Progress report on impact of cat trapping on prevalence of *Toxoplasma gondii* in one year old ewes as part of the Cape to City Initiative

Executive Summary

- Cape to City (C2C) is a collaborative landscape scale project covering 26,000 ha, between partners Hawkes Bay Regional Council, the Department of Conservation, Cape Sanctuary, Manaaki Whenua Landcare Research and various landowners and businesses. Project aims are to (1) leverage the native species and research success of privately owned Cape Sanctuary to deliver significant conservation outcomes within the project area; (2) trial large scale, low cost (<$3/ha) predator control (stoats, ferrets and feral cats) techniques for biodiversity enhancement within an agricultural landscape in Hawke's Bay; (3) drive a long term positive step change in regional biodiversity profile, funding, community engagement and conservation outcomes.
- This study's ultimate objective is to determine the impact of feral cat control on the prevalence of *Toxoplasma gondii* in ewes and associated abortion rates, through reducing opportunity for sheep infection by reducing the abundance of definitive cat hosts in the landscape.
- Here we present a progress report two years in to this five year project, after feral cat trapping was initiated at case farms from April 2017.
- 60 ewes are tested for exposure to *T. gondii* using a serological assay in each of 6 farms – 3 case farms within the C2C footprint that included trapping, and 3 control farms outside the footprint that had no trapping.
- During August – November 2017, observed *T. gondii* seroprevalence in ewes ranged from 48.3% to 93.3% across the 6 study farms.
- *T. gondii* seroprevalence was greater in both case and control farms in 2017 (an average of 69.2 %) than when previously assessed in 2015 (39.4 %). Mean relative increase in case farms (Δ = 27.2%) was slightly less than in control farms (Δ = 32.2%).
- In addition, monthly rainfall over 2015 and 2017 has been reviewed so that possible environmental drivers of *T. gondii* infection can be considered.

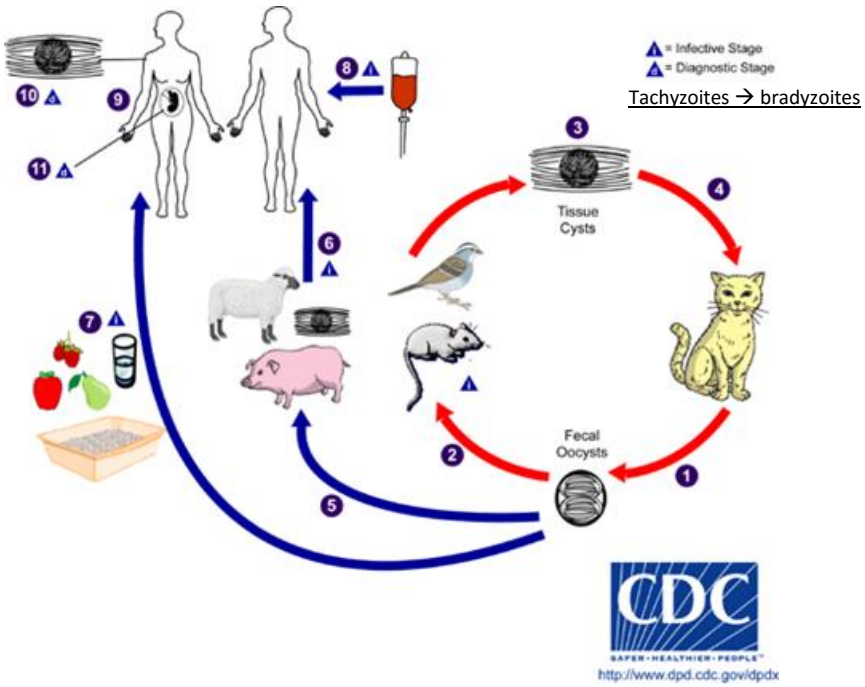
Background

Toxoplasma gondii is a protozoan parasite with a complex life cycle. Felids (domestic cats and their relatives) are the definitive hosts, which are the only place where the parasites can undergo sexual replication to generate oocysts (Figure 1). A large amount of unsporulated oocysts are shed in the cat's faeces; however, oocyst shedding occurs for only 1-2 weeks. Within 1-5 days, oocysts in the environment sporulate and become infective. Faecal matter that enters the soil can result in oocysts

being distributed by precipitation or can be taken up by earthworms which can carry the infective *T. gondii* [1]. All warm-blooded vertebrates can serve as intermediate hosts, which acquire infection through ingesting soil, water or plant contaminated with oocysts – this includes pigs, sheep and humans. Parasites in an intermediate host undergo asexual replications. First, oocysts transform into tachyzoites shortly after ingestion. Then, tachyzoites develop into tissue cyst bradyzoites, which often localize in neural and muscle tissue. While cats may be infected through ingestion of sporulated oocysts, the infection to cats is much more effective through consuming intermediate hosts containing tissue cysts. *T. gondii* can infect a wide range of intermediary hosts, infecting tissue in cysts, but felines are the primary host that pass the infective oocytes on in their faeces. It can cause a high rate of abortion in sheep on first infection.

Cape to City is a collaborative initiative with the objective of putting in place sustainable predator control in a 26000 ha region of Hawke's Bay ranging from Cape Kidnappers in the east to Havelock North in the West, and from Te Awanga in the north to as far south as Waimarama. Started in 2015 for an intitial five year period, the institutes involved include Hawkes Bay Regional Council, the Department of Conservation, Cape Sanctuary, Manaaki Whenua Landcare Research and various landowners and businesses. The ultimate objective is to eradicate invasive predators from the footprint, particularly stoats, ferrets, possums, rats and feral cats. It is believed that removing these pests will help native species recover from extinction risks and re-colonise their native habitats, which will help re-build ecosystems to how they once were before human arrival. The subsequent benefit to biodiversity will be high, as will the benefit to agriculture as these pests bring substantial costs to crop and livestock farmers. For example bovine tuberculosis can be spread to cattle from possums, as well as toxoplasmosis to sheep from cats, both of which incur substantial economic costs. A recent study estimated a 12-13% decrease in marked lambs as a result of toxoplasmosis infection in South Australia (with an average exposure rate of around 25%), costing the state $70 million Australian dollars per year (http://www.abc.net.au/news/rural/2017-02-07/toxoplasmosis-costs-south-australian-sheep-producers/8245676). In 2015, before the trapping programme was underway, we screened one-year-old ewes for *T. gondii* using serology on three case farms within the footprint and three farms outside the footprint, as well as develop a molecular assay for its detection in feral cats caught on the footprint farms, the latter as a means of describing possible routes of transmission. Seroprevalence in one-year-old ewes at that time ranged from 22% to 80%, and prevalence in feral cats ranged from 26% to 83%. In this study we review levels of toxoplasmosis in one-year-old ewes on the same farms two years later as a means of monitoring overall prevalence and to assess any detectable impact of the cat trapping that has been done to date.

Figure 1 The life cycle of *T. gondii* (figure from the Centers for Disease Control and Prevention)



Objectives

The objective of the toxoplasmosis programme within the Cape to City initiative is to test whether feral cat control can reduce the prevalence of *T. gondii* and the opportunity for infection of ewes, thereby reducing abortion rates. We previously reported seroprevalence on farms before any cat trapping, and the development of a molecular PCR assay for the detection of *T. gondii* from host brain tissue, to enable prevalence in various life history stages (namely feral cats as definitive hosts) to be estimated via PCR (Landcare Research Cape to City Consortium).

Here, we have monitored *T. gondii* seroprevalence in one-year-old ewes two years into the five year trapping programme, after feral cat trapping has commenced. The aim is to monitor ongoing toxoplasmosis prevalence on farms and look for early indications of whether feral cat trapping is reducing infection rates, in order to make recommendations around the extent and intensity of trapping efforts required for detection of any effect.

In 2019, we will repeat testing for *T. gondii* in cats and sheep to determine if the cat trapping programme has indeed resulted in reduced force of infection on farms, and confirm likely routes of transmission. We will also compare abortion rates in ewes between farms with an effective trapping programme and farms where feral cats are not controlled.

Methods

Specific cat trapping was carried out on all three case farms (Farm 1, Farm 2 and Farm 3) in 2015 between August and November. Trapping of feral cats has been ongoing (via kill traps) on two of the three footprint farms – Farm 2 and Farm 3 – since April and June 2017, respectively. Due to proximity to an urban cat population on the outskirts of Havelock North, limited trapping twice per year using live-capture traps has been done on Farm 1.

Blood from 60 one-year-old ewes was sampled between August and November 2017 from each of three case farms within the C2C footprint, and three control farms outside the footprint. Samples were collected by local vets and sent to Gribbles, Palmerston North, for serological testing and statistical analyses were performed on seroprevalence data.

Monthly rainfall data at five sites in the Southern Hawkes Bay region for 2015 and 2017 was acquired from the Hawkes Bay Regional Council. The sites are Mangaorapa, Waipoapoa, Ben Nevis, Wallingford and Maraetotara.

Results

Summary of *T. gondii* detection

Across all 6 farms *T. gondii* seroprevalence ranged from 48.3% to 93.3% in ewes. On all farms, seroprevalence was higher in 2017 than 2015 (Figure 1), and this difference was statistically significant (paired t-test, one-tailed, $p = 0.013$; Wilcoxon Signed Rank, $p < 0.05$). A summary is given in Table 1, including a comparison of the relative change in case farms vs control farms. The average relative increase was slightly less in case farms (27.2%) than in control farms (32.2%), though this difference was not significant.

Figure 1 Graph showing number of 60 one-year-old ewes that tested positive for *T. gondii*. Farm 1, Farm 2 and Farm 3 are case farms with feral cat trapping. Farm 4, Farm 5 and Farm 6 are control farms with no trapping.

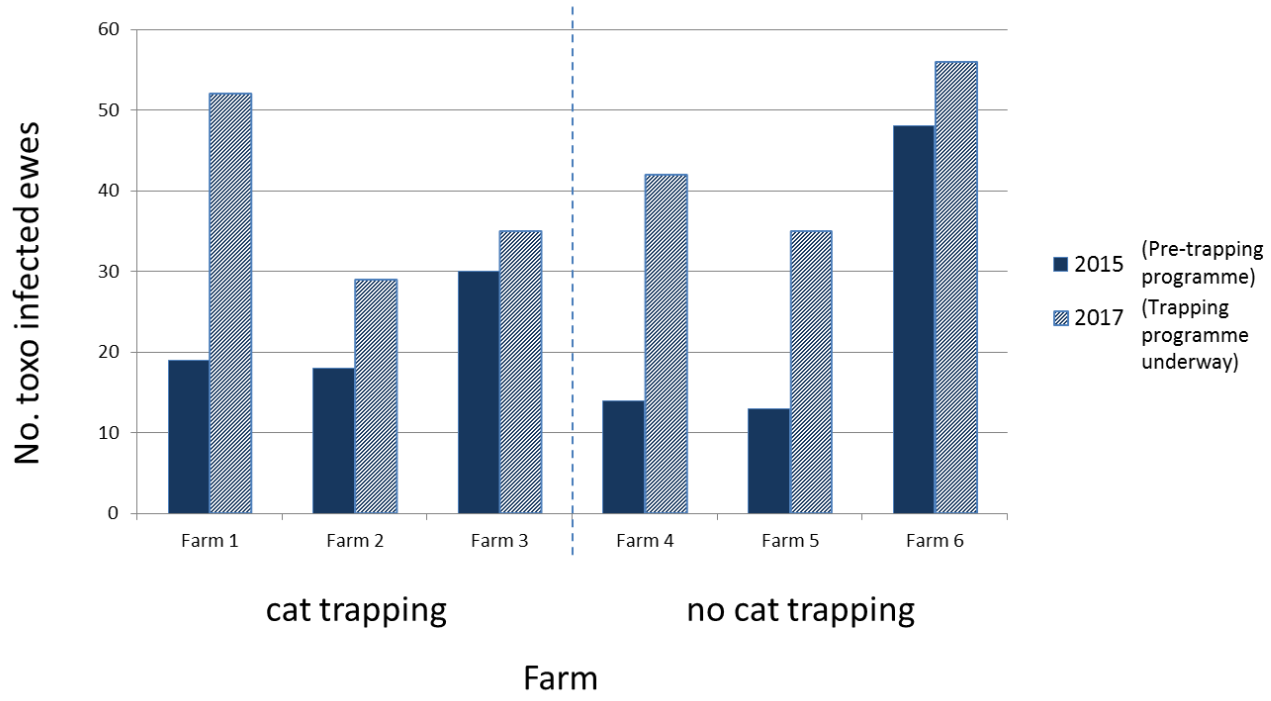


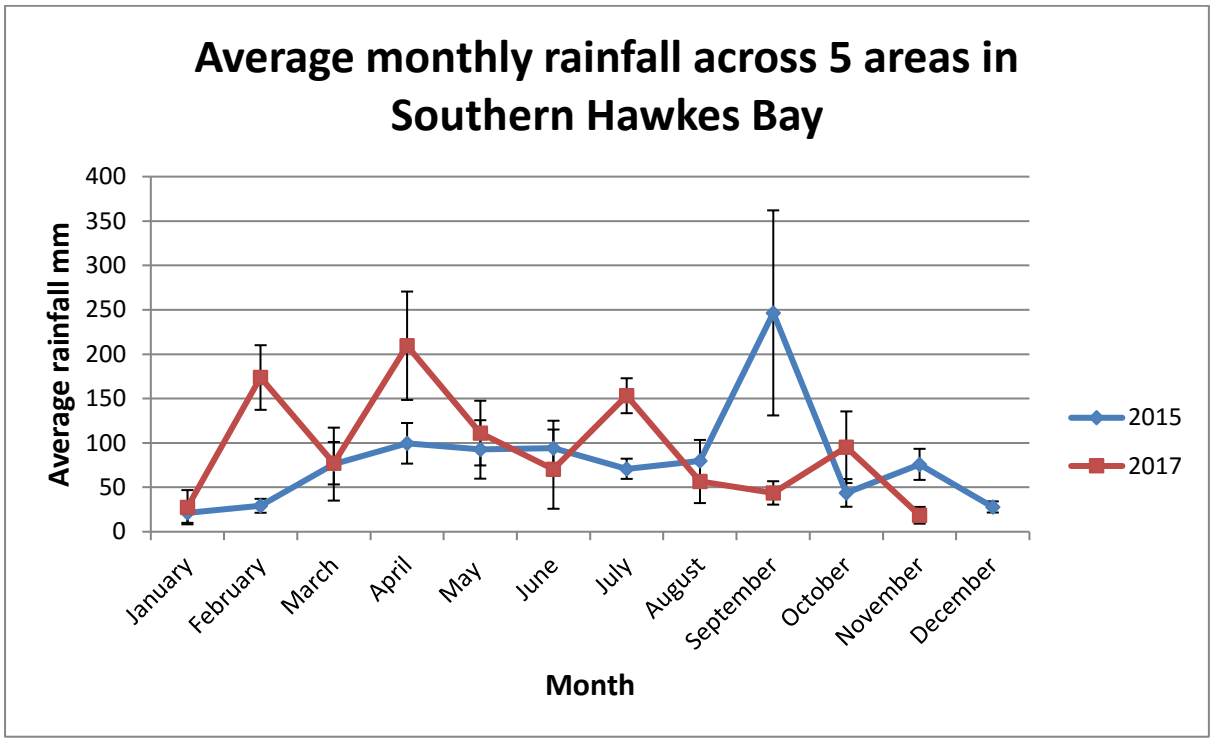
Table 1 Summary of the percentage of 60 one-year-old ewes infected with *T. gondii* on 3 farms with feral cat control (Farm 1, Farm 2, Farm 3), and 3 control farms (Farm 4, Farm 5, Farm 6) in 2015 and 2017, and an assessment of change in prevalence (delta, Δ) over these 2 years. [a]Δ is calculated as number of ewes infected in 2017 minus number infected in 2015.

		2015	**2017**	**Δ[a]**
Case	**Farm 1**	31.7	86.7	55.0
	Farm 2	30.0	48.3	18.3
	Farm 3	50.0	58.3	8.3
	Average	37.2	64.4	**27.2**
	(standard deviation)	(11.1)	(19.9)	(24.6)
Control	**Farm 4**	23.3	70.0	46.7
	Farm 5	21.7	58.3	36.7
	Farm 6	80.0	93.3	13.3
	Average	41.7	73.9	**32.2**
	(standard deviation)	(33.2)	(17.8)	(17.1)

Rainfall

There were relatively strong fluctuations in monthly rainfall during 2017. In particular there were greater than average spikes in February, April and July. In 2015 a large spike was seen in September but rainfall in other months was relatively low. The monthly mean rainfall over the year was greater in 2017 (94.3 ± 61.9) than in 2015 (79.8 ± 59.3).

Figure 2. Average monthly rainfall over 5 areas in Southern Hawkes Bay in 2015 and 2017.



Summary

Overall, there is a greater level of *T. gondii* in one-year-old ewes from all farms in 2017 than in 2015. The timing of the rainfall spikes in 2017 - about 4 to 6 months before sampling began - could have contributed to the greater seroprevalence, or other intrinsic factors may be involved. Further work is required to more fully understand the greater seroprevalence in 2017 than 2015 overall, and more specifically the relationship between rainfall and *T. gondii* seroprevalence in ewes. For example, it is possible wetter conditions increases the persistence of the environmentally occurring infective *T. gondii* oocysts by preventing desiccation, and could also lead to a greater spread across the farms – both of which would increase the opportunity for ewe infection.

The difference in infection rates was only marginally higher in farms that did not have feral cat trapping. While trapping effects appear to be modest, little trapping time had elapsed before the 2017 sampling. Further, in one farm (Farm 1) kill traps are not permitted due to its proximity to a built-up area, and trapping instead relies on specialist active control twice a year. As cat removal efforts increase and become more consistent, the modest effects shown here could be accentuated with time. In summary, an effect of cat trapping on reduction of *T. gondii* seroprevalence might be emerging but continued sampling and testing is required to verify this, and we recommend that future cat trapping be as thorough as possible. A meaningful comparison of abortion rates between footprint and control farms can then be made.

References

1. Krijger I, Cornelissen J, Wisselink H, Meerburg B (2014) Prevalence of Toxoplasma gondii in common moles (Talpa europaea). Acta Veterinaria Scandinavica 56: 1-4.

2. Landcare Research Cape to City Consortium (2016) Toxoplasma gondii screening in cats and mice using PCR as part of the Cape to City Initiative. Landcare Report