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15 August 2024

Dear MEEF Committee,

Re: Completion report MEEF2022009

Here I would like to submit our completion report for the period from 1st of July 2022 to 31st December 2023.

(i) Executive summary (1-2 pages)

Hong Kong is a bustling metropolis which went through intense anthropogenic coastal modification and pollution since the 1800s. Subtidal faunal turnovers had been recorded in the city waters during the 1970s – 1980s, but there is no information on baseline conditions and pre-1960s changes in benthic fauna composition in Hong Kong. To determine the natural baseline of the Hong Kong benthic mollusc community and its changes in distribution through time, we are performing Accelerator Mass Spectrometry (AMS) radiocarbon dating of dead shells of selected molluscan morphospecies from surface sediments around Hong Kong. Mollusc shells were collected by trawling from western, southern, and eastern Hong Kong waters. These shells were identified to the lowest taxonomic level possible. Environmental data showed high eutrophication in the Pearl River Delta in western Hong Kong and high heavy metal contamination in Tolo Harbour in eastern Hong Kong waters. Western, southern, and eastern Hong Kong waters had distinct species compositions. The eastern side of Hong Kong (Tolo Harbour) had low species diversity compared to the western (Lantau Island) and southern waters (south of Hong Kong Island), except for southeast waters, which had the lowest species diversity. A high abundance of shells from species considered opportunistic, such as the gastropods Murex trapa, Nassarius siquijorensis, Turritella bacillum, and the bivalve Placamen lamellatum, was found in all regions. Our radiocarbon dating analyses show that several species, such as Paphya euglypta, Pecten excavatus, P. lamellatum, Protapes gallus, P. lamellatum, and Timoclea micra, and snails in the family Naticidae have suffered populational declines or even local extinctions in areas around Hong Kong. Our environmental analyses show highly impacted Pearl River Delta and Tolo Harbour. The contrasting shell communities among Pearl River Delta, south of Hong Kong Island, and Tolo Harbour reflect the



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differences in water quality among these regions. Our results from the radiocarbon dating help picture baseline conditions of the subtidal fauna from Hong Kong waters and show that the eutrophication of the Pearl River Delta and the environmental degradation and pollution in the Tolo Harbour negatively affected the composition of local subtidal molluscan communities.

(ii) Project title and brief description of the Project.

Shells for understanding Lantau subtidal ecosystem history: Part 2. Hong Kong wide comparison

Whilst studies have shown a turnover and distributional shift in Hong Kong's subtidal macrofauna in 1970-1980s; there are no study records to document the changes in Hong Kong's subtidal ecosystem in the 1960s and before. It is likely, however, that substantial human-induced degradation of marine biota has been happening since early 20th Century or even before, but given the limited biological information available and the lack of comprehensive descriptions of historic changes, we are unable to determine what a predisturbance faunal distribution would look like. Hong Kong is a key place to understand the past marine ecosystem and biogeographical distribution because it is a major distributional boundary of key marine organisms. For example, Chinese White Dolphin mainly distributes in the northwestern side of Lantau and is much less common in the southeastern side. Finless porpoise and corals show opposite trends and found almost exclusively in the southeastern side. To determine the natural baseline of this marine ecosystem and how it differs from other regions in Hong Kong, we are performing Accelerator Mass Spectrometry (AMS) radiocarbon dating of dead shells of selected molluscan morphospecies from surface sediments around Hong Kong to piece together changes in their distributions with time. Age distributions of these shells will tell us about the natural baseline information of where they were distributed before human impacts, and the subsequent human-induced ecosystem/distributional changes. From this information we will illustrate and map the past distributional changes of key species in Hong Kong; illustrating the natural baseline and quantifying anthropogenic impacts on subtidal macrobenthic distributions.

(iii) Completed activities against the proposed Work Schedule;

We conducted morphospecies identification, selection, specimen inspection as well as size measurements. We also conducted analyses on size differences among the shells. 14C dating has been finished. We have done according to the proposed Work Schedule.



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Recruitment a senior research assistant: Done

Planning and species selection: Done

Specimen inspection: Done Size measurement: Done Radiocarbon dating: Done

Preparing the progress report: Done

Data analyses: Done as detailed below Section.

Paper writing: In progress. We are still developing the draft (on going version attached

FYI).

Preparing the final report: Done as detailed below Section.

We also prepared a poster to be put in SWIMS as attached. We are now asking a SWIMS officer to put there.

(iv) Results/descriptions on the completed activities with appropriate analysis, with the support of photos, videos, social media platform, etc., if any;

Shells were collected by trawling sampling performed at 6 regions in Hong Kong: southeastern (SL) and northwestern (NW) Lantau waters; Southwestern Hong Kong (Lamma Island, SW), Southeast Hong Kong (SE), Outer Tolo Harbour (EO) and Inner Tolo Harbour (EI) (Table 1, Figure 1). Two transects were sampled at each region. Sampling occurred twice at each transect, once in July and again in August 2018, except for one of the transects on NW Lantau, WT3, which was only sampled in August 2018. Sampling sites correspond those presented by (Tao, Lau, et al. 2020; Tao, Lui, et al. 2020). The trawler had both outriggers of 15 meters in length, towing ten nets of stretched mesh size 2 cm along the sea floor, each horizontally propped open by a metal bean of 2 m. For each transect, a trawl was conducted for 30 min at a speed of 5 to 7 km h⁻¹, surveying an area between 0.05 to 0.07 km². The obtained catch was sorted on board and the dead mollusk shells were transported to laboratory. The shells were cleaned and dried in a dry oven at 40°C. A total of 8347 shell were obtained. These shells were subsequently identified into 226 morphospecies, based on morphological characteristics and depictions on the books of Okutani (2017) and Zhang et al. (2016), and quantified. The shells had their length measured with a dial calliper (0.1 mm) and are being photographed. The



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photographs are being taken by a Nikon Z6II with a macro lens Nikkor MC 50mm f/2.8 mounted on a Cognisys Inc. Stackshot automated focus stacking rail, which is mounted in a copy stand (Figure 2).

We selected shells of representative mollusc species for radiocarbon dating, based on their abundance in our samples and previous information published about Hong Kong subtidal molluscs. The selected shells were from the gastropod species *Bufonaria rana*, *Nassarius siquijorensis*, *Turricula nelliae*, *Turritella bacillum*, and Babylonidae, Naticidae and Tonnidae families; and the bivalve species were *Anadara globosa*, *Anadara consociata*, *Paphia euglypta*, *Paratapes undulatus*, *Pecten excavatus*, *Placamen lamellatum*, *Protapes gallus*, *Ruditapes philippinarum* and *Timocle micra* (Figure 3). The radiocarbon dating of the shells was performed by a single-stage accelerator mass spectrometer (YS-AMS) at the Atmosphere and Ocean research Institute, the university of Tokyo, Japan (Yokoyama et al. 2019). The radiocarbon dates of the shells were calibrated with the package "rintcal" (Blaauw 2023) for the software R (R Core Team 2022). Shells with positive radiocarbon ages, i.e., dating before the present (BP, shells older than 1950 AD), were dated using the calibration curve marine20 (Heaton et al. 2020). Shells with negative radiocarbon ages (shells younger than 1950 AD) were dated using the post-bomb calibration curve nh3 (Hua et al. 2021; Reimer et al. 2020).

To assess water and sediment quality on the sampling sites, historical data were retrieved for the areas near the trawling transects. The data was obtained from the Hong Kong Environmental Protection Department (EPD)

(https://cd.epic.epd.gov.hk/EPICRIVER/marine/). The EPD sampling stations for water

and sediment quality were NM6 and NS6 (both at 22°20.366'N, 113°53.908'E), respectively, for NL, SM13 and SS6 (22°12.957'N, 113°57.724'E) for SL, MM8 and MS8 (22°12.021'N, 114°19.345'E) for SE, SM6 and SS3 (22°11.500N, 114°4.743'E) for SW, MM17 and MS17 (22°30.192'N, 114°20.960'E) for EO, andTM8 and TS5 (22°28.392'N, 114°18.003'E) for EI (Figure 1). The obtained data time interval ranged from 1986, period when the EPD was created and started monitoring the water quality, up to 2020. The parameters used to assess water quality were visibility, turbidity, suspended solids, total

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nitrogen, total inorganic nitrogen, NO₂-, NO₃-, NH₃, NH₄+, phosphorus, orthophosphate, dissolved oxygen (DO), faecal coliforms, *Escherichia coli* and Biochemical Oxygen Demand (BOD). For the sediment quality, low and high molecular weight polycyclic aromatic hydrocarbon (LPAH and HPAH respectively), polychlorinated biphenyls (PCB), the metalloid arsenic and the metals cadmium, chromium, copper, lead, mercury, nickel, silver and zinc concentration data was retrieved. The data was averaged by year.

Table 1. GPS coordinates with approximate initial and final transects location.

Region	Transect	Water depth (m)	Latitude (N)	Longitude (E)	
Southeastern Lantau	WT1	7.4 - 19.9	22°11.259' - 22°12.285'	113°55.232' - 113°59.086'	
	WT2	4.0 - 11.6	22°11.264' - 22°12.926'	113°51.358' - 113°54.570'	
Northwestern Lantau	WT3	6.2 - 27.3	22°14.517' - 22°17.062'	113°50.099' - 113°52.152'	
	WT4	13 - 32	22°20.502' - 22°25.125'	113°53.197' - 114°00.297'	
Southwestern Hong Kong (Lamma)	ST1	9.4 – 34	22°09.967' - 22°12.951'	114°16.442' - 114°18.552'	
	ST2	21.5 – 36.8	22°08.590' - 22°10.509'	114°11.272' - 114°17.976'	
Southeastern Hong Kong	ST3	18.8 – 27.3	22°08.256' - 22°10.777'	113°51.038' - 114°14.085'	
	ST4	12.1 – 24.2	22°08.441' - 22°11.684'	114°04.780' - 114°07.789'	
Inner Tolo Harbor	ET1	14.8 – 23.6	22°26.990' - 22°29.456'	114°16.843' - 114°24.886'	
	ET2	7.4 – 23.0	22°29.537' - 22°30.275'	114°20.802' - 114°22.018'	
Outer Tolo Harbor	ET3	4.0 – 11.6	22°29.169' - 22°30.070'	114°19.233' - 114°20.993'	
	ET4	9.7 – 21.8	22°27.094' - 22°28.676'	114°15.629' - 114°18.093'	

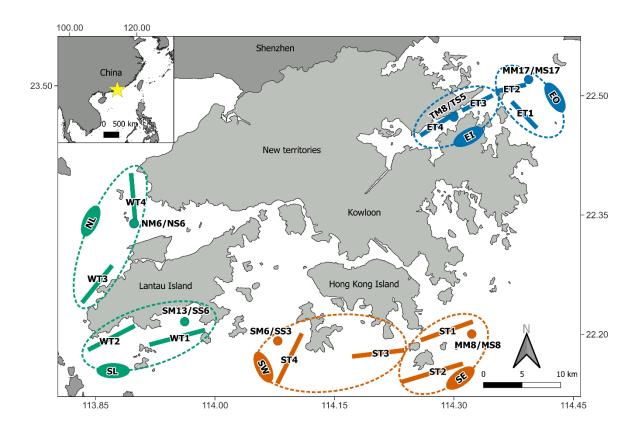


Figure 1. Sampling locations of Mollusc shells in Hong Kong. Each dashed ellipsis represents a sampling region, Northwestern Lantau (NL), Southeastern Lantau (SL), Southwestern Hong Kong (Lamma Island, SW), Southeastern Hong Kong (SE), Inner Tolo Harbor (EI) and Outer Tolo Harbor (EO). Lines represent the approximate location of sampling transects, and circles represent the location of EPD monitoring stations. Green represents Western Transects (WT), Orange represents Southern transects (ST), and blue represents Eastern Transects (ET).





Figure 2. Camera setup used to obtain photos of the shells.



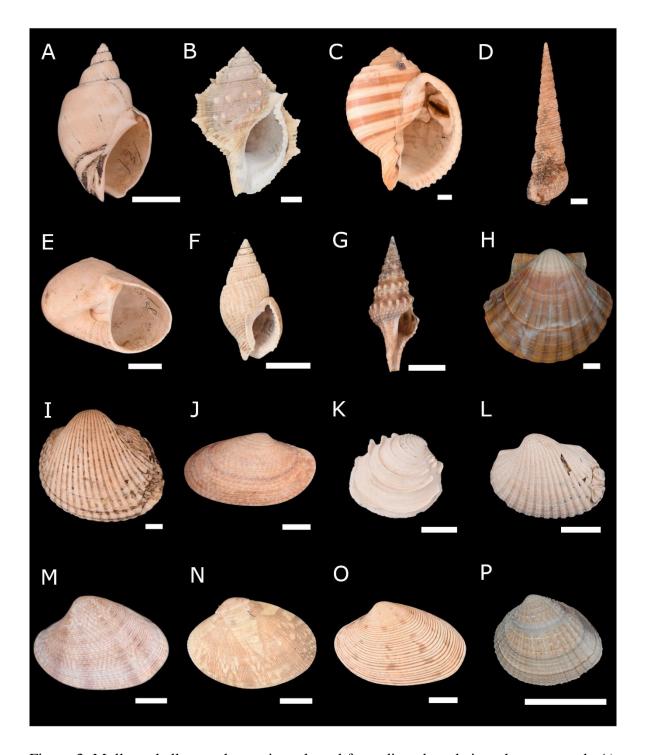


Figure 3. Mollusc shells morphospecies selected for radiocarbon dating: the gastropods A) Babylonidae, B) *Bufonaria rana*, C) Tonnidae, D) *Turritella bacillum*, E) Neritidae F) *Nassarius siquijorensis*, and G) *Turricula nelliae*; and the bivalves H) *Pecten excavatus*, I) *Anadara globosa*, J) *Paratapes undulatus*, K) *Placamen lamellatum*, L) *Anadara consociata*, M) *Protapes gallus*, N) *Ruditapes philippinarum*, O) *Paphia euglypta* and P) *Timoclea micra*. Scale bar = 10 mm.



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Statistical analyses were performed using the software R version 4.2.2 (R Core Team 2022). Shannon-Wiener diversity index were calculated for shell morphospecies composition at each sampling area. We compared the size of dead shells of representative species among sampling areas, the bivalves *A. consociata*, *P. undulatus*, *P. lamellatum* and the gastropods *B. rana*, *M. trapa* and *T. bacillum*. Fragmented shells which length could not be measured were excluded from the analyses. The shell size data were check for homogeneity and normality of the residual distribution by Levene and Shapiro-Wilks tests, respectively. Species which size data met the assumptions for parametric statistical tests were analysed by Analyses of Variance (ANOVA), with pairwise comparisons between groups performed by Tukey HSD tests with Holm's adjustment. Shell size data which did not met the assumptions for parametric tests even after transformation were analysed by Kruskal-Wallis tests, with pairwise comparisons between groups performed by Dunn's tests with Holm's adjustment.

The environmental data showed lower salinity on the western transects, with the lowest salinity at west Lantau (Figure 1A). There was a trend of warming waters over time in Hong Kong, with slightly warmer temperatures being found around Lantau (NL and SL), followed by southwest Hong Kong (Lamma Island, SW). Waters around Lantau and Lamma Island also had lower visibility. NL bottom waters historically have higher organic and inorganic nitrogen, NO₂-, NO₃- and orthophosphate (Figure 4). NL together with SW show higher Faecal coliforms and *E. coli* content compared to the other regions (Figure 5). The higher concentration of heavy metals such as cadmium, chromium, lead, nickel and zinc are in EI and EO, followed by SW and SE. NL sediments had higher concentration of the metalloid arsenic (Figure 6). LPAH, HPAH and TPCB are similar among areas (Figure 7).

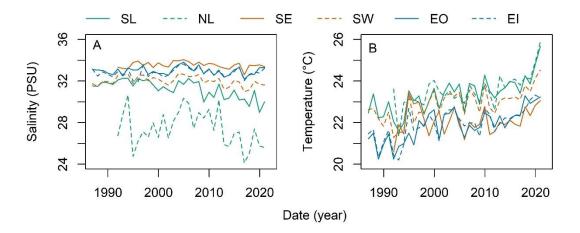


Figure 4. Salinity (A) and temperature (B) time series at the EPD water quality monitoring stations in the sampling areas. Southeast Lantau (SL), northwest Lantau (NL), Southeast Hong Kong (SE), Southwest Hong Kong (SW), Outer Tolo Harbour (EO) and Inner Tolo Harbour (EI).

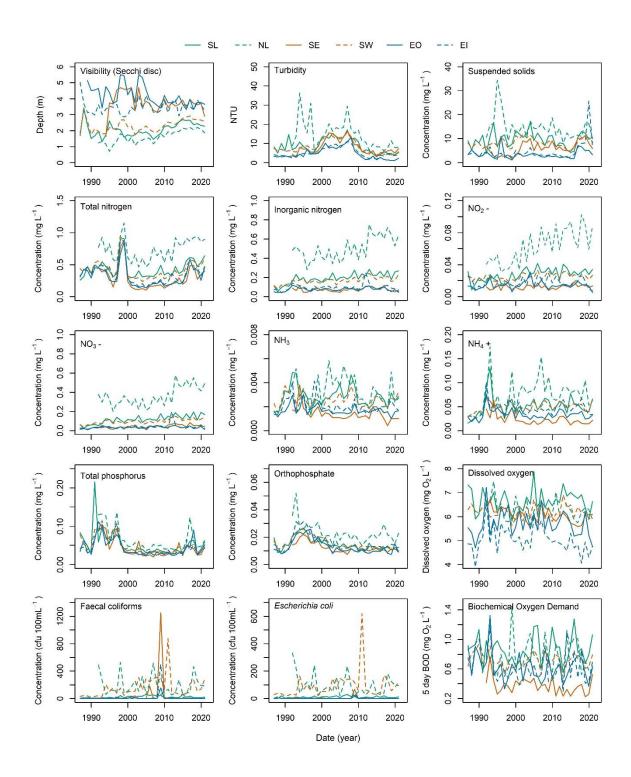


Figure 5. water quality parameters from bottom water at sampling sites near the trawling areas. Southeast Lantau (SL), northwest Lantau (NL), Southeast Hong Kong (SE), Southwest Hong Kong (SW), Outer Tolo Harbour (EO) and Inner Tolo Harbour (EI).

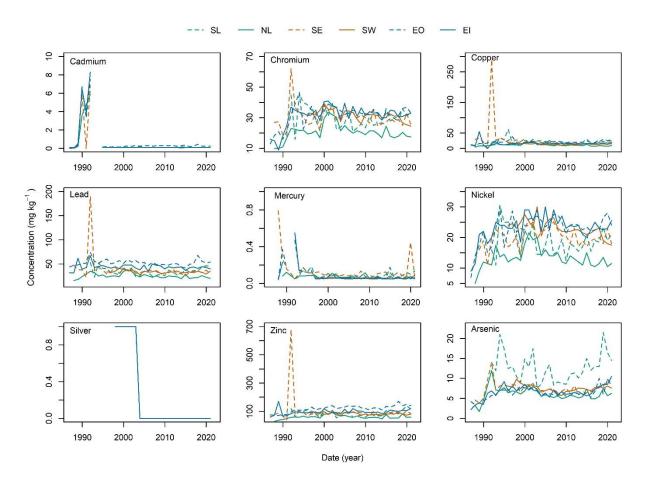


Figure 6. Concentration of metals and metalloids in marine sediments near the trawling areas, in the northwestern and southeastern Lantau waters. Southeast Lantau (SL), northwest Lantau (NL), Southeast Hong Kong (SE), Southwest Hong Kong (SW), Outer Tolo Harbour (EO) and Inner Tolo Harbour (EI).

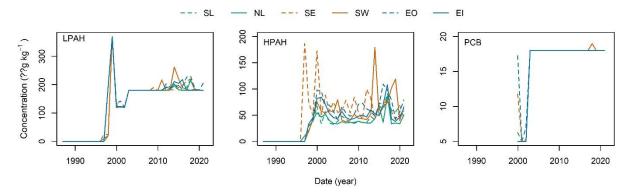


Figure 7. Concentration of low and high molecular weight polycyclic aromatic hydrocarbon (LPAH and HPAH, respectively) and total Polychlorinated biphenyls (PCB) in marine sediments near the trawling areas, in the northwestern and southeastern Lantau waters.



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Southeast Lantau (SL), northwest Lantau (NL), Southeast Hong Kong (SE), Southwest Hong Kong (SW), Outer Tolo Harbour (EO) and Inner Tolo Harbour (EI).

The highest diversity of shells was found in SW, followed by SL, NL, EI, EO, and the lowest diversity was found in SE (Shannon-Wiener index of 3.64, 3.39, 2.96, 2.34, 2.13 and 1.94, respectively). The shell composition of sampling areas could be distinguished into three different groups by analysis of similarity: west, south, and east of Hong Kong (Figure 8). The bivalve species A. consociata had larger shells in NL, followed by EO, and was smaller in the remaining transects (Kruskal-Wallis test, $\chi^2 = 42.510$, DF = 5, p < 0.001, Figure 9A). Paratapes undulatus was larger in SW, while smaller shells were found in SE and EI (Kruskal-Wallis test, $\chi^2 = 137.67$, DF = 5, p < 0.001, Figure 9B). There was no difference in size among sites in the bivalve P. lamellatum (Kruskal-Wallis test, χ^2 = 1.359, DF = 5, p = 0.93, Figure 9C). No differences in size was found for the gastropod B. rana (ANOVA, $F_{(5,118)} = 1.224$, SS = 1093.1, p = 0.302, Figure 10A). The ANOVA model accused a significant difference in the sizes of M. trapa (ANOVA, $F_{(5.51)} = 2.281$, SS = 3367, p = 0.037), however post-hoc Tukey HSD test adjusted by Holm's method showed no significant difference in size among sites (Figure 10B). No significant differences were found for N. siquijorensis sizes among sites (ANOVA, $F_{(5,114)} = 2.235$, SS = 116.05, p = 0.055, Figure 10C).

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nMDS Transects Dissimilarity

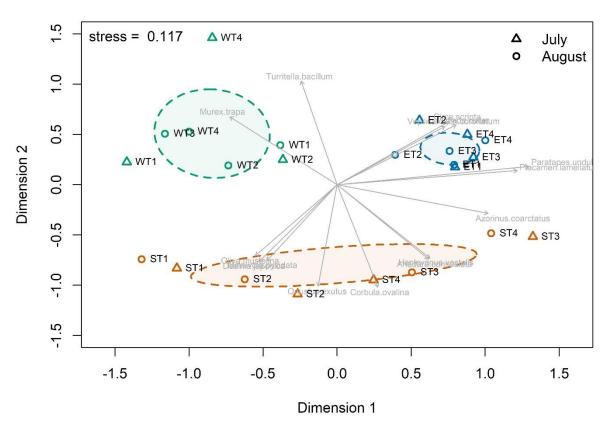


Figure 8. non-metric multidimensional scaling (nMDS) similarity analyses for the transects shell composition. Green are the western transects SL and NL, orange are the southern transects SE and SW, and blue are the eastern transects EO and EI.

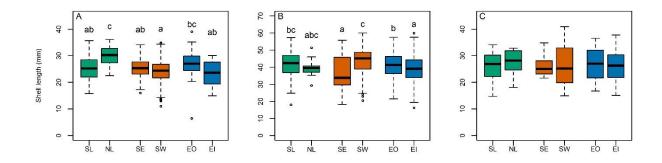


Figure 9. Comparison of the sizes of the bivalves *Anadara consociata* (A), *Paratapes undulatus* (B) and *Placamen lamellatum* (C). Green colour represents western transects, NL and SL, orange colour the southern transects SW and SE, and blue colour the eastern transects, EO and EI. Boxplots show the median, 25th and 75th percentiles, and whiskers are

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1.5 times the spread beyond the hinge. Different letters indicate significative differences among groups in each plot (Dunn test, p < 0.05).

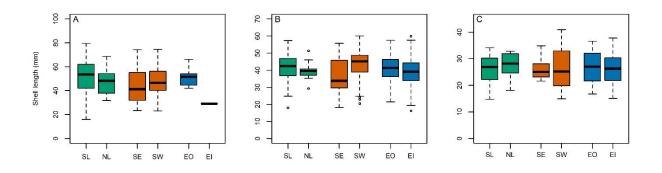


Figure 10. Comparison of the sizes of the gastropods *Bufonaria rana* (A), *Murex trapa* (B) and *Nassarius siquijorensis* (C). Green colour represents western transects, NL and SL, orange colour the southern transects SW and SE, and blue colour the eastern transects, EO and EI. Boxplots show the median, 25th and 75th percentiles, and whiskers are 1.5 times the spread beyond the hinge.

Table 2 and Figures 11 to 26 show the ages of the dated shells. All species had a mix of old and modern shells. Species such as Anadara consociata, Bufonaria rana, Paratapes undulatus had modern and old shells in all areas (Figures 11, 14 and 18), indicating They are baseline species across Hong Kong which are still present today. However, from these, Paratapes undulatus has larger number of older shells in both The Tolo Harbour and Northwest Lantau compared to in the other sampling areas, indicating possible populational decline due to environmental degradation (Figure 18). Anadara globosa has abundant number of shells in western waters of Hong Kong with both modern and old shells (Figure 12A and B). Only two shells of this species were found in south Hong Kong, with both being pre-1900s (Figure 12C and D). One shell of A. globosa was found in inner Tolo Harbour (Figure 12E). Babylonia spp. had old and young shells in Western Hong Kong (Figure 13A and B). In the south of Hong Kong, we only found one old shell of Babylonia sp. (Figure 13B and C). Old and young shells of Babylonia spp. were found in inner Tolo Harbour (Figure 13E), but no shells were collected from the outer Tolo Harbour (Figure 13F). The species N. siquijorensis had a combination of old and young shells, in the West, South and East Hong Kong waters and it is possibly a baseline species resistant to environmental degradation (Figure 15A to F). Shells from the Family Naticidae were



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found across all areas in Hong Kong with both old and young individuals (Figure 16A to F). However, in the Northwest Lantau (Figure 16A) and South of Hong Kong (Figure 16C) and D), only old shells were present, indicating possible impacts of environmental degradation on Naticidae moonsnails. Paphya euglypta was found in all sites but northwest Lantau (Figure 17A to F). This species only had modern shells in the Southeast waters of Hong Kong (Figure 17D), which is our least impacted site. The lack of shells from this species in other sites could indicate possible local extinction or populational decline due to the environmental degradation of Hong Kong shores. The species *P. excavatus* was present only in the East of Hong Kong (EI and EO) (Figure 19E and F). All dated shells from this species were old, indicating that *P. excavatus* was negatively affected by the degradation of the Tolo Harbour and may have been extinct in the region. The age of the shells of P. lamellatum show that this is also a baseline species in Hong Kong, with its shells displaying a broad age spam (Figure 20A to F). This species has no modern shells in Northwest Lantau (Figure 20A), indicating populational decline or extinction of the species in this area. Most of P. lamellatum shells in the inner and outer Tolo Harbour area are older, with few modern shells (Figure 20E and F), what may also be an indication of this species populational decline in the area. Protapes gallus also seems to be a baseline species in Hong Kong, with old shells being found in all sampled areas (Figure 21A to F). However, there are no modern shells from this species in the inner and outer Tolo Harbour, with possible local extinction of *P. gallus* in this area (Figure 21E and F). The oldest shell of R. philippinarum we dated is from 1896 (Table 2). The species has a majority of moderns shells and is present in most of our transects, except for Southeast Hong Kong and outer Tolo Harbour (Figure 22A to F). Timoclea micra was not present only in Northwest Lantau (Figure 23A). Only the outer Tolo Harbour had a modern shell of T. micra (Figure 23F), while the shells of this species in the other sites dates pre-1915 (Figure 23B to E). Thus, T. micra appears to be a baseline species which have suffered negative effects from Hong Kong marine waters degradation. Specimens of *Tonna* spp. were present in west and south Hong Kong, but not in the east (Inner and outer Tolo harbour) (Figure 24A to F). In the areas where it was present, its shells were not very abundant, but there was a mix of modern and old shells, showing that this genus was present in baseline conditions and is still present in Hong Kong waters. Turricula nelliae is present in West (Figure 25A and B) and East (Figure 25E and F) Hong Kong waters, but not in the south (Figure 25C and D). The ages are a mix of old and modern shells, indicating it is a baseline species which is still present in Hong Kong. Turritella bacillum has old and modern shells in all but one site, Southeast Hong Kong (Figure 26A to F). This shell is abundant in our samples and has large number of modern shells, indicating it thrives in impacted waters.



Table 2. Oldest (Min), youngest (Max) and median (Med) age of shells from the selected species in each area and respective number of replicates (N).

Area	Species	Min	Max	Med	N
	Anadara consociata	-5548.3	2013.1	2010.15	8
	Anadara globosa	1992	2015.8	2005.05	16
	Babylonia spp.	-5698.2	2009.9	1792.25	4
	Bufonaria rana	-4100.4	2009.2	1901.95	8
	Nassarius siquijorensis	2005.9	2018.9	2011.6	5
	Naticidae	-4508.8	1906.5	1808.6	4
NW	Paratapes undulatus	-4394.7	2014.5	517.35	10
	Placamen lamellatum	-5596.7	1901.4	555.7	11
	Protapes gallus	1603.4	2014.8	2009.6	15
	Ruditapes philippinarum	2010.5	2016.5	2013.9	7
	Tonna spp.	1920.5	2014.5	1967.5	2
	Turricula nelliae	1919.1	2013.4	2011.8	8
	Turritella bacillum	-5500.1	2017.1	2009.3	15
	Anadara consociata	1554.8	2009.6	2004.05	6
	Anadara globosa	1880.3	2016.3	2000.4	15
	Babylonia spp.	-801.4	2014.3	1917.7	14
	Bufonaria rana	1813.5	2010.5	1959.95	6
	Nassarius siquijorensis	1893.6	1916	1904.8	2
SL	Naticidae	658.7	2009.4	1994.05	10
	Paphia euglypta	1616.4	1917	1729.85	4
	Paratapes undulatus	31	2010.9	2006.15	10
	Placamen lamellatum	-3601.8	2003.1	1553.8	15
	Protapes gallus	1901.7	2015.8	2010.4	15
	Ruditapes philippinarum	1990.6	1990.6	1990.6	1
	Timoclea micra	1853.8	1853.8	1853.8	1
	Tonna spp.	-1605.9	2018.7	1916.9	12
	Turricula nelliae	1917.9	2013.9	2003.7	4
	Turritella bacillum	1866.8	2011.1	1994	15
	Anadara consociata	1898.6	2005.9	1999.3	8
	Anadara globosa	1870.4	1870.4	1870.4	1
	Babylonia spp.	1340.6	1340.6	1340.6	1
	Bufonaria rana	1900.4	2006.5	1915.5	7
	Nassarius siquijorensis	1828.7	2010.5	1999.5	15
	Naticidae	1661.3	1916.2	1880.6	8
SW	Paphia euglypta	625.8	1998	1890.6	3
	Paratapes undulatus	1280.8	2008.5	1910	15
	Placamen lamellatum	1900.7	2012.4	1998.1	15
	Protapes gallus	1914.8	2008.4	2008.1	3
	Ruditapes philippinarum	1896.5	2010.5	1915.4	3
	Timoclea micra	1849.7	1914.7	1905.5	14
	I IIIIOCICA IIIICIA	1077.1	1/17.1	1703.3	17



Table 2. Continuation.

Area	Species	Min	Max	Med	N
SW	Turritella bacillum	1915.1	2014.2	2009.7	15
SE	Anadara consociata	1887	2009.4	2000.8	8
	Anadara globosa	1811.9	1811.9	1811.9	1
	Bufonaria rana	1299	2012	1916.3	8
	Nassarius siquijorensis	1884.4	2013.5	2008.15	6
	Naticidae	-6265.7	2012.9	2006.45	8
	Paphia euglypta	-6137	2010.9	1384.3	10
	Paratapes undulatus	978.3	2013.3	2000.5	15
	Placamen lamellatum	-8206.8	2012.4	2001.5	13
	Protapes gallus	-6619.3	2012.6	-1613.9	15
	Tonna spp.	1251.5	2014.2	1637.05	4
	Anadara consociata	1916.6	2011.4	1998.35	8
	Anadara globosa	2000.4	2000.4	2000.4	1
	Babylonia spp.	1994.4	2011	2010.5	6
	Bufonaria rana	-1040.2	2010.1	1916.1	5
	Nassarius siquijorensis	1918.7	2002.1	1992.75	6
	Naticidae	295.1	2003.4	2001.5	7
	Paphia euglypta	-3570.5	-2247.3	-2908.9	2
EI	Paratapes undulatus	-5152.9	1999.9	1531.4	15
	Pecten excavatus	-100.3	1782.2	1721.25	6
	Placamen lamellatum	-316.9	2008.9	1915.8	15
	Protapes gallus	-1951	1918.3	577.1	3
	Ruditapes philippinarum	1915	2012.5	1992	9
	Timoclea micra	1905.5	1906.7	1906.1	2
	Turricula nelliae	-3940.4	1992.5	1920.6	3
	Turritella bacillum	1990.6	2010.7	1995.5	14
ЕО	Anadara consociata	1919.7	2012.2	2011.45	8
	Bufonaria rana	-4745.9	2012.1	1920	5
	Nassarius siquijorensis	1837.1	2012.2	1992.15	8
	Naticidae	1917.7	2010.7	2004.45	4
	Paphia euglypta	1695.7	1695.7	1695.7	1
	Paratapes undulatus	1303.6	2011.1	1996	15
	Pecten excavatus	-297.7	1897	1518.95	8
	Placamen lamellatum	1914.5	2009.9	1919.7	15
	Protapes gallus	482.1	482.1	482.1	1
	Timoclea micra	1998.3	1998.3	1998.3	1
	Turricula nelliae	1917.1	1996.6	1956.85	2
	Turritella bacillum	1991.7	2012.2	1998.4	15

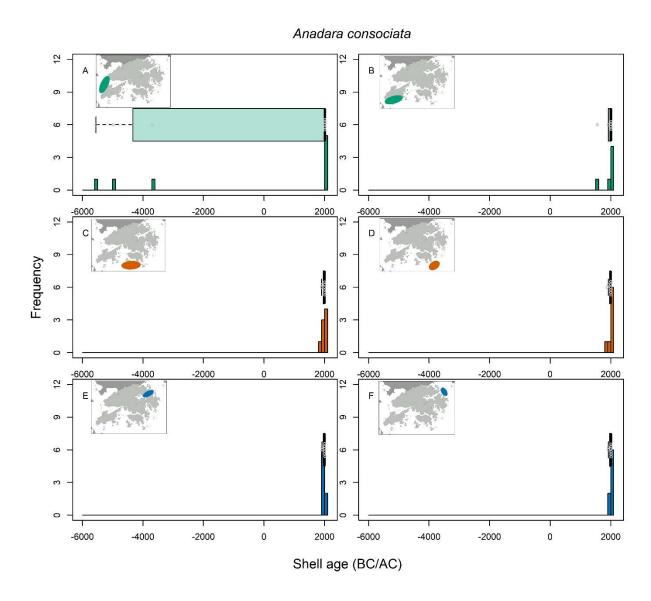


Figure 11. Histograms showing the shell age distribution of the species *Anadara consociata* in Northwest Lantau (A), South Lantau (B), Southwest Hong Kong Island (C), Southwest Hong Kong Island (D), Inner Tolo Harbour (E), and outer Tolo Harbour (F). Boxplots show the median, 25th and 75th percentiles, and whiskers are 1.5 times the spread beyond the hinge. Grey circles represent the data points.

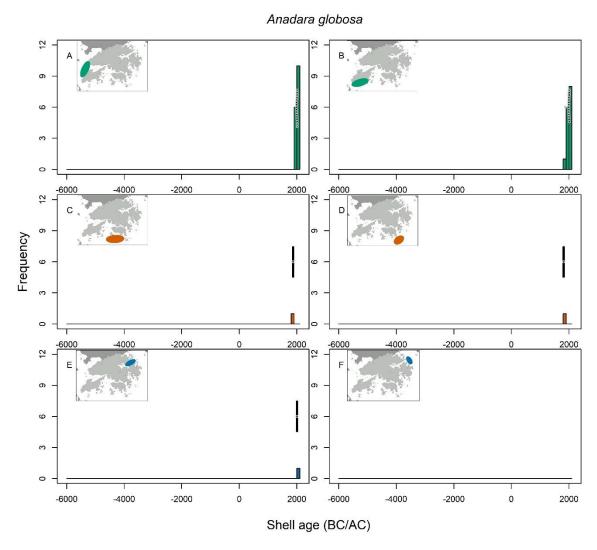


Figure 12. Histograms showing the shell age distribution of the species *Anadara globosa* in Northwest Lantau (A), South Lantau (B), Southwest Hong Kong Island (C), Southwest Hong Kong Island (D), Inner Tolo Harbour (E), and outer Tolo Harbour (F). Boxplots show the median, 25th and 75th percentiles, and whiskers are 1.5 times the spread beyond the hinge. Grey circles represent the data points.

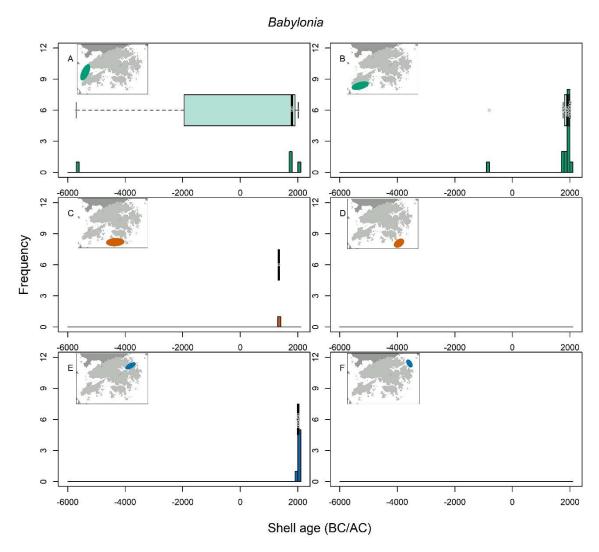


Figure 13. Histograms showing the shell age distribution of the species *Babylonia* spp. in Northwest Lantau (A), South Lantau (B), Southwest Hong Kong Island (C), Southwest Hong Kong Island (D), Inner Tolo Harbour (E), and outer Tolo Harbour (F). Boxplots show the median, 25th and 75th percentiles, and whiskers are 1.5 times the spread beyond the hinge. Grey circles represent the data points.

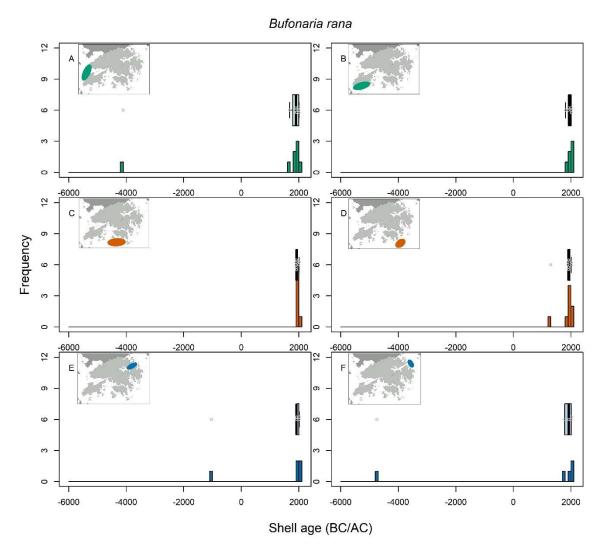


Figure 14. Histograms showing the shell age distribution of the species *Bufonaria rana* in Northwest Lantau (A), South Lantau (B), Southwest Hong Kong Island (C), Southwest Hong Kong Island (D), Inner Tolo Harbour (E), and outer Tolo Harbour (F). Boxplots show the median, 25th and 75th percentiles, and whiskers are 1.5 times the spread beyond the hinge. Grey circles represent the data points.

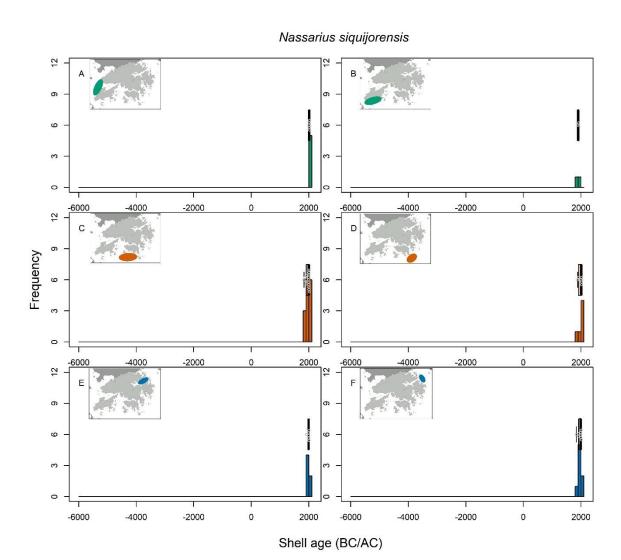


Figure 15. Histograms showing the shell age distribution of the species *Nassarius siquijorensis* in Northwest Lantau (A), South Lantau (B), Southwest Hong Kong Island (C), Southwest Hong Kong Island (D), Inner Tolo Harbour (E), and outer Tolo Harbour (F). Boxplots show the median, 25th and 75th percentiles, and whiskers are 1.5 times the spread beyond the hinge. Grey circles represent the data points.

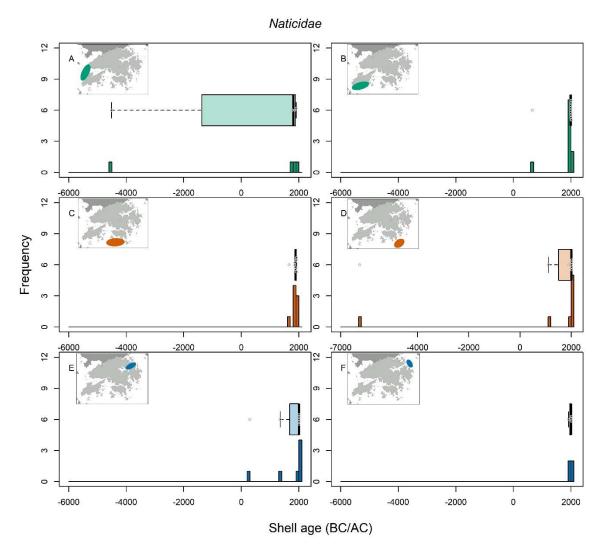


Figure 16. Histograms showing the shell age distribution of the family Naticidae in Northwest Lantau (A), South Lantau (B), Southwest Hong Kong Island (C), Southwest Hong Kong Island (D), Inner Tolo Harbour (E), and outer Tolo Harbour (F). Boxplots show the median, 25th and 75th percentiles, and whiskers are 1.5 times the spread beyond the hinge. Grey circles represent the data points.

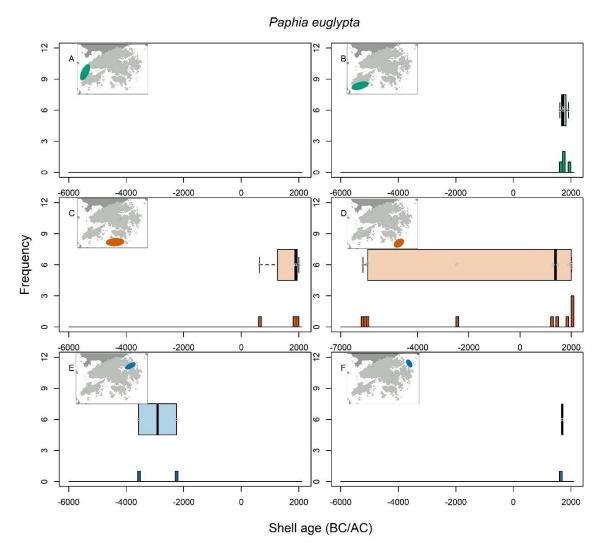


Figure 17. Histograms showing the shell age distribution of the species *Paphia euglypta* in Northwest Lantau (A), South Lantau (B), Southwest Hong Kong Island (C), Southwest Hong Kong Island (D), Inner Tolo Harbour (E), and outer Tolo Harbour (F). Boxplots show the median, 25th and 75th percentiles, and whiskers are 1.5 times the spread beyond the hinge. Grey circles represent the data points.

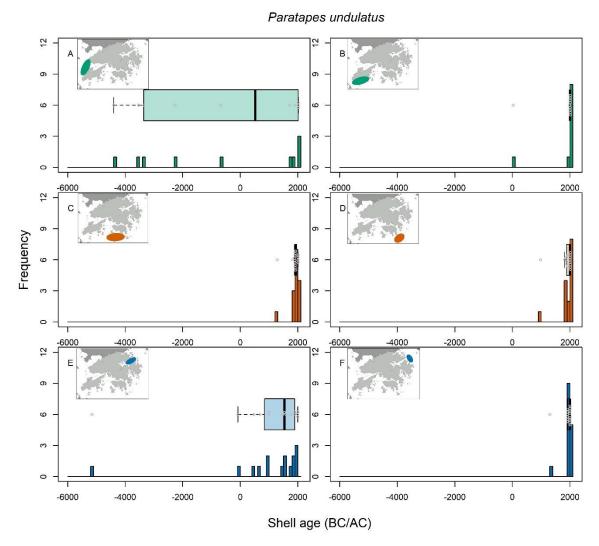


Figure 18. Histograms showing the shell age distribution of the species *Paratapes undulatus* in Northwest Lantau (A), South Lantau (B), Southwest Hong Kong Island (C), Southwest Hong Kong Island (D), Inner Tolo Harbour (E), and outer Tolo Harbour (F). Boxplots show the median, 25th and 75th percentiles, and whiskers are 1.5 times the spread beyond the hinge. Grey circles represent the data points.

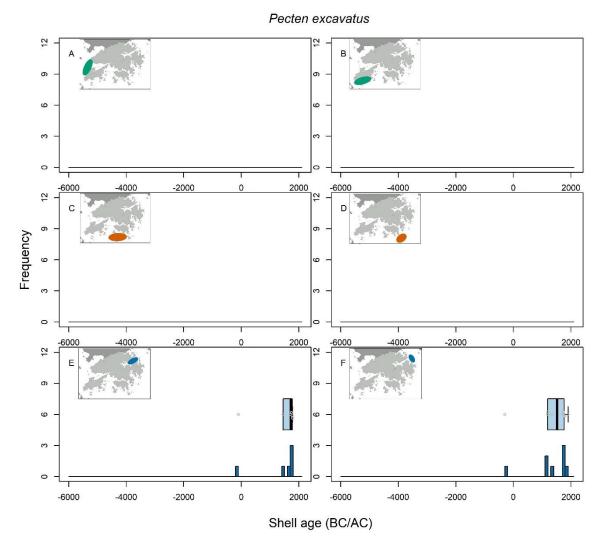


Figure 19. Histograms showing the shell age distribution of the species *Pecten excavatus* in Northwest Lantau (A), South Lantau (B), Southwest Hong Kong Island (C), Southwest Hong Kong Island (D), Inner Tolo Harbour (E), and outer Tolo Harbour (F). Boxplots show the median, 25th and 75th percentiles, and whiskers are 1.5 times the spread beyond the hinge. Grey circles represent the data points.

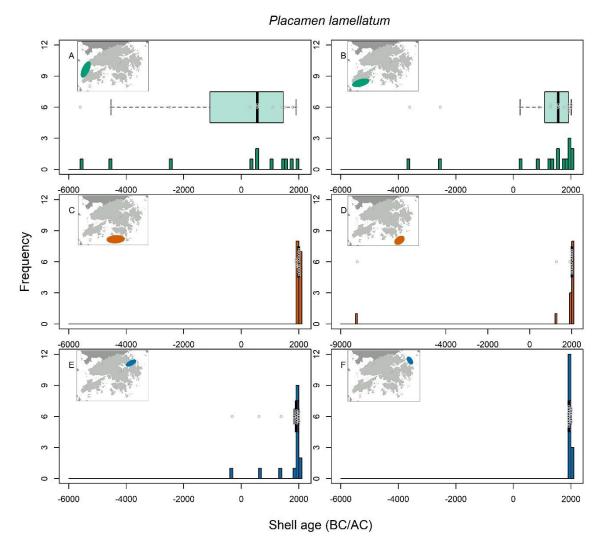


Figure 20. Histograms showing the shell age distribution of the species *Placamen lamellatum* in Northwest Lantau (A), South Lantau (B), Southwest Hong Kong Island (C), Southwest Hong Kong Island (D), Inner Tolo Harbour (E), and outer Tolo Harbour (F). Boxplots show the median, 25th and 75th percentiles, and whiskers are 1.5 times the spread beyond the hinge. Grey circles represent the data points.

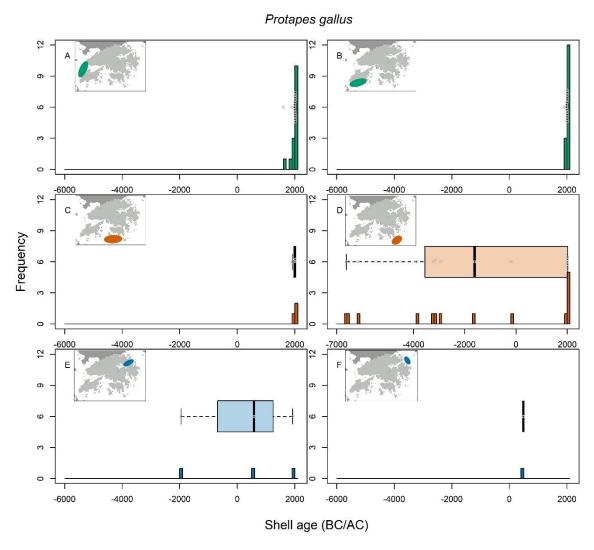


Figure 21. Histograms showing the shell age distribution of the species *Protapes gallus* in Northwest Lantau (A), South Lantau (B), Southwest Hong Kong Island (C), Southwest Hong Kong Island (D), Inner Tolo Harbour (E), and outer Tolo Harbour (F). Boxplots show the median, 25th and 75th percentiles, and whiskers are 1.5 times the spread beyond the hinge. Grey circles represent the data points.

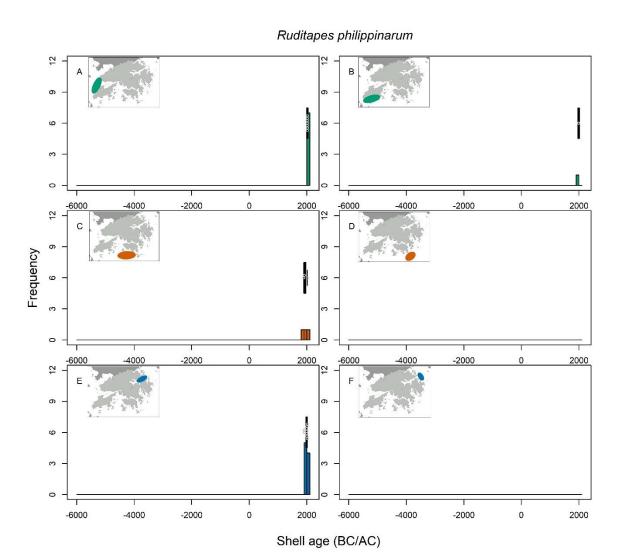


Figure 22. Histograms showing the shell age distribution of the species *Ruditapes philippinarum* in Northwest Lantau (A), South Lantau (B), Southwest Hong Kong Island (C), Southwest Hong Kong Island (D), Inner Tolo Harbour (E), and outer Tolo Harbour (F). Boxplots show the median, 25th and 75th percentiles, and whiskers are 1.5 times the spread beyond the hinge. Grey circles represent the data points.

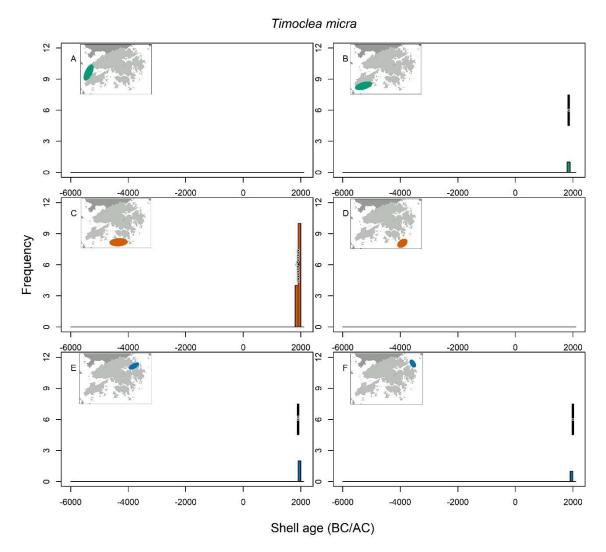


Figure 23. Histograms showing the shell age distribution of the species *Timoclea micra* in Northwest Lantau (A), South Lantau (B), Southwest Hong Kong Island (C), Southwest Hong Kong Island (D), Inner Tolo Harbour (E), and outer Tolo Harbour (F). Boxplots show the median, 25th and 75th percentiles, and whiskers are 1.5 times the spread beyond the hinge. Grey circles represent the data points.

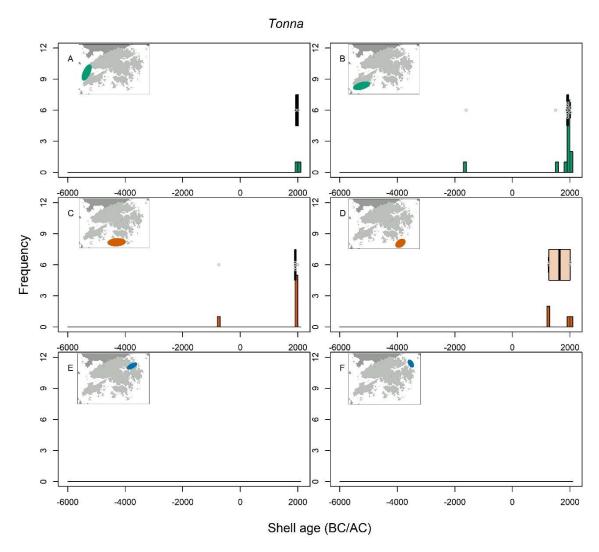


Figure 24. Histograms showing the shell age distribution of *Tonna* spp. in Northwest Lantau (A), South Lantau (B), Southwest Hong Kong Island (C), Southwest Hong Kong Island (D), Inner Tolo Harbour (E), and outer Tolo Harbour (F). Boxplots show the median, 25th and 75th percentiles, and whiskers are 1.5 times the spread beyond the hinge. Grey circles represent the data points.

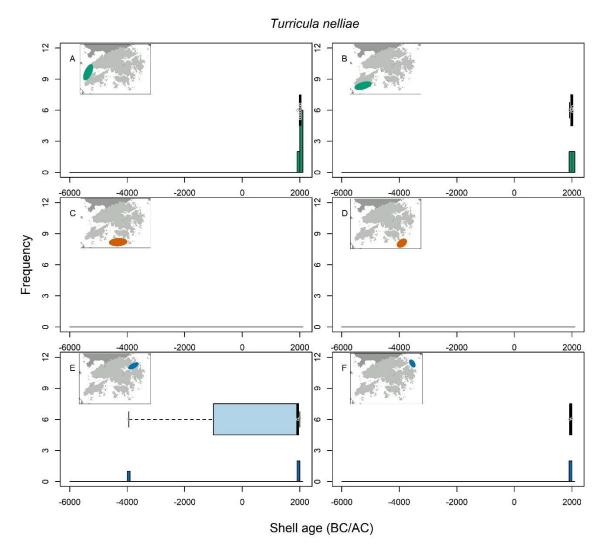


Figure 25. Histograms showing the shell age distribution of the species *Turricula nelliae* in Northwest Lantau (A), South Lantau (B), Southwest Hong Kong Island (C), Southwest Hong Kong Island (D), Inner Tolo Harbour (E), and outer Tolo Harbour (F). Boxplots show the median, 25th and 75th percentiles, and whiskers are 1.5 times the spread beyond the hinge. Grey circles represent the data points.

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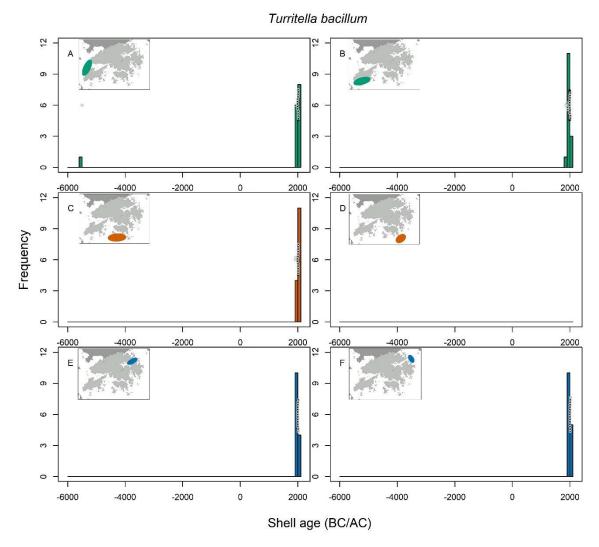


Figure 26. Histograms showing the shell age distribution of the species *Turricula nelliae* in Northwest Lantau (A), South Lantau (B), Southwest Hong Kong Island (C), Southwest Hong Kong Island (D), Inner Tolo Harbour (E), and outer Tolo Harbour (F). Boxplots show the median, 25th and 75th percentiles, and whiskers are 1.5 times the spread beyond the hinge. Grey circles represent the data points.

Our environmental and shell size analyses suggest (1) Highly impacted waters in Hong Kong, with high levels of nitrogen and phosphate compounds in northwestern Lantau and high levels of heavy metals in Tolo harbour, (2) different shell diversity, and therefore, mollusc communities among west, south and east Hong Kong. Our shell age analyses show different response between tolerant and sensitive mollusc species to environmental degradation in Hong Kong waters and bring evidence that the pollution and modification



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of shallow subtidal areas in the city led to the change in subtidal communities. The ages of our shells indicate that several subtidal mollusc species such as *P. euglypta, P. excavatus, P. lamellatum, P. gallus, P. lamellatum,* and *T. micra,* and moonsnails in the family Naticidae suffered populational decline or even local extinctions in certain areas in Hong Kong, with the Tolo Harbour being the area with largest number of species with lack of modern shells. Thus, the environmental degradation in Hong Kong caused severe impacts in the subtidal fauna, with many other species possibly having suffered similar impacts. Our results also bring insights on the subtidal fauna baseline conditions, showing species which were present in pre-anthropogenic impact Hong Kong waters.

As a form of divulgation, the project was presented at the 2nd Asian Paleontological Congress (APC2) held at the University of Tokyo, Japan on August 2023 (Figure 27).

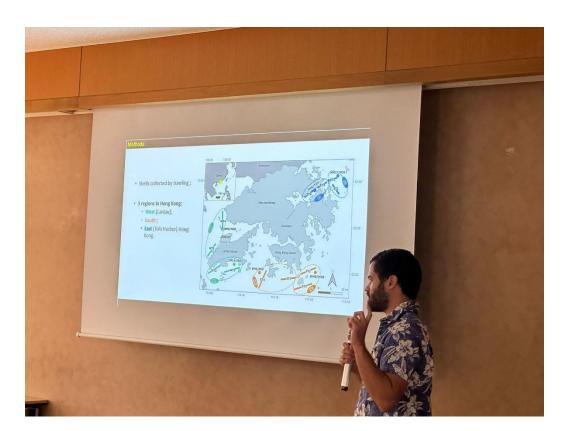


Figure 27. Project "Shells for understanding Hong Kong subtidal ecosystem history: a conservation baseline" being presented at the 2nd Asian Paleontological Congress in Tokyo, Japan.





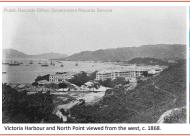
What dead molluscan shells can tell us about marine faunal changes in Hong Kong?



Authors: Pedro J Jimenez, Yuanyuan Hong, Rachel WC Chu, Jason KC Yau, Yusuke Yokoyama, Nicole Khan, David M Baker, Kenneth MY Leung & Moriaki Yasuhara

Hong Kong: a city with long history of coastal degradation

- Hong Kong has long history of occupation, with records of human presence dating back to the
 paleolithic.
- During the 1970s, the city witnessed a significant boost in economic growth, leading to rampant urbanization, which caused the degradation of marine environments.





- Urbanization and industrialization induced eutrophication (i.e. nutrients enrichment, such as Nitrogen) and pollution (such as from heavy metals, e.g. lead) of Hong Kong waters.
- Eutrophication causes harmful algal blooms and subsequent oxygen depletion in the water.
- Heavy metals can disrupt organisms' biology, ultimately leading to their death.

How the development of Hong Kong and derived water quality degradation affected its marine fauna?

Minimum oxygen concentration in the water** (mg L⁻¹) Lead concentration in the sediments (mg kg⁻¹)

Water quality (average for the period between 2009-2018)

Nitrogen* concentration in the sediments (mg kg-1)

Mollusc shells: witnesses to history

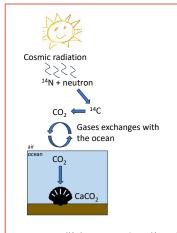
Mollusc shells are resilient and stay in the environment for hundreds and thousands of years, and even longer!



- Shells of ancient and living molluscs can be found together on the sea floor.
- The ratio of old/modern shells of a given species gives us clues about populational changes.
- By determining the age of these mollusc shells, we can:
 - Get insights into faunal baseline conditions.
 - · assess which species were affected by coastal degradation in Hong Kong.

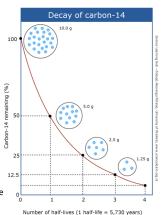
And how to determine the age of the shells? Radiocarbon dating

- Radiocarbon dating is a method that allows us to estimate the age of structures containing Carbon-14 (¹⁴C), such as Calcium Carbonate (CaCO₂) in mollusc shells.
- In this method, we measure the proportion of the unstable radioactive ¹⁴C in a sample to calculate its age:



- Nitrogen-14 (¹⁴N) is converted into ¹⁴C in the atmosphere.
- $^{14}\mathrm{C}$ is mixed into the ocean in the form of CO_2 .
- CO₂ is used by shellfish to produce their shells, composed of calcium carbonate (CaCO₂).

- After death, the animal do not uptake ¹⁴C anymore.
- 14C decays over time:

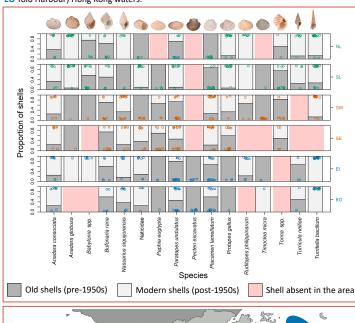


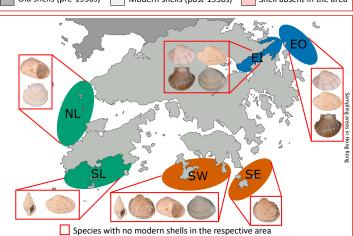
Measuring the proportion of the unstable ¹⁴C against the more common and stable ¹²C and the rare ¹³C allows us to calculate when the animal died.

Distribution of modern and old shells in Hong Kong waters

*Kjeldahl Nitrogen; **Bottom Wate

 We dated shells of representative species from West (Northwest, NW; and Southeast, SL Lantau), South (Southwest, SW; and Southeast, SE Hong Kong), and East (Inner, EI; and Outer, EO Tolo Harbour) Hong Kong waters.





- Some species lack modern shells in certain areas; They may have suffered populational declines or even local extinction.
 - Shells with mixed ages (old and modern) represent baseline species in Hong Kong waters.
- Eutrophication and pollution severely impacted Hong Kong waters and its fauna. It is uncertain
 if Hong Kong marine fauna will ever recover to baseline conditions.
- Mitigation plans to diminish pollution and modification of Hong Kong coast are fundamental to prevent further losses in diversity and subsequent losses of ecological functions.



























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(v) Evaluation of the project effectiveness in achieving the proposed objectives as well as the impact (benefits) of the Project;

The project was effective in achieving its proposed objectives. Our environmental analyses show differences in water and sediment quality among Western, Southern and Eastern Hong Kong waters, and our shell size analyses reveal different phenotypical responses of certain species to the different environmental conditions found among these areas. The radiocarbon dating of shells showed that the environmental degradation of Hong Kong Marine Ecosystems affected the marine fauna composition and caused local populational declines and even population extinctions. This project revealed some of the Hong Kong subtidal molluscan species present in natural baseline conditions and showed how they were impacted by the degradation of Hong Kong marine habitats. The results presented here improve our understanding about the urbanization impacts on the subtidal fauna from Hong Kong. The information we provide with this project can be used for conservation projects such as defining areas for preservation or restauration to conserve threatened species, or to pursue a more natural faunal composition.

(vi) Summary and Way Forward

In this project, we investigated the environmental degradation and dated mollusc shells from West, South and East Hong Kong waters. We identified baseline species which were present in pre-anthropogenic impact Hong Kong subtidal habitats and how they were affected by urbanization and subsequent environmental degradation. We used radiocarbon dating to infer the ages of 630 shells from 16 taxa including both gastropods and bivalves. The dating returned a mix of modern and old shells. We conducted an environmental analysis which showed that west Hong Kong waters are highly eutrophic while eastern waters in the Tolo Harbour contain heavy metal contaminants with high concentration of metals such as chromium, nickel, zinc and lead. The different environmental quality was reflected in different shell communities in the West, South and East Hong Kong waters. The radiocarbon dating of mollusc shells indicate that both West and East Hong Kong waters have species which have no modern individuals but that were present in the past, indicating possible extinction of these species in these areas. The radiocarbon dating of mollusc shells allowed us to identify several natural baseline species and how they were affected to environmental degradation. Future steps based on this project include



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identifying other species which suffered negative effects from environmental degradation and also species which are resilient to this degradation. The results of this project can be used in setting areas for conservation to avoid losing species which are sensitive to pollution and eutrophication.

(vii) Financial statement of the project

Financial statement is not disclosed due to confidentiality reasons.

(viii) Copies of supporting receipts

Supporting receipts are not disclosed due to confidentiality reasons.

(ix) Staff attendance record in accordance with the attendance monitoring plan

Staff attendance record is not disclosed due to confidentiality reasons.



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(x) If applicable, recruitment record for all project staff employed in accordance with the recruitment plan under the project.

Staff recruitment record is not disclosed due to confidentiality reasons.

(xi) A list of all project assets with photos enclosed as an appendix to the completion report.

NA. There are no project asset.

I hereby irrevocably declare to the MEEF Management Committee and the Steering Committee of the relevant Funds including the Top-up Fund, that all the dataset and information included in the completion report has been properly referenced, and necessary authorisation has been obtained in respect of information owned by third parties.

Any opinions, findings, conclusions or recommendations expressed in this report do not necessarily reflect the views of the Marine Ecology Enhancement Fund or the Trustee.

I hereby irrevocably declare, warrant and undertake to the MEEF Management Committee and the Steering Committee of the relevant Funds including the Top-up Fund, that I myself, and the Organisation:-

- 1. do not deal with, and are not in any way associated with, any country or organisation or activity which is or may potentially be relevant to, or targeted by, sanctions administered by the United Nations Security Council, the European Union, Her Majesty's Treasury-United Kingdom, the United States Department of the Treasury's Office of Foreign Assets Control, or the Hong Kong Monetary Authority, or any sanctions law applicable;
- 2. have not used any money obtained from the Marine Ecology Enhancement Fund or the related Top-up Fund (and any derived surplus), in any unlawful manner, whether



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involving bribery, money-laundering, terrorism or infringement of any international or local law; and

3. have used the funds received (and any derived surplus) solely for the studies or projects which further the MEEF Objectives and have not distributed any portion of such funds (including any derived surplus) to members of the Recipient Organisation or the public.

Sincerely yours,

Moriaki Yasuhara (PI)

--

Dr. Moriaki Yasuhara

Associate Professor

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