Testing the grouper biocontrol hypothesis: A response to Mumby et al. 2013

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Abstract

Biotic resistance is the idea that native species negatively affect the invasion success of introduced species. We tested the hypothesis that native grouper are controlling the abundance of exotic lionfish on Caribbean coral reefs by assessing the relationship between the density and biomass of lionfish and native predators at 71 reefs in three biogeographic regions. Our results indicated that: (a) the abundance of lionfish and large grouper are not negatively related, and (b) lionfish abundance is controlled by a number of physical site characteristics, and possibly by culling. Taken together, our results suggest that managers cannot rely on native grouper populations to control the lionfish invasion. Mumby et al. (2013) objected to several aspects of our analysis and conclusions. Here we address their criticisms and demonstrate that our original conclusions are valid.

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Overview

We recently published a paper (Hackerott et al. 2013) in which we tested whether biotic resistance by native predators influenced the invasion success of lionfishes (*Pterois volitans* and *Pterois miles*), piscivores from the Indo-Pacific. The following is the abstract of our paper:

"We surveyed the abundance (density and biomass) of lionfish and native predatory fishes that could interact with lionfish (either through predation or competition) on 71 reefs in three biogeographic regions of the Caribbean. We recorded protection status of the reefs, and abiotic variables including depth, habitat type, and wind/wave exposure at each site. We found no relationship between the density or biomass of lionfish and that of native predators. However, lionfish densities were significantly lower on windward sites, potentially because of habitat preferences, and in marine protected areas, most likely because of ongoing removal efforts by reserve managers. Our results suggest that interactions with native predators do not influence the colonization or post-establishment population density of invasive lionfish on Caribbean reefs."

Mumby et al. (2013) posted a critique of our manuscript. Here we respond to their main points.

Criticisms (in italics) with text from Mumby et al. 2013 in grey and our responses

1) Confounding effects of habitat, larval dispersal, and fishing

Mumby et al (2013): "Hackerott et al. aim to prove a 'negative' result of grouper on lionfish while failing to account for a multitude of processes that could likely obscure any relationship that might exist. The numbers of grouper and lionfish will vary from site to site for many reasons other than whether they exhibit a predator-prey relationship. Fishing has a major effect on grouper (Coleman et al. 2000) and increasingly, a significant effect on lionfish. Habitat quality, particularly rugosity, is a major driver of fish abundance and mediates predator-prey interactions (Hixon and Beets 1993). Larval dispersal is likely to be an important factor for lionfish, particularly given the very recent colonization of the species. All of these factors will vary dramatically among study sites around the Caribbean yet none are measured nor accounted for. The only attempt to control for fishing is to include reserve effects but it is well established that very few reserves have any significant effect on grouper (see AGRRA dataset, included in Mumby et al 2011)."

We agree with Mumby et al. (2013) that accounting for potential covariates is essential when evaluating the importance of any single factor in a spatial-comparative study. Fishing, larval dispersal, habitat quality, reef rugosity, depth, and myriad other factors control the population dynamics of lionfish, grouper, and all other reefs inhabitants. Work by Mumby et al. (2011) on the relationship between lionfish and grouper took place at only two locations, and did not present data on other processes that may have differed between the locations. The aim of Hackerott et al. (2013) was thus to examine the generality of Mumby et al. (2011)'s observation at a much broader and heterogeneous scale, while accounting for as many site-specific covariates as possible. The site-specific parameters included as covariates in our statistical model were wind exposure, habitat type, protection status, depth, and time since invasion (Figure 1). Accounting for these covariates, we found no relationship between predators and lionfish

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abundance (Hackerott et al. 2013). However, these covariates do appear to significantly influence lionfish density and biomass (Figure 1).

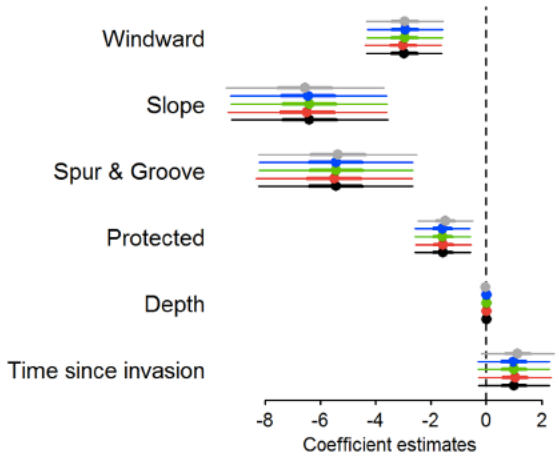

Figure 1. Portion of the glmmADMB models that show site-specific parameters reproduced from Fig. 3 of Hackerott et al. (2013). Coefficient estimates, ±1 standard deviation (thicker horizontal lines) and ± 95% confidence interval (thinner horizontal lines). Each color represents a model for either density or biomass of biotic groups. See original paper for details. The effect of biotic groups on lionfish is not shown.
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We have reanalyzed the data from Hackerott et al. (2013) with two additional covariates (reef rugosity and fishing pressure) to determine the effects of grouper biomass on lionfish abundance (Figure 2). When reef rugosity (structural complexity) and human population density/reef area (an accepted proxy for fishing pressure; Newton et al. 2007, Mora 2008, Stallings 2009) were included in the model, neither had an effect on lionfish abundance and there was still no significant relationship between lionfish and grouper abundance (Figure 2). The effects of other covariates (namely, wind exposure, protection status, and habitat type) on lionfish abundance remained the same (Figure 2) as in the previous models where reef rugosity and fishing pressure were not included (Figure 1). See Appendix 1 for methods and more detailed information on this new analysis.

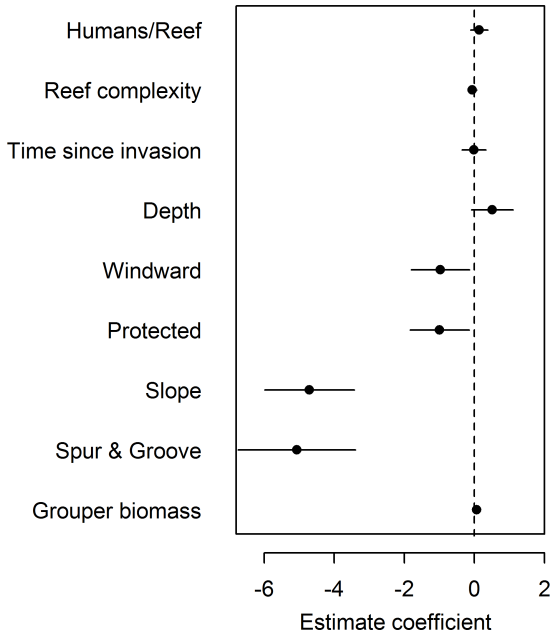

Figure 2. Coefficient estimates showing the effect of all the co-variables and grouper biomass on lionfish abundance based on a GLMM results. The 95% CI of some variables are very small because the scale of numerical and categorical cofactors are different. See Appendix 1 for effect values.

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We agree that the combined strength of these abiotic factors, relative to a potential effect of grouper, is likely why we did not observe a negative relationship between grouper and lionfish across sites. However, this reinforces our initial conclusion: factors other than predator abundance are playing a more important role in limiting lionfish abundance across the region. Indeed, the contribution of any one factor must be interpreted in the context of all other factors that are simultaneously acting in the system. Thus, the relative importance of a single factor can only be evaluated when a study's sampling scheme captures the true heterogeneity in conditions present within the system, as we have done. While predators may negatively impact lionfish under a particular set of local conditions (i.e., Mumby et al. 2011), this effect is undetectable on a wide range of sites across the Caribbean region.

Mumby et al (2013): "Larval dispersal is likely to be an important factor for lionfish, particularly given the very recent colonization of the species"

Given the context of our study, presumably, what Mumby et al. (2013) meant was that larval *supply* (i.e., settlement, rather than dispersal) could influence site-specific lionfish abundance. While undoubtedly true, such data is not available for our sites. Additionally, while measuring supply (or dispersal, recruitment, connectivity, larval export, etc.) would have been interesting, it was well outside the scope of our study, both in terms of the goal of the study, as well as the large number of sites included and the regional scale of the analysis. Although we have identified a number of factors that appear to influence local invasion success, the goal was not to identify or test every potential factor. Instead, the purpose of our study, as previously discussed, was to quantify the effect of predator abundance on lionfish abundance relative to other abiotic factors.

Mumby et al (2013): "Hackerott et al attempt to stratify data by reef zone is inadequate. It is well established that forereef slopes in the Caribbean encompass at least two contrasting habitats: structurally complex "Montastraea" reefs (i.e., Orbicella reefs) versus structurally simple hardbottom dominated by gorgonians (Chollett and Mumby 2012). The fish assemblages differ dramatically between these habitats (Harborne et al. 2008, Mumby et al. 2008) and failure to distinguish which sites lie in one habitat versus the other constitutes a genuine confounding effect."

We agree that there is a large degree of variation in what constitutes a "fore-reef" environment, in the Caribbean and elsewhere. As per Mumby et al. (2013)'s description, all of our "fore-reef" sites constituted high-profile slope. We distinguished between these forereef slopes and both spur-and-groove and patch reef sites, which are common habitat types in the Caribbean that were not considered by Mumby et al. (2011). Both lionfish and large-bodied grouper (classified as >30cm TL as per Mumby et al. 2011) are present in each of these habitats. It is important to determine whether a relationship also exists in these habitats to determine if native predators can control lionfish across the Caribbean region, as was the goal of our study.

Mumby et al (2013): "The authors also mix reef slopes with spur and groove zones, and again the additional large-scale complexity offered by coral spurs intersected by sand grooves can affect fish communities. The varying geomorphology between spur and groove zones and reef slopes are also typically driven by their location in different physical environments, which needs to be accounted for in any analysis pooling across reef zones".

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Our statistical models account for the varying geomorphology between reef habitats by including exposure, depth, rugosity, and habitat type (i.e., the three types described above) as covariates (see Figs. 1 and 2 above), therefore, we did not "pool across reefs zones." Also see text above about the purpose of the study: the aim was to explicitly incorporate physical variation among reefs as part of the design.

2) Insufficient grouper size

Mumby et al (2013): "Finally, 25 (35%) of the sites [in Hackerott et al. 2013] are shallow patch reefs from The Bahamas. Large grouper have well-established habitat preferences for deeper water and during extensive surveys on these patch reefs, one of us (ARH) has rarely seen large grouper comparable to those found in the ECLSP. Therefore, while high density might allow the total biomass of grouper on patch reefs to be high, the sizes of grouper - and therefore their predatory capacity - is substantially less than that of forereef populations and likely incomparable to our study of the ECLSP. Therefore, while high density might allow the total biomass of grouper on patch reefs to be high, the sizes of grouper - and therefore their predatory capacity - is substantially less than that of forereef populations and likely incomparable to our study of the ECLSP. Moreover, the fish assemblages on patch reefs, which include high densities of lionfish, tend to be concentrated and heavily influenced by patch size, shape (Acosta and Robertson 2002), and connectivity, all of which comprise confounding variables in the analysis of Hackerott et al. In short, any extrapolation of patterns seen on shallow patch reefs to deep forereefs should be made with caution."

The size distribution of groupers on our study sites indicates that grouper >30cm TL (deemed 'large-bodied' by Mumby et al. 2011) were frequently observed in patch reef habitats (Figure 3). Maximum predator biomass was 2-3 times higher in our study than Mumby et al. (2011). However, we cannot test the assertion that the sizes of the large predators at our high-biomass sites was lower than in the ECLSP because Mumby et al. (2011) did not published size distribution data for grouper in the ECLSP.

It is unlikely that high total grouper biomass would result from more abundant but smaller individual fishes. The opposite pattern is well documented in a wide range of habitat types for several fish species (Gust et al. 2001, Friedlander and DeMartini 2002, McClanahan et al. 2007). This seems to also be the case for groupers in our study (Figure 3 bottom panel). At sites with grouper biomass of at least $10gm^{-2}$ (i.e., the minimum observed in ECLSP; Mumby et al., 2013), there were relatively high frequencies of medium/large individuals with high predatory capacity. Across all sites, we found relatively low frequencies of small individuals.

Figure 3 (top panel) also shows that groupers at protected sites were generally larger than those at unprotected sites. This contradicts the statement by Mumby et al (2013) that 'very few reserves have any significant effect on grouper'.

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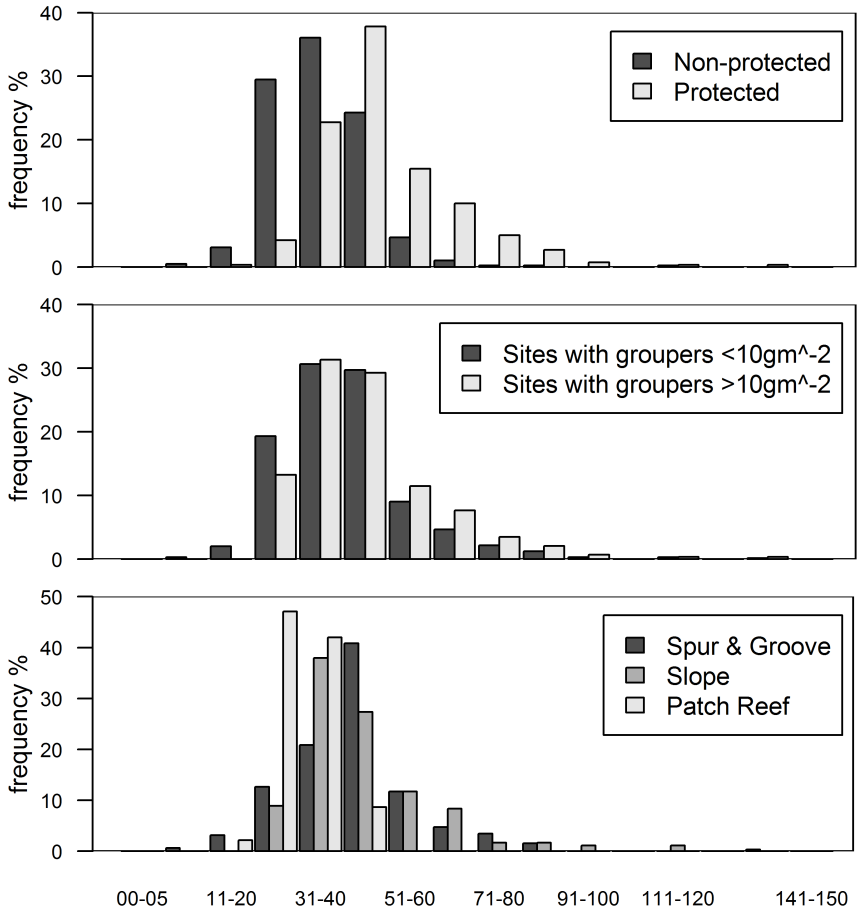

Figure 3. Class size distribution of groupers for protected and non-protected sites (top panel), for sites with over and under 10 gm^{-2} of grouper biomass (middle panel), and for each habitat type (bottom panel). Note that over 40% of protected sites and sites with >10 gm^{-2} of grouper biomass have individuals over 30 cm in total length. Class sizes are total length (cm) on the x-axis. Note that only every other class size has a label

In the one published record of grouper eating lionfish (Maljkovic et al. 2008) the two grouper caught with lionfish in their gut (note it could not be determined whether the lionfish were dead or alive when consumed) were modestly-sized (48 cm SL) Nassau grouper (*Epinephelus striatus*), and not especially large individuals of that species or larger grouper species, e.g., *Mycteroperca bonaci* and *Epinephelus itajara*. Many of the grouper at our sites were substantially larger than 48cm (Figure 3).

3) Do large grouper control lionfish abundance in the Caribbean?

Mumby et al (2013): "Re-examining the results of Hackerott et al., there appears to be a clear breakpoint in predator biomass and lionfish abundance which makes the reader wonder whether their interpretation is correct."

Mumby et al. (2013) re-plotted our results and interpreted variation in lionfish abundance solely as a function of predator biomass, i.e., without any analysis including covariates (see Figure 1 in

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Mumby et al., 2013). Although plotting and considering one's raw data is generally advisable for exploratory analysis, a thorough statistical model is necessary to make a valid interpretation of correlational data. This is particularly true when attempting to make causal inferences from such data. The underlying relationship between lionfish and grouper biomass is driven by multiple co-factors (Figure 2) that were accounted for in the statistical model, i.e., the graphic that truly represents the analysis is multidimensional.

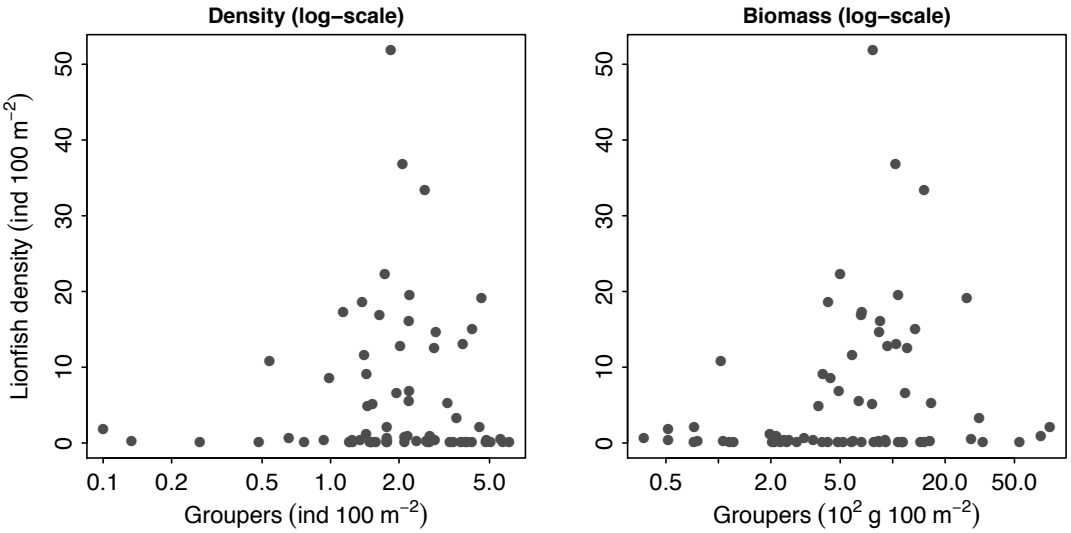

Figure 4. Relationship between lionfish density and log grouper density (left panel) and log grouper biomass (right panel).

Mumby et al. (2013) did not consider the correct null model when interpreting our species co-occurrence data, i.e., the null model is not a horizontal line. In fact, a log-normal distribution is precisely what is expected when plotting an independent factor (large grouper abundance in this case) against an unrelated variable (Figure 4). The lack of any relationship is also obvious in the log-log plot (see Hackerott et al., 2013), which is most relevant given the zero-inflated nature of the data and the statistical models we used. Also note that the untransformed lionfish-abundance axis obscures the large number of zero and very small values and higher sample size for lionfish abundance measurements at intermediate grouper biomass sites (Figure 4): this is likely why some sites with higher lionfish biomass were encountered at intermediate levels of grouper biomass (i.e., simply because far more of these sites were sampled). To play the devil's advocates, we could argue that Mumby et al. (2013)'s visual interpretation approach should lead to the conclusion that lionfish can be controlled by increasing grouper biomass beyond 20 gm^{-2} OR reducing it to < 5 gm^{-2} (Figure 4, right panel). This fallacy illustrates the perils of not considering the data structure, the null model, and the variety of factors influencing the relationship between two variables.

Mumby et al. 2013: "However, lionfish biomass is virtually zero at all higher levels of grouper biomass, such as the levels of biomass observed in the ECLSP (Fig. 1a). Lionfish density is also virtually zero at high levels of apex predator biomass (Fig. 1b). We interpret these results as a constraint upon lionfish under high predator biomass, though alternative interpretations should also be tested."

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Based on our updated statistical model, our results indicate that protection status (i.e., whether sites were located within a marine reserve) had a strong negative effect on lionfish abundance, while predator abundance did not (Figures 1 and 2). This is most likely due to targeted culling in protected areas. Morris and Whitfield (2009) suggested that lionfish removal efforts should be focused on ecologically important areas, including marine protected areas and reserves. Lionfish removals have since occurred in many marine reserves through organized citizen programs (Biggs and Olden 2011, López-Gómez et al. 2013) and by reef managers (e.g., author pers. comm. with Belize Audubon Society). This effort is paying off and has the potential to greatly reduce lionfish abundance, at least temporarily (Barbour et al. 2011, Frazer et al. 2012, Côté et al. 2013). In our dataset, of the six sites with grouper biomass over 20 gm^{-2} (the "clear breakpoint" in the effect of predator biomass on lionfish density proposed by Mumby et al., 2013), five were in protected areas (Figure 5) where lionfish culling is very likely occurring. This pattern supports the results of our statistical analysis that lionfish abundance is reduced in marine protected areas due to some factor other than predator abundance.

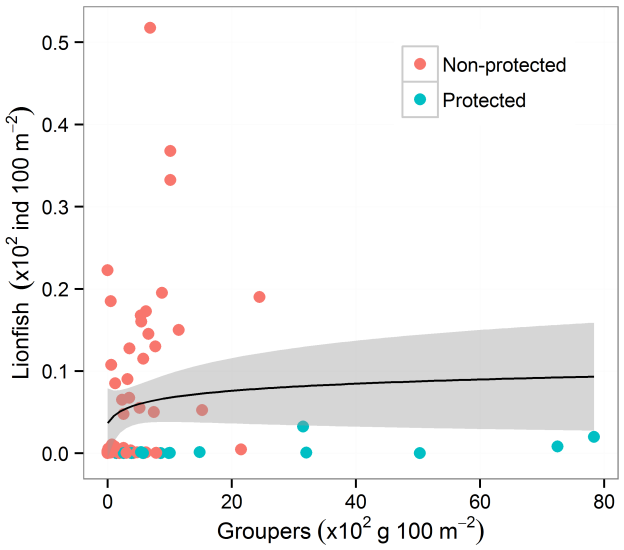

Figure 5. Relationship between large grouper biomass and lionfish density. Red dots are sites outside marine reserves, blue dots are sites inside marine reserves. The black line and shaded grey is the GLMM prediction (and the 95% confidence interval) of the relationship between grouper biomass and lionfish density after eliminating the effects of all the other co-factors. As is evident, large grouper biomass is not significantly related to lionfish density after the effect of all the co-variables are accounted for. In fact, at low values, the relationship between these two groups is positive.

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Conclusion

We addressed the main criticisms of Mumby et al. (2013), explained why they are erroneous, and provided additional analyses, the results of which were consistent with our original conclusions.

I. Mumby et al. (2013) argued that the analyses in Hackerott et al. (2013) omitted important covariates (namely, fishing, larval dispersal, and habitat complexity). Many factors varied among our surveyed reefs, as they would among any other collection of reefs. But that is precisely how we need to test the generality of the pattern observed by Mumby et al. (2011). Managers need to know whether the findings and solutions from local case studies will be effective elsewhere. In our initial paper, we included many parameters that are known to affect reef fish biomass. Here we expanded our original statistical model by including two additional covariates identified as potentially important by Mumby et al. (2013). After accounting for these additional processes, our original results still stand: there is no relationship between lionfish and predator abundance.

II. Mumby et al. (2013) argued that the predators in some habitat types considered in our analysis were too small to consume lionfish or influence their density and biomass. Our sites, including those with comparable total grouper biomass to sites in the ECLSP (~10 gm^{-2} or more), include many medium/large individuals with high predatory capacity.

III. Mumby et al. (2013) argued that our results indicate a "breakpoint" in predator biomass and lionfish abundance and interpreted this as a negative relationship between lionfish and predator abundance. We explained the fallacy in this interpretation. Our updated statistical model, including eight covariates, indicates a negative effect of protection and no effect of grouper biomass on lionfish abundance. Any visual interpretation based solely on a plot of the relationship between grouper and lionfish is misleading because it does not reflect the significant effects of the other cofactors.

Mumby et al. (2013) conclude that removals are the only feasibly mechanism for controlling lionfish. We agree. Based on an objective interpretation of all available evidence, it is clear that: (a) the abundance of lionfish and large grouper (or other large predators) are not negatively related, and (b) lionfish abundance is controlled by a number of physical site characteristics, as well as by human fishing. We suggest that these direct management efforts, such as lionfish removal, are necessary to control the lionfish invasion and should be promoted.

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Appendix 1. Methods and more information on reanalysis with additional covariates.

We ran a generalized linear mixed-effect model with a Poisson distribution and a log-link for lionfish count data. As this distribution is discrete we included an offset in the model to account for survey area, so that we could effectively analyze relationships between the density of lionfish and grouper, i.e.,

Log (LF Density) = Log(LF Counts) - Log (Survey Area)

Because lionfish density and biomass are highly correlated (~0.97), the results should be applicable to biomass as well.

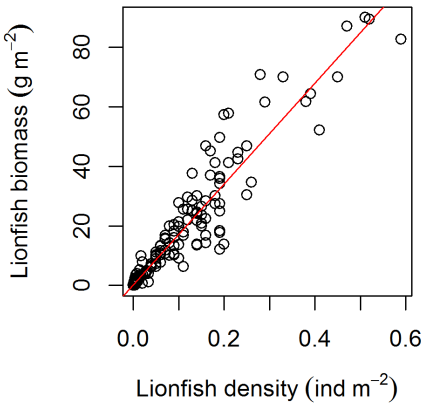

Figure A1. Relationship between lionfish biomass and density. Pearson correlation is ~0.97, p<0.001. See Figure S2 in original paper

We added two new variables to the model described in detailed in Hackerott et al. (2013): Humans/Reef and Reef Complexity (see below for detailed information on these two variables). For a detailed description of the analysis, see the R code below.

Human population: We calculated the number of humans within 50km (maximum number of people living within 50km of each site). We chose 50km as radius because it is a reasonable range of human influence on Caribbean reefs (see Mora 2008). Estimates of human population counts for the year 2010 were obtained from the Gridded Population of the World V.3 at 0.25 degree resolution (SEDAC 2010, see here for database http://sedac.ciesin.columbia.edu/). All calculations were done in ArcGIS v10.0.

Reef area: We calculated reef area within 10km radius of each site (as the distance a grouper might travel in a day). The area was calculated from the Global Distribution of Coral Reefs (2010) database as available at the Ocean Data Viewer (UNEP-WCMC; http://data.unep-wcmc.org/datasets/13). This database represents the global distribution of warm-water coral reefs compiled mostly from the Millennium Coral Reef Mapping Project validated and un-validated maps.

Humans/Reef Area = # Humans within 50km / Reef Area within 10 km/(10^2x3.14) (km^2) = humans/km^2 of reef.

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Reef Complexity: We used a rugosity index (0-5) after (Polunin and Roberts 1993) estimated at the transect level, where 0 is a flat substrate with no vertical relief and 5 is an exceptionally complex substrate with numerous caves and overhangs. Relief complexity for Eleuthera and New Providence was estimated by averaging measurements of reef height (i.e., the vertical distance between the lowest and highest point of the reef structure in cm), taken at five haphazard points within the survey area (either transect or rover diver area) (Wilson et al. 2007). To make reef complexity estimates homogenous for all sites we transformed the relief complexity estimates taken in Eleuthera and New Providence to the rugosity index, described by Polunin & Roberts (1993), by assigning a gradient of 0 cm to 0 and over 300 cm to 5. This resulted in a continuous rugosity index for Eleuthera and New Providence that is comparable with the rest of the sites.

Spatial autocorrelation: Spline correlograms constructed from the residuals of the GLMM model indicated that our mixed-effect modeling framework successfully accommodated spatial autocorrelation observed in the raw data (see similar result in Fig S3 in Hackerott et al.). Additionally, we used Mantel tests (Mantel 1967) to check for overall spatial autocorrelation between the Pearson residuals of the model and distance between sites (i.e., whether sites that are closer together were more similar), and found that the correlation coefficient for the model was low ($r = 0.073$, $p = 0.0001$). We performed the autocorrelation analyses in R version 3.0 using the package *ncf* version 1.1.4 (BjØrnstad 2012).

R Code:

```
#Set working directory
        setwd(.”/Data Analysis")

#Load Data
        fish=read.csv(.”/LFdata.csv")

#Check data
        head(fish)
        attributes(fish)

#Calculate Large Grouper Biomass with same species Mumby used
#include it in the dataframe
        fish <- within(fish, {Grouper.Biom = Black.Biom + Nassau.Biom +  Tiger.Biom + Yellowfin.Biom +
        Yellowmouth.Biom})
        fish <- within(fish, {Grouper.Abund = Black.Abund + Nassau.Abund + Tiger.Abund + Yellowfin.Abund +
        Yellowmouth.Abund})

#Attach fish data to make coding easier
        attach(fish)

#Calculate the log of the survey area to add it as an offset in the model
        LogArea= log(fish$Area4LF)

### Run the GLMM with ADMB
### Load first glmmADMB library

library(glmmADMB) # Load package from R.Forge server instead of R-Cran
```


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```
#Run model with all variables for LF Counts and negative binomial first
#Run a null model with structure first. Use sites as random effects

LFvsGrper.Biom.out0= glmmadmb(LF.Count ~ 1 + (1|Site.Code),
        data = fish,
        zeroInflation=F,
        #admb.opts=admbControl(shess=FALSE,noinit=F),
        family= "nbinom")
        summary(LFvsGrper.Biom.out0)
        AIC(LFvsGrper.Biom.out0)

#Run VIF to assure there is no correlation between numerical factors
#Load package (car)
        library(car)

#Run a glm (logistic model) model with all variables to get the VIF

LFvsGrper.Biom.glm= glm(LF.Count ~ Grouper.Biom + Time + Depth + Protection + Habitat + WindvsLee +
        Rugosity.t. + Hum.Reef + HumPopDen, data=fish)
        summary(LFvsGrper.Biom.glm)

#Run vif to see the variance of each factor and potential correlations problems
        vif(LFvsGrper.Biom.glm)

LFvsGrper.Biom.glm1= glm(LF.Count ~ Grouper.Biom
        + Time + Protection + Habitat + WindvsLee + Rugosity.t. + Hum.Reef + HumPopDen,
        data=fish)
        summary(LFvsGrper.Biom.glm1)

        vif(LFvsGrper.Biom.glm1)

#Rename LF biomass variable to make easier to code later

        LF.Biom=LF.Biom..g100m2.

#Use the same model as in the paper to make things comparable
#Add rugosity and humans/reef areas to the model
#Scale numerical variables to make easy to visualize factors effect on the Coefficient plots

# Run a glmm with ADMB and a negative binomial distribution first as in the original paper

library(glmmADMB)

LFvsGrper.Biom.out= glmmadmb(LF.Count ~ scale(Grouper.Biom)
        + Habitat + Protection + WindvsLee + scale(Depth)+ scale(Time)+ scale(Rugosity.t.)+ scale(Hum.Reef)
        + offset(log(Area4LF)) + (1|Site.Code),
        data = na.omit(fish),
        zeroInflation=T,
        corStruct="diag",
        admb.opts=admbControl(shess=FALSE,noinit=F),
        family= "nbinom")
        summary(LFvsGrper.Biom.out)
        AIC(LFvsGrper.Biom.out)

#No need to compare models using AIC in this analysis as we want to use the same model as in the original paper
```


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```
#Visualize the effects
#Load library
library (coefplot2)
coefplot2(LFvsGrper.Biom.out)

# Run a glmm with Poisson distribution and log-link because to see the differences between those two models
#Include a spatial autocorrelation structure

library (lme4) #install the new version from R Forge to get the prediction function
library (nlme)

LFvsGrper.Biom.glmer= glmer(LF.Count ~ scale(Grouper.Biom)
        + Habitat + Protection + WindvsLee + scale (Depth) + scale(Time)+ scale(Rugosity.t.)+ scale(Hum.Reef)
        + offset(log(Area4LF)) + (1|Site.Code),
        data = na.omit(fish),
        correlation=corAR1(form=~1|Site.Code),
        family= poisson(link="log"))
        summary(LFvsGrper.Biom.glmer)
        AIC(LFvsGrper.Biom.glmer)

#Visualize the data with coefplot

#Graphic the full model

        coefplot2(LFvsGrper.Biom.glmer)

#Calculate prediction for LF abundance vs Grouper biomass when all the cofactors are in.

        fish <- na.omit(fish) #omit NAs

        newdat <- expand.grid(
        Grouper.Biom = scale(fish$Grouper.Biom),
        HabitatSG = 0,
        HabitatSlope = 0,
        Protection = 0,
        WindvsLeeWindward = 0,
        Depth = 0,
        Time = 0,
        Rugosity.t. = 0,
        Hum.Reef = 0)

        mm <- model.matrix(terms(LFvsGrper.Biom.glmer),newdat)

        LF.Abundpred <-  mm %*% fixef (LFvsGrper.Biom.glmer)

#Back transform due to the Poisson and negative binomial distribution

        LF.Abundpredt <- exp(LF.Abundpred)

##Based on fixed effects uncertainity only
        pvar1 <- diag(mm %*% tcrossprod(vcov(LFvsGrper.Biom.glmer),mm))
##Based on fixed effects and random effects uncertainity
        tvar1 <- pvar1+VarCorr(LFvsGrper.Biom.glmer)$Site.Code[1]  ##Create data frame with predictions
        newdat <- data.frame(
        newdat$plo = LF.Abundpredt-2*sqrt(pvar1)
```


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```
newdat$phi = LF.Abundpredt+2*sqrt(pvar1)
newdat$tlo = LF.Abundpredt-2*sqrt(tvar1)
newdat$thi = LF.Abundpredt+2*sqrt(tvar1)

#Caculate means for LF biomass and Grouper biomass
#Add everything as dataframe

#First recode protection variable

fish2$Protection <- as.numeric(recode(fish2$Protection, "'n'='0';'y'='1'"))

newdata=as.data.frame(cbind(
Grouper.Biom.mean=tapply(fish$Grouper.Biom, fish$Site.Code, mean),
LF.Abund.mean=tapply(fish$LF.Abund,fish$Site.Code, mean),
LF.Biom.mean=tapply(fish$LF.Biom,fish$Site.Code, mean),
plo.mean=tapply(newdat$plo,fish$Site.Code,mean),
phi.mean=tapply(newdat$phi,fish$Site.Code,mean),
tlo.mean=tapply(newdat$tlo,fish$Site.Code,mean),
thi.mean=tapply(newdat$thi,fish$Site.Code,mean),
Protection.mean=tapply(fish$Protection,fish$Site.Code,mean)))
newdata=na.omit(newdata)

#### Build figure LF Abundance  ~ large Groupers with prediction#####

# save it as PDF format
pdf(."\FigRebglmm.pdf", height=4, width = 4)

# For TIFF figure use the following code
tiff("C:\FigRebglmm.tiff", width = 4, height = 4, units ="in", res =600, compression="lzw")

#Load ggplot Library to make pretty graphics
library (ggplot2)

#Build ggplot of Grouper and LF abundance
p1 <- ggplot(newdata, aes (Grouper.Biom.mean,LF.Abund.mean, colour=factor(Protection.mean))) +
xlab(expression('Groupers'~(x10^{2}~g~100~m^{-2}))) +
ylab(expression('Lionfish'~~(x10^{2}~ind~100~m^{-2}))) +
geom_point(pch=16, cex=2.8)+ theme_bw()+ theme(legend.position=c(0.75,0.85),
legend.title=element_blank())+
scale_colour_discrete(name = "",breaks=c("1", "2"),labels=c("Non-protected",
"Protected"))+
#legend.text = element_text("Non-Protected", "Protected"))+
theme(panel.grid.minor=element_blank(),
panel.grid.major=element_line(colour="grey99"),
axis.title.x=element_text(size=11),axis.title.y=element_text(size=11))+
#Add prediction based on CI on FE uncertainty and RE variance
geom_smooth(aes(ymin=plo.mean, ymax=phi.mean),method="glm", formula= y
~log(x+1), col="black")
#Plot Coeficient estimates from GLMM
par(mfrow=c(1,1), mai=c(0.7,0.4,0.1,0.1),tcl=-0.3, cex.axis=0.8, mgp=c(2,0.5,0.5))
p2 <-  coefplot2(LFvsGrper.Biom.glmer, h.axis=T,
varnames=c("Grouper biomass","Spur & Groove","Slope","Protected",
"Windward","Depth","Time since invasion","Reef
complexity","Humans/Reef"), col.pts="black",
top.axis=F, main="", cex.pts=0.8, lwd.1=1, xlim=c(-6.5,2),cex.axis=0.7,
cex.var=0.8, xlab="Estimate coefficient", cex.lab=0.8)
```


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```
                    rect(-6.8,0.6,2,9.4)

# close PDF and TIFF devices
dev.off()

#### Results of the GLMM (Poisson) #####
### DO NOT RUN. THIS ARE ONLY THE RESULTS

Generalized linear mixed model fit by the Laplace approximation
Formula: LF.Count ~ scale(Grouper.Biom) + Habitat + Protection + WindvsLee +   scale(Depth) + scale(Time) +
scale(Rugosity.t.) + scale(Hum.Reef) +      offset(log(Area4LF)) + (1 | Site.Code)
  Data: na.omit(fish)
  AIC   BIC logLik deviance
 629.1 671.7 -303.6    607.1
Random effects:
 Groups    Name        Variance Std.Dev.
 Site.Code (Intercept) 0.88519  0.94084
Number of obs: 355, groups: Site.Code, 68

Fixed effects:
                  Estimate Std. Error z value Pr(>|z|)
(Intercept)        -0.329881   0.665530  -0.496   0.6201
scale(Grouper.Biom)  0.070465   0.046957   1.501   0.1335
HabitatS&G          -5.063329   0.854005  -5.929 3.05e-09
HabitatSlope        -4.705014   0.652753  -7.208 5.68e-13
Protection          -0.986715   0.429519  -2.297   0.0216
WindvsLeeWindward   -0.966399   0.423198  -2.284   0.0224
scale(Depth)         0.513643   0.301654   1.703   0.0886
scale(Time)         -0.005651   0.173611  -0.033   0.9740
scale(Rugosity.t.)  -0.057312   0.065409  -0.876   0.3809
scale(Hum.Reef)      0.142201   0.124178   1.145   0.2522

(Intercept)
scale(Grouper.Biom)
HabitatS&G          ***
HabitatSlope        ***
Protection          *
WindvsLeeWindward   *
scale(Depth)        .
scale(Time)
scale(Rugosity.t.)
scale(Hum.Reef)
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1

Correlation of Fixed Effects:
            (Intr) s(G.B) HbtS&G HbttSl Prtctn WndvLW scl(D)
scl(Grpr.B)  0.050
HabitatS&G  -0.491  0.033
HabitatSlop -0.376  0.006  0.808
Protection  -0.665 -0.099 -0.263 -0.314
WndvsLWndwr -0.141  0.021 -0.493 -0.362  0.401
scale(Dpth)  0.719 -0.030 -0.845 -0.685 -0.056  0.138
scale(Time) -0.086 -0.012 -0.376 -0.394  0.402  0.355  0.156
scl(Rgst..)  0.141 -0.029 -0.071 -0.123 -0.096  0.053  0.024
scal(Hm.Rf) -0.226  0.016  0.078  0.128  0.127  0.166 -0.193
```


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scl(T) s(R..)
scl(Grpr.B)
HabitatS&G
HabitatSlop
Protection
WndvsLWndwr
scale(Dpth)
scale(Time)
scl(Rgst..) 0.088
scal(Hm.Rf) -0.171 -0.016
> AIC(LFvsGrper.Biom.glmer)
[1] 629.1131

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