Population genetic structure of common dolphins in the North-east Atlantic using microsatellite loci and mtDNA control region markers

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ABSTRACT

Short-beaked common dolphins (*Delphinus delphis*) are widespread in the North-east Atlantic, and have been regularly reported as bycatch in commercial fisheries. In this region, seasonal changes in abundance suggest movements on a regional scale and a high potential for dispersal. A number of studies have shown population segregation at a moderate to large geographical scale in the North Atlantic Ocean. However, population structure studies have been hampered by inaccessibility to offshore areas and sample availability, and the impact of bycatch on demographic parameters and genetic variability is currently unknown. Genetic variability among common dolphins stranded or bycaught in the North-east Atlantic waters off Portugal, France, Ireland, Celtic Sea, western English Channel and Scotland, were investigated using 25 microsatellite loci and 556bp of the mitochondrial DNA (mtDNA) control region. Results suggest that common dolphins show high levels of genetic variability, and do not appear to have been affected by strong recent demographic changes, such as a reduction of population size due to high incidental mortality rates. Population structure analyses indicate that common dolphins found in the sampled areas may be part of the same population as a result of strong gene flow (maintaining homogeneous gene frequencies throughout the sampled areas) or, alternatively but not exclusively, as a result of a recent population split (*i.e.* not enough time has passed to generate significant genetic differentiation). These findings suggest the presence of a large 'coastal' panmictic (random mating) population in the North-east Atlantic. These results corroborate previous genetic studies in the North-east Atlantic. However, we were unable to examine population structure in other areas within the same region, especially between inshore and offshore habitats. Furthermore, a lack of genetic structure does not mean that ecological (contemporary) differences should be ignored, as evidenced by non-genetic studies. Future studies should focus on incorporating animals from offshore waters, and should include dedicated survey data and strategic sampling approaches (e.g. temporal and spatial aggregations using remote biopsy darting systems).

Key words: common dolphin, *Delphinus delphis*, genetics, incidental catches, conservation

INTRODUCTION

The short-beaked common dolphin (*Delphinus delphis*) is among the most abundant cetacean species in the North-east Atlantic. They are widely distributed in coastal, continental shelf and open ocean waters, ranging from subtropical waters off Africa, into the Mediterranean Sea, northwards to approximately 65ºN latitude, west of Norway (Haug 1981; Weir *et al.* 2001; Reid *et al.* 2003; Bearzi *et al.* 2003; Murphy *et al.* 2008), and westwards to the mid-Atlantic ridge (Cañadas *et al.* 2008; Doksaeter *et al.* 2008). Common dolphins are regarded as extremely mobile with a high potential for dispersal. For instance, a female common dolphin fitted

with a radio transmitter and tracked for 72 hours in the Northeast Pacific, was found to have travelled approximately 270 nautical miles from her point of release over a ten days period (Evans, 1982), and even greater movements have been documented for this species in the eastern tropical Pacific (Danil & Chivers, 2006). Seasonal shifts in abundance and distribution have been noted throughout the North-east Atlantic (Evans, 1992; Pollock *et al*. 1997; O'Cadhla *et al*. 2003; Kiszka *et al*. 2004; Northridge *et al*. 2004, MacLeod *et al*. in press).

Generally, abundance estimates for common dolphins have shown relatively high numbers in waters west of Britain and Ireland, in the English Channel, and in continental shelf waters of France, Spain and Portugal. During July 2005, common dolphin abundance was estimated as 63,366 (CV = 0.46) individuals in continental shelf waters of the North-east Atlantic (SCANS-II 2006). In July 2007, 116,709 (CV = 0.34) individuals were estimated for European offshore waters by the CODA project (Cañadas *et al*. SC/61/SM6), which surveyed waters west of the shelf, within the jurisdictions of the UK, Ireland, France and Spain. Common dolphins are one of the main cetacean species frequently reported as bycatch by a number of different fisheries throughout the North-east Atlantic (*e.g*. Tregenza & Collet, 1998; Morizur *et al.* 1999; Lopez *et al*. 2003; Rogan and Mackey 2007), which has raised some concern about the status of the species. The pelagic albacore tuna (*Thunnus alalunga*) driftnet fisheries were closed in 2001 due to high mortality rates of non-target species, with common dolphins being one of the most affected species (Rogan and Mackey 2007). However, the impact of such mortality-rates on levels of genetic diversity and hence population viability is difficult to evaluate, mainly because of poor knowledge of population structure and relative abundance.

A cranial morphometric study undertaken in the North-east Atlantic concluded that morphologically, *D. delphis* appeared to be a very variable species. Further, common dolphins from this region should be classified as a larger-form of *D. delphis* found in the North-east Pacific and the Black Sea (Murphy *et al*. 2006). Genetic studies have also confirmed the existence of *D. delphis* in the North-east Atlantic (Natoli *et al*., 2006; Amaral *et al*. 2007). However, Amaral *et al*. (2007) did identify a group of highly divergent individuals (five out of 69 individuals) in this region, which were attributed to: introgressive hybridization between *D. delphis* and *D. capensis*, a separate species, or the existence of two divergent lineages within *D. delphis*.

The investigation of population structure of common dolphins throughout the North Atlantic and the Mediterranean Sea has been hampered by inaccessibility to offshore areas and sample availability, however, a number of studies have shown population segregation at a moderate to large geographical scale. Some population differentiation was reported within the North-east Atlantic, especially between the southern (*e.g*. Portugal) and northern (*e.g*. Scotland) distributional limits of the studies (Natoli *et al*. 2006; Amaral *et al*. 2007). Similarly, Murphy *et al*. (2006) revealed some population differentiation using cranial morphometric data, though it is recognized that morphological traits are influenced by the environment and so must be interpreted differently to genetic traits. Natoli *et al*. (2003) found no significant differentiation using nine microsatellite loci between common dolphins in the western Mediterranean (samples obtained from the Alboran Sea and Algeria) and the contiguous North Atlantic (samples obtained from the Strait of Gibraltar and Portugal), which may indicate gene flow between those areas. In contrast, significant differentiation was found between samples obtained from England and the western Mediterranean (Natoli *et al*. 2008). A recent genetic study indicated no structure among dolphins stranded along the south-west coasts of Ireland, summer concentrations in the Celtic Sea and winter concentrations in the western English Channel, although these were genetically distinct from dolphins from the North-west Atlantic (Mirimin *et al*. 2009). Common dolphins occurring in the north-West Atlantic were assumed to consist of a single population (Waring *et al*. 2007) and results from recent mitochondrial DNA control region and cranial morphometric studies provided evidence for this hypothesis (Westgate 2005; Westgate and Read 2007). Using both mtDNA and nuclear microsatellite loci, Mirimin *et al*. (2009) showed moderate but significant differentiation between western and eastern North Atlantic populations using both microsatellites (F_{ST} = 0.005, $P < 0.05$) and mtDNA control region data (F_{ST} = 0.018, $P < 0.001$), confirming the presence of at least two genetically distinct populations on either side of the North Atlantic, thus indicating that mid-Atlantic areas may not present a strong barrier to gene flow for this species.

A number of studies have provided some evidence of ecological variation, based on stable isotopes signatures and cadmium levels, highlighting some contemporary population structure in North-east Atlantic waters. Das *et al*. (2003) reported conservative trophic habitats between common dolphins that stranded along the Irish coastline and the French channel coast, based on variations in stable isotope signatures (Carbon-13 and Nitrogen-15). However, mean muscle and liver carbon-13 were significantly different for animals from the Irish coast (n = 8) compared to animals from the North-French coast (n = 14), which may reflect a more oceanic/offshore diet for common dolphins that stranded along the Irish coastline (Das *et al*. 2003). Analyses of cadmium levels in the Bay of Biscay revealed significantly higher levels in common dolphins from oceanic

waters as opposed to neritic waters off the French Atlantic coast, suggesting that two possible ecological stocks exist within the North-east Atlantic (*i.e.* neritic and oceanic) (Lahaye *et al*. 2005). It should be noted though that the sample size for the oceanic sample was small (n = 10) and obtained 10-15 years prior to the majority of the neritic sample. In contrast, analyses of lead concentrations and isotopic composition (206Pb/207Pb) revealed no geographical differences between common dolphins stranded on the north coasts of Brittany and Galicia, suggesting movements between these areas (Caurant *et al*. 2006).

The aim of the present study is to further investigate population structure of common dolphins in the North-east Atlantic by incorporating samples from adjacent areas from a continuous geographical range between Portugal and the British Isles, and using a large number of genetic markers in order to improve statistical power. In total, we included 192 common dolphins from six Atlantic areas (Portugal, France, west English Channel, Celtic Sea, Ireland and Scotland) and analysed up to 25 microsatellite loci and 556 base-pairs (bp) of the mitochondrial DNA (mtDNA) control region. We tested the hypothesis of genetic structure versus panmixia among the sampled areas and discuss the possible impact of bycatch on the genetic variability of common dolphins in North-east Atlantic coastal waters.

MATERIALS AND METHODS

A total of 192 individual samples were collected from animals stranded on the Atlantic coasts of Ireland (n = 50), France (n = 46), and Portugal (n = 39), and from individuals by-caught in the Irish albacore tuna driftnet fishery that operated in the Celtic Sea (n = 21) and in the UK sea bass (*Dicentrarchus labrax*) pelagic trawl fishery, which operates in the western English Channel (n = 36). Samples were obtained between 1991 and 2006 (Table 1; Figure 1). Sequences of the mtDNA control region from an additional 13 stranded individuals sampled in Scotland were obtained from a recently published study by Amaral *et al.* (2007).

Number of alleles, expected and observed heterozygosity, linkage between pairs of loci and Hardy-Weinberg equilibrium tests were performed for each sampled area (Ireland, Celtic Sea, English Channel, Scotland, France and Portugal), using ARLEQUIN 3.11 (Excoffier *et al*. 2005). Population structure was assessed using an analysis of molecular variance (AMOVA, Excoffier *et al*. 1992), which calculates the proportion of genetic variation due to population structure, taking into account sampling location. Two estimators of genetic differentiation, F_{ST} (Weir & Cockerham, 1984) (obtained from a locus-by-locus AMOVA) and Rho_{ST} (Goodman, 1997) (over all samples), were calculated for microsatellites. F_{ST} is calculated using allele frequencies only, whereas, Rho_{ST} takes into account both allele size (based on a specific model of mutation for microsatellites) and allele frequencies. These tests were carried out using the computer programs ARLEQUIN 3.11 (Excoffier *et al*. 2005) and RSTCALC 1.0 (Goodman 1997), respectively.

Mitochondrial DNA sequences were also analysed using an AMOVA framework using two estimators of genetic differentiation: F_{ST} is based on haplotype frequencies only, whereas Φ_{ST} incorporates distances between haplotypes (Tamura and Nei 1993). For mtDNA control region analyses, Ireland and Celtic Sea samples were pooled together, and samples from the English Channel (only two sequences were available) were not included in the analysis. In order to test for a possible bias in dispersal between sexes, analyses were repeated for males and females separately. All analyses of mtDNA sequence data were carried out using the computer program ARLEQUIN 3.11 (Excoffier *et al*. 2005). The significance level of all estimated values was tested after 10,000 permutations.

Using microsatellite data, the Bayesian approach implemented in the program STRUCTURE 2.0 (Pritchard *et al*. 2000) was used to evaluate the number of different genetic stocks/populations (K) that best explained the genetic variability observed across the whole dataset, without taking into account sampling location. After choosing appropriate burn in and run lengths (300,000 and 1,000,000, respectively), a number of independent runs were carried out for K = 1 to K = 4, using the admixture model, with the option of correlated allele frequencies. Additionally, we investigated each area for evidence of genetic bottlenecking (*i.e.* recent reduction of effective population size) using microsatellite loci and the computer program BOTTLENECK (Cornuet and Luikart 1996; Piry *et al*. 1999). In a recently bottlenecked population, the allele number at polymorphic loci is reduced faster than the gene diversity and, consequently, the observed gene diversity is higher than the expected equilibrium gene diversity (under the assumption of a constant-size population) (Luikart *et al*. 1998). A one-tailed Wilcoxon sign-rank test was carried out to determine whether each sample exhibited a significant number of loci with higher gene diversity than expected in populations in equilibrium; the distribution of gene diversity expected at equilibrium was estimated based on 10,000 simulations. For this test, the two-phase model of mutation (TPM; Di Rienzo *et al*. 1994) was chosen as it was more suitable for microsatellite loci (Piry *et al*. 1999). Further, the

parameters for TPM were set to 95% single-step mutations and 5% multiple-step mutations, with a variance among multiple steps set to 12, as suggested by Piry *et al*. (1999).

Using the mtDNA control region data, Fu's Fs neutrality test (Fu 1997) and mismatch distribution tests (Schneider and Excoffier 1999) were carried out to detect departures from population equilibrium (*e.g.* sudden demographic expansion) using the computer program ARLEQUIN 3.11 (Excoffier *et al*. 2005). The distribution of the observed number of differences between pairs of haplotypes is usually multimodal in samples drawn from populations at demographic equilibrium, as it reflects the highly stochastic shape of gene trees. However, it is usually unimodal in populations having passed through a recent demographic expansion (Rogers & Harpending 1992; Slatkin and Hudson 1991) or through a range expansion with high levels of migration between neighbouring demes (Ray *et al*., 2003, Excoffier, 2004). The sum of the squared differences (SSD) was used as a statistical test to accept/reject the hypothesis of sudden expansion, as described in Schneider and Excoffier (1999).

RESULTS AND DISCUSSION

In the present study, a total of 152 common dolphins from four geographic areas (Ireland & Celtic Sea, English Channel, France and Portugal) were genotyped at 20 microsatellite loci. Additionally, a subset of this sample (106 individuals) from three geographic areas (Ireland & Celtic Sea, English Channel and Portugal) was also genotyped at 5 extra microsatellite loci, raising the number of loci to 25 (Table 1). All sampled areas showed comparable levels of nuclear genetic diversity (Table 2), which were in the range of previously published studies (Natoli *et al*., 2006, Mirimin *et al*., 2009). No linkage between any pair of loci was detected, indicating that alleles segregate independently. Furthermore, no deviation from Hardy-Weinberg expected proportions was detected within each sampled area, suggesting random mating.

Sequences of a 556bp portion of the mtDNA control region were obtained from a total of 107 individuals from the Irish coast, Celtic Sea, French coast and Portuguese coast (Table 1). Overall, 48 polymorphic sites were identified (including 44 transitions, 6 transversions and 3 indels). Number of haplotypes, haplotype diversity and nucleotide diversity for each sampled area are shown in Table 2.

Analysis of molecular variance (AMOVA) and estimated fixation indexes indicated no significant genetic structure among all samples (*i.e*. most genetic variability resided within rather than between samples). This lack of genetic structure was observed using both microsatellite and mtDNA control region markers, and for all estimators calculated (Table 3). Similarly, using the Bayesian approach (STRUCTURE), the highest likelihood value was obtained for K=1 (K=1: -12,228.8, K=2: -12,252.5, K=3: -12,598.7, K=4: -12,682.4), which indicates that individuals from the sampled areas may belong to the same genetic stock (using genotypes from 20 microsatellite loci). Increasing the number of microsatellite loci to 25 (although not including the French sample) did not alter the result, with highest likelihood value still obtained for K=1 (K=1: -9,997.8, K=2: -10,004.6, K=3: -10,031.4, K=4: -10,023.7). Furthermore, no significant genetic differentiation was detected when the sexes were analysed separately (Table 3), suggesting that both sexes may contribute equally to gene flow and dispersal in the sample areas.

Results presented here are in agreement with previous genetic studies on common dolphins from North-east Atlantic areas (Natoli *et al*. 2006; Viricel *et al*. 2006; Amaral *et al*. 2007; Mirimin *et al*. 2009). In previous genetic studies, some differentiation has been reported between the northern and southern distribution range limits of this region (i.e. Scotland and Portugal). When common dolphins form these areas were analysed, differences in gene frequencies were found using microsatellite loci (Natoli *et al*. 2006) and also using mtDNA cyt*b* sequence data (Amaral *et al*. 2007), but both studies did not find significant differences among these areas using mtDNA control region data, as confirmed in the present study. Overall, Natoli *et al*. (2006) reported no difference in dispersal between sexes, and concluded that the low or non-significant levels of differentiation encountered in the North-east Atlantic are the result of strong gene flow. In contrast, Amaral *et al*. (2007) suggested sex-biased dispersal since levels of mtDNA differentiation were stronger among females than among males, although using limited samples sizes when geographical location and sex were taken into consideration. Furthermore, the present study also support findings from Viricel *et al*. (2006) investigation of dolphins stranded along the French coast of Brittany and the Bay of Biscay, using both mtDNA control region and eight microsatellite loci. Similar results were obtained in a study that included stranded dolphins from Irish coasts and also bycatch samples from the western English Channel and Celtic Sea, further indicating the lack of genetic structure in these areas.

Results using genetic analyses from the present study are in contrast with previous non-genetic studies (such as cranial morphology, contaminant profiles and stable isotopes signatures), where some evidence of population segregation was found among common dolphin aggregations throughout the North-east Atlantic (ranging from Scotland to Portugal) (Das *et al*. 2003; Lahaye *et al*. 2005; Murphy *et al*. 2006). In contrast to the current study, analysis of cranial morphometrics by Murphy *et al*. (2006) did reveal some evidence of population differention within the Northeast Atlantic, with segregation of female Portuguese common dolphins from more northerly sampled animals. Natoli *et al*. (2006) reported that habitat choice may be an important mechanism leading to population structure in common dolphins. Therefore, the inconsistent results from morphometric and the current genetic study could suggest that variations in morphological features (caused by adaptation to different habitats) may occur more rapidly than in genetic markers at the population level. In fact, low levels of genetic differentiation between populations may be maintained for many generations by recurrent gene flow or recent population subdivision (Waples, 1998), whereas morphological characters may change more rapidly under different environmental conditions (*e.g*. Kinnison *et al*. 1998). A separate Mediterranean Sea population has been proposed, with directional movement of females from the western Mediterranean Sea into the Northeast Atlantic Ocean (Natoli *et al*. 2008). This study corroborates the results of the cranial morphometric study, where it was suggested that female Portuguese common dolphins may not interbreed with common dolphins from other areas in the NE Atlantic, and/or common dolphins off the Portuguese coast are mixing with common dolphins in the Mediterranean Sea, and/or common dolphins inhabitating waters further south of the sampled region (Murphy *et al.* 2006).

The genetic bottleneck tests revealed that the number of loci showing excess heterozygosity was not significantly larger than expected in populations at equilibrium, indicating no evidence of recent reduction of effective population size in any of the sampled areas (Table 2). However, it must be noted that samples used in the present study were collected during the period when bycatch levels were at their peak, and not enough time has elapsed in order to reveal any signature in levels of genetic diversity. Therefore, it is difficult to predict whether human-related mortality will cause any long-term effect on demographic patterns of common dolphins from the North-east Atlantic. Bottlenecking events affect levels of genetic diversity and such a signal can be traced within a number of generations (less than $4N_e$), and thus, data presented here may serve as reference data for future studies monitoring patterns of genetic diversity.

For mtDNA sequence data, Fu's test of selective neutrality yielded significantly negative Fs values for all sampled areas (with the exception of Scotland) (Table 2). Further, the observed distribution of the number of differences between pairs of haplotypes (mismatch distribution) did not differ significantly from a unimodal distribution, as expected after a sudden historical population expansion (*i.e*. non-significant SSD values); for each sampled area and the whole dataset (Table 2). Large negative Fs values and unimodal mismatch distributions suggest that the population of common dolphins identified in the North-east Atlantic may have undergone a demographic expansion, which could have occurred following colonisation of the studied areas after the last glaciations, which ended some 10,000 years ago (Dynesius & Jansson, 2000). Simulation studies examining the effect of range/spatial expansion on the pattern of molecular diversity, under a coalescent approach, revealed that a range expansion with large levels of gene flow may lead to unimodal mismatch distributions, and that neutrality tests (*e.g*. Fu's Fs) will show very significant negative values after spatial expansion and a high number of migrants (Ray *et al*. 2003). Interestingly, the Scottish sample showed a unimodal distribution but not a significant negative Fs value, which suggests that its marginal position in the distributional range may have led to smaller exchange rates of migrants to neighbouring aggregations. In contrast, the same was not observed in the Portuguese sample, which may mix with southern (*i.e.* Eastern Central Atlantic), eastern (*i.e.* Mediterranean Sea) or more oceanic populations.

CONCLUSIONS

Findings from the present study revealed that common dolphins from the North-east Atlantic show high levels of genetic variability, which do not appear to have been affected by strong recent demographic changes, such as a reduction of population size due to high incidental mortality rates. Population structure analyses indicated that common dolphins found in the western English Channel and off the Atlantic coasts of Ireland, France and Portugal are part of the same population. These findings suggest the presence of a large "coastal" or neritic panmictic (random mating) population in the North-east Atlantic, maintained by strong gene flow (maintaining homogeneous gene frequencies throughout the sampled areas), though, the possibility of a recent population split cannot be ruled out - where not enough time has passed to generate significant genetic differentiation. Due to a lack of data from offshore waters in the Northeast Atlantic, we could not assess the possibility of some population differentiation between non-sampled oceanic (offshore) and neritic habitats. Furthermore, a lack of genetic structure in continental shelf and slope waters does not mean that ecological (contemporary) differences

should be ignored, as evidenced by non-genetic studies. Although the present study included one of the largest dataset ever analysed for this species, the high levels of genetic variability found at both nuclear and mtDNA control region markers may suggest that larger sample sizes are required to obtain more realistic population-wide estimates of gene frequencies (Carvalho and Hauser, 1994). In addition, the opportunistic sampling methods used in the present study (bycatch and strandings) may have generated some sampling biases at both temporal (*e.g.* seasonal sampling) and spatial (*e.g.* coastal areas) scales. Thus, as more samples are collected each year, genetic structure should be re-assessed using a larger dataset and testing different classes of markers (e.g. Single Nucleotide Polymorphisms and chromosome Y sequences). For a better understanding of population structure of common dolphins from the North-east Atlantic, future studies should focus on distribution, abundance and seasonal movements, with particular reference to offshore, northern and southern limits of the North-east Atlantic.

Acknowledgements

A special thank goes to all who provided samples, including fisheries observers and stranding networks workers. In Ireland, samples were collected with help from the Irish Whale and Dolphin group, and grants awarded to ER from the Heritage Council, National Parks and Wildlife Service, INTERREG Ireland – Wales, two EU funded projects: FAIR-CT95-0523: Assessment and Reduction of the By-catch of Small Cetaceans of Small Cetaceans (BY-CARE) and EVK3-2000-00027: Bioaccumulation of persistent organic pollutants in small cetaceans in European waters: transport pathways and impact on reproduction (BIOCET). Tissue samples from Scotland and Portugal were kindly provided by Jennifer Learmonth (SAC – Scottish Agricultural College and DEFRA) and by Marina Sequeira (Instituto para a Conservação da Natureza e Biodiversidade), respectively. Tissue samples from France were kindly provided by the Centre de Recherche sur les Mammifères Marins. Genetic analysis was carried out with funding from EU NECESSITY project.

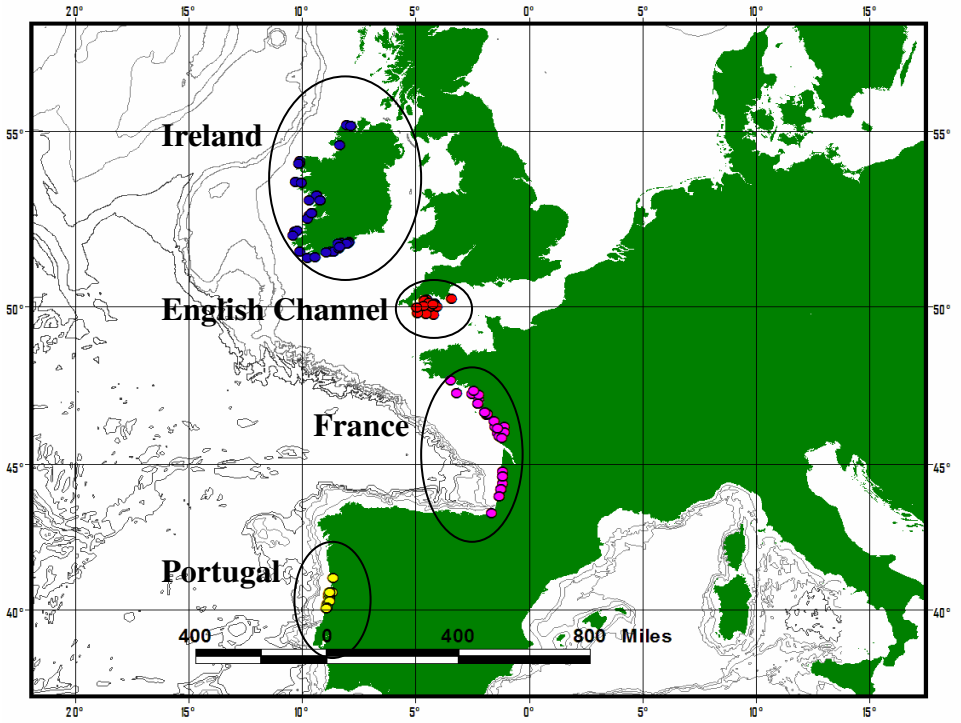

Figure 1a. Location of all individuals used in microsatellite loci analyses.

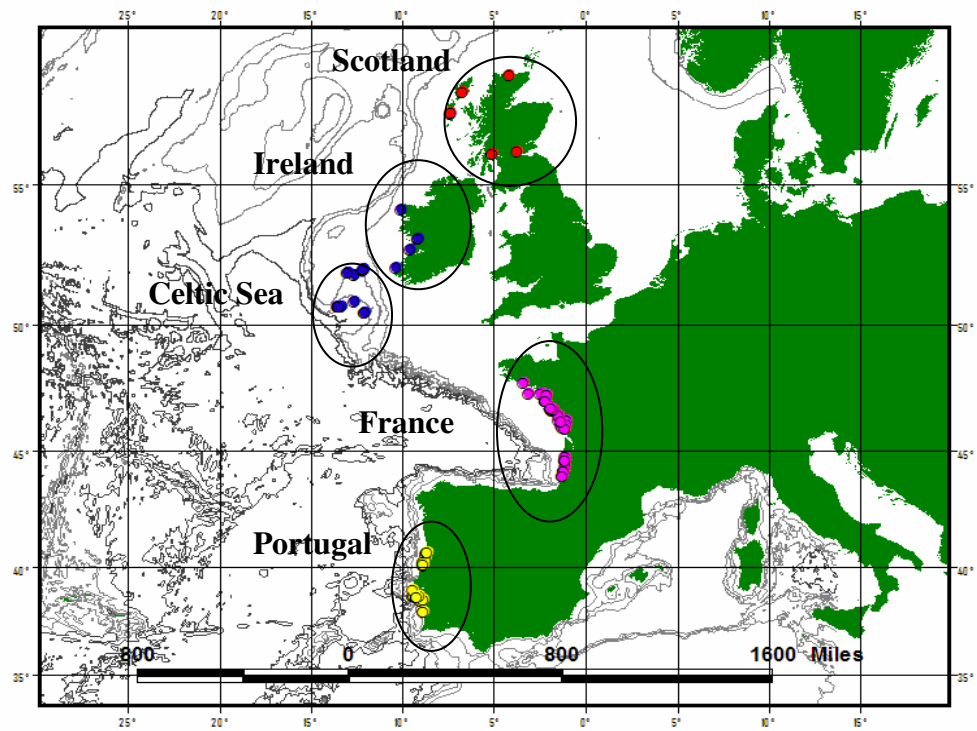

Figure 1b. Location of all individuals used in the mtDNA control region analyses.

Table 1. Number and period of collection of individuals used in the present study for 20 microsatellite loci, 25 microsatellite loci and 556bp of the mtDNA control region.

Summary	Date of collection	Microsatellite loci		mtDNA
		n (20 loci)	n (25 loci)	n (mtDNA)
Ireland	1991-2003	49	49	7
Celtic Sea	1996-1998	0	0	21
Portugal	1995-2005	21	21	25
English Channel	2001-2006	36	36	0
France	1995-2004	46	0	41
Scotland	2001-2003	0	0	13
Total		152	106	107

Table 2. Genetic variability statistics for 20 microsatellite loci, 25 microsatellite loci and 556bp of the mtDNA control region. n: number of individuals; N_A: mean number of alleles per locus; H_O: mean observed heterozygosity; H_E: mean expected heterozygosity; P (H excess): P-values for heterozygosity excess (one-tailed Wilcoxon) test under the TPM model; N_H: number of haplotypes; H: haplotype diversity; π: nucleotide diversity; Fs (P): Fu's test of selective neutrality with respective significance level (*: P<0.05, **: P<0.01, ***: P<0.001, ns: not significant); SSD (P): sum of squared deviation with respective significance level (*: P<0.05, **: P<0.01, ***: P<0.001, ns: not significant).

	All	Portugal	France	English Channel	Ireland/Celtic Sea	Scotland
20 Microsatellite loci						
n	152	21	46	36	49	-
N_A	13.9	9.6	11.4	10.7	11.9	-
H_O	0.818	0.837	0.812	0.794	0.830	-
H_E	0.823	0.821	0.827	0.824	0.822	-
P (H excess)	0.715	0.727	0.226	0.298	0.676	-
25 Microsatellite loci						
n	106	21	-	36	49	-
N_A	12.9	9.2	-	10.0	11.0	-
H_O	0.769	0.786	-	0.743	0.779	-
H_E	0.769	0.767	-	0.768	0.772	-
P (H excess)	0.890	0.802	-	0.532	0.755	-
mtDNA control region						
n	107	25	41	-	28	13
N_H	62	24	32	-	22	8
H	0.982 ± 0.004	0.997 ± 0.012	0.987 ± 0.009	-	0.973 ± 0.019	0.923 ± 0.050
π	0.015 ± 0.008	0.015 ± 0.008	0.015 ± 0.008	-	0.018 ± 0.009	0.012 ± 0.007
Fs (P)	-24.7 (***)	-17.6 (***)	-18.8 (***)	-	-7.8 (**)	-0.002 (ns)
SSD (P)	0.003 (ns)	0.004 (ns)	0.004 (ns)	-	0.007 (ns)	0.045 (ns)

Table 3. Measures of among-samples genetic structure for both microsatellite (20 or 25 loci) and mitochondrial DNA (556bp of the control region) markers. ns: not significant.

	Microsatellites (20 loci)
F_{ST}	-0.0009 (ns)
Males	-0.0011 (ns)
Females	-0.0015 (ns)
Rho_{ST}	-0.0017 (ns)
Males	-0.0062 (ns)
Females	0.0075 (ns)
	Microsatellites (25 loci)
F_{ST}	-0.0015 (ns)
Males	-0.0033 (ns)
Females	-0.0041 (ns)
Rho_{ST}	-0.0003 (ns)
Males	-0.0084 (ns)
Females	0.0003 (ns)
	mtDNA control region (556bp)
F_{ST}	-0.0011 (ns)
Males	-0.0013 (ns)
Females	0.0089 (ns)
Φ_{ST}	0.0036 (ns)
Males	0.0198 (ns)
Females	-0.0164 (ns)

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