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Impacts of sample size for stomach content analysis on the estimation of ecosystem indices

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Abstract

This study used Ecopath model of the Jiaozhou Bay as an example to evaluate the effect of stomach sample size of three fish species on the projection of this model. The derived ecosystem indices were classified into three categories: (1) direct indices, like the trophic level of species, influenced by stomach sample size directly; (2) indirect indices, like ecology efficiency (EE) of invertebrates, influenced by the multiple prey-predator relationships; and (3) systemic indices, like total system throughout (TST), describing the status of the whole ecosystem. The influences of different stomach sample sizes on these indices were evaluated. The results suggest that systemic indices of the ecosystem model were robust to stomach sample sizes, whereas specific indices related to species were indicated to be with low accuracy and precision when stomach samples were insufficient. The indices became more uncertain when the stomach sample sizes varied for more species. This study enhances the understanding of how the quality of diet composition data influences ecosystem modeling outputs. The results can also guide the design of stomach content analysis for developing ecosystem models.

Key words: computer simulation, Ecopath with Ecosim, ecosystem index, optimization sample size, stomach contents analysis

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1 Introduction

An increasing consideration of interactions among different components in the whole ecosystem, has prompted the call to develop ecosystem-based fisheries management (EBFM) (Plagányi, 2007). Instead of focusing solely on an individual component of an ecosystem (like single species approach fishery management), EBFM defines fisheries management portfolios for entire ecosystem (Ainsworth et al., 2010), which greatly promoted the development of ecosystem models. Various models have been developed, like Atlantis ecosystem model (ATLANTIS; Fulton et al., 2004), Ecopath with Ecosim (Christensen and Walters, 2004), and Object-oriented Simulator of Marine ecOSystem Exploitation (OSMOSE; Shin and Cury, 2001). These ecosystem models provide an overview of the ecosystem and even serve as operating models to represent the "real world" including the impact of fishing and other anthropogenic effects (Plagányi, 2007). Thus, these models can play an important role in management strategy evaluation (Halouani et al., 2016).

An ecosystem model commonly requires various types of data, such as species life history traits, trophic interactions among species, biomass and distribution information, and environmental factors (Masi et al., 2017; Raoux et al., 2017). Amongst them, diet compositions inform the quantitative relationship between the prey and predators, and contribute to the understanding of energy flow in the ecosystem and population dynamics (Ahlbeck et al., 2012). Therefore, diet compositions play a key role in many ecosystem models. The accuracy of diet composition can greatly influence the output of ecosystem models (Essington, 2007). For example, Guesnet et al. (2015) showed that the Finn's cycling index, the mean trophic level of captures and the system omnivory index were the most sensitive to less constrained diet compositions in the Ecopath model of the Bay of Biscay continental shelf. However, the development of most ecosystem models relies on sourcing dietary information from the literature and ignores the temporal and spatial variations associated with feeding habit, and the uncertainty of diet composition has been less well studied in modeling.

Stomach content analysis is the most commonly used method to estimate the diet composition of species, especially in the construction of ecosystem models. Stomach content analysis can provide quantitative and detailed compositions of each prey species "eaten" by predators, while methods like isotope analysis and fatty acid analysis pay more attention on the composition of prey types "assimilated" by predators (Davenport and Bax, 2002; Phillips et al., 2014). However, stomach content analysis is time-consuming and a large quantity of analysis may not be practical in many studies (Bock et al., 2017), whereas a small number of stomach samples could lead to the high uncertainty of diet composition. Therefore, an elaborate optimization of sample size may be beneficial in obtaining cost-effective information with

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constrained analysis effort to the construction of ecosystem model. Cumulative prey item curves and cumulative diversity curves are often used to investigate whether the stomach samples are enough to satisfy the stomach content analysis (McQueen and Griffiths, 2004). Power analysis could also determine the sample size of stomachs that can find the difference between diets (Ferry and Cailliet, 1996). However, both cumulative prey curves and power analysis focus on the performance of stomach sample size on dietary description, less research efforts have been concentrated on the influence of stomach sample size on ecosystem models.

Computer simulation studies are widely used to evaluate sampling strategies in the field survey (Simpson et al., 2001). Ideally, the performance measures should consider both accuracy and precision (Sokal and Rohlf, 2012). Relative estimation error (REE) and relative bias (RB) are used to compare the accuracy and precision of sampling designs with different sample sizes. In the fisheries research, most studies tend to focus on survey design to satisfy the requirement of stock assessment, while few studies focus on the adequate sampling strategy on feeding habit analysis (Ahlbeck et al., 2012; Xu et al., 2015; Li et al., 2015).

This study presents a computer simulation approach based on a previously developed Ecopath model in the Jiaozhou Bay (Han et al., 2017), and evaluated how the number of stomach samples influence the model projection. We focus on the diet data of three key species, and compare the influence of different sample sizes on the estimates of trophic level of predators, diversity of prey items, the contribution of dominant prey, and ecosystem model indices. In addition, we evaluate and compare the optimal sample size calculated from traditional methods (cumulative curves) and computer simulation. The objective of this study is to develop a framework for evaluating and optimizing the sample size of stomach content analysis which can meet the requirement of the ecosystem model development.

2 Materials and methods

2.1 *Study area*

This study was conducted in a marine ecosystem in the Jiaozhou Bay located in the southern Shandong Peninsula, China (Fig. 1) with an area of 350 km^2 and connecting the Yellow Sea through a 3.1 km wide mouth (Yuan et al., 2016). The associated fisheries resources were ever rich with a relative biomass of about 120 kg/haul per hour in the 1980s (Liu, 1992). Although fisheries resources of the Jiaozhou Bay have experienced a massive depletion due to overfishing and pollution in the past decades (Zeng, 2004; Yuan et al., 2016), the Jiaozhou Bay ecosystem is still an important feeding, spawning and nursery ground for many com-

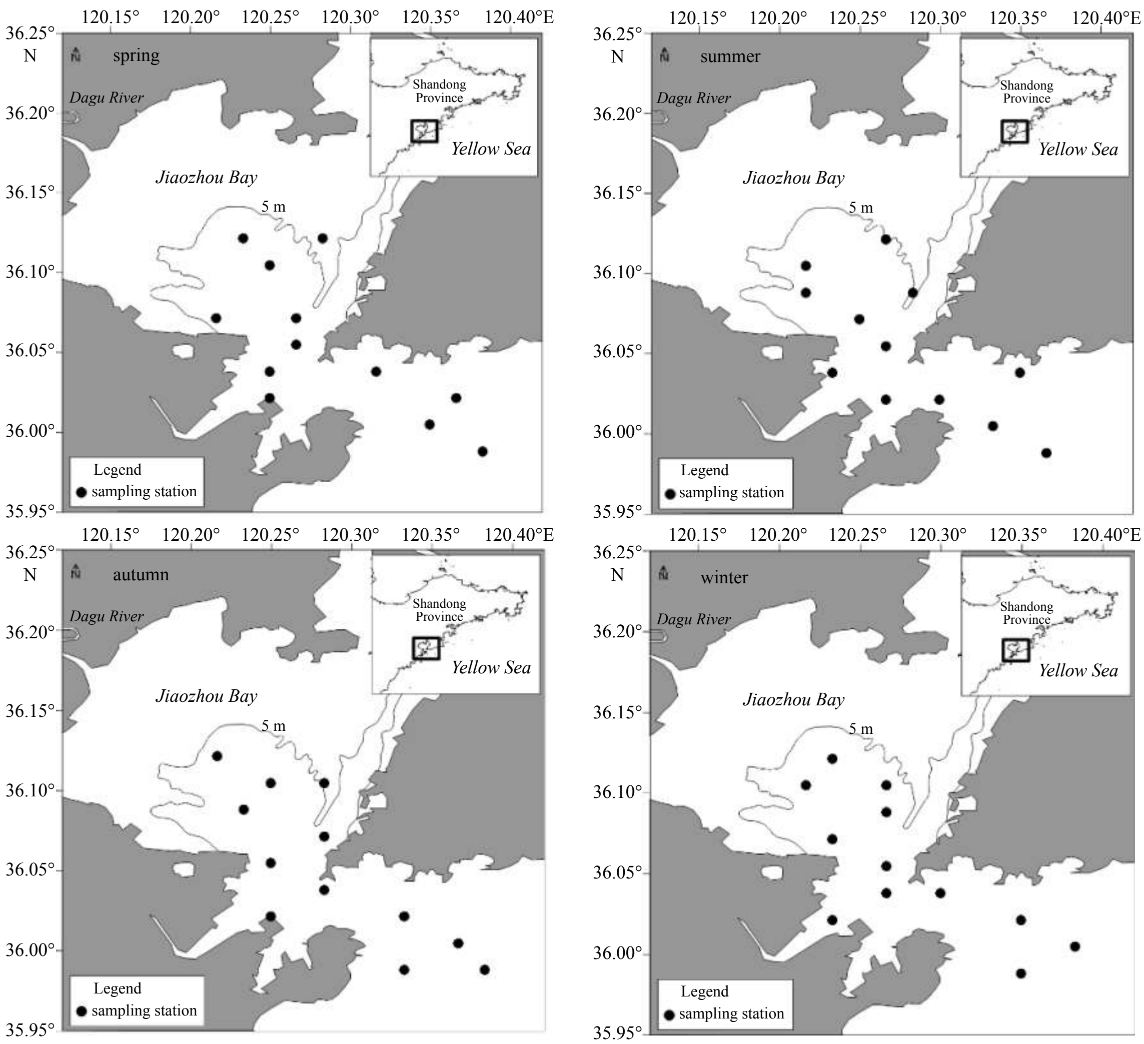

Fig. 1. Map of the Jiaozhou Bay showing the survey stations in four seasons.

mercially important species.

2.2 *Stomach analyses*

This study focused on three fish species and evaluated how their sample sizes might influence the ecosystem modeling. The three species included blenny *Pholis fangi*, pinkgray goby *Amblychaeturichthys hexanema*, and small yellow croaker *Larimichthys polyactis*. Both blenny and pinkgray goby were numerically dominant species in the Jiaozhou Bay ecosystem (Xu et al., 2013; Ma et al., 2017). Pinkgray goby mainly feeds on small benthic invertebrate and is fed by top predators (such as flounder and rockfish) in the Jiaozhou Bay ecosystem (Han et al., 2013), playing an important role in the benthic food chain in this ecosystem. Small yellow croaker was a commercially important species in coastal waters of China. A total of 1 684 stomachs representing three fish species were sampled and analyzed from four seasonal bottom trawl surveys in 2011 (Fig. 1), of which 1 125 had identifiable stomach contents and were used in the following simulation procedures (Table 1, Fig. 2). Samples collection were described with detail information in Han et al. (2017). Stomachs were frozen prior to dissection in the laboratory. In the laboratory, stomach contents were analyzed using a dissecting microscope, while dietary items were identified to the lowest taxonomic level. Biomass percentage was used to evaluate diet composition of each species. Although many prey items only had parts left, their actual weight was recorded. Correction procedures were not applied on those items because it is hard to find all the relationships between remnant body and whole body (Ainsworth et al., 2010). Additionally, the status of each prey item (complete or incomplete) before eaten was not clear.

Table 1. Summary of stomach samples of three species used in the study

Species	Body length range/mm	Number of stomachs	Number of empty stomachs
Blenny *Pholis fangi*	11–172	626	195
Pinkgray goby *Amblychaeturichthys hexanema*	11–137	659	272
Small yellow croaker *Larimichthys polyactis*	26–210	399	92

2.3 *Ecopath model construction*

An Ecopath model was developed for the Jiaozhou Bay (JZB) ecosystem to evaluate the influence of shellfish aquaculture on the ecosystem (Han et al., 2017). Due to data requirements for the ecosystem model, we identified prey items to the level of functional groups. Prey items were assembled into 16 groups (e.g., benthic fish, pelagic fish, shrimp, crabs, and bivalve) according to the JZB Ecopath model. We simulated the effect of stomach sample sizes by modifying the existing Ecopath model in the diet information of three species, i.e., blenny, pinkgray goby, and small yellow croaker. The modified model was constructed with R package "Rpath" (https://github.com/slucey/RpathDev). We denoted a "true ecosystem" based on "full diet information" defined for the full sample sizes for these three species, and all the indices produced by this model were set as the baseline indices.

2.4 *Simulation procedure*

For each species, the original stomach content data were resampled randomly at a defined sample size (from 1 to the total number of stomachs with identified contents) with replacement

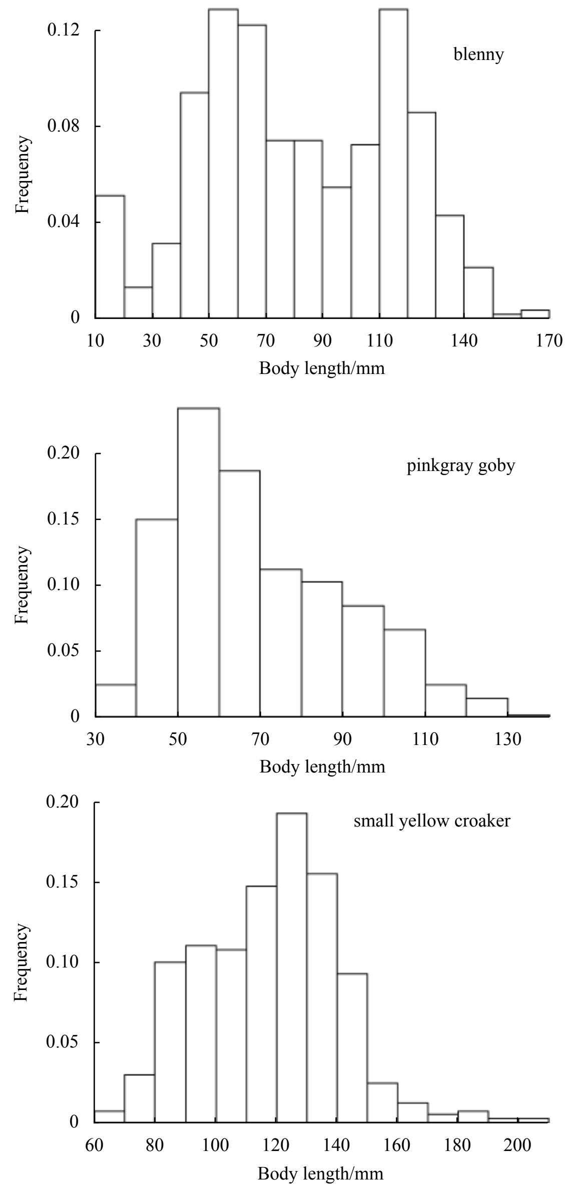

Fig. 2. Body length distribution of three species analyzed in this study.

for 1 000 times. Updated diet information of single or all three species was then applied in the modified JZB Ecopath model (Fig. 3). Thus, the effects of variations in diets for single or multiple species on modeling were evaluated.

For each simulation, the number of prey types and prey diversity of each species were calculated. Ecosystem indices were derived from the updated models and were used for subsequent analysis. The changes in these indices were used to evaluate the influence of each species and all three species on the Ecopath modeling (Fig. 3).

2.5 *Performance measures*

Ecosystem characteristics can be described by a set of ecological indices, which are widely used in ecosystem evaluation and fisheries management (González et al., 2016; Ofir et al., 2017). In this study, based on the potential impacts of diet composition, ecosystem indices calculated from modified models were classified into three levels: "direct index (Species level)", "indirect in-

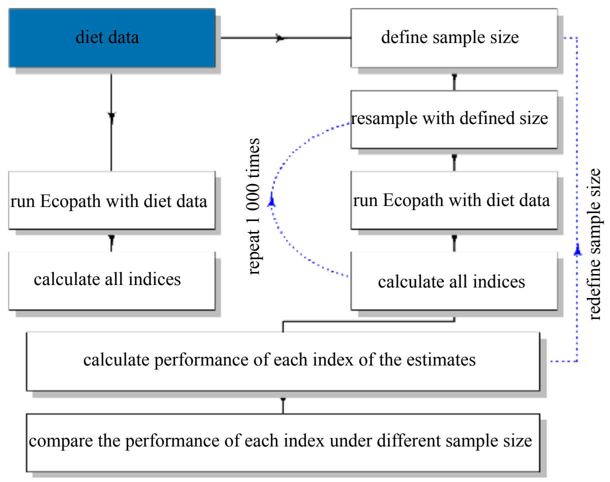

Fig. 3. Flowchart of the simulation study summarizing the framework for the evaluation of impact of stomach sample size on ecosystem indices.

dex (Multispecies level)" and "systemic index (Ecosystem level)" (Table 2). Direct indices, such as the number of prey type groups, prey diversity, and trophic level of predator, can be directly influenced by diet composition. Indirect indices, such as ecotrophic efficiency (EE) of prey groups in ecosystem, can be influenced by both diet information and predator-prey relationships. EEs of shrimp, crab, and polychaete were selected as the indirect indices due to their main contributions in the diet of high trophic level species in the Jiaozhou Bay ecosystem (Han et al., 2013). Systemic indices, such as total system throughout (TST), total primary production/total respiration (TPP/TR), are to describe the ecosystem status, and influenced by both diet information and complexity of trophic interactions.

2.6 *Measures for evaluating performance of different sample sizes of stomach content analysis*

Cumulative curves (Elliott, 1967) were used to evaluate the performance of direct indices under different sample sizes of each species. As sample size increases, the variations of prey group number and similarity between diet composition calculated from sampled stomachs and total stomachs tend to decrease. Finally, the curves reach an asymptote level as new prey types are being introduced into the diet only rarely (Ferry and Cailliet, 1996). We used Bray-Curtis similarity index to calculate the similarity between resampled diet composition and full diet composition of certain species.

For indirect and systemic indices, relative estimate error (REE) and relative bias (RB) were used to measure the accuracy and precision of estimation (Paloheimo and Chen, 1996; Li et al., 2015):

$$\mathrm{REE} = \frac{\sqrt{\sum_{i=1}^{R}\left(Y_i^{\mathrm{estimate}} - Y^{\mathrm{true}}\right)^2/R}}{Y^{\mathrm{true}}} \times 100\%,$$

$$\mathrm{RB} = \left(\frac{\sum_{i=1}^{R} Y_i^{\mathrm{estimate}}/R}{Y^{\mathrm{true}}} - 1\right) \times 100\%,$$

where Y_i^{estimate} is the estimated value of each index from the model with ith resampled data set; Y^{true} is the index value from the model with the whole data; and R is the times of sampling for a given sample size (i.e., 1 000 times in this study).

3 Results

3.1 *Baseline values of indices*

The key indices used in the impact of sample size for stomach content analysis on estimates of ecosystem indices in the simulation study were summarized in Table 2. The definitions and values for the three levels of indices were presented. Based on the trophic level values, all three species belong to the secondary consumer in the ecosystem. In the 24 functional groups of Ecopath model, the prey items of blenny, pinkgray goby, and small yellow croaker included 11, 10, and 13 groups, respectively. The main prey groups were shrimp for blenny and small yellow croaker, and polychaete for pinkgray goby. Blenny and pinkgray goby had relatively low prey diversity, mainly because they specifically fed on certain prey types.

3.2 *Variations of prey compositions and similarity*

The accumulation of the breadth of prey types with increas-

Table 2. Summary of the key indices for the three fish species considered in this study

Type of index	Specific index	Description	Small yellow croaker	Pinkgray goby	Blenny
Direct	*N*	number of prey groups	13	10	11
Direct	*H*	Shannon's diversity	2.03	1.28	1.36
Direct	TL	trophic level	3.86	3.74	3.15
Direct	*S*	Bray-Curtissimilarity index	similarity of food composition between certain sample size and full sample composition		
Direct	Mp	contribution of main prey item	shrimp 23.73%	polychaete 63.48%	shrimp 54.36%
Indirect	EE_shrimp	ecotrophic efficiency of shrimp in ecosystem		0.86	
Indirect	EE_crab	ecotrophic efficiency of crab in ecosystem		0.65	
Indirect	EE_poly	ecotrophic efficiency of polychaete in ecosystem		0.05	
Systemic	TST	total system throughout		11 781.62	
Systemic	TPP	total primary production		5 760.89	
Systemic	NSP	net system production		5 298.54	
Systemic	SOI	system omnivorous index		0.18	

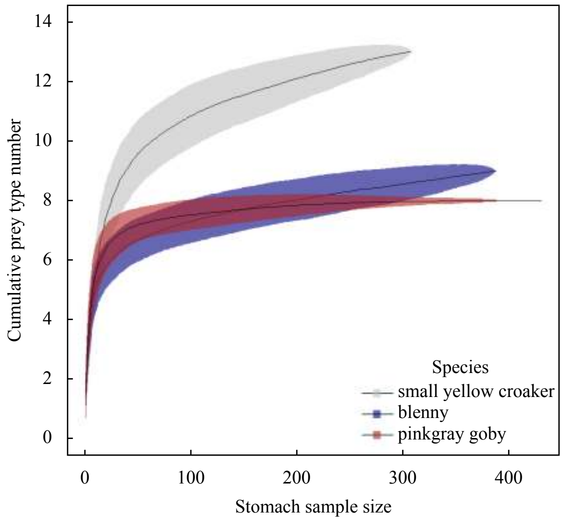

Fig. 4. Cumulative prey type number in relation to the stomach sample sizes of three fish species.

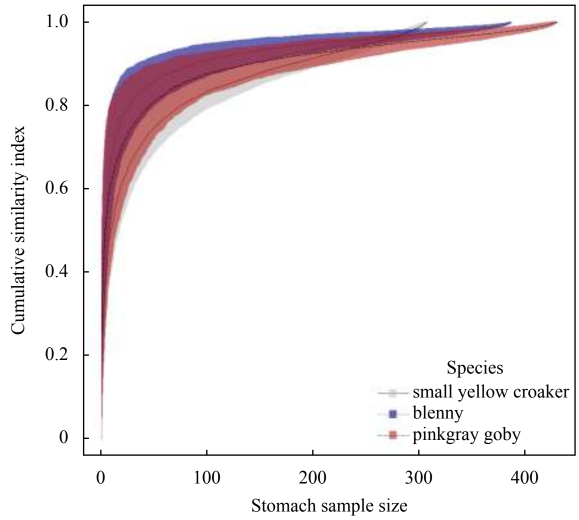

Fig. 5. Cumulative curves for similarity value between sampled diet composition and full sample data in relation to the stomach samples sizes of three fish species.

ing sample size for each species were shown in Fig. 4. The number of prey species appeared to be an asymptote when the stomach number increased. The mean level of stomach sample sizes, at which the cumulative prey type number reached was 323 (range 76–387), 232 (range 16–430), and 228 (range 52–307) for blenny, pinkgray goby, and small yellow croaker, respectively. The stomachs contained cumulative prey curves revealed that the number of stomachs were enough to describe the diet composition of these three species (Fig. 4).

Cumulative similarity curves revealed that the similarity of diet composition between the resampled data and full sample data increased with increasing number of stomach samples (Fig. 5). For example, mean similarity index between diet composition derived from 50 stomachs and full sample data was 85.7%, and could reach to 90% when sample size increased to 100 stomachs. The sample size at which the mean similarity value reached 80%, was 28 for blenny, 48 for pinkgray goby, and 76 for small yellow croaker, respectively.

3.3 *Impact of stomach sample size of single species on ecosystem model output*

For each species, the REE values of each index decreased gradually as the sample size of stomach content analysis increased, whereas the magnitude of changes differed among species and indices (Fig. 6). For most indices, the REE had a relatively distinct decrease with the increase of stomach number, and then remained relatively constant or slightly decreased with a further increase of stomach sample sizes. At the same sample size, the REEs for the direct indices such as the main prey contribution, trophic level and diversity were higher than those of indirect indices such as ecotrophic efficiency of shrimp and ecotrophic efficiency of crab. For example, when the sample size was 50 for small yellow croaker, the REE of main prey contribution was 24%, while the REE of ecology efficiency of shrimp was 0.5%. In addition, the variations of stomach samples sizes of single species had little impacts on the REE of most systemic indices such as TST and TPP, suggesting that the systemic indices were more robust than direct or indirect indices.

The trend of RBs of each index with increased sample size were similar to the performance of REE (Fig. 7). The RBs of direct indices were higher than indirect indices at same sample size. While most systemic indices were the most robust with different sample sizes, except the system omnivory index (SOI). With the increased sample size of each species from 50, RBs of some direct indices changed obviously. For example, the maximum value of RB for the Shannon-Wiener diversity of small yellow croaker was 12.5% when sample size was 50, indicating a large variation in diet composition at low stomach sample size.

3.4 *Combined impact of stomach sample sizes of three species on ecosystem model output*

When evaluating the impact of changing stomach samples size of three predators on the ecosystem model output, direct indices like contribution of prey type and trophic level of predator were not under consideration because they could only be impacted by the diet composition of each predator directly. When stomach sample sizes of all three species changed, the variations of RB and REE of indirect indices and systemic indices were similar to those of single species (Fig. 8). The RBs of indirect indices fluctuated around zero when stomach sample sizes of three species increased from 50 to 300. The REEs of indirect indices decreased gradually as the increase of stomach sample size. RBs and REEs of systemic indices like TST and TPP were close to zero and remained stable, indicating that most systemic indices were robust with increased sample size. However, RB of system omnivory index (SOI) was higher than other indices when stomach samples number of three species increased from 50 to 200, and close to 0 after then. Variations of REE of SOI were similar with RB of SOI, indicating obvious variations of food web structure when stomach samples number of three species under 200. Besides, when stomach sample sizes of three species changed, the estimated REE and RB of each index were larger than those of single species, indicating that the uncertainty of model output may raise when stomach number of more species changed.

4 Discussion

This study revealed that most systemic indices of the Ecopath model in the Jiaozhou Bay were robust and nonbiased regarding limited sample size, whereas specific indices related to species

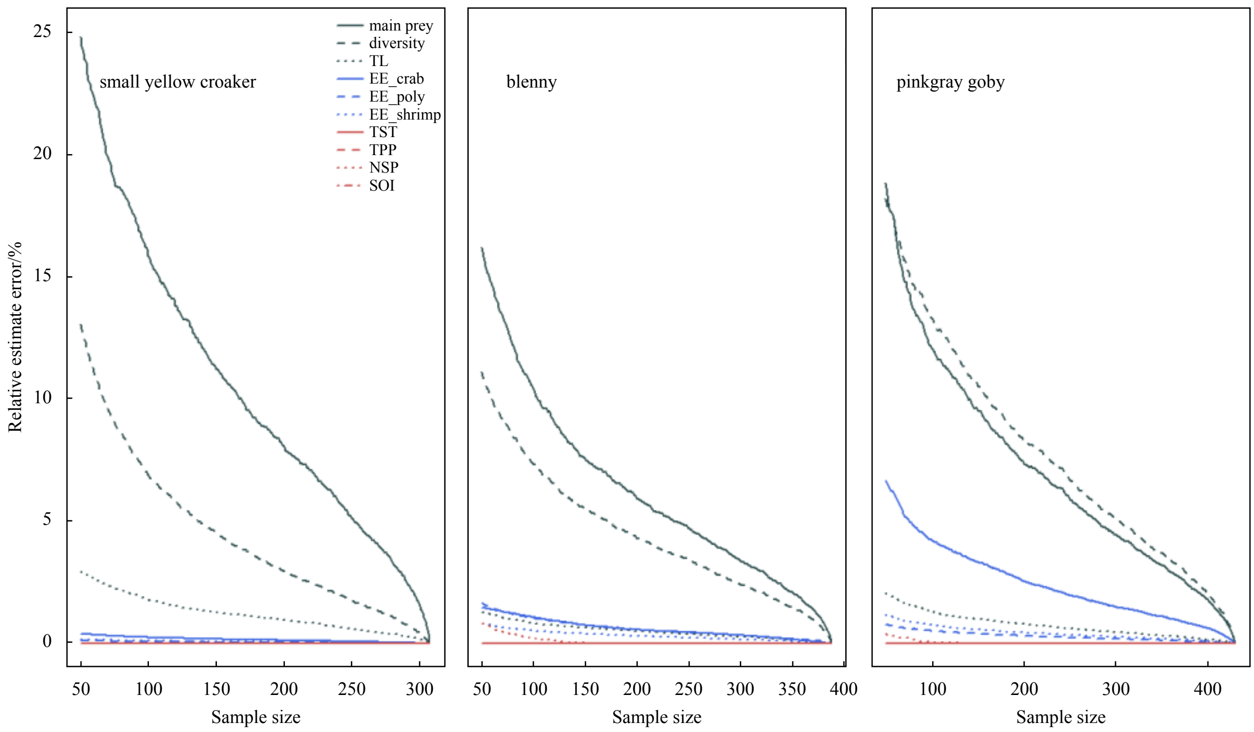

Fig. 6. Relative estimate error (REE) of each index with sample size of stomach content analysis for each fish species. See Table 2 for the detailed information of each index.

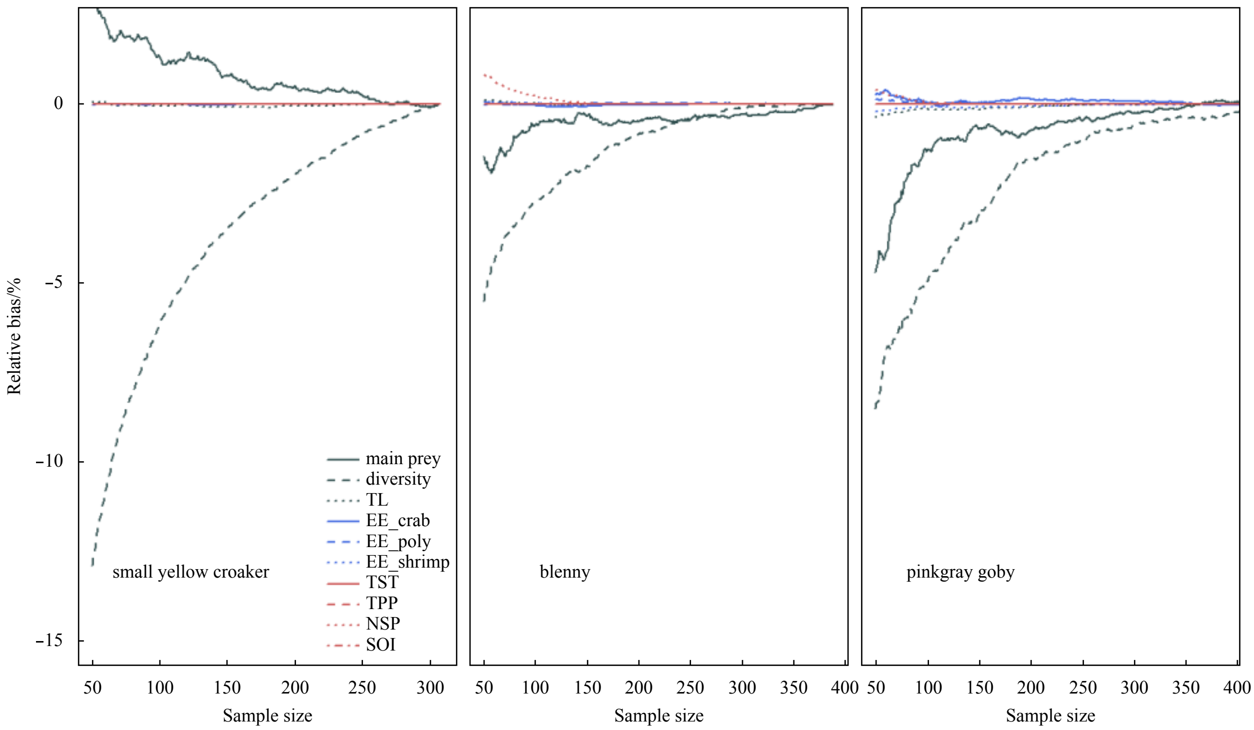

Fig. 7. Relative bias (RB) of each index with sample size of stomach content analysis for each fish species. See Table 2 for the detailed information of each index.

were indicated to be with low accuracy and precision when stomach sample sizes were insufficient. This indicates that appropriate stomach sample sizes need to be defined for different targets in ecosystem modeling. If a model aimed to examine the systemic status of the ecosystem, a few stomach samples may be enough. More stomach samples are required if the complex interactions among species and specific index of each group were under consideration. For example, systemic indices like total primary production, total system throughout are always used to evaluate the impact of climate or human activities on the whole

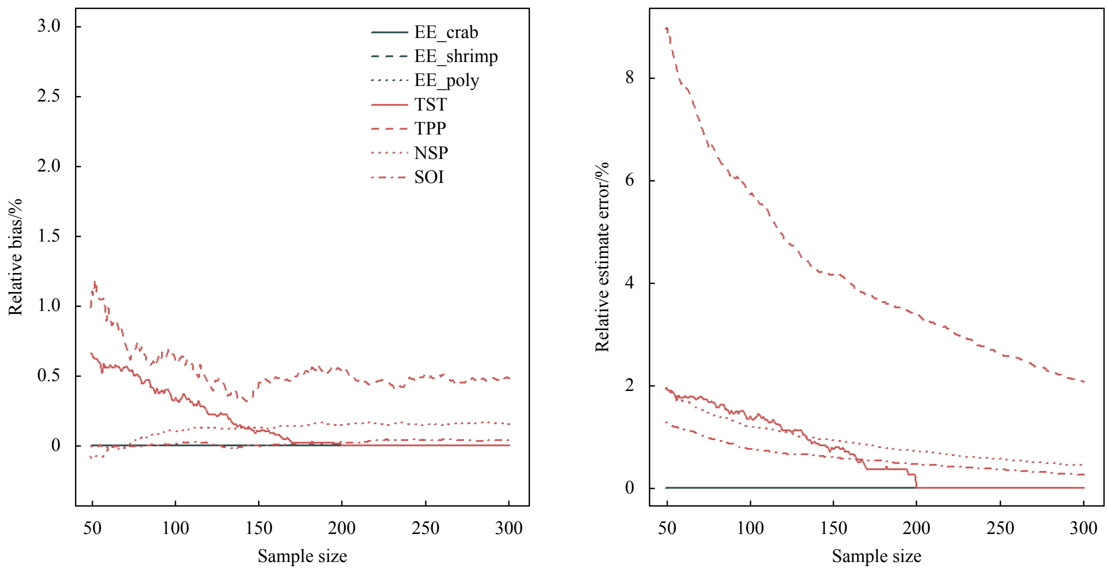

Fig. 8. Relative bias (RB) and relative estimate error (REE) of each index with the variations in stomach samples sizes of three species.

ecosystem (Plagányi, 2007; González et al., 2016; Cremona et al., 2018). In this situation, a small sample size might satisfy the development of systemic indices from ecosystem modeling, which would greatly save the time spent on stomachs analysis and reduce sampling effort. Thus, fishing mortality induced by the survey may also reduce, which is important in the protection of depleted stocks or rare stocks with low abundance in coastal waters (Xu et al., 2015). Cumulative prey item number curve is often used to evaluate whether the number of stomachs is sufficient to describe diet composition of species (Ferry and Cailliet, 1996; McQueen and Griffiths, 2004). The estimated value of the number of main prey groups and diversity index might be with low accuracy when the stomach sample size is too low. This mainly resulted from the absence of rare prey species in small stomach samples. With an increase of analyzed stomachs, the RB value decreased gradually. However, just like dominate species determining the trophic structure of an ecosystem, in most cases, the contribution of dominate prey items is the main factor influencing the ecology niche of the predators. The absence or presence of rare prey items showed small impact on trophic levels of analyzed species as well as ecosystem characters. As the contribution of main prey items was relatively robust with different stomach sample sizes, the trophic position of a predator calculated by low stomach samples were relatively stable, which made the ecosystem character more stable with less stomach samples. Since the purpose of cumulative prey item curve is to cover all the prey items in the analyzed stomachs, adequate sample size by this method may add extra workload in the development of ecosystem model.

In the Ecopath model of the Jiaozhou Bay, we assumed that the “true” feeding habit of each species as the weight percentage calculated by full stomach samples (Stergiou and Karpouzi, 2002; Heymans et al., 2016). Diet analyses can be conducted in numerous ways, like number, biomass or frequency of occurrence of prey items (Hyslop, 1980). However, how well each method describes the “true” feeding habit is still unknown. Some studies showed that frequency of occurrence provided the most robust and interpretable measurement of diet composition because it was not affected by the condition of prey items (Baker et al., 2014; Buckland et al., 2017). Ainsworth et al. (2010) argued that estimating fish diet compositions by bootstrap and likelihood methods from multiple data sources might be more robust. Ahlbeck et al. (2012) used an individual model and found that mass and points methods produced more accurate diet composition. In this study, we assumed that the mass methods could describe the “true” feeding habit of species since it can be used to represent the energy transport from prey to predators (Wilhm, 1968).

Based on the cumulative prey item curves, the adequate stomach samples size for the three species were all more than 200, which is larger than those in similar studies. Mean asymptotic stomach number of snoek (*Thyrsites atun*) in the southern Benguela was around 50 (McQueen and Griffiths, 2004), and 29 individuals were enough for stomach content analysis of *Notoliparis kermadecensis* (Gerringer et al., 2017). Within the three species, asymptotic value of small yellow croaker was larger than that of the other two species. One main reason of this phenomenon is the impact of mean prey items number per stomach. In this study, three fish species were all small sized fish and only one or two prey types were identified in each stomach, while a mean of 7 prey species were identified per sample of snoek (McQueen and Griffiths, 2004). Prey composition in each stomach tends to be variant when few items were eaten by the predator, and more stomachs were needed to describe accurate diet information for this predator. Another reason might come from the impact of total prey items. A total of 12 prey types were consumed by the species analyzed in this study, while only 8 prey types were consumed by Mariana liparid (Gerringer et al., 2017). Furthermore, we assumed that there should have certain relation between “the ratio of prey items per stomach and total prey items” and “the asymptotic stomach samples for species feeding habit analysis”, and high ratio meant small number of stomach samples are needed to describe the diet composition.

Effects of reducing stomach sample size of single species and all three species on ecosystem indices were evaluated in this study. Results showed that when stomach samples of three species changed, REE and RB values of each ecosystem index were larger than those caused by the change for a single species, indicating a complex combined effect of diet uncertainty on ecosystem model outputs. Impact of the body length of individuals on their diet compositions were not evaluated. Ontogenetic change in feeding habit of species and feeding shifting are common in marine ecosystems (Xue et al., 2004; Won et al., 2010; Buckland et al., 2017). Generally, Ecopath assumes the diet composition in each group is same and stable. Only group with sufficient feeding habit data and high-quality parameter can express the ontogenetic change in diet composition by divided one group into several ontogenetic stages (Christensen and Walters, 2004). In the base model, to account for ontogenetic diet shift in feeding patterns, diet composition for these species were calculated from stomach samples with all length size range. We assumed the ontogenetic change in feeding habit of species as one of the error sources in diet composition and sampled the stomach samples randomly in the following simulation process. As well as ontogenetic change of feeding habit, temporal and spatial variations of analyzed species also impact the adequate sample size for feeding habit analysis. Future study will focus on the adequate stomach samples for ecological modeling under different sampling strategy.

The uncertainty of other parameters was not considered in the simulation study. Interactions between each parameter could result in more complex uncertainty in the impacts of diet information on ecosystem model projections. The combined effect could be additive, synergistic, or antagonistic (Crain et al., 2008; Fu et al., 2018). Sensitivity analysis could be used to evaluate the possible consequence of each scenario and estimate the requirement of parameter accuracy. For example, a Matlab toolbox (ecopat_matlab) developed by Kearney (2017), can test the parameter uncertainty of Ecopath model (Guesnet et al., 2015). Besides, performance of stomach sample size of different functional groups on model projection may be different because of the role they played in ecosystem. Both pinkgray goby and small yellow croaker feed mostly on shrimp (Xue et al., 2004; Han et al., 2016). Since pinkgray goby is one of the most abundant species in the Jiaozhou Bay (Zeng et al., 2012; Han et al., 2016), the change in stomach sample size of pinkgray goby has caused more bias on the EE of shrimp. The role and function of each functional group in the ecosystem may influence its optimal stomach size for the model requirement, which needs more detailed analysis. Furthermore, since one main purpose of ecosystem models is to predict possible condition of ecosystem under different management strategies or climate change (Ofir et al., 2017; Raoux et al., 2017). A low level of uncertainty at the beginning of ecosystem modeling may result in a large change at the end of model simulation by iterations (Jopp et al., 2011), which calls for more accurate parameters and information in the construction of ecosystem models. Given the constraints associated with the sample sizes and needs for a better parameterized ecosystem model, future study should aim to optimize the stomach sample sizes that satisfy the modeling needs for data quality and overcome the issues of limitation for sampling efforts.

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References

Ahlbeck I, Hansson S, Hjerne O. 2012. Evaluating fish diet analysis methods by individual-based modelling. Canadian Journal of Fisheries and Aquatic Sciences, 69(7): 1184–1201, doi: 10.1139/f2012-051

Ainsworth C H, Kaplan I C, Levin P S, et al. 2010. A statistical approach for estimating fish diet compositions from multiple data sources: Gulf of California Case study. Ecological Applications, 20(8): 2188–2202, doi: 10.1890/09-0611.1

Baker R, Buckland A, Sheaves M. 2014. Fish gut content analysis: robust measures of diet composition. Fish and Fisheries, 15(1): 170–177, doi: 10.1111/faf.12026

Bock C, Wermter F C, Mintenbeck K. 2017. MRI and MRS on preserved samples as a tool in fish ecology. Magnetic Resonance Imaging, 38: 39–46, doi: 10.1016/j.mri.2016.12.017

Buckland A, Baker R, Loneragan N, et al. 2017. Standardising fish stomach content analysis: the importance of prey condition. Fisheries Research, 196: 126–140, doi: 10.1016/j.fishres.2017.08.003

Christensen V, Walters C J. 2004. Ecopath with Ecosim: methods, capabilities and limitations. Ecological Modelling, 172(2–4): 109–139, doi: 10.1016/j.ecolmodel.2003.09.003

Crain C M, Kroeker K, Halpern B S. 2008. Interactive and cumulative effects of multiple human stressors in marine systems. Ecology Letters, 11(12): 1304–1315, doi: 10.1111/j.1461-0248.2008.01253.x

Cremona F, Järvalt A, Bhele U, et al. 2018. Relationships between fisheries, foodweb structure, and detrital pathway in a large shallow lake. Hydrobiologia, 820(1): 145–163, doi: 10.1007/s10750-018-3648-2

Davenport S R, Bax N J. 2002. A trophic study of a marine ecosystem off southeastern Australia using stable isotopes of carbon and nitrogen. Canadian Journal of Fisheries and Aquatic Sciences, 59(3): 514–530, doi: 10.1139/f02-031

Elliott J M. 1967. The food of trout (*Salmo trutta*) in a Dartmoor stream. Journal of Applied Ecology, 4(1): 59–71, doi: 10.2307/2401409

Essington T E. 2007. Evaluating the sensitivity of a trophic mass-balance model (Ecopath) to imprecise data inputs. Canadian Journal of Fisheries and Aquatic Sciences, 64(4): 628–637, doi: 10.1139/f07-042

Ferry L A, Cailliet G M. 1996. Sample size and data analysis: are we characterizing and comparing diet properly? In: Mackinlay D, Shearer K, eds. Feeding Ecology and Nutrition in Fish, Symposium Proceedings. San Francisco: American Fisheries Society, 71–80

Fu Caihong, Travers-Trolet M, Velez L, et al. 2018. Risky business: the combined effects of fishing and changes in primary productivity on fish communities. Ecological Modelling, 368: 265–276, doi: 10.1016/j.ecolmodel.2017.12.003

Fulton E A, Smith A D M, Johnson C R. 2004. Biogeochemical marine ecosystem models I: IGBEM—a model of marine bay ecosystems. Ecological Modelling, 174(3): 267–307, doi: 10.1016/j.ecolmodel.2003.09.027

Gerringer M E, Popp B N, Linley T D, et al. 2017. Comparative feeding ecology of abyssal and hadal fishes through stomach content and amino acid isotope analysis. Deep Sea Research Part I: Oceanographic Research Papers, 121: 110–120, doi: 10.1016/j.dsr.2017.01.003

González J, Ortiz M, Rodríguez-Zaragoza F, et al. 2016. Assessment of long-term changes of ecosystem indexes in Tongoy Bay (SE Pacific coast): based on trophic network analysis. Ecological Indicators, 69: 390–399, doi: 10.1016/j.ecolind.2016.04.019

Guesnet V, Lassalle G, Chaalali A, et al. 2015. Incorporating food-web parameter uncertainty into Ecopath-derived ecological network indicators. Ecological Modelling, 313: 29–40, doi: 10.1016/j.ecolmodel.2015.05.036

Halouani G, Ben Rais Lasram F, Shin Y J, et al. 2016. Modelling food web structure using an end-to-end approach in the coastal ecosystem of the Gulf of Gabes (Tunisia). Ecological Modelling, 339: 45–57, doi: 10.1016/j.ecolmodel.2016.08.008

Han Dongyan, Ma Qiuyun, Xue Ying, et al. 2016. Feeding habits of *Amblychaeturichthys hexanema* in Jiaozhou Bay based on carbon and nitrogen stable isotope analysis. Periodical of Ocean

University of China (in Chinese), 46(3): 67–73

Han Dongyan, Xue Ying, Ji Yupeng, et al. 2013. Trophic and spatial niche of five gobiid fishes in Jiaozhou Bay. Journal of Fishery Sciences of China (in Chinese), 20(1): 148–156, doi: 10.3724/SP.J.1118.2013.00148

Han Dongyan, Xue Ying, Zhang Chongliang, et al. 2017. A mass balanced model of trophic structure and energy flows of a semi-closed marine ecosystem. Acta Oceanologica Sinica, 36(10): 60–69, doi: 10.1007/s13131-017-1071-6

Heymans J J, Coll M, Link J S, et al. 2016. Best practice in Ecopath with Ecosim food-web models for ecosystem-based management. Ecological Modelling, 331: 173–184, doi: 10.1016/j.ecolmodel.2015.12.007

Hyslop E J. 1980. Stomach contents analysis—a review of methods and their application. Journal of Fish Biology, 17(4): 411–429, doi: 10.1111/j.1095-8649.1980.tb02775.x

Jopp F, Reuter H, Breckling B. 2011. Modelling Complex Ecological Dynamics: an Introduction into Ecological Modelling for Students, Teachers & Scientists. Berlin, Heidelberg: Springer, 1–397

Kearney K A. 2017. ecopath_matlab: a matlab-based implementation of the Ecopath food web algorithm. Journal of Open Source Software, 2(9): 1–2, doi: 10.21105/joss.00064

Li B, Cao J, Chang J H, et al. 2015. Evaluation of effectiveness of fixed-station sampling for monitoring American lobster settlement. North American Journal of Fisheries Management, 35(5): 942–957, doi: 10.1080/02755947.2015.1074961

Liu Ruiyu. 1992. Ecology and Living Resources of Jiaozhou Bay. Beijing: Science Press (in Chinese), 1–429

Ma Qiuyun, Jiao Yan, Ren Yiping. 2017. Linear mixed-effects models to describe length-weight relationships for yellow croaker (*Larimichthys Polyactis*) along the north coast of China. PLoS One, 12(2): e0171811, doi: 10.1371/journal.pone.0171811

Masi M D, Ainsworth C H, Jones D L. 2017. Using a Gulf of Mexico Atlantis model to evaluate ecological indicators for sensitivity to fishing mortality and robustness to observation error. Ecological Indicators, 74: 516–525, doi: 10.1016/j.ecolind.2016.11.008

McQueen N, Griffiths M H. 2004. Influence of sample size and sampling frequency on the quantitative dietary descriptions of a predatory fish in the Benguela ecosystem. African Journal of Marine Science, 26(1): 205–217, doi: 10.2989/18142320409504058

Ofir E, Heymans J J, Shapiro J, et al. 2017. Predicting the impact of Lake Biomanipulation based on food-web modeling—Lake Kinneret as a case study. Ecological Modelling, 348: 14–24, doi: 10.1016/j.ecolmodel.2016.12.019

Paloheimo J E, Chen Y. 1996. Estimating fish mortalities and cohort sizes. Canadian Journal of Fisheries and Aquatic Sciences, 53(7): 1572–1579, doi: 10.1139/f96-077

Phillips D L, Inger R, Bearhop S, et al. 2014. Best practices for use of stable isotope mixing models in food-web studies. Canadian Journal of Zoology, 92(10): 823–835, doi: 10.1139/cjz-2014-0127

Plagányi É E. 2007. Models for an ecosystem approach to fisheries. Rome: FAO, 1–108

Raoux A, Tecchio S, Pezy J P, et al. 2017. Benthic and fish aggregation inside an offshore wind farm: which effects on the trophic web functioning?. Ecological Indicators, 72: 33–46, doi: 10.1016/j.ecolind.2016.07.037

Shin Y J, Cury P. 2001. Exploring fish community dynamics through size-dependent trophic interactions using a spatialized individual-based model. Aquatic Living Resources, 14(2): 65–80, doi: 10.1016/S0990-7440(01)01106-8

Simpson T W, Lin D K J, Chen W. 2001. Sampling strategies for computer experiments: design and analysis. International Journal of Reliability and Applications, 2: 209–240

Sokal R R, Rohlf F J. 2012. Biometry: the Principles and Practice of Statistics in Biological Research. 4th ed. New York: W.H.Freeman, 1–102

Stergiou K I, Karpouzi V S. 2002. Feeding habits and trophic levels of Mediterranean fish. Reviews in Fish Biology and Fisheries, 11(3): 217–254

Wilhm J L. 1968. Use of biomass units in Shannon's formula. Ecology, 49(1): 153–156, doi: 10.2307/1933573

Won N I, Kawamura T, Takami H, et al. 2010. Ontogenetic changes in the feeding habits of the abalone *Haliotis discus hannai*: field verification by stable isotope analyses. Canadian Journal of Fisheries and Aquatic Sciences, 67(2): 347–356, doi: 10.1139/F09-187

Xu Binduo, Zeng Huihui, Xue Ying, et al. 2013. Community structure and species diversity of fish assemblage in the coastal waters of Jiaozhou Bay. Acta Ecologica Sinica (in Chinese), 33(10): 3074–3082, doi: 10.5846/stxb201203040292

Xu Binduo, Zhang Chongliang, Xue Ying, et al. 2015. Optimization of sampling effort for a fishery-independent survey with multiple goals. Environmental Monitoring and Assessment, 187(5): 252, doi: 10.1007/s10661-015-4483-9

Xue Ying, Jin Xianshi, Zhang Bo, et al. 2004. Ontogenetic and diel variation in feeding habits of small yellow croaker *Pseudosciaena polyactis* Bleeker in the central part of Yellow Sea. Journal of Fishery Sciences of China (in Chinese), 11(5): 420–425

Yuan Yuan, Song Dehai, Wu Wen, et al. 2016. The impact of anthropogenic activities on marine environment in Jiaozhou Bay, Qingdao, China: a review and a case study. Regional Studies in Marine Science, 8: 287–296, doi: 10.1016/j.rsma.2016.01.004

Zeng Xiaoqi, Piao Chenghua, Jiang Wei, et al. 2004. Biodiversity investigation in Jiaozhou bay and neighbouring waters. Periodical of Ocean University of China (in Chinese), 34(6): 977–982

Zeng Huihui, Xu Binduo, Xue Ying, et al. 2012. Study on fish species composition and seasonal variation in the shallow waters of Jiaozhou bay. Periodical of Ocean University of China (in Chinese), 42(1): 67–74