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Identification of a previously unidentified endemic region for taeniasis in North Sumatra, Indonesia

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ABSTRACT

In North Sumatra, Indonesia, taeniasis has previously been reported solely from Samosir Island located in Lake Toba. In 2014, however, three individuals were identified with taeniasis after voluntarily reporting for treatment, stimulating a subsequent investigation conducted in 2017. This investigation indicated that a previously unidentified endemic area exists in Simalungun District. Molecular analysis showed the worms to be hybrid-derived descendants of *Taenia asiatica* and *Taenia saginata*, which is consistent with specimens identified previously from Samosir Island.

Human taeniasis, caused by *Taenia saginata, Taenia solium* and *Taenia asiatica*, occurs after eating beef, pork or pig viscera contaminated with metacestodes, the larval stage of these parasites, respectively (Ito et al., 2003). *T. asiatica* is considered a sister species of *T. saginata* based on molecular phylogeny (Nakao et al., 2013), with the two species being morphologically similar (Fan, 1988; Hoberg et al., 2000). The life cycles of the parasites do, however, differ with the predominate intermediate host for *T. asiatica* being the pig, in contrast to cattle for *T. saginata*. Molecular analyses have revealed that most *T. asiatica* circulating in Asia including Indonesia, Thailand, Vietnam, Lao PDR, China and Korea are not pure *T. asiatica*, but hybrid-derived descendants of *T. asiatica* and *T. saginata* (Okamoto et al., 2010; Sato et al., 2018; Yamane et al., 2013). It is now believed that pure *T. asiatica* may only be distributed in Taiwan and the Philippines (Okamoto et al., 2010; Yamane et al., 2013).

In Indonesia, taeniasis has been exclusively reported in residents of Samosir Island, which is located in Lake Toba, Samosir District, North Sumatra Province (Fig. 1) (Cross et al., 1976; Fan et al., 1988; Kosin et al., 1972, 1988). The population of North Sumatra consists largely of members of a variety of Batak tribes. Due to a local education program focused on boiling of meat prior to consumption, no *Taenia* tapeworm carriers were confirmed on the island from 2006 onwards (Wandra et al., 2006, 2017). However, in 2014, three taeniasis cases presented for treatment at Dr. Umar Zein Tropical Disease and Infection Clinic in Medan, the capital city of North Sumatra. Two of the carriers lived in Simalungun District, and the third lived in the bordering Serdang Bedagai District (Fig. 1B). These cases indicated to us the need to further investigate taeniasis in this region. The findings presented here are the results of a preliminary taeniasis survey conducted in Simalungun District during 2017.

The survey was carried out at the local health center in the village of Nagori Dolok in the Silau Kahaean Sub-District of Simalungun in

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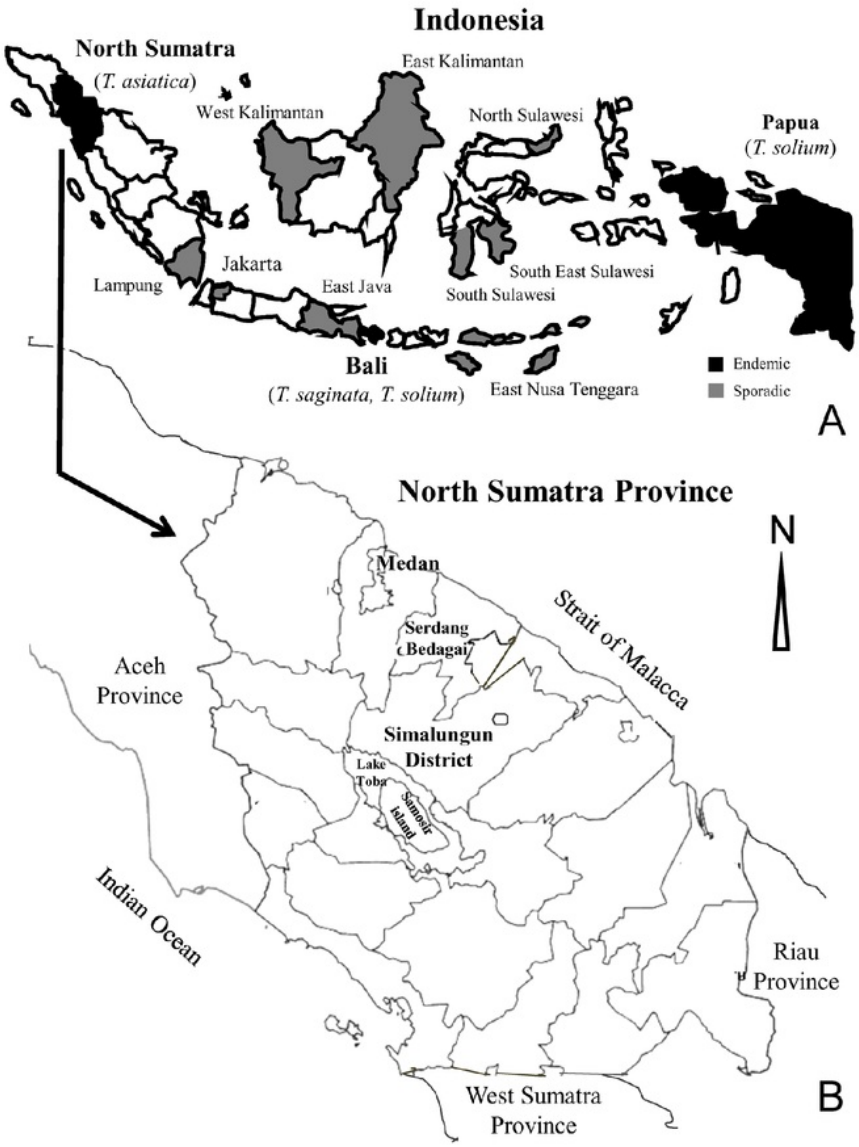

Fig. 1. Map of Indonesia (A), showing endemic areas of *T. solium* taeniasis and cysticercosis (Wandra et al., 2006, 2013) except North Sumatra where *T. asiatica*, exactly hybrid-derived descendants of *T. asiatica* and *T. saginata*, has been reported from 1970′, and Bali where *T. saginata* is rather common than *T. solium* (Wandra et al., 2006, 2013). All other sporadic areas shown in grey, and endemic Papua are exclusively on *T. solium*, and a close-up map of North Sumatra Province (B) showing both Samosir Island where hybrid-derived descendants of *T. asiatica* and *T. saginata* had been recorded, and Simalungun District where they were newly confirmed during 2017.

October and November of 2017. Communications were made to members of 7 villages (population: 10,068) in a province of 13 villages (population: 16,067) in Silau Kahaean, through health care workers one month prior to commencing fieldwork. Interested and willing volunteers were requested to come forward for testing and subsequent treatmenf for taeniasis.

In total, 250 individuals voluntarily participated in the survey, which consisted of a questionnaire used to collect demographic information and history of passing proglottids in the previous six months followed by microscopic stool examination and subsequent treatment for potential taeniasis cases. Treated individuals included those from whom taeniid eggs and proglottids were confirmed (n = 140), those who only expelled proglottids (n = 21), those who only expelled eggs (n = 8), and those without eggs or proglottids, but who requested treatment (n = 11). A total of 180 people were treated with a 600 mg tablet of praziquantel (10 mg/kg for an adult weighing 60 kg, no participants weighted over 60 kg) (Biltricide, Bayer, Leverkusen, Germany), followed 1–2 hours later with a 25 mg tablet of Dulcolax laxative (Boehringer Ingelheim, Ingelheim am Rhein, Germany). Of the 180 people treated, including two who had no evidence of infection upon microscopy, 171 were confirmed to have harbored tapeworms.

Proglottids from all discharged tapeworms were kept in 10% formalin-saline for morphological examination except for one which was fixed in ethanol. There were approximately 16 uterine branches in each gravid proglottid, indicating that the infecting species were all *T. asiatica* (11–32) or *T. saginata* (14–32), but not *T. solium* (5–11) (Fan, 1988). Molecular identification was performed on four specimens (one fixed in ethanol and three fixed in 10% formalin-saline). Genomic DNA was extracted from immature proglottids using the DNeasy Blood and Tissue Kit (QIAGEN, Hilden, Germany) in accordance with the manufacturer's instructions, and then used as a template for polymerase chain reaction (PCR). Both mitochondrial (mt) and nulcear genes were analyzed to evaluate whether the specimens were *T. asiatica*, *T. saginata*, or hybrid-derived descendants of the two species. Partial sequences of the mt *cox*1 gene were amplified using the universal primer set for cestodes (pr-a and pr-b) (Okamoto et al., 1995). The nuclear

polymerase delta (*pold*) gene was also sequenced as previously described (Yamane et al., 2013).

Partial sequences (361 bp) of the *cox*1 gene were obtained from all four tapeworms. The sequences were identical to each other and the BLAST search revealed that the sequence was the same as numerous previously submitted *T. asiatica* isolates, including an isolate from Samosir Island (AB465228). Therefore, based on mt gene analysis, these specimens are considered *T. asiatica*. Nuclear gene sequences (1097 bp) were obtained by direct sequencing of PCR product from two worms. Results showed that the *pold*B allele was the same as that found in previously described hybrid-derived descendants and differed from the alleles reported from pure *T. asiatica* (Yamane et al., 2013). This result indicated that these two tapeworms were hybrid-derived descendants of *T. asiatica* and *T. saginata*, which is consistent with specimens identified previously from Samosir Island (Okamoto et al., 2010; Yamane et al., 2013; Wandra et al., 2006).

Based on the questionnaire, all confirmed tapeworm carriers (n = 169) were Christians (100%) and most carriers were men (149/169, 88.2%) and palm farmers (141/169, 83.4%). Most confirmed carriers had a history of passing proglottids in the last 6 months (161/169, 95.3%). The youngest carrier was a 12-year-old boy, and the oldest was a 70-year-old man. All carriers (100%) reported eating raw or inadequately cooked pork as part of traditional dishes. In Batak culture, it is traditional for pieces of undercooked pig liver to be served to family leaders (men) as an honor at every wedding or cultural party. Also, almost all customers visiting restaurants where undercooked pig liver is served are men. None of the tapeworm carriers reported eating undercooked beef. Therefore, it is suspected that the infectious source, in the study area, is pork.

Although only a preliminary study, the data presented here indicate that there is a taeniasis endemic region in North Sumatra which had previously not been identified. The areas selected for previous studies and interventions, mainly Samosir Island and some lake side of Lake Toba, were easily accessible and of a higher socio-economic status, and it appears that the poorer communities in the large palm farming areas may have been overlooked when designing programs. Extensive, well designed, studies are now needed to clarify the causative species of taeniasis and to be better understand the epidemiology of the parasite in this locality so that appropriate control programs may be launched (Wandra et al., 2006, 2013).

As the prevalence of taeniasis in the 7 villages in Silau Kahaean Sub-District is assumed to be high based upon the data presented here, mass drug administration may be recommended for further studies. However, it is very difficult in Indonesia to get PZQ or niclosamide, and this barrier must be overcome to undertake a successful intervention.

In conclusion, we have documented the exisistance of a previously unidentified endemic foci of taeniasis in the Simalungun District of North Sumatra and call on the relevant stakeholders to undertake control strategies as a matter of urgency.

Author's contributions

UZ conducted the field work with SS, IJ, AHP.
UZ, SS, TWS worked for morphological observation.
MO, TY worked for molecular analysis.
AI coordinated this study when he got data from UZ with UZ, TW and KS.
AI prepared the MS with UZ, TW, MO, TY, KS, TWS, HL.

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Conflict of interest

The authors declare that there is no conflict of interest.

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