’ (number of contacts per day), duration (minutes per day) or the exposure index (corrected minutes per day). The base group in every case was an intensity of infection of 0 EPG (eggs per gram)

Factor	*n*	Value @ base (EPG = 0)	Incidence rate ratio	95% CI	*P*
A					
Model 1: frequency	239	4.4 contacts/day	1.00	0.99–1.00	0.44
Model 2: duration	239	65.0 min/day	1.00	0.99–1.00	0.74
Model 3: fast exposure index	222	93.5 cmin/day	1.00	0.99–1.00	0.29
B					
Model 1: frequency	147	4.2 contacts/day	1.00	0.99–1.00	0.5
Model 2: duration	147	63.8 min/day	0.99	0.99–1.00	0.47
Model 3: fast exposure index	141	92.3 cmin/day	1.00	0.99–1.00	0.12

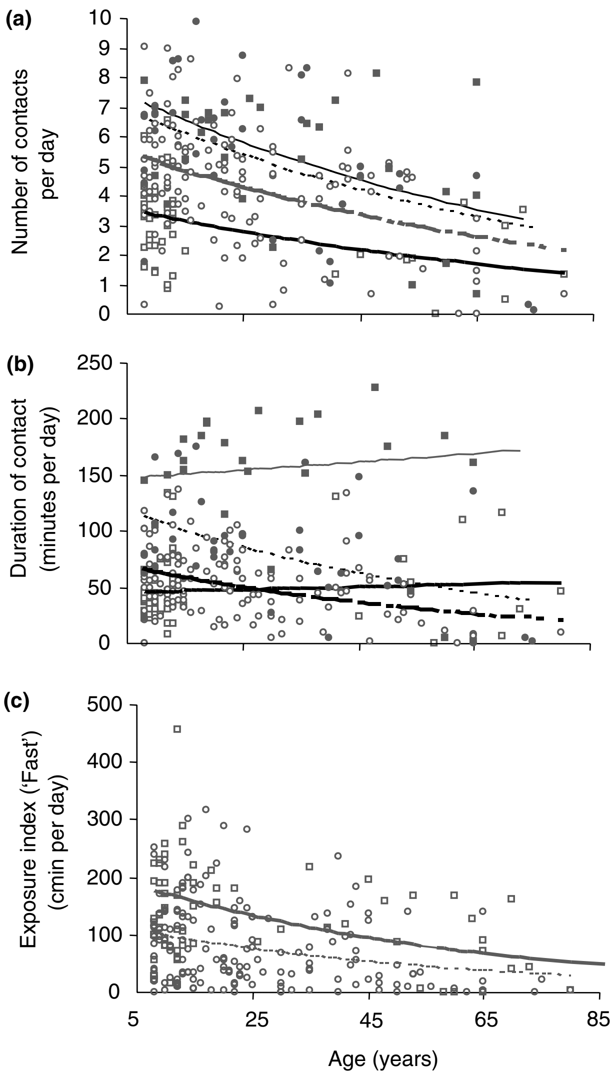


Figure 2 Water contact patterns at different ages, places of residence or sex. (a) Compares numbers of contacts per day with age (years), village and sex. Males resident in Buntabat are represented by square (data) or (model predictions). Females from Buntabat: • (data) (model predictions). Males resident in any of the other villages are represented by (data) or (model predictions). Females from other villages: (data) - - - (model predictions). The total sample size was 250. There were 48 males resident in younger epidemic foci, 137 females in younger epidemic foci, 27 males in order epidemic foci and 38 females in older epidemic foci. The model predicts that the base group for the analysis (8-year-old boys resident in the younger epidemic focus), had 3.44 contacts with water per day. The incidence rate ratio (IRR) for males in the younger epidemic focus was therefore 1. Females from the younger epidemic focus had more contacts than the base group (IRR: 1.545, 95% confidence interval (c.i): 1.29–1.85, P < 0.001), males from the older epidemic focus had the most contacts per day (IRR: 2.079, c.i: 1.64–2.63, P < 0.001) closely followed by females from the same focus (IRR: 1.953, c.i: 0.46–0.81 P < 0.001). Frequency of contact decreased in each group as people aged (IRR: 0.987, c.i: 0.98–0.99 P <0.001). (b) Compares total duration of contact with age (years), village and sex. The symbols and sample size are the same as for graph A. The model predicts that the base group (8-year-old boys resident in the younger epidemic focus), spent 46.3 min in the water per day. The incidence rate ratio (IRR) for males in the younger epidemic focus was therefore 1. Females from the younger epidemic focus spent more time in the water than the base group when young (IRR: 1.66, c.i: 1.22–2.25, P < 0.001), although this changed as they aged. Males from the older epidemic focus spent systematically much longer in the water than any other group (IRR: 3.21, c.i: 2.35–4.37, P < 0.001) and females from the older epidemic focus, when young spent much longer in the water than the base group, although again this dropped as they aged (IRR: 2.87, c.i: 0.37–0.79, P < 0.002). Duration in the water did not change for males as they aged (IRR: 1, c.i: 0.99–1.01, P < 0.5), but did for females (IRR = 0.98, c.i: 0.97–0.99 P < 0.001). (c) Compares exposure measured by the exposure index 'fast' with age and sex. Males are represented by (data) or (model predictions), females by: (data) - - - (model predictions). The model predicts that the base group for the analysis (8-year-old boys) spent 178 corrected minutes in the water per day. The IRR for males was therefore 1 (*N* = 67). Females had less exposure than the base group (*N* = 167, IRR: 0.59, c.i: 0.44–0.77, P < 0.001). Both sexes had less exposure as they aged (*N*=231, IRR: 0.98, c.i.: 0.98–0.99, P < 0.001).

of which assumption was used. Results have been reported here assuming that domestic water was of negligible risk.

Information in all three seasons for frequency and duration of contact with water was available for 250 people. Number of contacts per day were distributed normally around a mean of 4.4 (SD = 2). The normality of the distribution was tested using a one sample Kolmogorov–Smirnov test. Total duration of contact, and all the indices calculated from it were skewed to the right, and not easily transformable. Median duration in the water was 56.5 min per day [interquartile range (IQR): 32.6–85.2]. Height information was not available for 19 people, so the total sample size for the exposure index 'fast' was reduced to 231. The median value for this index was 85.8 corrected minutes (cmin) (IQR: 28.9–140.4).

Age, sex and place of residence were significantly associated with number of contacts per day as tested using a Poisson regression model. Number of contacts per day decreased in all villages and for both sexes as people got older. No significant differences were found between number of contacts per day in the more recently exposed villages (Maraye, Ndigue, Ravette or Rone); however, residents of Buntabat were in the water significantly more frequently than residents of other villages (*P* < 0.001). Within Buntabat there was no difference between the frequency of contact in males and females (IRR: 0.9, 95% CI: 0.8–1.2, $P = 0.5$). In the other villages, females were in the water more frequently than males ($P < 0.001$) (Figure 2a). The final model therefore included the categorical variables: 'sex' (male and female) and 'focus' (older epidemic: Buntabat, and younger epidemic: Maraye, Ndigue, Ravette and Rone), and an interaction factor between 'sex' and 'focus'. Age was included as a continuous variable.

Duration of contact with water altered according to age, sex and place of residence, when analysed using a negative binomial model. Age did not alter the time spent in the water by males ($P = 0.5$), however, the time spent in the water decreased as females aged ($P < 0.001$). Males resident in the older epidemic focus (Buntabat) spent longer in the water than any other group ($P < 0.001$), with females from Buntabat spending less time than males in their village, but more time than people in all other villages ($P < 0.002$). However, by middle age, females in Buntabat had similar levels of contact to people in other villages (Figure 2b). The final model describing duration of contact included all the same parameters as that describing frequency of contact, with the addition of an interaction factor between age and sex (Figure 2b).

Repeating the analysis using the 'fast' exposure index, the drop in exposure with age remained significant ($P < 0.001$). The difference between Buntabat and other villages however, was no longer apparent and did not contribute significantly to the model. A closer examination of all calculated exposure indices revealed that this change occurred only once when the correction of seasonality of transmission was applied (data not shown). In Buntabat, contact continued at very similar levels in all seasons, whereas in the other villages water contact dropped in the cold dry and hot wet seasons. The correction for seasonality of transmission weighed the index heavily in favour of the hot dry season, during which time any differences between villages were negligible. Using this index, males had significantly higher exposure than females ($P < 0.001$) (Figure 2c). The final model describing exposure measured using the 'fast' index included age as a continuous variable and sex as a categorical variable (Figure 2c).

Information regarding intensity of infection before treatment was available for 239 people and for 147 1 year later (mean and median values of water contact were not substantially different in the subset). Intensity of (re)infection was not significantly associated with either 'frequency' (pre-treatment: $P = 0.4$, 1 year post-treatment $P = 0.5$), total duration (pre-treatment: $P = 0.7$, post-treatment $P = 0.5$), or the 'fast' exposure index (pre-treatment: $P = 0.3$, post-treatment $P = 0.1$) tested using negative binomial models (Table 1). When intensity of infection or re-infection was included in models including age, sex and/or place of residence, it never contributed significantly to the model (data not shown).

Discussion

The relationship between age and intensity of infection has been observed in many different epidemiological situations; endemic (Hagan *et al.* 1998) and epidemic (Stelma *et al.* 1993; van Dam *et al.* 1996). Three causative mechanisms have been proposed (1) changing levels of exposure (Wilkins *et al.* 1987a; Butterworth *et al.* 1992), (2) resistance to infection acquired slowly as a result of experience of infection (Kloetzel & da Silva 1967; Woolhouse *et al.* 1991; Scott *et al.* 2001), and (3) resistance to infection acquired independently of experience of infection with increasing age (Gryseels 1994). We focused on the first mechanism.

In these epidemic foci in northern Senegal, there is no evidence that different levels of exposure contribute to intensity of infection before or 1 year after treatment, despite differences in water contact patterns depending on age, sex and place of residence. The general trend is for water contact to reduce gradually as people grow old, whereas the drop in intensity of infection occurs fairly suddenly, between 10 and 15 years of age depending on the focus. This concurs with previous studies in endemic areas (Butterworth *et al.* 1992), indeed, intensity of infection still dropped with age in communities with very different water contact patterns, even one where adults appeared to be more heavily exposed than children (Kabatereine *et al.* 1999). Other studies have found at least some relationship between water contact and intensity of infection, even if not to a scale sufficient to explain the relationship between age and intensity of infection (Kloos *et al.* 1983, 1998; Chandiwana & Woolhouse 1991). The lack of any discernible relationship between water contact and intensity of infection in the current study may be because water contact is at such a high level that it is not in this case a limiting factor for intensity of infection. Fulford and colleagues (Fulford *et al.* 1996) reported an average of 0.04–0.44 contacts per person per day in rural Kenya, and an average of 10 min spent in the water per day. Chandiwana and Woolhouse (1991) observed 0.43 contacts per day in Zimbabwe. In contrast, people in northern Senegal had a mean 4.4 contacts with water per day and spent a median 57 min per day in the water.

The possibility will always remain that the lack of evidence of a relationship between exposure and intensity of infection is because of either inaccurate or insufficient documentation of water contact, or an inadequate understanding of how water contact translates into exposure. Water contact information was collected using a questionnaire rather than via direct observation, because of the large number and wide dispersal of contact sites, although some direct observation was used as quality control. Many sites were out of sight from shore. For example, pathways cut in the tall aquatic reeds opened out to contact sites almost entirely surrounded by vegetation. In Brazil a questionnaire approach has also been favoured over direct observation, in areas of widely dispersed contact sites, where direct observation underestimated the frequency of contact (Gazzinelli *et al.* 2001). Questionnaires have the advantage that data management is much easier than for direct observation studies, reducing the probability of errors in the data collection phase of the study.

Different authors have approximated exposure from water contact behaviour in various ways. Frequency (Woolhouse *et al.* 2000) and total duration (Fulford *et al.* 1996) of contact are the simplest indices used. Body surface area has been approximated by some authors who used a grading system for each activity (Hagan *et al.* 1985; Wilkins *et al.* 1987a; Sama & Ratard 1994). Some exposure indices have incorporated many different factors, including soap use, place and time of day of water contact (Wilkins *et al.* 1987b). It should be noted that the use of one exposure index over another can potentially alter conclusions substantially. For example, Wilkins and colleagues (Wilkins *et al.* 1987a) reported that the mean exposure index of adult females was about one-third of 5–9-year-old children, whereas the mean duration of adult female water contact was larger than that of children. It is therefore important to verify conclusions by using different exposure indices.

We have repeated our analysis using different indices incorporating the factors most commonly used in existing literature. This avoids the problem of developing an exposure index, based on the combination of parameters that correlate most significantly with intensity of infection and then testing the hypothesis that there is a relationship between these two factors. The result of our statistical analysis was not influenced by the exposure index used, however, in other settings, this may not be the case. If the relationship between exposure and intensity of infection are to be investigated further, it may be necessary to improve understanding of what should be regarded as high risk behaviour and to have geographically and temporally appropriate data regarding densities of viable cercariae in water.

The conclusions of this study remain robust irrespective of the different assumptions made regarding the relationship between water contact behaviour and exposure to *S. mansoni*. In our opinion, we used the most appropriate means of collecting water contact information for these villages, and analysed it based on assumptions derived from the best available information regarding the relationship between water contact and exposure to *S. mansoni*. There is evidence that a person's age, sex and place of residence determines water contact behaviour, but none to suggest that exposure has an influence on the relationship between these factors and intensity of infection. It is reasonable to conclude therefore that in this population other factors such as resistance to infection acquired slowly as a result of the number of years' experience of the parasite or resistance linked intrinsically to host maturity have principal importance in determining intensity of infection.

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