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Microplastics in the Mottled Rabbitfish (*Siganus fuscescens*) in Negros Oriental, Philippines with Notes on the Siganid Fishery

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We reviewed the status of the Mottled Rabbitfish (*Siganus fuscescens* Houttuyn, 1782) as a major fishery product in Negros Oriental, including threats from microplastic pollution and overfishing. This species is often marketed as either fresh or dried “danggit”. Out of a total of 300 fish samples from four areas in Negros Oriental province, 91 (30%) of *S. fuscescens* ingested microplastics; the highest ingestion (39%) was observed in Dumaguete, a densely populated city. We also assessed the reproductive biology parameters of this species and compared them with the data gathered in 1979, roughly 40 years ago. The samples from Bais and Dumaguete had reduced sizes at sexual maturity and fecundity, suggesting negative effects from prolonged overexploitation. We therefore urge more studies on other parts of Negros Island and even elsewhere in the country, to determine the potential health hazards from microplastic pollution and the current threat to the sustainability of the siganid or “danggit” fishery.

Keywords: fecundity, fishery, microplastics, overfishing

INTRODUCTION

Rabbitfishes (Family *Siganidae*) are a popular and heavily consumed coastal food fish in the Philippines. Based on the 2017 fisheries profile of the Philippines, the country produced 194.31 tonnes of siganids per year, excluding unreported catch by local traditional gears. Fulton et al. (2020) pointed out an increasing trend in landings of siganids in the Philippines and Indonesia. This may be attributed to an increasing number of algal farms which have been reported as food sources of this fish species in these two countries.

In many parts of the Philippines, the most conspicuous species landed and often sold as fresh or as dried boneless “danggit” is *Siganus fuscescens*. This species is fast-growing, becoming sexually mature at around two years (Grandcourt et al., 2007). The fries and adults of this species often aggregate in schools of up to several thousands. As the fish matures, the number of individuals per school is reduced to about 60 individuals (Woodland, 1990). Diet of juveniles consists of filamentous algae, while adults feed on selected fleshy macroalgae (Woodland, 1990; Froese & Pauly, 2019). The species’ availability in coastal areas throughout the year, its relatively small home adult range, and its benthic feeding habits make it a good marine organism as an indicator of coastal pollution.

The *Siganus fuscescens* species complex consists of two similar species (*S. fuscescens* and *S. canaliculatus*) that differ subtly in spotting pattern (Woodland, 1990). Hsu et al. (2011) pointed out those specimens that were distinguishable morphologically as either *S. fuscescens* or *S. canaliculatus* were inconsistent in terms of their mitochondrial DNA and that both species should be regarded as synonyms. In this regard, following Ravago-Gotanco et al. (2018), we use the scientific name *S. fuscescens* (Houttuyn, 1782) to include *S. canaliculatus* (Park, 1797) which has been used by earlier studies (e.g., Alcala & Alcazar, 1979; Paraboles & Campos, 2018).

Globally, there is an emerging problem in marine plastic pollution (Walker, 2018). The Philippines ranks third among countries in terms of plastic pollution (Jambeck et al., 2015). Abreo et al. (2016) documented that large marine vertebrates such as turtles and whales ingested plastic debris. Many studies have shown that microplastics (i.e., plastic particles <5mm) are found in the aquatic environment (Deocaris et al., 2019; Pan et al., 2019). In the

Philippines, studies have shown that microplastics are present in beaches and benthic sediments (e.g., Kalnasa et al., 2019; Paler et al., 2019) and in edible mollusks (Argamino & Janairo, 2016). Espiritu et al. (2019) demonstrated the presence of microplastics in commercial fishes in Luzon. Bucol et al. (2020) documented the presence of microplastic particles in both marine sediments and the commercially important fish, *S. fuscescens*, along the coast of Negros Oriental in Central Visayas.

In Negros Oriental province, Bais Bay has been a major fishing ground for siganids since the 1970s (see Alcala, 1979). At present, there are at least 246 fishermen who use gears that frequently catch this species. Alcala and Alcazar (1979) studied aspects of the reproductive biology of the rabbitfish *Siganus canaliculatus*, now known as *Siganus fuscescens*, in Negros Oriental. Later, Silliman University researchers assessed the fishery profile of Bais Bay, including a stock assessment of *S. fuscescens* (Luchavez & Abrenica 1997a, 1997b). The present study includes an update on the fishery of *S. fuscescens*, using these two earlier studies as baseline.

Given the importance of this species to fisheries and coastal marine ecosystems, our objective was to document the potential impact of two of the most pressing threats (e.g., fisheries and plastic pollution) on regional populations, by documenting observed changes in sexual maturity and fecundity that were likely linked to overexploitation, as well as the presence and type of ingested microplastics that were likely linked to locally-sourced plastic pollution. As this species is of critical importance to the local human communities that rely on them for food, results aim to provide a deeper understanding of the potential impacts of overexploitation and plastic pollution on locally-caught marine fishes and human health, as well as to recommend additional studies and guidance for improved fisheries management and plastic pollution reduction policies in Negros Oriental.

MATERIALS AND METHODS

Sample Collection

Samples of *S. fuscescens* were directly purchased from either local fishers from landings or fish markets that only sold locally caught fish (< 5 km radius) in the following localities (arranged from North to South): 1) Tiguib,

Ayungon; 2) Campuyo, Manjuyod; 3) Olympia, Bais City; 4) Silliman Beach, Dumaguete City; and 5) Si-it Bay, Lutoban, Zamboanguita, all in Negros Oriental (Figure 1).

For the microplastic component of this study, we examined 300 fish samples (90 each from Ayungon, Bais, and Dumaguete, while only 30 in Manjuyod) between October 2018 and June 2019. Samples from Zamboanguita, however, were not included in the microplastic sub-study. At the laboratory, each fish was immediately processed measured in terms of standard length (cm) and total weight (g). Viscera were then excised by cutting a longitudinal slit at midbody and at right angle around the posterior edge of the body cavity.

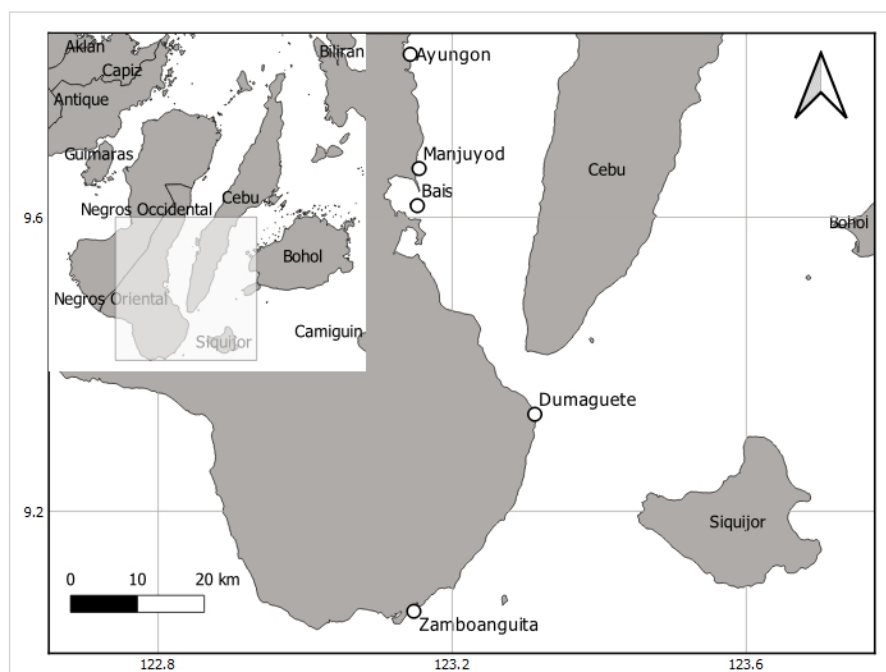


Figure 1. Map showing the location of the sampling sites for *S. fuscescens*.

Microplastics. Guts and gills were separated (excess fats and other organs removed) and soaked in 10% potassium hydroxide (KOH) for 3-5 days to undergo organic digestion (Lusher, 2017). Subsequently, digesta were filtered (Whatman, 2.5 microns pore size) and examined under dissecting microscopes (Zeiss®, 40x magnification) with built-in cameras at Silliman University-Angelo King Center for Research and Environmental Management

(SUAKCREM). Identification of plastic polymer type was conducted using fourier-transform infrared (FTIR) spectroscopy (Perkin Elmer, Spectrum 2) at NORSU Chemistry Laboratory. Confirmation of polymer type was determined using a library of polymers/compounds (built-in software in Perkin Elmer FTIR, Spectrum 2). Positive identification was considered only when correlation reached 0.6 and above.

Procedural blanks (controls) were frequently performed to determine the presence of contaminants. Microplastic particles that closely resembled the contaminants were excluded from the analysis.

Key Informant Interviews

To determine the scope and characteristics of the danggit fishery, key informant interviews with one key person per Barangay were conducted by a local student hired as an interviewer from 22-28, January 2020, from Barangay Campuyo (Manjuyod municipality) in the North to Barangay Luca (Tanjay City) in the South.

Reproductive Biology

Determination of sex and size at sexual maturity. The weight (to the nearest 0.1 g) and standard length (to the nearest 1.0 mm) of each fish sample was measured and dissected to determine the sex and gonad developmental stage. Each gonad was classified following the macroscopic gonad staging scale adapted from Kamakuru and Mgaya (2004). Using this scheme, gonads of immature fish were classified as Stages I and IIa, while those of sexually mature fish were classified as follows: Stage IIb – Resting; Stage III – Ripening; Stage IV – Ripe; and Stage V – Spent.

To determine size at maturity (Lm_{50}), the proportion (%) of mature and males was plotted and predicted using the logistic curve fit based on the formula:

$$Lm_{50} = a / (1 + e^{-b+cx})$$

where $a=100\%$, 1 and $e=2.71828$ were constants, b and c were the parameters to be estimated using the least-squares regression using the *car* package in R (R Core Team, 2015).

The size at sexual maturity is defined as the size class at which 50% of fish samples are classified as sexually mature (Sadovy, 1996). After determining the best logistic fit, Lm_{50} was computed using the formula $-b/c$.

Mean lengths (in SL mm) of mature male and female individuals from each of the four sampling sites were compared and tested for statistical significance using the Kruskal-Wallis test with Dunn's post hoc test. This was implemented using the *rstatix*, *ggpubr*, and *tidyverse* packages in R.

Estimation of batch fecundity. Ovaries that were classified into Stage III to V were fixed in 10% formalin. Batch fecundity (i.e., the number of eggs per spawn) was estimated using these preserved ovaries by applying the gravimetric method described by Hunter et al. (1985). In this method, subsamples of known weights (~0.01g) were obtained from each ovary. For consistency, subsamples were obtained from the left ovaries. To loosen the connective tissues, 2-3 drops of glycerine were added and after about 10 minutes, the sub-sample was macerated gently with a blunt end of a dissecting needle. The hydrated oocytes (i.e., eggs that are about to be released within 24 hours) were subsequently counted, and the total number of hydrated oocytes from each sampled ovary was extrapolated from average counts of hydrated eggs from the subsamples.

RESULTS

Microplastics

Of the 300 individuals that were examined, 91 fish samples (30%) had microplastics (both in guts and gills) (Figure 2). Among sites, Dumaguete ranked the highest (39%, $n = 90$) in terms of microplastic occurrence, followed by Manjuyod (37%, $n = 30$). However, in terms of the amount of particles per fish, the highest recorded density (1.97 particles/fish) was in Manjuyod (Fig. 3). One fish from Manjuyod had 44 fragments of microplastic, which was identified as polypropylene.

The proportion and types of microplastic ingested by *S. fuscescens* (Fig. 4) were different from those found in the sediment. This may suggest that the types of microplastics that were present in the sand have different densities compared to those attached to the seagrasses and algae and those consumed by the rabbitfish. The photomicrographs of microplastic particles are shown in Figure 5.

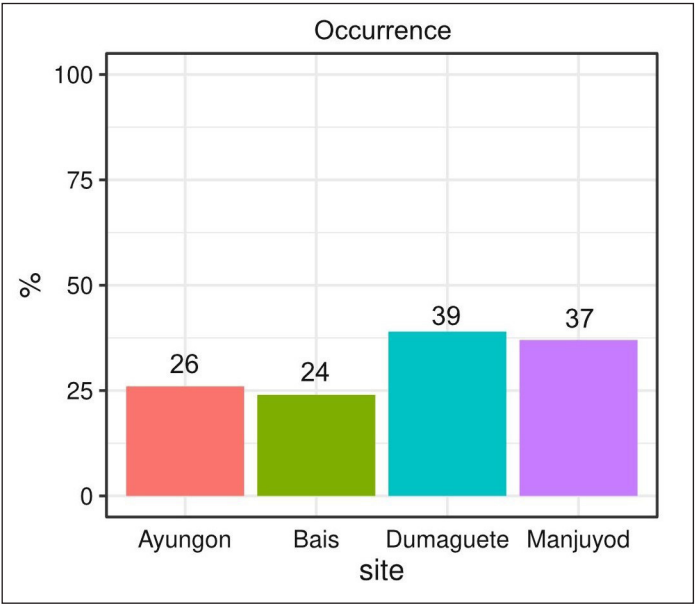


Figure 2. Occurrence of microplastics in *S. Fuscescens* across four sampling sites (n=90 in all sites, except Manjuyod, n=30)

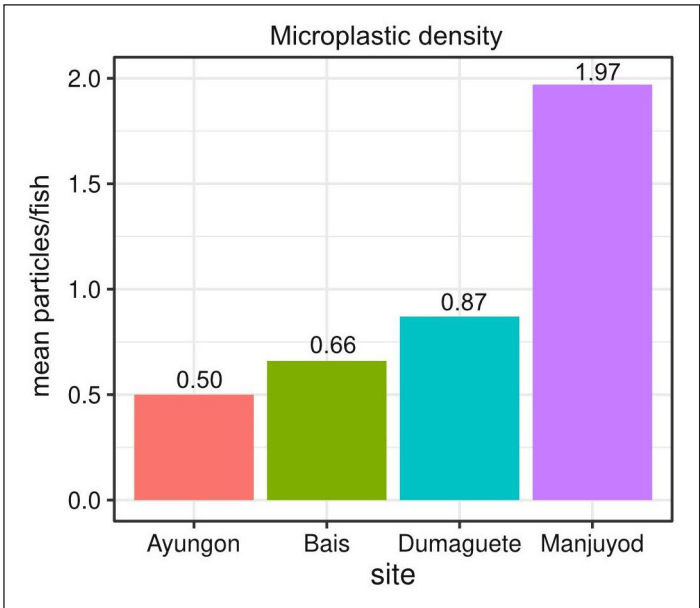


Figure 3. Average number of microplastic particles per fish across sampling sites (n=90 in all sites, except Manjuyod, n=30)

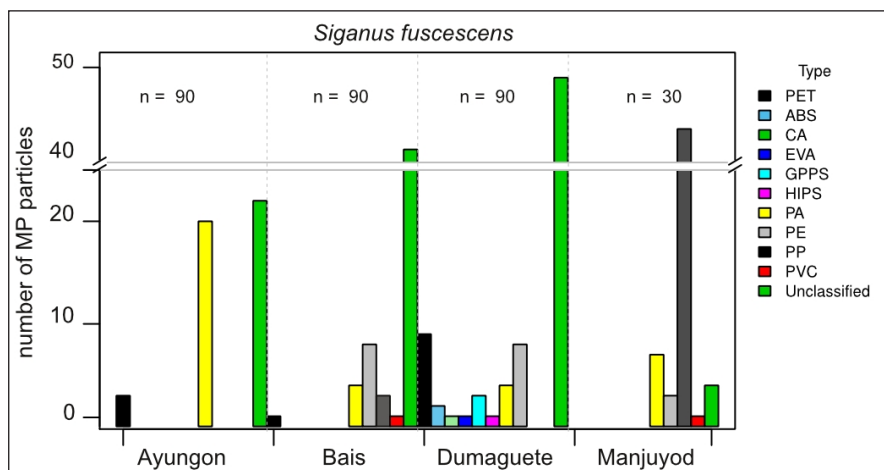


Figure 4. Classification of microplastic types from *S. fuscescens* (polyethylene terephthalate (PET), acrylonitrile butadiene styrene (ABS), cellulose acetate (CA), ethylene-vinyl acetate (EVA), general purpose polystyrene (GPPS), high impact polystyrene (HIPS), polyamide (PA), polyethylene (PE), polypropylene (PP), polyvinyl chloride (PVC) (Unclassified are those with FTIR correlation < 0.6.)

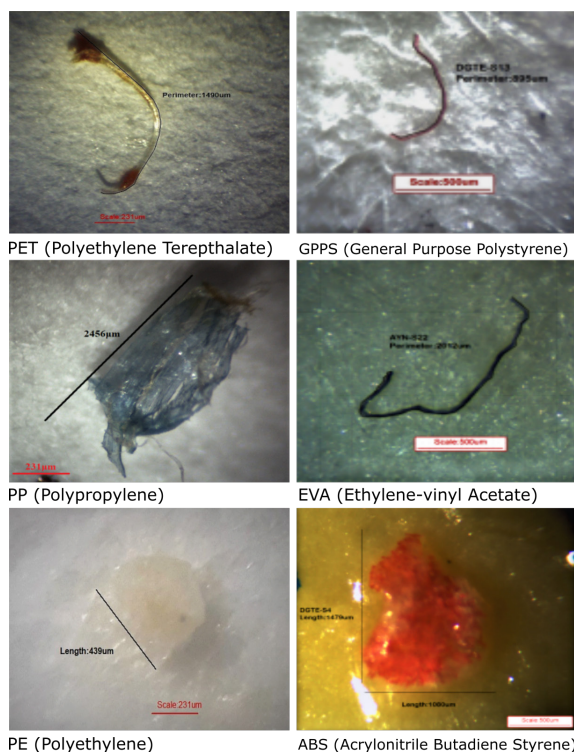


Figure 5. Examples of microplastic particles from the Rabbitfish *S. fuscescens*

Notes on “Danggit” Fishery

Results from the key informant survey revealed that out of about 853 fishermen who fished in both North and South Bais Bays, 179 primarily targeted *S. fuscescens*.

There were at least 246 fishing gears (number of units) deployed daily, with an estimated combined catch of 1,574 kg per day. Given that fishermen catch fish year-round, an annual harvest of around 500 metric tonnes is highly probable. This may still be an underestimation, given that fish corrals may sometimes catch up to 100 kg (normally 3-40 kg/day) during the new moon of each month. Most fishing gears that target this species were deployed in seagrass beds, while only fish pots (panggal) were deployed in coral reefs.

Based on the interview with one main dried-fish supplier from Bais City, preliminary data were collated on the volume of dried “danggit” (*S. fuscescens*) from Bais Bay. In two months (early 2020) orders from him amounted to 120-130 kg of dried “danggit” (about 7-9 kg of fresh “danggit” make up 1.0 kilogram of dried “danggit”) and between June and November 2019, he sold 300-400 kg of dried “danggit”. This means one supplier alone would need to buy 480-520 kg every month of fresh “danggit”. The months of December to May were considered lean season (low volume) for “danggit” but we do not have specific data for this. Other dealers (6-7 persons) also sold a combined volume of about 300 kg of dried “danggit”. This would mean an additional 400 kg of fresh “danggit” per month. Overall, the “danggit” fishery in Bais Bay may be estimated to have produced roughly about 880-920 kg/month of fresh “danggit” to supply the demand for dried fish alone. Considering that the peak season ran for six months in a year, there is an estimated annual harvest of 5.28-5.52 tonnes/year of “danggit”. Given that the fresh market price of “danggit” was Php 120/kg (range from Php 90-150/kg depending on size and location), the harvest is estimated to have a value of Php 633,600 to 662,400 as total annual income. This figure would still be an underestimation, given that some undetermined portions of the total catch was sold directly as fresh fish at the fish market.

Other islands/locations in the Visayas that produce dried “danggit” include Bohol, Cebu (mainly Bantayan island), and Negros Occidental. We have thus far no fishery data from these areas.

Size at Sexual Maturity

Table 1 shows a general trend in the attainment of male and female maturity of *S. fuscescens*. Males matured at 96.28 mm, which was slightly lower compared to the range reported in 1979 (100 - 105 mm SL) by Alcala and Alcazar (1979). Females matured at 115.48 mm SL, while Alcala and Alcazar (1979) reported 111 - 115 mm SL range for mature females. However, when specific locations were compared to the 1979 data, female samples from Bais matured at 100 mm SL, while those from Dumaguete matured at 96.86 mm SL. Those from Zamboanguita matured at 139.5 mm SL, while those from Ayungon matured at 116.35 mm SL. Note that the latter two locations had lower fishing pressure compared to Dumaguete and Bais. Males from Dumaguete matured at 78.13 mm SL, while those from Ayungon matured at 109.62 mm SL. Sizes of mature male and female *S. fuscescens* between sites showed significant difference based on the Kruskal-Wallis test ($p < 0.001$).

Table 1

*Size at Sexual Maturity (L_{m50}) and Comparisons of Sizes of Mature Males and Females of the Spotted Rabbitfish *Siganus fuscescens* from Four Sampling Locations along Negros Oriental, Central Philippines SL=Standard Length (mm)*

Parameters	Site				Total
	Ayungon	Bais	Dumaguete	Zamboanguita	
N (total)	232	349	162	75	818
N (male)	83	150	95	17	345
L_{m50}	109.62	88.62	78.13	89.87	96.28
mean size mature males (SL)	102.28	91.96	126.14	154.46	111.99
Kruskal-Wallis test	p-value < 0.001				
post-hoc (Dunn's test)	Dumaguete vs Zamboanguita not significant (p-value>0.05)				
N (female)	149	199	67	58	473
L_{m50}	116.35	100.00	96.86	139.50	115.48
mean size mature females (SL)	134.46	132.25	152.83	154.30	140.00
Kruskal-Wallis test	p-value < 0.001				
post-hoc (Dunn's test)	Ayungon vs Bais; Dumaguete vs Zamboanguita not significant (p-value>0.05)				

Fecundity

In this study, fecundity was generally lower compared to that in 1979 based on the data provided by Alcala and Alcazar (1979). The data in 2019 was based on the 22 mature female gonads examined from two sites (i.e., Dumaguete and Bais) (Figure 6). In 1979, fecundity ranged from 165, 727 to 650, 625 eggs/female (mean = 397, 680 eggs/female). In this study, fecundity ranged only from 11, 540 to 277, 550 eggs/female (mean = 141, 890 eggs/female). This difference is about three-fold decline in female fecundity within 40 years of harvesting this species.

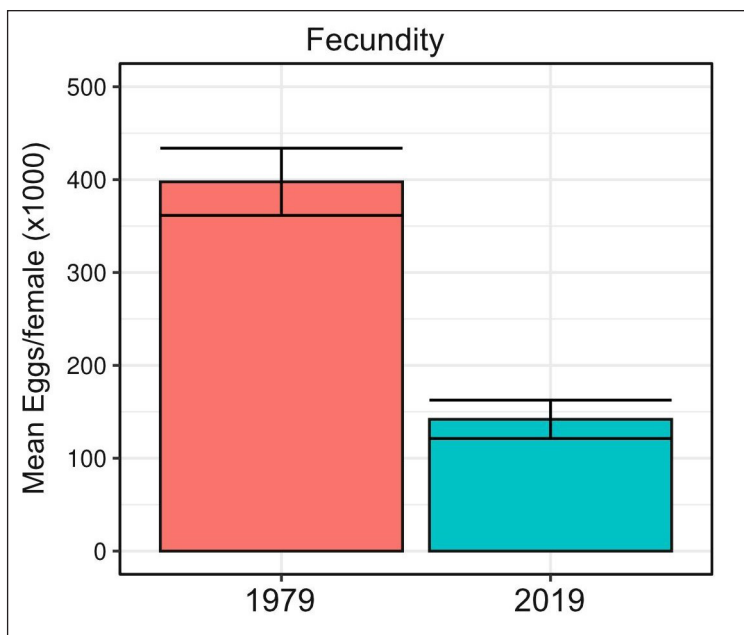


Figure 6. Comparison of the batch fecundity of *S. fuscescens* between 1979 (n=15) and 2019 (n=22) (Error bars indicate standard errors [S.E.].)

DISCUSSION

In this study, we present physiological evidence related to the potential impact of two major threats to the sustainability of the “danggit” fishery in Negros Oriental: 1) plastic pollution; and 2) overharvesting, which includes the harvest of small individuals. Plastic pollution has already reached the food

chain as shown by the presence of microplastics in the guts and gills of the rabbitfish *S. fuscescens*. In addition to the relatively unknown physiological, population, and community-level impacts to rabbitfish of ingestion of microplastics, there is also a possible danger that the microplastics could be ingested by humans through the widespread habit of eating fermented viscera (including guts, locally known as “dayok”) of this fish in the Visayas. Occurrence of fish with microplastics inside their guts and gills was highest in Dumaguete City. This city had the highest population (131,400 in 2015) in the province of Negros Oriental. Browne et al. (2011) showed a direct relationship between microplastic density and human population size.

Our recent estimate on microplastic ingestion by a siganid was lower (overall, ~30%) compared to cases in other neighboring countries like Indonesia which reported that 100% of sampled siganids had microplastics although we reported 46.7% occurrence in our previous paper (Bucol et al., 2020). In terms of microplastic density (i.e., particles/fish), our data ranged from 0.5 to 1.97 microplastic particles/fish, which is comparably lower than the reported 10-28 particles/fish in Luzon by Espiritu et al. (2019). A study in Indonesia reported a microplastic density ranging from 4-52 particles/fish for *S. canaliculatus* (Hastuti et al., 2019). A recent study by Palermo et al. (2020) on microplastic ingestion in a sardine species (*Sardinella lemuru*) from northern Mindanao reported a slightly higher microplastic density of 3.7 particles/fish and a high percent occurrence (85%). Microplastics have been found present in dried fish (Karami et al., 2017), canned sardines, and sprats (Karami et al., 2018).

This study highlights microplastic pollution in the Tañon Strait, a major fishing ground and conservation area in the Philippines. However, it is also noteworthy that another group of students from Arizona State University and Old Dominion University in 2018 also found that about 47% of the fish samples from both Bais and Dumaguete had microplastics inside their guts (Shire & Clark, 2018 unpublished report). This report also showed high concentrations of heavy metals (e.g., lead, arsenic, and aluminum) in their livers.

Evidence of overharvesting of this species could be gleaned from the relative reduction in size at sexual maturity and fecundity, as compared to the baseline data in the 1970s. This supports the findings of Paraboles and Campos (2018) on *S. fuscescens* in Palompon, Leyte, Eastern Philippines.

Other studies have documented overfishing of siganids in Lagonoy Gulf (Soliman et al., 2009; Soliman & Yamaoka, 2010). Our fecundity estimates were also lower than those reported by Jumawan-Nanual and Metillo (2008) for the same species in Pujada Bay, Southern Mindanao. This study noted a three-fold decline in female fecundity, suggesting a potential impact from overfishing.

More studies are needed to determine human-induced impacts on the siganid fishery in the Philippines. Fulton et al. (2020) noticed an increasing trend in siganid landings in both the Philippines and Indonesia, which was probably due to the presence of algal farms that provided food for the fish. This might also hold true in the case of Bais Bay. Anecdotal evidence based on an interview with a local fish vendor from Bais revealed that due to persistent harvesting of siganids in Bais Bay, the fishery almost collapsed some years ago if not for the algal farms that helped sustain the population of *S. fuscescens*.

While most of the highly destructive fishing gears (e.g., *muro-ami*) used in the 1990s (Luchavez & Abrenica, 1997a,b) have been eradicated in Negros Oriental and in most parts of the Visayas, certain gears such as beach seine and fine mesh gill-net are still in use, especially in Bais Bay. A visit to the Bais Fish Market, for example, revealed that juveniles (<7 cm SL) were being sold. It is also noteworthy that a few days after each new moon (probably coinciding with the known spawning period of this species), catch of fish corrals (sometimes reaching to about 100kg per unit) peaked (P. Tolelis, pers. obs.). McManus et al. (1992) documented that fish corrals targeted migrating *S. fuscescens* towards their spawning ground in Bolinao, Pangasinan.

In summary, Tañon Strait is a major fishing ground in Central Visayas, and seafood resources from this body of water, like in many parts of the world, may be at risk of both overexploitation and microplastic pollution. It has been well-established that the chemicals (ingredients) that comprise microplastic particles, in addition to other pollutants that are adsorbed by microplastic particles such as pesticides and heavy metals, can partition from the guts of fish into fish muscle (Zeytin et al., 2020; Zitouni et al., 2020). Lucas and Polidoro (2019) documented varying levels of contaminants such as the dibutyl phthalate (i.e., a suspected teratogen and endocrine disruptor) from the muscle tissues of recreationally-caught fish in Phoenix, Arizona. Moreover,

Deng et al. (2020) showed that microplastics could transport and release phthalate esters into the mice guts and cause aggravated toxic effects. Aside from conducting more research studies, extension activities and information dissemination should be done to target local stakeholders, such as fishermen and vendors, to inform them of the potential dangers of microplastic pollution and the of overfishing of “danggit” fishery resources to humans.

An ongoing study at SUAKCREM funded by the National Academy of Science and Technology Philippines (NAST), through National Scientist Dr. Angel C. Alcala of Silliman University, in collaboration with Negros Oriental State University and Dr. Beth Polidoro of Arizona State University in the USA, aims to quantify the levels of microplastics and potentially adsorbed pollutants in mangrove sediments, bivalves, and marketed seafood products (e.g., dried fish, salted anchovies “bagoong”, and bottled mussels). We are also investigating further as to the potential impact of “bunsod” or fish corral aimed at aggregating siganids especially during the spawning period (i.e., new moon). To address the potential negative impact of fish corrals and other gears (e.g., gill-net) on the stocks of *S. fuscescens*, we recommend setting up more no-take marine reserves (NTMRs) in sites where this species aggregate during spawning. Thus far, there is at least a 50-ha marine reserve in Bais Bay, covering three major ecosystems (i.e., mangrove, seagrass, and coral reef). It is hoped that this no-take marine reserve in South Bais City can protect part of the siganid population, which requires regular monitoring of this no-take marine reserve. Other interventions such as regulating the use of small-mesh nets (e.g., beach seines and gill-nets), that allow small and immature fish to escape capture, should also be implemented to ensure sustainability of the fishery resources.

We call on the offices and the government agencies (e.g., local government units, Department of Environment and Natural Resources, and the Bureau of Fisheries) to take note and validate our findings, and implement proper safeguards to keep the siganid fishery of the country sustainable for current and future generations of our people.

ACKNOWLEDGMENTS

We wish to thank the National Academy of Science & Technology Philippines (NAST) for funding the present study. We are also grateful to Pete John Tolelis

and Leizel Onte, both students from Negros Oriental State University, for their assistance during field interviews and fecundity analysis, respectively. Lyca Mae Siplon and Gianni Madrid served as research assistants for this research project. Additional specimens were purchased and examined through the Partnerships for International Research and Education Project (U.S. National Science Foundation award 1743711). Dr. Joel P. Limson, President of Negros Oriental State University (NORSU), is also thanked for allowing us to use the FTIR machine in the Chemistry Department of NORSU. Two units of Zeiss microscopes used by this study were kindly donated by Mr. Vic Mercado of MicroLab to A.C. Alcala.

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