

Little evidence for bioaccumulation or biomagnification of microplastics in a deep-sea food web

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Text S1: Nitrogen stable isotope data acquisition for Trophic level estimates.

Trophic levels for collected taxa were estimated from bulk nitrogen isotope compositions using Bayesian methods detailed in Quezada-Romegialli et al, (2018). *Euphausia* sp. (krill) collected from Monterey Bay was utilized as the food web baseline, along with a trophic discrimination factor of 3.4 after Post, (2002). For each taxon, a minimum of five specimens were utilized for stable isotope measurements. White muscle tissue was dissected and all skin and bones removed prior to freeze drying and homogenization. Bulk sample nitrogen isotope compositions were determined using an isotope ratio mass spectrometer (IRMS; Delta PlusXP) coupled to an elemental analyzer (Conflo IV/Costech ECS 4010) at the University of Hawaii's Stable Isotope Biogeochemistry Lab as in Choy et al. (2015).

Table S1: Information on species sampling dates, collection method and species trophic level.

Species	Common name	Dates collected	Collection method	Trophic level	Taxa
<i>Pleuroncodes planipes</i>	Tuna crab	July 2016	Pier dive	2	crustacean
<i>Doryteuthis opalescens</i>	Market squid	July 2019	Purse seining	3	cephalopod
<i>Stenobranchius leucopsarus</i>	Northern lampfish	2016	Midwater Trawl	3.5	fish
<i>Scomber japonicus</i>	Chub mackerel	July 2019	Bycatch with salmon trolling	3.5	fish
<i>Paralichthys californicus</i>	California halibut	Aug 2019	Trolling/jigging with rod and reel	3.5+	fish
<i>Oncorhynchus tshawytscha</i>	King salmon	Aug 2019	Trolling/jigging with rod and reel	3.5+	fish

Table S2: Habitat, migration behaviour (DVM = diel vertical migration), diet type, and primary feeding strategy of each species from Monterey Bay, California who had their gut content analysed for anthropogenic particles. Epipelagic is defined as 0–200 m, mesopelagic is defined as 200–1000 m.

Species	Habitat	DVM?	Diet type	Feeding strategy (primary)
Tuna crab (<i>Pleuroncodes planipes</i>)	Epipelagic/Upper mesopelagic	No	Zoo-/Herbivore	Filter feeder
Northern lampfish (<i>Stenobrachius leucopsarus</i>)	Mesopelagic	Yes	Zooplanktivore	Active Predator
Market squid (<i>Doryteuthis opalescens</i>)	Epipelagic/Upper mesopelagic	Yes	Zooplanktivore/Piscivore	Active Predator
Chub mackerel (<i>Scomber japonicus</i>)	Epipelagic/Upper mesopelagic	No	Zooplanktivore/Piscivore	Active Predator
California halibut (<i>Paralichthys californicus</i>)	Benthic	No	Piscivore	Ambush Predator
King salmon (<i>Oncorhynchus tshawytscha</i>)	Epipelagic	No	Piscivore	Active Predator

Table S3: Samples list analyzed per sequence on the Py-GC/MS.

Sequence #	Samples analyzed
A	Northern lampfish 01 to 08 (SL-01 to SL-08) Halibut 01 to 02 (HS-01 & HS-02)
B	Halibut 03 to 07 (HS-03 to HS-08) King salmon 01 to 05 (OT-01 to OT-05)
C	King salmon 06 to 06 (OT-06 to OT-08) Market squid 01 to 08 (DO-01 to DO-08)
D	Chub mackerel 01 to 08 (SJ-01 to SJ-08) Tuna crab 01 to 02 (PP-01 to PP-02)
E	Tuna crab 03 to 08 (PP-01 to PP-08)

Table S4: Calibration curve parameters ($y = ax + b$) for all sequence done for the Pyrolysis-GC/MS work.

For each calibration, 7 levels with concentration from 0.5 to 20 μg of polymer were analyzed.

Polymer	PE	PP	PS	PMMA	PVC	PC
Sequence A						
b	0.0166	0.1096	-0.0241	0.5457	0.0933	0.1629
a	0.0327	0.1982	0.3312	0.6267	0.035	0.1879
R²	0.9842	0.9943	0.9982	0.9312	0.9925	0.9796
Sequence B						
b	-0.0223	0.0004	-0.0747	0.3316	0.0422	-0.003
a	0.0393	0.2539	0.3392	0.6876	0.0455	0.2403
R²	0.9988	0.9981	0.9978	0.9689	0.9948	0.9802
Sequence C						
b	-0.0808	-0.0009	-0.2739	0.1360	0.0972	0.07
a	0.0926	0.1665	0.8336	0.2376	0.0841	0.5926
R²	0.9623	0.9964	0.9936	0.9936	0.9745	0.9904
Sequence D						
b	-0.0261	-0.0219	-0.018	0.1456	0.0206	-0.0631
a	0.0298	0.2333	0.2341	0.8343	0.0437	0.1147
R²	0.9767	0.9971	0.9783	0.9942	0.997	0.8494
Sequence E						
b	-0.0373	-0.015	-0.0661	0.4306	0.0508	-0.0096
a	0.0333	0.2586	0.3	0.6473	0.0433	0.0131
R²	0.9722	0.9762	0.9955	0.9717	0.9883	0.9325

Table S5: Plastic polymer recoveries (in %) obtained after MAE extraction and Py-GC/MS analysis from shrimp (expressed as %) Polyethylene (PE), polypropylene (PP), polystyrene (PS), poly(methyl-methacrylate (PMMA), polyvinylchloride (PVC) polyethylene terephthalate (PET) and polycarbonate (PC).

Polymer	Characteristic peak	Recoveries (%)				
		Rep1	Rep2	Rep3	Mean	SD
PE	1-Decene (C ₁₀)	89.48	75.20	98.75	87.81	11.86
	1-Heptadecene (C ₁₇)	100.28	69.13	81.22	83.55	15.71
PP	2,4-dimethyl-1-heptene	70.05	25.39	71.95	55.80	26.35
PS	Styrene trimer	84.66	74.26	86.39	81.77	6.56
PMMA	Methyl-methacrylate	96.13	49.51	0.00	48.55	48.07
PVC	Indene	120.49	95.81	95.50	103.93	14.34
PC	4-isopropenylphenol	68.30	47.63	51.99	55.97	10.90

Table S6: Suspected microplastic (MP) particles found in GI tracts and their classification depending on their morphology and their occurrence in all individuals for each species.

Species	Total number of suspected MPs	Number of suspected particles found (min.–max.)	Number of fibers (%)	Number of fragments (%)	Number of film (%)	Number of foam (%)	Number of Rubber (%)
Tuna crab (<i>Pleuroncodes planipes</i>)	111	2–21	103 (92.8)	7 (6.3)	1 (0.9)	0	0
California halibut (<i>Paralichthys californicus</i>)	222	7–79	219 (98.6)	2 (0.9)	0	1 (0.5)	0
Market squid (<i>Doryteuthis opalescens</i>)	13	0–6	13 (100)	0	0	0	0
Chub mackerel (<i>Scomber japonicus</i>)	84	0–38	82 (97.6)	2 (2.4)	0	0	0
Northern lampfish (<i>Stenobrachius leucopsarus</i>)	97	4–22	88 (90.7)	7 (7.2)	1 (1)	0	1 (1)
King salmon (<i>Oncorhynchus tshawytscha</i>)	145	4–40	141 (97.2)	1 (0.7)	1 (0.7)	0	2 (1.4)
Total	672		646 (96.13)	19 (2.82)	3 (0.45)	1 (0.15)	3 (0.45)

Table S7: Spectroscopy corrected number of microplastics (MP) found in species GI tracts.

Species	Spectroscopy corrected number of MP found per individual	
	(Min.–Max.)	(Mean ± S.D)
Tuna crab (<i>Pleuroncodes planipes</i>)	0.23–2.69	1.62 (±0.94)
California halibut (<i>Paralichthys californicus</i>)	0.47–9.24	3.71 (±3.47)
Market squid (<i>Doryteuthis opalescens</i>)	0–0.23	0.19 (±0.23)
Chub mackerel (<i>Scomber japonicus</i>)	0–4.45	1.23 (±1.47)
Northern lampfish (<i>Stenobranchius leucopsarus</i>)	0.47–2.57	1.42 (±0.78)
King salmon (<i>Oncorhynchus tshawytscha</i>)	0.35–4.68	2.12 (±1.41)

Table S8: Number of suspected microplastic particles according to color found in the GI tracts of sampled species.

Species	Blue	Black	Grey	Clear	Red	Brown	Pink	White	Purple	Orange	Yellow	Green
Tuna crab (<i>Pleuroncodes planipes</i>)	48	4	7	30	4	2	1	0	6	5	4	0
California halibut (<i>Paralichthys californicus</i>)	129	26	16	18	13	0	4	1	4	6	4	1
Market squid (<i>Doryteuthis opalescens</i>)	1	0	0	11	1	0	0	0	0	0	0	0
Chub mackerel (<i>Scomber japonicus</i>)	3	1	0	77	3	0	0	0	0	0	0	0
Northern lampfish (<i>Stenobranchius leucopsarus</i>)	30	9	2	25	5	3	2	1	5	5	10	0
King salmon (<i>Oncorhynchus tshawytscha</i>)	103	19	8	6	4	2	1	1	1	0	0	0
Total	314	59	33	167	30	7	8	3	16	16	18	1

Table S9: Number of suspected microplastic (MP) particles found in species GI tracts and their associated lengths and widths.

Species	Number of suspected MPs found per individual		Mean (\pm S.D) dimensions of suspected MPs (mm)	
	(Min – Max)	(Mean \pm S.D)	Length	Width
Tuna crab (<i>Pleuroncodes planipes</i>)	2 - 21	13.88 (\pm 8.04)	1.43 (\pm 1.17)	0.035 (\pm 0.071)
California halibut (<i>Paralichthys californicus</i>)	7 - 79	31.71 (\pm 29.66)	1.97 (\pm 1.65)	0.031 (\pm 0.068)
Market squid (<i>Doryteuthis opalescens</i>)	0 - 6	1.6 (\pm 2)	3.74 (\pm 4.29)	0.025 (\pm 0.009)
Chub mackerel (<i>Scomber japonicus</i>)	0 - 38	10.5 (\pm 12.56)	2.05 (\pm 1.63)	0.027 (\pm 0.030)
Northern lampfish (<i>Stenobrachius leucopsarus</i>)	4 - 22	12.13 (\pm 6.69)	1.90 (\pm 2.02)	0.033 (\pm 0.053)
King salmon (<i>Oncorhynchus tshawytscha</i>)	4 - 40	18.13 (\pm 12.05)	2.00 (\pm 1.82)	0.070 (\pm 0.357)

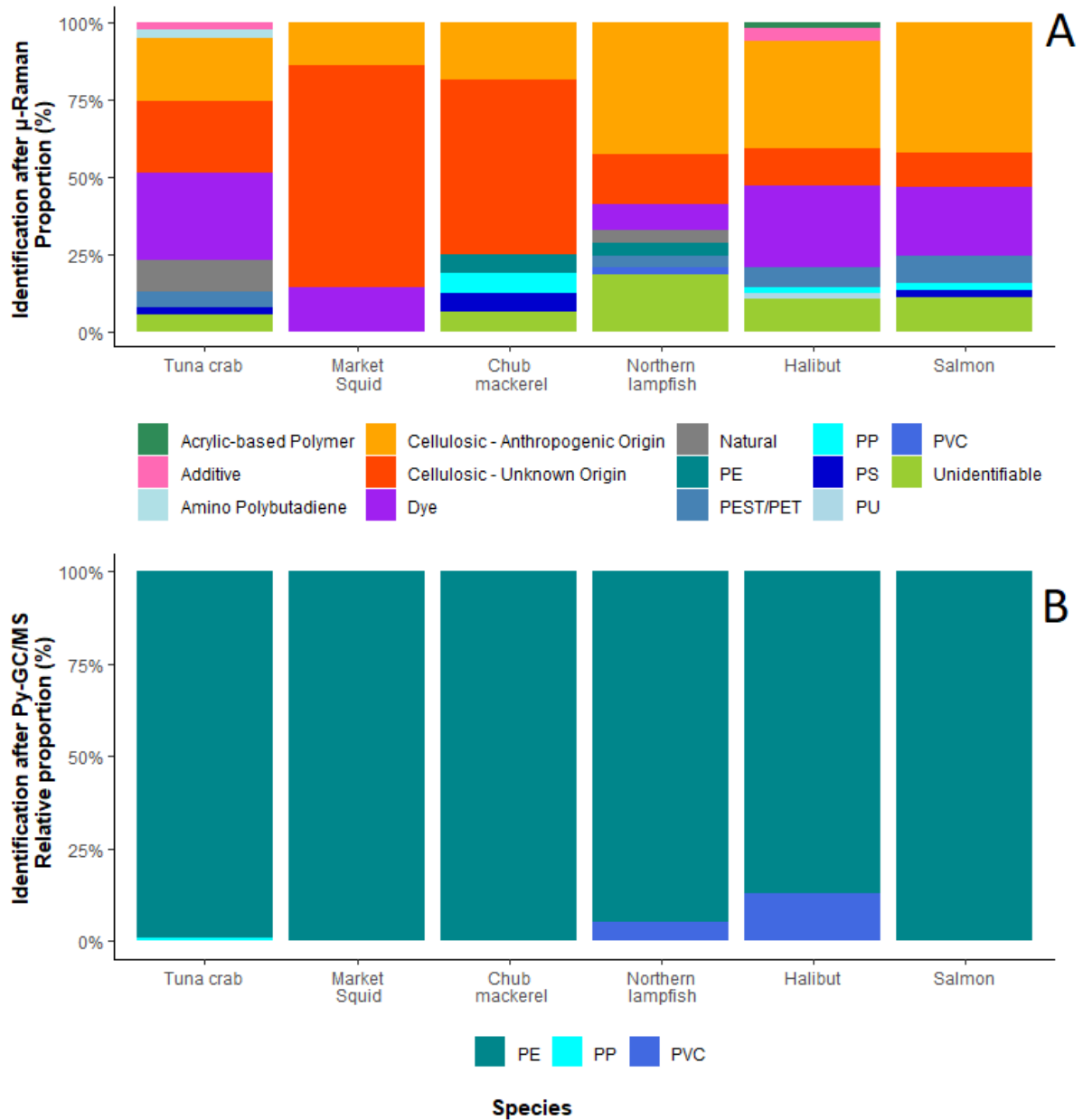
Table S10: Identification of suspected microplastic found in species GI tracts obtained after μ -Raman spectroscopy.

Species	Acrylic based Polymer	Additive	Amino Polybutadiene	Cellulosic – Anthropogenic origin	Cellulosic – Unknown origin	Dye	Natural	PE	PEST/PET	PP	PS	PU	PVC	Unidentified
Tuna crab <i>(Pleuroncodes planipes)</i>	0	1	1	8	9	11	4	0	2	0	1	0	0	2
California halibut <i>(Paralichthys californicus)</i>	1	2	0	17	6	13	0	0	3	1	0	1	0	5
Market squid <i>(Doryteuthis opalescens)</i>	0	0	0	1	4	0	0	0	0	0	0	0	0	1
Chub mackerel <i>(Scomber japonicus)</i>	0	0	0	3	9	0	0	1	0	1	1	0	0	1
Northern lampfish <i>(Stenobrachius leucopsarus)</i>	0	0	0	21	8	4	2	2	2	0	0	0	1	9
King salmon <i>(Oncorhynchus tshawytscha)</i>	0	0	0	19	5	10	0	0	4	1	1	0	0	5
Total	1	3	1	69	41	38	6	3	11	3	3	1	1	23

Table S7: Plastic polymer mass quantified in other tissues using Py-GC/MS.

Species	Mean mass of plastic polymer per individual ($\mu\text{g/g}$)		Plastic polymers quantified
	(Min. excl. zeros – Max.)	(Mean \pm S.D)	
Tuna crab (<i>Pleuroncodes planipes</i>)	0.022–61.40	21.52 (\pm 23.59)	PE, PP
California halibut (<i>Paralichthys californicus</i>)	0.011–87.76	19.32 (\pm 38.11)	PE, PP, PVC
Market squid (<i>Doryteuthis opalescens</i>)	0.0015–25.12	9.72 (\pm 11.28)	PE, PVC
Chub mackerel (<i>Scomber japonicus</i>)	0.009–11.12	5.89 (\pm 5.07)	PE, PP
Northern lampfish (<i>Stenobranchius leucopsarus</i>)	0.014–1009.11	713.71 (\pm 300.89)	PE, PVC
King salmon (<i>Oncorhynchus tshawytscha</i>)	54.21–1096.43	467.53 (\pm 378.16)	PE

Figure S1: Relative proportion of the material types of the subsampled particles measured using μ -Raman spectroscopy (A) and relative proportion of plastic types in samples analyzed with Pyrolysis-GC/MS (B)



References

- Choy, C.A., Popp, B.N., Hannides, C.C.S., Drazen, J.C., 2015. Trophic structure and food resources of epipelagic and mesopelagic fishes in the North Pacific Subtropical Gyre ecosystem inferred from nitrogen isotopic compositions. *Limnology and Oceanography* 60, 1156–1171. <https://doi.org/10.1002/lno.10085>
- Post, D.M., 2002. USING STABLE ISOTOPES TO ESTIMATE TROPHIC POSITION: MODELS, METHODS, AND ASSUMPTIONS. *Ecology* 83, 703–718. [https://doi.org/10.1890/0012-9658\(2002\)083\[0703:USITET\]2.0.CO;2](https://doi.org/10.1890/0012-9658(2002)083[0703:USITET]2.0.CO;2)
- Quezada-Romegialli, C., Jackson, A.L., Hayden, B., Kahilainen, K.K., Lopes, C., Harrod, C., 2018. tRophicPosition, an R package for the Bayesian estimation of trophic position from consumer stable isotope ratios. *Methods in Ecology and Evolution* 9, 1592–1599. <https://doi.org/10.1111/2041-210X.13009>