

## Research Article

**Distribution of intertidal rock oysters in the Pilbara, Western Australia**Fred E. Wells<sup>1,2</sup>, Sherralee S. Lukehurst<sup>1</sup>, Laura A. F. Fullwood<sup>1</sup> and Euan S. Harvey<sup>1</sup><sup>1</sup>School of Molecular and Life Sciences, Curtin University, Kent Street, Bentley, WA 6102 Australia<sup>2</sup>Negaunee Integrative Research Center, Field Museum of Natural History, Chicago, Illinois 60605 USACorresponding author: Fred E. Wells ([fred.wells@curtin.edu.au](mailto:fred.wells@curtin.edu.au))

**Citation:** Wells FE, Lukehurst SS, Fullwood LAF, Harvey ES (2024) Distribution of intertidal rock oysters in the Pilbara, Western Australia. *Management of Biological Invasions* 15(1): 131–143, <https://doi.org/10.3391/mbi.2024.15.1.08>

**Received:** 23 August 2023

**Accepted:** 7 December 2023

**Published:** 22 January 2024

**Handling editor:** Katherine Dafforn

**Copyright:** © Wells et al.

This is an open access article distributed under terms of the Creative Commons Attribution License ([Attribution 4.0 International - CC BY 4.0](https://creativecommons.org/licenses/by/4.0/)).

## OPEN ACCESS

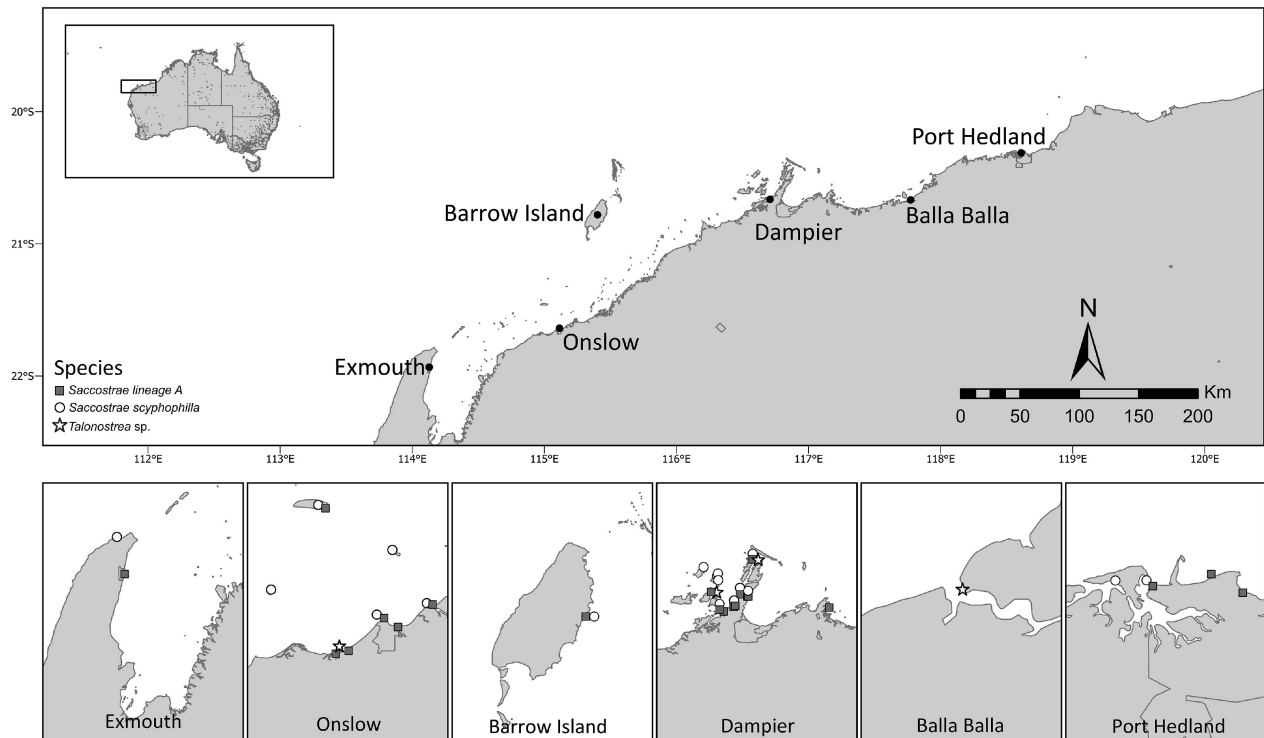
**Abstract**

Invasive marine species (IMS) are a major anthropogenic threat to global marine environments. To mitigate the threat, Australian federal and state governments have developed detailed quarantine programs to minimise IMS introductions, supported by monitoring programs to detect any IMS that penetrated the quarantine barriers. Considerable shipping movements occur between the Pilbara region of northwestern Australia and southeast Asia where a number of potential IMS oysters occur. eDNA techniques are being developed to rapidly scan biofouling and other samples for potential IMS, but there is limited information on oysters present in the Pilbara. We collected intertidal oysters in and near Pilbara ports and identified them using DNA sequences to determine if any IMS are present and to provide a baseline to prevent false positive results for IMS by native species. Only three species were detected: native *Saccostrea* lineage A and *S. scyphophilla*, both widespread in the Pilbara, and *Talonostrea* sp. nov. which had not previously been reported from the Pilbara. No IMS oysters were found. The study provides a solid basis for monitoring for any future occurrences of IMS oyster species in the Pilbara, an area with a very high known shallow water marine biodiversity, but where only a single IMS is known to occur.

**Key words:** Ostreidae, *Saccostrea*, *Magallana*, *Crassostrea*, *Talonostrea*, marine invasive species, ports, aquaculture, yachts

**Introduction**

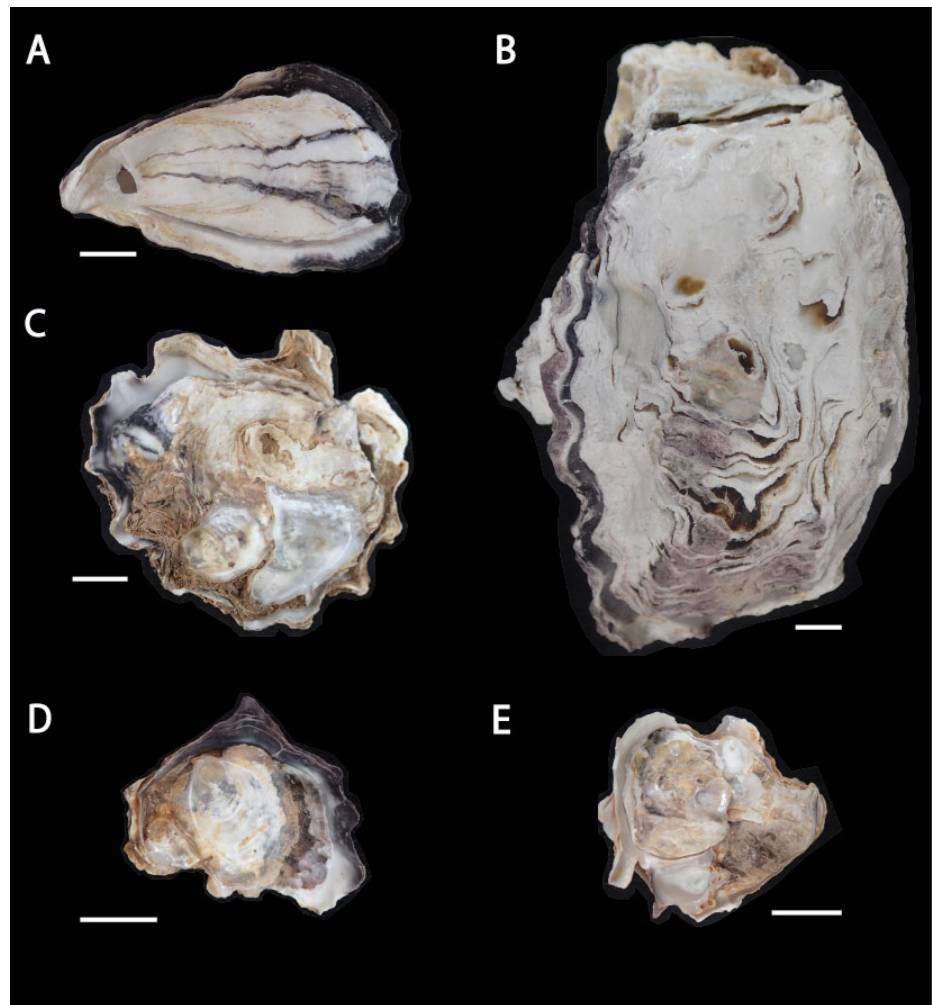
Invasive marine species (IMS) are a major anthropogenic threat to global marine environments (Johnson and Chapman 2007; Molnar et al. 2008; Katsanevakis et al. 2014; Crowe and Frid 2015; McDonald et al. 2020; Salimi et al. 2021). As an island continent, 98% of commerce into or out of Australia is on vessels (Ports Australia 2022). In recognition of the IMS threat, a strategy of quarantine barriers to prevent the introduction of IMS into Australia (e.g. Vessel-Check 2022) and monitoring for any that penetrated the barriers, was developed. A list of over 80 species of potential IMS concern was included in the strategy (NIMPCG 2009a, 2009b; DPIRD 2016). As the original monitoring strategy (NIMPCG 2009a, 2009b) was labour intensive and expensive a new strategy, the State-Wide Array Surveillance Program (SWASP) was developed based on passive settlement arrays deployed in ports. Marine growth on the arrays is collected and processed



**Figure 1.** Map of the Pilbara region of Western Australia showing ports mentioned in the text.

using Next-Generation Sequencing (NGS) to detect species on the potential IMS list (McDonald et al. 2020). As part of the SWASP program an extensive DNA database of potential IMS has been developed (Dias et al. 2017). One component of the BHP funded eDNA for Global Environment Studies (eDGES; Curtin 2022) program is continuing to develop the DNA database of potential IMS and related native species and use the database to search for species that may have already arrived in the Pilbara region of northwestern Australia, between North West Cape and Port Hedland (Figure 1). MarinePests (2023) records no marine pests on the north coast of Australia. However, their definition does not include the ascidian *Didemnum perlucidum*, which occurs throughout Western Australia, the Northern Territory and has recently been found in Queensland (Dias et al. 2021).

While there are a number of mechanisms for species introductions, shipping, either as biofouling or in ballast water, is the dominant source in most areas (Seebens et al. 2013; Salimi et al. 2021). Ports operated by the Pilbara Ports Authority, a government agency, exported 733.1 million tonnes, primarily iron ore, with 17,376 vessel movements in 2021–2022 (PPA 2022); there was significant additional export and vessel movements from private ports. Apart from commercial vessels, there is very little movement into northwestern Australia. Vessels, including the small number of private yachts, entering Western Australian waters from eastern Australia must adhere to the same reporting procedures as international commercial vessels. Only a few yachts enter WA from overseas. Those that do arrive are required to make Fremantle in southwestern WA as their first port of call (DAFF 2023), and are not permitted to first stop in the Pilbara. Aquaculture is another possible source



**Figure 2.** Intertidal rock oysters from the Pilbara, Western Australia. A: *Saccostrea* lineage A from a trial aquaculture farm (WAMS\_117501; GenBank OR466892). B: *Saccostrea scyphophilla* (WAMS\_117500; GenBank OR466930) C. Natural *Saccostrea* lineage A from a rocky shore (WAMS\_117506; GenBank OR466894). D, E. *Talonostrea* sp. nov. (WAMS\_117503; WAMS\_117504; GenBank OR466939). Scale bars are 1 cm. Photographs by Fred Wells.

of IMS introductions, but there is only one significant aquaculture industry in the Pilbara, for the pearl oyster *Pinctada maxima*. The fishery uses a combination of wild caught and hatchery grown pearl oysters. There are currently only three vessels in the wild caught fleet. A recent assessment of the *P. maxima* fishery scored the possibility of introducing IMS from overseas as very low risk (Smith et al. 2023).

The Pacific oyster (*Magallana gigas*, previously known as *Crassostrea gigas*) is a key potential IMS. It was introduced into Tasmania from Japan in 1949 (Thomson 1952, 1959). A successful and a thriving Pacific oysters aquaculture industry is now well established in south-eastern Australia. However, *M. gigas* escaped from the aquaculture leases and is now a feral pest species (NIMPIS 2022). The introduction of oysters for aquaculture also brings risks of introducing diseases and other species living on, or in, the oysters (Ruesink et al. 2005; Herbert et al. 2016).

For a number of years Pacific oyster spat were transported from Tasmania to Singapore and grown out in Singapore for the local market. *M. gigas* has



**Figure 3.** *Saccostrea* lineage A (left) and *Saccostrea scyphophilla* (center) and *Talonostrea* sp. nov. in their natural habitat in the Pilbara, Western Australia. Note that *Talonostrea* sp. nov. actually lived underwater attached to the bottom of the *Avicennia marina* branch but have been moved so they are visible. Photographs by Fred Wells.

been recorded in Singapore and neighbouring Malaysia (Lam and Morton 2009; Tan and Woo 2010), but whether it has actually established populations, or if *M. gigas* has been misidentified and there is an unknown local species has not been determined (Lam and Morton 2009). The importance of this question was demonstrated when an oyster found on 100m long crane barge *Mammoet 331* in Singapore in 2011 was tentatively identified as *M. gigas*. The vessel was drydocked, dewatered and cleaned. It was then refloated, moved 1.8m forward and dewatered a second time to clean drydock support strips from the first dewatering, causing expensive direct costs and construction delays in the project for which it was mobilising (Wells and Booth 2012).

Oysters are one of the target groups for the eDGES study in the Pilbara. Oyster taxonomy is notoriously difficult, with considerable morphological variations between adjacent individuals. Species usually cannot be reliably identified using shell and tissue morphology; DNA analysis is the most reliable method for species identification (Salvi and Mariottini 2016; Willan et al. 2021). The goals of the present research were to use genetic sequencing to determine the rock oyster species present in the Pilbara and their distribution, concentrating on ports and nearby areas where introduced oysters, if any, are most likely to be present. This will determine whether any introduced species are currently present in the Pilbara and provide a baseline for monitoring any future invasions by rock oysters.

## Materials and methods

### *Species taxonomy*

The taxonomy of oysters is in a considerable state of flux as DNA sequencing modifies our understanding of species limits. In particular the systematics of *Saccostrea* is undergoing modification. We have used the names *Saccostrea* lineage A and *S. scyphophilla* to be consistent with the recent paper on the group in Western Australia by Snow et al. (2023).

Voucher specimens have been deposited in the Western Australian Museum (WAM) and registered on GenBank.

### *Sample collection*

Four field surveys were conducted in and near major Pilbara ports (Figure 1) on spring tides between 2021 and 2023. Sites were chosen to cover a range of natural and artificial habitats. Shore sites were accessed by vehicle; islands were surveyed by boat. At each site 6-10 specimens were hand collected in the intertidal zone using a hammer and chisel. The oysters were taken to an onshore base camp and tissue samples collected or the animals were frozen for transport back to Perth. A total of 26 sites were examined (Table 1):

- Dampier, 21-22 October 2021: Withnell Bay and King Bay (2 sites) on the west side of the Burrup Peninsula and a small boat landing in Dampier.
- Onslow, 25-29 August 2022: Beadon Creek; old Onslow jetty at Beadon Point; Four Mile Creek; Ashburton Island; east end of Thevenard Island and two Port of Ashburton channel markers.
- Dampier, 27-30 October 2022: John's Creek Boat Harbour, Point Samson; Watering Cove on the east side of the Burrup Peninsula (2 sites) and islands in the Dampier Archipelago: West Lewis Island (2 sites); Rosemary Island; Malus Island (2 sites); Whittaker Island; East Intercourse Island.
- Port Hedland, 10-12 February 2023: Intertidal creek, Balla Balla. Port Hedland: public boat ramp; Port Hedland reef; Finucane Island; Pretty Pool.

### *DNA extraction, PCR amplification and Sequencing*

DNA was extracted from adductor muscle of each specimen using the DNeasy Blood and Tissue Kit (Qiagen Inc., USA) following the manufacturer's instructions. Polymerase Chain Reaction (PCR) amplification of approximately 400 bases of the mitochondrial ribosomal subunit 16S gene region was done for each specimen, using primers 16Sar (Simon et al. 1994) and 16S2R-degenerate (Deagle et al. 2007).

PCR reactions were conducted in 25µl containing 3 µl DNA (~20 ng), 1x Invitrogen Platinum Green Hot Start PCR master mix (containing 1.5mM MgCl<sub>2</sub> and 0.2mM of each dNTP), 0.5mg/ml bovine serum albumin (Fisher Biotec, Australia) and 0.6µM each primer. PCR conditions consisted of an initial incubation at 95°C for 3 min, followed by 35 cycles of 94°C for 45s, 52°C for 90s, 72°C for 45s; and a final extension step of 72°C for 10min.

Bi-directional sequencing of unpurified PCR products was performed using the Sanger sequencing service provided by the Australian Genome Research Facility (AGRF, Perth). Sequences were trimmed and edited using the Geneious Prime 2022.1.1 software (<http://www.geneious.com>). For each individual, species identification was verified by similarity-based searches on the NCBI BLAST database (Altschul et al. 1990). To assign species a minimum

**Table 1.** Sites searched for intertidal oysters in the Pilbara, Western Australia. SA, *Saccostrea* lineage A; Ss, *Saccostrea scyphophilla*; Ta, *Talonostrea* sp. nov.

Port	Location	Habitat	GenBank accession number	Latitude (°S)	Longitude (°E)
Dampier	Withnell Bay, Burrup Peninsula	Rocky shore and <i>Avicennia marina</i> mangroves	SA [OR466862; OR466863] Ss [OR466904, OR466905]	-20.58	116.79
Dampier	South entrance King Bay	Low tide rocks	SA [OR466864] Ss [OR466906; OR466907; OR466908; OR466909]	-20.63	116.75
Dampier	Eastern King Bay	<i>A. marina</i> mangroves	SA [OR466865; OR466866; OR466867]	-20.63	116.76
Dampier	Dampier boat landing	Artificial rock groyne	SA [OR466868; OR466869]	-20.65;	116.71
Dampier	Watering Cove, Burrup Peninsula	Rocky shore and <i>A. marina</i> mangroves	SA [OR466888; OR466891; OR466896; OR466897]	-20.59	116.81
Dampier Archipelago	Norbill Bay, Rosemary Island	Intertidal rocks	Ss [OR466921; OR466922; OR466930; OR466931]	-20.46	116.61
Dampier Archipelago	Karratha Bay, West Lewis Island	Trial oyster farm	SA [OR466887, OR466892; OR466893] Ta [OR466936]	-20.57	116.65
Dampier Archipelago	Whalers Bay, Malus Island	Intertidal rocks	Ss [OR466925; OR466926]	-20.52	116.68
Dampier Archipelago	Marney Bay, Malus Island	Intertidal rocks	Ss [OR466923; OR466924]	-20.51	116.68
Dampier Archipelago	High Point, Whittaker Island	Intertidal rocks	Ss [OR466927, OR466928]	-20.54	116.68
Dampier Archipelago	North side of East Intercourse Island	Old jetty infrastructure	SA [OR466890] Ss [OR466929]	-20.65	116.69
Ashburton	Hooley Creek	Low tide rocks	SA [OR466870; OR466871; OR466877; OR466883; OR466884] Ta [OR466937]	-21.87	117.79
Ashburton	Ashburton Island	Coral reef and beachrock platform	Ss [OR466910; OR466911; OR466912; OR466913]	-21.59	114.94
Onslow	Direction Island	Beachrock platform	Ss [OR466917; OR466918; OR466919]	-21.53	115.13
Onslow	East end Thevenard Island	Jetty pilings	SA [OR466875; OR466876] Ss [OR466916]	-21.46	115.02
Onslow	Salt creek	<i>A. marina</i> mangroves	SA [OR466878] Ss [OR466920]	-21.61	115.19
Onslow	Old Onslow jetty, Beadon Point	Intertidal rocks and jetty pilings	SA [OR466885; OR466886] Ss [OR466914; OR466915]	-21.63	115.11
Onslow	Beadon Creek jetty	Intertidal rocks and jetty pilings	SA [OR466879; OR466880; OR466881; OR466882]	-21.65	115.13
Onslow	Four Mile Creek	Intertidal rocks	SA [OR466872, OR466873; OR466874]	-21.68	115.06
Point Samson	John's Creek Boat Harbour	Artificial rock groyne	SA [OR466889; OR466894; OR466895]	-21.64	117.19
Balla Balla	Intertidal creek	Under live <i>A. marina</i> branches in tidepool	Ta [OR466938; OR466939; OR466940]	-20.65	117.76
Port Hedland	Town boat ramp	<i>A. marina</i> mangroves and rocks	SA [OR466901] Ss [OR466932; OR466933]	-20.31	118.58
Port Hedland	Finucane Island	Rocky cliffs	Ss [OR466934; OR466935]	-20.50	118.53
Port Hedland	Port Hedland reef		SA [OR466902; OR466903]	-20.30	118.62
Port Hedland	Pretty Pool		SA [OR466898; OR466899; OR466900]	-20.32	118.61
Total			79		

requirement of  $\geq 99.4\%$  similarity to sequences previously deposited in GenBank by Lam and Morton (2006) and Snow et al (2023) was required. All sequences were submitted to GenBank <https://www.ncbi.nlm.nih.gov/genbank/> and assigned individual accession numbers (Table 1).

## Results

Seventy-nine oysters from the 26 sites were sequenced (Table 1). Three native species of intertidal rock oysters were recorded in the present survey.

### *Saccostrea lineage A (Figures 2a, c, 3; Table 1)*

*Brief description of shell:* Often densely packed so shells grow to fit available space. Medium size, oval, length 5 cm or less, hinge to tip of shell. Margins crenulated, margins may have short lobes that can be worn off. Outer surface off white. Inner white, adductor scar on left valve purple. Oysters grown individually for aquaculture up to 6 cm, elongate, left valve deeper.

*Records in the present survey:* Ashburton Port area: East end Thevenard Island; Hooley Creek; Salt Creek; Beadon Creek jetty; Four Mile Creek. Dampier Port area: Withnell Bay, Burrup Peninsula; Southern entrance to King Bay; Eastern King Bay; Dampier boat landing; Watering Cove, Burrup Peninsula; Karratha Bay, West Lewis Island; North side of East Intercourse Island. Port Samson port area: Point Samson. Port Hedland Port area: Public boat ramp, Port Hedland reef, Pretty Pool.

*Previous records in the Pilbara verified by DNA sequences:* Lam and Morton (2006): Exmouth; Barrow Island; Watering Cove; Withnell Bay. Snow et al. (2023): Flying Foam Passage, north of Burrup Peninsula.

*Notes:* *Saccostrea* lineage A occurred on a wide variety of habitats including lying loose on intertidal sandflats, and attached to isolated rocks, intertidal rock platforms, mangroves and artificial structures on both protected and open shores, sometimes at high densities. It frequently co-occurred with *S. scyphophilla*, but was more common on protected shores than *S. scyphophilla*.

*Saccostrea cucullata* (Born, 1778) was regarded as a widespread Indo-Pacific species with a variable shell morphology (Harry 1985), but recent DNA sequencing has divided the “species” into a number of separate genetic lineages (e.g. Lam and Morton 2006; Sekino and Yamashita 2016; McDougall 2020; Snow et al. 2023). The taxonomic status of the various lineages is uncertain. Oysters previously identified as *S. cucullata* may in fact be a species complex, with each lineage representing a separate species (McDougall 2020). Lam and Morton (2006) referred to the lineage occurring in the Pilbara as *S. cucullata* A, but more recent studies (McDougall 2020; McDougall et al. 2020; Snow et al. 2023) have reported the lineages without a species name, a process followed here. Snow et al. (2023) reported that *Saccostrea* lineage A occurs only in Western Australia, from Shark Bay to Broome. It is thus probably an undescribed species. Until this is done we have listed the WA specimens as *Saccostrea* lineage A and deposited specimens in the Western Australian Museum..

### *Saccostrea scyphophilla (Peron and Lesueur, 1807) (Figures 2b, 3; Table 1)*

*Brief description of shell:* Shell large, up to 10cm in length, very heavy. Usually occurs as individuals that in dense assemblages do not extend over each other. Upper surface rounded. Shell off white, margins purplish. Interior whitish, adductor muscle scar on right valve may be a deep purple.

*Records in the present survey:* Ashburton Port area: Direction Island; East end Thevenard Island; Ashburton Island; Salt Creek; Old Onslow jetty, Beadon Point. Dampier Port area: Withnell Bay; Southern entrance to King Bay; Norbill Bay, Rosemary Island; Whalers Bay, Malus Island; Marney Bay, Malus Island; High Point, Whittaker Island; North side of East Intercourse Island. Port Hedland Port area: Public boat ramp; Finucane Island.

*Previous records in the Pilbara verified by DNA sequences:* Lam and Morton (2006): Vlamingh Head, north of Exmouth; Barrow Island; Withnell Bay, Burrup Peninsula (all as *S. mordax* (Gould, 1850). Snow et al. (2023): Flying Foam Passage, north of Burrup Peninsula.

*Published distribution (based on DNA analyses):* Okinawa, Taiwan, Hong Kong, China, Singapore, northern Australia (Western Australia, Northern Territory, Queensland, New South Wales) (Lam and Morton 2006 (as *S. mordax*); McDougall 2020; McDougall et al. 2020; Snow et al. 2023).

*Notes:* *Saccostrea scyphophilla* also occurred on a wide variety of habitats, from isolated rocks, intertidal rock platforms, mangroves and artificial structures on both open and protected shores, sometimes at high densities. It frequently co-occurred with *Saccostrea* lineage A, but was more common on open, exposed shores than *Saccostrea* lineage A.

Snow et al. (2023) commented that *S. scyphophilla* in WA was previously reported as two lineages, A and B by Lam and Morton (2006), with B being very rare. Snow et al. (2023) considered that the differences between lineages were relatively small and combined the two, a practice followed here.

*Talonostrea sp. nov. (Figure 2d, e)*

*Brief description of shell:* Shell small, 4cm or less in length, flattened, fragile, lack chomata, outer margin of left valve may be extended upwards. Occur as single individuals, shape variable to elongate depending on substrate. Outer shell colour variable, white to deep purple. Interior of shell chalky white. Adductor scar usually white, but may be light purple.

*Records in the present survey:* Ashburton Port area: Hooley Creek. Dampier Port Area: Karratha Bay, West Lewis Island. Balla Balla port area: Intertidal creek.

*Previous records in the Pilbara verified by DNA sequences:* None published. The WA Department of Primary Industry and Regional Development (DPIRD) has sequences from Flying Foam Passage, Dampier and Cone Bay, Kimberley collected between 2018-2020. The specimens were small and were obtained from spat collectors used during aquaculture trials.

*Published distribution (based on DNA analyses):* Moreton Bay to Cairns, Queensland (McDougall et al. 2020).



*Notes:* The Hooley Creek specimen was collected on an intertidal sandflat in the creek. The Karratha Bay specimen was found on a steel support for the aquaculture facility. Six specimens were collected at the base of mangroves (*Avicennia marina*) at Balla Balla.

McDougall et al. (2020) believe this is an undescribed species. WA specimens have been deposited in the Western Australian Museum.

## Discussion

The present study has achieved the primary goals of using genetic sequencing to determine the rock oyster species present in the Pilbara and their distribution. Seventy-nine oysters were genetically sequenced in the present study. Lam and Morton (2006) and Snow et al. (2023) provided an additional 31 DNA sequences from the Pilbara, for a total of 110 sequences. Three native species, *Saccostrea* lineage A, *S. scyphophilla*, and *Talonostrea* sp. nov. were detected and are found throughout the Pilbara, including ports and nearby localities. No IMS were detected. The study provides a solid baseline for monitoring for any future invasions of rock oysters.

The two genetically distinct lineages (*Saccostrea* lineage A and *S. scyphophilla*) previously known from the Pilbara (Lam and Morton 2006; Snow et al. 2023) have been shown to be widespread in the region. The records of *Talonostrea* sp. nov. are the first published records of the species outside Queensland. In addition to the present study the new *Talonostrea* has also been collected by DPIRD at Cone Bay in the Kimberley, WA and verified by DNA sequences, but no specimens were retained.

Several intertidal oyster species have been reported in marine biodiversity surveys conducted in the Pilbara at the Montebello Islands (Wells et al. 2000) and the Dampier Archipelago (Taylor and Glover 2004; Slack-Smith and Bryce 2004): *Saccostrea glomerata* (Gould, 1850) (as *Saccostrea commercialis* (Iredale and Roughley, 1933), *Saccostrea cucullata* (Born, 1778) and *Saccostrea echinata* (Quoy & Gaimard, 1832). None of these was confirmed by DNA sequenced material. The *S. echinata* record of Wells et al. (2000) was from a single site in the Montebello Islands and was erroneous. *Saccostrea commercialis* was reported from numerous sites (Taylor and Glover 2004; Slack-Smith and Bryce 2004). This is the well-known Sydney rock oyster that is a key aquaculture species in Australia. The only DNA validated record in WA is Albany on the south coast, where it was introduced through aquaculture (Lam and Morton 2006; Snow et al. 2023). The specimens recorded as *S. commercialis* are thus likely to be *Saccostrea* lineage A. ALA (2022) records an oyster valve (C.312696) in the Australian Museum identified as *Crassostrea bilineata* (Röding, 1798) (now *Magallana bilineata*) by the Japanese researcher Dr K. Torigoe. The valve was collected southeast of Exmouth homestead on Exmouth Gulf. Willan et al. (2021) examined the valve and concluded it is *Planostrea pestigris* (Hanley, 1846), but again this has not been confirmed by DNA sequencing.

Roberts et al. (2002) analysed the worldwide diversity of corals, fish, molluscs and rock lobsters on coral reefs and identified 18 global biodiversity hotspots. The west coast of WA, between North West Cape and Rottneet Island (32°S off Perth) was a key hotspot. It ranked second in restricted range species (56), seventh in species with broader ranges (768 in the four groups) and fifteenth in terms of anthropogenic threat. The Pilbara is located at the western edge of the Tropical Australian Province, part of the vast Indo-West Pacific (Wilson and Allen 1987; Morgan and Wells 1991). The WA hotspot lies immediately adjacent to the Pilbara. With 5532 known species in major taxa, including 1533 shallow water molluscs and 1565 fish (Wells 2018) there is a substantial basis for extending the WA west coast hotspot to include the Pilbara.

In this context it is surprising that only three native intertidal species of oysters were confirmed in the present study. Two (*S.* lineage A and *S. scyphophilla*) were reported from the adjacent Kimberley region of WA, along with *S.* lineage B and *S.* lineage J (Snow et al. 2023). *Talonostrea* sp. nov. was also recorded on spat collectors as discussed above. In comparison, a recent study (McDougall 2020) recorded 14 lineages in Queensland. Nineteen sites were listed, with a maximum diversity of 7 lineages in Cairns and 5 or 6 lineages at 8 other sites. A survey of Singapore oysters conducted in the same manner as the present study recorded two species of *Magallana* (*M. bilineata* and *M. belcheri* (G.B. Sowerby II, 1871) and several lineages of *Saccostrea* (Tan et al. unpublished). The limited intertidal oyster diversity in the Pilbara is not explained by a lack of habitat diversity. The offshore islands have a variety of habitats, including protected lagoons, bays, mangroves, channels, rocky and sandy shores and coral reefs (Wells and Berry 2000). Inshore, the continental coastline has extensive low diversity mangrove systems and rocky and sandy shores. The region is arid, so there are no major permanent rivers or estuaries (Wells and Walker 2003).

Huisman et al. (2008) reported that 60 introduced marine species have established populations in WA; 13 of these were recorded in the Pilbara, only one of which (the ascidian *Didemnum perlucidum*) is considered to be invasive. None of the identified introductions were oysters. The extraordinarily high known marine biodiversity (5532 known species; Wells 2018) combined with the single known IMS means that it is essential that all practical steps be taken to protect the Pilbara marine environment from the establishment of additional IMS.

### Authors' contribution

FEW: research conceptualization; sample design and methodology; investigation and data collection; data analysis and interpretation; and roles/writing – original draft; writing – review and editing. SSL: research conceptualization; sample design and methodology; investigation and data collection; data analysis and interpretation; and writing – review and editing. LAF: investigation and data collection; data analysis and interpretation; and writing – review and editing. ESH: investigation and data collection; data analysis and interpretation; funding provision; and writing – review and editing.

## Acknowledgements

This work was undertaken as part of the eDNA for Global Environment Studies (eDGES) program (<https://research.curtin.edu.au/scieng/edges/>) at Curtin University. We warmly thank BHP and their program manager, Dr Tim Cooper, for their invaluable support. David and Pip Arthur of Karratha Boat Hire kindly provided and skippered boats for fieldwork and access to a camp in the Dampier Archipelago. Louise Purkis and Anurodh Prasad of the Pilbara Ports Authority generously organised access to some of the sites in Onslow. We would also like to thank Dr Seema Fotedar (DPIRD) for providing sequence data and Warren Ure who provided information on the Pilbara oyster aquaculture trial. Collection permits were provided by the Western Australian Departments of Primary Industry and Rural Development (DPIRD) and Biodiversity Conservation and Attractions (DBCA). WAM registrations were done courtesy of Dr Lisa Kirkendale and Corey Whisson. Karl Schramm kindly helped prepare Figure 1. We very much appreciate the constructive comments made by reviewers.

## Funding declaration

The project was funded by BHP's Social Investment Framework, 'Environment' stream by contributing to "biodiversity conservation, water stewardship and climate change mitigation and adaptation". The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

## Permits

The project was undertaken with Fisheries exemption 250965722 of the Western Australian Department of Primary Industry and Regional Development (DPIRD) and permit and Department of Biodiversity Conservation and Attractions (DBCA) Fauna Taking (Scientific and Other Purposes) licence FO25000415-2.

## References

- Altschul SF, Gish W, Miller W, Myers EW, Lipman DJ (1990) Basic local alignment search tool. *Journal of Molecular Biology* 215: 403–410, [https://doi.org/10.1016/S0022-2836\(05\)80360-2](https://doi.org/10.1016/S0022-2836(05)80360-2)
- Crowe TP, Frid CLJ (eds) (2015) *Marine Ecosystems: Human Impacts on Biodiversity, Functioning and Services*. Cambridge University Press, Cambridge, UK, 409 pp, <https://doi.org/10.1017/CBO9781139794763>
- Deagle BE, Gales NJ, Evans K, Jarman SN, Robinson S, Trebilco R, Hindell MA (2007) Studying seabird diet through genetic analysis of faeces: a case study on Macaroni Penguins (*Eudyptes chrysolophus*). *PLoS ONE* 2: e831, <https://doi.org/10.1371/journal.pone.0000831>
- Dias PJ, Fotedar S, Munoz J, Hewitt MJ, Lukehurst S, Hourston M, Wellington C, Duggan R, Bridgwood S, Massam M, Aitken V, de Lestang P, McKirdy S, Willan R, Kirkendale L, Giannetta J, Corsini-Foka M, Pothoven S, Gower F, Viard F, Buschbaum C, Scarcella G, Strafella P, Bishop MJ, Sullivan T, Buttino I, Madduppa H, Huhn M, Zabin CJ, Bacela-Spychalska K, Wójcik-Fudalewska D, Markert A, Maximov A, Kautsky L, Jaspers C, Kotta J, Pärnoja M, Robledo D, Tsiamis K, Küpper FC, Žuljević A, McDonald JI, Snow M (2017) Establishment of a taxonomic and molecular reference collection to support the identification of species regulated by the Western Australian Prevention List for Introduced Marine Pests. *Management of Biological Invasions* 8: 215–225, <https://doi.org/10.3391/mbi.2017.8.2.09>
- Dias PJ, Lukehurst SS, Simpson T, Rocha RM, Tovar-Hernández MA, Wellington C, McDonald JI, Snow M, Kennington WJ (2021) Multiple introductions and regional spread shape the distribution of the cryptic ascidian *Didemnum perlucidum* in Australia: an important baseline for management under climate change. *Aquatic Invasions* 16: 297–313, <https://doi.org/10.3391/ai.2021.16.2.06>
- Harry HW (1985) Synopsis of the supraspecific classification of living oysters (Bivalvia, Gryphaeidae and Ostreidae). *The Veliger* 28: 121–158.
- Herbert RJH, Humphreys J, Davies CJ, Roberts C, Fletcher S, Crowe TP (2016) Ecological impacts of non-native Pacific oysters (*Crassostrea gigas*) and management measures for protected areas in Europe. *Biodiversity Conservation* 25: 2835–2865, <https://doi.org/10.1007/s10531-016-1209-4>
- Huisman JM, Jones DS, Wells FE, Burton T (2008) Marine introductions into Western Australian waters. *Records of the Western Australian Museum* 25: 1–44, [https://doi.org/10.18195/issn.0312-3162.25\(1\).2008.001-044](https://doi.org/10.18195/issn.0312-3162.25(1).2008.001-044)
- Johnson CR, Chapman RDO (2007) Seaweed invasions: Introduction and scope. *Botanica Marina* 50: 321–325, <https://doi.org/10.1515/BOT.2007.037>
- Katsanevakis S, Wallentinus I, Zenetos A, Leppäkoski E, Çinar ME, Oztürk B, Grabowski M, Golani D, Cardoso AC (2014) Impacts of invasive alien marine species on ecosystem services

- and biodiversity: a pan-European review. *Aquatic Invasions* 9: 391–423, <https://doi.org/10.3391/ai.2014.9.4.01>
- Lam K, Morton B (2006) Morphological and mitochondrial-DNA analysis of the Indo-West Pacific rock oysters (Ostreidae: *Saccostrea* species). *Journal of Molluscan Studies* 72: 235–245, [https://doi.org/10.1016/S0044-8486\(03\)00215-1](https://doi.org/10.1016/S0044-8486(03)00215-1)
- Lam K, Morton B (2009) Oysters (Bivalvia: Ostreidae and Gryphaeidae) recorded from Malaysia and Singapore. *Raffles Bulletin of Zoology* 57: 481–494
- McDonald JI, Wellington CM, Coupland GT, Pedersen D, Kitchen B, Bridgwood SD, Hewitt M, Duggan R, Abdo DA (2020) A united front against marine invaders: Developing a cost-effective marine biosecurity surveillance partnership between government and industry. *Journal of Applied Ecology* 57: 77–84, <https://doi.org/10.1111/1365-2664.13557>
- McDougall C (2020) Reinvigorating the Queensland oyster industry. Final Report to the Fisheries Research and Development Corporation, Griffith University, Brisbane, Queensland, November. 25 pp
- McDougall C, Nenadic N, Healy J (2020) Guide to Queensland's intertidal oysters. Griffith University, Brisbane, Queensland, Australia, 9 pp
- Molnar JL, Gamboa RL, Revenga C, Spalding MD (2008) Assessing the global threat of invasive species to marine biodiversity. *Frontiers in Ecology and the Environment* 6: 485–492, <https://doi.org/10.1890/070064>
- Morgan GJ, Wells FE (1991) Zoogeographic provinces of the Humboldt, Benguela and Leeuwin Current systems. *Journal of the Royal Society of Western Australia* 74: 59–69
- Roberts CM, McClean CJ, Veron JEN, Hawkins JP, Allen GR, McAllister DE, Mittermeier CG, Schueler FW, Spalding M, Wells F, Wynne C, Werner TB (2002) Marine biodiversity hotspots and conservation priorities for tropical reefs. *Science* 295: 1280–1284, <https://doi.org/10.1126/science.1067728>
- Ruesink JL, Lenihan HS, Trimble AC, Heiman KW, Micheli F, Byers JE, Kay MC (2005) Introduction of non-native oysters: ecosystem effects and restoration implications. *Annual Review of Ecology, Evolution and Systematics* 36: 643–689, <https://doi.org/10.1146/annurev.ecolsys.36.102003.152638>
- Salimi PA, Creed JC, Esch MM, Fenner D, Jaafar Z, Levesque JC, Montgomery AD, Salimi MA, Edward JKP, Raj KD, Sweet M (2021) A review of the diversity and impact of invasive non-native species in tropical marine ecosystems. *Marine Biodiversity Records* 14: 11, <https://doi.org/10.1186/s41200-021-00206-8>
- Salvi D, Mariottini P (2016) Molecular taxonomy in 2D: a novel ITS2 rRNA sequence-structure approach guides the description of the oysters' subfamily Saccostreinae and the genus *Magallana* (Bivalvia: Ostreidae). *Zoological Journal of the Linnean Society* 179: 263–276, <https://doi.org/10.1111/zoj.12455>
- Seebens H, Gastner MT, Blasius B (2013) The risk of marine bioinvasion caused by global shipping. *Ecology Letters* 16: 782–790, <https://doi.org/10.1111/ele.12111>
- Sekino M, Yamashita H (2016) Mitochondrial and nuclear DNA analyses of *Saccostrea* oysters in Japan highlight the confused taxonomy of the genus. *Journal of Molluscan Studies* 82: 492–506, <https://doi.org/10.1093/mollus/eyw022>
- Simon C, Frati F, Beckenbach A, Crespi B, Liu H, Flook P (1994) Evolution, weighting, and phylogenetic utility of mitochondrial gene sequences and a compilation of conserved polymerase chain reaction primers. *Annals of the Entomological Society of America* 87: 651–701, <https://doi.org/10.1093/aesa/87.6.651>
- Slack-Smith SM, Bryce CW (2004) A survey of the benthic molluscs of the Dampier Archipelago, Western Australia. *Records of the Western Australian Museum Supplement* 66: 221–245, <https://doi.org/10.18195/issn.0313-122x.66.2004.219-245>
- Smith KA, Brown S, Hart AM, Bissell A (2023) Ecological Risk Assessment for the Western Australian Silverlip Pearl Oyster (*Pinctada maxima*) Resource. Fisheries Research Report No. 330. Department of Primary Industries and Regional Development, Western Australia, 90 pp
- Snow M, Fotadar S, Wilson NG, Kirkendale LA (2023) Clarifying the natural distribution of *Saccostrea* Dollfus and Dautzenberg, 1920 (edible rock oyster) species in Western Australia to guide development of a fledgling aquaculture industry. *Aquaculture* 566: 739202, <https://doi.org/10.1016/j.aquaculture.2022.739202>
- Tan SK, Woo HPM (2010) A Preliminary Checklist of the Molluscs of Singapore. Raffles Museum of Biodiversity Research, National University of Singapore, Singapore, 78 pp
- Taylor JD, Glover EA (2004) Diversity and distribution of subtidal benthic molluscs from the Dampier Archipelago, Western Australia; results of the 1999 dredge survey (DA2/99). *Records of the Western Australian Museum Supplement* 66: 247–291, <https://doi.org/10.18195/issn.0313-122x.66.2004.247-291>
- Thomson JM (1952) The acclimatization and growth of the Pacific oyster (*Gryphaea gigas*) in Australia. *Australian Journal of Marine and Freshwater Research* 3: 64–73, <https://doi.org/10.1071/MF9520064>
- Thomson JM (1959) The naturalization of the Pacific oyster in Australia. *Australian Journal of Marine and Freshwater Research* 10: 144–149, <https://doi.org/10.1071/MF9590144>

- Wells FE (2018) A low number of invasive marine species in the tropics: a case study from Pilbara (Western Australia). *Management of Biological Invasions* 9: 227–237, <https://doi.org/10.3391/mbi.2018.9.3.05>
- Wells FE, Berry PF (2000) The physical environment, marine habitats, and characteristics of the marine fauna. In: Berry PF, Wells FE (eds), Survey of the marine fauna and habitats of the Montebello Islands, Western Australia. *Records of the Western Australian Museum Supplement* 59: 9–13
- Wells FE, Booth G (2012) Setting a new benchmark in managing biofouling on vessels in a sensitive marine area. Joint Health, Safety and Environment conference of the Society of Professional Engineers and the Australian Petroleum Producers and Exploration Association, Perth, September 2012. 11 pp, <https://doi.org/10.2118/156749-MS>
- Wells FE, Walker DI (2003) Introduction to the Dampier marine environment. In: Wells FE, Walker DI, Jones DS (eds), The Marine Flora and Fauna of Dampier, Western Australia. Western Australian Museum, Perth, pp 1–12
- Wells FE, Slack-Smith SM, Bryce CW (2000) Molluscs of the Montebello Islands. In: Berry PF, Wells FE (eds), Survey of the marine fauna and habitats of the Montebello Islands, Western Australia. *Records of the Western Australian Museum Supplement* 59: 29–46
- Willan RC, Nenadic N, Ramage A, McDougall C (2021) Detection and identification of the large, exotic, crassostreine oyster *Magallana bilineata* (Röding, 1798) in northern Queensland, Australia. *Molluscan Research* 41: 64–74, <https://doi.org/10.1080/13235818.2020.1865515>
- Wilson BR, Allen GR (1987) Major components and distribution of marine fauna. In: Dyne GR, Walton DW (eds), The Fauna of Australia. Volume 1A. General Articles. Bureau of Flora & Fauna, Australian Government Printing Service, Canberra, Australia, pp 43–68

## Web sites and online databases

- ALA (2022) *Saccostrea*. Atlas of Living Australia. <https://bie.ala.org.au/search?sortField=&dir=desc&q=Saccostrea> (accessed 21 November 2022)
- Curtin (2022) eDNA for Global Environment Studies (eDGES). <https://research.curtin.edu.au/scieng/edges/> (accessed 17 November 2022)
- DAFF (2023) Non-commercial vessel arrival biosecurity reporting - yachts and private superyachts. Department of Agriculture, Fisheries and Forestry, Canberra ACT. <https://www.agriculture.gov.au/biosecurity-trade/aircraft-vessels-military/vessels/non-commercial-vessels#where-in-australian-territory-can-noncommercial-vessel-operators-arrive> (accessed 12 November 2023)
- DPIRD (2016) Western Australian Prevention List for Introduced Marine Pests. Department of Primary Industry and Regional Development, Perth, WA. <http://www.fish.wa.gov.au> (accessed 25 May 2023)
- MarinePests (2023) Map of marine pests. <https://www.marinepests.gov.au/> (accessed 23 November 2023)
- NIMPCG National Introduced Marine Pests Coordinating Group (2009a) *Marine pests monitoring manual: Version 1*. National Introduced Marine Pests Coordinating Group, Department of Agriculture, Fisheries and Forestry, Canberra, Australia. [http://www.marinepests.gov.au/marine\\_pests/publications/Pages/monitoring\\_manual.aspx](http://www.marinepests.gov.au/marine_pests/publications/Pages/monitoring_manual.aspx) (accessed 15 October 2017)
- NIMPCG National Introduced Marine Pests Coordinating Group (2009b) Australian marine pests monitoring guidelines: version 1. National Introduced Marine Pests Coordinating Group, Department of Agriculture, Fisheries and Forestry, Canberra, Australia. [http://www.marinepests.gov.au/marine\\_pests/publications/Pages/monitoring\\_guidelines.aspx](http://www.marinepests.gov.au/marine_pests/publications/Pages/monitoring_guidelines.aspx) (accessed 15 October 2017)
- NIMPIS (2022) Species - *Magallana (Crassostrea) gigas*. National Introduced Marine Pest Information System, Canberra Australia. <https://nimpis.marinepests.gov.au/species/species/133> (accessed 18 November 2022)
- Ports Australia (2022) Trade statistics. <https://www.portsaustralia.com.au/resources/trade-statistics> (accessed 21 November 2022)
- PPA (2022) 2021-2022 Pilbara Ports Authority Annual Report. Pilbara Ports Authority. <https://www.pilbaraports.com.au/about-ppa/publications/annual-report> (accessed 25 November 2022)
- Vessel-Check (2022) Vessel-Check Aquatic Biosecurity Solutions. <https://vessel-check.com/> (accessed 17 November 2022)