Abundance of horseshoe crabs (*Limulus polyphemus*) in the Delaware Bay area

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In recent years, increasing commercial landings of horseshoe crabs (*Limulus polyphemus*) along the Atlantic coast of the United States have raised concerns that the present resource is in decline and insufficient to support the needs of its user groups. These concerns have led the Atlantic States Marine Fisheries Commission (ASMFC) to implement a fishery management plan to regulate the harvest (ASMFC[1]). In order to properly manage any species, specific management goals and objectives must be established, and these goals depend on the resource users involved (Quinn and Deriso, 1999). Horseshoe crabs present a distinct resource management challenge because they are important to a diverse set of users (Berkson and Shuster, 1999).

Horseshoe crabs lay their eggs on sandy beaches in spring and summer, and migrating shorebirds rely heavily on the eggs to supply the energy required to complete their migration (Rudloe, 1980; Shuster and Botton, 1985; Castro and Myers, 1993; Botton et al., 1994; Myers, 1996; Thompson, 1998; Tsipoura and Burger, 1999). Biomedical companies catch horseshoe crabs for their blood, from which they produce Limulus Amebocyte Lysate (LAL) (Novitsky, 1984; ASMFC[1]). LAL is used to detect contamination of injectable drugs and implantable devices by Gram-negative bacteria and is the most sensitive means available for detecting endotoxins (Novitsky, 1984). Finally, horseshoe crabs are harvested commercially for bait in the American eel (*Anguilla rostrata*), catfish (*Ictalurus* spp.), and whelk (*Busycon* spp.) fisheries (ASMFC[1]).

The goal of the ASMFC fishery management plan is to ensure a sustainable population level that will support the continued use by these diverse ecological, biomedical, and fishing interests (ASMFC[1]). Proper management of the resource requires information on the status and dynamics of the horseshoe crab population (Berkson and Shuster, 1999). However, the status of the population is poorly understood, and there is currently no reliable information on which to base any management scheme. Available fishery-independent surveys were not designed for horseshoe crabs, and are of little or no value in assessing their status (ASMFC[2]). Towards this end, the states of New Jersey, Delaware, and Maryland in conjunction with the ASMFC and the National Fish and Wildlife Foundation, funded a pilot benthic trawl survey for the fall of 2001. Data collected during this pilot trawl survey were used to estimate the horseshoe crab population size in the Delaware Bay area.

Methods

This study was conducted in the vicinity of Delaware Bay, which is the center of abundance for horseshoe crabs on the Atlantic coast (Shuster, 1982). The study area extended from north of Cape May, New Jersey, to south of Ocean City, Maryland (39°10′N to 38°10′N), and from shore out to 22.2 km (Fig. 1). The area was divided into four strata based on distance from shore and topography, both of which influence crab distribution. Distance from shore was considered important because horseshoe crab abundance decreases with depth (Botton and Ropes, 1987a). Therefore, the area was split into an inshore zone from 0 to 5.6 km (0 to 3 nautical miles [nmi]) from shore and an offshore zone from 5.6 to 22.2 km (3 to 12 nmi) from shore. Topography was also considered important because commercial fishermen stated that crabs are more abundant in troughs (Burke[3]; Eutsler[4]; Munson[5]). For this study, troughs were defined as at least 2.4 m deep, no more than 1.8 km wide, and more than 1.8 km long. These dimensions are common for troughs identified as important by the fishermen. The inshore and offshore zones were both further divided into trough and nontrough areas. The resulting strata were inshore trough, inshore nontrough, offshore trough, and offshore nontrough.

The study area was divided into grids of one-minute latitude by one-minute longitude. A grid was considered inshore if the majority of its area was in water and inshore of the 5.6-km dividing line. A grid was considered offshore if the majority of its area was offshore of the 5.6-km dividing line and inshore of the 22.2-km boundary. A grid was also considered a trough if the long axis of a trough passed through the grid.

[1] ASMFC (Atlantic States Marine Fisheries Commission). 1998. Interstate fishery management plan for horseshoe crab. Fishery management report no. 32, 58 p. Atlantic States Marine Fisheries Commission. 1444 Eye Street, NW, Sixth Floor, Washington, DC 20005.

[2] ASMFC. 1999. Horsehoe crab stock assessment report for peer review. Stock assessment report No. 98-01 (supplement), 47 p. Atlantic States Marine Fisheries Commission, 1444 Eye Street, NW, Sixth Floor, Washington, DC 20005.

[3] Burke, C. 2001. Personal commun. 25 Cove Drive, North Cape May, NJ 08204.

[4] Eutsler, J. 2001. Personal commun. 11933 Gray's Corner Road, Berlin, MD 21811.

[5] Munson, R. 2001. Personal commun. Box 358, Newport, NJ 08345.

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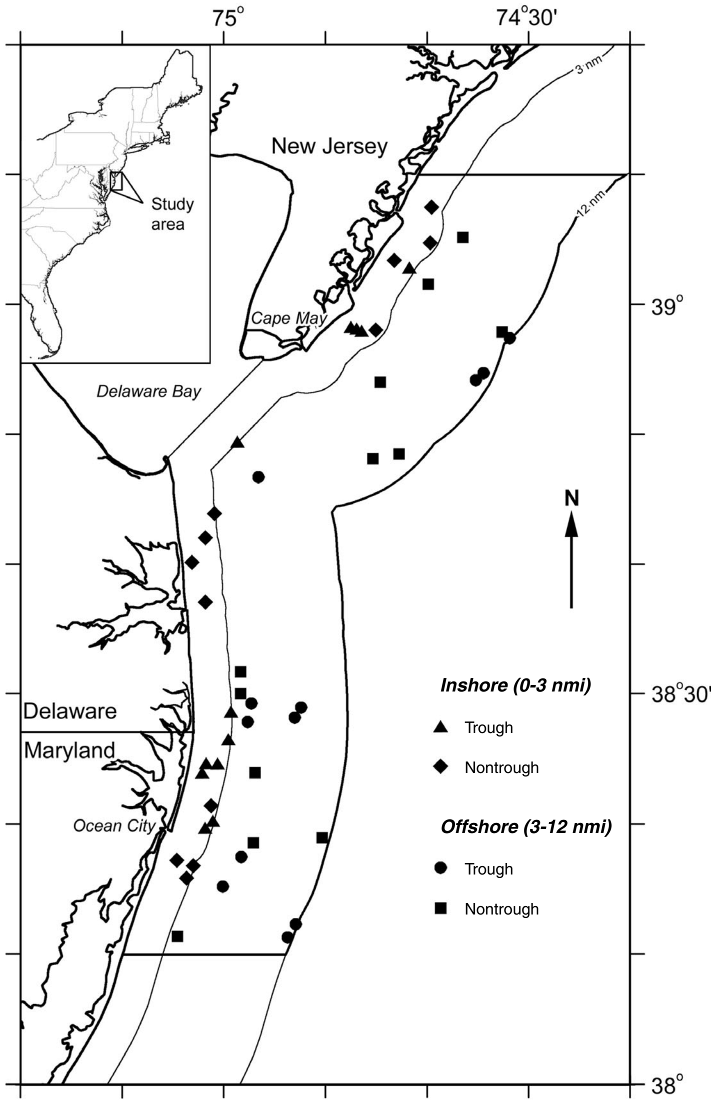

Figure 1

Study area and sampling locations. Symbols indicate type and location of strata. Day and night tows were made at each location.

A grid was considered nontrough if no trough long axis passed through it. Each grid was therefore assigned to one of the four strata. Twelve grids were randomly selected in each stratum, for a total of 48 unique sampling locations. The fishermen also stated that time of day influenced horseshoe crab catchability (Burke[3]; Eutsler[4]; Munson[5]). Therefore, grids were sampled both in daylight and at night. The second tow in a grid (day or night) was made near the location of the first to reduce location variability, but slightly offset to avoid possible influence of the first tow on the catch of the second. The second tow was also made more than 24 hours after the first to avoid interactions, but no more than four days later, to avoid introducing other unknown variability. Abundance estimates from the daytime and nighttime samples were calculated separately for comparison.

Our study was conducted in the fall, between 10 September and 16 October 2001. The stock assessment model adopted by the ASMFC requires abundance information on newly mature crabs, and identification requires that crabs have undergone a terminal molt. Crabs reportedly molt in the late summer and fall in the Delaware Bay area (Burke[3]; Eutsler[4]; Munson[5]).

Sampling was conducted from a chartered 16.8-meter commercial fishing vessel. For capturing horseshoe crabs, commercial fishermen typically use a flounder trawl equipped with a Texas sweep (Burke[3]; Eutsler[4]; Munson[5]; Michels[6]). This modified sweep consists of a chain line in-

[6] Michels, S. 2001. Personal commun. Delaware Department of Natural Resources and Environmental Control, Division of Fish and Wildlife, 89 Kings Hwy., P.O. Box 1401, Dover, DE 19901.

stead of rope, which runs from wing to wing of the net (Fig. 2).The net ropeline is attached behind the sweep chain. In addition, usually three rows of weight chain are attached behind the sweep chain. The chain sweep is considered more effective in digging crabs out of the bottom than the typical ground gear of most research trawls. We used a standard two-seam flounder trawl with an 18.3-m headrope and 24.4-m footrope. The net consisted of 14-cm stretched mesh polypropylene throughout and was equipped with chafing gear on the bag. The net was attached to the trawl doors by 91-m ground cables wrapped in rubber cookies. Tow duration was usually 15 minutes (bottom time), except for one tow in the Delaware Bay shipping channel, which was reduced to 7.5 minutes. We assumed that density was not affected by tow duration (e.g. gear saturation was not a factor).

All horseshoe crabs were culled from the catch, and either all or a subsample were examined. For subsamples of a large catch, 50 crabs greater than 150 mm prosomal width were examined, as well as all small, soft, and shedding crabs. Horseshoe crabs that were not examined were counted separately by sex. Examined crabs were measured for prosomal width and identified to sex and maturity. Maturity classifications were as follows: immature; primiparous (mature horseshoe crabs that had not spawned yet); and multiparous (crabs that had spawned at least once [Table 1]). When catches were subsampled, characteristics of examined crabs were extrapolated to all crabs in that tow. Abundance was estimated for each demographic group as well as for the total.

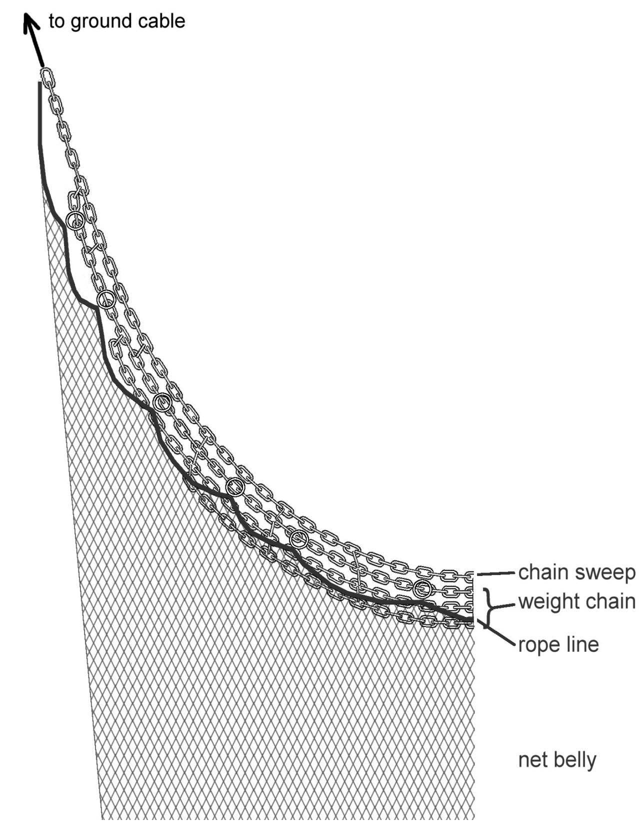

Figure 2

Diagrammatic representation of the Texas sweep used on the flounder trawl.

Tow distances were determined for most tows from beginning and ending positions and recorded by using Loran C. These are minima because they do not consider any deviations from a straight path. Distances were not recorded for three tows; therefore they were estimated as the mean distance of all other tows. Net width was estimated as half of the mean of the headrope and footline lengths (Fridman, 1986). The tow distance and net width were used to calculate the swept area to determine the density of horseshoe crabs. We assumed that the ground cables and trawl doors were not effective in catching crabs; therefore all fishing was done only by the net. No information is available on the efficiency of the ground cables or doors for horseshoe crabs, but we do not believe horseshoe crabs are mobile enough, nor swim fast enough, to be effectively herded by them.

The mean density (crabs/km^2) and variance in each stratum were calculated by assuming a Δ-distribution (Aitchison and Brown, 1957; Pennington, 1983), and these estimates were combined by using formulas for a stratified random design (Cochran, 1977). The Δ-distribution model is applicable to skewed data that consist of a portion of zero catches when the frequency of nonzero catches follows a lognormal distribution (Pennington 1983; Pennington 1996). With such skewed data, the estimator of the mean as defined for the Δ-distribution model is more efficient than the sample mean estimator derived from the normal distribution (Smith, 1988). Areas by stratum and total area were substituted for the numbers of grids per stratum and total number of grids for determining stratum weights (Table 2). Latitudinal and longitudinal distances, and therefore grid areas, differed by latitude; therefore grid areas were calculated separately for each minute of latitude. The total number of grids in each stratum was determined for each latitude to calculate the area by stratum and the total area. Ninety-five percent confidence intervals of the stratified mean density and population total were calculated by using the effective degrees of freedom (Cochran, 1977). Mean densities, totals, and confidence limits for demographic groups did not sum to the values calculated by using all horseshoe crabs combined because the stratum mean calculated by the Δ-distribution is a function of the stratum variance, which varies by demographic group.

Results

The mean abundance estimate for all crabs within the study area based on day sampling was 6.81 million crabs within the 2912-km^2 study area (Table 3). The mean abundance estimate for all crabs based on night sampling was 11.40 million crabs in the study area (Table 3).

Table 1

Criteria used in this study for classifying horseshoe crab maturity stage.

	Female	Male
Immature	Gonopores not hard and elevated, no modified pedipalps, soft, membranous area of ventral prosoma (doublure) pale colored.	Hard, elevated gonopores discernible on genital operculum, no modified pedipalps.
Primiparous	Soft, membranous area of ventral prosoma dark colored (indicating presence of eggs), no rub marks on upper opisthosoma.	Gonopores as above, modified pedipalps, both pedipalp digits intact on both sides.
Multiparous	Soft, membranous area of ventral prosoma dark colored, rub marks present on opisthosoma indicating previous amplexus.	Gonopores as above, modified pedipalps, smaller pedipalp digit broken off from at least one side.

Abundance estimates by stage class provided additional information. Multiparous males were estimated at 2.40 million for day sampling and 4.23 million for night sampling. Multiparous females were the next most abundant group, estimated at 1.63 million for day sampling and 2.25 million for night sampling (Table 3).

Primiparous males were uncommon during daylight sampling, estimated at only 84,000 during the day, as compared to 307,000 at night. In contrast, primiparous females were estimated at 338,000 and 361,000 for day and night sampling, respectively.

The estimated abundance of mature males (primiparous and multiparous combined) exceeded that of mature females: 2.48 million to 1.97 million for sampling during the day and 4.54 million to 2.61 million for night sampling. Estimates of immature horseshoe crabs showed that the opposite trend with greater numbers of females than males, 1.34 million to 0.38 million, respectively, for day sampling and 2.31 million to 1.19 million, respectively, for night sampling. With both mature and immature horseshoe crabs, estimates derived from night sampling were higher than those derived from day sampling (Table 3).

Confidence intervals for the estimates were wide, but informative. Confidence limits for total horseshoe crab abundance were 2.29 million to 11.33 million for day sampling and 5.95 million to 16.85 million for night sampling. The lower confidence limits provide useful reference points for conservative, risk-averse management schemes.

Table 2

Horseshoe crab survey stratum sizes. Sampling grids were one minute longitude by one minute latitude. The area of grids sampled in each stratum is denoted by a, the total area (km^2) of the stratum is A, n is the number of grids sampled, and N is the total number of grids in that stratum. Strata are the following: I NT = inshore nontrough, I TR = inshore trough, O NT = offshore nontrough, and O TR = offshore trough.

	Stratum				
	I NT	I TR	O NT	O TR	All
a	32.48	32.51	32.50	32.55	130.04
A	560.07	165.18	1964.87	222.06	2912.17
n	12	12	12	12	48
N	207	61	726	82	1076

Discussion

The study does not estimate actual population size, but rather the total number of horseshoe crabs available to the survey gear. Horseshoe crabs remain at the beaches where they were spawned for the first one to two years of life and gradually disperse offshore as they grow (Rudloe, 1981; Shuster, 1982). Crabs of these early age classes were undoubtedly in shallow shelf waters and coastal embayments beyond the reach of the vessel. Even if they were present, crabs of early age classes may have been too small to be caught in the gear. The study also excluded adults that may have been in shallow waters and embayments. It is also unlikely that 100% of the horseshoe crabs under the gear were in fact captured because some may have been buried too deep in the substrate to have been dug out by the gear. For all of these reasons, abundance estimates can legitimately be considered minimum population estimates. Results can be used as abundance indices for comparison between years, if the study is continued in the future.

The differences between day and night estimates may be the result of horseshoe crabs burying themselves during the day. Alternatively, the horseshoe crabs may be able to detect and avoid the trawl during the day. Night and day collections at individual locations were correlated (r=0.71) suggesting that both were a true reflection of horseshoe crab abundance at that site, although at different levels of efficiency. If the catches were uncorrelated, it would not be possible to determine which, if either, sample accurately represented true abundance. The larger catches and lower

Table 3

Stratified mean density (crabs/km²), standard deviation (SD), and coefficient of variation of the mean (CV) for horseshoe crab demographic groups and for all crabs combined. Estimated population totals by demographic group and for all crabs combined are given in thousands. UCL and LCL denote upper and lower 95% confidence limits, respectively. Estimates were determined separately for day and night sampling. Because the Δ-distribution was used to calculate stratum means, demographic group values do not sum to those calculated by using all crabs.

	Density (crabs/km²)			Population total (1000s)		
Demographic group	Mean	SD	CV	Total	UCL	LCL
Day						
Immature females	461	167	0.36	1341	2395	288
Primiparous females	116	40	0.34	338	588	88
Multiparous females	561	126	0.23	1634	2428	839
Immature males	129	45	0.35	377	659	95
Primiparous males	29	7	0.24	84	129	40
Multiparous males	823	207	0.25	2396	3699	1093
All horseshoe crabs	2338	718	0.31	6809	11,326	2291
Night						
Immature females	792	216	0.27	2308	3656	960
Primiparous females	124	26	0.21	361	522	199
Multiparous females	773	145	0.19	2250	3157	1343
Immature males	410	119	0.29	1193	1939	447
Primiparous males	106	40	0.38	307	555	60
Multiparous males	1453	353	0.24	4231	6434	2029
All horseshoe crabs	3915	873	0.22	11,400	16,853	5947

coefficients of variation from the night estimates suggest that the night estimates are more efficient and are probably better estimates of true abundance.

The results of the present study are intermediate between previous estimates of ocean abundance. Botton and Ropes (1987a) estimated that between 2.3 and 4.5 million adults occurred on the continental shelf between New Jersey and Virginia from National Marine Fisheries Service (NMFS) trawl surveys, in contrast to a mean of 7.1 million adults (primiparous and multiparous combined) estimated in the present study area. However, the trawl gear used in the NMFS surveys was inefficient for capturing horseshoe crabs, and the inshore extent was limited by the survey vessel size (Botton and Ropes, 1987a; ASMFC[2]). Botton and Haskin (1984) sampled within 5.6 km of the New Jersey coast using hydraulic clam dredges and obtained horseshoe crab densities of 14,600 to 23,000 per km². These densities are much higher than our nighttime estimate of 7900 horseshoe crabs per km² (weighted by stratum area) within 5.6 km. The gear we used was probably more efficient in capturing horseshoe crabs than that employed by the NMFS survey but may have been less efficient than the hydraulic dredge. Differing methods between the studies do not allow for a comparison over time.

It is interesting to note that in both the night-based and day-based estimates, females made up the majority of the immature animals, whereas males made up the majority of the mature animals. This could be due to the commercial fishery's preference for harvesting gravid females (Botton and Ropes, 1987b). The continual focused harvest of mature females may reduce their population enough to cause this change in sex ratios. Alternatively, mature females or immature males may have been more abundant outside the study area.

Conclusion

The continuation of annual trawl surveys could allow a full stock assessment to be conducted. The Horseshoe Crab Stock Assessment Subcommittee of the Atlantic States Marine Fisheries Commission has developed a stock assessment plan (HCSAS[7]) based on the catch-survey method derived by Collie and Sissenwine (1983). Unlike age-based stock assessment models, the catch-survey method requires only abundance of primiparous and multiparous horseshoe crabs (HCSAS[7]). The commercial fishery is selective for gravid females (Botton and Ropes, 1987b), and effort is biased toward areas of high abundance (Burke[3]; Eutsler[4]; Munson[5]); therefore commercial data are of limited use for stock assessment. A fishery-independent

[7] HCSAS (Horseshoe Crab Stock Assessment Subcommittee). 2000. Stock assessment of Atlantic coast horseshoe crabs: a proposed framework, 19 p. A report to the Horseshoe Crab Technical Committee, Atlantic States Marine Fisheries Commission, 1444 Eye Street, NW, Sixth Floor, Washington, DC 20005.

trawl survey is the best way to provide estimates of abundance while controlling catchability (Hilborn and Walters, 1992; Gunderson, 1993). This study demonstrates the utility of annual trawl surveys to obtain that information.

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