Biomedical bleeding may impact horseshoe crabs' spawning behavior and movement

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Horseshoe crabs that have undergone biomedical bleeding tend to reside in deeper water and approach mating beaches less often, according to a new study published in *The Biological Bulletin*. In "Effects of the Biomedical Bleeding Process on the Behavior of the American Horseshoe Crab, Limulus polyphemus in Its Natural Habitat," Meghan Owings and her colleagues report the results of an investigation of the behavioral and physiological effects that the bleeding process has on horseshoe crabs that are released back into their natural environment. The findings suggest biomedical bleeding may impact the reproductive output of female horseshoe crabs during the season in which they were bled.

Horseshoe crabs are harvested by the biomedical industry in order to create Limulus amebocyte lysate (LAL), which is used to test medical devices and pharmaceutical drugs for endotoxins. During the process, around 30 percent of a crab's blood is extracted before it is returned to its natural environment.

Owings' study is one of the first to examine the behavioral impacts that the bleeding process has on the horseshoe crabs once they are returned to the wild. "With the growing demand for LAL as the global population expands, medical advancements improve, and medical needs increase, it is critical to understand the consequences of the biomedical bleeding industry on horseshoe crabs' fitness and population dynamics," Owings writes.

Owings and her coauthors retrieved 28 horseshoe crabs from a spawning site in the Great Bay Estuary in New Hampshire. Half of the crabs were randomly selected to undergo the bleeding process and then all of them, both bled and controls, were fitted with acoustic transmitters and released where they had been captured. The transmitters were used to monitor the horseshoe crabs' movements, depth, and times when they were active. Data from the transmitters were logged by an array of acoustic receivers that were set up throughout the estuary.

All 28 horseshoe crabs were successfully tracked in the Great Bay Estuary from May 15 to December 6, 2016. Data was also obtained from 23 of the horseshoe crabs the following year, from April 14 and October 4, 2017.

The impact of biomedical bleeding on the horseshoe crabs was analyzed in terms of spawning activity, biological rhythms, overall range of movement, depth, and activity levels, while also taking into account the natural shifts in their behavior that come with the changing of the seasons in the Great Bay Estuary.

The authors found that in the first week that the animals were released back into the Great Bay Estuary, bled animals appeared to spawn less than the control animals. The difference was especially pronounced in females, with control females appearing to spawn on average 4.8 times while bled females appeared to spawn on average 2 times.

Another notable trend was that in May and June of 2017, the bled animals did not approach shallower areas in Great Bay but remained in the deeper channels. This poses a problem, since May and June are the months when the horseshoe crabs typically move into shallow water to spawn in the estuary.

For a possible explanation, Owings and her colleagues mention two

previous studies, one that took place in a laboratory and one in the field in Cape Cod, that found that biomedically bled horseshoe crabs had disrupted orientation and movements that were more random than control crabs. This disorientation could prevent horseshoe crabs from finding spawning beaches, they write.

They also suggest that the recovery from the bleeding process—crabs take three to seven days to regain their blood volume, and up to four months for amebocytes to return to baseline levels—could cause crabs to simply not have as much energy to put towards spawning as they would normally have.

These results are especially troubling since females are already favored in the bleeding process due to their larger size. "If the bled animals, especially females, have alterations in their biological rhythms and mating behaviors, it is likely to further alter the sex ratio on spawning beaches, reduce reproductive output, lower population levels, and decrease the fitness and survival of this keystone species," they write.

The authors recommend that further research is needed to better understand the impact of biomedical bleeding, both in terms of spawning behavior and seasonal movements, so that improvements can be made to reduce both the lethal and sublethal impacts of the process.

More information: Meghan Owings et al, Effects of the Biomedical Bleeding Process on the Behavior of the American Horseshoe Crab, Limulus polyphemus, in Its Natural Habitat, *The Biological Bulletin* (2019). DOI: 10.1086/702917

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