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## TROPHIC INTERACTIONS ASSOCIATED WITH INTRODUCTION OF THE INVASIVE QUAGGA MUSSEL IN LAKE MEAD, NEVADA

by

Eric Michael Loomis

Bachelor of Arts University of Nevada, Las Vegas 2003

A thesis submitted in partial fulfillment of the requirements for the

Master of Public Health Degree in the School of Community Health Sciences
Department of Environmental and Occupational Health
School of Community Health Sciences

Graduate College University of Nevada, Las Vegas May 2009



## Thesis Approval

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## **ABSTRACT**

## Trophic Interactions Associated with Introduction of the Invasive Quagga Mussel in Lake Mead, Nevada

by

## Eric Michael Loomis

Dr. Shawn Gerstenberger, Examination Committee Chair Chair and Associate Professor of Public Health University of Nevada, Las Vegas

The ecology of Lake Mead is experiencing an uncertain future since the documented arrival of the invasive quagga mussel. Interactions among constituents of the food web will undoubtedly be altered over time, adversely affecting the popular sport fishery. This study is the first in Lake Mead to present historic trends in lower trophic interactions among threadfin shad and zooplankton before, and shortly after, the arrival of quagga mussels. Shad stomach content analysis revealed cladocerans and copepods were the dominant identifiable food items in Las Vegas Bay and Overton Arm of Lake Mead. Baseline energetics data through the use of stable isotope analysis showed little variation in  $\delta^{13}$ C among larval and adult shad since quaggas were discovered. Fluctuations in  $\delta^{15}$ N were more variable over the same span, with 2008 showing highest nitrogen measurements. Historic relative shad abundance and zooplankton biomass data were gathered from multiple agencies for the purpose of comparative analysis. This project was funded by the National Park Service and is part of a long-term monitoring plan in response to quagga mussel influences in the Lake Mead watershed over time.

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## CHAPTER 1

## INTRODUCTION

The Lake Mead watershed has long provided valuable resources for not only the evergrowing population of the Las Vegas metropolitan area, but also to regions throughout the western United States. As the primary drinking water source for millions of people and a popular recreational area, Lake Mead is integral for a wide variety of human needs. It has been well documented that Lake Mead in the past decade has experienced the adverse affects of drought enveloping the Mojave Desert. Lake levels have dropped dramatically, threatening the water supply to a burgeoning human population. Moreover, the immense cost associated with constructing new delivery systems and the search for alternate drinking water resources puts a strain on the region's economy.

In addition to its value to the human population, Lake Mead has historically exhibited an aquatic ecological community conducive to supporting a world-class striped bass (*Morone saxatilis*) fishery. Vital to the success of any fishery is the complex interactions among various species in the aquatic ecosystem. Food chain dynamics are an essential element when considering the overall health of the fishery. A significant ecological disturbance has the potential to reduce resiliency in the system, leading to detrimental and irreversible damage to the aquatic environment.

The recent discovery of the invasive quagga mussel (*Dreissena bugensis*) has provided a new challenge to scientific research in the Lake Mead watershed. First

discovered in January 2007 in Boulder Basin of Lake Mead, quagga mussels have established in great numbers over a short time period (LaBounty & Roefer, 2007). Suspected to have reached Lake Mead from the Great Lakes from recreational boats over land, LaBounty & Roefer (2007) conclude the main invasion probably occurred in 2004 or possibly 2003.

The biological mechanisms of quaggas alter the food web in a number of ways. They are highly efficient water filterers, removing substantial amounts of phytoplankton, which in turn decrease the food source for zooplankton (Claxton et al., 1998). The potential impacts quagga mussels pose to the Lake Mead fishery are largely unknown at the present time. An implication of the potential decline in the zooplankton community which drives energetics is a growing concern. Extensive research in the Great Lakes region where both quaggas and the closely related zebra mussel has affected trophic dynamics may not adequately address the unique limnology of the arid southwest in relation to genetic adaptation. The quagga's ability to adapt to warmer climates and differences in food availability may prove irrelevant in relation to the Great Lakes.

In addition to changes in food webs, Snyder et al. (1997) found that the closely related zebra mussel (*Dreissena polymorpha*) produce a by-product known as pseudofeces that when decomposed create a threat to water quality at exponential levels relative to size. Moreover, organic pollutants created by pseudofeces can be readily passed up the food chain and consequently into wildlife communities. The question of pollutants and contaminants being introduced in Lake Mead due to the exotic quagga has the potential to adversely affect human health. The ability for these mussels to

biomagnify pollutants through the food web and the possibility of undesirable algal blooms should be a concern for recreation in areas where quaggas are abundant.

## Purpose of the Study

The purpose of this study is to determine a possible shift in food preference among lower trophic level species of fish, specifically threadfin shad (*Dorosoma petenense*), since the introduction of the invasive quagga mussel into Lake Mead. Since the introduction of quaggas is a relatively recent phenomenon, it is important to distinguish a potential alteration in diet caused by this highly invasive species of bivalve. Due to the lack of published historical count data from Lake Mead regarding threadfin shad in the scientific literature, this study will also attempt to document past trends with more recent data since the arrival of quaggas in the watershed.

The objectives of this study are to identify the foraging habits of threadfin shad in sampled locations in Lake Mead since the introduction of quagga mussels. Moreover, this paper evaluates population parameters of threadfin shad in specific areas of Lake Mead to determine if quaggas are a significant influence. Finally, this project serves as a baseline for further study into trophic energetics associated with the quagga mussel and attempts to glean insight into the sustainability of the Lake Mead fishery.

## **Research Questions**

With the introduction of any exotic species into an ecosystem, uncertainty abounds.

Questions this research attempts to answer include: Are threadfin shad utilizing quagga veligers as part of their diet and if so; are they a viable food source? By examining

stomach contents of the primary forage fish in Lake Mead, the question of what food items are available since quagga mussels arrived will attempt to be answered. Since this study examines larval, as well as adult threadfin shad, identifying if there is a particular size-class of shad that forages more effectively on quagga veligers is analyzed. Are their seasonal or water quality components involved in abundance of veligers causing adverse food web dynamics? The ecological conditions present in Lake Mead appear to be markedly different than from other water bodies where quagga mussels have been known to disrupt aquatic food webs. By analyzing original data gathered over the past several years, fisheries managers will hopefully be able to answer the question of whether quagga mussels have had an adverse affect on the sport fishery throughout portions of Lake Mead.

## Significance of the Study

If the primary food source supplying game fish is compromised—threadfin shad, the potential for mitigation measures may need to be addressed to sustain this valuable resource. A decline in zooplankton over time could result in a crash of shad populations where quagga mussels are ubiquitous. Such changes are of concern due to food web perturbations which could diminish equilibrium in the aquatic community. In addition, not only is the sport fishery in peril of exotic introductions. The impact to the endangered razorback sucker (*Xyrauchan texanus*) is of concern due to its specific habitat requirements in this ecosystem.

## CHAPTER 2

## REVIEW OF RELATED LITERATURE

## Quagga Mussels

The quagga mussel (*Dreissena bugensis*) and its close relative zebra mussel (*Dreissena polymorpha*) are native to the Dnieper River drainage in the Ukraine (Mills et al., 1996). Subtle morphological differences characterize *D. bugensis* from *D. polymorpha* in both larval and adult stages, most notably in the degree of roundness in shell shape, size, and color patterns (Mills et al., 1993). Extensive research has revealed that the genus *Dreissena* is highly polymorphic and is capable of producing millions of larvae (veligers). Furthermore, after a few generations rare alleles may increase greatly in frequency, leading to the adaptation to new environmental conditions after several generations (Mills et al., 1996). Ecological investigation concluded that quagga and zebra mussels rarely, if ever, overlap completely in the aquatic environment, owing to differences in life history parameters and explaining why the two species have different ranges (May & Marsden, 1992).

Spatial distribution in the water column varies greatly depending on geographic variability. Shevtsova (1968) observed in the Dnieper River drainage that with increasing water temperatures from north to south there was a higher occurrence of *D. bugensis*. In contrast, quaggas were initially found in deeper, colder waters of the Great Lakes (Mills et al., 1993). Based on contrast in temperature tolerance, it was later

hypothesized that *D. bugensis* was not limited to deep water habitats and could inhabit a wider range of depths in North American waters (Mills et al., 1996).

## Invasive Status

Great Lakes. Although difficult to determine with certainty, it is suspected that the quagga mussel first arrived in September 1989 in Lake Erie from ship ballast water originating from Eastern Europe. Among settled *Dreissena* species at this site, the quagga mussel population gradually increased from 2.3% during 1990 to 6.6% by October 1992 (Mills et al., 1993). Other researchers have speculated that the zebra mussel pre-dated quagga in the Great Lakes region based on its limited distribution upon arrival (May & Marsden, 1992). Genetic analysis confirmed that *D. bugensis* has two distinct phenotypes in the Great Lakes which suggests there is considerable plasticity in morphology and physiology within the species (Spidle et al., 1994). Verification that there is more than one extant species increases the taxonomic resolution of the genus, as concluded by Spidle et al. (1994).

There has been ongoing controversy among the scientific community surrounding the ecological impact dreissenid mussels have had since their introduction into the Great Lakes. It was postulated by Mills et al. (1993) that colonization by quaggas could provide a viable food source from the pseudofeces they produce. Moreover, the refugia provided amongst mussel colonies for macroinvertebrates such as *Mysis* and *Diporei* would facilitate important food sources for planktiverous fish like alewife (*Alosa pseudoharengus*), which in turn are significant food items for salmonids. This prognostication has been partially supported by a study conducted by Pothoven & Nalepa (2006) which found that large Lake Huron whitefish (*Coregonus clupeaformis*) prey

heavily on quaggas, accounting for 54% of the diet. The consequences of this diet shift may be cyclical in nature. In the short-term, however, commercial success of the lake whitefish fishery appears to be thriving. More recently, the Great Lakes Science Center has concluded that dreissenids have had a positive impact on parts of the Great Lakes ecosystem, providing native wildlife ample zebra mussels to prey upon (USGS, 2008). Due to the quagga's highly efficient ability to filter water in mass quantities, it has been intentionally stocked in lakes in the Netherlands as a management tool to remedy poor water quality (Reeders & Bij de Vaate, 1990).

In contrast to the apparent benefits the introduced dreissenids provide in the Great Lakes ecosystem are potential human health ramifications. The ability of the zebra mussel to biomagnify metal and organochlorine contaminants may serve as an efficient method to transfer contaminants to higher trophic levels (Mersch & Pihan, 1993). In addition, cyanobacteria blooms in Great Lakes basins supporting large mussel populations have been reported, further placing the public at risk in recreational areas (MacIsaac, 1996). Researchers admitting to benefits from dreissenid presence in some situations also realize the potential deleterious effects in regards to the suppression of zooplankton, a vital food source throughout the trophic ladder (Dermott & Munawar, 1993; MacIsaac, 1996; Mills et al., 1996; Mills et al., 1993).

The amount of uncertainty exhibited by the presence of *D. bugensis* and *D. polymorpha* in the Great Lakes is complex. Invasion success in this region has been thoroughly scrutinized with debate as to the apparent costs and benefits the mussels contribute to the ecosystem as a whole. How does research conducted in the Great Lakes

apply to the recent invasion of quaggas to Lake Mead and what amount of variability exists between these distinct watersheds?

Lake Mead. The first documented discovery of quagga mussels in the western United States occurred in January 2007 in Boulder Basin, Lake Mead (Figure 1), presumably from recreational boats brought overland from the Great Lakes (LaBounty & Roefer, 2007). Since the invasion sometime during 2003 or 2004 into Callville Bay (Dr.

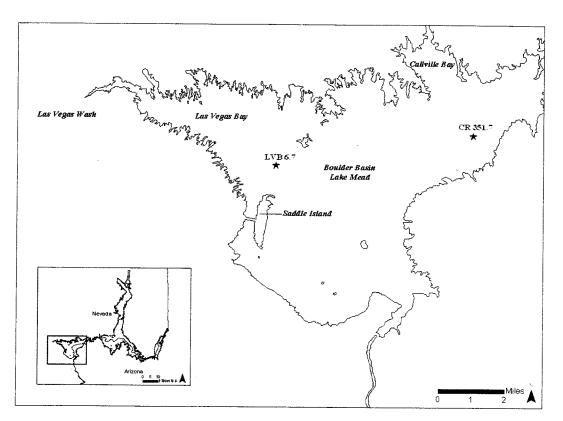


Figure 1. Boulder Basin, Lake Mead.

James LaBounty, personal communication) quagga larvae, or veligers, have multiplied exponentially based on preliminary water quality data analyzed by the Southern Nevada Water Authority (SNWA). At present very little data exists as to the impact quaggas

have had in and around the Lake Mead watershed, especially in regards to ecological impacts associated with trophic level dynamics.

Concentrations of quagga mussel veligers in Boulder Basin increased dramatically from 0.6/L from late February to late March to 126/L by July near Hoover Dam (LaBounty & Roefer, 2007). Water sample data collected as late as fall 2007 showed that veligers made up nearly 40% of zooplankton in the upper 5m of Boulder Basin. Prior to March 2007, no veligers were found in zooplankton samples taken from this location (John Beaver, BSA Environmental Services, Inc., Beachwood, Ohio).

Since the initial arrival of quaggas to Lake Mead, they have spread to various aquatic ecosystems throughout the southwest. In the Hoover Dam area of Black Canyon, quaggas reached highest densities in July 2007 (LaBounty & Roefer, 2007). Depth distribution ranged from the surface to >150 ft, with the highest densites observed at 25-35 ft. The dynamic distribution of quaggas in Lake Mead and their propensity to thrive in a new environment poses a challenge to researchers in attempting to control the invasion.

Veliger Characteristics and Life Cycle

Nichols & Black (1994) found that separating veligers of zebra mussels from those of quaggas is difficult and not always possible. Preshell larvae are virtually identical, owing to either hybridization or geographical variation among the two dreissenids. There are four types of shelled veligers: straight-hinged, umbonal, pediveliger, and plantigrade (Nichols & Black, 1994). Categories are defined based on hinge development, shell size, and the presence or absence of a foot and velum. In laboratory studies, Nichols & Black (1994) observed shell formation on the third day after fertilization at 22°C. Table 1

depicts approximate measurements of common Lake Mead plankton and quagga veligers during the developmental larval stages.

Table 1

Common plankton and developing quagga veligers present in Lake Mead depicting size comparisons

Category	Approximate Measurement of Organism
Quagga Mussel	
Straight-hinged larva	e 39-71µm
Early umbonal larvae	39-71µm
Older umbonal larvae	e 120-221μm
Pediveligers	150-228µm
Plantigrade veligers	222-410µm
Diatoms	
Golden-brown algae	10-100µm
Rotifers	0.15-1mm
Crustaceans	
Cladocerans	0.4-1.5mm
Copepods	2mm
Ostracods	2-3mm

The straight-hinged shell is the first type developed and is characterized by a D-shape, appearing translucent under the microscope and a foot or velum may or may not be present. The length at this D-shelled stage measured between 39  $\mu m$  and 71  $\mu m$ ,

respectively (Nichols & Black, 1994). As the veliger matures to the late plantigrade stage, it will typically measure between 222-410  $\mu m$ . An important distinction to make is that larval size is subject to local variation due to differences in brood-stock condition (Bayne et al., 1975). As the veliger metamorphoses, it gradually resembles an adult bivalve mussel.

## Lake Mead Energetics

## Trophic Interactions

Prior to the introduction of quaggas into the Lake Mead watershed, zooplankton abundance exhibited ideal ecological dynamics among taxa in Boulder Basin during the study period 2001-2004 (LaBounty & Burns, 2005). Data revealed that during 2003 zooplankton abundance were twice that of any other year in the investigation.

Concentrations of copepods and cladocerans both peaked in the spring of 2003. The significance of high zooplankton occurrence relates to success at higher trophic levels.

As a primary food source for threadfin shad (*Dorosoma petenense*), abundance of zooplankton during this time period in Boulder Basin provided the sport fishery with a boom-cycle of striped bass (*Morone saxatilis*) (LaBounty et al., 2004). The authors came to the conclusion that the presence of larger game fish is evidence that the food chain is healthy in an aquatic ecosystem.

Selected SNWA water quality sample sites in Boulder Basin include LVB6.7 and CR351.7 as shown in Figure 1. Zooplankton abundance (#/L) from April 2000 to October 2007 is shown in Figure 2 for weekly collection site LVB6.7. Taxa included in the data set were ostracods, rotifers, copepods, cladocerans, and veligers. The 2001 to

spring 2003 peak in copepods and cladocerans is an example of a desirable ecological balance because every phase in the trophic ladder was timed perfectly to coincide with growing season progression (LaBounty & Burns, 2005). The subsequent crash in zooplankton populations seen in June 2003, as depicted in Figure 2, is an expected natural event due to predation by shad—a healthy trophic level boom-bust cycle in the ecosystem (LaBounty & Burns, 2005).

Population modeling of zebra mussels in the Hudson River drainage suggests

# Zooplankton LVB6.7 B Ostracods S Veligers 160 Copepods 140 B Cladocerans 120 100 80 60 40 20 0 Ref B C C Ref B C Ref

Figure 2. Zooplankton density in Boulder Basin, Lake Mead (LVB6.7), April 2000 to October 2007. Source: SNWA.

uncertain variability when a new invasion has occurred (Strayer & Malcom, 2006).

Different populations of zebra mussels appear to follow different long-term trajectories including boom-bust cycling, stability, and irregular fluctuations (Strayer & Malcom, 2006). The consequences of quaggas exhibiting these uncertain population dynamics in

Lake Mead may rely on multiple factors—most notably temperature and biological adaptation over the long-term. Relatively predictable trophic interactions prior to the arrival of quaggas into this ecosystem could be significantly altered if quaggas show no distinct pattern in abundance spatially or temporally.

The onset of quagga veligers in the Lake Mead ecosystem threatens to alter food web dynamics. Figure 3 shows zooplankton concentrations together with the onset of veligers from the same location in Boulder Basin (LVB6.7) from January 2007 to October 2007. The data continues to be updated from the SNWA as it is collected (Warren Turkett, personal communication).

# Zooplankton LVB6.7 70 60 50 20 © Ostracods © Veligers © Rotifers © Copepods © Cladocerans

Figure 3. Zooplankton densities along with introduced quagga veligers from Boulder Basin (LVB6.7), January 2007 to October 2007. Source: SNWA.

Zooplankton abundance shows an increase from the inflow of Las Vegas Wash when flow from the Colorado River is below average (LaBounty et al., 2004). This is significant in that Las Vegas Wash drains into Las Vegas Bay, which in turn drains into Boulder Basin. As the drought and less than ideal runoff from the Colorado River continues, the exponential growth of veligers in the watershed could threaten the cyclical nature of the planktonic community.

The Boulder Basin sampling location of LVB6.7 used to depict trends in zooplankton data in Figures 2 and 3 is one of many sites the SNWA samples for water quality. For comparative purposes, CR351.7 (Figure 1) is located farther east in Boulder Basin and has experienced similar trends in veliger abundance (Sampled monthly). Figure 4 shows veligers at CR351.7 peaking at concentrations >90/L, as opposed to >70/L on the western end of the basin (LVB6.7).

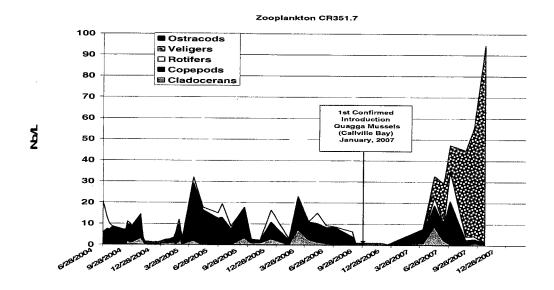


Figure 4. Zooplankton and veliger abundance on eastern side of Boulder Basin, Lake Mead (CR351.7) Colorado River inflow, June 2004 to October 2007. Source: SNWA.

The suspected initial invasion of quaggas found in Callville Bay (Figure 1) in

January, 2007 is depicted to show proximity to the CR351.7 sampling site and planktonic dynamics pre and post introduction. Trends in these two data sets

may suggest variance in trophic energetics caused by differing inflow parameters—

Las Vegas Bay inflow as opposed to Colorado River inflow, respectively.

Stable Isotope Analysis

Since carbon isotopic compositions of animals correspond closely to their food sources, carbon isotopic ratio is expressed by the following equation:

$$\delta^{13}C(\%e) = [(^{13}C/^{12}C)_{sample}/(^{13}C/^{12}C)_{standard} - 1] * 1000$$

where % is parts per thousand and  $^{13}\text{C/}^{12}\text{C}$  are atomic ratios of the number of atoms in the sample or standard (Mitchell et al., 1996). Due to the difficulties involved with observational data when interpreting diet and trophic interactions, useful insights can be gleaned from the stable isotopes of C and N in the feeding relationships of fish within a given food web (Mitchell et al., 1996; Gu et al., 1994). Current feeding by threadfin shad and young-of-the-year striped bass in Lake Mead will provide basic ecological information that may be used by fisheries managers since the introduction of quagga mussels in the watershed. Stable isotopes  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$ , analyzed together; serve as a valuable tool in determining energy sources contributing to trophic position in fish species (Vander Zanden et al., 1999; Gu et al., 1994).

As alluded to previously, stable isotopic composition of N also plays an important function between diet and trophic position. Preferential catabolism and excretion of the lighter form of  $^{14}N$  results in increased  $\delta^{15}N$  values for each trophic transfer along a food

chain (Peterson & Fry, 1987; Cabana & Rasmussen, 1994). Utilizing the properties for N discussed, isotopic analysis of nitrogen is expressed by the formula:

$$\delta^{15}N(\%) = [(^{15}N/^{14}C)_{sample}/(^{15}N/^{14}N)_{standard} - 1] * 1000$$

as with the equation for determining  $\delta^{13}C$ , the atomic ratios are expressed based on international standards for C and N in the Earth's atmosphere and PeeDee Belemnite, respectively (Mitchell et al., 1996).

## Seasonal Variation

Historical zooplankton density in Boulder Basin of Lake Mead has fluctuated in part by epilimnion temperature shifts on a seasonal basis. Background water temperatures revealed that the 10-year average was about 20°C from 1994-2004 (LaBounty & Burns, 2005). The average high water temperature for this data set was greatest at 28.5°C in 1998 and lowest at 27.5°C in 1999. LaBounty et al., (2004) reported highest population densities of zooplankton in the inner bay during April and May during the 1990-95 data set.

Trends in abundance were similar for the inner and outer bays, regardless of the April-May densities. Peaks in zooplankton in the outer bay occurred in both the May-June and September-October time periods (LaBounty et al., 2004). During the warmest months of July and August, the data showed a shift of zooplankton to deeper thermoclines, presumably due to high water temperatures near the surface.

Data on quagga mussel tolerance to warmer climates of North America is lacking.

Zebra mussel densities studies by the Army Corps of Engineers in El Dorado Lake,

Oklahoma from 1998 to 2005 showed veliger spikes in June and October. It was

observed that temperature was a key variable in monitoring of veligers in this region. In

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the past two years of monitoring, however, zebra mussel densities have been dramatically reduced, possibly due to hot, dry summers in the El Dorado Lake area. It has been suggested that zebra mussels may spawn throughout the summer months if water temperatures remain <30°C and may acclimate to temperatures >30°C over time (Laney, 2008).

From the 2001-04 data sets for Boulder Basin, copepods, cladocerans, ostracods, and rotifers for the most part peaked in spring and exhibited similar transitions to deeper water from July-August (LaBounty & Burns, 2005). The explosion in zooplankton populations during the spring of 2003 (see section on Trophic Interactions) greatly influenced the analysis during the four year study (LaBounty & Burns, 2005). The crash of zooplankton observed in June 2003 and lasting the rest of the summer months correlates with high predation rates by threadfin shad, especially in areas of the lake where shad densities are high (Pelle, 1989).

Background limnology of Boulder Basin preceding the introduction of quaggas to the ecosystem serves two functions for this study. First, the data provides an insight as to how primary producers like zooplankton have affected the food chain prior to the quagga arrival and second, the data will provide the ability to compare pre and post mussel invasion in regards to trophic level interactions.

## Threadfin Shad

It has been documented that threadfin shad (*Dorosoma petenense* (Gunther)) are a crucial element to the food web in Boulder Basin of Lake Mead (LaBounty et al., 2004). They constitute the primary food source for piscivorous game fishes such as largemouth

bass (*Micropterus salmoides*) and striped bass (*Morone saxatalis*), which are the most abundant game fish in the Lake Mead watershed (Miller, 1950; Haskell, 1959; Miller, 1961; Deacon et al., 1972; LaBounty et al., 2004).

Shad were first introduced into Lake Mead, Mojave, and Havasu in 1954-55 and were found throughout the lower Colorado River system by 1956 (LaRivers, 1962). Their rapid dispersal in a short time can be partially attributed to high fecundity rates (Pelle, 1989). Threadfin shad generally occupy the pelagic zone, or open water, of freshwater lakes and are native to the southeastern U.S. (Moyle & Cech, 1982). Pelle (1989) observed in the Overton Arm, Lake Mead that > 75% of shad occupied the top 1m of the water column with an exponential decrease in abundance with depth, as well as size range.

Threadfin shad are known to remain in constantly congregated schools, which explain a regularity of food types in multiple samples (Haskell, 1959). Under controlled conditions, threadfin shad were observed to both filter feed and particulate feed with vision not being a factor except in particulate prey selection (Holanov & Tash, 1978). In addition, it has been noted that shad can be attracted to and sometimes make filter feeding movements near water devoid of suspended matter, but previously inhabited by zooplankton (Holanov & Tash, 1978).

Diet Preferences. In the past, researchers familiar with Lake Mead have considered zooplankton and phytoplankton as the primary forage of threadfin shad (Deacon et al., 1972). Moreover, the observation was made in Lake Mead that when preferred plankton was unavailable, shad selected alternate forage with less nutritional value, such as plant debris, sand or detritus (see also Haskell, 1959 for similar findings in shad from central

Arizona lakes). Generally, diet composition of juveniles or adults was more dependent on availability of forage types (Minckley, 1973).

Reports of dramatic declines in zooplankton abundance have been observed in parts of Lake Mead where there are high shad densities (Wilde, 1984). This in part explains fluctuations in shad populations in Boulder Basin over approximate 5-yr boom/bust cycles directly associated with zooplankton abundance (LaBounty & Burns, 2005). The occurrence of periodic algal blooms, like the one in 2003, aid in returning the predator-prey relationship back into equilibrium. The relative balance once observed in foraging activities among threadfin shad in Lake Mead before the introduction of quagga mussels is potentially under threat of permanent alteration due to the voracious invasiveness of this species.

Stomach Anatomy and Function. Internal anatomy of the threadfin shad is characterized by a stomach that is an enlargement of the alimentary tract. The stomach is short and muscular and resembles the gizzard of a fowl (Miller, 1950). Often unidentified organic material found during analysis is attributable to the supposed grinding action of the alimentary organ (Haskell, 1959).

## Hypotheses

This study attempts to answer several questions regarding changes in the Lake Mead watershed related to the introduction of quagga mussels. The first hypothesis under investigation is to conduct a visual estimation of stomach contents of threadfin shad to determine if they eat quagga mussel veligers. If it is observed that a significant proportion of veligers are present in stomach contents, do they serve as a viable food

source? A second question that will attempt to be answered is where shad are feeding in Las Vegas Bay and Overton Arm since the arrival of quagga mussels? It is hypothesized that changes in zooplankton assemblages, as observed in other invaded waterways, will cause an energetics shift in patterns of shad foraging behavior since the discovery of quagga mussels in Lake Mead. Therefore, stable isotope analysis will reveal that, over time, food sources for threadfin shad have shifted from a predictable, pelagic feeding strategy, to a deep water benthic signal, owing to a consequent reduction in available food items due to quagga mussel impacts. A third hypothesis is that trends in historic larval shad trawl data have declined as a result of the presence of quagga mussels in the Lake Mead ecosystem. Finally, with the addition of quagga mussel veligers to the system, the composition and abundance of zooplankton has been reduced as veligers have increased, adversely affecting primary production dynamics.

## CHAPTER 3

## **METHODOLOGY**

## Collection of Data

## Field Collection Methods

Adult Threadfin Shad. In an attempt to examine the foraging habits of lower trophic level fish, a total of 197 adult threadfin shad were collected between April 2008 and February 2009 with the assistance of the Nevada Department of Wildlife (NDOW) and Bio-West, Inc. Of the total number of fish collected, 170 came from Las Vegas Bay and a representative sample of 27 were collected from Overton Arm, where quagga veliger numbers have yet to establish in large numbers.

Standard methods entailed setting gill or trammel nets overnight and returning next day for collection of larger fish. All nets were of variable mesh size, characterized by 3 inch at one end, down to 1-1.5 inch at the other. Dates for gill or trammel netting collections were dependent on the availability of NDOW and Bio-West resources. Fish collected through standard protocols were immediately fixed in 10% CaroSafe<sup>TM</sup> in the field in a container large enough to preserve multiple samples (ASTM, 1995; Haskell, 1959; Pelle, 1989).

Upon completion of field collection, samples were immediately transported to the laboratory for the purpose of measuring physical characteristics of each specimen and maintaining the integrity of the stomach contents. Standard length, total length, and

weight of whole fish were recorded for growth data analysis related to trophic interactions among fish and consumption of veligers (Krebs, 1999).

Larval Shad Trawls. Nevada Department of Wildlife (NDOW) shad trawl protocol consists of towing a cone-shaped net 6m long with a 10 inch collecting bucket on the end. The open end is 1m in diameter with a 1.6mm mesh screening. The net is towed approximately 20m behind the boat in 10 minute increments at a boat speed of 1,000 rpm and replicated three times to provide an average number. Flow volume is calculated using a flow meter at the mouth of the net in order to record water movement. Upon return to the laboratory, fish are counted and abundance converted to fish/100m<sup>3</sup> of water. Since fish >20mm tend to avoid capture in the net, the technique is an estimate of reproductive success and not a population estimate of threadfin shad (NDOW, 2006). Typical trawl monitoring by NDOW is carried out weekly in late spring and/or early summer. Data gathered for this study was collected in June, 2008 on a weekly basis in Las Vegas Bay and Overton Arm, respectively.

Figure 5 shows approximate locations of annual larval shad trawl sampling sites in Overton Arm and Las Vegas Bay, Lake Mead conducted by NDOW. Overton Arm sites are based on a Lake Mead fertilization study conducted in 1988 by Vaux and Paulson (1990) and were named F1, F2, F3, and F4, respectively. NDOW chose Las Vegas Bay sampling locations based on the Vaux and Paulson study protocol and are named ILV (Inner Las Vegas Bay), MLV (Middle Las Vegas Bay), and BB (Boulder Basin). All fish collected for this study were based on sampling locations shown in Figure 5. Regional sampling from Las Vegas Bay and Overton Arm was established in order to gather data

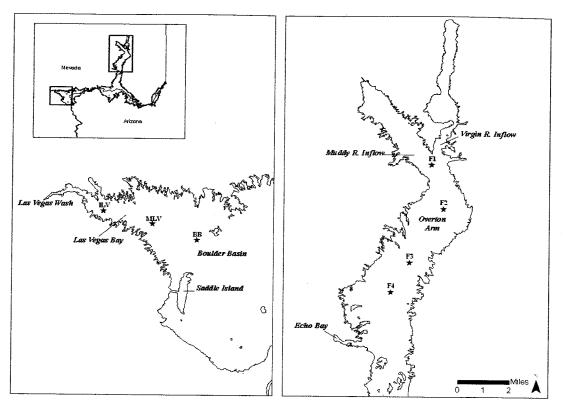


Figure 5. NDOW shad trawl sampling locations in Las Vegas Bay and Overton Arm, Lake Mead since 1988 (Vaux & Paulson, 1990; NDOW, 2006).

which compares fish diets from areas documented as containing representative foraging food with large volumes per liter of quagga veligers (Las Vegas Bay) and, conversely, relatively smaller volumes per liter of quagga veligers (Overton Arm) in the water column where shad and striped bass feed in the Lake Mead watershed.

## Stomach Extraction Procedures

Upon completion of length-weight recording the gut contents from each fish were removed from the esophagus to the anus and preserved in 10% CaroSafe™ individually in Whirl-Paks™ in order to minimize possible degradation of the stomachs (Blanco et al., 2003). Samples were archived after extraction under refrigeration until stomach contents analysis could be performed (Pinkas et al., 1971). Archived retention time varied from a

maximum of ten months from collections gathered in April, 2008 to as little as three weeks in the case of samples collected in February, 2009. Each stomach extracted was coded with date, location, and size of whole fish for later statistical analysis.

Stomach Contents Analysis

Microscopic Analysis. Stomach contents were examined individually upon completion of each sampling event (Hyslop, 1980). Each stomach was dissected vertically and the contents placed in a sedgwick-rafter cell with grid lines and suspended in 1ml distilled water (ASTM, 1995). Grid lines on the sedgwick-rafter provide efficiency of counts under the microscope and avoidance of over-estimating prey items. This methodology has been employed for water sample analysis conducted by the U.S. Bureau of Reclamation and the SNWA, respectively (G. Chris Holdren and Alan Sims, personal communication). A semi-quantitative visual estimation of abundance of dietary components for each stomach was recorded using a scale from 1-5. Empty stomachs were assigned a value of 1(0%) with a value of 5 indicating high abundance (100%) (Blanco et al., 2003; Karjalainen et al., 1999; Collares-Pereira et al., 1996). Stomach abundance from this method was incorporated into the statistical analysis.

Frequency of occurrence of three major taxons of zooplankton representative of Lake Mead and quagga veligers were recorded under a stereo dissecting microscope (Carl Zeiss SteREO Discovery.V8, Toronto, ON, Canada) fitted with a cross-polarized light (CPL) source at magnification suitable for positive identification (Johnson, 1995). The utilization of CPL aids in accuracy in counts of veligers due to the birefringent crystalline structure of the calcite in the larval shell (Johnson, 1995).

## Stable Isotope Analysis

Larval threadfin shad samples from June, 2008 annual trawls were donated by NDOW for the purpose of being subjected to stable isotope analysis. Young-of-the-year (YOY) shad were collected from Las Vegas Bay and Overton Arm along pre-existing trawl locations established in 1988 (Figure 5). Two tows from each location were performed in Las Vegas Bay and three tows in Overton Arm, respectively. Collection dates for Las Vegas Bay were conducted weekly for three consecutive weeks in June and Overton Arm collections took place over two consecutive weeks during the same month in 2008.

Five adult threadfin shad were collected from Overton Arm on 12 February 2009 and 21 adults were collected from Las Vegas Bay on 13 February 2009. The mean total length for all samples was 189 mm and mean total wet weight was 59.9 g, respectively. A sample of dorsal muscle from each fish was filleted and placed in a standard microcentrifuge tube and prepared for analysis.

Size-class for isotopic analysis was carried out by separating and measuring the length (mm) of each YOY fish of similar size, with a target number of ten specimens measured for each location and tow (Mitchell et al., 1996). In the case of adult samples, all fish were prepared individually for analysis. All samples were dried at 70°C for at least 24 h and ground into a fine powder using a mortar and pestle. Subsequent dried samples were weighed out between 1.0-1.2 mg and packaged into 5mm x 9mm tin cups. Replicate samples were taken from larval trawl samples and standard reference fish material was included every 20 samples for quality control (Vander Zanden et al., 2006).

Stable isotope analysis was performed using a NA2000 elemental analyzer (EA) at the Las Vegas Isotope Science Laboratory and interfaced to a Delta V Plus mass spectrometer through the Conflo III system. Elemental analysis was carried out using a flash combustion/chromatographic separation technique. The furnace temperature was kept at  $1000^{\circ}$ C; while the reduction oven was  $650^{\circ}$ C. Generated gas from consumption of the samples is carried in a helium stream into a GC column held at  $60^{\circ}$ C. Gases are then separated before being diluted in the Conflo III and passed to the mass spectrometer for analysis. Isotope ratios of  $\delta^{13}$ C are expressed in per mil (‰) notation relative to the VPDB scale, whereas  $\delta^{15}$ N values (‰) are reported relative to air-N<sub>2</sub>. Three in-house standards (ACET, CABG, and CORN) were directly calibrated against six international standards of Graphite (USGS24), Caffine (IAEA-600), Oil (NBS-22), and ammonium sulfate (IAEA-N-1, USGS25, USGS26) in order to create a three-point calibration curve to correct the raw data.

## Treatment of Data

A frequency of occurrence method was employed to analyze stomach contents for the determination of diet composition as described by Hyslop (1980):

$$% F_i = (N_i / N) \times 100$$

where  $F_i$  = percent frequency of prey type i,  $N_i$  = number of predators with prey i in the stomach, and N is total number of shad with stomach contents. Numerical counts of food items were recorded at the division level and included cladocerans, copepods, veligers, or rotifers. Percentage occurrence and percentage abundance of prey items was determined through modification of the Costello method for interpreting stomach contents data

(Amundsen et al., 1996). Prey-specific abundance is represented by the following equation:

$$P_i = (\Sigma S_i / \Sigma S_{ti}) \times 100$$

Proportional statistics were performed to test whether larger shad are utilizing selected prey items in the diet more than smaller shad in sampling locations in Lake Mead. Condition factor (K-factor) analysis was calculated to test for health in the samples collected (Blanco et al., 2003; Olsen & Ringø, 1999).

**K-Factor = W/L**<sup>3</sup>, where W = fish weight (g) and L = standard length (cm)

## **CHAPTER 4**

## RESULTS

## Stomach Contents Analysis

Threadfin shad from Las Vegas Bay ranged in size from 113 to 212 mm TL (mean = 184 mm; n = 170). Shad from Overton Arm ranged in size from 131 to 197 mm TL (mean = 150 mm; n = 27). The means represent all fish collected from the 2008 and 2009 sampling events. Table 2 summarizes the numerical counts of stomach contents from adult threadfin shad collected at each location. Identifiable shad stomach contents

Table 2
Summary of stomach contents collected from 197 adult threadfin shad in Las Vegas Bay and Overton Arm, Lake Mead in spring and summer 2008 and winter 2009.

Las Vegas Bay $(N = 170)$	Overton Arm $(N = 27)$	
N = 164	N = 16	
N = 49	N = 32	
N = 79 46%	N = 23 85%	
N = 91 54%	N = 4 15%	
	(N = 170) $N = 164$ $N = 49$ $N = 79$ $46%$ $N = 91$	(N = 170) $(N = 27)$ $N = 164$ $N = 16$ $N = 49$ $N = 32$ $N = 79$ $N = 23$ $46%$ $85%$ $N = 91$ $N = 4$

contained primarily cladocerans and copepods. Amorphous debris also contributed a significant portion to the total diet. No quagga mussel veligers were observed under cross-polarizing microscopy. Likewise, no rotifers were observed as food items identified at the division level in Las Vegas Bay or Overton Arm. Visual estimation of abundance revealed that 46% and 85% of stomachs contained either food or other ingested contents. A large proportion contained unidentified food items or other debris such as rocks, sand, and plant material. Those stomachs examined from Overton Arm exhibited more debris than those from Las Vegas Bay.

Frequency of occurrence and prey-specific abundance of stomachs which had identifiable cladocerans and copepods are depicted in Table 3. The proportional analysis shown includes only those stomach contents which contained either cladocerans and/or copepods, respectively. Testing among seasons revealed there was no significant difference in the proportion of cladocerans to copepods in shad stomachs for either spring (z = 0.70, P = 0.484) or winter (z = -0.91, P = 0.362) from comparative sampling events in Las Vegas Bay. Testing for seasonality was not performed for Overton Arm owing to a lack of comparison data.

Proportion of cladocerans differed significantly between spring and winter in Las Vegas Bay (z = 2.64, P = 0.008), whereas proportion of copepods counted in Las Vegas Bay stomach contents did not show statistically relevant difference for seasonality (z = 0.58, P = 0.562). Overall, frequency of occurrence in prey types taking seasonality into account is inconclusive at this time.

Table 3

Frequency of occurrence and prey-specific abundance percentages of cladocerans and copepods in the stomach contents of adult threadfin shad in Las Vegas Bay and Overton Arm, Lake Mead. Prey-specific abundance is a modification of the Costello method (Amundsen et al., 1996).

Location	Frequency of Occurrence				
Las Vegas Bay	% $F_{cladoceran} = (38_{cladoceran}/79) \times 100$	=	48.1%		
	$\% F_{copepod} = (24_{copepod}/79) \times 100$	=	30.4%		
Overton Arm	% $F_{cladoceran} = (9_{cladoceran}/23) \times 100$	=	39.1%		
	$\% F_{copepod} = (10_{copepod}/23) \times 100$	=	43.5%		
Location	Prey-Specific Abundance				
Las Vegas Bay	$P_{cladoceran} = (\Sigma 164/ \Sigma 208) \times 100$	=	78.9%		
	$P_{copepod} = (\Sigma 49/ \Sigma 150) \times 100$	=	32.7%		
Overton Arm	$P_{cladoceran} = (\Sigma 16/ \Sigma 38) \times 100$	=	42.1%		
	$P_{copepod} = (\Sigma 32/\Sigma 43) \times 100$	=	74.4%		

Taking prey-specific abundance into account by food type, a significant difference in proportion of cladocerans was observed among shad stomachs from Las Vegas Bay as opposed to Overton Arm (z = 4.32, P = <0.001). Similarly, copepod abundance also exhibited differing proportions between the two regions, with Overton Arm showing higher abundance (z = -5.44, P = <0.001). Of the prey items counted, cladocerans were more prevalent in stomachs of shad collected in Las Vegas Bay while copepods were more abundant in Overton Arm.

A comparison in prey-specific abundance by site was also analyzed. For Las Vegas Bay, cladocerans were significantly more abundant than copepods (z = 9.70, P = <0.001). An inverse relationship was observed in Overton Arm, where copepods were more

prevalent compared with cladocerans (z = -3.10, P = 0.002). These observations suggest the dominant food source for threadfin shad differs by location in Lake Mead.

## Stable Isotope Analysis

Results from bioenergetics analysis in adult threadfin shad revealed little variation in  $\delta^{13}$ C values in Las Vegas Bay for each of the years 2007-2009. Values ranged between -24.6% and -25.9%, as shown in Figure 6. Values for  $\delta^{15}$ N differed in 2009 (11.7%) from trends observed from the 2007 (15.2%) and 2008 (15.9%) data provided by Umek et al.(in press). Stable isotopic analysis suggests that from 2007-2009 threadfin shad in

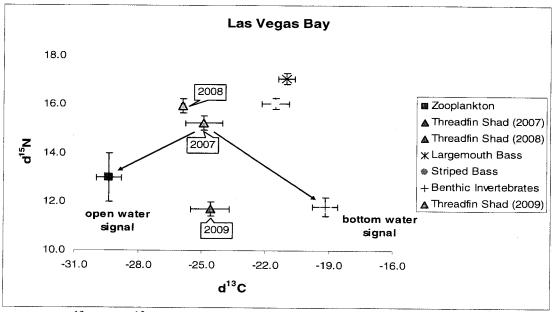


Figure 6.  $\delta^{13}C$  and  $\delta^{15}N$  comparative means for adult threadfin shad sampled from Las Vegas Bay, 2007-2009. Mean SE also depicted. Arrows show initial shad trophic position at the time of quagga discovery in Lake Mead. Zooplankton, bass, and benthic invertebrate data is representative of 2007 only. Data from 2007-2008 were provided by Umek et al., in press.

Las Vegas Bay have not altered their diets significantly towards an exclusively open water or bottom water regime shift, despite the presence of quagga mussels in the system.

Tissue samples analyzed from Overton Arm showed similar values in  $\delta^{13}$ C variability with those from Las Vegas Bay, ranging from -23.1‰ to -26.0‰ over the three-year dataset (Figure 7). Data from 2008 for  $\delta^{15}$ N spiked to 16.1‰. This differs from more consistent values observed from 2007 (12.7‰) and 2009 (12.2‰). A common trend in

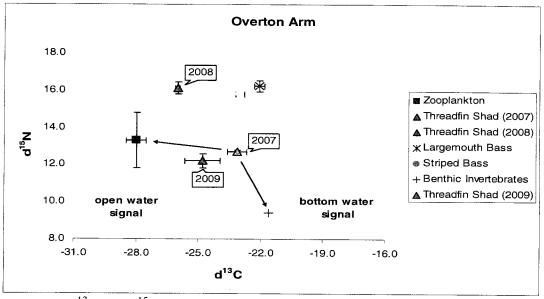


Figure 7.  $\delta^{13}$ C and  $\delta^{15}$ N comparative means for adult threadfin shad sampled from Overton Arm, 2007-2009. Mean SE also depicted. As with Las Vegas Bay data, organisms other than shad are representative of 2007 only from Umek et al., in press.

both Las Vegas Bay and Overton Arm isotopic analysis is that for each sampling location in Lake Mead, year 2008 showed the highest values in both  $\delta^{13}C$  and  $\delta^{15}N$ . The greatest shift occurred among  $\delta^{15}N$  values, with a more predictable pattern observed in  $\delta^{13}C$ .

#### Larval Shad

Results from June, 2008 NDOW larval shad trawls were compared with adult values from the same year (Table 4). Analysis revealed a mean  $\delta^{13}$ C signature of -25.8% for all trawls conducted in spring 2008 in Las Vegas Bay. This corresponds very closely with the adult value of -25.9% during the same year. Comparisons among size-classes in  $\delta^{15}$ N values, however, varied from a mean of 10.1% for larval and 15.9% for adults, respectively. Larval isotopic means were a composite of the inner, middle, and outer transects sampled by NDOW in Las Vegas Bay and thus represented the entire bay.

Table 4

Summarized mean stable isotope results comparing NDOW larval shad trawl samples with adult shad data collected by Umek et al., in press, for year 2008 in Las Vegas Bay and Overton Arm. (L) = Larval shad; (A) = Adult shad.

Location	$\delta^{13}C(L)$	$\delta^{13}C(A)$	$\delta^{15}N(L)$	$\delta^{15}N(A)$	
Las Vegas Bay N = 57 (L) N = 3 (A)	-25.8 % <sub>o</sub> SE = 0.55	-25.9 ‰ SE = 0.09	10.1 ‰ SE = 0.56	15.9 ‰ SE = 0.29	
Overton Arm N = 32 (L) N = 2 (A)	-25.6 % SE = 0.67	-26.0 % <sub>o</sub> SE = 0.11	11.4 ‰ SE = 0.49	16.1 ‰ SE = 0.31	

Similar trends were observed in Overton Arm in regards to isotopic profiles, as shown in Table 4. As was recorded in Las Vegas Bay,  $\delta^{13}$ C assimilation in larval shad (-25.6%) was remarkably close to adult counterparts (-26.0%). This indicates that for both Las Vegas Bay and Overton Arm, important dietary sources of nutrition remain constant regardless of size-class based on carbon isotope measurements (Peterson & Fry, 1987).

A difference in  $\delta^{15}N$  also was evident in Overton Arm in 2008 between larval and adult shad. Again, larval shad exhibited a lower value in nitrogen measurement of 11.4‰, as opposed to adults measuring 16.1‰. These results ( $\delta^{15}N$ ) correlate with smaller shad holding a different trophic position in the food web as opposed to their adult counterparts.

# Trend Analysis

Annual larval threadfin shad trawl protocol conducted by NDOW has remained consistent by location in Las Vegas Bay and Overton Arm since 1988. Figure 8 shows mean number of fish/100m<sup>3</sup> for all three sampling locations in Las Vegas Bay since 1988. No data is available for year 1990 and was omitted. The lowest average count occurred in 1989 at 2.78 fish/100m<sup>3</sup> and the highest was observed in 2007, with an average of 445.56 fish/100m<sup>3</sup>.

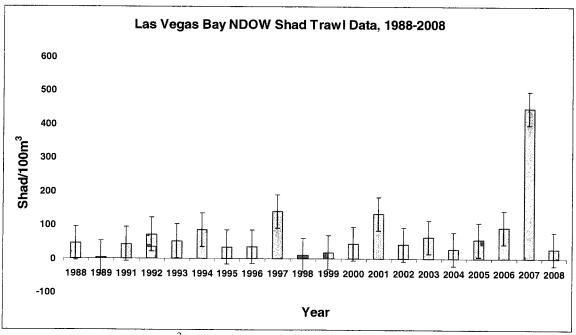


Figure 8. Mean shad/100m<sup>3</sup> from Las Vegas Bay trawls conducted by NDOW, 1988-2008 (No data available for 1990). Data is a composite of all trawling locations in Las Vegas Bay.

Trends in average shad trawl counts for Overton Arm are depicted in Figure 9 for all four sampling locations, with a minimum of 2.73 fish/100m<sup>3</sup> in 2000 and a peak of 256.07 fish/100m<sup>3</sup> in year 2007. Performing a one-way ANOVA of the data by year showed that for Las Vegas Bay, there was no difference among years in mean numbers of shad  $(F_{19,40,0.05} = 1.85$ , where  $F(1.38) < F_{\text{critical}}$  (1.85), P = 0.19). Post hoc analysis using a Least Squared Determination (LSD) t-test for Las Vegas Bay revealed a significant difference among years only for 2007, with no significance observed among all other years ( $\alpha = 0.05$ ).

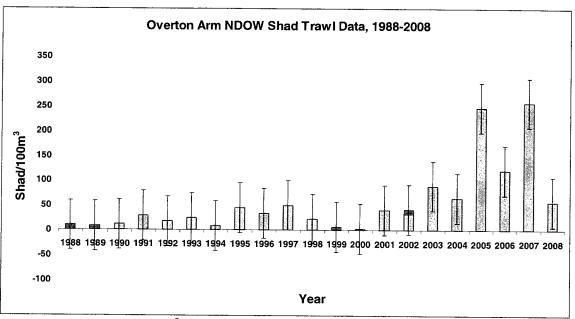


Figure 9. Mean shad/100m<sup>3</sup> from Overton Arm trawls, 1988-2008. Data is a composite of all trawl locations in Overton Arm. Data courtesy of NDOW.

Overton Arm showed a difference between years in the mean number of shad counted  $(F_{20,62,0.05} = 1.74)$ , where  $F(5.02) > F_{critical}(1.74)$ , P = <0.001. Post hoc ANOVA revealed no significant difference between years 2005 and 2007 at the 0.05 confidence

interval. The post hoc analysis LSD t-test for sample years 2005 and 2007 were however, significantly different compared with all other years in the dataset. A separate one-way ANOVA based on trawling locations, as opposed to year, revealed no significant difference in Las Vegas Bay ( $F_{2,57,0.05} = 3.16$ , where  $F(2.73) < F_{\text{critical}}(3.16)$ , P = 0.07). The same was observed for each of the four trawling locations in Overton Arm ( $F_{3,79,0.05} = 2.72$ , where  $F(0.39) < F_{\text{critical}}(2.72)$ , P = 0.76).

Trends in zooplankton biomass were compared with NDOW shad trawls, as shown in Figure 10, for sampling location Inner Las Vegas Bay (ILV) from 2000-2008.

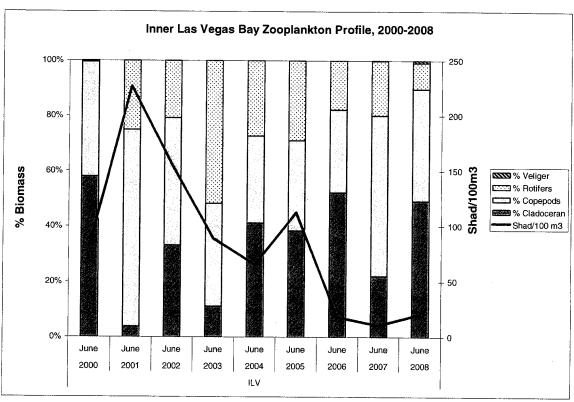


Figure 10. Percent biomass of four divisions of zooplankton compared with annual Juneshad trawl averages from 2000-2008 in Inner Las Vegas Bay sampling location.

Zooplankton data provided by SNWS was not readily available prior to year 2000 and thus was not included in the analysis. Copepod biomass was highest in 2001 comprising over 71% of all zooplankton. Subsequently, mean shad counts peaked with copepod availability during this time at 226.7 fish/100m<sup>3</sup>. Larval shad counts exhibited a precipitous decline through the decade despite predictable variability in prey availability.

Results from the outermost sampling location of Las Vegas Bay, named Boulder

Basin (BB), shows a marked difference in biomass and shad distribution from one side of
the bay to the other (Figure 11). Once again copepods dominated representative

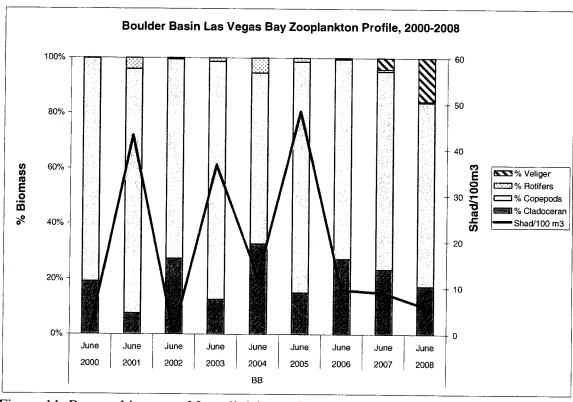


Figure 11. Percent biomass of four divisions of zooplankton compared with annual June shad trawl averages from 2000-2008 in Boulder Basin, Las Vegas Bay, sampling location.

zooplankton, comprising 76% of all zooplankton biomass over the nine-year dataset. Concurrent with copepod abundance was a predictable boom-bust trend in shad for most of the decade. However, from 2006-2008 shad counts/100m³ have declined as quagga veliger biomass has increased. Cladoceran biomass in relation to shad has shown an inverse oscillation from year-to-year which may suggest that shad gain better nutritional value and thus be more dependant on cladocerans such as *Daphnia* spp. than the more historically abundant copepods over this time-frame.

#### **CHAPTER 5**

# SUMMARY, CONCLUSIONS, AND RECOMMENDATIONS

### Discussion of Results

There continues to be a great amount of uncertainty for the future of the Lake Mead fishery following the introduction of quagga mussel. Based on data presented in this paper, the impact on the sport fishery in Lake Mead with the introduction of quagga mussels has yet to be revealed. Due to the complexities involved in aquatic food web dynamics, only time will reveal the true impact quaggas will have on Lake Mead ecology. The discovery in 2008 of gizzard shad (*Dorosoma cepedianum*) in Overton Arm may further complicate matters.

Results from stomach contents analysis of threadfin shad was largely inconclusive based in part on small sample size, especially from Overton Arm collections (N=27). Moreover, method of catch in this study utilized trammel and gill nets of varying mesh size. This factor probably allowed smaller shad to avoid capture in the two and three inch portions of the nets, respectively. The bulk of threadfin shad were located in the smallest one-third of nets, therefore limiting a representative sample. Moreover, results from condition k-factor may not be reliable, considering that smaller shad were most likely under-represented due to the aforementioned variability in mesh size.

Multiple studies have recognized the problem of rapid digestion in fish guts of soft-bodied larval tissue due to mechanical and chemical processes (Hunter, 1981; Folkvord,

1993; Kim & DeVries, 2001; Schooley et al., 2008). Therefore, the ability to efficiently quantify quagga veligers by this method has proved problematic. Although preservation techniques used in this study were adequate for large-bodied cladocerans and copepods, it was not practical for the delicate nature of veligers and may have explained the lack of identifiable rotifers in gut contents of shad as well. Based on previous studies, immediate examination of stomach contents is critical. Use of active capture methods such as electrofishing, angling, or seining could reveal better results than the passive method of capture conducted in this study—trammel and gill netting (Schooley et al., 2008).

Passive methods of capture most likely contribute to shad regurgitation of food contents, which might explain the lack of small-bodied, delicate prey items. The stress induced in threadfin shad while struggling to escape the nets could exacerbate this process.

As is evident with stomach contents analysis, available stable isotope data has yet to reveal quantifiable trends in either trophic position or assimilated dietary items in threadfin shad since quagga mussel invasion. Nevertheless, patterns in  $\delta^{13}C$  for both Las Vegas Bay and Overton Arm have shown remarkably similar signatures over the three-year dataset. This would suggest that quagga mussels may not as of yet had a deleterious influence on quality and/or quantity of available dietary components. The remarkable similarity in values of carbon isotope measurements between differing size-class of shad serves to strengthen this contention.

Results from  $\delta^{15}N$  are more problematic to discern based on the infancy of this investigation. Available nitrogen in the system may fluctuate due to a myriad of factors unrelated to the presence of quagga, shifting trophic position based on other environmental variables or disturbance. Inputs to Las Vegas Bay include nutrients

provided by the Las Vegas Wash, which differ markedly from the Muddy and Virgin Rivers contributing to Overton Arm. Distribution of quagga mussels is not equally representative from each region, furthering to complicate the data.

Background threadfin shad relative abundance gathered by NDOW prior to discovery of quagga mussels is much more substantial in gleaning future impacts of the invasive bivalve. The statistical significance in mean number of shad counted by year in Overton Arm is indicative of fluctuations unrelated to quagga influence. If environmental disturbance coupled with the impact caused by a highly invasive species coalesce, a colossal crash in the food web could occur, irreversibly damaging the Lake Mead fishery. Therefore, the ability to monitor background primary and secondary consumers over time is an invaluable tool for future fishery management initiatives.

The quantification of zooplankton biomass in relation to shad abundance can be compared spatially and temporally over the past decade. Although comparisons from the 1990s are lacking, valuable data is available to compare pre and post quagga invasion from throughout Las Vegas Bay. The observation of high percentages in biomass of copepods, for example, may change dramatically over time as quagga biomass continues to increase, as has recently been observed in the Boulder Basin reach of Las Vegas Bay. Threadfin shad and cladocerans have shown a clear cyclic pattern in abundance in the years leading up to quagga mussel discovery at trawl location BB (Figure 11). This observation suggests an important relationship in food web dynamics between threadfin shad and cladocerans historically. The influence caused by veligers has yet to produce quantifiable data into future perturbations in the shad/cladoceran cycle.

Prior to discovery of quagga mussels in Lake Mead, underestimation of rotifer counts was probable due to standard methodology employed at that time for plankton tows. Since quagga veliger sizes are substantially smaller in comparison with other zooplankton species (Table 1), a reduction in net mesh size of approximately  $20\mu m$  was implemented immediately following quagga discovery in 2007. An underestimation of rotifers prior to 2007 was likely owing to these organisms eluding capture in the larger mesh tow nets. Caution, therefore, must be observed in assessing rotifer biomass pre-2007.

Even though zooplankton data collection has been substantial by multiple agencies in Lake Mead, historical Overton Arm data was not readily available to include in this study. The acquisition of this data would be invaluable in making comparisons in food web interactions for two distinct regions in Lake Mead. The downward trend in shad abundance from 2000-2008 in Las Vegas Bay could be indicative of quagga influence, but it is still uncertain until further data are collected.

### Public Health Considerations

Threats to public health caused by the presence of quagga and zebra mussels have been identified by researchers in the Great Lakes (Fields, 2005). These mussels have the capability of bioaccumulating contaminants like polychlorinated biphenyls (PCBs) or methylmercury at rates 10 times that of native mussels. The result is accumulation of contaminants in sport fish which are commonly consumed by humans, leading to exposure of harmful pollutants known to pose a threat to human health. The high efficiency in which quagga mussels clarify water leads to promotion of increased plant

growth due to increased sunlight. When this new growth dies and decays, oxygen is depleted and provides ideal conditions for the bacteria *Clostridium botulinum* to grow. Similarly, zebra mussels have been linked to increases in blooms of the toxic algae *Mycrocystis*. The proliferation of pseudofeces releases by mussels has provided nutrients that feed the *Mycrocystis*.

Although not necessarily a direct threat to human health, the presence of unwanted aquatic organisms like *C. botulinum*, *Microcystis*, or the proliferation of cyanobacteria blooms do threaten water quality and promote toxins which, when bioaccumulated, pose serious concerns for those consuming contaminated fish. If it can be determined over time that threadfin shad utilize quagga mussels as a food source, the consequence could be bioaccumulation of toxins which could alter the food web forever in Lake Mead. The results observed between the round goby (*Neogobius melanostomus*) and quagga/zebra mussels by Great Lakes researchers is an example of such a deleterious food web interaction (Rutzke et al., 2000; Fields, 2005).

# Conclusions and Recommendations for Further Study

The impact on the Lake Mead sport fishery owing to the quagga mussel introduction is uncertain. Since little is known as to how quagga mussels will adapt to the desert southwest environment, only time will tell the true impact. This study attempted to create a baseline food web profile using threadfin shad as the key constituent in the future viability of a popular bass fishery. Included in the analysis was the attempt to inventory food items available to shad in two areas of Lake Mead—Las Vegas Bay and Overton Arm.

Research was conducted through stomach contents analysis and supplemented by stable isotope analysis. The goal being to identify whether shad could be utilizing quagga mussels as a dietary item, either directly through visual estimation of gut contents, or indirectly through the transfer of trophic energy through the aquatic ecosystem. Although visual estimations were lacking, shad may utilize mussels as part of the diet. Stomach contents analysis, however, may not provide an efficient or practical means of determining this end. Finally, a comprehensive database was developed which contains historical larval shad abundance and zooplankton biomass calculations which can be used in the future to monitor impacts caused by quagga mussels as it relates to fishing interests.

Results were inconclusive whether threadfin shad have the ability to assimilate quagga mussel veligers based on stomach contents analysis. Preservation technique remained consistent; however, the amount of time in which extracted stomachs remained archived may have led to considerable degradation of specific food items. In some cases, stomachs were analyzed >10 months after field collection concluded. Furthermore, the fragile morphological features of quagga veligers probably led to advanced chemical and mechanistic degradation even before each stomach was removed.

After performing stable isotope analysis on larval and adult threadfin shad post quagga mussel introduction, results were more telling. Based on comparison data from Umek et al., in press, larval and adult  $\delta^{13}$ C profiles varied very little (Table 4). This indicates the suspected shift towards a more benthic diet has not occurred with the coinciding presence of quagga mussels in either Las Vegas Bay or Overton Arm. Predictably,  $\delta^{15}$ N signals differed between larval and adult stages in threadfin shad.

Comparing life-stage data from 2008 suggests that larval shad hold a different trophic position than adult counterparts based on nitrogen content in the system. Overall, it appears quagga mussels have yet to influence a shift in shad energetics away from a pelagic, open water feeding strategy based on food availability constraints.

Results of stable isotope analysis from 2007-2009 were not uniform across seasons. Data gathered from Umek et al. (in press) was a combined summary of adult shad collected from March through November in both Las Vegas Bay and Overton Arm in year 2007. Conversely, data from 2008 was gathered from March only from the two lake regions under study. Threadfin shad samples from both areas in 2009 were collected in February. Annual comparisons regarding stable isotope signatures in the future would be more valuable if they were compared based on individual season, respectively. A seasonal analysis would provide a better understanding of food web dynamics by determining if there is an observed shift in  $\delta^{13}$ C and  $\delta^{15}$ N profiles from one season to the next. Environmental and ecological factors such as water temperature or diel patterns most likely fluctuate greatly depending on season, causing shad to shift feeding strategies accordingly.

This study was the first to develop a zooplankton biomass index specifically for Lake Mead for comparison with larval shad trawl data over time. Data was compiled from multiple sources to show the relationship of how relative shad abundance has decreased as a result of increasing quagga veligers. Although zooplankton data was not available for Overton Arm, comparisons could be made from shad trawls conducted in Las Vegas Bay from 2000-2008 (Figures 10-11). The historic record has shown a decline in mean shad counts from Inner Las Vegas Bay (ILV) and Boulder Basin (BB) sampling locations

since quaggas were identified. Copepod biomass coincided with peak shad numbers from one end of Las Vegas Bay to the other pre-quagga presence. In each of the two years quagga mussel biomass has increased at BB, a subsequent decline in shad has been recorded. It is too soon in the investigation; however, to conclude the decline in shad is related to quagga mussel increases, especially since copepod biomass has remained high throughout the decade. In summary, since abundance in quality dietary items such as copepods and cladocerans have remained proportionally similar pre and post quagga mussels, there is no evidence to suggest quaggas are impacting threadfin shad or zooplankton dynamics negatively at this time.

Since quagga mussel invasion is a recent phenomenon in this region of the U.S., many protocols and research needs have yet to be developed. One recommendation which should be investigated is the need to understand the assimilation of quagga veliger once it enters the stomach of the threadfin shad. The soft-bodied, miniscule properties of this organism make it difficult to identify. Studies related to degradation over time could be administered in a controlled environment in an attempt to evaluate the future plausibility of conducting gut content analysis in the targeted search for veligers.

When conducting field collections of adult shad, regardless of what hypotheses are set forth, using the appropriate net is critical. Whenever possible, utilizing a one inch mesh trammel or gill net maximizes total catch, enhancing sample sizes which have frequently hindered ecological studies.

Establishment of a quagga mussel stable isotope signature would greatly enhance the ability to recognize how the food web is structured in the context of other organisms in the ecosystem—specifically zooplankton, shad, and bass. Similarly, a possible

application to veliger detection in fish would be to incorporate Polymerase Chain Reaction (PCR) techniques employed to easily and efficiently detect the presence or absence of veligers, which recently have been used to detect veligers in plankton samples (Frischer et al., 2002; Livi, et al., 2006). The development of such technology would not eliminate the need to perform tedious stomach content analysis for abundance purposes, however. A field device that quickly identifies molecular composition and DNA sequencing would expedite research in the field of quagga mussel ecology in Lake Mead.

As a species of concern in Lake Mead, an expansion of research on the endangered razorback sucker habitat requirements should be considered. As quagga mussels are voracious filter feeders, able to deplete nutrients and reduce turbidity in the water column, an indication of sport fish health could be gleaned from razorback success or failure. Larval survivability could decrease with increased water clarity, leading to increased predation by species like striped bass in the short-term. Long-term consequences might include like-wise effects on the sport fish population.

Perhaps most critical to further study into influences caused by quaggas is continued monitoring and data collection post-invasion. Since quaggas were first discovered in 2007, very little data is available to gauge long-term impact. Larval shad trawls conducted by NDOW, coupled with zooplankton collections by the SNWA and USBR must continue if trends are to be determined. Ongoing stable isotope profiles in Las Vegas Bay and Overton Arm performed in this study, as well as others, need to continue in order gain insight into changes in energetics among primary and secondary consumers over time.

Long-term monitoring goals for Lake Mead based on quagga mussel influence need to be implemented if the sport fishery is to be sustained. Mapping spatial variability of quagga mussel veligers in Lake Mead needs to be established to track movements. In order to accomplish this goal, many variables need to be taken into consideration. Monitoring efforts should be geared towards potential seasonal migrations, either to different locations in the lake or through different zones throughout the water column. Finding a discernable pattern in quagga mussel movements would give researchers the much needed ability to understand the biology of quagga mussels in the arid southwest over time and how they may be evolving to fit the Lake Mead niche.

Fortunately, several agencies have been gathering critical Lake Mead biological data for at least a decade and even longer. Continued efforts in assessing recruitment of larval threadfin shad by NDOW must continue in historic sampling locations if any information is to be gleaned from quagga mussel impacts in the future. The addition of a more comprehensive trawling program in reaches other than Las Vegas Bay and Overton Arm should also be considered, especially in areas of Lake Mead where threadfin shad consistently congregate based on historic NDOW knowledge. The expansion of shad trawls would add significant data to an already comprehensive historical record into the primary food source for valuable species of game fish such as striped bass and largemouth bass.

Implementation of sampling stations specifically designed to monitor quagga mussel abundance should be spread lake-wide. Monitoring quagga mussel population dynamics is not only salient to the future of the Lake Mead fishery, but also to infrastructure and potential public health threats. Expansion of existing water quality and plankton

monitoring stations carried out by agencies such as the SNWA or USBR would expedite valuable biological information as Lake Mead learns to function in the presence of this highly invasive organism.

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