PREVALENCE OF HUMAN HOOKWORM SPECIES *(Ancylostoma duodenale* and *Necator americanus)* IN NASARAWA STATE, NIGERIA

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Received: February 20,2011; Accepted: July 24, 2011

Abstract

Two human hookworm species (Ancylostoma duodenale and Necator americanus) are endemic in many rural communities in the tropics causing human hookworm diseases. Both species are usually reported in routine diagnosis as 'hookworm eggs' without recourse to their speciation. Stool samples from 297 individuals with hookworm eggs were cultured at four varying temperatures to the third stage infective filariform (L3) larvae, using the Harada-Mori filter paper strip culture technique. A. duodenale (61.0%) alone recorded higher prevalence of hookworm infection than N. americanus (25.9%) and cases of mixed infection with both species (13.1%). Room temperature (30.5°C) gave the optimum temperature for the development of both species ofhookworm at seven days post-incubation period. The study highlights the influence of temperature and numbers of days on the development and prevalence of hookworm infection in the studied communities in Nasarawa State, Nigeria. These findings have implicationsforhookworm transmission and control in the area.

Key words: Ancylostoma duodenale] Necator americanus, Filariform Larvae.

INTRODUCTION

Two major hookworm species infect humans: the Old World hookworm, *Ancylostoma duodenale* and the New World hookworm, *Necator americanus*. According to Wallace and Herbert (1985), the adults of both species are morphologically distinguishable. The males of both species are slightly large in size than the females. A. *duodenale* is general large in size than *N. americanus.* Its shape is a single curve that makes the worm appear like letter C. The tail has one pair of copulatory spicule with separate endings and a caudal spine. N. *americnus* is comparatively smaller in size than *A. doudenale.* Its shape is a double curve that makes the worm looks like letter S. It has one pair jf copulatory spicules which unite to form a terminal hooklet and caudal spine is absent. *The* eggs of both species in fresh stools are morphologically indistinguishable and contain embryos at the four or eight cell stage. The first-stage (rhabditiform) larva develops within the egg. The rhabditifornvlarvae of the two species are also morphologicallyindistinguishable. Morphologically distinguishable characteristics of the two species only appears at the third-stage (filariform or L3) larvae. At this stage, N. *americanus* is characterized by dark, prominent buccal spears and a striated cuticle seen more clearly at the posterior end; these characteristics are absent in A. *duodenale* (John, 1999).

Adult hookworms are known to attach to the jejunal mucosa, where the females deposit eggs voided with feces (Ukoli, 1990). A female *A. duodenale* produce 10,000 to 20,000 eggs per day compared to 5,000 to 10,000 for N. *americanus.* In appropriate soil type and under favourable temperature and moisture conditions, the eggs hatch in 1 to 2 days and release rhabditiform larvae which feed on bacteria and organic debris. These molt twice and develop into slender infective filariform (L3) larvae in 5 to 8 days thereafter (Ukoli, 1990).

Infection via ingestion of larvae occurs more in A. *duodenale* and rarely in N. *americanus (Flores et al., 2001).* Estimates are that about a billion hookworm infections exist at any time with point prevalence as high as 70% (Bundy and de Silva, 1998). Infections are found equally in males and females, with the lowest prevalence rate in children (Quinnell *et.dl.,* 2001; Anne, 2004). Both N. *americanus* and *A. duodenale* are endemic in warm, tropical areas particularly where people defecate in the soil (Crompton, 2000). Frequent sites of infection include places where individuals defecate collectively, such as homes and schools without adequate toilet facilities and underground coal mines (Crompton, 2000). *N. americanus is* not confined to the Americas, neither is *A. duodenale* so *restricted in geographical distribution* (Bundy and de Silva (1998). The report of Flores *etal.* (2001) showed that 30% of cultured stool were positive *for* N. *americanus, 5%* for A. *duodenale, and* less than 1% for both. ' •

Temperatures between 25°C and 35°C anda shady, sandy or loamy soil with vegetation favour the development of hookworm eggs to filariform

larvae (Udonsi and Atata, 1987; Opara, 2005). Hookworm Infections are transmitted through soil dwelling L3 infective larvae that penetrate the skin (percutaneous transmission). Fecal soil in the neighbourhood of human habitations or on farmland is the source of infection for the barefooted inhabitants. The 13 larvae migrate through the epidermis into the dermis by mechanical and lytic actions (Wallace and Herbert, 1985). Whereas only percutaneous transmission is known to occur in *N. americanus* (Anne, 2004), lactogenic infection whereby infants acquire infection from infected mothers through breast milk, has also been reported in A. *duodenale* (Brown, 2005).

This study sought to determine the prevalence of hookworm species and to investigate the optimum temperature for hookworm development, as a basis for parasitological mapping of the species and parasite transmission and control in Nasarawa State, Nigeria.

MATERIALS AND METHODS Study Area

Nasarawa State in the middle belt of Nigeria lies J between latitudes 7° 50' and 9° 50'N and longitude 6° 54' and 9° 54'E. It is characterized by a tropical sub-humid climate with two distinct seasons: the wet season from about April to October, and the dry season between November and March. Annual rainfall figures range from 1100mm to about 2000mm. Temperatures are generally high during the day, particularly between the months of March and April (the hottest months) while the mean monthly temperature ranges from 28°C 38°C (Lyam,2001).

This study was conducted during both wet and dry seasons in three Local Government Areas of the State: Doma (south), Kokona (West) and Wamba (North).

Collection and Examination of Fecal Samples

Fecal samples identified by parasitological screening to be positive for hookworm ova were cultured unpreserved by the Harada-Mori culture technique as described by Bayoh *et al.* (1992). Twelve cultures in pre-numbered test tubes were prepared from each stool sample. A set of three cultures was incubated at each of the temperatures: 25°C, 35°C, 45°C and Toom temperature (30.5°Q. Cultures were examined for filariform larvae of *A. doudenale* and *N. americanus 6* 8 days post-incubation. The day on which filariform larvae were observed, their numbers and species were recorded appropriately. The filariform larvae of *N. americanus* were distinguished from those of *A. doudenale* by dark prominent anterior papillae and posterior cuticular striations, present in the former and absent in the later species (Ukoli, 1990).

RESULTS AND DISCUSSION

A total number of 297 fecal samples were cultured for speciation of hookworm in this study (Table 1). From this number, 181 (61.0%) had A. *doudenale* alone (plate 1), 77 (25.9%) had *N. americanus* alone (plate 2) while 39 (13.1%) had both species (mixed infection). This result shows that both species of hookworm (A. *doudenale* and N. *americanus)* occur in the study communities. A. *doudenale* however, appears to be a dominant species than *N. americanus* in single infection, but both can co-exist in mixed infection. Adenusi and Ogunyomi (2003) reported higher prevalence of *N. americanus* compared to A. *doudenale* in Sagamu, western Nigeria, while Adamu *et al.* (2005) reported only A. *doudenale* in Sokoto, northern Nigeria. Although several investigators find it easier to report hookworm infection without recourse to speciation, it would appear that there are differences in the geographical distribution of the two species in Nigeria. Three major factors known to influence

Table 1: Prevalence of hookworm species *(A. doudenale and N. americanus)* in Nasarawa State

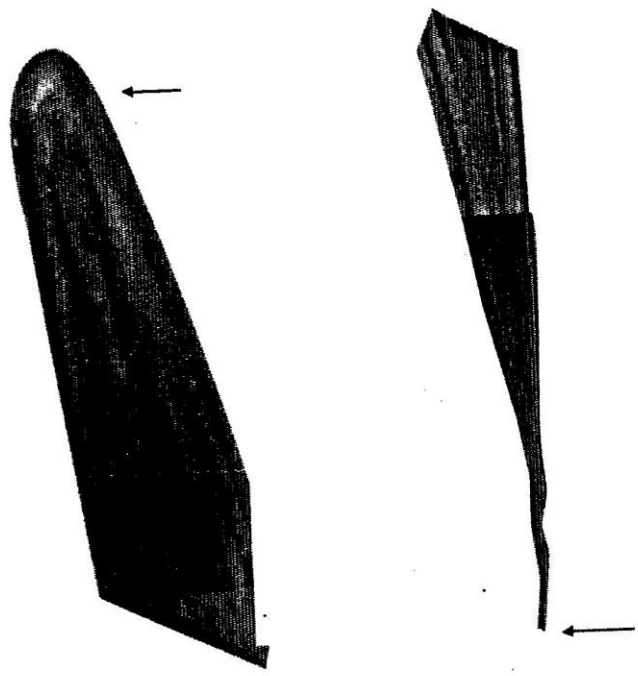
Hookworm Species	n=297
A. doudenale N. americanus	181(61.0)
	77(25.9)
Mixed Infection	39(13.1)

n=number of coprocultures

Table 2: Rate of hookworm development under different temperatures

Hookworm Species	Days of observation	No. (%) of hookworm at given Temperature (^{0}C)				Total
		25	30.5*	35	40	
A. doudenale	6	0 (0.0)	1 (0.3)	31 (10.4)	70 (23.6)	102 (20.4)
	7	1 (0.3)	175 (58.9)	23 (7.7)	15 (5.1)	214 (42.9)
	8	168 (56.6)	6 (2.0)	8 (2.7)	1 (0.3)	183 (36.7)
	Total	169 (33.9)	182 (36.5)	62 (12.4)	86 (17.2)	499 (61.0)
N. americanus	6	0 (0.0)	45 (15.2)	7 (2.4)	6 (2.0)	58 (27.4)
	7	0 (0.0)	66 (22.2)	20 (6.7)	0 (0.0)	92 (43.4)
	8	58 (19.5)	0 (0.0)	4 (1.4)	0 (0.0)	62 (29.3)
	Total	58 (27.4)	111 (52.4)	31 (14.6)	6 (2.8)	212 (25.9)
Mixed Infection	6	0 (0.0)	17 (5.7)	0 (0.0)	1 (0.3)	18 (16.7)
	7	0 (0.0)	16 (5.4)	40 (13.5)	0 (0.0)	56 (51.9)
	8	33 (11.1)	0 (0.0)	1 (0.3)	0 (0.0)	34 (31.5)
	Total	33 (30.6)	33 (30.6)	41 (38.0)	1 (0.9)	108 (13.2)

*Average room temperature of the study area at the time of making cultures;
Rate of development (%) in parenthesis



a b

PLATE 1: *Ancylostoma duodenale* (L3 larva) cultured from human feces collected inNasarawa State. Arrowed: (a) anterior portion (b) posterior portion

PLATE 2: *Necator americanus* (L3 larva) cultured from human feces collected in Nasarawa State. Arrowed: (a) anterior portion (b) posterior portion

hookworm development include temperature, soil moisture and pH. These factors have different geographical spread and could singly or collectively influence the distribution of hookworm species. The extent to which each species of hookworm response to these factors is yet to be elucidated.

Table 2 shows that both A. *doudenale* (36.5%) and N. *americanus* (52.4%) recorded high rate of development at room temperature (30.5°C) and on seven day post-incubation. Cases of mixed infection with both species, however, recorded higher rate of development at temperature of 35°C (38.0%) also on the seven day post-incubation. Six day post-incubation and 40°C recorded the least rate of development for both species but more of A. *doudenale* (17.2%) emerged than N. *americanus* (2.8%).

The optimum temperature for development of hookworm egg to filariform larvae in this study was 30.5°C. At this temperature, significant number of L3 larvae of both species of hookworm were recovered from coprocultures, especially on seven day post-incubation. This result of this study suggest that in Nasarawa State, Nigeria, hookworm development may require 30-5°C for seven days to develop to L3 larvae, when moisture and pH conditions are adequate.

Udonsi and Atata (1987) reported 30°C while Opara (2005) reported 27°C - 30°C as being the optimum temperatures for the development of hookworm ova. pH and moisture conditions were kept constant in this study by using sterile water for all cultures. Other soil factors that are known to affect

the rate of hookworm development were not considered in this study since soil samples were not part of the cultures.

CONCLUSIONS

The findings of this study show 61.0% of hookworm infections in the studied communities in Nasarawa State were due solely to *A. doudenale* while *N. americanus* alone accounted for 25.9%. Both species also occurred in mixed infection (13.1%). The study also showed that 30.5°C is the optimum temperature for hookworm development in the area. At this temperature, the eggs of both species of hookworm may require 7 days to develop to L3 larva.

REFERENCES

Adamu, T., Abubakar, U., Lawal, M. & Aliyu, B. A. (2005). Intestinal helminth parasites among school children in Wamakko LGA of Sokoto, State., Nigeria. The Zoologist, 3: 35-39.

Adenusi, A. A. & Ogunyomi E. O. A. (2003). Relative prevalence of the human hookworm species, *Necator americanus* and *Ancylostoma duodenale* in an urban community in Ogun State, Nigeria. *African Journal of biotechnology, 2:*470-473.

Anne, B. (2G04).Gastroenterology and Anaemia. *Medicine,* 35(3): 142-156.

Bayoh, M. N., Crompton, D. W. T., Stoddart, R. C. &Hodges, M. (1992). Identificaton of hookworm species prevalent in Sierra Leone. *Journal of Parasitology,* 78:1049-1054.

Brown, M. (2005). Intestinal Helminths. *Medicine,* 33(8): 54-59.

Bundy, D. A. P. & De Silva, N. R. (1998). Can we deworm this wormy world? *British Medical Bulletin,* 54(2): 421-432.

Crompton, D. W. T. (2000). The public health importance of hookworm disease. Parasitology Today, 121(16): 39-50.

Flores, A., Esteban, J. G., Angels, R. & Mas-coma, S. (2001). Soil-transmitted helminths at very high altitude in Bolivia. *Transactions of the Royal Society of Tropical Medicine and* Hygiene, 95:272-277.

John, H. C. (1999). Enteric nematodes of humans. Medical Microbiology ^ ed. Texas. 344 p.

Lyam, A. (2001). State survey of Nasarawa. A People united, a future assured 2:383-392.

Lyam, A. (2001). State survey of Nasarawa. A People united, a future assured 2:383-392.

Opara, F. N. (2005). A study of the human hookworm, *Necator americanus* in albino rat. The Zoologist, 3:82-88.

Quinnell, R. J., Griffin, J., Nowell, M. A., Raiko, A. &Pritchard, D. I. (2001). Predisposition to hookworm infection in Papau, New Guinea. *Transactions of the Royal Society of Tropical Medicine and Hygiene,* 95:139-142

Udonsi, J. K. & Atata, G. (1987). *Necator americanus:* Temperature, pH, light and larval development, longevity and desiccation tolerance. *Experimental Parasitology,* 63: 136-142.

Ukoli, F. M. A. (1990). Introduction to Parasitology in Tropical Africa. John Wiley. Ibadan, Nigeria. 3% p.

Wallace, P & Herbert, M. G. (1985). A Colour Atlas of Tropical Medicine and Parasitology. 2nd ed. ELBS Publ. Holland. 400 p.