How Environmental Policy Shapes The Aquarium Trade: Potter's angelfish (*Centropyge potteri*) Larval Growth and Improved Culture Methods

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Abstract

The Potter's angelfish (*Centropyge potteri*) is a striking reef fish endemic to the Hawaiian archipelago, highly sought after by aquarists for its vibrant colors and unique characteristics. This species was once one of the most exported aquarium fish from Hawaii before a ban on fish collection for the aquarium trade. This research project investigates the larval rearing and growth of captive bred Potter's angelfish within the context of Hawaii's role and history in the aquarium trade, while concurrently examining the influence of environmental policies on the trade's sustainability.

The study employs an interdisciplinary approach, combining laboratory trials with policy scenarios aimed at promoting sustainability. Five larval feeding regimes were tested, aiming to enhance the survival rates and growth trajectories of Potter's angelfish larvae. Morphometric and survival data were collected to monitor developmental milestones and assess the survival rate of reared individuals. Simultaneously, the research addresses the broader implications of Hawaii's environmental policies on the aquarium trade. The effects of Hawaii's aquarium ban were assessed with regard to supply, demand, and prices of previously heavily exported species, Yellow tang and Potter's angelfish. Interviews with stakeholders provide valuable insights into the perspectives and experiences surrounding the implementation and enforcement of these policies. A proposed policy scenario is presented with impacts on stakeholders described.

The project's outcomes aim to contribute essential data to the scientific understanding of Potter's angelfish larval biology and rearing requirements, thereby informing future larviculture research and potential conservation strategies for this iconic species. Additionally, the study assesses the role of environmental policies in shaping the sustainability of the aquarium trade in Hawaii, with implications for global marine conservation efforts. The integration of scientific findings and stakeholder perspectives ensures a holistic approach to addressing the complex interplay between the aquarium industry, marine ornamental aquaculture, and regulatory frameworks in Hawaii.

Introduction

Marine Ornamental Aquaculture and The Aquarium Trade:

Marine ornamental aquaculture (MOA) has emerged as a pivotal strategy to address the ecological concerns associated with the extraction of marine ornamental fish for the global aquarium trade. Each year, approximately 20 million marine ornamental fish are harvested from coral reefs worldwide, contributing to a burgeoning industry valued at an estimated $15 billion USD (Chen, 2019; Moorhead & Zeng, 2010). The Hawaiian Islands, renowned for their unique native and endemic species, notably the yellow tang (*Zebrasoma flavescens*) and Potter's angelfish (*Centropyge potteri*), have been a significant exporter with over 300,000 Yellow tang and 9,000 Potter's angelfish extracted each year (Walsh et al., 2004; Williams et al., 2009). The overreliance on wild collection, as seen in Hawaii, poses threats to the sustainability of coral reef ecosystems, necessitating alternative methods such as marine ornamental aquaculture. Due to concerns about overfishing in Hawaii, regulatory efforts such as Fish Replenishment Areas (FRAs) and collection bans have been implemented to protect coral reef fish abundance. Studies indicate the positive impact of FRAs on aquarium fish abundance, notably Yellow tang and Potter's angelfish, emphasizing the potential of conservation measures in conjunction with MOA for sustainable trade practices (Tissot et al., 2004). The State of Hawaii implemented a collection ban in 2021 prohibiting commercial collections of aquarium fish (Schaar & Cox, 2021). Currently, MOA is the only option available for the procurement of native and endemic Hawaiian species such as Yellow tang and Potter's angelfish.

While the majority of freshwater aquarium fish are now captive-raised, the marine aquarium trade predominantly relies on species harvested from environmentally sensitive coral reefs (Chen, 2019; Olivotto et al., 2011; Wabnitz & Taylor, 2003). Harmful collection methods, such as the use of cyanide and dynamite, continue to pose environmental hazards, exacerbating the challenges faced by reefs already impacted by climate change and other anthropogenic factors (Olivotto et al., 2017). Furthermore, the high mortality rates (60-80%) post-collection due to handling stress, disease, and shipping underscore the urgent need for sustainable alternatives like marine ornamental aquaculture (Wabnitz & Taylor, 2003).

MOA offers a promising avenue for procuring marine ornamental organisms while alleviating pressure on natural populations. The benefits extend beyond conservation, as MOA facilitates a deeper understanding of species' life histories, contributing valuable information for future fisheries management (Olivotto et al., 2011; Tlusty, 2002). However, despite recent successes in rearing certain species, the commercial-scale production of many key marine ornamental species remains a challenge (Callan et al., 2018; Moorhead & Zeng, 2010). Significant knowledge gaps persist in areas such as broodstock care, embryonic development, hatching, and the provision of specialized live feeds crucial for larval transition to exogenous feeding and further development (Moorhead & Zeng, 2010; Olivotto et al., 2011; Tlusty, 2002). While MOA has made significant strides, particularly in the successful rearing of the yellow tang, challenges persist, including obtaining high-quality eggs from broodstock, developing suitable live feeds for early larvae, and addressing species-specific nutritional requirements (DiMaggio et al., 2017; Groover et al., 2020; Pereira-Davison & Callan, 2017).

Dwarf angelfish:

Centropyge angelfish are some of the most highly valued fish within the marine aquarium trade. Within the Pomacanthidae family, the genus Centropyge stands out as one of the most sought-after and extensively traded marine angelfishes, owing to its vibrant colors and small size at maturity. Comprising the largest number of species within the marine angelfish family, Centropyge boasts 28 described species. The reproductive biology of Centropyge has been extensively studied, revealing striking similarities among the various species (Bauer and Bauer 1981; Moyer and Zaiser 1984; Lutnesky 1992; Hioki et al. 1990). These investigations cover diverse aspects, including reproductive strategy, histology, behavioral patterns, timing and frequency of spawning, egg and early larval development, sexual dimorphism and dichromatism, as well as social structure and territory. Research endeavors have managed to address some of the concerns about the quality of *Centropyge* eggs, appropriate embryo development, and hatching through the formulation of suitable diets and the utilization of advanced technologies (Laidley et al., 2008). Nonetheless, a significant hurdle that remains is the optimization of feeding schedules and environmental conditions in order to achieve commercial-scale success.

While the aquaculture of Centropyge presents an economically promising and environmentally conscientious alternative to wild collection, the technical challenges associated with their breeding are noteworthy. The larval rearing phase, particularly for species producing small pelagic eggs, poses inherent complications due to the delicate nature of the small larvae (Holt 2003). The artificial propagation of these individuals not only holds economic potential but also provides a unique opportunity to gather crucial information on the reproduction and early life history of these captivating fish species, potentially serving as an invaluable source of knowledge.

Potter's angelfish:

The Potter's angelfish (*Centropyge potteri*) is a strikingly beautiful species of marine dwarf angelfish that is highly valuable and sought after by aquarium enthusiasts. However, due to its limited range and the challenges associated with its collection from the wild, there has been growing interest in aquaculturing this species for the aquarium trade. Potter's angelfish are endemic to Hawaii, found exclusively on reefs along the island chain where they inhabit shallow reef environments at depths of 10 to 120 meters. They have a distinctive appearance, with a bright orange body that is marked with bold black and blue stripes and spots. The fins and tail are also edged in blue, creating a dramatic contrast with the orange body. Potter's angelfish are omnivorous and feed primarily on algae as well as small reef invertebrates such as copepods and amphipods (Randall, 1985; Pyle & Meyers, 2009).

Potter’s angelfish (*Centropyge potteri*) has social and reproductive behavior well understood, making it a suitable candidate for aquaculture efforts (Collier et al., 2003; Lutnesky 1992). Despite their popularity in the aquarium trade and reproductive habits well understood, Potter's angelfish early life history and rearing strategies have not been documented in depth. This endemic angelfish has been raised in small numbers by researchers in Hawaii prior to this study, however, they proved difficult to aquaculture in large numbers due to high larval mortality. The creation of a tailored larval rearing protocol for this species offers an important point for the commercialization of this species as captive-bred Potter's angelfish are more acclimated to aquarium conditions and therefore more resilient and long-lived than wild-caught specimens. Furthermore, the captive breeding of Potter's angelfish has the potential to stimulate the development of new aquaculture techniques and technologies that can be applied to other species of marine fish, furthering marine finfish aquaculture knowledge more broadly (Pouil et al., 2020).

Management of Hawaii’s Aquarium Fishery - A Brief History:

The aquarium fishery in Hawaii began as a small-scale, unregulated industry in the 1950s. As the industry grew, concerns over the sustainability of fish populations and environmental impacts began to emerge. In response, the State of Hawaii established a number of regulations in the 1960s and 1970s, including size limits on the fish that could be collected and restrictions on the number of fish that could be taken. These regulations were enforced by the Department of Land and Natural Resources (DLNR) and were periodically updated to address changing concerns (Walsh et al., 2004).

Despite the regulations in place, concerns over the impacts of the aquarium fishery on the marine ecosystem continued to grow in the 1980s and 1990s. In response, the DLNR conducted a series of studies to assess the impact of the aquarium fishery on fish populations and the environment. In 1990, the DLNR established a working group to develop a management plan for the aquarium fishery, which included recommendations for improving data collection, monitoring, and enforcement of regulations (Walsh et al., 2004).

During this period, controversy surrounding the aquarium fishery continued to grow, and environmental groups filed several lawsuits against the State of Hawaii seeking to halt the issuance of aquarium permits. In response, the state agreed to conduct a comprehensive environmental impact assessment (EIA) of the aquarium fishery. The EIA, completed in 2002, concluded that the aquarium fishery had a significant impact on the reef ecosystem and recommended a range of management strategies to reduce the impact. These strategies included limiting the number of permits issued, establishing size and bag limits, and improving monitoring and enforcement. Following the completion of the EIA, the State of Hawaii implemented a number of policies and regulations to manage the aquarium fishery. In 2004, the state established a new permit system that limited the number of permits issued for commercial aquarium collection. In 2008, the DLNR established a new rule limiting the use of fine-mesh nets to reduce the capture of juvenile fish.

Despite these policy changes, controversy surrounding the aquarium fishery continued, and concerns over the impact on the environment and fish populations persisted. In 2017, Hawaii became the first state to pass legislation to effectively end the aquarium fishery. The law, which was put into effect until 2024, prohibits the issuance of new permits for commercial aquarium collection and limits the renewal of existing permits. The law also required the DLNR to conduct a study on the impact of the aquarium fishery on the environment and fish populations. In September 2017, a circuit court judge ruled in favor of a coalition of environmental groups that had sued the DLNR for allowing the commercial collection of reef fish and aquarium trade species in West Hawaii without an environmental impact statement (EIS). The judge ordered the DLNR to conduct an EIS to analyze the fishery's environmental impacts and propose mitigation measures (DAR, 2017).

In August 2019, the Hawaii Supreme Court ruled that commercial aquarium fishing in the state requires an environmental review under the Hawaii Environmental Policy Act (HEPA). The ruling invalidated a lower court decision that had previously exempted the fishery from the HEPA requirements. The court's decision means that commercial aquarium fishing must undergo a thorough environmental review process before permits can be issued.

In January 2021, Circuit Court Judge Jeffrey Crabtree ruled that commercial aquarium fishing in Hawaii requires a valid EIS under the HEPA. The court also invalidated a previous EIS that had been used to issue permits for the aquarium fishery, ruling that it did not meet HEPA's requirements. The decision meant that the aquarium fishery would need to undergo a new, comprehensive environmental review before permits could be issued. This review was completed in January 2023, with Judge Crabtree then ruling to lift the injunction that was preventing fishing permits from being issued. Under the revised EIS, the number of permits issued would shrink from 10 to seven and only apply in the West Hawaii Regional Fishery Management Area with strict catch quotas in place for each collector (DLNR, 2023).

These court decisions and legislative actions highlight the ongoing controversy surrounding Hawaii's aquarium fishery and the need for comprehensive environmental assessments to be conducted before permits are issued. The rulings have also led to increased scrutiny and regulation of the aquarium trade in Hawaii, as well as potential implications for similar fisheries around the world.

Objectives:

The purpose of this project is to document and describe the early life cycle of Potter’s angelfish for the first time while investigating optimized rearing practices for this species and apply findings to form a case study that illustrates the interplay between MOA, environmental policy, and the aquarium trade. It is hypothesized that *C. potteri* survival and/or growth rate depends on rearing protocol, with increased survival in optimized protocols.

Is there an alternative policy scenario for Hawaii's aquarium fishery that provides overall positive outcomes in which all stakeholders benefit and ecosystem services are left unharmed? The effectiveness and potential benefits of such policies are unclear, and research is needed to evaluate their feasibility and potential impact. Therefore, this research project seeks to investigate: How do environmental policy changes, such as Hawaii's reef fish collection ban, impact the aquarium trade? Can improved aquaculture protocols for reef fish, such as the Potter's angelfish (*Centropyge potteri*), lead to feasible levels of production to replace the demand for wild-caught specimens in the aquarium industry?

Limitations:

Due to the limited nature of this project, the policy scenario set forth in this paper is not a policy recommendation, however, it seeks to investigate and describe the possible impacts on stakeholders within Hawaii's aquarium fish industry to illustrate the need for further research.

Methods

Potter's angelfish (*Centropyge potteri*) eggs were collected and raised using five different protocols to assess the impact of feed regimen on larval survival and growth. Microphotographs were taken periodically to document and measure larvae and survival at various ages was recorded and assessed.

Rearing Protocols and Trials:

Five rearing protocols were tested using six 200 L and one 1000 L tank in order to determine the effect of feeding regime on growth and survival. The tanks were left with constant flow and light aeration until 3 days post-hatch when algae and first feeds were added as the larvae transitioned to exogenous feeding. Each tank was prescribed a designated rearing protocol including flow rate, mesh standpipe size, the addition and density of algae, live feeds, and prepared feeds. The rearing protocol was followed by OI hatchery staff with deviations from the protocol being noted. All tanks were kept on a recirculating aquaculture system with a 12:12 hour photoperiod. Each tank was fitted with a small fan to agitate the surface water of the tank during the day to reduce larvae stress (Figure 1).

Six 200 L trial tanks were stocked in January and February of 2023 with a catastrophic hatchery die-off event in March, limiting the amount of data collected from these trials. Trials 1 & 2 were stocked on January 22 with 2,000 eggs in each tank and raised on protocol A, a feeding regime optimized for rearing Yellow tang (Figure 2A). The following day trials 3 & 4 were also stocked with 2,000 eggs and prescribed protocol B, a feeding protocol designed to raise eggs collected from public aquariums (Figure 2B). Trial 6 was stocked over February 9 and 10 with a total of 7,800 eggs and was prescribed protocol D (Figure 2D). Trial 5 followed with 5,200 eggs stocked on February 12 and 13. Both trials 5 and 6 originally followed protocol D, however, a significant die-off event witnessed in trial 6 when starting dry feed led to the delay of dry feed for trial 5 in hopes of avoiding this event, resulting in protocol C for trial 5 (Figure 2C). A hatchery-wide die-off event, believed to be copper toxicity from rainwater entering the hatchery system, occurred in mid-March of 2023 resulting in the loss of all fish in each six trial tanks.

The information gained from these six 200 L trials was then applied to 1000 L tanks, resulting in the first major success for the production of Potter's angelfish with a cohort of 61 juvenile fish from 15,000 eggs initially stocked in May of 2023. This tank was not a part of this project as it was also stocked with eggs collected from public aquariums, however, some larvae were sampled and photographed in later stages of development through settlement to juvenile coloration. The relatively high survival of this tank suggested that the information gained from previous trials was applied successfully. The protocol used for this tank was adapted into protocol E and attempted to be replicated as a trial for this project. Trial tank 7 was stocked from September 15 to 18, 2023 totaling 7,500 eggs in 1000 L and raised using protocol E (Figure 2E).

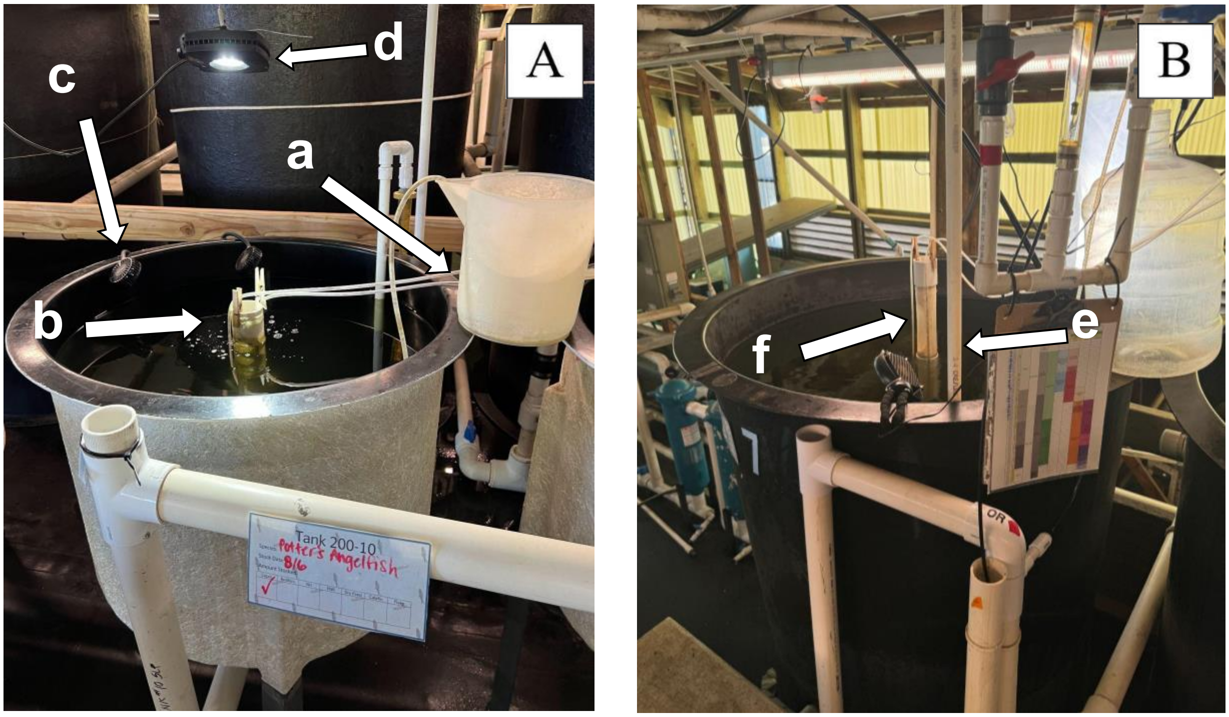

Figure 1. Photographs depicting 200 L (A) and 1000 L (B) larval rearing tank configurations used to culture Potter's angelfish. (a) Feed bucket, (b) airlines connected to airstones, (c) small fan, (d) AI Prime LED light 12:12 photoperiod, (e) water inflow pipe, (f) screened internal standpipe.

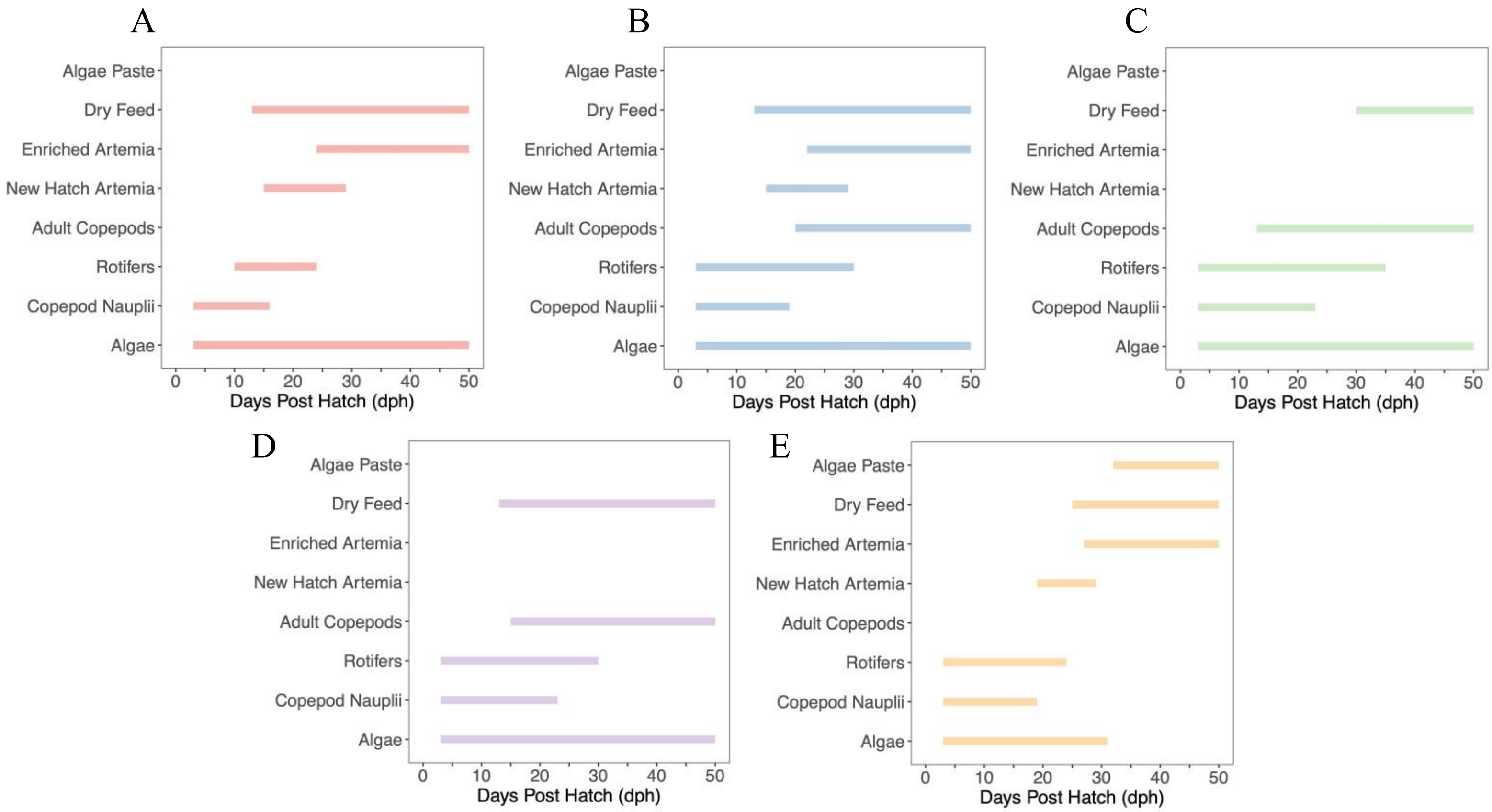

Figure 2. Outline of feeding regimens for all protocols including Algae, Algae Paste, Copepod Nauplii and Eggs, Adult Copepods, Rotifers, New Hatch Artemia, Enriched Artemia, and Dry Feed. (A) a feeding regime optimized for rearing Yellow tang. (B) a feeding protocol designed to raise eggs collected from public aquariums. (C) a feeding protocol without Artemia and with delayed dry feed. (D) a feeding protocol without Artemia and with normal dry feed. (E) a protocol considering observations seen in previous trials with a switch to algae paste at day 32 and no adult copepods.

Egg Collection and Stocking Procedures:

Eggs were collected and counted each morning to determine the total number of viable eggs produced from the previous night's spawn. Eggs were collected from Potter's angelfish broodstock tanks using a 200 micrometer mesh screen placed in the surface drains of the tanks then rinsed into a clean 1 L beaker and filled with saltwater to 1000 mL. The beaker was then moderately aerated for 2-3 minutes to mix the eggs homogeneously. While under aeration, ten random 1 mL samples were taken and combined into a 50 mL beaker. The 10 mL subsample was then poured into a 10 mL zooplankton counting wheel and counted under a dissecting microscope (Olympus SZ61). Eggs were characterized as either inviable or viable using the following criteria. Inviable: no clear cell division or other evidence of fertilization or clear cell division but abnormally developed. Viable: containing a fully developed embryo, ready to hatch (Figure 3). The viable egg count was extrapolated to determine the total number of viable eggs.

Eggs were then separated and stocked into larval rearing tanks to begin trials. Viable eggs were separated after turning off aeration for 20 to 30 minutes, allowing them to float while inviable ones sank. Viable eggs were carefully transferred to a clean 1 L beaker, transported to the hatchery, and added gently to either a 200 or 1000 L hatchery tank (Figure 1). No disinfection methods were employed as it had been seen previously that *C. potteri* eggs do not tolerate disinfection protocols using hydrogen peroxide. Stocking density (eggs/L) was calculated by dividing the total number of viable eggs stocked by the tank volume.

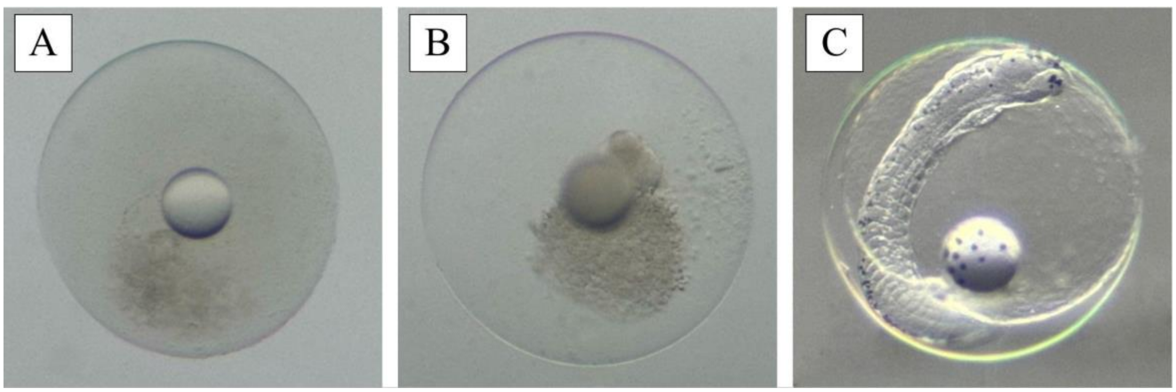

Figure 3. Photographs of representative egg quality categories for *C. potteri* eggs. (A) Unfertilized egg (B) Fertile-inviable egg (C) Viable egg.

Larval Growth, Development, and Survival:

Larval growth and development were captured to compare across feeding regimens. Larvae from each trial tank were periodically sampled using either 1) a 10 mL pipette or 2) a 50 mL beaker. The larvae were taken to Doherty Lab 206, transferred onto a Sedgewick Rafter counting slide using a pipette, and placed under a dissecting microscope (Olympus SZ61). Photographs were taken of the larvae on an iPhone 15 Pro using the ‘Manual Camera’ app by placing the phone’s camera over the microscope eyepiece to fully view the larvae, adjusting focus and magnification as needed. Photos were taken using DF and Oblique light sources on the microscope.

Microphotographs were then sorted, labeled, and larvae measured for body length (BL) and body depth (BD) in mm (Figure 4). Measurements were taken using ImageJ. Body length was measured from the anterior-most point of the larvae to the end of the notochord and in later developmental stages to the mid-caudal peduncle, at the posterior end of the vertebral column just anterior to the hypural plates. Body depth was measured across the myomeres at the anus. Larvae that survived sampling were returned to their respective tank. Images were used to assess developmental progress, recording major developmental milestones such as flexion, eye development, and gas bladder inflation (Figure 4). Measurements were plotted and compared using regression analysis and ANCOVA.

As tanks matured to ~30 days old, the number of fish was counted and recorded periodically to assess survival. Unfortunately, no fish in this experiment were able to reach metamorphosis due to catastrophic events and other phenomena that cannot be fully explained yet. All fish perished by 38 days post hatch in all trials.

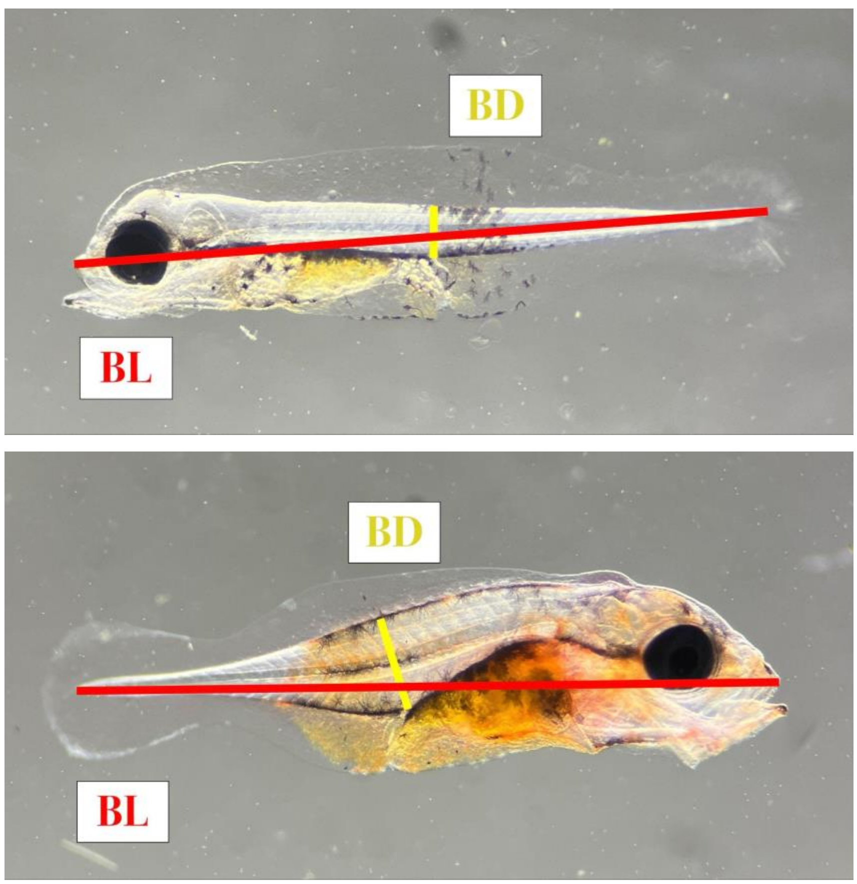

Figure 4. Morphometric measurements of *C. potteri* larvae. BL: body length, BD: body depth. Body length is measured from the most anterior point of the snout to the end of the notochord and to and in later developmental stages to the mid-caudal peduncle, at the posterior end of the vertebral column just anterior to the hypural plates. Body depth is measured across the myomeres at the anus.

The impacts of Hawaii's collection ban on the aquarium industry were assessed through collection of testimony and price data. Interviews with stakeholders were performed to gain valuable insights into the perspectives and experiences surrounding the implementation and enforcement of these policies. A proposed policy scenario incorporating state investment in OA was developed with impacts on stakeholders described, derived from Schaar & Cox, 2021. Average consumer prices for Yellow tang and Potter's angelfish from before and after the ban went into effect were obtained and compared.

Results

A total of 112 *C. potteri* larvae were sampled over the course of this study ranging from 0 to 38 days post hatch (dph). Body length and body depth increased steadily across all feeding regimens (Figure 5). Few discrepancies occurred in this trend likely due to random sampling error. Mean body length and depth increased consistently from 3 to 14 dph. Mean body length growth slowed as larvae entered flexion, from approximately 14 to 21 dph, while body depth increased rapidly during that critical period (Figure 6). After flexion, both body length and depth returned to steady increases. Linear regressions were performed on body length and body depth after flexion had begun (14 dph) to determine average growth rates post-flexion. Protocol A was found to have the highest average body length growth rate at 0.227 mm/day $R^2 = .94$, $t(19) = 18.27$, $p > .0001$. Protocol B was found to have an average body length growth rate of 0.159 mm/day $R^2 = .89$, $t(22) = 13.11$, $p > .0001$, and protocol E was the lowest at 0.097 mm/day $R^2 = .73$, $t(28) = 8.60$, $p > .0001$ (Figure 5A). Average body depth growth rates were found to follow similar trends. Average body depth grew at 0.195 mm/day for protocol A $R^2 = .96$, $t(19) = 20.77$, $p > .0001$. Protocol B followed with 0.166 mm/day $R^2 = .96$, $t(22) = 21.99$, $p > .0001$, and protocol E was found to grow at 0.132 mm/day $R^2 = .89$, $t(28) = 14.96$, $p > .0001$ (Figure 5B).

An ANCOVA was run to determine the effect of protocol on body length and body depth growth rate after 14 dph while controlling for time (dph). After adjustment for time, there was a statistically significant difference in body length growth rate between the protocols, $F(2, 71) = 31.64$, $p < 0.0001$. Multiple pairwise comparisons suggest that there are statistically significant differences in growth rates among all protocols (A-B, A-E, B-E: $p < 0.0001$). Similar significant results were found when comparing body depth growth rates across protocols, $F(2, 71) = 46.99$, $p < 0.0001$.

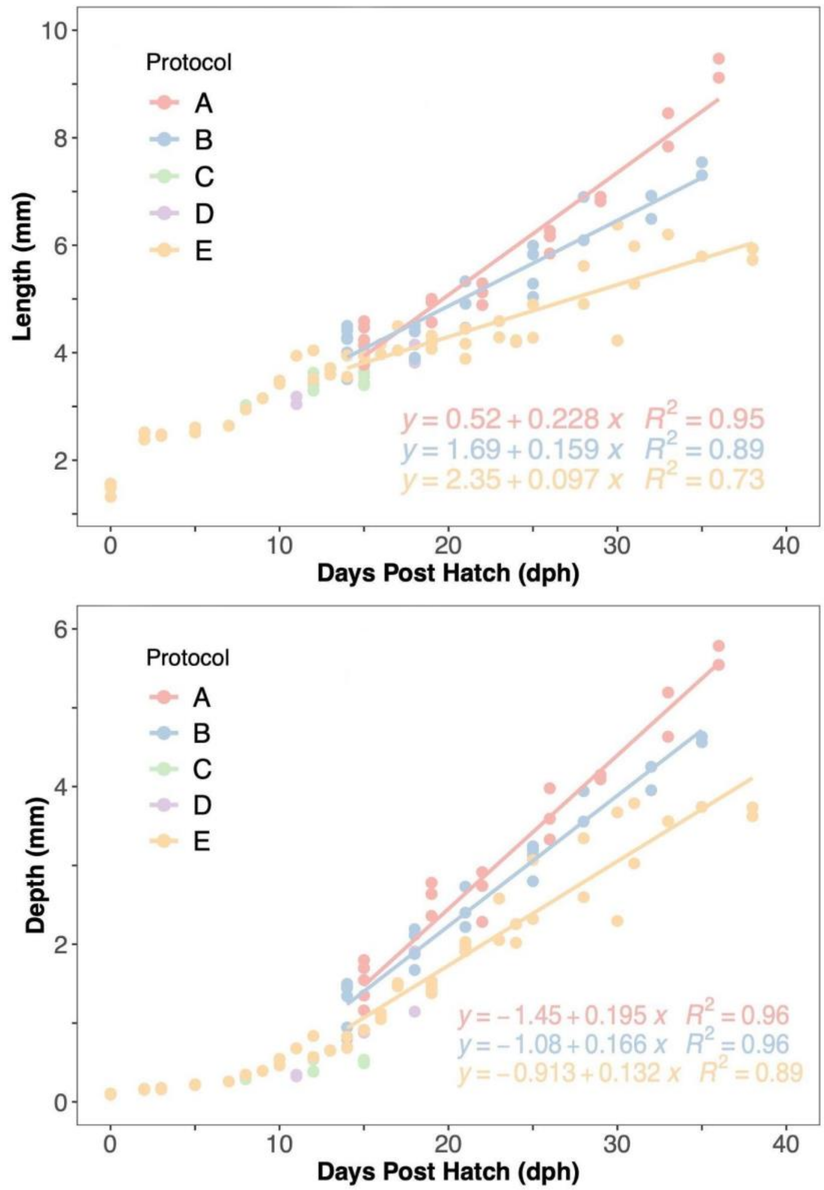

Figure 5. Body length (A) and body depth (B) measurements for all *Centropyge potteri* specimens sampled from 0 to 38 dph, n = 112. Linear regressions for protocols A, B, and E overlaid after 14 dph with equation and R^2 reported.

The critical period of flexion was observed from 14 to 22 dph for *C. potteri*. Flexion was first observed at 14 dph for protocols B and E, 15 dph for protocols A and D (all larvae sampled from protocol A had begun flexion), and no flexion was observed in protocol C at 15 dph when this trial was lost. The first larvae to complete flexion were seen in protocol A, with all larvae sampled at 19 dph having completed flexion. Complete flexion was observed at 21 dph in protocols B and E, with all fish sampled having completed flexion after 22 dph across every protocol.

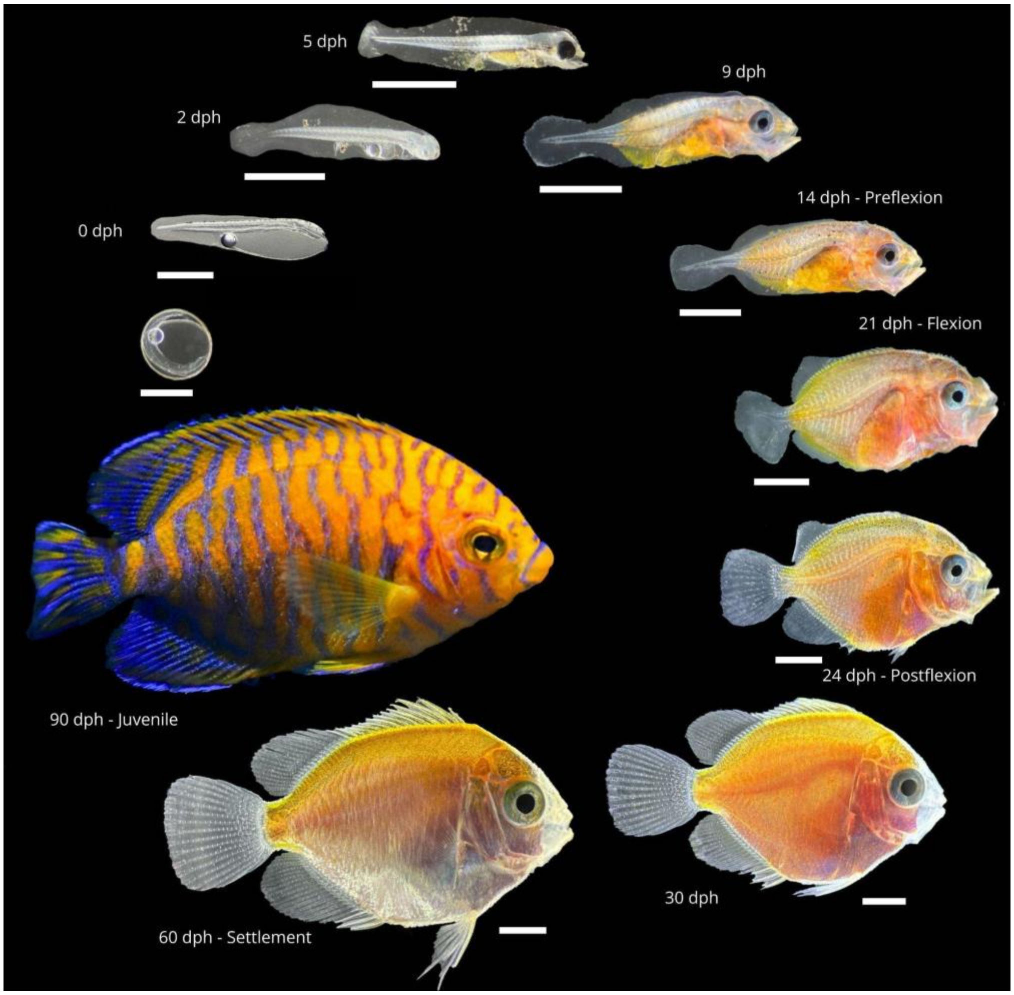

Figure 6. Images of representative egg and larvae at 0 through 90 dph across all protocols. Preflexion, flexion, postflexion, settlement, and juvenile stages are labeled as such. Juvenile photo courtesy of Jake Phillips of The Biota Group. Photographs correspond to egg at approximately 12 hours after fertilization, diameter = 0.68 mm, scale bar = 0.5 mm; larvae at 0 dph, BL = 1.49 mm, scale bar = 0.5 mm; 2 dph, BL = 2.47 mm, scale bar = 1 mm; 5 dph, BL = 2.56 mm, scale bar = 1 mm; 9 dph, BL = 3.15 mm, scale bar = 1 mm; 14 dph - onset of flexion, BL = 3.94 mm, scale bar = 1 mm; 21 dph, BL = 4.53 mm, scale bar = 1 mm; 24 dph, BL = 4.73 mm, scale bar = 1 mm; 30 dph, BL = 5.93 mm, scale bar = 1 mm; 60 dph, BL = 13.30 mm, scale bar = 2 mm.

All protocols maintained similar means until approximately 14 dph, after which growth rates began to diverge. Protocol A displayed the highest growth rate for body length and depth followed by protocol B and then protocol E (Figure 5). Not enough data was collected from protocols C and D to make comparisons. While protocol E began to level off for both length and depth around 30 dph, protocols A and B continued to increase sharply. The drop in growth rate seen in protocol E coincides with the addition of an algae paste, Nanno 3600, instead of live algae starting at 32 dph.

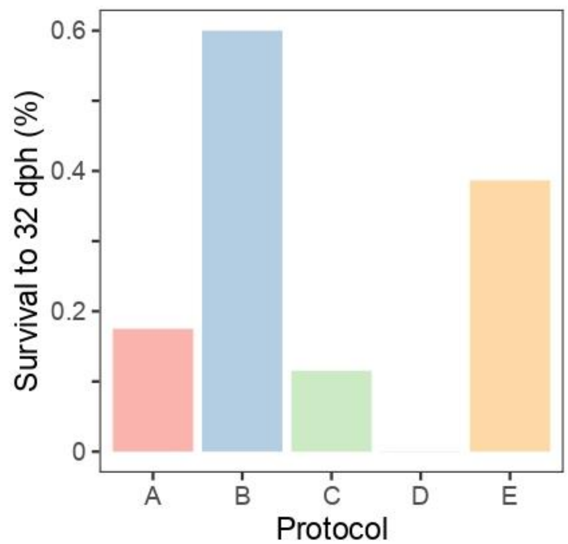

Figure 7. Percent survival to 32 dph for *Centropyge potteri* from feeding protocols A through E.

Many mass mortality events were observed throughout this experiment including when starting new feeds and a catastrophic hatchery-wide die-off. Large drops were seen when starting newly hatched Artemia on day 15 in protocols A and B, leading to protocols C and D which include adult copepods only instead of Artemia. No die-offs were seen in protocols C and D at 15 dph, however, a large mortality event was observed in protocol D when starting dry feeds at 13 dph, leading to the delay of dry feeds in protocol C. Protocol C experienced no major mortality events, indicating that the addition of Artemia and dry feeds too early was causing the die-offs seen previously. Protocol E did not experience any mass mortality events but did see a steady drop of 5 to 9 fish per day after starting algae paste on day 32. All trials active in March 2023 were cut short due to a hatchery-wide die-off event thought to be the result of copper toxicity from rainwater entering the recirculating aquaculture system.

Although 6 trial tanks were lost at once due to this major mortality event, survival to 32 dph was recorded for each protocol (Figure 7). Protocol D had no fish remaining at 32 dph while protocols A and C had survival of 0.12 and 0.18% respectively. Protocol E saw a survival of 0.39% while protocol B had 0.60% survival. Interestingly, while protocol A saw the highest growth rates, its survival rate is much lower compared to protocol B. In protocol B, the fish grew slightly slower compared to A but had much higher survival.

Prior to the implementation of the collection ban in Hawaii in 2021, the average retail price for wild-caught Yellow tang was $75. Post-ban, the scarcity of wild-caught specimens, coupled with increased demand, has led to a significant shift in pricing dynamics. Presently, captive-bred Yellow tangs are available in the market at an average retail price of $165, representing more than a twofold increase compared to their wild-caught counterparts. Similarly, the pre-ban retail price for wild-caught Potter's angelfish was approximately $250. Following the collection ban, the market has seen a substantial rise in the retail price of their captive-bred alternatives. Presently, captive-bred Potter's angelfish are available at an average retail price of $700, marking a nearly threefold increase compared to the pre-ban pricing of their wild-caught counterparts.

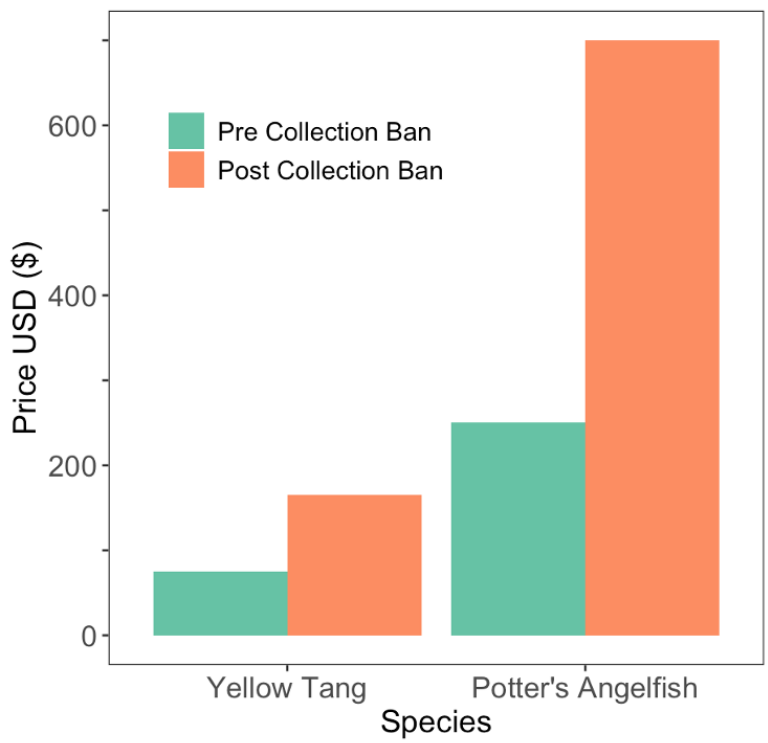

Figure 8. The average consumer price of wild caught Yellow tang and Potter’s angelfish specimens pre-collection ban compared to captive bred specimens post-collection ban.

I. A Win-Win Scenario?

Table 1: Proposed Policy Scenario derived from Schaar and Cox (2021)

Policy Scenario	Characteristics
Ban Collection for Direct Export, Permit to Collect for Captive Breeding, and Incentivise Aquaculture Programs	- Permits issued to stakeholders equipped with facilities that could house captive breeding operations for important aquarium species - Sample permit cost: $500 - Similar to the state's Special Activity Permits system - Standardized certification system suggested - Opportunity to export captive bred aquarium species from Hawaii - Unknown impacts on other fisheries, cultural resources, the environment, and on-reef tourism - State bares costs attempting to monitor and manage the fishery - State bares costs of incentives for aquaculture programs - Potential for non-compliance - Provides an alternate income for some stakeholders - Provides license fee revenues to state - Provides tax revenue to the state from created jobs - Encourages highly skilled workers and research institutions - Shifts value chain so that most value is accrued in the state, not to wholesalers and retailers - Costs of fishery management shifted to incentive options

Table 2: Costs and benefits associated with the Proposed Policy Scenario derived from Schaar and Cox (2021)

Proposed Policy Scenario Impacts	Cost or Benefit	Impacted Stakeholder Groups
Environmental costs avoided	+	Native Hawaiians, residents of Hawaii, tourists, all collectors and wholesalers in the state, and state government agencies

Tourism losses avoided	+	Residents of Hawaii, tourists, and state government agencies
Income benefits	+	Aquaculture programs, all collectors and wholesalers in the state
Management costs avoided	+	State government agencies
License/permit fees	+	State government agencies
Social benefits	+	Native Hawaiians, residents of Hawaii, all collectors and wholesalers in the state, and state government agencies
Possible indirect costs for other fisheries avoided	+	Native Hawaiians, residents of Hawaii, and state government agencies
Consumer interest in conservation	+	Aquaculture programs, native Hawaiians, residents of Hawaii, tourists, all collectors and wholesalers in the state, and state government agencies
Incentives for aquaculture programs	-	State government agencies

A. Cost and Benefit Descriptions

1. State Management Costs

Schaar and Cox (2021) found that DAR personnel in DLNR estimated their management costs of Hawaii's marine aquarium fishery to be USD $300,000 to $500,000 in 2019, a period when the fishery was not banned. This cost estimate includes the salaries of employees who work directly with the fishery, aquarium fish surveys, and any other costs associated with the fishery that DAR funds. In the proposed policy scenario, management costs should be eliminated over time or at least reduced as policies and regulations that support this scenario are refined.

2. Tourism Value

Estimates of tourism value and recreational benefits for Hawaii's near-shore reef environments have been investigated with estimates ranging from USD $304 million to USD $495.4 million per year (Schaar & Cox, 2021). It is assumed that wild collection of reef species will lower the tourism and recreational value of Hawaii's reefs.

3. Income and Revenue Accrued to Stakeholders

Aquaculture programs, local collectors, divers, and wholesalers would benefit from this policy scenario through subsidies and revenue from producing captive-bred fish for consumers. Local collectors and divers would not be able to export their fish, they would only be permitted to collect broodstock for aquaculture programs. It is assumed that collectors and divers benefit from this policy scenario as they can continue to profit from their profession legally compared to other policy options in which no collection is allowed.

4. License Fees

The proposed policy scenario explores an option where the permit cost is $500 annually for broodstock collectors with a cap of 100 permit holders statewide. This would bring revenue to the state in order to regulate and manage the broodstock collectors and divers to ensure compliance with permit regulations.

5. Exportation

Schaar and Cox (2021) discuss the value chain of exported species from Hawaii and found that the majority of the value of reef fish is accrued to the wholesaler and retailers. The proposed policy scenario brings forth regulations regarding the exportation of aquacultured fish from the state in which the value is accrued in the state. This could include creating a market for wholesalers or retailers within the state or regulating the export of fish to ensure benefit to the state.

6. Environmental Costs

Coral reefs are under a range of stressors such as climate change, ocean acidification, marine debris, nutrient pollution, algal overgrowth, and fishing pressure (Fujita et al., 2014; Friedlander et al., 2017). The marine aquarium fishery in Hawaii targets species that play important roles in their coral reef ecosystems, contributing to overall reef food webs and the epilithic algal matrices. Herbivorous fish play a crucial role in resisting regime shifts from coral to algal-dominated systems caused by climate change and local human-induced stressors (Graham et al., 2013). Studies have shown that spatial herbivore management through the establishment of Marine Protected Areas (MPAs) and Herbivore Fishery Management Areas (HFMAs) can maintain coral reef resiliency, particularly in severe and frequent bleaching events. Therefore, the over-harvesting of herbivores and corallivores in Hawaii's marine aquarium fishery could directly impede the state's reef recovery goals (Schaar & Cox, 2021).

7. Social Costs

The sustainability of the aquarium fishery in Hawaii has long been debated among various stakeholder groups, including aquarium fishers, hobbyists, scientists, native Hawaiians, resource managers, policymakers, and community members (McDermott, 2018). Concerns have been expressed over the social cost of the fishery, drawing in testimony from across the country and the world. A 2011 study showed that most fishers disliked the bureaucracy and conflict associated with the fishery (Stevenson et al., 2011). A 2017 survey of Hawaii residents found that 90% support further regulation of the fishery, and 83% support ending the trade altogether (Big Island Now.com, 2017). Due to these survey results, it is assumed that a ban on the aquarium fishery will benefit stakeholders socially.

8. Indirect Costs for Other Fisheries

Research has shown that overfishing is the main reason for the decline in reef fish populations in Hawaii, including food-fish species that overlap with commonly collected aquarium fish (Friedlander, 2017). Concerns have been raised about declining populations of some species targeted by the aquarium fishery in West Hawaii (DLNR, 2015). Of the top ten targeted aquarium species, three showed significant population declines, while five showed increases (DLNR, 2015). Overlap between target species in the aquarium fishery and sustenance-based fisheries is also a concern, including the Achilles tang (*Acanthurus achilles*), which is the third most collected species in Hawaii's marine aquarium fishery. The aquarium fishery in West Hawaii collects more reef fish than recreational and other commercial fisheries combined (DLNR, 2015).

9. Costs Associated with Collection of Broodstock and Wholesaling

Dierking (2002) found that local collectors face costs for boats, dive equipment, supplies, and license fees, while local wholesalers face costs for fish supply, facilities, boats, and shipping. The study indicates that the costs of local collection and wholesale business are high, and the profit margins are relatively slim. The proposed policy scenario provides subsidies to offset these high costs.

10. Costs Associated with Ornamental Aquaculture

Marine ornamental aquarium species fall into two main spawning types: demersal spawners and pelagic spawners. Demersal spawners lay clutches of eggs in nests or on substrates and form strong breeding pairs that display parental care, making them the preferred choice for most captive breeding aquarists. Pelagic spawners, such as Potter’s angelfish, release sperm and eggs into the water column to be carried freely by currents during peak spawning periods, making them notably more expensive and difficult to manipulate in simulated captive breeding settings. While technological advancements are being made by research institutions, there are many barriers that block the transfer of information in the field of marine ornamental fish rearing. Rhyne (2014) argues that reducing information access barriers can promote marine ornamental rearing, which can aid the marine aquarium community as well as the conservation of marine ornamental species. Due to the specialized knowledge and equipment required for the successful rearing of marine aquarium fish species, Schaar and Cox (2021) estimate the costs associated with collection and successful captive breeding to be much higher than those of a typical collection or wholesale operation in Hawaii. However, recent advances in culture technology have proven ornamental aquaculture to be a viable alternative as discussed previously.

11. Consumer Interest in Conservation

There are some who believe that the marine aquarium fish trade can contribute to the conservation of reef environments as owning aquarium fish is thought to increase awareness and interest in preserving their source habitats (Rhyne et al., 2014). It is assumed that increased numbers of Hawaiian fish within the aquarium trade will increase awareness and interest in conservation efforts of the reefs in Hawaii.

Discussion

The main goal of this study was to assess the larval growth of Potter's angelfish for the first time and discover a rearing protocol that optimizes both growth and survival of larvae to facilitate commercial-scale production of this species. While this study was limited in replicates (n = 2 for protocols A and B, n = 1 for protocols C, D, and E) and did not discover the most optimal protocol, observations and data gained throughout were integral in progressing culture efforts of Potter's angelfish. Rotifers and copepod nauplii and eggs were found to be suitable first feeds for *C. potteri* as larvae begin exogenous feeding at 3 days post hatch (dph) with live algae added twice per day to 300,000 cells/mL to provide contrast for larvae to see prey items, similar to established larviculture methods for Yellow tang (Pereira-Davison & Callan, 2017). Adult copepods and *Artemia* were found to be important in postflexion growth and survival.

Potter's angelfish were found to begin flexion at approximately 14 days post-hatch with growth rate impacting the onset and duration of flexion, consistent with the duration-stage hypothesis which states that an increased growth rate is advantageous for larval fish as it decreases the amount of time spent in critical and vulnerable stages (Anderson, 1998). The timing of flexion, 14 to 22 dph, is similar to that of other reef species including Yellow tang, *Zebrasoma flavescens*, and the Blue Mauritius angelfish, *Centropyge debelius* (Callan et al., 2018; Baensch & Tamaru, 2009). The early completion of flexion at 19 dph in protocol A indicates that the increased growth rate compared to other protocols leads to a shorter time to and duration of flexion. Body depth growth rate sharply increased with the onset of flexion while body length remained relatively constant. This is likely due to energy going into air bladder inflation and the development of a deeper, more developed body characteristic of postflexion angelfish larvae (Baensch & Tamaru, 2009).

Throughout the study, many mass die-off events were seen when starting new feed items, informing the creation of each subsequent protocol. The mitigation of these mass mortality events is what ultimately led to a tailored rearing protocol for Potter's angelfish. Large die-offs were observed when starting newly hatched *Artemia*, enriched *Artemia*, dry feeds (Otochime A1/A2), and algae paste (Nanno 3600). It is believed that Potter's angelfish larvae have difficulty fully digesting *Artemia* as stringy feces with visible *Artemia* exoskeletons were often observed hanging from the larvae's anus. While is unknown if the ability to fully digest *Artemia* is a function of development, gut microbiota, or other underlying mechanisms, it was observed that some individual larvae were able to digest *Artemia* earlier in development and more effectively than others indicating a genetic or developmental variation. The delay of newly hatched and enriched *Artemia*, to 19 and 27 days respectively, mitigated the die-off events previously observed further implicating development as an important factor in *Artemia* digestion.

The addition of other items such as dry feeds (Otochime A1 and A2) before the completion of flexion was also observed to cause mass mortality events. The underlying cause of this is unknown, however, we hypothesize that preflexion larvae either cannot digest prepared feeds or are sensitive to them chemically or physically. While a significant majority of fish still experience mortality during the early larval stage, consistent but small drops of postflexion larvae exert a notable influence on final production numbers. DiMaggio et al. (2017) reported comparable mortality rates, both in terms of daily occurrence and timing, while attempting to culture Pacific blue tang. This consistent pattern raises concerns about the vulnerability of postflexion-stage fish, particularly in the period following flexion and subsequent metamorphosis. Given the apparent stress during this phase, further exploration is essential to identify strategies for mitigating stressors and reducing associated mortality.

A significant dwindling of larvae along with a stagnation of growth was seen after switching from live algae to a concentrated algae paste (Nanno 3600) in protocol E. This switch was performed to conserve live algae usage while maintaining turbidity, however, the algae paste appeared to stress the larvae with a sudden increase in larvae 'fluttering' at the surface of the water. No significant drops or stress events were observed before this. The mechanism behind this stress reaction is unknown, however, increased stress has been observed in multiple species after the addition of algae paste.

While the survival and growth rate of larvae are thought to be directly related, this was not the case in protocol A, contradicting the "bigger is better" hypothesis (Litvak & Leggett, 1992). This could be explained by the limited number of replicates (n = 2), however, it is believed that this anomaly is due to rotifers not being offered until the larvae were 10 days old, causing high pre-flexion mortality. The small amount of remaining fish were subsequently larger on average compared to other protocols due to the lack of competition for prey items which compounded within the tanks on protocol A leading to an inflated growth rate in comparison to protocols B and E. The questionable nutritional benefits of *Artemia* could have also played a role in the protocol A anomaly, as *Artemia* have lower nutritional value than Copepods (Altaff & Vijayaraj, 2021). This was combatted with the enrichment of *Artemia* with essential amino acids and high concentrations of ascorbic acids, however, the effectiveness of enrichment can be uncertain. While *Artemia* offer a good source of energy for growth, the nutritional value of copepods can not be overstated. The significant differences in growth rates between protocols B and E could be attributed to the lack of adult copepods in protocol E, further indicating that copepods are important for the nutrition of postflexion larvae. However, adult copepods must be fed in conjunction with enriched *Artemia* to maximize growth and survival.

This study shows that Potter's angelfish require a multitude of live feeds throughout the rearing process including adult copepods and *Artemia*. The ideal protocol, given the results and observations of this study, would be Protocol E with adult copepods and consistent live algae. More research is needed to perfect the rearing of Potter's angelfish to mitigate postflexion-stage mortality, however, the results of this study led to the first cohort of *C. potteri* produced for consumers, clearly indicating that the continued commercial production of this species is possible. The advancements in culture methodology found in this study can be applied to other species, ultimately reducing the need for the wild collection of reef fish entirely.

The refined culture methods demonstrated in this research not only enhance the survival and growth rates of Potter's angelfish larvae but also provide valuable insights that contribute to the advancement of marine ornamental fish aquaculture that could have broader implications for the sustainable management of ornamental fish populations. The findings underscore the importance of optimizing larval rearing conditions, offering a practical approach for aquaculturists to improve the efficiency and success of larviculture programs. Moreover, the knowledge gained from this study may extend beyond Potter's angelfish to other species, benefiting the broader field of marine ornamental fish aquaculture, reducing the pressure on wild populations, and promoting the economic viability of this industry. As we continue to refine and implement these improved culture methods, the long-term impacts of increased efficiency on both the ornamental fish trade and marine ecosystems could be substantial, supporting the conservation of reef species while offering promising economic opportunities for sustainable aquaculture practices.

Impacts of Hawaii's Aquarium Collection Ban

The implementation of the collection ban had an immediate and noticeable impact on the supply of Hawaiian native and endemic species. As expected, the prohibition on wild collection led to a decrease in the availability of these species in the market. Meanwhile, the demand for these unique marine organisms remained constant, driven by the ongoing popularity of aquarium keeping. Prices for Hawaiian native and endemic fish surged to 2-3 times higher for captive-bred specimens (Figure 8). These findings underscore the economic impact of regulatory measures on the aquarium trade, with the increased prices reflecting not only market dynamics but also the costs associated with the research, development, and production of captive-bred alternatives. The stark contrast in pricing between wild-caught and captive-bred specimens highlights the potential economic viability and market acceptance of sustainable aquaculture practices in meeting the demand for marine ornamental species in a post-collection ban environment.

The significant increase in prices prompted a shift in the economic landscape of the aquarium trade, encouraging investment in the research and development of aquacultured alternatives. The ban acted as a catalyst for increased focus on the cultivation of marine ornamental species, particularly those endemic to Hawaii. This shift towards aquaculture aimed to meet the sustained demand for these species while mitigating the environmental impact associated with wild collection. One notable outcome of the increased investment in research and development is the successful commercial production of captive-bred Yellow tang. This achievement is a testament to the industry's adaptability and its ability to respond to regulatory changes. The aquacultured Yellow tangs not only serve as a sustainable alternative to wild-caught specimens but also contribute to the conservation of Hawaii's marine ecosystems.

Hawaii's 2021 collection ban had profound effects on the aquarium trade by reshaping the supply, demand, and pricing dynamics of native and endemic species. The scarcity of wild-caught specimens led to higher prices, prompting increased investment in the research and development of aquacultured alternatives. This shift has not only provided a sustainable solution to meet market demands but also contributed to the conservation of Hawaii's unique marine biodiversity. The success of the captive-bred Yellow tang and now Potter's angelfish exemplify the potential of aquaculture as a viable and environmentally conscious approach to sustaining the marine ornamental trade.

The success of ornamental aquaculture in Hawaii sets a precedent for global applications. The aquaculture techniques developed in response to the collection ban can be adapted and implemented in other regions facing similar challenges. The findings of this study emphasize the importance of investing in research and development to create viable and environmentally responsible alternatives to wild collection. As the global demand for marine ornamental species continues to grow, the lessons learned from Hawaii's policy changes can serve as a model for other regions to develop and implement sustainable aquaculture practices, contributing to both the economic viability of the industry and the conservation of marine biodiversity worldwide.

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