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A Baseline Study on the Quality and Safety of Consumption of a Pest Species (*Sarotherodon melanotheron*) in Bataan, Philippines: Basis for Its Productive Utilization in the Fisheries Sector

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ARTICLE INFO

Article history

Received: 28 February 2022

Accepted: 21 March 2022

Published Online: 01 April 2022

Keywords:

Tilapia

Black-chin Tilapia

Sarotherodon melanotheron

Proximate composition

Heavy metal load

Microbiological quality

Orani

Bataan

ABSTRACT

The baseline study profiled Black-chin Tilapia (*Sarotherodon melanotheron*), a fish farm pest species in Bataan, Philippines, in terms of yield (processing and fillet), proximate composition (moisture, ash, crude fat, and crude protein), heavy metal load (cadmium [Cd], lead [Pb], arsenic [As], and mercury [Hg]), and microbial count (aerobic plate, *Escherichia coli*, and *Staphylococcus aureus* counts). The purpose was to establish the species' safety and quality for consumption and potential utilization in the processing of higher value fishery products. A completely randomized experiment using two factors, fish size (standard and small sizes) and collection season (dry and wet seasons), was employed. The collected data were also compared against food consumption and processing standards and/or previous reports on more valuable species. The results showed that the species has a comparable yield and mineral load with the more popular farmed Nile Tilapia (*Oreochromis niloticus*). It has high moisture and protein compositions. It is a lean fish that can serve as a cheaper functional raw material for processed fishery products. Moreover, the results showed that the species have no As, Cd, and Pb contamination, although traces of Hg, far below the permissible limits, were detected. The Hg load varies across collection season and fish maturity suggesting its manageability. For the microbial contents, the species' aerobic plate, *Escherichia coli*, and *Staphylococcus aureus* counts were far below the standard limits, although best post-capture practices are still suggested due to the kind of microbial parameters measured. It was concluded that the *Sarotherodon melanotheron* infesting Bataan farm ponds can be consumed safely and has the quality of potential raw material for processed fishery products. However, further information is still needed to establish the best post-capture handling on the species. Also, more studies must be done to determine the impact of storage and processing on its stability.

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DOI: <https://doi.org/10.30564/jfs.v4i1.4483>

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1. Introduction

Black-chin Tilapia (*Sarotherodon melanotheron*) is a species native to tropical East Africa, originally occurring from Senegal to Zaire and southern Cameroon. It was introduced to several countries across Asia, North America, and Europe decades in the past [1]. In the Philippines, their introduction was believed to be through the country's illegal aquarium trade [2]. Naturally, *Sarotherodon melanotheron* can achieve sexual maturity at a very small body size, making it highly adaptive. Being a salt-tolerant and hardy fish, it can spread very rapidly in both saltwater and brackish water environments. These traits make for a very opportunistic invasive species, and its incursion to fish farm areas can be very damaging.

Sarotherodon melanotheron was reported to be infesting Philippine brackish water farm ponds [3]. Particularly in the province of Bataan, the "pest" has been causing losses to commercial fish farms. It was able to proliferate in the farm ponds from Manila Bay through its tributaries. It has been rapidly multiplying and competing with cultured fish species for food [4,5], compromising the carrying capacity of the aquacultural areas meant for higher-value species. The infestation has triggered calls from fish farm operators to curb their population and/or for their productive utilization in the fishery and food sectors [4,6].

While it is locally believed to be edible, literature on the fishery of *Sarotherodon melanotheron* is in paucity. The species was recently investigated for processing [7], growth and feeding behavior [8], breeding and culture [2], and sex-determination mechanisms [9] but the safety of its consumption and its nutritional profile has not been reported extensively, particularly those infesting Philippine fish farms. Recognizing these, the present study worked on profiling *Sarotherodon melanotheron* infesting Bataan fish farms in terms of yield, proximate composition, heavy metal load, and microbial counts. The purposes were to establish the species' safety and quality and to have baseline data for its potential utilization in the processing of higher value fishery products. Ascertaining the aforesaid information for any prospective fish raw material is important to fish processors for them to design and apply appropriate processing and storage methods.

Specifically, the baseline study's objectives were the following: measurement of yield indices of *Sarotherodon melanotheron* infesting Bataan farm ponds, analysis of its proximate composition (moisture, ash, crude fat, and

crude protein), and determining of its heavy metal load (cadmium, lead, arsenic, and mercury) and microbial count (aerobic plate, *Escherichia coli*, and *Staphylococcus aureus* counts). The data obtained were compared across fish size (standard and small sizes) and collection season (dry and wet seasons). All data obtained were also compared against food consumption and processing standards and/or against previous reports for more valuable tilapia species.

2. Materials and Methods

2.1 Materials

Fresh *Sarotherodon melanotheron* fish of standard and small sizes caught from infested farm ponds in Bataan, Philippines.

2.2 Material Sourcing, Post-harvest Processing, and Yield Computation

All fish samples used were sourced from two 2 m - 4 m deep tilapia grow-out ponds located in Orani, Bataan (14.811898, 120.537822). The town of Orani is known to be the province's aquaculture center being a major tiger prawn, milkfish, tilapia, and mud crab producer through brackish water fishery [10]. The pond sites were selected primarily due to their apparent infestation by *Sarotherodon melanotheron* and hence their capacity to supply the needed number of samples for the investigation. The same sourcing and post-harvest processing procedures were performed for acquiring samples for the yield, proximate composition, heavy metal load, and microbial count measurements. Samples were collected in two seasons: wet season (July-December 2019) and dry season (January-June 2020).

Fish samples were caught using net traps and gill nets. The caught fish were kept in insulated plastic coolers with a fish/ice ratio of 1:1 (w/w) to slow post-mortem decomposition and then were transported immediately to the laboratory in BPSU-Orani (20-30 minutes travel time) for sorting and post-harvest processing. The fish were sorted into standard size (4 pcs/kg-5 pcs/kg) and small size (13 pcs/kg-14 pcs/kg) samples and then were weighed per piece (see Figure 1 for reference sizes). Weighed fish were then scaled, de-headed, eviscerated, filleted, and then de-skinned by hand. The separated parts were weighed for each sample. Yield indices were then computed for both sample groups using the formula below. Fifteen replicates for each group were used.



Figure 1. Standard size and small size *Sarotherodon melanotheron* samples

$$\% \text{ Processing Yield} = \frac{\text{Weight of Whole Fish} - \text{Weight of Head, Skin, and Viscera}}{\text{Weight of Whole Fish}} \times 100$$

$$\% \text{ Fillet Yield} = \frac{\text{Weight of De-skinned Fillet}}{\text{Weight of Whole Fish}} \times 100$$

After the yield measurements, the samples were immediately kept in frozen storage (-15°C) until further analysis.

2.3 Proximate Compositional Analysis

Using standard methods, the *Sarotherodon melanotheron* samples were examined for moisture content (AOAC 952.08A), ash content (AOAC 938.08), crude fat content (acid hydrolysis [Soxhlet]), and crude protein content (automated Kjeldahl method [Buchi]). Values were expressed in % w/w. Three replicates for both standard and small size samples were used.

2.4 Heavy Metal Analysis

Heavy metal loads in the fish samples were analyzed using standard methods. The following were tested: cadmium (Cd) in ppm (AOAC 999.10), lead (Pb) in ppm (AOAC 999.10), arsenic (As) in ppb (AOAC 986.15), and mercury (Hg) in ppb (EPA 7473). Three replicates for each group were used.

2.5 Microbial Testing

The fish samples were examined for the following microbial parameters using standard culture techniques: aerobic plate count (FDA BAM-3, 2001, pour plate method), *Staphylococcus aureus* count (FDA BAM-12, 2001, spread plate method), and *Escherichia coli* count (FDA BAM-4, 2002, MPN method). All methods were acquired from the Bacteriological Analytical Manual, Online 2001 of the US Food and Drug Administration

(FDA). Aerobic plate count and *Staphylococcus aureus* count were expressed in CFU/g, while *Escherichia coli* count was expressed in MPN/g. The analyses were done in triplicates.

2.6 Statistical Analysis

The present study utilized a completely randomized experimental design where two factors, collection season and fish size, were investigated on their effect on yield, proximate composition, heavy metal load, and microbial count. Data for the yield, proximate composition, and heavy metal load were presented as mean \pm standard deviation. The microbial load meanwhile was presented in the mean total count. The tests for significant differences were done using One-way Analysis of Variance at $\alpha=0.05$. Post hoc analyses were performed using Fisher's Least Significant Difference Test. Statistical analyses were performed using IBM SPSS Version 20.0 for Windows.

3. Results and Discussion

3.1 Yield Indices of *Sarotherodon melanotheron* Samples

Knowledge of the yield of edible portions from fish is essential for its maximum utilization. As indicated in Table 1, the yield characteristics for *Sarotherodon melanotheron* revealed a fillet yield of $26.02\% \pm 0.23\%$ (dry season) and $25.88\% \pm 0.17\%$ (wet season) for the standard size samples and $25.15\% \pm 0.61\%$ (dry season) and $24.99\% \pm 0.40\%$ (wet season) for the small size samples versus the whole percentage of total fish weight. No significant difference ($P \leq 0.05$) was found in the values based on the statistical test. The processing yield of the present samples on the other hand were $66.65\% \pm 0.74\%$ (dry season) and $67.13\% \pm 0.66\%$ (wet season) for the standard size samples and $66.62\% \pm 0.66\%$ (dry season) and $66.96\% \pm 0.17\%$ (wet season) for the small size samples. No significant difference ($P \leq 0.05$) was found in the said values based as well based on the statistical test. The processing yield is the total weight of a whole fish minus the weight of its head, skin, and viscera. The fillet yield values for all present samples were slightly higher than that for farmed Nile Tilapia or *Oreochromis niloticus* (25.4%) in the previous study by Clement and Lovell^[11]. This suggests that the yield for *Sarotherodon melanotheron*'s edible portion can be comparable to that of the more commercially popular species.

Table 1. Yield indices of standard size and small size *Sarotherodon melanotheron* samples for dry and wet seasons

Form	Farmed <i>Oreochromis niloticus</i>	<i>Sarotherodon melanotheron</i>			
		Dry Season		Wet Season	
		Standard Size Samples	Small Size Samples	Standard Size Samples	Small Size Samples
Whole	100.00%	100.00%	100.00%	100.00%	100.00%
Processing Yield (whole fish weight minus weight of head, skin, and viscera)	51.00% ^[11]	66.65%±0.74%	67.13%±0.66%	66.62%±0.66%	66.96%±0.17%
Fillet	25.40% ^[11]	26.02%±0.23%	25.15%±0.61%	25.88%±0.17%	24.99%±0.40%

- Values are expressed as mean ± standard deviation of 15 measurements
- Values for *Sarotherodon melanotheron* in the same row were found to be not significantly different from each other at $\alpha = 0.05$

3.2 Proximate Composition of *Sarotherodon melanotheron* Samples

Determining the basic nutrients of the fish can justify its consumption and guide its utilization as a raw material for fish product processing. In this present study, the basic nutrients were described in terms of proximate composition. The corresponding mean percentage values for the *Sarotherodon melanotheron* samples are reported in Table 2.

As shown, the crude protein contents of the standard size samples were 19.30%±0.35% (dry season) and 16.54%±0.55% (wet season), while those of the small size samples were 19.23%±0.67% (dry season) and 15.93%±0.13% (wet season). The statistical test revealed that the samples collected during the dry season had values significantly higher ($P \leq 0.05$) than those collected in the wet season. The present figures for the dry season were also far higher than the values for farmed *Oreochromis niloticus* in the previous studies of Desta et al.^[12] and Anani and Agbeko^[13], which reported 14.77% and 16.37%-17.87% respectively. Meanwhile, the present values for the wet season were comparable to the figures from the same previous studies cited. All present samples had protein contents typical for raw tilapia (20.08%) as per the USDA^[14]. Based on the results, we can safely state that *Sarotherodon melanotheron* can be a rich protein source. Its high protein content may be due to its diet^[13,15,16]. For the present samples, such could have come from feeds intended for cultured fish in the ponds they have been infesting.

For crude fat, the standard size samples had mean percentages of 0.63%±0.06% (dry season) 0.50%±0.01% (wet season), while those for the small size samples had 0.57%±0.06% (dry season) and 0.50%±0.01% (wet season). There was no significant difference ($P \leq 0.05$) found among the values. All present values were lower than

that for farmed *Oreochromis niloticus* in the other studies cited earlier. Desta et al.^[12] and Anani and Agbeko^[13] reported 2.39% and 3.14%-4.20% for the species' crude fat respectively. Comparison of the present and previous results using the categories for fish by fat content outlined by Ackman^[17] tells that *Sarotherodon melanotheron* is a lean fish (<2%). Low fat, medium fat, and high fat fish has 2%-4%, 4%-8%, and >8% fat content respectively. Also, the present samples were lower than the average fat content (1.7%) for raw tilapia as per the USDA^[14].

The present crude protein and crude fat figures suggest that *Sarotherodon melanotheron* can be a cheaper alternative fish meat source and a functional raw material in the processing of various fishery products. For instance, the species has so much potential as a raw material for surimi-based goods which depend highly on the amount and strength of fish protein for quality. A high protein and low-fat fish flesh are a well-established surimi raw material standard^[18]. Surimi-based products are growing in demand locally^[19] and abroad^[20]. Apparently, due to its reputation as a pest and its flesh's characteristic bland taste, *Sarotherodon melanotheron* is sold very modestly at local markets in Bataan. A kilogram of the fish will cost only 0.2-0.4 USD.

As seen in Table 2, the major component of all *Sarotherodon melanotheron* samples was moisture. The moisture content of the standard size samples were 79.50%±0.26% (dry season) and 79.76%±0.54% (wet season), while those for the small size samples were 79.23%±0.12% (dry season) and 78.99%±0.94% (wet season). There was no significant difference ($P \leq 0.05$) between the four values. All were within the acceptable range of 60%-80% for tilapia previously mentioned by Tsegay et al.^[16] and were far higher than that for farmed *Oreochromis niloticus* in the previous work by Desta et al.^[12] and

Table 2. Proximate composition of standard size and small size *Sarotherodon melanotheron* samples for dry and wet seasons

Proximate Composition (% w/w)	Typical Values for Raw Tilapia	Farmed <i>Oreochromis niloticus</i>	<i>Sarotherodon melanotheron</i>			
			Dry Season		Wet Season	
			Standard Size Samples	Small Size Samples	Standard Size Samples	Small Size Samples
Crude Protein	20.08 ^[14]	14.77-17.87 ^[12,13]	19.30±0.35 ^a	19.23±0.67 ^a	16.54±0.55 ^b	15.93±0.13 ^b
Crude Fat	1.7 ^[14]	2.39-4.20 ^[12,13]	0.63±0.06	0.57±0.06	0.50±0.01	0.53±0.07
Moisture	78.08 ^[14] ; 60-80 ^[16]	73.62-76.65 ^[12,13]	79.50±0.26	79.23±0.12	79.76±0.54	78.99±0.94
Ash	0.93 ^[14]	1.5-1.96 ^[12,13]	1.30±0.10 ^b	1.23±0.06 ^b	2.25±0.19 ^a	1.05±0.00 ^b

- Values for *Sarotherodon melanotheron* are expressed as mean ± standard deviation of triplicate measurements
- Values for *Sarotherodon melanotheron* in the same row with different superscripts are significantly different at $\alpha = 0.05$

Anani and Agbeko^[13] where 73.61% and 75.61%-76.65% respectively were reported. The present moisture values were also close to the typical figure of 78.08% for raw tilapia^[14].

The high moisture content for the present samples may be attributed to the quality of their proteins and largely to their very small fat content. This is because moisture in fish flesh is associated with the myofibrillar proteins' capacity for water retention, while fat content in meats is proven to be inversely related to water content^[21]. Moisture content is a key quality measure of fishery products because it is linked to their important functional properties^[5]. Also, weight losses in fishery products during frozen storage are related to their capacity for water retention^[22].

In terms of ash content, the present standard size samples figured at 1.30%±0.10% (dry season) and 2.25%±0.19% (wet season). Meanwhile, the ash content of the small size samples had 1.3%±0.06% (dry season) and 1.05%±0.00% (wet season). The value obtained for standard size samples collected during the wet season was found to be significantly higher ($P \leq 0.05$) than the other values. Even then, all present values were comparable to the 1.5%-1.96% range for farmed *Oreochromis niloticus* previously reported by Desta et al.^[12] and Anani and Agbeko^[13]. Also, they were higher than the typical ash percentage of 0.93% for raw tilapia set by the USDA^[14]. Ash content is a measure of the samples' mineral load which may include but is not limited to magnesium, calcium, potassium, and zinc^[14]. It can be said that *Sarotherodon melanotheron* may be similar to *Oreochromis niloticus* by mineral load, and this must be confirmed by more specific mineral compositional analyses.

All in all, the proximate compositional results for the *Sarotherodon melanotheron* samples support the

reports that the chemical characteristics of fish flesh vary according to species, season, and size^[23,24]. The variations in protein and ash compositions by species and fish size found in the present study may be due to the combination of several morphological and physiological factors. Meanwhile, the variations by season may be linked to food availability and quality as well as the physical conditions of water where the pest has been thriving.

3.3 Heavy Metal Load of *Sarotherodon melanotheron* Samples

Heavy metals are elements of high densities and atomic weights that are hazardous even at very low concentrations^[25,26]. They can be introduced to the aquatic ecosystem through various anthropogenic and non-anthropogenic means^[27,28]. Because of their high degree of toxicity, As, Cd, Pb, and Hg rank among the priority heavy metals of public health significance^[28,29]. The presence of these heavy metals in the *Sarotherodon melanotheron* samples is indicated in Table 3. Data show that Pb, Cd, and As were not detected in all samples. While Hg was detected in both standard size (16.39±0.02 ppb and 14.16±0.08 ppb for dry and wet seasons respectively) and small size samples (11.57±0.46 ppb and 17.90±1.71 ppb for dry and wet seasons respectively), the levels were way below the permissible limits of 500 ppb outlined in FAO 210^[30]. Because Hg is ubiquitous in the environment^[28], its presence in the samples could have come from several sources, but it may be largely attributed to Hg additives in pesticides and fertilizers used in agricultural areas close to the sample collection site and to domestic discharges from nearby communities.

Also, it is worth noting that there were significant

Table 3. Heavy metals load of standard size and small size *Sarotherodon melanotheron* samples for dry and wet seasons

Heavy Metal	Permissible Limits for Fish Flesh	<i>Sarotherodon melanotheron</i>			
		Standard Size Samples		Small Size Samples	
		Dry Season	Wet Season	Dry Season	Wet Season
Pb (ppm)	0.2 ^[30]	ND	ND	ND	ND
Cd (ppm)	0.05 ^[30]	ND	ND	ND	ND
As (ppb)	10000-400000 ^[31]	ND	ND	ND	ND
Hg (ppb)	500 ^[30]	16.39±0.02 ^a	11.57±0.46 ^c	14.16±0.08 ^b	17.90±1.71 ^a

- Values are expressed as mean ± standard deviation of triplicate analysis
- Values with a different superscript in the same row are significantly different at $\alpha = 0.05$
- ND – Not detected

differences ($P \leq 0.05$) in the Hg loads across collection season and fish size, with the values for the wet season small size samples and dry season standard size samples figuring to be the highest. The seasonal variation can be explained by the different effects a season can bring on a water body's pollutant load, and hence on its heavy metal concentrations. The wet season for example can lower the heavy metal concentration via the dilution effect of rainwater run-off, while it can raise the concentration through the flushing-effect of run-off from areas with significant heavy metal sources^[32]. The water in the present study's collection site largely comes from the connecting rivers that directly receive run-offs from many sources. On the other hand, the variation by fish size may be due to the difference in swimming patterns and feeding behavior sometimes observed among different sizes or maturities of the same aquatic species^[33,34].

Overall, the results for the heavy metal load suggest that the *Sarotherodon melanotheron* collected from Bataan farm ponds has no risk of heavy metal contamination and therefore can be consumed safely. The results may also be an indication that Bataan's fish farms have no serious heavy metal concentrations.

3.4 Microbial Count of *Sarotherodon melanotheron* Samples

Analysis of the microbiological quality of fish is important to public health as it determines any presence of pathogenic bacteria. This becomes more necessary if *Sarotherodon melanotheron* will be considered for consumption and utilization as the microbial association is common in tilapia fish^[35-37].

The microbial load of the present study's samples is shown in Table 4. In terms of aerobic plate count, the

standard size samples had a mean count of 5,100.00 CFU/g (dry season) and 4,300.00 CFU/g (wet season), while the small size samples had a mean count of 25,333.33 CFU/g (dry season) and 16,333.33 CFU/g (wet season). All values were not significantly different ($P > 0.05$) from each other, and all were way below the permissible limits of 500,000 CFU/g set in FAO 210^[30]. The aerobic plate count indicates the bacterial populations in a sample^[38]. Their presence in fish flesh can be due to pre- and post-capture contamination^[29]. Similarly, for *Escherichia coli* and *Staphylococcus aureus* counts, the figures were far below the standard limits. All four samples had a mean *Escherichia coli* count of <3 MPN/g and a mean *Staphylococcus aureus* count of <10 CFU/g, which were way below the corresponding permissible counts of 11 MPN/g and 1,000 CFU/g as per FAO 210^[30]. The four values for each parameter were also found to be not significantly different ($P > 0.05$) from each other. *Escherichia coli* and *Staphylococcus aureus* are two of the most common causes of fish spoilage and seafood-borne diseases^[37,39]. Their occurrence in fish flesh is often associated with post-harvest handling^[29].

The present results suggest that the *Sarotherodon melanotheron* infesting Bataan fish farms have a minimal microbial load, hence, the risk of contracting microbial diseases upon consuming and processing the fish is low. This however still hinges on the way the fish will be handled as the occurrence of the microbial parameters measured depends highly on post-capture activities. According to Eltholth^[29], the post-capture contamination of fish could come from contaminated fishing tools, water, and ice; soiled surfaces and containers; and unhygienic handling practices. Nonetheless, further microbial risk assessments for the pest are recommended to further support the present results, considering bacterial growth

Table 4. Microbial load in standard size and small size *Sarotherodon melanotheron* samples for dry and wet seasons

Bacterial Culture	Permissible Limits for Fish Flesh	<i>Sarotherodon melanotheron</i>			
		Standard Size Samples		Small Size Samples	
		Dry Season	Wet Season	Dry Season	Wet Season
Aerobic Plate Count (CFU/g)	<500000 ^[30]	5100.00	25333.33	4300.00	16333.33
<i>Escherichia coli</i> Count (MPN/g)	11 ^[30]	<3	<3	<3	<3
<i>Staphylococcus aureus</i> Count (MPN/g)	1000 ^[30]	<10	<10	<10	<10

- Values are presented as a mean total count of triplicate analysis
- Values with a different superscript in the same row are significantly different at $\alpha = 0.05$

upon storage and their inactivation through processing, cooking, and consumption.

4. Conclusions

This study worked on profiling *Sarotherodon melanotheron* infesting Bataan fish farms in terms of yield, proximate composition, heavy metal load, and microbial count. The results showed that the species has a comparable yield and mineral load with the more commercially used farmed *Oreochromis niloticus*. The species has high moisture and protein contents but is very low in fat making it a lean fish. These characteristics offer a cheaper fish meat alternative and a potential functional raw material in the processing of fishery products. Meanwhile, results for the heavy metal load tests showed that the species have no As, Cd, and Pb contamination, although negligible traces of Hg were found. The Hg load varies across collection season and fish maturity, which suggests its manageability. Similarly, for the microbial contents, the species was found to be very minimal in microbial counts, although best post-capture handling is still suggested due to the kind of microbial parameters measured. It is concluded in this baseline investigation that the *Sarotherodon melanotheron* infesting Bataan farm ponds can be consumed safely and has quality potential for raw material in processed fishery products. However, further information is still needed to establish the best post-capture practices on the species. Also, more studies must be done to determine the impact of storage and processing on its stability.

Author Contributions

The first author was the one responsible for the study's conception and design and was the one who drafted the article and revised it critically for important intellectual content. All listed co-authors meanwhile contributed significantly through data collection, and analysis and interpretation of results. All authors reviewed the results and approved the final version of the manuscript.

Conflict of Interest

All authors hereby declare no conflict of interest for this article's content.

Funding

The study was funded by the Philippine Council for Industry, Energy and Emerging Technology Research and Development, one of the three sectoral planning councils of the country's Department of Science and Technology (DOST) through its Grants-in-Aid Program.

Acknowledgments

The authors would like to acknowledge the Bataan Peninsula State University's Institute of Fisheries and Aquatic Sciences and its Center for Research on Aquaculture and Aquatic Resources in Brackishwater Systems both in Orani, Bataan, Philippines for all the support and assistance in the conduct of this study. The authors would also like to acknowledge the Standards and Testing Division of the DOST-Industrial Technology Development Institute and the DOST-Food and Nutrition Research Institute for the technical assistance in the sample analyses.

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