AN EXTREMELY RARE REPORT OF VARIOUS LIGHT MICROSCOPIC IMAGES AND FEATURES OF FEMALE ANCYLOSTOMA DUODENALE FOUND WHILE DOING ENDOSCOPY IN A PATIENT WITH SEVERE ANAEMIA

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ABSTRACT
Before the introduction of upper gastro intestinal endoscope, the only way to diagnose hookworm infection is by doing stool examination for hookworm ova. But after the introduction of upper gastro intestinal endoscope there has been many reports of finding live adult hookworms in duodenum and rarely in stomach while doing endoscopy. Hence upper gastro intestinal endoscopy is an extremely useful investigation to demonstrate and diagnose the presence of live adult hookworms especially in patients with severe anaemia. One such adult hookworm retrieved out while doing endoscopy in a patient with severe anaemia in our institute was examined under light microscope and was identified as female Ancylostoma duodenale. Various light microscopic images and features of this hookworm and the scientific facts by which the hookworm was identified as female Ancylostoma duodenale are described below in detail as such reports are extremely rare in the literature.

Keywords: upper gastro intestinal endoscopy, adult hookworms, light microscopic images, severe anaemia.

INTRODUCTION
By doing stool examination only hookworm ova and its larvae can be seen after culturing the ova. Hence upper gastro intestinal endoscopy is the only possible investigation to demonstrate and diagnose the presence of live adult hookworms which are commonly seen in the duodenum (1-9) and rarely in stomach (10-11). Moreover while doing upper gastro intestinal endoscopy these adult hookworms can be retrieved out using biopsy forceps and can be studied in detail by examining them under light microscope (2,3, 6-11). Such study of the adult hookworms reveal important scientific facts about these hookworms which are described below in detail.

CASE REPORT
When Upper Gastro-intestinal Endoscopy was done for a 47 year old female patient with severe anaemia in our institute, multiple adult hookworms were found in the first part of duodenum. One of the hookworms was retrieved out using biopsy forceps and immediately sent for microbiological examination. After detailed examination of the hookworm under light microscope, the hookworm was identified as female Ancylostoma duodenale. The scientific facts about how to identify a hookworm as Ancylostoma duodenale or Necator americanus and how to identify a hookworm as male or female hookworm by examination under light microscope are described below in detail. Such detailed description of the various light microscopic images and features of an adult hookworm
retrieved out while doing endoscopy are extremely rare in the literature.

**DISCUSSION**

There has been many reports of finding live adult hookworms in duodenum while doing endoscopy especially in patients with severe anaemia (6-9). Similarly multiple adult hookworms were found in the first part of duodenum while doing endoscopy in a 47 year old female patient with severe anaemia in our institute. One of the hookworms was retrieved out using biopsy forceps in order to study about the hookworm in detail. Grossly by examination with the naked eye the hookworm was found to be very thin and small measuring only about 10mm in length. The hookworm was also found to be white in colour and was exactly looking like a small cotton thread (12). Hookworm belongs to the Phylum nematoda and nematoda means thread like. But by examination with the naked eye it is impossible to identify whether the hookworm is **Ancylostoma duodenale** or **Necator americanus** and whether the hookworm is a male or female hookworm. Hence the hookworm was immediately transferred to the microbiology department in order to study the hookworm in detail under light microscope. Under light microscope, by looking at the head and the buccal cavity of the hookworm we can identify whether the hookworm is **Ancylostoma duodenale** or **Necator americanus** and by looking at the tail of the hookworm we can identify whether the hookworm is a male or female hookworm. Hence the hookworm was immediately transferred to the microbiology department in order to study the hookworm in detail under light microscope.

Under light microscope, by looking at the head and the buccal cavity of the hookworm we can identify whether the hookworm is **Ancylostoma duodenale** or **Necator americanus** and by looking at the tail of the hookworm we can identify whether the hookworm is a male or female hookworm. Under light microscope we can identify **Ancylostoma duodenale** by the presence of sharp cutting teeth in its buccal cavity (12-14,16-18) which are absent in **Necator americanus** (Fig 1,2). In **Necator americanus** the buccal cavity has semilunar cutting plates (8,9,12-18) instead of the sharp cutting teeth. But the tail of both **Ancylostoma duodenale** and **Necator americanus** has almost similar features. In both the species the tail of the male hookworm has a broad, expanded copulatory bursa which gives the characteristic broad and expanded shape to the tail of the male hookworm (9,12,14,16-19). In both the species the tail of the female hookworm does not have the broad, expanded copulatory bursa and hence the tail of the female hookworm is narrow and pointed with tapered end (12,14,16-19) (Fig 3,4).

The hookworm of our patient was identified as **Ancylostoma duodenale** by the presence of sharp cutting teeth in its buccal cavity (Fig 1,2). The hookworm was also identified as female **Ancylostoma duodenale** due to its narrow and pointed tail with tapered end because of the absence of the broad, expanded copulatory bursa (Fig 3,4). In the tail of this female hookworm, a distinct notch is seen clearly (9,17) distal to which the tail becomes extremely narrow and pointed/ with tapered end(Fig 3,4). This notch is nothing but the anus of the female hookworm (17) which is an extremely important microscopic landmark in the tail of the female hookworm.(Fig 3,4).

There is a marked difference in the opening of anus between the male and the female hookworm. In the tail of the male hookworm we can see three important structures, the anus, the male genital opening- both of which open together in the cloaca and the broad copulatory bursa (17-19). But in the tail of the female hookworm we can see only one important structure -the anus alone (17-19)(Fig 3,4). The female genital opening or vulva opens separately away from the anus higher up in the body of the female hookworm(12,16-19). The broad, expanded copulatory bursa which gives the characteristic broad and expanded shape to the tail of the male hookworm is also absent in the tail of the female hookworm. Since the tail of the female hookworm neither has the female genital opening or vulva nor has the broad copulatory bursa and has only the anus, it is narrow and pointed with tapered end(12,14,16-19). (Fig 3,4)

The sharp cutting teeth of **Ancylostoma duodenale** or the semilunar cutting plates of **Necator americanus** are used to attach the hookworms to the intestinal mucosa and suck blood by causing...
injury to the intestinal mucosa (14-17). A single Ancylostoma duodenale sucks upto 300 μl (0.3ml) of blood per day whereas a single Necator americanus sucks upto 40 μl (0.04) ml of blood per day (20-22). Thus the sharp cutting teeth of Ancylostoma duodenale cause more intestinal damage and suck more blood than the semilunar cutting plates of Necator americanus (9,15,20-22). Both the species suck blood by creating negative pressure by contractions of their esophageal muscles(15,23). The muscular oesophagus is present below the buccal cavity.(Fig 1).

The entire hookworm is covered by an extremely tough outer coat called the cuticle(17,19)(Fig 1,3). In the light microscopic images we can clearly see the striations of the cuticle lining the outer aspect of the hookworm(Fig 4). The cuticle protects the hookworm from the attack by the digestive enzymes of the host (human beings). The cuticle also protects the hookworm from the attack by the immune system of the host (human beings). Thus in addition to the sharp cutting teeth the cuticle also plays an extremely important role in contributing to the pathogenicity of the hookworm.

Only few extremely important scientific facts helpful for the identification of the species and the gender of the hookworm along with the two important causes of the pathogenicity of the hookworm namely the sharp cutting teeth and the cuticle are described in this article. But there are also various other light microscopic features of the hookworm which are not described in this article due its extensive nature. However by further detailed study of the various other light microscopic features of the hookworm, we can do extensive scientific research about adult hookworms in the future which can be of great use to the mankind.

CONCLUSION
By doing stool examination only hookworm ova and its larvae (after culturing the ova) can be seen and studied (14). Unlike Ascaris lumbricoides which are passed out in human faeces, adult hookworms are not passed out in human faeces. Hence it is impossible to see and study the adult hookworms by doing stool examination. Hence upper gastro intestinal endoscopy is the only possible investigation to demonstrate and diagnose the presence of live adult hookworms especially in patients with severe anaemia. Moreover while doing upper gastro intestinal endoscopy these adult hookworms can be retrieved out using biopsy forceps and can be studied in detail by examining them under light microscope. In addition to the identification of the species and the gender of the hookworm, by doing such detailed study of the adult hookworms under light microscope, we can do extensive scientific research about adult hookworms in the future which can be of great use to the mankind.

REFERENCE
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Fig 1: Light microscopic image of the head and the upper part of the female Ancylostoma duodenale showing the sharp cutting teeth.

Fig 2: Magnified light microscopic image of the head showing clearly the sharp cutting teeth.
Fig 3: Light microscopic image of the female Ancylostoma duodenale showing the narrow, pointed tail and the anus.

Fig 4: Magnified light microscopic image of the female Ancylostoma duodenale showing the tapering end of the tail and the anus.