Journal of Environmental Management 230 (2019) 159–167

Contents lists available at ScienceDirect

Journal of Environmental Management

journal homepage: www.elsevier.com/locate/jenvman

Research article

Sampling methods and approaches to inform standardized detection of marine alien fouling species on recreational vessels

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ARTICLE INFO

Keywords:
Alien species
Biofouling
Secondary spread
Monitoring
Yachts

ABSTRACT

Recreational vessels are important contributors to the spread of marine alien species, particularly in relation to secondary spread within novel regions. As such, these vessels should be considered a monitoring priority. The aim of this study was to identify a preferred method for monitoring recreational vessels for alien species, while simultaneously developing a framework that enables managers to objectively choose the most effective sampling approach given their financial constraints. Divers and a remotely operated vehicle (ROV) were considered in relation to four sampling approaches i.e. meanders, transects, inspection of niche areas and the collection of quadrats. Each was applied to the same 53 vessels which represented a spectrum of hull fouling cover. The most effective methods were diver scrape quadrats (Range of alien species numbers per quadrat: 0–9, Total alien species: 20) and inspections of niche areas (Range of alien species numbers: 0–5, Total alien species: 9). All methods employed using an ROV had low efficacy and incurred high costs. While scrape samples were one of the most expensive methods, this was offset by the lowest cost per species detected. Thus, it is recommended that monitoring programmes utilize scrape samples and niche area inspections, but when faced with financial constraints, diver meanders and niche inspections offer sound alternatives for detecting alien species.

1. Introduction

Alien species are recognized as an important threat to biodiversity and ecosystem functioning in terrestrial, freshwater and marine environments (Mack et al., 2000; Marchi et al., 2011; Simberloff et al., 2013; Chan and Briski, 2017). In the marine environment, such species are transferred by a variety of vectors including ballast water (Adebayo et al., 2013), biofouling (Williams et al., 2013), aquaculture (Grosholz et al., 2015), the aquarium trade (Holmberg et al., 2015) and canals (Galil et al., 2015). Of these vectors, biofouling (i.e. the attachment or growth of biota on the submerged sections of hulls and niche areas of vessels (Coutts and Taylor, 2004)) has become dominant in recent years (Hewitt et al., 2009; Williams et al., 2013) with the role of fouling on recreational vessels becoming increasingly recognized (Davidson et al., 2010; Hewitt et al., 2007; Peters et al., 2017). Species are able to establish and accumulate on these vessels during their long stationary periods in marinas (Hewitt et al., 2009), a process that is aided by inefficient or ill-maintained anti-fouling paint (Floerl and Inglis, 2005). While it is not an easy task to assign an unequivocal link between an already established alien species and the vector through which it arrived (Minchin, 2007a), recreational vessels have played a key role in the spread of some marine algae and invertebrate species (Hewitt et al., 2007; Minchin et al., 2006). One of the best documented cases of yacht transfer is that of the mussel *Mytilopsis sallei* that was introduced to Darwin Harbour estuary (Willan et al., 2000). This introduction culminated in one of the first successful large-scale marine eradication attempts, following its early detection and the ensuing rapid response by authorities (Bax et al., 2002).

The example of *M. sallei* highlights the need to effectively survey small vessel fouling assemblages to detect alien species. Sampling of subtidal communities has been undertaken using a variety of techniques, ranging from visual to extractive approaches (Mallet and Pelletier, 2014). For fouling assemblages, approaches that have been used include visual observations from the surface (Brine et al., 2013; Floerl et al., 2005), subtidal visual surveys by divers that may include the taking of photographs and videos (Chapman et al., 2013; Coutts and Taylor, 2004; Davidson et al., 2009), the use of pole cameras operated from the surface (Brine et al., 2013; Davidson et al., 2010; Zabin et al., 2014), the use of remotely operated devices (Davidson et al., 2009; Lee and Chown, 2009; Needles and Wendt, 2013) and extractive sampling whereby samples are collected and then processed by taxonomic experts (Chapman et al., 2013; Coutts and Dodgsun, 2007; Davidson et al.,

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https://doi.org/10.1016/j.jenvman.2018.09.063
Received 22 February 2018; Received in revised form 5 September 2018; Accepted 20 September 2018

2010; Zabin et al., 2014). These methods require differing levels of expertise, with varying associated costs. Nonetheless, the success of detecting alien species may vary depending on the sampling approach applied, and this is unavoidably linked to the availability of resources (see Mallet and Pelletier, 2014). Non-invasive visual techniques, such as video surveys, enable the collection of large datasets that can be time-efficient (Lam et al., 2006). In contrast, the collection of samples allows for accurate identification and detection of smaller and inconspicuous organisms, ultimately resulting in higher species detection rates (Peters et al., 2014). Although these subtidal sampling techniques have been applied in various contexts, few studies have quantitatively compared the efficacy of these methods for assessing fouling assemblages (but see Beaumont et al., 2007), and none have considered their applicability for detecting alien species on recreational vessels. Notably, the relative costs involved with using these kinds of techniques have not been objectively compared (Mallet and Pelletier, 2014), despite the important implications that this has for management organizations tasked with monitoring for marine alien species.

Besides moving between countries, recreational vessels also connect main ports to more remote regions within country borders, constituting regional secondary vectors for species introduced through primary vectors, such as ballast water and ship fouling (Clarke Murray, 2012; Wasson et al., 2001). Despite this, and the recognition of the potential importance of this vector (Clarke Murray et al., 2011; Davidson et al., 2010), there is no systematic monitoring of recreational vessels for marine alien species anywhere in the world. A precursor for the development of such a system is the establishment of an effective and cost-efficient sampling approach. While the value of such a system is clear, the development of a standardized method that could be applied across regions would be extremely beneficial. The overarching aim of this study was thus, to identify a preferred method for monitoring recreational vessels for alien species, while simultaneously developing a framework that will enable managers to objectively choose the most effective sampling approach attainable within their financial and logistical constraints.

2. Materials and methods

2.1. Study region

This study considered yachts from four marinas in the Western Cape, South Africa, to experimentally compare methods for detecting alien species on the hulls of sea-faring recreational vessels. These marinas were; Port Owen Yacht Club (32º46′56.43″S; 18º08′53.60″E), Saldanha Bay Yacht Club (33º00′37.68″S; 17º56′56.75″E), Royal Cape Yacht Club (33º55′14.15″S; 18º26′34.84″E) and False Bay Yacht Club (34º11′32.99″S; 18º26′02.20″E). All marinas are situated within or adjacent to large ports and all marinas receive both local and international yacht traffic. This study considered sailing yachts because in the South African context motorized vessels rarely move among marinas due to rough sea conditions that typify this exposed coastline.

2.2. Fouling ranks

Data were collected between December 2015 and October 2016. To gain a measure of background fouling levels, all yachts in each marina (N = 638), were visually inspected and assigned a Fouling Rank (FR). Fouling Rank is an estimated measure of the amount of biofouling present on the submerged surface of a vessels hull, as visible from the surface. This ranking approach was adapted from the ordinal ranking system developed by Floerl et al. (2005) with the number of levels reduced to four for practical reasons (Table 1). A two-way Chi-Square test was used to determine if the number of boats differed across Fouling Ranks and marinas. All analyses, unless otherwise indicated, were undertaken in STATISTICA 13. A total of 53 yachts were sampled for alien species. This sample size was determined by the number of yacht owners who were willing to provide access to their vessels during the study period. While a balanced sampling design with equal numbers of yachts in each Fouling Rank would have been desirable, this was impossible at the level of individual marinas.

2.3. Sampling approaches

A total of eight sampling methods were applied to each yacht, using four sampling approaches that were undertaken by both a scientific diver and a VideoRay Pro 3 Remotely Operated Vehicle (ROV). The four approaches were the Meander, Transect, Niche and Quadrat. The Meander consisted of a diver searching the submerged hull area for alien species, for a period of 6 min. The Transect involved a diver searching the circumference of the vessel (i.e. all the around the edge of the entire hull) at a distance of 50 cm below the waterline. The Niche method included inspections of all submerged niche areas of each vessel (i.e. rudder, keel, water intakes, propeller and propeller shaft). The Quadrat method involved the collection of six randomly placed photo quadrats by the ROV and those same six quadrats were scraped and all fouling collected by divers as scrape quadrats. These scrapes were later identified in the laboratory. Divers also made use of a target list of 10 alien species when searching the hull and niche areas and this list was applied in the ROV methods as well. The use of a target list was implemented in order to ensure fast and cost-effective sampling (Minchin et al., 2016) as resources for marine biosecurity are generally limited. Excepting for the processing of scrape samples that needed to be done back in the laboratory, the various methods were randomly applied to ensure no effect of sampling order on the number of species detected. The fact that various people controlled the ROV, sorted the scrape samples and undertook the diver based methods, further avoided any sampling introduced bias. Additional details regarding these methods are provided in supplementary electronic Table S1 1. For each yacht, the total number of alien species detected by each method was recorded. Additionally, for the scrape quadrats, biomass (to the nearest 0.01 g) was recorded for each species, while for the ROV photo quadrats, percentage cover was estimated.

For each method Spearman's Rank Correlations were used to detect relationships between Fouling Ranks and the number of species recorded per yacht. For Scrape and Photo Quadrats, Spearman's Rank Correlations were also used to consider correlations between the Fouling Ranks and the mean percentage cover and biomass of alien species (per yacht) respectively. The number of species recorded per yacht was analyzed using a general mixed effects model (nlme package in R) with Fouling Rank (four levels: FR 0, FR 1, FR 2, FR 3) and method (six levels: diver meander, ROV meander, diver transect, ROV transect, scrape quadrat, photo quadrat) as fixed factors and yacht as a random factor. The unequal number of yachts in the various Fouling Ranks per marina precluded the inclusion of marina as random factor. The best fit model was chosen based on Akaike Information Criteria. A Wald test was used to assess the significance of the fixed factors in the final model (Bolker et al., 2009).

In order to assess if the various methods detected different suites of species, a two factor PERMANOVA was undertaken in Primer 6 (version 6.1.16), with method considered a fixed factor and marina a random factor. PERMANOVA offers a non-parametric approach to analysis of variance for multivariate datasets (Anderson, 2001). Because the detection of species was of interest, rather than their abundance, and the fact that some methods only yielded presence/absence data, the analysis was conducted on presence/absence data only, comparing each treatment in the various marinas. To ensure a balanced design for this analysis, four yachts were randomly selected as replicates (per treatment) for each marina.

2.4. Cost of sampling approaches

In addition to the collection of biological data, the cost of each