Analysis of the diet between an invasive and native fishes in the Peruvian Amazon. Anthony Mazeroll AMazeroll@soka.edu

Introduction

Non-native species are the second greatest threat to global species biodiversity after land development (Vitousek, D'Antonio, Loope, Rejmanek, & Westbrooks, 1997). Due to the magnitude of this threat, it is vital that the impacts of exotic species are understood. Fish biodiversity is especially threatened by the ecological changes caused by non-native species (Gozlan, Britton, Cowx, & Copp, 2010). Having higher species diversity allows more ecological processes to take place, protecting the resources of that system, and making it more resilient to change (Folke, 2006). Adding outside inputs, such as additional non-native species, into an ecosystem can have beneficial or compromising effects depending on the amount and type.

Invasive species have been shown to reduce native species richness and diversity of native organisms. Some have argued that the transportation of species from one ecosystem to another is actually beneficial for diversity, and non-native species are not really an environmental "problem". Introductions of non-native species increase diversity at a local level because in the short term there will be a lag time where both non-native and natives can coexist (Lodge, Stein, Brown, Covich, Bronmark, Garvey and Klosiewski, 1998).

Species entering a new ecosystem have to pass through a gauntlet of barriers before they can become established. The usual progression of invasion is: transportation to the new ecosystem, initial establishment, spread to a larger region, and then naturalization into the new community (Marchetti, Light, Moyle, and Viers, 2004). Most importantly, the majority of

invasions are unsuccessful (about 10% due to resource partitioning; Bohn, Amundsen, and Sparrow, 2008) and it may be through repeated introduction over long time periods that a population establishes itself (Sax and Brown, 2000). Success of invasion is determined by the naturalization (establishing a self-sustaining, naturally reproducing population; Falk-Petersen, Bohn, and Sandlund, 2006) Moving fish from one region to another, even within the same country can have dire consequences for biodiversity. In Brazil when two native species where moved from one watershed to another, to stock lakes and improve fishing in the area, species diversity and richness in the new watershed decreased significantly (Latini and Petrere, 2004).

One example of a non-native species taking advantage of multiple transportation vectors is the zebra mussel (Dresissena polymorpha) which are native to Russia and is known for its high rate of reproduction, genetic plasticity, and economic as well as environmental damage (Ludyanskiy, McDonald, and MacNeill, 1993). Arriving in ballast water from foreign ships, the zebra mussel quickly spread throughout most of the United States watersheds in a relatively short amount of time. It was able to do this because at each stage in the mussel's life it is able to easily move into new habitats. As a free-swimming larva, it can be easily carried through a watershed into a different location, forming a new colony and thus creating a new launch point for invasion. In its adult stage, it fixes itself to hard surfaces and from there can either clog water intake pipes for power plants or be easily transported from one watershed to the next via recreational water vehicles. Due to its genetic plasticity, the zebra mussel can survive in most aquatic environments, and then continue to spread. Zebra mussels degrade ecosystems by filtering out a significant portion of phytoplankton, thus reducing the amount of available food for young fish. They also impact other invertebrates by fixing themselves to their shells restricting movement, and in the case of other bivalves by crushing them and preventing them

from opening (Ludyanskiy, McDonald, and MacNeill, 1993). The destruction caused by the zebra mussel is uncommon; the majority of non-native species have unobservable or negligible impacts on their new ecosystems.

Given the current understanding of biology and evolution, a foreign invader should never be able to displace well-adapted native species. One popular term coined by researchers is the "paradox of invasion" (Sax and Brown, 2000). How does a relatively un-adapted new comer drive organisms, that have spent at least the last several thousand years adapting to an environment, to the point of extinction? One of the reasons, in aquatic systems, is that bodies of freshwater tend to act like biogeographic islands. The species in the water cannot leave, and most other species are prevented from entering the system. As a result, ecosystem niches can be left unexploited, leaving opportunities for invading generalists to fill in the gaps. Often the unexploited niches are predatory ones and a new invader may not have any predators in the region, or become the newest predator in a region historically absent of them (Sax and Brown, 2000).

Species can become threatened by an increase in competition for a specific food resource. In areas with diverse and plentiful food resources this is often not a problem, but in increasingly disturbed or isolated areas, resources can become scarce, and even scarcer by adding additional species. Usually the non-native does not have a natural ability to consume resources more efficiently; instead it has some other ecological aspect that allows it to outcompete native species. For example, when mosquito fish (*Gambusia affinis*) were introduced to Australia for mosquito control, they caused a decline in native rainbow fish species because they are more aggressive competitors than the rainbow fishes and resort to fin nipping to drive other species away from prey. Additionally, mosquito fish prey on similar food resources as native fish, changing the

ecological composition by decreasing the total size of prey items through selective feeding. This reduces the amount, and size of food items available for native fishes (Arthington, 1989).

Competition can also add additional ecological pressures on an already stressed population, assisting in its decline. For example, in Europe, competition from non-native brook trout (*Salvelinus fontinalis*) and brown trout (*Salmo trutta*) has displaced the native rainbow trout (*Onchorhynchas mykiss*) through competition for prey items. In addition, this new competition significantly reduced native invertebrates and amphibians as well (Vitousek, et al., 1997).

Predation of native species is the most visible and direct form of biodiversity decline. In Lake Victoria, Africa, Nile perch (*Lates niloticus*) were introduced for aquaculture and within a few decades the perch had escaped and decimated the over 500 species of native fish in that lake. Lake Victoria is an isolated rift valley lake that lacked large powerful predators such as the Nile perch, and by placing them in an environment with no other predators, it was easy for it to dominate the food chain (Goldschmidt, 1996). Now, the entire composition of the lake is different, the small fish that were removed by the Nile perch were believed to eat phytoplankton and algae. Because those fish no longer exist the turbidity of the lake has increased, and parts of the lake experience severe eutrophication.

Habitat degradation can occur when the foraging habits or the behavior of a non-native fish disturbs the environment to a significant degree that it disrupts ecosystem services. One example of this is in the United States where non-native Asian common carp (*Cyprinus carpio*) have been introduced. In bodies of water with carp the turbidity is significantly higher than areas without carp in the same region. As a result, areas with carp have more turbid waters due to their aggressive foraging habits that stir up the bottom layer of sediment. This reduces the ability of native fish to hunt by sight, and thus has reduced their populations in that area (Gozlan, et al.,

2010). Additionally, increased turbidity can reduce the effectiveness of primary production, which can have negative ecosystem wide effects.

Non-native species harm ecosystems by reducing their resilience, or the amount of change an ecosystem can undergo before it begins to have a different form or function (Folke, 2006). It is helpful to measure the effects of non-native species in this way, because the effects might be complex, indirect, or even beneficial. Additionally, it is important to look at ecosystems as having multiple stable regimes instead of a static equilibrium, after all the species may have been in this particular system for many generations. However, if the new species are changing the ecosystem into another state, where it no longer provides its essential functions, then that begins to have negative consequences for human livelihoods as well.

Currently there is much debate over whether non-native species are the direct cause of ecological change or are filling in gaps created by species loss and human development. Plants have shown to be in some cases passengers of disturbance and species loss rather than the direct cause of it (Didham, Tylianakis, Hutchison, Ewers, and Gemmell, 2005). Fishes however have been shown to be both driver and passengers of change, illustrating the complex relationship between human modification of an environment and the species that can persist there (Godinho and Ferreira, 1998). In some cases, non-native fish have been determined to be the best predictor of a decline in native species when compared with other common causes such as development (Hermoso, Clavero, Blanco-Garrido, and Prenda, 2011). More work must be conducted to further prove this relationship, but as of now it is highly likely that non-native fish species are directly responsible for this change.

Once established, non-native species are very difficult to extirpate from an environment and it is time consuming to study a species to determine whether or not it will be invasive. But

how can researchers predict if a species is going to become invasive? There are some general similarities detected about non-native fish that helps to focus research. Families of fish with smaller body sizes tend to establish easier, as well as fish that are genetically similar to native species. More interestingly, some factors such as the date of first introduction, and native species richness are not associated with successful establishment of a population. Species most likely to establish are generally rapid reproducers, generalists, or omnivores who are introduced into isolated environments (Ruesink, 2005). Many scholars have previously believed that the more diverse an ecosystem is, the better it is at deterring establishment. However, this has shown not to be true, tropical biodiverse ecosystems are at great risk, and have much more to lose then less diverse ecosystems (Sax and Brown, 2000). This is especially true for aquatic environments.

Of the 3,120 described freshwater fish species that there is sufficient population data for, 37% of these are threatened with extinction (IUCN, 2009). In the United States since 1890, over half of the fish species that have gone extinct were negatively affected by non-native fish (Vitousek, et al., 1997). More concerning is that non-native fish introductions have doubled in the last 30 years, and the primary method of introduction is through aquaculture (Gozlan, et al., 2010). Aquaculture has become the main vector for non-native dispersion because the globalization of trade has made it much cheaper to raise fish in developing countries and export them to developed ones for sale. As a result, many of these facilities lack the proper equipment to prevent fish from escaping into the wild. Additionally, unlike one-time introductions such as stocking of a lake or accidental releases during transportation, aquaculture facilities can continually release non-native fish, in effect seeding a new population.

Due to the connection between human development and non-native fish, the decline of native fishes has most often occurred in disturbed or polluted habitats. Fish raised for aquaculture are generally more tolerant of poorer water quality, and have been shown to easily reproduce in disturbed waters (Arthington, 1989). Thus, development makes it more difficult for native fish populations to survive and easier for non-native species to establish populations. The link with regulation and trade is especially clear in Italy where controls on fish introductions are almost non-existent, 70% of its native species have gone extinct due to the ecological impacts of non-native species (Copp, Bianco, Bogutskaya, Eros, Falka, and Ferreira, 2005).

Poor countries are especially sensitive to non-native species because these countries generally lack the infrastructure, funds, and policies to manage them. As a result, countries with much to lose are seriously unprotected and understudied. Peru is a prime example of this, over 855 species of freshwater fish have been described, with several hundred more estimated to be discovered in the future (Ortega, Guerra, and Ramirez, 2007). Brazil, which has similarly high rates of freshwater biodiversity and ecosystem composition, has already observed native species decline, just by transporting native species between watersheds within the same country (Latini and Petrere, 2004). On local or regional spatial scales, invasion success has little correlation with endemic species richness (Marchetti, et al., 2004). Much of the diversity in Peru is divided into regional areas meaning that the overall diversity of the region will most likely not prevent it from being invaded by non-native species. This makes the water of Peru sensitive to both invasion and its effects, as it does not have the resources to manage non-native species once they become established. This justifies the study of the non-native fish in Peru, and whether or not they are having observable ecological impacts.

One non-native fish species found in Peru, which has not been previously studied, is the blue gourami (*Trichogaster trichopterus* Fig. 1). Fish in the belontiidae family (of which blue gourami are a member of) have a labyrinth organ which allows them to breathe atmospheric oxygen, thus allowing them to living in normally hypoxic environments (Helfman, Collette, Facey, and Bowen, 2009). They grow to a maximum size of 15 cm. The have a large geographic range in South East Asia throughout the Mekong: Thailand, Laos, Cambodia, Vietnam, Malaysia, Sumatra, Borneo, Java, and Madura. Male blue gourami create bubble nests, which are a collection of bubbles, saliva, and mucus that floats at the surface of the water and holds the eggs (Cheal and Davis, 1974). During this time, male gourami are very territorial and will behave aggressively towards other fish in order to protect their nest.

Detailed research of blue gourami introductions is rare but there are several scant reports of introductions in several countries; there is one report that the blue gourami was in competition with and led to a decline of *Puntius semifasciolata*, a cyprinid in Taiwan (Liao and Liu, 1989). No specific reason is given as to how the blue gourami caused the decline of *P. semifasciolata*, instead the authors just state that where the blue gourami were found, *P. semifasciolata* populations were in decline or extinct from their historical ranges. Blue gourami have also been found as a non-native fish in Jamaica and Australia but with no information on any effects it may be having there (Olden, Kennard, and Pusey, 2008; Geheber, McMahan, and Piller, 2010). It has apparently been in India long enough to become naturalized in some areas (Knight, 2010). The introduction took place approximately 25-30 years ago when fish from an aquaculture facility repeatedly escaped; the fishermen in the area now collect and sell the gourami to museums and in the pet trade (Daniels and Rajagopal, 2003). Current available research is conflicting or anecdotal, and offers no practical information for researchers, policy makers, or managers. This



lack of evidence demands that further ecological research is done to determine the current ecological affects that the blue gourami are having.

The aim of this research was to determine if blue gourami have the same diet as the native fishes it co-occurs with in the Peruvian Amazon.

Figure 1: A preserved blue gourami used for gut content analysis collected in Iquitos, Peru.

Materials and Methods

The study site was located in the Pampa Chica area of Iquitos, Peru (3°45'00.32" S, 73°16'37.04" W). This area is within the Rio Nanay flood plain and contains both standing water areas and running water that originate from the open sewage system canals used in this area. Fish were captured using a 7 m x 2 m sine with a mesh size of 3 mm. Immediately after capture, all fish were preserved in AFA. In order to make sure the stomach contents were preserved, a cut was made into the peritoneal cavity to insure influx of AFA for at least 3 days before gut content analyses were conducted.

Gut contents were analyzed by removing the entire gastro-intestinal tract from the anterior portion of the gut to the anus. Gut contents were placed into a petri dish and examined using a dissecting microscope while viewing on 10 to 40X magnification. Contents were recorded based on ten categories: detritus, plant fiber, insect larvae, ant, fly, chrionomus larvae, tubifex worm, fish, fish parts, and algae. The categories were made based on observations of items inside the gut contents of all fish. Any undistinguishable food item was categorized as detritus; this included sediment ooze, significantly digested food, indistinguishable masses, charcoal, and rice. Plant fiber refer to easily identifiable plant material found inside stomachs; this could be pieces of sticks, leaves, aquatic plants, or any kind of vegetative material that had a solid structure. Insect larvae was used to signify juvenile insects of unidentifiable species. Ant and fly categories were used for those insects that could be easily identified as such. Chrionomus larvae were easily distinguished but in some of the gut contents, the bodies of the larvae were digested but the easily identifiable head would remain. Tubifex worms are small red worms found in sewage sediments, or other waters with high levels of decomposing organic matter. Individual species were impossible to identify so all small red worms were indicated as

tubifex. Any fish specimen with over 50% of its body intact was indicated as fish while parts of fish such as scales, fins, or barbells were indicated as fish parts. Algae refer to any green photosynthetic material that lacked shape, yet was identifiable as aquatic "plant" material that was not fibrous.

The relative importance of food items in a fish's stomach was determined using the frequency of occurrence method (Hyslop, 1980) which is calculated by the number of stomachs food occurs expressed as a % of the total number of stomachs examined for each species. Due to its simplicity, this method does have some disadvantages. It does not account for the amount of the food item consumed, the volume it takes up in the stomach, or the importance of the food item to that species and only demonstrates what the fish are feeding on. This method has been used in the past as an indicator of interspecific competition (Johnson, 1977). We additionally determined the relative abundance of each food item by taking the number of each food item in the stomachs of the fish divided by the number of food items in all stomachs for each species.

To compare the diets of each of the native species to that of the blue gourami, we used Schoener's Index (Schoener, 1970). This index is calculated with a minimum of 0 (meaning no diet overlap) and a maximum of 1 (meaning complete overlap). Statistical analysis of Schoener's index is not possible, but typically, values greater than 0.6 are interpreted as a significant degree of dietary overlap between species (Zaret & Rand, 1971; Wallace, 1981; Wallace and Ramsey, 1983).

Results

A total of 2090 fish were collected and their gut contents analyzed for this study comprising 15 different species; *Trichogaster trichopterus* (46.6% of the fish examined), *Dianema longibarbis* (18.1% of the fish examined), *Astyanax bimaculatus* (11.8% of the fish examined), *Cichlasoma amazonarum* (9.8% of the fish examined) *Pimelodus pictus* (6.9% of the fish examined), *Aequidens matae* (4.5% of the fish examined), *Triportheus angulatus* (4.1% of the fish examined), *Bunocephalus knerii* (2.1% of the fish examined), *Gymnocorymbus temetzi* (1.3% of the fish examined), *Curimata aspera* (0.9% of the fish examined), *Rineloricaria sp.* (0.8% of the fish examined), *Sorubim lima* (0.8% of the fish examined), *Chalceus macrolepidotus* (0.7% of the fish examined), *Erythrinus erythrinus* (0.7% of the fish examined), and *Cichla monoculus* (0.3% of the fish examined), and *Crenicichla sp.* (0.3% of the fish examined),

Of the 974 gourami sampled, the three food items with the highest, frequency of occurrences were; plant fibers (38.1%), insect larvae (30.2%) and chrionomus (21.0%; Table 1). In total, blue gourami had a diet composed of 8 different food items. The most numerous prey items were plant fibers (46.75%), insect larvae (21.43%) and chrionomus (13.38%; Table 2). Thirty (3.0%) had no items in their GI tract upon examination. We did find items that were clearly from human activities. Pieces of rice, charcoal and fish scales were found in 29 of the gourami sampled (a total of 230 pieces). Many people living in this area use the local water to wash their dishes and use the water sources as their garbage bin. Clearly, some of the fish are utilizing these items as a food source.

The diet of *D. longibarbis* was dominated by plant fibers (45.8%), chrionomus (39.7%) and tubifex worms (30.7%). A total of 6 food items were identified in the stomach contents with

the most abundant item in the diet were tubifex worms (24.59% of the total number) followed by plant fibers (22.95%) and chrionomus (21.31%), respectively (Table 2).

The diet composition of *A. bimaculatus* consisted of five types of prey and plant fibers (Table 1). Flies were identified as the most abundant prey item consisting of 45.24% of all stomach contents (Table 2), but were only found in 44.1% of the stomachs (Table 1). It appears that *A. bimaculatus*, in our study is a terrestrial and aquatic insect specialist as 87.08% of the diet consisted of both terrestrial and aquatic insects.

Three members of family Cichlidae were sampled during this study. *Cichlasoma amazonarum* were the most abundant in the sampling with 95 individuals examined. Plant fibers (31.07%) and chrionomus (22.45%) had the highest relative abundances of the food eating (Table 2). In addition, these two food types also were found in the highest frequency of the gut contents (46.6% plant fiber and 30.68% chrionomus; Table 1). *Crenicichla* sp. and *Cichla monoculus* were collected in low numbers (7 and 6 individuals, respectively; Table 1). Tubifex worms were of the highest relative abundance in *Crenicichla* sp. (36.36%), whereas, fish had the highest relative abundance in *Cichla monoculus* (Table 2).

The 87 *Triportheus angulatus* that were analyzed had a total of 195 food items (Table 2). The majority of the gut contents were composed of insect larvae (93.9% of the individuals sampled) and tubifex worms (28.75; Table 2). Despite this, insect larvae (30.77%) and ants (25.13%) were of the highest relative abundances in the stomach contents.

Curimata aspera, Sorubim lima, Rineloricaria sp, Chalceus macrolepidotus, Erythrinus erythrinus each composed less than 1% of the fish collected (Table 1). *Curimata aspera* ate fish parts in highest abundances (75.0%) with the remaining 25% consisted of plant fibers (Table 2). *Sorubim lima* had insect larvae (46.0% of food items) and fish (fish parts, 29% and fish, 25%) in

its gut contents. The majority of gut contents of *Rineloricaria* sp. contained plant fibers (64.29% relative abundance; Table 2). , *Chalceus macrolepidotus, Erythrinus erythrinus* had frequency (64.29% for both species; Table 1) and the highest relative abundance of insect larvae in there gut contents (34.29% and 34.88%, respectively; Table 2).

The dietary analyses of blue gourami when compared to the other species analyzed in this study showed a high level of overlap as calculated by the Schoener index (Table 3). Eight of the fourteen species sampled showed overlap with the diet of blue gourami: *Cichlasoma amazonarum* (81.60%), *Triportheus angulatus* (78.90%), *Pimelodus pictus* (77.60%), *Dianema longibarbis* (72.90%), *Hemigrammus pulcher* (67.00%), *Bunocephalus knerii* (63.60%), *Erythrinus erythrinus* (62.70%) and, *Chalceus macrolepidotus* (61.70%). On the other hand, six of the native species sampled showed a low percentage of overlap with blue gourami: *Crenicichla* sp. (55.30%), *Sorubim lima* (50.10%), *Astyanax bimaculatus* (49.20%), *Cichla monoculus* (36.60%), *Rineloricaria* sp. (32.90%), and *Curimata aspera* (17.50%).

Discussion

Invasive species have many characteristics in common: polyphagous (Moyle & Light, 1996), high dispersal rates (Rehage & Sih, 2004), large native range (Novak & Mack, 1993), high genetic variability (Kolbe et al., 2004), r-strategy (high reproductive potential; McMahon, 2002), human commensalism (Sol et al., 2002), eurytopy (high environmental tolerance; Casatti et al., 2006). Blue are a perfect candidates for being an invasive a new environment. In their native range this species typically occurs in heavily vegetated, shallow, sluggish or standing water and in seasonally flooded habitats. They can tolerate wide ranges of several water parameters including hardness (5° to 35° dGH), pH (6.0 to 8.8), temperature (21°C to 31°C), salinity (tolerate brackish from water 5-20 ppm) and low dissolved oxygen conditions (0-1 ppm; Priest (2002). They are omnivorous, feeding mainly on zooplankton, (e.g. copepods, cladocerans, ostracods), macroinvertebrates (insect larvae), detritus and occasionally terrestrial macrophytes (Conlu, 1986; Chung et al., 1994; Talde et al., 2004). Blue gourami's have a promiscuous mating system and capable of year round spawning (Hails and Abdullah, 1982). They breed when the water temperature is between 18 and 29°C (Axelrod and Shaw, 1967) with spawning being enhanced in acidic water with a pH range between 5.5 and 6.5 (Reyes-Bustamente and Ortega-Salas, 2002). They are bubble nest spawners with extended male parental care with the male becoming increasing aggressive towards other conspecifics including the recently-spawned female (Hodges and Behre, 1953; Miller, 1964; Picciolo, 1964; Pollak et al., 1981). Fecundity varies with female size (300 for smaller females and 2000-4000 from larger females; Zukal, 1983; Rich, 1988). Under experimental conditions, a mean absolute fecundity of 8,021 and a maximum value of 9,104 was recorded (Reves-Bustamante and Ortega-Salas, 2002). Combined

with multiple spawnings, this enables rapid population growth with a doubling time estimated at less than 15 months (Froese & Pauly, 2007).

Blue gourami are by far the most abundant species found in the sampled area. These areas are highly impacted by humans. The water is used as a garbage dump, toilet, and washing area (clothes and dishes; per obs.) Others have found that invasive species are closely linked to human development (Olden, Kennard, and Pusey, 2008). In addition, blue gourami are able to exploit a habitat where most other fishes can not. Gourami densities in the open sewage canals around Iquitos are tremendous (pers obs.). Here, the gourami are free from most aquatic predators and are able to reproduce unaffected by interspecific competition.

Knowledge on a non-native fish's niche is generally pre-requisite for management decisions involving the future of a non-native species (Townsend, 2002). The first aspects of a non-native species that must be studied are its foraging habits (Olaf and Lewis, 2006). Foraging habits are important because it determines if the non-native species is in direct competition with other species, or if it is consuming them directly. Gut content analysis and fish surveys were used in combination to gain a clearer understanding of the ecological interactions taking place. If only one or the other was done then no further explanation would be possible. Only by knowing what the fish is eating, where it is, and how many there are can researchers develop policies to remove it from the ecosystem, or allow it to persist.

Blue gourami were found to have diet overlap with eight native species. Although diet overlap does not necessarily mean the species are in competition, in other studies diet overlap between non-native species and native species has been correlated with decreased native species abundance (Karlson, Almqvist, Skora, & Appelberg, 2007). Competition is believed to cause ecosystem instability because prey items lack ways of avoiding predation by non-native species,

thus dramatically lowering prey item numbers and negatively impacting species at higher trophic levels such as fish (Mandrak & Cudmore, 2010). A non-native generalist is able to consume a more diverse selection of prey items than native species, creating displacement pressures on the fish for feeding area and limiting food resource access for other fish (Figueroa, Ruiz, Berrios, Villegas, & Andreu-Soler, 2010). This could plausibly be happening with the gourami due to its territorial breeding nature and the high densities found in the survey areas which seemed to nearly block out the surface of the water in certain areas.

The original goal of measuring niche overlap was to infer interspecific competition (Schoener 1974). But, the relationship between niche overlap and competition is poorly defined in the literature. The particular resources being studied may not always be limiting populations, and species may overlap with no competition. Conversely, MacArthur (1968) pointed out that zero niche overlap did not mean that interspecific competition was absent. Abrams (1980) pointed out that niche overlap does not always imply competition, and that in many cases niche overlap should be used as a descriptive measure of community organization. The relationship between competition and niche overlap is complex (Holt 1987).

Fish surveys are not a perfect method for understanding population composition and instead offer a very narrow view into the complex ecological interactions taking place. Often surveys of non-native fish over time reveal fluctuating populations, or range expansions that may not reflect the stable size of the population (Trexler, Loftus, Jordan, Lorenz, Chick, and Kobza, 2000). Despite these limitations fish surveys are still one of the only ways to estimate the size of a fish population. And when these limitations are taken into account potential inaccuracies can be accounted for. Gut content analysis also has its own limitations. Importance, nutritional value, nor volume was measured during this study. It is unclear whether the gourami was consuming a surplus of food items, or they were directly competing with native species over a scarce item. The only information collected was whether or not the food item was present inside the fish's stomach. Another limitation was being unable to identify the majority of the gut contents of the fish, what was unidentifiable may have actually been a very important part of that fish's diet.

The gourami here are also clearly opportunistic feeders. Gut content not only contained "natural foods" but foods anthropogenic food sources (fish scales, rice, and charcoal). It could not be determined if the fish were intentionally eating scales or if they were consuming fish and the scales were the only thing left in their gut undigested. In addition, some of the detritus found in the gut contents of many of the fish (including blue gourami) contained rice and charcoal. This demonstrates that many of the fish are taking what is readily available in the environment as many of the people dispose of their trash and food remnants in the water.

Change in diet, population density, and life history have been documented in non-native fishes (Bohn, Sandlund, Amundsen, & Primicerio, 2004). Initially density for non-natives is low, yet it increases over time until intraspecific competition becomes too intense resulting in a density dependent life history change, usually in declining population density. This change in life history, called the pioneer stage, is believed to be because species that have recently invaded a region will invest more resources into early reproduction and high fecundity to ensure future generations survival. It is possible that the gourami may be moving out of this pioneer stage, or was just in it because the gourami found in both sites were in greater numbers than the native fishes, and many had signs of reproductive development. This plastic life history, in addition to

the gourami's generalist water quality and food requirements could help to explain why they can be found in such large numbers in the sample areas.

The gourami's ability to persist in different water qualities and consume a wide range of food items including scavenging means that it may be able to outcompete native fishes in the Pampa Chica area. Negative ecological impacts are possible given that non-native species, less related to native species, pose a greater risk of becoming invasive, then those species more related to native species (Strauss, Webb, and Salamin, 2006). Gourami evolved in Southeast Asia and are distantly related to cyprinodontiformes found in the Americas and have no close relatives in Peru (Nelson, Grande, and Wilson, 2016). Thus, they are not bound by the evolution of the group in Peru.

Additionally, non-native species often have a "lag-time" where it is establishing a population, and is in such small quantities that it has not caused any obvious ecological alterations. However, species have been known to cause in population level extinctions several years after its introduction, with no prior indication that they would (Ricciardi, 2004). Again, there is insufficient data to determine if the blue gourami is past this lag time, in it, or just about to start it. This is similar to the ecological impacts of the Nile perch in Lake Victoria, where by the time biodiversity of the lake was being studied, the Nile perch had already been introduced and was already having an impact (Goldschmidt, 1996).

Of even greater concern is that if the gourami had impacts in the past, it would likely have similar ones in the future, should it enter new ecosystems. Endemic specialized species with restricted habitat requirements are most susceptible to extinction due to non-native species (Trexler, Loftus, Jordan, Lorenz, Chick, and Kobza, 2000). Many of the fishes of Peru are highly specialized and isolated into different river systems, yet during the rainy season, these

river systems merge giving the gourami access to previously isolated and sensitive new systems. This is further demonstrated by the fish surveys which showed a diverse group of fish each only consuming a few different food items, with a few exceptions. Due to seasonal flooding, the area of Pampa Chica connects to the Nanay River which connects to the Amazon. Seasonal migrations from one watershed to another are not only possible it is likely, especially with a hardy prolific fish like the gourami. Once in the Amazon River, there would be no barriers preventing it from spreading throughout the entire Amazon basin, with uncertain ecological impacts.

Disturbed areas, such as the areas found adjacent to aquaculture facilities, allow non-native species to adapt to a new environment easier and have a higher change of establishing and spreading (Sax and Brown, 2000). In 1965, the Peruvian government devised a feeding program for Arapaima gigas by using guppies (Poecilia reticulata) as food fish. But this species was insufficient for the feeding requirements of Arapima.gigas, so the government brought in Tilapia rendalli in 1968 (Ortega, Guerra, and Ramirez, 2007). Both species are now found in Peruvian rivers, with uncertain ecological impacts. As of 2007, twenty non-native species have been found in the watersheds of Peru (Ortega, Guerra, and Ramirez, 2007). However, if the nonnative fishes released into Peru follow similar trends as other non-native species then extinctions are inevitable. If this trend of importing non-native fishes continues then soon the economically worthless, yet ecologically valuable species of fish in Peru will be significantly threatened. Worse yet, creating protected areas around these watersheds does not reduce non-native species numbers, meaning that once populations become established there is little that can be done (Marchetti, Light, Moyle, and Viers, 2004). That is why it is crucial that further research is undertaken, and that further non-native fish species are not imported, because the consequences

of a negative impact means severely damaging one of the most beautiful and biodiverse regions in the world.

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Species	Total No	Detritus	Plant Fiber	Insect Larvae	Chrinomus	Tubifex	Fly	Algae	Fish Parts	Ant	Fish	Other*	Empty
Trichogaster trichopterus	974	881	360	285	198	129	48	45	36	13	0	29	30
Dianema longibarbis	379	287	167	100	145	112	87	0	0	90	0	3	14
Astyanax bimaculatus	246	213	30	30	52	0	104	0	0	15	0	2	10
Pimelodus pictus	145	107	32	27	11	16	5	15	23	0	0	7	5
Cichlasoma amazonarum	95	85	41	19	27	8	19	3	8	22	2	0	7
Triportheus angulatus	87	77	32	32	4	23	10	0	0	9	0	3	5
Bunocephalus knerii	44	36	12	16	12	12	32	0	7	28	0	1	7
Hemigrammus pulcher	28	28	14	31	0	8	11	3	0	0	0	2	0
Curimata aspera	18	18	2	0	0	0	0	0	12	0	0	1	0
Sorubim lima	17	15	0	11	0	0	0	0	6	0	12	0	0
<i>Rineloricaria</i> sp.	16	15	12	2	0	2	1	0	0	0	0	0	0
Chalceus macrolepidotus	14	4	5	9	0	0	5	0	0	5	0	0	0
Erythrinus erythrinus	14	0	0	9	4	0	0	0	3	0	8	0	0
Crenicichla sp.	7	7	0	0	4	6	4	0	0	0	2	0	0
Cichla monoculus	6	3	0	0	0	0	0	0	3	0	6	1	0

Table 1: Species, number of individuals examined, and number of GI tracts that contained each food item.

• other include identifiable food items that appeared to have an anthropogenic source: fish scales, pieces of carbon, and rice. This category was not used as part of the any analysis.

Species	Total	Plant	Insect							
Species	No	Fiber	Larvae	Chrinomus	Tubifex	Fly	Algae	Fish Parts	Ant	Fish
Trichogaster trichopterus	1232	46.75	21.43	13.38	11.04	1.46	3.25	2.19	0.49	0.00
Dianema longibarbis	427	22.95	13.11	21.31	24.59	9.84	0.00	0.00	8.20	0.00
Astyanax bimaculatus	294	12.93	12.93	22.45	0.00	45.24	0.00	0.00	6.46	0.00
Pimelodus pictus	182	25.82	28.02	7.69	15.38	4.95	2.75	4.95	10.44	0.00
Cichlasoma amazonarum	103	31.07	15.53	22.33	1.94	15.53	1.94	0.00	11.65	0.00
Triportheus angulatus	195	16.41	30.77	2.56	13.85	11.28	0.00	0.00	25.13	0.00
Bunocephalus knerii	182	10.99	3.30	7.14	10.99	17.58	0.00	10.44	39.56	0.00
Hemigrammus pulcher	28	28.57	17.86	0.00	17.86	28.57	7.14	0.00	0.00	0.00
Curimata aspera	16	25.00	0.00	0.00	0.00	0.00	0.00	75.00	0.00	0.00
Sorubim lima	24	46.00	0.00	0.00	0.00	0.00	0.00	29.00	0.00	25.00
Rineloricaria sp.	14	64.29	14.29	0.00	14.29	7.14	0.00	0.00	0.00	0.00
Chalceus macrolepidotus	35	22.86	34.29	0.00	0.00	20.00	0.00	0.00	22.86	0.00
Erythrinus erythrinus	43	0.00	34.88	25.58	0.00	0.00	0.00	16.28	0.00	23.26
Crenicichla sp.	11	0.00	18.18	18.18	36.36	18.18	0.00	0.00	0.00	9.09
Cichla monoculus	14	0.00	0.00	0.00	0.00	0.00	0.00	28.57	0.00	71.43

Table 2: Species, total number of food items and relative abundance of each food item in the diet of the fish examined in Iquitos, Peru.

Table 3:Results of diet comparisons by Schoener's Index between blue gourami and the species that co-occurred with blue gourami in Pampa Chica,
Iquitos, Peru. Indices above 60% denote significant degree of dietary overlap between species

Species	Schoener's Index
Cichlasoma amazonarum	81.60%
Triportheus angulatus	78.90%
Pimelodus pictus	77.60%
Dianema longibarbis	72.90%
Hemigrammus pulcher	67.00%
Bunocephalus knerii	63.60%
Erythrinus erythrinus	62.70%
Chalceus macrolepidotus	61.70%
Crenicichla sp.	55.30%
Sorubim lima	50.10%
Astyanax bimaculatus	49.20%
Cichla monoculus	36.60%
Rineloricaria sp.	32.90%
Curimata aspera	17.50%