



VIABLE CULTIVABLE BACTERIA WITHIN TICKS AFTER HOMOGENIZATION WITH PRECELLYS 24

Tropical Infectious Diseases Research & Education Centre, University of Malaya, 50603 Kuala Lumpur, Malaysia.

/ CONTEXT

Disease transmission from rat to humans can occur through direct contact with rat excretion, contaminated environment or through ectoparasite vectors such as ticks. *Ixodes* sp. in particular, are three-host ticks commonly encountered in wild rats and have been documented to harbour *Anaplasma* sp. and *Babesia* sp. pathogens. Among those pathogens that are commonly found in the environment, bacterial pathogens of the genus *Bacillus* and *Paenibacillus* for example, are ubiquitous and several rare species have recently surfaced in the clinical setting as causative agents of human infections.

Wild rodents are often attracted to human dwellings by waste produced by human activity, this may aid the spread and transmission of tick-borne pathogens.

In light of this, an effort was initiated to study the cultivable bacteria within ticks collected from wild rodents.

/ MATERIALS AND PROTOCOL

- Rodents were captured (Sungai Congkak Recreational Forest, Selangor, Malaysia) and identified (Table 1).
- Ticks were collected from trapped rodents. Engorged adult ticks were collected from the rats and identified using morphological keys.
- Collected ticks were individually surface sterilized with 70 % ethanol and rinsed thoroughly with nuclease-free water to remove environmental contaminants before homogenization.
- Clean ticks were homogenized using Precellys® 24 as follows:
 - Precellys® Lysing kit CK28, ref. P000911-LYSK0-A.0 (2.8 mm ceramic beads),
 - 5500 rpm, 40 seconds in 500µl of sterile PBS.
- Tick homogenates were inoculated onto Columbia agar with 5 % sheep blood and incubated at 37°C for 48 h under aerobic condition.
- Bacterial cultures were purified until single colonies were obtained, followed by 16S rDNA sequencing for identification.

/ RESULTS

One unique bacterial isolate originating from an engorged adult female *Ixodes granulatus* Supino (Fig.1, B et C) collected off a *Sundamys muelleri* rat (Fig. 1, A) was identified as *Paenibacillus lautus* via 16S rDNA, *ctpA* sequencing and biochemical tests. *P. lautus* isolate was susceptible to amikacin, ciprofloxacin, imipenem and meropenem. The isolate displayed resistance toward ampicillin, penicillin, clindamycin, chloramphenicol, rifampicin and trimethoprim-sulfamethoxazole, and intermediate-resistance toward erythromycin and gentamycin.

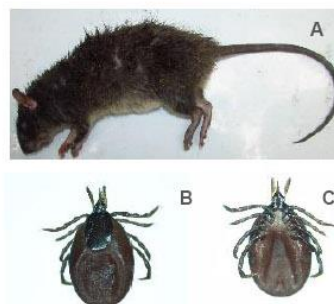


Fig. 1. *Sundamys muelleri*, Müller's giant Sunda Rat (A) and the collected engorged female adult *Ixodes granulatus*, dorsal (B) and ventral view (C).

Host		Number of ticks (n)
Common name	Scientific name	
Müller's giant Sunda rat	<i>Sundamys muelleri</i>	25
Whitehead's spiny rat	<i>Maxomys whiteheadi</i>	1
Rajah spiny rat	<i>Maxomys rajah</i>	1
Black rat	<i>Rattus rattus</i>	3

Table 1. Host of collected engorged female adult *Ixodes granulatus*.

/ CUSTOMER

Shih-Keng Loong, Siti-Nabilah Ishak, Fang-Shiang Lim, Jing-Jing Khoo, Farah-Shafawati Mohd-Taib, Sazaly AbuBakar.



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/ CONCLUSION

The Precellys® 24 coupled to the right set of lysing kits and parameters allow the extraction of living bacteria from ticks or infected tissue samples, ensuring their culture downstream to homogenization.

Paenibacillus lautus carried by ticks could potentially cause disease to humans thorough tick bites. In order to reduce the probability of interactions between wild rodents and human dwellings, an organized and monitored waste disposal could take place in public recreational areas.



エムエス機器株式会社

□東京 〒162-0805 東京都新宿区矢来町 113 番地 TEL (03)3235-0661(代) / FAX (03)3235-0669

□大阪 〒532-0005 大阪市淀川区三国本町2丁目12番4号 TEL (06)6396-0501(代) / FAX (06)6396-0508

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