E-ISSN 2586-9027
Homepage : *https://tci-thaijo.org/index.php/SciTechAsia* Science & Technology Asia
Vol. 30 No.1 January - March 2025 Page: [290-297]
Original research article

Identification of Gut Microbiota in Blue Swimming Crabs Collected from the Eastern Coast of the Gulf of Thailand Containing Gill Net Debris

Ichaya Paijitpimuk[1], Patarapong Kroeksakul[1], Thayat Sriyapai[1], Wirongrong Duangjai[2], Praepilai Mittrarath[1], Arin Ngamniyom[1,*]

[1]*Faculty of Environmental Culture and Ecotourism, Srinakharinwirot University, Bangkok 10110, Thailand*
[2]*Department of Silviculture, Faculty of Forestry, Kasetsart University, Bangkok 10900, Thailand*

Received 26 October 2024; Received in revised form 1 January 2025
Accepted 10 February 2025; Available online 24 March 2025

ABSTRACT

The aim of this study was to investigate the gut microbiota of blue swimming crabs (*Portunus pelagicus*) in which a piece of gill net debris was found. Next-generation sequencing was performed to analyse the V1–V3 sequences of the 16S rRNA gene for bacteria and the internal transcribed spacer sequences for fungi. Samples of stomach crabs were collected from the coastal wetlands of eastern Thailand. Four fragments of gill nets were found (a single piece per gut sample), with lengths of 5.2–12.5 mm. Stomachs without gill net fragments from a total of four samples comprised Group A, whereas stomachs containing gill net fragments in the four samples comprised Group B. Groups A and B shared 131 OTUs (Operational Taxonomic Units), while they contained 51 and 26 OTUs, respectively. *Photobacterium* was the predominant *Vibrionaceae* present in both groups, but *Marinobacter* of *Alteromonadaceae* was present at high levels in Group A. Interestingly, a single sample in Group B was dominated by *Vibrio*. For fungi, 276 and 195 OTUs were included in Groups A and B, respectively, whereas 224 OTUs were shared by Groups A and B. *Malassezia* was predominant in both groups. *Moesziomyces*, *Ustilago*, *Erythrobasidium* and *Schizophyllum* were more common in Group B than in Group A. In contrast, *Cladosporium*, *Ramicandelaber*, *Claroideoglomus* and *Stachybotrys* were more common in Group B than in Group A. These results provide the first evidence of the microbiota in blue swimming crabs that have gill nets in their stomachs.

Keywords: Crab guts; Monofilament; Microorganisms; *Portunus*

***Corresponding author:** arin@g.swu.ac.th

1. Introduction

Resulting from more than a decade of global pollution, plastic waste is an environmental issue that affects the health and wellness of humans and wildlife [1, 2]. Plastic waste can be degraded and distributed in various environments; for example, plastic contamination in aquatic ecosystems can cause disease and economic loss in aquatic animals [3-5].

In decapod species, blue swimming crabs (*Portunus pelagicus*) belonging to the infraorder Brachyura are famous marine crustaceans species for human consumption and economic aquatic animals in many countries of Southeast Asia including Thailand [6, 7]. Blue swimming crabs mainly inhabit coastal ecosystems [6, 7]. Many studies have reported chemical contaminations and microbial diversity in *Portunus* crabs [7-9]. However, the blue swimming crab is one of several marine species that may be at risk of plastics pollution in natural environments or aquaculture [10].

Nylon monofilaments are synthetic filament lines made from plastic materials that are used in gill nets [11]. Rochman [12] reported the first findings of plastic debris of monofilaments in marine animals. Nylon was found in 38% of fish stomachs from the open waters of the Beibu Gulf, South China Sea [13]. Furthermore, Bordbar et al. [14] showed evidence of nylon filaments in shrimp stomachs from the Mediterranean Sea. However, very little is known for gill nets or monofilaments with the profile of microbial communities in stomachs of aquatic animals.

In molecular biology and microbiology, next generation sequencing has been used to analyse DNA sequences in massive data sets [15, 16]. The 16S rRNA gene is frequently utilized to provide microbial sequences for the purpose of bacterial identification and understanding endosymbiotic interactions in digestive tracts of many aquatic species [17, 18], including crab gut [19]. In fungi, internal transcribed spacer (ITS) regions are standard for performing fungal identification, including from environmental samples [20, 21].

The aim of this study, therefore, was to investigate the microbiota and pieces of debris from gill nets in the stomachs of blue swimming crabs captured from the eastern coast of the Gulf of Thailand.

2. Materials and Methods

Blue swimming crabs (*Portunus pelagicus*) were purchased from fishermen who had collected them from the coast of Chanthaburi-Trat, Gulf of Thailand using crab traps. Forty individual crabs were collected (22 males and 18 females). Living crabs were euthanized by immersing them in cooled artificial seawater with isoeugenol for 30 min [22]. The average weight of these 40 crab bodies was 97.25 g. The guts of the crabs were dissected and stored in separate 50 ml sterilized centrifuge tubes in a cooled plastic box at 4 °C.

After the gill net debris was removed, the crabs' stomachs were washed with isopropanol and then dried overnight. Pieces of gill nets were found in the stomachs of four male crabs.

The study crabs were assigned to two groups. Group A consisted of male crabs without gill nets in their stomachs whereas, Group B consisted of male crabs whose stomachs contained gill nets. The crabs selected for Group A had similar weights and sex to those in Group B to allow for comparing the microbiota profiles between the two groups.

Plastic material types of gill nets were identified under the Fourier-transform infrared spectroscopy (FT-IR) (PerkinElmer, US) from 450-4000 cm^{-1}. In addition, the small fragments of gill nets after dehydration were put on conductive carbon tape on an aluminum stub holder, coated with platinum/palladium using a sputter coater, and then imaged using a scanning electron microscope (SEM) (SEM-HITACHI SU-8010) with an acceleration of 5.0-10.0 kV.

Exterior stomach surfaces were treated with antiseptic solution. Surface tissues from inside the stomachs were moved into 1.5 ml sterile Eppendorf tubes using a scalpel with sterile blades and sterile forceps, immediately followed by genomic extraction.

Total genomic DNA from gut tissues with or without gill nets were extracted using DNeasy Blood & Tissue Kits (Qiagen, Germany) according to the manufacturer's protocol. The PCR mixtures for amplicon were performed by using *Pfu* DNA polymerase MasterMix (Bioneer, South Korea) and added the universal primers for amplifying the V1–V3 hypervariable region of 16S rRNA gene for bacteria and the internal transcribed spacer (ITS) rRNA gene for fungal organisms. PCR cycles were conducted by the following program: 95°C for 3 min to pre-denature, followed by 30 cycles at 95°C for 45 s to denature, 56°C for 45 s to anneal, 70°C for 2 min for extension and a final extension for 10 min. The PCR products were purified by QIAquick Gel Extraction Kit (Qiagen, Germany) and were measured by Qubit®dsDNA HS Assay Kit. The amplicon generation and library preparation, in the sequencing library was constructed using a MetaVX Library Preparation Kit. The library was purified with magnetic beads and qualified by Infinite® 200 PRO microplate reader. Next generation sequencing was conducted on an Illumina/HiSeq 2500.

To generate high-quality clean reads, raw data were filtered using the iTools Fqtools fqcheck software (v.0.25), and a consensus sequence was created by the Fast Length Adjustment of Short reads (v1.2.11). The ITS rRNA data was analysed by QIIME data. Sequences were identified into operational taxonomic units (OTUs) by VSEARCH (1.9.6), the 16s rRNA reference database and the UNITE ITS database with pre-clustered at 97% of sequence identity. The sequences and chimeras were screened and filtered by mapping to gold database (v20110519) and UNITE (v20140703), respectively. Ribosomal Database Program (RDP) was classified to assign taxonomic category of all OTUs at a confidence threshold of 0.8 for predicted taxonomic categories of the genus level. The Venn Plot were performed by Venn Diagram software R (v3.1.1).

3. Results and Discussion

Forty blue swimming crabs were collected to determine if they had gill net debris in their stomachs. Four gill nets were found in each crab stomach (the ratio of the gill net to the sample was 1:1). The lines ranged in length from 5.2–12.5 mm, and they were light blue and slightly transparent monofilaments (Fig. 1a). The gill net surfaces were rough throughout the samples. FT-IR revealed that all the gill nets matched polyamide or nylon 66, with scores greater than 0.90. Gill nets were present only in the male crabs, not in any of the female crabs. Su et al. [23] reported that male *Portunus* crabs were more aggressive than the females, and so the finding that pieces of gill nets were found only in male swimming crabs might be indicative of male crabs damaging fragments of gill nets as an aggressive behaviour.

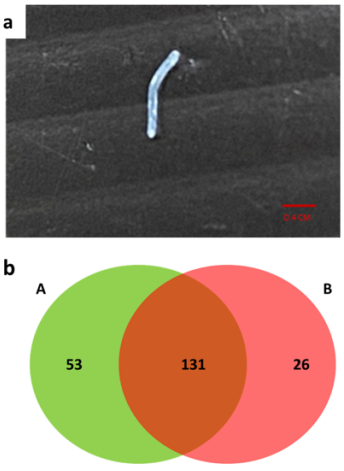

Fig. 1. Piece of debris of gill nets in stomachs of blue swimming crabs (a). Operational taxonomic unit of bacteria represented by the Venn Plot in stomachs without gill nets of the Group A and stomachs with gill nets of the Group B (b).

During microbial identification, 53 OTUs were specifically identified in the stomachs of crabs without gill nets from four individuals (Group A). In contrast, 26 OTUs were specifically detected from the four individuals with gill nets in their stomachs (Group B). Groups A and B shared 131 bacterial OTUs in their stomachs (Fig. 1b).

In bacterial genera, *Marinobacter*, *Photobacterium* and *Vibrio* were detected in Groups A and B. *Photobacterium* was predominant *Vibrionaceae* in both groups, but *Marinobacter* of *Alteromonadaceae* was high in group A. However, it was found that a single sample in Group B was dominated by *Vibrio* (Fig. 2). In addition, the other genera (relative abundance < 0.05) of both groups were composed of *Tenacibaculum*, *Halobacteriovorax*, *Halioglobus*, *Shewanella*, *Sedimenticola*, *Alcanivorax*, *Rhodopirellula*, *Ilumatobacter*, *Blastopirellula*, *Gimesia*, *Roseivivax*, *Maribacter*, *Haliea*, *Pseudoalteromonas*, *Pseudahrensia*, *Propionigenium*, *Pseudomonas*, *Aquihabitans*, *Nautella*, *Actibacter*, *Legionella*, *Aliiroseovarius*, *Mycobacterium*, *Roseibacillus*, *Owenweeksia*, *Blastopirellula*, *Winogradskyella*, *Nonlabens*, *Pelagibius*, *Microbacterium*, *Brevundimonas*, T*etrasphaera*, *Staphylococcus*, *Diaphorobacter*, *Ruegeria*, *Spongiimonas*, *Psychrosphaera*, *Formosa* and *Serratia*.

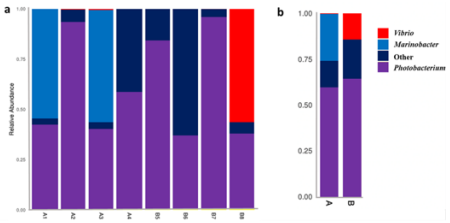

Fig. 2. Operational taxonomic unit of bacterial genera in crab stomachs without gill nets are indicated as A1–4 and stomachs with gill nets are indicated as B5–8 (a). Comparison between Groups A and B (b).

For fungi, there were 276 OTUs specific to Group A and 195 specific to Group B. Groups A and B shared 224 fungal OTUs (Fig. 3).

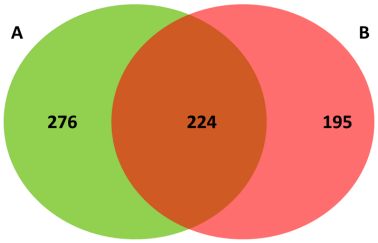

Fig. 3. Operational taxonomic unit of fungi represented by the Venn Plot in stomachs without gill nets of the Group A and stomachs with gill nets of the Group B.

The dominant bacterial community of *Malassezia* was found in both Groups A and B. *Mortierella* followed by *Fusarium* were also dominant at Groups A and B. *Moesziomyces*, *Ustilago*, *Erythrobasidium* and *Schizophyllum* were higher in Group A than in Group B. In contrast, *Cladosporium* was predominant in Group B. In addition, *Ramicandelaber*, *Claroideoglomus* and *Stachybotrys* were higher in Group B than in Group A. In addition, the other genera (relative abundance < 0.05) of both groups consisted of *Cryptodiscus*, *Phaeophleospora*, *Zygoascus*, *Clavaria*, *Bartalinia*, *Rhizopogon*, *Spissiomyces*, *Trichothecium*, *Dialonectria*, *Ovatospora*, *Gibellulopsis*, *Inocybe* and *Xylaria* (Fig. 4).

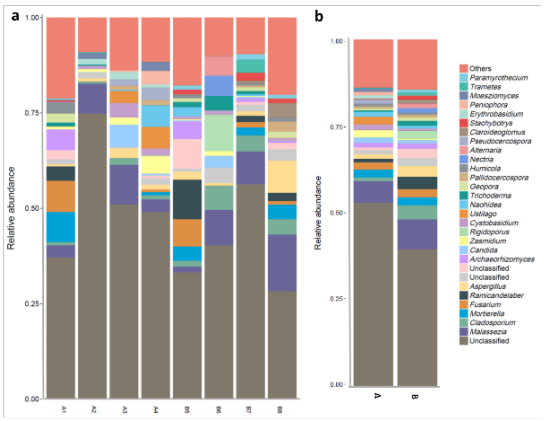

Fig 4. Operational taxonomic unit of fungal genera in crab stomachs without gill nets are indicated as A1–4 and stomachs with gill nets are indicated as B5–8 (a). Comparison between Groups A and B (b).

It is known that polyamide is the main plastic material used for fishery equipment, such as nylon fishing nets or fishing lines [24]. In this study, a piece of gill net from the stomachs of blue swimming crabs was classified as a type of plastic, such as polyamide or nylon 66; this confirmed that marine plastic debris might be from monofilament lines, crab traps or crab gill nets. The results also suggest that major sources of monofilament lines may be nets from fisheries. Furthermore, Yin et al. [25] reported that fewer microplastics were found in edible crabs than in nonedible crabs. Therefore, the occurrence of monofilaments might be a minor item of plastic waste in the stomachs of blue swimming crabs captured from the eastern coast of Thailand.

D'Costa [26] highlighted that plastic particles could induce toxicity in decapods. These particles might contaminate seafood at relatively high trophic levels, including humans. Moreover, it is well known that plastic debris is broken down by photodegradation, hydrolytic degradation and biodegradation, leading to environmental damage and contamination in microorganisms [27]. Thus, the observation of gill nets in blue swimming crabs may be associated with the highest risk of contamination of edible crabs for human consumption.

Microplastics were recently found to have altered the microbial community in the gut of Javanese medaka (*Oryzias javanicus*) [28]. This finding was consistent with our study, in which the microbiota differed between crabs in which gill nets were or were not detectable in the gut. Therefore, gill nets may affect bacterial diversity in the stomachs of blue swimming crabs. In addition, many species of *Vibrio* that are found in a wide variety of aquatic environments can cause infections in animals [29]. These findings suggest that predominance of *Vibrio* associated with gill nets in the stomachs of blue swimming crabs may concern not only gill net contamination but also the risk of pathogenesis in those crabs.

Wei et al. [19] reported that *Arcobacter*, *Photobacterium*, *Vibrio*, *Shewanella* and *Desulfovibrio* were the dominant genera in guts of the mud crab (*Scylla paramamosain*). This is consistent with our results showing that stomachs of blue swimming crabs were dominated by the genus of "*Proteobacteria*" *Photobacterium*. In this study, it thus suggests that *Photobacterium* may be a core gut microbiota of blue swimming crabs inhabiting the coastal wetland in Trat province.

In the genus *Alteromonadaceae*, *Marinobacter* mainly inhabits marine environments, including sediments and seawater [30]. Moreover, *Marinobacter* is also found in the digestive system of marine animals such as fish [31, 32]. In this study, *Marinobacter* was identified in crab stomachs, which may be used as an alternative biomarker or bioindicator for monitoring the absence of nylon monofilament contamination in these crab stomachs.

Recently, Shaumi et al. [33] reported that the dominant fungi identified in the gut of three-spot swimming crabs (*P. sanguinolentus*) from Taiwan were *Candida,* followed by *Apiotrichum*, *Rhodotorula* and *Fusarium*. In coculture systems, *Aspergillus* was the dominant genus, including *Penicillium* and *Talaromyces,* in the gut of Chinese mitten crab (*Eriocheir sinensis*) according to a previous study by Xu et al. [34]. In the present study, *Malassezia* was the dominant genus, followed by *Mortierella* and *Fusarium*. For the trophic mode, *Candida*, *Rhodotorula*, *Fusarium*, *Aspergillus* and *Penicillium* were identified as pathotrophs, saprotrophs, and symbiotrophs, respectively, whereas *Apiotrichum* was identified as a saprotroph. *Talaromyces* and *Malassezia* were pathotrophic and saprotrophic. *Mortierella* is considered a saprotroph-symbiotroph [35]. The fungal communities in the guts of blue swimming crabs differ from those in the guts of three-spot swimming crabs [33] and Chinese mitten crabs [34], which might involve the environment and crab species

being assayed. Therefore, pathotroph-saprotroph interactions may constitute the main trophic mode in the gut of blue swimming crabs. *Malassezia* may constitute a core fungal genus of blue swimming crabs collected from coastal areas in this study.

High relative abundances of *Moesziomyces* and *Ustilago* were detected in Group A, whereas high relative abundances of *Cladosporium* were detected in Group B. *Moesziomyces* and *Ustilago* are pathogens in plants [35]. Among microfungi, *Cladosporium* has been reported to be an animal pathogen and a plant pathogen [33, 36]. Therefore, it could be assumed that the dominance of *Cladosporium* in the gut of blue swimming crabs may be caused by contamination with gill nets. However, it is difficult to determine the relationship between the microbiome and gill nets in stomach crabs because this study did not include a control experiment. In addition, the types of fungal pathogens present in the crab gut are not well understood because this study did not provide fungal identification data at the species level. These results revealed the diversity of gut-associated fungi in stomachs contaminated with fragments of gill nets and normal stomachs of blue swimming crabs.

4. Conclusion

In summary, our investigation reveals the microbial communities of bacteria and fungi present in the guts of blue swimming crabs from the eastern coast of the Gulf of Thailand. Furthermore, the present study may support the understanding of the core microbiota and microsymbiosis of blue swimming crabs in natural environments.

Acknowledgements

This research was funded by the grant of Srinakharinwirot University no. 642/2563 and Fundamental fund 022/2565.

References

[1] Thompson RC, Moore CJ, vom Saal FS, Swan Plastics, the environment and human health: current consensus and future trends. Philos Trans R Soc B. 2009; 364(1526): 2153-66.

[2] Daltry A, Merone L, Tait P. Plastic pollution: why is it a public health problem? Aust N Zeal J Public Health. 2021; 45(6): 535-37.

[3] Danso D, Chow J, Streit WR. Plastics: Environmental and Biotechnological Perspectives on Microbial Degradation. Appl Environ Microbiol. 2019; 85(19): e01095-19.

[4] Cássio F, Batista D, Pradhan A. Plastic Interactions with Pollutants and Consequences to Aquatic Ecosystems: What We Know and What We Do Not Know. Biomolecules. 2022; 12(6): 798.

[5] Beaumont NJ, Aanesen M, Austen MC, Börger T, Clark JR, Cole M, Hooper T, Lindeque PK, Pascoe C, Wyles KJ. Global ecological, social economic impacts of marine plastic. Mar Pollut Bull. 2019; 142: 189-95.

[6] Wiloso EI, Romli M, Nugraha BA, Wiloso AR, Setiawan AAR. Henriksson P.J. G. Life cycle assessment of Indonesian canned crab (Portunus pelagicus). J Ind Ecol. 2022; 26(6): 1-14.

[7] Olatunde O.O., Chantakun K., Benjakul S. Microbial, chemical qualities and shelf-life of blue swimming crab (Portunus armatus) lump meat as influenced by in-package high voltage cold plasma treatment. Food Biosci. 2021; 43: 101274.

[8] Sarasiab AR, Hosseini M. Study on PCB 101 PCB 153, mercury and methyl mercury content in blue crab Portunus Pelagicus from Khuzestan shore (Persian Gulf). J Toxicol Environ Health Sci. 2014; 6: 81-6.

[9] Talpur AD, Memon AJ, Khan MI, Ikhwanuddin M, Danish Daniel MM, Abol-Munafi AB. A Novel of Gut Pathogenic Bacteria of Blue Swimming Crab Portunus pelagicus (Linneaus, 1758) and Pathogenicity of Vibrio harveyi a

Transmission Agent in Larval Culture under Hatchery Conditions. Res J Appl Sci. 2011; 6(2): 116-27.

[10] Daniel DB, Ashraf PM, Thomas SN, Thomson KT. Microplastics in the edible tissues of shellfishes sold for human consumption. Chemosphere. 2021; 264(Pt 2): 128554.

[11] Matos MR, Santos-Bezerra DP, Correa-Giannella ML. Reproducibility of a nylon fishing line as a screening test for diabetic foot ulceration risk. Clinics (Sao Paulo). 2020; 75: e1658.

[12] Rochman CM, Tahir A, Williams SL, Baxa DV, Lam R, Miller JT, Teh FC, Werorilangi S, Teh SJ. Anthropogenic debris in seafood: Plastic debris and fibers from textiles in fish and bivalves sold for human consumption. Sci Rep. 2015; 5: 14340.

[13] Koongolla JB, Lin L, Pan YF, Yang CP, Sun DR, Liu S, Xu XR, Maharana D, Huang JS, Li HX. Occurrence of microplastics in gastrointestinal tracts and gills of fish from Beibu Gulf, South China Sea. Environ Pollut. 2020; 258: 113734.

[14] Bordbar L, Kapiris K, Kalogirou S, Anastasopoulou A. First evidence of ingested plastics by a high commercial shrimp species (Plesionika narval) in the eastern Mediterranean. Mar Pollut Bull. 2018; 136: 472-76.

[15] Slatko BE, Gardner AF, Ausubel FM. Overview of Next-Generation Sequencing Technologies. Curr Protoc Mol Biol. 2018; 122(1): e59.

[16] Zhou X, Ren L, Meng Q, Li Y, Yu Y, Yu J. The next-generation sequencing technology and application. Protein Cell. 2010; 1(6): 520-36.

[17] Ngamniyom A, Sriyapai T, Duangjai W, Sriyapai P. Report on microbial communities with gene functions and distribution of elements in Echinomuricea (Anthozoa: Holaxonia) from Thailand. Agric Na Resour. 2020; 54(6): 657-64.

[18] Ngamniyom A, Sriyapai T, Sriyapai P, Panyarachun B. Diversity of gut microbes in freshwater and brackish water ricefish (Oryzias minutillus and O. javanicus) from Southern Thailand. Agric Na Resour. 2021; 55(2): 311-18.

[19] Wei H, Li X, Tang L, Yao H, Ren Z, Wang C, Mu C, Shi C, Wang H. 16S rRNA gene sequencing reveals the relationship between gut microbiota and ovarian development in the swimming crab Portunus trituberculatus. Chemosphere. 2020; 254: 126891.

[20] Raja HA, Miller AN, Pearce CJ, Oberlies NH. Fungal Identification Using Molecular Tools: A Primer for the Natural Products Research Community. J Nat Prod. 2017; 80(3): 756-70.

[21] Schoch CL, Seifert KA, Huhndorf S, et al. Nuclear ribosomal internal transcribed spacer (ITS) region as a universal DNA barcode marker for Fungi. Proc Natl Acad Sci USA. 2012; 109(16): 6241-46.

[22] de Souza Valente C. Anaesthesia of decapod crustaceans. Vet Anim Sci. 2022; 14: 100252.

[23] Su X, Sun Y, Liu D, Wang F, Liu J, Zhu B. Agonistic behaviour and energy metabolism of bold and shy swimming crabs Portunus trituberculatus. J Exp Biol. 2019; 222: jeb188706.

[24] Srimahachota T, Yokota H, Akira Y. Recycled Nylon Fiber from Waste Fishing Nets as Reinforcement in Polymer Cement Mortar for the Repair of Corroded RC Beams. Materials (Basel). 2020; 13(19): 4276.

[25] Yin J, Li JY, Craig NJ, Su L. Microplastic pollution in wild populations of decapod crustaceans: A review. Chemosphere. 2022; 291(Pt 2): 132985.

[26] D'Costa AH. Microplastics in decapod crustaceans: Accumulation, toxicity and impacts, a review. Sci Total Environ. 2022; 832: 154963.

[27] Liu L, Xu M, Ye Y, Zhang B. On the degradation of (micro)plastics: Degradation methods, influencing factors, environmental impacts. Sci Total Environ. 2022; 806(Pt3): 151312.

[28] Usman S, Razis AFA, Shaari K, Azmai MNA, Saad MZ, Isa NM, Nazarudin MF. Polystyrene microplastics induce gut microbiome and metabolome changes in Javanese medaka fish (Oryzias javanicus Bleeker, 1854). Toxicol. 2022; 9: 1369-79.

[29] Baker-Austin C, Oliver JD, Alam M, Ali A, Waldor MK, Qadri F, Martinez- Urtaza J. Vibrio spp. infections. Nat Rev Dis Primers. 2018; 4(1): 8.

[30] Raddadi N, Giacomucci L, Totaro G, Fava F. Marinobacter sp. from marine sediments produce highly stable surface-active agents for combatting marine oil spills. Microb Cell Fact. 2017; 16(1): 186.

[31] Gupta S, Lokesh J, Abdelhafiz Y, Siriyappagouder P, Pierre R, Sørensen M, Fernandes JMO, Kiron V. Macroalga-Derived Alginate Oligosaccharide Alters Intestinal Bacteria of Atlantic Salmon. Front Microbiol. 2019; 10: 2037.

[32] Walter JM, Bagi A, Pampanin DM. Insights into the Potential of the Atlantic Cod Gut Microbiome as Biomarker of Oil Contamination in the Marine Environment. Microorganisms. 2019; 7(7): 209.

[33] Shaumi A, Cheng UC, Guo SY, Jones EB, Chan TY, Pang KL. Diversity of fungi isolated from carapace and gut of the marine crab Portunus sanguinolentus in northern waters of Taiwan. Bot. Mar 2023; 66(4): 301-7.

[34] Xu S, Wang X, Nageen Y, Pecoraro L. Analysis of gut-associated fungi from Chinese mitten crab Eriocheir sinensis. All Life. 2021; 14(1): 610-21.

[35] Nguyen NH, Song Z, Bates ST, Branco, S, Tedersoo L, Menke J, Schilling JS, Kennedy PG. FUNGuild: an open annotation tool for parsing fungal community datasets by ecological guild. Fungal Ecol. 2016; 20: 241-8.

[36] Põlme S, Abarenkov K, Henrik Nilsson R. et al. FungalTraits: a user-friendly traits database of fungi and fungus-like stramenopiles. Fungal Divers. 2020; 105: 1-16.