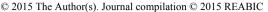
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Research Article

Rapid assessment of marine non-native species in northern Scotland and a synthesis of existing Scottish records

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Abstract

In this study, we compiled existing records of fouling marine non-native species in Scotland, and created a national checklist of these species. We then targeted a selection of these species (excluding those that could not be reliably identified) in a rapid assessment survey of 27 harbours in the north of Scotland. Collation of existing records revealed that 23 fouling marine non-native species were known to be present in Scotland. The geographic distribution of these records was not uniform, and they were largely underrepresented in the north and east of mainland Scotland, likely as result of lack of survey effort. In the rapid assessment survey of north Scotland, 9 out of 18 targeted species were found: Austrominius modestus (Darwin, 1854); Botrylloides violaceus Oka, 1927; Caprella mutica Schurin, 1935; Codium fragile fragile (Suringar) Hariot, 1889; Corella eumyota Traustedt, 1882; Heterosiphonia japonica Yendo, 1920; Neosiphonia harveyi (Bailey) Kim, Choi, Guiry and Saunders, 2001; Schizoporella japonica Ortmann, 1890; and Tricellaria inopinata d'Hondt and Occhipinti Ambrogi, 1985. The non-native bryozoan Bugula simplex Hincks, 1886, which was not targeted, was also found, and this constituted the first confirmed Scottish record. The surveys provided 60 new records and extended the northward national range for most of the species found. The number of fouling non-native species in the surveyed harbours was positively associated with the presence of floating structures and vessel activity indices. Our study presents an overview of the current status of fouling marine non-native species in Scotland, and the results of the first comprehensive survey of these species in the north of Scotland. The latter provides a baseline dataset for monitoring future changes, which may occur as a result of the development of the wave and tidal energy industry in the north of Scotland. The wave and tidal energy industry has the potential to facilitate the invasion of fouling marine non-native species through the provision of habitat and by increasing vector activity.

Key words: invasive species, fouling, checklist, harbour properties, marine renewable energy

Introduction

Non-native species (NNS) are considered one of the greatest threats to biodiversity (Sala et al. 2000; Millennium Ecosystem Assessment 2005) and can cause severe ecological and economic damage (Vila et al. 2010; Williams et al. 2010). In the marine environment, the introduction and spread of NNS is largely mediated by anthropogenic activities (Eno et al. 1997; Streftaris et al. 2005; Wonham and Carlton 2005; Seebens et al. 2013). Baseline datasets of NNS distribution are therefore important to assess the potential impact of future anthropogenic activities on the introduction and spread of marine NNS. In some countries, including Scotland, there are also legislative requirements for reporting and monitoring the presence of marine

NNS (Scottish Government 2012; European Commission 2013; DEFRA 2014). Furthermore, monitoring is vital for informing management efforts, for example the attempt at eradicating a population of the carpet sea squirt *Didemnum vexillum* Kott, 2002 close to a large mussel farm in north Wales (Holt and Cordingley 2011).

A large proportion of marine NNS are fouling organisms (Minchin 2007a; DAISIE 2009; Minchin et al. 2013); these include sessile organisms that grow on hard substrata and vagile clinging organisms that live within or on sessile communities. Fouling NNS are well adapted for transfer to new locations via anthropogenic vector movement such as vessel traffic (Clarke Murray et al. 2012) or aquaculture processes (Mineur et al. 2007). These species inhabit the water-exposed surfaces

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of vessels and other structures, and can be introduced to recipient habitats through dislodgement, fragmentation, or the release of propagules (Gollasch 2002; Coutts and Taylor 2004; Clarke Murray et al. 2011). The addition of artificial structures (e.g. harbour structures, aquaculture lines and cages, and marine energy devices) to the marine environment also creates hard habitat for fouling NNS to colonise (Glasby et al. 2007; Mineur et al. 2012). These structures can act as 'stepping stones' for fouling NNS to spread across natural dispersal barriers such as soft sediment habitats (Bulleri and Airoldi 2005; Sheehy and Vik 2010).

One of the world's largest developments for commercial wave and tidal energy is planned in the Pentland Firth and Orkney waters, northern Scotland. The proposed development represents the addition of a substantial amount of artificial structure to the local marine environment. This will include the installation of a large number of energy capture devices (hundreds to thousands), and the expansion of harbours to provide logistical support (Scottish Enterprise 2010; The Crown Estate 2011). The addition of artificial habitat could aid the establishment of fouling NNS in the area (Kerckhof et al. 2011; Mineur et al. 2012), and if a network of suitable habitats is formed this may promote the spread of NNS through the 'stepping stone' effect (Miller et al. 2013). Increased vector movement, in the form of vessel and towed-structure traffic during maintenance and construction phases of the development, could also facilitate the introduction and spread of NNS. However, the potential for the wave and tidal energy industry to facilitate the invasion of NNS has not been considered in the Scottish Strategic Environmental Assessment for wave and tidal power (Scottish Executive 2007). At present it is only a concern in some literature (Inger et al. 2009; Mineur et al. 2012; The Crown Estate 2014). In order to monitor this potential impact, it is important to know the presence/absence and distribution of fouling NNS in the Pentland Firth and Orkney waters and surrounding area (referred to as "north Scotland" in this paper), prior to the development of this industry.

In an initial literature search we found there was a paucity of both presence and absence records for fouling NNS in north Scotland. A survey for the presence and absence of fouling NNS in the area would therefore provide the baseline dataset required for monitoring potential effects of the wave and tidal energy industry. It would also provide the first comprehensive dataset

of fouling NNS in north Scotland. This has important applications for the statutory environmental bodies in charge of managing NNS and for local industries that can be adversely affected by the presence of fouling NNS, such as aquaculture facilities (Durr and Watson 2010). A baseline dataset could also be useful for monitoring the effects of other potential coastal developments, such as the proposed expansion of aquaculture production throughout Scotland (Scottish Government 2013). Due to its northerly latitude, this dataset will be similarly useful for monitoring northward range shifts of fouling NNS that could occur as a result of climate change (Mieszkowska et al. 2006; Sorte et al. 2010).

Before a regional baseline survey is carried out, it is useful to know what fouling NNS are present within a country. This can inform the selection of 'target species' to be searched for in field surveys. Whilst peer-reviewed inventories for the British Isles have been created previously (Eno 1996; Eno et al. 1997; Maggs and Stegenga 1999; Minchin 2007a; Minchin et al. 2013), none exist specifically for Scotland (although see incomplete lists in Saunders (2004) and Baxter et al. (2011)). The first aim of this study was therefore to collate all known records of fouling NNS in Scotland from both unpublished and published literature, and to create a national checklist. As well as determining which species should be targeted in a survey of north Scotland, this also provided a platform to discuss the fouling NNS present in Scotland and the geographical distribution of the records.

The second aim of the study was to determine the presence/absence and distribution of targeted fouling NNS in north Scotland, through field surveys of harbours in the area. Vessel activity has been shown to have a positive relationship with the rate of biological invasion (Ricciardi 2001; Floerl et al. 2009), and fouling NNS may be more prevalent on floating structures, such as pontoons, than on fixed structures, such as harbour walls (Glasby et al. 2007; Dafforn et al. 2009). Therefore, an additional aim of this study was to examine whether or not these factors influenced the number of non-native species found in harbours.

Methods

Synthesis of Scottish records

We compiled a list of marine hard-substrate fouling species from the most recent marine and brackish NNS inventory of Great Britain (Minchin et al.

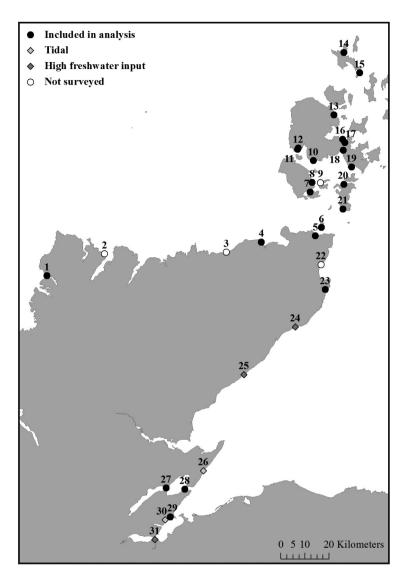


Figure 1. Locations of sampled harbours. Site names: 1. Kinlochbervie, 2. Loch Eriboll - Rispond, 3. Sandside, 4. Scrabster, 5. Gills Bay, 6. Stroma, 7. Hoy Longhope, 8. Hoy Lyness, 9. Flotta - Sutherland Pier, 10. Houton, 11. Stromness Polestar, 12. Stromness Marina, 13. Tingwall Harbour, 14. Westray Pierowall, 15. Westray Rapness, 16. Kirkwall Hatson, 17. Kirkwall Marina, 18. Scapa, 19. St Mary's Holm. 20. St. Margaret's Hope. 21. Burwick, 22. Kiess, 23. Wick, 24. Lybster, 25. Helmsdale, 26. Balintore, 27. Invergordon, 28. Cromarty, 29. Fortrose, 30. Avoch, 31. Inverness Marina. The shade of the dots indicate which harbours were included in analysis and those that were not as a result of the harbour being tidal (dry at low tide), having high freshwater input (sites with < 27 salinity) or not being surveyed. See Supplementary material Table S1 for harbour coordinates, harbour properties and number of non-native species found in them.

2013). Only strictly marine fouling species were identified from this inventory. 'Fouling' species were defined as sessile organisms on hard substrata and associated vagile species; we excluded any brackish, saltmarsh, or wood boring species. We then carried out a literature search for Scottish records of these species, searching published literature and the publically accessible database, National Biodiversity Network (NBN) gateway. Unpublished records acquired from the Scottish Association of Marine Science were also added to the dataset (Rodgers 2012; Cook EJ unpublished data). We also searched for records of the non-Schizoporella native encrusting bryozoan japonica Ortmann, 1890; this fouling NNS was

first recorded in Scotland in 2011 (Porter et al. 2012) and was not included in the Minchin et al. (2013) inventory. Coordinates, year of record, and the source reference were extracted for each record

Confirmations were requested from record providers when individual NBN gateway records appeared unusual. There was no specific definition of an 'unusual' record; some examples included a single record of a species in an area distant from other records or old records in areas with no recent sightings. Duplicate records were also removed; these had been created as a result of simultaneous records occurring in published literature and the NBN gateway.

Table 1. List of targeted fouling NNS for the north Scotland surveys and the harbours where they were detected. Records for untargeted species (both NNS and cryptogenic species), and whether or not species were found in the subtidal or intertidal are also presented. Harbour numbers are defined in Figure 1. * = cryptogenic species \mathbf{X} = Recorded after survey and not included in analysis.

Species	Harbours where they were detected	\sum no. of harbours	Subtidal	Intertidal
Targeted species				
Asparagopsis armata Harvey, 1855	-	0	-	-
Austrominius modestus (Darwin, 1854)	26, 27, 28, 29, 30	5	✓	✓
Bonnemaisonia hamifera Hariot, 1891	-	0	-	-
Botrylloides violaceus Oka, 1927	17, 27, 28	3	✓	×
Bugula neritina (Linnaeus, 1758)	-	0	-	-
Caprella mutica Schurin, 1935	1, 4, 7, 8, 11, 12, 14, 17, 20, 27, 28, 29	12	✓	×
Codium fragile fragile (Suringar) Hariot, 1889	8, 10, 11, 12, 13, 15, 17, 19, 20, 21, 23	11	✓	✓
Corella eumyota Traustedt, 1882	4 ^x , 8, 11, 12, 16, 20, 23	7	✓	×
Crassostrea gigas (Thunberg, 1793)	-	0	-	-
Crepidula fornicata (Linnaeus, 1758)	-	0	-	-
Diadumene lineata (Verrill, 1869)	-	0	-	-
Didemnum vexillum Kott, 2002	-	0	-	-
Heterosiphonia japonica Yendo, 1920	6, 8, 11, 12, 15, 19, 20, 23, 28	9	✓	✓
Neosiphonia harveyi (Bailey) Kim, Choi, Guiry and Saunders, 2001	1, 4	2	✓	×
Sargassum muticum (Yendo) Fensholt, 1955	-	0	-	-
Schizoporella japonica Ortmann, 1890	4, 11, 12, 14, 16, 17, 27, 28	8	✓	✓
Styela clava Herdman, 1891	-	0	-	-
Tricellaria inopinata d'Hondt and Occhipinti Ambrogi, 1985	11, 12, 17	3	✓	×
Untargeted species				
Bugula simplex Hincks, 1886	17	1	✓	×
Bugula fulva Ryland, 1960*	4, 7, 8, 11, 12, 16, 17, 27	8	✓	×
Jassa marmorata Holmes, 1905*	16, 20, 27, 28, 29	5	✓	×

A distribution map of Scottish records was created using ArcMap 10 (ESRI Ltd.). This was split into geographical regions based on those used in the Marine Nature Conservation Review (MNCR, Hiscock 1996), and the number of records and fouling NNS richness in each region was reported. The year of first detection in Scotland was compared with the year of first detection in Great Britain. We also noted whether or not a species was 'established'. We considered a species to be 'established' if it appeared from the records to be present in the same area for at least four years. This definition follows Roy et al. (2012) and assumes that persistent presence is the result of self-sustaining reproduction and not repeated introduction. If there was no evidence of a species being recorded in the same place for at least four years, but it had been recorded in many different locations, we recorded it as being 'probably established'.

North Scotland survey

Site selection

The north Scotland field survey sites included harbours in the Pentland Firth and Orkney waters as well as harbours on surrounding coasts. We selected harbours as survey sites because they are known hotspots for NNS (Minchin et al. 2006; Glasby et al. 2007). In total 31 harbours (including marinas) were selected for sampling (Figure 1, supplementary Table S1). The selected harbours spanned a geographical area from Inverness marina (57.4926°N, 4.2346°W) in the south, up to Westray Pierowall Harbour (59.3233°N, 2.9750°W) in the north, and Kinlochbervie Harbour (58.4583°N, 5.0650°W) in the west. Surveys were conducted between July and August 2012. Unfortunately four harbour surveys had to be abandoned due to poor weather or for logistical reasons.

Rapid assessment survey

We used rapid assessment methods to target the fouling NNS highlighted from the literature search (Table 1). Rapid assessment surveys are commonly used to assess the presence and absence of fouling NNS in harbours (Cohen et al. 2005; Ashton et al. 2006; Minchin and Nunn 2013) and involve the inspection of fouling assemblages on harbour structures for targeted NNS. Focusing search effort on targeted species decreases the likelihood of missing NNS, and this has proved effective when used by small teams of surveyors (Ashton et al. 2006; Minchin 2007b; Beveridge et al. 2011).

Training in identification of the targeted fouling NNS was undertaken by the main surveyor involved studying (CRN). This taxonomic descriptions, photographs, and voucher specimens (acquired from the Scottish Association of Marine Science). For the algae species with heteromorphic life histories (Asparagopsis armata Harvey, 1855 and Bonnemaisonia hamifera Hariot, 1891), identification training only included the gametophyte form. Native species similar in morphology to targeted species were also studied to avoid misidentification. A similar strategy of familiarisation with species in advance of the surveys was successfully adopted by Minchin (2007). We also conducted training surveys in marinas on the west coast of Scotland. During these training surveys new sightings of Styela clava Herdman, 1891 (in Troon Yacht Haven, March 2012) and Schizoporella japonica (in Croabh Yacht Haven, March 2012) were recorded.

Rapid assessment surveys were undertaken by two people (one of whom was always CRN) and were completed when we could find no more additional fouling NNS; this usually took between one and two hours. We inspected a number of harbour features, these included: floating marina pontoons; smaller floating submerged structures (e.g. mooring buoys and fenders used for berthing protection); and fixed structures (e.g. pilings and harbour walls). When necessary, we used a small boat to access harbour features. Subtidal samples were collected from floating structures between the depths of 0-1m by hand or using a purposebuilt scraper (a 15 cm wide scraper blade with detachable collection bag welded to the end of a 1.5 m pole). Where possible, items where pulled out of the water for thorough inspection. Surveys were conducted around low tide to allow access to the intertidal zone on fixed structures. We recorded whether each fouling NNS was observed in the intertidal zone, in the subtidal zone, or both. Samples of all species thought to be fouling NNS were collected and fixed in phosphatebuffered formalin (4% formaldehyde in distilled water). Samples were later transferred to 80% industrial methylated ethanol for preservation and storage. Identifications were verified in the laboratory with microscopy and, where necessary, we sought confirmations from relevant taxonomic

In each harbour we recorded the following harbour properties: salinity (at 0.5 m depth); presence of pontoons; presence of smaller floating structures (mooring buoys and fenders);

and whether or not the harbour was dry at low tide. Two proxy measures were also taken to estimate vessel activity in harbours: estimated length of quayside, and the number of vessels moored. These were measured from aerial images (taken within the last 3 years) on Google Earth (version 6.2). The length of harbour walls and pontoons (not including pontoon fingers) were measured in metres using the Google Earth path function tool

Data analysis

Using ANOVA and Spearman rank-correlation analyses we evaluated relationships between the number of fouling NNS found in harbours and the following harbour properties: presence of pontoons, presence of smaller floating structures, and the vessel activity indices, length of quayside and number of vessels. The number of fouling NNS found was natural $\log (x + 1)$ transformed for the ANOVA analysis. All statistics were run with SPSS version 21.0 (IBM Corp. 2012). Harbours that were dry at low tide or had a high freshwater input (salinity < 27) were removed numerical analyses (Figure supplementary Table S1). These harbours had very small numbers of NNS; either none at all or only 1 NNS. Stromness Polestar pier was also removed from analyses as it was deemed not to be independent of Stromness marina due to its close proximity.

Results

Synthesis of Scottish records

From the literature search of Scottish records we revealed that 23 fouling NNS had previously been recorded in Scotland (Table S2). This comprised 1,034 records (a full list of these records with coordinates, year of record, and the source reference is available in Table S3). Fifteen of the 23 fouling NNS present in Scotland had established self-sustaining populations (Table S2). Species which were not established included those which had arrived in the country too recently for establishment to be determined (Botrylloides violaceus Oka, 1927, Didemnum vexillum, and Schizoporella japonica), and those which had reports of single or few incidences without any recent records (Aulacomya atra (Molina, 1782), Amphibalanus amphitrite (Darwin, 1854), Bugula neritina (Linnaeus, 1758), Crepidula fornicata (Linnaeus, 1758), and Crassostrea gigas (Thunberg, 1793)).

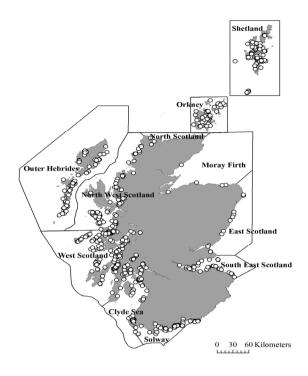


Figure 2. Distribution of fouling non-native species (NNS) records in Scotland including an overlay of the Scottish Marine Nature Conservation Review (MNCR) regions. The records displayed here are exclusively from the literature search and do not include records from the north Scotland survey.

More than half (15/23) of the fouling NNS present in Scotland were first detected within the last 30 years and 8 of those 15 species were first detected within the last decade (Table S2). With the exception of *Codium fragile fragile* (Suringar) Hariot, 1889, Caprella mutica Schurin, 1935, and Aulacomya atra, all species were detected in Great Britain prior to being detected in Scotland. In some cases the time difference between first detection in Great Britain and first detection in Scotland was very large: Crepidula fornicata (116 yrs), Bugula neritina (95 yrs), Diadumene lineata (Verrill, 1869) (83 yrs), Neosiphonia harveyi (Bailey) Kim, Choi, Guiry and Saunders, 2001 (82 yrs), Bonnemaisonia hamifera (76 yrs), Crassostrea gigas (52 yrs), Amphibalanus amphitrite (51 yrs), and Styela clava (34 yrs) (Table S2).

The distribution of fouling NNS records in Scotland was not uniform (Figure 2, Figure 3a). MNCR regions with the largest number of records included 'West Scotland', 'Clyde Sea', and 'Shetland' (297, 152, and 150 records, respectively). MNCR regions with a comparatively low number of records included 'East Scotland', 'Moray Firth',

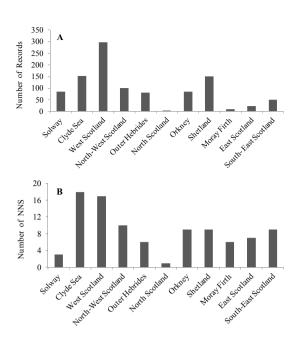


Figure 3. Number of records for fouling non-native species in each MNCR region between 1891–2012 (A); fouling NNS richness in each MNCR region between 1891–2012 (B). The records displayed here are exclusively from the literature search and do not include records from the north Scotland survey.

and 'North Scotland' (23, 10, and 3 records, respectively). The fouling NNS richness was also unevenly distributed across the MNCR regions (Figure 3b). However, this was less pronounced than the number of records, because certain fouling NNS were widespread across Scotland. Widespread species included Asparagopsis armata, Austrominius modestus (Darwin, 1854), Bonnemaisonia hamifera, Caprella mutica, Colpomenia peregrina Sauvageau, 1927, and Codium fragile fragile. The 'Clyde Sea' and 'West Scotland' regions had the highest fouling NNS richness (18 and 17, respectively; Figure 3b). In the north Scotland field survey area there was a total of 96 records for 10 different species (Table S3). This comprised only 9% of all the Scottish records found in the literature search. The majority of these records were from Orkney with only 13 records from the mainland. Of the 10 recorded species, 5 were algae (Asparagopsis armata, Bonnemaisonia hamifera, Codium fragile fragile, Colpomenia peregrina, and Heterosiphonia japonica Yendo, 1920) and 5 were invertebrates (Aulacomya atra, Caprella mutica, Crassostrea gigas, Diadumene lineata, and Schizoporella japonica).

North Scotland survey

Although 23 fouling NNS were reported in Scotland from the literature search, 18 species were targeted in the rapid assessment survey (see Table 1). Five fouling NNS (Amphibalanus amphitrite, Aulacomya atra, Antithamnionella spirographidis (Schiffner) Wollaston, 1968, Antithamnionella ternifolia (Hooker and Harvey) Lyle, 1922, and Colpomenia peregrina) were omitted from the target list because identification training was not undertaken for these species before the survey.

Nine of the 18 targeted fouling NNS were found in the rapid assessment surveys (Table 1). Five of these NNS (Austrominius modestus, Botrylloides violaceus, Corella eumyota Traustedt, 1882, Tricellaria inopinata d'Hondt Occhipinti Ambrogi, 1985, and Neosiphonia harveyi) represented the first records in north Scotland. The untargeted non-native bryozoan Bugula simplex Hincks, 1886 was also found in Kirkwall Marina (6th August 2012), and this represents the first confirmed record of this species in Scotland (JS Ryland pers. comm.). Two additional untargeted species were also found: the cryptogenic (as defined by Carlton 1996) species Bugula fulva Ryland, 1960 and Jassa marmorata Holmes, 1905. There were 60 sightings of fouling NNS and 13 sightings of cryptogenic species during the survey (Table 1, Table S4). The frequency with which each fouling NNS was found varied (Table 1). Caprella mutica and Codium fragile fragile were the most common fouling NNS in the surveys; these species were found at 12 and 11 harbours respectively.

The number of fouling NNS found at each site ranged from 0 to 6 (Table S1). Stromness marina/Stromness Polestar pier (6 species), Kirkwall marina (6 species), and Cromarty marina (5 species) had the highest number of NNS. The number of fouling NNS found was positively correlated with both of the vessel activity indices: number of vessels (Spearman's r_s = 0.534, n = 21, P = 0.013); and length of quayside (Spearman's $r_s = 0.479$, n = 21, P = 0.028). Presence of floating structures within harbours also influenced the number of fouling NNS found. The mean (± SE) number of fouling NNS was significantly greater (ANOVA, $F_{1.19}$ = 22.051, P < 0.001) in harbours with small floating submerged structures (3.29 ± 0.425) than in harbours without (0.86 \pm 0.261). There was a similar difference (ANOVA, $F_{1,19} = 4.781$,

P=0.041) between the mean number of fouling NNS in harbours with pontoons (3.30 ± 0.597) compared to harbours without (1.73 ± 0.407) . All species detected in the rapid assessment survey were found subtidally on floating structures; *Codium fragile fragile*, *Heterosiphonia japonica*, *Schizoporella japonica*, and *Austrominius modestus* were also found on the intertidal areas of fixed structures (Table 1). In harbours categorised as having high freshwater input, no fouling NNS were found; these harbours were situated near freshwater outflows and had salinities < 10.

Discussion

Synthesis of Scottish records

Twenty-four fouling NNS have been recorded in Scotland; this includes the addition of *Bugula simplex* which was first recorded in our north Scotland field surveys. Although only 15 species appear to have self-sustaining populations, the recently arrived species *Didemnum vexillum, Botrylloides violaceus,* and *Schizoporella japonica* may become established soon because they are reported to be abundant where present (Beveridge et al. 2011; Porter et al. 2012). *Schizoporella japonica* and *Botrylloides violaceus* were also found at a number of sites in our north Scotland field surveys (in 8 and 3 harbours respectively).

Fewer marine fouling NNS are known to be present in Scotland (24 species) than in Great Britain (59 species, including *Schizoporella japonica*) and Ireland (34 species) (Minchin 2007a; Holt and Cordingley 2011; Minchin et al. 2013). The lower number of fouling NNS recorded in Scotland may be the result of undersampling or because there are actually fewer fouling NNS. Some fouling NNS are likely to be absent from Scotland because their seawater temperature requirements restrict them to lower latitudes. For example, the non-native serpulid polychaete *Ficopomatus enigmaticus* (Fauvel, 1923), requires a minimum seawater temperature of 18°C to reproduce (Dixon 1981).

Within Scotland, the distribution of records and fouling NNS richness was not uniform across MNCR regions. The 'West Scotland' and 'Clyde Sea' regions had the highest fouling NNS richness and, along with the 'Shetland' region, had disproportionately more records. This disparity could be the result of non-uniform survey effort across Scotland, with many previous surveys focused on the west coast (Ashton et al. 2006, 2007; Beveridge et al. 2011). Another possibility

could be a difference in vector activity between the regions. For example, recreational boat movement is more frequent in the 'West Scotland' and 'Clyde Sea' regions than in other regions of Scotland (RYA 2008). Regional differences in habitat availability may also explain the disparity in the distribution of records and fouling NNS richness; aquaculture facilities and marina pontoons are known habitats for fouling NNS, and the west coast of Scotland, and the Shetland Islands contain the majority of aquaculture sites in Scotland (Baxter et al. 2011). The west coast of Scotland also has a relatively large number of marinas (RYA 2008).

Species that have been reported in Scotland on only single or a few occasions (with no recent records), may have failed to establish and may no longer be present. These species include Aulacomya atra, Crepidula fornicata, Crassostrea gigas, Amphibalanus amphitrite, and Bugula neritina. Seawater temperatures around Scotland do appear to be suitable for the southern hemisphere mussel, Aulacomya atra (Davenport and Davies 1984). However, there has only been one record of this species (in 1994, see Table S3) and it is unlikely that it has established a population. Establishment may have been prevented by low propagule pressure, low density of introduced organisms, and by predation and competition from native species. The other species may require warmer seawater temperatures for reproduction than are available in Scotland (Qiu and Qian 1999; Dutertre et al. 2009; Ryland et al. 2011; Bohn et al. 2012). For example, Crassostrea gigas is thought to require temperatures of at least 18°C for spawning (Dutertre et al. 2009), and it must remain relatively warm for successful larval development and settlement (Ruesink et al. 2005). This is 3-4°C higher than the maximum annual temperature observed in most Scottish waters (Baxter et al. 2011). Following warm summers Crassostrea gigas has, however, settled in Scandinavian waters as far north as 60°N (Wrange et al. 2010). Moreover, predicted surface seawater temperature (SST) rises due to climate change (IPCC 2007) could mean that more species may establish in Scotland in the future (Cook et al. 2013). Climate change may have already induced range shifts of fouling NNS in Scotland (Cook et al. 2013), and this may explain the large time difference some species have shown between being detected further south in Great Britain and being detected in Scotland.

The compilation of records presented in this study provides a checklist of fouling NNS within

Scotland. Subsequent to the completion of the literature search (May 2012), a newly introduced non-native ascidian, *Asterocarpa humilis* (Heller, 1878), was found in Oban marina, west Scotland (56.4174°N, 5.4975°W) in October 2013 (observation made by EJC). This discovery takes the number of Scottish fouling NNS up to 25 and highlights the importance of maintaining up-to-date national checklists of NNS. We suggest that target species lists for future NNS surveys should include species that might be introduced in the near future. These species can be identified through horizon scanning (Minchin and Nunn 2013; Roy et al. 2014).

North Scotland survey

The rapid assessment surveys have detected the presence of 6 new fouling NNS in the north Scotland field survey area. The surveys also confirmed the continued presence of 4 of the 10 fouling NNS previously known in the area. All incidences were new spatial records, with the exception of Schizoporella japonica in Stromness and Kirkwall marinas (Porter et al. 2012). The presence of new fouling NNS and the numerous records (60) from the field surveys contrasts with the relatively small number of records and NNS richness previously reported from the north Scotland field survey area. This confirms that the non-uniform distribution of fouling NNS records and NNS richness exhibited across Scotland in the literature search could be a result of a lack of survey effort, although it could alternatively be a result of recent introductions to north Scotland.

Range expansions

Records from the field survey have extended the national ranges northwards for all the fouling NNS found, apart from Austrominius modestus, Caprella mutica, and Codium fragile fragile, which have previously been recorded further north in the Shetland Islands (Table S3; Hiscock et al. 1978; Ashton et al. 2007; Provan et al. 2007). The greatest range expansion was for Bugula simplex, a species native to Mediterranean whose previous most northerly record in Great Britain was in Holyhead harbour. Wales (Ryland 1958). Only a few records of Bugula simplex exist in the British Isles and northern Europe (Blauwe and Faasse 2001; Ryland et al. 2011) so it is difficult to ascertain the importance of this finding. Its lack of detection is probably due to a combination of it rarely

being searched for, its close resemblance to native *Bugula* spp., and because it is seasonal in occurrence (Ryland et al. 2011). If, however, its detection now represents a genuine range expansion, this may have been mediated by an increase in SST over the last century in Scotland (Hughes et al. 2010). Either way, the large range expansion suggests this species could be present elsewhere in Scotland and Europe, and future NNS surveyors in Scotland and Europe are advised to add this to their list of target species.

Climate change can result in poleward shifts in the distribution of some species (Thomas 2010), and there are reports suggesting that SST rise may have caused some marine NNS to spread northwards in the British Isles and Europe (Maggs et al. 2010; Wrange et al. 2010; Pederson et al. 2011; Cook et al. 2013). The long established non-native barnacle Austrominius modestus was previously thought to be restricted by temperature to southern Scotland (Crisp 1958; Barnes and Barnes 1960), and its gradual northward range expansion and increased abundance in Scotland has been attributed to SST rise (Evans 2008; O'Riordan et al. 2009). Our record of Austrominius modestus in Balintore (57.75393°N, 3.91225°W) is the most northerly record of the species on the UK mainland and the distribution of Austrominius modestus within our field survey area showed a latitudinal boundary, with no detection of the species north of Balintore. However, this species has previously been detected in the Shetland Islands (Table S3; Hiscock 1978) and it therefore seems unlikely that its northwards spread has been limited only by temperature. Other potential limiting factors include: a relative lack of vector activity; unfavourable southwards residual currents on the UK east coast preventing natural dispersal northwards (McCubbin et al. 2002) and lack of suitable sheltered habitats on the open, exposed north and east coasts of Scotland. Austrominius modestus was abundant (field observation: between 0.1-1.0 per cm² in the upper inter-tidal zone) at the harbours, Balintore, Cromarty, Invergordon, and Avoch. These abundances may be supported by large breeding populations within the sea inlets, Cromarty Firth and Beauly Firth. Austrominius modestus is mainly found in sheltered waters (Crisp 1955, 1958; Foster 1971; Allen et al. 2006) and is tolerant of the lower salinities (Jones 1961; Barnes and Barnes 1974; Harms 1986) characteristic of these sea inlets. It is also known to out-compete native species in similar estuarine areas (Muxagata et al. 2004; Gomes-Filho et al. 2010; Witte et al. 2010).

The range expansions displayed in the north Scotland survey could also be explained by a lack of survey effort in the area, or by a lack of detection of certain species due to them being difficult to identify. The red algae Neosiphonia harvevi has been known in the British Isles for a long time but has relatively few records (Maggs and Stegenga 1999), and it is similar in morphology to some native species (Maggs and Hommersand The apparent range expansion Neosiphonia harveyi could therefore be an example of lack of detection and survey effort in the past. Re-processing of NNS survey samples taken by one of us (EJC) in 2006-2008 has identified Neosiphonia harveyi in various marinas on the west and east coast of Scotland (Table S3; Rodgers 2012).

Other species displaying range expansions to the north Scotland survey area (Botrylloides violaceus. Corella eumyota, Schizoporella japonica, and Tricellaria inopinata) are likely to be newly introduced to the area because these species are recent arrivals to both Scotland and Great Britain (Table S2). They are likely to have been secondarily introduced into the surveyed harbours through human-mediated dispersal. Introduction through hull fouling is presumed to be the most likely vector, especially as these species have short lecithotrophic larval periods and a low natural dispersal potential (Grosberg and Quinn 1986; Occhipinti Ambrogil and D'Hondt 1994; Watts et al. 1998; Lambert 2004; Marshall et al. 2006; Watts and Thorpe 2006).

Species not found

The species targeted but not detected in the north Scotland surveys were Asparagopsis armata, Bonnemaisonia hamifera, Sargassum muticum (Yendo) Fensholt, 1955, Bugula neritina, Crassostrea gigas, Crepidula fornicata, Didemnum vexillum, Styela clava, and Diadumene lineata. There are a number of potential reasons why they were not detected: their environmental requirements may not be met in the area; they may have not been introduced vet; they could be present in localised populations in areas or habitats that were not surveyed; or the survey techniques used may not have been suitable for their detection. Reasons for lack of detection will be species specific. Bugula neritina, Styela clava, Crepidula fornicata, and Crassostrea gigas are likely to be restricted to lower latitudes because they require warmer temperatures (Davis and Davis 2007; Dutertre et al. 2009; Ryland et

al. 2011: Bohn et al. 2012). Sargassum muticum and Didemnum vexillum are not restricted by the SST in north Scotland (Norton 1977; Rueness 1989; Bullard et al. 2007). They have also been detected during other rapid assessment surveys (Arenas et al. 2006; Beveridge et al. 2011) so their absence from our survey is not likely to be the result of limitations in the rapid survey technique. Although undetected during our surveys, Asparagopsis armata, Bonnemaisonia hamifera and Diadumene lineata have previously been reported in north Scotland. These species may not have been detected because they were not present at the sites surveyed, or because of methodological limitations. One methodological limitation for the detection of Bonnemaisonia hamifera is the timing of our surveys. The gametophyte stage of B. hamifera is present between February - July (Breeman et al. 1988). This does not completely coincide with our survey time (July-August); Bonnemaisonia hamifera may have already undergone senescence before the surveys were carried out and would not have been detected.

Harbour properties

Commercial and recreational vessels are important vectors for fouling NNS (Gollasch 2002; Minchin et al. 2006; Clarke Murray et al. 2011; Seebens et al. 2013). In our study, there were significant positive correlations between the number of fouling NNS detected and the size of harbour and number of vessels. Assuming that length of quayside and the number of vessels moored are reliable measures of vessel activity, this result is in agreement with previous studies that have shown an association between vessel activity and the likelihood of NNS introduction (Ricciardi 2006; Floerl et al. 2009). However, the parameters used here are only indirect measures of vessel activity. Direct measures of vessel activity can be difficult and expensive to obtain and the indices chosen for this study are logical alternative measures.

The majority of fouling NNS were found exclusively on floating structures, and although some were found in the intertidal areas of fixed structures these species were also present on floating structures. A greater number of fouling NNS were also found in harbours with floating structures. This is consistent with studies that have found a greater proportion of fouling NNS on floating structures than on fixed harbour structures (Glasby et al. 2007; Dafforn et al. 2009).

Fouling NNS may be more adapted for settlement on floating structures as they are analogous to vessel hulls (Neves et al. 2007), an important vector for fouling NNS. These findings from our study could however be biased by methodology because the subtidal areas of fixed structures were not surveyed, and therefore are not strictly comparable.

Implications of these non-native species in north Scotland

Some NNS can have negative impacts on native ecosystems and on economic activity: these species are termed 'invasive' and they are often capable of rapid spread (Occhipinti Ambrogi and Galil 2004: Keller et al. 2011). Of the species found in our surveys there is evidence that Austrominius modestus, Caprella mutica, Codium fragile fragile, Heterosiphonia japonica, and Tricellaria inopinata are invasive elsewhere and therefore may also be invasive in the north of Scotland. These NNS are known to spread rapidly and compete with native species (Crisp and Southward 1958; Trowbridge 1998; Occhipinti Ambrogil 2000; Lawson et al. 2004; Husa et al. 2004; Scheibling and Gagnon 2006; Cook et al. 2007; Shucksmith et al. 2009; Witte et al. 2010; Drouin et al. 2012; Johnson et al. 2012; Newton et al. 2013). Codium fragile fragile, Heterosiphonia japonica, and Austrominius modestus have also been seen to alter ecosystem function (Baird et al. 2012; Drouin et al. 2012; Krumhansl and Scheibling 2012), and Codium fragile fragile is considered one of the most invasive seaweeds in existence (Nyberg and Wallentinus 2005; Vila et al. 2010).

There is no current evidence to suggest the other five fouling NNS found in our surveys (Bugula simplex, Botrylloides violaceus, Corella eumyota, Neosiphonia harveyi, and Schizoporella japonica) could be invasive. However, their invasiveness may vet to be described. For example, although newly introduced into the British Isles, Schizoporella japonica is already showing signs of being invasive. After first being recorded in 2011 in north Wales (Holt and Cordingley 2011) it has rapidly expanded its distribution, with sightings from south west England (CA Wood, Marine Biological Association, UK pers. comm.) to the west and north coast of Scotland (Porter et al. 2012; this study). In Alaska, Schizoporella japonica may out-compete native encrusting bryozoans (Dick et al. 2005) and other Schizoporella species have been shown to prevent settlement

(Sutherland 1978) and overgrow other organisms, in some cases causing mortality (Turner and Todd 1994; Cocito et al. 2000). In our surveys, *Schizoporella japonica* was abundant where present and in Kirkwall Hatson Pier it dominated the low intertidal zone.

Conclusions

This study provides an overview of the current status of fouling NNS in Scotland and it will aid the assessment of temporal and spatial changes in the presence of fouling NNS. Monitoring the presence and absence of NNS is important for assessing the effects of anthropogenic activities, but it is also necessary for management and legislative purposes. For instance, EU member states are required to assess the success of Marine Strategy Framework Directive nonindigenous species targets (DEFRA 2014). The results of the north Scotland survey are particularly important, as they can serve as a baseline dataset to monitor changes in the presence and distribution of fouling NNS, which could be facilitated by the wave and tidal energy developments planned within the survey area. Our study also showed that busier and larger harbours hosted more fouling NNS; further suggesting that the increased vessel activity and harbour expansion associated with the wave and tidal energy industry, could promote the invasion of fouling NNS. There is a growing body of evidence that the invasion of NNS is facilitated by maritime industries similar to the wave and tidal energy industry (Kerckhof et al. 2011; Bouma and Lengkeek 2012), and this needs to be considered as a potential environmental impact by the industry and its regulators.

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The following supplementary material is available for this article:

- Table S1. Harbours sampled in the rapid assessment survey of north Scotland.
- Table S2. Checklist of marine fouling non-native species in Scotland.
- Table S3. List of marine fouling non-native species records in Scotland.
- **Table S4.** Marine fouling non-native species records from rapid assessment surveys of north Scotland.

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