**Diagnosis of human taeniasis**

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*Taenia solium, T. saginata and T. asiatica* are taeniid tape-worms that cause taeniasis in humans and cysticercosis in intermediate host animals. *T. solium* can also cause cysticercosis in humans. A number of diagnostic methods have been developed to diagnose *Taenia* species that infect humans. This article is aimed at providing an overview of currently available diagnostic methods for human taeniasis.

Human taeniasis is an important zoonotic health problem and is caused by adult stages of taeniid cestodes, including *Taenia solium*, *T. saginata*, and *T. asiatica*. These parasites have indirect life cycles where humans act as definitive hosts whereas pigs (for *T. solium* and *T. asiatica*) and cattle (*T. saginata*) serve as intermediate hosts. Humans become infected by ingesting parasite cysts present in raw or undercooked infected meat/liver; when the cyst reaches the human intestine, it develops into an adult tapeworm, releasing segments and/or eggs in the stools or motile segments (e.g. *T. saginata*) are expelled actively. Intermediate hosts such as cattle and pigs become infected when they ingest taeniid eggs via contaminated feed or water and the larval stage (cysticercus) forms in muscular and sometimes other tissues. The adult tapeworm stage of *Taenia* spp. is relatively innocuous and does not cause pathogenic effects in humans; however, the intermediate stage of *T. solium* can also develop in human brains causing neurocysticercosis, a major cause of neurological disease in many developing countries as well as other organs causing intramuscular, ocular, subcutaneous and spinal cysticercoses. *T. saginata* is endemic in Australia, whereas *T. solium* and *T. asiatica* are exotic. However, people coming and/or returning to Australia from endemic countries can be infected with *T. solium*, leading to the possibility that infected individuals may pass segments of the parasite in their stools, which can serve as a source of infection for human cysticercosis.

Therefore, differentiation of *Taenia* species becomes significant for surveillance and control of human taeniasis.

A number of diagnostic methods have been used to differentiate the common human cestodes, *T. saginata* and *T. solium*; however, each method has its advantages and disadvantages, and careful attention should be paid to determining which particular test is best to use for differentiation of the two species. The following sections provide a quick rundown on various diagnostic methods available to differentiate *Taenia* spp. that infect humans.

**Microscopic diagnosis**

Traditionally, diagnosis of taeniasis has been based on the detection of eggs by microscopic examination but this method lacks sensitivity and specificity as *T. solium* and *T. saginata* eggs are morphologically identical, making species identification impossible. However, morphological examination of gravid proglottids can allow the differentiation of *T. solium* and *T. saginata* provided the internal structures (i.e. uterine branches) are intact. Sometimes, even the morphological examination of proglottids does not allow the differentiation of *T. solium* and *T. saginata*, thus requiring alternate methods for the differentiation of human *Taenia* spp.

**Immunodiagnosis**

The first method of coproantigen (parasite antigens in human stool) detection using enzyme-linked immunosorbent assay (ELISA) was developed by Allan et al. Although the test displayed a higher sensitivity and specificity than microscopic diagnosis for the detection of *Taenia* spp., it did not allow differentiation of *T. saginata* and *T. solium*. Recently, Guezala et al. developed another coproantigen ELISA and successfully differentiated *T. solium* from *T. saginata*. These tests were developed in individual labs using...
in-house reagents and have not been independently validated in different laboratories nor have they been widely used in diagnostic laboratories.

The first serological assay to detect specific antibodies against *T. solium* infection in humans was developed by Wilkins *et al.*9 Subsequently, a number of studies reported various immunocassays for the diagnosis of human taeniasis, primarily caused by *T. solium*10,11 and these studies used either native excretory-secretory products collected from adult tapeworms9 or cloned and expressed excretory-secretory products of adult *T. solium*10,11. Like coproantigen ELISAs, the detection of *Taenia* species-specific antibodies are also more specific and sensitive than microscopic techniques. However, these tests have been found to have some degree of cross-reactivity in sera from patients with cystic echinococcosis, ascariasis, and schistosomiasis11. Furthermore, currently available immunodiagnostic tests may give false positive results as specific circulating antibodies in taeniasis patients could possibly remain detectable for some time either after treatment and recent past infections4.

**Molecular diagnosis**

A number of molecular methods using PCR-based technologies have been developed to either determine the presence of *Taenia* species-specific DNA in human stools or differentiate *Taenia* spp. (*T. solium, T. saginata* or *T. asiatica*) based on the analysis of DNA extracted directly from tapeworm12. PCR-based methods have higher sensitivity in the detection of taeniasis cases (i.e. the detection of parasite DNA in human stools) than microscopy alone. In addition, a combined use of PCR and microscopy have been found to improve diagnostic sensitivity where Yamasaki *et al.*13 showed that some proven egg-positive cases were negative by PCR. Specificity of PCR is high with control faecal samples, including samples from patients with other parasitic infections, being almost always negative in PCR.

To date, predominantly conventional and multiplex PCR, and PCR-restriction fragment length polymorphism (PCR-RFLP) have utilized various markers, including internal transcribed spacer, mitochondrial cytochrome c oxidase subunit I gene as well as 12S rDNA, cathepsin L-like cysteine peptidase, *T. saginata*-specific repetitive sequence (HDP1) and cestode-specific sequence (HDP2) to discriminate between human *Taenia* spp13–21 (Table 1). Mayta *et al.*19 developed a nested PCR utilizing two rounds of PCR amplification of the Tso31 gene, which is more sensitive than conventional PCR but pose technical difficulties. A field DNA-based test known as loop mediated isothermal amplification (LAMP) was developed by Nkouawa *et al.*20, which amplifies the cox1 gene. To date, only one real-time PCR to discriminate *T. solium* and *T. saginata* has been developed which targets the internal transcribed spacer 1 of the nuclear ribosomal RNA11. The majority of DNA-based methods have utilised DNA isolated from proglottids for *Taenia* species identification while relatively fewer studies isolated DNA from stool samples (Table 1). In addition, almost all studies have been tested only on small numbers of usually known positive and negative samples.

**Conclusions and future perspectives**

Human taeniasis is a worldwide parasitic disease and detection and discrimination of *T. solium, T. saginata* and *T. asiatica* remains a

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RFLP, restriction fragment length polymorphism; ITS, internal transcribed spacer; cox1, mitochondrial cytochrome oxidase subunit I gene; Clp, cathepsin L-like cysteine peptidase; HDP1, *T. saginata*-specific repetitive sequence; HDP2, cestode-specific sequence. Adapted from Verweij and Stensvold12.
public health concern. Microscopic, immunological and molecular methods have been used to detect and differentiate *Taenia* spp., and a combination of two or more methods appears to provide higher sensitivity. Almost all of the immunological and molecular methods developed so far have not been independently tested and validated on controlled negative and positive samples as well as field samples. Among existing molecular methods, the nested PCR provides the highest sensitivity and specificity of those methods that have been developed and validated using unselected faecal samples from parasitological proven taeniasis carriers. Although the method is technically challenging and could be expected to be relatively expensive to use, the reagents required are available worldwide and laboratories capable of undertaking PCR competently can run this PCR.

To the best of our knowledge, no standardised PCRs are available in Australia to discriminate human *Taenia* spp. Future studies should focus on independent validation of the nested PCR and the LAMP technique as these two DNA-based tests offer higher sensitivity and a user-friendly option without sophisticated equipment, respectively. In addition, future studies should be aimed at extracting DNA from sodium acetate-acetic acid-formalin (SAF) fixed proglottids using different methods as the current DNA extraction protocols do not provide reliable and consistent DNA yield for PCRs (Jabbar, unpublished data). To date only one study has reported the extraction of DNA from long-term stored *Taenia* specimens in formalin and this has not been independently verified.

### References


### Biographies

**Dr Abdul Jabbar** is a Senior Lecturer in Veterinary Parasitology at The University of Melbourne. His main research interests cover epidemiology and diagnosis of parasites of socioeconomic importance using next-generation molecular tools.

**Dr Charles Gauci** is a Senior Research Fellow at The University of Melbourne. He has worked with Prof Marshall Lightowlers throughout his career and his research interests focus on recombinant vaccines for prevention of transmission of the parasite causing neurocysticercosis and the related parasite that causes hydatid disease.

**Professor Marshall W Lightowlers** has been a full-time research scientist supported by medical research funding for more-or-less all of his working life. He currently holds appointments as Laureate Professor at the University of Melbourne’s Faculty of Veterinary and Agricultural Sciences, and Principal Research Fellow with the NHMRC.