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SHORT COMMUNICATION

Morphology of Medicinal Leeches in Melghat Region

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ABSTRACT
Leeches have been used in the treatment of certain diseases since ancient times. There are a few species of medicinal leeches. Secretion of the salivary glands of leeches contains more than 100 bioactive substances. These secretions include vasodilators, bacteriostatic, analgesic, anti-inflammatory and anticoagulants, anti-edematous, which eliminate microcirculatory disorders, restore the damaged vascular permeability of tissues and organs, eliminate hypoxia, reduce blood pressure, increase immune system activity, resolving the cause of pain and improve the bioenergetics status of the organism. U.S. Food and Drug Administration (Food and Drug Authority of USA-FDA) have allowed the sale of leeches in this country, the use for plastic surgery, the general purposes and microsurgery in 2004. At the same time, the difference of teeth and jaw structures of leeches may be a criterion in the classification of medicinal leeches. Medicinal leeches use their suction discs for locomotion, adhesion to the host and, in the case of the anterior disc, also for blood ingestion. We investigated the functional morphology of the anterior and posterior suckers of medicinal Leeches by using light and scanning electron microscopy. Identification of leech species is an important research area to understand the mechanism of therapeutic gains and exploring more probable benefits.
Keywords: *Hirudotherapy, Medicinal leeches, Salivary secretion, Suckers, Scanning electron microscope.*

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INTRODUCTION

Leeches (Hirudinea) constitute a relatively small monophyletic group of highly specialized annelids and play important roles as invertebrate predators in freshwater, while others are infamous for their ectoparasitic bloodsucking. They are globally distributed on all continents with one-half of all continental species, known for their local endemism [1-3]. Leeches are hermaphroditic annelids with totally reduced chetae and parapodia which are dominant in other classes of Phylum. Leeches have unpaired male and female genital pores in the region of clitellum which is glandular region in the body of leech. Leech has anterior and posterior sucker from which the anterior sucker is used for attachment and useful in blood sucking while the posterior sucker is only for the attachment to host body. The body of leech is annulated in appearance; each annulus or somite is superficially divided into usually three to five sub annuli or segments sometimes these annuli may reach to ten in number. Most of the leeches are sanguivorous on vertebrates or invertebrates' others are predatory mainly and very few are scavengers in their mode of feeding [4,5].

There are more than 700 species of leeches, with many notable examples of diversity and interesting evolutionary transitions in habitat preference, feeding behavior, and morphological adaptations. Leeches can live in a variety of environments, including aquatic and moist terrestrial regions. Some species live in freshwater, estuaries, rivers, ponds, lakes, and sea [6-9]. Leeches are carnivorous, hermaphroditic, and wormlike invertebrates with a sucker at each end of their bodies, belonging to the class Hirudinea of phylum Annelida. Leech classification is arranged so that families and genera tend to include members with similar ecological and physiological conditions. Leech biologists congregated recently to discuss the medical and scientific uses of the leech, primarily the European species *Hirudo medicinalis*, and its habit of feeding on mammalian, particularly human, blood'. Hungry leeches bite through the skin of mammals

with their three jaws and make tri-radiate incisions [10,11]. In the present study, we describe the morphology of medicinal leeches.

MATERIAL AND METHODS

Leech specimens were collected from various villages of Melghat regions (Table 1). Leeches were kept in the laboratory in starved condition at 20 ^{0}C away from direct sunlight. They were kept in appropriate glass jars with adequate de-chlorinated water. The water was changed every day. Leeches are amphibious animals and like to crawl about, hence the jar containing leeches was covered by a lid or cloth was covered tightly with string, rubber band or tape over a jar. Not more than twenty leeches were kept in a large container. The leeches were first examined alive and later were stunned with 10% ethyl alcohol and fixed in 70% ethyl alcohol or 4% formaldehyde. After fixation, the internal morphology of leeches was examined by dissecting them under a stereo microscope and Sample preparation for Scanning Electron Microscopy (SEM). SEM preparations generally involves immediate fixation of samples to avoid autolysis, putrefaction and drying effects, which may destroy the ultrastructural integrity of tissues. Small pieces of tissues were taken and fixed in glutaraldehyde. SEM analysis was performed at Visvesvaraya National Institute of Technology, South Ambazari Road, Nagpur. On the day of SEM dehydration of samples were carried out in a graded series of alcohol. After that samples were dried and handed over to technician for further process.

Table 1: The collection sites of medicinal leeches.

S. No.	Area	Location	Altitude (Feet)
1.	Chikhaldara	210 21.42'N 77022'.26 E	3500
2.	Harisal	21°35'.242"N 77°07'631"E	1574
3.	Semadoh	21034'.52"N 77016'.12"E	1764
4.	Dharni	21°32'60.0"N 76°52'60.0"E	1057.05
5.	Churni	21.41061"N 77.28076"E	1272.97

RESULTS AND DISCUSSION

The general form of this species is similar to that of *P. granulosa,* colour was comparatively more greener and pattern disintegrated more completely earlier in life. The black pattern is chiefly limited to transverse rows of midmetameric spots (Fig. 1). Dorsum is bright olive-green and venter in greyish-green, sharply separated by bright marginal orange stripes (Fig. 2). Dorsum usually with supra marginal black spots on b2 and b5 and often with paramedian spots on b6 and b1, and more or less of a median dorsal line; no definite submarginal black stripes. Head was observed a little broader. Jaw prominent, bearing on each side about sixty outstanding rounded papillae 0.05 to 0.095 mm. in diameter (Fig. 3); the median jaw somewhat larger, with 100-105 teeth, the paired with 90-92 teeth (Fig. 4). The lip marked on the venter by a median furrow (Fig. 2). Oral chamber spacious. Caudal sucker more or less small (Fig. 2). eyes, sense organs and annulation as in *H.granulosa*. clitellum as in other species of the genus, a thick, glandular layer extending from X b5 to XIII a2 inclusive, but little evident externally. Gonopore at XI b5/b6 and XII B5/B6, nephropores 17 pairs on the caudal border of b2 from VIII to XXIV inclusive. Integument furrow regularly and strongly developed. Integuments nearly smooth on some, very rough on other individuals, in which the areas stand out as papillae roughened by the protruding sense-organs, which on still other specimens may be retracted to form minute pits. Each area bears a central larger sense –organ with smaller ones cluster about it. These areas are rather more numerous than on *H. granulosa*, there being from forty-four to fifty six on each annulus of the middle region. Sensillae disposed as in *H. granulosa* but, though appearing conspicuously on the dark background, they are smaller and less elongated, generally white spots on a clear elliptical area; dorsal intermediate largest [12,13].

Figure 1: Dorsal view of *Hirudinaria* species.

Figure 2: Ventral view of *Hirudinaria* species.

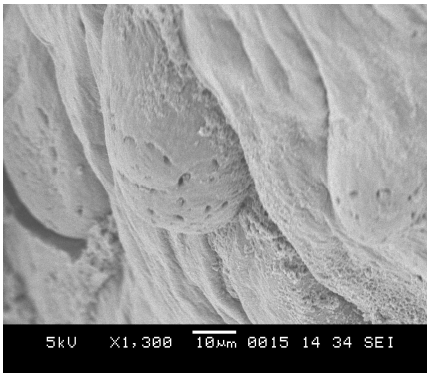

Figure 3: S.E.M. large salivary papillae with openings of the salivary ductules of the leeches belonging to the *Hirudinaria* species.

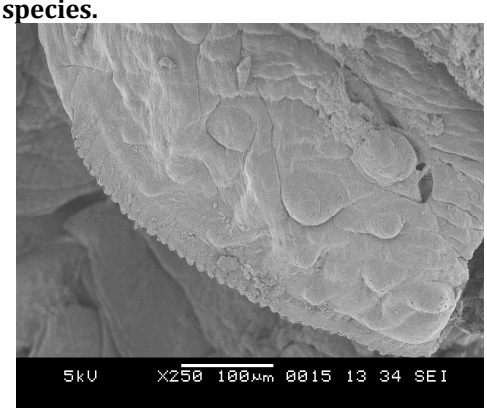

Figure 4: S.E.M. rows of denticles of the leeches belonging to the *Hirudinaria* species,

CONCLUSIONS

In this study, the triple jaws and suckers of the leeches belonging to the *Hirudo* (Annelida; Clitellata; Hirudinida) were examined using the stereomicroscopy and scanning electron microscopy. In this study suckers are seen on the first annulus and last annulus of the body. The mouth opens in the center of the front suction cup, and behind this opening is a movable triple jaw apparatus with many teeth. The posterior sucker disc consists of the last seven body segments and lacks an opening. The shape of the jaw is trignatous. The pharynx is equally located around of the three muscular jaws. The jaws are muscular covered with cuticle and carry a row of teeth arranged at the tip. In this study, it was determined that secretory canal holes were identified between the teeth. The results show that the size of teeth determines long-term bleeding so revealing the structure and working mechanism of the teeth has importance for medicinal leeches. At the same time, the difference of teeth and jaw structures of leeches may be a criterion in the classification of medicinal leeches.

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CONFLICT OF INTEREST

The authors claim no conflicts of interest because none financial support was received from any government, non-government agency or organization to conduct this research work.

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