A Case Study of Human Infection with Necator americanus (Nematoda: Ancylostomatoidea) Found in Okayama Prefecture, Japan

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ABSTRACT. A rare case of human necatoriasis found in Okayama Prefecture, Japan is reported. The patient was an 81-year-old woman residing in Soja City, Okayama Prefecture, Japan. She had repeatedly been admitted to Department of Medicine, Okayama Municipal Kibi Hospital and discharged from the hospital due to regurgitant esophagitis, cerebral infarction, superficial burn and provisional diagnosis of headache, constipation, thoracic discomfort, palpitation of the heart, and other On June 17th, 1996, adult worms of complaints since 1991. hookworm-like nematode were incidentally discovered on the mucous membrane of duodenal bulb of the patient by gastroenteroscopic examination when she readmitted to the hospital in order to take therapy for severe headache and palpitation. Two adult nematodes, one each of male and female were carefully removed from duodenal mucous membrane with a biopsy forceps. The removed nematodes were identified as adult worm of Necator americanus (Stiles, 1902) Stiles, 1906 based on morphological characteristics of cutting plates, male copulatory bursa and rays, and female caudal end. No characteristic symptom of the patient caused by adult N. americanus infection was recognized, and species specific eggs were not found in feces of the patient. The patient had a habit to eat fresh green vegetables, although a reliable route of infection

To our best knowledge, the present report deals with the first finding of human infection with N. americanus in Okayama Prefecture, Japan.

Key words: necatoriasis — Necator americanus — hookworm — Nematoda — Okayama Prefecture

Two particular species of hookworm belonging to the Superfamily Ancylostomatoidea, *Ancylostoma duodenale* (Dubini, 1843) Creplin, 1845 and *Necator americanus* (Stiles, 1902) Stiles, 1906 are known to be parasitic to human intestinal tracts. Other 2 spices, *A. ceylanicum* (Looss, 1911) Leiper, 1915 and *A. malayanum* (Alessandrini, 1905) Lane, 1926 are very rarely found from man.¹⁻⁴⁾ *N. americanus* has frequently been called "New World hookworm" or "American hookworm".^{5,6)}

The adult worm of *N. americanus* parasitic only to man was first discovered in Texas, U.S.A..⁷⁾ *N. americanus* are widely distributed throughout the world, particularly in southern part of North America, Central and Northern South America, Equatorial Africa, South and Southeast Asia, Oceania, Australia, East India, Central and South China and Japan.^{4–6)}

In Japan, *N. americanus* infection tends to be found more in south than north during past years, however, human infection with *N. americanus* is markedly diminished or almost completely absent in the recent years. In this paper, a very rare case of human infection with *N. americanus* found in Okayama Prefecture is described with photomicrographic analysis of the agential worms by light and scanning electron microscopy.

CASE NOTE

Patient: Y. I., an 81-year-old woman living in Soja City, Okayama Prefecture, Japan.

Family history: Nothing noticeable.

Main complaints: The patient has been troubled with thoracic discomfort, severe headache, constipation and palpitation or pounding of the heart since 1991. The symptoms continue to date.

Personal habits: She lives with her eldest daughter ever since she suffered from cerebral infarction which occurred 10 years ago. She is exceedingly fond of green vegetables and often eats numerous kinds of fresh vegetables.

Past history: The patient was admitted to Department of Medicine, Okayama Municipal Kibi Hospital for 3 months in the summer of 1991 to take medical therapy for cerebral infarction. On October 1995, she visited her home doctor at Fukai Clinic complaining severe thoracic pain, then hospitalized for 2 months. Very soon after discharge from the clinic, she had frequently repeated admission and discharge from the hospital with the main complaints of regurgitant esophagitis caused by esophageal hiatus hernia and superficial burn on her right forearm and rump.

Present illness: June 13th, 1996, the patient was again admitted to the hospital for further detailed examinations of severe headache, stubborn constipation, palpitation or pounding of the heart and thoracic discomfort.

Four days after admission, adult worms of hookworm-like nematode were then incidentally discovered on the mucous membrane of her duodenal bulb by a series of gastroenteroscopic examination. Two adult worms of nematode

TABLE 1. Clinical laboratory data of patient Y. I. (June 13, 1996)

WBC 4400/mm³ GOT 29 HI/I

WBC	$4.400/\text{mm}^3$	GOT	29 IU/L	
Neu	54.0%	GPT	25 IU/L	
Lym	36.4%	LDH	348 IU/L	
Mon	6.7%	γ-GTP	14 IU/L	
Eo	2.4%	LAP	37 IU/L	
Ba	0.5%	ChE	359 IU/L	
RBC	$436\times10^4/\text{mm}^3$	BUN	10.8 mg/dl	
Hb	13.4 g/dl	UA	3.7 mg/dl	
Ht	40.4%	CRN	0.49 mg/dl	
Plt	$23.8\times10^4/\text{mm}^3$	Na	142 mEq/L	
T-Bil	0.3 mg/dl	K	4.6 mEq/L	
TTT	7.4 KU	Cl	109 mEq/L	
ZTT	16.8 KU	Ca	4.3 mEq/L	
TG	124 mg/dl			

were carefully removed from duodenal mucous membrane with a biopsy forceps. The findings of blood examination revealed as follows: red blood cell count $436 \times 10^4/\text{mm}^3$, hemoglobin 13.4 g/dl, hematocrit 40.4%, platelet count $23.8 \times 10^4/\text{mm}^3$, white blood cell count $4400/\text{mm}^3$ with 54.0% segmented neutrophils, 36.4% lymphocytes, 6.7% monocytes, 2.4% eosinophils and 0.5% basophils as shown in Table 1. No significant abnormality was found in the patient's blood. The fecal examination for eggs of the helminthic parasites was negative in the stools of the patient as well as her family.

DESCRIPTION OF THE WORMS

The adult worms of hookworm-like nematode removed from the patient were relatively small in size, cylindrical in form and grayish-white in color. The male was rather shorter and smaller than the female. The male worm measured about 9.0 mm in length and 0.5 mm in maximum width with a characteristic copulatory bursa and a pair of spicules at the caudal end. The female worm measured about 10.5 mm in length and 0.6 mm in maximum width with a conical tail (Fig 1). And the vulva was recognized somewhat anterior to the middle of the body. Both the male and female worms had 2 pairs of cutting plates in the buccal capsule at the anterior end, and of these a pair of crescent-shaped large ones were the ventral cutting plates and that of smaller ones were the dorsal cutting plate, respectively (Fig 2). The outer halves of each ventral cutting plate were slightly thick, and the dorsal cutting plates were very small in size, and it is barely visible at the dorsal margin of mouth opening.

On external observation of the worm bodies, transverse striation was clearly recognized on the surface of tough cuticle of the entire body, showing single, straight and somewhat deep appearance (Fig 3). The distance between each transverse cuticular striation was about 3.0 μ m. The copulatory bursa situated on both sides of the male caudal end was a paired structure, and it was evidently longer in the direction of the lateral rays than of the dorsal and ventral rays (Fig 4). On detailed examination of arrangement of rays, both

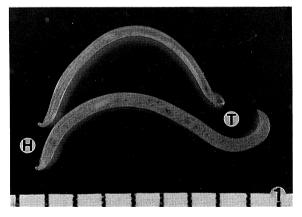


Fig 1. A male (upper) and a female (lower) of adult Necator americanus removed from duodenal mucous membrane of the patient (Scale=1 mm) H: head, T: tail

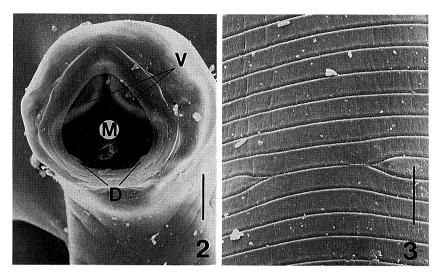


Fig 2. Scanning electron micrograph of anterior end of adult *N. americanus*, showing 2 pairs of cutting plates in the buccal capsule, anterodornal view (Scale=30 μ m)

D: dorsal cutting plate, M: mouth opening, V: ventral cutting plate

Fig 3. Scanning electron micrograph of adult *N. americanus*, showing transverse cuticular striation on the body surface (Scale= $10 \mu m$)

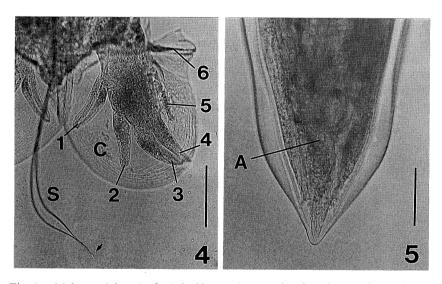


Fig 4. Male caudal end of adult *N. americanus*, showing the copulatory bursa and rays of the right side, lateral view (Scale=0.2 mm)

C: copulatory bursa, S: spicule, 1: ventral ray, 2: externolateral ray, 3: mediolateral ray, 4: posterolateral ray, 5: externodorsal ray, 6: dorsal ray, Arrow: harpoon-like hook

Fig 5. Female caudal end of adult *N. americanus*, showing no a minute spine on the tip, ventral view (Scale=0.05 mm)
A: anus

mediolateral and posterolateral rays were fairly united to each other as shown in Fig 4. Two copulatory spicules of the male worm had a characteristic harpoon-like hook on the tip of each spicule (Fig 4).

A characteristic caudal minute spine (mucron) of the female worm was not-existed (Fig 5). The fresh eggs removed from the uterine ducts of the female were ellipsoidal in shape with a thin transparent shell, measuring 54.9 to 72.6 μ m (av. 65.5) in length and 36.2 to 46.2 μ m (av. 40.9) in maximum width. Judging from these morphological features, the present nematodes were identified as adult worm of *Necator americanus* (Stiles, 1902) Stiles, 1906.

DISCUSSION

As mentioned above, among the intestinal nematodes so-called hookworm, particularly two species *N. americanus* and *A. duodenale*, are ordinarily parasitic and found in the human intestinal tracts. The eggs of *N. americanus* and *A. duodenale* are very similar to each other in shape and they are almost indistinguishable from the external appearance.⁸⁾ In addition, *A. ceylanicum*, normally a parasite to cats and dogs, has been recorded from humans in Taiwan, Southeast Asia and Surinam.^{2,5)} *A. malayanum*, a parasite to bears in Malaysia and India, has also been recorded once in a human.^{2,9)} Although the hookworms parasitic in domestic and wild animals do not become sexually mature in the human body, very typical symptoms of cutaneous larve migrans or creeping eruptions appear on skin surface where the infective stage larvae show invasion or infection.

In Japan, the first human case of *N. amemicanus* infection was reported by Watanabe (1913)¹⁰⁾ and Kitamura (1913).¹¹⁾ Thereafter, human infection with *N. americanus* had been found mostly in local farming regions before 1960, although the infection has been diminished in recent years. More recently Inaba *et al* (1998)¹²⁾ have reported one case of *N. americanus* infection found in Iwate Prefecture, furthermore, a total of 26 cases of hookworm infection in Japan reported between 1986 and 1997 has also been summarized by them. According to their description, the patients infected with *N. americanus* were 13 except 2 cases of a Filipino and a Brazilian of Japasese parentage. Although a majority of these patients are distributed widely in farming areas, mostly south of Tohoku District. Prefectural and local distribution of those 13 patients are: 3 in Wakayama; 2 in Iwate and one each in Yamagata, Niigata, Ibaragi, Aichi, Hiroshima, Shimane, Kyushu and Okinawa.

In Okayama Prefecture on the other hand, there was no reliable literature for *N. americanus* infection. Therefore, it is highly probable that the present report is the first description of human infection with *N. americanus* in Okayama Prefecture.

The adult worms of *N. americanus* live in the small intestine of the definitive host. The worms are attached to mucous membrane of the intestinal wall with dorsal and ventral cutting plates in the buccal capsule (Fig 4) whereby so-called hookworm anaemia may occur in the patients. The anaemia has a close relationship with blood loss, namely sucking blood by the infected worms and consecutive hemorrhages at the site of attachment. The amount of blood loss is 5 times or more in infection with *A. duodenale* than *N. americanus*. Hence, the patients infected with a limited number of adult *N.*

americanus are usually asymptomatic, but heavy infection lapses into the state of iron-deficiency anaemia.^{5,9)} In the present case, the adult worms found in the patient body were so few in number that no characteristic symptom cased by *N. americanus* infection was recognized.

The individuals of N. americanus develop into adult by 4 molting passages through 3 developing stages; those stages are egg, rhabditiform and filariform larvae. The filariform larvae are the infective stage worms which are locating on the surface of vegetables and in habitable soil. The human infection of N. americanus is caused by invasion of the filariform larvae through the skin including buccal mucous membrane. In the present case, it is conceivable that the filariform larvae got into the patient's body with food vegetables, perhaps because the present patient had customarily been eating fresh vegetables.

The adult body of hookworm is principally characterized by the intrinsic structures of the buccal capsule, male copulatory bursa and female caudal end. As stated above, the adult worm of N. americanus has a pair of crescent-shaped ventral cutting plates (Fig 2), while somewhat larger species A. duodenale has 2 pairs of ventral teeth in the buccal capsule.^{4,8)} The male copulatory bursa of A. duodenale is longer along direction of the dorsal and ventral rays than of the lateral rays.^{4,8,13)} At the caudal end of female worm there is a minute spine about $21 \mu m$ in length.¹⁴⁾ Moreover, the lateral rays of A. duodenale fall into 3 branches at the same angle.^{4,8,13)}

The morphological studies of N. americanus by scanning electron microscopy have been reported by two investigators in Japan. Ishii $(1971)^{15}$ showed scanning electron photomicrographs of the buccal capsule and the transverse striation very briefly. On the contrary, Yoshida $et\ al\ (1974)^{16}$ demonstrated more detailed information on the surface structure, such as the ventral and dorsal cutting plates, transverse striations, male copulatory bursa and female caudal end. The morphological characteristics of adult N. americanus shown in our present study are practically identical to the findings reported by the above investigators, and the reliability of the study by Ishii $(1971)^{15}$ and Yoshida $et\ al\ (1974)^{16}$ was well confirmed by the present study.

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