BioInvasions Records (2020) Volume 9, Issue 3: 461–470

Rapid Communication

First record of the plumose sea anemone, *Metridium senile* (Linnaeus, 1761), from the Falkland Islands

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Citation: Glon HE, Costa M, de Lecea AM, Goodwin C, Cartwright S, Díaz A, Brickle P, Brewin PE (2020) First record of the plumose sea anemone, *Metridium senile* (Linnaeus, 1761), from the Falkland Islands. *BioInvasions Records* 9(3): 461–470, https://doi.org/10.3391/bir.2020.9.3.02

Received: 20 February 2020
Accepted: 19 May 2020
Published: 30 July 2020

Handling editor: Linda Auker
Thematic editor: Andrew David

Abstract

Metridium senile is a circumboreally distributed sea anemone (Cnidaria: Anthozoa: Actiniaria) native to the northern hemisphere, and has been presumed as introduced to several locations in the southern hemisphere. Although the sea anemone fauna of the Falkland Islands is not well known, to date no historical records of *Metridium senile* exist. In 2019, we conducted biodiversity surveys to gather information about sea anemones present in the islands. During these 2019 surveys, we detected *M. senile* from three locations: Stanley (eastern Falklands), the Bird Island (south-west Falklands), and in the northern Jason Islands (north-west Falklands). The species was well established at each location, with evidence for asexual reproduction occurring in the western sites. We confirmed the identification by morphology and DNA analysis. Stanley populations are distinct in color variation from the southwestern and northwestern populations, which may indicate separate introduction waves or pathways and the potential for additional, undetected locations of *M. senile*.

Key words: introduced species, Actiniaria, benthic community, marine invasions, marine biology, non-native

Introduction

Sea anemones (Actiniaria: Anthozoa: Cnidaria) are ubiquitous components of benthic marine communities, exhibiting a higher species diversity in temperate (30–40°N) latitudes (Fautin et al. 2013). A prominent cold-water species of sea anemone, *Metridium senile* (Linnaeus, 1761), has a native circumboreal range in the northern hemisphere. Members of *Metridium* de Blainville, 1824 are distinctive as they have lobed oral discs and short tentacles, with individuals not easily confused with species outside of the

genus once mature. However, outside of these key characteristics within *M. senile* there is a high level of morphological plasticity, as individuals can range widely in size, shape, and color. Because of the wide variation and challenging taxonomic history within the genus, there has been taxonomic confusion particularly in the usage of *M. senile* and *Metridium dianthus* (Ellis, 1768). For the purpose of this study, we treat these names as synonyms and use the older name, *M. senile*. An investigation into their systematics is ongoing by several of the authors.

Metridium senile prefers hard substrate of rocks, bivalves, docks, shipwrecks, and pilings and are abundant from the intertidal zone to depths of 100 m (Carlgren 1933; Hoffmann 1976). These anemones are primarily found in temperatures from −1 to 20 °C, though individuals can tolerate short exposures to temperatures of up to 24 °C (Stephenson 1935; Glon et al. 2019). Individuals can withstand a broad range of salinity from 14.8 to 37.5 ppt, though they are commonly found at salinities between 20 and 37 ppt (Shumway 1978; Glon et al. 2019). Reproduction in *M. senile* can occur either through sexual means or via asexual reproduction through pedal laceration, the latter of which can occur prolifically when conditions (temperature, salinity, substrate) are within the preferred range (Hoffmann 1986; Anthony and Svane 1995; Glon et al. 2019).

Metridium senile is confirmed as introduced in the southern hemisphere. In South Africa, *M. senile* was first detected at shallow depths in Table Bay Harbour in Cape Town (Griffiths et al. 1996). Since then, it was recorded along pipelines at 115 m deep, and on oil rigs at 126 m deep on the Agulhas Bank (Mead et al. 2011; Laird and Griffiths 2016). In South America, *Metridium* was first described from Chile in 1904 as *M. parvulum* (McMurrich 1904), but was later synonymized with the *M. senile* subsp. *lobatum* described in Argentina (Riemann-Zürneck 1975). Currently, *M. senile* dominates fouling communities in southern Patagonia, Argentina in Bahía San Julián and surrounding natural areas, as well as slightly north in the intertidal and subtidal of Ría Deseado (Martin et al. 2015). In Chile, however, *Metridium* was not documented again until 2006, despite intensive surveys in the areas where it had been originally described from a wide region and depth gradient, leaving doubts as to the original identification and description (Häussermann and Försterra 2005; Häussermann 2006). *Metridium senile* is currently well-established south of the Peninsula Taitao in several of the Chilean Patagonian fjords to the point of displacing the native *Anthothoe chilensis* (Lesson, 1830) (Häussermann 2006).

The Falkland Islands are located on the Patagonian shelf in the southwest of the Atlantic Ocean, about 500 km off the coast of South America on the Falkland Plateau. This shallow region is heavily influenced by the effects of the Cape Horn Current (e.g. dispersal of organisms, temperature, and salinity) from the southeast Pacific, which flows past the southern tip of South America to the east where it branches in the cold

Eastern and Western Falkland Currents in the Atlantic, both also intensified by the Antarctic Circumpolar Current (Howe et al. 1997; Arkhipkin et al. 2013). Despite their remote location, climatic models have suggested that the Falkland Islands are susceptible to invasive terrestrial species under a warming climate (Duffy et al. 2017) and thus may become more susceptible to marine invasions over time as well. Dispersal of organisms is hypothesized to be more likely to occur from Chile rather than from Argentina, closely aligning their faunal biodiversity, despite the presence of the Argentine drift traveling south (Riemann-Zürneck 1986; Häussermann and Försterra 2005). Riemann-Zürneck (1986) studied benthic fauna from around South America including the Falklands, detailing nineteen species of sea anemones collected. Though the shallow-occurring *Antholoba achates* (Drayton in Dana 1846), which prefers similar habitat to *Metridium senile*, was among those occurring in the Falklands listed in this survey, no presence of *M. senile* was detected near the Falkland Islands.

Suspected individuals of *M. senile* in Stanley Harbour in a site known as the "Narrows", which connects the Stanley Harbour basin to Port William, were first photographed in large numbers at a single dive survey location, and reported as yet unidentified in a technical report (Davidson 2013). *Metridium* was also suspected as occurring at Navy Point in Port William, but the identification was never confirmed (Brewin *pers. obs.*). This study documents the first confirmed record of *M. senile* for the Falkland Islands, and summarizes our current understanding of its presence there.

Materials and methods

Collections

The first synoptic survey of the subtidal marine flora and fauna of the Falklands was conducted in 1996, and since 2006 has been regularly surveyed via SCUBA (Beaton et al. 2020), with a particularly large effort in the vicinity of Stanley including Port William and Berkeley Sound (unpublished reports can be found at https://smsg-falklands.org/). In November 2019, we undertook marine biodiversity surveys to document the species of actiniarians present. Using either SCUBA (down to 20 m depth), snorkeling, or intertidal surveys, we surveyed twenty-two locations (Figure 1). All specimens were collected by hand or with a small spatula. Preservation of anemones for identification occurred in 10% formalin or in 96% ethanol, with a full change of the ethanol within one week of collection.

Genetics

We extracted total genomic DNA from two individuals, one each from Stanley Harbour and Bird Island, using the E.Z.N.A. Tissue DNA Kit (Omega Bio-Tek, Norcross, GA, USA) according to the manufacturer's instructions. We amplified the mitochondrial 16S rRNA (468 bp) and nuclear

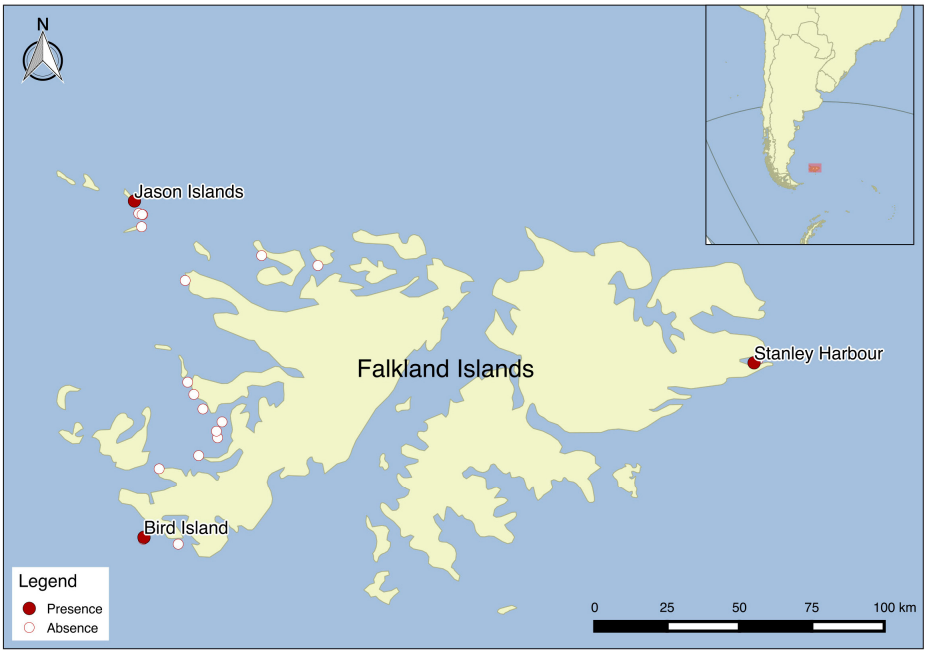

Figure 1. Locations surveyed throughout Falkland Islands for biodiversity surveys. Red points designate locations where *Metridium senile* was encountered and collected, white points designate sites surveyed via SCUBA where *M. senile* was not encountered.

18S rRNA (1792 bp) regions using primers according to Apakupakul et al. (1999) and Geller and Walton (2001) and described in Daly et al. (2008). We cleaned PCR products using ExoSAP-IT (Affymetrix, Santa Clara, CA, USA) and performed standard Sanger sequencing at the Ohio State University Biomedical Research Tower (subsidized shared resource P30CA016058). Post-processing, we used Geneious R11 (www.geneious.com; Kearse et al. 2012) to visually edit and trim the forward and reverse reads for quality before assembling our resulting DNA sequences. For all of the sequences, we ran a BLAST query against the NCBI nucleotide database.

Results

In total, *M. senile* was recorded from three locations in the Falkland Islands between the depths of 6 and 20 m (Supplementary material Table S1, Figure 1). All locations had a high number and density of individuals, indicating that reproduction was actively occurring.

Stanley Harbour

The occurrence of *Metridium senile* in Stanley Harbour at the site previously surveyed has now been confirmed by our subsequent survey and collection dive in 2019. The substrate at this site is soft silt, though generally *M. senile* prefers hard substrate (Hoffmann 1976). However, the presence of *Macrocystis* and *Lessonia* (kelp) blades laying across the sediment surface originating from drift, as well as bivalves (e.g. *Aulacomya ater*), provide suitable substrate. The density of *M. senile* rarely exceeded one individual per square meter. Visibility was very poor (< 2 m), which is

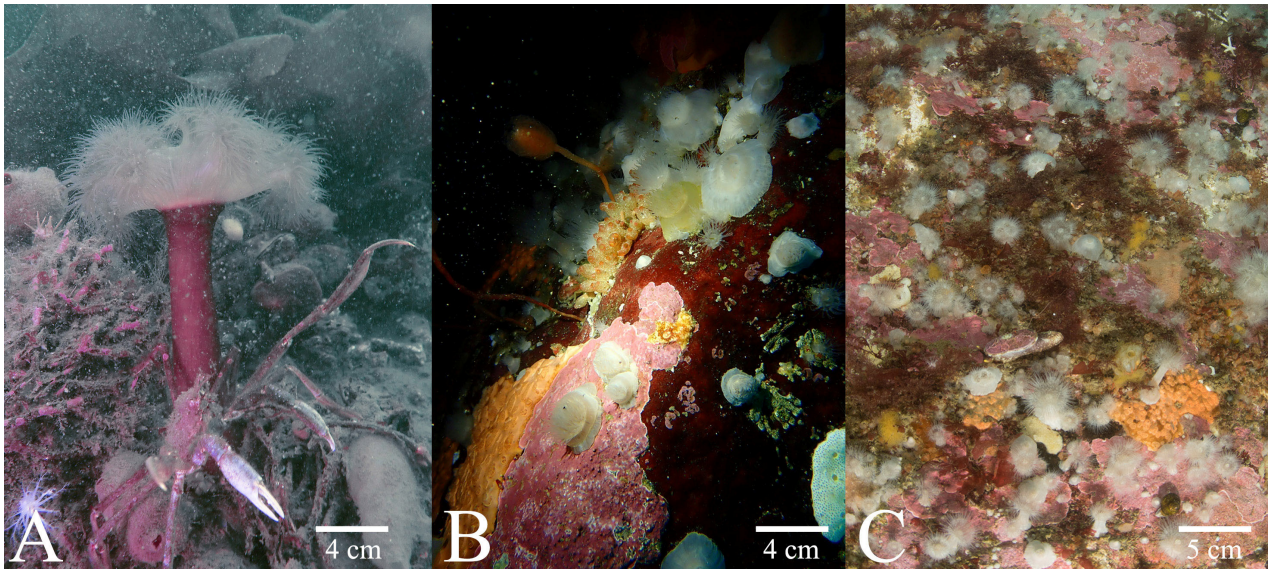

Figure 2. Photographs of *Metridium senile* from 2019 surveys recorded from (A) Stanley Harbour, (B) Bird Island, and (C) Flat Jason Island. Scale bars are approximate. Photos by Heather Glon and Claire Goodwin.

typical of the semi-closed small basins found along the coastline of the Falkland Islands. All individuals observed at this location had brown columns with white tentacles, and were taller than they were wide (Figure 2A). Several individuals exceeded 20 cm in height, and most had strongly lobed oral discs. There were no clonal groups encountered; rather, they were virtually all solitary individuals, with no evidence of nearby propagules originating from pedal laceration.

Bird Island

The waters surrounding Bird Island are under consideration for conservation protection. Prior to these surveys, few surveys of marine benthos had been completed in the waters surrounding the island. The habitat at the site where *M. senile* was discovered is characterized by large boulders and stands of kelp (*Macrocystis pyrifera* (Linnaeus) C. Agardh, 1820). These provided suitable hard substrate for *M. senile*, encompassing a variety of horizontal and vertical surfaces. Visibility was good (> 10 m) at the site, as no main source of freshwater discharge was present. Column color included white, tan, and orange, which is typical of *M. senile* throughout its native range, and showed more color variation than the individuals in Stanley Harbour. Polyps were generally wider than they were tall, though the size difference may be due to different environmental conditions (Figure 2B). Because of their smaller size, not all had strongly lobed oral discs. Additionally, smaller individuals at the base of the pedal disc of multiple observed specimens, as well as the prevalence of dense groups (> 30 individuals/m^2) in some areas, indicated the presence of asexual reproduction. The population was isolated to an area roughly 100 m^2.

Flat Jason Island

At Flat Jason Island, a small group of *M. senile* was observed on a small boulder. An extensive and dense area of *M. senile*, indicating potential asexual reproduction, was observed on a nearby steeply sloping bedrock wall. Both areas had dense kelp cover (*Lessonia vadosa* Searles, 1978). The overall physical appearance of the animals at Flat Island resembled those of Bird Island (Figure 2C), although only the white color morph was observed.

Genetics

Sequence identification with our sequences (Table S2) from Stanley Harbour and Bird Island using NCBI BLAST displayed 100% identity with established *M. senile* 16S genes within the mitochondrial genome (e.g. KJ482950.1, HG423143.1, JF832972.1, JF832971.1) and 18S genes (e.g. AF052889.1, KJ483035.1). As the individuals at Bird Island and Flat Jason were morphologically similar, only an individual from Bird Island was sequenced.

Discussion

The data collected in this study suggest that *M. senile* is established at multiple locations in the Falkland Islands, which increases the introduced range. Populations at two of the three locations sampled suggest active asexual reproduction. The majority of individuals discovered exhibited the characteristic lobed oral disc that is shared, but not easily confused with, *A. achates*.

Shipping traffic is considered to be one of the leading causes for introductions of invasive species worldwide, and introductions are expected to increase in the future with heightened boat traffic (Mills et al. 1993; Ruiz et al. 2000; McGee et al. 2006). Ballast water is a well-known vector for the transport of invasive species, as pelagic species (including larvae) can be brought in with the intake of water at the initial port and released subsequently at a different location (Carlton 1985, 2009; Carlton and Geller 1993). *Metridium senile* can be a dominating component of fouling communities within both its native (e.g. Nelson and Craig 2011) and introduced range (e.g. Martin et al. 2015), and has been recorded in the fouling communities on both the hulls of ships (Visscher 1928) and in ballast water (Carlton and Geller 1993).

Individuals from Stanley Harbour appear morphologically (e.g. brown color) distinct from those at Bird Island and Flat Jason Island, indicating a potential for genetic divergence and thus separate waves or paths of introduction. Shipping traffic, including fishing vessels, cargo vessels, cruise ships, private crafts, and military vessels, are routine visitors to the harbour, as well as to the Jason Islands. Traffic to the harbour has regular connections largely from the North Atlantic and locally from South

America (Montevideo and Punta Arenas), but can be from many parts of the world, while Bird Island and the Jason Islands are more heavily visited by South American traffic. In Argentina, the growing dominance of *M. senile* in fouling communities is closely correlated with shipping traffic increasing since the post-1980s (Martin et al. 2015). Interestingly, *M. senile* was not detected around the Falkland Interim Port and Storage System (FIPASS) present in Stanley Harbour; this may have been due to the dominating presence of ascidians, including the invasive *Ciona* sp. Linnaeus, 1767, excluding other species from establishing on the docks. The apparent absence of prolific asexual reproduction raises questions about whether there is a source population contributing to the stability of the Stanley Harbour population, or if this population is undergoing sexual reproduction but with a slow settling and population growth rate to sustain itself. Regardless, the lack of color and morphological variation at Stanley Harbour is uncharacteristic of the diversity observed within the native range of *M. senile*; this suggests limited contact with the original source population.

The smaller individuals at Bird Island and Flat Jason Island appear to have colonized from only one or very few settlement events, as they are highly localized via asexual reproduction at each site despite the high presence of suitable substrate. The restricted nature of both of these populations, particularly at Flat Jason Island, suggests that *M. senile* has arrived recently. The Stanley Harbour populations show differences in coloration from the other western populations, but since the genetics of coloration remain unknown, it is possible that this is the source population for the smaller and more morphologically variable populations at Bird and Flat Jason Islands. As the size and shape of *M. senile* is highly plastic and influenced by habitat factors, these populations may be related to each other despite the discrepancy in size and shape. Individuals from Bird Island and Flat Jason Island may have been transported via anthropogenic means (e.g. vessels coming from South America visiting the Jason Islands) or introduced from other populations nearby. Alternatively, currents around the tip of South America strongly flow eastward; thus, *M. senile* could disperse through the Beagle Channel from the Pacific coast of South America to the Atlantic, but likely not vice versa, if they were able to withstand the temperature and salinity differences between the Atlantic and Pacific Oceans (Riemann-Zürneck 1986; Häussermann and Försterra 2005).

Further surveys around the Falkland Islands are necessary to document the extent of *M. senile* in the Falkland Islands, as there are very likely to be other individuals or populations present in other locations. The impact of *M. senile* on the native community is largely unknown as it is not classified as harmful or fully invasive (see Blackburn et al. 2011) in the Falklands, though in Chile it has replaced the native *Anthothoe chilensis* covering mytilid banks (Häussermann 2006). Thus, there is a potential for the colonization of alien species which would be detrimental to native fauna in

the future. The position of the Falkland Islands as a stop for ships prior to visits to the sub-Antarctic island of South Georgia and the Antarctic Peninsula places greater urgency for the detection of non-native species in the Falkland Islands under a warming climate and increasing pressures from tourism and industrial development (McCarthy et al. 2019). Early detection is key for a rapid response towards eradication of localized introduction events, with monitoring critical in identifying which potential vectors and routes require greater biosecurity.

Acknowledgements

The authors would like to thank members of the Shallow Marine Surveys Group (including Steve Brown and Sacha Cleminson), and Dion Poncet and Juliette Hennequin (Golden Fleece) for providing logistic and collecting support. Heather Glon would also like to thank Meg Daly for being a supportive advisor during field work planning and writing. We would also like to thank anonymous reviewers for their constructive comments during the revision of our manuscript.

Funding Declaration

Funding sources for the expedition included the Shackleton Scholarship Fund, Darwin Initiative through UK Government Funding (through the DPLUS071 project – Fine Scaling of the Marine Management Area of the Falkland Islands), and The Falkland Islands Government. In-kind support came from the South Atlantic Environmental Research Institute and the Shallow Marine Surveys Group.

Ethics and Permits

Collection of sea anemones took place under The Falkland Islands Government Research Licence No: R33/2019 entitled "Biogeography of the plumose anemone *Metridium senile*: potential marine invasive in the Falklands?"

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Supplementary material

The following supplementary material is available for this article:

Table S1. Known locations of *Metridium senile* occurring in the Falkland Islands.

Table S2. Sequence data and accession numbers on NCBI GenBank for *Metridium senile* collected in the Falkland Islands.

This material is available as part of online article from:
http://www.reabic.net/journals/bir/2020/Supplements/BIR_2020_Glon_etal_SupplementaryMaterial.xlsx