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***Giardia* in New Zealand Animals**
**Prevalence, Viability in the Environment and
Preservation**

A thesis presented in partial fulfilment of the
requirements,

for the degree,

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ABSTRACT

Little is known about the epidemiology of giardiasis in New Zealand. Most interest has been focused on the occurrence of *Giardia* cysts in the water ways of this country and in particular in municipal water supplies. Little is known about the occurrence of asymptomatic or even symptomatic giardiasis in people as this is not a notifiable disease, even less is known about giardiasis in domestic or wild animals.

Giardia can now be cultured in the laboratory. *G. intestinalis* may be cultured *in vitro* while *G. muris* is often cultured in the mouse. *G. intestinalis* cysts when harvested are often only a portion of the total cells present with trophozoites and incompletely formed cysts being a larger portion of the cell population. For ease of counting cysts it was desirable to destroy the trophozoites and incompletely formed cysts. On average 64% of the trophozoites can be destroyed when harvested cultures are incubated in double distilled water overnight. Trophozoites were found to remain present in the water for weeks when stored at 4°C. Sonication destroyed trophozoites within two minutes while incompletely formed cysts persisted in suspension with completely formed cysts. The only way to quickly and easily destroy trophozoites and incompletely formed cysts was to incubate the cell suspension in 0.1% SDS for approximately two minutes and then wash the cysts remaining by slow centrifugation. The result is a clean suspension of non-viable completely formed cysts.

New Zealand animals shown to harbour the parasite *Giardia* include farm animals; cattle, sheep, dogs and chickens, of importance to anyone using animal manure on their gardens especially on vegetables that do not require cooking before consumption. Domestic animal wastes can enter the water supplies on farms and get into rivers that supply town water supplies. Wild animals infected with *Giardia* studied in more detail were the possum, house mouse and ship rat. These animals may be a reservoir for contaminating water ways in less populated areas of New Zealand though they were more likely to just maintain the infection within their own population due to little contact with running water. Other wild animals defecating near water ways could serve as sources of infection.

Little is known about the zoonotic potential of *Giardia* found in animals. In past trials with *Giardia* cysts from beavers were shown to infect 2 out of 3 people. Dogs and cats have been implicated as possible sources of household infections and it is recommended when treating a family for giardiasis to also treat household pets.

Giardia intestinalis cysts cultured *in vitro* are commonly used for experimental work due to the ease of harvesting large numbers. It was not known if the *in vitro* cultures truly reflected the characteristics of *Giardia* isolates from people and animals. Morphologically, few cysts harvested from the flask are elliptical in shape, most being round. It is thought

in vitro culturing is highly selective and resulting cultures would only represent a small portion of the wild population.

G. intestinalis cysts cultured *in vitro* were compared to *G. intestinalis* isolated from human faeces and *G. muris* cultured in the mouse. Cysts could not survive in the absence of water. Laboratory trials found that *Giardia* cysts were able to survive and remain viable for months in cold water (4°C) and for shorter periods of time at higher storage temperatures. Cysts suspended in water free of faecal matter were viable and detectable for a longer period of time than those cysts exposed to faecal matter. Cysts incubated at 4°C had the best survival rate with respect to viability and the length of time cysts were present.

Preservation of cysts in 10% formalin did not necessarily prolong the length of time *Giardia* cysts could be stored for. Cysts stored in water at 4°C survived for as many months as cysts fixed in 10% formalin. *Giardia* cysts studied in the laboratory are often fixed in 10% formalin or Schaudin's fixative (PVA) to enable long term storage of specimens in suspension or on slides. Using Schaudin's fixative, *Giardia* cysts were destroyed and the internal morphology of the cysts was greatly distorted. Fixing with osmium tetroxide was not found to distort internal morphology as no difference in morphology was viewed under Nomarski optics between viable organisms and those fixed with osmium tetroxide.

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