An Assessment of Native and Non-Native Crayfish Populations in Six Tributaries of the West Branch of the Susquehanna River

Presented to the faculty of Lycoming College in partial fulfillment of the requirements for Departmental Honors in Biology

by
Michelle Herman
Lycoming College
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Approved by:

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Dr. Peter Petokas (Advisor)

____
Dr. Melvin Zimmerman (Advisor)

____
Dr. Michelle Briggs

____
Dr. Jeremy Ramsey

____
Dr. Len Cagle

Honors Project Thesis:

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Submitted by: Michelle Herman

Advisors: Drs. Peter Petokas and Melvin Zimmerman

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INTRODUCTION

Crayfish Classification, Biology, Ecology and Current Conservation Status

Crayfish belong to order Decapoda, a diverse group of crustaceans that also includes the familiar lobsters, crabs and shrimps. Decapods have a worldwide distribution with an estimated 17,635 described species in over 200 families (Reynolds and Souty-Grosset 2012). Defining characteristics of all decapods include: a chitinous exoskeleton, a body with two distinct sections (the cephalothorax and abdomen), five pairs of pereioods (walking legs) attached to the thoracic region and six pairs of pleopods (swimmerets) attached to the abdomen, a head with paired, usually stalked compound eyes, two pairs of sensory antennae and three pairs of mouthparts (mandibles and first and second maxillae, Crocker and Barr 1968), an open circulatory system with a dorsal heart, and gills located internally in the branchial chamber of the cephalothorax (Reynolds and Souty-Grosset 2012, Thorp and Rogers 2011). Crayfish (superfamily: Astacoidea) are distinguished from other decapods by several morphological adaptations. These include the last segment of the elongated and flexible muscular abdomen consisting of a well-developed telson with several uropods, forming the tailfan. This structure is important for the backwards swimming behavior seen in many species to escape danger; it is greatly reduced in other decapods such as the crabs (Reynolds and Souty-Grosset 2012). The first three pairs of pereiopods are also modified with chelae, with the first pair greatly enlarged (Thorp and Rogers 2011). Chelae serve many important functions in crayfish, including capture and manipulation of prey items, defense against predators, inter- and intraspecific interactions and reproductive activities (Reynolds and Souty-Grosset 2012).

Among freshwater crayfish, Cambaridae is the largest and most diverse family, with over 440 described species among 12 genera in North America and eastern Asia (Reynolds and Souty-

Grosset 2012). In North America, cambarid crayfish occur east of the Rocky Mountains, extending north into southern Canada and south through Mexico (Reynolds and Souty-Grosset 2012). An extremely important feature of male cambarid crayfish is the modified first pair of pleopods (gonopods). Used for sperm transfer during copulation, gonopods are conspicuous for their larger size, more heavily calcified appearance, and position. Unlike the other thoracic appendages, gonopods are tight against the ventral surface of the cephalothorax and point towards the head (Rogers and Hill 2008). When male crayfish in this family molt, they alternate between a reproductive form I and a non-reproductive form II, which often have radically different gonopod morphologies. Form I males generally possess larger chelae and rigid, corneous gonopods; form II males exhibit blunter and non-corneous gonopods. Besides their reproductive function, form I gonopods are also critical for proper crayfish species identification (Rogers and Hill 2008). Female crayfish lack gonopods and instead possess an *annulus ventralis* (seminal receptacle), a disc-shaped structure that receives sperm, on the posterior ventral surface of the carapace (Jezerinac *et al.* 1995). Besides the presence of the gonopods or the *annulus ventralis*, crayfish do not exhibit extreme sexual dimorphism. Other, more subtle differences between the sexes include larger body size and longer, heavier chelae in males and broader abdomens in females (Thorp and Rogers 2011, Reynolds and Souty-Grosset 2012).

In population ecology, it is important to understand the life cycle of the study organism. A cambarid crayfish's life cycle involves a surprising array of complex behaviors and chemical signals. For example, adults can distinguish the sex of another crayfish, as well as its reproductive status (*i.e.*, male form I or form II), using a polysaccharide hormone (Reynolds and Souty-Grosset 2012). Most male crayfish in the northeastern United States molt to the reproductive form I by mid to late summer (*i.e.*, July – August, Jezerinac *et al.* 1995). During this time, form I males use their larger and more colorful chelae for signaling, excluding competitors and grasping the female during copulation, which typically occurs during early fall (*i.e.,* August – September, Reynolds and Souty-Grosset 2012, Jezerinac *et al*. 1995). During this ritual, the male usually lies on top and the couple embraces anywhere from a few minutes to several hours (Jezerinac *et al*. 1995, Reynolds and Souty-Grosset 2012). Upon release, the female acquires a whitish sperm plug that protrudes from the *annulus ventralis* (Jezerinac *et al*. 1995). Many females will store the sperm for several months until the following spring. At this time the female seeks cover and releases a viscous substance (glair) which becomes glued to the hairs of the abdominal pleopods, forming a sack (Reynolds and Souty-Grosset 2012). The sperm plug is dissolved and both the egg and sperm are extruded into the sack to be externally fertilized (Jezerinac *et al.* 1995). The eggs remain attached to the female's pleopods and are incubated for several weeks to months (Reynolds and Souty-Grosset 2012).

These brooding females, said to be "*in berry*", will not feed or molt until after the young have dispersed (Reynolds and Souty-Grosset 2012). The young hatch on the female and cling to the pleopods with their chelae. They become free living after two or three molts, but will initially return to the mother for protection from predators; in some species, this is accomplished by the brooding female emitting a pheromone which enables the young to identify and return to her (Reynolds and Souty-Grosset 2012). The juveniles molt several times during the summer, increasing their carapace length by an average of 2-3 mm per molt (Jezerinac *et al*. 1995, Crocker and Barr 1968); in comparison, adult crayfish molt increments are usually about 15% of the total body length and ecdysis occurs only once or twice a year (Reynolds and Souty-Grosset 2012). Most crayfish do not reach sexual maturity until one or two years of age. As adults, crayfish may participate in one or several breeding periods during their lifetime (Reynolds and

Souty-Grosset 2012). In temperate regions, longevity ranges from one to four years, depending on the species and the particular environment (Jezerinac *et al*. 1995, Reynolds and Souty-Grosset 2012).

In the northeastern United States, crayfish occur in a variety of lentic and lotic freshwater habitats. Among stream-dwelling species, important abiotic factors include pH, temperature, dissolved oxygen and substrate type (Reynolds and Souty-Grosset 2012). A stream's pH can affect a crayfish's molting cycle and limit populations because sufficient calcium ions are required for exoskeleton formation and fortification; these ions are less readily available in waters with low pH (Olden *et al*. 2006, Reynolds and Souty-Grosset 2012). Water temperature serves as a cue for various growth and reproduction-related activities (Reynolds and Souty-Grosset 2012). The size (order) of the waterway is probably less important, as lotic crayfish are found in rough, high gradient headwaters to rivers over 40 m wide, although most species seem to prefer moderately wide or small streams (Jezerinac *et al.* 1995, Ortmann 1906). The underlying critical feature of crayfish habitat appears to be shelter (Crocker and Barr 1968). This can include cobbles, gravels, undercut banks, organic debris piles, etc., although a mostly rocky substrate with limited vegetation appears to be optimal for many species (Crocker and Barr 1968, Jezerinac *et al*. 1995, Flinders and Magoulick 2007). Many stream dwelling crayfish also seek out shallower riffle areas, most likely because of higher dissolved oxygen levels and lower densities of large aquatic predators (Flinders and Magoulick 2007, Jezerinac *et al.* 1995). Many crayfish, especially burrowing species, are also adapted to surviving periods of drought because their gills are enclosed within the carapace and therefore do not collapse out of water. This allows the animal to switch from water to air as an oxygen source as long as the air remains moist, as it does in a burrow (Reynolds and Souty-Grosset 2012).

Crayfish consume, and are consumed by, many organisms. Most crayfish species are opportunistic omnivores and scavengers, eating mostly animal matter as juveniles and adopting a more herbivorous diet as adults (Crocker and Barr 1968, Reynolds and Souty-Grosset 2012). These animals will typically emerge at dusk and feed on snails, insects, worms, small fish, tadpoles, dead aquatic animals, fish eggs, algae, vegetation, and other crayfish (Crocker and Barr 1968, Reynolds and Souty-Grosset 2012, Jezerinac *et al*. 1995). Crayfish in turn are important prey items for both aquatic and terrestrial predators. Crocker and Barr (1968) estimate that at least 46 species of fishes, 10 amphibians, 20 reptiles, 38 birds and 6 mammals feed on crayfish. Common fish predators include smallmouth bass (*Micropterus dolomieu*), rock bass (*Ambloplites rupestris*), brook trout (*Salvelinus fontinalis*), fallfish (*Semotilus corporalis*), various sunfish (*Lepomis* spp.), catfish (*Ameiurus* spp.) and creek chubs (*Semotilus* spp.). Reptiles such as the queen snake (*Regina septemvittata*) and the common snapping turtle (*Chelydra serpentina*) also consume crayfish, while examples of important amphibian predators include the aquatic salamanders known as mudpuppies (*Necturus*) and the eastern hellbender (*Cryptobranchus alleganiensis*). Examples of common bird predators include green heron (*Butorides virescens*), great blue heron (*Ardea herodias*), belted kingfisher (*Megaceryle alcyon*) and barred owl (*Strix varia*). Raccoons (*Procyon lotor*), muskrat (*Ondatra zibethicus*), northern river otter (*Lontra canadensis*) and American mink (*Neovison vison*) are important mammalian predators of crayfish (Crocker and Barr 1968, Jezerinac *et al*. 1995).

The hellbender is one of the primary crayfish consumers in northcentral Pennsylvania streams. Crayfish are a critical food item for this animal, accounting for the vast majority of its diet; studies have indicated a 59-87.5% crayfish-based diet via stomach and intestinal content analysis, but this range is probably conservative (Nickerson and Mays 1973). Due to this salamander's relatively large size (up to 74 cm in total length), longevity (about 30 years in captivity) and entirely aquatic lifestyle (Nickerson and Mays 1973), arguably no other predator spends as much time in the water consuming as many crayfish as the hellbender does over its lifetime. This unique giant salamander is currently experiencing sharp declines throughout its geographical range for reasons not yet fully understood (Wheeler *et al.* 2003, Petokas *et al.* 2012), but the species clearly depends on healthy crayfish populations for survival.

Crayfish perform many ecological functions in stream food webs and are unique among most other stream macroinvertebrates. First, crayfish are one of the largest macroinvertebrates in a stream and often account for the greatest biomass among benthic invertebrates (Flinders and Magoulick 2007). They are also among the most long-lived macroinvertebrates, as some individuals reach three or four years of age in temperate regions (Reynolds and Souty-Grosset 2012). Relative to other stream macroinvertebrates, the crayfish body form is unspecialized and highly versatile, allowing for much greater activity (*i.e.,* pereiopods for locomotion upon a substrate; a muscular abdomen with pleopods and a tailfan for swimming) and seizing and manipulating a wider range of food items (*i.e.,* chelae; three distinct mouthparts) (Crocker and Barr 1968, Jezerinac *et al.* 1995, Thorp and Rogers 2011, Reynolds and Souty-Grosset 2012). Studies appear to support this idea of versatility; for example, Flinders and Magoulick (2007) comment that crayfish consume more animal matter than any other benthic macroinvertebrate while also processing as much coarse particulate organic matter (CPOM) as a specialized shredder; this activity converts CPOM to a form that filtering and gathering organisms can use.

Taking into account these characteristics, it is not surprising that some North American species are considered to occupy trophic positions intermediate to those of fish and other typical macroinvertebrates (Taylor and Soucek 2010). Crayfish subsequently exert a great amount of control over their environment. For example, when crayfish feed on CPOM or primary producers (*e.g.,* algae), the diatom and aquatic insect community may subsequently change in terms of composition and abundance (Flinders and Magoulick 2007). As secondary consumers preying on other macroinvertebrates, crayfish in turn may reduce their populations and promote primary production (Flinders and Magoulick 2007). Fish populations can also be impacted by crayfish predation on eggs or fry or by competition for resources (Crocker and Barr 1968, Reynolds and Souty-Grosset 2012). Crayfish can therefore function as keystone species and promote biodiversity by preying on commoner species and keeping these populations in check (Reynolds and Souty-Grosset 2012).

In spite of their ecological importance, freshwater crayfish are unfortunately a very threatened group of organisms. Additional research and conservation efforts are especially needed in North America, where about 80% of the world's crayfish diversity is contained. This diversity peaks in the southeastern United States (Thorp and Rogers 2011, Reynolds and Souty-Grosset 2012) with Mississippi as the "hotspot" boasting 63 different crayfish species among 6 genera (Reynolds and Souty-Grosset 2012). This crayfish diversity is mainly threatened by habitat loss, pollution, overexploitation and introduction of non-indigenous species (Taylor *et al.* 2007). These threats may prove especially detrimental to crayfish compared to other aquatic taxa due to the limited natural range of these animals. As Taylor *et al.* (2007) note, crayfish are highly endemic organisms, with approximately 43% of crayfish species in the United States distributed entirely within one state's political boundaries. As previously discussed, crayfish also rely on chemical signaling for many activities related to communication and reproduction, and water pollution in the form of herbicides has been shown to interfere with antagonistic behaviors in at least one species (Cook and Moore 2008).

A prominent issue in recent times is non-native crayfish invaders, which threaten more than half of all declining North American crayfish through competition, predation and hybridization (Olden *et al.* 2006). This problem is compounded since the spread of invasive crayfish species is facilitated by aquaculture, aquarium and pond trades, the biological supply trade and the live bait trade. Many non-native species released into new waterways from these sources tend to spread rapidly and become well established, potentially displacing native crayfishes (Olden *et al.* 2006).

Historical Crayfish Population Studies in the Susquehanna River Basin

Previously published scientific literature on crayfish populations in the Susquehanna River Basin is discontinuous and neglects the West Branch sub-basin in northcentral Pennsylvania. Much of what is known about crayfish in the West Branch is still based on surveys conducted in the region over a century ago by Arnold Edward Ortmann. An early 20th century malacologist and astacologist, Ortmann (1906) published a landmark statewide study of Pennsylvania crayfish based on three years of field collections and archival review. Working as then-Curator of Invertebrate Zoology for the Carnegie Museum, most of Ortmann's (1906) field collections were based around Pittsburgh, Pennsylvania and the extreme western part of the state, with less sampling in more eastern waterways near Harrisburg and Philadelphia. Ortmann (1906) largely ignored the central, northern and northeastern portions of the state, citing a lack of crayfish species diversity that apparently made surveying waterways in those regions pointless. The only West Branch waterways surveyed were the headwaters of the West Branch and several of its westernmost tributaries in Cameron, Clearfield, Cambria, and Indiana Counties (Ortmann 1906). Ortmann (1906) sampled over 100 sites from waters in thirty-nine of the state's sixtyseven counties, resulting in well over 1800 specimens of seven different species: *Cambarus limosus* (now *Orconectes limosus*), *C. propinquus* (*O. propinquus*) *C. obscurus* (*O. obscurus*), *C. bartonii* (with one subspecies, *C. b. robustus*, now *C. robustus), C. carolinus*, *C. monongalensis*, and *C. diogenes*. *Cambarus bartonii* (common or Appalachian brook crayfish) was the only crayfish species encountered by Ortmann (1906) in the West Branch sub-basin. Despite his simplistic methods of capture (mostly handnetting and excavation of crayfish burrows) and the disproportionate attention given to the extreme western and eastern drainages of the Commonwealth, Ortmann (1906) is credited for setting the precedent for future astacological studies in both Pennsylvania and all of North America (Lieb *et al*. 2011a). Unfortunately, coordinated research that assesses the statewide distribution of crayfishes in Pennsylvania has since been lacking. As a result, many contemporary crayfish studies still acknowledge Ortmann's (1906) work and value it as one of few historical references available for comparing past and present crayfish species distribution (e.g. Lieb *et al*. 2011a, Lieb *et al.* 2011b, Killian *et al*. 2010, Jezerinac *et al.*1995).

While it has been argued that no other astacological studies in Pennsylvania or North America have since matched Ortmann's (1906) in terms of importance and scale (Lieb *et al.* 2011a), several contemporary crayfish studies within the Susquehanna River basin provide much needed updates to Ortmann's (1906) work. Mangan and Stocker (2011) sampled several sites along the North Branch as well as the main stem of the Susquehanna River to measure mercury contamination in crayfish abdominal tissue. The authors used baited wire traps at 11 sampling sites along 410 km of the Susquehanna River in 2008. The non-native rusty crayfish (*O. rusticus*) was the predominant species at five sites; these included the three most downriver sites located in the main stem just below the confluence of the West and North branches and two sites in the upper portion of the North Branch. The non-native Allegheny crayfish (*O. obscurus*) was the predominant crayfish species at the other sampling sites (Mangan and Stocker 2011).

Recent crayfish population studies conducted in other portions of the Susquehanna River Basin also suggest crayfish species are undergoing changes in distribution and diversity. Kuhlmann and Hazelton (2007) sampled the upper Susquehanna River watershed near Oneonta, New York from 1999 to 2005 to determine the distribution of crayfish species. Sampling included the River, its headwaters at Otsego Lake, and many smaller tributaries in the region. Thirty-eight sampling locations yielded close to 4000 crayfish, consisting of four species: *O. rusticus*, *O. propinquus*, *O. obscurus*, and *C. bartonii*. *O. rusticus* was the most abundant and widely-distributed species, occurring throughout most of the upper Susquehanna River, the upper Unadilla River, and the lower reaches of most other tributaries. The authors commented that the upper Susquehanna River crayfish community appeared to have changed considerably within the last century. One example was the study's failure to yield two crayfish species historically abundant in the area (*O. limosus* and *O. immunis),* suggesting that populations now occur at much lower densities or have been extirpated. *O. obscurus* and *O. rusticus* were also considered recent additions to the region, not known to occur in the upper Susquehanna River watershed prior to 1991 (Kuhlmann and Hazelton 2007). *C. bartonii*, historically abundant in both larger streams and headwaters, was restricted to a few sites in the upper reaches of the streams sampled (Kuhlmann and Hazelton 2007).

Killian *et al*. (2010) provide a summary of astacological studies in the state of Maryland since the late 19th century while also including an update on the distribution of the state's fourteen species, nine of which are native and are potentially threatened by invasive crayfishes. From 2006 to 2007, crayfish data were collected from 446 sites in streams, rivers, seepage wetlands, and floodplains. *C. bartonii*'s status was considered stable. The invasive *O. rusticus*, first discovered in the Monocacy and Susquehanna rivers in 2007, was predicted to spread throughout Maryland. Killian *et al.* (2010) contend that this invasion will be highly detrimental to several of the state's native species, including *O. virilis*, *O. obscurus* and *O. limosus* (all of which also occur in Pennsylvania).

Hypotheses and Objectives of Current Study

When Ortmann (1906) surveyed western portions of the West Branch of the Susquehanna River and its tributaries, only one crayfish species (*C. bartonii*) was identified, and this was presumed to be the only species in the entire West Branch drainage. Since then, there has been no published research attempting to "resurvey" northcentral Pennsylvania's crayfish populations. In the century that has passed since Ortmann's (1906) work, crayfish populations are not likely to have remained static in terms of diversity and distribution. This is especially true considering the recent documentation of *O. obscurus* and *O. rusticus* in other portions of the Susquehanna River drainage (*i.e.,* Kuhlmann and Hazelton 2007, Mangan and Stocker 2011) and extreme southeastern Pennsylvania (Leib *et al.* 2011a). In preparation for the current study, it was hypothesized that *C. bartonii* was no longer the sole crayfish species in northcentral Pennsylvania. To determine the current crayfish species composition of northcentral Pennsylvania, crayfish populations of several West Branch tributaries were surveyed by selecting sample sites ranging from the mouth to the headwaters of each waterway. In addition to determining species occurrence, the current study also considered the demographics of the region's crayfish populations, something that Ortmann (1906) never examined. In particular, sampling methods provided quantitative data allowing relative crayfish densities and sex ratios at each site to be calculated. This information can gauge the health of the region's crayfish populations. For example, low densities or an overabundance of one sex could have negative implications for future reproduction in a population.

New survey work assessing the region's crayfish populations is especially critical at a time when non-native crayfish species introductions and invasions are wreaking havoc on native populations throughout North America (e.g. Lieb *et al.* 2011a, Swecker *et al.* 2010, Taylor *et al.* 2007, Olden *et al.* 2006). Invasions of non-native crayfish species have been documented across Pennsylvania. For example, Lieb *et al.* (2011a) recently surveyed waters in southeastern Pennsylvania and found that five of the eight crayfish species caught were non-natives not present in the region at the time of Ortmann's (1906) study. This research marks the first published record of *P. clarkii* (red swamp crayfish) in the state and the first published records of *O. rusticus*, *O. virilis* and *O. obscurus* occurring in southeastern Pennsylvania The authors concluded that native crayfish species in southeastern Pennsylvania such as *O. limosus* are in significant decline (Lieb *et al.* 2011a). Another study by Lieb *et al.* (2011b) expressed concern that introduced crayfishes besides *O. rusticus* (*i.e., P. acutus*, *C. robustus*, *O. obscurus*, and *O. virilis*) remained unregulated in Pennsylvania and could be purchased or collected elsewhere and released legally into the state's waters. Based on the changes documented both across the state and within the Susquehanna River Basin (e.g. Mangan and Stocker 2011, Kuhlmann and Hazelton 2007, Killian *et al*. 2010), I hypothesized that native crayfishes in northcentral Pennsylvania are likely undergoing, or are at risk of undergoing, declines as non-native species expand their range and possibly exclude natives. Within the proposed study area it is common knowledge that at least one non-native crayfish species is present (*i.e., Orconectes rusticus*) in Loyalsock Creek, Lycoming County. As previously mentioned this species has also been documented in the main stem of the Susquehanna River just below its confluence with the West Branch and also appears throughout the North Branch of the Susquehanna River (Mangan and Stocker 2011, Kuhlmann and Hazelton 2007). The exact extent of this species' penetration into West Branch watersheds, however, is currently unknown aside from Loyalsock Creek. In order to provide a clearer picture regarding the status of *O. rusticus* in northcentral Pennsylvania, the Loyalsock Creek and the major West Branch tributaries immediately up-river (*i.e.,* Lycoming Creek) and downriver (*i.e.,* Muncy Creek) were included in the surveys for crayfish species occurrence.

In addition to surveying West Branch tributaries for crayfish species occurrence, another objective of the current study was to examine the morphometry of the region's crayfishes. If non-native species have indeed penetrated a significant portion of West Branch watersheds, and these non-native crayfishes are replacing natives, then exploring differences in body size may help predict future consequences of this takeover. Select body measurements will be taken and compared across factors such as species, sex, and tributary to determine whether morphometry differs significantly among the region's crayfish.

One additional reason to study northcentral Pennsylvania crayfish populations concerns the ecological relationship between crayfish and the eastern hellbender (*Cryptobranchus alleganiensis*). As previously mentioned, the hellbender is a unique giant salamander that feeds primarily on crayfish. Hellbender populations still persist in select West Branch tributaries that meet the species' habitat requirements of clean, cool, swift flowing streams with an abundance of large, flat cover rocks. However, this animal's continued existence is potentially threatened as numbers continue to decline throughout its geographic range (e.g. Wheeler *et al.* 2003). Within the proposed study area, sharp population declines have been documented in Loyalsock Creek, Lycoming County (Petokas *et al.* 2012). The link between crayfish population composition and structure and the health of the eastern hellbender has not been previously investigated. The information obtained from surveying northcentral crayfish populations in tributaries where hellbenders also occur potentially provides more insight into the hellbender's decline. Perhaps the composition and health of this species' food base (*i.e.,* proportion of native versus non-native species or crayfish density) are contributing factors that threaten the survival of the hellbender. This study will hopefully provide pertinent baseline information on the region's crayfishes that will aid in research seeking to identify the cause of the hellbender's decline.

METHODS

Description of the West Branch and Six Watersheds

The West Branch sub-basin of the Susquehanna River has a drainage area of approximately 4.5 million acres (PADEP 2009, WBSRTF 2005). It spans several northcentral counties in the state, including Cambria, Clearfield, Elk, Cameron, Potter, Clinton, Centre, Tioga, Sullivan, Lycoming, Union, and Montour Counties (West Branch 2005). The watershed is characterized as mostly forested land with minimal urban development, with approximately 1.4 million acres of State Forest Land, 250,000 acres of State Game Lands, and 29,000 acres of State Park Land. Expressed as percentages, land use in the watershed breaks down to 83% forested, 10% agricultural, and 7% developed and disturbed (PADEP 2009). Urban centers are generally few and far between in the sub-basin and include cities such as Williamsport (Lycoming County), State College (Centre County), Lock Haven (Clinton County), and Clearfield (Clearfield County). The sub-basin's total population was last estimated at 580,000 people (WBSRTF 2005). The West Branch of the Susquehanna River originates in West Carroll

Township, eastern Cambria County, and flows more or less east past scattered urban areas such as Renovo (Clinton County), Lock Haven (Clinton County), Williamsport (Lycoming County) and Muncy (Lycoming County). At Muncy, the West Branch makes a sharp bend to the south and reaches its confluence with the North Branch of the Susquehanna River at Northumberland in Northumberland County (PADEP 2009).

Historically, the sub-basin was deep-mined for bituminous coal beginning in the late 1800s, with the industry peaking around the mid 1900s and declining in subsequent decades. Surface strip mines largely replaced deep mines by the 1970s, although both types of coal mining operations currently take place within the watershed (PADEP 2009). This legacy of mining has caused many water quality problems within the West Branch sub-basin in the form of abandoned mine drainage (AMD). AMD currently surpasses agriculture as the leading source of pollution to impaired West Branch waterways, especially in the western portion of the drainage near the Cambria County headwaters (WBSRTF 2005). Approximately 1,205 stream miles (including both the main stem and tributaries) are considered degraded by AMD, which accounts for 66% of the total AMD-impaired mileage in the entire Susquehanna River Basin (PADEP 2009).

Kettle Creek was the westernmost and most up-river tributary in the study area, and has perhaps the most isolated and least developed watershed of all the streams surveyed. The Kettle Creek watershed contains 690 stream kilometers (Pennsylvania State University 2001) and drains about 637 km^2 in Potter, Tioga, and Clinton counties (PA DEP 2001). Most (92%) of the watershed consists of Northern Hardwood forest managed by the state (*i.e.*, Elk, Sproul, Susquehannock and Tioga state forests and Kettle Creek and Ole Bull state parks), with few agricultural areas. Human development within the stream valley is minimal, consisting primarily of hunting camps and small villages with few year-round residents (Pennsylvania State

University 2001). Kettle Creek originates in a remote forested area in Elk Township, western Tioga County, near the Potter-Tioga County line (Pennsylvania State University 2001). The headwaters flow southwest through the Susquehannock State Forest in Potter County, eventually passing through relatively pristine sections of forest that include the F. H. Dutlinger Natural Area and Hammersley Wild Area. The mouth of Kettle Creek is located at the village of Westport (Clinton County), about 10.1 km upstream from Renovo. Kettle Creek is not a free flowing tributary, as three state park impoundments (Ole Bull Dam, Kettle Creek Lake and Kettle Creek Recreation Dam) are present on the stream (Pennsylvania State University 2001). The Ole Bull Dam and Kettle Creek Dam are relatively small in height (less than 5 m) and are used primarily for recreation purposes. The Alvin R. Bush Dam forms Kettle Creek Lake in Kettle Creek State Park and is about 50 m in height. It was constructed in 1961 to provide flood control and recreation opportunities in the park (Pennsylvania State University 2001). About five kilometers below the Bush Dam, Kettle Creek becomes impacted by mine drainage, but the upper portion of the watershed above the Bush Dam to the headwaters remains fairly pristine (Pennsylvania State University 2001). Kettle Creek is considered one of the most intensively stocked streams in the Commonwealth, and is managed chiefly as a catchable trout fishery (Pennsylvania State University 2001).

Pine Creek is the second largest tributary to the West Branch of the Susquehanna River (Schwarz 2005) and had the largest watershed in this study at about 2541 km^2 (PA DEP 2001). The Pine Creek watershed includes 17 major sub-basins (*e.g.*, Marsh Creek, Babb Creek, Cedar Run, Slate Run and Little Pine Creek) (Schwarz 2005). The two largest tributaries are Marsh Creek and Babb Creek, together accounting for almost a quarter of the total drainage area of the Pine Creek watershed (Schwarz 2005). Despite Pine Creek's relatively large size, nearly three quarters of the streams in its drainage area are smaller first and second order streams (Schwarz 2005). A significant portion of the Pine Creek watershed is also surrounded by the Tiadaghton State Forest, and this along with the Pine Creek Gorge (a steeply carved section of the stream valley), various state park facilities (e.g. Colton Point and Leonard Harrison State Parks) and a Rails-to-Trails bike path along the creek make the watershed a popular recreational destination (Schwarz 2005). Pine Creek originates in Ulysses Township, Potter County (Schwarz 2005) and is approximately 135.8 km long, passing through Potter, Tioga and Lycoming Counties and flowing more or less southeast towards the West Branch near Jersey Shore. The headwaters first flow southeast to Galeton, where the creek bends to the east for roughly 20.9 km to Ansonia (Tioga County); Marsh Creek, a major tributary, also joins Pine Creek at this point. Pine Creek then bend sharply to the south and passes through the Pine Creek Gorge. Pine Creek continues to flow more or less south, with its other major tributary, Babb Creek, draining into the waterway at the village of Blackwell near the Tioga-Lycoming County line. In Lycoming County, Pine Creek parallels the western county line before forming the boundary between Lycoming and Clinton counties near Jersey Shore. The creek empties into the West Branch of the Susquehanna just south of the Jersey Shore borough. One notable impoundment along Pine Creek is the Galeton / Centertown Lake, which is a 12 acre stop log construction impoundment of Pine Creek itself. While mainly used for flood control, it also provides recreational purposes (Schwarz 2005).

Little Pine Creek is a tributary of Pine Creek located primarily in Lycoming County. It drains an area of 466 km^2 and is formed by the confluence of Blockhouse Creek and Texas Creek in northern Lycoming County. Little Pine Creek flows for about 24.5 km in a mostly southwest direction and is dammed about 6.4 km upstream from its confluence with Pine Creek at Waterville. The 94 acre Little Pine Lake was constructed in 1949 and serves as a flood control and recreational reservoir in Little Pine State Park (Schwarz 2005).

The Larry's Creek watershed was the smallest among the six included in this study, encompassing about 231 km^2 (PA DEP 2001). It is entirely contained within Lycoming County. Larry's Creek originates near Steam Valley in Cogan House Township and flows primarily south for about 34.6 km before draining into the West Branch just east of Jersey Shore, Pennsylvania. State Routes 287 and 184 parallel the Creek near its mouth and headwaters, respectively, while the middle portion of Larry's Creek passes through State Game Lands No. 114 where no public roads are present.

The Lycoming Creek watershed drains about 704 km^2 (PA DEP 2001). Lycoming Creek originates close to where Bradford, Tioga and Lycoming Counties touch, and flows for approximately 56.8 km south, passing through a parcel of Tiadaghton State Forest and State Game Lands No. 133 before draining into the West Branch near Williamsport, Pennsylvania. State Routes 15 and 14 parallel much of the stream's length.

The Loyalsock Creek watershed is located primarily in Lycoming and Sullivan Counties and drains 1279 km^2 (PA DEP 2001), making it the second-largest watershed in the study area. Loyalsock Creek is about 100.5 km long and originates in western Wyoming County near the Sullivan-Wyoming County line. It flows west across Sullivan County until the Little Loyalsock Creek, a major tributary, joins it at the village of Forksville. Here the Loyalsock bends to the southwest and flows into Lycoming County, eventually reaching its confluence with the West Branch at Montoursville, Pennsylvania.

Muncy Creek was the most downriver West Branch tributary surveyed. The watershed drains 559 km^2 (PA DEP) and is located in Sullivan and Lycoming Counties. Muncy Creek is about 56.5 km long and its source is located in a remote corner of Davidson Township, Sullivan County. It flows more or less southwest, with U.S. Highway 220 paralleling the stream for much of its length. In Lycoming County Muncy Creek passes through several small boroughs such as Picture Rocks and Hughesville before reaching its confluence with the West Branch at Muncy.

Field Sampling Procedures

A total of six tributaries of the West Branch of the Susquehanna River in northcentral Pennsylvania were surveyed for crayfish species occurrence (Fig. 1). The study area consisted of five counties (Tioga, Potter, Clinton, Lycoming and Sullivan) and included, moving down river along the West Branch: Kettle Creek, Pine Creek (plus Little Pine Creek, a tributary of Pine Creek), Larry's Creek, Lycoming Creek, Loyalsock Creek, and Muncy Creek. Crayfish were collected from a total of twenty-eight sites among the seven streams, with sampling occurring from 6 June to 14 August 2012 (Table 1).

In order to determine crayfish species occurrence, each stream was sampled at various points starting near the mouth and working upstream toward the headwaters, although sampling was not always done in a linear fashion up or down a tributary. Specific sampling locations were mostly selected based on access and habitat rather than sampling regular intervals along each stream. This was because carrying the necessary sampling equipment down to a desired reach was not always practical, or a reach was surrounded by private land. Habitat was another factor because crayfish need adequate cover to survive in streams and thus tend to occupy places that have plenty of shelter (Crocker and Barr 1968, Reynolds and Souty-Grosset 2012). If a locality did not appear to have ideal substrates, in the interest of time and effort it was presumed to not have adequate numbers of crayfish for sampling purposes.

To collect data on crayfish population demographics (*i.e.*, density and sex ratios), a quantitative yet fairly rapid sampling method was needed. While quadrat sampling is considered to be a very accurate method for measuring densities (Dorn *et al.* 2005), this method is known to be biased towards collecting smaller individuals in a population (Rabeni 1997, Price and Welch 2009) and is perhaps more time consuming. Mangan *et al.* (2009) successfully utilized traps in the Susquehanna River to capture crayfish, but the shallower and swifter waters of these West Branch tributaries make trapping problematic. Trapping is also known to be biased towards certain species, larger individuals, and males (e.g. Price and Welch 2009, DiStefano *et al.* 2003, Kutka *et al.* 1992, Dorn *et al.* 2005). Another criticism of trapping is that it measures density over an unknown area and thus really gauges relative abundance and animal activity levels (Dorn *et al.* 2005). Electrofishing can be made into a semi-quantitative method with time constraints, and is noted for its unbiased results and high success rate *(i.e.,* high number of individuals caught) (Price and Welch 2009, Rabeni 1997). However, eletrofishing was not considered a viable method in this study due to the need for proper training and equipment and the inconvenience of transporting the necessary equipment to and from samples sites.

Considering the type of waterways to be surveyed and the biases and limitations of quadrants and trapping, semi-quantitative seining was the primary method selected for this study. In a review of crayfish population sampling methods, Price and Welch (2009) commented that seine netting was a good choice for documenting species diversity and collecting larger individuals compared to other semi-quantitative techniques. Kuhlmann and Hazelton (2007) also successfully utilized a similar semi-quantitative kicknet protocol to sample crayfish populations in the upper Susquehanna River. This method allows for fairly rapid sampling (maximizing the potential number of sites surveyed) while also allowing one to record the number of individuals in a known sample area. Most sites were ideal for seining, as they usually consisted of relatively shallow water (averaging less than 40 cm deep at the seine sampling locations) in a reach consisting of a glide, run and/or riffle area and a bottom with a mix of easily disturbed smaller particle sizes (*i.e.,* gravel, cobble and/or small boulder).

When seining (Fig. 1a), one person stood at either end of a 10' x 4' seine with 3/8" sized mesh and formed a semicircular pocket facing upstream. At least one person stood immediately upstream of the seine and vigorously kicked and agitated the substrate, lifting rocks and scouring the area so that any organisms would be swept by the current into the net. For each attempt, the sample area was estimated by measuring net width (the distance in centimeters between the poles) with a retractable measuring tape. To assess potential habitat preferences, water depth, distance to shore, water velocity and habitat type were recorded. Water depth (cm) was taken at the center of the pocket using a standard measuring staff. Distance to shore was classified as "near shore" if the seine sample was 7 m or less from the closest bank and "far shore" if greater than 7 m. In the interest of time and equipment, water velocity was a purely qualitative measurement and classified by the data recorder as slow, medium or fast. Water velocity was considered slow if the surface of the water was completely smooth and only a weak current existed to maintain the net pocket. Medium water velocity consisted of a strong enough current to fully maintain the net pocket, and the surface of the water may or may not have been disturbed as in a riffle or run area. Water velocity was considered fast if apparent physical effort by the persons holding the poles was required to keep the seine in place and /or the water surface was highly agitated, as in a riffle area or rapids. Habitat type was recorded as one of six possible categories based on the predominant substrate within the sampling area: sand and gravel (SG) (pieces <5 cm long); tight cobble (TC) with little to no interstitial spaces (embedded) or loose cobble (LC) with interstitial spaces (cobble = 5-20 cm); small boulder (SB) (20-50 cm); medium boulder (MB) (50-100 cm); and large boulder (LB) (>100 cm). The number of crayfish of each sex caught in each seine sample was recorded with all previously described measurements, as well as the species and number of any other macroinvertebrates or fish caught in the net. The mid-channel width of the reach and length of the reach sampled were recorded in yards to estimate the total area sampled. Where seining was not feasible, such as in a reach with deep, slow water, qualitative sampling was done with a diving mask and snorkel (Fig. 1b). The stream bottom was methodically searched for crayfish by several researchers and animals were captured by hand.

When sufficient numbers of large, mature crayfish were captured (at least 15 – 20 of each species and/or sex), all individuals were brought on shore and sorted into separate containers by sex and species. Species identification was according to the regional key by Rogers and Hill (2008) and site totals for the number of males and females of each species were recorded. For morphometric analysis, a sample size of ten of the largest males and ten of the largest females of each species were transported back to the Lycoming College Biology Department in Williamsport, Pennsylvania. The designation of "largest" was purely visual and based on the judgment of the person selecting the crayfish. Only the largest individuals were kept to facilitate the body measurement process. When numbers were adequate, five additional male crayfish of each species (form I when possible) were also brought back from the field for later preservation as voucher specimens. While crayfish sorting was occurring, other researchers recorded site water chemistry (pH, specific conductivity (µs), water temperature (°C), turbidity (NTU) and dissolved oxygen (mg/l-)). GPS coordinates, elevation (ft), reach type (e.g. run or riffle vs. pool), bottom type (e.g. cobble), riparian zone type for both banks (e.g. bare vs. tree covered) were among other ecological data collected.

Field Sampling Procedures: Tables and Figures

Figure 1: Crayfish Sampling Locations and Species Distribution in Six Watersheds of the West Branch of the Susquehanna River in Northcentral Pennsylvania, 2012.

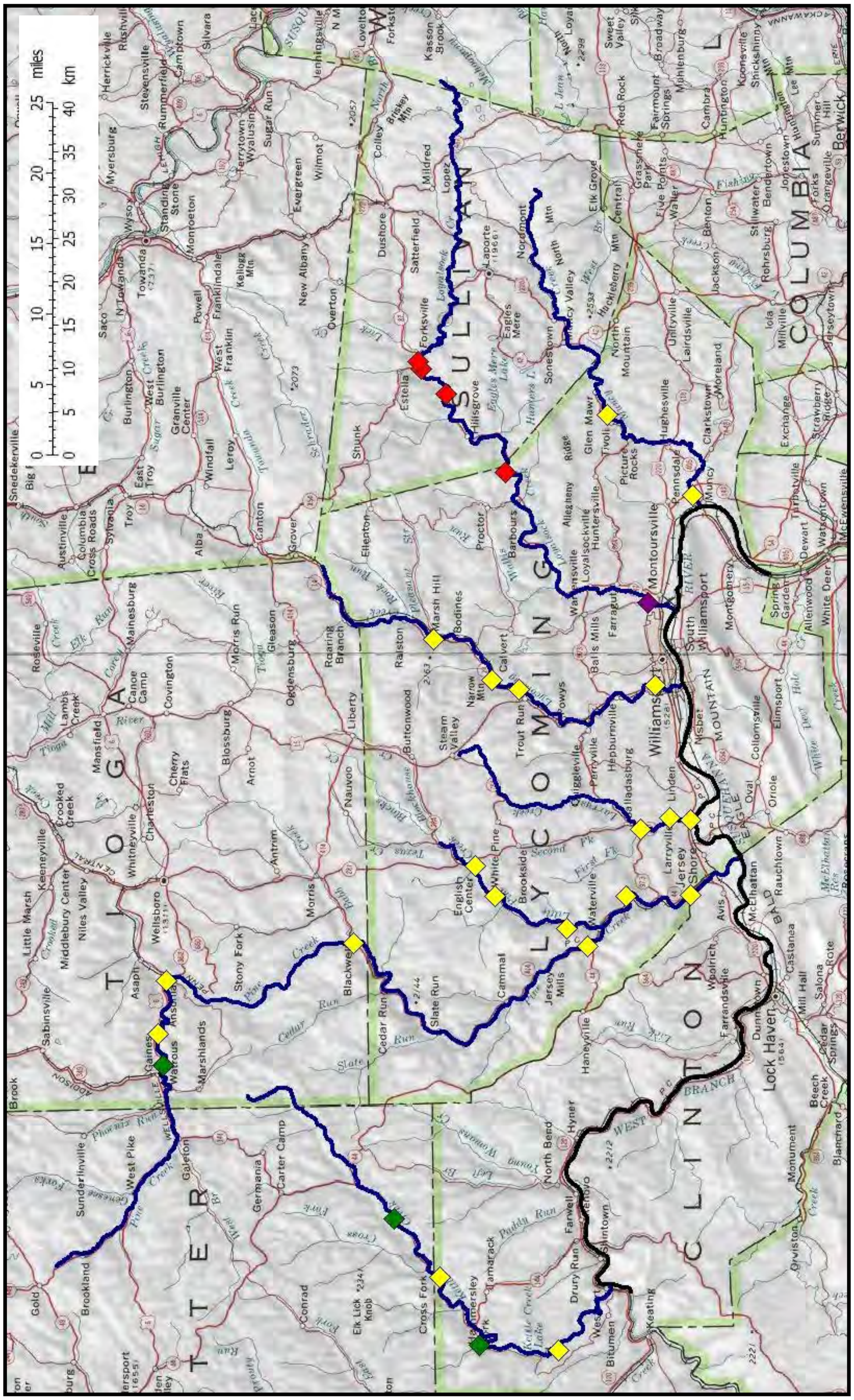


Fig. 1: Topographic map displaying the 28 sampling locations and crayfish species collected from six watersheds of the West Branch of the Susquehanna River in northcentral Pennsylvania. **Key:** Black Line = West Branch; Blue Line = tributary; Yellow Diamond = *Orconectes obscurus*; Green Diamond = *O. obscurus + Cambarus bartonii*; Red Diamond = *O. rusticus*; Purple Diamond = probable *O. rusticus x obscurus*.

Table 1: Crayfish Sampling Locations and Methods.

Waterway	Site	Location	Stream Kilometer	Sample Method(s)	Date Sampled
Kettle Creek	KET01	41.21089°N 77.55505°W	12.6	S-Q Seine	08/14/12
	KET02	41.2583°N 77.55119°W	22.7	S-Q Seine	06/19/12
	KET03	41.28241°N 77.49711°W	36.8	Mask and Snorkel	08/12/12
	KET04	41.51387°N 77.75170°W	46.3	S-Q Seine; Mask and Snorkel	08/10/12
Pine Creek	PC01	41.21920°N 77.32082°W	7.7	S-Q Seine	06/26/12
	PC02	41.28348°N 77.32140°W	16.9	S-Q Seine	06/26/12
	PC03	41.32170°N 77.38647°W	24.8	S-Q Seine	06/26/12
	PC04	41.55598°N 77.38247°W	65.4	S-Q Seine	08/02/12
	PC05	41.74396°N 77.43317°W	92.4	S-Q Seine	07//24/12
	PC06	41.75269°N 77.50539°W	100.1	S-Q Seine	07/24/12
	PC07	41.74756°N 77.54597°W	104.1	Mask and Snorkel	07/24/12
Little Pine Creek (Tributary of Pine Creek)	LPC01	41.20564°N 77.21863°W	4.6	Non S-Q Seine	06/12/12
	LPC02	41.41434°N 77.32018°W	15	Non S-Q Seine	06/13/12
	LPC03	41.44093°N 77.27992°W	19.4	Non S-Q Seine	06/13/12
Larry's Creek	LAR01	41.13124°N 77.1319°W	0.24	S-Q Seine	08/02/12
	LAR02	41.23961°N 77.21661°W	2.9	S-Q Seine	06/15/12
	LAR03	41.26860°N 77.23208°W	7	S-Q Seine	06/15/12
Lycoming Creek	LYC01	41.15265°N 77.2447°W	3.5	Non S-Q Seine	06/06/12
	LYC02	41.23496°N 77.2782°W	24.7	Non S-Q Seine	06/07/12 06/08/12
	LYC03	41.25089°N 77.2019°W	28.4	Non S-Q Seine	06/11/12
	LYC04	41.28584°N 76.58805°W	38.2	Non S-Q Seine	06/11/12
Loyalsock Creek	LOY01	41.26337°N 76.92579°W	3.3	S-Q Seine	08/08/12
	LOY02	41.39731°N 76.75936°W	35.4	Non S-Q Seine	06/14/12
	LOY03	41.46384°N 76.65405°W	52.1	Non S-Q Seine	06/14/12
	LOY04	41.48937°N 76.61974°W	57.5	S-Q Seine; Mask and Snorkel	06/27/12
	LOY05	41.49310°N 76.60447°W	58.3	Mask and Snorkel	08/01/12
Muncy Creek	MUN01	41.21736°N 77.49711°W	1.5	S-Q Seine	08/01/12
	MUN02	41.30243°N 76.68106°W	21.5	Mask and Snorkel	07/02/12

Table 1: Twenty-eight locations among seven streams were sampled for crayfish in northcentral Pennsylvania. The primary capture method was a 10' x 4' seine with 3/8" sized mesh used both non-semi-quantitatively (Non S-Q Seine) and semi-quantitatively (S-Q Seine). Where seining was not feasible, a diving mask and snorkel were used. Stream kilometer is measured beginning at the mouth of the waterway.

Figure 1a: Semi-Quantitative Crayfish Sampling via Seining.

Figure 1b: Non-Quantitative Crayfish Sampling via Mask and Snorkel.

Figures 1a and 1b: The primary method for collecting crayfish was a 10’ x 4’ seine with 3/8” sized mesh used semi-quantitatively as pictured in Fig. 1a. Where seining was not feasible, a mask and snorkel were used to survey the stream bottom as pictured in Fig. 1b.

Lab Sampling Procedures

Most crayfish specimens were processed in the lab within 24 hours of their collection in the field. Six body measurements were taken for each crayfish: blotted wet mass (BWM) (g), total body length (TBL) (mm), carapace length (CL) (mm), areola length (AL) (mm), areola width (AW) (mm) and palm width (PW) (mm). To keep all individual data separate, crayfish were placed in separate glass dishes filled with tap water during processing. Specimens were then designated as male #1-10 and female #1-10 for the appropriate site. BWM was measured as a total (all ten males or females of one species) and also individually. Specimens were blotted several times with a paper towel to remove excess water before weighing in an A&D ER-182A analytical balance. The remaining five measurements were taken with a needle point dial caliper accurate to 0.05 mm and are shown in Figure 3. TBL was measured as the length from the tip of the rostrum to the posterior edge of the telson. CL was defined as the length from the tip of the rostrum to the posterior edge of the cephalothorax. AL was measured from the most posterior point of the cervical groove, where the two concave lines that define the areola begin, to the end of these lines on the posterior dorsal surface of the cephalothorax. AW was measured at the narrowest point between the two concave lines of the areola. PW was defined was the widest point of the chelae perpendicular to the finger tips. The left cheiliped was measured to obtain PW whenever possible; if missing or in an early stage of regeneration, the right cheiliped was measured instead. Additional data were collected on whether a female had eggs/young, whether a male was form I, and other miscellaneous information such as missing or abnormal body parts.

After morphometric data were collected, both gonopods were removed from all male crayfish with forceps and placed in a 1.5 mL Eppendorf tube filled with 70% ethanol; each tube was then labeled by sample site and crayfish species. Using a scalpel sterilized in 70% ethanol and thoroughly rinsed with distilled water, the abdomen was removed and the muscle tissue extracted. After removing any unwanted materials from the tissue (*e.g.,* exoskeleton fragments or intestine), the sample was placed in a labeled 1.5 mL Eppendorf tube and frozen. Any additional male crayfish from a site were also placed in 70% ethanol and labeled by site, species and date collected in the field. Preserving all gonopods of processed individuals along with whole specimens served as vouchers for species identification. Abdominal tissue samples were collected for future analysis of total mercury concentration.

All statistical analyses of morphometric data were performed using SPSS version 20. Mean values for BWM, TBL, CL, AL, AW, and PW were compared across waterways, species and sex using ANOVA with an alpha significance level of 0.05. Sample size and variance were not equal across groups, so the nonparametric Games-Howell post hoc test was chosen to determine significant differences in body measurements across variables. Weight-length relationships (CL and BWM; BWM and PW) were graphed for each species and an r^2 value for both sexes was obtained to measure the strength of the relationship. Crayfish density at each site was calculated taking the area of a semicircle (1/2 πr^2 where r = half the mean seine net width) to first estimate the area sampled. The number of crayfish caught at the site was then divided by this value to obtain crayfish per meter squared. Crayfish density could not be calculated for certain sites where seining was not semi-quantitative or qualitative hand capture with a mask and snorkel was used instead. Sex ratios were determined using the total number of male and female crayfish recorded for each species at each site. To determine whether a given sex ratio differed significantly from a 1:1 male to female ratio, a chi square goodness of fit test with alpha significance level at 0.05 was performed.

Lab Sampling Procedures: Tables and Figures

Figure 3: Crayfish Morphometric Measurements.

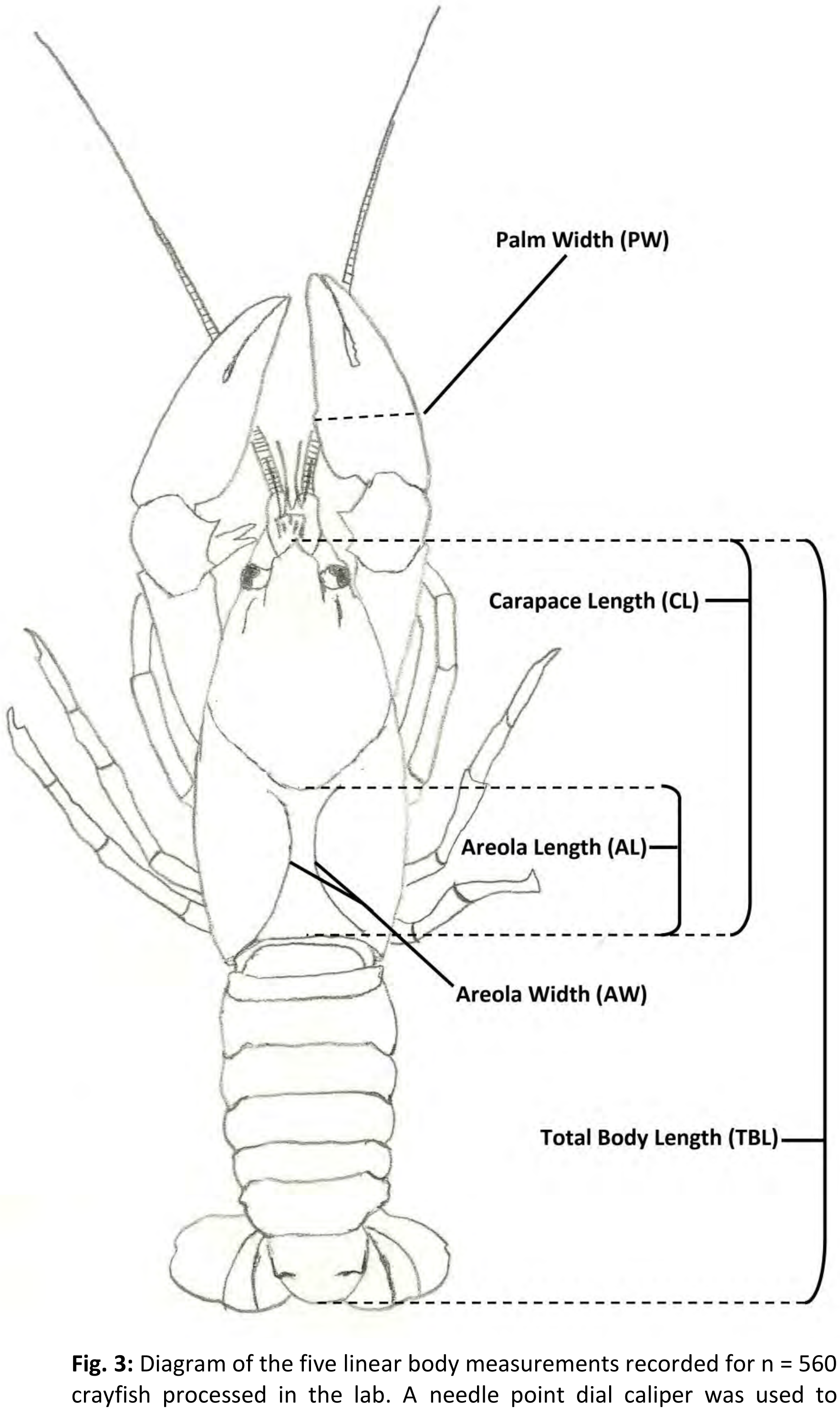


Fig. 3: Diagram of the five linear body measurements recorded for n = 560 crayfish processed in the lab. A needle point dial caliper was used to record each measurement to the nearest one-tenth of a millimeter.

RESULTS

Species Occurrence

Surveys at 28 samples sites among seven West Branch waterways yielded a total of 1888 crayfish consisting of three species: *Orconectes obscurus*, *O. rusticus* and *Cambarus bartonii*. Among the total crayfish caught, 1508 (79.9%) were *O. obscurus*, 308 (16.3%) were *O. rusticus* and 72 (3.8%) were *C. bartonii*. As seen in Fig. 1, *O. obscurus* was the most commonly-encountered species in the study area, present in six of seven streams surveyed and 23 of the 28 total samples sites. Of these 23 sites, 20 (yellow diamonds in Fig. 1) featured *O. obscurus* as virtually the only species present (negligible numbers of *C. bartonii* were occasionally recorded). The only sites that did not yield *O. obscurus* were the five sampling points on Loyalsock Creek (LOY01 – LOY05; purple and red diamonds in Fig. 1).

O. rusiticus was found to be the predominant species in Loyalsock Creek, and this species was not collected in any other stream in this study. No individuals of *O. obscurus* and fewer than ten total individuals of *C. bartonii* were recorded among the five sample sites on Loyalsock Creek. The most downstream sampling location (LOY01 at stream kilometer 3.3) yielded possible *O. rusticus x obscurus* hybrids (purple diamond, Fig. 1). Many specimens from this location had missing or indistinct features typically present in rusty crayfish, making species identification difficult (see Appendix I, Table 22). For the purposes of statistical analysis these individuals were considered *O. rusticus*.

C. bartonii was the least frequently caught species in this study, only present in large enough numbers for morphometric analysis at KET02 (stream km 22.7) and KET04 (stream km 46.3) on Kettle Creek and PC07 (stream km 104.1) on Pine Creek (green diamonds, Fig. 1). Stray individuals of this species were caught in Lycoming and Little Pine Creeks and were infrequently observed in Loyalsock Creek. *C. bartonii* was never the only crayfish species present at a sample site, and was never more numerous than *O. obscurus* at KET02, KET04 and PC07.

Morphometry

A total of n = 560 crayfish were processed in the lab for morphometric and total mercury analysis. Pine Creek, the largest tributary surveyed in this study, contributed the most specimens (26.79%) followed by Kettle Creek (17.14%); Muncy Creek (7.14%) and Little Pine Creek (8.93%) contributed the least number of crayfish (Fig. 4). The species composition of the samples processed in the lab was n = 435 (77.68%) *O. obscurus*, n = 86 (15.36%) *O. rusticus* and n = 39 (6.96%) *C. bartonii* (Fig. 5). Table 2 provides a summary of the morphometric data by crayfish species and sex. In all three species, males had higher averages than females for all body measurements with the exception of AW (mm) in *O. obscurus* ($x = 1.7 \pm 0.37$ for males and $x = 1.7 \pm 0.38$ for females). Analyzing these differences with a nonparametric ANOVA at $\alpha = 0.05$, between male and female *C. bartonii* the only average not significantly higher for males was TBL, with $p = 0.068$ (Table 3). Male and female *O. obscurus* did not differ significantly in two body measurements: TBL ($p = 0.110$) and AW ($p = 0.368$) (Table 4). AW was the only average value not significantly higher in *O. rusticus* males compared to females ($p = 0.062$) (Table 5).

Among male crayfish, *O. rusticus* had the highest average for all body measurements, followed by *O. obscurus* and *C. bartonii* (Table 2). Among females, *C. bartonii* also had the lowest average for all measurements but as seen in Table 2 female *O. obscurus* had a higher average than female *O. rusticus* for BWM (g) ($x = 10.0 \pm 3.81$ vs. 9.2 ± 3.09), TBL (mm) ($x = 65.5 \pm 8.68$ vs. 64.1 ± 7.28), CL (mm) ($x = 31.3 \pm 4.27$ vs. 30.3 ± 3.18) and AL (mm) ($x = 8.1 \pm$

1.34 vs. 7.6 ± 1.1). The distribution of BWM, CL and PW are represented with box plots among species and sex in Figs. 6a-c.

Body size differences among the males of each species were analyzed using nonparametric ANOVA tests at $\alpha = 0.05$ (Tables 6a-c). In male crayfish, only AW was not significantly higher in *O. rusticus* males compared to *C. bartonii* males ($p = 0.112$) (Table 6a). Comparing male *O. obscurus* with male *C. bartonii* (Table 6b), only BWM ($p = 0.014$), TBL ($p<0.001$) and CL ($p <0.001$) were significantly higher in *O. obscurus*. *O. rusticus* had a significantly higher mean than *O. obscurus* for all body measurements except AL ($p = 0.160$) (Table 6c). Tables 7a-c show the results of ANOVA tests comparing female crayfish of each species. *O. rusticus* females had significantly higher averages compared to *C. bartonii* females excluding AL ($p = 0.432$) (Table 7a), while female *O. obscurus* had significantly higher averages than female *C. bartonii* for all six measurements (Table 7b). Comparing the females of the two *Orconectes* species (Table 7c), only AL was significantly different, being higher for *O. obscurus* ($p = 0.027$).

Comparing chelae width among male crayfish, *O. rusticus* had a higher average PW than *C. bartonii* (x = 11.7 mm ± 2.4 vs. x = 10.2 mm ± 1.31, respectively) and this difference was found to be statistically significant ($p = 0.01$); PW was also significantly higher for *O. rusticus* (x = 11.7 mm ± 2.4) than for *O. obscurus* (x = 10.5 mm ± 2.52) ($p = 0.005$). Between male *O. obscurus* and *C. bartonii*, average PW for *O. obscurus* was also higher (x = 10.5 mm ± 2.52 vs. x =10.2 mm ± 1.31, respectively); however, this difference was not found be statistically significant ($p = 0.593$).

Blotted wet mass was plotted with carapace length and palm width for each species and sex in Figures 7a-b, 8a-b and 9a-b. The r^2 values ranged from 0.607 (Fig. 9b) to 0.897 (Fig. 7a).

BWM and PW in *O. rusticus* males and females (r^2 = 0.607 and r^2 = 0.695, respectively), and *O. obscurus* males and females (r^2 = 0.663 and r^2 = 0.675, respectively) showed a weaker relationship compared to *C. bartonii* (r^2 = 0.888 for males and r^2 = 0.852 for females). BWM and CL generally yielded higher r^2 values compared to the respective BWM and PW relationship, with the lowest at r^2 = 0.729 for male *O. rusticus* (Fig. 9a). In all but one comparison (*C. bartonii* BWM and PW, Fig. 7b), female crayfish had slightly higher r^2 values compared to males.

Areola length versus width and the proportion of the carapace consisting of the areola were calculated from the morphometric data in Table 2. In *O. obscurus* the areola was x = 4.9 times longer than wide, and the areola comprised x = 26.3% of the carapace length (x = 26.8% in males; x =25.9% in females). In *O. rusticus* the areola was x = 4.9 times longer than wide and comprised x = 26.1% of the carapace length (x = 26.7% in males; x = 25.1% in females). In *C. bartonii*, the areola was x = 5.3 times longer than wide and accounted for x = 29% of the carapace length (x = 29.4% in males; x = 28.1% in females).

Table 7 summarizes crayfish morphometric data by stream. Among waterway totals, crayfish from Larry's Creek (n = 60) and Lycoming Creek (n = 78) had the lowest BWM (g) (x = 8.4 ± 2.19 and x = 8.4 ± 3.68, respectively), while crayfish from Muncy Creek (n = 40) had the highest BWM (x = 13.9 ± 4.88). Crayfish collected from Larry's Creek had the smallest CL (mm) (x = 29.6 ± 2.12) and individuals from Lycoming Creek had the smallest PW (mm) (x = 7.8 ± 2.26); crayfish from Muncy Creek had both the largest CL (x = 34.5 ± 3.81) and PW (x = 11.5 ± 3.16). Among male *O. obscurus*, individuals from Larry's Creek had the lowest BWM (g) (x = 8.5 ± 2.49) and smallest CL (mm) (x = 29.5 ± 2.04). Male *O. obscurus* from Lycoming Creek had the smallest PW (mm) (x = 8.9 ± 1.64). Among female *O. obscurus*, individuals from

Lycoming Creek had the lowest BWM (x = 6.9 ± 4.17) and smallest CL (x = 26.7 ± 4.47) and PW (x = 6.7 ± 2.27). Comparing male *C. bartonii* from Kettle Creek and Pine Creek, individuals from Pine Creek had a higher average BWM but mean CL and PW were higher in males from Kettle Creek (Table 7). Female *C. bartonii* were only collected for processing from Pine Creek, so no comparisons across waterways could be made; *O.rusticus* was only collected from a single waterway as well (Loyalsock Creek).

Tables 8a-c combine crayfish totals for each waterway and compare means for BWM, CL and PW using a nonparametric Games-Howell test at $\alpha = 0.05$. Crayfish from Pine Creek had significantly higher means for BWM, CL and PW compared to crayfish from every other stream except for Loyalsock Creek and Muncy Creek. Crayfish from Loyalsock Creek had a significantly lower average for CL ($p<0.001$, Table 8b) compared to Pine Creek but BWM and PW were not statistically different. In crayfish from Muncy Creek, no significant differences were observed from Pine Creek crayfish regarding BWM ($p = 0.532$, Table 8a), CL ($p = 0.967$, Table 8b) or PW ($p = 0.432$, Table 8c). Muncy Creek crayfish had significantly higher averages for BWM, CL and PW compared to all other streams except for the previously mentioned Pine Creek and Loyalsock Creek (no significant difference in PW, $p = 0.244$). Mean values for crayfish from Kettle Creek were not significantly different from those collected in Larry's Creek for BWM ($p = 0.125$), CL ($p = 0.999$) or PW ($p = 0.997$).

Morphometry: Tables and Figures

Figure 4: Total Number and Percentage of Crayfish Processed by Waterway.

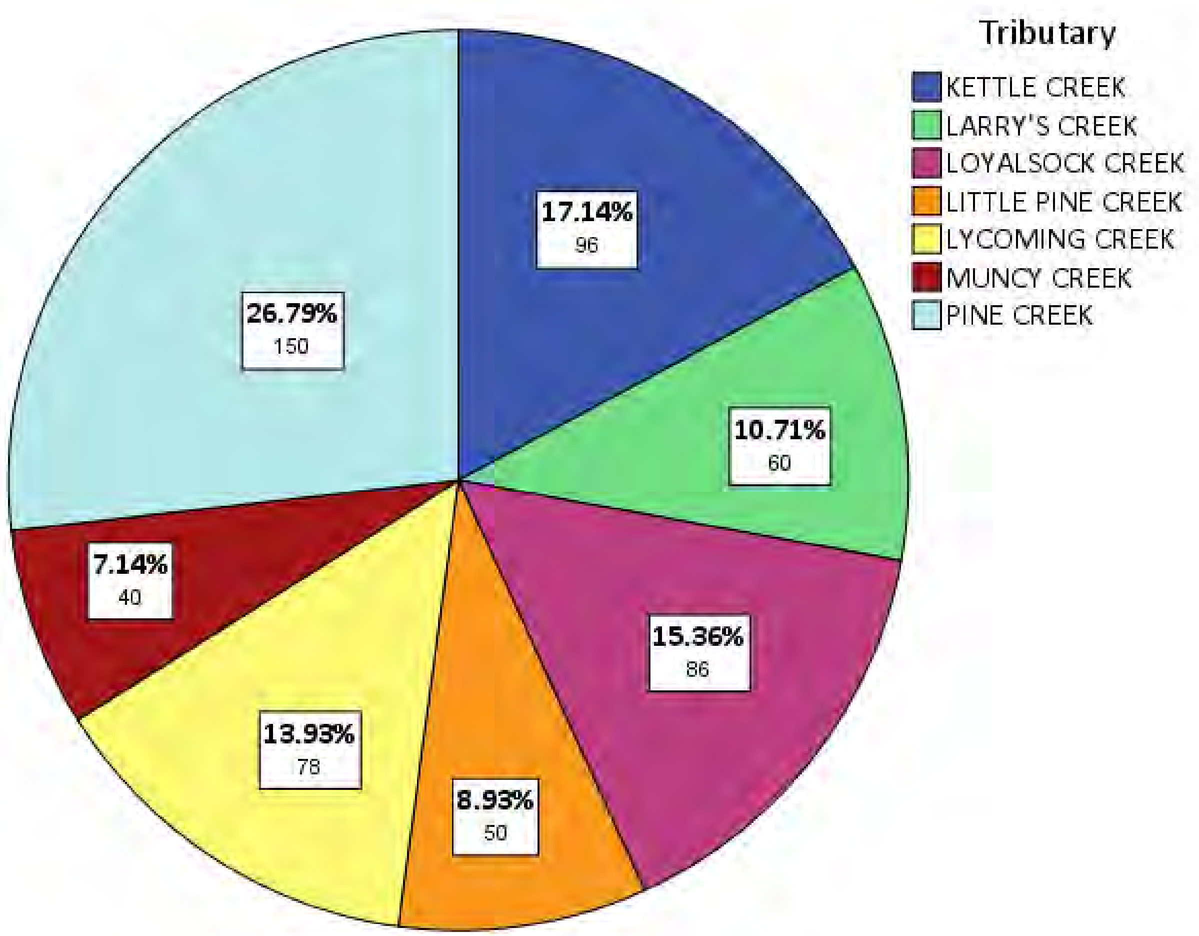


Fig. 4: Pie chart displaying the distribution of crayfish processed in the lab by stream of origin. Kettle Creek, Pine Creek, Larry's Creek, Lycoming Creek, Loyalsock Creek and Muncy Creek are major tributaries of the West Branch of the Susquehanna River in northcentral Pennsylvania; Little Pine Creek is a major tributary of Pine Creek. A total of n = 560 crayfish were processed for morphometric analysis.

Figure 5: Total Number and Percentage of Crayfish Processed by Species.

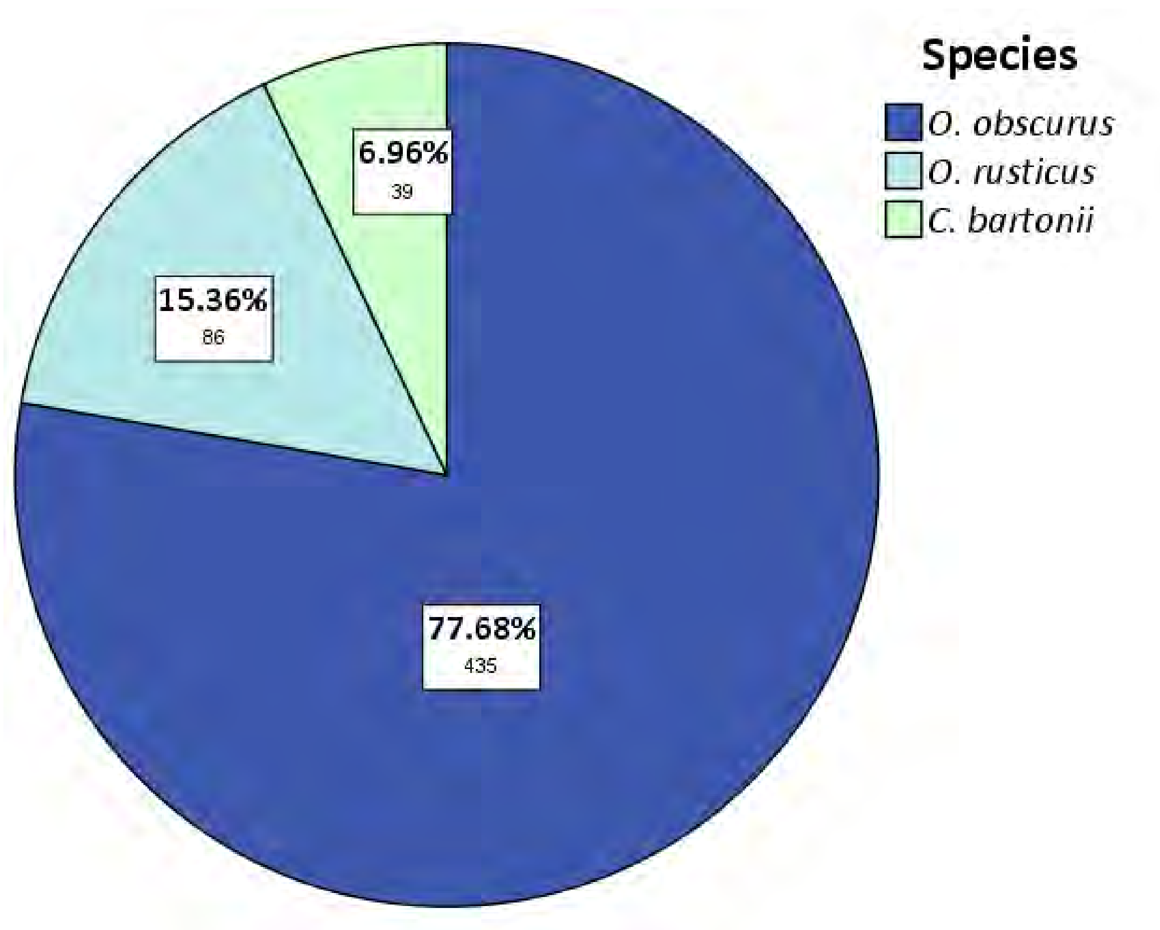


Fig. 5: Pie chart displaying the species composition of the n = 560 crayfish processed in the lab for morphometric analysis. Crayfish species consisted of the non-natives *Orconectes obscurus* (Allegheny crayfish) and *O. rusticus*, (rusty crayfish) and the native *Cambarus bartonii* (common/Appalachian brook crayfish).

Table 2: Differences in Crayfish Size Among Species.

Species	Sex		Blotted Wet Mass (g)	Total Body Length (mm)	Carapace Length (mm)	Areola Length (mm)	Areola Width (mm)	Palm Width (mm)
O. obscurus	Male (n = 217)	Mean ± SD	11.3 ± 3.89	66.7 ± 6.41	32.5 ± 3.42	8.7 ± 1.32	1.7 ± 0.37	10.5 ± 2.52
		Min.	4.1	49.3	23.1	5.6	0.8	0
		Max.	25.8	89.4	45.6	14.0	2.9	17.8
	Female (n = 218)	Mean ± SD	10.0 ± 3.81	65.5 ± 8.68	31.3 ± 4.27	8.1 ± 1.34	1.7 ± 0.38	8.8 ± 2.1
		Min.	1.5	35.4	18.6	4.2	0.8	0
		Max.	20.0	83.5	40.5	11.2	2.9	13.5
	Total (n = 435)	Mean ± SD	10.6 ± 3.9	66.1 ± 7.64	31.9 ± 3.91	8.4 ± 1.36	1.7 ± 0.38	9.7 ± 2.47
		Min.	1.5	35.4	18.6	4.2	0.8	0
		Max.	25.8	89.4	45.6	14.0	2.9	17.8
O. rusticus	Male (n = 40)	Mean ± SD	13.5 ± 3.74	68.8 ± 4.63	33.7 ± 2.38	9.0 ± 0.95	1.8 ± 0.39	11.7 ± 2.4
		Min.	7.6	60.4	29.9	6.9	1.0	7.4
		Max.	20.9	79.2	40.1	10.7	2.9	16.1
	Female (n = 46)	Mean ± SD	9.2 ± 3.09	64.1 ± 7.28	30.3 ± 3.18	7.6 ± 1.1	1.7 ± 0.37	8.8 ± 2.15
		Min.	2.8	45.6	21.6	4.7	0.9	0
		Max.	16.3	79.0	37.7	10.3	2.4	12.0
	Total (n = 86)	Mean ± SD	11.2 ± 4.02	66.3 ± 6.6	31.8 ± 3.3	8.3 ± 1.24	1.7 ± 0.39	10.1 ± 2.7
		Min.	2.8	45.6	21.6	4.7	0.9	0
		Max.	20.9	79.2	40.1	10.7	2.9	16.1
C. bartonii	Male (n = 20)	Mean ± SD	9.1 ± 2.31	57.8 ± 3.77	28.9 ± 2.25	8.5 ± 0.76	1.6 ± 0.38	10.2 ± 1.31
		Min.	5.8	53.0	25.8	7.0	1.1	8.4
		Max.	16.8	68.4	36.1	9.8	2.6	14.0
	Female (n = 19)	Mean ± SD	6.8 ± 3.24	54.3 ± 7.27	26.3 ± 3.76	7.4 ± 1.33	1.4 ± 0.29	7.2 ± 2.1
		Min.	2.7	42.1	20.9	5.2	1.0	3.1
		Max.	12.9	66.6	33.0	9.8	1.9	10.7
	Total (n = 39)	Mean ± SD	8.0 ± 3.0	56.1 ±5.94	27.6 ± 3.31	8.0 ± 1.2	1.5 ± 0.36	8.7 ± 2.29
		Min.	2.7	42.1	20.9	5.2	1.0	3.1
		Max.	16.8	68.4	36.1	9.8	2.6	14.0

Table 2: A total of n = 560 crayfish from six watersheds of the West Branch of the Susquehanna River in northcentral Pennsylvania were processed for morphometric analysis. Crayfish species consisted of the non-natives *Orconectes obscurus* (Allegheny crayfish) and *O. rusticus* (rusty crayfish) and the native *Cambarus bartonii* (common/Appalachian brook crayfish).

Table 3: ANOVA Results for Body Size Differences between Male and Female C. bartonii.							
			Sum of Squares	**df**	**Mean Square**	**F**	**P (α = 0.05)**
Blotted Wet Mass	Between Groups	(Combined)	52.277	1	52.277	6.656	0.014
	Within Groups		290.615	37	7.854		
	Total		342.892	38			
Total Body Length	Between Groups	(Combined)	116.664	1	116.664	3.533	0.068
	Within Groups		1221.839	37	33.023		
	Total		1338.503	38			
Carapace Length	Between Groups	(Combined)	65.867	1	65.867	6.957	0.012
	Within Groups		350.280	37	9.467		
	Total		416.147	38			
Areola Length	Between Groups	(Combined)	11.694	1	11.694	10.127	0.003
	Within Groups		42.723	37	1.155		
	Total		54.417	38			
Areola Width	Between Groups	(Combined)	.490	1	0.490	4.202	0.048
	Within Groups		4.313	37	0.117		
	Total		4.803	38			
Palm Width	Between Groups	(Combined)	86.466	1	86.466	28.506	0.000
	Within Groups		112.232	37	3.033		
	Total		198.697	38			

Table 3: Results of a nonparametric ANOVA (α = 0.05) comparing differences in body size between the native male (n = 20) and female (n = 19) *Cambarus bartonii* (common/Appalachian brook crayfish). For all measurements males had a higher average than females.

Table 4: ANOVA Results for Body Size Differences between Male and Female *O. obscurus*.							
			Sum of Squares	**df**	**Mean Square**	**F**	**P (α = 0.05)**
Blotted Wet Mass	Between Groups	(Combined)	174.672	1	174.672	11.784	0.001
	Within Groups		6418.054	433	14.822		
	Total		6592.725	434			
Total Body Length	Between Groups	(Combined)	149.161	1	149.161	2.562	0.110
	Within Groups		25206.226	433	58.213		
	Total		25355.387	434			
Carapace Length	Between Groups	(Combined)	166.751	1	166.751	11.147	0.001
	Within Groups		6477.556	433	14.960		
	Total		6644.307	434			
Areola Length	Between Groups	(Combined)	41.972	1	41.972	23.834	0.000
	Within Groups		762.519	433	1.761		
	Total		804.491	434			
Areola Width	Between Groups	(Combined)	0.116	1	0.116	0.814	0.368
	Within Groups		61.730	433	0.143		
	Total		61.846	434			
Palm Width	Between Groups	(Combined)	316.164	1	316.164	58.785	0.000
	Within Groups		2328.823	433	5.378		
	Total		2644.987	434			

Table 4 Results of a nonparametric ANOVA (α = 0.05) comparing differences in body size between non-native male (n = 217) and female (n = 218) *Orconectes obscurus* (Allegheny crayfish). Except for areola width, males had a higher average than females for body measurements.

Table 5: ANOVA Results for Body Size Differences between Male and Female O. rusticus.							
			Sum of Squares	**df**	**Mean Square**	**F**	**P (α = 0.05)**
Blotted Wet Mass	Between Groups	(Combined)	400.998	1	400.998	34.557	0.000
	Within Groups		974.746	84	11.604		
	Total		1375.744	85			
Total Body Length	Between Groups	(Combined)	478.302	1	478.302	12.477	0.001
	Within Groups		3220.207	84	38.336		
	Total		3698.509	85			
Carapace Length	Between Groups	(Combined)	247.742	1	247.742	30.811	0.000
	Within Groups		675.414	84	8.041		
	Total		923.155	85			
Areola Length	Between Groups	(Combined)	41.857	1	41.857	39.493	0.000
	Within Groups		89.028	84	1.060		
	Total		130.884	85			
Areola Width	Between Groups	(Combined)	0.521	1	0.521	3.567	0.062
	Within Groups		12.257	84	0.146		
	Total		12.777	85			
Palm Width	Between Groups	(Combined)	187.788	1	187.788	36.423	0.000
	Within Groups		433.085	84	5.156		
	Total		620.873	85			

Table 5: Results of a nonparametric ANOVA (α = 0.05) comparing differences in body size between non-native male (n = 40) and female (n = 46) *Orconectes rusticus* (rusty crayfish). For all body measurements males had a higher average than females.

Figure 6a: Distribution of Blotted Wet Mass (g) among Three Crayfish Species.

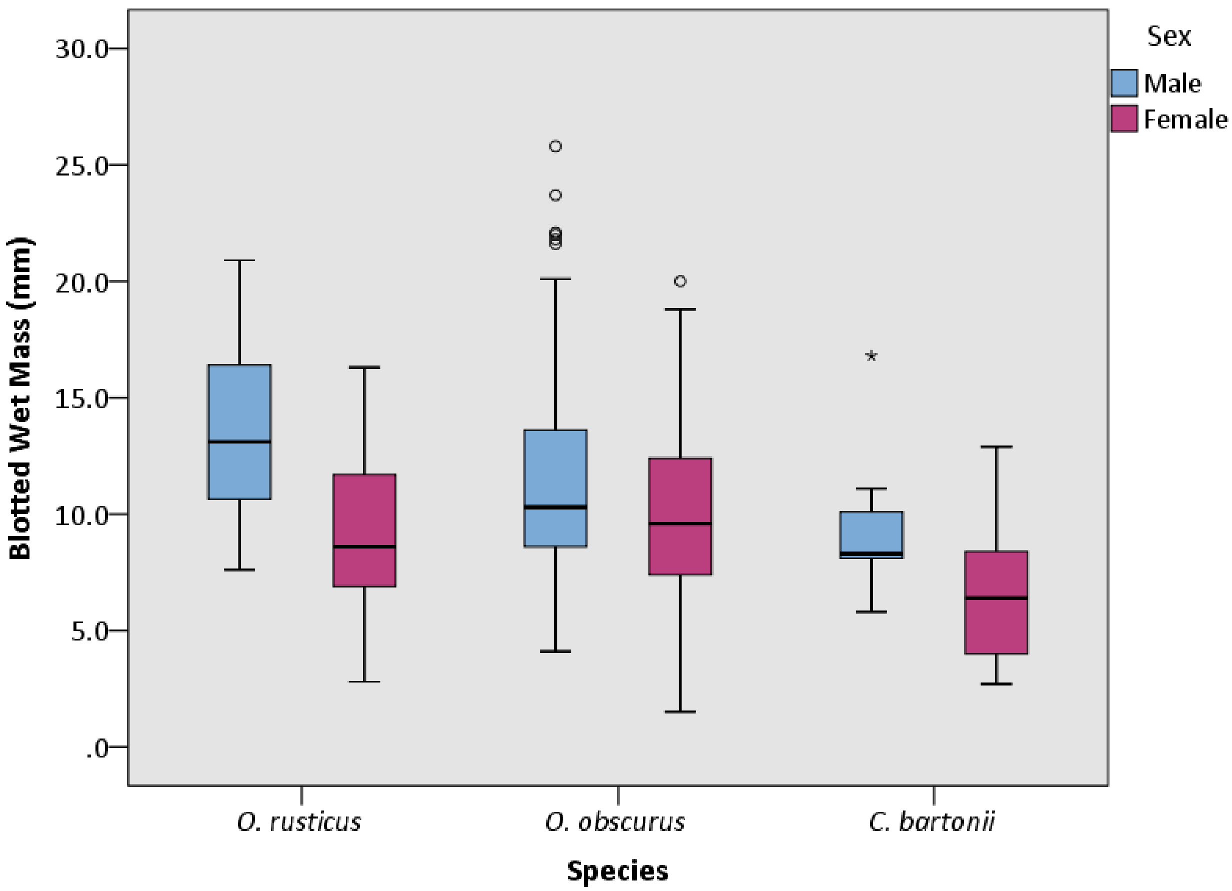


Figure 6a: Box plots displaying the distribution of blotted wet mass (g) among non-native *Orconectes rusticus* (rusty crayfish) and *O. obscurus* (Allegheny crayfish) and native *Cambarus bartonii* (common/Appalachian brook crayfish) from the West Branch sub-basin of the Susquehanna River in northcentral Pennsylvania.

Figure 6b: Distribution of Carapace Length (mm) among Three Crayfish Species.

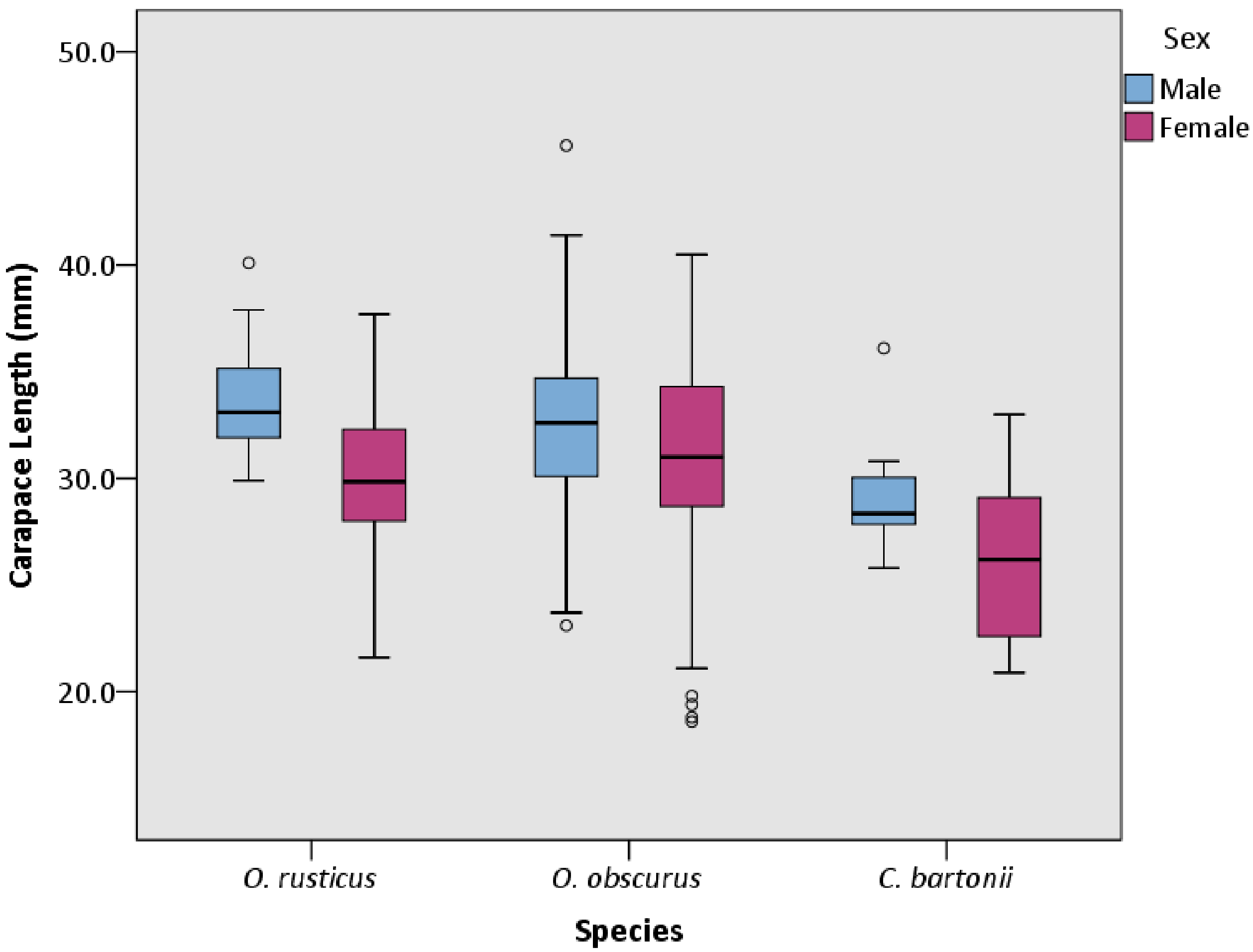


Figure 6b: Box plots displaying the distribution of carapace length (mm) among non-native *Orconectes rusticus* (rusty crayfish) and *O. obscurus* (Allegheny crayfish) and native *Cambarus bartonii* (common/Appalachian brook crayfish) from the West Branch sub-basin of the Susquehanna River in northcentral Pennsylvania.

Figure 6c: Distribution of Palm Width (mm) among Three Crayfish Species.

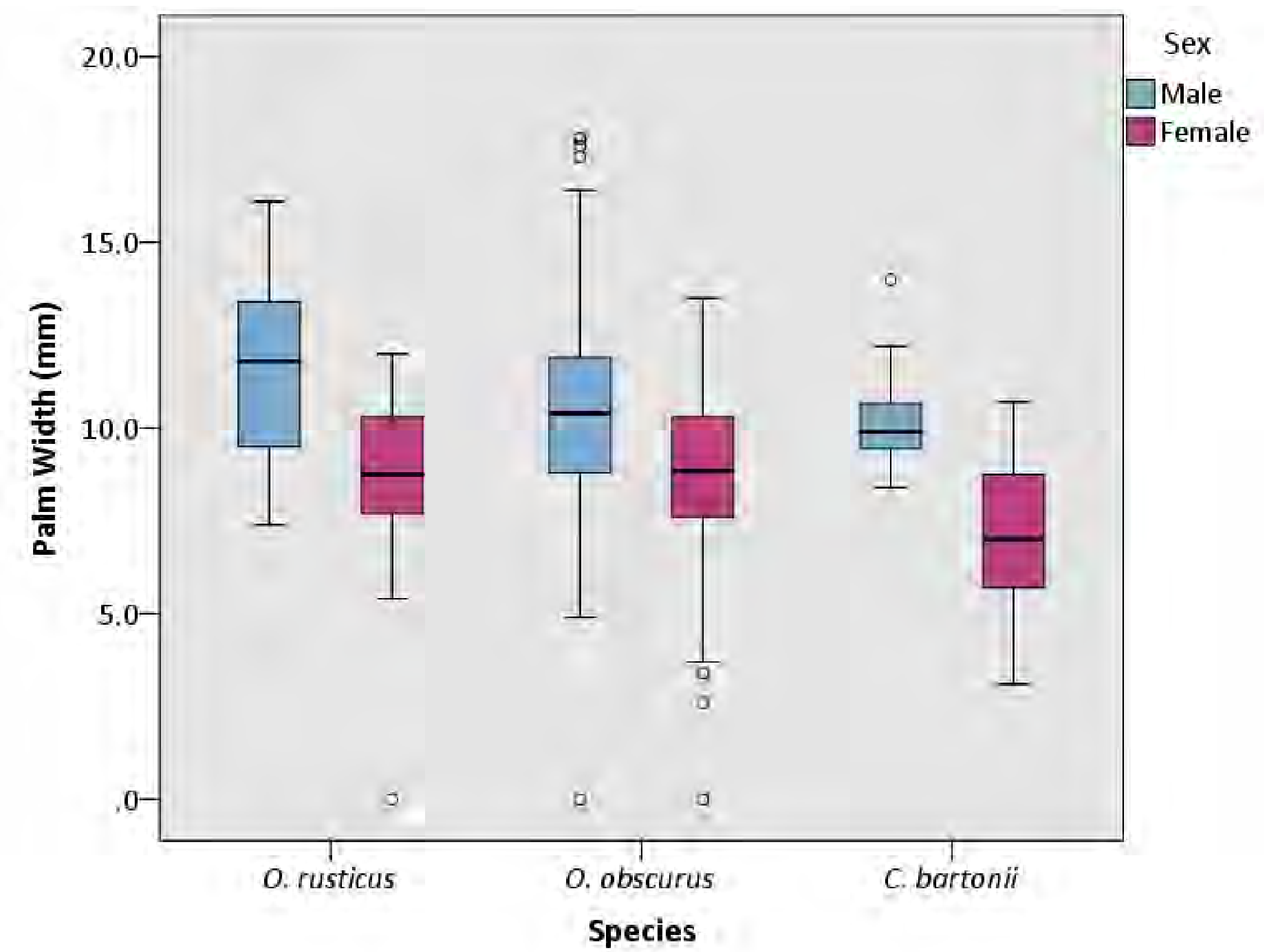


Figure 6c: Box plots displaying the distribution of palm width (mm) among non-native *Orconectes rusticus* (rusty crayfish) and *O. obscurus* (Allegheny crayfish) and native *Cambarus bartonii* (common/Appalachian brook crayfish) from the West Branch sub-basin of the Susquehanna River in northcentral Pennsylvania.

Table 6a: ANOVA Results for Body Size Differences between Male *C. bartonii* and *O. rusticus.*							
			Sum of Squares	**df**	**Mean Square**	**F**	**P (α = 0.05)**
Blotted Wet Mass	Between Groups	(Combined)	260.191	1	260.191	23.371	0.000
	Within Groups		645.726	58	11.133		
	Total		905.917	59			
Total Body Length	Between Groups	(Combined)	1636.147	1	1636.147	85.827	0.000
	Within Groups		1105.677	58	19.063		
	Total		2741.824	59			
Carapace Length	Between Groups	(Combined)	301.467	1	301.467	55.361	0.000
	Within Groups		315.839	58	5.445		
	Total		617.306	59			
Areola Length	Between Groups	(Combined)	4.107	1	4.107	5.178	0.027
	Within Groups		46.002	58	0.793		
	Total		50.108	59			
Areola Width	Between Groups	(Combined)	0.397	1	0.397	2.611	0.112
	Within Groups		8.812	58	0.152		
	Total		9.208	59			
Palm Width	Between Groups	(Combined)	31.212	1	31.212	7.036	0.010
	Within Groups		257.284	58	4.436		
	Total		288.496	59			

Table 6a: Results of a nonparametric ANOVA (α = 0.05) comparing differences in body size between n = 20 male *Cambarus bartonii* (common/Appalachian brook crayfish) and n= 40 male *Orconectes rusticus* (rusty crayfish). For all body measurements *O. rusticus* had a higher average than *C. bartonii*. *O. rusticus* is non-native to the West Branch sub-basin.

Table 6b: ANOVA Results for Body Size Differences between Male *C. bartonii* and *O. obscurus.*							
			Sum of Squares	**df**	**Mean Square**	**F**	**P (α = 0.05)**
Blotted Wet Mass	Between Groups	(Combined)	88.064	1	88.064	6.143	0.014
	Within Groups		3368.905	235	14.336		
	Total		3456.969	236			
Total Body Length	Between Groups	(Combined)	1468.994	1	1468.994	37.777	0.000
	Within Groups		9138.211	235	38.886		
	Total		10607.205	236			
Carapace Length	Between Groups	(Combined)	243.441	1	243.441	21.874	0.000
	Within Groups		2615.399	235	11.129		
	Total		2858.841	236			
Areola Length	Between Groups	(Combined)	1.124	1	1.124	0.686	0.408
	Within Groups		384.763	235	1.637		
	Total		385.887	236			
Areola Width	Between Groups	(Combined)	0.030	1	0.030	0.215	0.643
	Within Groups		33.027	235	0.141		
	Total		33.057	236			
Palm Width	Between Groups	(Combined)	1.709	1	1.709	0.286	0.593
	Within Groups		1404.993	235	5.979		
	Total		1406.703	236			

Table 6b: Results of a nonparametric ANOVA (α = 0.05) comparing differences in mean body measurements between n = 20 male *Cambarus bartonii* (common/Appalachian brook crayfish) and n= 217 male *Orconectes obscurus* (Allegheny crayfish). For all body measurements *O. obscurus* had a higher average than *C. bartonii*. *O. obscurus* is non-native to the West Branch sub-basin.

Table 6c: ANOVA Results for Body Size Differences between Male *O. obscurus* and *O. rusticus.*							
			Sum of Squares	df	Mean Square	F	P (α = 0.05)
Blotted Wet Mass	Between Groups	(Combined)	167.136	1	167.136	11.182	0.001
	Within Groups		3811.595	255	14.947		
	Total		3978.731	256			
Total Body Length	Between Groups	(Combined)	151.936	1	151.936	3.993	0.047
	Within Groups		9702.509	255	38.049		
	Total		9854.445	256			
Carapace Length	Between Groups	(Combined)	41.532	1	41.532	3.866	0.050
	Within Groups		2739.638	255	10.744		
	Total		2781.170	256			
Areola Length	Between Groups	(Combined)	3.189	1	3.189	1.989	0.160
	Within Groups		408.874	255	1.603		
	Total		412.063	256			
Areola Width	Between Groups	(Combined)	0.587	1	0.587	4.125	0.043
	Within Groups		36.302	255	0.142		
	Total		36.890	256			
Palm Width	Between Groups	(Combined)	50.639	1	50.639	8.085	0.005
	Within Groups		1597.237	255	6.264		
	Total		1647.876	256			

Table 6c: Results of a nonparametric ANOVA (α = 0.05) comparing differences in mean body measurements between n = 217 male *Orconectes obscurus* (Allegheny crayfish) and n= 40 male *Orconectes rusticus* (rusty crayfish). For all body measurements *O. rusticus* had a higher average than *O. obscurus*. Both species are non-native to the West Branch sub-basin.

Table 7a: ANOVA Results for Body Size Differences between Female C. bartonii and O. rusticus.							
			Sum of Squares	**df**	**Mean Square**	**F**	**P (α = 0.05)**
Blotted Wet Mass	Between Groups	(Combined)	77.746	1	77.746	7.905	0.007
	Within Groups		619.635	63	9.835		
	Total		697.381	64			
Total Body Length	Between Groups	(Combined)	1293.903	1	1293.903	24.433	0.000
	Within Groups		3336.369	63	52.958		
	Total		4630.271	64			
Carapace Length	Between Groups	(Combined)	210.025	1	210.025	18.640	0.000
	Within Groups		709.855	63	11.268		
	Total		919.879	64			
Areola Length	Between Groups	(Combined)	0.853	1	0.853	0.627	0.432
	Within Groups		85.749	63	1.361		
	Total		86.602	64			
Areola Width	Between Groups	(Combined)	0.779	1	0.779	6.328	0.014
	Within Groups		7.758	63	0.123		
	Total		8.538	64			
Palm Width	Between Groups	(Combined)	32.152	1	32.152	7.032	0.010
	Within Groups		288.033	63	4.572		
	Total		320.185	64			

Table 7a: Results of a nonparametric ANOVA (α = 0.05) comparing differences in mean body measurements between n = 19 female *Cambarus bartonii* (common/Appalachian brook crayfish) and n= 46 female *Orconectes rusticus* (rusty crayfish). For all body measurements *O. rusticus* had a higher average than *C. bartonii*. *O. rusticus* is non-native to the West Branch sub-basin.

Table 7b: ANOVA Results for Body Size Differences between Female C. bartonii and O. obscurus.							
			Sum of Squares	**df**	**Mean Square**	**F**	**P (α = 0.05)**
Blotted Wet Mass	Between Groups	(Combined)	183.681	1	183.681	12.925	0.000
	Within Groups		3339.764	235	14.212		
	Total		3523.445	236			
Total Body Length	Between Groups	(Combined)	2210.189	1	2210.189	30.040	0.000
	Within Groups		17289.854	235	73.574		
	Total		19500.043	236			
Carapace Length	Between Groups	(Combined)	438.284	1	438.284	24.451	0.000
	Within Groups		4212.437	235	17.925		
	Total		4650.721	236			
Areola Length	Between Groups	(Combined)	9.110	1	9.110	5.092	0.025
	Within Groups		420.479	235	1.789		
	Total		429.589	236			
Areola Width	Between Groups	(Combined)	1.547	1	1.547	11.011	0.001
	Within Groups		33.017	235	0.140		
	Total		34.564	236			
Palm Width	Between Groups	(Combined)	43.596	1	43.596	9.889	0.002
	Within Groups		1036.062	235	4.409		
	Total		1079.658	236			

Table 7b: Results of a nonparametric ANOVA (α = 0.05) comparing differences in mean body measurements between n = 19 female *Cambarus bartonii* (common/Appalachian brook crayfish) and n= 218 female *Orconectes obscurus* (Allegheny crayfish). For all body measurements *O. obscurus* had a higher average than *C. bartonii*. *O. obscurus* is non-native to the West Branch sub-basin.

Table 7c: ANOVA Results for Body Size Differences between Female O. obscurus and O. rusticus.							
			Sum of Squares	**df**	**Mean Square**	**F**	**P (α = 0.05)**
Blotted Wet Mass	Between Groups	(Combined)	26.632	1	26.632	1.948	0.164
	Within Groups		3581.205	262	13.669		
	Total		3607.838	263			
Total Body Length	Between Groups	(Combined)	78.330	1	78.330	1.096	0.296
	Within Groups		18723.924	262	71.465		
	Total		18802.254	263			
Carapace Length	Between Groups	(Combined)	42.328	1	42.328	2.513	0.114
	Within Groups		4413.332	262	16.845		
	Total		4455.660	263			
Areola Length	Between Groups	(Combined)	8.397	1	8.397	4.970	0.027
	Within Groups		442.673	262	1.690		
	Total		451.070	263			
Areola Width	Between Groups	(Combined)	0.122	1	0.122	0.851	0.357
	Within Groups		37.684	262	0.144		
	Total		37.807	263			
Palm Width	Between Groups	(Combined)	0.042	1	0.042	0.009	0.923
	Within Groups		1164.671	262	4.445		
	Total		1164.713	263			

Table 7c: Results of a nonparametric ANOVA (α = 0.05) comparing differences in mean body measurements between n = 218 female *Orconectes obscurus* (Allegheny crayfish) and n= 46 female *Orconectes rusticus* (rusty crayfish). *O. obscurus* had a higher average for all body measurements than *O. rusticus* except for areola width and palm width. Both species are non-native to the West Branch sub-basin.

Figure 7a: Relationship between Carapace Length (mm) and Blotted Wet Mass (g) in *C. bartonii.*

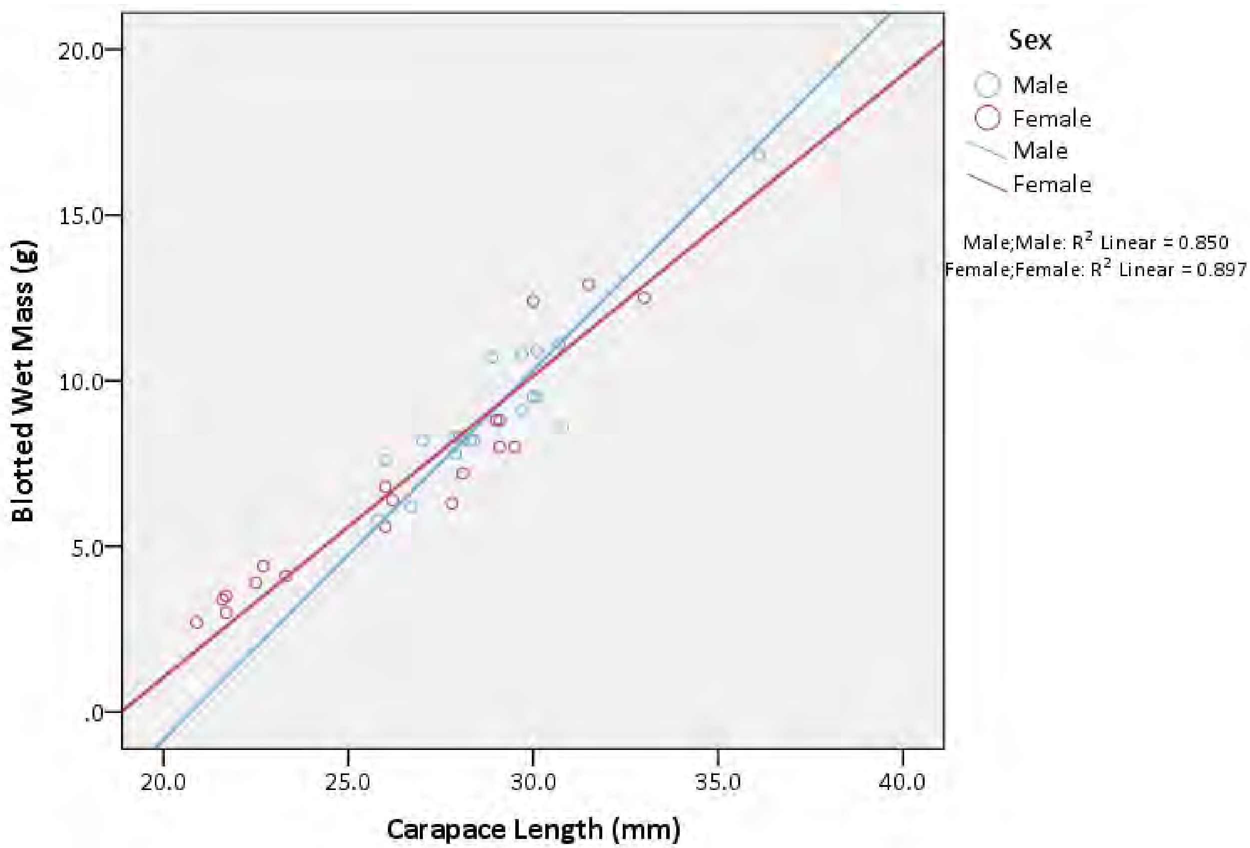


Figure 7a: The relationship between carapace length (mm) and blotted wet mass (g) in native male and female *Cambarus bartonii* (common/Appalachian brook crayfish) from the West Branch sub-basin in northcentral Pennsylvania.

Figure 7b: Relationship between Palm Width (mm) and Blotted Wet Mass (g) in *C. bartonii.*

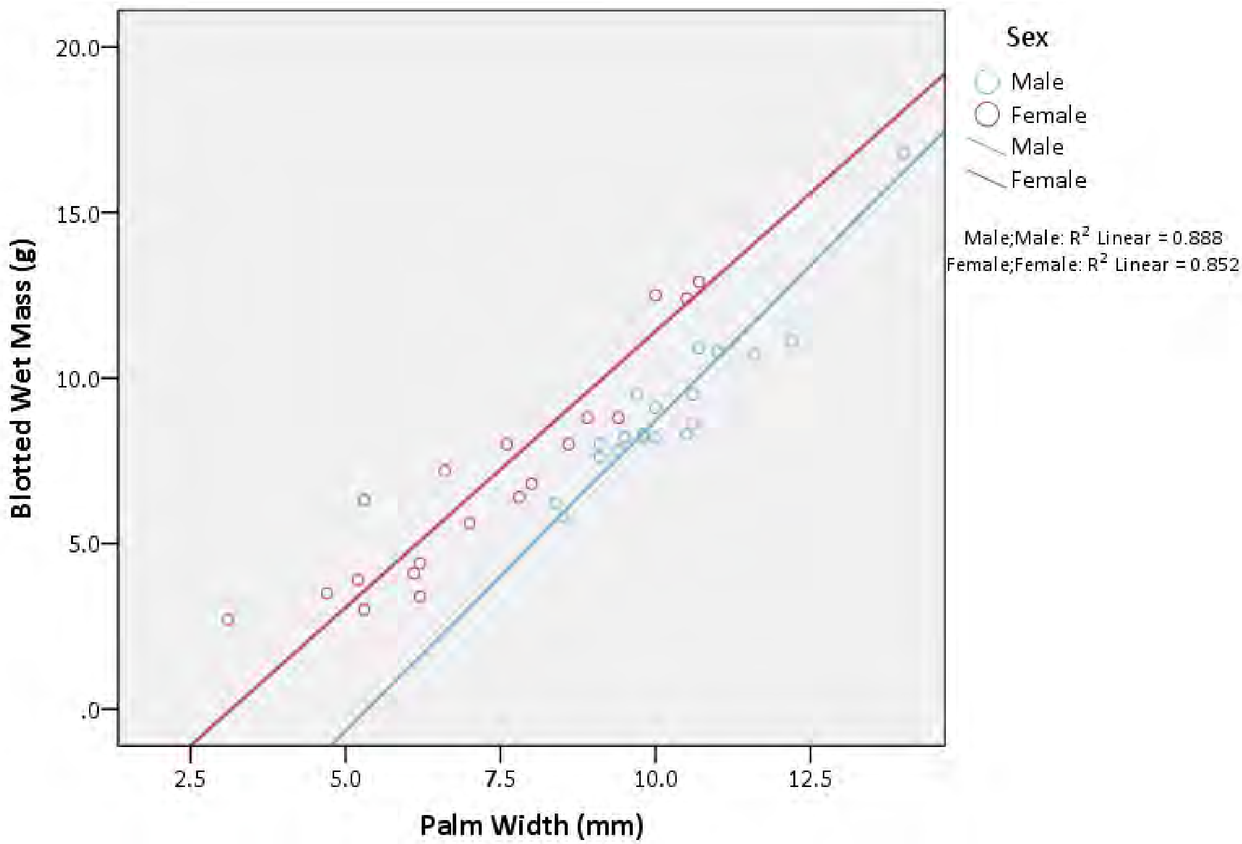


Figure 7b: The relationship between palm width (mm) and blotted wet mass (g) in native male and female *Cambarus bartonii* (Common/Appalachian brook crayfish) from the West Branch sub-basin in northcentral Pennsylvania.

Figure 8a: Relationship between Carapace Length (mm) and Blotted Wet Mass (g) in *O. obscurus.*

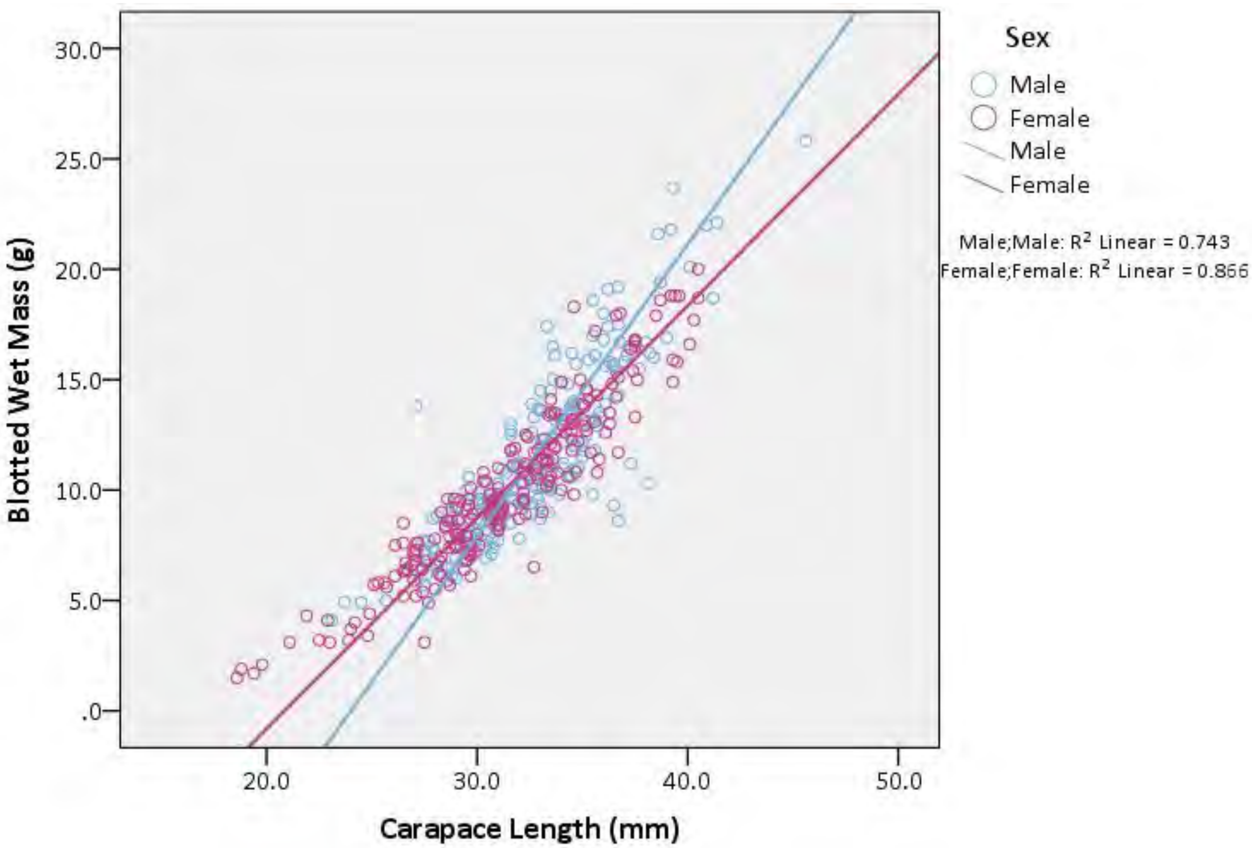


Figure 8a: The relationship between carapace length (mm) and blotted wet mass (g) in non-native male and female *Orconectes obscurus* (Allegheny crayfish) from the West Branch sub-basin in northcentral Pennsylvania.

Figure 8b: Relationship between Palm Width (mm) and Blotted Wet Mass (g) in *O. obscurus.*

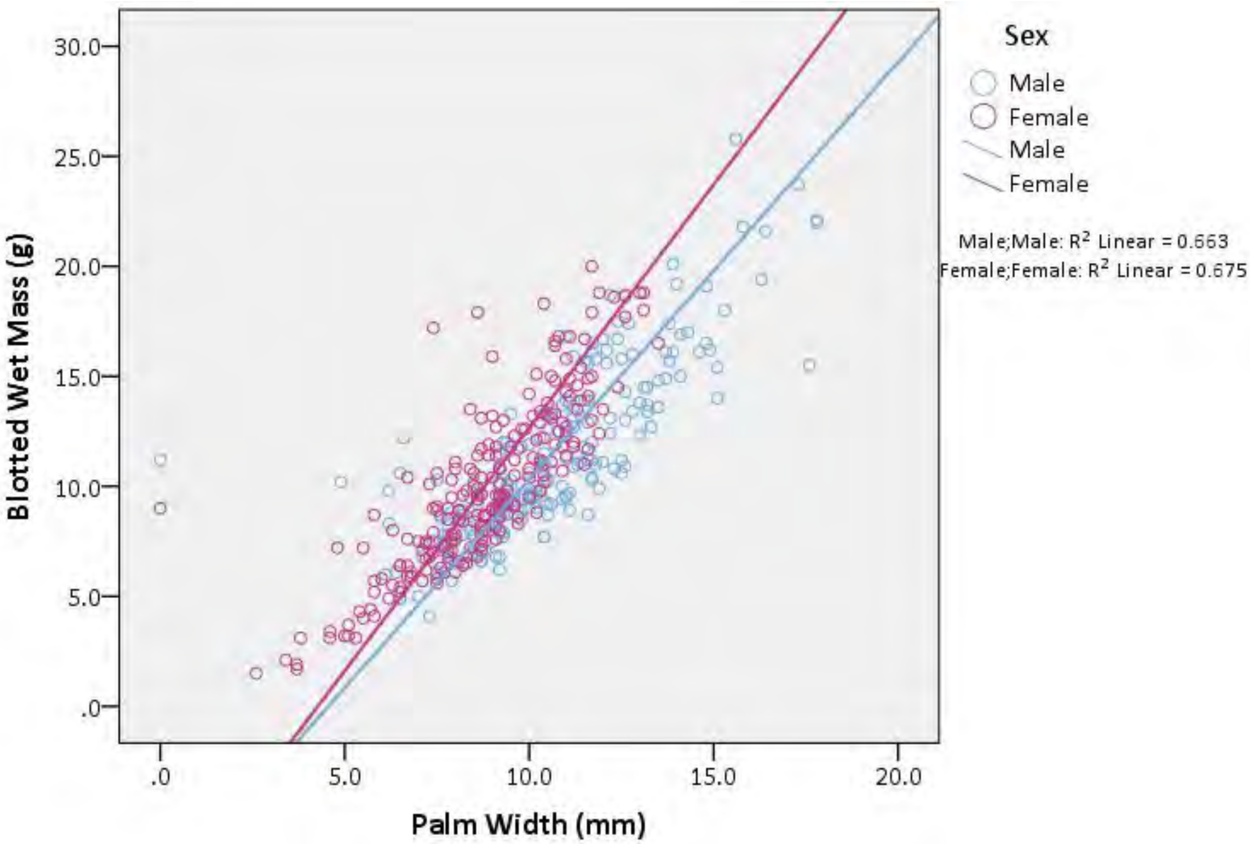


Figure 8b: The relationship between palm width (mm) and blotted wet mass (g) in non-native male and female *Orconectes obscurus* (Allegheny crayfish) from the West Branch sub-basin in northcentral Pennsylvania.

Figure 9a: Relationship between Carapace Length (mm) and Blotted Wet Mass (g) in *O. rusticus.*

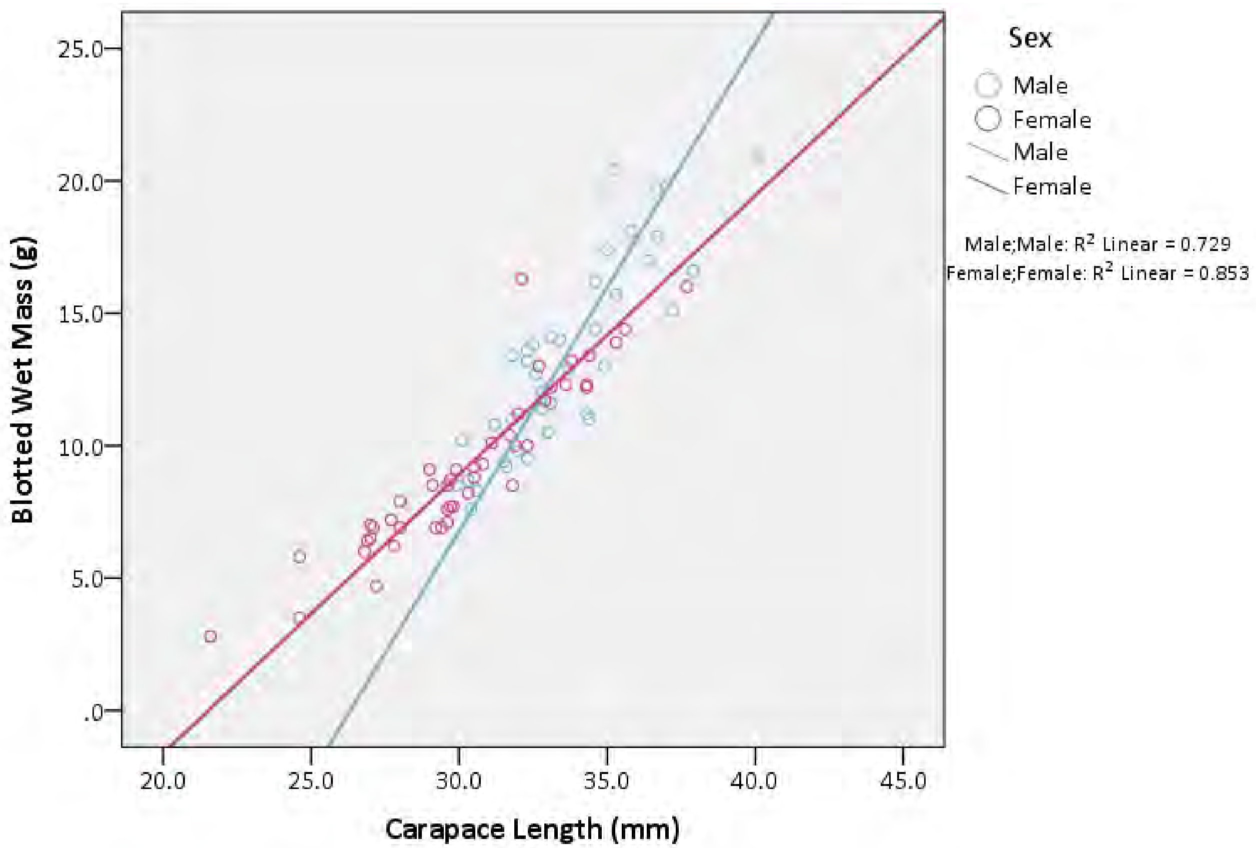


Figure 9a: The relationship between palm width (mm) and blotted wet mass (g) in non-native male and female *Orconectes rusticus* (rusty crayfish) from the West Branch sub-basin in northcentral Pennsylvania.

Figure 9b: Relationship between Blotted Wet Mass (g) and Palm Width (mm) in *O. rusticus.*

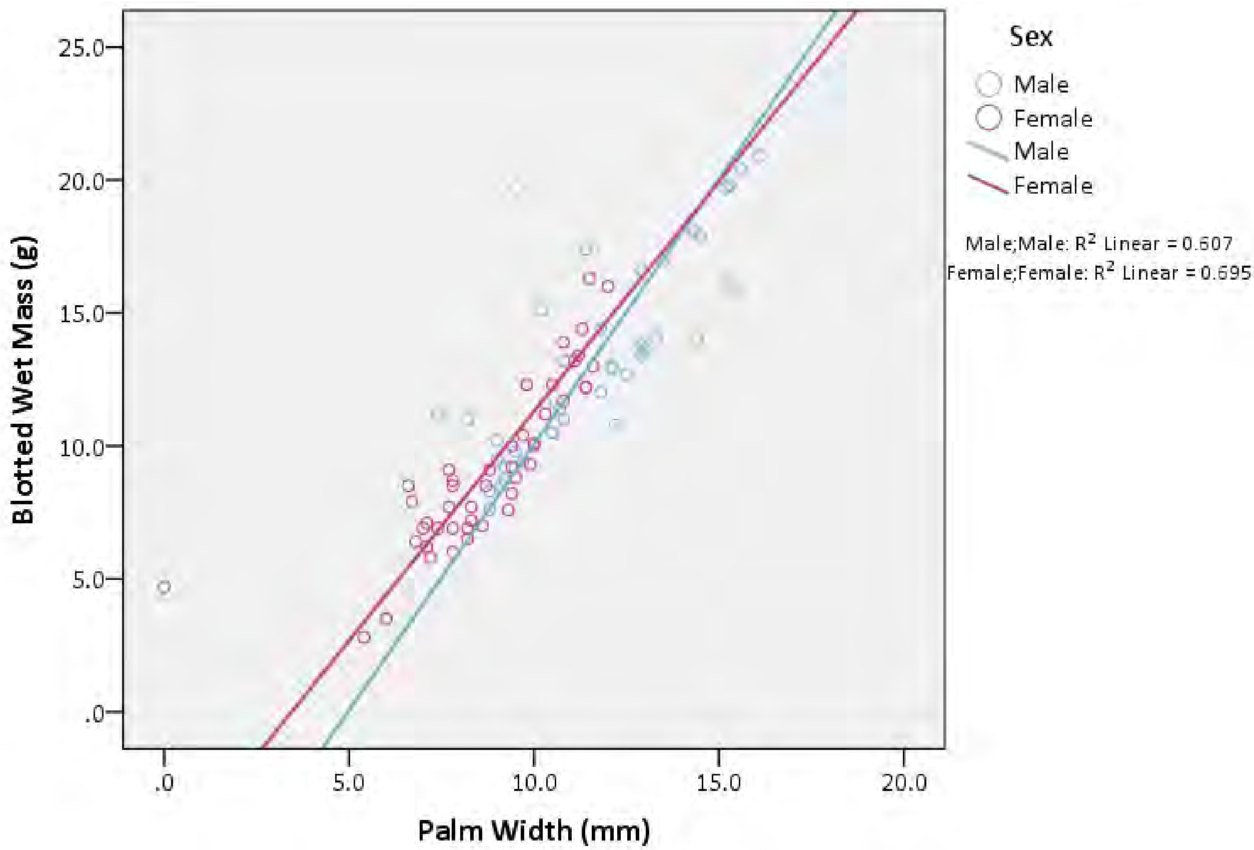


Figure 9b: The relationship between palm width (mm) and blotted wet mass (g) in non-native male and female *Orconectes rusticus* (rusty crayfish) from the West Branch sub-basin in northcentral Pennsylvania.

Table 8: Differences in Crayfish Body Size among Six Watersheds of the West Branch of the Susquehanna River, Northcentral Pennsylvania.									
Waterway	**Species**	**Sex**		**Blotted Wet Mass (g)**	**Total Body Length (mm)**	**Carapace Length (mm)**	**Areola Length (mm)**	**Areola Width (mm)**	**Palm Width (mm)**
KETTLE	***O. obscurus***	Male (n = 27)	Mean ± SD	11.7 ± 4.61	64.8 ± 7.73	31.0 ± 3.92	8.5 ± 1.33	1.4 ± 0.23	11.2 ± 2.55
			Min.	4.1	49.3	23.1	6.2	0.8	6.5
			Max.	23.7	78.7	39.3	10.7	1.8	17.3
		Female (n = 40)	Mean ± SD	10.1 ± 2.61	65.9 ± 5.23	31.2 ± 2.85	8.2 ± 0.92	1.6 ± 0.3	9.1 ± 2.0
			Min.	5.6	54.3	25.7	6.7	1.1	0
			Max.	16.7	78.0	37.5	10.8	2.2	13.5
	C. bartonii	Male (n = 10)	Mean ± SD	7.9 ± 1.1	56.9 ± 3.78	28.1 ± 1.49	8.7 ± 0.45	1.5 ± 0.21	9.6 ± 0.74
			Min.	5.8	53.0	25.8	8.1	1.1	8.4
			Max.	9.5	65.9	30.8	9.3	1.9	10.6
		Female (n = 19)	Mean ± SD	6.8 ± 3.24	54.3 ± 7.27	26.3 ± 3.76	7.4 ± 1.33	1.4 ± 0.29	7.2 ± 2.1
			Min.	2.7	42.1	20.9	5.2	1.0	3.1
			Max.	12.9	66.6	33.0	9.8	1.9	10.7
	TOTAL (n = 96)		Mean ± SD	9.6 ± 3.74	62.3 ± 7.87	29.8 ± 3.79	8.2 ± 1.17	1.5 ± 0.28	9.4 ± 2.5
			Min.	2.7	42.1	20.9	5.2	0.8	0
			Max.	23.7	78.7	39.3	10.8	2.2	17.3
PINE	***O. obscurus***	Male (n = 70)	Mean ± SD	12.3 ± 3.53	69.0 ± 5.8	33.6 ± 2.97	8.9 ± 1.27	1.5 ± 0.28	10.8 ± 2.33
			Min.	6.1	57.1	27.9	5.6	1.0	0
			Max.	22.1	84.0	41.4	14.0	2.2	17.8
		Female (n = 70)	Mean ± SD	12.8 ± 2.95	72.8 ± 5.13	34.9 ± 2.75	9.1 ± 0.96	1.8 ± 0.38	10.0 ± 1.48
			Min.	8.2	63.4	29.6	6.9	1.0	7.5
			Max.	20.0	83.5	40.5	11.2	2.6	13.1
	C. bartonii	Male (n = 10)	Mean ± SD	10.3 ± 2.61	58.6 ± 3.75	29.8 ± 2.61	8.3 ± 0.98	1.8 ± 0.43	10.9 ± 1.46
			Min.	7.6	55.3	26.0	7.0	1.3	9.1
			Max.	16.8	68.4	36.1	9.8	2.6	14.0
	TOTAL (n = 150)		Mean ± SD	12.4 ± 3.26	70.1 ± 6.44	33.9 ± 3.11	9.0 ± 1.13	1.7 ± 0.36	10.4 ±1.96
			Min.	6.1	55.3	26.0	5.6	1.0	0
			Max.	22.1	84.0	41.4	14.0	2.6	17.8
LITTLE PINE (Tributary of Pine)	***O. obscurus***	Male (n = 30)	Mean ± SD	9.7 ± 2.02	65.3 ± 4.54	32.0 ± 2.58	8.5 ± 1.08	1.8 ± 0.31	9.7 ± 2.07
			Min.	6.1	57.3	27.6	6.3	1.3	6.2
			Max.	15.5	76.4	37.7	10.3	2.6	17.6
		Female (n = 20)	Mean ± SD	7.6 ± 1.39	61.2 ± 3.69	29.0 ± 1.88	7.7 ± 0.9	1.8 ± 0.23	7.9 ± 1.25
			Min.	5.4	54.5	26.5	6.0	1.3	4.8
			Max.	10.5	67.0	32.9	9.1	2.2	10.0
	TOTAL (n = 50)		Mean ± SD	8.8 ± 2.06	63.6 ± 4.65	30.8 ± 2.73	8.1 ± 1.08	1.8 ± 0.28	9.0 ± 1.99
			Min.	5.4	54.5	26.5	6.0	1.3	4.8
			Max.	15.5	76.4	37.7	10.3	2.6	17.6
LARRY'S	***O. obscurus***	Male (n = 30)	Mean ± SD	8.5 ± 2.49	61.5 ± 4.44	29.5 ± 2.04	7.7 ± 1.11	1.6 ± 0.29	9.7 ± 2.09
			Min.	5.0	52.7	25.7	6.0	1.0	6.1
			Max.	16.2	74.0	34.5	11.2	2.2	14.9
		Female (n = 30)	Mean ± SD	8.3 ± 1.88	62.6 ± 4.4	29.6 ± 2.24	7.5 ± 0.87	1.6 ± 0.46	8.7 ± 1.5
			Min.	4.9	54.5	25.3	6.0	0.8	5.5
			Max.	12.3	71.1	33.4	8.9	2.9	11.5
	TOTAL (n = 60)		Mean ± SD	8.4 ± 2.19	62.1 ± 4.41	29.6 ± 2.12	7.6 ± 0.99	1.6 ± 0.38	9.2 ± 1.86
			Min.	4.9	52.7	25.3	6.0	0.8	5.5
			Max.	16.2	74.0	34.5	11.2	2.9	14.9
LYCOMING	***O. obscurus***	Male (n = 40)	Mean ± SD	9.8 ± 2.45	65.3 ± 4.19	32.5 ± 2.69	8.9 ±1.01	2.0 ± 0.41	8.9 ±1.64
			Min.	6.8	58.1	27.9	6.5	1.0	4.9
			Max.	20.1	80.1	40.1	12.0	2.9	13.9
		Female (n = 38)	Mean ± SD	6.9 ± 4.17	55.7 ± 9.92	26.7 ± 4.47	6.8 ± 1.44	1.7 ± 0.44	6.7 ± 2.27
			Min.	1.5	35.4	18.6	4.2	1.0	2.6
			Max.	18.6	79.4	38.7	11.1	2.7	12.3
	TOTAL (n = 78)		Mean ± SD	8.4 ± 3.68	60.6 ± 8.91	29.7 ± 4.67	7.8 ± 1.62	1.9 ± 0.45	7.8 ± 2.26
			Min.	1.5	35.4	18.6	4.2	1.0	2.6
			Max.	20.1	80.1	40.1	12.0	2.9	13.9
LOYALSOCK	***O. rusticus***	Male (n = 40)	Mean ± SD	13.5 ± 3.74	68.8 ± 4.63	33.7 ± 2.38	9.0 ± 0.95	1.8 ± 0.39	11.7 ± 2.4
			Min.	7.6	60.4	29.9	6.9	1.0	7.4
			Max.	20.9	79.2	40.1	10.7	2.9	16.1
		Female (n = 46)	Mean ± SD	9.2 ± 3.09	64.1 ± 7.28	30.3 ± 3.18	7.6 ± 1.1	1.7 ± 0.37	8.8 ± 2.15
			Min.	2.8	45.6	21.6	4.7	0.9	0
			Max.	16.3	79.0	37.7	10.3	2.4	12.0
	TOTAL (n = 86)		Mean ± SD	11.2 ± 4.02	66.3 ± 6.6	31.8 ± 3.3	8.3 ± 1.24	1.7 ± 0.39	10.1 ± 2.7
			Min.	2.8	45.6	21.6	4.7	0.9	0
			Max.	20.9	79.2	40.1	10.7	2.9	16.1
MUNCY	***O. obscurus***	Male (n = 20)	Mean ± SD	16.8 ± 3.45	74.2 ± 5.62	36.5 ± 3.18	10.1 ± 1.21	1.9 ± 0.31	14.0 ± 1.8
			Min.	10.9	66.3	32.8	8.5	1.4	10.6
			Max.	25.8	89.4	45.6	13.2	2.3	17.8
		Female (n = 20)	Mean ± SD	11.0 ± 4.35	66.9 ± 7.05	32.6 ± 3.45	8.6 ± 1.12	1.8 ± 0.33	9.0 ± 1.92
			Min.	5.2	55.6	27.1	6.8	1.2	6.5
			Max.	17.9	78.1	39.3	10.2	2.4	12.4
	TOTAL (n = 40)		Mean ± SD	13.9 ± 4.88	70.6 ± 7.3	34.5 ± 3.81	9.3 ± 1.4	1.8 ± 0.33	11.5 ±3.16
			Min.	5.2	55.6	27.1	6.8	1.2	6.5
			Max.	25.8	89.4	45.6	13.2	2.4	17.8

Table 8: A total of n = 560 crayfish from seven waterways in northcentral Pennsylvania were processed for morphometric analysis. Streams are listed in a "headwaters to mouth" order moving down the West Branch excluding Little Pine Creek (a tributary of Pine Creek). Crayfish species consisted of the non-natives *Orconectes obscurus* (Allegheny crayfish) and *O. rusticus* (rusty crayfish) and the native *Cambarus bartonii* (common/Appalachian brook crayfish).

Table 9a: Results of Games-Howell Test for Differences in Crayfish Blotted Wet Mass (BWM) Among Waterways.						
(I) Waterway	(J) Waterway	Mean Difference (I-J)	Std. Error	P (α = 0.05)	95% Confidence Interval	
					Lower Bound	Upper Bound
Kettle	Pine	-2.751*	0.4655	0.000	-4.139	-1.363
	Little Pine	0.811	0.4805	0.625	-0.626	2.248
	Larry's	1.249	0.4751	0.125	-0.171	2.669
	Lycoming	1.262	0.5649	0.283	-0.424	2.948
	Loyalsock	-1.543	0.5781	0.112	-3.267	0.182
	Muncy	-4.249*	0.8613	0.000	-6.877	-1.620
Pine	Kettle	2.751*	0.4655	0.000	1.363	4.139
	Little Pine	3.562*	0.3946	0.000	2.381	4.743
	Larry's	4.000*	0.3880	0.000	2.841	5.159
	Lycoming	4.013*	0.4939	0.000	2.535	5.490
	Loyalsock	1.208	0.5089	0.217	-0.313	2.729
	Muncy	-1.497	0.8164	0.532	-4.008	1.013
Little Pine (Tributary of Pine)	Kettle	-0.811	0.4805	0.625	-2.248	0.626
	Pine	-3.562*	0.3946	0.000	-4.743	-2.381
	Larry's	0.438	0.4059	0.933	-0.782	1.658
	Lycoming	0.451	0.5081	0.974	-1.072	1.974
	Loyalsock	-2.354*	0.5227	0.000	-3.919	-0.789
	Muncy	-5.059*	0.8251	0.000	-7.594	-2.525
Larry's	Kettle	-1.249	0.4751	0.125	-2.669	0.171
	Pine	-4.000*	0.3880	0.000	-5.159	-2.841
	Little Pine	-0.438	0.4059	0.933	-1.658	0.782
	Lycoming	0.013	0.5030	1.000	-1.494	1.520
	Loyalsock	-2.792*	0.5177	0.000	-4.341	-1.243
	Muncy	-5.498*	0.8220	0.000	-8.023	-2.972
Lycoming	Kettle	-1.262	0.5649	0.283	-2.948	0.424
	Pine	-4.013*	0.4939	0.000	-5.490	-2.535
	Little Pine	-0.451	0.5081	0.974	-1.974	1.072
	Larry's	-0.013	0.5030	1.000	-1.520	1.494
	Loyalsock	-2.805*	0.6012	0.000	-4.600	-1.010
	Muncy	-5.510*	0.8769	0.000	-8.182	-2.839
Loyalsock	Kettle	1.543	0.5781	0.112	-0.182	3.267
	Pine	-1.208	0.5089	0.217	-2.729	0.313
	Little Pine	2.354*	0.5227	0.000	0.789	3.919
	Larry's	2.792*	0.5177	0.000	1.243	4.341
	Lycoming	2.805*	0.6012	0.000	1.010	4.600
	Muncy	-2.706*	0.8854	0.048	-5.400	-0.011
Muncy	Kettle	4.249*	0.8613	0.000	1.620	6.877
	Pine	1.497	0.8164	0.532	-1.013	4.008
	Little Pine	5.059*	0.8251	0.000	2.525	7.594
	Larry's	5.498*	0.8220	0.000	2.972	8.023
	Lycoming	5.510*	0.8769	0.000	2.839	8.182
	Loyalsock	2.706*	0.8854	0.048	0.011	5.400

Table 9a: Results of a nonparametric Games-Howell Post Hoc Test (α = 0.05) comparing differences in mean blotted wet mass (BWM) among crayfish by waterway. Streams are listed in a "headwaters to mouth" order moving down the West Branch excluding Little Pine Creek (a tributary of Pine Creek). For Kettle Creek n = 96 crayfish; Pine Creek n= 150; Little Pine Creek n = 50; Larry's Creek n = 60; Lycoming Creek n = 78; Loyalsock Creek n = 86; and Muncy Creek n = 40. Significant mean differences are followed by an asterisk.

Table 9b: Results of Games-Howell Test for Differences in Crayfish Carapace Length (CL) Among Waterways.						
(I) Waterway	(J) Waterway	Mean Difference (I-J)	Std. Error	P (α = 0.05)	95% Confidence Interval	
					Lower Bound	Upper Bound
Kettle	Pine	-4.111*	0.4629	0.000	-5.493	-2.730
	Little Pine	-0.969	0.5469	0.570	-2.607	0.669
	Larry's	0.244	0.4743	0.999	-1.173	1.661
	Lycoming	0.165	0.6550	1.000	-1.793	2.123
	Loyalsock	-2.006*	0.5255	0.003	-3.573	-0.439
	Muncy	-4.716*	0.7163	0.000	-6.888	-2.544
Pine	Kettle	4.111*	0.4629	0.000	2.730	5.493
	Little Pine	3.143*	0.4623	0.000	1.750	4.535
	Larry's	4.356*	0.3736	0.000	3.240	5.471
	Lycoming	4.277*	0.5862	0.000	2.517	6.036
	Loyalsock	2.106*	0.4367	0.000	0.802	3.409
	Muncy	-0.604	0.6540	0.967	-2.607	1.399
Little Pine (Tributary of Pine)	Kettle	0.969	0.5469	0.570	-0.669	2.607
	Pine	-3.143*	0.4623	0.000	-4.535	-1.750
	Larry's	1.213	0.4736	0.151	-0.215	2.641
	Lycoming	1.134	0.6546	0.596	-0.828	3.096
	Loyalsock	-1.037	0.5249	0.436	-2.611	0.538
	Muncy	-3.747*	0.7159	0.000	-5.922	-1.572
Larry's	Kettle	-0.244	0.4743	0.999	-1.661	1.173
	Pine	-4.356*	0.3736	0.000	-5.471	-3.240
	Little Pine	-1.213	0.4736	0.151	-2.641	0.215
	Lycoming	-0.079	0.5952	1.000	-1.866	1.708
	Loyalsock	-2.250*	0.4488	0.000	-3.592	-.0908
	Muncy	-4.960*	0.6621	0.000	-6.986	-2.934
Lycoming	Kettle	-0.165	0.6550	1.000	-2.123	1.793
	Pine	-4.277*	0.5862	0.000	-6.036	-2.517
	Little Pine	-1.134	0.6546	0.596	-3.096	0.828
	Larry's	0.079	0.5952	1.000	-1.708	1.866
	Loyalsock	-2.171*	0.6368	0.015	-4.076	-0.265
	Muncy	-4.881*	0.8015	0.000	-7.296	-2.466
Loyalsock	Kettle	2.006*	0.5255	0.003	0.439	3.573
	Pine	-2.106*	0.4367	0.000	-3.409	-.0802
	Little Pine	1.037	0.5249	0.436	-0.538	2.611
	Larry's	2.250*	0.4488	0.000	0.908	3.592
	Lycoming	2.171*	0.6368	0.015	0.265	4.076
	Muncy	-2.710*	0.6996	0.004	-4.837	-0.584
Muncy	Kettle	4.716*	0.7163	0.000	2.544	6.888
	Pine	0.604	0.6540	0.967	-1.399	2.607
	Little Pine	3.747*	0.7159	0.000	1.572	5.922
	Larry's	4.960*	0.6621	0.000	2.934	6.986
	Lycoming	4.881*	0.8015	0.000	2.466	7.296
	Loyalsock	2.710*	0.6996	0.004	0.584	4.837

Table 9b: Results of a nonparametric Games-Howell Post Hoc Test (α = 0.05) comparing differences in mean carapace length (CL) among crayfish by waterway. Streams are listed in a "headwaters to mouth" order moving down the West Branch excluding Little Pine Creek (a tributary of Pine Creek). For Kettle Creek n = 96 crayfish; Pine Creek n= 150; Little Pine Creek n = 50; Larry's Creek n = 60; Lycoming Creek n = 78; Loyalsock Creek n = 86; and Muncy Creek n = 40. Significant mean differences are followed by an asterisk.

Table 9c: Results of Games-Howell Test for Differences in Crayfish Palm Width (PW) Among Waterways.						
(I) Waterway	(J) Waterway	Mean Difference (I-J)	Std. Error	P (α = 0.05)	95% Confidence Interval	
					Lower Bound	Upper Bound
Kettle	Pine	-1.057*	0.3010	0.010	-1.955	-0.158
	Little Pine	0.437	0.3796	0.911	-0.702	1.575
	Larry's	0.206	0.3505	0.997	-0.842	1.254
	Lycoming	1.553*	0.3612	0.001	0.475	2.631
	Loyalsock	-0.753	0.3873	0.455	-1.908	0.403
	Muncy	-2.102*	0.5609	0.007	-3.813	-0.392
Pine	Kettle	1.057*	0.3010	0.010	0.158	1.955
	Little Pine	1.493*	0.3235	0.000	0.516	2.471
	Larry's	1.263*	0.2887	0.001	0.396	2.129
	Lycoming	2.610*	0.3016	0.000	1.707	3.512
	Loyalsock	0.304	0.3324	0.970	-0.691	1.299
	Muncy	-1.046	0.5245	0.432	-2.660	0.569
Little Pine (Tributary of Pine)	Kettle	-0.437	0.3796	0.911	-1.575	0.702
	Pine	-1.493*	0.3235	0.000	-2.471	-0.516
	Larry's	-0.231	0.3700	0.996	-1.344	0.882
	Lycoming	1.116	0.3801	0.060	-0.025	2.257
	Loyalsock	-1.189	0.4050	0.059	-2.403	0.024
	Muncy	-2.539*	0.5732	0.001	-4.285	-0.793
Larry's	Kettle	-0.206	0.3505	0.997	-1.254	0.842
	Pine	-1.263*	0.2887	0.001	-2.129	-0.396
	Little Pine	0.231	0.3700	0.996	-0.882	1.344
	Lycoming	1.347*	0.3510	0.004	0.296	2.398
	Loyalsock	-0.959	0.3778	0.154	-2.088	0.171
	Muncy	-2.308*	0.5544	0.002	-4.002	-0.614
Lycoming	Kettle	-1.553*	0.3612	0.001	-2.631	-0.475
	Pine	-2.610*	0.3016	0.000	-3.512	-1.707
	Little Pine	-1.116	0.3801	0.060	-2.257	0.025
	Larry's	-1.347*	0.3510	0.004	-2.398	-0.296
	Loyalsock	-2.306*	0.3877	0.000	-3.463	-1.148
	Muncy	-3.655*	0.5612	0.000	-5.367	-1.943
Loyalsock	Kettle	0.753	0.3873	0.455	-0.403	1.908
	Pine	-0.304	0.3324	0.970	-1.299	0.691
	Little Pine	1.189	0.4050	0.059	-0.024	2.403
	Larry's	0.959	0.3778	0.154	-0.171	2.088
	Lycoming	2.306*	0.3877	0.000	1.148	3.463
	Muncy	-1.350	0.5783	0.244	-3.108	0.409
Muncy	Kettle	2.102*	0.5609	0.007	0.392	3.813
	Pine	1.046	0.5245	0.432	-0.569	2.660
	Little Pine	2.539*	0.5732	0.001	0.793	4.285
	Larry's	2.308*	0.5544	0.002	0.614	4.002
	Lycoming	3.655*	0.5612	0.000	1.943	5.367
	Loyalsock	1.350	0.5783	0.244	-0.409	3.108

Table 9c: Results of a nonparametric Games-Howell Post Hoc Test (α = 0.05) comparing differences in mean palm width (PW) among crayfish by waterway. Streams are listed in a "headwaters to mouth" order moving down the West Branch excluding Little Pine Creek (a tributary of Pine Creek). For Kettle Creek n = 96 crayfish; Pine Creek n= 150; Little Pine Creek n = 50; Larry's Creek n = 60; Lycoming Creek n = 78; Loyalsock Creek n = 86; and Muncy Creek n = 40. Significant mean differences are followed by an asterisk.

Demographics

As indicated in Table 10, crayfish density never exceeded 4/m^2 and was highest on Pine Creek at PC04 (stream kilometer 65.4) with a value of 3.38/m^2. At PC04 the habitat sampled consisted of ~83% loose cobble (LC) (5-20 cm long) with remainder being small boulder (SB) (20-50 cm long); the vast majority (~85%) of crayfish was caught in LC (Fig. 10). The lowest crayfish densities occurred at PC06 on Pine Creek (0.43/m^2) and KET04 on Kettle Creek (0.44/m^2). At PC06 only about 40% of samples were LC, the rest a mix of SB, MB (medium boulder, 50-100cm long) and LB (large boulder, >100 cm long); however, over 60% of all crayfish collected at that site came from LC habitat (Fig. 11). At KET04, samples consisted of a roughly equal proportion of LC and SB followed by MB and LB (Fig. 12). The highest proportion of crayfish came from LC, followed by SB, MB, and LB. The only value for both Loyalsock Creek and *O. rusticus* is taken from LOY01 (stream km 3.3) at 0.95/m^2 (Table 10).

In Larry's Creek, crayfish density increased moving upstream from LAR01, LAR02 and LAR03 (0.94/m^2, 2.02/m^2, and 3.26/m^2; Table 9). Figures 13a-c display the habitat composition of LAR01, LAR02 and LAR03. Samples at LAR01 were ~60% LC with greater than 50% of all crayfish caught in that habitat. The majority (~58%) of samples at LAR02 were of MB habitat, but this category contributed less than 50% of all crayfish captured. LAR03 had the second-highest crayfish density value in this study (3.26/m^2). Roughly 60% of the sample habitat at LAR03 was MB, followed by 30% SB and 10% LC; the proportion of crayfish caught in each habitat closely follows this trend (Fig. 13c).

No density data exists for several locations in Table 9. All sites on Lycoming Creek and Little Pine Creek as well as two sites on Loyalsock Creek (LOY02 and LOY03) were seined prior to developing a semi-quantitative protocol. The sample sites MUN02, PC07, LOY04 and LOY05 included non-quantitative hand capture to collect specimens.

Table 11 lists crayfish sex ratios by species. Although the overall M:F ratio did not differ significantly from 1:1 (1:1.05, $p = 0.311$), the ratios for *O. obscurus* and *O. rusticus* did. A total of 693 males and 815 females of *O. obscurus* yielded a male:female sex ratio of 1:1.18 ($\chi^2 = 9.87$, $p = 0.002$). For *O. rusticus*, 186 males and 122 females gave a ratio of 1:0.66 ($\chi^2 = 13.298$, $p < 0.001$). For C. bartonii, 43 males and 29 females resulted in a ratio of 1:0.67 ($\chi^2 = 2.722$, $p = 0.099$).

Combining crayfish totals by waterway (Table 12), only Kettle Creek (total) and Pine Creek (total) had a higher proportion of females (1:1.91 and 1:2.03, respectively). Both ratios were significantly different from the expected 1:1 M:F ratio (Kettle Creek $\chi 2 = 20.88$, $p < 0.001$ and Pine Creek $\chi 2 = 56.8$, $p < 0.001$). Lycoming and Loyalsock Creeks yielded significantly more male crayfish than female crayfish ($p <0.001$, both waterways), while Larry's Creek, Muncy Creek, and Little Pine Creek did not differ significantly from the expected 1:1 M:F ratio.

Further separating crayfish totals by sample site within a waterway (Table 13), sex ratios ranged from 1:6.50 M:F ($p = 0.005$) for *O. obscurus* at KET02 to 1:0.22 M:F ($p <0.001$) for *O. obscurus* at MUN02. Among site totals, only nine locations for which data is available yielded a M:F ratio that differed significantly from the expected 1:1 ratio. Some highly skewed ratios other than the two previously listed were PC03 (1:6.11, $p <0.001$), LYC02 (1:0.42, $p<0.001$) and KET01 (1:5.91, $p<0.001$).

Demographics: Tables and Figures

Table 10: Crayfish Density (org/m^2) among Sampling Locations.

Waterway	Sample Site	Species	Density (org/m^2)
Kettle Creek	KET01	*O. obscurus*	0.67
	KET02	*O. obscurus*	ND
		C. bartonii	
	KET03	*O. obscurus*	ND
	KET04	*O. obscurus*	0.44
		C. bartonii	
Larry's Creek	LAR01	*O. obscurus*	0.94
	LAR02	*O. obscurus*	2.02
	LAR03	*O. obscurus*	3.26
Lycoming Creek	LYC01	*O. obscurus*	ND
	LYC02	*O. obscurus*	ND
	LYC03	*O. obscurus*	ND
	LYC04	*O. obscurus*	ND
Muncy Creek	MUN01	*O. obscurus*	1.35
	MUN02	*O. obscurus*	ND
Pine Creek	PC01	*O. obscurus*	0.48
	PC02	*O. obscurus*	1.02
	PC03	*O. obscurus*	0.88
	PC04	*O. obscurus*	3.38
	PC05	*O. obscurus*	0.46
	PC06	*O. obscurus*	0.43
	PC07	*O. obscurus*	ND
		C. bartonii	ND
Little Pine Creek (Tributary of Pine Creek)	LPC01	*O. obscurus*	ND
	LPC02	*O. obscurus*	ND
	LPC03	*O. obscurus*	ND
Loyalsock Creek	LOY01	*O. rusticus*	0.95
	LOY02	*O. rusticus*	ND
	LOY03	*O. rusticus*	ND
	LOY04	*O. rusticus*	ND
	LOY05	*O. rusticus*	ND

Table 10: Crayfish density (org/m^2) by sampling location among the seven waterways surveyed in the West Branch sub-basin. For each waterway, sampling sites are listed in order from "mouth" to "headwaters". Species include the non-natives *Orconectes obscurus* (Allegheny crayfish) and *O. rusticus* (rusty crayfish) and the native *Cambarus bartonii* (common/Appalachian brook crayfish). ND = no density data available, *i.e.,* non-quantitative hand capture or seining was used to sample crayfish at that location.

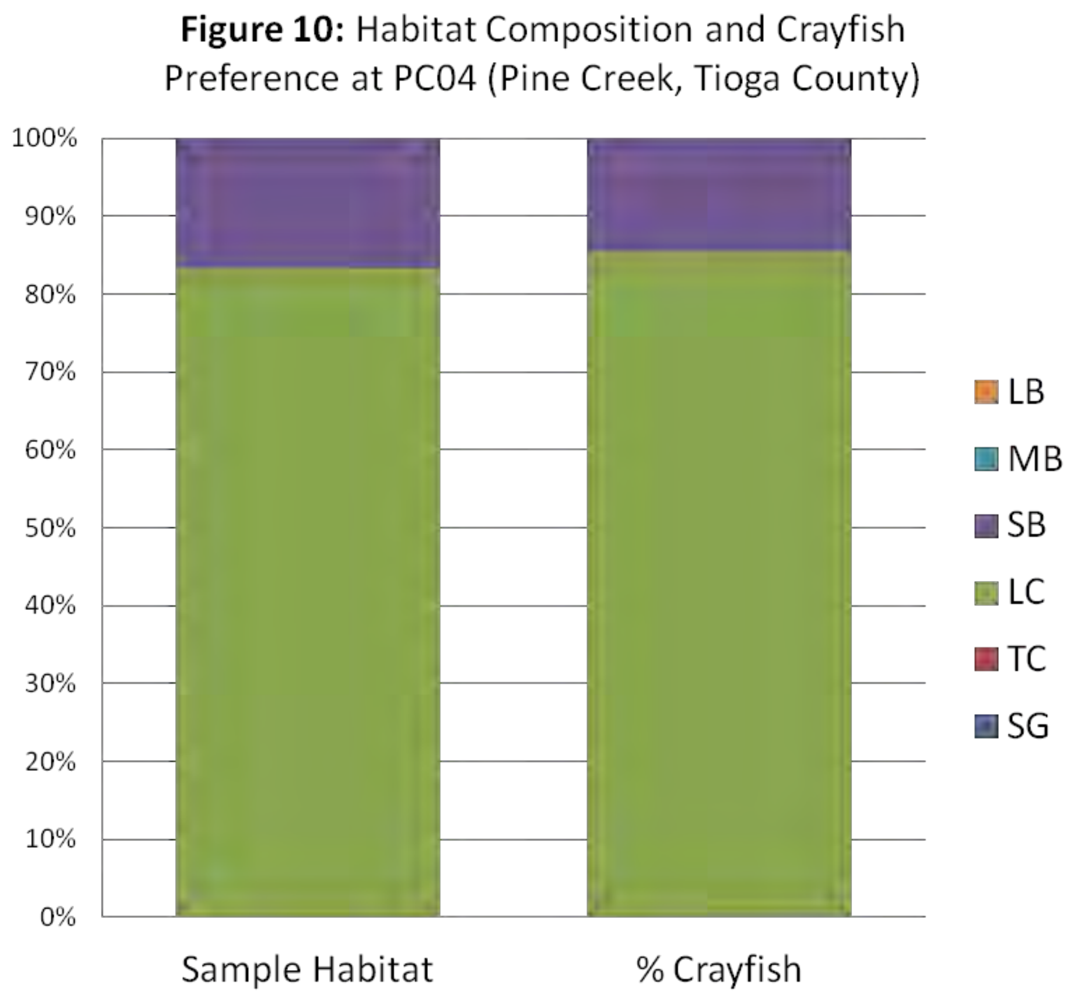


Fig. 10: Semi-quantitative seining at PC04 (stream km 65.4) on Pine Creek, a tributary of the West Branch of the Susquehanna River (Tioga County), allowed each seine sample to be classified by the predominant substrate type and the number of crayfish caught in each habitat to be recorded. PC04 yielded the highest value for crayfish density in this study (3.38/m^2). SG = sand and gravel; TC = tight cobble (5-20cm); LC = loose cobble; SB = small boulder (20-50cm); MB = medium boulder (50-100cm); LB = large boulder (>100cm).

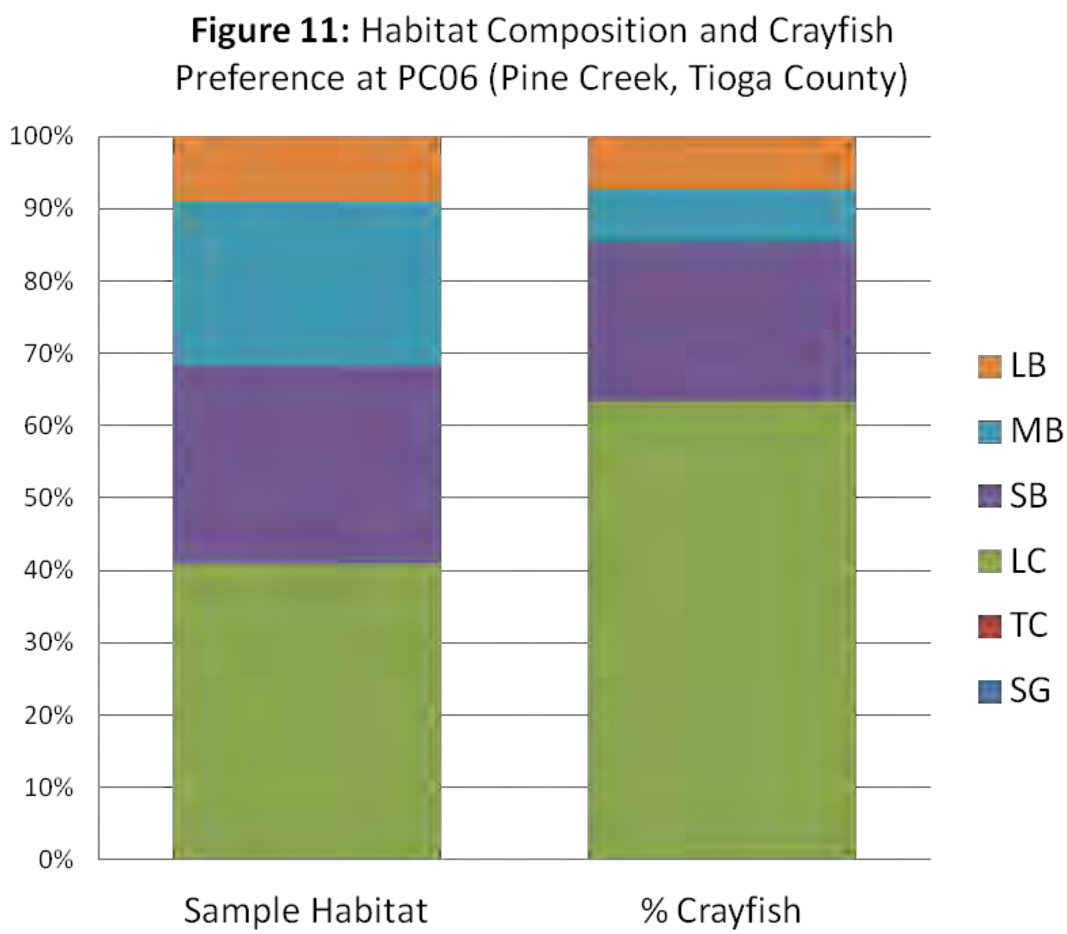


Fig. 11: Habitat composition and percentage of crayfish caught in each habitat at PC06 (stream km 100.1) on Pine Creek, a tributary of the West Branch (Tioga County). PC06 yielded the lowest value for crayfish density in this study ($0.43/m^2$). SG = sand and gravel; TC = tight cobble (5-20cm); LC = loose cobble; SB = small boulder (20-50cm); MB = medium boulder (50-100cm); LB = large boulder (>100cm).

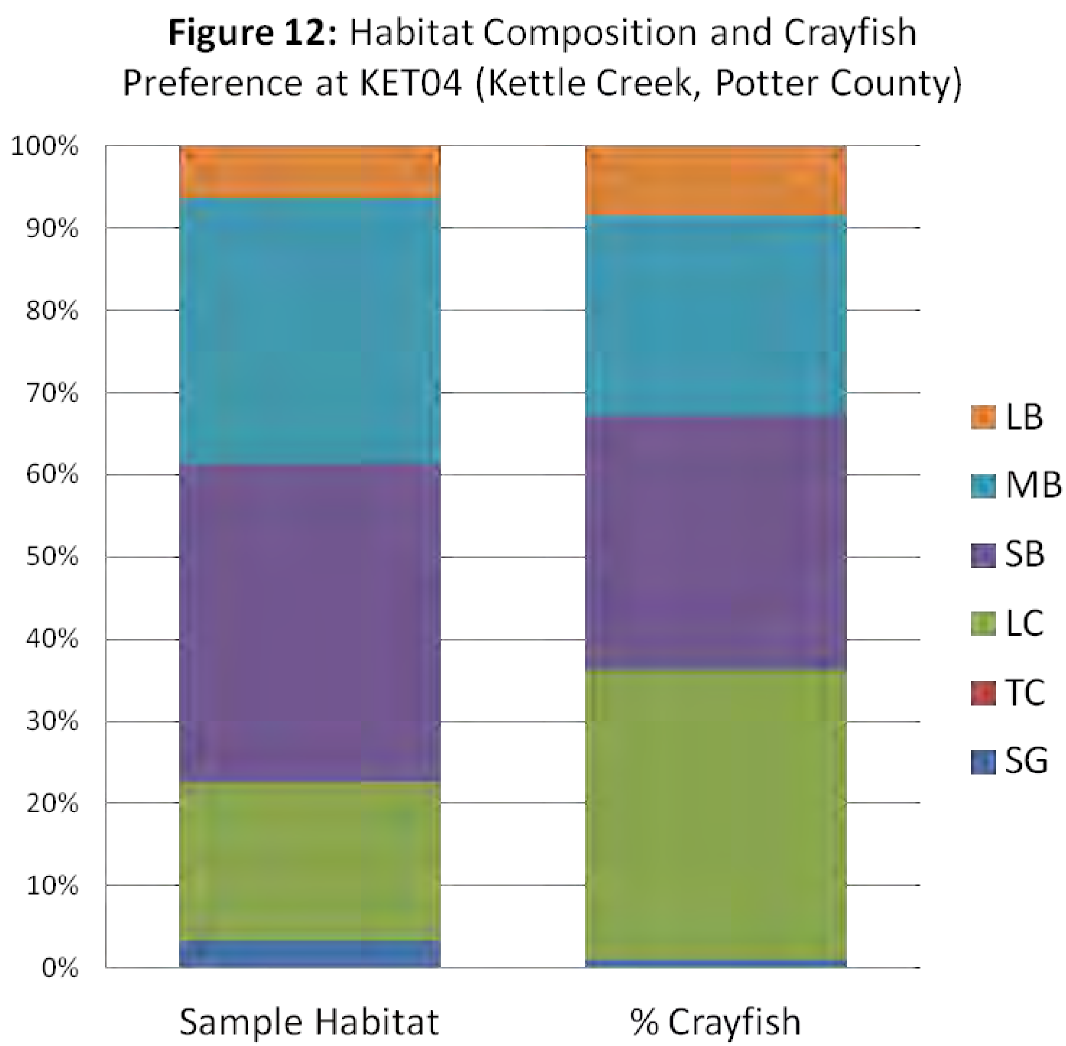
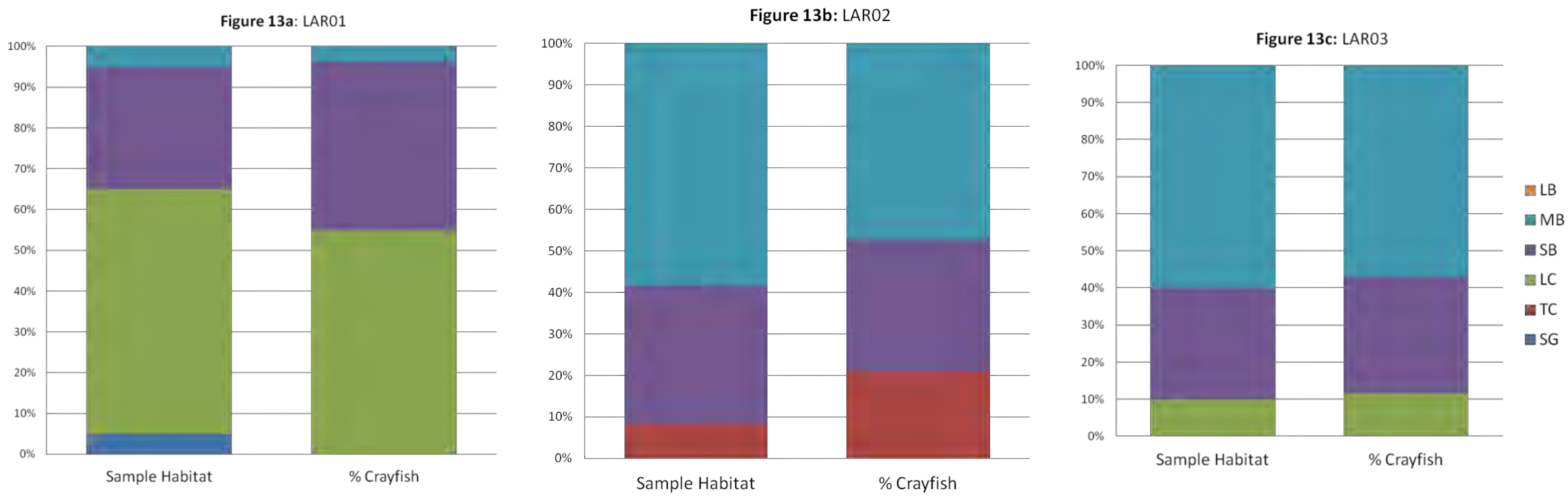


Fig. 12: Habitat composition and percentage of crayfish caught in each habitat at KET04 (stream km 46.3) on Kettle Creek, a tributary of the West Branch (Potter County). KET04 yielded the second-lowest value for crayfish density in this study ($0.44/m^2$). SG = sand and gravel; TC = tight cobble (5-20cm); LC = loose cobble; SB = small boulder (20-50cm); MB = medium boulder (50-100cm); LB = large boulder (>100cm).

Figures 13a-c: Habitat Composition and Crayfish Preference at Three Locations on Larry's Creek.



Figs. 13a-c: Habitat composition and percentage of crayfish caught in each habitat at LAR01 (stream km 0.24), LAR02 (stream km 2.9), and LAR03 (stream km 7) on Larry's Creek, a tributary of the West Branch (Lycoming County). Crayfish density increased moving upstream from LAR01 to LAR03. SG = sand and gravel; TC = tight cobble (5-20cm); LC = loose cobble; SB = small boulder (20-50cm); MB = medium boulder (50-100cm); LB = large boulder (>100cm).

Table 11: Male to Female Sex Ratios of Three Crayfish Species.

Species	# ♂	# ♀	♂ : ♀ Sex Ratio	χ^2	P (α = 0.05)
O. obscurus	693	815	1 : 1.18	9.87	0.002
O. rusticus	186	122	1 : 0.66	13.298	<0.001
C. bartonii	43	29	1 : 0.67	2.722	0.099
TOTAL	922	966	1 : 1.05	1.026	0.311

Table 11: Male:Female sex ratios by crayfish species. Species include the non-natives *Orconectes obscurus* (Allegheny crayfish) and *O. rusticus* (rusty crayfish) and the native *Cambarus bartonii* (common/Appalachian brook crayfish). A chi-square (χ2) goodness of fit test with α = 0.05 was used to determine whether ratios deviated significantly from the expected 1:1 ratio.

Table 12: Crayfish Male to Female Sex Ratios by Waterway.

Waterway	Species	# ♂	# ♀	♂ : ♀ Sex Ratio	χ^2	P ($\alpha = 0.05$)
Kettle Creek	*O. obscurus*	51	121	1 : 2.37	28.488	<0.001
	C. bartonii	23	20	1 : 0.87	0.21	0.647
	TOTAL	74	141	1 : 1.91	20.88	<0.001
Larry's Creek	*O. obscurus*	162	162	1 : 1	<0.001	>0.95
Lycoming Creek	*O. obscurus*	172	81	1 : 0.47	32.732	<0.001
	C. bartonii	0	2	1 : 2	2	0.157
	TOTAL	172	83	1 : 0.48	31.062	<0.001
Muncy Creek	*O. obscurus*	92	70	1 : 0.76	2.988	0.084
Pine Creek	*O. obscurus*	145	325	1 : 2.24	68.936	<0.001
	C. bartonii	17	4	1 : 0.24	8.048	0.005
	TOTAL	162	329	1 : 2.03	56.8	<0.001
Little Pine Creek (Tributary of Pine Creek)	*O. obscurus*	71	56	1 : 0.79	1.772	0.183
	C. bartonii	3	3	1 : 1	<0.001	>0.95
	TOTAL	74	59	1 : 0.8	1.692	0.193
Loyalsock Creek	*O. rusticus*	186	122	1 : 0.66	13.298	<0.001

Table 12: Crayfish male:female sex ratios after combining totals from all sample sites on a stream. Species include the non-native *Orconectes obscurus* (Allegheny crayfish) and *O. rusticus* (rusty crayfish) and the native *Cambarus bartonii* (common/Appalachian brook crayfish). A chi-square ($\chi 2$) goodness of fit test with $\alpha = 0.05$ was used to determine whether a ratio deviated significantly from the expected 1:1 ratio.

Table 13: Crayfish Male to Female Sex Ratios among Sampling Locations.

Waterway	Site	Species	# ♂	# ♀	♂ : ♀ Sex Ratio	χ^2	P (α = 0.05)
Kettle Creek	KET01	*O. obscurus*	11	65	1 : 5.91	38.368	<0.001
	KET02	*O. obscurus*	2	13	1 : 6.50	8.066	0.005
		C. bartonii	9	7	1 : 0.78	0.25	0.617
		TOTAL	11	20	1 : 1.82	2.612	0.106
	KET03	*O. obscurus*	ND	ND	ND	-	-
	KET04	*O. obscurus*	38	43	1 : 1.13	0.308	0.579
		C. bartonii	14	13	1 : 0.93	0.154	0.695
		TOTAL	52	56	1 : 1.08	0.148	0.7
Larry's Creek	LAR01	*O. obscurus*	33	47	1 : 1.42	2.45	0.118
	LAR02	*O. obscurus*	52	50	1 : 0.96	0.04	0.841
	LAR03	*O. obscurus*	77	65	1 : 0.84	1.014	0.314
Lycoming Creek	LYC01	*O. obscurus*	ND	ND	ND	-	-
	LYC02	*O. obscurus*	97	41	1 : 0.42	22.724	<0.001
	LYC03	*O. obscurus*	48	28	1 : 0.58	4.664	0.031
	LYC04	*O. obscurus*	27	12	1 : 0.44	5.77	0.016
		C. bartonii	0	2	1 : 2	1	0.317
		TOTAL	27	14	1: 0.52	4.122	0.042
Muncy Creek	MUN01	*O. obscurus*	49	58	1 : 1.18	0.758	0.384
	MUN02	*O. obscurus*	43	12	1 : 0.22	17.472	<0.001
Pine Creek	PC01	*O. obscurus*	18	28	1 : 1.56	2.174	0.14
	PC02	*O. obscurus*	17	38	1 : 2.24	8.018	0.005
	PC03	*O. obscurus*	9	55	1 : 6.11	33.062	<0.001
	PC04	*O. obscurus*	30	140	1 : 4.61	71.176	<0.001
	PC05	*O. obscurus*	25	21	1 : 0.84	0.348	0.555
	PC06	*O. obscurus*	17	24	1 : 1.41	1.196	0.274
	PC07	*O. obscurus*	29	19	1 : 0.66	2.084	0.149
		C. bartonii	17	4	1 : 0.24	8.048	0.005
		TOTAL	46	23	1: 0.5	7.666	0.006
Little Pine Creek (Tributary of Pine Creek)	LPC01	*O. obscurus*	ND	ND	1 : 0.47	-	-
	LPC02	*O. obscurus*	49	44	1 : 0.9	0.268	0.605
		C. bartonii	2	0	1	2	0.157
		TOTAL	51	44	1: 0.86	0.516	0.473
	LPC03	*O. obscurus*	22	12	1 : 0.55	2.942	0.086
		C. bartonii	1	3	1 : 3	1	0.317
		TOTAL	23	15	1 :0.65	1.684	0.194
Loyalsock Creek	LOY01	*O. rusticus*	38	29	1 : 0.76	1.208	0.272
	LOY02	*O. rusticus*	53	42	1 : 0.44	1.274	0.259
	LOY03	*O. rusticus*	29	29	1 : 1.00	<0.001	>0.95
	LOY04	*O. rusticus*	13	6	1 : 0.46	2.578	0.108
	LOY05	*O. rusticus*	53	16	1 : 0.30	19.84	<0.001

Table 13: Crayfish male:female sex ratios by sample sites along each waterway. Species include the non-native *Orconectes obscurus* (Allegheny crayfish) and *O. rusticus* (rusty crayfish) and the native *Cambarus bartonii* (common/Appalachian brook crayfish). A chi-square (χ2) goodness of fit test with α = 0.05 was used to determine whether a ratio deviated significantly from the expected 1:1 ratio. ND = no data available for male and female crayfish totals.

DISCUSSION

Species Occurrence

I found the native Appalachian brook crayfish *C. bartonii* was no longer the only crayfish species in northcentral Pennsylvania; furthermore, this species is now largely limited to the upper reaches of the tributaries surveyed where it occurs in relatively low numbers (Fig. 1). This species has been replaced by the non-native *O. obscurus* (Allegheny crayfish), currently the most predominant and widespread species throughout the study area. *O. obscurus* was present in six out of the seven streams surveyed and was found at 23 out of 28, or 82%, of the samples sites (Fig. 1). This species accounted for 1508 of 1888 (79.9%) total crayfish captured and 435 of 560 (77.68%) total crayfish processed in the lab (Fig. 5).

O. obscurus is native to Pennsylvania, but not to the Susquehanna River Basin. At the time of Ortmann's (1906) study, the Allegheny crayfish naturally occurred only in extreme western Pennsylvania within the upper Ohio River drainage, and Ortmann's (1906) eastern-most collecting points were in the headwaters of the Genesee River in northern Potter County and in Wills Creek, Bedford County (which was a suspected human introduction). The region including both *O.obscurus* and *C. bartonii* was mapped as the northwestern corner of the state, including northern Jefferson, northern Armstrong, northeastern Butler, northern Mercer, Crawford, Venango, Clarion, Forest, western Elk, northern Potter, Mckean, Warren, and southeastern Erie Counties; a small area is also marked in eastern Westmoreland County (Ortmann 1906). Ortmann (1906) could not find evidence of *O. obscurus* occurring in the region surveyed in the present study (southern Potter, Clinton, Lycoming, Sullivan and Tioga Counties) or in the entire West Branch sub-basin during his surveys. He hypothesized that the geographic divide between the Ohio drainage in the west and the Susquehanna and Potomac drainages in the east presented a natural barrier to the further eastern expansion of *O. obscurus* in Pennsylvania. Evidence for the species' restriction to extreme western Pennsylvania was supported by its absence from West Branch watersheds immediately east of the divide, including Sinnemahoning Creek (Cameron County), Clearfield Creek (Cambria County), the West Branch itself and several other tributaries in Clearfield, Cameron, Cambria, and Indiana Counties.

The now widespread distribution of *O. obscurus* in northcentral Pennsylvania indicates a major species expansion has occurred in the region since the early 20th century, but the occurrence of *O. obscurus* in West Branch watersheds in 2012 is not entirely unexpected. The divide mapped by Ortmann (1906) is very close to the headwaters of Pine Creek in Potter County as well as the Sinnemahoning Creek in Cameron and Clinton Counties (including the Bennett Branch and First Fork). The proximity of this so-called barrier to such major east/southeast flowing streams makes it likely that the Allegheny crayfish would eventually migrate into the West Branch sub-basin. Mangan and Stocker (2011) had also recently identified *O. obscurus* at several sites along the adjacent, more eastern North Branch watershed, and Leib *et al.* (2011a) documented *O. obscurus* in the Delaware drainage in extreme southeastern Pennsylvania. The presence of this species in the study area and in nearby and more eastern watersheds suggest it was inevitable that *O. obscurus* would cross the barrier described by Ortmann (1906).

Determining exactly where or how the Allegheny crayfish penetrated the six West Branch watersheds in this study is challenging considering the present pervasiveness of this species and a general lack of historical information since the early 1900s. A combination of natural migration from the Ohio River drainage in the west facilitated by multiple points of human introduction is a likely scenario. A gradual migration eastward is certainly likely when one considers the physiological adaptations most crayfish posses to survive short periods out of water (Reynolds and Souty-Grosset 2012); this could allow the species, over time, to travel via a network of smaller streams and rivers and reach the West Branch. All seven streams in the present study are additionally large enough for permanent year round flow and feature at least one public roadway paralleling most of the stream's length, making these waterways ideal for fishing. Anglers using *O. obscurus* as bait in any of these streams could have accidently introduced individuals at any time since Ortmann's (1906) surveys.

Examining the crayfish species composition above and below barriers to upstream migration is potentially helpful in establishing a tentative timeline for the Allegheny crayfish's expansion. Two such examples in the study area are the Alvin R. Bush Dam on lower Kettle Creek and Little Pine Lake on Little Pine Creek. As previously mentioned the 50 m high Alvin R. Bush Dam was constructed in 1961 and forms Kettle Creek Lake in Kettle Creek State Park (Pennsylvania State University 2001). Examining the crayfish species collected at points downstream and upstream of the lake (Fig. 1), *O. obscurus* is present at all locations. Assuming the introduction of the Allegheny crayfish to Kettle Creek occurred via upstream migration from West Branch, the species must have already been present in these upstream reaches prior to 1961. Little Pine Lake is a 94 acre impoundment 6.4 km upstream from the confluence of Little Pine Creek and Pine Creek at Waterville, and the dam has been in place since 1949 (Schwarz 2005). All sample sites on this stream yielded *O. obscurus* as well, suggesting that any upstream migration from lower Pine Creek into Little Pine Creek must have occurred prior to 1949, pushing back the estimated arrival of the Allegheny crayfish in the region since Little Pine Creek is further east than Kettle Creek.

Sources published around this time cannot confirm these tentative timelines. For example, Crocker and Barr (1968) make no indication that the distribution of *O. obscurus* in the eastern United States had changed since Ortmann (1906). This is likely because no new survey work in northcentral Pennsylvania had occurred since that time; as a result, Crocker and Barr's (1968) distribution for *O. obscurus* in Pennsylvania is clearly based on Ortmann's (1906) and is not particularly helpful. Even more recent sources such as Jezerinac *et al.* (1995) still appear to use Ortmann's (1906) distribution, as only extreme western Pennsylvania is included in the species' range. However, introduction by human agency could have easily occurred in the years before or after the installation of these two dams, making the presence of these structures significant only if upstream migration of *O. obscurus* is assumed to be the primary means of introduction into these watersheds. Due to the popularity of both watersheds (and lakes) among anglers (Pennsylvania State University 2001, Schwarz 2005, personal observation), coupled with a lack of historical crayfish data from the region, basing the arrival of *O. obscurus* around the construction dates of these dams may or may not be valid. Ample opportunity has existed for anglers to bring in exotic crayfish as live bait and release them at various points above and below these structures.

The relationship between *C. bartonii* and *O. obscurus* is certainly different in northcentral Pennsylvania in comparison to other regions where the species are considered native and have historically co-occurred together. For example, Jezerinac *et al.* (1995) report *C. bartonii* is the most common crayfish associate of *O. obscurus* in the Potomac River drainage; Ortmann (1906) reported the same for a large portion of western Pennsylvania. These associations probably reflect niche partitioning which has developed over time. Niche partitioning is adaptive because two species with similar habitat requirements can minimize costly direct competition for food, shelter and space by utilizing the environment in slightly different ways (Crocker and Barr 1968). When *O. obscurus* was first introduced in northcentral Pennsylvania, only one such niche would have existed for crayfish and would have been occupied by the established resident populations of *C. bartonii*. *C. bartonii* apparently could not withstand direct competition with *O. obscurus*, and has subsequently been in decline ever since. This would explain why in most of the streams surveyed *O. obscurus* was the predominant species (yellow diamonds in Fig. 1) and why at virtually all locations yielding *C. bartonii* and *O. obscurus, O. obscurus* was present in significantly greater numbers. The future status of *C. bartonii* in all seven streams surveyed is questionable at best in the presence of the dominant Allegheny crayfish.

The fact that *C. bartonii* was the least frequently caught species in the study area (Fig. 1) could reflect a number of different things besides displacement by *O. obscurus*. Both sampling methods (seining and hand capture) could have been biased toward non-burrowing species such as *O. obscurus* and *O. rusticus*, as neither method is ideal for sampling the shoreline burrows of *C. bartonii* (Ortmann 1906). However, *C. bartonii* is considered a facultative burrower at best, mostly digging during periods of extreme drought to avoid desiccation. In streams with a permanent water supply, such as all the waterways surveyed in this study, this species is known to behave more like the surface dwelling crayfish of genus *Orconectes* and occupy shallow depressions under stones (Ortmann 1906, Crocker and Barr 1968). Burrowing behavior has not been observed at all by Jezerinac *et al.* (1995) in the West Virginia portion of *C. bartonii*'s range or in Ontario, Canada by Crocker and Barr (1968).

Low sample sizes of *C. bartonii* may also be attributed to this species persisting mostly in smaller, rougher feeder streams not surveyed, as the species is noted for its ability to live in small, high gradient mountain streams that *O. obscurus* tends to avoid (Ortmann 1906, Jezerinac *et al.* 1995). However, none of the three sample sites where *C. bartonii* was collected in relatively high numbers (KET02, KET04, and PC07) were rough or high gradient compared to other, more downstream locations where *C. bartonii* was not collected. This suggests that *C. bartonii* could have, and probably did, occupy these similar downstream reaches in greater numbers before the arrival of *O. obscurus*. While *C. bartonii* can survive in small mountain streams, it probably fared just as well (if not better) in the larger tributaries that were surveyed; the low numbers observed in the present study therefore likely reflect displacement and not habitat preference. Two of the three sites that yielded *C. bartonii* (PC07 on Pine Creek and KET04 on Kettle Creek) were also relatively high in their respective watersheds and were the most upstream localities sampled on those tributaries (stream km 104.1 and 46.3, respectively). PC07 and KET04 could therefore represent transitional stages between a reach where native *C. bartonii* still persists in relatively high numbers and a reach dominated by expanding populations of non-native *O. obscurus*.

The invasive, non-native rusty crayfish (*O. rusticus*) was only found in Loyalsock Creek (Lycoming and Sullivan Counties). The native range of *O. rusticus* is the Ohio River drainage, where it occupies lakes and rivers through much of Ohio and parts of Indiana and Kentucky (Phillips 2010). Ortmann (1906) briefly mentions reports of the species near the extreme western portion of Lake Erie in Ohio, but was doubtful *O. rusticus* occurred in Pennsylvania. It was first discovered in Wisconsin in the 1970s, and has since rapidly spread across several mid-western and eastern states from Maine south to Tennessee and west to New Mexico as well as throughout southern Canada (Phillips 2010, Olden *et al*. 2006). The spread of this species is mostly attributed to human introductions (*i.e.,* anglers using it as live bait; the biological supply trade in schools and universities) (Reynolds and Souty-Grosset 2012, Phillips 2010). It has been illegal to possess, sell, transport or culture *O. rusticus* in Pennsylvania since 2005 (Lieb *et al.* 2011b), but human introduction likely explains the presence of the rusty crayfish in Loyalsock Creek. The introduction is probably recent (within the last decade) considering its confinement to Loyalsock Creek despite the species' reputation for rapid dispersal (Reynolds and Souty-Grosset 2012). Multiple introduction points are also possible given the rusty crayfish's presence from the mouth up to at least the confluence with Little Loyalsock Creek at LOY05 (Fig.1). The introduction probably did not originate from the West Branch itself (*i.e.,* up-river migration) since Muncy Creek, another major West Branch tributary approximately 13 km downriver from Loyalsock Creek, did not yield any individuals of *O. rusticus.* Up-river migration into the Loyalsock was a possibility because the rusty crayfish has been documented at the confluence of the West Branch and the main stem of the Susquehanna River at Northumberland (Mangan and Stocker 2011), and it is unknown how long the species had previously been there. It is possible that *O. rusticus* is indeed making its way up the West Branch from the main stem, but simply has not made it as far as Muncy Creek. Up-river migration of *O. rusticus* from Loyalsock Creek is likely temporarily hindered by the presence of a dam about 6 km up-river from the mouth at the city of Williamsport. However, given that the dam is a fairly popular local fishing spot (personal observation), it will likely not remain a barrier for long if anglers use *O. rusticus* as live bait above the dam. If human introduction occurs in the West Branch at any point above this structure, other tributaries will likely be invaded over time as well, such as Lycoming Creek situated just 3 km up-river from the dam. Muncy Creek, which drains into the West Branch about 13 km downriver from Loyalsock Creek, should be considered at risk of invasion by *O. rusticus* within the next few years. Continued monitoring of such West Branch tributaries is imperative.

The marked absence of *O. obscurus* from Loyalsock Creek likely reflects exclusion by *O. rusticus*. Since *O. obscurus* was present in significant numbers in all other streams surveyed, including Lycoming Creek (approximately 9 km up-river) and Muncy Creek (about 13 km downriver), there is no reason to believe that this species never occurred in Loyalsock Creek as well. This is supported by the fact that *O. obscurus* occurs in several tributaries of Loyalsock Creek (personal observation). Based on Ortmann's (1906) records, the original predominant crayfish species of Loyalsock Creek was likely *C. bartonii*. *C. bartonii* may have eventually been displaced by *O. obscurus* (the situation currently observed in the other six streams surveyed), at which point both species were subsequently excluded with the arrival of *O. rusticus*. Interestingly, whereas no *O. obscurus* were captured in Loyalsock Creek, stray individuals of *C. bartonii* persisted at certain sites. Both species appear to be inferior competitors to *O. rusticus* in this waterway, but *C. bartonii* may have a slight advantage that explains its presence. As a species observed to burrow in Pennsylvania (Ortmann 1906, personal observation), *C. bartonii* perhaps has the option of avoiding some direct competition with *O. rusticus* for shelter and space by constructing shoreline burrows. While this may be suboptimal habitat for *C. bartonii*, it could allow the species to persist where *O. obscurus*, strictly a surface dwelling species like *O. rusticus* (Ortmann 1906, Crocker and Barr 1968), cannot. Shelter from predators is known to be critical for crayfish survival (Reynolds and Souty-Grosset 2012), and the larger, more aggressive *O. rusticus* likely drove out *O. obscurus* from Loyalsock Creek itself by occupying these shelters. Populations of *O. obscurus* may still persist in the smaller, rougher tributaries of the Loyalsock because *O. rusticus* appears to favor larger, lower gradient streams and rivers (Turner 1926, Lieb *et al.* 2011a). This displacement of both a native (*C. bartonii*) and a previously established non-native (*O. obscurus*) by *O. rusticus* is similar to what Hill and Lodge (1999) observed in northern Wisconsin lakes. The authors attributed superior competitive ability and lower mortality rates from predation as major factors allowing the invasive *O. rusticus* to replace both *O. virilis* (a native) and *O. propinquus* (a previous invader).

The presence of *O. rusticus* in additional West Branch watersheds will likely prove highly detrimental. As previously discussed, the rusty crayfish has decreased crayfish biodiversity in the invaded Loyalsock Creek, similar to other parts of the invaded range in mid-western and other eastern states as well as Canada (Hill and Lodge 1999, Reynolds and Souty-Grosset 2012). Introduced rusty crayfish are believed to force native species from shelter, leaving them exposed to predators (Hill and Lodge 1999). This is supported by Klocker and Strayer (2004), who found *O. rusticus* was clearly dominant over a native species (*O. limosus*) in a laboratory setting by winning a majority of aggression trials and obtaining shelter more frequently. Typical of many other exotic nuisance species, *O. rusticus* also appears to tolerate a wide range of habitats and environmental conditions (especially temperature), increasing the probability of individuals surviving in new waterways. *O. rusticus* additionally has a higher metabolic rate compared to other crayfish species of similar size, which leads to the decimation of aquatic plant beds and macroinvertebrate prey (Phillips 2010, Reynolds and Souty-Grosset 2012). Introduced individuals of *O. rusticus* are known to grow significantly larger and faster than individuals from native populations, possibly because bait bucket introductions select for high aggressiveness and associated high growth rates due to the stressful conditions of the bait trade (Pintor and Sih 2009). Another advantage that rusty crayfish have over most other congeners in temperature regions is the ability to lay eggs at lower temperatures, encouraging greater population growth as the *O. rusticus* offspring develop earlier and exploit food and shelter resources first (Phillips 2010).

Hybridization is one additional consequence of an invasion by the rusty crayfish, and one which may already be occurring within the study area. While genetic testing of individuals to confirm hybridization is not available at this time, the individuals captured near the mouth of Loyalsock Creek at LOY01 (purple diamond, Fig. 1) are suspected *O. rusticus x obscurus* hybrids based on several observed atypical physical characteristics. For example, common features of rusty crayfish include a rust colored spot on either side of the carapace, a black band near the fingertips of each cheiliped, mandibles with a smooth cusp, and a rostrum with slightly concave margins (Crocker and Barr 1968, Roger and Hill 2008, Phillips 2010). While one or more of these characteristics are not always present in certain *O. rusticus* populations (Phillips 2010), the crayfish captured at LOY01 exhibited such a confusing mixture of so-called "rusty" and "non-rusty" traits that species identification was difficult and uncertain (see Appendix I Table 22 for a detailed description of processed individuals). It is hypothesized that Allegheny crayfish originating from the West Branch are migrating up Loyalsock Creek and breeding with rusty crayfish that occur near the mouth; specifically, *O. rusticus* females are likely producing hybrids with *O. obscurus* males based on previous studies (*e.g,.* Perry *et al.* 2001). This is detrimental because hybrid crayfish are typically superior competitors to both parent species and eventually cause the elimination of genetically pure individuals from the population (Perry *et al.* 2001, Reynolds and Souty-Grosset 2012). Hybridization coupled with direct competition may be preventing Allegheny crayfish from the West Branch from further penetrating Loyalsock Creek at this time. Future work should include crayfish sampling at additional points closer to the mouth of Loyalsock Creek as well as upstream of LOY01 to get a clearer picture of this phenomenon.

Hybridization is not likely to directly impact native *C. bartonii* populations in this region, as mating between *Orconectes* and *Cambarus* species has not been reported and is unlikely due to mechanical isolation (Ortmann 1906). Crayfish of the genus *Cambarus* possess distinctly different gonopod morphology than those in genus *Orconectes*. The terminal elements of gonopods in genus *Cambarus* are robust and curved, sometimes forming a 90 degree angle, whereas in genus *Orconectes* the terminal elements tend to be straight and fine-tipped (Ortmann 1906; Jezerinac *et al.* 1995; Rogers and Hill 2008). These differences probably prevent mating between the two species.

It is interesting to note that the estimated arrival of the rusty crayfish in Loyalsock Creek within the past decade or so roughly corresponds with the eastern hellbender's documented decline in that tributary over the past six years (Petokas *et al.* 2012). While a direct cause and effect relationship cannot be determined between the two events, it is quite possible that the arrival of the larger, more aggressive *O. rusticus* in Loyalsock Creek stressed these animals further and contributed to their rapid disappearance in the waterway. Hellbenders swallow prey whole when feeding (Nickerson and Mays 1973), and perhaps struggle more with rusty crayfish, eventually avoiding this species even as it becomes more numerous and replaces native crayfishes. Therefore, hellbenders in Loyalsock Creek may be slowly starving in the midst of exploding *O. rusticus* populations, leaving them more susceptible to pathogens, pollution or other stressors that increase mortality. Although hellbenders will ingest *O. rusticus* (personal observation), their preference for one crayfish species over another has not been studied. It is logical that these animals would prefer native crayfish species (*i.e., C. bartonii*) over large, aggressive exotics like the rusty crayfish. Containing the rusty crayfish and preventing it from spreading into other West Branch tributaries that harbor hellbender populations is therefore critical.

Morphometry

Male crayfish of all three species were larger on average than females (Table 2). This was expected and agrees with the literature because females do not undergo ecdysis and grow when *in berry* or when young are present, leading to a size disparity between the sexes (Turner 1926; Crocker and Barr 1968; Reynolds and Souty-Grosset 2012). However, total body length (TBL) was not significantly higher for males in *C. bartonii* and *O. obscurus* ($p = 0.068$ and $p = 0.110$, respectively) compared to females. It is interesting that carapace length (CL) was significantly larger in the males of all three species (Tables 3-5) but TBL (which includes CL) was not except in *O. rusticus*. This suggests that the relative proportion of carapace to abdomen in the males and females of these species is different but ultimately does not affect total body length. Female crayfish of all three species in this study had a smaller average carapace length than males, and the carapace made up a slightly smaller proportion of the total body length. In *O. obscurus*, carapace length was x = 48.7% of the total body length in males and x = 47.8% of the total body length in females; in *O. rusticus* x = 49% in males and x = 47.2% in females; and in *C. bartonii* x = 50% in males and x = 48.4% in females (Table 2). Abdomen length was therefore slightly longer in females. Abdominal width, not measured in this study, is known to be slightly larger in female crayfish compared to males, presumably reflecting the female abdomen’s role in incubation of eggs and young (Reynolds and Souty-Grosset 2012); perhaps a difference in abdominal length occurs also.

Significant body size differences were observed between the three crayfish species collected in this study, but crayfish sex appears be an important factor in these differences. Figures 6a-c graph the distribution of BWM, CL, and palm width (PW) among the three species, and a trend is seen in males where the average for each measurement decreases moving from *O. rusticus*, *O. obscurus* and *C. bartonii*; however, this pattern does not hold for female crayfish. Comparing just two species at a time, in Table 6b male *C. bartonii* and *O. obscurus* differed significantly in three measurements (BWM with $p = 0.014$; TBL with $p = 0.000$; CL with $p = 0.000$) while female *C. bartonii* had significantly lower means for all measurements compared to female Allegheny crayfish (Table 7b). Between the two *Orconectes* species, there is a stark lack of significant differences between female Allegheny and female rusty crayfish except for areola length (AL) ($p = 0.027$) (Table 7c) but as shown in Table 6c male *O. rusticus* had significantly higher means for all body measurements compared to male *O. obscurus*. Both sexes of *C. bartonii* appear to be significantly smaller in body size compared to *O. rusticus* (Tables 6a and 7a), as both male and female rusty crayfish had five significantly higher averages (the non-significant difference in males and females being areola width (AW) and AL, respectively). These results suggest that *C. bartonii* and *O. rusticus* differ the most in body size in both sexes (*i.e.*, five out of six body measurements significantly different). Male *C. bartonii* and *O. obscurus* appear to differ less than their female counterparts, while female *O. obscurus* and *O. rusticus* are very similar in body size but males are not.

Invasive rusty crayfish that are significantly larger (*i.e.*, longer body length and heavier mass) compared to the native *C. bartonii* could potentially throw local ecosystems out of balance, as larger crayfish logically require more space for shelter and more food. *O. rusticus* is known to attain a larger size in invaded ranges and in effect remove itself from food webs because few native predators can consume it (Pintor and Sih 2009, Reynolds and Souty-Grosset 2012). Therefore, invasive *O. rusticus* often have few controls on their population growth. High population densities coupled with higher metabolic demands in *O. rusticus* leads to habitat degradation as food sources are decimated (Phillips 2010, Reynolds and Souty-Grosset 2012). *O. obscurus* may also prove to be detrimental in these regards, as both male and female Allegheny crayfish were significantly larger in terms of BWM, TBL and CL compared to their *C. bartonii* counterparts. Literature on the behavior and impacts of *O. obscurus* as an invasive species is scarce, as the species is more commonly discussed as a native in the eastern United States. *O. rusticus* is probably the more aggressive and harmful of the two species in this region, based on the apparent exclusion of established *O. obscurus* populations in Loyalsock Creek.

Chelae are especially important in male crayfish, as they are used for a variety of activities related to competition with rivals and courtship rituals (Reynolds and Souty-Grosset 2012). *O. rusticus* had a significantly higher average PW than *C. bartonii* ($p = 0.01$), but *O. obscurus* did not ($p = 0.593$). Between male *O. rusticus* and *O. obscurus*, the difference between average PW was also significant ($p = 0.005$), being higher in *O. rusticus*. Average PW probably did not differ significantly between *O. obscurus* and *C. bartonii* (despite *C. bartonii* having significantly lower averages for both TBL and CL with $p < 0.001$) by virtue of its burrowing tendencies. As Reynolds and Souty-Grosset (2012) note, burrowing crayfish species like *C. bartonii* must possess relatively robust chelae to facilitate digging, whereas crayfishes in the genus *Orconectes* typically do not burrow and tend to have longer and more slender chelae. Male rusty crayfish chelae were significantly wider compared to those of *C. bartonii* despite not being a burrowing species. A possible explanation is that *O. rusticus* is simply able to attain a greater size, with proportionally larger (wider) chelae, than *C. bartonii*. This is supported by both the literature (*e.g.*, Phillips 2010) and the fact that comparisons of the six body measurements between males of these two species yielded a relatively high number of significant differences (five), with *O. rusticus* having higher average values (Table 6a). The significant difference between chelae width in *O. rusticus* and *O. obscurus* was again likely reflective of the rusty crayfish's overall larger size and proportionately larger chelae compared to many other species of crayfish (Phillips 2010). Significantly wider chelae in *O. rusticus* could pose problems to native predators (such as the eastern hellbender) when attempting to consume it, perhaps increasing the risk of injury to the predator and leading to rejection of the prey item. Reduced predation would eventually lead to higher *O. rusticus* population densities.

Body weight and length appear to be strongly related in crayfish. When blotted wet mass was plotted with carapace length and palm width for each species and sex, r^2 values ranged from 0.607 to 0.897, indicating that crayfish mass is a good predictor of certain aspects of body size. BWM was not as strongly related to PW in *O. rusticus* males and females (r^2 = 0.607 and r^2 = 0.695, respectively), and relatively low r^2 values were also obtained when *O. obscurus* BWM was plotted with PW (r^2 = 0.663 and r^2 = 0.675 for males and females, respectively). These values contrast with those obtained for *C. bartonii*, which were r^2 = 0.888 for males and r^2 = 0.852 for females (Fig. 7b). A reason why palm width showed a generally weaker relationship to blotted wet mass in the two *Orconectes* species may concern their invasive status and the fact that crayfish constantly lose and regenerate their chelae (Turner 1926, Crocker and Barr 1968). As two invasive species, the Allegheny and rusty crayfish are presumably more aggressive and may engage in behaviors that cause them to lose their chelae more frequently compared to the native *C. bartonii*. Therefore, even as these crayfish continue to molt and increase regularly in mass and body size, chelae size may be temporarily independent of body size and thus different than expected depending on the stage of regeneration. The lower r^2 values relative to *C. bartonii* may also be interpreted as *O. rusticus* and *O. obscurus* simply having larger, wider chelae than predicted for their body size, which has been documented in the rusty crayfish (*e.g.,* Phillips 2010). Both explanations (*i.e.,* losing chelae more frequently and possessing larger than predicted chelae) could apply to the males within both species as well. Male crayfish compete with one another for females (potentially losing one or both chelae) and are known to have slightly larger chelae than females of the same species and body size (Reynolds and Souty-Grosset 2012). These characteristics could weaken the relationship between BWM and PW, resulting in lower r^2 values for males. BWM was a slightly stronger predictor of CL in all three species, with the lowest value of $r^2 = 0.729$ for male *O. rusticus* (Fig. 9a). This is logical because CL does not fluctuate like PW does due to regeneration. When a crayfish molts, mass as well as body length should increase proportionately, which is supported by the relatively high r^2 values obtained for each species and sex.

Areola width also did not differ significantly between male and female *O. obscurus* and *O. rusticus* with $p = 0.368$ and $p = 0.062$, respectively (Tables 4 and 5). Areola width is a very small measurement that stays relatively fixed within certain species of crayfish (*e.g.*, Rogers and Hill 2008), making these results typical. Areola length versus width is also a fairly static ratio that distinguishes between crayfish species (e.g. Rogers and Hill 2008, Jezerinac *et al.* 1995). Comparing areola length versus width among the three species, the two *Orconectes* species both had a length x = 4.9 times longer than the width, compared to x = 5.3 times longer than wide in *C. bartonii*. For *O. obscurus* and *O. rusticus*, Jezerinac *et al.* (1995) report the areola as x = 5.6 times longer than wide and x = 7.4 times longer than wide, respectively; the authors' mean value for *C. bartonii* was identical to the one obtained in the present study (x = 5.3 times longer than wide). The higher means obtained for *O. obscurus* and *O. rusticus* by Jezerinac *et al.* (1995) may reflect differences in measuring technique or perhaps regional variation in crayfish populations.

The relationship between waterway and crayfish size (Table 8) appears to be relatively weak, as statistical analyses of crayfish BWM, CL and PW by waterway (Tables 9a-c) did not reveal many discernible patterns. However, crayfish from Muncy Creek and Pine Creek had no significant differences in BWM, CL or PW and were thus relatively similar in these aspects of body size. Crayfish collected from both of these streams also had significantly higher averages for BWM, CL and PW compared to all other streams except for Loyalsock Creek. The only non-significant difference between crayfish from Muncy Creek and Loyalsock Creek was PW. Two non-significant differences existed between Pine Creek and Loyalsock Creek crayfish (BWM and PW). Examining the crayfish species composition of each stream, the only species collected from Muncy Creek was *O. obscurus*, and the vast majority (n =140 out of 150 total, Table 8) of crayfish from Pine Creek were also *O. obscurus* with a small fraction of *C. bartonii*. The only species collected from Loyalsock Creek was *O. rusticus*. It appears that *O. obscurus* collected from Muncy and Pine Creeks were simply larger relative to those in the other streams surveyed so as to be comparable in size to *O. rusticus* from Loyalsock Creek, a species generally expected to be larger in size (*e.g.,* Phillips 2010, Reynolds and Souty-Grosset 2012) and to have significantly higher means for all body measurements. Crayfish from Kettle Creek and Larry's Creek also appeared to also be relatively similar, with no significant differences in the three analyzed body measurements. Crayfish from Pine Creek and its tributary Little Pine Creek differed significantly in all three measurements, perhaps reflecting a significant difference in stream size and characteristics (*e.g.,* channel width, water depth, substrate size, predators) that makes both streams favorable for different size classes of crayfish. Sample size differences could also be an issue, as n = 150 for Pine and n = 50 for Little Pine (Table 8).

Several sources of error are possible in these morphometric analyses. First, the six body measurements taken are by no means comprehensive, and at best give a rough picture of the animal's dimensions. Future work should include additional morphometric measurements such as palm length, abdominal width, length of male gonopods, etc. to be able to generate more meaningful comparisons. The sample sizes of each species were also very different (Table 2), with *C. bartonii* making up a very small portion (3.8%) of all processed crayfish; perhaps these individuals were not typical specimens of the species. Another possible flaw in this analysis was that both form I and form II males were combined together; form I males have been shown to be generally larger than form II individuals (e.g. Jezerinac *et al.* 1995). This could have acted to decrease the size difference between males and females. Additionally, a measurement such as areola width consists of such a small area on the animal that it is of limited use in morphometric analyses attempting to compare body size differences. As previously discussed, areola length and width are primarily used for calculating a length-to-width ratio that distinguishes between certain species of crayfish (Rogers and Hill 2008); they are unlikely to have any implications for aquatic ecosystem health and function.

Demographics

Crayfish density varied considerably among sample sites, ranging from 0.43 to 3.38/m^2 (Table 10). This range is comparable to the 0.4 to 6.2/m^2 densities reported by Taylor and Soucek (2010) in Illinois streams for *O. propinquus* and *O. rusticus*. Interestingly, the minimum and maximum density values both came from Pine Creek (PC06 and PC04, respectively), separated by a distance of approximately 34.8 stream kilometers. Crayfish densities are known to show remarkable variability, ranging from less than $1/m^2$ to more than $70/m^2$ (Reynolds and Souty-Grosset 2012), so this is perhaps not entirely unusual.

A major factor known to influence crayfish density is the substrate, with crayfish generally preferring complex habitats that offer shelter from predators (Reynolds and Souty-Grosset 2012, Taylor and Soucek 2010, Crocker and Barr 1968). The size of the substrates may also be important, as Taylor and Soucek (2010) found crayfish densities were typically highest in riffle habitats with small cobble and gravel substrates. Comparing the habitat composition of the sites that yielded the two highest crayfish densities (PC04 and LAR03, respectively) with the sites that yielded the two lowest crayfish densities (PC06 and KET04, respectively), there are several unexpected differences. For example, sample habitats from PC06 and KET04 actually appeared to be more heterogeneous in terms of the number of different substrate types sampled (Figs. 11 and 12). PC06 had four substrate classes (loose cobble (LC), small boulder (SB), medium boulder (MB) and large boulder (LB)) and KET04 had five (sand and gravel (SG), LC, SB, MB and LB); PC04 (Fig. 10) had only two (LC and SB) and LAR03 (Fig. 13c) had three (LC, SB and MB). Most (~85%) of the samples at PC04 were also of just one substrate (LC), and roughly the same proportion of crayfish was caught in that habitat. LAR03 consisted of mostly MB, and most crayfish were also caught from this habitat. Examining the low-density sites, the PC06 samples were about 40% LC with over 60% of all crayfish caught at this habitat type; the largest proportion of KET04 samples was SB (almost 40%) followed by MB and LC, with LC yielding the highest percentage of crayfish (~35%).

While habitat heterogeneity was not greater at the two high crayfish density sites, the substrate that yielded the highest proportion of crayfish at three of these four sites was the smaller loose cobble, which agrees with Taylor and Soucek's (2010) findings. Loose cobble is likely the preferred habitat when larger boulders are absent or scarce due to the number of small interstitial spaces in which crayfish can hide and escape from larger predators. If boulders happen to be few and far between, then these habitats are also likely the sites of fierce competition between the few largest crayfish in the population, which smaller individuals may wish to avoid by utilizing cobbles instead (Crocker and Barr 1968). It is notable that at KET04 loose cobble made up a relatively small portion of habitat sampled, and yet that substrate yielded the highest proportion of crayfish; most crayfish at PC06 were also caught in loose cobble despite that category accounting for less than half of the total.

The only clear pattern in crayfish density was observed in Larry's Creek, where density increased moving upstream (see Table 10). This could perhaps reflect a gradual increase in habitat heterogeneity, as crayfish prefer and are more likely to survive in areas with a complex mixture of substrate types (Crocker and Barr 1968, Reynolds and Souty-Grosset 2012). The primary bottom substrates at LAR01 were LC followed by SB and a very small proportion of MB (Fig. 13a); LAR02 was mostly MB followed by SB and a small proportion of TC (Fig. 13b); and LAR03 was characterized by mostly MB with SB and LC (Fig. 13c). The general increase in substrate size moving upstream (*i.e.,* mostly LC at LAR01 versus mostly MB at LAR02 and LAR03) may indicate better shelter for crayfish that leads to higher densities, as crayfish tended to be caught in the largest substrate size class at each site except for LAR01. Since LAR01 did not have an abundance of small or medium boulders, most crayfish probably preferred to utilize the various interstitial spaces between the more common loose cobble. This strategy possibly avoids costly direct competition over scarce boulders and/or avoids sharing of space and resources.

Although only one density value was obtained for rusty crayfish in Loyalsock Creek ($0.95/m^2$, Table 10), this value was relatively low compared to population densities known to exceed $21/m^2$ for the species (Klocker and Strayer 2004). Because this site was in close proximity to the mouth and yielded possible *O. rusticus x obscurus* hybrids, this single value may not be representative. However, the unexpectedly low value may be also attributed to the catastrophic flooding that occurred in Loyalsock Creek from Tropical Storm Lee in September 2011. A 500 year flood event, stream flow on 8 September 2011 at the USGS Loyalsockville gauge (41.325000° N, 76.912778° W) was recorded at 69,100 cfs, which was significantly higher compared to 47,900 cfs seen during Hurricane Agnes, another major flooding event in 1972. Water levels on the creek crested at 19.78 ft, surpassing the flood stage level of 12 ft (NOAA 2011). Many crayfish may have been flushed out of Loyalsock Creek or simply did not survive this event, resulting in a lower than expected density value. It is therefore possible that rusty crayfish populations are still recovering a year later. Although no density data exists for rusty crayfish in Loyalsock Creek prior to the flooding, future work should include monitoring rusty crayfish density in Loyalsock Creek to verify increases as the stream continues to rebound from Tropical Storm Lee.

Crayfish density has several implications for the health of aquatic ecosystems. As discussed earlier, crayfish exert great control over other macroinvertebrate taxa through predation, competition, habitat alteration, etc. (Flinders and Magoulick 2007). Extremely low crayfish densities may decrease biodiversity in a section of stream, as one or two commoner organisms are not kept in check by a keystone species (*i.e.,* crayfish). At the other end of the spectrum, extremely high densities of crayfish may lead to the decimation of aquatic plant beds and other essential food items for other aquatic taxa, also decreasing biodiversity (Flinders and

Magoulick 2007, Phillips 2010). The range of crayfish densities in the present study (0.43 to 3.38/m^2) does not appear to be unusually high or low in comparison to other published values (e.g. Taylor and Soucek 2010, Reynolds and Souty-Grosset 2012), and does not appear to be concerning for the streams surveyed.

The overall crayfish sex ratio of 1:1.05 M:F did not differ significantly from the expected 1:1 ratio. When broken down by species, waterway and sample site, however, some significant deviations from a1:1 ratio were observed. For example, the sex ratio of 1: 1.18 M:F obtained for *O. obscurus* in this study differed significantly from a 1:1 ratio ($\chi^2 = 9.87$, $p = 0.002$) , as did the 1 : 0.66 M:F ratio for *O. rusticus* ($\chi^2 = 13.298$, $p <0.001$) (Table 11). Looking at demographics by waterway, in most (four out of seven) streams the sex ratio differed significantly from the expected 1:1 ratio, with two waterways yielding more males (Loyalsock and Lycoming) and two yielding more females (Kettle and Pine) (Table 12). Larry's Creek yielded an exact 1:1 M:F ratio, while Muncy Creek and Little Pine Creek did not differ significantly from a 1:1 ratio. The crayfish sex ratios among samples sites (Table 13) also show a remarkably wide range of variation, with few clear patterns in the proportion of one sex moving up or down a stream. One interesting pattern was noted among the Larry's Creek collection sites, however. Moving upstream from LAR01 to LAR03, the number of males to females steadily increased, although none of the ratios were statistically different from 1:1. Of the sex ratios that differed significantly from 1:1, most yielded less females than males (*i.e.*, LYC02, LYC03, LYC04 (total), MUN02, PC07 (total) and LOY05).

Most other studies report crayfish sex ratios close to 1:1 M:F. For example, Jezerinac *et al.* (1995) reported remarkably similar M:F sex ratios in West Virginia for the three crayfish species collected in the present study, with 1:1.1 for *O. obscurus*, 1.1:1 for *O. rusticus* and 1.1:1

for *C. bartonii*; none of these ratios were statistically significant from a 1:1 ratio. Fielder (1972) studied populations of *O. obscurus, O. sanborni* and *O. propinquus* in Ohio rivers using a similar sampling technique to the one in the present study (minnow seine), and also found that sex ratios of all three species were approximately 1:1 most of the time. Fielder (1972) noted that this ratio varied on a seasonal basis, in which twice as many males were captured in the fall and spring for one species. The author suggested that this pattern corresponded with a particular stage of the crayfish life cycle when ovigerous females remain hidden and are harder to capture. In the present study, crayfish were captured within a fairly narrow window of time (June through August) during one season (summer), so it is unknown if similar seasonal variations also occur in crayfish sex ratios in northcentral Pennsylvania. From June to August in Pennsylvania, most juvenile crayfish are probably leaving the female, allowing her to molt and feed for the first time in weeks; meanwhile, most adult males are molting to form I to prepare for a new breeding season that begins in late summer and early fall (Jezerinac *et al.* 1995, Reynolds and Souty-Grosset 2012).

The summer months therefore appear to be a time of transition for both sexes, and potentially a time when mortality increases if individuals are not well hidden. Molting represents a very dangerous time for a crayfish, as it must seek shelter and wait for the new exoskeleton to harden; until this occurs, it is easy prey for many other animals (Turner 1926, Crocker and Barr 1968, Reynolds and Souty-Grosset 2012). As a result, the majority of individuals of one sex may have been hidden in places inaccessible to the sampling methods used, depending on the molting cycle.

Sites such as PC03 on Pine Creek (1:6.11 M:F) or KET01 on Kettle Creek (1:5.91 M:F) (Table 13) that yielded highly skewed sex ratios of mostly female crayfish may reflect a more long term situation. Perhaps the relative size and behavior of males at these locations led to their increased mortality and removal from the population. Male crayfish are generally larger than females as discussed in the present study and in the literature (e.g. Jezerinac *et al.* 1995, Reynolds and Souty-Grosset 2012); perhaps male crayfish were more conspicuous and/or desirable to predators at these locations. Male crayfish also compete aggressively with each other for shelter, food and mates (Reynolds and Souty-Grosset 2012), possibly reducing their overall fitness and increasing their mortality. This is plausible considering most sampling took place shortly before most crayfish breed in temperate states such as Pennsylvania, when intense competition among males could have been occurring (Jezerinac *et al.* 1995). At sites with highly skewed ratios in favor of male crayfish (*e.g.*, MUN02 M:F 1:0.22), perhaps the energy associated with incubating and caring for offspring also acts to decrease female fitness and survival.

These highly skewed sex ratios may change throughout the course of a year as observed by Fielder (1972) and may thus reflect temporary situations that pose no serious long term ecological consequences. Future work could include sampling these same locations at different times of year that correspond to different stages of the crayfish life cycle. If equalization between the sexes was not observed at other times of the year, then skewed sex ratios may represent a more permanent situation that could impact the future reproduction of these crayfish populations. For example, too few females among an excess of males (e.g. MUN02 M:F 1:0.22; Table 13) may act to slow population growth. The variable sex ratios could also be attributed to the sampling methods used; perhaps one sex prefers a specific microhabitat within a reach that seining and hand capture could not adequately sample, such as the bottom of extremely deep pools.

CONCLUSIONS

The crayfish species composition of northcentral Pennsylvania has undergone great changes since Ortmann's (1906) surveys of the West Branch sub-basin. The native Appalachian brook crayfish (*C. bartonii*) is no longer the only crayfish species occurring in this region, as the non-native Allegheny crayfish (*O. obscurus*) is now widespread across the study area and the rusty crayfish (*O. rusticus*) has overtaken Loyalsock Creek in Lycoming and Sullivan Counties. *C. bartonii* has been largely displaced by these species and is limited mostly to the upper reaches of the streams surveyed, where it occurs in much lower numbers. Significant differences exist in certain aspects of body size between the three species, which could prove detrimental to the area's aquatic food webs as the generally smaller *C. bartonii* is replaced by the two larger *Orconectes* species. Crayfish densities were relatively low among the sample sites but not out of the ordinary range for temperate region streams. Crayfish male to female sex ratios were found to vary widely among each waterway and sample site with few clear patterns; however, the overall sex ratio was not significantly different from the expected 1:1 ratio. It is unknown whether the highly skewed sex ratios observed among sampling locations represent biases in sampling methods, natural fluctuations linked to the crayfish life cycle, or a more permanent and potentially serious situation for future crayfish population growth.

Two non-native crayfish species are now present in the same streams where the eastern hellbender salamander (*Cryptobranchus alleganiensis*), a species of special concern, also occurs. Thus, continued monitoring of crayfish populations is an imperative. It is unknown at this time whether the hellbender, a top crayfish predator in the region, has been or will be negatively affected by this major shift in crayfish species composition, but the spread of rusty crayfish into tributaries besides Loyalsock Creek would likely be devastating for those other aquatic ecosystems and hellbender populations. The results of this study will hopefully provide valuable baseline information for the continued study and monitoring of crayfish.

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APPENDIX I

Crayfish Morphometric Data

Table 1: Crayfish Morphometric Measurements for KET01 (Kettle Creek).

Site: KET01 (Bush Dam Outlet) **Date Sampled:** 08/14/2012 **Species:** *Orconectes obscurus*								*Males Form I unless noted otherwise
Male/Female Specimen #	**Blotted Wet Mass (g)**	**Total Body Length (mm)**	**Carapace Length (mm)**	**Areola Length (mm)**	**Areola Width (mm)**	**Palm Width (mm)**	**Eggs/Young (♀)**	**Notes**
M01	4.9	50.5	23.7	6.6	0.8	6.5	-	
M02	4.9	50.7	24.5	6.8	1.6	6.5	-	
M03	9.6	61.6	30.1	7.1	1.2	11.0	-	
M04	8.2	60.2	29.3	8.0	1.4	8.9	-	
M05	8.6	59.1	28.7	7.5	1.6	9.2	-	
M06	4.1	49.3	23.1	6.2	1.3	7.3	-	
M07	7.0	56.1	27.1	6.4	1.3	8.7	-	
F01	11.4	70.6	33.3	8.2	1.5	11.0	N	
F02	12.5	68.6	32.3	8.7	1.6	10.8	N	
F03	11.2	69.0	32.3	7.7	1.5	10.3	N	
F04	10.8	67.5	32.2	9.1	1.9	10.0	N	
F05	13.4	70.6	33.4	8.4	1.5	10.3	N	
F06	11.9	69.0	33.7	8.2	1.5	8.9	N	
F07	12.9	71.1	34.1	8.8	1.6	10.3	N	
F08	14.6	71.8	35.2	8.8	2.0	11.3	N	
F09	9.1	64.7	31.6	7.4	1.5	7.5	N	
F10	11.1	66.4	31.7	8.7	1.3	10.3	N	

Table 2a: Crayfish Morphometric Measurements for KET02 (*Orconectes obscurus*) (Kettle Creek).

Site: KET02 (Trout Run)
Date Sampled: 06/19/2012
Species: *Orconectes obscurus*

Male/Female Specimen #	Blotted Wet Mass (g)	Total Body Length (mm)	Carapace Length (mm)	Areola Length (mm)	Areola Width (mm)	Palm Width (mm)	Eggs/Young (♀)	Notes
F01	13.2	72.8	34.6	9.8	1.6	10.1	N	
F02	6.8	59.5	28.2	6.8	1.5	7.8	N	Right claw regenerating
F03	16.5	75.3	37.5	10.8	1.3	13.5	N	Left claw regenerating; right used
F04	12.3	71.4	34.5	9.1	1.8	9.6	N	
F05	10.2	71.0	33.3	8.9	1.7	10.4	N	Right claw missing
F06	9.2	64.4	30.7	8.5	1.4	9.6	N	
F07	7.9	64.0	29.5	7.0	1.1	7.4	N	Left claw missing; right used
F08	9.0	66.7	30.8	7.5	2.0	N/A	N	Both claws missing
F09	9.6	65.8	31.0	7.5	2.1	8.7	N	
F10	9.0	67.6	33.1	9.0	1.9	9.0	Y	

Table 2b: Crayfish Morphometric Measurements for KET02 (*Cambarus bartonii)* (Kettle Creek)

Site: KET02 (Trout Run)
Date Sampled: 06/19/2012
Species: *Cambarus bartonii*

Male/Female Specimen #	Blotted Wet Mass (g)	Total Body Length (mm)	Carapace Length (mm)	Areola Length (mm)	Areola Width (mm)	Palm Width (mm)	Eggs/Young (♀)	Notes
F01	6.3	57.0	27.8	7.9	1.9	5.3	N	
F02	2.7	42.1	20.9	5.5	1.0	3.1	N	
F03	4.1	48.1	23.3	6.0	1.3	6.1	N	
F04	8.8	58.3	29.1	7.3	1.8	9.4	N	Softshell
F05	8.0	58.6	29.1	8.1	1.7	8.6	N	Wound on left side of abdomen
F06	5.6	53.1	26.0	6.9	1.4	7.0	N	
F07	3.4	46.2	21.6	5.2	1.1	6.2	N	Right claw missing
F08	3.0	44.6	21.7	5.5	1.5	5.3	N	
F09	8.8	57.6	29.0	8.6	1.2	8.9	N	Branchiobdellid worm on left claw
F10	7.2	57.3	28.1	8.0	1.2	6.6	N	

Table 3: Crayfish Morphometric Measurements for KET03 (Kettle Creek).

Site: KET03 (DEB)
Date Sampled: 08/12/2012
Species: *Orconectes obscurus*
*Males Form I unless noted otherwise

Male/Female Specimen #	Blotted Wet Mass (g)	Total Body Length (mm)	Carapace Length (mm)	Areola Length (mm)	Areola Width (mm)	Palm Width (mm)	Eggs/Young (♀)	Notes
M01	16.8	74.0	36.0	10.1	1.1	11.0	-	
M02	23.7	78.7	39.3	10.7	1.8	17.3	-	
M03	16.1	71.4	35.6	9.7	1.3	13.9	-	
M04	15.7	72.4	34.7	9.8	1.6	13.8	-	
M05	19.1	76.0	36.2	9.9	1.8	14.8	-	
M06	16.1	70.4	33.7	10.1	1.4	13.7	-	
M07	13.4	69.2	33.7	8.9	1.1	12.8	-	
M08	13.9	68.8	32.6	8.8	1.5	11.6	-	
M09	14.5	72.2	33.5	10.2	1.6	13.1	-	
M10	12.5	68.1	31.6	9.3	1.5	11.0	-	
F01	9.1	64.5	29.5	7.8	1.4	9.6	N	
F02	8.7	63.5	29.7	7.8	1.4	8.8	N	
F03	7.8	62.7	29.1	7.4	1.8	7.9	N	
F04	8.4	62.2	28.5	8.3	1.6	8.5	N	
F05	9.1	65.1	31.2	8.6	2.1	9.2	N	
F06	9.3	64.8	29.6	8.5	1.5	9.4	N	
F07	9.6	65.0	31.1	9.0	1.6	9.3	N	
F08	8.6	59.7	28.6	8.2	1.5	8.8	N	
F09	9.6	64.4	30.8	7.3	1.5	9.1	N	
F10	16.7	78.0	37.5	10.3	2.2	11.5	N	

Table 4a: Crayfish Morphometric Measurements for KET04 (*Orconectes obscurus*) (Kettle Creek).

Site: KET04 (Bundle bridge)
Date Sampled: 08/10/2012
Species: *Orconectes obscurus*
*Males Form I unless noted otherwise

Male/Female Specimen #	Blotted Wet Mass (g)	Total Body Length (mm)	Carapace Length (mm)	Areola Length (mm)	Areola Width (mm)	Palm Width (mm)	Eggs/Young (♀)	Notes
M01	8.9	67.2	29.8	7.9	1.5	9.8	-	
M02	9.0	60.5	28.7	7.7	1.3	10.0	-	
M03	14.8	69.5	34.3	9.4	1.7	13.5	-	
M04	9.6	61.4	29.0	8.2	1.5	10.0	-	
M05	10.0	62.8	29.5	7.5	1.3	10.9	-	
M06	13.0	65.3	31.6	9.1	1.4	12.6	-	
M07	14.5	69.2	33.0	9.7	1.5	13.2	-	
M08	9.2	59.7	29.0	7.9	1.1	10.5	-	
M09	9.5	64.2	30.0	8.5	1.2	11.0	-	Left claw regenerating; right used
M10	9.2	59.8	28.8	6.8	1.5	10.6	-	
F01	12.7	71.6	34.5	8.7	1.9	11.0	N	
F02	10.1	65.5	31.0	8.0	1.3	9.2	N	
F03	6.6	57.2	26.9	7.3	1.1	8.1	N	
F04	5.9	56.3	27.0	6.7	1.3	6.7	N	Left claw regenerating
F05	5.6	54.3	25.7	7.5	1.1	7.5	N	Left claw regenerating
F06	8.9	66.0	30.6	7.7	1.7	9.2	N	
F07	9.5	68.3	29.2	7.4	1.2	10.0	N	
F08	9.2	61.8	29.1	7.8	1.8	9.3	N	
F09	6.7	58.2	26.6	7.3	1.9	7.2	N	
F10	7.3	59.0	27.1	6.9	1.1	8.7	N	

Table 4b: Crayfish Morphometric Measurements for KET04 (*Cambarus bartonii*) (Kettle Creek).

Site: KET04 (Bundle bridge)
Date Sampled: 08/10/2012
Species: *Cambarus bartonii*
*Males Form I unless noted otherwise

Male/Female Specimen #	Blotted Wet Mass (g)	Total Body Length (mm)	Carapace Length (mm)	Areola Length (mm)	Areola Width (mm)	Palm Width (mm)	Eggs/Young (♀)	Notes
M01	8.3	55.3	27.9	8.7	1.5	10.5	-	
M02	8.2	54.5	28.4	8.7	1.4	10.0	-	Left claw missing; right used
M03	5.8	53.0	25.8	8.2	1.3	8.5	-	
M04	8.2	56.0	28.1	8.3	1.5	9.5	-	
M05	8.0	57.3	27.8	8.3	1.1	9.1	-	
M06	8.6	59.6	30.8	9.3	1.3	10.6	-	Left claw missing; right used
M07	9.5	65.9	30.1	9.2	1.4	9.7	-	Atypical dark green/grayish exoskeleton - superficially resembles *O. obscurus*
M08	7.8	58.4	27.9	9.2	1.9	9.4	-	
M09	6.2	53.8	26.7	8.5	1.6	8.4	-	
M10	8.2	54.9	27.0	8.1	1.5	9.8	-	
F01	8.0	57.3	29.5	8.5	1.0	7.6	N	
F02	6.8	56.5	26.0	7.6	1.2	8.0	N	Branchiobdellid worm on dorsal surface of carapace
F03	3.5	46.6	21.7	6.6	1.5	4.7	N	
F04	12.9	65.8	31.5	9.3	1.2	10.7	Y	Well developed young
F05	4.4	48.9	22.7	6.6	1.8	6.2	N	
F06	3.9	47.6	22.5	6.7	1.7	5.2	N	
F07	12.4	64.8	30.0	9.8	1.8	10.5	Y	Well developed young
F08	6.4	54.6	26.2	7.3	1.4	7.8	N	
F09	12.5	66.6	33.0	9.0	1.2	10.0	N	Branchiobdellid worm on walking leg
F10	8.0	57.3	29.5	8.5	1.0	7.6	N	

Table 5: Crayfish Morphometric Measurements for PC01 (Pine Creek).

Site: PC01 (Torbert)
Date Sampled: 06/26/2012
Species: *Orconectes obscurus*

Male/Female Specimen #	Blotted Wet Mass (g)	Total Body Length (mm)	Carapace Length (mm)	Areola Length (mm)	Areola Width (mm)	Palm Width (mm)	Eggs/Young (♀)	Notes
M01	11.2	69.6	33.6	9.1	1.3	11.6	-	
M02	9.3	64.9	33.0	9.2	1.7	9.4	-	
M03	11.9	69.2	33.1	9.8	1.6	10.0	-	
M04	12.9	71.9	35.7	10.7	2.1	11.6	-	
M05	11.0	68.2	32.7	8.6	1.4	10.4	-	
M06	10.2	68.2	33.5	8.5	1.2	9.2	-	
M07	18.7	79.4	41.2	11.2	1.5	12.2	-	
M08	15.6	75.0	36.1	9.8	1.4	11.6	-	
M09	12.5	70.5	35.2	9.6	2.0	10.8	-	
M10	7.7	62.6	30.4	7.9	2.1	8.5	-	
F01	8.9	63.4	31.1	8.2	2.0	8.1	N	
F02	11.2	72.0	33.3	8.8	1.3	9.6	N	Right eye damaged
F03	13.0	74.6	36.3	10.3	2.3	9.3	N	
F04	11.4	71.7	33.1	10.0	1.8	8.9	N	
F05	13.5	76.0	36.3	9.8	1.4	8.4	N	
F06	9.5	65.6	32.2	8.2	1.4	9.3	N	
F07	13.0	72.6	34.6	10.0	2.3	11.7	N	
F08	12.2	70.8	34.8	9.1	1.8	10.4	N	
F09	8.4	65.6	31.0	7.8	1.5	8.2	N	
F10	11.4	70.7	33.6	8.5	1.6	9.1	N	

Table 6: Crayfish Morphometric Measurements for PC02 (Pine Creek).

Site: PC02 (Ramsey)
Date Sampled: 06/26/2012
Species: *Orconectes obscurus*

Male/Female Specimen #	Blotted Wet Mass (g)	Total Body Length (mm)	Carapace Length (mm)	Areola Length (mm)	Areola Width (mm)	Palm Width (mm)	Eggs/Young (♀)	Notes
M01	7.9	63.1	29.5	8.0	1.2	9.2	-	
M02	10.1	67.4	33.3	7.6	1.3	9.8	-	
M03	7.8	62.1	30.7	8.1	1.7	7.7	-	
M04	10.1	67.2	33.1	9.0	1.5	9.4	-	
M05	8.5	63.8	30.1	7.4	1.5	9.8	-	
M06	8.4	61.9	31.1	7.7	1.1	8.1	-	
M07	6.1	57.1	28.4	7.4	1.4	6.4	-	
M08	6.9	60.1	29.0	7.6	1.4	8.5	-	
M09	8.7	63.6	31.1	8.0	2.0	8.2	-	
M10	8.4	63.0	30.9	7.7	1.9	9.0	-	
F01	11.7	75.6	36.7	9.0	1.3	8.7	N	
F02	8.9	65.9	32.3	8.4	1.3	7.5	N	
F03	10.8	70.3	33.8	8.5	2.2	8.4	N	
F04	10.0	70.5	34.0	9.2	1.6	8.5	N	
F05	9.6	67.1	32.2	8.1	1.5	9.2	N	
F06	12.9	73.3	35.1	8.9	1.8	10.9	N	
F07	15.8	79.9	39.5	10.6	1.9	11.0	N	
F08	13.3	73.9	37.5	9.4	1.7	10.7	N	
F09	11.4	72.1	35.8	9.0	1.6	8.6	N	
F10	10.8	73.6	35.7	9.0	1.0	9.2	N	RCM

Table 7: Crayfish Morphometric Measurements for PC03 (Pine Creek).

Site: PC03 (Upper Pine Bottom)
Date Sampled: 06/26/2012
Species: *Orconectes obscurus*

Male/Female Specimen #	Blotted Wet Mass (g)	Total Body Length (mm)	Carapace Length (mm)	Areola Length (mm)	Areola Width (mm)	Palm Width (mm)	Eggs/Young (♀)	Notes
M01	12.2	71.5	35.0	9.3	1.3	10.3	-	
M02	14.3	75.5	36.7	10.1	1.6	12.6	-	Left claw deformed; softshell; blue coloration
M03	11.2	75.8	37.3	10.4	2.0	N/A	-	BCM
M04	11.2	65.6	31.9	9.0	1.5	12.5	-	Softshell; Form I
M05	9.6	66.8	32.2	8.7	1.5	10.2	-	
M06	12.3	67.1	33.1	10.0	2.1	13	-	Form I
M07	10.2	67.1	32.6	8.0	1.6	10.5	-	
M08	12.0	71.0	34.7	9.0	1.5	9.4	-	
M09	9.5	63.5	31.1	7.6	1.3	10	-	
M10	8.7	63.3	31.0	7.5	1.1	9.2	-	
F01	10.6	69.1	34.3	9.0	2.0	8.5	N	
F02	8.9	64.9	31.0	8.2	1.5	9.1	N	
F03	18.7	82.9	40.5	11.2	2.0	12.6	N	
F04	8.7	65.8	31.0	8.4	1.5	8.8	N	
F05	18.8	81.4	39.2	10.7	2.0	13.1	N	
F06	9.9	68.3	32.2	8.1	1.6	8.6	N	
F07	8.7	66.0	32.0	8.0	1.8	8.6	N	
F08	8.2	64.4	31.0	7.5	1.4	8.7	N	
F09	12.6	73.2	36.1	10.6	2.1	9.8	N	
F10	14.1	74.1	35.3	9.7	2.1	11.1	N	

Table 8: Crayfish Morphometric Measurements for PC04 (Pine Creek).

Site: PC04 (Blackwell)
Date Sampled: 08/02/2012
Species: *Orconectes obscurus*
*Males Form I unless noted otherwise

Male/Female Specimen #	Blotted Wet Mass (g)	Total Body Length (mm)	Carapace Length (mm)	Areola Length (mm)	Areola Width (mm)	Palm Width (mm)	Eggs/Young (♀)	Notes
M01	21.6	78.0	38.6	10.3	2.2	16.4	-	
M02	22.1	84.0	41.4	14.0	1.7	17.8	-	Left claw missing
M03	16.5	74.9	37.2	10.3	1.5	11.7	-	
M04	15.0	69.5	33.6	9.0	1.9	14.1	-	
M05	14.9	71.0	34.0	10.4	1.4	13.7	-	
M06	13.4	68.4	34.0	9.5	1.4	13.2	-	
M07	13.7	67.1	32.9	9.2	1.2	13.2	-	
M08	11.1	62.6	31.7	9.5	1.4	12.0	-	
M09	10.3	61.8	31.0	8.2	1.6	10.5	-	
M10	8.7	59.4	27.9	8.5	1.5	10.5	-	
F01	18.0	76.7	36.8	9.0	1.3	13.1	N	
F02	13.2	71.0	34.0	9.6	1.8	10.5	N	
F03	15.0	76.6	37.6	9.6	1.6	11.7	N	Right claw missing
F04	13.5	70.4	33.7	8.1	2.0	11.3	N	
F05	13.9	71.0	35.0	9.0	1.9	11.4	N	
F06	13.5	69.4	33.5	9.1	1.8	12.0	N	
F07	12.0	71.1	33.7	8.8	1.6	11.2	N	
F08	12.4	67.2	32.4	8.8	1.9	11.9	N	
F09	12.1	69.7	33.5	8.2	1.2	10.2	N	
F10	11.8	68.3	31.6	8.7	2.0	10.9	N	

Table 9: Crayfish Morphometric Measurements for PC05 (Pine Creek).

Site: PC05 (Colton Point Rd. bridge)
Date Sampled: 07/24/2012
Species: *Orconectes obscurus* — *Males Form I unless noted otherwise

Male/Female Specimen #	Blotted Wet Mass (g)	Total Body Length (mm)	Carapace Length (mm)	Areola Length (mm)	Areola Width (mm)	Palm Width (mm)	Eggs/Young (♀)	Notes
M01	16.7	74.1	36.8	9.9	1.4	12.4	-	
M02	13.3	74.6	34.7	7.9	1.4	9.5	-	
M03	15.8	75.7	37.0	9.4	1.5	12.5	-	
M04	12.7	68.5	31.6	9.1	1.9	11.2	-	
M05	12.6	67.2	32.6	8.9	1.3	10.5	-	
M06	13.9	72.7	36.1	10.1	2.1	11.5	-	
M07	12.0	66.1	33.0	8.0	1.3	11.7	-	
M08	15.6	74.8	36.5	9.7	1.6	12.1	-	
M09	13.2	70.6	33.7	8.2	1.5	11.2	-	
M10	9.3	62.3	30.5	7.7	1.5	9.5	-	
F01	17.7	80.4	40.3	11.1	2.1	12.6	N	
F02	15.1	76.9	36.7	8.4	1.9	10.2	N	Green-blue coloration
F03	18.8	79.3	39.6	11.2	2.4	13.0	N	
F04	11.1	75.5	32.8	7.6	1.9	8.0	N	
F05	10.3	69.2	32.1	8.0	1.5	7.9	N	
F06	10.6	69.2	33.5	9.1	2.0	7.5	N	
F07	9.6	65.5	29.6	6.9	1.1	8.6	N	
F08	9.5	69.1	30.7	8.0	2.1	7.9	N	
F09	9.4	65.1	31.0	7.9	1.5	8.6	N	
F10	10.5	70.2	32.5	7.7	1.5	10.4	N	

Table 10: Crayfish Morphometric Measurements for PC06 (Pine Creek).

Site: PC06 (Rexford/Lick Run bridge)
Date Sampled: 07/24/2012
Species: *Orconectes obscurus* — *Males Form I unless noted otherwise

Male/Female Specimen #	Blotted Wet Mass (g)	Total Body Length (mm)	Carapace Length (mm)	Areola Length (mm)	Areola Width (mm)	Palm Width (mm)	Eggs/Young (♀)	Notes
M01	10.8	68.8	32.9	7.9	1.4	12.3	-	Left claw missing, right used
M02	10.9	65.8	31.2	8.1	1.4	11.2	-	
M03	10.3	67.1	32.2	7.0	1.3	9.5	-	
M04	9.4	63.9	30.1	7.8	1.5	10.9	-	
M05	9.3	63.0	29.6	8.1	1.4	10.4	-	
M06	16.0	76.5	38.4	10.3	1.6	12.8	-	
M07	13.2	72.4	35.6	9.2	1.1	11.4	-	
M08	7.5	60.0	29.0	5.6	1.2	9.0	-	Softshell
M09	9.8	65.1	35.5	6.8	1.3	10.2	-	
M10	6.7	60.4	28.0	7.2	1	7.9	-	
F01	14.9	81.0	39.3	9.5	1.3	11.1	N	Right claw regenerating
F02	15.4	79.0	37.4	10.5	2.6	11.4	N	
F03	13.1	73.1	34.5	8.4	2.0	10.6	N	Left claw regenerating, right used
F04	10.8	71.6	34.7	9.3	1.5	8.0	N	
F05	13.2	73.6	35.0	9.3	2.3	9.0	N	
F06	14.2	78.0	36.6	10.0	2.2	10.0	N	
F07	16.6	80.4	40.1	10.7	2.6	10.7	N	
F08	12.6	72.6	34.2	8.8	1.3	9.9	N	
F09	13.1	73.7	35.6	8.7	1.5	8.7	N	
F10	11.7	74.0	35.4	9.4	1.6	9.7	N	Right claw regenerating

Table 11a: Crayfish Morphometric Measurements for PC07 (*Orconectes obscurus*) (Pine Creek).

Site: PC07 (Rt. 6 Rest Area)
Date Sampled: 07/24/2012
Species: *Orconectes obscurus* — *Males Form I unless noted otherwise

Male/Female Specimen #	Blotted Wet Mass (g)	Total Body Length (mm)	Carapace Length (mm)	Areola Length (mm)	Areola Width (mm)	Palm Width (mm)	Eggs/Young (♀)	Notes
M01	19.2	80.3	36.7	10.2	1.9	14.0	-	
M02	15.9	73.8	35.3	9.7	2.1	11.2	-	
M03	13.8	72.1	34.6	9.3	1.4	11.1	-	
M04	13.9	72.5	34.5	9.5	1.3	11.0	-	
M05	15.8	74.8	36.3	9.6	1.5	11.8	-	
M06	16.7	75.8	38	10.9	1.2	12.0	-	
M07	13.3	69.4	32.7	9.0	1.6	10.6	-	
M08	18.6	78.6	35.5	9.6	1.5	12.6	-	
M09	17.5	76.7	36.7	10.0	1.5	12.4	-	
M10	13.1	71.7	35.3	9.2	1.6	12.2	-	Left claw regenerating, right used
F01	18.8	82.7	39.4	9.5	2.6	11.9	N	
F02	17.9	83.5	38.5	10.8	1.6	11.7	N	
F03	14.8	76.3	36.4	8.9	1.3	10.7	N	
F04	14.3	73.4	35.7	9.3	2.4	11.0	N	
F05	16.8	79.3	37.5	9.4	2.3	11.1	N	
F06	20.0	82.7	40.5	10.1	1.8	11.7	N	
F07	13.8	74.0	35.1	9.0	2.3	10.5	N	
F08	16.4	78.3	37.3	8.8	1.8	10.7	N	
F09	12.7	72.5	35.2	9.5	1.8	9.1	N	
F10	11.8	69.1	32.9	8.2	1.4	9.1	N	

Table 11b: Crayfish Morphometric Measurements for PC07 (*Cambarus bartonii*) (Pine Creek).

Site: PC07 (Rt. 6 Rest Area)
Date Sampled: 07/24/2012
Species: *Cambarus bartonii*

Male/Female Specimen #	Blotted Wet Mass (g)	Total Body Length (mm)	Carapace Length (mm)	Areola Length (mm)	Areola Width (mm)	Palm Width (mm)	Eggs/Young (♀)	Notes
M01	8.2	55.6	28.3	8.6	1.8	9.5	-	
M02	8.3	55.3	28.0	7.5	1.5	9.8	-	Form I; Lots of black pigment on ventral surface
M03	10.7	58.0	28.9	8.1	1.9	11.6	-	Form I
M04	9.1	58.5	29.7	7.0	1.3	10.0	-	
M05	7.6	55.6	26.0	7.4	1.4	9.1	-	Form I
M06	9.5	58.5	30.0	7.4	1.5	10.6	-	
M07	16.8	68.4	36.1	9.4	1.9	14.0	-	Form I; Lots of black pigment
M08	11.1	59.6	30.7	8.8	2.6	12.2	-	Form I
M09	10.9	58.3	30.1	9.2	2.4	10.7	-	Form I
M10	10.8	58.6	29.7	9.8	2.0	11	-	Form I

Table 12: Crayfish Morphometric Measurements for LPC01 (Little Pine Creek).

Site: LPC01 (Lower Picnic Ground)
Date Sampled: 06/12/2012
Species: *Orconectes obscurus*

Male/Female Specimen #	Blotted Wet Mass (g)	Total Body Length (mm)	Carapace Length (mm)	Areola Length (mm)	Areola Width (mm)	Palm Width (mm)	Eggs/Young (♀)	Notes
M01	8.9	62.6	31.0	8.1	2.2	8.2	-	Left claw missing; right used
M02	8.7	61.8	31.0	8.1	1.7	9.3	-	Softshell
M03	6.2	57.3	27.6	8.7	1.5	9.2	-	Softshell
M04	11.8	69.7	35.0	9.2	2.0	11.5	-	
M05	9.7	64.0	30.9	8.1	2.0	11.1	-	Left claw regenerating; right used
M06	8.8	61.2	28.1	8.5	1.5	7.9	-	
M07	12.2	68.1	34.1	9.6	1.5	6.6	-	
M08	12.4	66.6	33.5	9.7	2.6	12.2	-	
M09	7.1	58.2	27.6	6.4	1.5	8.8	-	
M10	15.5	76.4	37.7	10.3	2.2	17.6	-	
F01	7.1	61.1	27.0	7.7	1.6	8.7	N	
F02	7.6	62.5	29.6	7.6	2.0	6.7	N	
F03	6.4	57.9	26.6	6.0	1.3	6.7	N	
F04	7.4	61.5	28.7	8.2	2.1	7.2	N	Left claw missing; right used
F05	10.4	66.0	30.3	9.1	1.7	10.0	N	
F06	8.6	67.0	28.8	8.6	2.0	9.2	N	
F07	7.2	61.1	29.9	8.0	1.8	4.8	N	
F08	8.5	62.1	30.7	9.0	2.0	7.8	N	
F09	9.0	64.1	31.1	9.1	2.0	7.4	N	
F10	6.4	59.1	27.4	7.9	2.0	6.5	N	

Table 13: Crayfish Morphometric Measurements for LPC02 (Little Pine Creek).

Site: LPC02 (Carsontown)
Date Sampled: 06/13/2012
Species: *Orconectes obscurus*

Male/Female Specimen #	Blotted Wet Mass (g)	Total Body Length (mm)	Carapace Length (mm)	Areola Length (mm)	Areola Width (mm)	Palm Width (mm)	Eggs/Young (♀)	Notes
M01	10.9	70.7	34.6	9.7	1.3	10.4	-	
M02	9.5	66.3	30.6	8.9	1.6	10.1	-	
M03	8.9	64.6	32.6	8.7	2.0	9.1	-	Softshell
M04	11.3	66.9	33.4	9.0	2.0	11	-	
M05	9.4	65.1	32.2	9.1	1.7	8.6	-	
M06	10.7	68.7	33.5	9.1	2.2	10.6	-	
M07	9.6	70.2	32.9	8.1	1.6	9.6	-	Softshell; right claw missing
M08	9.6	67.3	31.8	9.5	1.6	10.1	-	Softshell
M09	9.8	66.6	33.9	8.1	2.1	6.2	-	Left claw missing; right regenerating
M10	11.2	66.6	34.1	8.6	1.8	11.7	-	
F01	6.3	59.3	26.5	7.1	1.6	7.6	N	
F02	5.4	57.0	27.4	6.7	1.8	6.5	N	
F03	10.5	65.6	32.9	8.7	2.1	9.6	N	
F04	6.5	66.8	32.7	6.6	2.2	8.3	N	
F05	8.0	61.4	29.0	7.7	1.8	8.8	Y	
F06	6.1	57.1	28.3	6.9	1.7	7.7	N	
F07	8.9	63.8	29.8	7.0	1.8	9.0	N	
F08	7.5	59.6	29	7.1	2.0	7.9	N	
F09	6.8	55.5	27.5	7.0	1.5	8.6	N	
F10	6.8	54.5	27.1	7.2	1.6	8.3	N	

Table 14: Crayfish Morphometric Measurements for LPC03 (Little Pine Creek).

Site: LPC03 (English Center)
Date Sampled: 06/13/2012
Species: *Orconectes obscurus*

Male/Female Specimen #	Blotted Wet Mass (g)	Total Body Length (mm)	Carapace Length (mm)	Areola Length (mm)	Areola Width (mm)	Palm Width (mm)	Eggs/Young (♀)	Notes
M01	9.2	62.6	32.1	7.8	1.8	8.6	-	
M02	11.0	69.8	34.8	10.2	1.6	11.3	-	Right claw missing
M03	6.1	59.1	28.8	7.1	1.5	7.5	-	
M04	9.1	64.1	30.5	7.8	1.5	9.1	-	
M05	7.8	62.2	29.2	7.0	1.5	8.5	-	
M06	10.6	69.5	34.5	9.6	2.1	10.0	-	Right claw missing
M07	8.2	58.7	30.0	8.3	1.6	9.0	-	
M08	8.2	65.5	30.5	6.3	1.5	8.8	-	
M09	11.5	69.6	35.0	8.0	1.9	9.2	-	
M10	6.6	57.5	28.1	6.5	1.5	8.7	-	
F01	7.1	60.0	29.3	6.6	1.0	7.3	N	
F02	7.8	58.9	28.4	7.0	1.6	8.2	Y	
F03	3.6	48.6	22.8	5.6	1.6	4.7	N	
F04	3.5	50.2	24.6	5.6	1.3	5.1	N	

Table 15: Crayfish Morphometric Measurements for LAR01 (Larry's Creek).

Site: LAR01 (Susquehanna Campground)
Date Sampled: 08/02/2012
Species: *Orconectes obscurus* *Males Form I unless noted otherwise

Male/Female Specimen #	Blotted Wet Mass (g)	Total Body Length (mm)	Carapace Length (mm)	Areola Length (mm)	Areola Width (mm)	Palm Width (mm)	Eggs/Young (♀)	Notes
M01	9.9	63.0	30.1	7.7	2.1	11.9	-	Right tip of claw broken off
M02	10.6	61.1	29.6	7.9	1.6	12.5	-	
M03	6.8	56.8	27.6	7.5	1.6	9.2	-	
M04	7.7	57.3	27.7	8	1.3	10.4	-	
M05	8.7	59.1	29.1	7.2	1.9	11.6	-	Left claw regenerating, right used
M06	7.7	58.0	27.4	6.7	1.9	10.4	-	
M07	16.2	69.3	34.5	9.4	1.5	14.9	-	
M08	11.0	62.1	31.9	8.4	1.5	11.8	-	
M09	10.4	65.1	32.2	8.5	1.2	11.7	-	
M10	10.3	61.1	30.5	8.4	1.3	11.7	-	
F01	11.0	68.1	31.0	8.4	1.2	11.5	N	
F02	10.7	68.4	32.8	8.7	1.6	10.9	N	Left claw regenerating, right used
F03	9.5	64.9	30.8	8.0	1.6	9.2	N	
F04	9.8	65.6	30.6	7.6	1.6	10.3	N	
F05	8.8	62.7	30.7	7.6	1.4	10.2	N	
F06	12.3	71.1	33.2	8.6	2.1	11.0	N	
F07	11.3	67.9	33.3	8.9	2.9	10.1	N	
F08	6.4	57.6	26.7	6.3	0.8	8.2	N	
F09	7.5	62.3	30.1	8.1	1.8	7.0	N	
F10	6.1	55.8	26.1	7.1	1.2	8.0	N	

Table 16: Crayfish Morphometric Measurements for LAR02 (Larry's Creek).

Site: LAR02 (Larryville)
Date Sampled: 06/15/2012
Species: *Orconectes obscurus*

Male/Female Specimen #	Blotted Wet Mass (g)	Total Body Length (mm)	Carapace Length (mm)	Areola Length (mm)	Areola Width (mm)	Palm Width (mm)	Eggs/Young (♀)	Notes
M01	9.8	69.0	30.7	6.8	1.5	10.3	-	Softshell
M02	6.5	60.0	28.8	6.4	1.8	8.0	-	
M03	7.7	61.6	31.0	8.2	2.2	9.3	-	Right claw regenerating
M04	5.7	56.3	27.5	6.7	1.8	7.9	-	
M05	6.0	59.1	29.0	6.8	1.2	6.1	-	
M06	7.5	62.2	29.0	7.0	1.5	7.8	-	
M07	7.2	60.1	28.0	8.2	1.6	7.7	-	Right claw regenerating
M08	7.8	65.6	32.0	9.2	1.6	7.8	-	Right claw missing
M09	6.5	58.4	28.5	6.8	1.2	7.8	-	
M10	5.0	52.7	25.7	6.1	1.7	7.0	-	
F01	7.0	61.5	28.3	7.3	1.3	7.4	N	
F02	9.8	66.2	31.0	8.1	1.4	8.3	N	
F03	7.9	61.5	28.7	8.0	1.3	8.7	N	
F04	8.0	62.7	29.1	6.2	1.1	9.2	N	
F05	6.4	60.3	29.4	7.0	1.5	6.5	N	
F06	7.6	59.6	27.1	7.5	1.6	9.1	N	
F07	8.9	64.0	30.7	8.1	1.5	8.1	N	
F08	7.2	56.7	27.0	7.5	1.6	8.4	N	
F09	8.2	61.4	29.0	7.5	1.5	9.1	N	
F10	7.5	62.7	29.7	7.9	1.4	8.8	N	

Table 17: Crayfish Morphometric Measurements for LAR03 (Larry's Creek).

Site: LAR03 (State Route 973 bridge)
Date Sampled: 06/15/2012
Species: *Orconectes obscurus*

Male/Female Specimen #	Blotted Wet Mass (g)	Total Body Length (mm)	Carapace Length (mm)	Areola Length (mm)	Areola Width (mm)	Palm Width (mm)	Eggs/Young (♀)	Notes
M01	13.8	74.0	27.1	11.2	1.8	13.0	-	Left claw missing
M02	6.0	57.7	27.3	6.0	1.0	7.5	-	
M03	8.7	63.8	30.0	8.0	1.6	9.4	-	
M04	7.1	61.8	30.7	7.4	1.3	7.6	-	
M05	6.9	61.7	30.4	7.5	1.7	8.6	-	
M06	6.6	57.5	28.1	7.0	2.0	8.7	-	
M07	10.0	65.7	31.7	8.1	1.6	10.2	-	
M08	11.4	66.1	33.1	9.1	1.8	11.3	-	
M09	6.5	58.5	27.4	7.0	1.6	8.2	-	
M10	7.9	61.4	29.7	7.0	1.2	9.3	-	
F01	10.4	69.5	33.4	8.8	1.6	9.0	N	
F02	7.2	63.1	29.5	6.4	2.0	5.5	N	
F03	5.5	57.7	28.0	6.4	1.8	6.3	N	
F04	9.2	64.5	31.2	8.0	2.5	9.4	N	
F05	5.8	54.5	25.3	6.0	1.7	7.5	Y	Right claw regenerating
F06	8.2	64.8	31.0	7.6	2.2	8.7	N	
F07	7.6	59.0	26.5	6.5	1.0	8.7	Y	
F08	4.9	56.3	27.7	6.1	1.4	6.2	N	
F09	11.1	68.4	32.4	8.0	2.5	10.6	N	
F10	8.3	59.3	28.5	6.4	1.5	9.7	Y	

Table 18: Crayfish Morphometric Measurements for LYC01 (Lycoming Creek).

Site: LYC01 (Newberry)
Waterway: Lycoming Creek
Date Sampled: 06/06/2012
Species: *Orconectes obscurus*

Male/Female Specimen #	Blotted Wet Mass (g)	Total Body Length (mm)	Carapace Length (mm)	Areola Length (mm)	Areola Width (mm)	Palm Width (mm)	Eggs/Young (♀)	Notes
M01	20.1	80.1	40.1	12.0	2.9	13.9	-	
M02	7.5	62.2	30.4	9.5	1.9	7.7	-	
M03	8.3	62.0	30.9	7.9	2.3	6.2	-	
M04	13.7	71.6	34.4	9.6	2.6	10.9	-	
M05	9.4	60.7	30.3	10.3	2.3	9.8	-	
M06	10.2	66.5	32.8	10.1	1.6	8.5	-	
M07	10.8	64.7	31.4	9.1	2.1	9.2	-	
M08	10.2	68.0	33.1	9.4	2.5	4.9	-	Left claw missing; right regenerating
M09	8.6	58.6	30.2	8.3	1.9	8.5	-	
M10	11.5	63.7	31.7	8.5	1.4	10.4	-	
F01	18.6	79.4	38.7	11.1	1.4	12.3	N	
F02	14.1	71.3	33.5	9.3	2.3	11.6	N	
F03	7.4	61.4	29.0	6.7	2.0	7.3	N	
F04	7.5	55.8	26.1	7.7	2.7	7.2	N	
F05	8.0	64.4	30.0	7.0	1.8	6.3	N	
F06	8.6	62.5	29.1	6.4	2.2	9.7	N	
F07	9.6	61.7	28.6	7.3	1.8	9.2	N	
F08	11.8	67.7	31.6	7.7	2.0	9.5	N	
F09	7.1	59.7	29.6	7.4	1.6	7.9	N	Left claw missing; right used
F10	7.8	57.6	28.0	7.6	1.7	8.0	N	Left claw regenerating; right used

Table 19: Crayfish Morphometric Measurements for LYC02 (Lycoming Creek).

Site: LYC02 (Trout Run)
Date Sampled: 06/07/2012
Species: *Orconectes obscurus*

Male/Female Specimen #	Blotted Wet Mass (g)	Total Body Length (mm)	Carapace Length (mm)	Areola Length (mm)	Areola Width (mm)	Palm Width (mm)	Eggs/Young (♀)	Notes
M01	11.8	68.4	35.6	10.6	2.7	9.8	-	
M02	8.6	66.2	36.7	8.7	2.1	7.6	-	
M03	9.3	65.5	36.5	9.4	2.5	9.2	-	
M04	10.6	68.9	34.4	9.2	1.8	6.5	-	
M05	11.1	67.9	32.1	9.7	2.2	11.4	-	
M06	12	65.7	34.6	9.0	2.5	9.3	-	
M07	13.5	68.5	35.8	9.6	2.2	11.2	-	
M08	8.5	63.9	31.6	8.2	1.8	7.8	-	
M09	8.9	68.6	31.3	8.2	2.2	11.1	-	
M10	10.3	66.5	38.1	9.4	2.2	7.5	-	
F01	5.7	58.1	25.1	5.6	1.7	7.1	N	
F02	4.4	58.5	24.9	5.3	2.2	5.7	N	
F03	3.7	49.1	24.0	6.3	1.4	5.1	N	
F04	4.1	48.3	22.9	5.0	1.3	5.8	Y	
F05	4.0	56.0	24.2	5.8	1.6	5.5	N	
F06	18.3	73.3	34.6	7.7	2.4	10.4	N	Caught 06/08/12
F07	10.4	66.2	30.4	8.1	2.0	6.7	N	Caught 06/08/12
F08	9.0	57.0	28.3	8.1	2.4	7.8	N	Caught 06/08/12
F09	9.6	58.7	28.9	7.3	1.7	8.2	N	Caught 06/08/12
F10	10.1	59.5	29.7	7.4	2.0	7.3	N	Caught 06/08/12

Table 20: Crayfish Morphometric Measurements for LYC03 (Lycoming Creek).

Site: LYC03 (Camp Susque Road bridge)
Date Sampled: 06/11/2012
Species: *Orconectes obscurus*

Male/Female Specimen #	Blotted Wet Mass (g)	Total Body Length (mm)	Carapace Length (mm)	Areola Length (mm)	Areola Width (mm)	Palm Width (mm)	Eggs/Young (♀)	Notes
M01	9.5	64.3	32.3	9.1	1.8	8.4	-	
M02	9.0	65.6	33.4	7.8	2.3	10.2	-	
M03	8.7	62.0	30.7	7.8	2.5	9.4	-	
M04	9.4	65.7	32.6	8.5	2.0	9.6	-	
M05	7.4	61.1	30.8	7.3	1.5	7.9	-	
M06	8.0	61.4	30.0	7.8	1.3	8.4	-	
M07	6.8	58.1	29.3	6.5	1.6	9.1	-	
M08	10.0	67.2	33.5	9.2	1.0	9.4	-	
M09	9.7	65.5	31.6	8.2	2.2	8.7	-	
M10	13.3	72.6	35.9	9.3	1.9	10.4	-	
F01	8.5	61.3	26.5	7.5	1.5	8.1	Y	
F02	3.1	46.7	27.5	6.0	1.4	4.6	N	
F03	4.3	50.0	21.9	6.1	1.4	5.4	Y	
F04	10.8	64.2	30.3	8.2	2.2	9.2	Y	
F05	3.1	46.4	23.0	6.0	1.0	5.3	N	
F06	5.2	57.0	26.5	7.5	1.5	5.8	N	
F07	5.7	48.6	28.7	5.4	1.6	5.8	Y	
F08	3.2	44.5	22.5	5.1	1.5	5.0	N	
F09	3.4	48.2	24.8	6.0	2.0	4.6	N	
F10	3.2	48.6	23.9	5.5	1.0	5.1	N	

Table 21: Crayfish Morphometric Measurements for LYC04 (Lycoming Creek).

Site: LYC04 (Pleasant Stream Road bridge)
Date Sampled: 06/11/2012
Species: *Orconectes obscurus*

Male/Female Specimen #	Blotted Wet Mass (g)	Total Body Length (mm)	Carapace Length (mm)	Areola Length (mm)	Areola Width (mm)	Palm Width (mm)	Eggs/Young (♀)	Notes
M01	7.1	67.2	29.6	8.8	2	7.8	-	
M02	7.5	59.3	28.9	8	1.7	7.4	-	
M03	9.4	65.4	32	9.6	1.5	10	-	
M04	8.2	65.1	30	7.8	1.5	7.8	-	
M05	6.8	61.5	29.5	8.3	2.1	7.1	-	
M06	7.4	60	27.9	7.8	2.1	8.6	-	
M07	7.8	60.7	29.9	8.4	1.9	8.2	-	
M08	8.7	65.3	33	8.4	2.1	7.6	-	
M09	10.9	68.8	32.8	9.5	2.2	9.2	-	
M10	11	66.8	34.1	9.6	1.8	10.4	-	
F01	8.7	61.5	30.7	8.4	1.5	5.8	Y	
F02	7.6	58.8	27.2	8.4	1.5	8	Y	
F03	3.1	44.1	21.1	5.5	1.5	3.8	N	
F04	5.8	55.3	25.6	7.3	2.6	6	N	
F05	1.9	40.5	18.8	4.9	1.1	3.7	N	
F06	1.7	39.2	19.4	4.2	1	3.7	N	
F07	1.5	35.4	18.6	4.9	1.2	2.6	N	
F08	2.1	40.1	19.8	5.1	1.4	3.4	N	

Table 22: Crayfish Morphometric Measurements for LOY01 (Loyalsock Creek).

Site: LOY01 (Mill Creek Lane)
Date Sampled: 08/08/2012
Species: *Orconectes rusticus x obscurus* (?)

*Males Form I unless noted otherwise

Male/Female Specimen #	Blotted Wet Mass (g)	Total Body Length (mm)	Carapace Length (mm)	Areola Length (mm)	Areola Width (mm)	Palm Width (mm)	Eggs/Young (♀)	Notes
M01	19.8	76.1	37.0	10.0	2.3	15.3	-	No rust spots; smooth mandibles; straight rostrum; 8 chela tubercles; no black chelae bands
M02	20.9	79.2	40.1	10.7	2.0	16.1	-	No rust spots; smooth mandibles; straight rostrum; 9 tubercles; no black bands
M03	15.7	71.0	35.3	9.0	1.8	15.4	-	No rust spots; smooth mandibles; straight rostrum; 8 tubercles; no black bands
M04	18.1	76.6	35.8	10.4	1.8	14.3	-	Rust spots; smooth mandibles; curved rostrum; 8 tubercles; black bands
M05	13.4	66.4	31.8	9.7	1.5	12.9	-	Rust spots; smooth mandibles; curved rostrum; 6 tubercles; black bands
M06	12.0	69.0	32.8	9.0	1.5	11.8	-	Rust spots; smooth mandibles; curved rostrum; 8 tubercles; black bands
M07	12.9	70.8	33.7	8.8	1.7	12.1	-	No rust spots; smooth mandibles; straight rostrum; 8 tubercles; no black bands
M08	14.1	66.9	33.1	8.7	2.1	13.3	-	Rust spots; smooth mandibles; straight rostrum; 8 tubercles; no black bands
M09	13.6	66.7	32.3	9.0	1.6	13.0	-	Rust spots; smooth mandibles; straight rostrum; 8 tubercles; black bands
M10	14.0	68.0	33.4	9.1	1.4	14.4	-	No rust spots; smooth mandibles; curved rostrum; 6 tubercles; black bands
F01	16.3	78.0	32.1	9.6	1.9	11.5	N	Rust spots; smooth mandibles; straight rostrum; 8 tubercles; no black bands
F02	13.2	73.5	33.8	8.0	1.4	11.1	N	Rust spots; smooth mandibles; curved rostrum; 7 tubercles; black bands
F03	12.2	72.6	33.1	8.4	1.4	11.4	N	No rust spots; smooth mandibles; straight rostrum; 9 tubercles; no black bands; left claw regenerating
F04	11.7	72.2	32.9	9.0	1.8	10.8	N	No rust spots; smooth mandibles; straight rostrum; 6 tubercles; black bands
F05	12.2	73.3	34.3	7.8	2.4	11.4	N	No rust spots; smooth mandibles; curved rostrum; 7 tubercles; black bands; left claw regenerating
F06	14.4	74.5	35.6	8.9	1.8	11.3	N	No rust spots; smooth mandibles; curved rostrum; 9 tubercles; black bands
F07	12.3	71.9	34.3	8.5	2.2	9.8	N	No rust spots; smooth mandibles; curved rostrum; 9 tubercles; black bands
F08	13.4	74.5	34.4	8.6	2.4	11.2	N	No rust spots; smooth mandibles; straight rostrum; 8 tubercles; no black bands
F09	16.0	79.0	37.7	9.5	1.9	12.0	N	No rust spots; serrated mandibles; straight rostrum; 8 tubercles; black bands
F10	13.0	70.9	32.7	8.0	2.0	11.6	N	No rust spots; serrated mandibles; straight rostrum; 7 tubercles; no black bands

Table 23: Crayfish Morphometric Measurements for LOY02 (Loyalsock Creek).

Site: LOY02 (Sandy Bottom)
Date Sampled: 06/14/2012
Species: *Orconectes rusticus*

Male/Female Specimen #	Blotted Wet Mass (g)	Total Body Length (mm)	Carapace Length (mm)	Areola Length (mm)	Areola Width (mm)	Palm Width (mm)	Eggs/Young (♀)	Notes
M01	8.5	60.4	29.9	6.9	2.3	9.1	-	
M02	7.6	60.9	30.4	7.8	1.8	8.8	-	
M03	9.8	63.9	32.0	8.8	1.7	9.5	-	
M04	11.2	69.2	34.3	8.8	1.2	7.4	-	
M05	9.2	64.5	31.6	7.7	2.0	9.2	-	
M06	8.7	61.1	30.3	8.6	1.7	9.2	-	
M07	10.8	65.4	31.2	8.1	1.6	12.2	-	Softshell
M08	11	67.2	34.4	9.0	1.8	8.2	-	
M09	10.2	64.2	30.1	8.4	2.1	9.0	-	
M10	15.1	73.1	37.2	10.5	2.3	10.2	-	
F01	6.0	56.9	26.8	6.8	1.3	7.8	N	
F02	6.2	54.9	27.8	6.5	1.3	7.1	N	
F03	6.4	57.1	26.9	5.6	1.3	6.8	N	
F04	7.2	58.5	27.7	7.5	1.2	8.3	N	
F05	5.8	52.7	24.6	5.8	1.6	7.2	N	
F06	7.7	62.9	29.7	7.5	1.2	7.7	N	
F07	9.1	65.3	29.9	7.5	1.6	8.8	N	
F08	6.5	55.9	27.0	7.0	1.2	8.2	N	
F09	9.2	64.0	30.5	7.7	1.2	9.4	N	
F10	10.4	65.6	31.7	8.1	1.8	9.7	N	

Table 24: Crayfish Morphometric Measurements for LOY03 (Loyalsock Creek).

Site: LOY03 (Splash Dam Road)
Date Sampled: 06/14/2012
Species: *Orconectes rusticus*

Male/Female Specimen #	Blotted Wet Mass (g)	Total Body Length (mm)	Carapace Length (mm)	Areola Length (mm)	Areola Width (mm)	Palm Width (mm)	Eggs/Young (♀)	Notes
M01	11.4	68.3	32.8	8.1	1.5	10.7	-	Softshell
M02	13.0	72.2	34.9	9.7	2.2	12.1	-	
M03	10.5	64.6	33.0	8.8	2.3	10.5	-	
M04	14.4	70.4	34.6	9.8	2.6	11.8	-	
M05	11.6	67.1	33.1	9.1	1.2	10.3	-	
M06	11.0	66.7	31.8	9.1	2.1	10.8	-	
M07	9.4	64.3	31.5	7.8	1.6	9.7	-	
M08	16.6	75.3	37.9	10.5	1.7	12.9	-	
M09	8.3	64.5	30.6	7.6	1.6	8.8	-	
M10	9.5	64.7	32.3	9.1	1.4	9.0	-	
F01	10.0	65.4	31.9	7.6	1.6	9.4	N	
F02	10.0	69.4	32.3	8.6	1.2	10.0	N	
F03	9.1	64.1	29.0	7.8	1.8	7.7	N	
F04	11.2	68.3	32.0	7.7	1.7	10.3	N	
F05	7.6	60.2	29.6	7.6	1.6	9.3	N	
F06	7.9	62.1	28.0	7.4	1.5	6.7	N	
F07	8.5	65.6	29.6	8.5	1.8	6.6	N	
F08	6.9	58.5	27.1	6.6	1.6	8.2	N	
F09	12.3	72.2	33.6	10.3	1.6	10.5	N	
F10	9.3	63.9	30.8	7.5	2.1	9.9	N	

Table 25: Crayfish Morphometric Measurements for LOY04 (Loyalsock Creek).

Site: LOY04 (Almost Heaven Campground)
Date Sampled: 06/27/2012
Species: *Orconectes rusticus*
*Males Form I unless noted otherwise

Male/Female Specimen #	Blotted Wet Mass (g)	Total Body Length (mm)	Carapace Length (mm)	Areola Length (mm)	Areola Width (mm)	Palm Width (mm)	Eggs/Young (♀)	Notes
F01	8.5	64.3	31.8	8.3	1.9	8.7	N	
F02	2.8	45.6	21.6	4.7	1.0	5.4	N	
F03	4.7	59.2	27.2	6.3	1.5	N/A	N	Both claws missing; Softshell
F04	6.9	60.0	29.2	7.3	1.1	7.8	N	
F05	3.5	49.6	24.6	6.0	0.9	6.0	N	Right claw missing
F06	7.7	60.9	29.8	7.6	2.0	8.3	N	

Table 26: Crayfish Morphometric Measurements for LOY05 (Loyalsock Creek).

Site: LOY05 (Forksville)
Date Sampled: 08/01/2012
Species: *Orconectes rusticus* *Males Form I unless noted otherwise

Male/Female Specimen #	Blotted Wet Mass (g)	Total Body Length (mm)	Carapace Length (mm)	Areola Length (mm)	Areola Width (mm)	Palm Width (mm)	Eggs/Young (♀)	Notes
M01	19.7	75.2	36.7	9.1	2.9	9.5	-	
M02	17.0	73.6	36.4	10.5	2.0	13.5	-	
M03	19.7	74.1	34.9	9.2	1.4	15.2	-	
M04	17.9	73.2	36.7	9.3	1.8	14.5	-	
M05	20.4	72.8	35.3	10.1	2.1	15.6	-	
M06	16.2	72.9	34.6	10.0	1.4	15.3	-	Left claw regenerating, right used
M07	17.4	71.5	35.0	10.2	1.0	11.4	-	Gonopods deformed/damaged
M08	13.8	68.0	32.5	9.3	1.9	12.9	-	
M09	12.7	67.7	32.6	7.7	1.6	12.5	-	
M10	13.2	65.6	32.3	7.6	2.0	10.8	-	
F01	10.1	65.2	31.1	9.0	2.2	10.0	N	
F02	8.7	62.0	29.7	7.4	1.5	7.8	N	
F03	8.2	61.9	30.3	7.3	1.9	9.4	N	
F04	8.8	62.5	30.5	7.1	1.9	9.5	N	
F05	6.9	59	28.0	6.8	1.6	7.0	N	
F06	13.9	71.4	35.3	9.0	2.4	10.8	N	
F07	7.1	60.5	29.6	7.2	1.5	7.1	N	Missing right 1/3 of telson
F08	7.0	58.2	27.0	6.3	1.6	8.6	N	
F09	6.9	61.8	29.4	7.7	1.8	7.4	N	Left claw regenerating, right used
F10	8.5	62.8	29.1	7.6	2.0	7.8	N	

Table 27: Crayfish Morphometric Measurements for MUN01 (Muncy Creek).

Site: MUN01 (N Main Street Bridge)
Date Sampled: 08/01/2012
Species: *Orconectes obscurus* *Males Form I unless noted otherwise

Male/Female Specimen #	Blotted Wet Mass (g)	Total Body Length (mm)	Carapace Length (mm)	Areola Length (mm)	Areola Width (mm)	Palm Width (mm)	Eggs/Young (♀)	Notes
M01	18.0	76.7	36.0	9.6	2.1	15.3	-	
M02	13.6	70.4	33.1	8.5	1.6	10.6	-	Rostrum tip broken off
M03	16.5	70.8	33.6	9.0	2.0	14.8	-	
M04	16.1	71.7	35.6	10.1	2.1	11.7	-	
M05	12.7	67.8	34.0	9.4	2.1	13.3	-	Left claw regenerating; right used
M06	17.0	73.2	35.5	9.4	1.5	14.3	-	
M07	13.6	66.3	33.0	9.7	1.6	13.5	-	
M08	14.0	71.8	34.6	9.6	1.6	15.1	-	Right claw missing
M09	10.9	67.1	32.8	8.6	2.0	12.6	-	
M10	17.4	74.2	36.2	9.7	1.4	13.8	-	
F01	17.9	75.7	36.6	9.4	1.9	8.6	N	
F02	14.9	71.3	34.0	9.2	2.0	11.6	N	
F03	17.2	74.2	35.6	9.6	2.0	7.4	N	
F04	15.9	78.1	39.3	10.1	2.2	9.0	N	Left claw regenerating, right used
F05	16.8	75.6	37.5	10.0	2.4	10.8	N	
F06	15.0	73.7	34.9	9.5	2.1	10.6	N	
F07	14.5	73.1	35.2	10.2	1.7	12.4	N	
F08	11.8	70.7	34.5	9.4	2.1	11.2	N	Left claw regenerating, right used
F09	11.7	66.0	32.7	9.5	1.5	11.6	N	
F10	11.9	67.2	31.8	7.8	2.0	10.7	N	

Table 28: Crayfish Morphometric Measurements for MUN02 (Muncy Creek).

Site: MUN02 (Barto Hollow Road Bridge)
Date Sampled: 07/02/2012
Species: *Orconectes obscurus* — *Males Form I unless noted otherwise

Male/Female Specimen #	Blotted Wet Mass (g)	Total Body Length (mm)	Carapace Length (mm)	Areola Length (mm)	Areola Width (mm)	Palm Width (mm)	Eggs/Young (♀)	Notes
M01	16.9	76.5	39.0	9.5	2.3	14.1	-	Form II
M02	21.8	79.6	39.2	11.7	2.3	15.8	-	
M03	15.7	74.5	36.5	10.3	2.0	11.5	-	
M04	16.1	75.5	37.1	10.1	2.3	14.6	-	
M05	15.4	73.0	36.6	9.1	1.5	15.1	-	
M06	19.4	75.6	38.7	10.9	2.2	16.3	-	Left gonopod deformed/damaged
M07	25.8	89.4	45.6	13.2	2.1	15.6	-	Form II
M08	22.0	84.9	40.9	12.2	2.3	17.8	-	
M09	17.4	70.3	33.3	10.3	1.9	12.7	-	Rostrum tip broken off
M10	16.2	75.4	38.2	11.3	1.6	12.1	-	Form II
F01	9.8	69.1	34.6	8.8	1.2	9.4	N	Left claw missing; right used
F02	7.3	59.7	29.7	8.2	1.6	7.7	N	
F03	6.2	58.7	28.2	7.3	1.9	7.2	N	
F04	7.0	60.4	29.7	7.6	1.4	7.8	N	
F05	5.2	55.6	27.1	6.8	1.5	6.5	N	
F06	7.1	62.1	29.7	7.2	1.7	7.1	N	
F07	6.1	60.2	29.7	7.6	1.3	6.8	N	
F08	5.9	58.6	28.6	7.0	1.9	6.8	N	
F09	6.8	59.3	29.5	7.9	1.4	7.3	N	
F10	10.4	69.0	33.4	8.1	1.5	8.7	N	

APPENDIX II

Sample Site Water Chemistry Data

Table 1: Summary of Water Chemistry Data from Kettle Creek.

Parameter	KET01 (08/14/12)	KET02 (06/19/12)	KET04 (08/10/12)
Water Temperature (°C)	20.5	17.5	21.6
Dissolved Oxygen (mg/l)	5.86 (65.2% Sat.)	8.98 (94.0% Sat.)	7.57 (85.9% Sat.)
Specific Conductivity (μs)	104.1	49.1	63.8
Turbidity (NTU)	14.2	2.05	5.38
pH	6.40	6.62	7.60

Table 2: Summary of Water Chemistry Data from Pine Creek.

Parameter	PC01 (06/26/12)	PC02 (06/26/12)	PC03 (06/26/12)	PC04 (08/02/12)	PC05 (07/24/12)	PC06 (07/24/12)	PC07 (07/24/12)
Water Temperature (°C)	23.9	19.9	22.1	22.9	25.8	27.0	28.0
Dissolved Oxygen (mg/l)	8.13 (95.8% Sat.)	7.56 (83.1% Sat.)	8.22 (94.3% Sat.)	8.02 (93.3% Sat.)	6.58 (80.7% Sat.)	7.60 (95.1% Sat.)	7.99 (102.2% Sat.)
Specific Conductivity (µs)	95.6	96.0	79.6	106.7	92.8	92.7	97.0
Turbidity (NTU)	2.06	2.16	2.70	4.34	3.02	2.61	2.33
pH	7.31	6.71	7.70	7.66	7.05	7.92	8.54

Table 3: Summary of Water Chemistry Data from Larry's Creek.

Parameter	LAR01 (08/08/12)	LAR02 (06/15/12)	LAR03 (06/15/12)
Water Temperature (°C)	23.5	16.0	16.5
Dissolved Oxygen (mg/l)	7.62 (89.7% Sat.)	9.54 (96.8% Sat.)	9.24 (94.6%)
Specific Conductivity (μs)	101.9	65.7	65.2
Turbidity (NTU)	4.81	2.17	2.36
pH	7.54	6.93	6.76

Table 4: Summary of Water Chemistry Data from Loyalsock Creek.

Parameter	LOY01 (08/08/12)	LOY02 (06/14/12)	LOY03 (06/14/12)	LOY04 (06/27/12)	LOY05 - UP (08/01/12)	LOY05 - DOWN (08/01/12)
Water Temperature (°C)	22.4	18.1	18.1	20.8	23.4	23.1
Dissolved Oxygen (mg/l)	6.97 (80.4% Sat.)	8.12 (86.1% Sat.)	8.42 (89.1% Sat.)	8.08 (90.2% Sat.)	7.1 (83.3% Sat.)	7.43 (86.8% Sat.)
Specific Conductivity (µs)	81.8	66.6	61.3	66.2	52.9	65.5
Turbidity (NTU)	1.66	0.63	0.95	1.0	1.23	0.81
pH	6.43	6.63	6.43	7.26	6.83	7.06

Table 5: Summary of Water Chemistry Data from Muncy Creek.

Parameter	MUN01 (08/01/12)	MUN02 (07/02/12)
Water Temperature (°C)	21.8	24.3
Dissolved Oxygen (mg/l)	6.91 (78.8% Sat.)	6.96 (83.2% Sat.)
Specific Conductivity (µs)	154.6	98.6
Turbidity (NTU)	3.06	1.21
pH	6.31	7.48