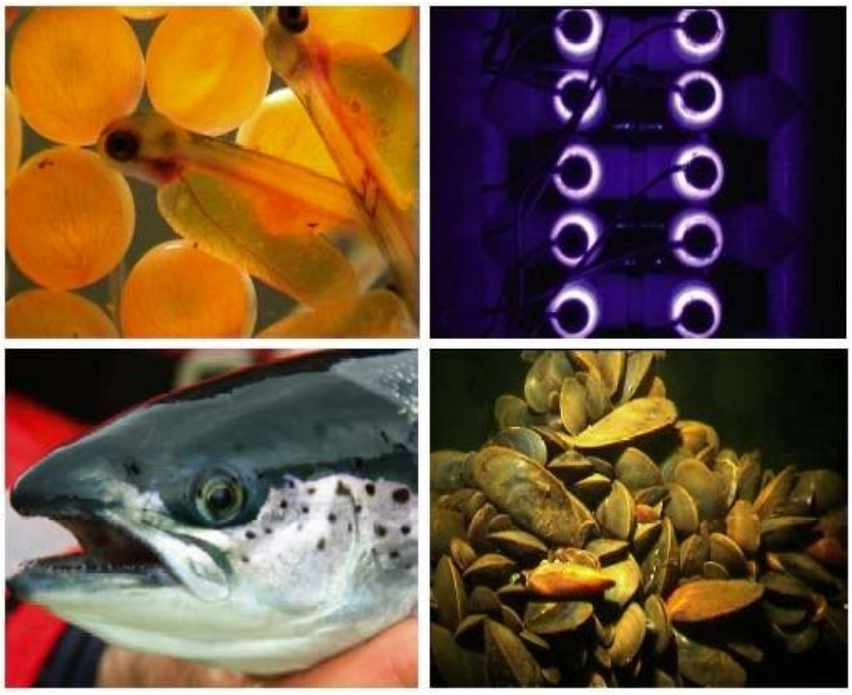
SARF099 - Survey of Pacific oyster in Scotland



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SURVEY OF WILD PACIFIC OYSTER *CRASSOSTREA GIGAS* IN SCOTLAND

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Figure 1 (Front Page). Wild Pacific oyster *Crassostrea gigas* found in Balcary Bay, Solway Firth (Photo: C. Beveridge, SAMS)

EXECUTIVE SUMMARY

SURVEY OF WILD PACIFIC OYSTER *CRASSOSTREA GIGAS* IN SCOTLAND

Contractor: Scottish Association for Marine Science (SAMS)

Year of publication: 2015

Background

Global sea surface temperatures have accelerated in the last few decades (IPCC, 2007). Some of the most rapid increases have been seen in the NE Atlantic (IPCC, 2007), where increases of up to 1 ^{o}C have been recorded (Hawkins *et al.*, 2003, Inall *et al.*, 2009) and mean seawater surface temperatures have risen by 0.57 ^{o}C per decade since 1975 (Inall *et al.*, 2009). Furthermore, an increase in magnitude and frequency of short-lived, high temperature events is predicted (Meehl & Tebaldi, 2004), which could cause significant changes in marine hard substrate communities (Sorte *et al.*, 2010). Periodic increases of 2 ^{o}C have been observed off the west coast of Scotland (Inall *et al.*, 2009). This warming seawater has been linked to the northwards spread of certain non-native species in the UK, such as the Pacific oyster *Crassostrea gigas* (Thunberg, 1793) (Maggs *et al.*, 2010). A recent report suggests that it is highly likely that 'wild' populations of *C. gigas* will continue to expand northwards and become established on the west coast of Scotland by the 2020s (Cook *et al.*, 2013).

Until recently, it was thought that the northern most 'wild' populations of *C. gigas* in Great Britain were located in Lough Foyle, North coast of Northern Ireland (Kochmann *et al.*, 2012). However, *C. gigas* was recently reported from the Firth of Forth on the east coast (Smith *et al.*, 2014) and the Solway Firth (Clair McFarlan, Solway Firth Partnership, pers. comm.) and Loch Fyne (J. Khan-Marnie, SEPA, pers. comm.) in south-west Scotland.

The presence of wild populations of the Pacific oyster *Crasssostrea gigas* was assessed in eleven regions in Scotland. An extensive, co-ordinated survey programme was undertaken between March and September 2014. Sixty locations were surveyed on the west and east coasts of Scotland using a standardised protocol and specific factors, previously associated with the presence of *C. gigas,* were recorded. Sites were chosen based on regions; (i) where 'wild' *C. gigas* had already been found , (ii) where *C. gigas* was commercially cultivated and (iii) where suitable habitat and environmental conditions (e.g., summer seawater temperatures and water retention rates) existed for spawning, thus increasing the probability of the region supporting 'wild' populations.

Main findings

- Wild *C. gigas* were found at 5 out of the 60 sites surveyed.
- All of these sites were located in the Solway Firth, south-west Scotland and the oyster densities were extremely low, ranging from single individuals to < 5 individuals per m^2. Analysis of the size-frequency distributions in the three sites, where > 3 individuals were recorded, suggested the possibility of more than one recruitment event in the region over the last few years.
- This survey provides the Scottish aquaculture industry and the environmental agencies with baseline information on the presence of 'wild' *C. gigas* populations in Scottish waters, together with a repeatable methodology that can be used for future monitoring.
- Statistical analysis was unable to be performed to determine the relationship between the occurrence of the wild *C. gigas* and the substrate type, residence times of the embayments or lochs surveyed and proximity to aquaculture due to the absence of this species in ten of the eleven regions surveyed.
- From observations of substrate type, however, exposed bedrock and large boulders which were devoid of macroalgae, appeared to provide suitable habitat for wild *C. gigas* in the Solway Firth region. In many cases, the oyster shells and the surrounding hard substrata were heavily fouled with barnacles. Substrate with dense macroalgal coverage was deemed unsuitable for the settlement and/or establishment of wild *C. gigas*.
- This information can be used to assist in the development of policies that balance and support the continuing development of the Scottish Pacific oyster farming industry and the maintenance of status of sites designated under Natura 2000.

Note: Supplementary information and photographs available with accompanying files.

Table of Contents

1. INTRODUCTION

Global sea surface temperatures have accelerated in the last few decades (IPCC, 2007). Some of the most rapid increases have been seen in the NE Atlantic (IPCC, 2007), where increases of up to 1 ^{o}C have been recorded (Hawkins *et al.*, 2003, Inall *et al.*, 2009) and mean seawater surface temperatures have risen by 0.57 ^{o}C per decade since 1975 (Inall *et al.*, 2009). Furthermore, an increase in magnitude and frequency of short-lived, high temperature events is predicted (Meehl & Tebaldi, 2004), which could cause significant changes in marine hard substrate communities (Sorte *et al.*, 2010). Periodic increases of 2 ^{o}C have been observed off the west coast of Scotland (Inall *et al.*, 2009). This warming seawater has been linked to the northwards spread of certain non-native species in the UK, such as the Pacific oyster *Crassostrea gigas* (Maggs *et al.*, 2010). A recent report suggests that it is highly likely that 'wild' populations of *C. gigas* (i.e., found attached to the substratum) will continue to expand northwards and become established on the west coast of Scotland by the 2020s (Cook *et al.*, 2013).

Until recently, it was thought that the northern most 'wild' populations of *C. gigas* in Great Britain were located in Lough Foyle, North coast of Northern Ireland (Kochmann *et al.*, 2012), however, wild *C. gigas* was recently reported from the Firth of Forth on the east coast (Smith *et al.*, 2014) and the Solway Firth (Clair McFarlan, Solway Firth Partnership, pers. comm.) and Loch Fyne (J. Khan-Marnie, SEPA, pers. comm.) in south-west Scotland.

The expansion of 'wild' populations of *C. gigas* in Scotland will have complex implications for both the oyster growing industry, which forms a key component of the Scottish aquaculture industry and for the environment agencies, which are responsible for safeguarding biodiversity driven by legislative instruments, such as the EU Habitats Directive (92/43/EEC), the Water Framework Directive (2000/60/EC), the Marine Strategy Framework Directive (2008/56/EC) and the Council Regulation concerning the use of alien species in aquaculture (Reg. No, 708/2007) (see review in Herbert *et al.*, 2012), hence the need for a baseline survey to determine the full extent of *C. gigas* in Scotland.

The aim of this research, therefore, was to conduct a survey for populations of 'wild' *C. gigas* on the east and west coasts of Scotland, concentrating on the areas; (i) where wild *C. gigas* had already been found, (ii) where *C. gigas* was commercially cultivated and (iii) where suitable habitat and environmental conditions (e.g., summer seawater temperatures and water retention rates) existed for spawning, thus increasing the probability of the region supporting 'wild' populations.

2. METHODOLOGY

2.1 Site Selection

The survey sites were chosen based on a thorough understanding of the habitat preferences and physiological tolerances of *Crassostea gigas*, together with the physical properties of the embayment (i.e., wave exposure, embayment residence time, intertidal area) and proximity to either existing or former oyster farming activity. In the case of the former, *C. gigas* is known to have a particular preference for hard substratum including bedrock, boulders, cobbles, pebbles and biogenic reefs (Kochmann *et al.*, 2013), shell fragments, aquaculture trestles and harbour walls (Reise *et al.*, 1998, Nehls & Büttger, 2007, Kochmann *et al.*, 2012) in the lower intertidal zone (Kochmann *et al.*, 2013). This species is also known to prefer sites with an intertidal width ≥ 50m and extended water residence times (Kochmann *et al.*, 2013).

In terms of physiological tolerances, seawater temperature is a key factor in determining the length of the conditioning, spawning and recruitment phase in *C. gigas* (Syvret *et al.*, 2008). The number of days that the temperature is above a certain threshold (i.e. degree days) being critical in determining the timing of these three phases (Mann, 1979). For example, at 18 ^{o}C, *C. gigas* requires 30 days (or 224 degree days above 10.55 ^{o}C) for successful recruitment (Magoon & Vining, 1981). In addition, *C. gigas* prefers salinities over 11, so this was also accounted for in site selection. Based on the above knowledge, plus additional information from the Scottish Sea loch catalogue (Edwards & Sharples, 1986), the FRS sealoch database, the SAMS Sanitary Survey database, which monitors shellfish harvesting areas throughout Scotland for the Food Standards Agency, and personal contacts, a list of 60 survey sites in 11 regions was produced, including the regions where *C. gigas* has recently been found (Fig. 2; Appendix 1).

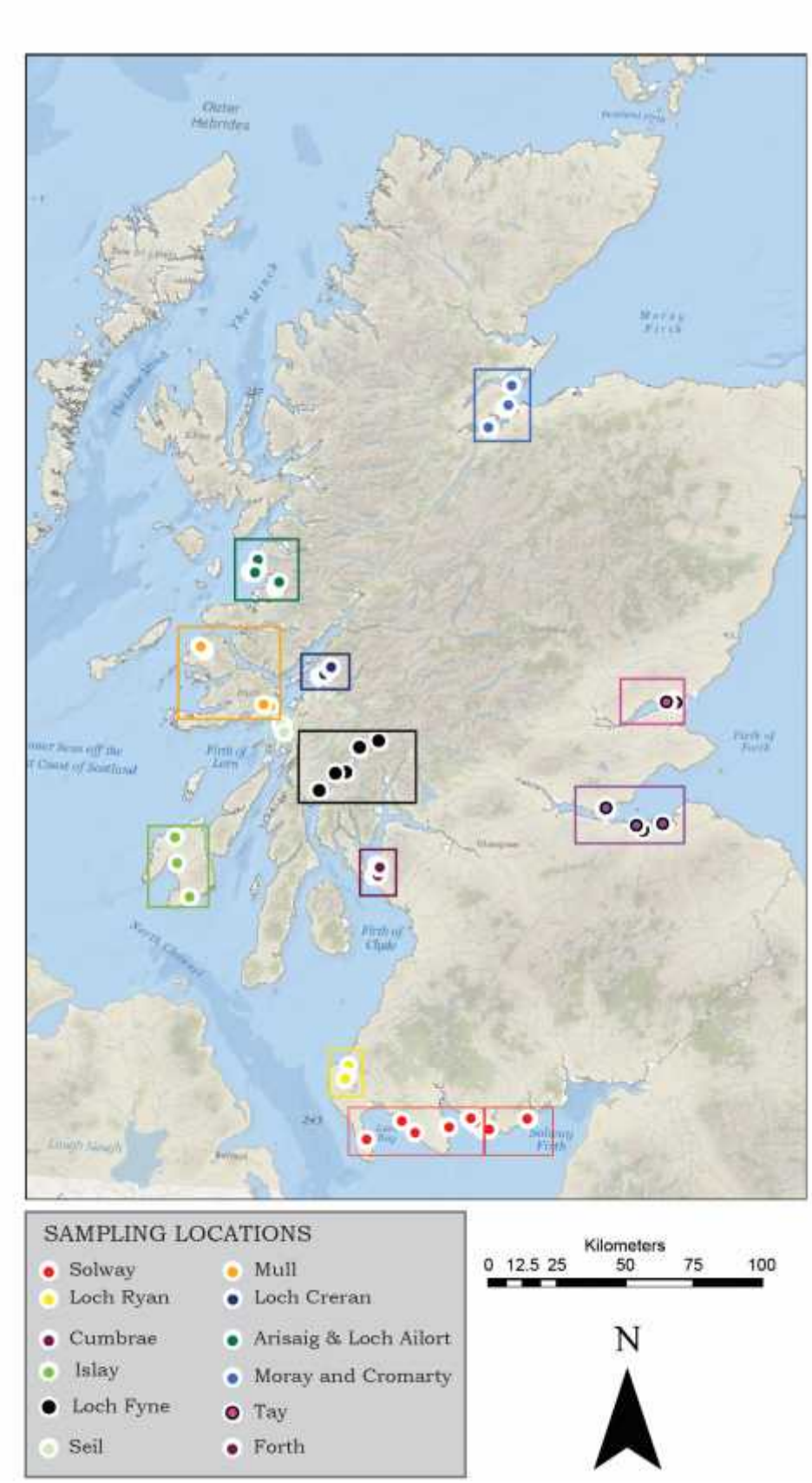

Figure 2. Map showing the regions and sites for the Scottish Pacific oyster survey 2014.

Once the sites were agreed with the SARF steering group, the sampling programme was based on a modified version of Kochman et al. (2013), Burrows et al. (2008) and the NSW Department of Primary Industries (2012). The sampling of a specified region was undertaken within 2 hours either side of Mean Low Water Springs between 31 March and 12 September 2014. Intertidal width for each site was measured from the Mean High Water Spring tide mark to the Mean Low Water Spring tide mark using GPS and residence time was determined, where possible using the formula developed by Hartnett et al. (2011), in conjunction with the physical data provided in the sea loch catalogue (Edwards & Sharples, 1986), the MSS sealoch database and the SARF017 report (Gillibrand *et al.*, 2006). Initially, a 1 m wide transect line parallel to the waterline in the lower intertidal zone was followed by the survey team. However, due to the uneven terrain and absence or low abundance of wild *C. gigas* at most of the sites, the surveyors increased the width of this transect to >5m.

The presence or absence of wild *C. gigas* was recorded, together with the predominant substrate type, associated species present at each site (e.g., native oyster *Ostrea edulis*, mussel *Mytilus edulis*) and any other non-native species present were noted. A handheld GPS (Garmin GPS72) was used to record the start and finish points of each transect and the position of individual *C. gigas*, if present, during the survey. If the density of *C. gigas* exceeded 'Frequent' (small patches, 5%; ≥10 small ind. per 0.1 m^2; ≥1 large ind. per 0.1 m^2), then the extent of the 'patch' was to be recorded using waypoints on the handheld GPS. In addition, quadrats of 0.25 m^2 would be used to estimate overall abundance, and abundance of individuals of a reproductively viable age (i.e., >40mm).

Shell length and width of *C. gigas* was measured using digital Vernier callipers and where a positive sighting was recorded, specimens were collected for confirmation of identification and a sample of mantle and gill tissue was preserved in 100% alcohol for future genetic analysis by Marine Scotland Science, Aberdeen.

2.3 GIS Mapping of Sites

ArcGIS maps were produced for each of the sites visited, including positive and negative records of wild *C. gigas* using ArcGIS 10.0.

2.4 Statistical Analysis

Logistic regression, using a mixed model to account for random factors was to be used to predict the probability of finding wild *C. gigas,* as a function of the environmental variables recorded. In addition, in areas where wild *C. gigas* was already present, logistic regression, using a nested and mixed model, was to be used to model the probability of finding individuals of a reproductively viable age, as a function of the environmental variables. However, as a result of the absence of wild *C. gigas* from the majority of the sites and lack of any spatial replication this detailed analysis could not be performed.

2 RESULTS

A total of 60 locations were visited during the survey, covering sites within eleven regions in Scotland including; (i) Solway Firth (ii) Loch Ryan, (iii) Isle of Cumbrae, (iv) Loch Fyne, (v) Seil Island, (vi) Isle of Mull, (vii) Loch Creran, (viii) Arisaig and Loch Ailort on the west coast and (ix) Moray and Cromarty, (x) Tay Estuary and the (xi) Firth of Forth on the east coast of Scotland (Fig. 2; Appendix I).

3.1 Location of *Crassostrea gigas* recorded during Scottish Survey

A total of 49 live wild *Crassostrea gigas* were found attached to the substratum in 5 sites surveyed in the Solway Firth, a large estuary in south-west Scotland (Fig. 3a, b; Table 1). Live, wild *C. gigas* were not found at any of the other sites (see Appendix II for detailed maps), even though wild *C. gigas* had been found in previous surveys in the Firth of Forth (verified sightings) and Loch Gair (unverified; Loch Fyne) (see Appendix III for previous sightings of *C. gigas*). The greatest number of wild *C. gigas* was found at 3 sites in the Solway Firth including; Ross Bay, Balcary Bay and Brighouse Bay (Fig. 3b). Apart from the individuals that were removed for genetic analysis, all other individuals were left *in situ*. The native oyster, *Ostrea edulis* was also found at a number of the survey sites (Appendix I) and it should be noted that this species was typically attached by the convex or 'cup' valve, whereas wild *C. gigas* was always found attached with the flattened valve.

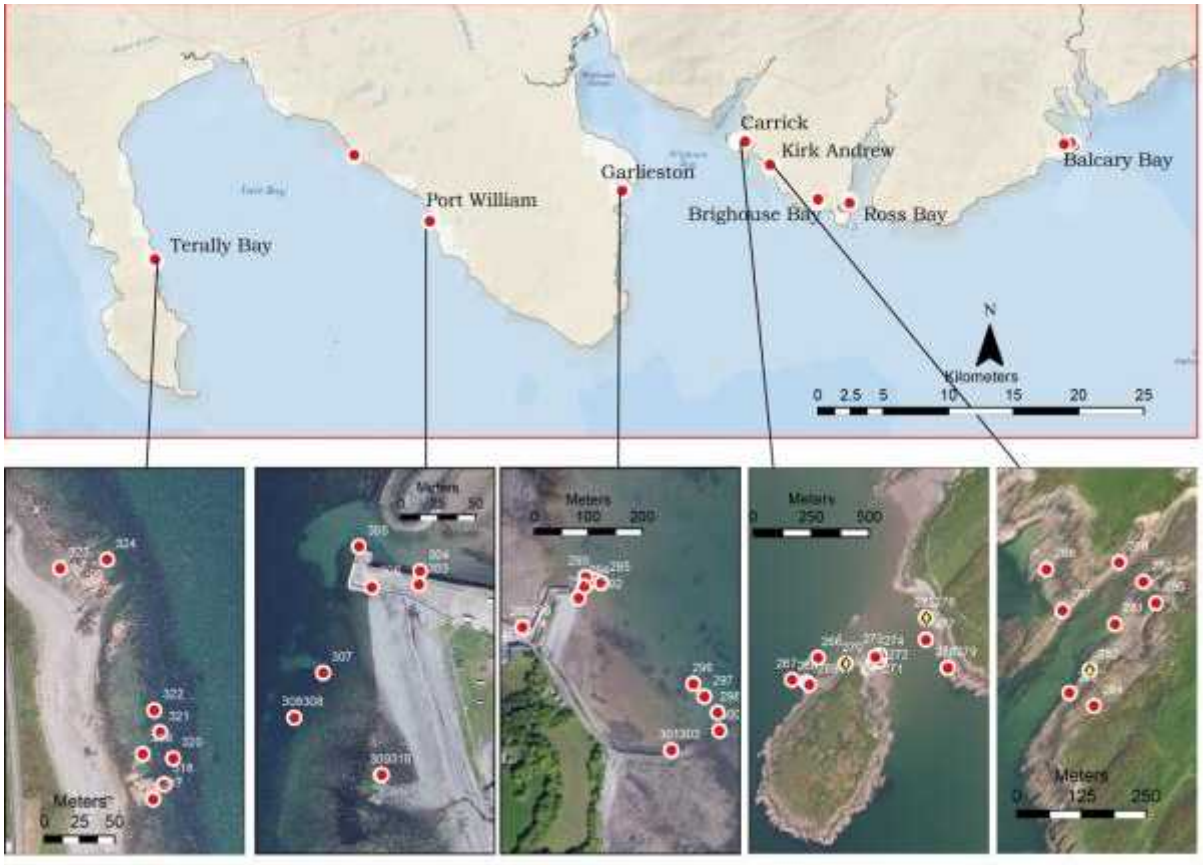

Figure 3a. Survey sites in the north-western region of the Solway Bay (as shown by red dots). Detailed maps are shown below of survey sites from Kirk Andrew to Terally Bay, where multiple surveys were conducted with corresponding unique survey code (shown next to red dot). The position of live, wild *C. gigas* is shown (yellow diamond).

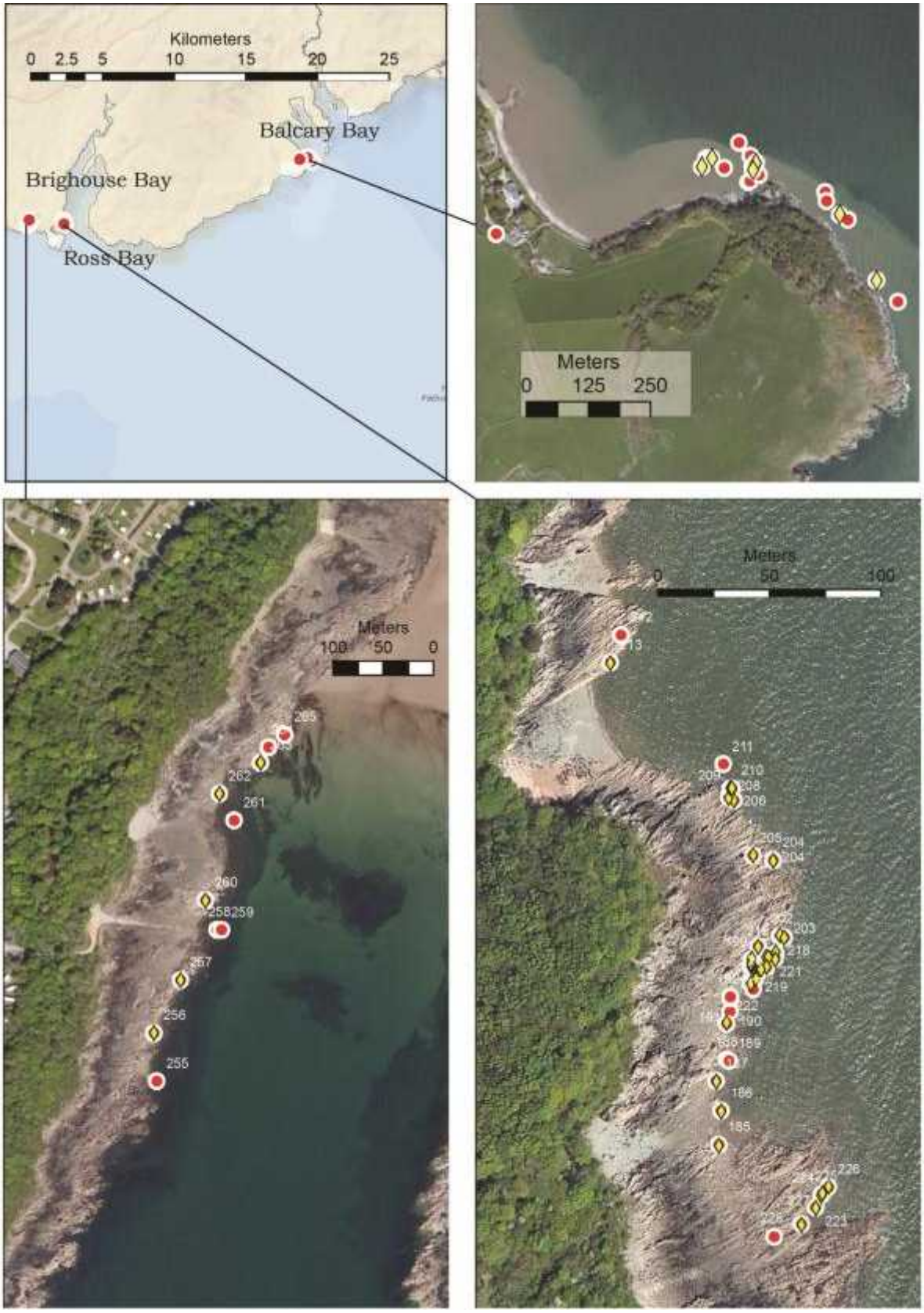

Figure 3b. Survey sites in the north-eastern region of the Solway Bay (as shown by red dots) from Balcary Bay to Brighouse Bay. Detailed maps of the survey sites are shown to the right and below, with

corresponding unique survey code (shown next to red dot). The position of live, wild *C. gigas* is shown (yellow diamond).

Table 1. Locations and abundance of 'live' wild *Crassostrea gigas* found on Scottish Survey. B = Bedrock; BB = Bedrock encrusted with barnacles; BO = Boulder; SB = Small boulder; C = Crevice in bedrock.

Region	Site Name	Date	Intertidal Width (m)	Residence Time (d)	Total No. per site	Latitude	Longitude	Dimensions (L x W; mm)	Habitat Type
Solway Firth	Ross Bay	13/07/14	40 – 80	< 1	37	54.78255	4.090222	75.0 x 70.0	BB
						54.78269	4.090213	104.2 x 78.8	BB
						54.78281	4.090253	90.5 x 91.2	BB
						54.78321	4.090034	96.0 x 88.2	BB
						54.78328	4.09002	87. 1 x 68.6	C
						54.78328	4.090031	93.4 x 69.8	BB
						54.78329	4.090033	75.1 x 75.1	BB
						54.78331	4.090032	70.1 x 57.7	BB
						54.78331	4.090029	75.8 x 46.6	BB
						54.78336	4.089988	84.0 x 70.4	BB
						54.78332	4.08993	103.1 x 88.0	BB
						54.78341	4.089839	94.5 x 86.9	BB
						54.7834	4.089807	69.8 x 59.8	BB
						54.78371	4.089901	62.5 x 70.4	BB
						54.78373	4.090042	62.2 x 36.0	BB
						54.78395	4.090186	62.5 x 62.3	BB
						54.78371	4.089901	58.0 x 56.7	BB
						54.78396	4.090225	74.3 x 69.3	BB
						54.784	4.090215	95.2 x 93.2	BB
						54.784	4.090205	116.4 x 80.0	BB
						54.78449	4.091082	92.0 x 70.0	BB
						54.78334	4.089864	82.1 x 73.5	BB
						54.78331	4.089867	111.1 x 133.9	BB
						54.78332	4.089913	55.4 x 46.0	BB
						54.78328	4.089924	85.1 x 78.5	BB
						54.78327	4.089962	69.3 x 51.7	BB
						54.78323	4.089998	81.8 x 75.9	BB

						54.78323	4.089997	62.0 x 55.7	BB
						54.78305	4.090194	143.9 x 96.8	BB
						54.78231	4.089531	70.6 x 61.2	BB
						54.78236	4.089496	94.0 x 63.3	BB
						54.78237	4.089487	86.7 x 88.1	BB
						54.7824	4.089446	103.5 x 69.0	BB
						54.78224	4.089627	102.2 x 62.0	BB
	Balcary Bay	14/07/14	60 – 120	<1	7	54.82802	3.830733	79.7 x 66.9	SB
						54.82786	3.831037	76.7 x 53.8	BO
						54.8279	3.829387	90.0 x 80.0	C
						54.82796	3.829354	69.0 x 59.7	BO
						54.82707	3.8267	68.4 x 73.7	BO
						54.8259	3.825497	76.8 x 69.1	BO
						54.82783	3.829443	76.7 x 67.2	C
	Brighouse Bay	14/07/14	50 – 100	<1	3	54.78154	4.129339	84.2 x 76.2	B
						54.78274	4.128602	65.8 x 48.2	B
						54.7837	4.128441	112.2 x 72.4	B
	Carrick/ Ardwell Isle	15/07/14	60 – 125	<1	1	54.82414	4.218084	34.6 x 39.3	B
	Kirk Andrew	15/07/14	60 – 250	<1	1	54.80519	4.184687	58.4 x 40.0	C
TOTAL LIVE OYSTERS					**49**				

3.3 Density of wild *Crassostrea gigas*

The live wild *C. gigas* found during the survey were typically widely dispersed at the five sites where they were recorded. The greatest number of *C. gigas* (n = 37) was found in Ross Bay, Solway Firth, but even at this site the density was below the threshold level of 'frequent' (i.e. small patches, 5%; ≥10 small ind. per 0.1 m2; ≥1 large ind. per 0.1 m2) and so, 'patch' size was not measured (Fig. 4). At Balcary Bay and Bridghouse Bay, seven and five live individuals were recorded respectively, whereas at Carrick and Kirk Andrew only a single individual was recorded at each site.

Figure 4. Single *Crassostrea gigas* on barnacle, encrusted bedrock (indicated by arrow) in Ross Bay, Solway Firth (Photo: P. Lamont, SAMS).

3.4 Habitat associated with wild *Crassostrea gigas*

The live wild *C. gigas* recorded in this survey were typically found in the lower intertidal, inhabiting bedrock, bedrock encrusted with barnacles, large and small boulders and crevices found in the bedrock (Figs. 5-7). The substrate inhabited by *C. gigas*, appeared to be site specific (Table 1), however, due to the low densities of oysters found at only a small number of sites, no statistical analysis could be performed to determine any association between the presence of oysters and their habitat. *Crassostrea gigas* was not found in habitats dominated by bedrock or boulders, which were overgrown by macroalgae (Fig. 8). No wild *C. gigas* were found directly attached to macroalgae.

Figure 5. Typical substrate type (i.e. bedrock encrusted with barnacles) where live, wild *C. gigas* (indicated by arrow) were recorded in Ross Bay, Solway Firth (Photo: C. Beveridge, SAMS).

Figure 6. Typical substrate type (i.e. boulders encrusted with barnacles) where live, wild *C. gigas* (indicated by arrow) were recorded in Balcary Bay, Solway Firth (Photo: C. Beveridge, SAMS).

Figure 7. Typical substrate type (i.e. boulders) where live, wild *C. gigas* (indicated by arrow) were recorded in Brighouse Bay, Solway Firth (Photo: C. Beveridge, SAMS).

Figure 8. The probability of sighting of wild *C. gigas* on boulders covered with macroalgae was considered low by the survey team based on their observations throughout the survey (Photo: C. Beveridge, SAMS).

3.5 Size distribution of wild *Crassostrea gigas* in the Solway Firth

At Ross Bay, Solway Firth where the greatest abundance of wild *C. gigas* was observed in this survey, the oysters ranged in size from 55.4 to 143.9 mm (n = 37). Smaller size ranges were observed at the other two sites, where multiple live individuals were found. In Balcary Bay, the oysters ranged from 68.4 to 90.0 mm (n = 7) and in Brighouse Bay (n = 3) the size range was from 65.8 to 112.2 mm (Fig. 9). The smallest live oyster measured 34.6 mm and was a single individual recorded in Carrick.

3.6 Intertidal width

Intertidal width at 90 % (n = 54) of the survey sites was > 40 m, with the exception of sites where specific structures (i.e., pilings, harbour walls, marina pontoons) were surveyed (Appendix I). All five sites in the Solway Firth, where live wild *C. gigas* were recorded, had an intertidal width of over 40 m (Table 1).

3.7 Residence time

Residence times for the sites surveyed ranged between <1 to 223 days (Appendix I). At the five sites, where live wild *C. gigas* was recorded, an average residence time of < 1 day was calculated (Table 1).

3.8 Commercial *Crassostrea gigas* cultivation

Commercial *C. gigas* cultivation was recorded within 10 km of 57 % (n = 34) and 50 km of 0.05 % (n = 3) of the sites surveyed. Sites in Loch Ryan (south west Scotland) and Moray & Cromarty, Tay Estuary and the Firth of Forth on the east coast of Scotland were all > 500 km from commercial *C. gigas* cultivation areas (Appendix I). In the Solway Firth, the five sites, where live wild *C. gigas* were recorded, were between 28 and 95 km from a commercial *C. gigas* cultivation site in Silloth, south Solway coast.

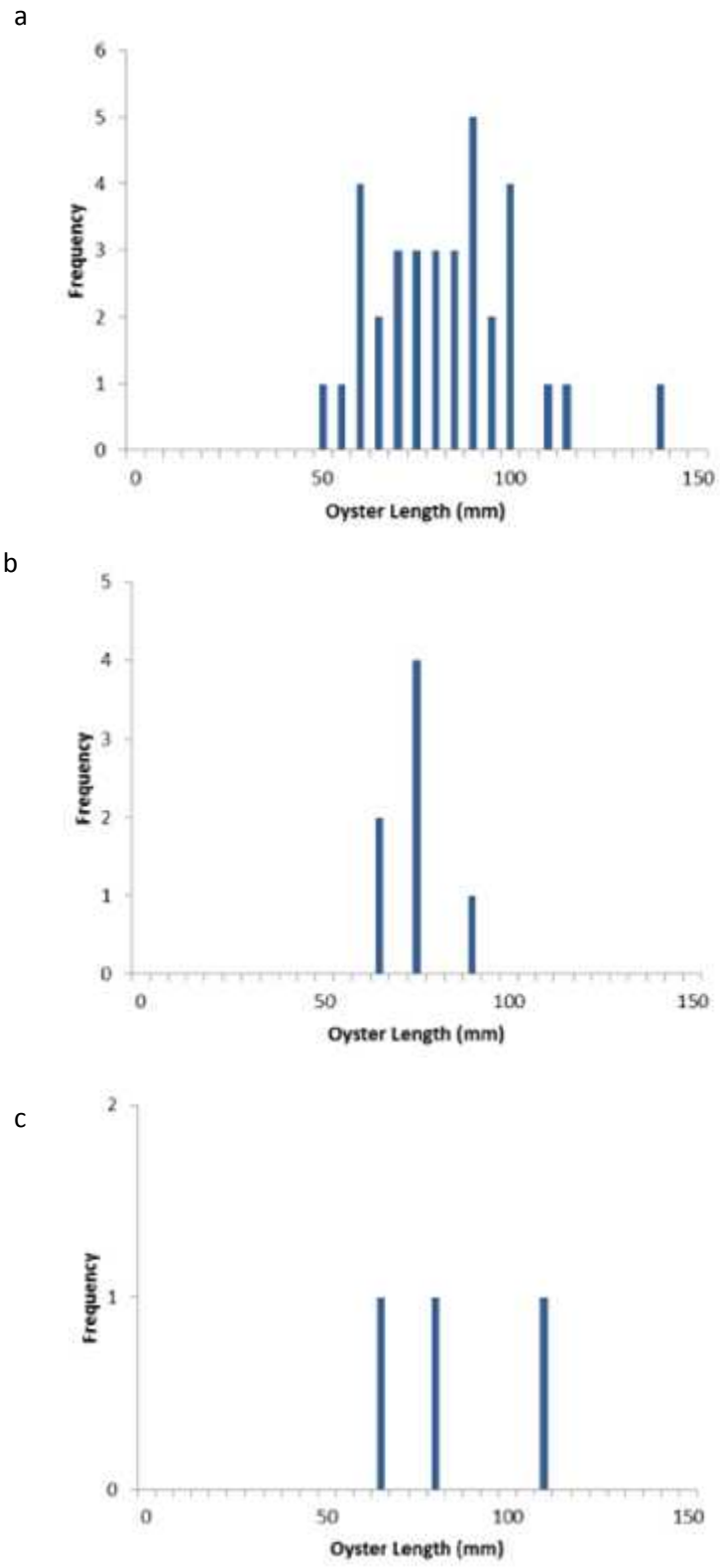

Figure 9. Size-frequencies of wild *Crassostrea gigas* in 5 mm size intervals at (a) Ross Bay; n = 37, (b) Balcary Bay; n = 7 and (c) Brighouse Bay; n = 3.

4. DISCUSSION

No intertidal reefs of the Pacific oyster *Crassostrea gigas* were found in Scotland, unlike the extensive reefs recorded in southern UK and other European countries (see review in (Herbert *et al.*, 2012). Wild *C. gigas* were found in the intertidal zone at 5 of the 60 sites surveyed. These sites were all located in the Solway Firth, a large estuary in south-west Scotland. Densities of wild *C. gigas* at the five sites were from single individuals to less than 1 large individuals per 0.1 m^2. In other regions of Europe, similar densities to these were recorded at various time intervals before *C. gigas* intertidal reefs became established, including the Wadden Sea (Diederich *et al.*, 2005), Argentina (Orensanz *et al.* 2002), Sweden and Denmark (Wrange *et al.*, 2010).

The largest wild *C. gigas* found in this study reached between 112 and 144 mm in length. These sizes are comparable to the largest oysters found in recent studies in Northern Ireland, where the largest oysters reached lengths of 155 mm (Guy & Roberts, 2010) and 146 mm (Kochmann *et al.*, 2013). An estimate of age, based on an age-size relationship study on populations in Strangford Lough, Northern Ireland (Guy & Roberts, 2010), suggests that the largest oysters found in this study are approximately 6 years old. Diederich (2005), however, stresses that the age-size relationship can vary between sites and so this estimate must only be used tentatively as an indication of age.

The majority of the live wild *C. gigas* that were recorded in this survey, were between 50 and 100 mm in length, which is comparable to the size of the oysters recently found at Whitehouse Bay, Firth of Forth (Smith *et al.*, 2014). Previous studies in the Wadden Sea have shown that *C. gigas* in the first year after settlement typically reach 20 – 50 mm and in their second year reach 30 – 80 mm on mussel beds (Reise *et al.*, 1998, Fey *et al.*, 2010). Smith et al. (2014), however, found using the acetate-peel method of Richardson et al. (1993), that *C. gigas* recorded in the Firth of Forth, measuring between 84 – 91 mm were approximately 4 years of age. This suggests, therefore, that the wild *C. gigas* recorded in this survey were potentially between 2 – 4 years old. No evidence of recent recruitment was found in the three main areas, where multiple individuals were found, although at one site (Carrick, Wigtown Bay, Solway Firth) a single individual measuring 34 mm in length was recorded. Interestingly, two *C. gigas* of < 25 mm in length were also found in Ravenshall, which is located approximately 6 km north of Carrick in 2013 (Nic Coombey, Solway Firth Partnership, pers. comm.), suggesting a recruitment event had taken place within recent years. The size range at one of the sites surveyed (Ross Bay; > 80 mm), is comparable to populations in Northern Ireland, where a similar range of sizes has been recorded in several bays in Lough Swilly (Kochmann *et al.*, 2013).

Live wild *C. gigas* in this current study were all found associated with either bedrock or boulders of varying sizes. Typically, the substrate and the oysters were covered with barnacles. No oysters were found under macroalgae. The settlement of oysters onto hard substratum has widely been documented (Diederich *et al.*, 2005, Nehls *et al.*, 2006) and the positive association of 'Hardreef' with oyster presence was found to be a good indicator for the settlement of *C. gigas* based on an Irish survey in 2009 (Kochmann *et al.*, 2013). The lack of settlement in areas with dense macroalgal coverage was also observed by Kochmann et al (2013). Inhibiting metabolites exuded by the macroalgae (Brock *et al.*, 2007) and the disturbance to the underlying substratum by the movement of the fronds (Jenkins *et al.*, 2009) were both suggested as likely to limit the settlement of invertebrate larvae and may have affected the recruitment and mortality of the oysters.

Kochman et al (2013) found a positive correlation between the width of the intertidal area and oyster presence, with a greater likelihood of oyster presence being recorded on extensive, intertidal shores (> 50 m). Similarly, in the current study, live *C. gigas* were all found at sites in the Solway Firth, where the intertidal width was greater than 40 m. This suggests that shores with a width of >40 m could be used as a potential indicator for the presence of *C. gigas* in the future, in conjunction with other environmental parameters, such as substrate type.

The larvae of *C. gigas* can remain in the water column for 3 – 4 weeks prior to settlement (Quayle, 1988). However, this is highly dependent on seawater temperature and nutrition and Magoon and Vining (1981) predicted that at 18 ^{o}C, *C. gigas* would require at least 30 days (or 224 degree days above 10.55 ^{o}C) for the larvae to be competent to settle. Only survey sites in the upper basins of Loch Fyne, Loch Ailort and Loch Creran were found to exceed the 30 days residence times and no oysters were found at these sites. Whereas, live wild *C. gigas* were found in five bays in the Solway Firth, where residence times were < 1 day. A commercial oyster farm operates at Silloth, south Solway coast and a significant population of large *C. gigas* (> 300 individuals), inhabits the stone piers at the entrance to Ramsey Harbour, north-east Isle of Man (Manx Wildlife Trust, pers. comm., E. Cook, pers. obs.). The eastern side of the Irish Sea is typically shallower (< 50 m) and has a longer residence time compared with the western side. The currents in the eastern region are also weaker, more varied in circulation and more influenced by wind strength and direction than those to the west (Kennington & Rowlands, 2005). In this region, the predominant wind direction is from the south-west, which *may* assist with the transfer of oyster larvae from more southerly populations, such as the ones in Ramsey, Isle of Man and Silloth and with the retention of oyster larvae within the northern section of the Solway Firth. It could be suggested, that in certain cases, such as the Solway Firth, the average residence time of the specific bay, in which the *C. gigas* have been found may not be as critical, as other environmental parameters for larval recruitment. These parameters may include; optimum seawater temperatures, availability of suitable substrate, orientation of the site to the prevailing wind direction and increased retention time of a site, caused by specific weather conditions, together with biological variables, such optimal larval nutrition, predation and competition.

From production to ultimate consumption, the annual Gross Output for British reared *C. gigas* has recently been estimated at over £13 million (Humphreys *et al.*, 2014). In Scotland, the production of *C. gigas* is an important source of income, particularly for rural communities on the west coast of Scotland, with approximately 1.9 million *C. gigas* produced for the table and 6.2 million individuals produced for on-growing (Munro & Wallace, 2014). The use of non-natives, however, for aquaculture can increase the risk of unintentional escapes (Cook *et al.*, 2008). Many species are cultivated in regions where they do not originate and increasingly, these countries are having to deal with the environmental and economic consequences of the establishment (i.e. able to survive and reproduce) of these species in the wild (Cook *et al.*, 2008). In this current study, the majority of the sites on the west coast of Scotland were specifically chosen to be within a short distance (<20 km) of oyster cultivation, due to the potential for these cultured oysters to act as source populations. No *C. gigas* were found within 20 km of any oyster culture activity and in fact, live wild oysters were found in a previous survey on the east coast of Scotland (Firth of Forth), where cultivation has not been attempted since the late 1970s or 1980s (Smith *et al.*, 2014). In the case of the latter, it was hypothesised that the wild *C. gigas* were either (i) related to the earlier cultivated stock, (ii) discards from oysters bought for consumption but not consumed, (iii) introduced as larvae from the ballast water of commercial shipping or recreational craft, or spawned by adults introduced to the region

attached to ships' hulls (see (Smith *et al.*, 2014)). As seawater temperatures increase over the next few years, however, it will be important to closely monitor seawater temperature, together with gonadal development, spawning and frequency of larvae in the water column and recruitment of wild *C. gigas* to hard substratum around cultivation sites, particularly in southern Scotland. In The Netherlands, Germany and France, populations of *C. gigas* were intentionally introduced, as water temperature was considered too low for establishment. Within 20 years, however, wild populations had become established in each of these countries (Ruesink *et al.*, 2005) and more recently southern Britain (Herbert *et al.*, 2012) and Ireland (Kochmann *et al.*, 2013).

Kochman et al. (2013) proposed a number of criteria for determining the likelihood of *C. gigas* establishment at a particular location based on their survey of 69 sites throughout Ireland, where over 730 individuals were found at 18 sites. The sites with the greatest probability of supporting populations of wild *C. gigas* would have; (i) oyster aquaculture, (ii) temperature regimes that supported oyster spawning and larval development, (iii) long residence times, (iv) hard substrata (v) intertidal areas > 50 m and (vi) low percentage cover of macroalgae. The results of this survey are in general agreement with the last three criteria, however, a lack of adequate spatial data for *C. gigas* in the current Scottish survey, meant that a similar analysis to that of Kochmann et al. (2013) could not be achieved. Combining the data from the Scottish and Irish studies, however, may provide greater insight into the environmental variables that govern the early life-stages of *C. gigas* and could help to predict where this species is likely to become established in Scotland in the future.

To conclude, the northernmost sightings of wild *C. gigas* were recorded in this survey at 54.8^{o} N on the west coast of Scotland and this survey suggests that wild *C. gigas* in Scottish waters are currently at very low densities. These populations, however, are further south than wild populations of this species recorded recently in Ireland (55.1^{o} N) (Kochmann et al., 2013) and on the east coast of Scotland (56^{o} N) (Smith et al., 2014). Since, several populations of wild *C. gigas* have been reported from Scandinavia at 60^{o} N (Wrange et al., 2010), it is highly likely that this species will continue to spread northwards throughout mainland Britain, as a consequence of increasing seawater temperatures (Cook et al., 2013). It is likely that further spread will have complex implications in the future for both the oyster growing industry, which forms a key component of the Scottish aquaculture industry and for the environment agencies, which are responsible for safeguarding biodiversity driven by legislative instruments, such as the EU Habitats Directive (92/43/EEC), the Water Framework Directive (2000/60/EC), the Marine Strategy Framework Directive (2008/56/EC) and the Council Regulation concerning the use of alien species in aquaculture (Reg. 708/2007) (see review in (Herbert et al., 2012)). It is envisaged that the results of this survey will directly inform the industry and the relevant authorities' discussion of the legislative implications and aid in the development of biosecurity measures or best practice for wild *C. gigas* in Scotland.

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APPENDIX I – SURVEY SITE ADDITIONAL DATA

Site No.	Region	Location	Survey site	OS Grid Ref	Residence time (d)	Intertidal width (m)	Proximity to oyster culture (km)	General Habitat Description^
1	Mull	Loch a' Chumbainn	Inner	NM 42702 51944		50	2	*Ascophyllum*, no *F. serratus*. Boulders with occasional barnacles, mud. Occasional rocky outcrops.
2			Outer*	NM 40982 53452		200	<0.5	Native oyster found on small boulder.
3		Loch Spelve	Croggan	NM 70404 27401	11.6	60	1	Pier supports covered with mussels, barnacles and limpets. End of pier, dense *Ascophyllum*, barnacles, *F. serratus*, boulders and cobbles/pebbles.
4			Rubha Iain MhicAilein	NM 68338 28409		50	2	Small boulders in gravel / cobbles, with *F. serratus* attached. Native oysters and shells present.
5	Islay	Loch Gruinart		NR 30251 71214	1	100 - 200	2	Boulders, gravel, sand, bedrock and *F. serratus*. Old trestles, half buried oyster basket and native oyster shells.
6		Port Ellen		NR 36681 45170	<1	25 - 100	75 km (Lough Foyle, N. Ireland)	Marina pontoons – macroalgal dominated. Rocky outcrop, *Ascophyllum* with *F. serratus* and barnacles underneath. Concrete pipes, surrounded by sand and *F. vesiculosus*.
7		Bowmore		NR 31078 59956	<1	10 - 25	90 km (Lough Foyle, N. Ireland)	Harbour wall, abundant *Littorina*. Native oyster shells. Large *Anomia*. Barnacles and mussels abundance at end of harbour outer wall.
8	Seil	Ardencaple		NM 77298 17735	<1	550	<0.5	Boulders covered with barnacles and mussels, bedrock, *F. vesiculosus* and *Ascophyllum.* Native

								oysters and unattached *C. gigas* (consequence of storm damage to nearby oyster farm in 2012).
9		Balvicar		NM77195 16242		50	<0.5	Anoxic mud with Arenicola casts. *Fucus mackayi* and *F. ceranoides* in area of FW runoff. Bedrock outcrops with barnacles, *F. serratus* and *Ascophyllum.*
10	Moray	Cromarty		NH 75268 65808		250	<500	Old oyster trestles, mud, occasional boulders with clumps of mussels. Native shells.
11		Rosemarkie		NH 73874 57763		250	<500	End of outfall pipe, mussels, barnacles and *L. digitata*. Sand and boulders.
12		Fortrose	Bay	NH 73697 56190		50	<500	Outfall pipe. Abundant *L. digitata*, Rhodophyta and sponges. Mixed coarse sand and small boulders, occasional native shell fragments, *F. serratus*, L. *saccharina, Halidrys, Palmaria, Chondrus, Membranoptera, Furcellaria, Corda filum*, *Entromorpha.*
13			Harbour	NH 72441 56290		125	<500	Harbour wall, barnacles, limpets. Mud, sand and small boulders. Mussel beds, gravel and native oyster shell.
14		North Kessock		NH 65301 47954		250	<500	Cobbles, Enteromorpha, F. serratus, Palmaria, Chondrus. Occassional large boulders/ concrete block covered with barnacles. *Austrominius modestus*. Gravel bank, small pebbles covered by *L. digitata, S. latissima, F. serratus, Chondrus.* Jetty, slipway, modern pier. Native oyster shells.
15	Solway	Ross Bay		NX 65514 44631		40 – 80	45	Barnacle encrusted bedrock
16		Balcary Bay		NX 82352 49591		60 – 120	28	Mixed boulders, cobbles and pebbles
17		Brighouse Bay		NX 63152 45099		50 – 100	50	Bedrock outcrops with sand at lower shore

18		Ardwell Isle		NX 57212 49647		60 – 125	58	Rocky outcrop with extensive dense covering of large mussels, barnacles and *F. vesiculosis*, surrounded by mud with shell debris
19		Carrick		NX 57544 50674		60 - 125	60	Rocky outcrop with mussels and barnacles and boulders
20		Kirkandrews		NX 59555 47612		60 - 250	58	Barnacle and mussel bedrock and boulders adjacent to stream with cobbles and pebbles.
21		Garlieston		NX 48297 45984		200	65	Sloping harbour wall, *F. vesiculosis, F. serratus* and *Ascophyllum*. Cobbles.
22		Port William		NX 33711 43743		100	90	Harbour wall with mixed sediment below.
23		Terally Bay		NX 12677 40598		50 – 100	95	West side of rocky outcrop. Mixed old and new barnacles with new spat on scoured lower bedrock. Outcrops surrounded by rounded boulders.
24	Loch Ryan	Kirkcolm Spit		NX 04237 67890	10	80	>500	Sand spit out into the loch, survey area of boulders at midshore with *F. vesiculosus. F. serratus* some barnacles and abundant *Littorina.* Abundant native oyster shells, *Styela clava, Austrominius modestus* and attached *Sargassum muticum*.
25		Wig Bay		NX 03484 66812	10	80	>500	End of ex-military stone pier. *F. serratus* on boulders. Attached *Sargassum muticum,* Mearl, mud and gravel. Abundant native oyster shells, *Styela clava.*
26		Glen App		NX 05079 72427	10	60	>500	Round boulders with rocky outcrops near LW. Abunant barnacles, *Patella*. Abundant native oyster shells. Attached *Sargassum muticum*.
27	Cumbrae	Millport	West	NS 15857 54531		100	4	Red sandstone bedrock with barnacles to rocky outcrop with older barnacles, plus winkles, mussels and limpets.
28			Middle	NS 16327 54842		100	3.5	Stone pier *F. vesiculosus*, barnacles and small boulders with barnacles and winkles.

29			East	NS 17167 54756		20	3	Boulders at base of red sandstone bedrock. Macroalgal coverage over dense barnacle, limpets. Small mussel spat settlement mid shore bedrock.
30		Keppel Pier		NS 17634 54462		30	2	Under Keppel pier. Dense mussel spat and barnacles.
31		Watersports Centre		NS 18318 57944		60	4	Breakwaters. *F. vesiculosus* and barnacles on boulders. Shelly sand and pebbles.
32		North Shore		NS 16761 58896		50	7	Red sandstone bedrock. Top of the *F. serratus* zone. *F. vesiculosus* and green filamentous algae.
33	Loch Creran	Rubha Garbh		NM 92710 40868	14	50	<0.5	Barnacle covered boulders with gravel and cobble under *F. vesiculosus*.
34		Sealife Centre		NM 94553 41309	14	40	3	Boulders with 100% algal cover *Ascophyllum, F. serratus*, no barnacles beneath. Rocky outcrop covered with *Ascophyllum* over *Cladophora* with some large boulders with barnacles midshore.
35		Creran House Headland		NM 97184 43831	21	50	2	Rocky outcrop covered with barnacles and old mussels, dense *Ascophyllum* and *F. serratus* at lower shore. Area to east with mud. Native oysters.
36		Creagan	South	NM 97664 44304	21	50	2	*Ascophyllum, F. serratus* and *F. vesiculosus* with dense hydroids attached over cobbles with a few barnacles. Also dense sponge on *Fucus.*
37			North	NM 97560 44496	21	40	2	Rocky outcrops with *Ascophyllum* and *F. vesiculosus.*
38	Arisaig	Urchair Fhada		NM 64366 86499		50	<0.5	Rocky outcrops with *Ascophyllum* and *F. vesiculosus Cladophora*, barnacles. Sand/gravel in other areas. Extensive area of *F. mackii* in FW runoff. *Codium fragile subsp. fragile* and drift *Sargassum muticum*. Live and shells of native oysters.
39		Boatyard		NM 65677 86414		40	2	Jetty, new pier and pontoon. 100% macroalgal coverage of area - *Ascophyllum, F. vesiculosus, F. serratus.* High densities of native oysters on small

								pebbles, rock or other shells, found in gravel areas. Drift *Sargassum muticum* and attached *Codium fragile subsp. fragile.*
40	Arisaig Rhu	Torr Mor	West	NM 62833 85447		50	2	Lagoon behind natural rock wall. *Ascophyllum, F. serratus, F. vesiculosus*. Barnacles and limpets on rocky outcrops. Small boulders with dense barnacle coverage. Drift *Sargassum muticum*. *Colpomenia peregrina* and *F. mackii*. Native oysters.
41			East	NM 63848 85513		40	1	Boulders, *F. vesiculosus*, native oysters and shells. Damaged oyster trestles. Rocky outcrop, *Ascophyllum* with barnacles and mussels. *F. serratus* and *Cladophora, Halidrys, Chondrus*, fine filamentous reds
42		Port Mhuilion		NM 64671 85229		50	2	Mud, occasional boulders. Native oysters. Abundance of *Ascophyllum* and *F. serratus* over silt.
43	Loch Ailort	East of Eilean na Gualainn		NM 73471 78783	1	50	3	*Ascophyllum, F. vesiculosus, F. serratus.* Algae dominated over boulders with barnacles, red filamentous algae binding sand, *Cladophora, Entromorpha, F. mackii.* Native oysters. Bedrock with barnacles and dense algae. Drift *Sargassum muticum*.
44		Site 2		NM 73839 79271	1.1	50	2	Bedrock and boulders, some barnacles and mussels covered with *Ascophyllum, F. serratus, F. vesiculosus*. Anoxic mud at base of rocks. Small boulders, native oyster shells.
45		Layby with blocks and slipway		NM 74248 80565	126	40	1	100% algal cover – *Ascophyllum, F. serratus, F. vesiculosus, Entromorpha, S. latissima* over unstable boulders, short area of coarse gravel, boulders. Abundance of native oysters.
46		Council Pier		NM 75924 81401	126	20	<0.5	Stone pier, dense macroalgal coverage on vertical sides.

47	Loch Fyne	Newton		NS 04135 98417	223.4		14	Gravel and small cobbles over sand. F. vesiculosus, some cockle shells. Barnacles on stones and boulders, occasional larger boulders, mussels.
48		St Catherines		NN 12140 07589	223.4	100	6	Jetty, mussel shells. Large boulders mid shore. Barnacles. Gravel with *F. vesiculosus, F. serratus* on occasional small boulders. Algae with *Ectocarpacae*. Native oyster shells.
49		Cairndow		NN 18288 11193	223.4	250	<0.5	Abundance mussel shells and some large live mussels over sandy gravel. Very occasional *F. vesiculosus*.
50		Head of Loch		NN 18313 12188	223.4	40	0.5	80% algal cover over boulders, pebbles and cobbles (*Ascophyllum, F. vesiculosus, F. ceranoides, Cladophora, Entromorpha, Chondrus*). No barnacles or mussels.
51		Loch Gair		NR 92567 90631	223.4	125	6.3	*F. vesiculosus* on small boulders/cobbles over gravelly mud with shells and bivalves. Large live mussels on surface of mud over large area. *Ascophyllum*. Large mussels attached to concrete supports for plastic pipe. Stone jetty, barnacles and large mussels. Three native oysters, plus shells.
52		Crarae		NR 99524 97911	223.4	50	16.5	Cobbles and boulders with a few barnacles in sandy mud. Rusty metal grid leading down shore to large angular boulder covered in barnacles. *Pomatoceros, Ascophyllum* and *F. vesiculosus* over barnacles on boulders. Lower shore *F. serratus* with spirorbids, *Psammechinus miliaris* abundant.
53		Inveraray		NN 09807 08772	223.4	250	9.6	Wooden Pier, *F. vesiculosus* over boulders with some old barnacles and new settlement. Sand with occasional *Arenicola* casts and gravel beneath the boulders, some FW run off with *Entromorpha*. Boulder covered with barnacles. Native oyster shell.

54	Firth of Forth	Port Seton		NT 40833 76127	<1	250	>500	Bedrock, covered in barnacles, some *F. vesiculosus*. Abundance of native oyster and razor shells. Mussels on lower shore with shell debris.
55		Joppa		NT 32158 73428	<1	125	>500	Bedrock with barnacles and sand at base. Bands of rock perpendicular to the shoreline, mussels and mature barnacles on boulders. *Entromorpha* lower shore over silt. Dense mussel spat. Native oyster shell.
56		Craigentinny		NT 29165 75639	<1	150	>500	Boulders with sand at base. Mussel spat settlement forming ridges with trapped sediment. *Entromorpha* and *Porphyra* on top of mussel spat. Native oyster shells.
57		Donibristle Bay		NT 15850 82623	<1	250	>500	Boulders, few barnacles, *F. vesiculosus* with silt and shell debris. Native oyster shells. Rocky outcrop, boulders with *F. serratus* and *F. vesiculosus*. Barnacle covered bedrock.
58		Dalgety Bay		NT 16533 83153	<1	125	>500	Small boulders with barnacles and *Chondrus*, silt/mud, boulders with barnacles, abundant mussel shells. Outcrop of bedrock - older mussels attached. Boulders with *F. vesiculosus*.
59	Tay	Greenside Scalp		NO 42723 28949		20	>500	Boulders with macroalgae and occasional barnacles. *Ascophyllum, F. vesiculosus*. Soft mud at LW. Rocky outcrop, *Ascophyllum* and *Cladophora.* Small boulders with barnacles and *Ascophyllum*.
60		Tayport		NO 46246 28850		750	>500	Harbour, jetty. Lower walls covered with *Ascophyllum Fucus* and *Enteromorpha. Ascophyllum* on harbour wall. Extensive sand area with *Arenicola* casts dotted with a few small boulders with *F. vesiculosus* and *Ascophyllum* over barnacles. Pebbles/ cobbles with *Enteromorpha* and *Porphyra*. Zostera. Wooden posts with barnacles and *F. spiralis*. Posts set in hard substrata (pebbles)

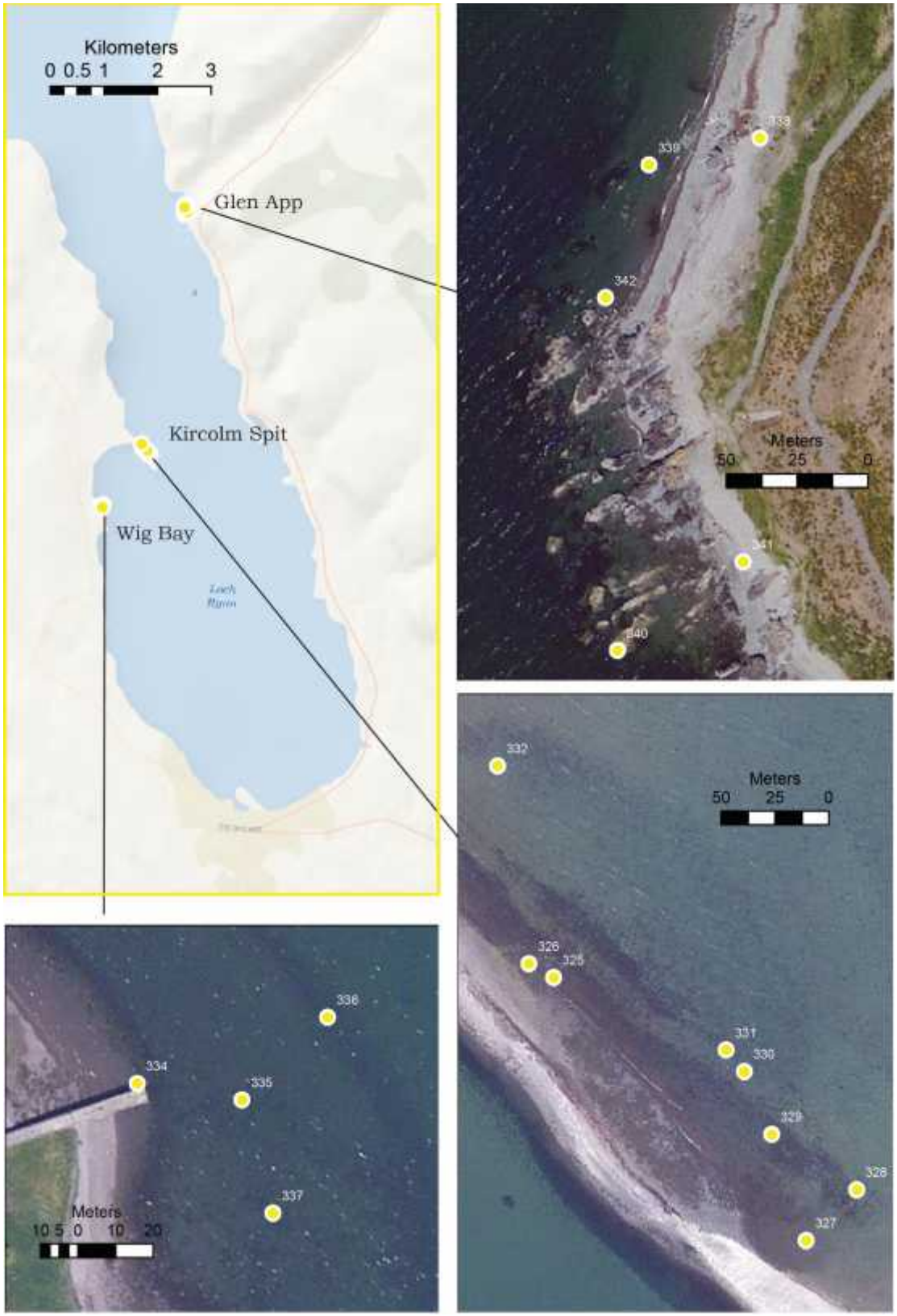
								covered in barnacles with some small mussels and *F. vesiculosus*.

*Preliminary observations – test run of survey technique.

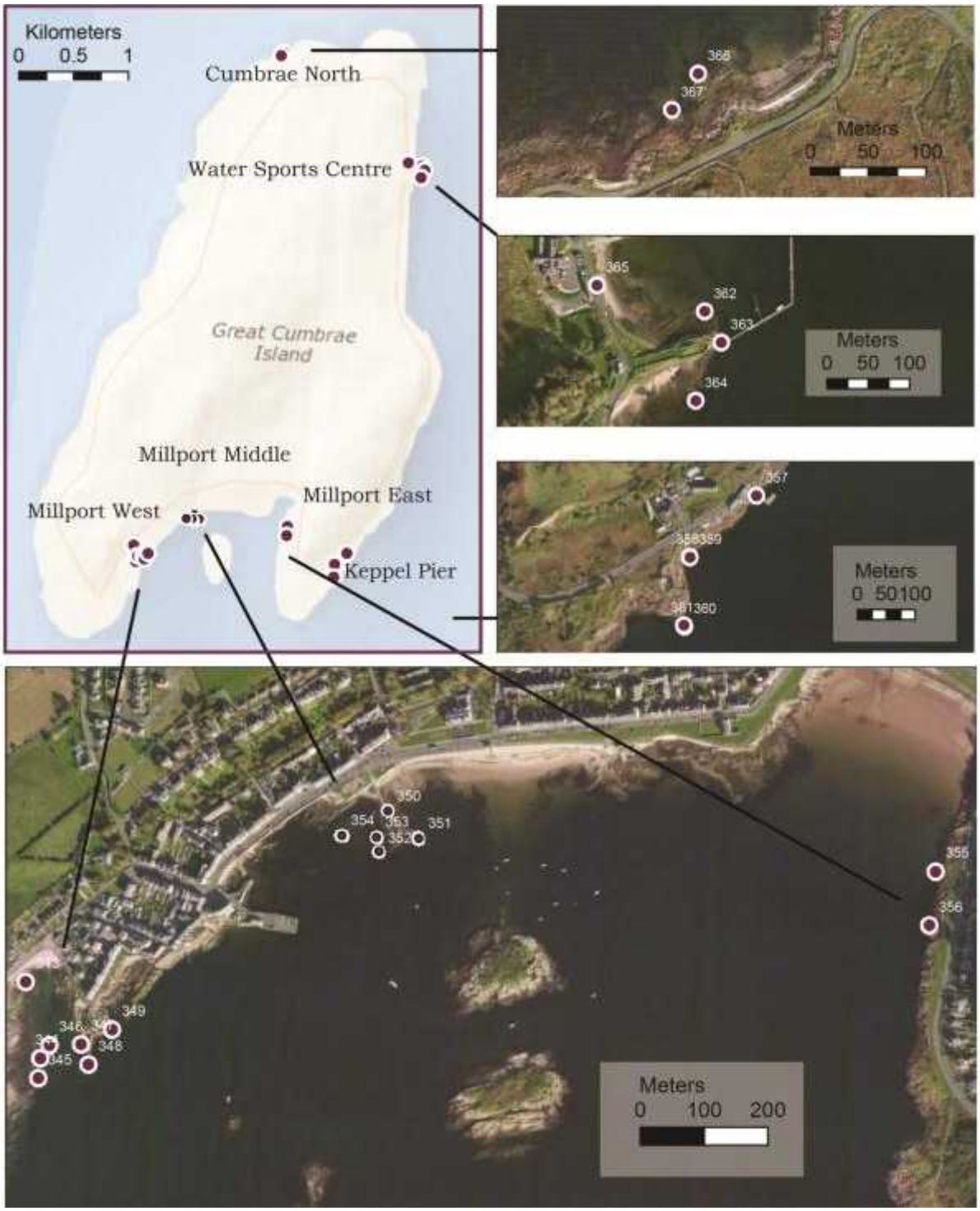
^See supplementary information, including GPS waypoints of each transect and photographs of each site for further detailed information.

APPENDIX II – SURVEY SITE LOCATIONS

4.1 Loch Ryan



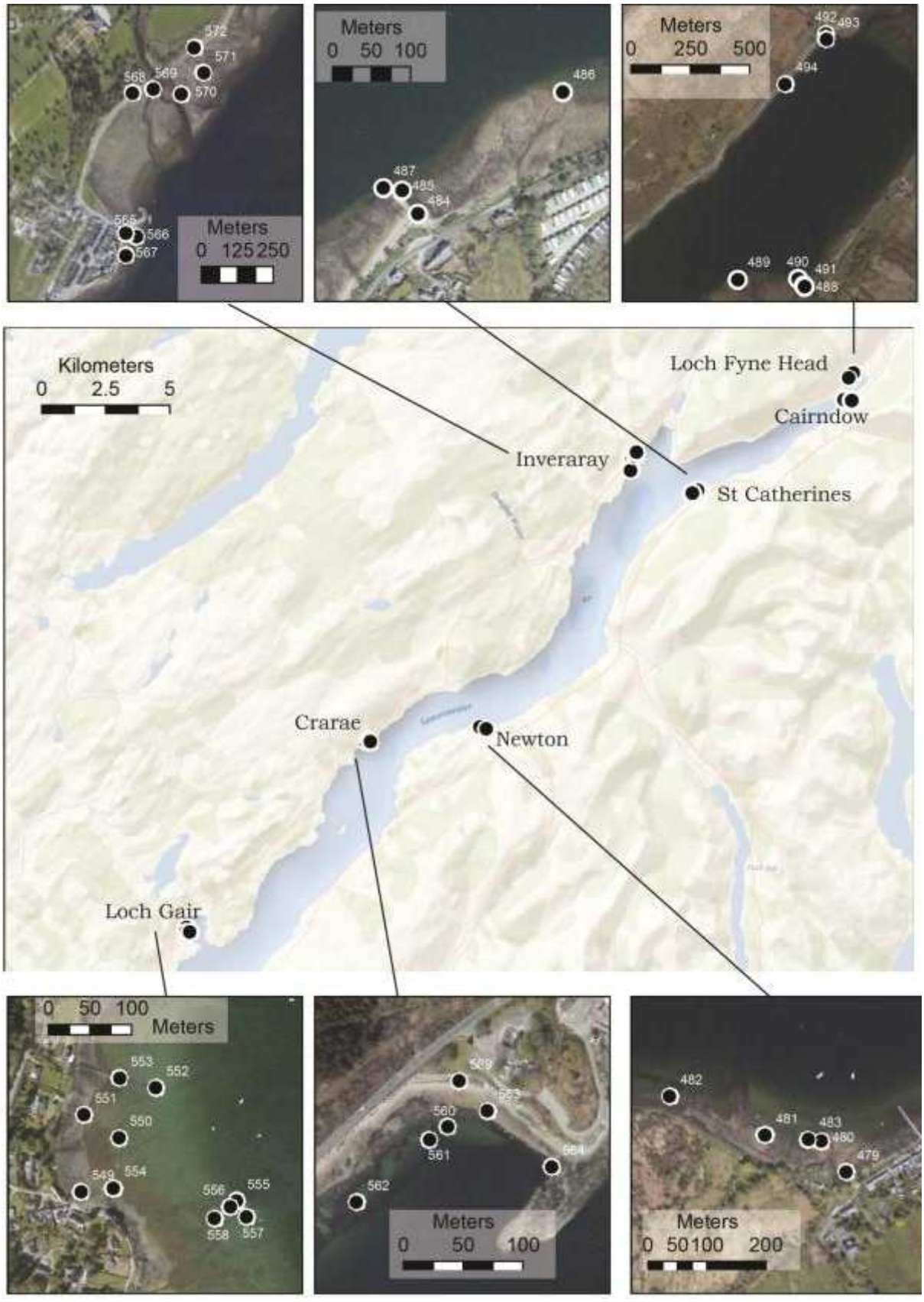
4.2 Isle of Cumbrae



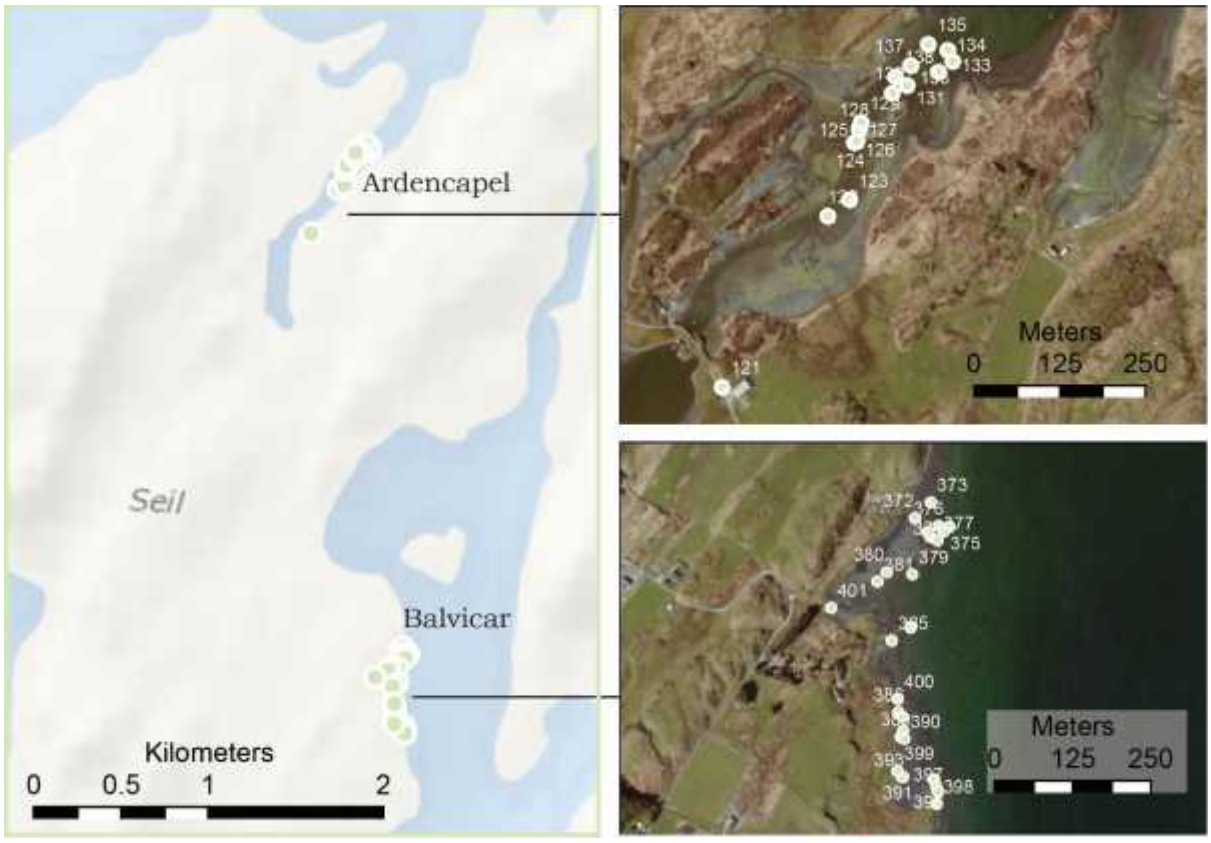
4.3 Islay


Loch Gruinart
Bowmore
Port Ellen
Kilometers
10 5 0
Meters
0 25 50 100 150
Meters
0 25 50
Meters
0 50 100

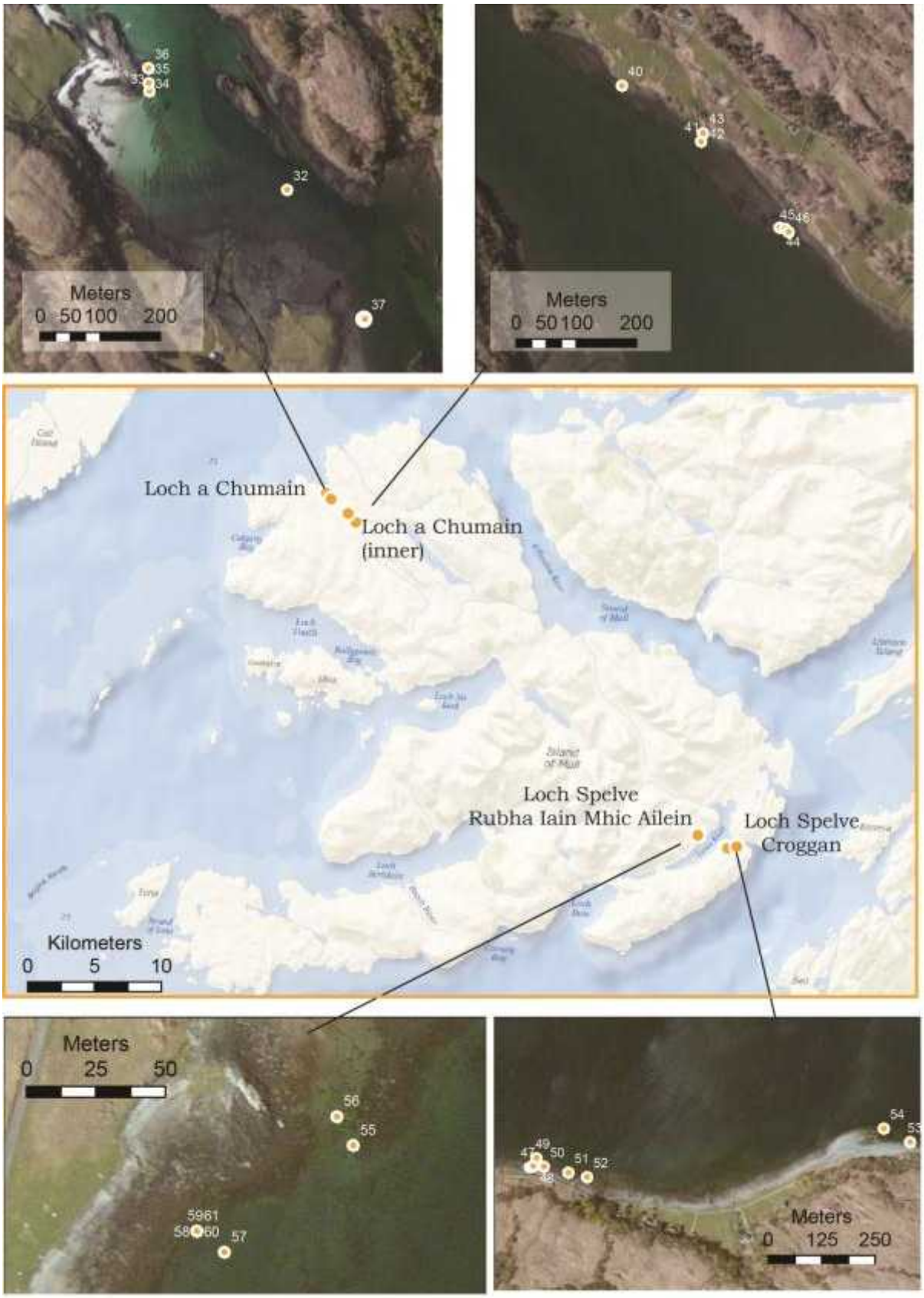
4.4 Loch Fyne


572
571
568
569
570
565
566
567
Meters
0 125 250
Meters
0 50 100
486
487
489
484
Meters
0 250 500
492
493
494
489
490
491
488
Kilometers
0 2.5 5
Loch Fyne Head
Cairndow
Inveraray
St Catherines
Crarae
Newton
Loch Gair
0 50 100
Meters
553
552
551
550
549
554
556
555
558
557
560
561
562
Meters
0 50 100
482
481
483
480
479
Meters
0 50100 200

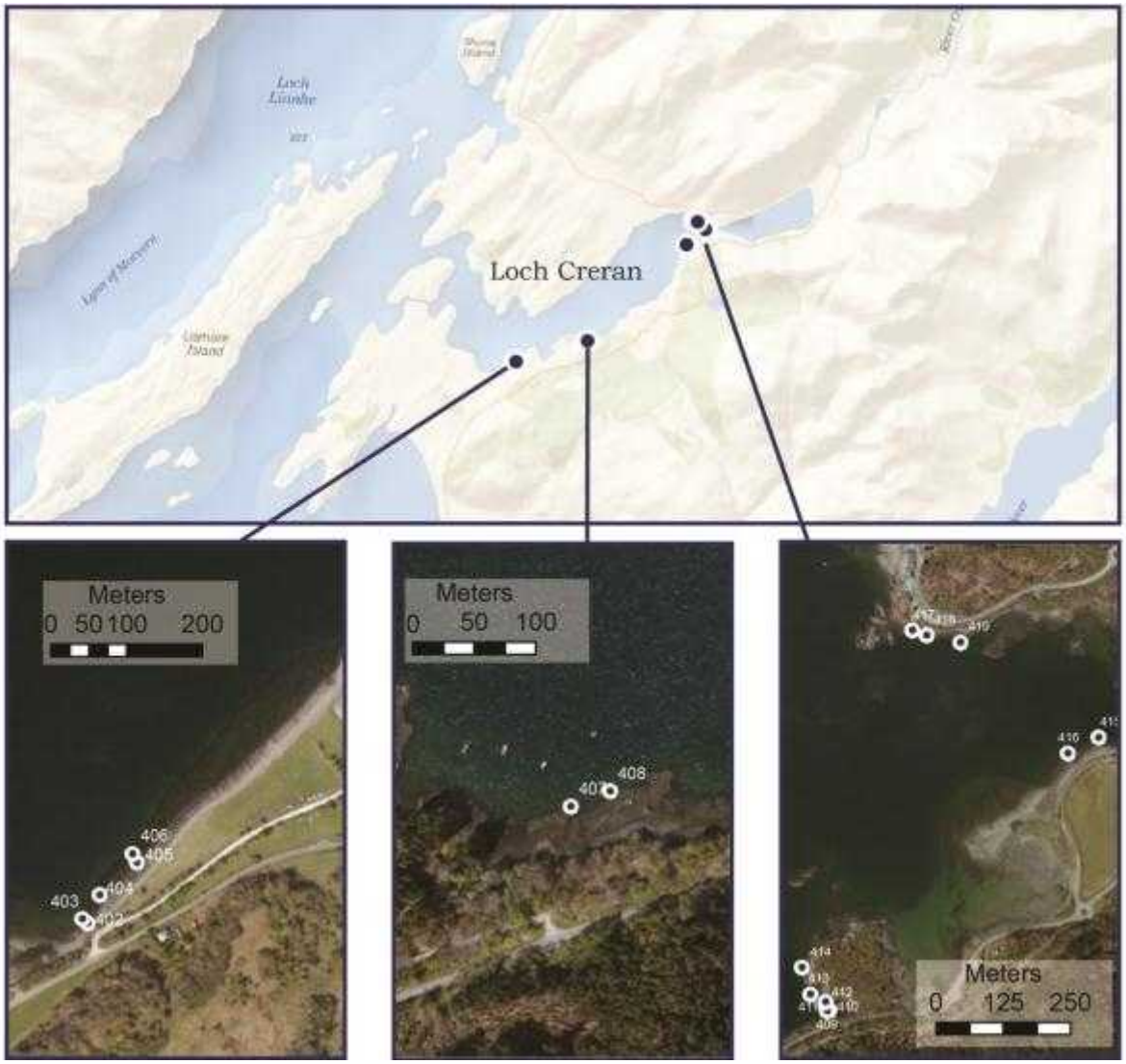
4.5 Seil Island



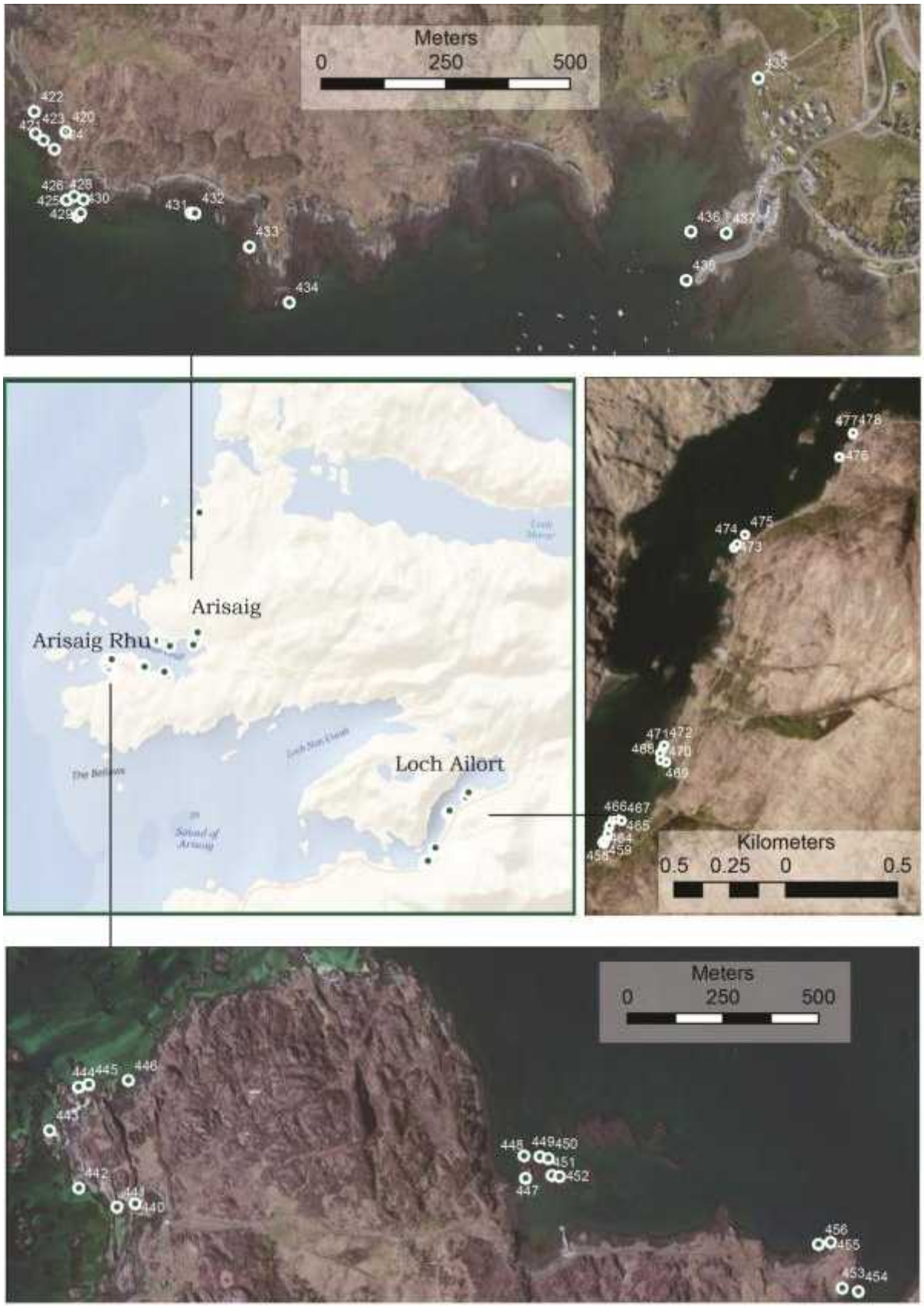
4.6 Isle of Mull



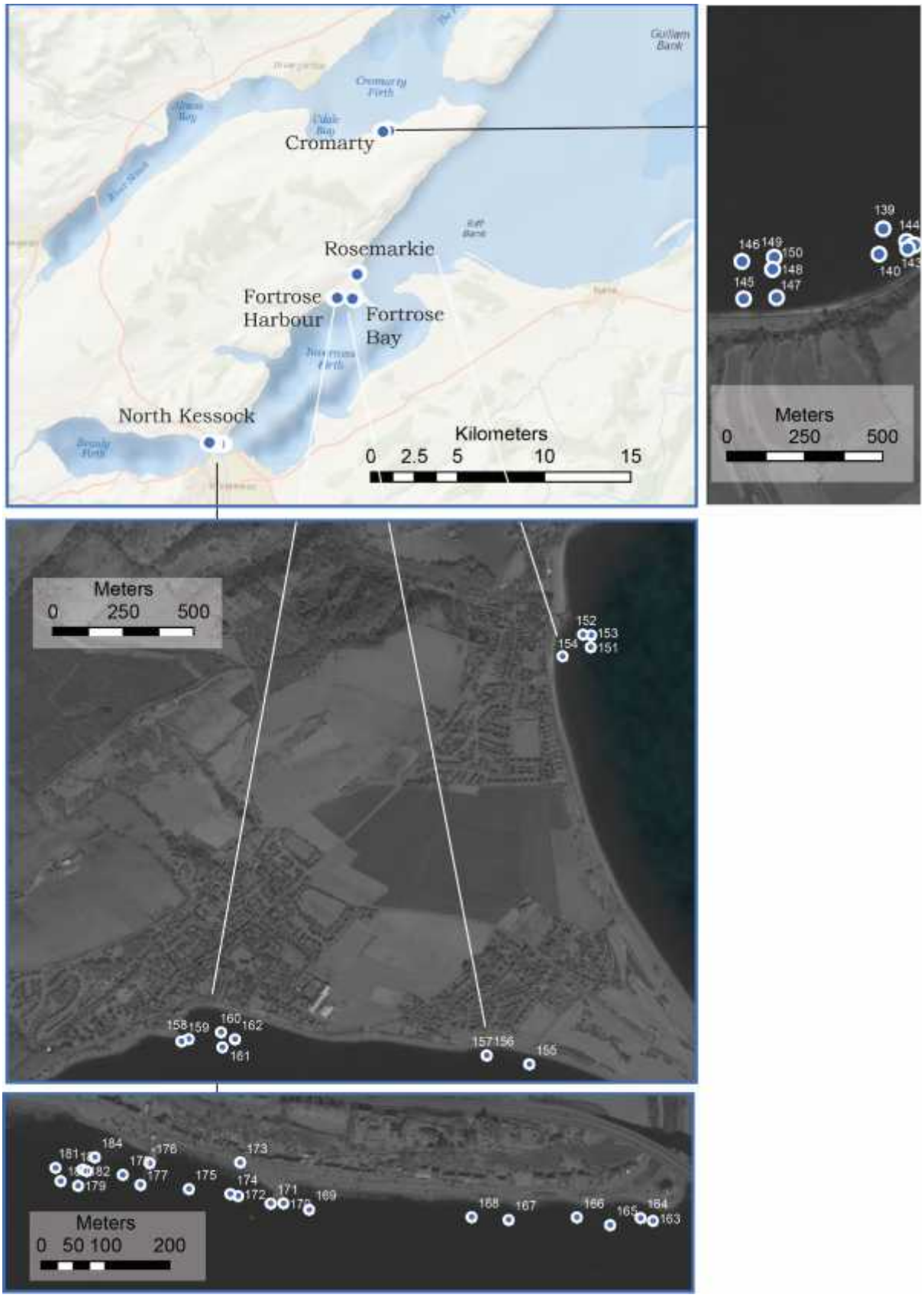
4.7 Loch Creran



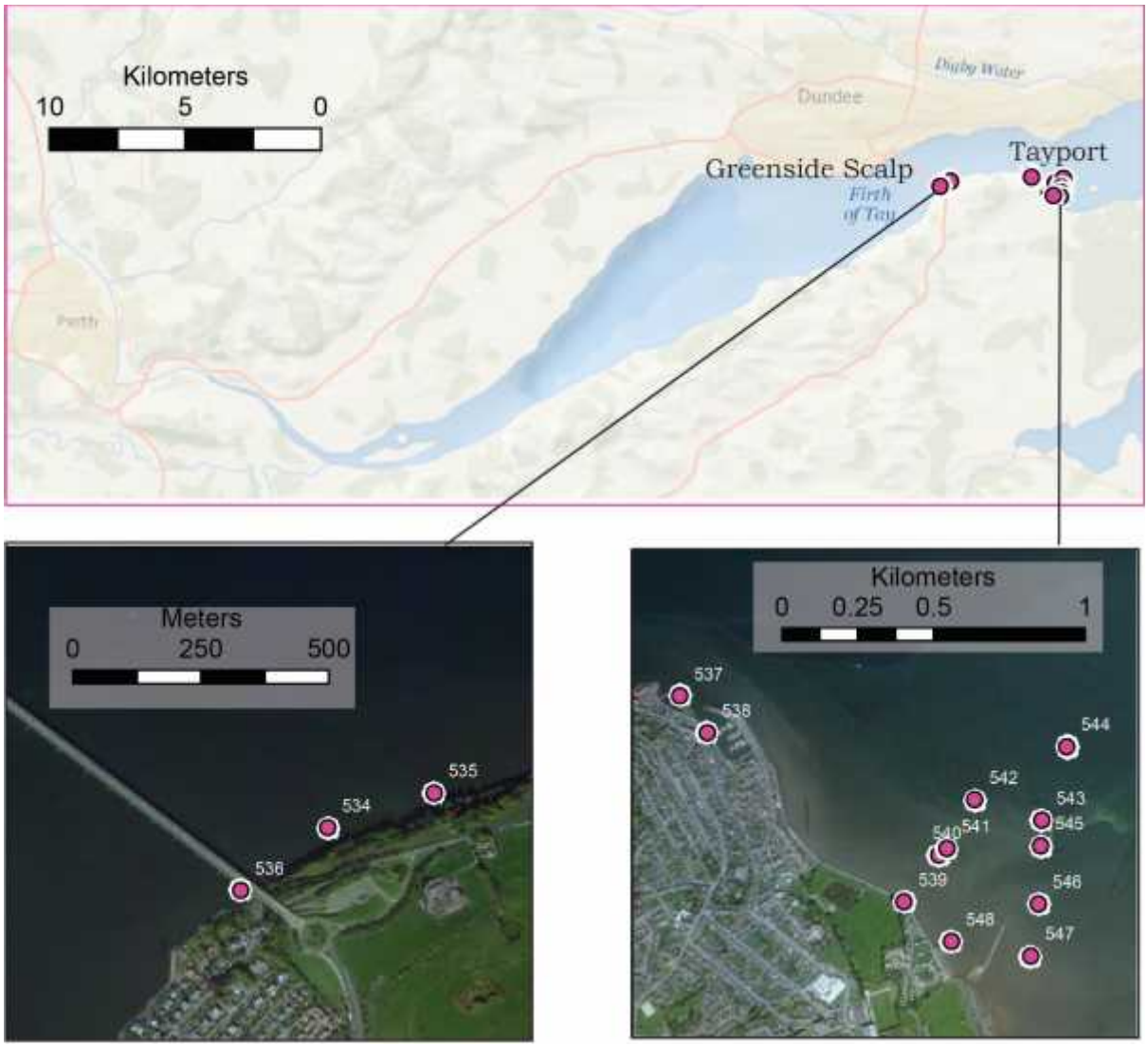
4.8 Arisaig and Loch Ailort



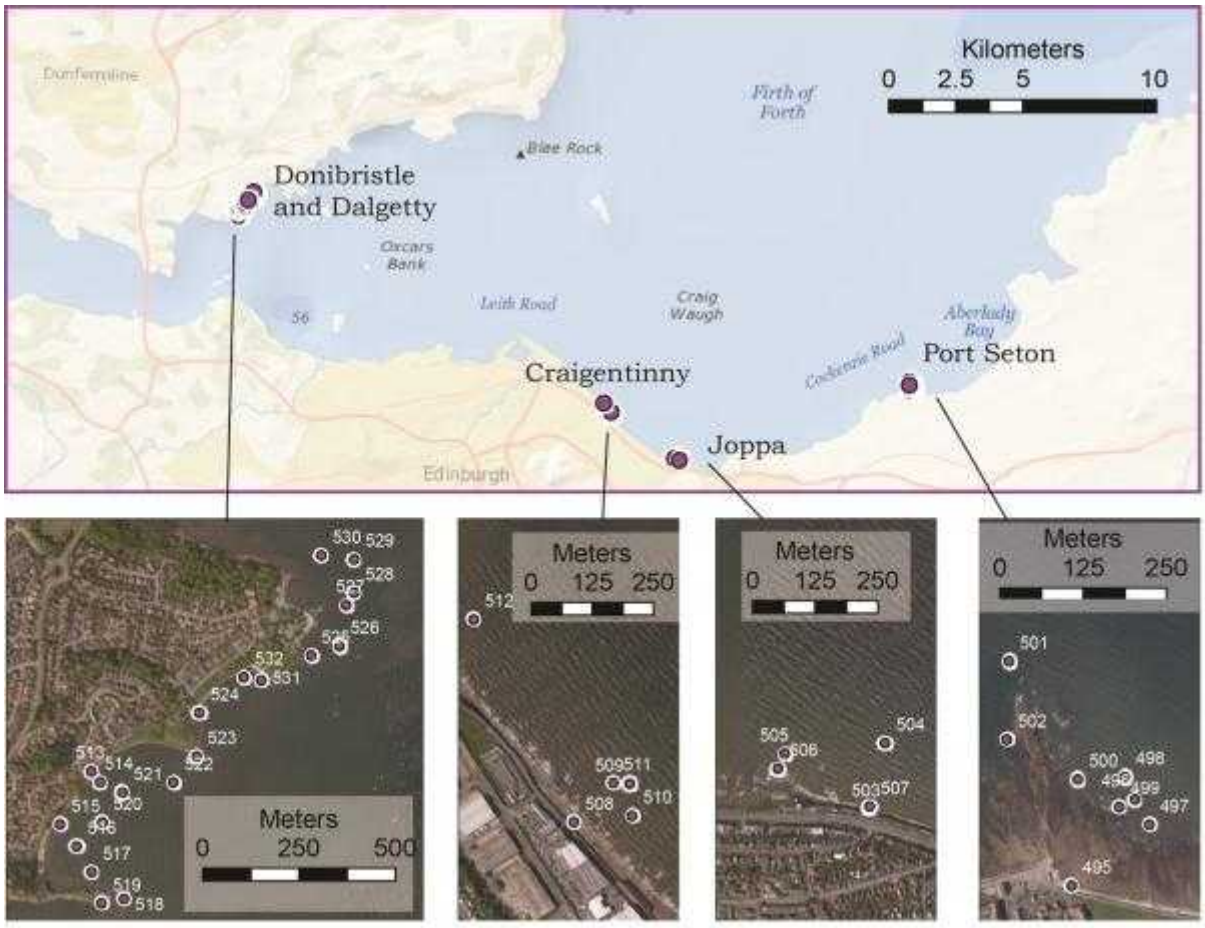
4.9 Moray and Cromarty



4.10 Tay Estuary



4.11 – Firth of Forth



APPENDIX III – ADDITIONAL SIGHTINGS OF LIVE WILD *CRASSOSTREA GIGAS* IN SCOTLAND

Region	Site Name	Date	No. oysters per site	Grid Ref	Oyster Length (mm)	Habitat Type	Reference	Comments
Loch Fyne	Loch Gair	13/04/12	1	NR 92465 90614	227.4	Small cobbles	Janet Khan-Marnie, SEPA (pers. comm.).	Visual record only, not confirmed with genetic analysis.
Solway Firth	Ravenshall Point	28/09/2012	6	NX524522	74 102 91 85 79		Solway Firth Partnership (pers. comm.)	Visual record, only.
	Ross Bay	06/01/2013	11	NX655448			Solway Firth Partnership (pers. comm.)	Awaiting genetic verification.
	Ravenshall point nr Dirk Hatteraicks cave	23/01/2013	13	NX518526			Solway Firth Partnership (pers. comm.)	Visual record only.
	Balcary Bay	24/01/2013	4	NX825497			Solway Firth Partnership (pers. comm.)	Awaiting genetic verification.

	Torr Point nr Auchencairn	04/12/2013	1	NX823517			Solway Firth Partnership (pers. comm.)	Visual record only.
	Ravenshall	03/04/2013	35	NX523522	50-75 (n=33) <25 (n=2)		Solway Firth Partnership (pers. comm.)	Visual record only.
	Carrick	27/01/2014	1	NX571497			Solway Firth Partnership (pers. comm.)	Visual record only.
	Carrick	27/02/2014	1	NX571497			Solway Firth Partnership (pers. comm.)	Visual record only.
	Kirkandrews	09/03/2014	2	NX595477	44 62		Solway Firth Partnership (pers. comm.)	Awaiting genetic verification.
	Carrick	27/03/2014	2	NX571497	216	Mussel beds	Solway Firth Partnership (pers. comm.)	Visual record only.
	Ravenshall	19/03/2014	5	NX523522			Solway Firth Partnership (pers. comm.)	Visual record only.
Firth of Forth	Whitehouse Bay	12/04/2013	5	NT 14495 78770	91 64	Top of boulder Top of boulder	(Smith *et al.*, 2014)	Verified.

					84 82 101	Small stone Side of boulder Upper surface of dolerite sill		
	Whitehouse Bay	27/05/2013	3	NT 14495 78770	109 88 74	Unattached Small rock Mudstone slab outcrop	(Smith *et al.*, 2014)	Verified.
North Berwick	Aberlady Bay (South)	08/10/2014	1	NT448803		Unattached, but showing signs that had previously been attached	D. Donnan, SNH (pers. comm.)	Visual record only.

Scottish Aquaculture Research Forum

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SARF - Member Organisations

Industry

SEAFISH
Association of Scottish Shellfish Growers
Scottish Salmon SSPO
British Trout Association

Government and Regulators

Scottish Natural Heritage
SEPA Scottish Environment Protection Agency
marinescotland
The Scottish Government
HIE Highlands and Islands Enterprise
Iomairt na Gaidhealtachd 's nan Eilean

Non-Governmental Organisations

The Crown Estate
Scottish Environment LINK
Rivers & Fisheries Trusts of Scotland
Safeguarding Scotland's Rivers & Lochs
ASFB The Association of Salmon Fishery Boards