Groups of related belugas (*Delphinapterus leucas*) travel together during their seasonal migrations in and around Hudson Bay.

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Social structure involving long-term associations with relatives should facilitate the learning of complex behaviours such as long-distance migration. In and around Hudson Bay (Canada), three stocks of beluga whales form a panmictic unit, but have different migratory behaviours associated with different summering areas. We analysed genetic variation at 13 microsatellite loci among 1524 belugas, to test hypotheses about social structure in belugas.

We found significant proportions of mother–offspring pairs throughout the migratory cycle, but average relatedness extended beyond close kinship only during migration. Average relatedness was significantly above random expectations for pairs caught at the same site but on different days or months of a year, suggesting that belugas maintain associations with a network of relatives during migration. Pairs involving a female (female–female or male–female) were on average more related than pairs of males, and males seemed to disperse from their matrilineal group to associate with other mature males. Altogether, our results indicate that relatives other than strictly parents, and especially females, play a role in maintaining a social structure that could facilitate the learning of migration routes. Cultural conservatism may limit contributions from nearby summer stocks to endangered stocks such as the Eastern Hudson Bay beluga.

1. Introduction

The maintenance of complex animal behaviours is likely favoured when individuals have the opportunity to gain information from conspecifics. Social learning has been documented for song structure in songbirds [1], foraging tactics in bottlenose dolphins [2], tool manufacture in New Caledonian crows [3] and migratory behaviour in light-bellied Brent geese [4]. Extended associations between offspring and parents (i.e. associations extending beyond periods of parental care), as well as social networks consisting of kin may be particularly important when juveniles learn complex behaviour, such as tool manufacturing [5] or long-distance migratory routes [4,6,7].

Extended associations between parent and offspring, as well as kinship in general, can vary considerably among groups of cetaceans. In baleen whales (Mysticeti), low levels of relatedness (beyond mother–calf) have been documented (e.g. humpback whales, *Megaptera novaeangliae* [8,9]; minke whales, *Balaenoptera acutorostrata* [10]). In toothed whales (Odontoceti), some long-term associations among close kin have been documented, but exceptions exist [11]. For example, both male and female resident killer whales (*Orcinus Orca*) are philopatric to their matrilineal group [12,13]. Likewise, significant associations among kin have been found among females in striped dolphins.
Transmit killer whales, as well as some sperm whales (Physeter macrocephalus), exhibit matrilineal philopatry, however, older males eventually disperse and become solitary [16–18]. Some populations of bottlenose dolphins show weak associations among kin [19], and on the other end of the continuum, some populations of dolphins [20], as well as sperm whales [21], show no significant associations with kin.

The beluga whale, Delphinapterus leucas, is a circumpolar, highly mobile-toothed whale. Sixteen management stocks are distinguished worldwide by the area where belugas home in the summer, along with behavioural, morphometric and genetic characteristics [22,23]. Three stocks are found in and around Hudson Bay (Canada) that are named by their specific and distinct summering areas, i.e. the Cumberland Sound, the western Hudson Bay and the eastern Hudson Bay (EHB) stocks (figure 1). The EHB stock is small (approx. 3300 individuals in 2009 [24]) and is listed as endangered in Canada [23]. In autumn and spring, each stock migrates along distinct routes to and from their specific summering areas [25–27], but they probably overlap in and around Hudson Strait (figure 1). In the winter, these stocks are found in Hudson Strait, Ungava Bay, the Labrador Sea and southwest Davis Strait, where their distributions are largely unknown [22,28–30] (figure 1). Mating is thought to occur during winter or early spring, and calving probably occurs before or just after arrival in the summering areas [28]. During the summer, belugas are often found in shallow estuarine waters, where they find food resources [31,32] and possibly escape predation by killer whales [32,33]. All three stocks are subject to subsistence harvest by Inuit communities along migration routes and in summering areas. The harvest of the endangered EHB stock has been under quota regulation since 1986 [23].

Previous work has shown that for both males and females, there is clear mtDNA differentiation, but no nuclear genetic differentiation among these three beluga stocks [27,34]. Such patterns are likely due to matrilineages showing fidelity to summering areas, with gene flow occurring on
common wintering areas [27,35]. Processes by which site fidelity and distinct migration routes are maintained are undocument ed. Fidelity to the natal site (or the first summering area) may be acquired while a juvenile depends on its mother. Indeed, weaning occurs after nearly two years, such that juveniles may stay with their mother for two or three migration cycles. Long-lasting associations with conspecifics, and potentially relatives, could also contribute to the maintenance of distinct migratory routes. Belugas are seen in aggregations of several individuals near estuaries during summer as well as during migration [22,23,36]. Also, pod composition is not well studied, but they can consist of mature females with several young individuals of both sexes [37–39].

Here, we analyse nuclear genetic variation among belugas to test hypotheses about the social structure along migratory pathways and in summering areas. If juveniles learn routes from their mother and disperse away from family upon weaning, relatedness should not extend beyond mother and (young) offspring found in close proximity throughout the migratory cycle. Alternatively, but not exclusively, if social association with kin contributes to the maintenance of migration routes, extended relatedness should be apparent during migration. In addition, we test whether sexes differ in contributing to kin structure. If so, the sex composition of a pair would influence the degree of relatedness. Finally, we examine whether sexes tend to aggregate separately, in which case the sex composition of samples from a social group would more likely be homogeneous i.e. made up of a single sex, either male or female.

2. Material and methods

(a) Sample collection and genetic analyses

A total of 1524 samples were obtained between 1982 and 2004 from 46 Inuit communities engaged in subsistence harvesting across Hudson Bay, Hudson Strait and southern Baffin Island (figure 1; see the electronic supplementary material for a description of the general methods employed for this legal harvest). Sampling sites are located along belugas’ migratory pathways, as well as in summer areas. We analysed multilocus genotypes at 13 microsatellite loci as reported by Turgeon et al. [27]. This study indicated that there is no evidence of genetic structure within and among stocks at these markers. Polymorphism level was high with a mean of 15.7 alleles per locus (range 9–27), and a mean heterozygosity of 0.678. The sex of 95 per cent of the individuals was determined with a molecular sexing technique. We classified individuals based on spatio-temporal and sex criteria. First, individuals sampled in the summering areas (northern Quebec) according to Stewart et al. [41] for 471 animals were random re-pairing of real pairs of belugas (genetic simulations were also used to test whether these statistics differed among pairs with different sex compositions. A detailed description of each permutation procedure is available in the electronic supplementary material).

(b) Relatedness analyses

We classified individuals based on spatio-temporal and sex criteria. First, individuals sampled in the summering areas (493 samples) were analysed separately from those sampled during migration (1031 samples). Second, we defined four categories comprising pairs of individuals captured at the same site, but on different temporal scales, i.e. on the same day (day), the same month (month), the same year (year) or at the same site but on different years (site). Finally, we also classified pairs of individuals by the sex of pair members: all pairs (all), female–female pairs (ff), male–female pairs (mf), and male–male pairs (mm). This classification resulted in a large number of pairs in each of 16 spatio-temporal and sex-based classes. Those numbers, computed on the basis of the empirical data available from the 1524 sampled specimens, are provided in the electronic supplementary material, table S1. Therefore, the number of pairs involved in each permutation test (see below) may be found under the appropriate class identification in the electronic supplementary material, table S1.

We wished to evaluate whether parent and offspring remain in close association and if kinship of pairs extends beyond the parent–offspring relationship. To do so, we first identified pairs of close kin (including parent–offspring), and then tested whether such pairs were found in proportions exceeding random expectations in each class. Then, we estimated the mean relatedness within each class while including or excluding close-kin pairs. Excluding close-kin pairs allowed estimating relatedness beyond parent–offspring relationship. Again, we tested whether mean relatedness was above random expectations in each class.

Our identification of closely related individuals was based on the number of allelic mismatches (AM) between two individuals. An allelic mismatch occurs when members of a pair do not share an allele at a given locus. Simulations show that pairs of individuals with zero allelic mismatches (AM0) are mostly parent–offspring pairs, whereas pairs with zero or one allelic mismatch (AM1) are either parent–offspring or half-sibling pairs (close kin), and very rarely unrelated (see electronic supplementary material). We validated the use of the AM statistic by comparing it to results obtained by KINGROUP [42] (see the electronic supplementary material).

Average pairwise relatedness (Rxy) was based on fM, the coancestry relatedness index between two individuals [43–46]. This statistic is robust [46]. As with most estimators, the relatedness index fM differs substantially from the pedigree relatedness but this difference is unrelated to accuracy [46]. Rxy is the average of fM values computed over a collection of pairs of individuals (see electronic supplementary material for an example).

Because both proportions of AM0 and AM1, as well as Rxy values, are based on pairwise statistics, a lack of independence between observations prevented us from using parametric tests. Instead, p-values were computed by running permutation procedures to evaluate whether observed values were above random expectations. First, a distribution of artificial values was obtained by random re-pairing of real pairs of belugas (genotypes). Then, the p-value of the observed values (AM0, AM1, or Rxy) was estimated based on the constructed distribution. Permutations were also used to test whether these statistics differed among pairs with different sex compositions. A detailed description of each permutation procedure is available in the electronic supplementary material.

(c) Sex composition of captures

We determined if captures consisting exclusively of one sex occurred more often than expected by chance. To do this, we first classified individuals harvested at the same site on the same day (i.e. individuals with the highest probability of being in the same pod, see the electronic supplementary material on hunting technique) into ‘male-only’, ‘male–female’ and ‘female-only’ captures. We then estimated (see the electronic supplementary material for an example).
3. Results

(a) Relatedness patterns during migration and in summering areas

When belugas were sampled during migration, the proportion of parent–offspring pairs ($AM_0$) and close kin ($AM_{01}$) was significantly higher than expected by chance for the day category (figure 2a,b). This was true for all sex categories except for parent–offspring pairs involving two males (figure 2b), which comprised a significantly lower proportion of related pairs ($p = 0.04$). For all other spatio-temporal categories, the proportions of $AM_0$ and $AM_{01}$ did not differ significantly from random expectations. In the summering areas, there was a significant proportion of parent–offspring for pairs involving a female ($ff$ and $mf$), for the day category only (figure 2c). The proportion for $ff$ pairs was significantly higher than for the other sex classes ($p = 0.03$). The proportions of close-kin pairs were never significant in the summering areas (figure 2d).

Patterns of mean relatedness ($R_{xy}$) during migration were also different from those in summering areas (figure 3). For belugas sampled during migration, mean relatedness was significantly higher than expected by chance for the categories day, month and year, but never for site (figure 3a). This was true for all sex classes, with the exception of pairs of males for the year category. The exclusion of parent–offspring and close-kin pairs did not affect the significance of $R_{xy}$ estimates (see the electronic supplementary material). Pairs of males caught on the same day were less related, on average, than were mixed-sex pairs ($p = 0.04$). Differences in $R_{xy}$ values among sex categories were more apparent for year, where average relatedness of pairs involving a female ($ff$ and $mf$) was significantly higher than for pairs of males ($p = 0.03$ and $p = 0.04$, respectively). It is worth noting that $R_{xy}$ values decreased in categories involving wider temporal windows, i.e. from day to month to year and site (figure 3a). This was true for all sex classes except $ff$, which slightly increased from month to year.

In contrast, mean relatedness for belugas sampled in the summering areas was largely non-significant (figure 3b). For the day category, pairs of females were significantly related, and more so than pairs of males ($p = 0.03$), but this result vanished when parent–offspring and close-kin pairs were excluded (see electronic supplementary online material). Otherwise, these results hold whether or not closest kin were excluded (see also the electronic supplementary material).

It must be noted that given the large panmictic beluga population (more than 50,000 individuals, [23]), we must expect that in each class, most pairs will involve unrelated individuals. Therefore, a relatively small proportion of more closely related pairs will usually be sufficient for an observed value of $R_{xy}$ to be significantly different from random expectation. This means that the differences between significant $R_{xy}$ and non-significant $R_{xy}$ values may seem small even at very high significance levels.
well as in the summering areas. For pairs which we had data on sex, there was always a female involved. That parents, and especially females, travel and summer with very young offspring is not surprising. Given that weaning occurs after at least 2 years [38,48,49], female belugas necessarily remain with their calves for one or more migratory cycles. However, only 1.5 per cent of the 471 individuals of known age were possibly of pre-weaning ages (less than or equal to 3 years), indicating that parent–offspring pairs most probably consisted of older offspring sampled with their mother. This is congruent with reports of females sometimes being accompanied by several juvenile individuals of different ages [37], which are likely half-siblings.

Our results also indicate that relatedness extends beyond the mother–offspring relationship between belugas travelling together. Indeed, a significant proportion of pairs of close kin, most probably half-siblings, were caught together, but only during migration. For migrating animals, the significant average relatedness ($R_{xy}$) for the day category is probably boosted by parent–offspring pairs (especially female–offspring, see below), but mean relatedness remained significant when the closest kin categories were excluded. Therefore, during the migratory period, belugas also travel with relatives other than their parents and siblings. Social structures in which groups of related individuals remain in close association over extended periods of time has been reported for several other toothed whales [11,12,50].

Our results also strongly suggest that belugas form networks of related individuals which aggregate in space and time during the migratory period. This is supported by significant $R_{xy}$ for the month and year categories (figure 3a). That is, when individuals are sampled at a given site along migration routes, other individuals sampled at the same site but after some period of time (in the same month or in the same year) are, on average, significantly related. Such extended spatio-temporal associations between related individuals could indicate that relatives using the same migration route are temporally separated by several days to several months. Although not tested statistically, average relatedness tended to decrease as temporal classes became larger (e.g. from day to month). Thus, for a network of relatives, individuals that are more closely related are probably more tightly aggregated in space and time.

In contrast, relatedness does not appear to extend beyond associations among females and their offspring when in summering areas. During this period, mean relatedness was significant for pairs involving a female sampled on the same day, but only if the closest kin categories were included, further indicating that mother and offspring remain together during summer.

There are two possible explanations for such strong differences in relatedness patterns between the summer and migratory periods. First, it is possible that upon arriving in summering areas, family groups (other than females and offspring) dissociate from each other, such that in the summering areas belugas tend to associate with non-relatives. This would require that family groups re-associate prior to departing for migration at the end of summer. Alternatively, several groups of relatives might intermingle in the same general area during the summer. This would increase the probability that Inuit hunters sample individuals from different groups of relatives, decreasing overall levels of relatedness. Although we feel the second explanation is more likely, we are currently unable to distinguish among these possibilities.
and female-only samples are disproportionately common summering areas [27]. In contrast, we find that male-only bites the same pattern of mtDNA differentiation among the distinct summering areas, because males and females exhibit a geographical area [59], but they may also result from social segregation of males and females within an area [38,60]. There is no evidence for male biases in dispersal away from a sex-based bias in outright dispersal away from a certain period of time, but eventually leave to associate primarily with other mature males, leading to differences in levels of relatedness among the sexes. The significant proportion of female-only captures that were neither particularly young nor old may then be viewed as a by-product of male segregation resulting in other groups being female-biased. Because the sampling of pods was probably never fully inclusive (not all animals of the groups were harvested or sampled for genetics), we cannot formally conclude to social segregation of the sexes in belugas. Nevertheless, this portrait of social structure is in agreement with previous field observations about the sex composition of beluga herds [61–63] in summering areas and during migration. It seems to resemble the social structure of transient killer whales [16]; however, in transient killer whales, older males become solitary, whereas in belugas, older males probably associate with other adult males. This interpretation matches telemetry data for Canadian High Arctic and Beaufort Sea beluga indicating that during summer, males travel back and forth between shallow and deeper waters, whereas female beluga, particularly those accompanied by calves, spend most of their time in estuarine embayments or closer to shore [61,64].

<table>
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<th>total no. individuals in capture</th>
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<td>14</td>
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<tr>
<td>total</td>
<td>128 45 25 16 9 2 1 3 1 1 1 231</td>
</tr>
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</table>

Finally, it is worth noting that we did not find evidence for fidelity to specific summer sites, as reported in some whales [51,52], including beluga [53]. If belugas showed site fidelity in the study area, we would have expected significant mean relatedness \((R_{xy})\) for the site category. This would be because from one year to the next, closely related individuals should be found at the same site. This result was never observed. Thus, relatives likely migrate together or along the same route, but they do not appear to show strict annual fidelity to specific migratory sites. Satellite transmitters deployed on EHB belugas at two estuaries (Nastapoka and Little Whale rivers) show that belugas in this area make frequent inshore–offshore movements with some movement between summer estuaries [54]. Thus, belugas probably show fidelity to general summering areas [27,34], as is the case for many other migratory cetaceans [39,55–58].

(b) Sex-biased social structuring

Sex-based differences in relatedness indicate that beluga social structure is strongly influenced by associations involving female relatives. These differences first begin with parent–offspring associations. Pairs of males were never identified as parent–offspring, indicating that mixed-sex parent–offspring pairs can be interpreted as consisting of mothers and sons. Pairs of males were also least often represented among close-kin pairs. Finally, even when pairs of males are found to display significant mean relatedness (figure 3a, day), mean relatedness was lower than when a male was matched with a female.

Sex-based differences in beluga relatedness patterns may result from a sex-based bias in outright dispersal away from a geographical area [59], but they may also result from social segregation of males and females within an area [38,60]. There is no evidence for male biases in dispersal away from the distinct summering areas, because males and females exhibit the same pattern of mtDNA differentiation among summering areas [27]. In contrast, we find that male-only and female-only samples are disproportionately common among captures, indicating that the sex composition tends to be more homogeneous than expected if sexes were distributed at random among those groups. This suggests that some aggregating/segregating mechanism based on sex may be operating. Moreover, males from male-only captures are older than males in mixed-sex samples. Thus, males may remain with their relatives for a certain period of time, but eventually leave to associate primarily with other mature males, leading to differences in levels of relatedness among the sexes. The significant proportion of female-only captures that were neither particularly young nor old may then be viewed as a by-product of male segregation resulting in other groups being female-biased. Because the sampling of pods was probably never fully inclusive (not all animals of the groups were harvested or sampled for genetics), we cannot formally conclude to social segregation of the sexes in belugas. Nevertheless, this portrait of social structure is in agreement with previous field observations about the sex composition of beluga herds [61–63] in summering areas and during migration. It seems to resemble the social structure of transient killer whales [16]; however, in transient killer whales, older males become solitary, whereas in belugas, older males probably associate with other adult males. This interpretation matches telemetry data for Canadian High Arctic and Beaufort Sea beluga indicating that during summer, males travel back and forth between shallow and deeper waters, whereas female beluga, particularly those accompanied by calves, spend most of their time in estuarine embayments or closer to shore [61,64].

(c) Maintenance of migration routes and conservation issues

Our results suggest that young belugas remain in close association with their mother, very probably beyond weaning. Some individuals, and especially females, remain faithful to their group of matrilineal relatives, establishing a network of related individuals. Relatedness patterns are most evident while belugas are travelling, as is expected if
associating with relatives is important to learn migratory routes. Males probably do not play as strong a role as females in maintaining networks of relatives, in part, because mature males seem to form separate associations apart from closely related females and offspring. These networks move along the migratory pathway to and from their summering and wintering areas. While networks are not faithful to particular sites, they are faithful to the distinct summering areas [27,34].

As such, young individuals have ample opportunity to learn the specific migratory route of their mother and close relatives. It has been suggested that the much longer nursing period of odontocetes when compared with mysticetes could be associated with a ‘sophisticated communicational–navigational training’ between mothers and calves [65]. Therefore, the migratory route may well be socially transmitted, with relatives other than strictly parents potentially playing a role in maintaining social structure and facilitating learning. It cannot be formally excluded that belugas first learn the migration route as young juveniles; being dependent upon their mother, they perform at least two complete migrations between the winter and summering areas before they are weaned. However, post-weaning parent–offspring associations during migration strongly support that travelling with close family is at stake. Yet again, learning migration routes may not be the sole, and perhaps not the main, benefit associated with extended kin associations during migration. Indeed, cryptic kin selection has been associated with sharing of resources while foraging and anti-predator behaviour [66]. For example, while migrating in open waters, individuals, and in particular calves, may be better protected if they swim in groups [67]. Group formation may also aid navigation in pack ice, as for bowhead whales which assess ice conditions from the echoes of other whales’ vocalizations [68]. Additional unknown factors promoting kin associations during migration may also be at work.

If indeed migratory routes are socially transmitted, this may have important conservation and management implications. Indeed, it could impede the re-colonization of extirpated summering areas, and limit the dispersal between stocks using different migration routes. Thus, contributions to the endangered EHB stock from neighbouring stocks may be limited. Unfortunately, historical observations attest to this scenario. For example, extensive hunting in the nineteenth and twentieth centuries practically extirpated belugas from Mucaluc River (Ungava Bay), Great Whale River and Nowalapik River (EHB) and probably also the Manicouagan Bank (St Lawrence River beluga population). Some 150 years later, these areas have never been fully re-colonized [69–71]. Such migratory conservatism is also seen in southern right whales, where maternally directed site fidelity limits populations to two distinct feeding areas despite the availability of alternative feeding areas with abundant resources [72]. For belugas, the primary function of reaching shallow estuarine embayments is not fully understood. In delphinids, food distribution and predator avoidance are the main driving factors of sociogenetic structure and kin association [73]. Both factors are likely involved in explaining why belugas swim back to shallow estuarine embayments every summer [31–33]. As for many other taxa, fidelity to a route leading to predictable and favourable environment may be adaptive [74]. At times, it can also limit the potential for range extension and re-colonization. Overall, our findings, therefore, encourage continued monitoring and managing belugas on the basis of summering stocks to recommend quotas supporting subsistence hunting by Inuit communities [75].

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References


31. de March BGE, Postma LD. 2003 Molecular genetic stock discrimination of belugas (*Delphinapterus leucas*) hunted in eastern Hudson Bay, northern Quebec, Hudson Strait, and Sanikiluaq (Belcher Islands), Canada, and comparisons to adjacent populations. *Arctic* 56, 111 – 124.


37. Stewart REA. 1998 Estimation of relatedness between individuals in general populations with a focus on their use in conservation programs. *Genetics* 173, 483 – 496. (doi:10.1534/genetics.105.049940)


75. Doniol-Valcroze T, Hammill MO, Lesage V. 2010 Information on abundance and harvest of eastern Hudson Bay beluga (Delphinapterus leucas). Canadian Science Advisory Secretariat, Fisheries and Oceans Canada, Research document 2010/121, Canada.