# Limulus Spawning Activity on Delaware Bay Shores on June 5, 1993

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## Introduction

The horseshoe crab has existed for millions of years. The largest population of the American horseshoe crab, *Limulus polyphemus*, exists today in the Delaware Bay, but the numbers used to be larger. The baseline for abundance records in the Delaware Bay comes from harvesting reports in the mid 19th century and these contain high numbers. In 1880, for example, 4.3 million horseshoe crabs were harvested. The population declined sharply in the late 19th and early 20th centuries due to over harvesting for fertilizer and poultry feed.

Since then, the population has slowly rebounded, particularly when chemical fertilizers became available and commercial harvesting stopped. But the use of the horseshoe crabs for eel and conch bait, has revived concerns about the population and this has led to regulations adopted in Delaware in January 1992, and in New Jersey in May 1993, that limit the time and location of harvesting

Beginning in 1990, an annual census, a largely voluntary undertaking, has gathered information about the horseshoe crab population in the Delaware Bay to learn more about population trends.

#### **Ecological Value**

The horseshoe crab's fisheries value is supplemented by its significant ecological role and medical application. The ecological link between shorebirds and horseshoe crab eggs is of worldwide magnitude. The Delaware Estuary is the largest staging site for shorebirds in eastern North America. Every spring, millions of migratory shorebirds from the coasts of Brazil, Patagonia, and Tierra del Fuego, from desert beaches of Chile and Peru and from mud flats in Suriname, Venezuela, and the Guyanas stop at Delaware Bay and feed on horseshoe crab eggs (Bryant and Pennock, 1988). Many birds double their weight during the feeding frenzy along the Delaware bayshore before finishing the rest of their flight to their Arctic breeding grounds. Without the horseshoe crab eggs, the migratory bird population would be devastated.

#### **Medical Value**

The medical product derived from the horseshoe crab white blood cells detects pyrogens in pharmaceutical drugs. Pyrogens are fever-inducing substances and the most significant pyrogens for the pharmaceutical industry are bacterial endotoxins. Endotoxins are high molecular-weight proteins associated with the outer membrane of gram-negative bacteria. Endotoxins are harmless to humans except when they are in direct contact with the blood stream. In the blood stream, endotoxins are extremely toxic; causing fever, shock, hemorrhage and even death. In the mid-1920's, studies conducted on rabbits proved that fevers associated with intravenous solutions were the result of bacterial products. This discovery enabled pharmaceutical companies to implement procedures to prevent contamination of their intrave-

nous solutions. In 1942, the Food and Drug Administration (FDA) incorporated the first official rabbit pyrogen test in the United States Pharmacopoeia (USP) guidelines.

The test involves measuring the rise in temperature of rabbits after an intravenous solution is injected into their marginal ear vein (Pearson, 1985).

For 37 years, the rabbit test was the only practical pyrogen test, however the need for a simpler, more accurate test was recognized.

In 1964, Dr. Frederick Bang and Jack Levin discovered that the material inside the white blood cells of the horseshoe crab could detect small quantities of gram-negative bacteria. This material, called Limulus Amebocyte Lysate (LAL), was investigated for use in the pharmaceutical industry as an alternative to the rabbit pyrogen test (Pearson, 1985). In 1979, the FDA issued draft guidelines for the use of LAL as an end product pyrogen test for endotoxin in medical devices and injectable drugs.

test for endotoxin in medical devices and injectable drugs. Effective in May 1993, the bacterial endotoxin test in the United States Pharmacopoeia was revised to accord with the 1987 FDA final guidelines for the use of LAL. The simplest procedure using LAL is the gel clot method. A small amount of the drug to be tested is incubated with a small amount of LAL for one hour. The test is positive if the mixture coagulates. The LAL test is more accurate, more sensitive, simpler and less expensive to perform than the rabbit test.

The horseshoe crab population in Delaware Bay is important to our modern day existence and continued monitoring of their population trends is necessary to protect them. Four factors could negatively affect the population: loss of suitable spawning habitat due to bulkheading, jetties, erosion of the shoreline, etc.; overfishing of the crab for eel and conch bait; incidental loss of horseshoe crabs due to dredging; and oil spills in Delaware Bay.

# Methods

The fourth annual horseshoe crab census was conducted on June 5, 1993. Volunteers assigned to beaches along the Delaware bayshore counted the number of crabs spawning during the AM and PM high tide. A landmark stake was located at each beach, and each volunteer was given a 100 meter string which was stretched along the beach to the left and right of the landmark stake. The survey method used depended on the abundance of spawning crabs. The surveyor could either count the individuals along the entire 200 meters (moderate density) or in three randomly selected five meter segments within the 200 meter strand (heavy density). A rapid survey of the entire beach was also conducted by

identifying the density of crabs every 15 meters: zero, moderate, heavy and maximum densities.

To obtain the estimate of the Delaware Bay spawning population, the average number of crabs within five meters was calculated for each surveyed beach. This number was then multiplied by the number of five meter segments along the spawning length of the beach. Each beach estimate was then added to obtain the final estimate of the spawning population along the Delaware bayshore. The rapid survey estimate was obtained by adding the number of crabs for each density identified in the 15 meter area.

Table 1. Census Data Summary 1990 - 1993

	June 8, 1990	May 25, 1991	May 16, 1992	June 5, 1993
Estimate of Spawning Individuals	1,240,679	1,224,771	399,147	394,039
Percentage of Spawning Individuals in DE	19	43	28	88
Percentage of Spawning Individual s in NJ	81	57	72	12
Individuals During AM High Tide*	317,399	318,153	133,952	16,846
Individuals During PM High Tide*	923,280	906,618	265,195	377,193
Ratio of Individuals PM vs. AM Tide	2.91	2.85	2.04	22.39
Beaches Surveyed in Delaware	22	15	13	9
Beaches Surveyed in New Jersey	13	8	9	9
Ratio of Male Individuals to Female	2.19	2.64	3.13	2.49
Ratio of Male to Female Delaware	1.76	2.78	2.12	2.77
Ratio of Male to Female New Jersey	2.62	2.51	4.14	2.29
Main Spawning Beaches in Delaware	Cedar Beach	North Bowers	Pickering	Big Stone
	South Bowers	Kitts Hummock		South Bowers
	Kitts Hummock	Pickering		Kitts Hummock
	Pickering	Port Mahon		Pickering
	A CONTRACTOR OF THE	Kelly Island		Port Mahon
Main Spawning Beaches in New Jersey	Cooks Beach	Fortescue	Kimbles	Kimbles
	Highs Beach	Moores Beach	Highs Beach	Highs Beach
	Moores Beach	East Point		
	Thompsons			
	East Point			

\* The AM tides were during daylight hours and the PM tides were at night; all shoreline surveys were made at the time of peak high tide.

## Results

The 1993 census was conducted on June 5th during the morning high tide (9:59 am at Breakwater Harbor, Delaware) and the night high tide (10:18 pm). Eighteen beaches were surveyed, nine in New Jersey and nine in Delaware. The New Jersey beaches were North Cape May, Norbury's Landing, Rutgers Marine Lab, Highs, Kimbles, Cooks, Reeds, Fortescue and Sea Breeze. The nine beaches in Delaware were Broadkill, Prime Hook, Big Stone, South Bowers, Bowers, Kitts Hummock, Pickering, Port Mahon and Kelly Island. In addition to the June 5th date, some beaches were surveyed on other dates (May 20th, 29th, 30th, 31st, June 3rd, 19th, 21st, July 2nd and July 3rd).

The estimated number of spawning crabs on June 5th was 394,039 individuals. Eighty-eight percent of the crabs were

found spawning on Delaware beaches, only 12 percent were found spawning on the New Jersey side. The number of crabs spawning on Big Stone beach in Delaware was estimated to be 243,458 during the night high tide. This number constitutes the greatest proportion of the estimated total count. The number of crabs spawning at night was considerably higher than in the morning.

The results from the 1993 survey are listed in Table 1 with the results from the previous three years. The 1993 estimated spawning number is the lowest of all four years. The rapid survey method resulted in a lower estimate than the random sampling method. The 1993 percentage of crabs spawning on the Delaware side was higher than the New Jersey for the first year. The ratio of male to female crabs was 2.49 males to one female.

#### Discussion

The low number of spawning individuals in 1993 can be partially attributed to a week of stormy weather preceding and on the census date, when strong winds (15-20 mph) blew along the Delaware side of the bayshore and the northern side of the New Jersey bayshore. The high winds created a rough surf which may have caused the crabs not to spawn. The Delaware PM observations showed less wind and calmer waters. The southern New Jersey side exhibited calm waters during the AM count. Light rain, thunder and lightening were occurring during the PM count.

Wave action caused by winds governs the spawning activity of the crabs; rough waters will deter crabs from coming ashore. The surveys conducted on other dates when the weather was not stormy, however, also revealed low numbers. There was no indication of any estimates (except at Big Stone beach) resembling the numbers found during the 1990 (June 8, 1990) and 1991 (May 25, 1991) censuses.

For the 1994 census, certain beaches will be surveyed closest to the new and full moon phases (May 7th, June 11th and June 25th) to supplement the one day census count (May 21, 1994). The survey will also include estimating the age of the adult spawners. Over time, probably at least six to ten years,

a survey of the age groups (year classes) of females participating in a spawning season should provide an index to the "health" of that portion of the total adult population. Surveying the age of the males would provide a baseline reference since they are harvested less frequently.

#### Age Categories for the Adult Spawners

(Note: Adults begin spawning at 9 to 11 years; the years below are the years beyond this.)

- 1 2 years clean, lustrous carapace;
- 2 6 years moderate erosion of carapace usually with epibionts;
- 6 plus years carapace nearly or entirely blackened with epibionts.



Horseshoe Crabs Spawning on Thompsons Beach, NJ, 1989.

# Summary

Improvements have been made in each of the four years that the census has been held. The census was designed to obtain a baseline estimate of the spawning crab population. The numbers from the census suggest that spawning individuals have decreased considerably over the four years. Low numbers can be attributed to weather conditions, mainly wind direction and velocity. Two consecutive years of low estimates of spawning individuals (1992 and 1993), make the results of the fifth (1994) horseshoe crab census instructive for revising existing and proposed fisheries management regulations.

The Delaware Bay volunteer horseshoe crab census is a pioneer study and a large undertaking. Emphasis has been placed on using methods that would enhance the statistical value of the estimated numbers. An important additional source of data are the trawl surveys conducted by the Delaware Division of Fish and Wildlife in Delaware Bay during April to December for the years 1990 - 1993 (Table 2). The 1992 and 1993 trawling data revealed approximately fifty percent fewer crabs than the previous two years. Both the census and the trawling data have revealed decreases in the Delaware Bay horseshoe crab population.

The male to female ratio is another important piece of information that is obtained from the census and the trawl

Table 2. Delaware Division of Fish and Wildlife, Trawl Data Summary 1990 - 1993

Number of horseshoe crabs collected in 30-foot otter trawls in Delaware Bay (Jeff C. Tinsman and Stewart F. Michels, DNREC; personal communication).

Estate of the same	1990	1991	1992	1993
Males	537	498	203	277
Females	463	541	202	172
Immature	21	17	7	6
Unnclassified	0	0	1	0
Total	1021	1056	413	455
Male to Female Ratio	1.16	0.92	1.00	1.61

surveys. Since the male to female ratio obtained from the census survey is biased due to the mating behavior of the horseshoe crabs and tends to exaggerate the number of males, the trawl data (Table 2) provides a better clue to the ratio of male to female and the number of adult females. The 1993 trawl data shows an increase in the male to female ratio from approximately 1:1 to 1.6:1. A marked decrease in the number of females may be the first sign of a population decline.

(see "Summary," page 4)

("Summary," from page 3)

The volunteer census has been successful in educating the public about the horseshoe crab and in assisting management agencies by producing a data bank on the spawning horseshoe crab population. Fisheries management personnel from Maryland and Virginia have expressed interest in implementing the census project along their coastal waters. Concern about the horseshoe crab and the information provided by the Delaware Bay census has inspired other states to monitor and protect their horseshoe crab populations. The census is creating a horseshoe crab population data bank and a new interest in the environment!

#### Acknowledgements

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Benjie Lynn Swan is involved in research of the horseshoe crab and manufactures a medical product from its blood.

William R. Hall, Jr. is a marine education specialist with the University of Delaware Sea Grant Program.

Carl N. Shuster, Jr. is an adjunct professor of biological marine science at the Virginia Institute of Marine Studies at the College of William and Mary. He has studied the horseshoe crab for over 50 years.

#### The Horseshoe Crab Hotline

This year, the horseshoe crab census survey will be implemented along the shores of the state of Maryland. Since the horseshoe crabs spawning on the shores of New Jersey, Delaware, Maryland and Virginia are believed to be one population of horseshoe crabs along the Atlantic Coast, the data collected from the Maryland beaches will aid the overall census. A horseshoe crab hotline was established by the State of Maryland Department of Natural Resources to obtain public information on the spawning areas of horseshoe crabs within the Chesapeake Bay and coastal bays of Maryland. Please call (410) 974-2241 and ask for Tom O'Connell if you have horseshoe crab information.

#### References

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#### 1994 Census Announcements

The date selected for the 1994 census is Saturday, May 21st. The daylight savings times of the high tides at the reference station for Delaware Bay (Breakwater Harbor, DE) are 5:32am and 6:09pm. The further up-bay, the later the time of occurence of the high tides.

Four preparatory workshops will be conducted: (1) Sunday, May 1st, at 1:00 pm in the Agriculture Extension building of Delaware State College, Dover, DE (2)Saturday, May 7th, at 2:00 pm in classroom 1 of the Thomas H. Kean New Jersey State Aquarium, Camden, NJ; (3) Monday, May 9th, at 7:30 pm in the Emergency Operation Center, 100 Airport Rd. in Sussex County, Georgetown, DE. (4) Thursday, May 19th, at 7:00 pm in the Nature Center of Cape May, 1600 Delaware Ave., Cape May, NJ.

For additional information call or write: Benjie Lynn Swan (Limuli Laboratories, 7 Bay Avenue, Dias Creek, NJ 08210 or (609) 465-6552) or William Hall (Sea Grant Program, College of Marine Studies, University of Delaware, DE 19958-1298 or (302) 645-4253).



A view of a large holding pen on the Delaware Bay shore of Cape May, New Jersey. This pen, estimated at 1.8 meters high x 3m wide x 24 m long, could have held over 18,500 horseshoe crabs. Harvested crabs were stored in many such pens, of all sizes, until ready for processing into fertilizer. (Source: Fowler, Henry W. 1908. The King Crab Fisheries in Delaware Bay. Amnual Report, New Jersey State Museum, 1907).