

Abundance and population structure of the Atlantic horseshoe crab *Limulus polyphemus* in Pleasant Bay, Cape Cod

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ABSTRACT: Populations of horseshoe crabs *Limulus polyphemus* are widely distributed from Maine to the Gulf of Mexico, are commercially harvested, and are thought to have decreased in abundance in recent decades. To provide information needed to manage exploited populations of horseshoe crabs, we conducted comprehensive field sampling in a major shallow estuary containing preferred habitat for horseshoe crabs. The data we obtained are the first to comprehensively define abundance, determine sex ratios of horseshoe crabs across an entire estuary, and estimate cohort-specific growth and mortality of juvenile and adult horseshoe crabs. We found that juveniles were more abundant and suffered greater mortality than adults. Adults were largely found on the sediment surface, but 20% were buried shallowly in sediments. The male:female sex ratio in juveniles was 1.4:1, but the adult sex ratio was 2.3:1. Juveniles grew faster than adults, and adult crabs may plausibly molt as frequently as once per year rather than have a terminal molt. Spawning appeared to span late March to mid-July, and juveniles hatched at semilunar intervals during Year 0 and grew to 16.6 ± 0.9 mm prosomal width by the start of Year 1. The distinct semilunar cohorts of Year 0 coalesced into annual cohorts after Year 0. Females deposited large numbers of eggs, but only 0.001% survived to the end of Year 0, and approximately 78% of these juveniles reached adulthood.

KEY WORDS: Population dynamics · Growth · Sex ratio · Mortality · Spawning · Recruitment

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INTRODUCTION

The Atlantic horseshoe crab *Limulus polyphemus* is an important component of the economy and ecosystems of the Atlantic coast. Crabs are currently harvested for scientific research, for bait in the eel and conch fisheries, and for the multimillion-dollar biomedical industry that uses *Limulus* lysate, obtained from crab blood, as a screening medium for endotoxins (ASMFC 1998, Berkson & Shuster 1999). Horseshoe crab eggs provide a major food source to migrating shorebirds (Shuster 1982, Castro & Myers 1993, Clark 1996). Horseshoe crabs effectively consume bivalves and other benthic species (Botton 1984a,b, Botton & Haskin 1984, Botton & Ropes 1989), bioturbate sediments (Rudloe 1985, Commito et al. 1995), and provide habitat to epibiotic organisms (Turner et al. 1988, Dietl et al. 2000, Grant 2001).

Limulus polyphemus populations on the East coast of the USA have decreased in recent years. According to spawning censuses, trawl surveys, and anecdotal data, abundance of crabs in New Jersey and Delaware decreased by an order of magnitude from 1970 to 1990 (Michels 1996, Swan et al. 1996). Declines have also been reported in other East Coast sites, including Cape Cod, Massachusetts (Rudloe 1982, Widener & Barlow 1999). These reports have prompted public interest in the conservation and management of horseshoe crab populations (Berkson & Shuster 1999, Eagle 2001), which require specific data on abundance and population structure (Rudloe 1982, ASMFC 1998).

There is limited information on the population dynamics of horseshoe crabs. Horseshoe crabs are found in <30 m of water from the continental shelf to inshore estuaries (Shuster 1982) at the sediment surface or buried in sediments (Rudloe 1979, 1980). Mature

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horseshoe crabs migrate to inshore beaches in early spring to spawn (Shuster 1982, Sekiguchi 1988). Peak spawning activity typically occurs during nighttime full- and new-moon high tides in May and June (Rudloe 1980, Shuster & Botton 1985, Barlow et al. 1986). Females may lay at least 88 000 eggs per year, usually in clutches of 3000 to 8000 (Shuster 1982, Shuster & Botton 1985, Sekiguchi 1988). The extent to which egg production may vary with female size or through the spawning season is not known. Eggs develop in nests buried 5 to 20 cm in coarse intertidal sand (Rudloe 1979, Shuster 1982, Brockmann 1990) and hatch at semilunar intervals approximately 2 wk after spawning (Jegla & Costlow 1982, Sekiguchi et al. 1982, Sekiguchi 1988). Juvenile horseshoe crabs spend their first few years inshore on nursery grounds, moving to deeper waters as they age (Rudloe 1979, 1981, Shuster 1982), up to ~20 yr (Ropes 1961, Shuster 1982, Botton & Ropes 1988).

Abundance of horseshoe crabs has not been well defined. A trawling survey conducted offshore in more than 9 m of water estimated 2.3 to 4.5 million crabs along the Atlantic coast (Botton & Ropes 1987). In contrast, a more recent trawling survey, which included shallower waters, estimated 6 to 17 million horseshoe crabs off the coast from New Jersey to Maryland (D. Hata & J. Berkson unpubl.). Other studies conducted on individual beaches sampled only adult spawning crabs (Baptist et al. 1957, Sokoloff 1978, Rudloe 1980, Shuster & Botton 1985, Widener & Barlow 1999, Smith et al. 2002), which make up only a portion of the population. More comprehensive whole-estuary and whole-population studies are needed in the shallow water, where horseshoe crabs are abundant.

No studies have assessed sex ratios of horseshoe crabs in a way that is not biased by spawning strategy or sampling method. Spawning surveys find more males than females because more males congregate on the beach during spawning (Shuster 1950, 1953, Rudloe 1980). Trawling surveys may count more females than males if some females remain longer in deep water while males are on the beach during the spawning season (Rudloe 1980) or if females are more easily captured by the trawl because of their larger mean size compared to males.

Defining age classes and estimating growth within horseshoe crab populations is difficult because there is no known method to age a horseshoe crab (Botton & Ropes 1988, Grady et al. 2001). Only 2 studies have provided consecutive growth increments for juveniles. Sekiguchi et al. (1988) followed juvenile growth through 14 instars in the laboratory, and Shuster (1982) measured growth from preserved exuviae from a single horseshoe crab reared in captivity. Horseshoe crabs have been considered to reach a terminal molt at

sexual maturity. The evidence for this assertion is that there has been only 1 report of a castoff molt from a sexually mature male, and no one has reported seeing a mature crab molt (Shuster 1955).

Annual recruitment and mortality across life stages have not been measured. Predation on eggs and hatchlings may be intense (Botton 1984c, Castro & Myers 1993, Loveland et al. 1996), but survival rates are unknown (Rudloe 1979, Botton et al. 1992, 1994). Although various causes of mortality have been discussed, only adult mortality from stranding and industry-specific harvest has been estimated (Rudloe 1983, Botton & Loveland 1989, Botton 2000).

To manage horseshoe crab populations subject to harvest, we will need data on abundance and size structure of populations, sex ratios of juvenile and non-spawning adults, growth rates of individual size classes, mortality, and recruitment, among other variables (Rudloe 1978b, 1982, 1983, ASMFC 1998, D. Hata & J. Berkson unpubl.). In response to these needs, we set out to obtain data on abundance and structure of a horseshoe crab population in an estuary with a significant crab population. We counted and measured male and female horseshoe crabs of all sizes within Pleasant Bay, the largest coastal embayment on Cape Cod, Massachusetts. We chose this embayment because it supports a significant population of actively spawning adult horseshoe crabs and furnishes substantial feeding and nursery grounds for them (RMP 1998). We construct a first description of the population dynamics of a horseshoe crab population in an entire estuary.

MATERIALS AND METHODS

Abundance. We sampled abundance of horseshoe crabs on a 500 m grid throughout Pleasant Bay. We sampled additional sites in smaller outlying subestuaries to include each major subestuary of the Bay, for a total of 111 sites (Fig. 1).

To determine the abundance of adult horseshoe crabs in the Bay, we counted crabs in a 100 m × 1 m transect at each grid point. We counted crabs from a boat or while snorkeling, using a 50 cm² view box or towing a modified 50 cm clam rake with collection basket. To include crabs buried in sediments, we returned to 14 sites representative of the full range of surface densities to resample surface transects and simultaneously rake each site to sample buried crabs. From these data, we calculated the ratio of surface to buried crabs and used this ratio as a correction factor to apply to our surface abundance data. The corrected number of adult crabs per 100 m² at each grid point was then multiplied by the area of the Bay sampled (26.3 km²). For

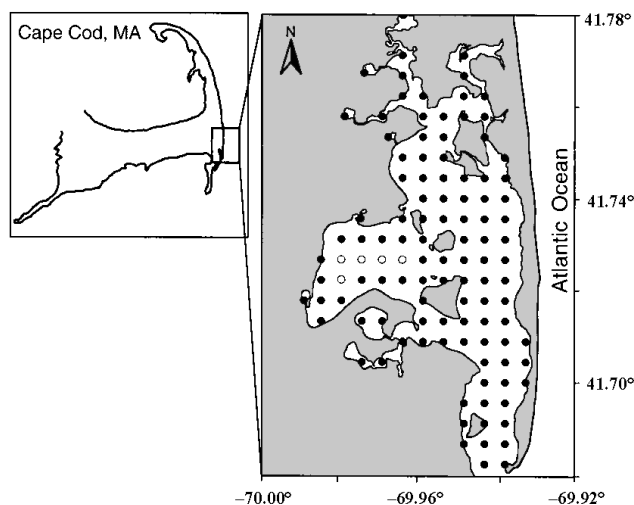


Fig. 1. Location of survey sites in Pleasant Bay on Cape Cod, Massachusetts, USA. (○) Grid points that could not be sampled because of depth or poor visibility. (●) Sampled grid points

sites where surface transects were resampled, we used the mean density of crabs sampled. An additional 1.2 km² was not sampled because of depth or poor visibility (Fig. 1).

To determine the abundance and distribution of juveniles within the bay, we sampled intertidal areas adjacent to grid sites where adults were counted. We used bathymetry to estimate the portion of the Bay most likely to provide nursery habitat. We chose sampling sites within these areas nearly randomly, depending on accessibility. Four randomly oriented 2 m × 2 m quadrats were sampled at 17 sites throughout the Bay during low and intermediate tides. To capture emerging crabs without disturbing unhatched eggs (Rudloe 1979), we systematically sampled the surface top 5 mm, 10 mm, and 5 cm of sediment by hand-picking. To determine abundance of hatchlings and small juveniles (<25 mm prosomal width), which are likely to stay in intertidal areas, we multiplied abundance of these juveniles (per m²) by the estimated area of nursery habitat in the Bay. We calculated abundance of older juveniles (>25 mm prosomal width), which are likely to move throughout the Bay, using the total area sampled in the Bay.

We chose our distribution of sampling sites and times of sampling to avoid potential biases, including differential depth distribution, spawning aggregations on intertidal beaches and recounting crabs. The spatial distribution of our grid points was such that the depths of sampling sites closely matched the bathymetry of the Bay (Fig. 2). This distribution assured that our sampling of the Bay floor was representative of horseshoe crab habitats across the Bay. We recorded data on

salinity ($33 \pm 0.2\text{‰}$) and water temperature ($24 \pm 3^\circ\text{C}$) to confirm that these features are largely seasonally uniform across the Bay (RMP 1998). We collected data from mid-May through 1 September 2001, during daylight hours, across the full range of tidal cycles, when adult crabs are less likely to spawn (Rudloe 1979, 1980), but juveniles are likely to be emerging (Rudloe 1981). The 500 m distance between sampling sites, randomly chosen starting locations, and sampling as many adjacent sites as possible per day minimized the chance of recounting crabs.

Population structure. Size and sex: To determine the size structure of the population, we measured prosomal width (Shuster 1955, Riska 1981) to the nearest 1 mm for adults and the nearest 0.1 mm for juveniles. We determined sex by the presence or absence of monodactylus pedipalps and the structure of genital pores (Shuster 1982, Sekiguchi 1988). We determined sex of juveniles in the field for most animals >16 mm prosomal width and distinguished subadult females by the absence of mating scars on the posterior marginal angles of the opisthosoma (Shuster 1955).

Cohort analysis: We used both direct and indirect methods to assess cohorts. To be certain that cohorts represented individuals hatched at semilunar intervals during the spawning season and to directly follow changes in cohort abundance and growth, we recorded the number and size of juveniles through 3 semilunar cycles at a single location in Pleasant Bay. This real-time sampling was effort-intensive and could not be done at many sites. To cover the entire Bay synoptically, we pooled data across sampling sites to create an aggregate depiction of size frequency distributions for the population. We used the aggregate cohort data to define instar stages and estimate growth, mortality, and recruitment.

To obtain a sufficient number of crabs to define cohorts, we collected crabs throughout the Bay (N = 803 juveniles and N = 980 adults) simultaneously with and in addition to the survey of abundance. We used Mix

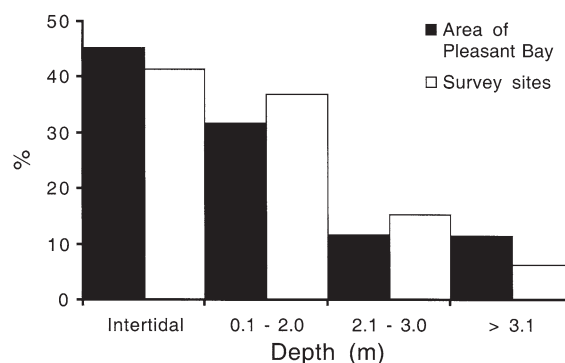


Fig. 2. Depth distribution of areas in Pleasant Bay compared to the depth distribution of sampled areas. $\chi^2 = 4.16$, $p = 0.25$ ns

3.1.3 software to determine the statistically best-fit χ^2 distributions to our size frequency data and to calculate a mean size for each cohort. This procedure involved entering an estimated number of cohorts, as well as estimated mean, variance, and proportion for each distribution. Then the software bootstrapped combinations of variables until the best-fit χ^2 value was found.

Growth: We calculated growth from the direct sampling of cohorts and from the indirect aggregate data. To determine growth rates for juvenile horseshoe crabs in Pleasant Bay, we compared the mean prosomal width of each cohort to either the real time of growth (directly measured cohorts) or to estimated mean age (indirectly aggregated cohorts). We used data of Sekiguchi (1988) regarding the mean duration of each instar (among juveniles reared at salinities close to that of Pleasant Bay) to estimate mean age of aggregated cohorts as $A_X = I_X + A_{X-1}$, where A_X = age at instar X , I_X = mean duration of instar X , and A_{X-1} is age at instar $X - 1$. For juvenile instars 11+ and adults, we assumed annual molt increments (Rudloe 1978a, Sekiguchi 1988). To describe growth of individuals across all life stages, we plotted % increase in size from hatchling through adulthood. For comparison with growth of crustaceans, we also compared the difference in mean size from one cohort (pre-molt) to the next (post-molt) according to the Hiatt growth model (Hiatt 1948, Kurata 1962).

Mortality: We roughly estimated juvenile and adult mortality from reductions in abundance at successive cohorts. Mortality of juveniles and adults was calculated from different sets of data. There were sufficient data to determine juvenile abundance directly from the frequency of crabs in each size class collected in quadrats during our abundance survey. Adult abundance was estimated from aggregate data by multiplying the proportion of individuals represented by each cohort (provided by Mix software) by the total abundance of adult horseshoe crabs in the Bay. To partially compensate for potential differences in annual recruitment into adult cohorts, we regressed changes in abundance in successive instars, starting with the cohort of peak abundance. For juveniles and adults, we calculated % mortality as $(A_{\text{Instar } X} - A_{\text{Instar } X+1})/A_{\text{Instar } X}$, where $A_{\text{Instar } X}$ = abundance at a given instar stage and $A_{\text{Instar } X+1}$ = abundance in the next instar.

Recruitment: To estimate recruitment, we first estimated reproductive output in terms of periodicity, length, and peak of spawning, the number of mature females likely to spawn, and the number of eggs likely produced in the Bay. Second, we compared survival at different life stages to the number of eggs produced.

To determine length of spawning season in Pleasant Bay we compared the occurrence of juvenile cohorts across semilunar periods from 21 June through 1 Sep-

tember 2001. We then extrapolated to estimate approximate time of first and last spawning from the estimated age of juveniles of oldest and youngest instars observed during our first and last sampling dates respectively. We identified semilunar intervals, starting with the full moon and ending with the new moon, since larvae generally emerge from nests on the full moon (Rudloe 1979). To estimate duration of the spawning season, we counted the number of adults in coupled pairs at each grid point during our abundance survey and calculated % of the adult population paired during each semilunar cycle, assuming that the number of animals paired in the general population was a proxy for spawning intensity. The number of adults in pairs over time provided a corroboration of the seasonal extent of spawning season.

To estimate the portion of adult females that may be spawning, we calculated the number of adult females in the total population that had prosomal widths within the range of sizes observed among paired crabs collected during our abundance survey and found in spawning aggregates in Pleasant Bay.

To estimate the number of eggs that may be produced annually in the Bay, we assumed that each female capable of spawning does so seasonally and deposits 88 000 eggs. We then estimated recruitment by comparing this reproductive output to the abundance of juveniles at different life stages to adulthood.

RESULTS AND DISCUSSION

Abundance

Juveniles

There were 1.35×10^7 juvenile horseshoe crabs in Pleasant Bay (Table 1). Juveniles <25 mm prosomal width, which inhabited intertidal nursery areas of the Bay, were more numerous than subadult juveniles (>25 mm) distributed throughout the Bay (Table 1, Fig. 3). Our estimates of juvenile abundance were calculated from the density of juveniles in the quadrats, across an estimated 8.2 km² of the Bay that may provide nursery habitat for juvenile crabs from hatchling to 25 mm. These estimates are likely conservative since more of the Bay may be used as nursery habitat than we allowed, and newly hatched juveniles from deeper nests may not have emerged from sediments.

Adults

The total estimated population of adult horseshoe crabs in Pleasant Bay was $5.0 \times 10^5 \pm 27\%$ (Table 1).

Table 1. *Limulus polyphemus*. Estimated abundance (\pm SE) of juvenile and adult horseshoe crabs in Pleasant Bay. Juveniles are categorized by prosomal width and adults by location relative to the sediment surface. CV = coefficient of variation (standard error/mean \times 100)

	Mean no. (100 m ⁻²)	Suitable habitat (km ²)	Total no.	CV (%)
Juveniles				
<25.0 mm	158.1 \pm 88.1	8.2	1.3 \times 10 ⁷	56
>25.0 mm	1.9 \pm 1.1	26.3	4.9 \times 10 ⁵	58
Total juveniles	–	–	1.35 \times 10 ⁷	
Adults				
Surface	1.5 \pm 0.41	26.3	4.0 \times 10 ⁵	27
Buried	0.4 \pm 0.30	26.3	1.0 \times 10 ⁵	75
Total adults	1.9 \pm 0.51	26.3	5.0 \times 10 ⁵	27

Table 2. *Limulus polyphemus*. Mean (range) prosomal width (mm) and sex ratios of juvenile and adult horseshoe crabs in Pleasant Bay. We determined sex of juveniles larger than 16.0 mm, and assumed smaller sizes were present in both sexes based on our calculated sex ratio

	Prosomal width (mm)		M:F
	Males	Females	
Juveniles	2.9–168	2.9–150	1.4
Adults	180 (132–230)	216 (135–288)	2.3

Average density of adult crabs in Pleasant Bay was 1.9 per 100 m² (Table 1). Approximately 20% of adult crabs in the Bay were buried shallowly in sediments (Table 1). Juveniles were over an order of magnitude more abundant than adults, but the density of adults was comparable to that of juveniles >25 mm (Table 1, Fig. 3).

Our estimates of abundance are larger than suggested by Shuster in the early 1950s for Pleasant Bay (pers. comm.) or estimated by offshore trawls near Cape Cod (Botton & Ropes 1987). Our abundances are likely higher, in part, because they include buried crabs and non-spawning adults, and because we sampled in the type of shallow embayment favored by crabs during the spawning season. Spawning censuses miss non-spawning crabs spread out over the estuary, and trawl surveys typically focus on deeper waters where crabs may be sparse.

Population structure

Size

Juveniles ranged in prosomal width from 2.9 to 168 mm (Table 2). Adult horseshoe crabs ranged in size

from 132 to 288 mm, and males were an average of 17 \pm 1% smaller than females. Juveniles overlapped in size with adults between 132 and 168 mm, indicating this is the size range at which horseshoe crabs in Pleasant Bay reach maturity. The range of prosomal widths observed in this study is similar to that of crabs recorded in Pleasant Bay almost 50 yr ago (Shuster 1955) and collected throughout Massachusetts waters in recent years (Massachusetts Division of Marine Fisheries unpubl.).

All of the largest juveniles (>150 mm prosomal width) in Pleasant Bay were males, suggesting that juvenile males in Pleasant Bay can reach at least the same size as juvenile females (Table 2). Shuster (1955) and Sekiguchi et al. (1988) suggested the size difference between adult male and female horseshoe crabs results from males maturing at a smaller size than females, but we did not find this to be the case in Pleasant Bay.

Sex ratio

There was a shift in sex ratio from juveniles to adults (Table 2). Juvenile sex ratio was nearly equal, while adult sex ratio was male-dominated, suggesting a selective loss of females compared to males after sexual maturity in Pleasant Bay. Juvenile sex ratios have not been previously reported, but the adult results fall within the range of sex ratios reported by spawning

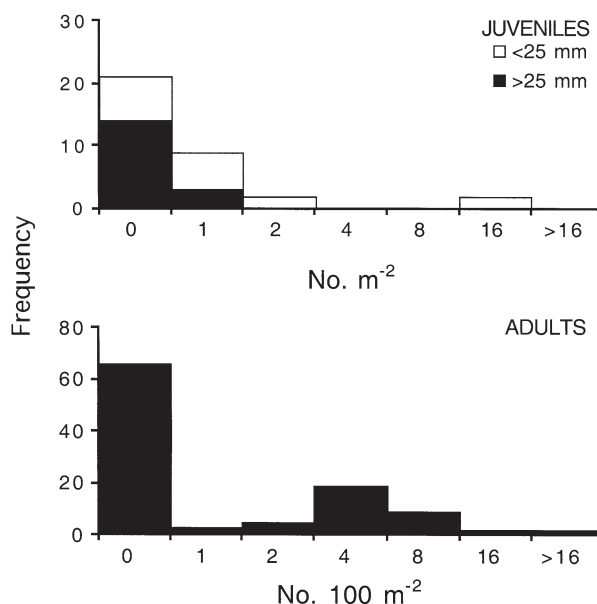


Fig. 3. *Limulus polyphemus*. Frequency distribution of the number of sampling sites in different density categories for juvenile (top) and adult (bottom) horseshoe crabs in Pleasant Bay. Juveniles are separated by prosomal width (mm)

Table 3. *Limulus polyphemus*. Male to female sex ratios of juvenile (J) and adult (A) horseshoe crabs in this study (**bold**) and for spawning (S) and offshore (T) populations along the Atlantic coast. Offshore populations were sampled by trawling

Location	M:F	Category	Comments	Source
Pleasant Bay, MA	1.4	J	–	This study
Pleasant Bay, MA	2.3	A	Non-spawning	This study
Delaware Bay	1–6	A	S	Rathbun (1884)
Barnstable, MA	1.5–2.4	A	S	Shuster (1950)
Pleasant Bay, MA	2.5	A	S	Shuster (1955)
Cape May, NJ	1–12	A	S	Shuster (1955)
Cold Spring Harbor, NY	4	A	S (1957)	Sokoloff (1978)
Apalachee Bay, FL	1–14	A	S	Rudloe (1980)
Delaware Bay, NJ	3–5	A	S (1977–1979)	Shuster & Botton (1985)
Mashnee Dyke, MA	2.5	A	S	Barlow et al. (1986)
MD	2–4	A	S	DNR ^a (1998) ^c
SC	3.5	A	S	Thompson (1998) ^c
Delaware Bay, DE	0.8–5.9	A	S	Smith et al. (2002)
Delaware Bay, NJ	1–9.4	A	S	Smith et al. (2002)
Apalachee Bay, FL	0.5–0.6	A	T	Rudloe (1980)
Delaware Bay	0.8	A	T	Swan et al. (1993) ^c
NJ	0.7	A	T	DFG&W ^b (1997) ^c

^aMaryland Division of Natural Resources; ^bNew Jersey Division of Fish, Game, and Wildlife; ^cfrom ASMFC (1998)

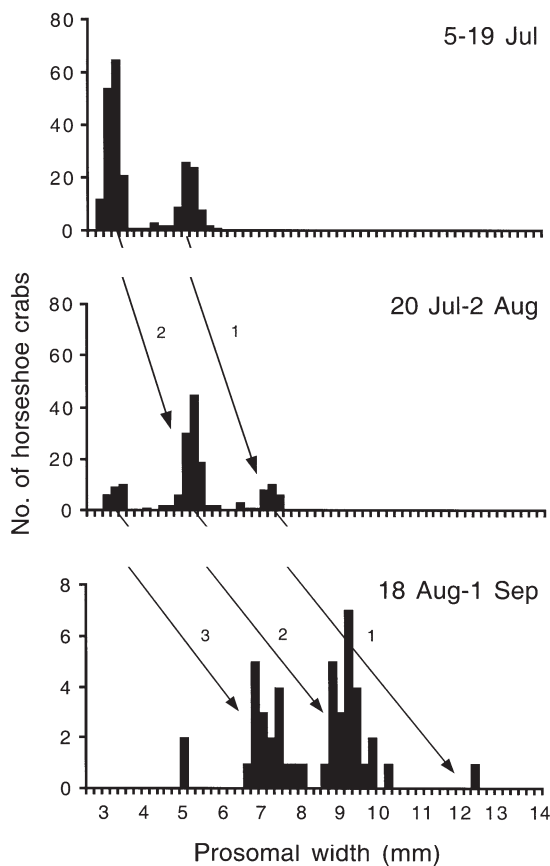


Fig. 4. *Limulus polyphemus*. Juvenile cohorts across 3 semi-lunar periods during the spawning season at a single location in Pleasant Bay. Lines show progression of cohorts across size classes as time passes. Numbers refer to cohort descriptions in Table 6

and trawl surveys of horseshoe crab populations along the Atlantic coast (Table 3).

Since sampling biases may affect sex ratios reported by spawning and trawl studies (Rudloe 1980, Cohen & Brockman 1983, Shuster 1996), our data may be the first to demonstrate a male-dominated sex ratio in a horseshoe crab population, outside of spawning aggregates. A shift in sex ratio almost identical to that observed among adult horseshoe crabs in Pleasant Bay also has been observed in snow crab populations. In these populations, adult abundance is dominated by the sex that molts less because molting is associated with greater mortality (Tester & Carey 1986). Thus, a greater number of molts among adult female horseshoe crabs in Pleasant Bay may explain the observed shift in sex ratio. Rudloe (1980) suggested males and females may be spatially segregated during the spawning season, with females remaining in or returning to offshore areas unless they are actively spawning. If true, this behavior could account for a male-dominated sex ratio even among the general population of inshore embayments. Further study is needed to determine which of these or other factors may account for the apparent relative loss of females from the adult horseshoe crab population in Pleasant Bay.

Cohort analysis

Juveniles. Juveniles monitored at a single site in Pleasant Bay (approximately 41.72° N, 69.94° W),

across 3 semilunar periods during the spawning season, showed distinctive cohorts at each sampling date, and the modal size of cohorts shifted to the right as time passed (Fig. 4). The cohorts represent individuals that hatched during 3 semilunar intervals of the 2001 spawning season.

The aggregated data show identifiable juvenile cohorts for horseshoe crabs that can be assigned age classes by using information from Sekiguchi (1988) (Fig. 5). Each juvenile cohort was composed of crabs collected during any of our sampling times, at which they had achieved a certain size. We interpret these cohorts as representing individuals of the same age at the time of sampling.

The frequency distribution of our cohorts indicate horseshoe crabs in Pleasant Bay likely molted 5 times in Year 0 after hatching, resulting in up to 6 distinct cohorts (Instars 1 to 6) (Fig. 5, top). A conservative interpretation of the data is that the 5th molt occurred in the winter of the first year or spring of the second year, in accordance with data from Sekiguchi (1988), and crabs in all Year 0 cohorts continued to grow until they reached the 6th instar, so that they entered their second year as a single cohort with a prosomal width of 16.6 ± 0.9 mm. During Years 1 to 4 crabs molted 4 times

Table 4. *Limulus polyphemus*. Instar stages and mean prosomal width (mm) (\pm SE) for juvenile cohorts defined in this study and reported in previous studies. Instar stages of horseshoe crabs measured by Shuster (1955, 1982) have been estimated based on similarity to prosomal widths of cohorts in this study

Instar stage	Mean prosomal width (mm)		Shuster (1955, 1982)
	This study	Sekiguchi (1988)	
1	1	3.1 \pm 0.2	3.3
2	2	5.0 \pm 0.3	5.0
3	3	7.1 \pm 0.4	7.0
4	4	9.1 \pm 0.4	9.1
5	5	12.5 \pm 0.7	11.3
6	6	16.6 \pm 0.9	14.5
	7		18.6
7	8	21.7 \pm 0.3	23.5
8	9	29.6 \pm 0.2	29.5
9	10	40.9 \pm 1.5	38.0
	11		43.7
10	12	49.2 \pm 1.6	50.2
11	13	62.6 \pm 3.3	64.4
12	14	77.3 \pm 4.2	81.2
13		91.1 \pm 3.3	
14		103.4 \pm 4.4	
15		115.5 \pm 3.6	
16		134.0 \pm 3.3	
17		159.7 \pm 5.1	

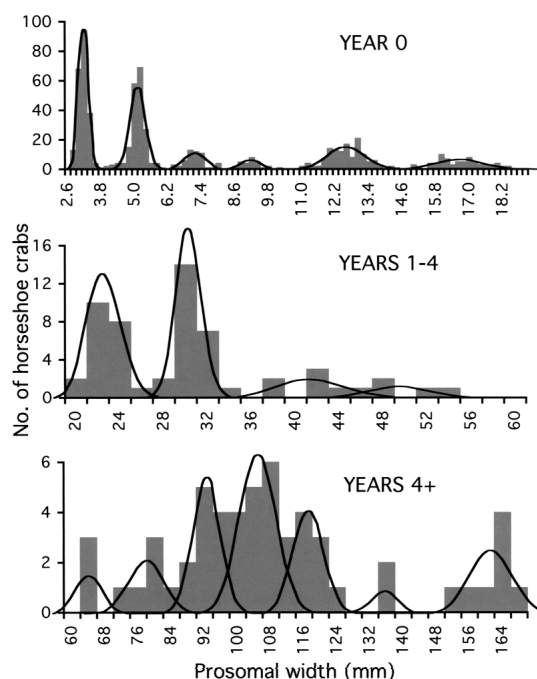


Fig. 5. *Limulus polyphemus*. Juvenile cohorts based on size (prosomal width) classifications. Cohorts are distributions statistically best fit to the size frequency data (gray bars) for horseshoe crabs in Year 0 (top), Years 1 to 4 (middle), and Year 4 to sexual maturity (bottom). Year class groupings are based on information from Sekiguchi et al. (1988)

to reach a size of 49.2 ± 1.6 mm (Fig. 5, middle). Horseshoe crabs in Pleasant Bay likely molted another 5 to 7 times before reaching sexual maturity (Fig. 5, bottom). Hence, juveniles took up to 11 yr and 17 instars to reach sexual maturity, at a prosomal width of 159.7 ± 5.1 mm.

There might be other interpretations of the aggregated cohort data of Fig. 5. Our interpretation assumes that there were approximately annual molts between Years 1 and 11. Sekiguchi (1988) found that horseshoe crabs in the laboratory molted more than once in Years 1 to 4. His crabs occasionally had smaller molt increments than crabs we measured in Pleasant Bay (Table 4), and other comprehensive field data are not available for comparison. Since we cannot be certain when each molt occurred in Pleasant Bay, our assessment is the parsimonious and conservative given the distinctiveness of the cohorts and the relative increase in size from one cohort to the next. The mean prosomal widths of successive juvenile cohorts are similar to those reported for juveniles reared continuously in the laboratory (Sekiguchi et al. 1988), for a few horseshoe crab cohorts previously measured in Pleasant Bay (Shuster 1982) (Table 4), and for snow crabs (Comeau et al. 1998).

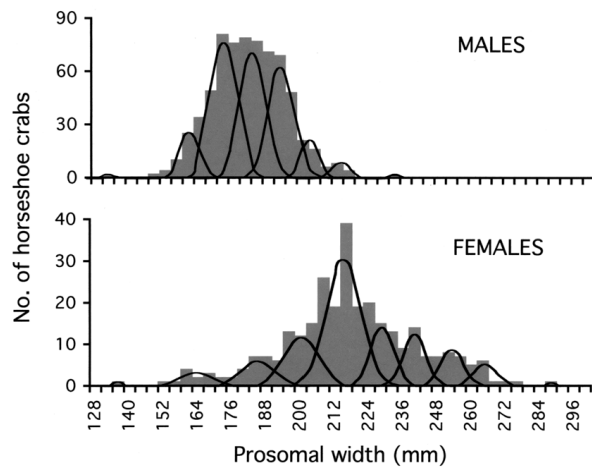


Fig. 6. *Limulus polyphemus*. Adult male (top) and female (bottom) cohorts based on size (prosomal width) classifications. Cohorts are distributions statistically best fit to the size frequency data (gray bars) for horseshoe crabs measured in Pleasant Bay

Adults. Adult male horseshoe crabs in Pleasant Bay were sorted into 8 cohorts, while adult females were sorted into 10 cohorts by the Mix predictions (Fig. 6). The distribution of cohorts clearly shows the larger size achieved by females compared to males at each instar (Table 5), and the greater number of cohorts among females suggests that adult females molt more than adult males.

The identifiable cohorts among adult crabs and the wide range in sizes of these cohorts make it implausible to think that horseshoe crabs in Pleasant Bay are limited to a terminal molt at sexual maturity. It seems rather unlikely that horseshoe crabs of 150 mm (mean size at which juveniles reach maturity in Pleasant Bay) could reach the mean or maximum prosomal widths for adult males or females measured in this study in 1 molt. Such a feat would require very large increases in

Table 5. *Limulus polyphemus*. Instar stages and mean prosomal width (mm) (\pm SE) for adult cohorts defined in this study

Instar	Mean prosomal width \pm SE (mm)	
	Males	Females
16	130 \pm 1.8	134 \pm 1.8
17	157 \pm 3.5	161 \pm 6.0
18	169 \pm 4.5	183 \pm 5.8
19	179 \pm 4.5	198 \pm 6.0
20	189 \pm 4.5	213 \pm 6.4
21	199 \pm 3.2	226 \pm 3.8
22	210 \pm 3.0	238 \pm 4.2
23	230 \pm 1.8	251 \pm 4.2
24		263 \pm 4.1
25		286 \pm 1.8

prosomal width. Juvenile males would have to make a 20 to 53 % prosomal increase in a single molt to maturity. Juvenile females would have to make a 44 to 92 % prosomal increase in a single molt. These seem dauntingly large given that a prosomal increase of approximately 20 % has been suggested as the largest possible for adult snow crabs, a species with a mean size of instars and size at sexual maturity almost identical to those of horseshoe crabs (Dawe et al. 1991, Comeau et al. 1998). Even sea spiders, the taxon that has the largest reported % increase associated with a single molt to maturity ($41.5 \pm 6.2\%$ for males and $57.2 \pm 3.1\%$ for females), do not achieve the size increase necessary to account for the largest horseshoe crabs in the Bay (Tomaschko et al. 1997).

Growth

Growth rates. We calculated juvenile growth rates directly from the real-time series in Fig. 4, and indirectly from the aggregated data in Fig. 5. The directly estimated growth rates for juveniles 3.0 to 12.0 mm (Table 6) were reasonably similar to growth rates obtained from aggregated data among juveniles in the same size range (Instars 1 to 5, Table 7).

We estimated juvenile growth rates for aggregated data by comparing the mean prosomal width of instars with estimated mean age of instars calculated from intermolt time increments described in the literature (Sekiguchi et al. 1988; Table 7). These relationships are depicted in Fig. 7, and the slopes of each line pro-

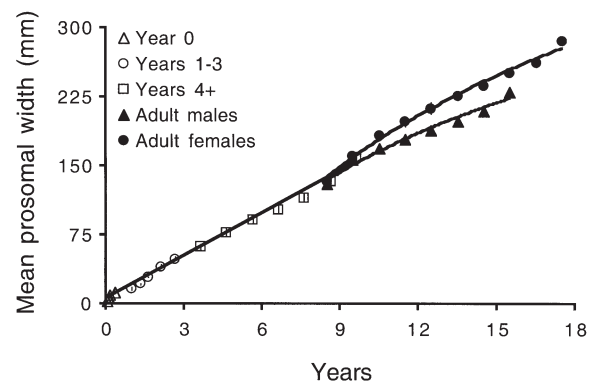


Fig. 7. *Limulus polyphemus*. Growth rates of juvenile and adult horseshoe crabs in Pleasant Bay, compared as increase in prosomal width \pm SE over time. Juvenile growth rates were determined based on size to age relationships in Table 7. Adult growth rates assume 1 molt per year. We used Model II regression to derive equations for the best-fit line describing relationships for juveniles: $y = 15.48x + 5.65$, $R^2 = 0.99$, $F_{reg} = 3263.51$, adult males: $y = 150.38 \ln(x) - 189.76$, $R^2 = 0.98$, $F_{reg} = 230.77$, and adult females: $y = 196.65 \ln(x) - 286.20$, $R^2 = 0.99$, $F_{reg} = 492.95$. All F_{reg} are significant to $p < 0.001$

Table 6. *Limulus polyphemus*. Direct estimates of growth (\pm SE) and mortality of juvenile cohorts of Fig. 4, sampled in real-time across 3 semilunar periods during the spawning season at a single location in Pleasant Bay. No. is total abundance, and size is mean prosomal width (\pm SE) of crabs in each cohort. We estimate 44 d of growth for Cohorts 1 and 2, and 30 d of growth for Cohort 3

Cohort	1		2		3		4	
	No.	Size (mm)	No.	Size (mm)	No.	Size (mm)	No.	Size (mm)
Semilunar interval								
5–19 Jul	77	5.0 \pm 0.03	155	3.1 \pm 0.01	–	–	–	–
20 Jul–2 Aug	29	7.0 \pm 0.06	108	5.1 \pm 0.02	26	3.2 \pm 0.04	–	–
18 Aug–1 Sep	1	12.3	24	9.2 \pm 0.07	18	7.1 \pm 0.09	2	5.0 \pm 0.05
Growth rate (mm d ⁻¹)	0.17 \pm 0.03		0.14 \pm 0.07		0.13 \pm 0.10		–	
% mortality	99		85		31		–	

Table 7. *Limulus polyphemus*. Approximate mean duration, age, and growth rates (\pm SE) of instars from Fig. 4 cohorts. Mean instar duration was calculated from information in Sekiguchi (1988) and was used to estimate age of instars in this study. Growth rates were calculated from the mean prosomal widths in Table 6 and age of each instar

Instar	Instar duration (d)	Age (yr)	Growth rate (mm d ⁻¹)
1	16.6	0.05	0.17 \pm 0.01
2	10.9	0.08	0.17 \pm 0.01
3	12.5	0.11	0.18 \pm 0.01
4	15.8	0.15	0.17 \pm 0.01
5	71.0 (22–120) ^a	0.35	0.10 \pm 0.01
6	182.5	0.97	0.05 \pm 0.003
7	121.7	1.3	0.05 \pm 0.001
8	121.7	1.6	0.05 \pm 0.001
9	182.5	2.1	0.05 \pm 0.002
10	182.5	2.6	0.05 \pm 0.002
11	365.0	3.6	0.05 \pm 0.003
12	365.0	4.6	0.05 \pm 0.003
13	365.0	5.6	0.04 \pm 0.002
14	365.0	6.6	0.04 \pm 0.002
15	365.0	7.6	0.04 \pm 0.001
16	365.0	8.6	0.04 \pm 0.001
17	365.0	9.6	0.04 \pm 0.001

^aRange of days crabs may spend in Instar 5

vide estimates of growth rate for juveniles, adult males, and adult females (Fig. 7). The growth rate of juveniles was significantly faster than that of adults and growth rates for adults of both sexes slowed as crabs aged (Fig. 7). There was no significant difference in growth rate between adult males and females (ANOVA, $F = 4.43$, $p = 0.057$ ns) (Fig. 7). These results suggest that males may live up to 16 yr and females up to 18 yr, consistent with the 14 to 19 yr horseshoe crab life span estimated by Ropes (1961) and Botton & Ropes (1988).

If there were more than 1 yr between molts, we might find crabs in each size class that are older. For example, 1 molt per 3 yr would result in a life span of up to 26 yr for males and 29 yr for females. Such life spans may be possible; lobsters, for example, continue

to molt after maturity and may reach 50 to 70 yr of age (Govind 1995). Some primitive spiders live more than 20 yr, with females having the longest life expectancy (Foelix 1982). Since we lack more information on molt frequency of adult horseshoe crabs, we limit ourselves to the parsimonious annual interpretation of cohorts.

Percent growth. Percent growth was highest among juvenile horseshoe crabs under 40 mm, and diminished as crabs became larger (Fig. 8), as is common in many animals (Mauchline 1976, Miller & Watson 1976, Fogarty 1995, Valiela 1995, Hart 2001). The magnitude of this decrease is comparable to that observed for Cape Cod horseshoe crabs reared in the laboratory (R. S. Jackson unpubl.) and is virtually identical to that of snow crabs (Comeau et al. 1998).

There are some anomalies in Fig. 8, particularly a growth spurt at sexual maturity. This sudden increase in prosomal width could be due to missing cohorts, but the change in growth was found independently and consistently among subadults, males, and females, and is consistent with increments measured in the field by Shuster (1955). This growth spurt is also associated with larger % increase in growth at Instars 16 to 19, just after sexual maturity, in females compared to males (Table 5, Fig. 8). A growth spurt at sexual maturity occurs in sea spiders, where females increase in body size more than males (Tomaschko et al. 1997), resulting in a difference in size among sexes of this species. Since growth did not differ between sexes during other stages of life, this period of increased growth in females just after maturity could account for the size difference between male and female horseshoe crabs in Pleasant Bay.

Hiatt growth. Growth increments were significantly larger in juveniles than in adults and among adult females than adult males in our Hiatt growth comparisons (Table 8). Hiatt analysis compares growth increments between juveniles and adult males and females by relating post-molt to pre-molt sizes, and is necessary for comparison of horseshoe crab growth to growth of many crustaceans. This method shows a dif-

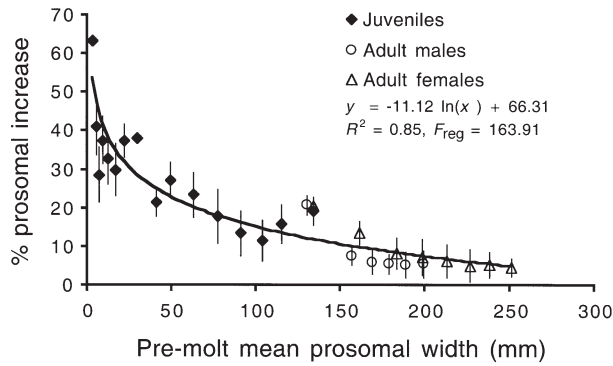


Fig. 8. *Limulus polyphemus*. Percent prosomal increase (\pm SE) versus pre-molt size of each successive cohort from hatchling through adult. Cohort sizes are given in mean prosomal width (mm). We performed Model II regression on the log-transformed values of x to generate an equation of the best-fit line describing % growth for the population. F_{reg} is significant to $p < 0.001$

ference in growth between adult males and females that is not found when comparing growth in terms of increase in size over time (Fig. 7). The growth increments of horseshoe crabs in Pleasant Bay overlap extensively with Hiatt growth relationships among different populations of lobsters (values compiled in Fogarty 1995) and juvenile snow crabs (Comeau et al. 1998).

Mortality

Juveniles. We calculated mortality directly from the real-time series in Fig. 4 and indirectly from the abundance data in Table 9, which were aggregated from our abundance transects. We found similar patterns of mortality for the instars which overlap between the real-time and aggregated cohorts (Tables 6 & 9), where total mortality was generally close to 99% by Instar 5 (ca. 12 mm). Comparing across all juvenile size classes in the aggregated data, mortality was highest in Year 0 and remained roughly constant after proso-

Table 8. *Limulus polyphemus*. Hiatt growth statistics, comparing mean pre-molt to mean post-molt sizes among juveniles (hatchling to maturity) and adult males and females. Type II regression was used to derive each growth relationship. Each F_{reg} was significant to $p < 0.0001$. Slopes and y -intercepts were significantly different among groups (ANCOVA: $F_{reg} = 1105.78$, $p < 0.001$; Fisher's PLSD, $p < 0.0001$)

	Juveniles	Males	Females
y :	$1.14x + 2.81$	$0.92x + 28.01$	$0.94x + 30.30$
R^2 :	0.99	0.98	0.94
F :	4759.05	81.01	447.25

mal width 21.7 mm (Instar 7, Fig. 9, top). Abundance of juveniles decreased exponentially with prosomal width during Year 0, and the slope of this line provides a rough estimate of mortality rate for these crabs. Similar patterns of high size-dependent mortality among juveniles are common to invertebrates (Gosselin & Qian 1997, Wang & Haywood 1999).

Estimated mortality at each juvenile instar ranged from 0 to 80% (Table 9). The range of % mortality among juvenile instars for which we have data overlaps with the range of annual % natural mortality for juvenile rock crabs (65 to 85%) and American lobsters (2 to 70%) (Reilly & Saila 1978, Fogarty 1995).

Adults. Adult abundance of both sexes decreased exponentially with increasing prosomal width (Fig. 9, bottom & Table 10), and the slopes of these lines provide rough estimates of mortality rate for each sex.

Table 9. *Limulus polyphemus*. Abundance and % mortality of juvenile horseshoe crabs at Instars 1 to 14 in Pleasant Bay nursery grounds. % mortality was calculated at each successive instar and accumulated across instars. CV = coefficient of variation (standard error / mean \times 100)

Instar	Abundance ($\times 10^6$)	CV (%)	% mortality	
			Per instar	Cumulative
1	8.00	75	58	–
2	3.38	80	80	92
3	0.66	48	27	94
4	0.48	88	63	98
5	0.18	84	67	99
6	0.06	100	–	–
7	0.15	89	36	99.6
8	0.10	100	–	–
13	0.20	100	0	–
14	0.20	100	–	–

Table 10. *Limulus polyphemus*. Abundance and % mortality of adult horseshoe crabs within Pleasant Bay. % mortality was calculated for each successive instar. Standard error of adult abundance at each instar is $\pm 27\%$, propagated from error of total abundance

Instar	Abundance		% mortality	
	Males	Females	Males	Females
16	853	505	–	–
17	27344	6356	–	–
18	105355	11397	8	–
19	96883	22275	12	–
20	85644	57962	75	69
21	21211	18029	60	13
22	8514	15738	90	29
23	853	11247	–	40
24		6702		92
25		506		–

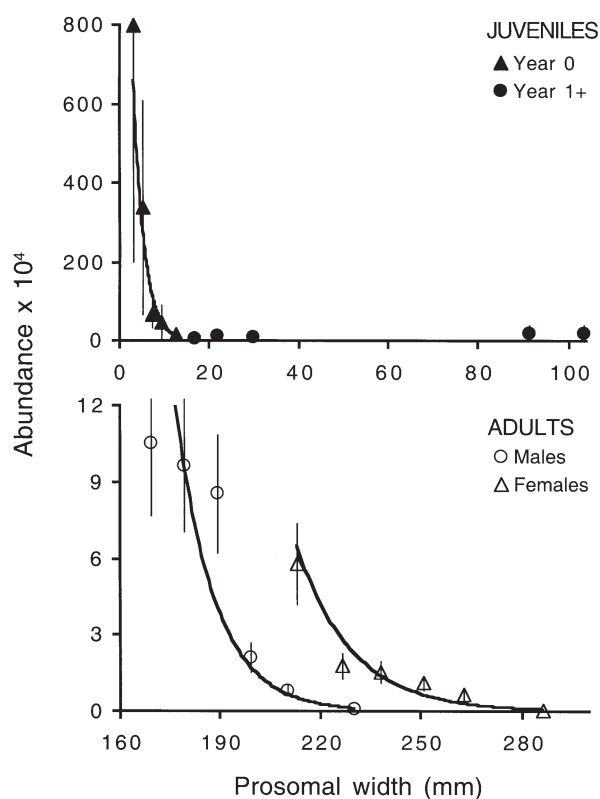


Fig. 9. *Limulus polyphemus*. Change in abundance ($\times 10^4$) of juvenile (top) and adult (bottom) horseshoe crabs with increasing prosomal width. We performed Model II regression on log-transformed values of y to obtain the best-fit line describing mortality rate for juveniles: $y = 2379.5e^{-0.42x}$, $R^2 = 0.94$, $F_{\text{reg}} = 45.95$, adult males: $y = 5 \times 10^7 e^{-0.09x}$, $R^2 = 0.93$, $F_{\text{reg}} = 49.83$, and adult females: $y = 3 \times 10^6 e^{-0.06x}$, $R^2 = 0.91$, $F_{\text{reg}} = 38.12$. All F_{reg} are significant to $p < 0.01$

Adult horseshoe crabs in Pleasant Bay had a lower estimated rate of mortality than juveniles (ANCOVA, $F = 4.36$, $p < 0.05$; Fisher's PLSD, $p < 0.0001$), and there was no significant difference in estimated mortality rate between adult males and females (ANCOVA, $F = 2.91$ ns). Estimated mortality rates were calculated only for prosomal widths >169 mm for males and >213 mm for females because these are the mean sizes of cohorts of peak abundance for each sex (Table 10).

Percent mortality increased with size among adult males and females, with an estimated 90% of crabs lost in the transition to last instar (Instar 23 for males and 25 for females, Table 10). Mortality at intermediate instars ranged from 8 to 75% in males and 13 to 69% in females (Table 10). High mortality at the last instar may have been due to senescence. Mortality of intermediate instars may be due in part to stranding, which can account for 10% loss of adult horseshoe crabs annually (Botton & Loveland 1989), and older horseshoe crabs may be most susceptible to

mortality by stranding and parasitism (Penn & Brockmann 1995).

Females experienced highest % mortality at Instar 20 (Table 10). We do not know the reason for a spike in mortality at this instar. Adult abundance may have been affected by differences in annual recruitment or migration of individuals in and out of the local population (Gosselin & Qian 1977), neither of which can be separated from mortality.

Our sex ratio data suggested selective loss of females compared to males after sexual maturity. Because mortality rate is similar among sexes at the prosomal widths we examined (>213 mm, Instar 20 for females), these data do not explain the male-dominated abundance of crabs in Pleasant Bay. A difference in mortality between sexes that affected adult sex ratio may have occurred at smaller adult prosomal widths, between sexual maturity and peak abundance (134 to 198 mm, Instars 16 to 19 for females).

Recruitment

Recruitment of juvenile crabs into the adult population depends on the number of young produced in the Bay each year and their survival. To estimate recruits into the population, we determined the number of eggs produced per year by first estimating length and peak of spawning period and fecundity of females during this period. We then used this information to estimate survival from egg to hatchling.

Spawning period. The spawning period in Pleasant Bay may have started as early as late March and continued through mid-July 2001 (Table 11). We back-calculated the approximate time of first spawning in the Bay from our estimates of juvenile growth rates and assumed crabs hatched into Instar 1 in the semilunar interval after they were spawned (14 d). In the first sampling interval in which we found Year 0 juveniles,

Table 11. *Limulus polyphemus*. Range of juvenile instars in Year 0 and Years 1+ that were sampled in Pleasant Bay during semilunar intervals of the 2001 spawning season. Estimated spawning dates cover the full range of semilunar intervals in which juveniles were spawned based on their rates of growth

Sampling interval	Instars		Estimated spawning dates for Year 0 instars
	Year 0	Years 1+	
5–20 Jun	–	8–17	
21 Jun–4 Jul	1–6	7–17	24 Mar–4 Jun
5–19 Jul	1–3	–	22 May–20 Jun
20 Jul–2 Aug	1–6	7–11	23 Apr–4 Jul
3–17 Aug	–	–	
18 Aug–1 Sep	2–6	7–11	22 May–19 Jul

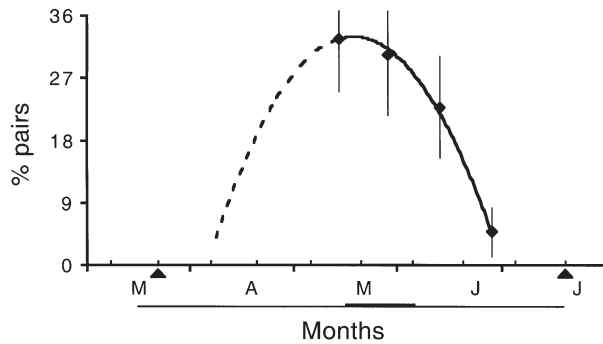


Fig. 10. *Limulus polyphemus*. Percentage of paired horseshoe crabs in the general adult population (\pm SE) compared to months of the spawning season ($y = -0.02x^2 + 4.90x - 306.72$, $R^2 = 0.99$). % pairs in each month is plotted on the mean day of each ~2 wk semilunar cycle (inner tick marks) during which we sampled. Also shown are estimated % pairs for the portion of the spawning season not sampled (---), beginning and end of spawning season predicted from juvenile growth (\blacktriangle), and estimated length (—) and peak (\blacktriangle) of spawning seasons in New Jersey, Delaware, and South Carolina

we estimated that crabs at Instar 5 represented the largest crabs <1 yr old (Table 11). We did not estimate crabs in Instar 6 to be <1 yr old because some of these juveniles may have been spawned last season (cf. Fig. 4, cohort description). We used the minimum duration of this instar (Table 7) to calculate age because back-calculating from the mean estimated age would predict spawning in February, well outside the spawning season. We estimated the time of last spawning from the smallest juveniles (Instar 2) found during our last sampling interval (Table 11). From mean age of this instar we estimate that spawning in Pleasant Bay ended in early to mid-July, accounting for the absence of a 3 mm cohort in the bottom panel of Fig. 4.

We found the greatest percentage of animals in pairs during mid- to late May (Fig. 10). We determined an approximate peak in spawning by comparing the % of paired crabs in the general population across semilunar periods of the spawning season. This method also confirmed our estimate of length of spawning season. We used a polynomial curve fit to the % pairs and semilunar interval data to extrapolate to the start and end of the spawning season (early April to early July, peaking in late May). The resulting estimate (Fig. 10) is close to the spawning period predicted from our observations of juvenile growth (Table 11), and is consistent with length and peak of spawning season in New Jersey, Delaware, and South Carolina (Shuster & Botton 1985, Thompson 1998, ASMFC 1998, Smith et al. 2002).

Female fecundity. We estimate that 99.8% (1.5×10^5 individuals) of the female population in the Bay was likely able to spawn and could have produced $1.3 \times 10^{10} \pm 27\%$ eggs during the 2001 spawning season. We

made these calculations by assuming each mature female between 157 mm and 295 mm prosomal width deposited 88 000 eggs in intertidal areas (cf. Fig. 2). We estimated the proportion of adult females potentially spawning in the Bay by comparing the size range of females in the general population with the size range of females in pairs during this study (157 to 265 mm) and in spawning aggregates (up to 295 mm; James-Pirri et al. unpubl.) in Pleasant Bay. We combined these sizes to estimate that prosomal widths of 157 to 295 mm represent the range of sizes of adult females that were actively spawning in the Bay.

We estimate that 1.7×10^9 eggs were deposited per semilunar cycle, assuming that the total number of eggs produced in the Bay was deposited evenly among the 8 periods. From our extrapolation of length of spawning season, the time over which eggs were deposited includes 8 semilunar intervals from late March through early July (Fig. 10). We expect, however, the magnitude of egg production to follow spawning frequency, so that most eggs were deposited in the Bay during the peak of spawning.

Just over 4×10^9 eggs were likely produced in the Bay during the 2 semilunar intervals of peak spawning (Table 12). We used the equation relating % pairs and spawning intervals in Fig. 10 to estimate egg production in each semilunar interval of the 2001 spawning season, assuming our data on % pairs represent the % of females spawning in the Bay. Our data from Fig. 10 sum to $134 \pm 28\%$. Since this number may not be significantly higher than 100%, it suggests that a minority of females spawned their eggs in multiple clutches at different times and possibly different locations in the Bay (Cohen & Brockmann 1983, Shuster & Botton 1985). The majority of females appear to have deposited their eggs during a single lunar cycle.

Table 12. *Limulus polyphemus*. Estimated egg production per spawning interval in Pleasant Bay during the 2001 spawning season. Mean number of females paired at each interval was estimated from the equation for the line in Fig. 10. Standard error of egg production per interval is $\pm 39\%$ and was propagated from error of % pairs and total annual egg production

Spawning interval	% pairs	% SE pairs	Eggs/interval
24 Mar–6 Apr	0	0	0
7–22 Apr	13	4	1.7×10^9
23 Apr–6 May	27	8	3.6×10^9
7–21 May	33	9	4.4×10^9
22 May–4 Jun	32	9	4.2×10^9
5–20 Jun	23	6	3.0×10^9
21 Jun–4 Jul	6	2	7.9×10^8
5–19 Jul	0	0	0
20 Jul–2 Aug	0	0	0

Table 13. *Limulus polyphemus*. Survival of juvenile horseshoe crabs at different life stages from egg through sexual maturity in Pleasant Bay. Adult refers to the first adult instar after maturity that did not overlap in size with juvenile instars

Stage of life	% survival
Egg–hatchling (Instar 1)	0.06 ± 0.02
Egg–Year 1 (Instar 7)	0.001 ± 0.001
Egg–subadult (Instar 14)	0.001 ± 0.001
Subadult–adult (Instar 18)	78 ± 27

Egg density and shorebird predation. From our estimate of total annual egg production, egg density in the Bay would have been approximately 1.1×10^5 eggs m^{-1} of shoreline. This estimate assumes eggs were distributed evenly across the 114.8 km of shoreline in Pleasant Bay (RMP 1998). Actual density of eggs was likely higher in areas where spawning was more intense and lower in areas of the Bay in which little or no spawning occurred. The average density of eggs was comparable to egg densities that were observed on Delaware Bay beaches in May and June in the early 1990s and could have fed 1.4×10^4 shorebirds d^{-1} during the 16 wk (112 d) spawning season in Pleasant Bay (Botton et al. 1994). This estimate is based on the assumption of Botton et al. (1994) that each bird eats 8300 eggs d^{-1} . The actual number of shorebirds that might exploit horseshoe crab eggs in Pleasant Bay during the spawning season has not been documented. This information is needed to estimate of the portion of egg mortality that may be due to shorebird predation.

Survival to juvenile stages. We estimate 0.06% of the total eggs produced in the Bay survived to hatchling stage, 0.001% survived to Instar 7 (Year 1), and roughly this same number survived to Instar 14 (Year 6), the oldest juvenile instar for which we have abundance data (Table 13). Hence, the greatest loss of recruits occurred between egg and hatchling stage. We estimated survival by comparing the changes in abundance from egg to hatchling, Instar 7 and Instar 14, using the data in Table 9.

Survival to maturity. We estimate that $78 \pm 27\%$ of subadult juveniles at Instar 14 survive to reach adulthood (Instar 18) (Table 13). We chose to estimate recruitment to adulthood by estimating survival to Instar 18 because this was the first adult instar that did not overlap with juvenile stages (Tables 4 & 5).

Although females produced many eggs, few survived their first year in which mortality was the highest, but most juveniles surviving to Year 1 survived to reach adulthood. These results suggest that the first year of life is a critical period for horseshoe crabs and survival during this year may significantly affect recruitment into the adult population.

CONCLUSIONS

We found that juveniles were more abundant and suffered greater mortality than adults. Adults were largely found on the sediment surface, but 20% were buried shallowly in sediments. The male:female sex ratio in juveniles was 1.4:1, but the adult sex ratio was skewed to 2.3:1. Juveniles grew faster than adults, and adult crabs may plausibly molt as frequently as once per year rather than have a terminal molt. The spawning season may have spanned late march to mid-July, and juveniles hatched at semilunar intervals during Year 0, growing to 16.6 ± 0.9 mm prosomal width by the start of Year 1. The distinct semilunar cohorts of Year 0 coalesced into annual cohorts after Year 0. Females deposited large numbers of eggs, but only 0.001% survived to the end of Year 0, and approximately 78% of these juveniles reached adulthood. Despite its taxonomic distinctiveness, *Limulus polyphemus* is ecologically similar to other arthropods to which we compared our data, including primitive spiders, sea spiders, various crustaceans, and other marine invertebrates.

The results discussed above document a first attempt at defining the population structure and dynamics of horseshoe crabs in an entire estuary. The results of this paper provide information essential for any plans for monitoring, evaluating, and sustaining this important economic and ecological resource. Collection of similar data in other areas could provide substantial information to determine the extent to which local populations differ in abundance and structure, assisting local managers in identifying those areas that may be most susceptible to pressures such as over-harvesting, disease or other sources of mortality. These data may also be extended to developing models to better define population dynamics for horseshoe crab populations coast-wide. Further study to improve these estimates should include a better definition of fecundity, in terms of egg production per female and the extent to which different local populations may be genetically distinct.

Acknowledgements. We are grateful for support from the Friends of Pleasant Bay and their contributors. Many thanks to Tony Davis and the crew at the Arey's Pond Boat Yard, who donated and maintained the boat, the Massachusetts Audubon Society for providing lodging near the Bay, and the Cape Cod National Seashore for access to areas within their boundary. Thanks to Dr. George Hampson for guidance with development of our view box, and Atlantic Aquaculture in Bristol, MA, for design collaboration and custom-building our sampling rake. Thanks also to Frank Germano at the Massachusetts Division of Marine Fisheries, Dr. Carl Shuster for sharing his literature, and to the many residents and fishermen living and working on the Bay who provided insight and access to the Bay.

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Editorial responsibility: Kenneth Tenore (Contributing Editor), Solomons, Maryland, USA

Submitted: June 11, 2002; Accepted: October 2, 2002
Proofs received from author(s): December 9, 2002