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Relative Prevalence of the Human Hookworm Species, Necator americanus and Ancylostoma duodenale in Jos-North Local Government Area of Plateau State

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Abstract: A study was conducted to determine the relative prevalence of the two species of hookworms (Necator americanus and Ancylostoma duodenale) in 144 hookworm-positive stool samples from 2,800 school children attending 7 primary schools in Jos-North Local Government Area of Plateau State. Hookworm eggs, in about 4 g of the freshly passed faeces, were cultured to the third stage (13) larvae by coproculture at 24-28°C using the Harada and Mori Test Tube Method. Of the 144 hookworm positive coprocultures, 138 filariform larvae, representing 95.83% were recovered for N. americanus and 41 filariform larvae representing 28.47% were recovered for A. duodenale. One hundred and three (71.5%) of the coprocultures had only N. americanus filariform larvae, 6 (4.2%) had only A. duodenale larvae in them while 35 (24.3%) were mixed infections (that is had the filariform larvae of both species. In all mixed infections, the filariform larvae of N. americanus were recovered from a higher proportion of the coprocultures when compared to that of A. duodenale in a ration of 8:1. Although, N. americanus was the predominant hookworm species in the present study, A. duodenale also occurred.

Key words: Relative prevalence, coproculture, Harada-Mori method, *Necator*, *Ancylostoma*

INTRODUCTION

Hookworm infection in humans is caused by an infection with the helminth nematode parasites *Necator americanus* and *Ancylostoma duodenale* and is transmitted through contact with contaminated soil. The greatest number of hookworm cases occurs in Asia followed by sub-Saharan Africa (De Silva *et al.*, 2003) and is one of the most common chronic infections in areas of rural poverty. The two species of hookworm currently infect about 1.2 billion persons worldwide (Chan *et al.*, 1994). They occur in predictable areas where sanitary and environmental conditions favour the development of filariform larvae and infection of the host. They also cause significant gastro-intestinal blood loss.

Necator americanus and Ancylostoma duodenale are sympatric over much of their distribution and people are often simultaneously infected with both species in endemic areas. Despite significant differences in their life histories, these two species have traditionally been considered to be identical for treatment and control strategies (Hoagland and Schad, 1978). Failure to consider these differences is probably responsible for reports of failed drug treatments and rapid re-infection rates following anti-helminthic treatment. Report of different anti-helminthic susceptibilities between the species (Rossignol, 1990) indicates that administration of only 1 anti-helminthic may not be sufficient to kill both species. However,

the rational design of hookworm control strategies requires, among other things, knowledge of the species infecting human population in order for treatment to be effectively successful in the long term.

Previous reports from other parts of Nigeria (Adenusi and Ogunyomi, 2003) have shown that *N. amricanus* is the most common and dominant hookworm species and that infections with *A. duodenale* represents only a small proportion of the local hookworm infections over the years.

In order for treatment and control to be successful, there is the need to assess and report the relative prevalence of these two species in Nigeria and Jos in particular, where the weather favours the development of these parasites. Moreover, the epidemiological situation may not be the same as previously reported. For example, the relative distribution of the 2 species may vary from one endemic locality to the other. Therefore, similar studies need to be carried out in other parts of the country.

This study is designed to estimate the relative prevalence of hookworm species in Jos-North LGA of Plateau State. It was part of an earlier investigation to report the prevalence of hookworm infection in pupils attending 7 primary schools in Jos-North area of the state.

MATERIALS AND METHODS

This study was part of an earlier survey conducted to determine the prevalence of hookworm infection among 2,800 pupils attending 7 primary schools in Jos-North LGA of Plateau State. The survey which was carried out between March 2008 and June 2009 revealed that 144 pupils had hookworm eggs in their faeces, therefore the faeces were cultured to determine the respective hookworm species. The first part was presented at the 33rd Annual Conference of the Parasitology and Public Health Society of Nigeria, which took place in Sokoto State, Nigeria.

Identification of Hookworm Species

Hookworm species (N. americanus and A. duodenale) were determined by morphological identification of the third stage (l_3) filariform larvae of 144 hookworm-positive stool samples. Hookworm eggs, in about 4 g of the freshly passed faeces, were cultured by coproculture for 7-10 days at 24-28°C by the Harada and Mori Test Tube Method (Thienpoint *et al.*, 1986). The cultured larvae were concentrated by centrifugation and stained with Lugol's iodine for proper identification. Two hundred (l_3) were examined in subjects with a single hookworm species while $100 l_3$ were examined in all mixed hookworm infections. Larvae were identified microscopically by WHO (1981).

RESULTS

Result from this study showed that *N. amricanus* was the predominant hookworm species found in the study population as determined by morphology of l_3 larvae recovered from hookworm-positive stool samples by coproculture. The relative percentage on *N. americanus* and *A. duodenale* is shown in Table 1. Out of the 144 hookworm positive coprocultures, 138 and 41 filariform larvae were recovered for *N. americanus* and *A. duodenale*, respectively. The 103 (71.5%) of the coprocultures had only *N. americanus* filariform larvae, while 6 (4.2%) had only *A. duodenale* larvae in them. Among the 144 hookworm-infected stool cultures examined, 35 (24.3%) were mixed infections (that is had

Table 1: Percentage prevalence of Necator americanus and Ancylostoma duodenale in hookworm-positive stool samples

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Hookworm species	No. of cases observed	Prevalence (%) (n = 144)
Necator americanus	138	95.83
Ancylostoma duodenale	41	28.47
Total	144	100.00

Table 2: Relative distribution of Necator americanus and Ancylostoma duodenale in hookworm-positive stool samplesHookworm speciesNo. of cases observedPercentageNecator only10371.53Ancylostoma only64.16Mixed infection3524.30

the filariform larvae of both species) Table 2. In all mixed infections, the filariform larvae of *N. americanus* were recovered from a higher proportion of the coprocultures when compared to that of *A. duodenale* in a ration of almost 8:1 (*N. americanus* to *A. duodenale*).

DISCUSSION

It is evident from the study that *N. americanus* was the dominant hookworm species in the study area as it accounts for 95.83% of all the hookworm infections. This result confirms other works (Muller and Muller, 2002; Adenusi and Ogunyomi, 2003; Mbaso *et al.*, 2004) that *N. americanus* is the ubiquitous and dominant hookworm species.

It is also evident that apparently all infections with *A. duodenale* occurred almost always in association with *N. americanus*. This is in line with the study of Adenusi and Ogunyomi (2003), who suggested that *A. duodenale* seldom occurs solely in human hosts.

In the present study, although, *A. duodenale* was solely responsible for 4.2% of all hookworm infection, it nevertheless occurs concurrently with *N. americanus* (mixed infections) in 24.3% of the subjects. These were probably heavily exposed to hookworm infections as to have been infected with *A. duodenale* whose overall prevalence was about a quarter that of *N. americanus*.

Despite the report by Pierkersku (1989), that a female A. duodenale lays an average of 30, 000 eggs per day, in all mixed infections, much higher numbers of N. americanus infective (l₃) larvae were recovered in coprocultures when compared to A. duodenale (8:1 ratio). This is perhaps indicative of the fact that much higher numbers of adult female egg-laying N. americanus worms were present compared to A. duodenale in mixed infections, since all the eggs were subjected to the same culture conditions.

Epidemiological assessment of the public health significance of hookworm infections should not, as has been the case over the years, be focused only on the estimation of the number of hookworm infections, which occur in a given population (prevalence). Rather, it should also include the identification of the infecting hookworm species. This is vital to the evaluation of hookworm infection as a public health problem where therapy and control of the disease should be specific and targeted at the infecting hookworm species. It may be worth mentioning here, that the 2 hookworm species differ in susceptibility to the same anti-helminthic and dosage regimen. Thus, efficacy of anti-helminthic therapy is dependent on the infecting species of hookworm (Pierkersku, 1989). Well-known anti-helminthics (like Alcopar and Pyrantel) are known to be comparatively less effective against *N. americanus* than *A. duodenale* (Rajasekariah *et al.*, 1986).

Also in hookworm infections, the degree of severity varies with the infecting hookworm species (WHO, 1998). Because infection with *A. duodenale* causes greater blood loss than those infections with *N. americanus*, the degree of iron-deficiency anaemia induced by

hookworms also depends on the species. *A. duodenale* is the more pathogenic of the two species as about 0.02 mL of blood is lost per worm per day with *N. americanus* compared to about 0.1 mL with *A. duodenale* (Pierkersku, 1989). For instance, in Zanzibar, among children who were infected with *N. americanus* hookworms, the prevalence of hypoferritinaemia was 33.1%, whereas in children who were also infected with *A. duodenale* hookworms, the prevalence was 58.9% (Stolzifus *et al.*, 1997). It has also been conjectured that in China and other regions where *A. duodenale* occurs, hookworm infection during pregnancy could result in vertical transmission to neonates, possibly through ingestion of *A. duodenale* third-stage larvae in milk and colostrums (Yu *et al.*, 1995). Because of the aforementioned therefore, where *A. duodenale* is more prevalent, the effect on anaemia might be greater. This is of great public health significance.

In conclusion, although *N. americanus* was the predominant hookworm species in the present study, *A. duodenale* also occurred. The results of the present study were reliably guaranteed by the Harada-Mori Test Tube Culture Method, which is commonly used as for the culture of hookworm eggs and the WHO (1981) identification scheme.

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