

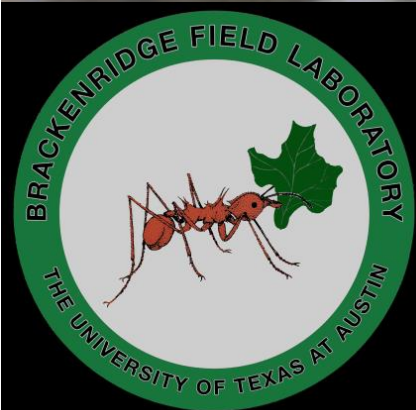
# The intersection of social organization and pathogen biology in an invasive ant: tawny crazy ants and their microsporidian pathogen

**Edward LeBrun**

**collaborators:**

**Larry Gilbert, Rob Plowes, Patricia Folgarait, Martin Bollazi, and Melissa Jones.**

**Brackenridge Field Laboratory  
Department of Integrative Biology  
University of Texas at Austin**



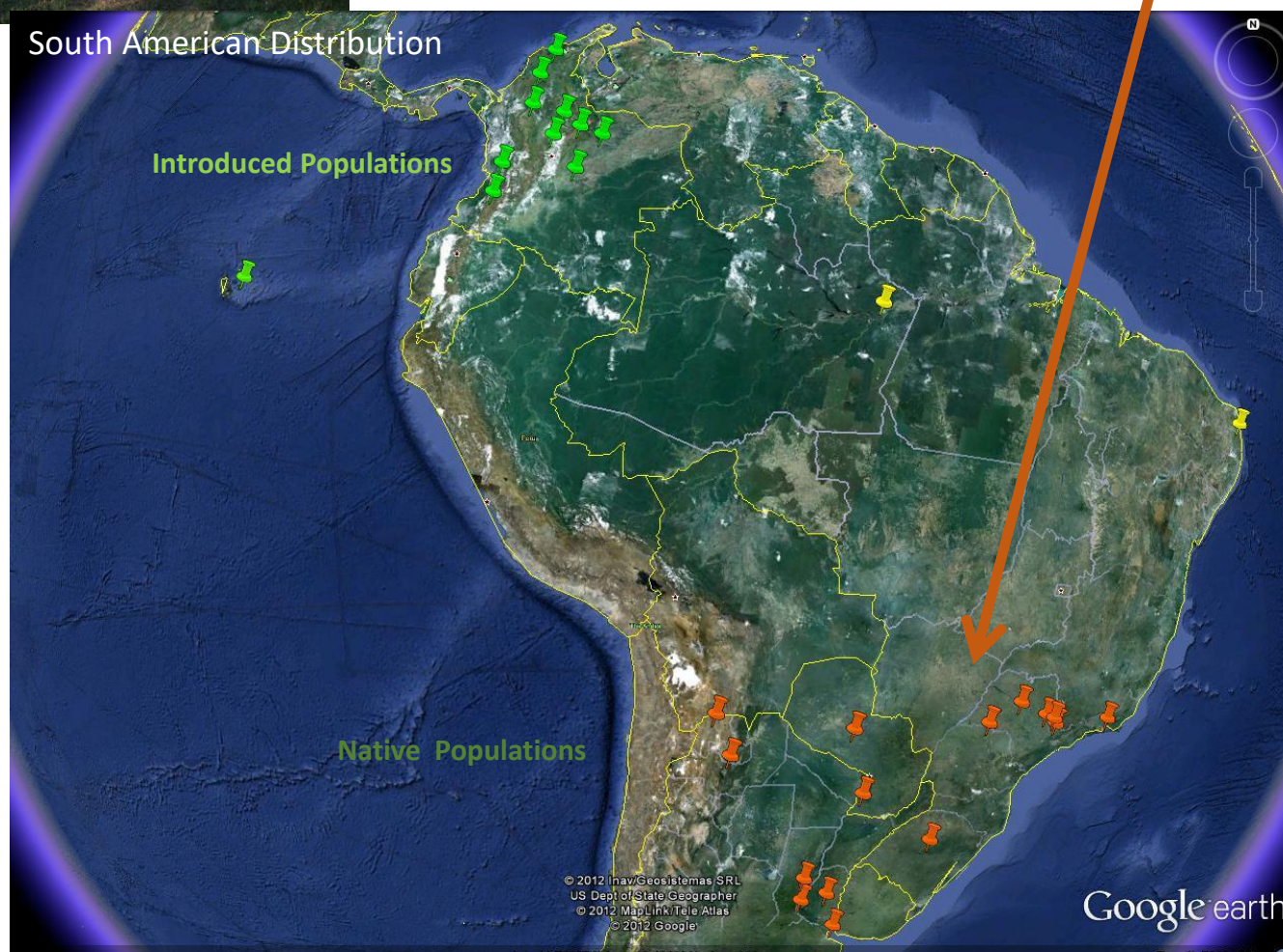




## Tawny Crazy ant: Native Range

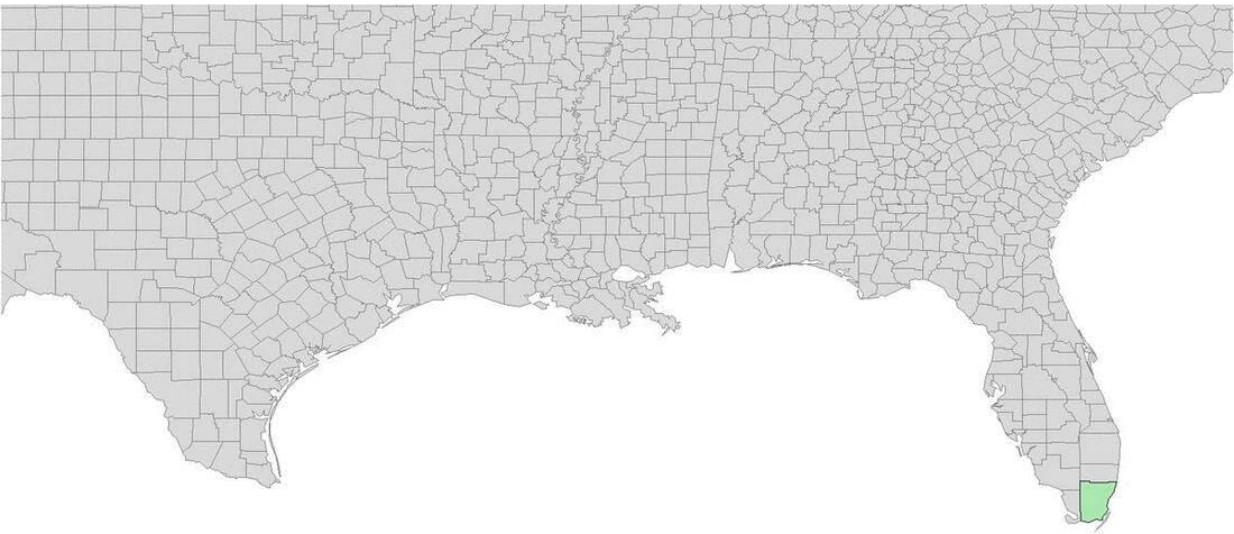


## South American Distribution





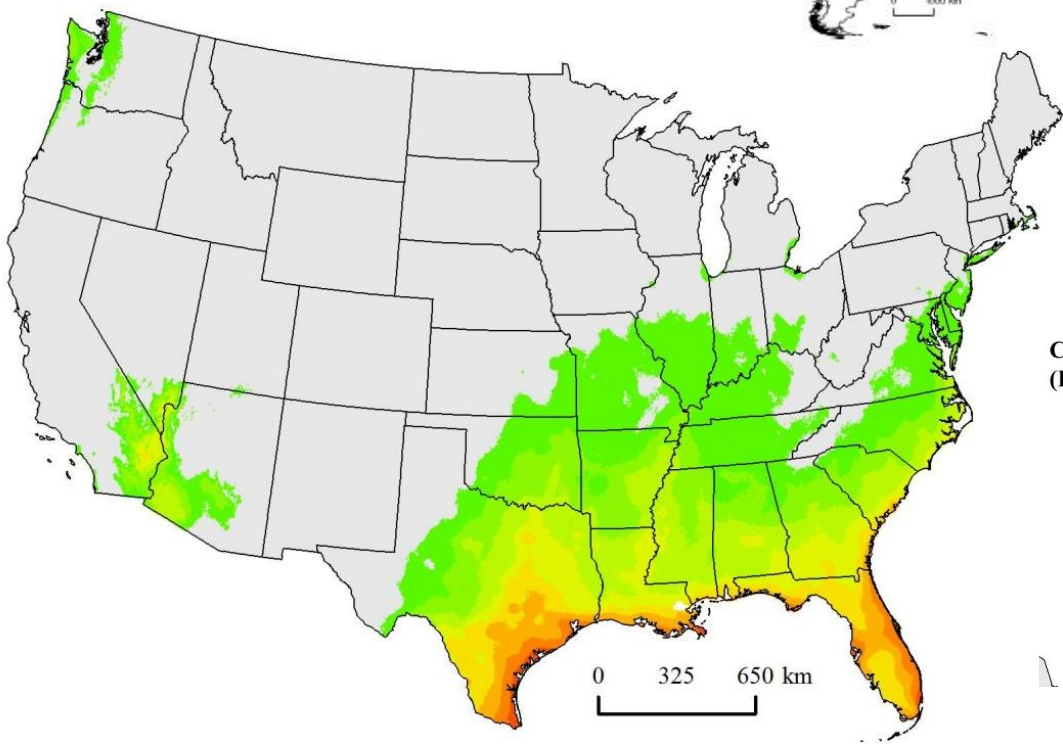
# TCA Spread and Climate Modeling Projection of Suitable Regions



Tawny Crazy Ants (*Nylanderia fulva*) Distribution

- 1990
- USA Counties

1990



Climatic suitability  
(Probability of presence)

- < 0.1
- 0.1 - 0.2
- 0.2 - 0.3
- 0.3 - 0.4
- 0.4 - 0.5
- 0.5 - 0.6
- 0.6 - 0.7
- 0.7 - 0.8
- 0.8 - 0.9
- 0.9 - 1

Extrapolation based upon North and South American occurrence records.

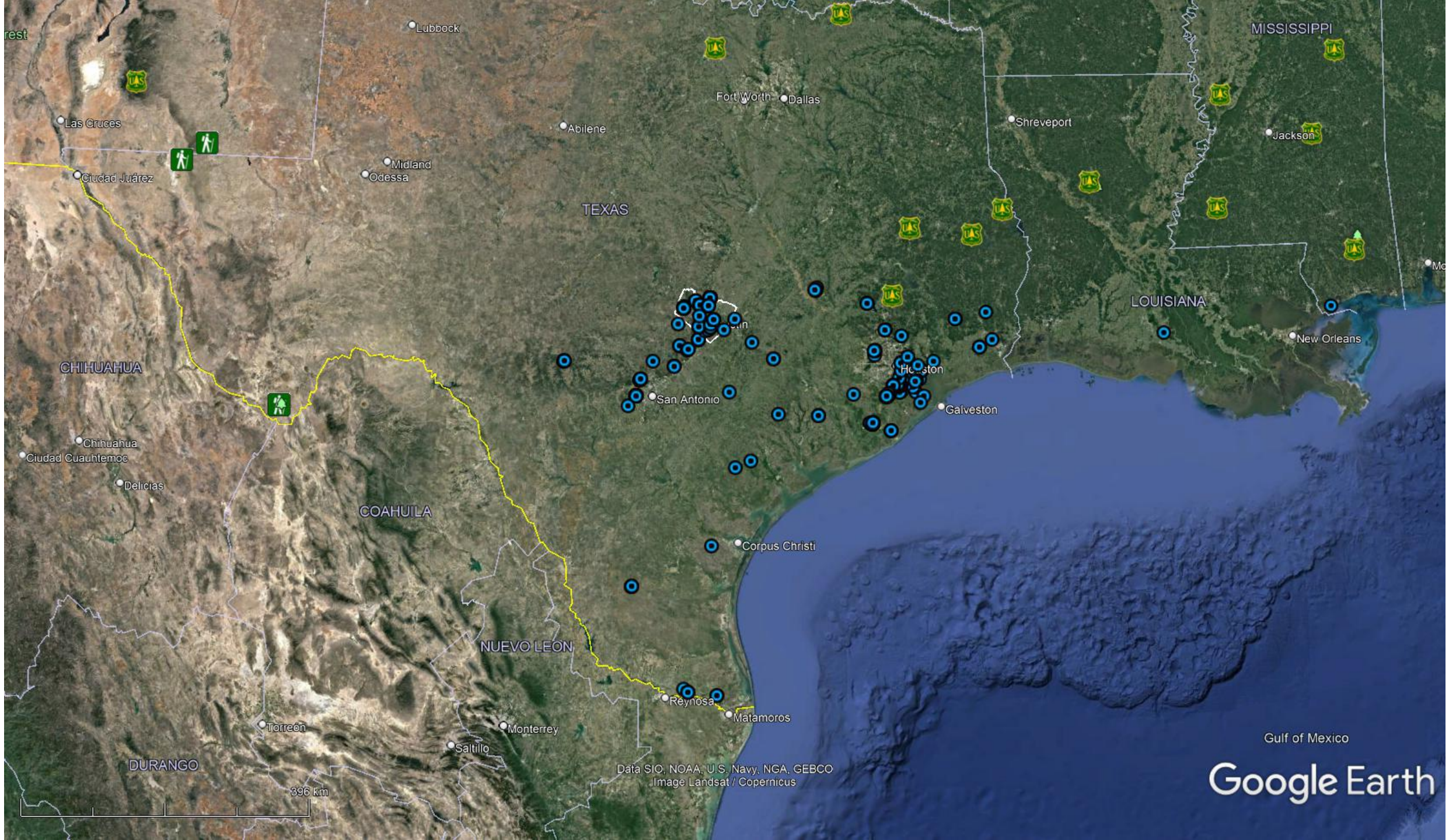
Ecology and Evolution

Open Access

Evidence of niche shift and global invasion potential of the Tawny Crazy ant, *Nylanderia fulva*

Sunil Kumar<sup>1,2</sup>, Edward G. LeBrun<sup>3</sup>, Thomas J. Stohlgren<sup>1,2</sup>, Jared A. Stabach<sup>1</sup>, Danny L. McDonald<sup>4</sup>, David H. Oi<sup>5</sup> & John S. LaPolla<sup>6</sup>

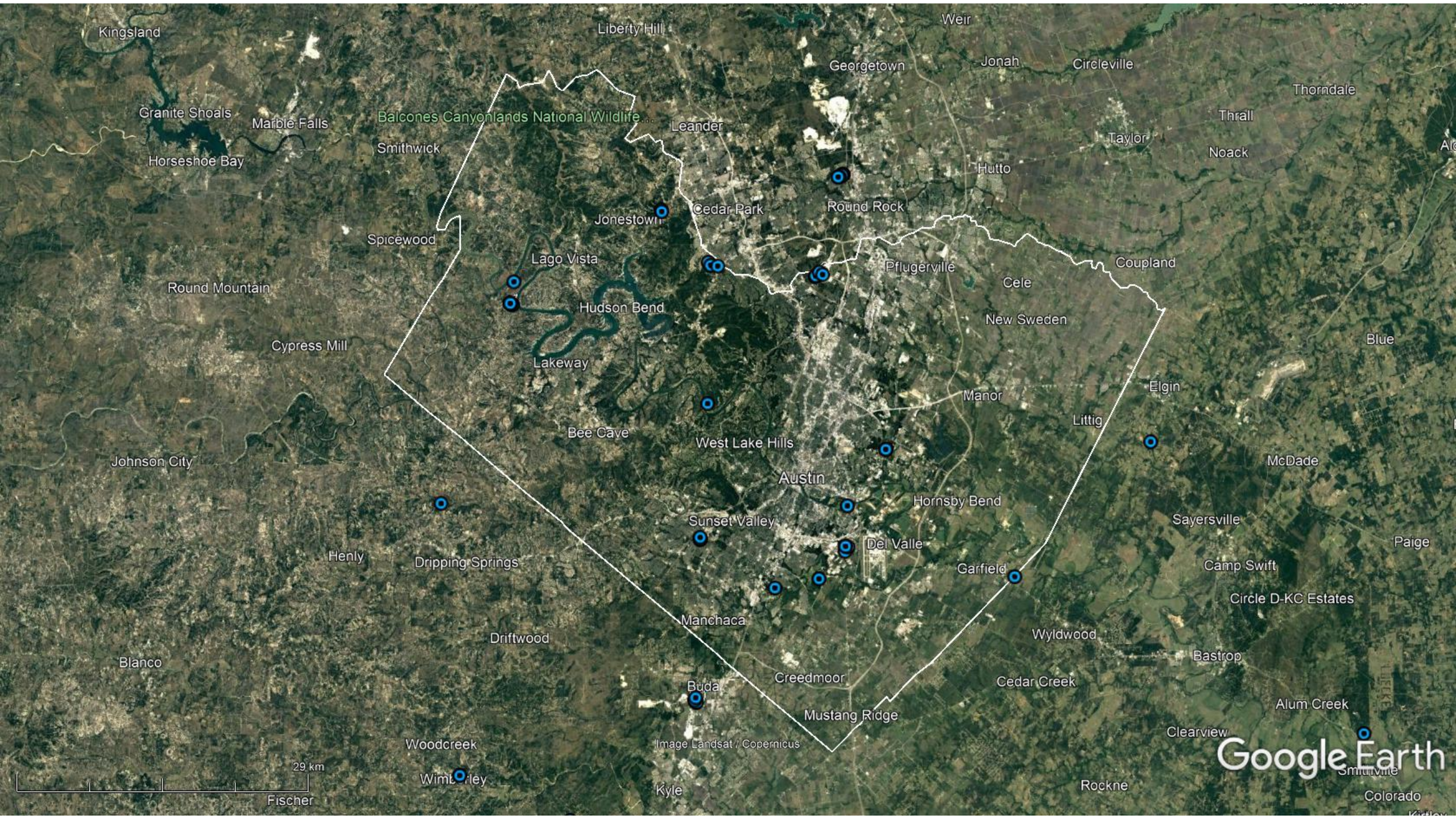




Data SIO, NOAA, U.S. Navy, NGA, GEBCO  
Image Landsat / Copernicus

Gulf of Mexico  
Google Earth







# Natural History

## Foraging

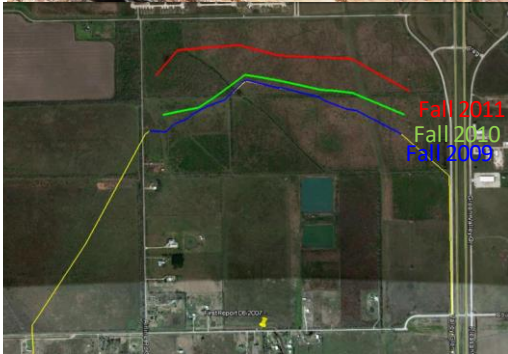
- Omnivorous. Scavenging and preying on insects and feeding on insects and honeydew.
- Ecologically dominant. Extremely good at monopolizing food resource base.

## Social Biology

- Multiple sometimes 100s of queens per nest.
- Opportunistic nester.

## Reproduction

- Females do not fly. Apparently all mating is within the nest and spread is through nest fission.
- Long distance dispersal is human mediated.
- Populations expand radially 0 – 200 m a year, forming a circular “plaque” on the landscape with a discrete boundary separating invaded from uninvaded.



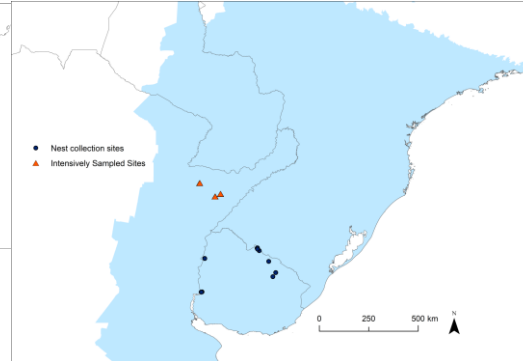
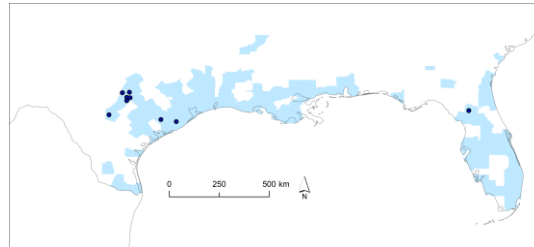
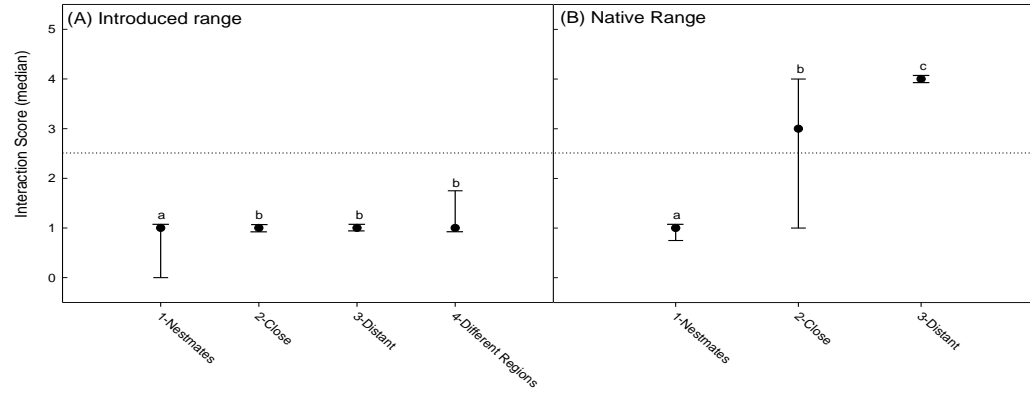
# Extreme Densities



Whirlpool Cave, October 2017  
Video: R. Zarria

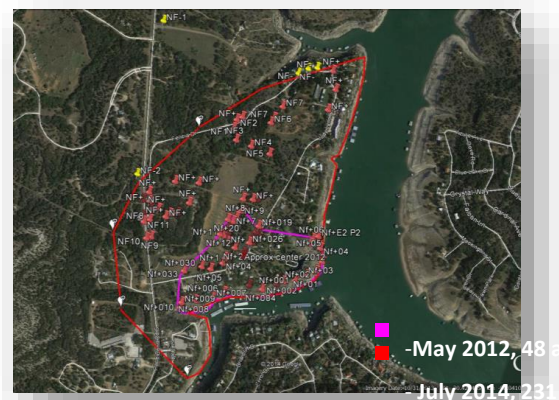
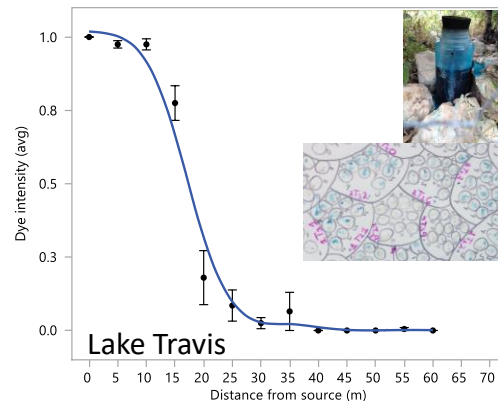


# Tawny crazy ant social organization : Supercolonial



**PLOS ONE**  
RESEARCH ARTICLE  
November 22, 2019  
Ritualized aggressive behavior reveals distinct social structures in native and introduced range tawny crazy ants  
Edward G. LeBrun<sup>1\*</sup>, Robert M. Plowes<sup>1</sup>, Patricia J. Folgarait<sup>2</sup>, Martin Bollazzi<sup>3</sup>, Lawrence E. Gilbert<sup>4</sup>

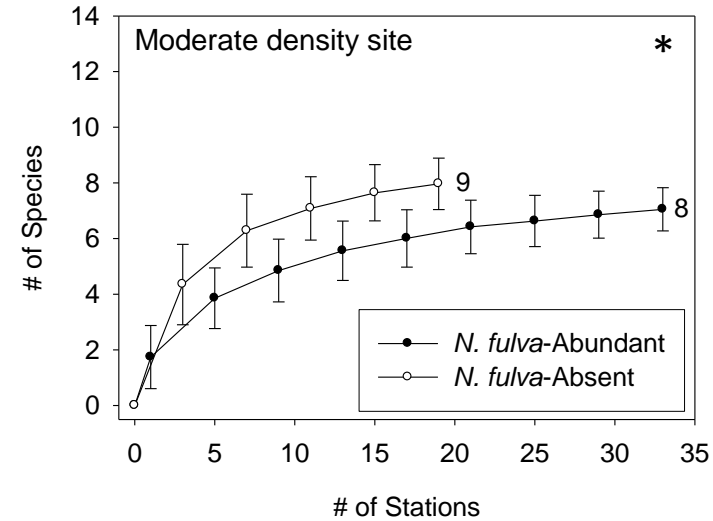
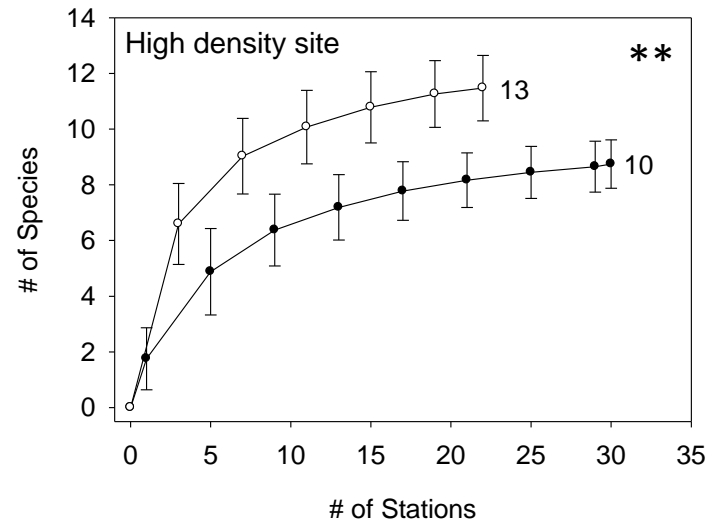
- All tawny crazy ants in the SE USA are members of the same, highly-fragmented supercolony.
  - Eyer et al. 2018
  - Lawson and Oi 2020
- Not true in South America. Highly polygyne, multinest, expansive but still multi-colonial.



- Local populations are functionally interconnected networks of nests spanning square kilometers.



# Impacts Other Ants: Species Richness

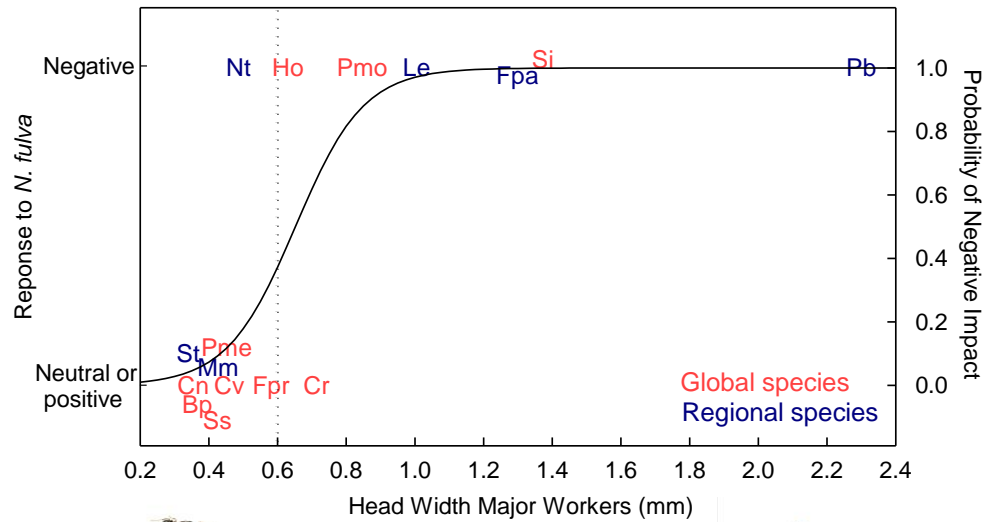


- Reduced ant assemblage species richness in both high and moderate density sites in areas invaded by *N. fulva*.
- Species loss is non-random.

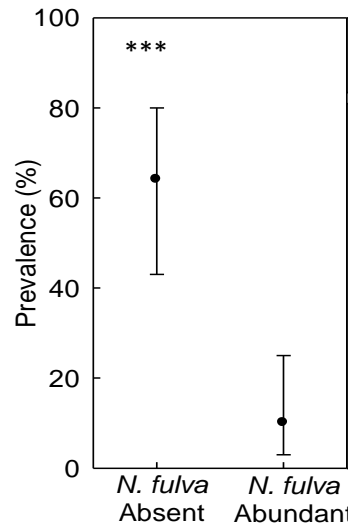




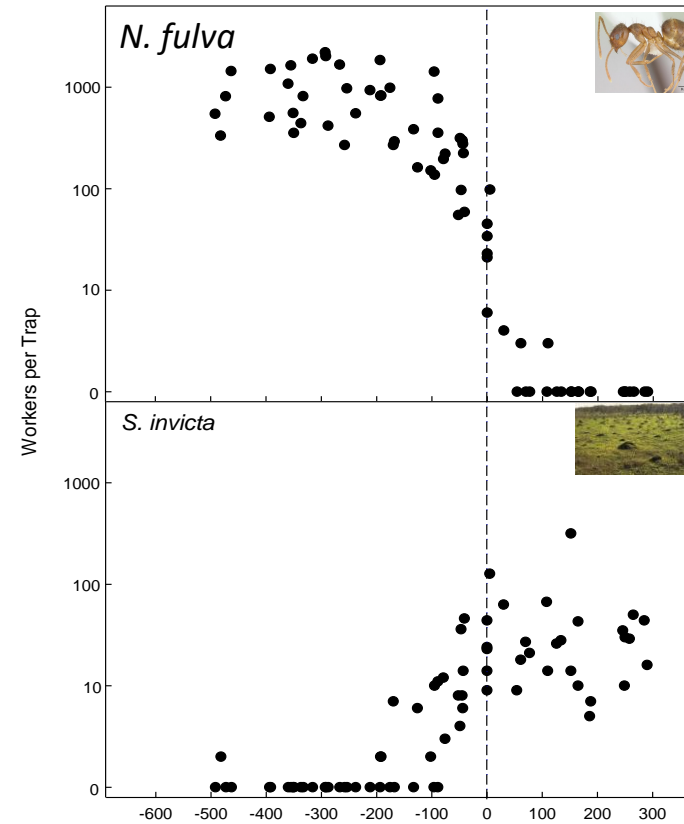
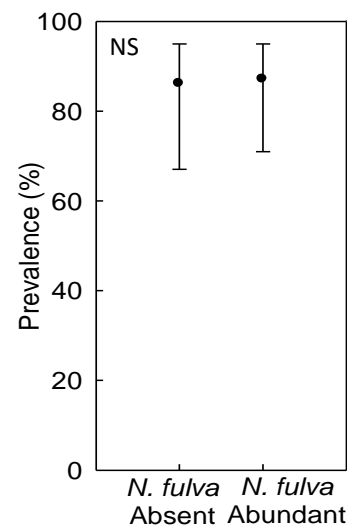
# Impacts on Other Ants



Regional Species

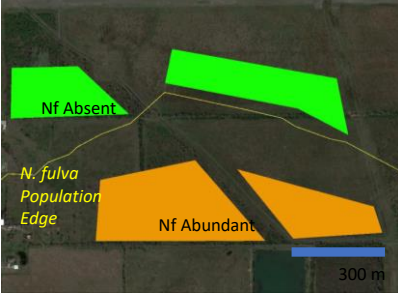


Global Species



- Native ants are preferentially displaced.
- Imported fire ants are displaced.

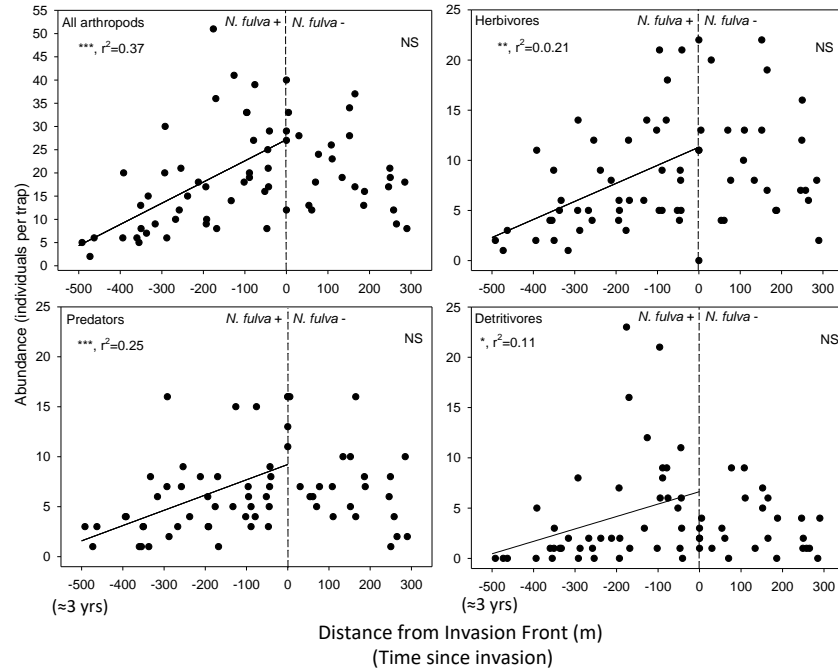




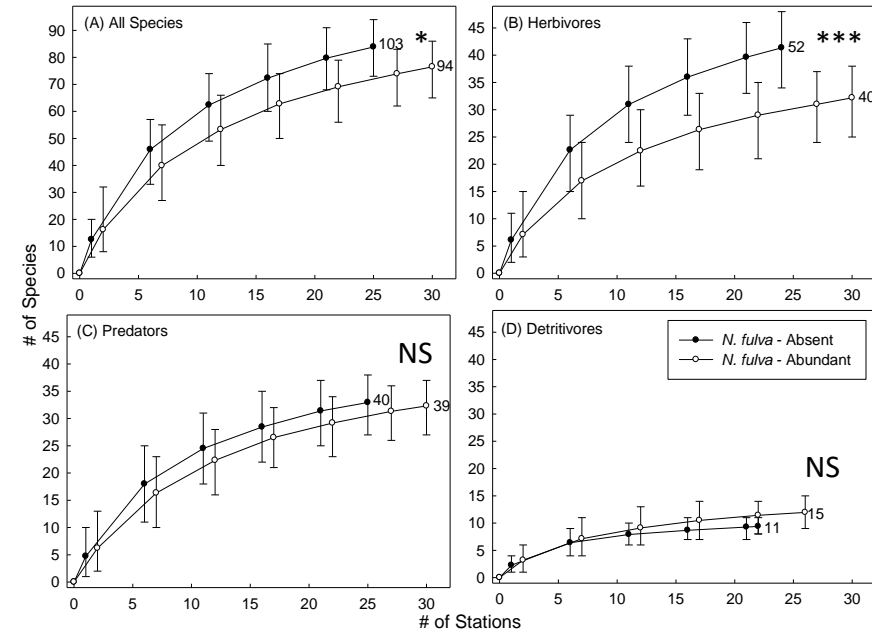
# TCA Impacts: Other Arthropods



## Abundance



## Species Richness





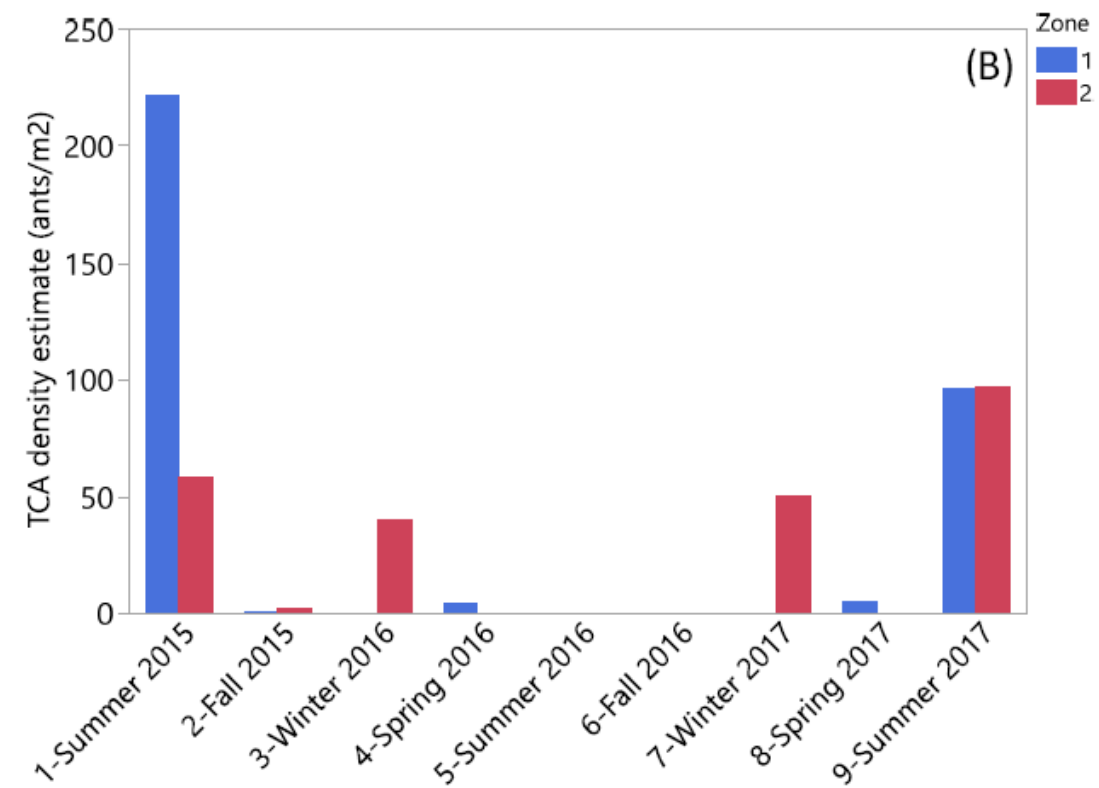






Figure 20: TCA preying on cave invertebrates in Whirlpool Cave. Top: *Cryptachaea porteri*. Bottom: *Oxidus gracilius*.

Table 2: Contrasts of abundances from time periods when TCA were present at No Rent Cave with time periods when they were absent.

Species <sup>1</sup>	TCA Status <sup>2</sup>	N	Median (IQR) Abundance <sup>3</sup>	DF	$\chi^2$	P value <sup>4</sup>
<i>Cicurina buwata</i> - spider	Absent	10	0.75 (0.47, 0.84)	1	1.2	0.27
	Present	5	0.34 (0.28, 0.71)			
<i>Cicurina varians</i> - spider	Absent	10	0.55 (0.4, 0.89)	1	4.6	0.03
	Present	5	0.23 (0.05, 0.45)			
<i>Pseudosinella violenta</i> - springtail	Absent	10	0.21 (0.07, 0.41)	1	0.1	0.81
	Present	5	0.09 (0.07, 0.46)			
<i>Texella reyesi</i> - harvestman	Absent	10	0.04 (-0.17, 0.32)	1	1.5	0.22
	Present	5	-0.13 (-0.19, -0.03)			



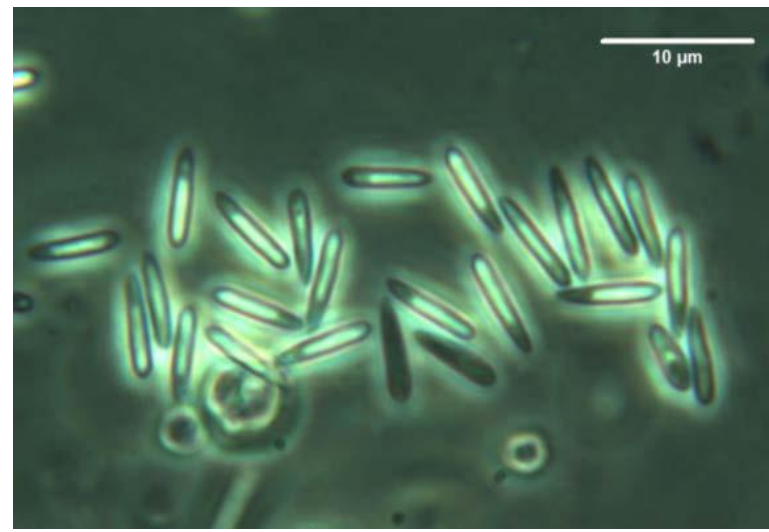
# Impacts

- **Economic**
  - Invade homes and yards, becoming an extreme nuisance
  - Cause major damage to electrical components and computer systems
  - Serious agricultural pests damaging plants and invading commercial honeybee colonies

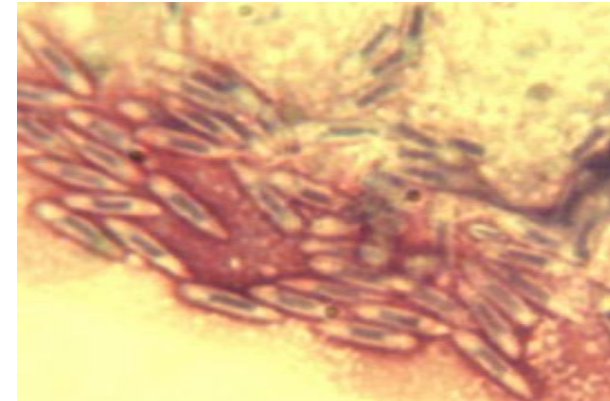




# ***Myrmecomorba nylanderiae*: microsporidian pathogen of tawny crazy ants**



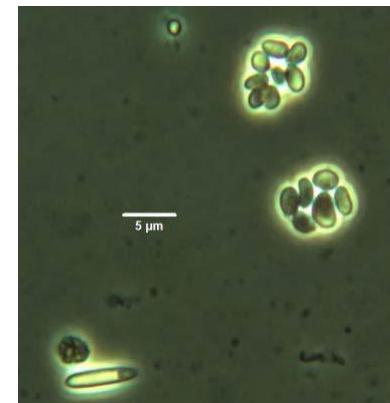
- First documented pathogen of tawny crazy ants.
- Four spore types
- Primarily infects the worker caste, brood, and males.
- Documented in scattered populations in Florida and Texas.
- Unknown origin.
- Unknown dispersal mechanism.



*Myrmecomorba nylanderiae* gen. et sp. nov., a microsporidian parasite of the tawny crazy ant *Nylanderia fulva*

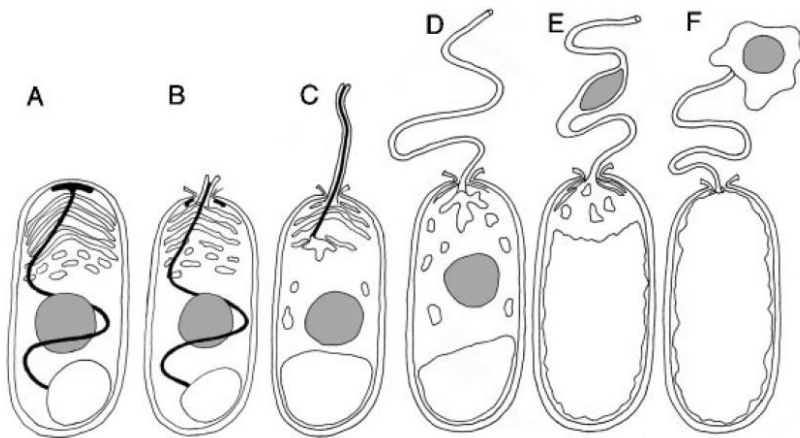
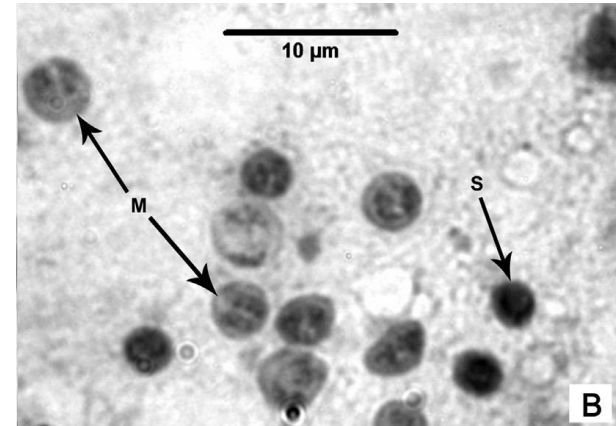
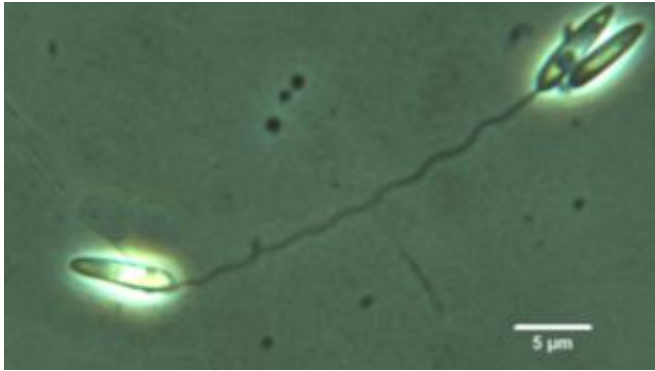
Robert M. Plowes<sup>a,\*</sup>, James J. Becnel<sup>b</sup>, Edward G. LeBrun<sup>a</sup>, David H. Oi<sup>b</sup>, Steven M. Valles<sup>b</sup>, Nathan T. Jones<sup>a</sup>, Lawrence E. Gilbert<sup>a</sup>

[Journal of Invertebrate Pathology 129 \(2015\) 45–56](#)

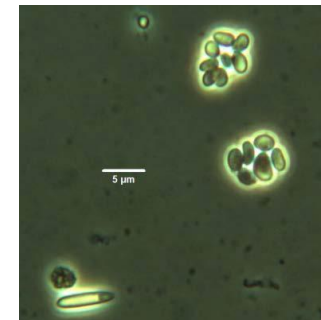
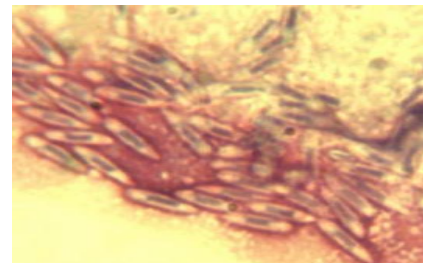


# Microsporidian pathogens

- Specialized single celled, intracellular, pathogen
- Virus like biology

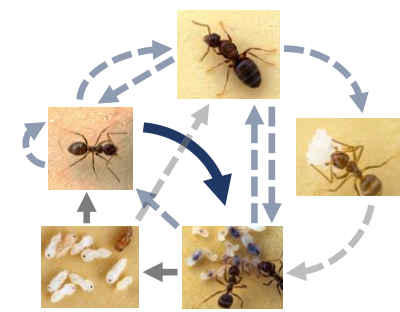


Keeling & Fast 2002





# Intra-nest transmission and impacts of *M. nylanderiae*

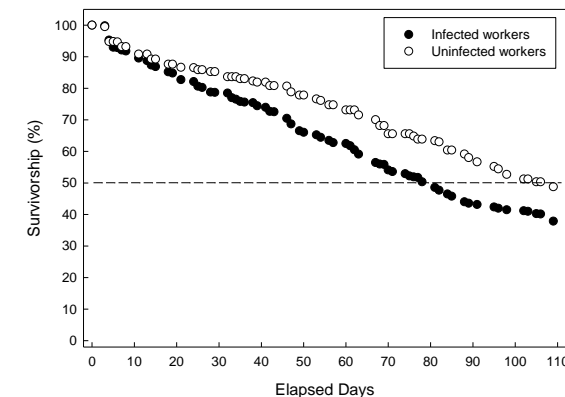
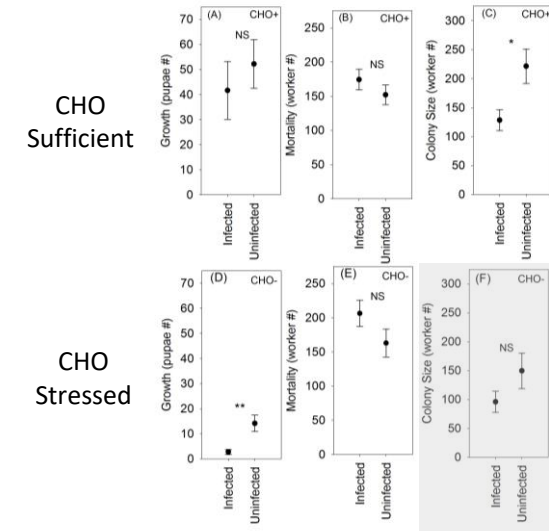


## Transmission

- Queens in natural populations are largely uninfected.
- Infection appears to be acquired primarily or exclusively in the larval stage.
- Infection appears to be largely passed in a closed loop amongst worker caste.

## Virulence

- *M. nylanderiae* impacts colony growth by impeding larval development and shortening worker life-span.
- It does so most strongly under carbohydrate stressed conditions suggesting its impacts on ant colonies will be magnified in winter and periods of drought..



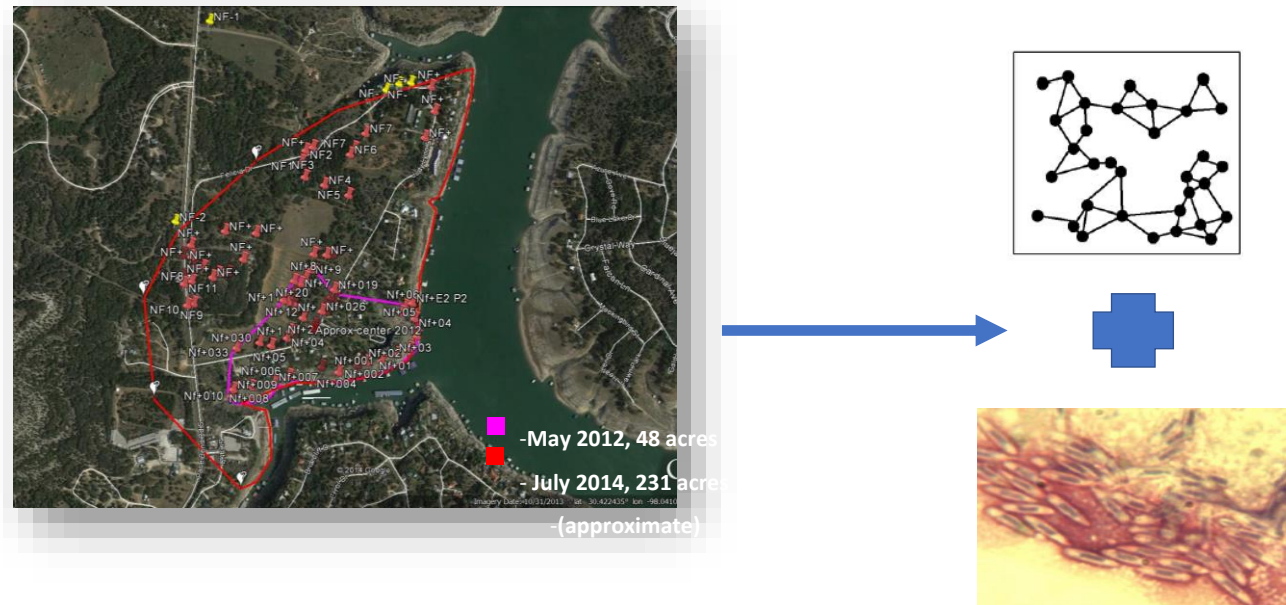
ORIGINAL CONTRIBUTION

WILEY JOURNAL OF APPLIED ENTOMOLOGY

The microsporidian pathogen *Myrmecomorba nylanderiae*:  
Intracolony transmission and impact upon tawny crazy ant  
(*Nylanderia fulva*) colonies

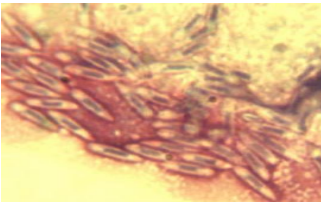
E. G. LeBrun<sup>1,2</sup> | K. J. Ottens<sup>1</sup> | L. E. Gilbert<sup>2</sup>

# Population scale impacts?

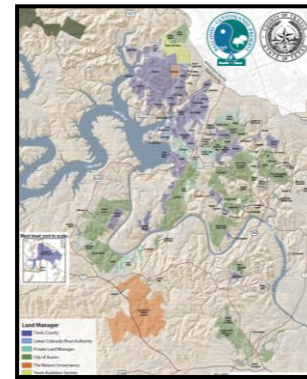
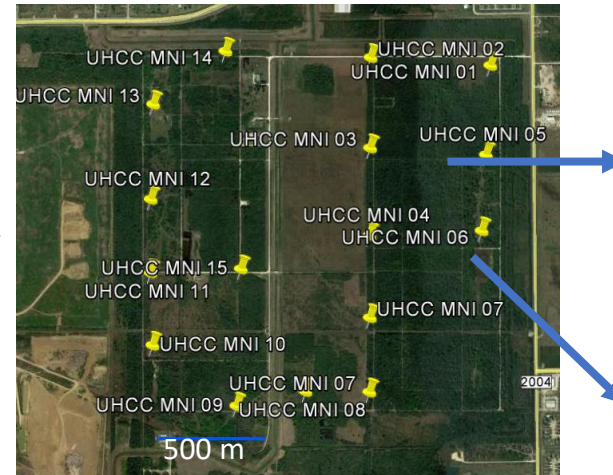
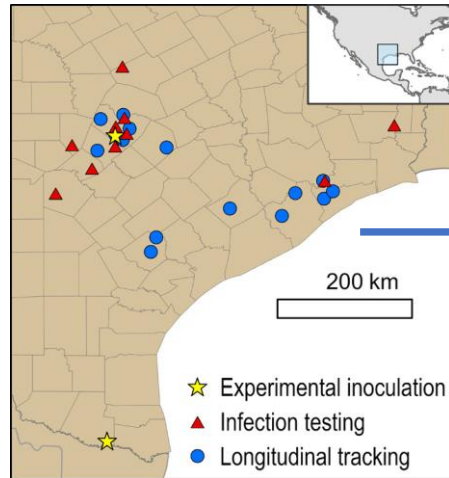


- Supercolonial ants thought to be both vulnerable to pathogen acquisition and potentially more able to tolerate pathogen infection (S. Cremer 2019).

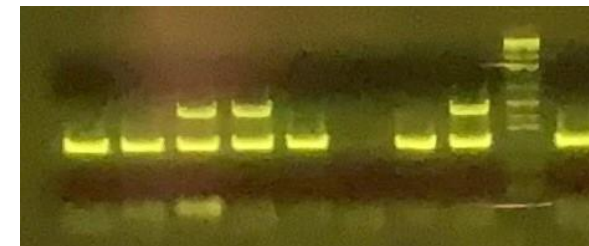
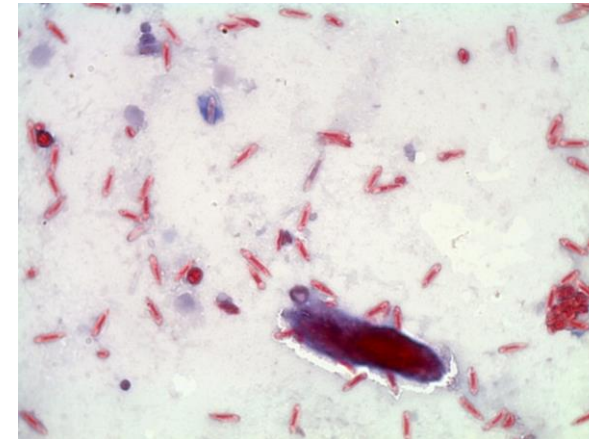




# Population Impacts: Longitudinal study of natural infections



- Pitfall trapping to quantify abundance
- Spore counting and PCR to quantify infection intensity
- 12 widely scattered sites, 52 year to year transitions in TCA abundance and disease parameters (2009-2020)



**PNAS**

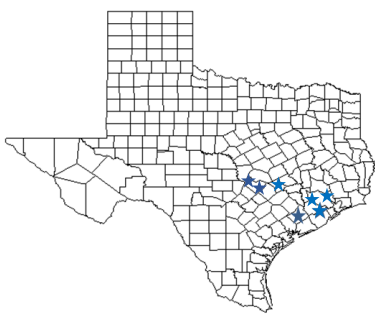
RESEARCH ARTICLE

APPLIED BIOLOGICAL SCIENCES

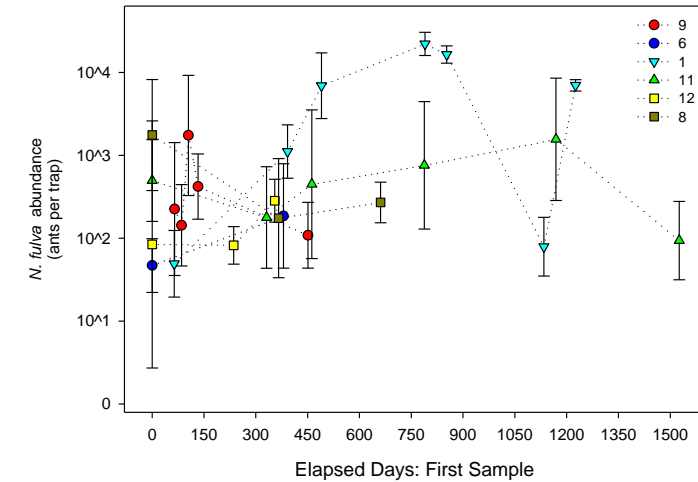
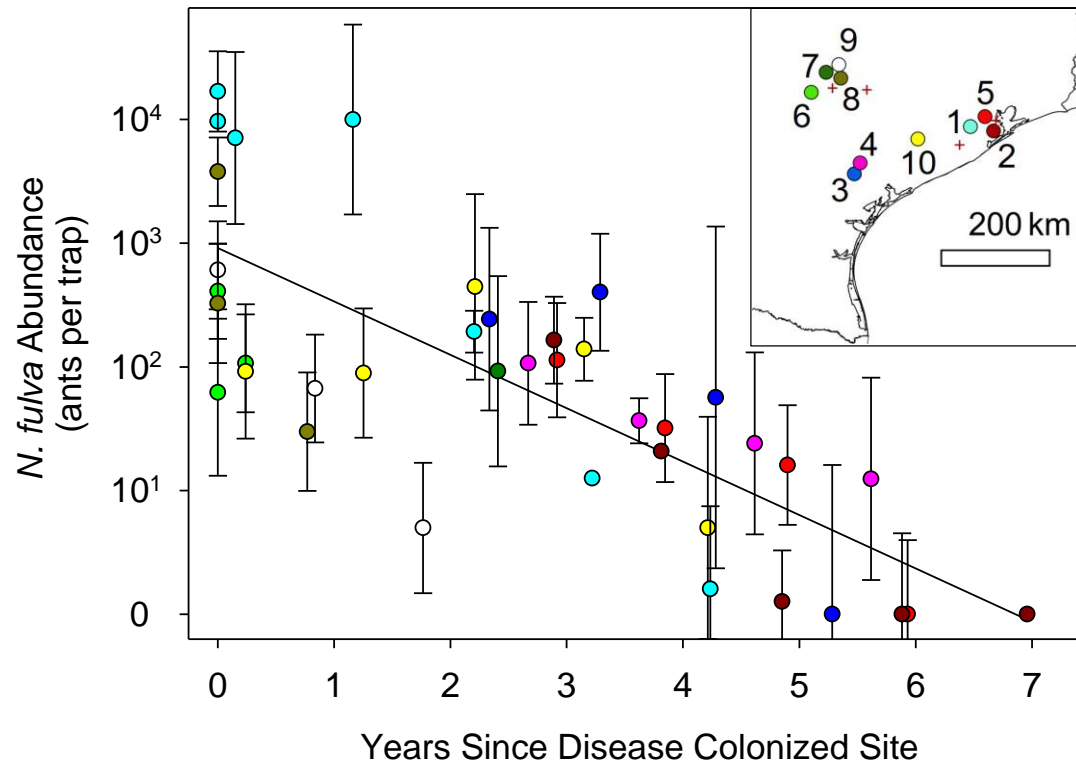
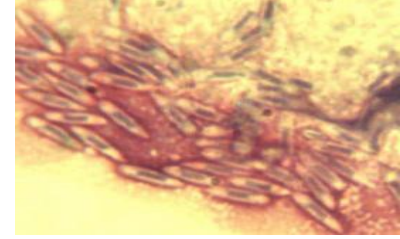
## Pathogen-mediated natural and manipulated population collapse in an invasive social insect

Edward G. LeBrun<sup>a,1</sup>, Melissa Jones<sup>b</sup>, Robert M. Plowes<sup>a,1</sup>, and Lawrence E. Gilbert<sup>c</sup>

Edited by Daniel Simberloff, University of Tennessee, Knoxville, TN; received August 6, 2021; accepted February 7, 2022



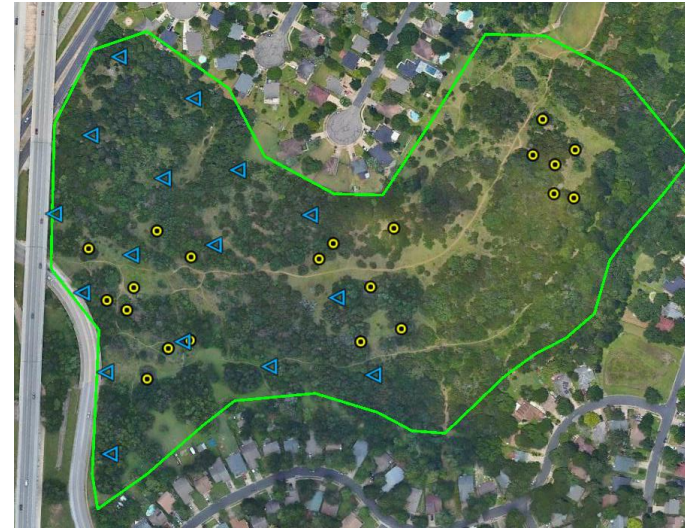
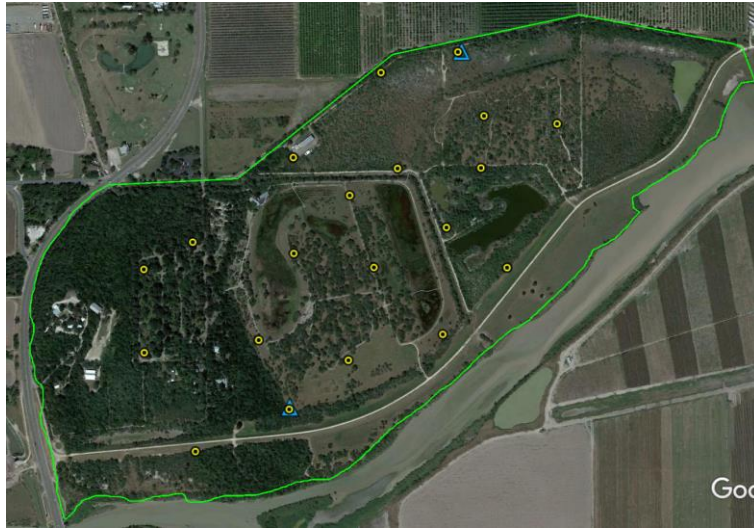
## Population Impacts: Longitudinal study of natural infections




- Infected TCA populations universally decline in abundance, often substantially.
- Some populations have declined to local extinction.



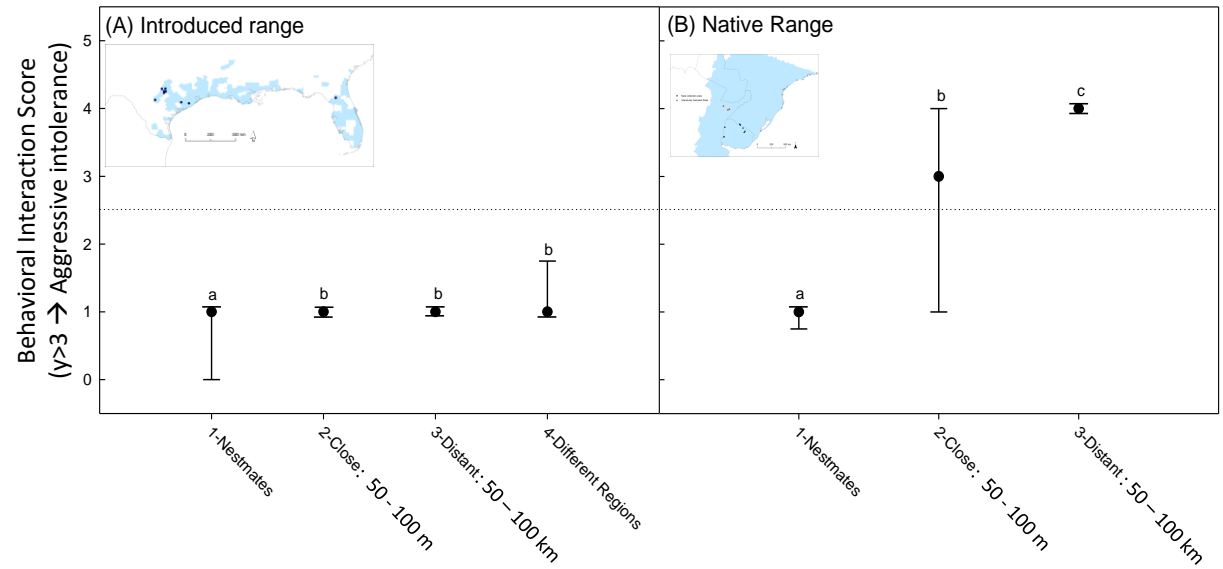
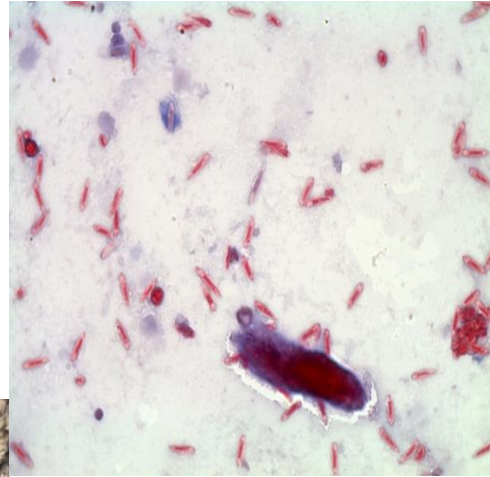
## Experimental inoculations of TCA population with *M. nylanderiae*



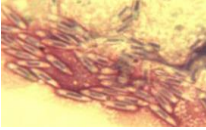
- 
- ▲ Inoculation stations
  - Pitfall traps

- Spring of 2017: 2 inoculations sites
- Site 1: 4 nest boxes with 800 infected workers and 0.25 cc of brood each installed at 2 inoculation stations. Response followed over 91 hectares.
- Site 2: 250 infected workers and 0.25 cc of brood released onto foraging trails at 15 inoculation stations. Response followed over 12 hectares.
- Both sites tested negative for the disease pre-inoculation.
- But,, at Site 1,, Mn found present at low levels at time of inoculation. In samples collected at the time of inoculation.

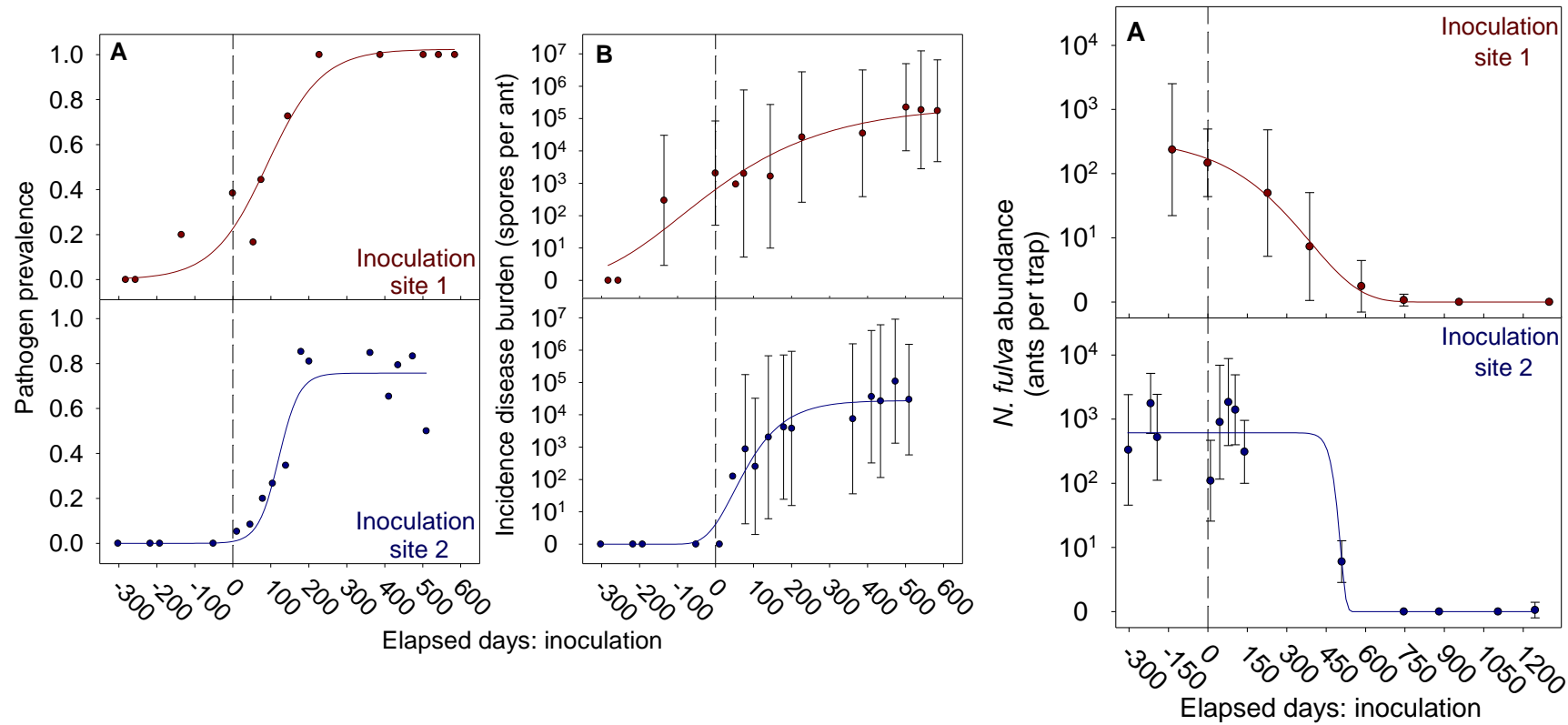
# Experimental Inoculations of uninfected populations



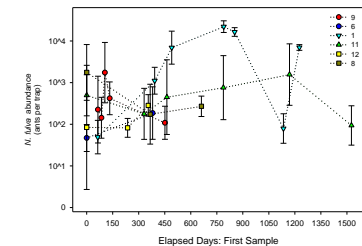




# Impact of Inoculation on TCA Populations

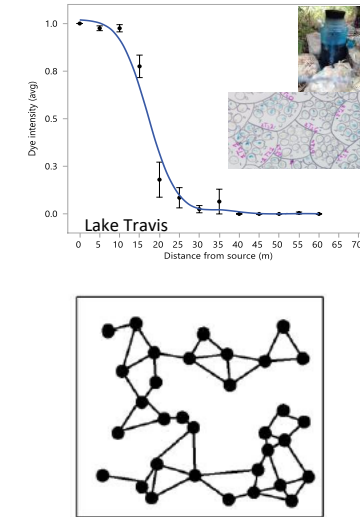
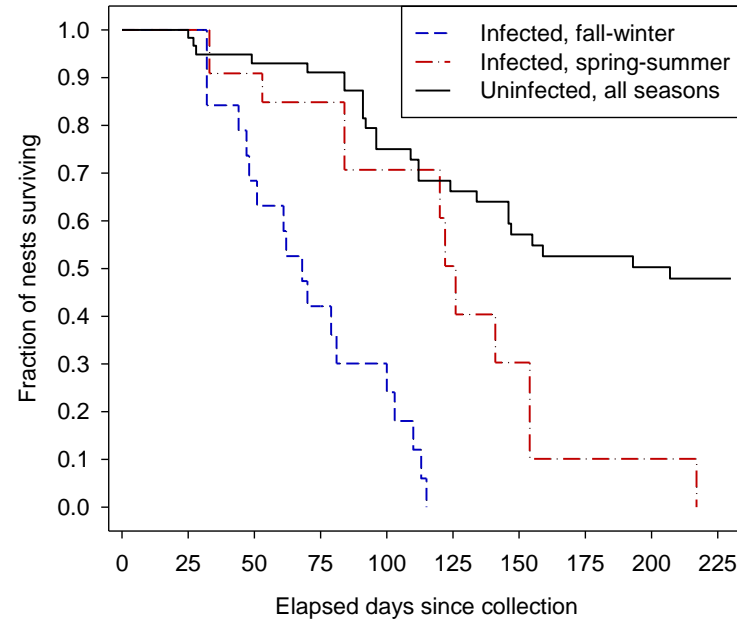
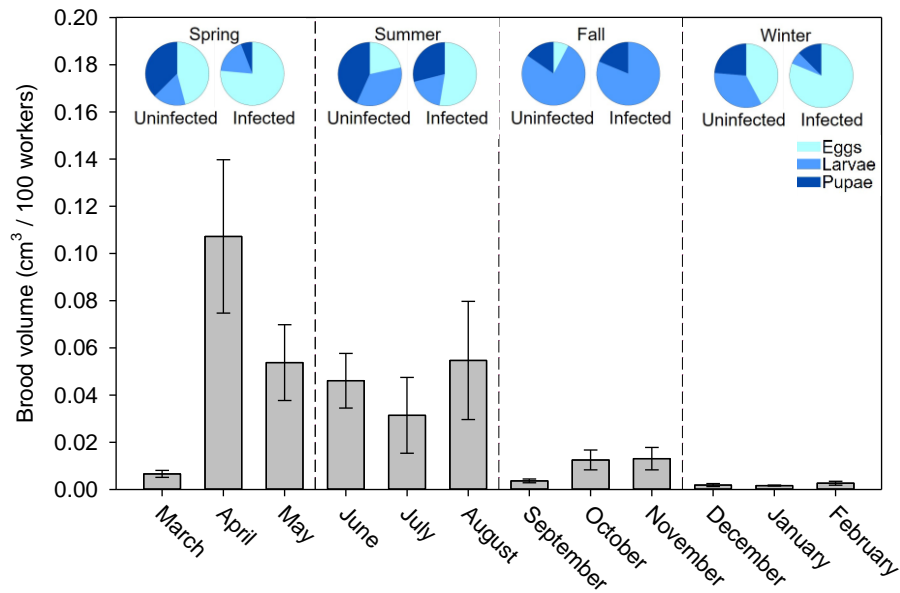


- 65% decline in incidence in pitfall traps.
- 99% decline in median abundance where present.



# Why do supercolonies decline to local extinction?

## Mind the gap! (hypothesis)



- All colony fragments cease brood production in September and do not resume until April: a 7 month gap.
- Colony fragments collected in fall decline rapidly dying out in about 4 month: insufficient to bridge the gap in brood production.
- Extreme connectivity short circuits negative density dependent regulation of disease transmission.



Site	Inoculation Year	Inoculation Season	Method - primary	Method - Secondary	Inoculations	Successes	Failures	% Success
Convict Hill	2017	Spring	Foraging Trail	Workers vs brood	15	6	9	40%
ELGSP	2017	Spring	Nest	Nest Box	2	1	1	50%
Lake Travis	2017	Spring	Nest	Nest Box	2	0	2	0%
Lake Travis	2018	Spring	Nest	Nest Box	2	0	2	0%
Anderson Mill	2018	Fall	Foraging Trail	Workers+brood	16	0	13	0%
Lake Travis	2018	Fall	Foraging Trail	Workers+brood	16	0	16	0%
Anderson Mill	2019	Spring	Foraging Trail	Workers vs workers+brood vs nest box	16	0	24	0%
Lake Travis	2019	Spring	Nest	Nest Box	2	0	2	0%
Anderson Mill	2019	Fall	Nest	Natal nest material	1	0	1	0%
Pace Bend	2020	Fall	Nest	Laboratory nest fusion	7	1	6	14%
Pace Bend	2021	Spring	Nest	Laboratory nest fusion	5	3	2	60%
Roy Guerero	2021	Fall	Foraging Trail	Workers vs workers+brood	7	0	7	0%
Walnut Creek	2021	Fall	Mixed	Workers vs workers+brood vs nest box	15	??	??	??

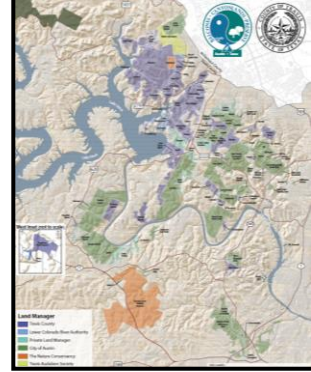
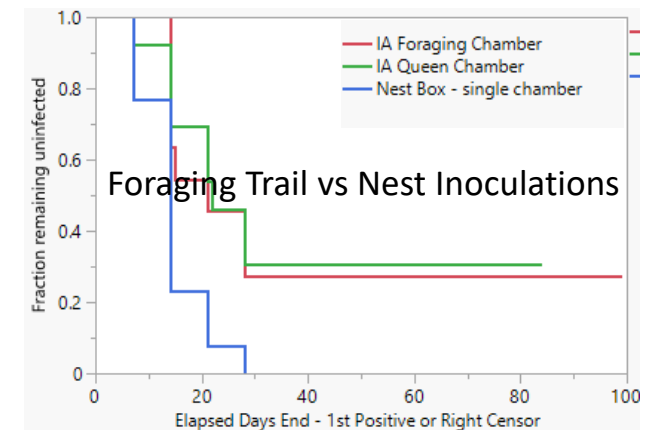
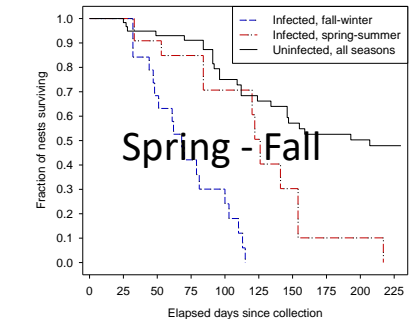
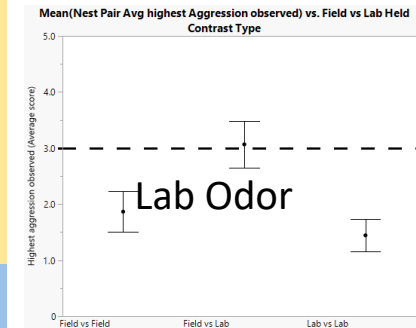
Lab Odor Controls

No Lab Odor Controls

Inoculation methods development: trial and error process with 6-month time lag. Insights from lab provide path forward.

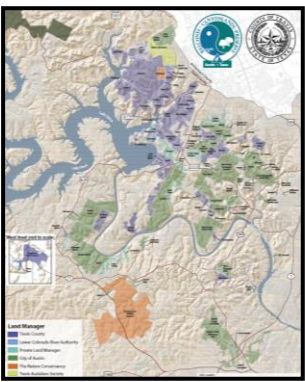
- Spring to early summer inoculations
- Controlling laboratory odor exposure
- Homogenizing odor cues

Laboratory nest fusion or introduction onto foraging trails?









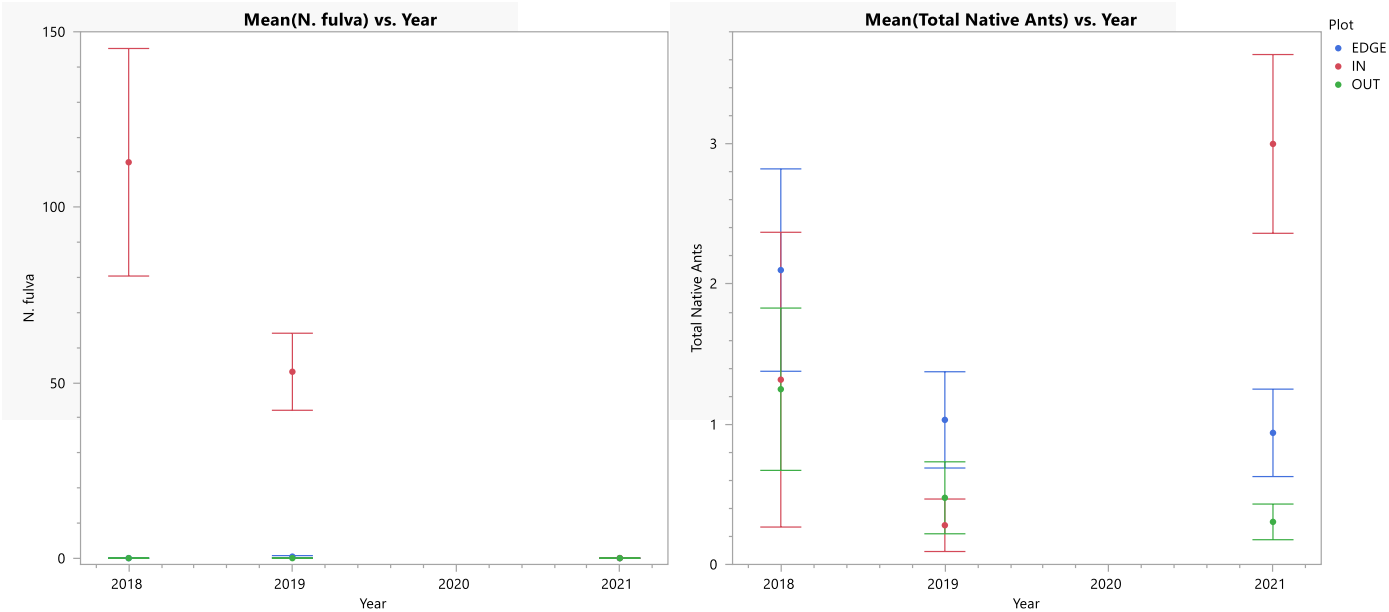
# Canopy Arthropod Study: GCW prey base, direct impact to nestlings



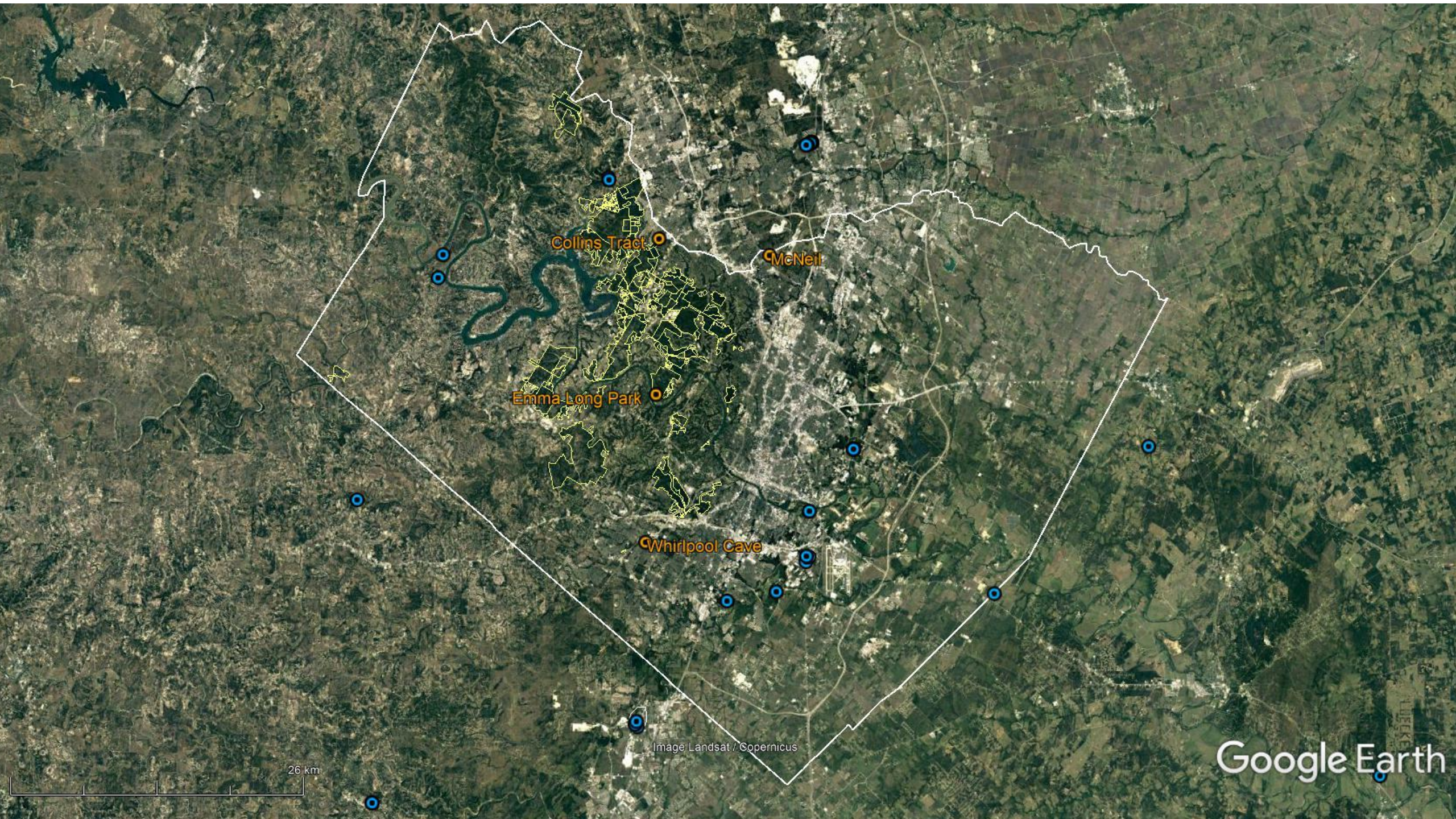
Initiated in 2018 to study how Golden Cheek Warbler prey base responds to TCA invasion.

Lack of impact on edge habitat due to collapse of TCA population.

Processing samples to see if a recovery of prey base is occurring at the longer-term invaded plot.







Collins Track

McNeil

Emma Long Park

Whirlpool Cave

Image Landsat / Copernicus

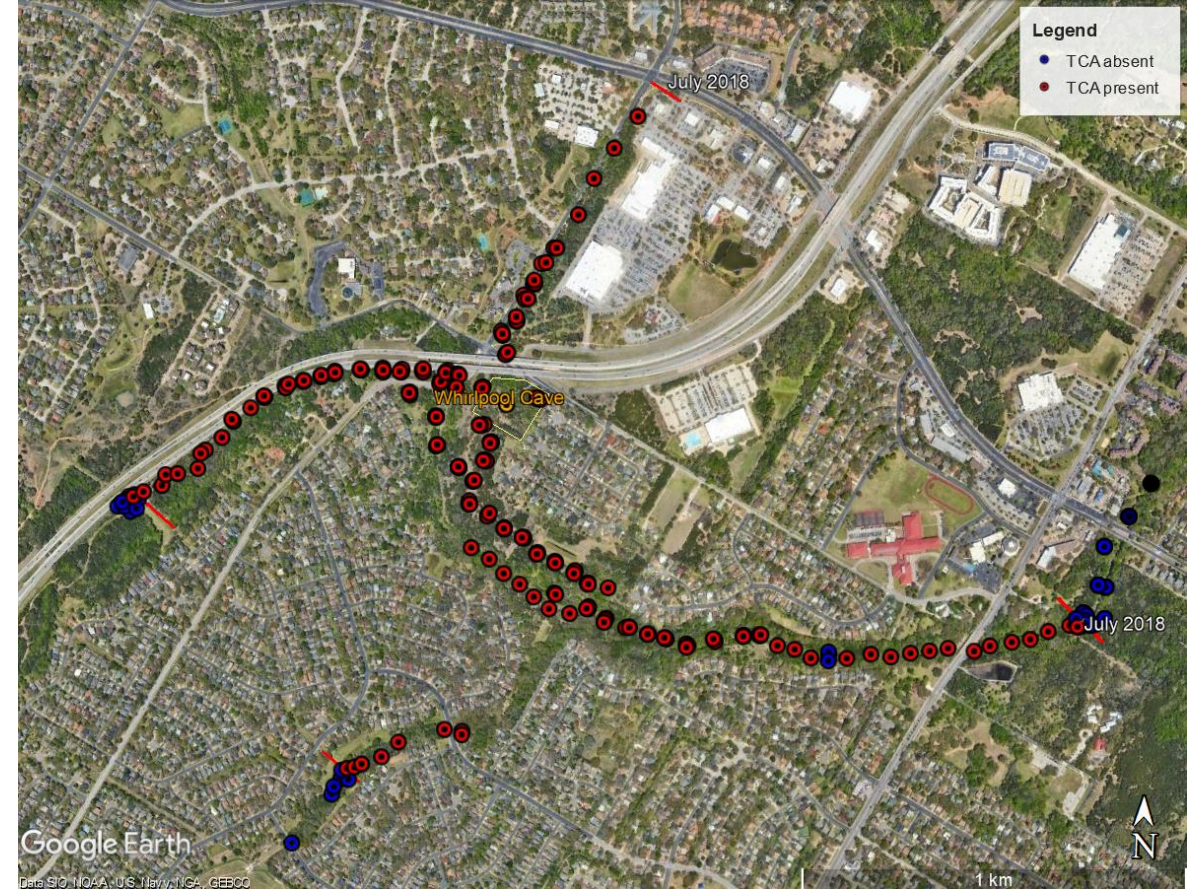
Google Earth

26 km



## Whirlpool Cave

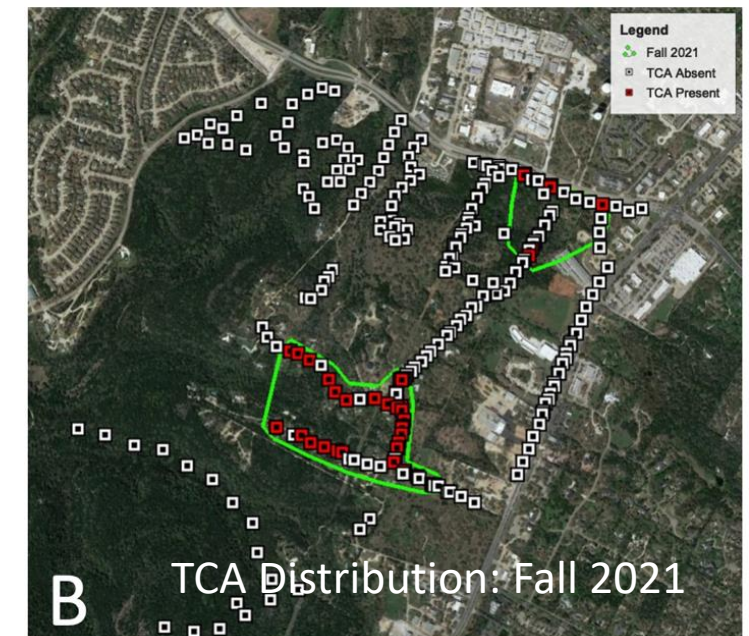
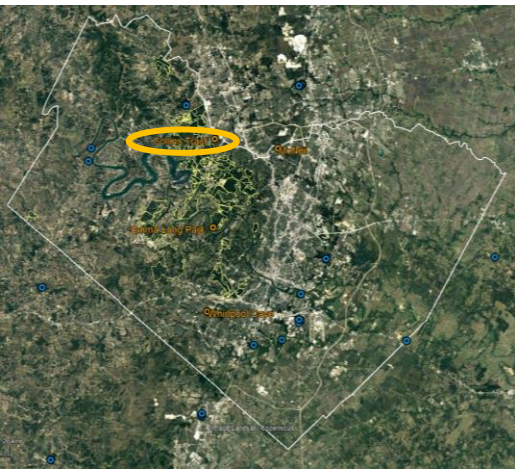
- TCA infestation first reported in July 2013
- Successfully introduced microsporidian into TCA population in Spring 2017.
- Maximum TCA population extent in summer of 2018.
- TCA population locally undetectable by 2020.
- Re-emergent TCA in area, infected and declining.





## Collins Tract:

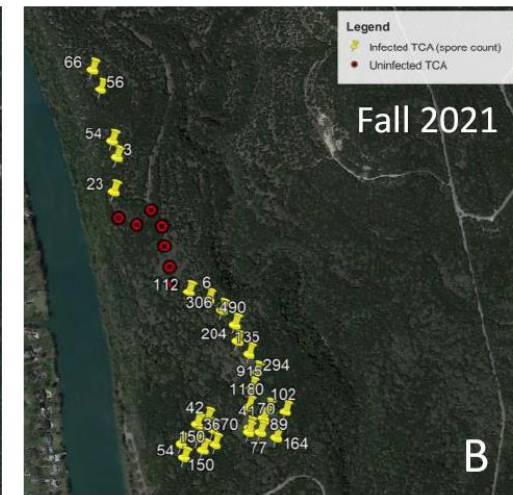
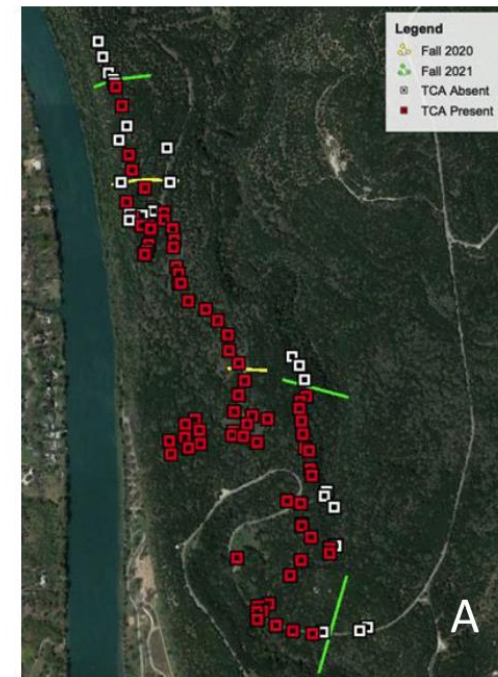
- Tract invaded in 2017
- Mn infection emerged in 2018. Naturally colonized TCA from outside of preserve.
- Inoculation experiments conducted in 2018, and 2019.
- Infection became universal in 2020.
- TCA eliminated from the areas of the preserve they originally invaded by fall of 2021.





## Emma Long Metropolitan Park

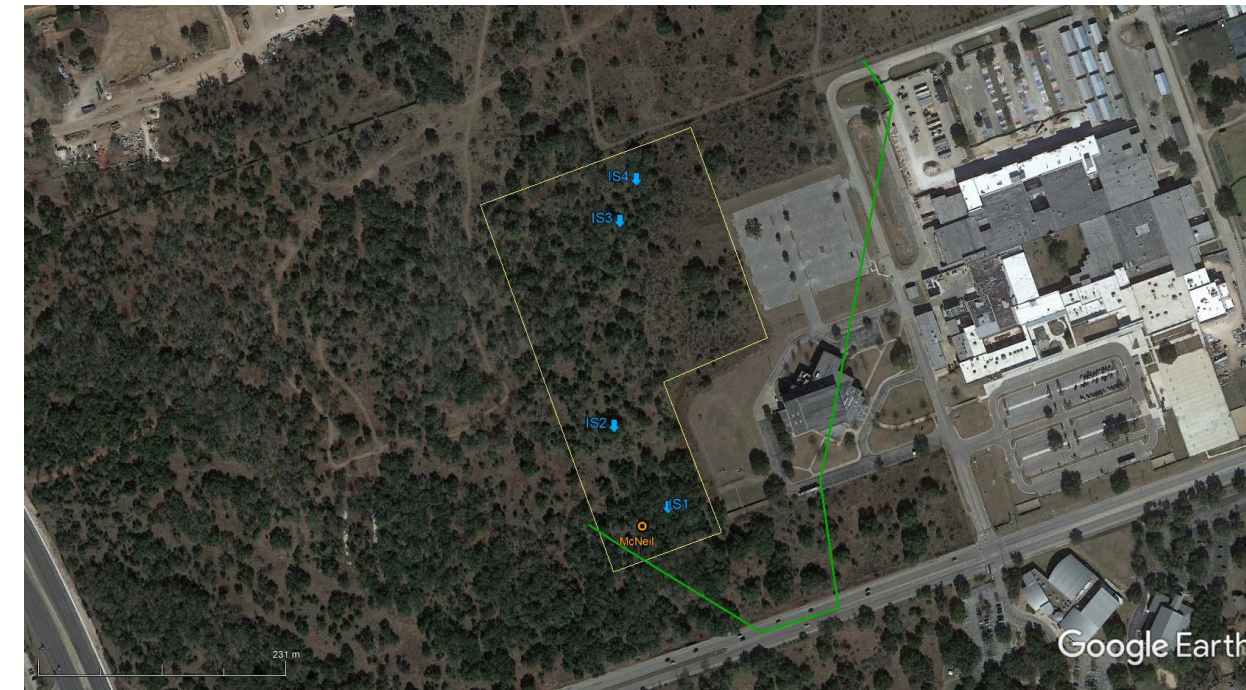
- TCA infestation discovered in July 2020.
- TCA infected with microsporidian at time of first discovery.
- TCA population expanding
- Microsporidian infection is spreading within the TCA and infection was almost universal by fall 2021.





## McNeil

- Reported June 2013
- Uninfected with microsporidia
- Mysterious decline in 2016
- Re-invaded preserve in Spring of 2022
- Microsporidian inoculations in progress.





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