Diagnostic evaluation of unknown white-tailed deer morbidity and mortality in New York State: 2011–2017

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White-tailed deer *Odocoileus virginianus* are the most popular big game animal in the United States. Recreational harvest of these animals is a critical tool in population management, as well as an important financial resource for state economies and wildlife agencies. Thus, herd health evaluations can provide information to wildlife managers tasked with developing sustainable harvest practices while monitoring for emergent problems. The purpose of our study was to document causes of illness and natural mortality in New York white-tailed deer submitted for post mortem evaluation. Animals were presented by members of the public and wildlife management personnel due to abnormal behavior or unexplained death. We describe demographic and seasonal associations among gross and histologic evaluation and diagnostic testing. Post mortem examinations were performed on 735 white-tailed deer submitted for necropsy in New York from January 2011 to November 2017. Causes of euthanasia or mortality were classified into nine categories. The most common findings were bacterial infections, trauma not evident at time of collection, and nutritional issues, primarily starvation. Using a multinomial logistic regression model, we looked for associations between the mortality categories and age, sex and season. Compared to the baseline of bacterial deaths, adults were less likely to have died from nutritional and parasitic causes, males were less likely to have died from other causes, and risk of death from nutritional reasons decreased from season to season, with lowest risk in winter. These methods can help wildlife biologists track changes in disease dynamics over time.

Keywords: infectious disease, malnutrition, mortality, *Odocoileus virginianus*, white-tailed deer
DEC may be notified of deer meeting these criteria by members of the public or law enforcement and can submit the animal for necropsy and diagnostic testing. Because the surveillance program specifically excludes deer that died from obvious predation, hunting, and deer-vehicle collisions, animals collected do not represent the New York population as a whole; however, they are valuable for assessing the breadth of diseases affecting wild deer and establishing a standardized baseline for future assessment. A benefit of this program is that these animals can serve as sentinels for emerging diseases. This type of opportunistic surveillance is a widely used method for states to prioritize deer that could be infected by CWD (Joly et al. 2009). Providing a basis for comparison will allow states to refine their surveillance systems to be better informed about white-tailed deer diseases by demographic categories and seasonality.

For the present study, records from deer presented for necropsy through the surveillance program from 2011 to 2017 were compiled to retrospectively evaluate disease occurrence in a subset of the New York deer population. Our objectives were to: 1) describe causes of morbidity and mortality in wild deer and; 2) evaluate differences in cause of morbidity and mortality according to sex, age and season.

Study area

Free-ranging wild white-tailed deer were collected through the DEC surveillance program throughout New York (141 300 km²), which is composed of two major topographical regions: the Appalachian Mountains and the Adirondack Highlands. No regions of New York were excluded for this study. Wildlife health surveillance activities were overseen by DEC statewide.

Methods

Beginning in 2011, DEC biologists and staff were trained through a series of bi-annual workshops run by DEC to respond to observations or reports from the public and law enforcement of wild white-tailed deer that appeared to be sick, were emaciated, behaving abnormally, or were found dead without obvious cause. Explicit instructions were given not to collect animals that appeared to have died from obvious causes such as predation, hunter harvest, or vehicle collisions. Written procedures for specimen submission and expert consultation were available to DEC field staff both online and over the phone. The form for specimen submission included field history, finder/submitter information, reason for submission, and other circumstances, such as unusual weather events or environmental contamination (<http://nwdc.wildlifesubmissions.org>). DEC Bureau of Wildlife or law enforcement staff euthanized or humanely killed sick deer using physical (gunshot) or chemical methods according to the recommendations of the American Veterinary Medical Association Guide to Euthanasia in Animals (Underwood et al. 2013). Carcasses or diagnostic specimens that could not be delivered to a laboratory immediately were stored frozen or chilled at DEC facilities.

Gross and histopathological examination

DEC field personnel delivered carcasses to either the Cornell University Animal Health Diagnostic Center (AHDC, Ithaca, NY) or the DEC Wildlife Health Unit (WHU, Delmar, NY) for full necropsies and diagnostic testing. Though infrequent, fresh and/or formalin-fixed tissues were also accepted for submission if the entire carcass was unavailable. All animals received a full gross examination of major organs by a diagnostician (trained biologist or board-certified anatomic pathologist) and, when appropriate, samples of lesions and major organs were collected in 10% neutral buffered formalin for histopathologic processing; ancillary veterinary diagnostic testing (i.e. virology, bacteriology, toxicology, parasitology, endocrinology and clinical pathology) was done at the diagnosticians’ discretion. Sections of obex and retropharyngeal lymph node were submitted for CWD testing, and sections through the cerebellum and brainstem were submitted for rabies testing at the New York State Department of Health in Slingerlands, NY for all animals where tissues were available. In rare cases of a rabies-positive result, no further gross examination or diagnostics were performed. Processing of tissues for histopathology, CWD testing, and ancillary testing were conducted using standard methods established at the AHDC (<www.vet.cornell.edu/animal-health-diagnostic-center>, accessed 15 April 2021).

In addition to gross lesions, the diagnostician performing the necropsy recorded the sex and age of the animal. Age was categorized as neonate (< 1-month-old), juvenile (1–18 months old) or adult (≥ 18 months old) based on coat, size, and tooth wear pattern (Severinghaus 1949, Shuman et al. 2017). We chose these age classes because we could not determine precise age by month for many deer, and these age classifications were of highest utility for management purposes. All gross, histopathologic, and laboratory findings were archived in a wildlife health database (Canadian Wildlife Health Center, SK, Canada). The cases used in this study were identified by searching for all white-tailed deer in this database within the timeframe of 2011–2017. Cases were further refined to exclude non-natural causes of death, including hunting, vehicular trauma, and research purposes.

Study design

Deer included in this study were opportunistically collected as part of an ongoing surveillance program of wild white-tailed deer and do not represent a random sampling strategy. Therefore, we had strict inclusion criteria for study animals. All deer were free-ranging, recovered in New York and were not killed by hunters or euthanized for management purposes. We supplemented our neonate category before searching the database through an ongoing radio-collared study started in 2015 at Fort Drum, NY examining urban white-tailed deer survival (<https://fortdrumdeer.org>, accessed January 2018). These animals fit submission criteria in that they were found dead without an obvious cause. Collection dates of deer were classified by season as follows: 1) autumn (hunting and rutting seasons) was October–December; 2) winter (post-rut) was January–March; 3) spring (fawning season) was April–June and; 4) summer was July–September.
Analysis of post mortem data

A board-certified anatomic pathologist (E. L. Buckles) reviewed all pathology reports, laboratory results, submission histories, and when needed, case photos and histologic slides from white-tailed deer that met inclusion criteria to determine a final cause of death. Determination of the cause of death was based on analysis of the damage to the tissue and if such damage was significant enough to lead to fatal organ dysfunction, systemic illness and/or reported signs prior to euthanasia. In some cases, gross findings were sufficient for determination, but histopathology was conducted in all animals in order to rule out lesions not detectable at gross examination, such as disease, that may have predisposed an animal to trauma or resulted in emaciation. When possible, the etiology of the lesions was recorded based on combinations of gross, histologic, and laboratory findings. After analysis, deer were classified into nine mortality categories: traumatic, bacterial, viral, parasitic, fungal, nutritional, unknown, toxin, or other (Table 1). Traumatic death was defined as the clear presence of injury, such as hemorrhages and fractures, in the absence of other lesions that may have predisposed the animal to traumatic death. Infectious diseases were recognized by gross and histologic detection of inflammation and degenerative changes, results of veterinary diagnostic testing, or by a combination of morphologic analysis and diagnostic testing. Nutrition-related morbidity/mortality included emaciation, ruminal acidosis or its sequelae, such as fungal rumenitis, and vitamin E/selenium deficiency as determined by toxicological testing. Emaciation was interpreted to be the cause of death if no other significant lesions were present or if emaciation was found in conjunction with significant dental attrition. The category of 'other' included cases of spontaneous abortion, congenital abnormalities, foreign bodies, drowning, dystocia, cardiac diseases and neonate mortality. We defined death from neonate mortality as an encompassing term for fawns with no significant lesions along with either a poor body condition, an empty gastrointestinal tract indicating a lack of feeding, or being found dead after harsh environmental conditions, such as periods of excessive cold or heat. This category encompassed metabolic issues (hypoglycemia), environmental (hyperthermia, hypothermia), or failure of maternal care, none of which would be evident at gross or histopathological assessment. The cause of death was deemed unknown if there were no significant postmortem or laboratory findings or not enough information was provided by the submitter.

Statistical analysis

Difference in average weight by sex was assessed using the Kruskall–Wallis rank sum test at $\alpha \leq 0.05$. Differences in cause of death between sex, age and season of death were evaluated using a multinomial logistic regression model at $\alpha \leq 0.05$ to assess significance. The outcomes of interest in this study are the eight nominal (non-ordered) causes of death, and the exposure variables are sex, age and season of death. Statistical analysis and figures were performed or generated using R ver. 3.6.3 (www.r-project.org).

Results

From January 2011 to November 2017, a total of 534 deer out of 735 met the criteria for inclusion in the study. Study animals were submitted from all New York counties with the exception of New York County and Kings County. Most submissions (> 95%) were full carcasses. Deer that died from obvious, non-natural causes, including deer killed for diagnostic tests (9), forensic studies (102), research (21), hunter killed (49), obvious vehicular trauma and predation (20) were excluded. The study population consisted of 230 females, 169 males, and 135 animals of unknown sex. There were 227 adults, 157 juveniles, 17 neonates, and 133 deer of unknown age. Weight data was available for 215 cases in which full carcasses were submitted. On average, adult males weighed 60.7 kg (95% CI: 45.4, 76.0 kg), juvenile males 23.6 kg (95% CI: 5.8, 41.4 kg), adult females 48.5 kg (95% CI: 38.6, 58.4 kg), and juvenile females 24.9 kg (95% CI: 7.6, 42.4 kg). There was no weight difference between all males and females ($\chi^2 = 1.45$, $p = 0.23$). The majority of the animals for which sex, age and weight data were unavailable were in the ‘unknown’ cause of death category (n = 253). Of these, a diagnosis could not be obtained in 168 because of insufficient histories and lack of appropriate tissue collection. Missing data in the remaining cases was due either to missing tissues, autolysis or failure of the prosector to record the data.

A total of 387 deer had both known sex and age, and cause of death was determined in 281 of these animals (Table 1). More deer were submitted in autumn compared to other seasons (Fig. 1). There were more female carcass submissions than males throughout the year. Male deaths increased during autumn, specifically during October and November (Fig. 2). Excluding unknown cause of death,

Table 1. Description of mortality categories based on analysis of gross, histologic, and laboratory findings and distribution of free-ranging white-tailed deer deaths from unspecified causes submitted in New York State between 2011–2017, n = 534

| Cause of Mortality | Description                                                                 | Count |
|--------------------|-----------------------------------------------------------------------------|-------|
| Bacterial          | Animal died or euthanized due to bacterial infection                          | 129   |
| Fungal             | Animal died or euthanized due to fungal infection                            | 3     |
| Nutritional        | Death due to starvation/emaciation, fermentation problems                    | 28    |
| Other              | Death or euthanasia due to etiologies that do not fit in other categories    | 37    |
| Parasitic          | Animal died or euthanized due to parasitic infection                          | 21    |
| Toxin              | Animal died or euthanized with toxin as a primary diagnosis                   | 1     |
| Traumatic          | Cause of death or euthanasia was traumatic without clear relation to underlying condition; obvious vehicle collisions were excluded | 44    |
| Unknown            | Cause of death undetermined                                                  | 253   |
| Viral              | Animal died or euthanized due to viral infection                             | 18    |
Figure 1. Free-ranging white-tailed deer of natural, unspecified mortality submitted for necropsy examination and cause of death determination by month of death in New York, 2011-2017 (n = 534)

Figure 2. Distribution of free-ranging white-tailed deer dying of natural, unspecified mortality in male and female animals by month, New York 2011–2017 (n = 399; sex: F, female; M, male)
adults generally died from bacterial and traumatic causes, while juveniles died primarily from bacterial and nutritional causes, with nutritional death making up a greater fraction of all juvenile deaths (Fig. 3).

Of the infectious causes of death, bacterial infections were the most frequent \((n = 129)\) (Fig. 4). The common primary diagnoses in this category were suppurative or necrotizing respiratory infections including pneumonia, pleruitis or pleruapneumonia \((n = 49)\), and meningoencephalitis with or without accompanying pituitary abscesses \((n = 33)\), sepsis/vasculitis \((n = 10)\), arthritis/osteomyelitis \((n = 4)\), and unspecified abscesses \((n = 5)\). Two deer were euthanized due to skin lesions, which were identified as *Dermatophilus*. Of the deer that died from bacterial infections, tissues were submitted for bacterial culture from 83 animals. The facultative anaerobic bacteria *Trueperella pyogenes* \((n = 38)\) was isolated most frequently. Infections with this pathogen were characterized by pockets of viscous to casous yellow/green pus. In cases of pleuritis, the suppurative areas extended into the pleura and formed pockets between the body wall and the lungs. Encapsulation and tenacity of pleural adhesions varied according to chronicity (Fig. 4). Other isolates included *Escherichia coli* \((n = 9)\), *Fusobacterium* spp. \((n = 9)\), *Pasteurella multocida* \((n = 7)\), *Bibersteinia* spp. \((n = 5)\), *Dermatophilus* spp. \((n = 4)\), and *Bacteroides* pyogenes \((n = 2)\). Twenty cultures resulted in either no bacterial growth or in growth of mixed bacteria interpreted to be contaminants. In these cases, the lesions were interpreted to be bacterial in origin based on the histologic character of the lesions.

The only obligate anaerobic bacteria isolated were *Fusobacterium necrophorum* and *F. nucleatum*, which were isolated or identified microscopically from lesions of eight individuals. These bacteria were isolated from cases of stomatitis, esophagitis, meningoencephalitis, and pneumonia, either alone or in combination with other isolates, such as *T. pyogenes*.

Mycoplasma infection was suspected in one case of pneumonia based on the histologic features including airway centric inflammation surrounding central cores of brightly eosinophilic, acelluar material. Unfortunately, culture was unrewarding due to sample contamination and immunohistochemistry for *Mycoplasma bovis* was inconclusive.

Parasitic infections accounted for 21 of the deaths, and 12 of these were due to infections of the central nervous system or optic nerve either by aberrant nematode migration \((n = 11)\) or *Neospora* infection \((n = 1)\). Other diagnoses included *Fascioloides* hepatitis \((n = 2)\), presumptive Coenurosis \((n = 1)\), an unidentified Apicomplexa in the heart and brain \((n = 1)\), *Setaria* peritonitis \((n = 1)\), and non-specific verminous pneumonia \((n = 1)\). Studies to characterize the central nervous system nematodes and the Apicomplexa are ongoing.

Of the 18 animals identified with viral infections, the most common was rabies \((n = 8)\) followed by epizootic hemorrhagic disease \((n = 5)\) by PCR. Four individuals from this latter group were submitted from a localized outbreak. Two animals were infected and died from West Nile virus (PCR), two animals had extensive cutaneous papillomatosis, and one animal was infected with a herpesvirus, which we were unable to further characterize beyond PCR by virus isolation and sequencing.

Figure 3. Causes of death in free-ranging white-tailed deer of unspecified mortality determined by necropsy subsetted by age group in New York, 2011–2017 \((n = 387)\).
Only three animals died as a result of fungal disease, one from pneumonia due to an unidentified organism, one from gastric infection by an organism histologically compatible with a zygomycete, and one due to *Mucor* sp. infection of the sinuses and brain. In contrast to the lesion in the lungs of animals dying of bacterial pneumonia, the lesions of fungal pneumonia were nodular, very firm, and caseous. One animal died from exposure to a toxin (tetanus). No positive cases of CWD were detected out of 254 animals tested, despite intensive efforts to sample high risk animals.

**Statistical model**

In the multinomial model, bacterial deaths was set as the baseline because it was the most common cause of death in all deer and was relatively constant across seasons. The one toxin case were excluded from statistical analysis because of the small sample size. In comparison to bacterial deaths, risk of death from nutritional (OR = 0.27, p = 1.5e-5) causes decreased from spring through winter, with lowest risk during winter. Males were less likely than females to die from ‘other’ causes of death (OR = 0.25, p = 0.02); sex was not significant for remaining death categories. Age was a significant risk factor for nutritional (OR = 0.20, p = 1.1 × 10^-3) and parasitic (OR = 0.22, p = 6.8 × 10^-3) deaths, where adults were significantly less likely to die from these causes compared to juveniles and neonates. Age, sex and season were not significant risk factors for any other mortality categories (Table 2).

**Discussion**

White-tailed deer are an important resource for New York State and play a prominent role in One Health as a source of recreation and food for humans and in their interactions with...
livestock across the landscape. Our retrospective analysis of common causes of morbidity and mortality in free-ranging white-tailed deer establishes a baseline against which emerging issues can be evaluated. Like many areas, this surveillance system was initially set up to detect CWD in the New York deer population, though no positive cases were detected in this dataset of complete necropsies or in the 54,000 primarily hunter-harvested deer tested since 2002 (Walsh and Miller 2010, Schuler et al. 2018). Given the data available, we determined that more females died from ‘other’ causes of death than males, which could be related to parturition as evidenced by dystocia and presence of a large or malpositioned fetus in several females (Galindo-Lean and Weber, 1994, Audigé et al. 2001, Zele et al. 2006). Fewer adults died due to nutritional and parasitic issues than juveniles, but only adults died from vitamin E/selenium deficiency although not all animals were tested for these deficiencies.

High mortality from hunting and predation in adults and juveniles is reiterated by many studies, however understanding non-hunter and non-predation causes of mortality is also important to assess herd health of wild deer populations (Van Deelen et al. 1997, Ballard et al. 1999, Delgiudice et al. 2002). Compared to other studies of deer mortality, this study provides detailed cause of death determination based on comprehensive specimen histories and postmortem examination, including both diagnostic testing and histopathology.

This retrospective analysis allowed us to evaluate the consistency and thoroughness of these diagnostic evaluations, and to highlight shortcomings that we will be rectifying moving forward. Despite concerted efforts, cause of death could not be determined for all animals. Common causes for inability to determine a cause of death included lack of information from the field to inform diagnostics or circumstances under which the animal was found, such as an advanced state of decomposition or missing tissues, or inappropriate samples submitted for laboratory testing. Missing information from the field was particularly difficult in neonate death because diagnoses are based on lack of evidence of underlying disease in the context of environmental conditions or no evidence of feeding. In situations where necropsies are to be performed by field personnel if sending the entire animal to the diagnostic laboratory is not possible, biologists should be trained in how to provide relevant history, proper sample collection, including documentation of abnormalities either photographically or in written form, and tissue fixation. It is incumbent upon diagnosticians to make this training readily available to field biologists. Likewise, diagnosticians should consider standardized diagnostic panels and tissue collection by species to ensure consistent data collection across laboratories and a wider suite of differentials, which would facilitate comparison and statistical analyses. Additional laboratory diagnostic testing, such as bacteriology and virology, was useful in determining the cause of death of an animal. Autolysis, scavenging, and artifacts due to freezing can hinder laboratory and histologic analysis, interpreting all results in combination greatly enhances diagnostic certainty.

Though histopathology has limitations, particularly with regards to autolytic change, it is an essential tool in any disease monitoring program. In cases where laboratory testing yields no or equivocal results, histopathology can characterize the nature of the lesions and provide a probable etiology based on morphology. When laboratory testing yields a positive result, histopathology can be used to confirm the significance of the result. Histopathology may also detect agents or lesions suggestive of agents that are not part of routine laboratory screening panels, and can thus play an important role in recognizing emergent diseases. These points are illustrated by the findings of fungi in pneumatic lesions and detection of herpesvirus inclusions and Apicomplexa in tissues that would have been missed without inclusion of histopathology (Duncan Jr et al. 2000, Kleinboeker et al. 2002). Given the frequency of bacterial infections in this dataset, we recommend aerobic culture in most cases where lesions are consistent with a purulent exudate, as the majority of significant organisms in this study were aerobic organisms. However, the presence of Fusobacterium spp. indicates anaerobic culture is warranted in cases where oral or esophageal lesions are present, or when necrotizing, malodorous lesions are present in any organ. Though fungal agents were not a common cause of disease in our group, pneumonia is of particular interest as the caseous, nodular, firm lesions are also compatible with mycobacterial infections. Though it is rare, cases of bovine tuberculosis have been documented in humans after exposure to infected deer (Sunstrum et al. 2019). Given the regulatory significance of mycobacterial diseases, lesions such as these should always be submitted for examination for acid-fast bacteria, though fresh or frozen tissues are necessary to perform follow up culture or molecular testing in the case of a positive test.

Viral infections, though rare in this study, are significant due to zoonotic potential. Although the frequency of rabies was low, it was greater than expected, as deer are not a traditional rabies vector (Petersen et al. 2012). Possible human exposure is significant in the public, particularly hunters. Similarly, West Nile virus, another zoonotic agent, was also found in the population, though deer are not a known source for West Nile transmission to humans (Colpitts et al. 2012). Thus, personnel handling tissues of white-tailed deer should be aware of these zoonoses; biologists handling wildlife life should be vaccinated for rabies and wear appropriate personal protective gear (Bosch et al. 2013, Tarrant et al. 2020).

We found female white-tailed deer faced additional risks due to pregnancy, pregnancy-related nutritional deficiencies, and parturition, which could partially explain the increased frequency of female animals in the mortality category designated ‘other’ (Bishop et al. 2009). Nutritional issues were common in young animals, and starvation/emaciation represented over half of the nutritional cases. Nutritional deaths were less frequent during spring and summer, which is logical given scarce food and harsh weather in New York between January and March (Verme 1969, Mautz 1978).

Wild, free-ranging species, such as white-tailed deer, present unique challenges in diagnostic evaluation for cause-of-death determination. Importantly, response to public inquiries and recovery of carcasses from the field in a timely manner by wildlife agency can be limited by staff availability and training. Access to appropriate storage and distance to a trained diagnostician can also contribute to sample deterioration in carcasses that often have an unknown time of death. Once at a lab, many diagnostic tests are not optimized...
for wildlife species or for novel pathogens and can be further hampered, along with histopathology, by autolyzed tissues. Because our cases were collected through passive surveillance and focused on abnormal and unknown circumstances, this study may not be completely representative of the entire deer population in New York and is subject to selection bias. For example, trauma may be underrepresented in this study since people are less likely to question the cause of death of an animal found alongside a road, and the surveillance plan instructs field staff not to submit deer that died directly as a result of vehicle collisions. Previous studies on death in free-ranging white-tailed deer relied primarily on tracking collars, which provides faster detection of mortality (Schuler et al. 2018). Predators may remove diseased deer from the landscape before they can be recovered by humans, thus biasing postmortem estimates of mortality sources (Schuler et al. 2018). New York is home to mid- and large-sized predators such as black bears, coyotes, and bobcats that could have killed or scavenged deer, skewing our study population, especially in fawns and juveniles (Joyner 1983). Other studies of death in captive deer also documented a high frequency of bacterial deaths. These results have lower relevance to wildlife management agencies because bacterial infections in captive animals may be the result of overcrowding and/or poor management (Hattel et al. 2004, Haigh et al. 2005).

Severity of illness may have also biased our sample population, because clearly abnormal animals are more likely to be noticed by the public and submitted for analysis. A similar bias applies to time of year, because deer may be less likely to be encountered in the field outside of hunting season. Because animals were classified by primary death diagnosis, this might not reflect multifactorial causes of death since animals were often found with multiple issues. It is not possible to determine if a starved fawn with a bacterial infection was infected before the adult female withdrew milk, if the bacterial infection caused the fawn to stop nursing, or if the infection occurred after nursing ended. Therefore, studies of radio-collared fawns may combine mortalities due to apparent disease and starvation as sickness-starvation (Haskell et al. 2017). Interpretation of complete post mortem results by an attending pathologist familiar with wildlife is vital in making the best determination of the primary cause of death.

Management implications

This study represents a snapshot in time for the New York free-ranging deer population and the effectiveness of DEC, Cornell, and surveillance efforts to find and diagnose causes of morbidity and mortality in white-tailed deer. We have identified areas of improvement are necessary in recording data and further development of our system to evaluate causes of morbidity/mortality in free-ranging species. By providing a baseline assessment, diagnosticians and biologists can prioritize differentials and subsequent testing most likely to yield useful results in diagnosis. More states are conducting disease surveillance in wildlife, particularly for CWD in white-tailed deer, so baseline probabilities of common deer maladies are important for wildlife veterinarians and biologists to consider in their necropsy assessments. The introduction of emerging pathogens may be detected with routine surveillance, as part of a larger goal to track population health trends over time.

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Conflicts of interest – The authors have no competing interests to declare.

Data availability statement

Data are available from the Dryad Digital Repository: <http://doi.org/10.25338/B89D1S> (Zhu et al. 2021).

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