

# Big Brown Bat (*Eptesicus fuscus*) Mass Changes During White-Nose Syndrome Invasion

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## EEB313: Quantitative Methods in R for Biology

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### Abstract

This study analyzes mass changes in Big Brown Bats (*Eptesicus fuscus*) exposed to White-Nose Syndrome (*Pseudogymnoascus destructans*). Using 30 years of data, we applied a Generalized Linear Mixed Model (GLMM) with a Gamma distribution to assess predictors like sex, pregnancy status, and disease stage. Results showed significant site-specific variability, with males weighing 87.3% of females and pregnant bats having higher mass. Post-invasion, slight log-mass declines were observed, especially in pregnant bats. These findings emphasize the need to refine analytical methods to better understand WNS impacts.

### 1.0 Introduction

Environmental, physiological and behavioral factors multiply the complexities of host immune responses to pathogens. White Nose Syndrome (WNS), a disease caused by the fungal pathogen *Pseudogymnoascus destructans* (*Pd*), has impacted the health of hibernating North American bat species, such as the Big Brown Bat (*Eptesicus fuscus*), with an ongoing invasion originating from 2006 (Simonis et al., 2023; Davy et al., 2017). North American bats are heterothermic and are vulnerable during hibernation periods when their immune function is suppressed, and since *Pd* is a cryophilic fungus, it thrives in winter hibernation sites, and upon infection leaves an uncomfortable white fuzz on the epithelial tissues of bats, causing arousal that prematurely burns the bats stored brown fat and water leading to associated losses in mass (Simonis et al., 2023; Davy et al., 2017). The energy expenditure of *Pd* immune responses decreases available energy for emergent seasonal behaviors such as migration to summer hibernacula and initiation of reproduction; with differing energy necessities between adult and juvenile male and female bats. WNS often leads to the mortality of smaller species, such as the 95% mortality rate of the Little Brown Bat (*Myotis lucifugus*), yet, recovered individuals show carry-over impacts in the larger context of bat seasonal and reproductive behaviors and physiological responses (Davy et al., 2017). Species such as *E. fuscus* are integral in understanding mass changes over an exposure period, as they are more likely to survive and exist over a large latitudinal gradient with expected mass differences given Bergmann's rule (Simonis et al., 2023). Previous research by Simonis and colleagues (2023) used linear-mixed effects models to quantify the changes in Big Brown Bat mass over the invasion period of WNS. Yet, in this study we show their model could be improved upon.

Our null hypothesis followed that age and sex are not correlated with the effect of *Pd* on the mass of big brown bats. Our alternative hypothesis followed that age and sex are correlated with the effect of *Pd* on the mass of big brown bats. If the change in mass pre- and post-invasion of *Pd* remains constant throughout the different age and sex groups, the null hypothesis would be accepted. Otherwise, we would reject the null hypothesis.

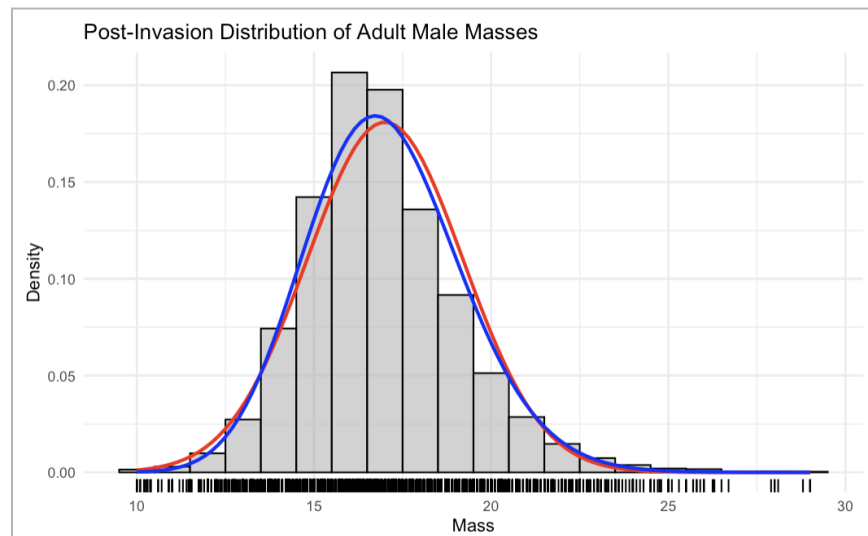
### 2.0 Methods

#### 2.1 Data Description

This dataset originates from Big brown bat (*E. fuscus*) capture records from researchers and wildlife agencies over 30 years, from 1990-2020. The data contains 30,497 individual records across 3,797 unique capture sites across the eastern United States, including both juvenile and adult bats across different reproductive stages.

## 2.2 Data Analysis

We started with data wrangling, simplifying the reproductive status and disease time step into two categories each: pregnant/non-pregnant and pre-/post-invasion. We grouped the bats by different combinations of sex, age, and reproductive status, such as male juvenile, female adult non-pregnant, etc., and plotted the raw mass distribution of each group on histograms with rug plots. The mass distributions were close to normal, but slightly positively skewed with more data on the higher end of the mass range, suggesting alternative distributions might better capture the variability in the data.



**Figure 1:** Distribution modeled using a **normal distribution (red)** and **gamma distribution (blue)** for Post-Invasion Adult Male Masses.

Our analysis involved multiple iterations. Initially, we assumed a Gamma distribution and estimated parameters using maximum likelihood. However, this was difficult to execute without a specific package in R, which led to a shift in our approach. Our second attempt was to explore other distributions, trying to model our data on as many distributions as we could and compare the maximum likelihood estimators. However, we ultimately found that the data was best described by gamma distribution. Building on the original study, which used linear mixed models with normal distributions, we applied generalized linear mixed models (GLMMs) with a Gamma distribution using the `glmer()` function from the **lme4** package in R. We essentially conducted a variation of the original analysis with a different error distribution to check that the relationship between the variables is also best described by a gamma distribution.

The final GLMM with a Gamma family and log link function analyzed bat mass in relation to sex, pregnancy status, and disease group, with site-specific variability accounted for using a random intercept for `site_mask`. Elevation was considered but excluded due to data limitations. To determine the most suitable random effect structure, we tested three models with different random intercepts: year, `site_mask`, and state. The Akaike Information Criterion (AIC) was used to evaluate model fit, with a lower AIC indicating a better balance between accuracy and complexity. Each model was fitted to assess how temporal, site-specific, and regional variability influenced bat mass.

### 3.0 Results

Model	Random Effect	AIC
Model 1	year	138844.5
Model 2	site_mask	132567.2
Model 3	state	137431.1

In determining the most suitable random effect structure (Table 2), **Model 2** with a random intercept for site\_mask had the lowest AIC (132,567.2) compared to **Model 1** (year, AIC = 138,844.5) and **Model 3** (state, AIC = 137,431.1). These results identify site-specific spatial variability as the most significant factor in explaining bat mass differences, aligning with

**Table 1: Model Comparison** of random effects findings from the original study (Simonis et al., 2023).

The Summary of the Generalized Linear Mixed Model (GLMM) in Figure 2 showed a random effect variance of 0.003733 and a standard deviation of 0.0611, indicating low site-specific variability in bat mass. The intercept estimate of 2.937757 represents the baseline log-mass for non-pregnant female bats post-invasion, corresponding to a mass of approximately 18.86. The fixed effects showed that male bats had a significantly lower log-mass than females, with a difference of 0.1347 units ( $p < 2e-16$ ), indicating males weighed 87.3% of female mass when determined through exponentiation. Pregnant females had higher log-mass than non-pregnant females (0.1726 units,  $p < 2e-16$ ), or 118.8% of non-pregnant female mass. A slight log-mass decline was observed post-invasion, with pregnant bats being marginally more affected during the pre-invasion period (-0.0264 units,  $p = 0.0163$ ). The interaction between sex (male) and disease group (pre-invasion) was not significant ( $p = 0.454$ ), indicating no notable differential effect of invasion status on males compared to females.

The interaction plot (Figure 3), created using ggplot2, showed two time periods ("Pre-Invasion" and "Post-Invasion") on the x-axis and predicted log-mass on the y-axis, faceted by pregnancy status (columns) and sex (rows). The plot showed that pregnant bats had higher predicted log-mass than non-pregnant bats, and females generally showed higher log-mass than males, particularly among pregnant individuals. Across all groups, there was a slight decline in log-mass from pre-invasion to post-invasion, with the decline being less significant in pregnant bats. Random effects, represented by colored lines, captured site-specific variability, with some sites showing minimal change and others more noticeable shifts.

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Generalized linear mixed model fit by maximum likelihood (Laplace Approximation) ['glmerMod']
Family: Gamma ( log )
Formula: mass ~ sex * pregnancy_status * disease_group + (1 | site_mask)
Data: data_new

      AIC      BIC   logLik deviance df.resid
139176.3 139242.9 -69580.2 139160.3    30488

Scaled residuals:
      Min       1Q   Median       3Q      Max
-4.6609 -0.6113 -0.0060  0.6144  6.4627

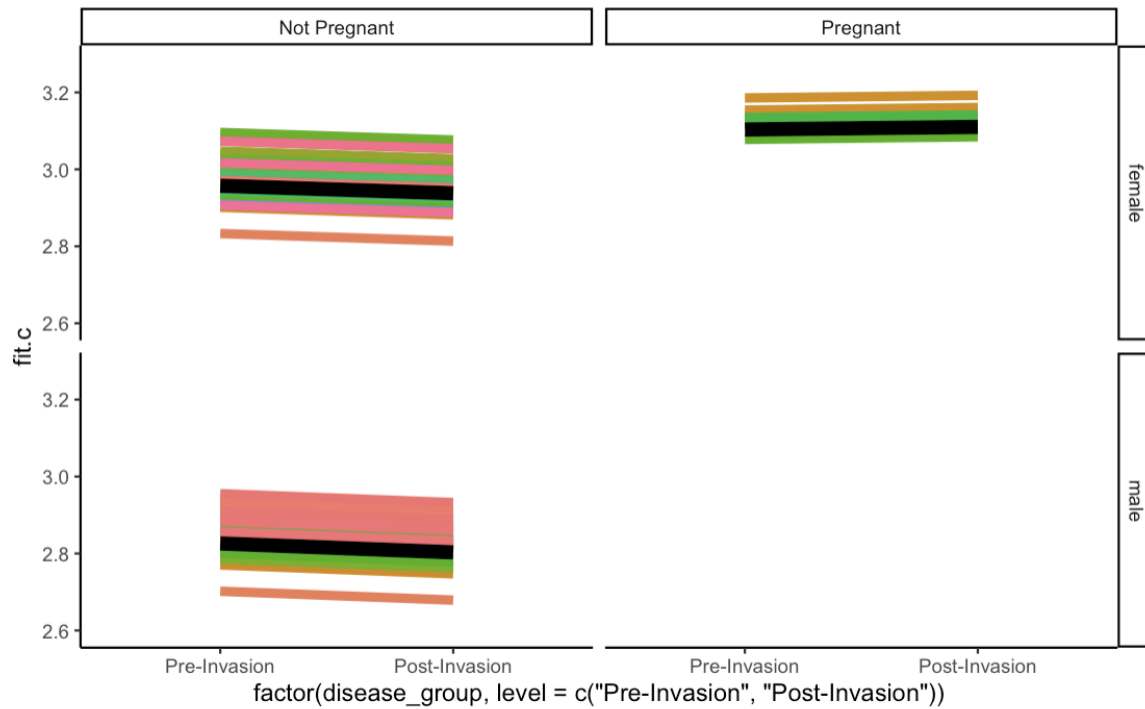
Random effects:
Groups   Name      Variance Std.Dev.
site_mask (Intercept) 0.003733 0.0611
Residual              0.016579 0.1288
Number of obs: 30496, groups: site_mask, 3797

Fixed effects:
                                Estimate Std. Error t value Pr(>|z|)
(Intercept)                    2.937757    0.002520 1165.905 < 2e-16 ***
sexmale                        -0.134676    0.001767  -76.215 < 2e-16 ***
pregnancy_statusPregnant        0.172564    0.003925  43.970 < 2e-16 ***
disease_groupPre-Invasion        0.019582    0.005182   3.779 0.000158 ***
sexmale:disease_groupPre-Invasion 0.003668    0.004901   0.748 0.454187
pregnancy_statusPregnant:disease_groupPre-Invasion -0.026366    0.010979  -2.401 0.016329 *
---
Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Correlation of Fixed Effects:
      (Intr) sexmal prgn_P ds_P-I s:_P-I
sexmale      -0.373
prgnncy_stP  -0.203  0.229
dss_grpPr-I  -0.345  0.179  0.116
sxml:ds_P-I   0.127 -0.357 -0.081 -0.514
prgn_P:_P-I   0.068 -0.084 -0.359 -0.275  0.233
fit warnings:
fixed-effect model matrix is rank deficient so dropping 2 columns / coefficients

```

**Figure 2:** Summary of predicted log-mass by disease group, sex, and pregnancy status.



**Figure 3:** Interaction plot of predicted log-mass by disease group, sex, and pregnancy status.

#### 4.0 Discussion

Through our analysis, we rejected the null hypothesis that age and sex are not correlated with the effect of *Pd* on mass and demonstrated that the Gamma distribution better represents the biological reality of body mass by effectively modeling strictly positive, variable, and skewed data. Comparisons between LMM and GLMM validated the Gamma distribution's suitability, as LMM assumes normality, which is unsuitable for this study (Lo & Andrews, 2015). Since *E. fuscus* shows resilience to WNS due to its size, measurable mass changes can be leveraged to better understand host responses, where the Gamma distribution fits the nature of mass-changes (Simonis et al., 2023).

The GLMM analysis showed low variance (0.003733) in site-specific variability, indicating that most variation in bat mass is explained by fixed effects. Fixed effects showed males weighed less than females, likely due to females foraging up to 100% of their body weight in insects to support gestation (Hood et al., 2006). The site-specific model better captures localized effects, while the state-specific model, with lower variability (0.0001), is better suited for broader patterns. Post-invasion declines in bat mass support the hypothesis that pathogen exposure causes energetic stress (Davy et al., 2017). Interaction plots highlighted site-specific mass changes, emphasizing local environmental factors, likely due to the site plasticity of pre-hibernating bats (Jung & Kalko, 2010).

The model had limitations, including the removal of two fixed-effects columns due to collinearity. While pregnancy status and disease group showed slight variations in invasion impact, no interaction between sex and disease group indicated similar effects on males and females. These results show site-specific spatial variability as the key factor in explaining bat mass differences, aligning with the original study (Simonis et al., 2023). A slight post-invasion decline in log-mass was observed, with pregnant bats marginally more affected during the

pre-invasion period (-0.0264 units,  $p = 0.0163$ ). This study adds to the large body of research to improve analytical methods in WNS invasion research specific to mass changes, a large part impacting mortality as well as measuring changes in a complex environmental, physiological, and behavioral driven fungal-caused disease spread.

## References

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## Supplementary Material

Data file from original study: <https://datadryad.org/stash/dataset/doi:10.5061/dryad.ngf1vhhvv>

Table x: Description of every column in data file

Column	Description
state	capture site state abbreviation
rMapState	capture site state
rMapCounty	capture site county
month	month of capture, numerical
day	day of capture, numerical
year	year of capture, numerical
age	age group (adult/juvenile)

sex	sex (male/female)
repstat	reproductive status (non-reproductive/pregnant/lactating/post-lactating/testes-descended)
mass	mass in grams
fa	forearm length in milimeters
Pd_intro	earliest year of confirmed or suspected <i>Pd</i> occurrence in each state
years_Pd	years since <i>Pd</i> introduction calculated by subtracting <i>Pd_intro</i> from <i>year</i> of capture
disease_time_step	groupings based on <i>Pd</i> exposure time (pre-invasion: < 0 years; invasion: 0-1 years; epidemic: 2-4 years; establishment: 5+ years)
county_centroid_lon	longitude of centroid point of county of capture
county_centroid_lat	latitude of centroid point of county of capture
site_mask	site names masked in format of 'state abbr._unique identifier'
fips	county-level FIPS code