Identification and morphological characterisation of spiny rat louse (*Polyplax spinulosa*) from a laboratory rat

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Abstract
Lice infestation is uncommon in laboratory animals but accidental entry to animal facility may occur through wild rodent admittance or breach in strict quarantine procedures. A case of lice (*Polyplax spinulosa*) infestation was detected in rat during health monitoring of quarantined animals in experimental animal house facility of Institute of Life Sciences, Bhubaneswar, Odisha, India. A male rat of 8 weeks age, procured from another lab animal facility by road, was isolated to sick animal room upon observation of alopecia and restlessness. The observed clinical symptoms were decreased grooming behaviour, scratching, restlessness, debilitation, alopecia and paleness of eyes and mucous membrane. The parasite was isolated and identified as Spiny Rat Louse (*Polyplax spinulosa*) upon detailed microscopic examination. All stages of life cycle of the louse i.e. nit, nymph and adult were identified and detail morphological characterisation was done.

Keywords: Characterisation, microscopic, morphological, Spiny Rat Louse, *Polyplax spinulosa*

Introduction
Lice infestation though common in wild rodents but is a rare condition in laboratory animals due to strict bioexclusion measures. Access of wild rodents to lab animal rooms may pose a great risk of transmission of lice either by direct contact with an infested host or through contaminated not autoclaved bedding material [1]. Another source of parasitic infestation may be exchange and transport of animal within institutes [2]. Thus strict bioexclusion measures need to be addressed to keep animals free from diseases. Parasitized animals are undesirable experimental subjects causing deviation in animal health and thus research implications. *Polyplax spinulosa* and *Gliricola porcelli* are sucking and biting lice, respectively, found on rat skin [3]. *Polyplax spinulosa* feeds on host blood and causes symptoms that includes purities and anaemia. The ectoparasites also act as a biological vector of some pathogenic virus, bacteria and protozoa [4].

In the current study, we have reported a detailed case study of Spiny Rat Louse (*Polyplax spinulosa*) infestation in a male Sprague Dawley rat which was detected during routine health monitoring of quarantined animals. This louse is host specific and completes the life cycle in a single host. The female louse lay eggs (nits) on hair follicle near the skin of the host that hatch in 5-6 days. The nymphs emerge from the eggs and undergo three molts to become sexually mature adult in 26 days [5]. Thus it is very important to strictly include this arthropod parasite in regular health monitoring programme for laboratory rodents.

Materials and Methods
Laboratory animals at Experimental Animal House facility of ILS, were group housed on autoclaved corn cobb bedding (Sparconn Life Sciences, Bangalore, India) in solid-bottom polycarbonate cages and maintained on a 12:12-h light:dark cycle at 22 ± 0.5 °C with 40% to 60% relative humidity. Rats were provided with autoclaved rodent diet (Hypro feed, Pranav Agro Industries Limited, Maharashtra, India) and deionized water treated by reverse osmosis ad libitum. All animals were maintained as per guidelines of Committee for the Purpose of Control and Supervision of Experiments on Animals (CPCSEA), Government of India. All the incoming animals to Animal Facility. ILS are quarantined and screened for many diseases before entering to main facility. The animals with signs of disease/stress are segregated to sick animal room till further diagnosis or recovery.
A male SD (Sprague Dawley) rat of 8weeks age, procured from another institute was observed to be restless with marked alopecia during quarantine and thus was segregated and kept under observation in sick animal room. Further examination revealed light tan, brown, or reddish colour “dot like lesions” on the skin and hair coat of the animal. Upon close observation, by parting the hair follicles, silvery coloured nits were found attached to hair shafts. Live animal was observed under magnifying lamp for the presence of stationary/moving lice and live/hatched egg cases. Small amount of blood (0.5 ml) was collected from saphenous vein [6] followed by isoflurane inhalation anaesthesia and haemoglobin level in blood was estimated by acid haematin method. Looking at the clinical conditions, the rat was euthanized by injecting Thiopentone @ 150 mg/kg body weight intraperitonially. Following euthanasia the detection of the ectoparasite was done by direct examination of skin pelage of rat under magnifying LED table lamp. Cellophane tape impression of skin scrapping was taken on a slide and observed microscopically under low power for identification of parasite based upon morphological features [7]. Fur pluck examination for ectoparasites (tape test) was done for diagnosis and identification of nits that stick hair follicles [7].

Results and Discussion
The animal was observed to be dull, with lustreless soiled dirty hair coat and marked alopecia over the dorsum, nuchal crest area of skull, dorsal neck and area between scapulae (Fig. 1). The skin over the area was thick scaly and slightly inflamed. The eyes and mucous membrane were pale in colour. The Hb level in blood was 6.3 g/dL against a normal value of 12.9 g/dL [8], depicting severe anaemia. The animal was inactive with absence of grooming behaviour. Generally infestation with small number of parasites goes unnoticed with no clinical signs. Animals also keep a check on numbers of ectoparasites by regular grooming of their body; however heavy infestation of blood sucking lice may cause anaemia, dermatitis, pruritus, alopecia, inflammed or scaly skin, unthriftiness, decreased growth rates [9]. Examination of slides with taped impression of skin revealed nits appearing as bunch of grapes attached to the hair shaft which measured about 0.5mm (Fig. 2a, 2b). The abdomen of lice was engorged with blood. The louse was slender and yellowish brown coloured, apterous (wingless), soft bodied, and dorso-ventrally flattened. The female was observed to be 1.5mm to 1.7mm long and the male measured 0.6mm to 0.75mm in length (Fig.2d, 2e). The head was rounded with two five-segmented antennae of 0.1mm length (Fig. 2f). There were three pair of appendages with pointed hook like claws adapted for grasping hair shafts. The appendages were 0.2mm to 0.3mm long. Developing nymphs resembled smaller versions of the adults. Seven lateral plates on each side and thirteen dorsal plates were observed on the abdominal region (Figure 2f). The above morphological appearance of lice was consistent with that of Polyplax spinulosa [9].

Spiny rat louse is an obligate parasite of rat and cosmopolitan in distribution [10] but less common in laboratory rats housed in modern animal facilities with strict bioexclusion measures. These parasites may gain entry through wild rodents or if undiagnosed during routine quarantine measures. However spread is not rapid as the only means of transmission is by direct contact [1]. Animals housed in main facility were sampled for presence of parasite by accidental exposure and were found to be free from this disease. However as a prophylactic measure we dusted the animals as well as the bedding material with Carbaryl 5% dusting powder. In addition to the rats, colonies of mice were maintained in the facility. The mice were consistently lice-negative on the basis of results of visual inspections, fur pluck exam (tape test), lack of scratching and host specificity of lice. Therefore, they were not included in the treatment programme.

Conclusion
The identification and elimination of parasite remains important due to its impact on animal health and research studies. They can also act as vector for transmission of various bacterial, viral and protozoan diseases. This parasite can be effectively controlled by preventing entry of wild
rodents and screening incoming animals. *Polyplax spinulosa* does not have any direct public health risk. However as an arthropod vector of some of the human blood-borne pathogens, caution may be taken while working with laboratory rats infested with wild population of lice.

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**References**