عدد خاص
بالمؤتمر الليبي الدولي للعلوم التطبيقية
و الهندسية
27-28- سبتمبر 2022

Microplastic contamination in Gills and Gastrointestinal Tract of Fish Collected from the Tobruk Coast, Eastern Libya

Amani Fitori*[1] , Ibrahim A. Ishag[2] , Ashraf Naji Masoud[3], Aboubakar M. Mahjoub[1], Hamid I. Hamid[1], Haneen M. Hashem[1], Rawan J. Al-zaroog[1], Meran O. Abd Alhafeed[1]

1. Marine Resources Department, Natural Resources Faculty, Tobruk University, Libya.

2 Department of Zoology, Faculty of Science, Tobruk University, Libya.

3.Environmental Science Department, Natural Resource Faculty, Tobruk University, Libya.

Email:amanifitori1@gmail.com, abutuga2006@hotmail.com

الملخص

تدخل اللدائن (البلاستيك) الدقيقة جسم الأسماك عبر الفم والخياشيم. ويسبب التلوث بالدائن الدقيقة العديد من المخاطر والعواقب المتنوعة في جسم الأسماك مثل زيادة مستوى السمية وتوقف النمو. أجري هذا البحث بهدف دراسة اللدائن الدقيقة في أجسام الأسماك المجمعة من حوض خليج طبرق. جمع عدد61 عينة من الأسماك. واستخدمت صناديق الثلج لحفظ ونقل العينات من المصائد إلى المختبر. فحصت الخياشيم والجهاز الهضمي لتحديد شكل وحجم اللدائن الدقيقة في الأسماك المجمعة. حيث فصلتا الخياشيم والجهاز الهضمي عن أجسام الأسماك، واستخدم المجهر الضوئي فيفحص اللدائن الدقيقة لتحديد الشكل والحجم. أظهرت نتائج الدراسة فحص بأن جميع الأسماك المدروسة (100.0٪) ملوثة باللدائن الدقيقة في الخياشيم والجهاز الهضمي معا. كشفت نتائج اختبار مربع كاي عن عدم وجود ارتباط معنوي بين حجم اللدائن الدقيقة الموجودة في الخياشيم أو الجهاز الهضمي مع أنواع الأسماك المدروسة، وكذلك كشفت نتائج الدراسة عن عدم وجود ارتباط معنوي بين حجم اللدائن الدقيقة في الخياشيم أو الجهاز الهضمي مع حجم الأسماك المدروسة. توصي الدراسة بإجراء المزيد من البحوث حول التلوث باللدائن الدقيقة في الكائنات البحرية والأسماك.

الكلمات المفتاحية: اللدائن الدقيقة، الجهاز الهضمي، خياشيم، أسماك، حوض خليج طبرق.

Abstract

The gills and mouth of the fish allow microplastics to enter the body. Microplastic contamination of the fish body can have a variety of consequences, including increasing toxicity and growth suppression. This research was conducted to study microplastics in fish collected from Tobruk Bay basin. A total of 61 fish samples were caught and collected. An icebox was used to keep and transport the samples to the laboratory. The form and size of microplastics were discovered and examined in each gill and gastrointestinal tract of the collected fish samples. Samples' gills and gastrointestinal tracts are separated from their bodies. Microplastics were observed under the light microscope. The results showed that the investigation of the gills and digestive gut revealed that all studied fishes (100.0%) were contaminated by microplastics. The chi-square test revealed the insignificant association between the size of microplastics found in fish gill and digestive tract with fish species. Also, the results showed an insignificant association between the size of microplastics in fish gill and gut with fish size. This study recommends further research on microplastic contamination in marine organisms and fish

Keywords: Microplastic, Gastrointestinal tract, Gills, fish, Tobruk Bay basin

1. Introduction

Marine plastic garbage entered the water as a result of deliberate or unintentional activity on land. Microplastics (MPs) contamination is a concern to the marine ecosystem from the tropics to the poles[1]. This pollution has a significant impact on the Mediterranean Sea, which is a semi-closed basin[2]. MPs can be found floating on the surface, in the water column, in sediments, and all the way down to the deep sea[3]. MPs can be consumed by aquatic biota and can serve as a carrier for other pollutants such as persistent organic pollutants (POPs), which can be absorbed and concentrated from the surrounding saltwater. As a result, MPs may be able to transport a variety of harmful substances. Pollutant transmission across the food chain[4]. Based on their morphology, MPs can be classified into four groups. Fragments, fiber, film, and granules are all present[5]. MPs produced by the breakdown of plastic debris can have a wide range of negative effects on numerous marine species disturbances in the biota, such as feeding disruptions and reproductive disruptions impairment, altered metabolism, and interaction with other pollutants[6].Tobruk bay basin is in the south-east of Tobruk city, which is located in northeast Libya. Some economic activity, such as Turkish port (for fishing boats), commercial port (for commerce ships, fishing and rescue boats), and the Cornice resort (for entertainment), are centered on both sides of the bay. Direct sewage discharge is a problem in the Bay basin[7].Not only does plastic trash come from plastic products like bags and bottles, but numerous synthetic components from fishing nets are also sources of microplastics in the marine environment[8]. MPs can clog the digestive tracts as well as hinder digestive processes and affect nutrition absorption[9, 10]. There are increased concerns about the number of microplastic particles in commercial fish gills and gastrointestinal tracts around the world, as well as their impacts. However, no research has been done on the amount of microplastic found in fish gills and gastrointestinal tracts off the coast of Tobruk. This study aimed to provide information about the amount and shapes of microplastics found inside the gills and gastrointestinal tract of some fish that is collected from Tobruk coast. The form and amount of MPs discovered in each gill and gastrointestinal tract of the fish samples were investigated in this study.

2. Materials and Methods

2.1 Study area and sampling location

Tobruk Bay basin (Fig.1) lies in the south-east of Tobruk city, which is located in northeast Libya at (longitude 23.59 and 13.06 E and latitudes 32.04 and 09.46 N). Some economic activities, such as Turkish port (for fishing boats), commercial port (for commerce ships, fishing and rescue boats), and the Cornice resort (for entertainment), are centered on both sides of the bay. Direct sewage discharge is a problem in the Bay basin. A total of 61 fish samples belonging to different species (more frequent in the region) were collected from various locations around Tobruk Bay basin*(Sparus aurata*,*Sphyraena chrysotaenia*,*Mugil cephalus,Epinephelus marginatus*,*Seriola fasciata*,*Oblada melanura*), during the period of April and May. 2022 . An icebox was used to keep and transport the samples to the laboratories of Natural Resources Faculty, Tobruk University. Gills and gastrointestinal tracts were separated from their bodies. Microplastic form and size were investigated and observed under the light microscope (Fig. 2).

2.2 Statistical Analysis

The data were analyzed using Statistical Package for Social Sciences (SPSS Version 23, IBMCorp., Armonk, NY). The data were analyzed in terms of frequency. Also the Pearson's chi-square test for contingency tables with Yates' continuity correction were used for comparisons between categorical variables to determine whether there were associations between (i) the frequency of microplastic size in gills or digestive tract with fish species, and (ii) the frequency of microplastic size in fish gill or digestive gut with fish size. The results were presented as tabular frequency.

Figure. 1.Tobruk Bay basin

3. Results

3.1 Microplastics in fish gills and digestive gut:

The results of Table 1 revealed that all investigated fishes (100.0%) were contaminated by microplastics. Moreover; the results showed the majority of fish gills (86.9%) were contaminated by microplastics with size <100 μm, while only 13.1% of fish gills were contaminated by microplastics with size ≥ 100 μm.On the other hand, the results in Table 2explored the investigation of the digestive gut and showed that all studied fishes (100.0%) were contaminated by microplastics. Most of the fishes (98.4%) were contaminated by microplastics with a size ≥100 µm; while only 1.6% were contaminated by microplastics with a size < 100 µm.

Table 1. Microplastic contamination and its size in fish gills

Size of microplastics	N(number of samples)	Percentage (%)
< 100 µm	53	86.9
≥ 100 µm	8	13.1
Total	61	100.0

Table 2. Microplastic contamination and its size in fish digestive gut

Size of microplastics	N (number of samples)	Percentage (%)
< 100 µm	1	1.6
≥ 100 µm	60	98.6
Total	61	100.0

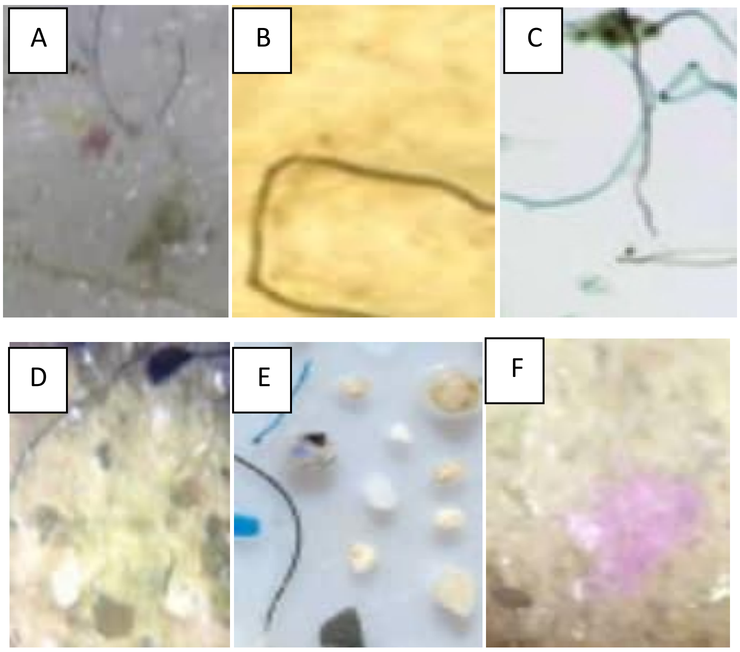

Figure.2 Microplastic forms in gills (A= Fragment, B= Fiber, C= Film) and in digestive gut (D= Fragment, E= Fiber, F= Film)

The results in Table 3 explained that the highest percentage of microplastic type or form (Fig. 2) in gills was found as fragment form(32.8%), then film form(13.1%), while the other forms and types were recorded with the lowest percentages. However, the results in Table 4revealed that the fragment form recorded the greatest frequency (60.7%) as microplastics form in the digestive gut of the studied fishes, followed by together fragment and fiber from with the percentage of 16.4%, then fiber form(9.8%). While the other forms such as rope, pellet and film, pellet, film, and fragments recorded the lowest percentage (1.6%).

Table 3. Form of microplastics which found in fish gill

Form of microplastics	N (number of samples)	percentage (%)
Fiber	2	3.3
Fiber and film	1	1.6
Film	8	13.1
Film and fiber	4	6.6
Film and fragment	4	6.6
Film and rope	1	1.6
Fragment	20	32.8
Fragment and fiber	4	6.6
Pellet	4	6.6
Pellet and film	4	6.6
Rope	4	6.6
Rope and fragment	5	8.2
Total	61	100.0

Table 4. Form of microplastics in the gut of the studied fishes

From of microplastics	N (number of samples)	Percentage (%)
Fiber	6	9.8
Fragment	37	60.7
Fragment and fiber	10	16.4
Fragment and film	3	4.9
Fragment and rope	2	3.3
Pellet and film	1	1.6
Pellet, film and fragment	1	1.6
Rope	1	1.6
Total	61	100

3.2The associations of microplastic size with fish species and size

The Pearson chi-square test in Table 5 showed an insignificant association between the size of microplastics found in fish gills and the size of fish (χ^2=0.002; P>0.05). Moreover, the Pearson chi-square test in Table 6revealed an insignificant association between the size of microplastics found in fish gill and fish species (χ^2=3.0; P>0.05).The results of the Pearson chi-square test showed an insignificant association between the size of microplastics found in the digestive gut and size of fish (χ^2=0.98; P>0.05) (Table 7). Moreover, the Pearson chi-square test in Table 8 revealed an insignificant association between the size of microplastics and fish species (χ^2=5.18, P>0.05).

Table5. The association between the size of microplastics found in fish gill and the size of fish

Size of fish	Size of microplastic			
	< 100 µm		≥ 100 µm	
	N	%	N	%
≤ 30 cm	26	86.7	4	13.3
> 30 cm	27	87.1	4	12.9
Total	53	86.9	8	13.1
Chi-square test	χ^2=0.002; P>0.05			

Table 6. The association between the microplastic size found in gills and fish species

Fish species	Size of microplastics			
	< 100 µm		≥ 100 µm	
	N	%	N	%
Sparus aurata	8	80.0	2	20.0
Mugil cephalus	8	80.0	2	20.0
Epinephelus marginatus	9	90.0	1	10.0
greater amberjack	8	80.0	2	20.0
Sphyraena chrysotaenia	10	100.0	0	0.0
Oblad melanura	10	90.9	1	9.1
Total	53	86.9	8	13.1
Chi-square test	χ^2=3.0; P>0.05			

Table 7. The association between the size of microplastics found in the digestive gut and the size of fish

Size of fish	Size of microplastics			
	< 100 µm		≥ 100 µm	
	N	%	N	%
≤ 30 cm	0	0.0	30	100.0
> 30 cm	1	3.2	30	96.8
Total	1	1.6	60	98.4
Chi-square test	χ^2=0.98; P>0.05			

Table 8. The association betweenthe microplastic size in gut and fish species

Fish species	Size of microplastics			
	< 100 µm		≥ 100 µm	
	N	%	N	%
Sparus aurata	**1**	**10.0**	**9**	**90.0**
Mugil cephalus	**0**	**0.0**	**10**	**100.0**
Epinephelus marginatus	**0**	**0.0**	**10**	**100.0**
greater amberjack	**0**	**0.0**	**10**	**100.0**
Sphyraena chrysotaenia	**0**	**0.0**	**10**	**100.0**
Oblad melanura	**0**	**0.0**	**11**	**100.0**
Total	**1**	**1.6**	**60**	**98.4**
Chi-square test	**χ^2=5.18, P>0.05**			

4. Discussion

Microplastic can be mistakenly consumed by marine biota, such as fish. Microplastics can be consumed in two ways: directly and indirectly. Plastic particles are directly swallowed by fish when they are unable to distinguish between their prey and plastic. When the fish ingested plastic that was already polluted inside their prey's body or adhered to their prey's body, this is known as indirect contamination[11]. The following microplastic forms such as fragment, fiber, film, pellet, and rope were detected in the gills and gastrointestinal tracts of the collected fish from Tobruk Bay basin. The overall results in Tables 1 and 2revealedthat microplastics infected all the investigated fishes (100.0%). Microplastic contamination in the gastrointestinal tract of commercial fish inTobruk Bay basin can be significantly connected tothe depth and the fish habitat where they feed[12]. Fish intake of microplastics and the presence of microplastic particles in seawater had no effect on fish-eating behavior. The size of the microplastic had an impact on the amount of microplastic discovered in the gastrointestinal system. The smaller the microplastic, the more the fish swallow it. The fish can distinguish and avoid certain sizes of microplastic[13].A large amount of microplastic particles are accumulating inside the fish body, on the other hand, can clog the digestive tract, disrupt digestion, and hinder absorption processes[14]. Microplastic particles in the digestive tract can potentially alter a fish's appetite or transport chemicals[15].The results in Tables 3 and 4 explained that the fragment(32.8%) is the most common form of microplastic found in fish gills, followed by the film form(13.6%) in gills. Also, the fragment form recorded the highest frequency (60.7%) of microplastic form in the digestive guts of the examined fishes, followed by combined fragment and fiber forms (16.4%), and then fiber form(9.8%). The high frequency of fragment was also reported by [16] and they explained that the microplastics in the gill and gastrointestinal tract of canting groupers are mostly found in the form of fragments. The lowest frequencies were found in rope, pellet, and film forms. The fragmentedform of microplastics is derived through the decomposition of plastic trash through photolysis or biodegradation mechanisms[17].Microplastic fibers are made from waste from the textile industry or various synthetic materials found in fishing rods and nets[5].The chi-square test revealed an insignificant relationship between the size of microplastics discovered in fish gills or digestive gut and the size of the fish. Furthermore, the chi-square test demonstrated an insignificant relationship between the size of microplastics discovered in fish gills or digestive gut and fish species. Microplastic concentrations in the gastrointestinal tract of hardhead catfish rose dramatically with increasing body length, according to prior research. In comparison to the sole published study, and found increased quantities of microplastics in the stomachs of southern flounders [18]. Because microplastics may emerge indirectly from their prey or accidently from the water column, an ambush feeding behavior may contribute to the apparently random nature of microplastics of various sizes. In addition to the foraging guild, at least one study found that greater tropic levels have higher levels of microplastics[19].Microplastic burdens in the environment and in fish may vary throughout time scales, and evidence of seasonal changes has already been discovered[20].Size limitations, one of the most frequent fisheries management techniques, are based on length. The needing to start addressing characteristics within species, such as length, that are connected to microplastic burdens, and how such aspects may be considered for natural and managed populations given that we know most fish species ingest microplastics[21]. This study recommends further research on microplastic contamination and ingestion in marine organisms and fish.

5. References

[1] Waller, C. L., Griffiths, H. J., Waluda, C. M., Thorpe, S. E., Loaiza, I., Moreno, B., et al.. Microplastics in the Antarctic marine system: an emerging area of research. Sci. Total Environ.(2017), 598, 220–227. doi: 10.1016/j.scitotenv.2017.0 3.283.
[2] Llorca, M., Alvarez-Munoz, D., Abalos, M., Rodrìguez-Mozaz, S., Santos, L., Leon, V. M., et al. Microplastics in Mediterranean coastal area: toxicity and impact for theenvironment and human health. Trends Environ. Anal. Chem. (2020). 27:e00090. doi: 10.1016/j.teac.2020.e00090.
[3] Van Cawenberghe, L., Vanreusel, A., Mees, J., and Janssen, C. R. Microplastic pollution in deep-sea sediments. Environ. Pollut. 182, (2013), 495–499. doi: 10.1016/j.envpol.2013.08.013.
[4] Fytianos, G., Ioannidou, E., Thysiadou, A., Mitropoulos, A. C., and Kyzas, G. Z. Microplastics in mediterranean coastal countries: a recent overview. J. Mar. Sci. Eng. 9:98. (2020),doi: 10.3390/jmse9010098.
[5] Ding, J.;Li, J.; Sun,C.; Jiang,F.; Ju,P.; Qu,L.; Zheng,Y.;& He, C.. Detection of microplastics in local marine organisms using a multi-technology system. Anal. Methods. 11 (2019) 78–87.
[6] Anbumani, S.& Kakkar, P. Ecotoxicological effects of microplastics on biota: a review. Environ. Sci. Pollut. Res. 25 (2018) 14373–14396.
[7] Fitori, A.A.; Ishag, I.A.; Al-Shobaki, K.F.; Balal1, D.M.; Jaballah, A.; Khaled, Souad A. and Alkhawaja, H. Microbial contamination in the Tobruk Bay basin. International Journal of Multidisciplinary Sciences and Advanced Technology Special Issue 1(2021) 663–667.
[8] Dabrowska,A.; Lopata,I.& Osial, M.. The ghost nets phenomena from the chemical perspective. Pure. Appl. Chem. 93 (2021) 479–496.

[9] Yudhantari, C. I. S.; Hendrawan, I.G.L. &. Puspita, N.L.P.R.. Kandungan mikroplastik pada saluran pencernaan ikan lemuru protolan (Sardinella lemuru) hasil tangkapam di Selat Bali. JMRT 2 (2019) 47–51.
[10] Zhang,F.; Man,Y.B.; Mo,W.Y.; Man, K.Y. & Wong,M. H.. Direct and indirect effects of microplastics on bivalves, with a focus on edible species: A mini-review. Critical reviews in environmental science and technology (2019) 1– 35.
[11] Cole, M.; Lindeque,P.; Fileman,E.; Halsband,C.; Goodhead,R.; Moger, J.& Galloway, T. S.. Microplastic ingestion by zooplankton. Environ. Sci. Technol. 47 (2013) 6646-6655.
[12] Merle,.M. B. R.; Edgar, F. M. F.; Griselda,E. S.& Jaime, R. O.. Plastic density as a key factor in the presence of microplastic in the gastrointestinal tract of commercial fishes from Campeche Bay Mexico. Environmental Pollution. 267 (2020).
[13] Critchell,K. & Hoogenboom, M. O.. Effects of microplastic exposure on the body condition and behaviour of planktivorous reef fish (Acanthochromis polyacanthus). PLoS ONE. 13 (2018).
[14] Wright,S.L.; Thompson,R.C.& Galloway, T.S.. The physical impacts of microplastics on marine organisms: a review. Environmental pollution 178 (2013) 483-492.
[15] 15. Hirai, H.; Takada, H.; Ogata, Y.; Yamashita, R. et al.. Organic micropollutants in marine plastics debris from the open ocean and remote and urban beaches. Marine pollution bulletin 62(8) (2011) 1682-1683.
[16] Afra, N. and Mufti, P. P. Microplastics abundance in gills and gastrointestinal tract of Epinephelus fuscoguttatus-lanceolatus at the Coastal of Pulau Panjang, Serang, Banten. E3S Web of Conferences 324, 01002 (2021). https://doi.org/10.1051/e3sconf/20213240100.
[17] Reisser,J.; Shaw, J.; Wilcox, C.; Hardesty,B. D.;Proietti, M.;Thums, M.& Pattiaratchi, C.. Marine plastic pollution in waters around Australia: Characteristics, concentrations, and pathway. PLoS ONE. (2013).
[18] Phillips, M.B.; Bonner, T.. Occurrence and amount of microplastic ingested by fishes in watersheds of the Gulf of Mexico. Mar. Pollut. Bull. 2015, 100, 264–269.
[19] Zhang, F.; Wang, X.; Xu, J.; Zhu, L.; Peng, G.; Xu, P.; Li, D.. Food-web transfer of microplastics between wild caught fish and crustaceans in East China Sea.Mar. Pollut. Bull. 2019, 146, 173–182.
[20] Ferreira, G.V.; Barletta, M.; Lima, A.R.A.. Use of estuarine resources by top predator fishes. How do ecological patterns affect rates of contamination by microplastics? Sci. Total Environ. 2018, 655, 292–304.
[21] Gad, A.K.; Midway, S.R.. Relationship of Microplastics to Body Size for Two Estuarine Fishes. Microplastics. (2022), 1, 211–220. https://doi.org/10.3390/ microplastics1010014.