

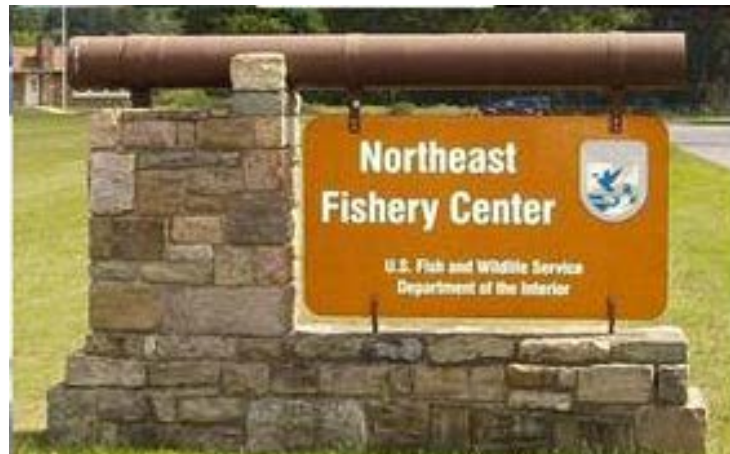
Survey of Amphibian Emergent Infectious Diseases in Mitigated and Reference Wetlands

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Study Objectives

1. Determine the status and extent of *ranavirus* in Pennsylvania's amphibian populations
2. Determine the status and extent of chytrid fungus *Batrachochytrium dendrobatidis* (*Bd*) in amphibian populations
3. Examine the relationship between disease parameters and wetland condition



Ranavirus

- Hemorrhaging of skin and organs
- Mortality in larvae and adults
- Spread via water, contact, scavenging
- Mortality events in >20 species turtle and amphibian
- Infects fish, amphibians, and reptiles



Chytrid Fungus (*Bd*)

- Hyperkeratosis - impaired electrolyte exchange, excess sloughing
- Mortality in post-metamorphic individuals
- Contracted by >350 species
- Declines in >200 species
- Species range in susceptibility to pathogenic effects

Disease-related responses and predictors

Responses

- Occurrence
 - % infected populations
- Prevalence
 - % individuals infected
- Intensity
 - Zoospores (spores) per infected individual

Predictors

- PA Ecoregions
- Wetland condition
 - Reference vs Mitigated
 - % Forest within 1km
 - Level 1 Assessment
 - Anthropogenic stressors
 - Level 2 Assessment

Green frog

Lithobates clamitans melanotus

- State-wide distribution, abundant populations, generalist species
- Breed in permanent and semi-permanent bodies of water
 - Tadpoles must over-winter
 - Co-inhabit ponds with fish
- Higher tolerance to chytrid loads
- Metamorphs and juveniles “visit” seasonal wetlands



2013 Sampling Season

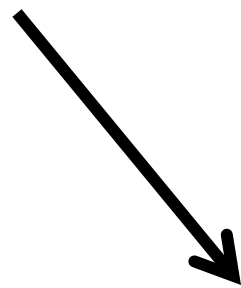
- 20 wetlands
- Tadpoles sampled per wetland
 - N= 60 for 70% of all wetlands
 - Mean = 52, Min 13

	Pennsylvania Ecoregion			
	Allegheny Plateau	Glaciated	Piedmont	Ridge and Valley
Mitigated	3	3	1	3
Reference	2	4	1	3

Pathogen Screening Techniques

Bd Screening

Ranavirus Screening



Tissue Culture

Quantitative PCR (qPCR)



Bd prevalence and *Ranavirus* presence

● Reference

● Mitigation

+ *Ranavirus* present



• *Bd* absent

○ 5-10% w/*Bd*

○ >50% w/*Bd*

Bd intensity and *Ranavirus* presence

● Reference

● Mitigation

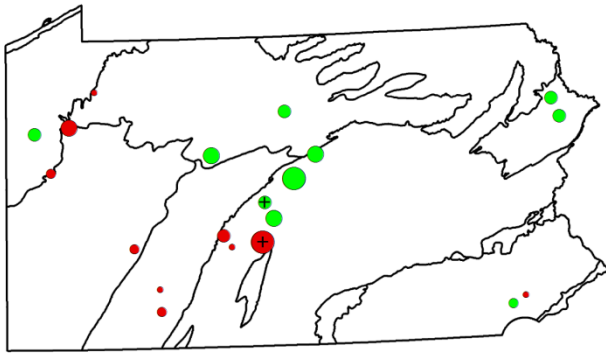
+ *Ranavirus* present



• *Bd* absent

○ 50-200 spores

○ >1000 spores



Allegheny
Plateau
(N = 5)

Glaciated
Allegheny
(N = 5)

Glaciated
Poconos
(N = 2)

Piedmont
(N = 2)

Ridge &
Valley
(N = 6)

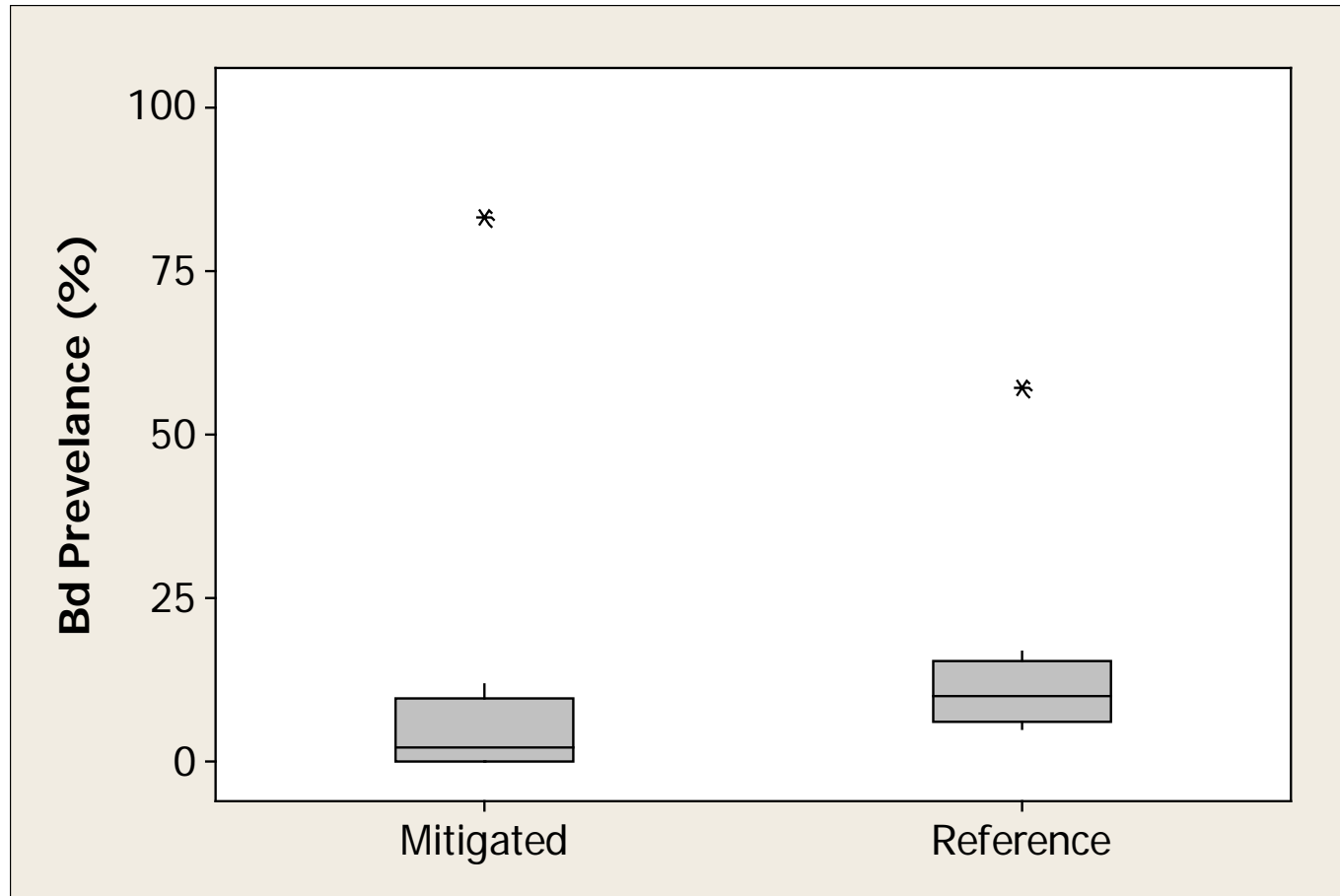
Chytrid fungus (<i>Bd</i>)	Occurrence	80%	80%	100%	50%	83%
	Prevalence*	7.8%	10.3%	8.9%	5.0%	33.5%
	Intensity	346 ±87	764 ±512	177 ±39	358 ±na	177 ±39
<i>Rana- virus</i>	Occurrence	0%	0%	0%	0%	33.3%
	Prevalence*	-	-	-	-	5.9%

*mean values among disease-present sites

		Mitigated Wetlands (N = 10)	Reference Wetlands (N = 10)
Chytrid fungus (<i>Bd</i>)	Occurrence	60%	100%
	Prevalence*	18.9%	14.5%
	Intensity	585 ±341	279 ±64
<i>Ranavirus</i>	Occurrence	10%	10%
	Prevalence	1.7%	8.5%

*mean values among disease-present sites

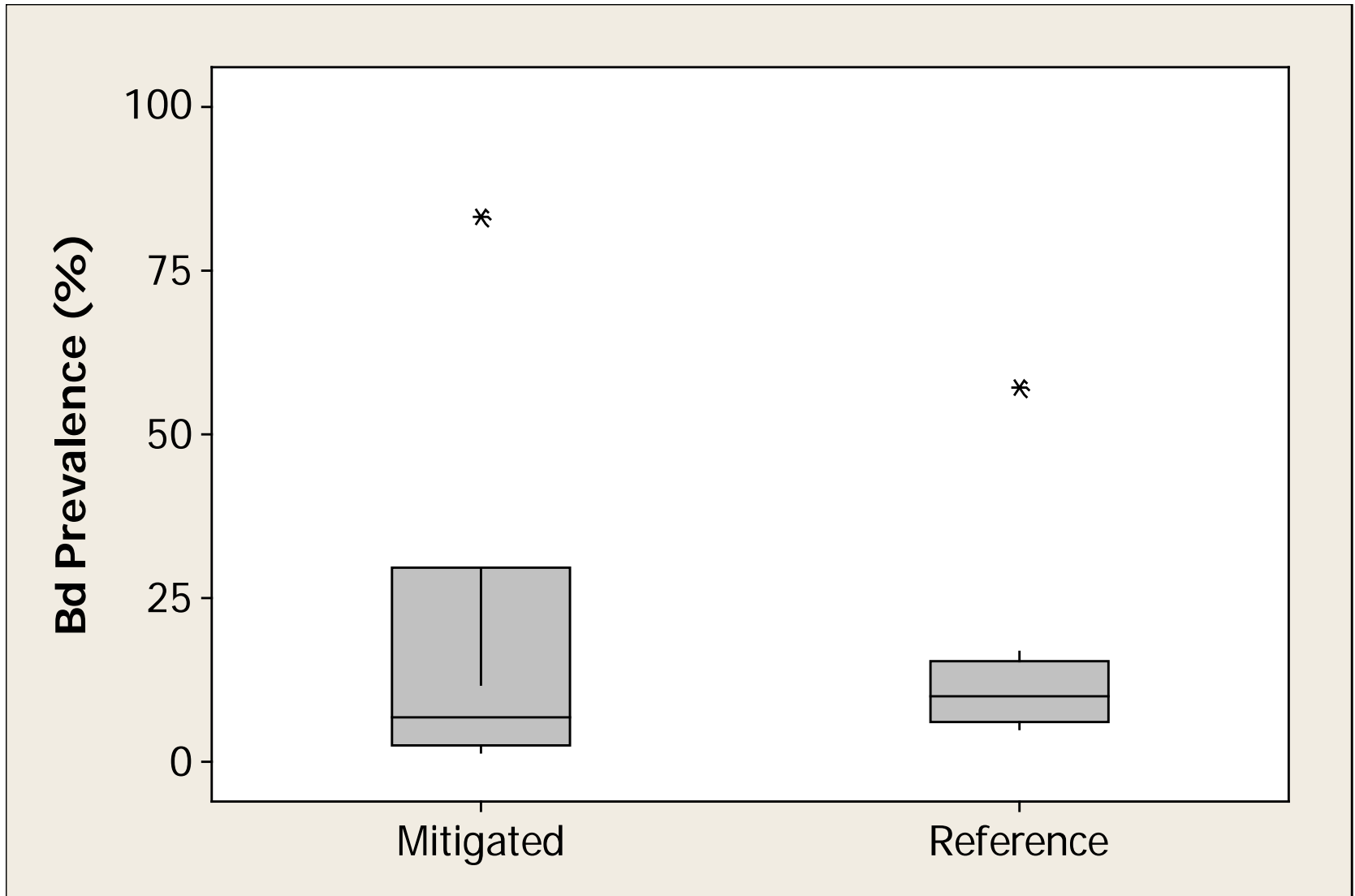
Mean *Bd* prevalence for all populations



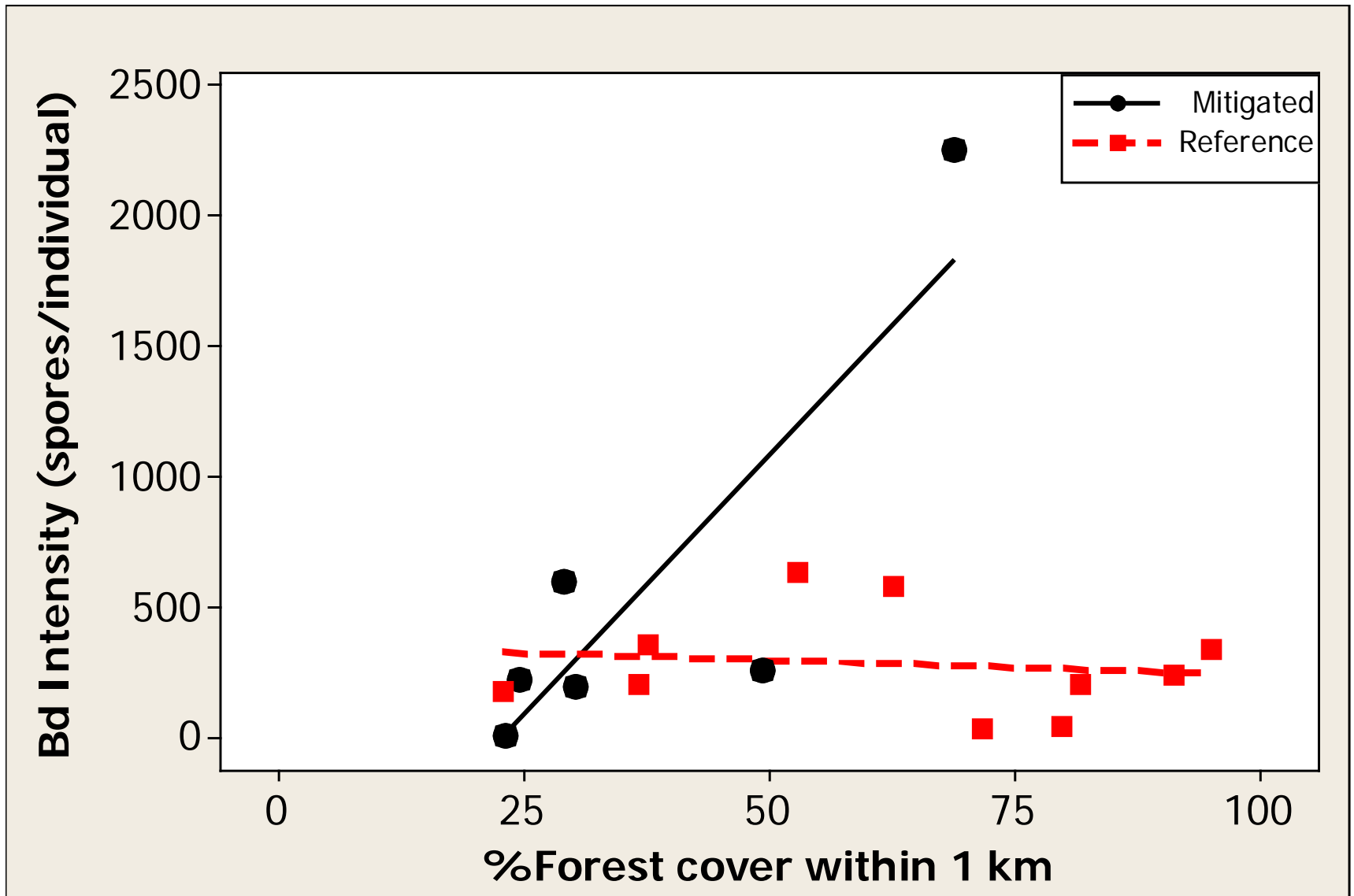
Kruskal-Wallis Test using ranked prevalence values

$H = 4.68$, $DF = 1$, $P\text{-value} = 0.03$

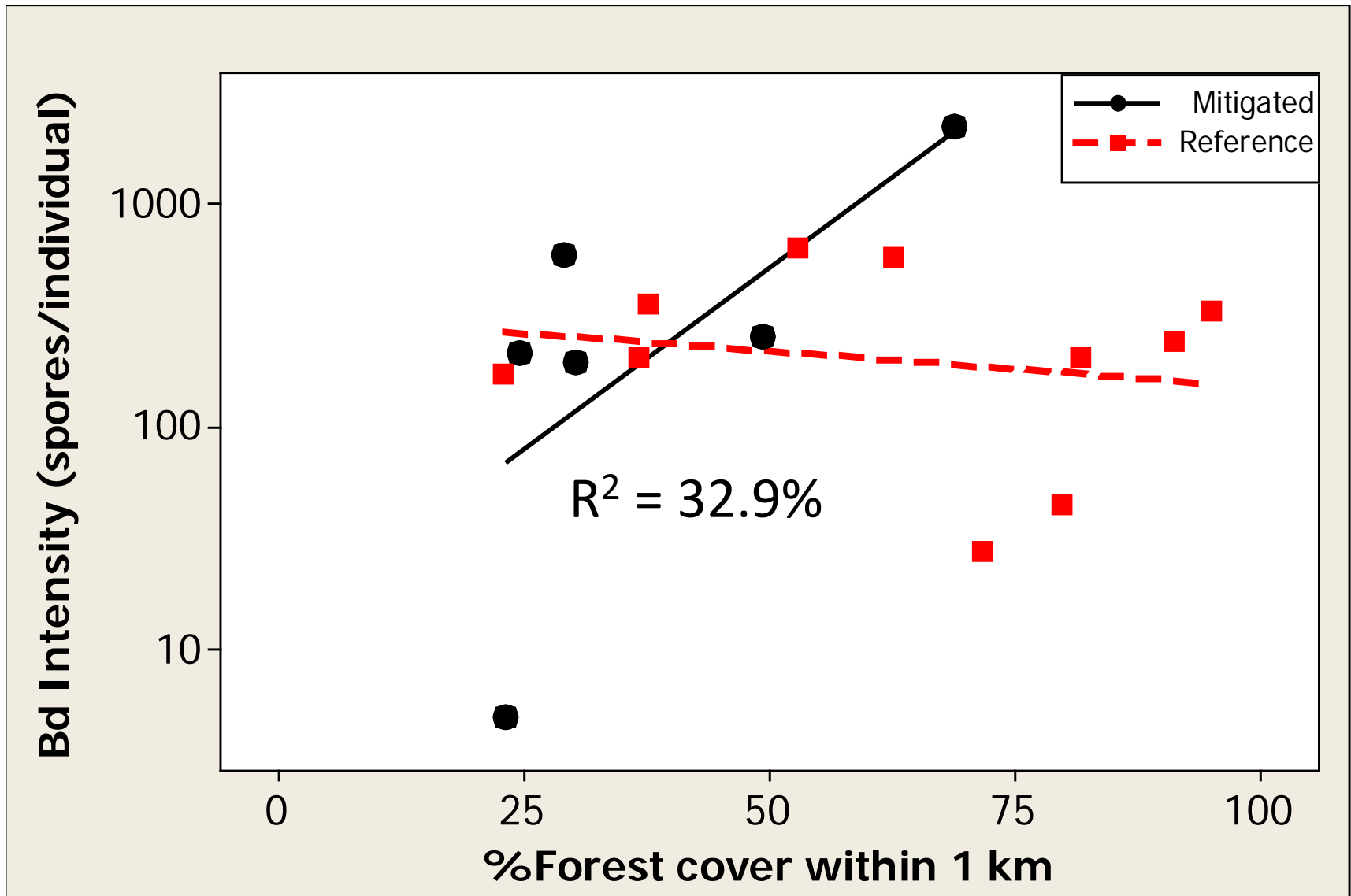
Mean prevalence for *Bd*-positive populations



Bd intensity as a function of an interaction between forested landscape and wetland type



Bd intensity as a function of an interaction between forested landscape and wetland type



Conclusions

- ***Ranavirus occurred*** only in Ridge and Valley, and in low prevalence
- ***Bd occurred*** in ALL reference wetlands and most of mitigated wetlands
- ***Bd prevalence*** has a higher range in mitigated wetlands than reference wetlands
- ***Bd intensity*** may be (+) correlated with forest cover among mitigated wetlands

Future Directions

- 2014 Sampling to target Piedmont region and mitigated wetlands with higher % forest
- Use Level 2 Assessments of wetlands for anthropogenic stressors
- Amphibian community influences on *Bd* prevalence and intensity
- Differences in prevalence among tadpole cohorts