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Research Communication

Finding the pulse of healthy native freshwater mussels (order Unionida): Insights from propagation biologists

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ABSTRACT

Objective: This study engaged representative native freshwater mussel propagation biologists in focus group discussions to learn their views and priorities related to the development and adoption of freshwater mussel health metrics. It provides a case study on how early consultation with practitioners can better inform research questions and improve imperiled species conservation.

Methods: Two focus group discussions were conducted with freshwater mussel propagation biologists from across the United States to understand the needs of conducting mussel health assessments, current approaches, technical capacity, and where advances may be most beneficial.

Results: Propagation biologists identified how they currently measure freshwater mussel health, listed the largest threats to health, explained similarities among approaches, and made recommendations for future research.

Conclusions: Propagation biologists called for quantitative metrics that complement current procedures that could be measured nonlethally and were reflective of health and resilience rather than presence or absence of disease. Further conservation research could benefit from early engagement with propagation biologists to better ensure adoption of research products and tools.

KEYWORDS: conservation, focus group, unionid

LAY SUMMARY

This study explored the challenges and needs of assessing the health of freshwater mussels, a group of highly imperiled species. We conducted focus group discussions with mussel propagation biologists who highlighted the need for standardized health metrics to avoid health declines.

INTRODUCTION

Unionid mussels (class Bivalvia) comprise a highly imperiled, species-rich, environmentally important family of freshwater mussels facing enigmatic die-offs and widespread population declines (Graf & Cummings, 2007; Haag, 2010; Vaughn, 2018). Of the approximately 300 species in North America, 10% have gone extinct in the past century. An additional 65% are in some state of imperilment and are only represented by an insufficient number of fragile individuals persisting in isolated or captive populations (Haag & Williams, 2014).

Direct intervention has gained momentum in response to the urgent need for protection, management, and recovery of mussels (Patterson et al., 2018). Propagation, augmentation, and reintroduction of mussel populations can serve to recover extirpated species or isolate populations from impending impacts (Araujo et al., 2015; Peck et al., 2014; Strayer et al., 2019), and state and federal recovery plans increasingly include imperiled species propagation in necessary conservation actions (U.S. Fish and Wildlife Service, 2019).

Freshwater mussel propagation is an effective management strategy that began over 100 years ago, with rapid advances in measured and assumed success based on production rates and recapture surveys (Aldridge et al., 2023). Propagation facilities serve as a frontier for research into mussel autoecology and population dynamics while conserving diversity, but standardized practices and goal assessment lag the propagation of other fauna (Rytwinski et al., 2021; Strayer et al., 2019). Propagation, augmentation, and reintroduction impact assessments on genetic health and diversity have been proposed, but incorporation of health and fitness metrics, particularly beyond expected behavior or the presence and transport of potential pathogens, is severely lacking (Jones et al., 2006).

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This case study used focus group discussions to engage representative mussel propagation biologists to gain a better understanding of their experiences with freshwater mussel health in a propagation setting to inform future research priorities and to develop applied tools and techniques for quantifying mussel health. Discussions provided an in-depth exploration of the demand for mussel health assessments and current approaches and identified topics where advances toward mussel health metrics may be most beneficial for conservation.

Conservation and biodiversity research has relied extensively on qualitative research methods to make inductive inferences from collective ideas about complex topics that would otherwise not be captured with quantitative methods (Roller & Lavrakas, 2015). Focus groups can integrate social and biological knowledge for more effective, adaptive decision making. This includes identifying shared objectives, measuring the value of new standardized programs, evaluating limitations, weighing consequences, and contextualizing differences (Nyumba et al., 2018) Accounting for these differences decreases the risk of opposition among resource managers to researchers' recommendations and places management activities on a timeline that reflects the urgency of implementation. Further, researchers and practitioners can share information that might not be well documented in published literature.

Table 1. Demographics of the focus group participants.

Characteristic	Number (percentage) of participants
Gender	
Male	5 (42)
Female	7 (58)
Facility affiliation	
Academic	2 (17)
Federal	3 (25)
State	6 (50)
Tribal	1 (8)
Facility region	
Midwest	3 (25)
Northeast	2 (17)
Pacific	1 (8)
Southeast	6 (50)
Highest education	
Bachelor's degree	2 (17)
Master's degree	6 (50)
Doctorate	4 (33)
Focal propagation species	
Only threatened and endangered	1 (8)
A mix of common and threatened and endangered species[a]	11 (92)

[a]Some common species are considered imperiled but are not listed yet.

METHODS

Mussel propagation biologists were recruited to participate in two focus group discussions. Criteria for participation included conducting freshwater mussel propagation, routinely engaging in hands-on interactions with mussels, and having the authority to make decisions relevant to mussel heath assessments within their programs. Individuals were identified based on prior knowledge of employment and expertise, with care given to convene mussel propagation biologists from across geographic regions of the United States and representative of different affiliations (state agency, federal agency, tribal community, and academic institution). Potential participants were personally invited via email to participate in a conversation about perceptions related to mussel health, mussel health metrics and monitoring, and necessary research initiatives.

The facilitator used a semi-structured guide with questions related to participants' experiences and recommendations. Focus group participants shared current techniques and operating procedures as well as opportunities and barriers to adopting additional metrics. Study participants consented to participate using a protocol approved by the Institutional Review Board of North Carolina State University (Protocol Number 25873).

The two discussion sessions lasted approximately 90 min each and were audio-recorded and transcribed. The research team convened immediately after each focus group to debrief to summarize major elements of the discussion. We took a systematic approach to analyzing the discussion transcripts; participants' responses captured in the transcripts were assigned codes from a codebook. The first two authors developed a codebook from (1) a priori codes, or expected concepts, derived from research questions and the discussion guide (DeCuir-Gunby et al., 2011) and (2) emergent codes, or concepts that arose during the conversations, derived from a line-by-line reading of the transcripts (Saldaña, 2009). The codebook consisted of 11 code families (e.g., Health) and 21 total codes (e.g., positive health, negative health); codes within code families shared common concepts or themes. The first two authors co-coded a subset of a transcript in a series of iterative meetings to ensure intercoder reliability, reconcile any differences, and adjust a priori codes. The first author coded and analyzed the remaining transcripts and confirmed interpretations through review by the other authors. Codes and prevailing themes were then compared within and between focus groups. Exemplary and representative quotations of frequently expressed views are reported.

RESULTS

This case study engaged 12 mussel propagation biologists in two focus group discussions. The first focus group included two propagation biologists who helped refine questions and delivery, and the second focus group included 10 propagation biologists. The participants had an average of 16 years of experience (range = 1–30 years) in the profession. They represented propagation facilities that had at least two permanent full-time staff members (average = 3; range = 2–6). Additional participant demographics, including affiliation, region represented, and education, are reported in Table 1.

Three major themes emerged from the focus group discussions related to current freshwater mussel health assessments and future needs (Table 2). Participants described how they record indicators of health and disease, shared concerns about threats to health, and compared their approaches across facility maintenance and conditions. They also identified priorities for future research that would aid in the development of health indicators, tools, and methods that are accessible to and relevant for mussel propagation facilities.

Table 2. Code families, codes, and exemplary quotations from focus group discussions with freshwater mussel propagation biologists.

Code family and code	Exemplary quotation(s)
Health	
Positive	"Active cilia movement in young juveniles, intense cilia movement with brooding females once the glochidia have been extracted, especially in *Lampsilis* that the glochidia are fully developed and whole. They're usually fully fertilized, which you know, are stable populations." (participant 8 [P8])
Negative	"In a sick adult, especially more, but almost on their mantle tissue, it starts to pull away from the shell. Once it completely pops off both sides, they're dead. That's it." (P2)
Approach	
Preventative	"We do all of our culturing in these beakers, and so we don't use any pond water or any kind of filtered water. All our water is 100 hard [*sic;* 100 mg/L hardness] that we create in tanks. Everything that we do, all of our systems, is pristine conditions. Everything is super clean. So, I think that helps a lot of our survival and growth." (P1)
Monitoring	"We're always making notes on how many external other types of animals are attached to their shells or maybe attaching to the live tissue." (P3)
Responsive	"You just leave them alone, and sometimes they seem to be able to come back from it." (P5)
Threats	"About 100 things will kill a juvenile mussel." (P4)
Biotic	"Well, we try to take extra precautions with biosecurity, like with the broodstock. We keep them separate. We disinfect water from, like before we release water from other places, we'll disinfect it first with bleach and neutralize it with sodium thiosulfate. So, we do try to be really cautious about biosecurity." (P10)
Abiotic	"We test the water quality almost every day because ammonia I think is a big problem, especially for the young, newly transformed, juvenile mussels. They're very sensitive to the ammonia." (P7)
Needs	"I think kind of our only really big wish list item would be to have some sort of mechanism to do a check-up, like an annual check-up on our brood mussels, especially the ones that we're planning to hold indefinitely without killing them." (P8)
Limitations	"Populations are so small and fragmented that we can't even source the mussels that we need." (P7)

Code family Health: "Your perfect, healthy mussel looks … alive."

Throughout the conversations, propagation biologists shared their perspectives on freshwater mussel health among the three life stages (glochidia [larvae], juveniles, and adults), including current protocols, metrics, and available tools. Comments were roughly balanced across juveniles or glochidia and adults or broodstock. Female adults were referenced more than males or adults in general, with additional emphasis on the connection between females and their brood. For example, a participant explained, "I see a big difference between the quality of juveniles coming from different females. … The condition of those larvae in the mom is fairly important" (participant 1 [P1]).

Participants discussed both indicators of fitness (or positive health) and of diminished health and disease (or negative health). Most participants reported currently using metrics that (1) rely on mussel behavior that is observed nonlethally, (2) are primarily qualitative, and (3) require experience to differentiate between health states. For example, one participant stated, "If you handle them a lot, you'll kind of get a feel for that. But quantifying that is going to be maybe a different story" (P2).

Propagation biologists described using both anatomy and physiology to grade mussel health, and most measurements were qualitative. There was agreement among the participants that the strength of the adductor muscle was a common metric of health used in adults, but all other comments about adults were framed as indicators of diminished health, including lack of burrowing and filtering activity, slow shell closure response to stimuli, diminished quality of glochidia produced from a female, and abnormalities observed on the mantle margin, foot, or apertures. The only common quantitative measurement mentioned was growth. One person mentioned glycogen content; however, they reported that they "haven't personally looked at [glycogen], but [they] worked with several people that have" (P3). Similarly, feeding and filtration rates were cited, but as one participant reported, they "think some people have worked on it, but we hear variation that what people say in an article is 20 L a day, is that every kind of species is like that, or is it one?" (P2). Another person also reported that they are "not an expert on the adults" and opted to discuss their experience with glochidia and juveniles (P4).

There was broad consensus that "early juvenile [growth] and survival" comprised the overarching goal of propagation (P1). The main test for glochidium health reported to be used among participants was the salt test for viability, but participants indicated that juvenile "heart rate [*sic*; heartbeat]" can also be quantified (P1). Participants expressed their beliefs that juveniles are assumed to be healthy if they are active, their guts are full, their crystalline style indicates feeding, and their cilia are moving.

Code family Threats: "How much time do you have?"

Threats to mussel health identified by participants could be categorized as biotic threats and abiotic threats. The described biotic threats included human handling, algal blooms, invasive species, and competition. Pathogens, such as flatworms, trematodes, parasites, fungi, bacteria, and viruses, were a common concern among participants, but one propagation biologist expressed that they "don't think we ever 100% know exactly what we're looking at" in terms of pathogens and "they've yet to connect it with disease" (P3). One individual said, "Of course, when you start looking, and you've never looked, you're going to find everything in the world under the sun is in these animals because freshwater is just full of that stuff and they're vacuuming it in" (P5).

Abiotic threats described included water quality measures, such as ammonia, temperature, alkalinity/pH, and calcium. Participants indicated that these conditions are prepared, monitored for, and adjusted based on species needs and mussel development goals.

Code family Approach: "There's not a uniform target across the different facilities"

Participants described propagation tactics focusing predominantly on accomplishing higher growth and survival, but captive management tactics varied by facility. Throughout the discussions, propagation biologists frequently used words such as "unique" (P1), "different/differences" (shared among several participants), and "the oddball" (P3) to describe their systems and facilities. Largely, the propagation biologists reported relying on preventing declines in health (38 unique mentions), with some tools to monitor their success (23 unique mentions). Tools and knowledge for responding to (or predicting) further problems once they started were limited (9 unique mentions), and participants emphasized that personnel time was a limiting factor to their approach.

Preventing declines to mussel health: "Don't let them get to that point"

Primarily, participants described preventive care that relies on planning and substantial investment of time. One participant said

> I can pre-plan and get my broodstock on site and ready to go. If you're not prepared, yeah, that's going to be a big problem for you. But if you know what you're doing, that shouldn't be a limiting factor unless the species is just sort of hard to find. (P1)

Participants spoke confidently about preventing declines by "monitoring our water quality all the time … can't say enough about having proper water quality" (P6), with shared water quality concerns among propagation biologists, but subsequent discussion revealed that accepted limits or desired optimums varied among participants based on the system, species, and source water. Similarly, propagation biologists expressed that there could be a trade-off between growth and survival that can be managed through controlling density-induced competition, but they reported that nominal mussel density differs according to the system.

Biosecurity, specifically flatworms and rotifers, was a shared prevailing concern among participants. They described that live parasite introduction is minimized by "never exposing from animals collected out in the field" (P1), containing broodstock in separate systems, and never mixing water sources that supply the rest of the hatchery system. Additionally, some propagation biologists indicated that they fully sterilize their sediment and keep their system and growth conditions in "pristine conditions" (P2).

Monitoring mussel health: "We want it to just be awesome in there and they grow"

Participants described that, once implemented, many of the preventive measures require consistent monitoring of water and mussel quality. Some mussel characteristics that participants indicated are or can be monitored include growth, survival, weight, behavior, glycogen, gamete production, feeding rate, and filtration rate. Participants reported that feeding and filtration rates are measured with Coulter (particle) counters. One propagation biologist asked, "I've tried for a long time to get at and never really succeeded: How many cells (of food) should go into a recirculating system?" (P7). Participants revealed that visible threats are also monitored, including trematodes, flatworms, and fungi. Some biologists reported employing histology to confirm parasitism or impacted physiology, but participants generally expressed reluctance to kill mussels for analysis.

Responding to declines in mussel health: "You cry"

Propagation biologists spoke least about how to respond to health problems. Even when asked directly about how they respond when health problems start, their initial comments included "You cry" (P8) and "Sometimes it's not even science" (P2). Responses then returned to discussing preventive measures. Participants described some common interventions, including isolating the sick mussel, treating fungi with fluconazole, or changing the substrate. Otherwise, the propagation biologists reported that they "stop, drop, and reset" (P3), "leave them alone" (P6), or "keep an eye on things … because if they get sick … you're not going to come back" (P1).

Research needs

Research needs were emphasized for future health monitoring, including "a mechanism to do an annual check-up on brood mussels" (P7), "protocols for sending animals off and having them checked periodically" (P8), "check the health of mussels before we put them out" (P9), and a "standard way to evaluate each of the facilities with equal metrics" (P5).

Participants identified some fundamental questions that are still unanswered in freshwater mussel care. For instance, a basic definition of healthy baselines is still not established, as reflected by one propagation biologist: "What does healthy mean? What does abnormal or what does unhealthy mean, and defining that more clearly, and giving us the tools to help us understand what that means" (P9). Several participants described the lack of nutritional studies or assessments based on indices generated by morphology (length, width, height, and weight), growth curves, or metabolism. Participants described that the complexity in natural systems and often latent mortality of mussels confound the ability to diagnose issues, causing uncertainty about the combined effects of reintroduction sites and propagation techniques. One propagation biologist said,

> We need to identify and describe the threats and understand the role, their prevalence and their role in the systems, and then we need to be able to translate that into something meaningful so it can be worked into management. (P9)

Participants explained that further complicating the widespread application of mussel health indicators is the evident species-specific variability. One propagation biologist sternly articulated, "We have to accept that as a group that they may all have different best ways to raise them" (P1). Another participant agreed: "Sometimes we're guilty too, even as experts, lumping them all as one. They're all different species" (P2). Additionally, participants raised concerns about poorly understanding the intrinsic year-to-year variability of brood quality.

Participants overwhelmingly preferred nonlethal, noninvasive methods to assess mussel health, though some were still reluctant to interrogate females or withdraw hemolymph. However, some participants indicated that they would consider withdrawing hemolymph. For example, one participant stated, “Eventually maybe it'll be like humans. You do bloodwork and bam, this one's got this, this one's got that” (P3). Participants stressed that health metrics need to be connected to long-term survival; those that care for imperiled species must report and justify any losses.

Specifically, propagation biologists were interested in the mussel microbiome; the presence or absence of other organisms, especially the utility for developing therapeutics in response to declining health; and ways to assess fitness and resilience to disease. They also shared their perceptions of the benefits of collaborative endeavors with experts. Participants agreed that other propagation biologists serve as a repository of wisdom and knowledge by “sharing information [with] … all these people here” (P4). External experts identified by participants included veterinarians and other scientists, mostly associated with academia. In total, 23 other people were specifically named by participants as partners or as having completed informative studies, with the caveat that some studies have begun but propagation biologists are still waiting for “proof” of pertinence.

DISCUSSION

The focus group discussions reported in this case study represent an effort to engage practitioners in the formulation of future research questions, tools, and methods to better assess and promote freshwater mussel health. We found that underutilized opportunities exist for applied research to be guided and improved by actively engaging with mussel propagation biologists early in the development process. Further, focus group discussions like those reported herein can provide new and robust repositories of information with less resource investment on the part of researchers than other methods. Engaging practitioners early in the research process likely increases the willingness to share information across facilities and the likelihood of adopting research products and outcomes.

Through focus group discussions, we explored the knowledge and beliefs of freshwater mussel biologists from across the United States and found them to be attentive to freshwater mussel health and receptive to health metric advances, with stipulations. For example, given the imperiled status of the organisms they work with, mussel propagation biologists emphasized that while current advances in histopathology and biomarker monitoring are valuable (McElwain & Bullard, 2014), there is a need for nonlethal, noninvasive techniques for any new health assessment tools and techniques. Moreover, the limited manpower and uniqueness of approaches among the facilities highlighted the need for future health assessment procedures with low demands on employee capacity, that are complementary to their current protocols, and that are relatively inexpensive to improve feasibility. We learned that prevailing qualitative measurements, which are reliant on experience, introduce subjectivity in mussel care and generate an urgent need for standardization across facilities that can be taught, transferred, and learned by those entering the profession, especially as pioneers in propagation contemplate retirement or employment transition.

Our findings show that propagation biologists currently rely on monitoring protocols for the presence of disease, such as flatworms or fungi, that would negatively affect growth or survival. Thus, there is an opportunity to monitor for—and improve mussel resilience and durability to—a range of threats. Propagation biologists actively participate in and lead studies for constructing and maintaining hatchery facilities; advancing juvenile transformation; and understanding mussel ecology, reproduction, life history, and toxicology. However, conversations towards measuring mussel health and fitness in the absence of obvious disease are less formally documented or the methods are not readily accessible (Augspurger et al., 2007; Geist et al., 2021; Lopes-Lima et al., 2014; Patterson et al., 2018; Popp et al., 2018; Ryan et al., 2022).

While qualitative methods, such as our case study with focus groups, provide a richer understanding of propagation biologists' priorities and willingness to implement new protocols, they also have several limitations. For example, we included most of the major propagation facilities across the United States in this study, but we conducted only two focus group discussions and cannot confirm saturation (i.e., more discussions might have yielded additional themes). Additionally, findings and interpretations might not be generalized to other settings, conditions, and biologists, particularly those in other countries. Propagation biologists provided valuable perspectives that were occasionally shared across facilities and illuminated areas where differences could be identified.

Planning for advances in conservation: “What if it's the last one on Earth?”

The extensive declines and loss of freshwater mussel populations are expected to continue, although the impacted species, geographic area, or driving perturbation is difficult to predict (Haag & Williams, 2014). Taxonomic shifts, emerging threats, catastrophic events, and changing climate (McIver et al., 2023) could exacerbate the prospects of finding sufficient broodstock to maintain populations. In this case study, we found that mussel health and resilience should be considered for broodstock, propagated individuals, and wild populations beyond absence of disease.

Participants reported that difficulty in finding broodstock creates a significant bottleneck to successful propagation. Contrary to some recommendations, participants highlighted that propagation should be considered a first-line defense with resources committed to maintaining whole communities, including currently “common” species, with attention given to recommendations for assessing augmentation success (McMurray & Roe, 2017; Smith et al., 2015; U.S. Fish and Wildlife Service, 2019). They also urged that expediency should be factored into all future plans to combat any impending future losses of these rare species.

In summary, we learned from our case study of focus groups with mussel propagation biologists that they care deeply about improving the health and fitness of the organisms they are tasked to conserve and that they are open to implementing new knowledge and tools from researchers under certain conditions. Future research on mussel health would benefit from early and frequent consultation with individuals involved in propagation during proposal development and study design.

DATA AVAILABILITY

All data are available from the first author or corresponding author upon request.

ETHICS STATEMENT

This research meets all ethical guidelines and legal requirements. Study participants consented to participate using a protocol approved by the Institutional Review Board of North Carolina State University (Protocol Number 25873).

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CONFLICTS OF INTEREST

The authors declare no conflicts of interest.

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