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Ophidiomyces Detection in Free-ranging Snakes on DoD Installations in the U.S. and Puerto Rico

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Ophidiomyces Detection in Free-Ranging Snakes on Department of Defense Installations in the U.S. and Puerto Rico

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Introduction

- Ophidiomycosis (formerly Snake Fungal Disease, or SFD) is an emergent condition caused by the fungus *Ophidiomyces ophiodiicola*
- It is characterized by a range of symptoms including skin lesions, nodules, hyperkeratosis, and scale deformities; and it can damage deeper tissue in muscle and bone
- The disease can be fatal to individuals and may pose a risk at the population level as well

Background

- Ophidiomycosis documented in more than 20 states in over 30 species, including viperids and colubrids
- Many questions still remain including those regarding the current and potential distribution, species affected, and origin of the fungus
- Military lands are home to 131 snake species (Petersen et al. 2018), including several either currently listed or candidates for listing by the USFWS (e.g. Eastern Indigo Snake, Louisiana Pinesnake, Black Pinesnake, Giant Gartersnake, Eastern Massasauga Rattlesnake)
- As such, understanding potential threats to these animals is of conservation interest and may have implications for military activities




Research Goals and Objectives

- Close information gaps in the scientific understanding of this disease
  - Spatial distribution
  - Species affected
  - Environmental conditions
- Raise awareness and train/educate personnel on military installations nationwide
- Prevention of negative impacts to military readiness as a result of degrading ecosystem health

Methods

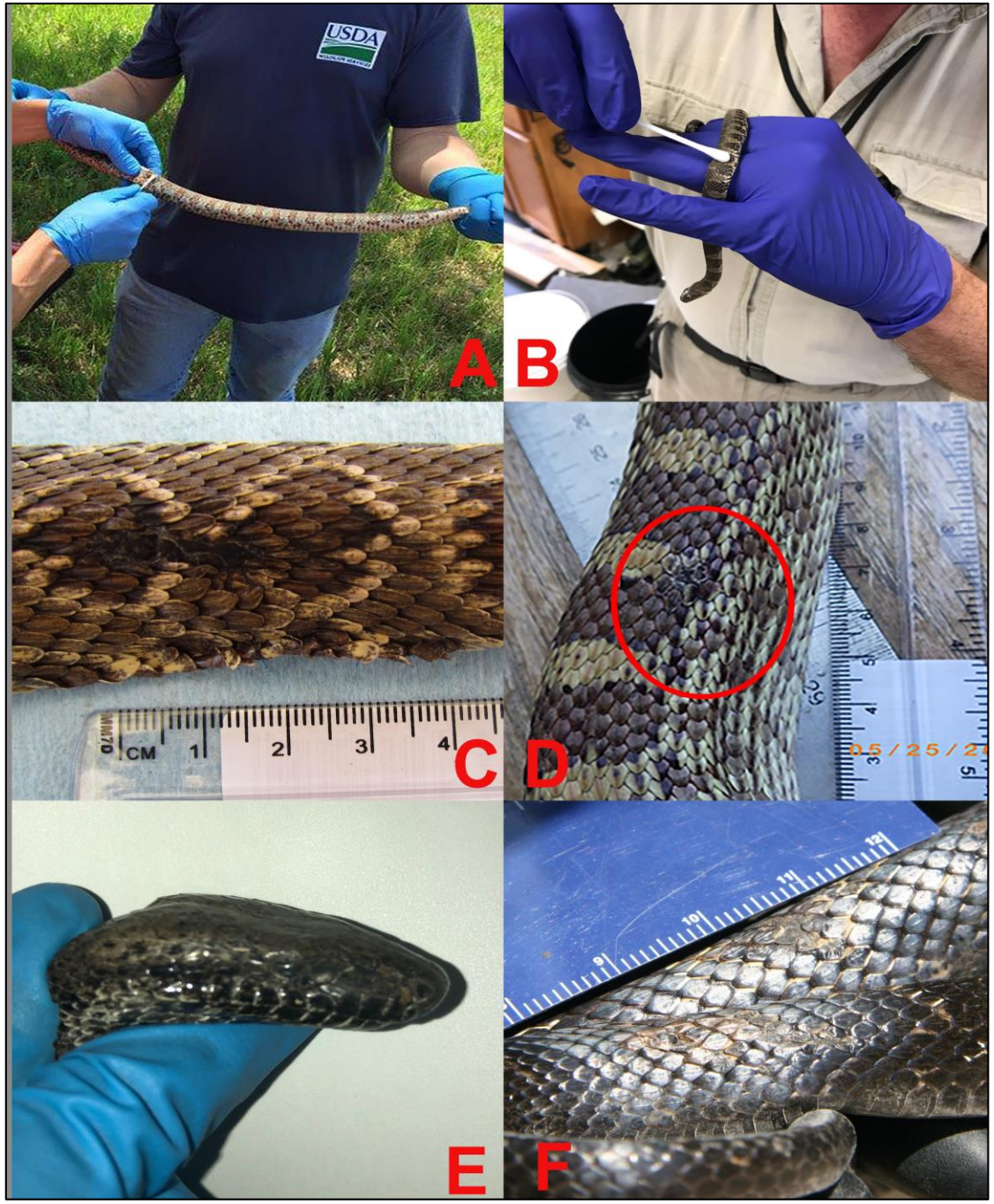
This project provided the training and materials needed to conduct field sampling at their respective installations

- We provided a standardized field datasheet, sampling protocol, and biosecurity procedures
- We provided swabbing kits (swabs, sample tubes, etc.)
- We conducted a training session at 2017 NMFWA meeting and developed an online training video
- **DoD Training Video 2018 - YouTube**
- Participants sent field samples to University of Illinois for analysis
- We provided a summary report and installation-specific report of results

Field Sampling

Photographs of swab sampling **(A,B)** of snakes for detection of Ophidiomyces on military installations in 2018

A ophidiomycosis (Snake fungal disease) lesion in *Crotalus oreganus helleri* **(C),** *Pituophis melanoleucus* **(D),** *Chilabothrus inornatus* **(E)**, and *Pantherophis spiloides* **(F)**



Results

- Kits sent to 68 military installations, 56 (82%) returned results
- 657 individuals from 58 species in 31 states/territories were observed and tested
- 25 species from 19 states/territories were detected with *O. ophiodiicola*

qPCR Results

1) Negative (no clinical signs or qPCR detection of *O. ophiodiicola* DNA; 462 individuals)

2) *Ophidiomyces* present (qPCR detection in absence of clinical signs; 64 individuals)

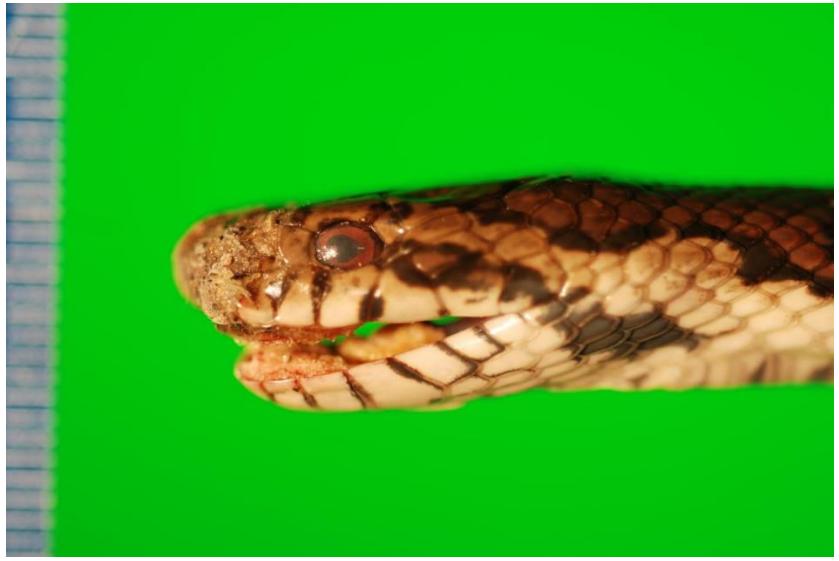
3) Possible ophidiomycosis (presence of clinical signs in absence of qPCR detection; 82 individuals)

4) Apparent ophidiomycosis (presence of clinical signs and qPCR detection; 49 individuals)

Categories based on Baker et al. 2019

Results

- Multinomial multivariable logistic regression indicated adults had 2.38 greater odds of being diagnosed with ophidiomycosis vs. sub-adults
- Snakes from GA, MA, PA, VA had greater odds of being ophidiomycosis diagnosed
- Snakes from ID were less likely to have ophidiomycosis diagnosed
- We report the first detections of *O. ophiodiicola* in ID, OK, and PR



Species Detected With *O. ophiodiicola* in our Study for the First Time

Common Name	Scientific Name	Location
Puerto Rican Boa	*Chilabothrus inornatus*	Puerto Rico
Great Plains Ratsnake	*Pantherophis emoryi*	Kansas
Western Milksnake	*Lampropeltis gentilis*	Kansas
Western Foxsnake	*Pantherophis ramspotti*	Wisconsin

Akaike Information Criteria (AIC) Rankings of the top 5 Logistic Regression Models Predicting Ophidiomycosis Classification in Snakes Sampled on Military Installations

Model	K	AICc	Delta_AICc	AICcWt	Cum.Wt	LL
S+A	33	637.56	0	0.96	0.96	-283.78
S	31	644.62	7.06	0.03	0.99	-289.55
S+A+Sp	59	647.12	9.55	0.01	1	-257.94
S+Sp	57	649.04	11.48	0	1	-261.36
A+Sp	30	656.31	18.75	0	1	-296.51

Sp = species

A = age class

S = state. K=number of parameters

AICc = AIC for small sample sizes

Delta AICc = change in AICc from the top model

AICc weight = proportion of variance in the data that the model explains (ideal =1.0)

Cum. Wt = the cumulative weight of this model and all above it that explains the variance in the data

LL = log likelihood, the natural logarithm of the likelihood of the model (ideal values are lower)

Odds Ratios for States Significantly Associated With Ophidiomycosis Sampled in Different States

State	Odds ratio	95% CI	p value
Georgia	5.28	1.31-21.51	0.027
Massachusetts	6.0	1.26-28.55	0.041
Pennsylvania	8.75	1.76-43.6	0.009
Virginia	3.64	0.91-14.61	0.033

CI = Confidence interval.

AIC Rankings of the top 5 Logistic Regression Models Predicting Ophidiomycosis Classification Detection in Snake Species

Model	K	AICc	Delta_AICc	AICcWt	Cum.Wt	LL
I + A	58	636.08	0	0.96	0.96	-253.65
I	56	642.53	6.45	0.04	1	-259.33
A + Sp	30	656.31	20.23	0	1	-296.51
I + A + Sp	83	660.5	24.42	0	1	-233.61
I + Sp	81	662.22	26.14	0	1	-237.16

Sp = species

A = age class

I = installation

K = number of parameters

AICc = AIC for small sample sizes

Delta AICc = change in AICc from the top model

AICc weight = proportion of variance in the data that the model explains (ideal =1.0)

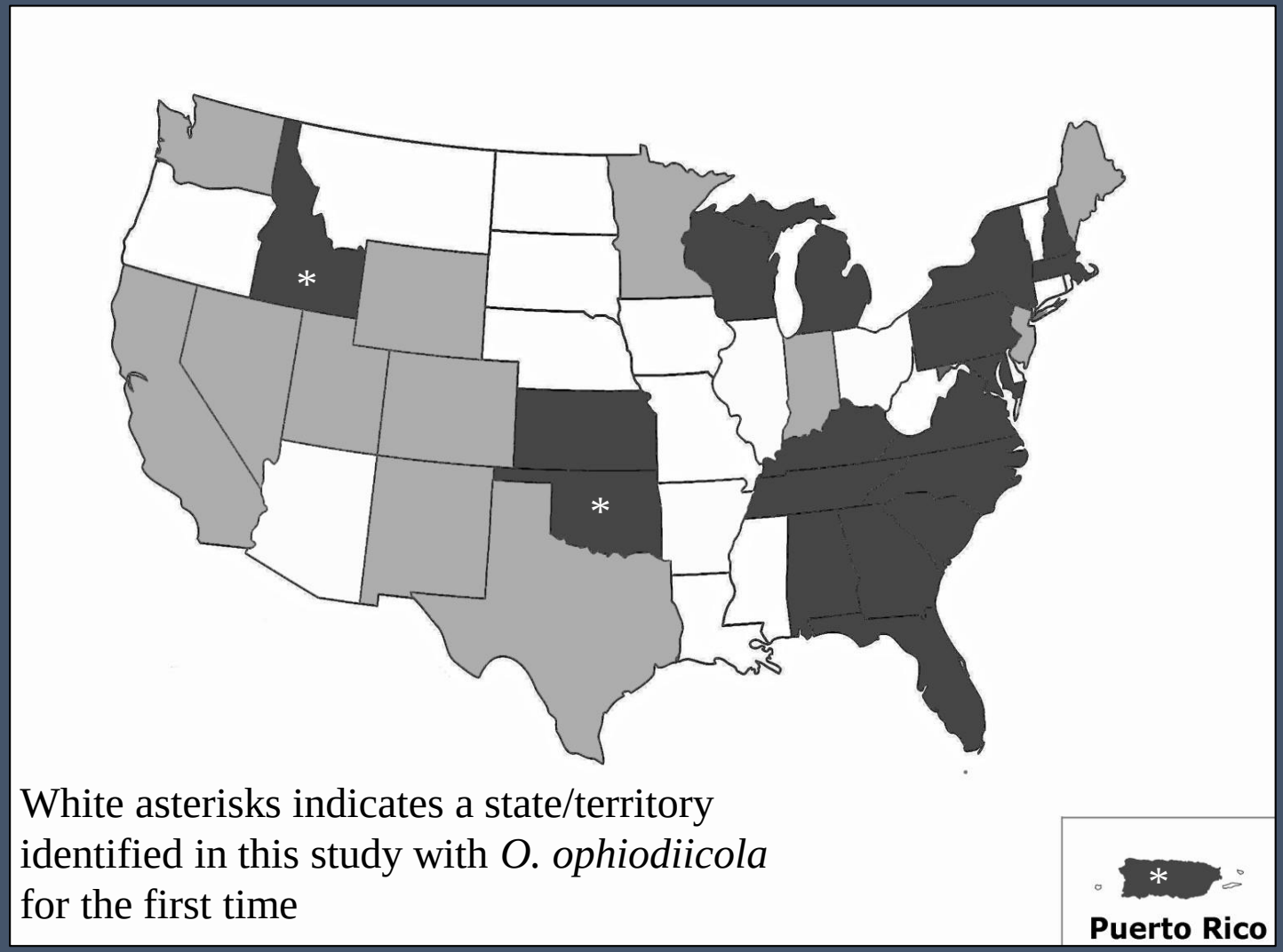
Cum. Wt = the cumulative weight of this model and all above it that explains the variance in the data

LL = log likelihood, the natural logarithm of the likelihood of the model (ideal values are lower)

Spatial Distribution of *O. ophiodiicola* Detection in Snakes on Military Installations Sampled in 2018

Map Legend

White = states not sampled
Light Grey = states with no detection of *O. ophiodiicola*,
Dark Grey = states detected with *O. ophiodiicola*



White asterisks indicates a state/territory identified in this study with *O. ophiodiicola* for the first time

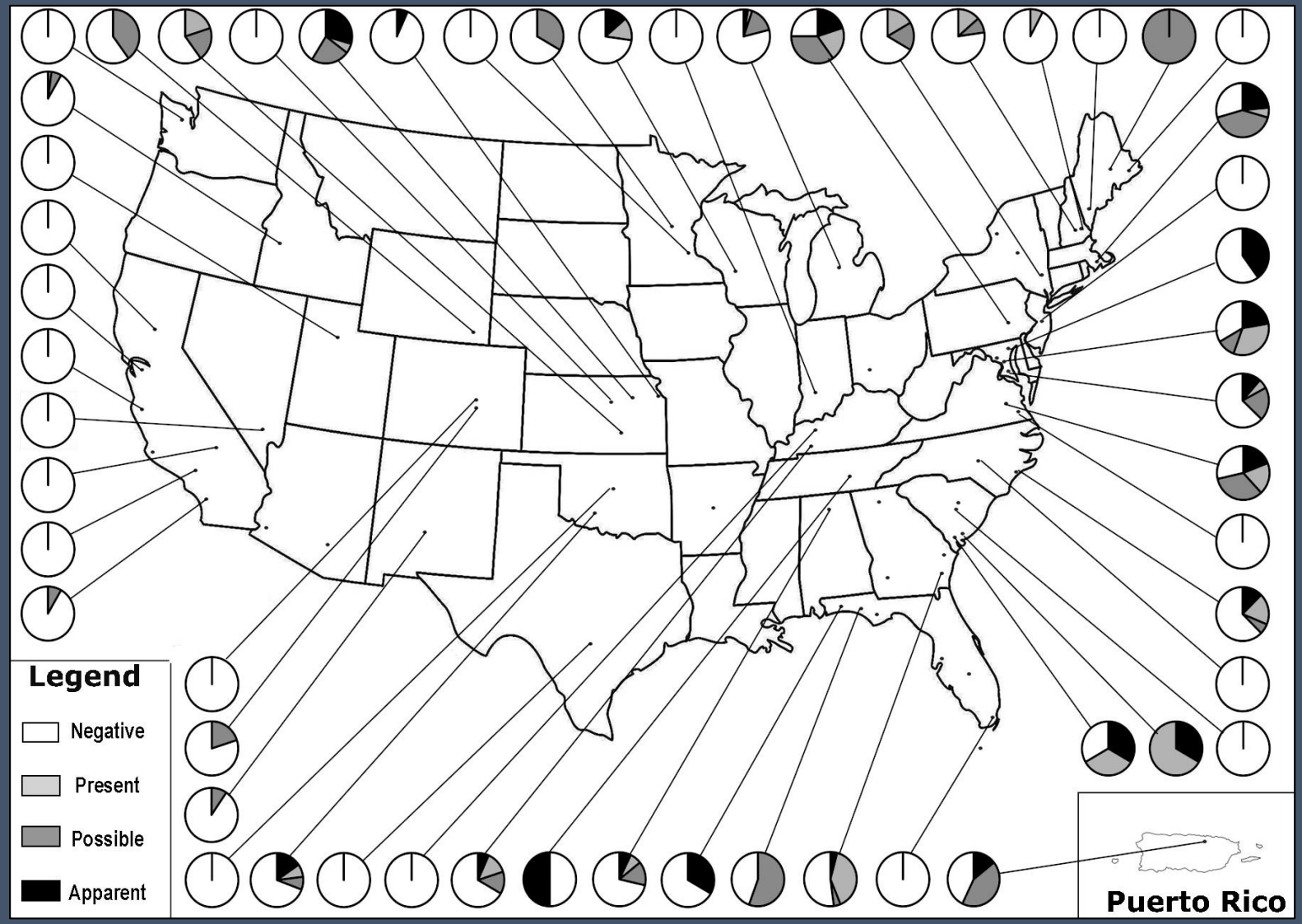
Spatial Distribution and Prevalence of Ophidiomycosis in Snakes on Military Installations Sampled in 2018

Negative = no clinical signs and no qPCR detection of *Ophidiomyces ophiodiicola*

Present = qPCR positive AND no clinical signs

Possible = clinical signs present AND no qPCR detection

Apparent = qPCR detection AND clinical signs



Phylogenetic Analysis of Snake Species Detected with *Ophidiomyces ophiodiicola* Using qPCR From Sampling on Military Installations in 2018

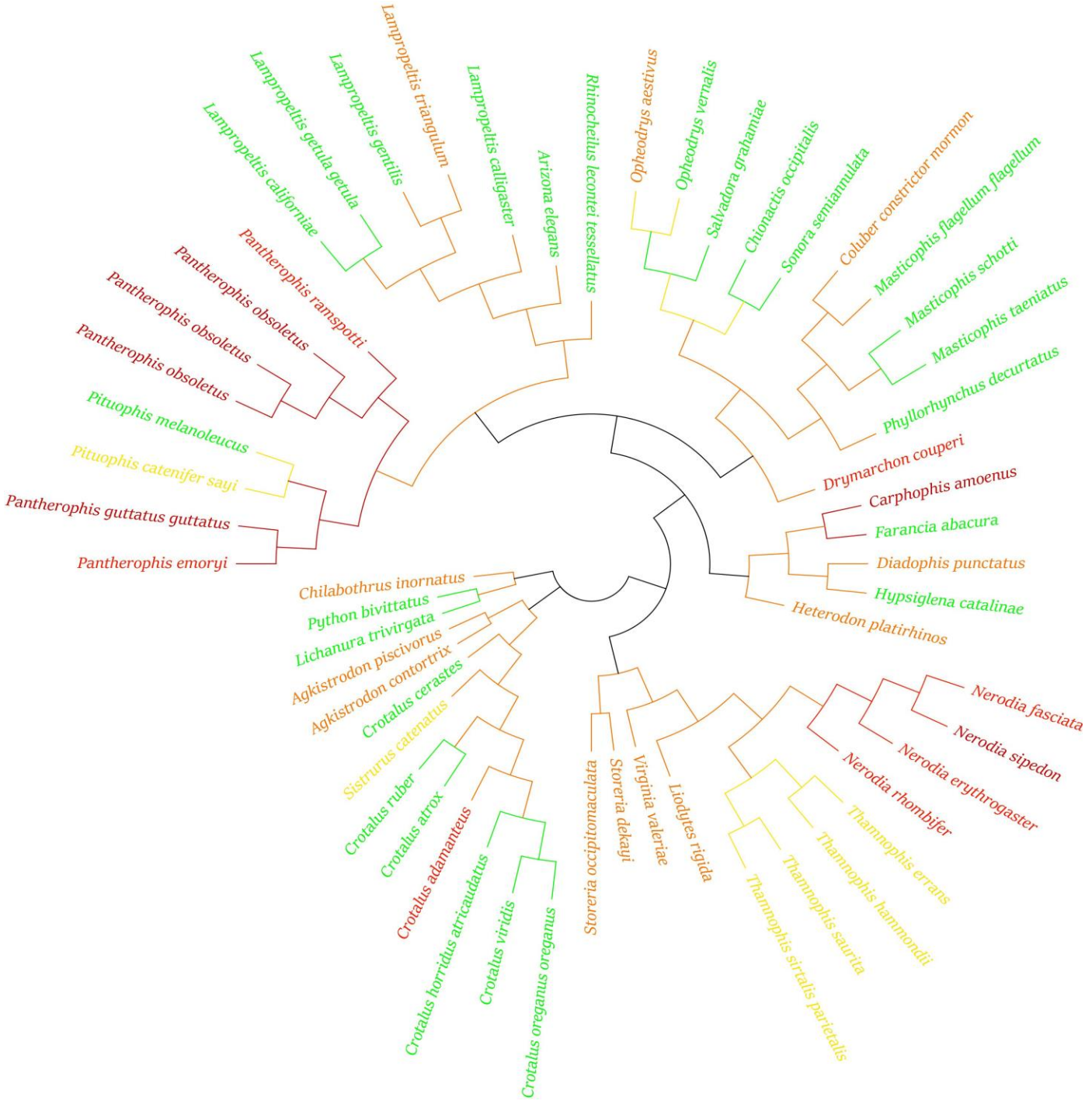
Green = 0% prevalence

Yellow = 0-9.9%

Light Orange = 10-24.9%

Dark Orange = 25-49.9%

Red = >50%



Summary

- *O. ophiodiicola* is relatively widespread on military installations
- 657 individuals from 58 species in 31 states/territories were observed and tested
- 25 species from 19 states/territories were detected with *O. ophiodiicola*
- We report the first detections of *O. ophiodiicola* in ID, OK, and PR
- Lots of work to be done to further research *O. ophiodiicola* , develop monitoring and handling protocols, and educational materials both on, and off military lands

Questions?

Prevalence Percentages of Ophidiomycosis Classifications in Snake Species Sampled

Variable		Total n	Negative *Ophidiomyces* present	Possible ophidiomycosis	Apparent ophidiomycosis
Age class					
	Unknown	106	75.5%	6.6%	16.0%	1.9%
	Juvenile	121	82.6%	9.9%	6.6%	0.8%
Adult		430	65.6%	10.5%	13.3%	10.7%
Lesions					
	No	526	87.8%	12.2%	0%	0%
	Yes	49	0%	0%	62.6%	37.4%