# The Effects of Venting and Decompression on Yellow Tangs (*Zebrasoma flavescens*) in the Ornamental Aquarium Fish Trade

## Prepared By

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#### Abstract

Ornamental aquarium fish catch volume may be affected by collection methods that cause mortality. To evaluate the effects of collection methods on fish stress and mortality, we examined three decompression treatments (fast(F), intermediate(I), slow(S)) coupled with(+V) or without venting(-V) (puncturing the swim bladder to release gas that has expanded during ascent from the reef to the surface) in a fully crossed orthogonal design on yellow tang (*Zebrasoma flavescens*) mortality and sublethal effects, as elucidated through histology and serum cortisol. In *Z. flavescens* subjected to F-V, post-collection mortality of 6% occurred within 24h of capture. Popular collection methods in the fishery (F+V and I+V) resulted in no mortality. Histopathology of heart, liver, head kidney, and swim bladder tissues in fish 0d and 21d post-collection revealed no significant inflammation or other lesions in any treatment group. F resulted in significantly higher serum cortisol than S, and venting alone did not affect cortisol. Future work should examine links in the supply chain following collection to determine if further handling and transport stressors affect survivorship and sublethal effects.

## Introduction

Each year, over 45 countries remove and export up to 30 million fish from coral reefs as part of the ornamental marine aquarium trade (Bruckner 2005; Wood 2001). Although ~90% of freshwater aquarium fish are successfully cultivated in aquaculture facilities, most tropical marine aquarium fish are wild-caught (Wood 2001) and their removal can have negative effects on coral reefs (Tissot et al. 2010). Mortality that occurs after live fish are removed from depth as part of the collection process is one issue that affects the impact of this fishery on aquarium fish populations. Although prior studies have examined the response of deep-water (20-152 m) fish to forced removal from depth, no studies have investigated the effects on shallow (15-18 m) tropical reef fish collected for the live ornamental aquarium fish, in which more fish are removed from the reef to replace losses (Stevenson et al. 2011). Live fish collection for the aquarium trade involves removal of reef fish from depth to the surface while transport encompasses the movement of collected fish to an export facility, where they are held prior to being shipped to an importer, and finally to an aquarium retail store where a consumer can purchase the fish for their home aquarium.

An important yet understudied component of live fish collection is the removal of fish from depth, subsequent transport to the surface, and the methods that must be implemented for fish to survive the pressure transition. As fish are brought to the surface, decreasing external pressure can result in barotrauma. Barotrauma signs in fish manifest both externally and internally and may include positive buoyancy (bloating caused by overexpansion of the swim bladder), bulging of the eyes (exophthalmia) and protrusion of the intestines from the cloaca. In deep water fish, protrusion of the esophagus from the mouth is common (Parker et al. 2006; Pribyl 2010; Wilde 2009). In addition, internal signs have been observed in deep water temperate fish which include swim bladder rupture, internal bleeding, compression damage to and displacement of organs surrounding the swim bladder, stretching of optic nerves, emphysema of the heart ventricle, and gas emboli in the rete mirabile and kidney caused by gas leaking from the swim bladder (Gotshall 1964; Bruesewitz et al. 1993; Parker et al. 2006; Rogers et al. 2008; Pribyl 2010).

Pribyl (2010) found that sublethal effects from barotrauma-related injuries such as rupture of the outer layer of the swim bladder (tunica externa) persisted for at least one month after collection in rockfish (genus *Sebastes*) without causing mortality. In addition, Hannah and Matteson (2007) determined that barotrauma could reduce post-release survival of fish through behavioral impairment. These findings indicate that sublethal signs may persist long after barotrauma occurs, and suggest that fish intended for the live ornamental trade may suffer sublethal effects that remain undetected.

Venting and decompression are two common methods that mitigate or prevent barotrauma, respectively, in both deep water food fishes and aquarium fishes. Though the use of these procedures in aquarium fisheries has been documented (Randall 1987; Pyle 1993; LeGore et al. 2005), the efficacy of each of these procedures in preventing aquarium fish mortality and sublethal effects has not been evaluated. Decompression at a rate that is natural to fish is determined by the fish's ability to remain neutrally buoyant in the water column while being decompressed. In deep-water temperate fish, natural decompression can take days (Parker et al.

2006; Pribyl 2010). Performing one or several decompression stops (pause in the water column at an intermediate depth before removal to the surface) is a technique implemented in aquarium fisheries such as those in Puerto Rico and West Hawaii (LeGore et al. 2005; Stevenson et al. 2011), though its effectiveness at preventing mortality has not been tested. Decompression can be a more time-consuming process that allows fish adequate time to naturally remove the greater volume of air in its swim bladder, thus preventing barotrauma. In contrast, venting allows for fish to be brought rapidly to the surface by puncturing the swim bladder with a hypodermic needle to release excess air in the swim bladder, thus mitigating barotrauma, but potentially causing organ and tissue damage.

Studies on deep-water food fisheries show conflicting results on venting as an effective practice to mitigate barotrauma and increase fish survival (Gotshall 1964; Keniry et al. 1996; Collins et al. 1999; Kerr 2001; Nguyen et al. 2009; Wilde 2009). Different depth ranges appear to present different survival rates among fish treated with venting (Gotshall 1964; Collins et al. 1999; Wilde 2009), though it is difficult to determine a strong depth pattern associated with venting mortality because few studies have closely examined this interaction, which may also be species-specific. The outcomes of these studies also appear to be based on whether short- or long-term fish survival is examined (Keniry et al. 1996), which is a product of the experimental design: either short-term holding, or long-term mark and recapture. We expect that long-term captive observation of fish post-collection will resolve this issue.

Recently proposed legislation in Hawaii reflects the attempts of a local animal rights group to ban the harvest of marine species for the aquarium trade based on animal cruelty claims and concerns about post-collection mortality (Lauer 2011; Talbot 2012a; Wintner 2010, 2011). Venting is popular among aquarium fishers, but is a disputed collection method in West Hawaii. Opponents claim venting inflicts suffering and mortality on fish, while collectors maintain that venting benefits fish brought up from depth and is necessary for fish survival. Those opposing venting have suggested that slow decompression be used in its stead. This controversy highlights the paucity of data surrounding collection practices in the aquarium industry, not only in Hawaii, but globally. Thus, research on mortality as it relates to collection is warranted. Legislation should be based on sound science, which should also lay the groundwork for best management practices that are both ecologically sustainable and economically practical.

Although aquarium fisheries where destructive fishing practices like cyanide are used may have high rates ( $\geq 90$  %) of fish mortality (Rubec and Cruz 2005), Stevenson et al. (2011) estimated the short-term mortality rate of fish collected in West Hawaii as <1 % (from collection to harbor). Although short-term mortality is low, it is possible that delayed mortality occurs as a result of sublethal effects and remains undetected in the long-term. Because of the rapid movement of fish through the supply chain, fishers and exporters in West Hawaii may be unaware of specific connections between collection or handling methods and mortality further down the supply chain. Moreover, delayed mortality could shift the burden of fish death and economic loss from the collector to an exporter, or to the importer, retailer, or hobbyist. Importantly, improvements in the collection and transport methods for fish could potentially reduce overall mortality of aquarium fish, thus reducing the numbers of fish that must be removed from the reef to compensate for losses in various stages of the supply chain.

Given the conflict surrounding collection practices in Hawaii and the paucity of data regarding barotrauma mitigation practices in the aquarium trade, it is clear that a scientific study is needed to investigate the practices used by fishers, while providing recommendations for best management practices. To meet these goals, we address the following objectives:

- 1. Determine short- and long-term mortality of reef fish caught for the aquarium trade subjected to barotrauma prevention and/or mitigation practices: decompression and venting, respectively.
- 2. Examine sublethal effects of collection that could result in delayed mortality: gross and histologic lesions, and stress.

# **Materials and Methods**

#### **Experimental Design**

To examine short- and long-term mortality of ornamental aquarium fish as it relates to collection practices, fish were subjected to different collection methods and subsequently held for 21 days (d) for observation. Fish suffering mortality were examined by histopathology to identify specific causes of death. A subset of surviving fish were also examined to determine if sublethal injuries at the tissue level were present. Serum cortisol concentration was also measured as a proxy for stress in fish (Donaldson 1981).

*Z. flavescens* was selected as the study animal because it is the most common species targeted by aquarium fishing in West Hawaii, making up between 65-80% of the total catch of aquarium fish (Cesar et al. 2002; Tissot and Hallacher 2003; Walsh et al. 2004; Williams et al. 2009). Understanding how collection practices affect yellow tang survivorship and health is especially relevant to the West Hawaii aquarium fishery.

A fully crossed factorial experiment was conducted, with three levels of decompression treatment (fast, intermediate, slow), and two levels of venting (no, yes) in all possible combinations (k=6 treatments). Each treatment was replicated three times, with n=20 fish in each treatment combination for a total of 360 individuals. Fish were subsampled (n=5) immediately following collection in each treatment replicate to establish a baseline for histopathology and assess post-collection cortisol level. Fish were collected between 15-18 m depth, reflecting the range typically utilized by West Hawaii collectors (Stevenson et al. 2011). In order to accurately reflect the methods actually used in the fishery, fish collection was performed by a licensed aquarium fisher on SCUBA with a barrier net as described by Stevenson et al. (2011). When the desired quantity of fish (n=40) was caught, fish were transferred to containers assigned to each decompression treatment. Following the decompression treatment, half (n=20) of the fish were subjected to the venting treatment and half were not.

The fast decompression treatment involved bringing the fish directly to the surface from 15-18 m at a rate of 0.25 m·s<sup>-1</sup> (recommended SCUBA ascent rate) resulting in ~1 min total decompression time. In the intermediate decompression treatment, the collection container was hung off the fishing vessel and brought up at a rate of 0.25 m·s<sup>-1</sup> to half the maximum depth. Fish

were allowed to decompress for 45 min before being brought to the surface at a rate of  $0.25 \text{ m} \cdot \text{s}^{-1}$ . In the slow decompression treatment, fish were brought up 3 m every 15 min until the collection container was at 10 m (2 atm), then 1.5 m every 15 min from 10 m to the surface. Fast decompression followed by venting is a method applied most frequently by fishers on yellow tangs in West Hawaii and 100% of fishers interviewed reported using venting, ranging from occasionally to always, depending on depth and target species.

Venting was performed by the fisher on the surface vessel using a 20 G hypodermic needle, replaced after  $\sim$ 50 fish. Each fish was held out of water for  $\sim$ 3-5 s by the fisher while the needle was inserted through the body wall toward the swim bladder, caudal to the pectoral fin and ventral of the lateral line.

Fish were collected over a period of 2 d. For each collection day, the duration of transport from the collection site to port (<1 h) and from port to the holding facility was similar (<1 h) to reduce uncertainty associated with differences in fish transport time. During transport, each replicate treatment group was held separately in the collector's live well. During collection and while in transit from the collection site to port, the water in the live well was continuously exchanged with fresh seawater.

#### **Holding Period**

Post-collection, fish were observed for 21 d at an aquaculture facility located at the Natural Energy Laboratory Hawaii Authority in West Hawaii provided with surface seawater. The experiment duration was chosen because it represents a reasonable time period for a fish to be transferred from the reef to a retailer or hobbyist in the aquarium supply chain. In addition, swim bladder healing in rockfish has been observed after 21 d (Parker et al. 2006) and is sufficient time to allow skin and muscle regeneration in fish (Roberts 2010). Therefore, fish exhibiting lesions after 21 d may not have fully recovered in a supply chain environment and could be categorized as having sublethal effects from collection.

Fish were held in 1 m diameter mesh floating cages within three ~10 000 l pools, which served as replicate blocks, each containing all six treatments. Incoming seawater was filtered to  $5\mu$ m, and set to flow through each pool at a rate of 1 volume·d<sup>-1</sup>. Regular skin scrapes were collected and if parasites were detected, a hyposalinity treatment was implemented (30-14°/°) for the entire pool. Pools were exposed to natural sunlight, and minimum and maximum daily temperatures were monitored.

All fish were fed a natural algae diet (*Ulva fasciata*) rich in nutrients (primarily nitrogen) absorbed from food fish outflow in the aquaculture facility. Aquaculture facilities use algae such as *Ulva spp*. for biofiltration (Vandermeulen and Gordin 1990; Jiménez del Río et al. 1996) and the algae accumulates nutrients and can serve as a nutrient rich food source for herbivorous fish.

Fish health was observed, and any mortality was recorded. Standard length (SL) (from snout to start of caudal fin) of each fish was measured. Following mortality, fish were placed in 10% neutral buffered formalin for histopathology; the operculum was removed and body cavity

opened to facilitate flow of formalin fixative over tissues. Moribund fish were humanely euthanized using an overdose solution (> 250 mg $\cdot$ l<sup>-1</sup>) of tricaine methanesulfonate (MS-222).

## Histopathology

To determine the sublethal effects of decompression and venting treatments, fish (n=5) were chosen randomly from each replicate treatment group immediately upon arrival to the holding facility (0 d) and at the end of the holding period (21 d) for histopathology. Fish used for histopathology were euthanized using an overdose solution of MS-222, placed on ice, and shipped within 48 h to Oregon State University's (OSU) Veterinary Diagnostic Laboratory (VDL) for analysis. Fish that perished during the experiment were fixed in 10% neutral buffer formalin and sent to the VDL for histopathology.

Formalin-fixed fish were immersed for 24 h in Cal-Ex II (Fisher Scientific) to decalcify bone, after which serial cross sections of the fish were placed in plastic cassettes and processed using standard histologic techniques. Paraffin embedded specimens were sectioned at 5  $\mu$ m and stained with hematoxylin and eosin. Brown-Hopps Gram stain was used to assess bacterial growth. Using a Nikon Eclipse 50i microscope, gill, heart, kidney, liver, swim bladder, and intestine tissues were examined for evidence of histologic lesions.

#### Cortisol

Because of the potential for cortisol concentrations to decrease when a stressor subsides, blood samples were collected from fish immediately upon arrival to the holding facility (0 d). Fish (n=2) were anesthetized from each treatment replicate group using MS-222 prior to drawing 0.3-1.0 ml blood from the heart using a 25G 2.54 cm needle and 3 ml syringe. To determine *Z. flavescens* ocean baseline cortisol concentration, blood was collected from fish (n=4) underwater on SCUBA at capture depth within 3 min of capture. Blood was injected into 3 ml vacutainer tubes with no additive (Becton-Dickinson), placed on ice, and centrifuged at 3,000 rpm for 10 min <1 h later. Serum supernatant was transferred to a clean vacutainer tube with no additive, placed on ice, and frozen <1 h later for  $\leq$  40 d in a non-frostless freezer, and transported overnight on dry ice to the OSU Department of Fisheries and Wildlife for analysis.

Serum cortisol concentrations were determined using radioimmunoassay (RIA) as described by Redding et al. (1984). Total binding, the ratio of the radiolabeled cortisol bound to the antibody to the total amount of radiolabeled cortisol in the sample, was 40-50%. Samples showed adequate parallelism, and 3.9-500.0 ng/ml cortisol standards were used.

# **Statistical Methods**

Statistical analyses were performed using Minitab 15 Statistical Software program. To meet assumptions of normality and homogeneity of variance, data were transformed to square root (fish SL) or log (cortisol). A two-way ANOVA was used to compare mean cortisol concentrations, with decompression treatment and venting as fixed factors and replicate block as a random factor. Tukey's multiple comparisons test was used to determine significant differences between levels within each factor.

# Results

# Mortality

Sizes of Z. *flavescens* in this study ranged from 5.0-10.0 cm SL with a mean value of 7.2 cm (SE=0.05 cm), similar to sizes previously reported in the fishery (Stevenson et al. 2011). Mortality occurred <24 h post-collection in fish subjected to fast decompression with no venting, with a mean mortality of 6.2% (SE=0.6%). No mortality occurred in the other experimental treatments, negating need for statistical tests.

The incidence of mortality was consistent with observations of the frequency and severity of external barotrauma signs. These included high frequency of positive buoyancy, bloating, exophthalmia, and protrusion of the intestines from the cloaca in fish subjected to fast decompression (Figure 1). The intermediate decompression treatment resulted in some fish being bloated and positively buoyant, but the other symptoms were not as frequently observed. Fish subjected to slow decompression did not exhibit these signs.

# Histopathology

Histopathology of gill, heart, kidney, liver, swim bladder, and intestinal tissues failed to detect significant inflammation, necrosis, or gas embolism associated with barotrauma or venting in any treatment either pre- or post-collection. However, in one case a venting wound was detected in a fish subjected to slow decompression and venting, which was sampled immediately after collection. In this sample (Figure 2), there was locally extensive necrosis of body wall musculature and a localized influx of neutrophils surrounding the needle track.

#### Cortisol

The mean ocean baseline cortisol concentration was 8.9 ng·ml<sup>-1</sup> (SE= 4.96 ng mL<sup>-1</sup>) and in some cases was at or below the detection limit for the assay (3.9 ng·ml<sup>-1</sup>). Decompression treatment significantly affected cortisol concentration (Two-way ANOVA: F=4.26; df= 2,10; p=0.03), with fast decompression resulting in a significantly higher mean cortisol concentration (M=58.8 ng mL<sup>-1</sup>, SE=8.7 ng mL<sup>-1</sup>) than slow decompression (M=35.5 ng mL<sup>-1</sup>, SE=5.3 ng mL<sup>-1</sup>), with neither treatment being significantly different from intermediate decompression (M= ng mL<sup>-1</sup>, SE= ng mL<sup>-1</sup>) (Figure 3). Fast decompression produced the highest observed cortisol concentration (101.49 ng mL<sup>-1</sup>), whereas the highest observed cortisol concentrations in fish subjected to intermediate and slow decompression were 59.09 and 68.03 ng mL<sup>-1</sup>, SE=6.9 ng mL<sup>-1</sup>) than the no venting treatment (M=38.2 ng mL<sup>-1</sup>, SE=4.3 ng mL<sup>-1</sup>), but differences were not statistically significant. In addition, there was no significant interaction between decompression and venting.

# Discussion

The conflict between fishers and opponents of aquarium collecting in Hawaii is not only driven by the competition for tropical fish, but also by values differences (Capitini et al. 2004; Tissot 2005; Stevenson et al. in review). Because values are involved, science alone may not be sufficient to settle the dispute. However, because conflict surrounding the aquarium trade also suffers from scientific uncertainty (Capitini et al. 2004), we aim to provide the science necessary for full evaluation of potential management solutions directed at collection practices.

With the objective of informing management on collection practices in the West Hawaii aquarium trade, our study focused on the short- and long-term mortality of reef fish subjected to two different barotrauma mitigation practices: venting and decompression. Overall, we found that varying decompression methods produced little to no mortality and that venting reduced mortality in fish subjected to fast decompression. Furthermore, we found no evidence of significant tissue inflammation associated with venting, or lesions linked to barotrauma immediately after collection and after a 21 d holding period. Finally, decompression resulting in significantly higher serum cortisol concentrations than slow decompression. However, consistent with mortality observations, venting did not significantly affect cortisol concentration. In the following sections, we discuss possible factors influencing our results, future research recommendations, and implications for management of aquarium fisheries.

#### Mortality

A popular method used to collect yellow tangs and other aquarium fish species, fast decompression followed by venting, resulted in no mortality. Only fish subjected to fast decompression with no venting suffered post-collection mortality. Our finding that mortality rates in *Z. flavescens* are low is consistent with prior observations of the fishery by Stevenson et al. (2011). Our results indicate that venting following decompression does not cause short- (0 d post-collection) or longer-term (21 d post-collection) mortality. Venting likely improved survivorship in fish following fast decompression because venting causes fish to become neutrally or negatively buoyant, allowing them to control position in the water and avoid colliding with the transport container during transport to the holding facility. This is contrasted with fish subjected to fast decompression with no venting, which were positively buoyant and therefore at risk of acquiring secondary injuries during transport, as well as aerial exposure.

Additional factors that influence post-collection fish mortality include collection depth, body size, and species. Our study examined fish collected from 15-18 m depths, which is typical for most West Hawaii fishers, although some dive to  $\geq 27$  m for different species (Stevenson et al. 2011). At these deeper depths, the effects of decompression rate and venting may differ, and it is known that fish mortality and occurrence of barotrauma increases with capture depth (Collins et al. 1999; St John and Seyers 2005; Hannah et al. 2008; Jarvis and Lowe, 2008; Campbell et al. 2010). Interviews with West Hawaii fishers indicate that fish collected from >25 m require more decompression time and venting at depth, or several venting applications during ascent. Fishers have also mentioned that larger fish exhibit more severe external barotrauma symptoms than smaller fish of the same species, which is similar to findings in studies on deep water fish (Hannah et al. 2008; St John and Seyers 2005). Just as different deep water fish species exhibit different responses to decompression (Hannah and Matteson 2007; Jarvis and Lowe 2008; Pribyl 2010), aquarium fish species reportedly react differently to decompression and venting. These differences are likely caused by variation in body shape and durability, and swim bladder volume between species. Methods used by fishers reflect these species differences, with practices such as performing venting on more delicate, soft-bodied fish like angelfish (Pomacanthidae) underwater to prevent (rather than mitigate) swim bladder expansion. Examining differences among aquarium fish species of varying sizes and investigating the variety of techniques employed by

fishers during collection would provide further insight into the prevalence and effectiveness of aquarium fish barotrauma prevention and mitigation methods.

# Histopathology

Histopathology did not detect significant widespread inflammation, organ damage or infection caused by venting. Only one case of a needle wound was found that showed some localized inflammation, with no visible bacteria. Because the microtome used in the histological process sliced tissue into a 5  $\mu$ m sections, there may have been other fish with similar minor wounds that were undetected. However, the object of histopathology in our study was to determine if widespread inflammation or tissue damage were present in fish, indicating serious sublethal injury. If such injuries were present, they would have been detected in multiple sections of the tissues surrounding the venting wound. The fish in which the venting wound was detected was sampled 0 d post-collection, and serious infection is not be expected within hours of collection. Rather, inflammatory and epithelial cells mainly occupy the area of the wound within a few hours of injury (Roberts 2010), and invading bacteria would not have spread far from the wound in this time.

If fish were unable to heal in the 21 d holding period because of wound severity, we would have detected significant infection in fish sampled after 21 d. Because we did not, our results indicate that the venting procedure does not pose a significant threat to fish survival post-collection, nor cause significant sublethal effects. However, we caution that the fish in our study were held in an aquaculture facility for 21 d without additional handling and transport stressors as they normally would in the supply chain, thus potentially promoting recovery from injuries inflicted during collection. Future studies should investigate if additional stressors of the supply chain diminish the efficacy of venting in promoting long-term fish survival.

Because aquarium fish exhibited external signs of barotrauma similar to those observed in deep-water temperate fish, we expected internal barotrauma symptoms to be present similar to those described by Pribyl (2010). We did not detect tissue-level signs of barotrauma, even in fish subjected to fast decompression. However, externally observable signs indicated that barotrauma did occur and that aquarium fish likely also experience some internal signs of barotrauma. Because positively buoyant fish were bloated and showed protrusion of the intestines from the cloaca, with some presenting exophthalmia, organ displacement by the swim bladder and stretching of the optic nerve caused by exophthalmia were likely occurring immediately following collection, internal barotrauma signs detected in rockfish (Parker et al. 2006; Rogers et al. 2008). Determining if organ displacement occurs, and if venting relieves this issue in aquarium fish would further our understanding of the mechanisms with which venting reduces mortality in fish subjected to fast decompression.

# Cortisol

Fast decompression produced significantly higher cortisol levels in fish than slow decompression. This higher cortisol level may be driven by a higher frequency and severity of barotrauma signs in fish subjected to fast decompression. Because fish endured transport to the

holding facility prior to blood collection, fish exhibiting positive buoyancy (those not subjected to venting) should have produced higher cortisol concentrations because they were more susceptible to injury and stress from collision with the transport container. Venting, applied prior to transport, should reduce exposure to this transport stressor by eliminating positive buoyancy. However, though venting mitigated the effects of fast decompression by relieving positive buoyancy and reducing mortality, our results show cortisol concentrations were not significantly affected by venting. This suggests that other signs of barotrauma not relieved by venting (i.e. intestinal protrusion and exophthalmia), or experiencing positive buoyancy even if only for a short period of time, are likely driving the higher cortisol concentrations observed in fish subjected to fast decompression.

Comparisons with stressed (45-65 ng•ml<sup>-1</sup>) and non-stressed (10-25 ng•ml<sup>-1</sup>) plasma cortisol concentrations in a closely related acanthurid (*Ctenochaetus striatus*) (Soares et al. 2011) suggest that venting increases stress in fish subjected to fast decompression, but our results show that it also reduces mortality. However, because cortisol concentrations vary widely between fish species, these values should be interpreted cautiously. Because fast decompression followed by venting is a popular method used to collect yellow tangs, other ornamental aquarium fish, and food fish species, further investigation of stress following collection is warranted.

Handling in and transport between export, import, and retail facilities may exacerbate stress caused by collection. Chronic stress results in immune system suppression (Barton 2002), increasing susceptibility to infection and disease and the probability of delayed mortality. Because mortality occurring in hobbyist aquariums followed by fish replacement is one driver of the demand for more aquarium fish (Tissot et al. 2010), future studies should address stress as it relates to handling in and transport between each link in the supply chain beyond collection. In addition, tracking supply chains originating both inside and outside the US and investigating how fish stress relates to water quality, fish behavior, sublethal effects, and mortality would provide insight into drivers of stress and mortality in the aquarium fish and live food fish trade and lead to improved methods.

#### **Implications for Management**

Venting did not cause mortality or sublethal injuries in yellow tangs, and banning venting may increase mortality rates if fishers continued to implement fast decompression. However, only allowing intermediate or slow decompression might be economically detrimental to fishers.

Our results further our overall understanding of the effects of venting. Previous studies show conflicting results on venting's effectiveness, both supporting and refuting the efficacy of its use for reducing fish mortality (Gotshall 1964; Keniry et al. 1996; Nguyen et al. 2009; Wilde 2009). Our results indicate that when performed properly, venting does not cause mortality or inflict significant sublethal injuries, although our inference is limited to a single species and within a relatively shallow depth range.

In contrast to other aquarium fisheries such as in the Philippines where cyanide is used and increases fish and coral reef mortality (Hall and Bellwood 1995; Hanawa et al. 1998; Rubec et al. 2001; Jones and Hoegh-Guldberg 1999; Jones and Steven, 1997), fishers in West Hawaii appear to implement non-destructive collection practices. Although the Hawaii fishery contributes fewer ornamental aquarium fish to the global exported catch relative to Indonesia and the Philippines (Wood 2001; Walsh et al. 2004), if methods used by fishers in West Hawaii were to be implemented by these large scale aquarium fisheries where destructive fishing methods are popular, sustainability of the global trade would improve. To this end, we recommend that other aquarium fisheries adopt methods used in West Hawaii, which are economically feasible and foster sustainability.

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# **FIGURES**

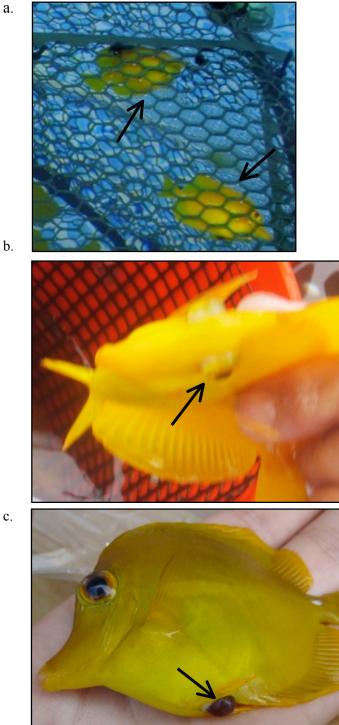


Figure 1: Barotrauma signs observed in yellow tangs following collection: (a) positive buoyancy (b) exophthalmia and (c) intestinal protrusion from the cloaca.

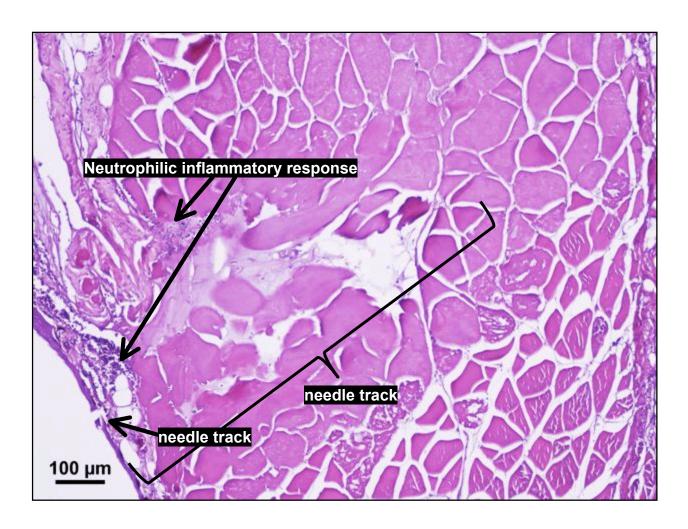


Figure 2: Histological section of needle track in a yellow tang subjected to venting showing muscle cell necrosis, edema, and neutrophilic inflammation, at 10x magnification.

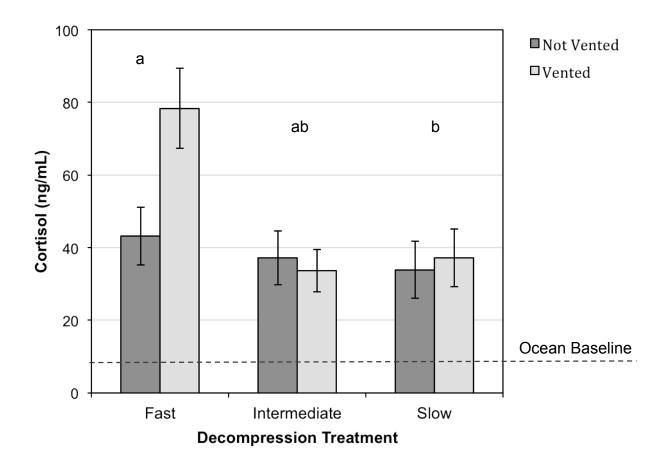


Figure 3: Cortisol concentration (mean  $\pm$ -SE) by each treatment. Letter groups represent Tukey's multiple range test results comparing means between decompression treatments. All treatment groups are above the ocean baseline concentration of 8.9 ng mL<sup>-1</sup>.



Figure 3: Comparison of buoyancy in yellow tangs subjected to the fast decompression treatment, before and after venting was applied by an experienced aquarium fisher.