Effects of Land Use and Physicochemical Water Quality on Grass Shrimp, *Palaemonetes pugio*, and its Parasitic Isopod, *Probopyrus pandalicola*, in South Carolina, USA Tidal Creeks



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Effects of Land Use and Physicochemical Water Quality on Grass Shrimp, *Palaemonetes pugio*, and its Parasitic Isopod, *Probopyrus pandalicola*, in South Carolina, USA Tidal Creeks

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Abstract

Grass shrimp, Palaemonetes pugio, are a common inhabitant of US East and Gulf coast salt marshes and are a food source for recreationally and economically important fish and crustacean species. Due to the relationship of grass shrimp with their ecosystem, any significant changes in grass shrimp population may have the potential to affect the estuarine system. Land use is a crucial concern in coastal areas where increasing development impacts the surrounding estuaries and salt marshes and has made grass shrimp population studies a logical choice to investigate urbanization effects. Any impact on tidal creeks will be an impact on grass shrimp populations and their associated micro-environment whether predator, prey or parasitic symbiont. Anthropogenic stressors introduced into the grass shrimp ecosystem may even change the intensity of infections from parasitic symbionts. An ectoparasite found on *P. pugio* is the bopyrid isopod Probopyrus pandalicola. Little is known about factors that may affect the occurrence of this isopod in grass shrimp populations. The goal was to analyze the prevalence of *P. pandalicola* in grass shrimp in relation to land use classifications, water quality parameters, and grass shrimp population metrics. Eight tidal creeks in coastal South Carolina were sampled monthly over a three year period. The occurrence of P. pandalicola ranged from 1.2% to 5.7%. Analysis indicated that greater percent water and marsh coverage resulted in a higher incidence of bopyrid occurrence. Analysis also indicated that higher bopyrid incidence occurred in creeks with higher salinity, temperature, and pH but lower dissolved oxygen. The land use characteristics found to limit bopyrid incidence were limiting to grass shrimp (definitive host) populations and probably copepod (intermediate host) populations as well.

Introduction

Grass shrimp, *Palaemonetes pugio*, are a common inhabitant of salt marshes along the East and Gulf coast of the US and are an important food source for recreationally and commercially viable fish and crustaceans. Grass shrimp are abundant in their preferred habitat and can account for up to 56% of the total macrofauna in South Carolina estuarine tidal creeks (Leight et al. 2005). These tidal creeks are not only productive grass shrimp habitat, but also feeding grounds, spawning sites, and nurseries for fish, shellfish, birds and mammals (Sanger et al. 2008). Grass shrimp, which help to accelerate the breakdown of detritus, are thus instrumental in transferring energy from lower levels to higher levels in the estuarine food chain (Anderson 1985). Due to the relationship of grass shrimp with their ecosystem, any significant changes in grass shrimp population may have the potential to affect the estuarine system. This, along with the fact that land use is a crucial concern in coastal areas where increasing development (urbanization, golf courses) impacts the surrounding estuaries and salt marshes, has made grass shrimp population studies a logical choice for urbanization effects (Key et al. 2006).

Research has shown that urban and suburban growth surrounding tidal creeks can reduce grass shrimp populations as compared to populations in non-urbanized estuaries (Vernberg et al. 1996; Leight et al. 2005; Daugomah 2007). Tidal creeks can be the first area of impact from non-point source runoff resulting in contaminant levels higher in these areas than in the open water environment downstream (Sanger et al. 2008). Increasing stormwater runoff resulting from increases in impervious covers, characteristic

of human development, can not only cause increased contaminant and bacterial loads but alter food webs and salinity as well (Holland et al. 2004).

Characterizing the impact of stresses resulting from urbanization on estuaries is needed to provide a scientifically valid basis for making land-use management decisions in the coastal zone. Any impact on tidal creeks will be an impact on grass shrimp populations and their associated micro-environment whether predator, prey or parasitic symbiont. The effects of suburban and urban development on grass shrimp parasites are not known outside of the effects themselves on grass shrimp populations. Parasites can create direct and indirect effects on such parameters as grass shrimp reproduction, growth and ultimately, viability of the shrimp (host) population. Anthropogenic stressors introduced into the grass shrimp ecosystem may even change the intensity of parasitic infections thus affecting the previously mentioned parameters. One of the most common parasites found on *P. pugio* in South Carolina is the bopyrid isopod, *Probopyrus pandalicola*. This parasite is known to cause reproductive sterility in shrimp (Beck 1980a), decreased respiration and decreased metabolic activity (Anderson 1975). Bopyrid prevalence in grass shrimp has recently been reported to be over 3% in a one year study of four southeastern US creeks (Chaplin-Ebanks and Curran 2007).

The overall goal of the present research was to collect grass shrimp, *Palaemonetes pugio*, at monthly intervals over a three year period and relate grass shrimp parasite prevalence to surrounding land use and water quality parameters. Land use characterization around seven estuarine creeks at Kiawah Island, SC and one reference estuarine creek at

Wadmalaw Island, SC was determined. Shrimp were collected monthly to investigate the relationship between land use classifications, physicochemical parameters, grass shrimp population metrics and the prevalence of the parasitic isopod, *P. pandalicola*.

Materials and Methods

Study Sites

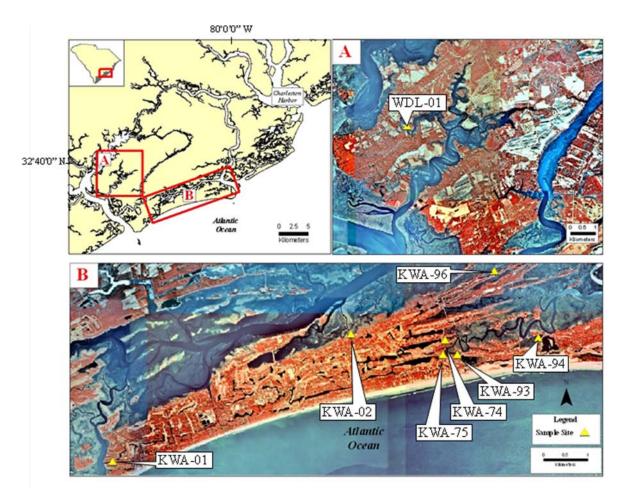
Grass shrimp were collected from seven sites on Kiawah Island, SC (KWA-01, 02, 74, 75, 93, 94, 96) and one reference site on Wadmalaw Island, SC (WDL-01) by pushnet from January 2004 through December 2006 (Figure 1). Kiawah Island is a 29 km² barrier island located directly on the southeastern SC coast. It is a tourist destination composed of over 3000 housing units (106/km²) and five golf courses with a permanent resident population density of 38 people/ km² (City-Data.com 2010a). Wadmalaw Island is a 108.5 km² sea island located just west of Kiawah Island. It is a rural unincorporated area composed of just over 1060 housing units (10/ km²) with a population density of 27 people/km² (City-Data.com 2010b).

Sampling Design

All creeks sections sampled were 95 m in length to accommodate 3 transects (each 25 m long with 10 m buffers) on each bank. Sampling began on an ebb tide two to three hours prior to predicted low tide when the water level had just fallen out of the marsh and most of the water was in the main creek channel. All of the Kiawah Island sample sites were located within 100 m of retention pond overflows. Sampling was performed by foot using a pushnet method accomplished by holding the net vertically in the water near the

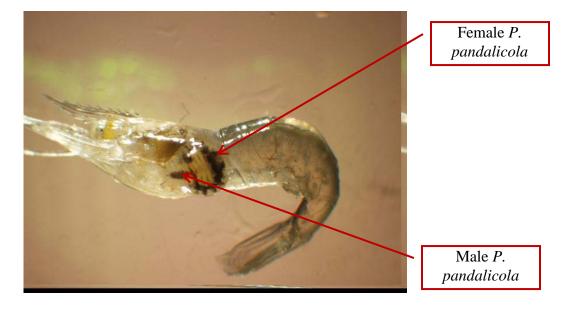
marsh edge and water line. The net had a mouth opening of 1000 cm^2 (40 x 25 cm) with a mesh size of 3 mm. The net was submerged and care was taken to prevent the net from contacting the sediment bottom. This sampling protocol is described in detail by Leight et al. (2005). Grass shrimp samples were stored on ice until processed at the lab and then transferred to 90% ethanol within 24 hours. Each sample was analyzed under a

Figure 1. Maps of sampling sites in South Carolina. Map A shows the Wadmalaw Island, SC site. Map B shows the seven sites located on Kiawah Island, SC.



dissecting microscope and individual grass shrimp were separated based on sex. For large samples of a 100 shrimp or greater, a subsample of 30 individuals was randomly selected for analysis. Individuals were then measured for length (mm) and weight (mg). Only grass shrimp determined to be *Palaemonetes pugio* according to Holthuis (1952) were counted.

Figure 2. Grass shrimp, *Palaemonetes pugio*, with bopyrid isopod parasite, *Probopyrus pandalicola*, attached to branchial (gill) chamber.



All shrimp analyzed were noted for the presence of the bopyrid isopod, *P. pandalicola*. These parasites were easily determined by their attachment under the carapace to the branchial chamber of the host shrimp (Figure 2). As Figure 2 shows, the large mass is the female *P. pandalicola* while the much smaller male *P. pandalicola* is located among the female's pleopods at her posterior end. When parasites were counted on a shrimp, only the large female was counted. The first *P. pandalicola* to reach a host shrimp's gill chamber develops into a female with all subsequent ones becoming male (Anderson 1990). However, if both gill chambers were occupied by *P. pandalicola*, then both parasites were counted.

At the time of sampling, water quality measurements of temperature (°C), salinity (‰), and dissolved oxygen (mg/L) were made using a YSI 85 meter (YSI Inc, Yellow Springs, OH). pH was measured with an Oakton phTestr (Vernon Hills, IL).

ArcGIS 8.x, Image Analysis Extension, and ERDAS (ESRI, Charlotte, NC) were used to delineate land use classifications on a National Aerial Photography Program image along with Image Analysis Seed Tool Properties and Find Like Areas function. Land use classes were merged using ArcGIS geoprocessing tools. Random points within the maps were selected and verified in situ to determine the overall accuracy of the maps. Land use classification delineations were calculated using an error matrix method that was 95% accurate (ESRI, Charlotte, NC). Land use classifications were divided into three categories: % urban which included paved and gravel roads, buildings, cultivated vegetation and docks; % woods which was all noncultivated vegetation; and % marsh which consisted of marsh vegetation and water.

Statistics

PROC FREQ and PROC MEANS (SAS, Cary, NC) were used to generate the tabular information. A generalized linear model [PROC GENMOD (SAS, Cary, NC)] was used to examine the relationship between the bopyrid parasite prevalence and the following variables: physical water quality parameters (temperature [°C], salinity [‰], dissolved oxygen [mg/L], pH); grass shrimp population metrics (% males, % females, % gravid females, length, weight); seasons (Winter, Spring, Summer, Fall); and specific land use characteristics as discussed previously. Analyses were performed using SAS 9.1 with

alpha = 0.05. PROC GENMOD (distribution; binomial; link: logit) assumes the relationship between coefficient and probability is $\beta_0 + \beta_1 x = \log (\pi/1-\pi)$ where β_0 is the intercept, β_1 is the coefficient, *x* is the independent variable and π is the probability of incidence of parasites.

Results

The land use cover (Table 1) reveals that sites KWA-01 and KWA-75 had the largest urban coverages. Sites KWA-94 and KWA-96 had the most marsh coverages. The highest percent woods coverage occurred at KWA-02. The reference site, WDL-01, had urban coverage toward the low end (6%) and woods and marsh coverages toward the high end (25% and 69%, respectively).

Table 1. Land use cover immediately surrounding grass shrimp collection sites onKiawah and Wadmalaw Islands, SC and associated 3-year average water qualityparameters with standard errors in parentheses.

Site	Urban [%]	Woods [%]	Marsh [%]	D.O. [mg/L]	Temp. [°C]	pН	Salinity [‰]
KWA-01	70	10	20	6.8	20.9	7.8	30.5
				(0.4)	(1.1)	(0.06)	(0.6)
KWA-02	25	44	31	7.1	20.8	7.6	30.2
				(1.5)	(1.1)	(0.03)	(0.5)
KWA-74	31	20	49	6.5	22.3	8.0	13.5
				(0.4)	(1.1)	(0.04)	(0.9)
KWA-75	59	6	35	10.3	22.2	8.2	14.7
				(2.7)	(1.2)	(0.04)	(0.9)
KWA-93	32	0	68	8.8	21.3	8.1	18.9
				(2.7)	(1.2)	(0.04)	(0.9)
KWA-94	11	6	83	7.7	21.5	7.8	26.6
				(1.9)	(1.1)	(0.04)	(0.8)
KWA-96	2	18	80	5.2	21.1	7.6	30.1
				(0.4)	(1.1)	(0.04)	(0.6)
WDL-01	6	25	69	6.3	21.4	7.4	27.0
				(0.8)	(1.2)	(0.03)	(0.9)

The average dissolved oxygen measurements over the three year period ranged from a high of 10.3 mg/L for KWA-75 down to 5.2 mg/L for KWA-96 with WDL-01 averaging 6.3 mg/L. Average temperatures ranged from 22.3°C for KWA-74 down to 20.8°C for KWA-02 with WDL-01 averaging 21.4°C. Average pH ranged from 8.2 for KWA-75 down to 7.4 for WDL-01. Average salinities were the most variable water quality parameter in these tidal creeks ranging from 30.5‰ for KWA-01 down to 13.5‰ for KWA-74 with WDL-01 at 27.0‰ (Table 1).

The parasite prevalence in grass shrimp averaged 3.17% for all eight sites (Table 2). The highest parasite prevalence was 5.67% at site KWA-93 which had the lowest woods cover. This parasite prevalence was statistically higher (p<0.0001) than the reference site WDL-01 (2.4%). Site KWA-74 with the second highest prevalence (statistically higher [p<0.0001] than the reference site WDL-01) had the lowest average salinity and the highest average temperature. The lowest parasite prevalence was 1.24% at KWA-01 which had the highest urban land cover (70%). The site with the second lowest incidence (KWA-02, 1.86%) had the highest woods cover (44%). Both KWA-01 and KWA-02 parasite prevalences were statistically lower (p=0.0005 for each) than WDL-01.

The highest probability of parasite incidence occurred in land use areas of higher percent marsh and lower percent woods (Table 3). There were no statistical differences (p=0.3318) in parasite prevalence directly associated with urban coverage. All four water quality parameters were found to have an effect on parasite prevalence with a higher probability

Table 2. Summary of enumerating the incidence of the bopyrid parasite, *P. pandalicola*, in grass shrimp, *P. pugio*, at eight tidal creek sites over a three year period and associated statistics. Female (\bigcirc) and male (\bigcirc) percents included.

Site	Total	# Male	# Female	# Gravid	# shrimp	%shrimp	Site
	shrimp	shrimp	shrimp	female	with	with	notes
	collected			shrimp	parasite	parasite	
KWA-01	1207	623	504	80	15	1.24	Lower prob. of
					53.4%♀		shrimp parasite
					46.6%∂		than WDL-01
							(p=0.0005).
		~ = ~			~~~		Most urban.
KWA-02	1876	978	755	143	35	1.86	Lower prob. of
					60%♀ 40% ⊅		shrimp parasite
					40%♂		than WDL-01
							(p=0.0005). Most woods.
KWA-74	1933	931	871	131	89	4.60	Higher prob. of
K W A-74	1755	931	0/1	151	62.8%♀	4.00	shrimp parasite
					38.2%∂		than WDL-01
					50.2700		(p<0.0001).
							Lowest salinity.
							Highest temp.
KWA-75	1907	889	791	227	51	2.67	Highest pH.
					58.8% ♀		Highest D.O.
					41.2%	_	_
KWA-93	1797	901	788	108	102	5.67	Higher prob. of
					58.8%♀		parasitized
					41.2%♂		shrimp than
							WDL-01
							(p<0.0001).
WWA 04	1050	1027	5 00	224	<u> </u>	2 22	Least woods.
KWA-94	1959	1037	588	334	65 56 0% ()	3.32	Most marsh.
					56.9%♀ 43.1%♂		
KWA-96	1820	706	835	279	53	2.91	
IX (117)0	1020	700	055	21)	69.8% ♀	2.71	
					30.2%♂		
WDL-01	1708	953	612	143	41	2.40	
					46.3%♀		
					53.7%		
							Higher prob. of
					451		parasitized
Total	14207	7018	5744	1445	59.2% ♀	3.17	females than
					40.8% ♂		males
							(p<0.0001).

Parameter Category	Variable	Estimate	P value	Result
Grass shrimp	Male	-0.0731	0.0004	Higher probability
characteristic	Length			as male shrimp
				lengths decrease
	Female	-0.0394	0.0003	Higher probability
	Length			as female shrimp
_				lengths decrease
	Male		0.73	No differences
	Weight			
	Female		0.08	No differences
	Weight			
	Sex	0.5726	< 0.0001	Higher probabilit
				in female shrimp
Water quality	Salinity	-0.0212	0.0002	Higher probabilit
parameter	-			in creeks with
				lower salinity
	D.O.	0.0141	< 0.0001	Higher probabilit
				in creeks with
				higher D.O.
	Temp.	0.0616	< 0.0001	Higher probabilit
	-			in creeks with
				higher temp.
—	pН	0.4462	0.0006	Higher probabilit
	±			in creeks with
				higher pH
Land use characteristic	Urban		0.3318	No differences
_	Marsh	0.0087	0.0001	Higher probabilit
				in creeks with
				higher % marsh.
_	Woods	-0.0166	< 0.0001	Higher probabilit
				in creeks with
				lower % woods.

Table 3. Significant probabilities of incidence of bopyrid parasite on grass shrimp versus variables of grass shrimp characteristics, water quality parameters and land use characteristics in the eight tidal creek sites sampled over a three year period.

period.	•			
Comp	oarison	P value	Results	
Winter	Spring	0.0017	Significantly less parasitized shrimp in Winter.	
Winter	Summer	<0.0001	Significantly less parasitized shrimp in Winter.	
Winter	Fall	0.0005	Significantly less parasitized shrimp in Winter.	
Spring	Summer	0.0266	Significantly less parasitized shrimp in Spring.	
Spring	Fall	0.7351	No difference	
Summer	Fall	0.0334	Significantly less parasitized shrimp in Fall.	

Table 4. Statistical comparisons between seasons of the year of incidence of bopyrid parasite prevalence in grass shrimp from eight tidal creek sites over a three year period.

in sites with lower salinity but higher D.O., temperature and pH (Table 3). Length of the shrimp was significantly related with parasite prevalence. As shrimp length increased, parasite prevalence decreased in male and female shrimp. There was not a significant relationship with the weight of male or female shrimp (Table 3). Since the parasite reproductively sterilizes the host shrimp (Beck 1980a), gravid females were not included in analysis as it would have skewed the weight data and therefore gravid female data were excluded as well. A significantly higher probability of parasite incidence occurred in female shrimp as compared to male shrimp (Table 3).

Seasonally, over the three year sampling period, the highest probability of parasite occurrence was in summer with the lowest occurrence in winter (Table 4). During Winter, the reference site, WDL-01, contained significantly less parasitized shrimp than six of the seven Kiawah Island sites (Table 5). The differences between the Kiawah Island sites and the reference site at other seasons were fewer (Table 5).

Season	P values	Results
Winter	< 0.0001	KWA-74, 75, 93, 94, 95
		96 significantly more parasitized shrimp.
Spring	< 0.0001	KWA-74 significantly
		more parasitized shrimp.
Summer	< 0.0001	KWA-74 and 93
		significantly more
		parasitized shrimp.
Summer	0.01	KWA-01 significantly
		less parasitized shrimp.
T-11	0.0042 0.0056	
Fall	0.0043 - 0.0056	
		significantly more
		parasitized shrimp.
Fall	0.0253	KWA-02
		significantly less
		parasitized shrimp.

Table 5. Probabilities of incidence of bopyrid parasite on grass shrimp in seven tidal creeks during each season as compared to the reference site WDL-01 over a three year period.

Discussion

The bopyrid ectoparasite, *P. pandalicola*, is an Epicaridean isopod which is well-known ectoparasite of many shrimp species (Sinderman 1990). These parasites penetrate an uninfected shrimp near the base of the shrimp's pereiopods as a cryptoniscus larva. The parasite then becomes endoparasitic in the host's cephalothorax for up to two weeks. After this postinfection period, the parasite moves into the branchial chamber becoming ectoparasitic (Anderson 1990). It then attaches to the host with mouthparts forming a suctorial cone used to feed on blood (Kensley and Schotte 1989). The first parasite to reach one the host's branchial chambers develops into a female with both chambers susceptible to parasitic infection. All later parasites do not go through an endoparasitic period and instead move directly to the branchial chamber, attach to the female *P. pandalicola* and develop into males (Anderson 1990).

P. pandalicola has been found to infect over 16 palaemonid species of shrimp ranging from Brazil, to the Pacific Coast of Panama and up the Atlantic coast of the US to New Hampshire (Beck 1980a; Bunkley-Williams and Williams 1998). It has also been found to infect freshwater shrimp from several Caribbean islands (Bunkley-Williams and Williams 1998).

This present study indicated that bopyrid prevalence over a three year period ranged from 1.24% to 5.67% in *P. pugio*. This was similar to findings by Chaplin-Ebanks and Curran (2007) who measured prevalence over a one year period ranging from 1.3% to 3.1% in grass shrimp collected from four creeks in South Carolina and Georgia. In a study of

grass shrimp population metrics over a 10 year period, Leight et al. (2005) incidentally noted that bopyrid parasite prevalence remained "below 3%" in four South Carolina tidal creeks. An unnamed *Probopyrus* species infected 50% of a population of the shrimp *Hippolysmata wurdemanni* in Biscayne Bay, FL. Other bopyrid isopods, such as *Argeia pugttensis*, can cause infection rates of 3 to 5% in black-tailed shrimp (*Crago nigricauda*) of the Pacific coast of the US. While *Epipenaeon japonicus* can infect the Japanese red prawn (*Panaeopsis akayebi*) at rates up to 70% (Sinderman, 1990). Another Asian shrimp, *Metapenaeopsis dalei*, was infected by the bopyrid *Parapenaeon consolidatum* at rates of up to 68% (Choi et al. 2004). Of the shrimp hosts for *P. pandalicola*, some are considered to be economically important as food to humans (Beck 1980b).

All measured water quality parameters were shown to significantly affect parasite incidence (Table 3). There was a statistically higher probability of occurrence in creeks with lower salinity, higher D.O., higher temperatures and higher pH. Chaplin-Ebanks and Curran (2007) found no relationship between bopyrid occurrence and salinity or any other water quality parameter. The significant increase in bopyrid incidence in lower salinity waters is consistent with the *Probopyrus* genus as it is the only bopyrid genus that can be found in fully freshwater habitats (Markham 1986). *P. pandalicola* has been found to release its larvae both in freshwater habitats and in estuaries though it needs saline waters for larval development (Conner and Bauer 2010). Several freshwater shrimp species are parasitized by *P. pandalicola* (Bunkley-Williams and Williams 1998), including the freshwater *Palaemonetes* species *P. paludosus* (Beck 1980a).

Correlations between water quality parameters and parasite prevalence have been studied in other crustaceans. Clams (Anodonta piscinalis) subjected to low oxygen levels for 25 days were found to be more intensively infected by a parasitic copepod (*Paraergasilus* rylovi) than unstressed clams (Saarinen and Taskinen 2005). In the intertidal snail, Zeacumantus subcarinatus, increased temperatures were found to increase the cercariae emergence of one parasitic trematode species while inhibiting emergence of another trematode species (Koprivnikar and Poulin 2009). Survival of parasitic trematodes of another intertidal snail, *Cerithidea californica*, was found to be mixed upon exposure to high salinity (40 ‰) with one trematode species faring well while the other showed low survival (Koprivnikar et al. 2010). For this present study, the two sites (KWA-01 and 02) with the highest salinities also exhibited the lowest parasite occurrences. Temperature, dissolved oxygen and pH levels found at KWA-01 and 02 were neither higher nor lower than the other sites. While water quality parameters may play a part in affecting P. *pandalicola* incidence, they may also impose limits on the calanoid copepod intermediate host (usually Acartia tonsa; Anderson and Dale 1981). All these studies show that further work is needed to fully assess the impacts of changes in water quality parameters on crustacean parasites.

The highest probability of bopyrid occurrence in this present study was found to be in female grass shrimp (Table 2). Of the infected shrimp, 59% were female while 41% were male (Table 2). The sex ratio of the collected shrimp for all sites was 51% females to 49% males. Beck (1979, 1980b) collected the freshwater grass shrimp, *P. paludosus*, from several sites in Florida, USA. Females were found to be significantly more

parasitized than males with bopyrid infection levels so high that grass shrimp reproduction was minimal. The results were not quantified however (Beck 1979, 1980b). A higher female shrimp presence in a population would not necessarily explain higher female infection rates. Site KWA-02 consisted of 48% females over the sampling period yet 60% of the infected shrimp were female (Table 2). It may seem that females, which are larger by weight and length than males (Anderson, 1985) could be a larger "target" for the infective cryptoniscus stage bopyrid larva. However in laboratory studies, Anderson (1990) found that the highest infection rates occurred before sexual differentiation in grass shrimp larvae followed by postlarvae (due to the frequency of ecdysis as the parasite infects near the time of ecdysis). Cash and Bauer (1993) stated that there was a high correlation between the size of the parasite and the size of the shrimp host leading to the conclusion that grass shrimp must be parasitized at an early life stage with the parasite growing and molting with the host.

The parasitized female and male shrimp in this present study were statistically significantly shorter in length than non-parasitized shrimp (Table 3). It may be that growth in parasitized shrimp is affected by the bopyrid. Walker (1977) determined that one bopyrid parasite ingested enough grass shrimp hemolymph to constitute a significant loss to the host. Anderson (1975, 1977) found that the bopyrid parasite significantly affected *P. pugio* energetics to point of decreased respiration and metabolic activity (which may be the cause of shrimp reproductive sterility). Grass shrimp weights in this present study showed no differences between those shrimp with parasites and those

without. This was similar to the findings of Beck (1980b) who determined that parasite presence did not increase the host shrimp weight.

The statistically highest probability of parasite infection occurred in summer (Table 4) and, for the three summers sampled, parasitized shrimp constituted from 3.5 to 4.7% of the population. In the sites studied by Chaplin-Ebanks and Curran (2007), seasonal bopyrid occurrence was not consistent in their one year study. At one site, peak parasite incidence occurred in spring and at a second site the peak occurred in winter. For this present study, the summer peaks were consistent for all eight sites during the three year period. Summer is the grass shrimp peak reproductive season and, when combined with the parasite's higher probability of occurrence in female grass shrimp (Table 3), this may lead to effects on grass shrimp populations.

In addition to quantifying parasite incidence, the relationship between parasite incidence and human induced changes in those areas surrounding the sampled tidal creeks was investigated. The importance of parasitic infections in grass shrimp is the ability of the parasite to deplete grass shrimp energetics leading to changes in growth, reproduction and survival. Also, changes in grass shrimp behavior as a result of the parasitic infection may lead to altered predator-prey relationships (Bass and Weis 1999). Several researchers have addressed the effects of anthropogenic inputs on grass shrimp population parameters. These findings indicated that agricultural runoff, stormwater runoff from anthropogenic sources and industrial point sources can cause population declines in grass shrimp (Scott et al. 1999; Leight et al. 2005; Fulton et al. 2006). With respect to land use effects on parasite incidence, anthropogenic contaminant effects on

freshwater and marine fish parasites have been reviewed, but any association between parasite prevalence and contaminant level was considered to be weak (Lafferty 1997). A later review by Williams and MacKenzie (2003) found that two fish parasites could be useful as indicators of hydrocarbon pollution in the North Sea. Mixed results in parasite abundance in fish were reported in comparisons of an industrialized estuary to a nonindustrialized estuary (Dzikowski et al. 2003).

Changes in land areas surrounding water bodies have been implicated in changes observed in several other parasite-host relationships. A difference in amphibian parasites was observed in which six parasite species increased in abundance in three host frog species after surrounding forest was converted to pasture (McKenzie 2007). The oyster (*Crassostea virginica*) parasite, *Perkinsus marinus*, showed an increase in infection rates in host oysters from urbanized estuaries when compared to oysters from pristine estuaries (Vernberg et al. 1996). In a Mexican estuary surrounded by agriculture and offshore oil extraction, pink shrimp (Farfantepenaeus duorarum) parasites were studied. Endoparasitic larval helminths were not found to be significantly correlated with contaminant load increases; however, gill symbionts were found to be negatively correlated (Vidal-Martinez et al. 2006). The shrimp Palaemon concinnus was studied in pristine and urbanized African mangrove ecosystems. Shrimp from the urbanized mangrove forest were infected with the bopyrid isopod *Pseudione elongata* at frequencies up to 60%. In contrast, shrimp from the pristine mangrove forest showed infection frequencies significantly lower of between 5 and 10% (Penha-Lopes et al. 2011). In a previous study of bopyrid prevalence in grass shrimp, it was noted that the lowest

prevalence occurred at a site that the authors considered to be the most impacted by anthropogenic inputs (Chaplin-Ebanks and Curran 2007). However, another site exposed to abundant nonpoint source runoff had the highest bopyrid prevalence leading the authors to surmise that other factors need to be considered (Chaplin-Ebanks and Curran 2007).

Searching for relationships in this present study between land use characteristics and bopyrid incidence was not clear even after three years of sampling. Site KWA-01, which had the lowest bopyrid incidence, had the lowest % marsh coverage along with the highest % urban coverage and the highest salinity. This fits well with the probabilities of bopyrid incidence in which higher probabilities were found with higher % marsh coverage and lower salinity. This site had the least suitable grass shrimp habitat (i.e. lowest % marsh cover) and also resulted in the least number of grass shrimp collected over the three year period. Site KWA-93, with the highest bopyrid incidence, had the least % woods coverage and the third highest % urban coverage. This site was only fourth highest in % marsh coverage and its average salinity ranked it as the third lowest. The one creek water quality characteristic in this study that seemed to be limiting to bopyrid incidence was salinity. The high salinity sites, but with more marsh and woods coverage (KWA-02, KWA-96), contained low bopyrid incidence. As stated previously, researchers (Vernberg et al. 1996; Leight et al. 2005; Daugomah 2007) have found a negative correlation between grass shrimp densities and an increase in urban development. The land use characteristics found to limit bopyrid incidence are also

limiting to grass shrimp (definitive host) populations and probably copepod (intermediate host) populations as well.

Conclusions

Long term monitoring of grass shrimp populations is important in developing fundamental understandings of both natural and human caused effects on ecosystems including parasites, disease, climate change and nonpoint source runoff. In addition to population changes caused by natural stressors, is the complexity of differentiating human caused effects in aquatic ecosystems from natural background effects. There is a clear need to develop accurate ecological forecasts using long term data sets. As land use characteristics can change due to increased urbanization, water quality can change due to increases in contaminant loadings and altered salinity regimes. The level of bopyrid prevalence needed to have an effect on grass shrimp population density is not known. While other researchers (Beck 1979, 1980b; Anderson 1990) have suggested that this parasite can affect grass shrimp density, it was not observed in this present study. A significant increase in the prevalence above background levels (1 to 5%) in these eight creeks studied may have some effect. A report of the National Research Council on urban stormwater management recently stated that as land is converted to urban and suburban uses, water flow from storm events results not only in increased runoff of pollutants into surrounding estuaries but also increased volumes of non-saline water itself (NRC 2008). While increased contaminants can effect grass shrimp populations, changes in water quality parameters may affect bopyrid prevalence as well.

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