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Original Article

Does population genetic structure support present management regulations of the northern shrimp (*Pandalus borealis*) in Skagerrak and the North Sea?

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Population structuring in the northern shrimp (*Pandalus borealis*) in the North Sea area (including Fladen and Skagerrak) was studied by microsatellite DNA analyses. Screening 20 sample locations in the open ocean and Skagerrak fjords for nine loci revealed low, but significant genetic heterogeneity. The spatial genetic structure among oceanic samples of Skagerrak and the eastern North Sea was weak and non-significant, consistent with the current management regime of one single stock. However, Skagerrak fjord samples generally displayed elevated levels of genetic differentiation, and significantly so in several pairwise comparisons with other fjords and oceanic samples. Although the Skagerrak fjord populations are of less economic value, some of them are regulated separately (e.g. the Gullmarsfjord) and local stocks may prove important to uphold genetic variability and biocomplexity in a changing environment.

Keywords: fisheries management, fjord populations, genetic structure, microsatellites, *Pandalus borealis*, shrimp.

Introduction

Uncertainty about the geographic extent of populations impedes proper monitoring, management, and conservation of marine resources. The spatial scale of population structuring depends on several factors, including dispersal patterns of early life history stages (pelagic eggs and larvae) and the active migratory behaviour of juveniles and adults [reviewed by Ciannelli *et al.* (2013)]. Management strategies are often criticized for not considering population units at an appropriate biological scale (Hutchinson, 2008; Reiss *et al.*, 2009); however, this is often due to the lack of available knowledge.

A variety of methods are available to characterize the spatial distribution and population structure of marine organisms, but not all are suitable for every species and circumstances (Cadrin *et al.*, 2013). In particular, physical tagging requires large enough individuals to support tags, is difficult to recapture in species with very large population sizes, and has a limited usefulness in species that moult their shell: all of which applies to shrimps. Genetic methods, on the other hand, are available for all organisms and might convey information on population structure beyond what is possible with more traditional approaches [e.g. reviewed by Waples *et al.* (2008) and Funk *et al.* (2012)]. Genetic markers have

the advantage of being intimately linked to reproduction, allowing for detection of dispersal and intermixing at all life history stages, including eggs and larvae, which typically are inaccessible to commonly applied ecological methods.

The trawl fishery for northern shrimp (*Pandalus borealis*; hereafter synonymous with shrimp) is one of the most important fisheries in Skagerrak (Figure 1). The economic impact of this traditional fishery goes far beyond those actively participating in the fishery, as shrimp in this region is typically processed and sold locally. The shrimp fishing fleets in the three Scandinavian countries are quite different, with larger fleets of heterogeneous vessel sizes in Sweden and Norway, and a few large vessels in Denmark. Shrimp trawlers in Sweden and Norway operate both inside fjords and sheltered waters as well as in offshore areas. Catches from the fjords are small compared with those taken in the open sea. In the Skagerrak and Norwegian Deep region, shrimp is found at depths between 100 and 500 m (Figure 1). Formerly, shrimp was also fished in two smaller areas in the western North Sea (Figure 1), the Farn Deeps and the Fladen Ground, but these stocks seem to have collapsed (ICES, 2004, 2013). Along the Danish coast and in the southern and central North Sea, shrimp is absent due to too shallow water.

The International Council for the Exploration of the Sea (ICES) currently considers shrimp from the Farn Deeps, Fladen Ground, and Skagerrak/Norwegian Deep to belong to three separate stocks (Figure 1), based on (1) geographic separation and oceanic current patterns and (2) differences in length frequency distributions (LFDs), where the shrimp stock on the Fladen Ground is observed to have fewer age groups than the Skagerrak/Norwegian Deep stock (ICES, 2003). The rationale behind point (2) is that a different number of age groups in two areas could imply a lack of exchange of adults. Furthermore, comparison of Landings Per Unit Effort (LPUE) time-series from the Danish shrimp fishery indicates that the Fladen stock differs in abundance fluctuations and therefore may be demographically independent (i.e. display a different recruitment pattern) from the stock in the Norwegian Deep and Skagerrak region (cf. Figure 2). Differences in mortality and recruitment exist not only between, but also within, the ICES defined shrimp stocks: LFDs show that shrimp in Skagerrak only reach a maximum age of 3–4 years, whereas shrimp in the Norwegian Deep may live for 4–5 years, and mean size-at-age identified from modal analysis is consistently higher in Skagerrak than in the Norwegian Deep, indicating faster growth in the former area (Tveite, 2000, Søvik and Thangstad, 2013). In Skagerrak, the number of juvenile shrimp shows large annual fluctuations, compared with lower and more constant numbers in the Norwegian Deep (Figure 3).

Since 2003, ICES has recommended genetic studies to clarify the shrimp stock structure in the North Sea and Skagerrak region (ICES, 2003). However, until now, no genetic study has been directed towards disentangling whether shrimp in offshore areas in the North Sea and Skagerrak belong to the same biological population or not, or if shrimp in the fjords represent separate biological populations and how they relate to the shrimp in offshore areas. Meanwhile, the overall stock biomass has declined substantially from an average stable level since the mid-1990s to its currently historical low (cf. Figure 2b). Hence, information on the population structure is urgently for improving management of this important resource.

Here, we combine information on 30 years of survey and commercial catch data in Skagerrak and the North Sea with spatial and temporal genetic information from nearly 1800 individuals of shrimp collected from Skagerrak fjords and offshore areas, to resolve the population structure of shrimp in this region.

Material and methods

The study species

Northern shrimp (*P. borealis*) is a cold water species living on soft mud or sand/silt on the continental shelves in the North Atlantic, usually at depths between 50 and 500 m (Shumway *et al.*, 1985). In the Northeastern Atlantic it is found in Skagerrak and the North Sea, along the Norwegian coast, in the Barents Sea, and around Jan Mayen and Iceland (Bergström, 2000; Hvingel, 2006). The species is probably adapted to local sea temperatures, ranging from 6–9°C in the southernmost part of its distribution range (Schlüter and Jerosch, 2009) to 0–4°C in the Barents Sea (Furevik, 2001; Boitsov *et al.*, 2012).

Shrimp is a protandric hermaphroditic species, hatching as male and then developing into female after spawning as male once or twice (Bergström, 2000). Age at sex change is negatively correlated with temperature. In Skagerrak and the North Sea, spawning and mating takes place in October/November and females carry the fertilized eggs under the abdomen until hatching takes place in March (Bøhle, 1977), the following year. The species has five pelagic larval stages (Ouellet and Allard, 2006), which drift with ocean currents for 45–90 days depending on the ambient sea temperature before settling on the bottom (Shumway *et al.*, 1985). Shrimp in Skagerrak and the Norwegian Deep grow relatively rapidly and there is little size overlap between year-classes (Shumway *et al.* 1985). Thus, it is possible to discriminate up to 4 year-classes based on frequency distributions of carapace length (CL).

Stratification and sampling

Based on existing knowledge of geographic variation in shrimp abundance, growth, and reproduction, three potentially different open sea populations were identified for sampling (Skagerrak, the Norwegian Deep, and Fladen Ground), together with seven fjord sites. The original plan was to include also the Farn Deeps in the study, but we were not able to obtain any shrimp from this area. Neither in 2010 nor in 2011 were any shrimp caught in this area by the Danish International Bottom Trawl Survey (IBTS). Shrimp samples were collected by trawl in 2007 and 2010–2012, by research vessels, in offshore Skagerrak, Fladen Ground, and the Norwegian Deep, and by small commercial vessels in the fjord locations (Figure 1 and Table 1). At each location, we sampled 3–5 kg of shrimp, from which 96 females were subsampled. Only female shrimp, which comprise >1 year-class, were chosen to ensure that individuals from multiple spawning events were represented in each sample, thus minimizing possible temporal effects of a single cohort. In addition, temporal replicate samples were obtained from different years at Fladen Ground (2007, 2011), Skagerrak (A and C; 2011, 2012), and from four fjord locations: Stolsfjord, Håøya, Oslofjord, and Gullmarsfjord (2010, 2012), and used to check on temporal stability of observed genetic structure.

Survey data

A trawl survey which covered the whole distributional area of shrimp in Skagerrak and the Norwegian Deep north to 60°N (Figure 1) is conducted annually by the Norwegian Institute of Marine Research (IMR). The survey data consisted of four different time-series (Søvik and Thangstad, 2013): (i) October/November 1984–2002 with RV *Michael Sars*; (ii) a point estimate in October/November 2003 with RV *Håkon Mosby (HM)*; (iii) May/June 2004–2005 with RV *HM*; and (iv) January/February 2006–2014 with RV *HM*. A survey bottom trawl (Campelen) with

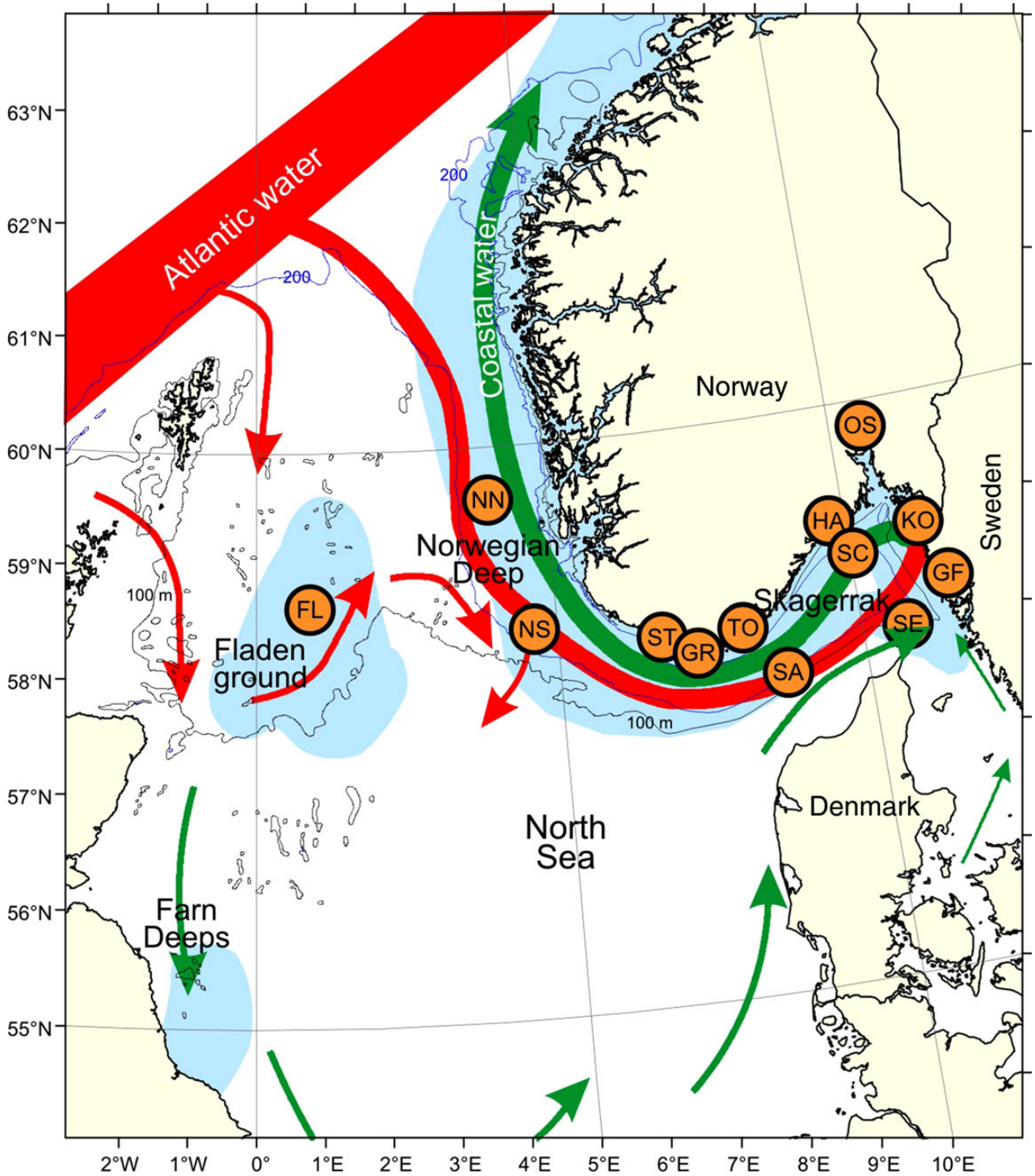

Figure 1. Distribution of northern shrimp (*P. borealis*) in Skagerrak and the North Sea (blue), prevailing ocean currents (arrows), and sampling locations (circles). See Table 1 for sample abbreviations.

codend mesh size of 20 mm was used. For comparing population trends, two separate survey biomass indices were calculated here: (i) Skagerrak and (ii) the Norwegian Deep (Figure 2b). Samples of 300 shrimp were taken from each trawl haul, sorted by maturity stage characteristics, and measured to the nearest mm below (CL). LFDs are partitioned into age groups by modal analysis (Bhattacharya, 1967; software: FISAT). As eggs hatch in March (Bøhle, 1977), the first modal group in the LFD (time-series from 2006 until present) consisted of shrimp almost 1 year old. The number of these is defined as a recruitment index.

Commercial fishery data

Danish commercial landings and effort data from 1987 to 2013 provide the basis for four LPUE time-series: (i) Fladen Ground,

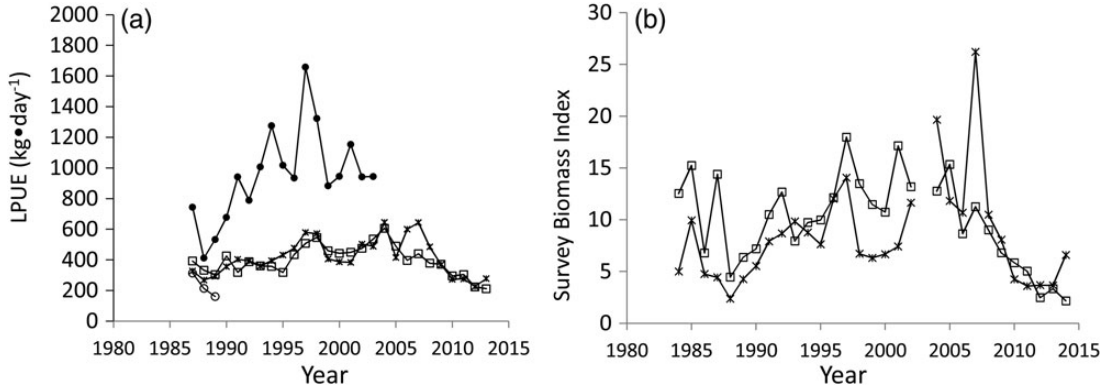

Figure 2. (a) Danish LPUE time-series from 1986 to 2013: Fladen Ground (dots), 1987 – 2003; the Norwegian Deep (squares), 1987 – 2013; Skagerrak (crosses), 1987 – 2013; and Farn Deeps (circles), 1987 – 1989; (b) Norwegian survey biomass indices for the Norwegian Deep (squares), and Skagerrak (crosses).

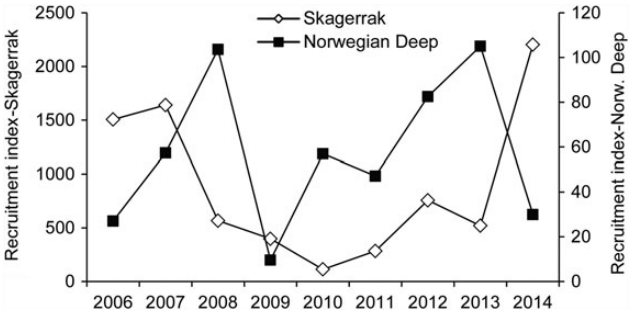

Figure 3. Recruitment index (abundance in million) of 1-year-old shrimp (open diamonds) in Skagerrak and the Norwegian Deep (solid squares), 2006 – 2014.

(ii) Skagerrak, (iii) Norwegian Deep, and (iv) Farn Deeps. A generalized linear model procedure is used to correct official logbook data for fleet renewal, trawl-size changes, ICES rectangle differences, and seasonal variation (Eigaard and Munch-Petersen, 2011). The standardized time-series are plotted together in Figure 2 to allow inferences of differences and similarities in shrimp population dynamics in the whole area and for comparison with the genetic results.

Genetic analysis

After length measurements (CL) and maturity stage classification, a small piece of the tail from each of the 96 females subsampled at each location was taken for genetic analysis and stored at 4°C in pure ethanol. Genomic DNA was extracted using the E.Z.N.A. Tissue DNA Isolation Kit (Omega Bio-Tek) and 11 microsatellite loci (Pereyra *et al.*, 2012) were amplified by PCR. The PCRs were performed on MyCycler (Bio-Rad) in 10 μl volume; using Qiagen Taq DNA Polymerase (0.8 U/sample), 10× buffer, 0.3 mM of each dNTP and primers (Life Technologies), diluted in molecular grade water. The loci PbC105, PbC8, Sd2 – 14, PbA110, PbC109, PbA1, SD1-41, SD2-68, and SD3-62 had primer concentrations of 0.35, 0.5, 0.1, 0.4, 0.4, 0.2, 0.35, 0.5, and 0.04 μM, respectively. Cycling conditions included an initial 5-min phase of denaturation at 95°C, followed by 35 cycles of 95, 56, and 72°C for 30, 90, and 60 s, respectively, ending with a single elongation step at 60°C for 30 min [modified from Pereyra *et al.* (2012)]. Microsatellite fragment sizes were determined after separation on ABI 3130xl automated sequencers (Life Technologies) with the genotyping software Genemapper 4.0. To prevent potential genotyping errors, all capillary traces were scored independently by two trained persons, and disagreements were re-analysed with new PCR products to minimize misclassifications of alleles and genotypes. Genotypes that could not reliably be solved by this procedure were left as missing. Individuals with six or more missing genotypes were considered of poor DNA quality and excluded from further consideration, leaving a total of 1797 individuals for analysis (see Supplementary Appendix for detailed information on the number of scored individuals at each locus and sample locality).

Table 1. Sample data denoting locality (abbreviations in parenthesis), year of sampling, sample size, coordinates and expected heterozygosity (H_E).

Sample	Year	Sample size	Coordinates	H_E
Fjord samples				
Stolsfjord (ST)	2010	96	N58.22 E6.69	0.695
Stolsfjord (ST)	2012	96	N58.22 E6.69	0.688
Grønnsfjord (GR)	2010	96	N58.04 E7.04	0.708
Topdalsfjord (TO)	2010	94	N58.82 E8.18	0.672
Håøya (HA)	2010	96	N59.02 E9.80	0.692
Hå(HA)	2012	96	N59.022 E9.80	0.693
Oslofjord (OS)	2010	96	N59.81 E10.59	0.685
Oslofjord (OS)	2012	94	N59.81 E10.59	0.694
Kosterfjord (KO)	2010	96	N58.88 E11.10	0.707
Gullmarsfjord (GF)	2010	80	N58.27 E11.48	0.677
Gullmarsfjord (GF)	2012	96	N58.27 E11.48	0.712
Oceanic samples				
Skagerrak A (SA)	2011	96	N57.80 E8.82	0.705
Skagerrak C (SC)	2011	95	N58.87 E9.80	0.698
Skagerrak E (SE)	2011	95	N57.99 E10.82	0.702
Skagerrak A (SA)	2012	94	N57.80 E8.82	0.704
Skagerrak C (SC)	2012	96	N58.87 E9.91	0.706
Fladen Ground (FL)	2007	96	N58.59 E10.68	0.698
Fladen Ground (FL)	2011	94	N58.38 E10.49	0.698
Norwegian Deep S (NS)	2011	95	N58.33 E4.86	0.696
Norwegian Deep N (NN)	2011	96	N59.48 E4.18	0.710

Temporal samples were taken at or very close to the same location (see below).

Statistical analysis

The amount of genetic variability was characterized by heterozygosity (observed heterozygosity, H_O, within samples and expected heterozygosity, H_E, in the total material for each locus; Nei and Chesser, 1983) and by observed number of alleles, with the FSTAT software ver. 2.9.3.2 (Goudet, 2001). Deviations from Hardy–Weinberg genotype proportions were estimated by F_{IS} (Weir and Cockerham, 1984) and assessed using the exact probability test in the GENEPOP software ver. 4.0 (Rousset, 2008). We adopted the false discovery rate (FDR) approach (Benjamini and Hochberg, 1995) when interpreting significances (at the 5% level) in tables with multiple tests. We tested for null alleles using the software MICROCHECKER (Van Oosterhout *et al.*, 2004).

Genetic differences among localities were quantified by F_{ST}, using Weir and Cockerham's (1984) estimator θ overall samples and between pairs of sample localities. Allele frequency differences over all samples and among pairs of localities were tested with exact G-tests in the GENEPOP software, using 10 000 dememorizations and batches and 10 000 iterations per batch. The reported p-values for each locus were summarized over loci, following Fisher's procedure (i.e. sum of twice the negative logarithm of p-values), to test for genetic divergence jointly overall loci. Tables of multiple pairwise tests were assessed for significance with the FDR approach, at the 5% level, following Benjamini and Yekutieli (2001). Temporal stability of spatial structure was tested for with analysis of molecular variance (AMOVA) analyses of spatial samples and temporal replicates, using the Arlequin software ver. 3.5 (Excoffier and Lischer, 2010).

The statistical power of our sampling design for exact tests given different levels of F_{ST} was estimated using the POWSIM software ver. 4.1 (Ryman and Palm, 2006), exploring a range of different values for F_{ST} by varying the number of generations of drift (t) while keeping effective population size (N_e) constant at 1000. The power to detect a predefined level of genetic differentiation from given numbers of individuals and loci is affected by the polymorphism in the genetic data (Ryman *et al.*, 2006). A sampling scheme corresponding to the empirical data for each marker and species was used, and the analyses were conducted using 1000 dememorizations, 100 batches, and 1000 iterations per batch (POWSIM settings). The percentage of significant outcomes (at alpha = 0.05) for a range of predefined F_{ST}-values was interpreted as the power of the tests for detecting the defined level of genetic divergence. To set the present results for shrimp into a wider perspective, we likewise calculated power for currently available data from other species in Skagerrak: the European lobster (*Homarus gammarus*; Huserbråten *et al.*, 2013), the Atlantic cod (*Gadus morhua*; Knutsen *et al.*, 2003), and the corkwing wrasse (*Symphodus melops*; Knutsen *et al.*, 2013).

To visualize spatial patterns of genetic differentiation, estimates of F_{ST} among sample pairs were analysed with the BARRIER software ver. 2.2 (Manni *et al.*, 2004). We performed separate tests for each locus and overall loci (all with four barriers) to ensure that spatial patterns were robust. The test applying all loci denotes the rank of importance of these barriers, whereas the number of loci supporting a given barrier is provided by single locus tests. Coordinates for temporal samples were shifted slightly (when identical), relative to positions in Table 1, to force the software to treat them as separate samples.

Results

Genetic results

We successfully scored 1797 individuals in at least 6 of the 11 microsatellite loci. Locus PbD9 did not amplify at all localities and together with locus PbA104a evidenced large deficit of heterozygotes apparently due to the presence of null alleles (according to MICROCHECKER; Van Oosterhout *et al.*, 2004). These two loci were omitted from further analyses, which were thus based on nine loci (Table 2; Supplementary Appendix). Genetic variability at the nine loci spanned from 6 alleles at locus SD3-62 to 58 alleles at locus PbC109, with a range of expected heterozygosity (H_E) of 0.179 (SD3-62) to 0.928 (PbC109; Table 2). There was a slight tendency for excess homozygotes relative to Hardy–Weinberg expectations, with 25 estimates being significant before FDR correction and 5 of 180 (i.e. 9 loci in 20 samples) F_{IS} estimates significant at 5% level under the FDR (cf. Supplementary Appendix). Significant estimates were not clustered to specific loci or samples and most likely reflected artefacts.

Overall spatial genetic structure was weak, but statistically significant ($F_{ST} = 0.0009$, $p < 0.0001$) and did not change when lumping temporal replicates vs. including them as separate samples (not shown). Pairwise estimates among all samples of F_{ST} ranged from −0.0028 to 0.0046, and 32 of these estimates (17%) were significant under FDR (Table 3). The significant pairs refer to different localities and none of the temporal comparisons were significant. Instead, most significant comparisons were either among fjords and oceanic localities (21), or among fjords (5), while only six significant comparisons were among oceanic localities (one between Fladen and Skagerrak, three between Norwegian Deep and Skagerrak locations, and two between the Norwegian Deep and Fladen Ground).

On average, genetic differentiation among fjords was at the same level ($F_{ST} = 0.0010$, $p = 0.0002$) as the overall estimate, while comparing fjords and oceanic/offshore samples of Skagerrak also revealed similar genetic divergence ($F_{ST} = 0.0011$). The three oceanic/offshore Skagerrak samples were genetically more homogeneous ($F_{ST} = 0.0006$). The AMOVA suggested temporal stability in the spatial genetic structure (among spatially grouped samples $F_{CT} = 0.0010$, $p < 0.0001$; among temporal samples within groups $F_{SC} = 0.0003$, $p = 0.23$).

An evaluation of the statistical power estimated an 89% probability of detecting structure for a F_{ST} value of ca. 0.0020 (Figure 4). This result suggests that our tests should most likely detect a real population structure if true estimates of F_{ST} were at this level or above. Several pairwise estimates span at about this level (Table 3), illustrating that statistical power in our study was at the limit or higher of

Table 2. Summary statistics for each microsatellite locus and averages over loci.

Loci	*A*	H_E	F_{ST}
Pbc105	17	0.85	0.0007
Pbc8	29	0.721	0.0037
Sd2-14	26	0.706	−0.0007
PbA110	16	0.694	−0.0002
PbC109	58	0.928	**0.0010**
PbA1	18	0.767	0.0022
SD1-41	27	0.855	−0.0005
SD2-68	15	0.579	0.0014
SD3-62	6	0.179	−0.0008
Average	23.5	0.698	0.0009

A is the observed number of alleles at each locus, H_E is expected heterozygosity in the pooled sample, and F_{ST} quantifies genetic differences among all 20 samples (no pooling of temporal replicates). Bold estimates (F_{ST}) indicate significance (at 5% FDR or higher) for the null hypothesis of no allele frequency differentiation among samples.

Table 3. Estimates of pairwise genetic differentiation (F_{ST}) among samples.

	FL07	FL11	SA11	SC11	SE11	SA12	SC12	ST10	ST12	GR10	TO10	HA10	HA12	OS10	OS12	KO10	GF10	GF12	NS
FL07																			
FL11	0.0002																		
SA11	−0.0001	0.0011																	
SC11	0.0018	0.0004	0.0000																
SE11	0.0003	0.0009	0.0010	0.0004															
SA12	0.0007	−0.0007	0.0001	−0.0010	0.0004														
SC12	**0.0031**	−0.0001	0.0024	0.0000	0.0007	−0.0005													
ST10	0.0008	−0.0003	**0.0019**	0.0013	0.0029	**0.0009**	0.0016												
ST12	0.0011	−0.0006	0.0021	0.0002	0.0001	0.0007	0.0001	0.0013											
GR10	0.0032	0.0008	0.0026	−0.0017	0.0003	0.0007	−0.0005	0.0029	0.0004										
TO10	0.0045	**0.0036**	**0.0042**	**0.0014**	0.0030	**0.0023**	**−0.0001**	0.0033	**0.0027**	**0.0017**									
HA10	0.0018	−0.0002	0.0009	−0.0028	0.0003	−0.0009	−0.0013	0.0011	0.0004	−0.0008	0.0002								
HA12	−0.0004	0.0005	−0.0001	−0.0012	−0.0005	−0.0009	−0.0001	0.0009	0.0006	0.0003	0.0002	−0.0014							
OS10	0.0021	0.0001	0.0002	−0.0012	0.0013	0.0001	**0.0022**	0.0024	0.0006	**0.0003**	**0.0046**	0.0002	0.0012						
OS12	0.0003	0.0001	0.0000	0.0012	0.0007	0.0010	0.0002	0.0001	0.0016	0.0014	0.0021	0.0008	−0.0016	0.0012					
KO10	0.0005	0.0007	−0.0007	−0.0011	−0.0011	−0.0017	−0.0004	0.0012	0.0002	−0.0012	0.0008	−0.0014	−0.0005	0.0013	0.0001				
GF10	0.0037	0.0014	0.0030	−0.0009	0.0013	−0.0002	−0.0012	0.0007	−0.0005	−0.0012	−0.0020	−0.0015	−0.0012	0.0011	0.0013	−0.0004			
GF12	0.0034	**0.0030**	0.0015	0.0012	**0.0034**	**0.0015**	0.0003	0.0039	0.0043	0.0025	0.0030	0.0010	−0.0007	**0.0030**	0.0015	0.0014	0.0027		
NS	**0.0025**	0.0006	**0.0039**	**0.0024**	0.0011	**0.0008**	0.0006	0.0024	0.0003	0.0021	**0.0026**	**0.0021**	−0.0002	**0.0035**	**0.0026**	**0.0018**	0.0000	**0.0038**	
NN	**0.0018**	−0.0002	0.0024	0.0007	0.0009	0.0001	0.0006	0.0015	0.0004	0.0014	**0.0030**	**0.0007**	0.0008	**0.0027**	0.0026	0.0011	0.0011	**0.0034**	−0.0009

Statistically significant comparisons at 5% level or more (after FDR correction, Benjamini and Yekutieli, 2001) in bold (from exact *G*-tests of allele frequency heterogeneity).

detection. For detecting divergence at 0.0010 or lower power dropped rapidly, indicating that some structure could go unnoticed in our tests.

The Barrier visualization (Figure 5) revealed the following general pattern: (i) fjord populations are generally divergent from each other and from offshore areas. Some fjords deviate from this pattern [Håøya (HA) and Stolsfjord (ST)]. (ii) No apparent structure among oceanic samples collected in Skagerrak and the Norwegian Deep. (iii) Temporal replicates group generally within the same barrier, supporting the results of the AMOVA analysis of a larger spatial than temporal component of genetic divergence.

Survey and commercial fisheries data

In general, trends in the survey stock biomass indices for Skagerrak and the Norwegian Deep followed the same pattern. Both indices increased from 1988 to 1997 and both have decreased to a very low level in recent years (Figure 2b). The two indices differ, however, in some years, with the most notable difference being the distinctive peak in biomass in 2007 in Skagerrak, which was not observed in the Norwegian Deep. Recruitment has remained at a steady low level in the Norwegian Deep in the period 2006–2014, while it has shown large fluctuations in Skagerrak (Figure 3).

Commercial Danish shrimp fishery in Farn Deeps only took place in 3 years, from 1987 to 1989, and was terminated on the Fladen Ground in 2003 (Figure 2a). Daily catch rates (LPUE) were substantially higher in the latter area compared with Skagerrak, the Norwegian Deep, and Farn Deeps, suggesting higher densities of shrimp on the Fladen Ground. In some years, the Fladen Ground time-series differs considerably from those of both Skagerrak and the Norwegian Deep, with directly opposite trends from 1992 to 1996 and again from 1998 to 2002. However, the large peak in biomass in 1997–1998 is seen in all three time-series. Furthermore, the Fladen Ground LPUE is strikingly similar to the survey biomass index from the Norwegian Deep, with both time-series having peaks in 1991, 1997, and 2001 (cf. Figure 2a and b). Only for the years 1992–1995, do these two time-series diverge. The Skagerrak and Norwegian Deep LPUE time-series agree reasonably well throughout the period, but show the same 2007 scaling difference as the survey biomass indices.

Discussion

The spatial structuring of shrimp in the North Sea and Skagerrak area has long been the subject of scientific discussion and speculation (ICES, 2003, 2013) as the geographic extent of populations may suggest how local or regional fishing impacts the stocks. Overall, we find a low, but statistically significant genetic structure ($F_{ST} = 0.0009$): samples collected from open waters within Skagerrak and the Norwegian Deep show little or no genetic structure, despite the finding of contrasting recruitment patterns and different size-at-age distribution (Figure 3). However, shrimp from the Fladen Ground in the western North Sea and several Skagerrak fjords seem to be genetically divergent.

Our finding of a general lack of genetic divergence between Skagerrak and the Norwegian Deep is in accordance with the present assessment and management regime of shrimp in this area, which defines the entire Skagerrak and Norwegian Deep as one single management unit (ICES, 2003, 2013). Thus, as judged from the genetic data, and similar patterns in abundance fluctuations (LPUE and survey data, cf. Figure 2), shrimp in these two areas may constitute one common biological population. As mentioned above, shrimp in the two areas do show some differences

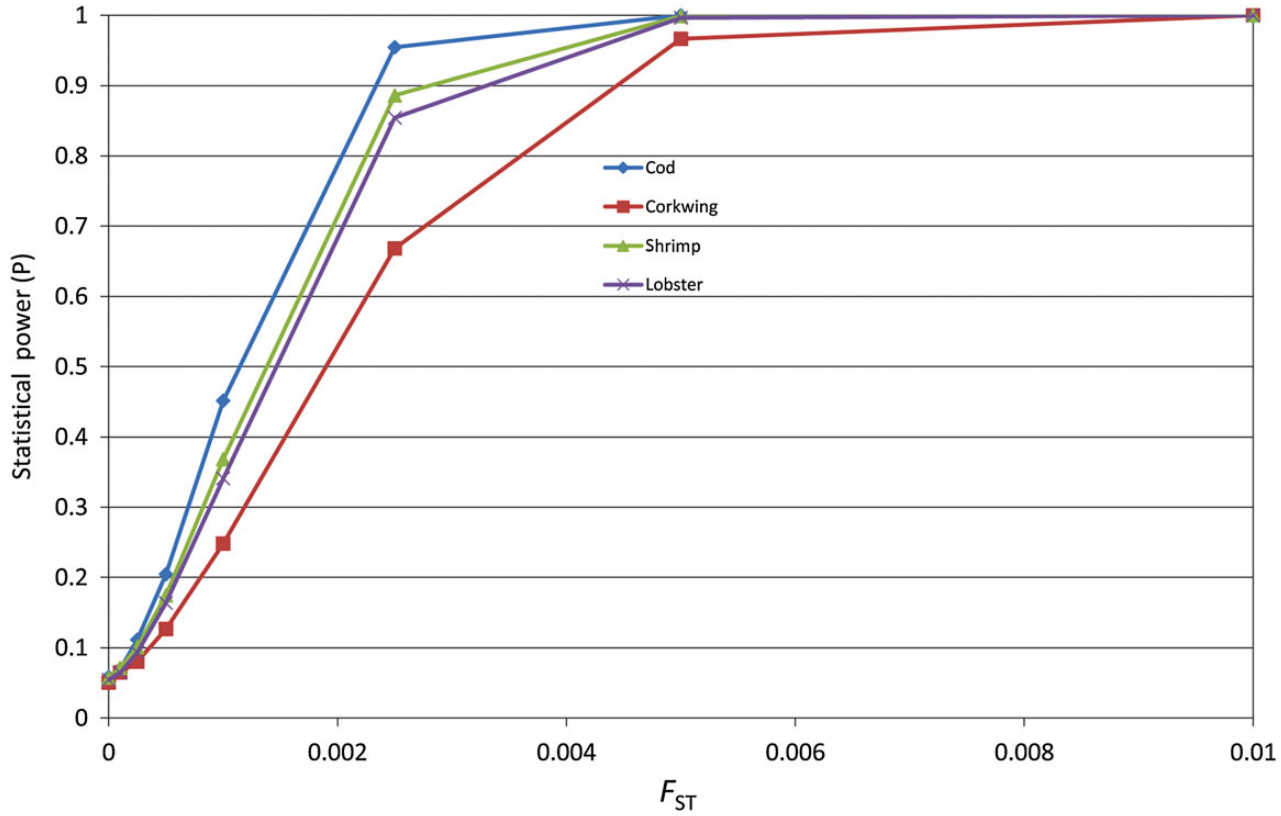

Figure 4. Comparison of statistical power (*y*-axis) with the level of genetic differentiation F_{ST} (*x*-axis) among four species sampled ($n = 96$ for all species) along the Skagerrak coast over a comparable geographic range: Atlantic cod (Knutsen *et al.*, 2003), European lobster (Huserbråten *et al.*, 2013), and corkwing wrasse (Knutsen *et al.*, 2013).

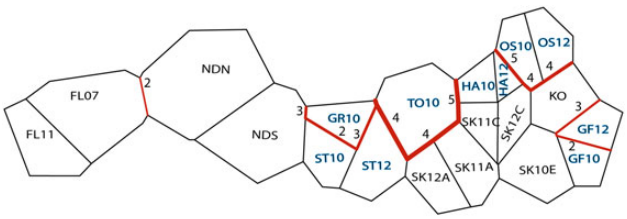

Figure 5. BARRIER-inferred restrictions or "barriers" to gene flow among sample localities, based on nine microsatellite loci. Thickness of red line denotes the rank of importance of the respective barrier, and numbers denote the number of loci supporting each barrier. Sample abbreviations (with sample years appended) are given in Table 1. Blue abbreviations indicate fjord samples.

in growth and longevity, but the genetic basis for these differences is unclear and may simply reflect different environmental conditions, e.g. in food availability.

The Norwegian Deep and Skagerrak are connected by strong oceanic currents. A branch of the North Atlantic Current flows into the Norwegian Deep from the west and follows the trench into Skagerrak where the current joins with coastal water from Kattegat and finally flows out of Skagerrak as the Norwegian Coastal Current (Figure 1). Shrimp could be transported by the strong current, thereby joining shrimp in the two areas into a common gene pool. The large difference in recruitment (Figure 3) suggests that Skagerrak constitutes a nursery ground for the Skagerrak/Norwegian Deep stock and that juvenile shrimp gradually move westwards, as has been claimed by local fishers for years. Spatial variation in the distribution of different size (age) groups is known from other shrimp stocks and may be explained by ontogenetic migration (Simard and Savard, 1990; Aschan, 2000).

Our genetic results are consistent with this notion of a common biological population within oceanic Skagerrak and the Norwegian Deep. However, it should be kept in mind that statistical power in detecting very small genetic differences (0.0010 or below) was found to be low, and so we cannot exclude the possibility of as yet undetected genetic differences between areas going unnoticed (cf. Figure 4), should there be any.

In contrast to the Skagerrak/Norwegian Deep situation, shrimp from the Fladen Ground tended to differ genetically from other locations (Table 3 and Figure 5) and might represent a separate population. A similar explanation may be given for the Danish LPUE time-series, which suggest different population dynamics on the Fladen Ground compared with in Skagerrak and the Norwegian Deep. The average catch rate on the Fladen Ground was roughly twice as high as in Skagerrak and the Norwegian Deep in overlapping periods (Figure 2a), although LPUE standardization may have been biased. Bias could arise from a combination of (i) different fleet compositions of the areas (only the largest vessels are capable of fishing on the Fladen Ground) and (ii) technological creep (increasing vessel catchability from advances in, e.g., navigational aids and gear design) taking place at a higher pace on-board larger vessels (Eigaard, 2009; Eigaard *et al.*, 2014). Furthermore, as only daily catch rates exist, we cannot rule out the possibility that vessels on the Fladen Ground fished for longer days (more hours)

compared with vessels in Skagerrak and the Norwegian Deep, where fishing grounds are closer to harbour. Yet, the catch rate differences are of such magnitude that they would most likely not be balanced out by further standardization, thus strongly indicating real population differences. Also the periodically opposite trends of the Fladen Ground LPUE time-series compared with the much more similar Skagerrak and Norwegian Deep LPUE time-series support the view of different population dynamics. Contrary to this, the similar trends in the Fladen Ground catch rate and the survey index from the Norwegian Deep may indicate some connection between these two areas. Ocean currents linking the Fladen Ground and the Norwegian Deep and Skagerrak area are weak compared with the strong currents flowing into and out of the Skagerrak basin (Figure 1). The combination of genetic and time-series data together with ocean current information suggests that the Fladen Ground shrimp constitute a separate population from both Norwegian Deep and Skagerrak open and fjord shrimp.

The inclusion of fjord samples in the analysis slightly increased the level of genetic differentiation (from $F_{ST} = 0.0006$ among oceanic samples to 0.0010 among fjords), indicating some genetic structuring at the fjord level. The BARRIER plot (Figure 5) revealed fjord shrimp as the most heterogenous, thus indicating the presence of local populations within at least some Skagerrak fjords. Hence, shrimp in local fjords exploited by small boats appear genetically more structured than those living in open waters. Structure between fjords/bays and open coast has also been found in other organisms (e.g. mussels: Nicastro *et al.*, 2008, Teske *et al.*, 2012), including cod in the Skagerrak region (Knutsen *et al.*, 2011). Oceanographic features, such as bathymetry, temperature, and salinity boundaries, and habitat heterogeneity may all act as barriers to population connectivity in marine species in this region (e.g. cod, *G. morhua*, Jorde *et al.*, 2007; corkwing wrasse, *S. melops*, Knutsen *et al.*, 2013; and sprat, *Sprattus sprattus*, Glover *et al.*, 2011). This finding of local fjord populations contradicts current management regulations, which assume shrimp in the entire region to represent a single stock, except for the Gullmarsfjord shrimp stock which is managed separately. In general, fjord populations are probably of less economic value, but may help maintaining genetic variability and biocomplexity of the species in this region (cf. Ruzzante *et al.*, 2006; Olsen *et al.*, 2008; Schindler *et al.*, 2010). Considering that Skagerrak is already among the warmest part of the species' range, and that temperatures are currently increasing there (e.g. Knutsen *et al.*, 2013), genetic variability for further adaptations to changing conditions may prove crucial to the species' continued presence in this region.

Supplementary material

Supplementary material is available at the *ICESJMS* online version of the manuscript.

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References

Aschan, M. 2000. Spatial variability in length frequency distribution and growth of shrimp (*Pandalus borealis* Krøyer 1838) in the Barents Sea. Journal of Northwest Atlantic Fishery Science, 27: 93–105.

Benjamini, Y., and Hochberg, Y. 1995. Controlling the false discovery rate: a practical and powerful approach to multiple testing. Journal of Royal Statistical Society, 57: 289–300.

Benjamini, Y., and Yekutieli, D. 2001. On the control of false discovery rate in multiple testing under dependency. Annals of Statistics, 29: 1165–1168.

Bergström, B. I. 2000. The biology of *Pandalus*. Advances in Marine Biology, 38: 55–244.

Bhattacharya, C. G. 1967. A simple method of resolution of a distribution into Gaussian components. Biometrics, 23: 115–135.

Boitsov, V. D., Karsakov, A. L., and Trofimov, A. G. 2012. Atlantic water temperature and climate in the Barents Sea, 2000–2009. ICES Journal of Marine Science, 69: 833–840.

Bøhle, B. 1977. Vekst og utvilking av larver og postlarver av dypvannsreke (*Pandalus borealis* Krøyer) ved eksperimentelle forhold. Fisken og Havet, Serie B 1977 Nr. 10. 33 pp.

Cadrin, S. X., Kerr, L. A., and Mariani, S. 2013. Stock identification methods: an overview. *In* Stock Identification Methods, Applications in Fishery Science, 2nd edn. Ed. by S. X. Cadrin, L. A. Kerr, and S. Mariani. Academic Press, London, UK. 588 pp.

Ciannelli, L., Fisher, J. A. D., Mauritzen, M. S., Hunsicker, M. E., Hidalgo, M., Frank, K. T., and Bailey, K. M. 2013. Theory, consequences and evidence of eroding population spatial structure in harvested marine fishes: a review. Marine Ecology Progress Series, 480: 227–243.

Eigaard, O. R. 2009. A bottom-up approach to technological development and its management implications in a commercial fishery. ICES Journal of Marine Science, 66: 916–927.

Eigaard, O. R., and Munch-Petersen, S. 2011. Influence of fleet renewal and trawl development on landings per unit effort of the Danish northern shrimp (*Pandalus borealis*) fishery. ICES Journal of Marine Science, 68: 26–31.

Eigaard, O. R., Marchal, P., Gislason, H., and Rijnsdorp, A. D. 2014. Technological development and fisheries management. Reviews in Fisheries Science and Aquaculture, 22: 156–174.

Excoffier, L., and Lischer, H. E. L. 2010. Arlequin suite ver 3.5: a new series of programs to perform population genetics analyses under Linux and Windows. Molecular Ecology Resources, 10: 564–567.

Funk, C. F., McKay, J. K., Hohenlohe, P. A., and Allendorf, F. W. 2012. Harnessing genomics for delineating conservation units. Trends in Ecology and Evolution, 27: 489–496.

Furevik, T. 2001. Annual and interannual variability of Atlantic Water temperatures in the Norwegian and Barents Seas: 1980–1996. Deep-Sea Research I, 48: 383–404.

Glover, K. A., Skaala, Ø., Limborg, M., Kvamme, C., and Torstensen, E. 2011. Microsatellite DNA reveals population genetic differentiation among sprat (*Sprattus sprattus*) sampled throughout the Northeast Atlantic, including Norwegian fjords. ICES Journal of Marine Science, 68: 2145–2151.

Goudet, J. 2001. FSTAT, a program to estimate and test gene diversities and fixation indices version 2.9.3. www2.unil.ch/popgen/softwares/fstat.htm.

Huserbråten, M. B., Moland, E., Knutsen, H., Olsen, E. M., André, C., and Stenseth, N. C. 2013. Conservation, spillover and gene flow within a network of Northern European Marine Protected Areas. PLoS ONE, 8. doi:10.1371/journal.pone.0073388.

Hutchinson, W. F. 2008. The dangers of ignoring stock complexity in fishery management: the case of the North Sea cod. Biology Letters, 4: 693–695.

Hvingel, C. 2006. Construction of biological advice for the management of a northern shrimp fishery—the West Greenland example. Doctoral thesis. Greenland Institute of Natural Resources, Nuuk,

Greenland/Norwegian College of Fishery Science, University of Tromsø, Norway.

ICES. 2003. Report of the *Pandalus* Assessment Working Group. ICES CM 2004/ACFM: 05. 26–29 August 2003. Lysekil, Sweden. 37 pp.

ICES. 2004. Report on the *Pandalus* Assessment Working Group. ICES CM 2005/ACFM: 05. 27 October—5 November 2004. Copenhagen, Denmark. 74 pp.

ICES. 2013. NAFO/ICES *Pandalus* Assessment Group Meeting, 12–19 September 2013. NAFO SCS Doc. 13/19 Serial No. N6235. ICES CM 2013/ACOM: 14. 70 pp.

Jorde, P. E., Knutsen, H., Espeland, S. H., and Stenseth, N. C. 2007. Spatial scale of genetic structuring in coastal cod *Gadus morhua* and geographic extent of local populations. Marine Ecology Progress Seriens, 343: 229–237.

Knutsen, H., Jorde, P. E., André, C., and Stenseth, N. C. 2003. Fine-scaled geographic population structuring in a highly mobile marine species: the Atlantic cod. Molecular Ecology, 12: 385–394.

Knutsen, H., Olsen, E.M., Jorde, P.E., Espeland, S.H., André, C., and Stenseth, N.C. 2011. Are low but statistically significant levels of genetic differentiation in marine fishes "biologically meaningful"? Molecular Ecology, 20: 768–783.

Knutsen, H., Jorde, P. E., Gonzalez, E. B., Robalo, J., Albretsen, J., and Almada, V. 2013. Climate change and genetic structure of leading edge and rear end populations in a northwards shifting marine fish species, the corkwing wrasse (*Symphodus melops*). PLoS ONE, 8: e67492.

Manni, F., Guérard, E., and Heyer, E. 2004. Geographic patterns of (genetic, morphologic, linguistic) variation: how barriers can be detected by using Monmonier's algorithm. Human Biology, 76: 173–190.

Nei, M., and Chesser, R. K. 1983. Estimation of fixation indices and gene diversities. Annals of Human Genetics, 47: 253–259.

Nicastro, K. R., Zardi, G. I., McQuaid, C. D., Teske, P. R., and Barker, N. P. 2008. Coastal topography drives genetic structure in marine mussels. Marine Ecology Progress Series, 368: 189–195.

Olsen, E. M., Knutsen, H., Gjøsæter, J., Jorde, P. E., Knutsen, J. A., and Stenseth, N. C. 2008. Small-scale biocomplexity in coastal Atlantic cod supporting a Darwinian perspective on fisheries management. Evolutionary Applications, 1: 524–533.

Ouellet, P., and Allard, J. P. 2006. Vertical distribution and behavior of shrimp *Pandalus borealis* larval stages in thermally stratified water columns: laboratory experiment and field observations. Fisheries Oceanography, 15: 373–389.

Pereyra, R. T., Westgaard, J.-I., Dahl, M., Johansen, T., Knutsen, H., Ring, A.-K., Søvik, G., *et al.* 2012. Isolation and characterization of nuclear microsatellite loci in the northern shrimp, *Pandalus borealis*. Conservation Genetics Resources, 4:109–112.

Reiss, H., Hoarau, G., Collas, M. D., and Wolff, W. J. 2009. Genetic population structure of marine fish: mismatch between biological and fisheries management units. Fish and Fisheries, 10: 361–395.

Rousset, F. 2008. GENEPOP'007: a complete re-implementation of the GENEPOP software for Windows and Linux. Molecular Ecology Resources, 8: 103–106.

Ruzzante, D., Mariani, S., Bekkevold, D., André, C., Mosegaard, H., Clausen, L., Dahlgren, T., *et al.* 2006. Biocomplexity in a highly migratory marine pelagic fish. Proceedings of the Royal Society London Series B, 273: 1459–1464.

Ryman, N., and Palm, S. 2006. POWSIM: a computer program for assessing statistical power when testing for genetic differentiation. Molecular Ecology Notes, 6: 600–602.

Ryman, N., Palm, S., André, C., Carvalho, G., Dahlgren, T., Jorde, P. E., Laikre, L., *et al.* 2006. Power for detecting genetic divergence: differences between statistical methods and marker loci. Molecular Ecology, 15: 2031–2045.

Schindler, D. E., Hilborn, R., Chasco, B., Boatright, C. P., Quinn, T. P., Rogers, L. A., and Webster, M. S. 2010. Population diversity and the portfolio effect in an exploited species. Nature, 465: 609–613.

Schlüter, M., and Jerosch, K. 2009. Digital atlas of the North Sea. Alfred Wegener Institute for Polar and Marine Research. 91 pp. hdl:10013/epic.34893.d001.

Shumway, S. E., Perkins, H. C., Schick, D. F., and Stickney, A. P. 1985. Synopsis of biological data on the pink shrimp, *Pandalus borealis*, Krøyer, 1838. NOAA Technical Report NMFS 30. FAO Fisheries Synopsis No. 144. 57 pp.

Simard, Y., and Savard, L. 1990. Variability, spatial patterns and scales of similarity in size-frequency distributions of the northern shrimp (*Pandalus borealis*) and its migrations in the Gulf of St. Lawrences. Canadian Journal of Aquatic and Marine Science, 47: 794–804.

Søvik, G., and Thangstad, T. 2013. The Norwegian Fishery for Northern Shrimp (*Pandalus borealis*) in Skagerrak and the Norwegian Deep (ICES Divisions IIIa and IVa east), 1970–2013. NAFO SCR Doc. 13/066. 33 pp.

Teske, P., Papadopoulos, I., Barker, N., and McQuaid, C. 2012. Mitochondrial DNA paradox: sex-specific genetic structure in a marine mussel—despite maternal inheritance and passive dispersal. BMC Genetics, 13: doi:10.1186/1471-2156-13-45.

Tveite, S. 2000. Suitability of a fixed station shrimp abundance survey in the Skagerrak-Norwegian Deeps for stock assessments and associated research. Journal of Northwest Atlantic Fishery Science, 27: 177–182.

Van Oosterhout, C., Hutchinson, W. F., Wills, D. P. M., and Shipley, P. 2004. MICRO-CHECKER: software for identifying and correcting genotyping errors in microsatellite data. Molecular Ecology Notes, 4: 535–538.

Waples, R. S., Punt, A. E., and Cope, J. M. 2008. Integrating genetic data into management of marine resources: how can we do it better? Fish and Fisheries, 9: 423–449.

Weir, B. S., and Cockerham, C. C. 1984. Estimating *F*-statistics for the analysis of population structure. Evolution, 38: 1358–1370.

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