ABSTRACT

TROPHIC ECOLOGY OF SARDINES: Sardinella lemuru feeding strategies, vulnerability to marine microplastic pollution, and endoparasitism in northern Mindanao

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Sardine is one of the most important marine resources in the Philippines and the dominant species caught in the waters of Northern Mindanao Region (NMR) is Sardinella lemuru. However, the sardine fisheries are dwindling and under threat due to overfishing, habitat degradation, and data necessary for effective management are limited. Understanding the complexity of diet is vitally important to manage the fisheries effectively as fish survival, growth, reproduction, and assimilation of environmental pollutants are associated to food uptake. There has been no detailed investigation of S. *lemuru* diet in the country, particularly in NMR. This dissertation aimed to fill this gap and has 4 major objectives: (1) to better understand the dynamics of sardine catch data from NMR in relation to the remotely sensed environmental data coupled with climatological conditions and to validate the putative trophic link between the sardines and phytoplankton, (2) to assess the implication of various feeding strategies of S. lemuru to microplastic ingestion in the upwelling system of Northern Zamboanga Peninsula (NZP) and other non-upwelling sites along the NMR during the spawning periods of July to September and November to March from 2014 to 2016, (3) to determine the susceptibility of S. lemuru to microplastic pollution through visual inspection of stomach contents, and (4) to establish a discrete stock using parasite data to aid in the formulation of effective management unit of sardine fisheries in NMR.

A Generalized Linear Model (GLM) was developed to determine the important variables in hindcasting and forecasting predictions of S. lemuru catch data in NMR. Ingested phytoplankton pigments were correlated to the ambient phytoplankton standing stock (chl-a) and POC derived from MODIS/Aqua Ocean Color. Results of the sardine catch data for 2009 to 2013 training period showed that 80% of the variance can be accounted using GLM. Location (province) and time (quarterly period) were the important predictors in the model that contributed significantly (59% and 12% respectively, p=0.00) to the variance of the sardine catch data while chl-a and sea surface temperature only accounted 9% in total. The linear relationship between the observed and predicted sardine catch data were all highly significant for the 2002 to 2008 hindcasting (r^2 =0.69, p=0.00) and 2014 to 2016 forecasting $(r^2=0.93, p=0.00)$ periods. Thus, suggesting that the model developed was robust enough to predict the spatial distribution and seasonality of S. lemuru catch data in NMR. However, there was no apparent trophic link between S. lemuru and phytoplankton standing stock in the surface water based on the following results: (1) RS chl-a contributed only 1% of the explained variance in GLM, (2) no correlation between sardine catch and RS chl-a, and (3) no correlation between ingested chl-a and phaeopigment in the stomach of S. lemuru versus RS chl-a and POC. Therefore, the trophic condition of S. lemuru during the spawning seasons in the Northern Mindanao Region cannot be fully explained by RS parameters. It is hypothesized that S. lemuru population potentially explore food reserves below the mixed layer, particularly in the depths of chl-a or phaeopigment maxima characterized by marine snow. Stomach contents of 142 adult *S. lemuru* were identified under the microscope and COI minibarcode was used to identify ingested muscle, scale, and vertebra as 100% *S. lemuru* in BOLD Systems. The graphical analysis of 96 classified food items indicated that marine snow was the most important food while also exhibiting generalist foraging preferences. The 72 plankton species were relatively eaten rarely by the sardines. Results of GLM indicated that 45.05 % of the microplastic variance was accounted by the relative % abundance of scale (25.27 %, p=0.02), muscle (14.15 %, p=0.024), and Conidia filiform (5.63%, p=0.138). The dietary size spectra of the sardine ranged from 5 to 7,698 μ m overlapping with the ingested microplastic sizes. There was no correlation between ingested microplastics versus sardine feeding intensity and relative condition index.

The stomach contents of 600 sardines were examined visually under a microscope, stained with Rose Bengal, and tested with hot needle technique to identify ingested microplastics. These anthropogenic particles were measured and physically classified into fibers, fragments, and films. Results of this study showed that 85% of *S. lemuru* were already contaminated with $3.74 \pm 3.92 \#$ of microplastics even before being processed into various sardine products. These microplastics ranged from 0.12 to 21.30 mm and 80 % were mostly < 2.5 mm size classes. The dominant microplastics were 97.94 % in the form of fibers while 1.52 % and 0.54 % were respectively classified into fragments and films. Method validation by isolating microplastics from spiked samples (n = 30) with three retrieval attempts showed 100% recovery efficiency. While results from Canonical Correspondence Analysis of ingested microplastic data had no relationship with the standard lengths of the sardine and the masses of ingested food materials at varying size classes, the total number of ingested microplastics from 2014 to 2016 were directly correlated (r^2 =0.91, p=0.003) with the human population at the landing sites along the coastline of northern Mindanao.

About 225 sardines were examined for endoparasites in the stomachs and intestines while undertaking dietary study of S. lemuru from Patawag, Sindangan, Dipolog, Iligan, Macajalar, Gingoog, and Butuan Bays. A total of 103 parasites were recovered from the 23.1% of all the surveyed sardines. Three parasites belonging to Phyla Nematoda, Acanthocephala, and Platyhelminthes were enumerated microscopically with the latter 2 as new records. The parasitic prevalence showed discontinued geographic distribution patterns; 11 % of sardines were infected with anisakid in NZP while 12% of sardines were infected with acanthocephalan in Bohol Sea System (BSS). Additionally, the parasitic intensity data showed similar geographic discontinuity between NZP and BSS at 75% remaining information of the Two-way Cluster Analysis dendrogram. Thus, distinct sardine stock formation between NZP and BSS is being hypothesized. This hypothesis was tested using One-way ANOVA, where a marked difference was observed in the mean standard lengths of sardines between NZP and BSS (p = 0.00). Therefore, the use of anisakid and acanthocephalan parasites as biological tags were successful enough to delineate the sardine stocks in NZP and BSS. The results, thus, support the closed fishing season in NZP (DA-DILG Administrative Order No. 1 series of 2011) as well as the recently established 12 Fisheries Management Areas (FMA, BFAR FAO # 263) for the conservation and management of fisheries particularly for FMA 4 and 9 that correspond respectively to NZP and BSS.

It is hoped that the studies will provide bases in furthering the effective management of sardine fisheries while also highlighting the vulnerability to the ubiquitous nature of the emerging marine pollutant in NMR and the importance of targeting microplastics in future pollution control efforts in the country.