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RESEARCH ARTICLE

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Higher fat stores contribute to persistence of little brown bat populations with white-nose syndrome

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Abstract

1. The persistence of populations declining from novel stressors depends, in part, on their ability to respond by trait change via evolution or plasticity. White-nose syndrome (WNS) has caused rapid declines in several North America bat species by disrupting hibernation behaviour, leading to body fat depletion and starvation. However, some populations of *Myotis lucifugus* now persist with WNS by unknown mechanisms.
2. We examined whether persistence of *M. lucifigus* with WNS could be explained by increased body fat in early winter, which would allow bats to tolerate the increased energetic costs associated with WNS. We also investigated whether bats were escaping infection or resistant to infection as an alternative mechanism explaining persistence.
3. We measured body fat in early and late winter during initial WNS invasion and 8 years later at six sites where bats are now persisting. We also measured infection prevalence and intensity in persisting populations.
4. Infection prevalence was not significantly lower than observed in declining populations. However, at two sites, infection loads were lower than observed in declining populations. Body fat in early winter was significantly higher in four of the six persisting populations than during WNS invasion.
5. Physiological models of energy use indicated that these higher fat stores could reduce WNS mortality by 58%–70%. These results suggest that differences in fat storage and infection dynamics have reduced the impacts of WNS in many populations. Increases in body fat provide a potential mechanism for management intervention to help conserve bat populations.

KEYWORDS
emerging infectious disease, evolution, plasticity, resistance, tolerance, trait change

1 | INTRODUCTION

Anthropogenic processes have resulted in rapid environmental change, resulting in novel environments and multiple stressors (Vitousek, Mooney, Lubchenco, & Melillo, 1997). The persistence of populations in the face of anthropogenic change depends, to some degree, on their ability to respond demographically or by trait change at the population level—whether through selection or phenotypic plasticity (Cattau, Fletcher, Kimball, Miller, & Kitchens, 2017; Darimont et al., 2009; Daszak, Cunningham, & Hyatt, 2001; Dirzo et al., 2014; Gomulkiewicz & Holt, 1995; Hendry, Farrugia, & Kinnison, 2008; Kilpatrick et al., 2006; Williams, Shoo, Isaac, Hoffmann, & Langham, 2008) . Novel pathogens can exert strong selective pressures on populations and cause rapid evolutionary responses in hosts (Altizer, Harvell, & Friedle, 2003) and have allowed some species to persist and recover (e.g., rabbits and *Myxoma virus* (Ratcliffe, Myers, Fennesy, & Calaby, 1952), house finches and *Mycoplasma gallisepticum* (Bonneaud et al., 2011)), while other species have been driven extinct (e.g., chytridiomycosis; Skerratt et al., 2007). Identifying host traits that enable persistence can be critical to help facilitate management efforts to recover populations following perturbations (Hobbs, Hallett, Ehrlich, & Mooney, 2011).

In the past decade, the disease white-nose syndrome (WNS), caused by the fungal pathogen, *Pseudogymnoascus destructans* (*Pd*), has caused widespread declines in hibernating bats in North America and threatens several species with extinction (Frick et al., 2010, 2015, 2017; Langwig et al., 2012). *Pd* infects the epidermis of hibernating bats during winter causing gross lesions in infected skin tissue (Meteyer et al., 2009), a cascade of physiological responses (including hypotonic dehydration, hypovolaemia and metabolic acidosis), disruption of hibernation patterns and sickness behaviour (Bohn et al., 2016; Verant et al., 2014; Warnecke et al., 2013). In healthy bats, arousals during winter hibernation are infrequent (representing <1% of hibernation time expenditure) but necessary in order to counteract costs of torpor, including the build-up of metabolic waste and dehydration, despite also being energetically costly (using 80%–90% of total stored body fat throughout the winter) (Kayser, 1965; Thomas, Dorais, & Bergeron, 1990). *Pd*-infected bats arouse approximately twice as frequently as uninfected bats (Lilley et al., 2016; Lorch et al., 2011; Reeder et al., 2012), which results in premature depletion of body fat and starvation.

While many colonies of multiple species have been extirpated or continue to decline from WNS, some colonies of the little brown bat *Myotis lucifugus* in the north-eastern United States appear to have stabilized following initial declines (Langwig et al., 2012, 2017; Maslo, Valent, Gumbs, & Frick, 2015). The mechanisms allowing each colony to persist with *Pd* are not fully known but may include: (a) reduced transmission from density-dependent disease dynamics (Anderson, & May, 1992), (b) attenuated virulence of the pathogen (Alizon, Hurford, Mideo, & Van Baalen, 2009), (c) environmental refugia that reduce pathogen growth rate, such as lower temperatures or humidity (Langwig et al., 2012; Verant et al., 2014), (d) resistance to infection by bats (e.g., via the immune system or cutaneous barriers, including antimicrobial peptides or the skin microbiome (Hoyt et al., 2015)), as suggested by recent work showing lower pathogen loads in some colonies (Langwig et al., 2017); or (e) tolerance of infection (Frick et al., 2017), either via altered hibernation behaviour (Lilley et al., 2016) or greater fat stores, which could allow animals to tolerate more frequent arousals or sickness behaviour.

Given the impact of WNS on hibernation energetics (Reeder et al., 2012; Warnecke et al., 2012) and the importance of body fat in overwinter survival (Kunz et al., 1998; Supporting Information Figure S5), greater body fat of WNS-affected bats should also be associated with higher overwinter survival, but evidence for fatter bats following WNS declines has been mixed. A single analysis that grouped all data across 13 sites in Virginia suggested that body mass index (BMI) in females of three bat species, *M. lucifugus*, *Perimyotis subflavus* and *M. sodalis*, during early hibernation showed little temporal variation over a 5-year period following WNS detection (Powers, Reynolds, Orndorff, Ford, & Hobson, 2015). In contrast, at a single site in Kentucky, body mass and BMI of *M. lucifugus* in fall swarm were significantly higher in year following WNS detection than the three previous years (Lacki et al., 2015).

Here, we examine whether *M. lucifugus* in colonies that are now persisting at least 8 years after WNS detection have greater early winter body fat than bats from these same colonies nearly a decade earlier when WNS was first detected. We hypothesized that bats in persisting colonies would have greater early winter fat stores than when WNS emerged, and we used energetic models to examine whether differences in fat could explain the persistence of little brown bats at these sites. We also tested the alternate hypotheses that bats in persisting sites were escaping infection, or resistant to infection, by measuring infection prevalence and intensity of *Pd*. Finally, we gathered data on temporal patterns of variation in body mass for *M. lucifugus* to put our results in a broader spatial and temporal context.

2 | MATERIALS AND METHODS

2.1 | Sites

We studied bats at six sites in the north-eastern United States during initial WNS invasion (2009) and again roughly 8 years later (2016) in colonies persisting with WNS (Supporting Information Table S1). Specific dates of WNS arrival at each of these sites are unknown, but *Pd* is presumed to have invaded between 2008 and 2009, with the most severe declines occurring when many sick bats were observed leaving the site during the first 2–3 years following *Pd* arrival (Supporting Information Figure S1; Frick et al., 2010; Langwig et al., 2012).

2.2 | Data collection

We measured body fat in early winter (November to December) and again in late winter (February to March) to assess fat loss over

hibernation (Supporting Information Table S1). We sampled an average of 22 ± *SD* = 8 (range: 2009: 4–39; 2016: 19–20) *M. lucifugus* per site per visit with a roughly even sex ratio (Supporting Information Table S1).

In the winter of 2009, which encompasses the hibernation season from October 2008 to April 2009 (except for late winter sampling at Williams conducted in February 2008), we collected bats and measured fat mass using Soxhlet analyses. We collected bats during early (November through December) and late winter (February through March) from roosting locations inside hibernacula, recorded body mass and killed them via overdose of isoflurane followed by cervical dislocation. We stored specimens at 4°C for up to 2 weeks and then transferred them to Boston University where we measured total body fat mass by lipid extraction with a Soxhlet apparatus using 3:1 solution of 95% ethyl alcohol and petroleum ether (Reynolds & Kunz, 2001). We calculated fat mass (g) as the difference between dry mass before lipid extraction and lean dry mass after lipid extraction. We also calculated proportion body fat as the total body fat (g) divided by body mass (g) at time of collection.

In the winter of 2016 (October 2015–April 2016), we first sampled bats for *Pd* infection at their roosting location by swabbing bats using a polyester-tipped swab (Fisher Scientific 23-400-116) five times across the forearm and five times across the muzzle (Langwig et al., 2015). We stored swabs in RNAlater until processing to preserve genomic DNA (Puechmaille, Fuller, & Teeling, 2011). We measured *Pd* prevalence and fungal loads on bats from swabs using quantitative polymerase chain reaction (Muller et al., 2013) and quantified Pd DNA from C_t scores as described previously (Frick et al., 2017). After collecting swab samples and UV photographs of wings in situ, we transported bats in cloth bags inside coolers (maintained at room temperature, 17–24°C) from the hibernacula to a sprinter van housing a quantitative magnetic resonance (QMR) scanner to measure body fat. We used QMR, rather than Soxhlet analyses for measuring fat mass in winter 2016 sampling because it can be done on live bats and in a relatively short period of time, which minimizes disturbance to bats. QMR has been validated for a range of species, including little brown bats (Guglielmo, McGuire, Gerson, & Seewagen, 2011), providing body composition measurements (in our case, body fat) that are directly comparable (1:1) to Soxhlet analyses (McGuire & Guglielmo, 2010). The QMR scanner was maintained at 17–24°C inside the temperature-controlled van. We calibrated and validated the accuracy of the QMR using 5.02 and 15 g canola oil standards before and after sampling at each site. Prior to QMR measurement, we measured forearm length of each bat using digital callipers (±0.01 mm) and weighed each individual using a digital scale (±0.01 g). We allowed each bat to fully arouse and reach euthermic body temperature prior to QMR processing. Once aroused, each bat was gently inserted into and restrained by a 3-cm internal diameter plastic tube outfitted with breathing holes (EchoMRI-Bird-Mini-HLDR-ASM, product number 600-000533B). The tube was then placed inside the QMR scanner. Each scan took 2–3 min, and we scanned each bat twice. We averaged fat measurements from repeated scans to produce a single estimate of total body fat for each bat (the mean difference between the two measurements was 0.03 g). We calculated the per cent body fat as the amount of fat measured by QMR divided by the body mass of each bat. We measured body fat using QMR at all sampling visits in 2016 except for late hibernation sampling at Hibernia Mine where we measured forearm length and body mass. We estimated body fat for this visit based on the relationship between measured QMR fat and body mass (Supporting Information Figure S2). After QMR sampling, we placed bats in bags inside coolers, and transported them back to their roosting location and released them.

2.3 | Statistical analyses

All analyses were run in program R version 3.3.0 (Brack & Twente, 1985; R Core Team, 2016; Twente, Twente, & Brack, 1985).

2.3.1 | *Pd* prevalence and fungal load on bats in 2016

We examined *Pd* prevalence using a generalized linear mixed-effects model (with a binomial distribution and a logit link) and *Pd* load using a linear mixed-effects model using package *lme4*. We first examined whether *Pd* prevalence and load increased over hibernation time (defined as days since the fall equinox—September 22 or 23 depending on the year) as a continuous fixed effect and site as a random effect. We examined whether *Pd* prevalence or load differed between sexes or was correlated with body fat of a bat within each hibernation sampling time period (early or late winter), and included body fat, sex and hibernation time as fixed effects and site as a random effect. We used the Akaike information criteria with a correction for small sample size (function AIC_c in package *MuMIn*; Bartoń, 2016) to compare models.

We also compared trends in *Pd* prevalence and *Pd* loads over winter hibernation at our six sites with continent-wide patterns (Frick et al., 2017) to determine whether differences in *Pd* transmission (lower *Pd* prevalence in early winter) or host resistance (decreased change in *Pd* loads over hibernation and lower *Pd* loads at the end of hibernation; Langwig et al., 2017) were contributing to persistence of bats at our six sites. We compared *Pd* prevalence and loads at our six sites with >5,600 *Pd* samples taken across 130 sites across North America (Frick et al., 2017). We used the best-supported models describing continental trends in *Pd* prevalence and loads (generalized linear mixed-effects model with binomial distribution and linear mixed-effects model using package *lme4* and *lmerTest*), which included site as a random effect and days in hibernation, years since first *Pd* detection at a site and species as fixed effects (Frick et al., 2017). In order to compare trends in *Pd* prevalence and loads at our sites against continental trends (Frick et al., 2017), we included an additional grouping variable with one level for each of our six sites plus an additional grouping variable for continental data (designated as "continental"). We used this grouping variable as a fixed effect in this model with "continental" as the reference

level. For *Pd* prevalence analysis, we subtracted 32 days from the date (the mean hibernation day in early winter when our samples were taken) in order to compare the early winter *Pd* prevalence at our sites against the continent-wide dataset on this date. For *Pd* load analysis, we adjusted days in hibernation to the end of hibernation (day 192) in order to compare late winter *Pd* loads at our sites with the continent-wide dataset on this date.

2.3.2 | Site-specific differences in body condition between sampling periods

We compared body fat between bat colonies sampled in winter 2009 and 2016 at each site to account for site-specific differences in winter body fat in bats. For each of the six sampled sites, we examined predictors of body fat with linear beta regression models (function *betareg* in package *betareg*; Cribari-Neto & Zeileis, 2010). We included the two sampling time periods (2009 and 2016), sex of the sampled bat, hibernation time (also defined as days since the fall equinox) and interactions between these terms as fixed effects (Supporting Information Table S8). We used AIC_c, as described above, to compare models. We used beta regression because the response variable (fraction body fat) was constrained between 0 and 1 and because analyses with log- or arcsine-square-root-transformed data resulted in residuals that deviated significantly from normality using a Shapiro–Wilk normality test (function *shapiro.test*).

2.4 | Predicted WNS survival from greater body fat

In order to examine how increased body fat might affect survival of bats with WNS, we used an energetic model (Thomas et al., 1990) to estimate WNS mortality prior to, during WNS invasion in 2009 and after WNS invasion in 2016 (Supporting Information Appendix S1). We calculated overwinter mortality for bats starting with early hibernation body fat from our measurements in 2009 versus 2016, and expending energy over the course of hibernation at pre-WNS and peak-WNS rates (Lilley et al., 2016; Supporting Information Appendix S1). We compared overwinter mortality for three scenarios: (a) bats starting with early hibernation body fat observed in 2009 and experiencing no WNS impacts, (b) bats with early hibernation body fat observed in 2009 and experiencing higher arousal frequencies from WNS and (c) bats with early hibernation fat observed in 2016 and also experiencing higher arousal frequencies from WNS.

3 | RESULTS

3.1 | *Pd* prevalence and fungal loads on bats in 2016

In early winter of 2016, prevalence averaged 82.3% ± 7.8%, which was not significantly lower than prevalence observed at 5,659 sampling events across continental North America (Frick et al., 2017) where *Pd* has become established (Figure 1a; Table S3). Prevalence increased to >90% at five of six sites by late winter (Figure 1a), and changes in *Pd* prevalence over hibernation at our six sites did not differ from

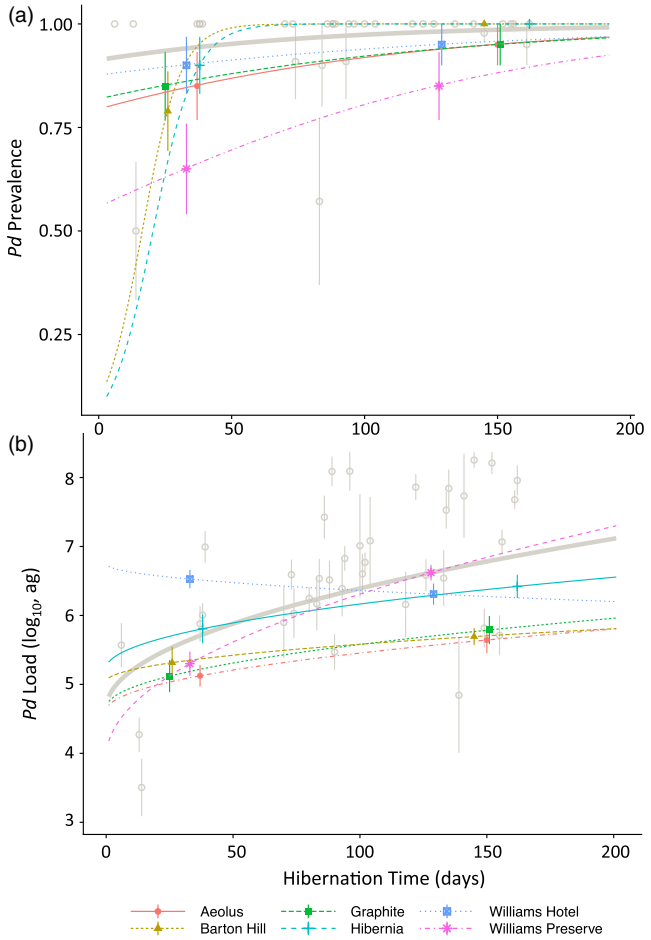

FIGURE 1 *Pd* prevalence and fungal loads in bats sampled in 2016 in early and late winter in six persisting *Myotis lucifugus* populations. Points show *Pd* prevalence ±1 *SE* based on a binomial distribution (a) and mean $\log_{10}$-transformed fungal loads in attograms ±1 *SE* (b) at our six sites (colours). Grey points and grey dashed lines represent sampling from a previous study of 130 sites across North America (Frick et al., 2017)

continental trends (Frick et al., 2017; Supporting Information Table S3). *Pd* loads in early winter were similar to those from declining sites in the continental study (Frick et al., 2017), but were lower than expected at the end of hibernation in two (Aeolus and Barton Hill) out of six sites (Figure 1b and Supporting Information Table S4). *Pd* loads increased over hibernation at comparable rates to continental trends except at Barton Hill where *Pd* loads increased at a lower rate compared to continental trends (Figure 1b and Supporting Information Table S4). At Williams Hotel, *Pd* loads decreased over hibernation (Figure 1b and Supporting Information Table S4), but these data may not be comparable as bats in early and late winter were sampled from different sections of the mine. *Pd* presence or load did not vary significantly with body fat or sex of the bat (Supporting Information Table S5).

3.2 | Site-specific changes in body condition

Fat stores in early winter were significantly greater in bats sampled in 2016 than in 2009 at four out of six sites (Figures 2 and 3, Supporting

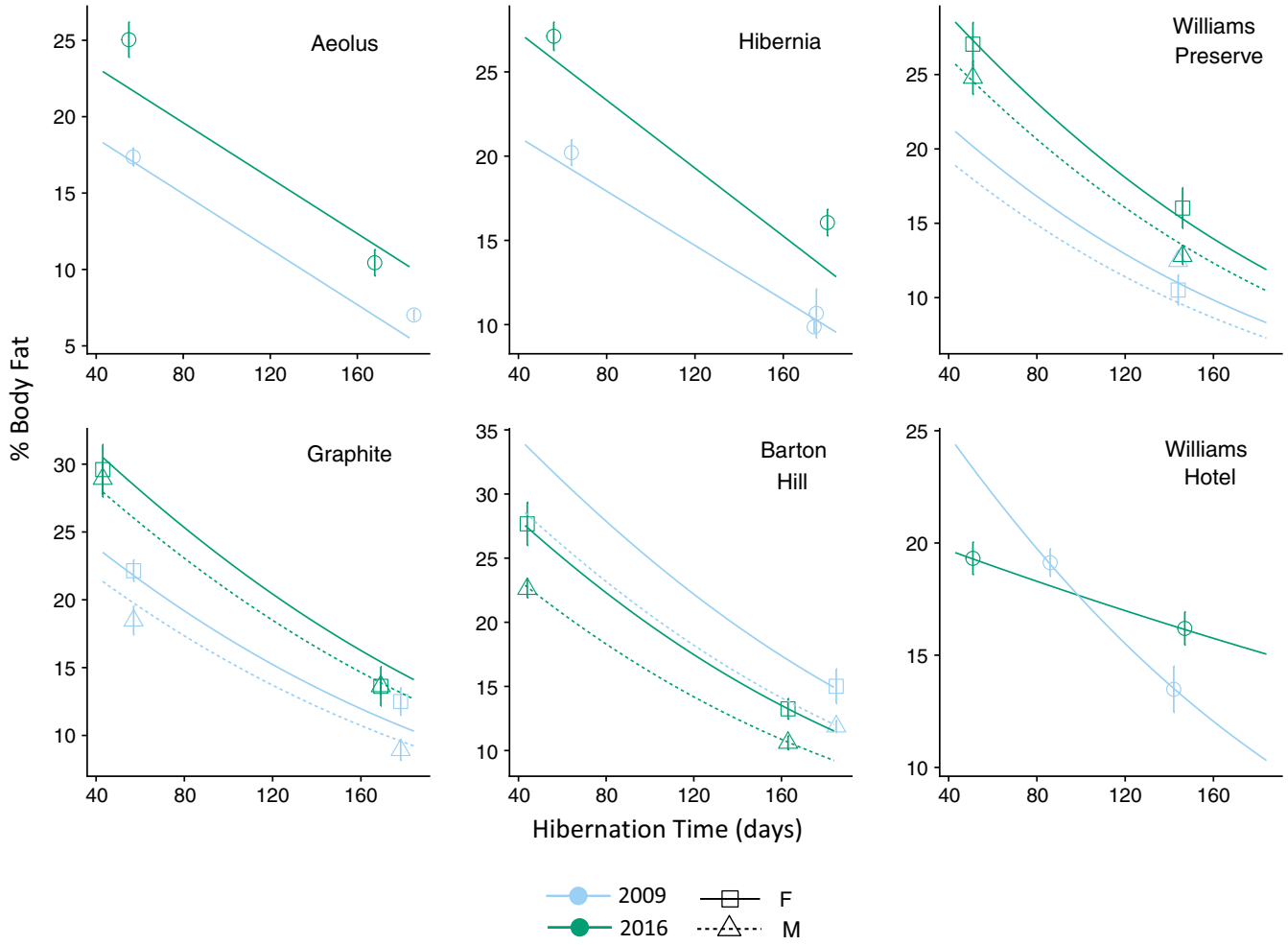

FIGURE 2 Mean per cent body fat (±1 *SE*) of *Myotis lucifugus* over hibernation time (days since fall equinox) sampled in 2009 and 2016 at Aeolus Cave, Hibernia Mine, Graphite Mine, Williams Preserve Mine, Barton Hill Mine, Williams Hotel Mine. In each panel, fitted lines are based on best-supported beta regression models (Supporting Information Table S6), and include variation between sexes where significant

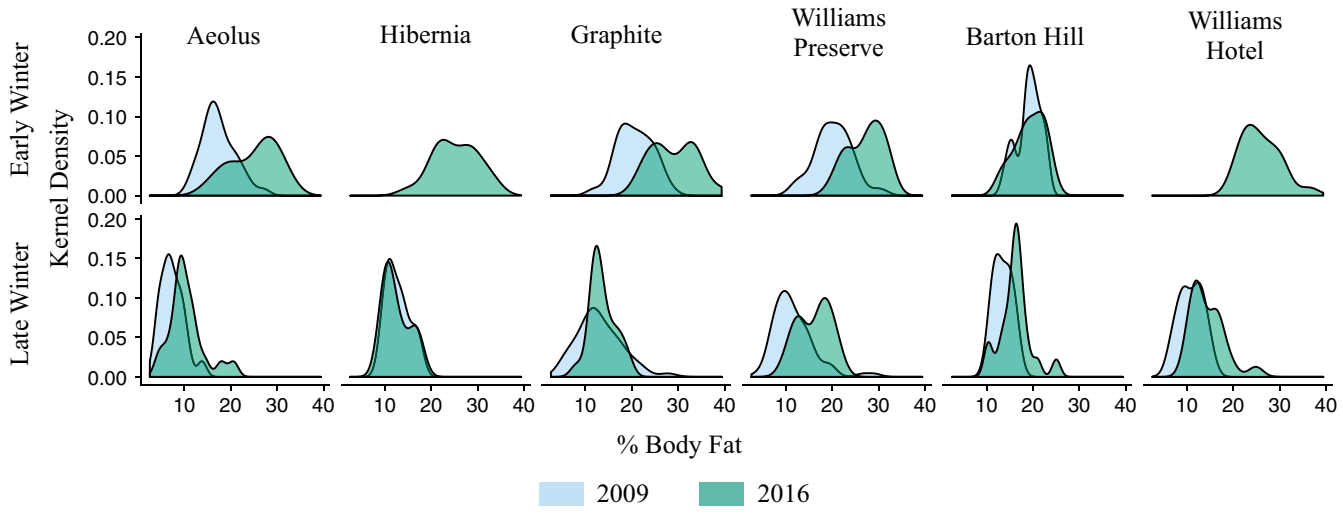

FIGURE 3 Kernel density plots of body fat measured in bat populations from 2009 and 2016 in early winter (first row) and late winter (second row). No measurements were made in early winter in 2009 at Williams Preserve or Barton Hill. Note that bats were measured for body fat on different dates at Barton Hill and Williams Hotel in 2009 and 2016, which makes comparisons between years (2009 and 2016) using the kernel density plots difficult

Information Tables S6 and S8) and when examined collectively across all sites (Supporting Information Table S8). At sites where bats were fatter, they were fatter by 0.8 ± 0.009 g (mean ± *SE*; Aeolus, Graphite, Hibernia, Williams Preserve) and had greater body mass by 1.08 ± 0.07 g (mean ± *SE*; Aeolus, Graphite, Hibernia), exceeding natural inter-annual variation in body mass, which was 0.19 ± 0.05 g (mean ± *SE*; Bay City Mine, Wise1, Williams Preserve) (Figure 4 and Figure S3). At three of these four sites (Aeolus, Graphite, and Williams Preserve), the rate of fat loss over winter did not differ between 2009 and 2016 (Supporting Information Tables S6 and S7), whereas at Hibernia Mine, bats lost fat more slowly over winter in 2016 than 2009 (Supporting Information Tables S6 and S7). At two sites (Barton Hill and Williams Hotel), fitted models suggested that early winter body fat was lower in 2016 than 2009 (Figure 2; Supporting Information Tables S6 and S7). In addition, at Williams Hotel, the best-fit model suggested that 2016 bats started with less fat in early winter but lost fat more slowly than 2009 bats (Figure 2; Supporting Information Tables S6 and S7). Body fat was significantly higher in female bats than males at three of the six sites (Supporting Information Figure S4; Tables S6 and S7).

3.3 | Predicted WNS survival from greater body fat

Across all sites, we estimated that average winter mortality prior to WNS arrival was 6% ± 3%, increased to 52% ± 12% during initial mortality from WNS in 2009 and decreased to 36% ± 9% in 2016 (Figure 5). At sites where bats were fatter in 2016 (Aeolus, Graphite, Hibernia, Williams Preserve), mortality prior to WNS arrival was estimated at 4% ± 3%, but rose to 69% ± 16% during WNS invasion in 2009. In 2016, estimated mortality decreased to 25% ± 4% for bats experiencing sustained impacts from WNS but with greater body fat (Figure 5). Thus, these energetic models suggest that higher body fat observed in persisting bats would reduce WNS mortality by an average of 64% (Figure 5).

4 | DISCUSSION

White-nose syndrome has caused widespread declines and extirpations in many colonies of hibernating bats as it has spread across North

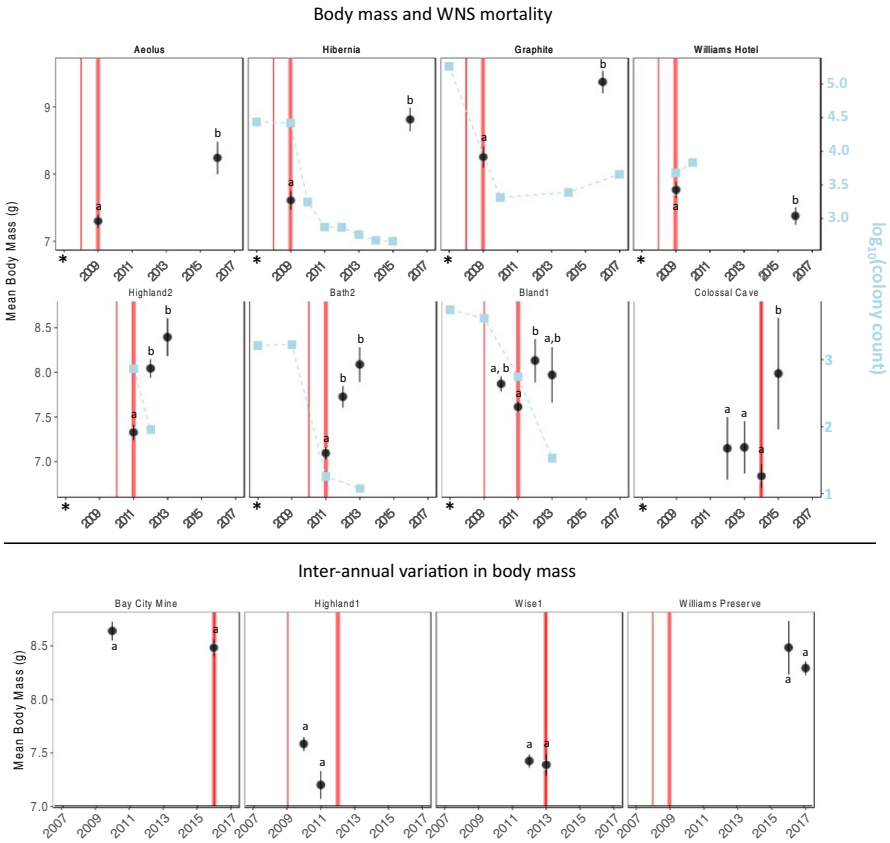

FIGURE 4 Body mass and colony counts of *Myotis lucifugus* over time. Body mass and White-nose syndrome (WNS) mortality (top panel): mean body mass with standard error (solid shapes, left axes) and colony count (log-transformed; blue squares, right axes) of *M. lucifugus* are plotted by winter year (e.g., winter year 2009 includes the hibernation season from September 2008 to April 2009). The thin red bar indicates the first detection of WNS, and the thick red bar indicates the first year of mass mortality at each site. Letters above solid shapes denote comparisons in body mass among years, with significant differences ($\alpha = 0.05$) indicated by different letters. Historical counts (prior to winter year 2009) are denoted by asterisks. Inter-annual variation in body mass (bottom panel): mean body mass with standard error (closed circles, left axes) is shown for different years either before or after WNS detection (thin red line) or WNS mortality (thick red line). Letters above solid shapes denote comparisons in body mass among years, with significant differences ($\alpha = 0.05$) indicated by different letters

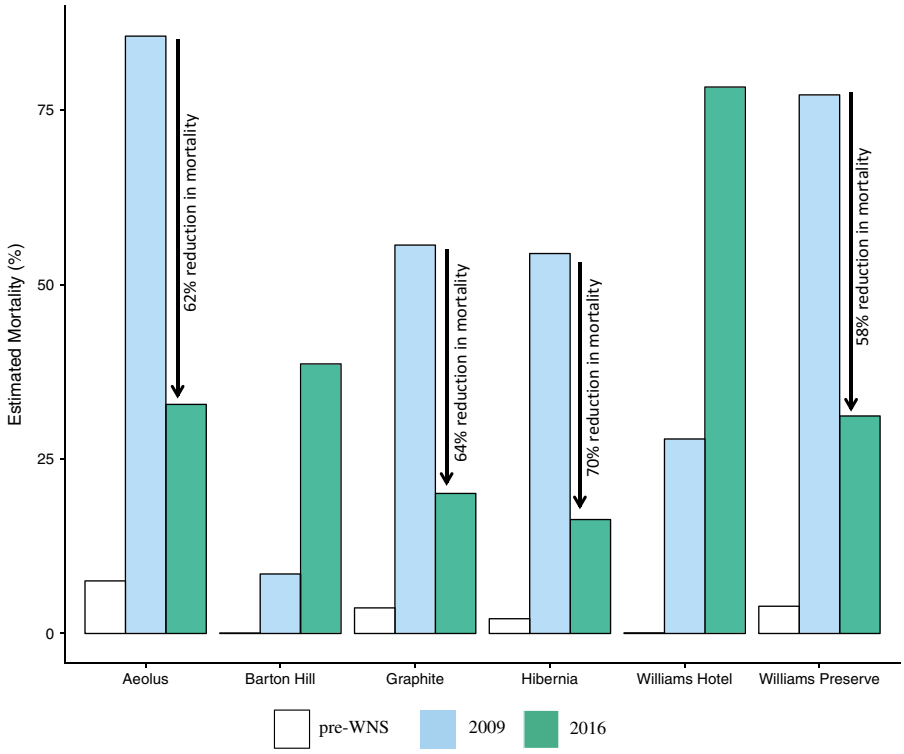

FIGURE 5 Estimated winter mortality from energetic models based on fat and hibernation energetics for bat populations prior to *Pd* invasion (pink bars), during *Pd* invasion in 2009 (green bars), and following *Pd* invasion in 2016 (blue bars). Arrows and text indicate where greater early winter body fat in 2016 resulted in reduced White-nose syndrome (WNS)-related mortality compared to peak-WNS mortality in 2009

America, but some colonies of *M. lucifugus* are now persisting with the fungus in parts of the north-eastern United States (Frick et al., 2015, 2017; Langwig et al., 2012, 2017). We found that infection with the fungus in six persisting colonies was not lower than expected, but that infection intensity was lower than in declining populations at two persisting colonies at the end of winter, and at four of six sites bats were significantly fatter in early hibernation in 2016 than bats at these sites nearly a decade earlier during initial invasion of the fungus. Energetic models suggested that the increased fat stores observed at these sites could reduce WNS mortality by 58%–70%.

Although increased body fat likely reduced WNS mortality, other mechanisms are necessary to fully explain persistence of bats with WNS at our sites. *Pd* prevalence in bats sampled during early hibernation was 10%–35% lower than sites where *Pd* has been present for at least 5 years (Frick et al., 2017). These results suggest that *Pd* transmission was lower in early winter at these sites, either due to density-dependent mechanisms (Anderson & May, 1979), reduction of the environmental reservoir (and therefore environment-to-bat transmission) over time, or increased resistance in bats (Langwig et al., 2017). Lower transmission would increase survival by (a) allowing some bats to escape infection entirely (5%–15% of bats were *Pd*-negative in late winter at our sites) and (b) delaying infection in bats, some of which could survive until spring since death often occurs 70–100 days after infection in the laboratory (Warnecke et al., 2012). We did not find evidence that *Pd* transmission was lower at persisting sites than in declining populations. However, we found that late winter *Pd* loads were lower than expected at two sites and increased at a slower rate over hibernation at one of these sites compared to declining populations (Frick et al., 2017), suggesting that some bats may be exhibiting resistance, as has been documented in four colonies of *M. lucifugus* in New York (Langwig et al., 2017), including two that were studied here (Williams Preserve where fat stores were higher, and Williams Hotel where they were not). Increased torpor bout duration could also contribute to continued persistence of WNS-affected bats, and has been suggested as an explanation for persistence at one site (Lilley et al., 2016). Although temperatures were also cold at this site (and torpor bout duration decreases loglinearly with roosting temperature (Brack & Twente, 1985; Twente et al., 1985)), the torpor bout duration was 29% longer than expected based on temperature (Supporting Information Figure S6). However, bats at this site had much longer arousal durations; thus, it is not clear whether their energy expenditure would be lower than bats exhibiting disturbed hibernation behaviour characteristic of WNS (Reeder et al., 2012; Warnecke et al., 2012). Finally, *M. lucifugus* in hibernacula with lower temperatures had less severe declines due to WNS (Langwig et al., 2012), possibly due to reduced *Pd* growth (Verant et al., 2012) or lower energy expenditure by bats during hibernation (Brack & Twente, 1985; Twente et al., 1985), suggesting that cooler hibernacula temperatures could also facilitate bats persisting with *Pd*. Additional research is needed to determine the relative contribution of these different mechanisms to persistence of *M. lucifugus* with WNS.

Our results are consistent with previous studies showing increased body condition in early winter in *M. lucifugus* following WNS declines at the majority of sites, but not all. We re-examined differences in body mass from a study in Virginia of *M. lucifugus* (Powers et al., 2015), which had data from three sites where bats were sampled in early winter both before and after WNS detection and had >4 bats per sample (Supporting Information Appendix S2). We found that body mass was 15%–23% higher in two (Highland 2, Bath 2) out of three

colonies following declines of 88%–99% (Figure 4 and Supporting Information Figure S3), and 7% higher in another (Bland 1; Figure 4 and Supporting Information Figure S3). A separate study in Kentucky (Colossal Cave) found that body mass of *M. lucifugus* measured during fall swarm was higher by 15%–20% 1 year following WNS detection and an 80% decline (Figure 4 and Figure S3; Lacki et al., 2015). These data suggest that body fat increases following mass mortality due to WNS in many, but not all, colonies of *M. lucifugus*, and are contributing to persistence of bat colonies with WNS. Whether the variation in increased fat stores is due to differences in site quality near hibernacula (e.g., higher insect abundance), or the strength of selection due to WNS mortality, or other factors remains to be determined.

Several mechanisms could result in bats having higher fat composition in early winter following WNS declines. First, inter-annual variation in prey availability in fall, due to natural variability or decreased resource competition, could lead to higher fat composition if bats exhibit phenotypic plasticity in body fat and forage to higher fat stores when prey abundance is higher. However, temporal data from colony counts in New York, Virginia, Wisconsin and Kentucky suggest that inter-annual variation in early winter body mass is lower than we observed (Figure 4 and Supporting Information Figure S3) and the weather (precipitation and temperature) was similar in 2009 and 2016 (Supporting Information Figure S7). Second, if levels of fat storage in fall is a heritable trait, our results suggest that natural selection would increase fat stores through increased survival with WNS, and possibly increased reproduction due to higher fat stores at the end of winter. Although both phenotypic plasticity and evolutionary change could increase fat stores, if higher fat stores post-WNS declines are due to reduced competition, then bat populations are less likely to reach their previous pre-WNS abundance. Regardless, management actions that allow bats to obtain higher fat stores in fall before beginning hibernation will aid in conservation and recovery.

Thus far, most conservation efforts for WNS have been centred on treating bats or reducing *Pd* in the environment during winter hibernation (Cheng et al., 2016; Cornelison et al., 2014; Hoyt et al., 2015; Zhang, Chaturvedi, & Chaturvedi, 2015) when WNS most heavily impacts bats (Langwig et al., 2015). Our study suggests that management efforts that increase bats' ability to increase fat stores in autumn may help facilitate population persistence. *M. lucifugus* are dietary generalists, primarily consuming emerging aquatic insects (Anthony & Kunz, 1977; Belwood & Fenton, 1976; Clare et al., 2013). Management actions that protect and restore aquatic habitats, especially near hibernacula, may facilitate greater fat accumulation during the fall swarm period (Parker et al., 2018). For example, states could provide incentives for landowners not to remove beavers and beaver dams which create wetlands, and they could incentivize or implement wetland or riparian restoration by reducing river channelizing, and by reducing invasive plants and restoring native vegetation which has recently been shown to be critical for some insectivorous bird populations (Narango, Tallamy, & Marra, 2018). Managing habitats to increase the ability of bats to increase fat stores before winter has several advantages as a management strategy. First, there is no associated risk with causing additional mortality due to disturbance during hibernation. Second, it can be done in combination with other conservation efforts occurring during winter. Improving body condition may facilitate both increased survival and reproduction, and this can facilitate the evolution of resistance or tolerance (Kilpatrick, 2006; Maslo et al., 2015). Employing these conservation strategies may provide critical relief and facilitate recovery as WNS impacts continue to threaten North American bat populations.

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AUTHORS' CONTRIBUTIONS

T.L.C. conceived of the study idea following a presentation by G. Turner at a white-nose symposium; T.L.C., A.M.K. and W.F.F. designed and coordinated the study, conducted data analyses and drafted the manuscript; T.L.C., J.D.S. and A.G. collected the data; A.G. and C.K.R.W. advised the study design and participated in data analysis; J.D.R. and M.S.M. helped with study design and data collection. All authors contributed to the final version of the manuscript and gave final approval for publication.

DATA ACCESSIBILITY

Data used in this study are from the Dryad Digital Repository: https://doi.org/10.5061/dryad.sh487nh (Cheng et al., 2019).

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SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section at the end of the article.

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