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Hybrids Between Gray Seals (*Halichoerus grypus*) and Spotted Seals (*Phoca largha*): A Case of Xeno-Breeding Preference in Pinnipeds

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Abstract

Generally, a species is defined as an independent unit that is reproductively isolated from others. However, deviations from this definition are not uncommon. For example, cross-fertilization in pinnipeds has been reported among at least 10 species. Herein, we describe an unexpected hybridization that occurred between female gray seals (*Halichoerus grypus*) and male spotted seals (*Phoca largha*) in the presence of adult male gray seals in a mixed-species seal population under human care. To our knowledge, these are the first cases of gray and spotted seal hybrids ever reported. The three hybridized offspring had the appearance of a gray seal body and a spotted seal head. Microsatellite DNA markers were employed as genetic evidence to further support the hybridization events. Our study suggests a natural preference for interspecies hybridization between female gray seals and male spotted seals in human care. Following that, potential causes of cross-species hybridization, including female preference, male competition, and some other factors, are discussed.

Key Words: human care, gray seal, *Halichoerus grypus*, hybridization, spotted seal, *Phoca largha*

Introduction

Conventional view dictates that accumulated genetic variation is a barrier to separate two different species, otherwise known as the "isolation mechanism" (Mayr, 1963). Isolation mechanisms prevent the exchange of genetic information between different species by genetic incompatibility after reproduction, non-viability of zygotes, or the sterility of hybrid offspring (Mayr, 1963; Pascarella, 2007), in addition to the incompatibility of two species' reproductive organs and differences in the mating cycles of two species (Barnard et al., 2017). Though it is common in other vertebrates, such as fish and birds (Grant & Grant, 1992; Scribner et al., 2000), interspecies hybridization is extremely uncommon in mammals due to their higher ratio of regulatory evolution to protein evolution and the rapid rate of change in chromosome number (Wilson et al., 1974).

Cetaceans, however, are an exception. An exceedingly high level of consistency can be seen in the karyotypes of cetaceans, and hybridization has been observed in roughly 20% of these species (Kingston & Gwilliam, 2007; Caballero & Baker, 2010; Crossman et al., 2016; Espada et al., 2019). For pinnipeds, karyotypes of 16 or 17 pairs of chromosomes in phocids and 18 in otariids likewise demonstrate extraordinarily high consistency within a family (Caballero & Baker, 2010); and at least 10 species of pinnipeds have been documented as having cross-fertilization (Brunner, 2002; Kovacs et al., 2006; Lancaster et al., 2007; Franco-Trecu et al., 2016; Savriama et al., 2018; Rohner et al., 2020; Sinclair et al., 2021; Lopes et al., 2023). Interspecific mating can either involve voluntary or coerced "practice" mating. Alternatively, it may occur due to varying forms of social interaction between species or as a result of one species exhibiting behavioral dominance over another (Vasey, 1995). These animals are often difficult to access and observe in the wild, and these hybridization events may leave no signs of fertile offspring; therefore, we hypothesize that hybridization occurs more often in marine mammals than what current data suggest. Hybridization in populations under human care is much easier to recognize. Apart from morphological evidence, various methods can be used to identify hybrids (Taylor et al., 2006). Vocal signature and genetic methods based on mtDNA genotype and nuclear genotype are all potential methods (Goldsworthy et al., 1999; Hindell, 2001; Page et al., 2010; Zhang, 2014; Zhang et al., 2016).

Gray seals (*Halichoerus grypus*) are affiliated with pinnipeds. Male gray seals are 2.25 m in length and weigh 300 to 350 kg. They are polygynous, but males do not defend territories or herd females (Jefferson et al., 2015). Partner fidelity and polygyny operate synchronously in this species, which means that female preference plays an important role during mating. Female gray seals tend to mate with the same individual, and male seals close to females approaching estrus are more likely to reproduce successfully (Amos et al., 1995; Twiss et al., 2006). Spotted seals (*Phoca largha*) are another kind of pinniped species. Adult male spotted seals are 1.61 to 1.7 m in length and weigh 85 to 110 kg. They are annually monogamous and territorial. They haul out with females and their pups during the nursing season and wait for the subsequent post-weaning mating (Jefferson et al., 2015).

In the present study, in a mix population of gray and spotted seals under human care in Qujiang Polar Ocean Park in Xi'an, China, the female gray seal was expected to mate with adult male gray seals, but interspecies hybrids always occurred. Utilizing genetic data in addition to morphological evidence, we investigated and documented the hybridization cases between gray and spotted seals. In addition, we further discussed the possible causes of the unusual phenomenon of interspecies breeding selection in gray seals rather than intraspecies breeding.

Methods

Qujiang Polar Ocean Park is a modern aquarium in Xi'an, China. There are six gray seals and six spotted seals kept in one enclosure for exhibition purposes. The enclosure consists of a 6 m × 150 m^2 pool with 500 m^3 of water and 50 m^2 of haul-out ground. Three seal pups were born in Qujiang Polar Ocean Park between 2018 and 2021. Because all three pups had a gray seal body and spotted seal head, we assumed that all three of them were hybrids (Figure 1); however, more evidence was warranted to back up our speculation because hybridization between gray and spotted seals had not yet been documented. The detailed information of all the seals kept in Qujiang Polar Ocean Park is presented in Table 1.

Microsatellite DNA markers were used to determine paternity. Since the three seal pups in the current study were all born under human care, we knew their dam was "Benben." To conduct the paternity study, we sampled 11 individuals: the three pups, the dam Benben, and the seven candidate male sires (see animal details in Table 1). For each individual, a 2 mL blood sample was collected by venipuncture from veins in the hind flippers and stored in an EDTA-K2 anticoagulant tube. All blood samples were stored at -20°C before DNA extraction. Genomic DNA was extracted using a Solarbio® Blood Genomic

Figure 1. External morphology and coloration of the seal pups born under human care: a gray seal (*Halichoerus grypus*) body and a spotted seal (*Phoca largha*) head. Photograph provided by the Qujiang Polar Ocean Park.

Table 1. Detailed information of the seals housed in Qujiang Polar Ocean Park

Species	Name	Gender	Age	Source
Spotted seal (*Phoca largha*)	Xiaowu	Male	11	Liaodong Bay, China
	Tangmu	Male	11	Liaodong Bay, China
	Lulu	Male	11	Liaodong Bay, China
	Chuangwei	Female	21	Liaodong Bay, China
	Kaisa	Female	21	Liaodong Bay, China
	Ali	Male	11	Liaodong Bay, China
Gray seal (*Halichoerus grypus*)	Nunu	Male	11	Republic of Latvia
	Xiaoliu	Female	11	Republic of Latvia
	Benben	Female	11	Republic of Latvia
	Dangdang	Female	11	Republic of Latvia
	Halin	Male	11	Republic of Latvia
	Xiaohei	Male	11	Republic of Latvia
Pups born in human care	Buding	Male	4	Born in human care
	Chunjuan	Male	1	Born in human care
	Tangyuan	Male	3	Born in human care

DNA Extraction Kit (Beijing, China) according to the manufacturer's instructions. To amplify the alleles, we employed 24 pairs of microsatellite DNA primers with high polymorphism and stable amplification (Table 2) (Gao et al., 2020). PCR reactions were conducted on an ABI VeritiTM 96 well machine in 25 µL reactions, containing 20 to 50 ng DNA, 10 mM Tris-HCl (pH 8.8), 50 mM KCl, 1.5 mM MgCl2, 200 µM dNTPs, 0.2 µM of each primer, and 1 U of Taq DNA polymerase with a final volume of 25 µL containing 1× PCR buffer. Touchdown PCR was used, and the cycling conditions were as follows: 95°C (5 min); 10 cycles of 94°C (30 s); 60 to 55°C (-0.5°C per cycle; 30 s); 72°C (30 s); 30 cycles of 94°C (30 s), 55°C (30 s), and 72°C (30 s); and a final extension of 72°C (10 min). Genotypes were examined using an ABI 3037XL sequenator (Thermo Fisher Scientific, Waltham, MA, USA). Paternities were verified manually by the direct exclusion method.

Results

The genotypes of all the seals surveyed are listed in Table 3. Of the 24 primers employed, four pairs of primers were workable on the direct determination of pup paternity, and the other 20 pairs were useless for direct paternity exclusion but are still presented with our results. Microsatellite genotyping analysis revealed that "Xiaowu" was the unequivocal sire of both "Buding" and "Tangyuan" as evidenced by the respective primer pairs PL68, PL7, and SSR11 (Table 4), marking the first two births. By utilizing the SSR10 primer pair, we were able to directly determine that "Tangmu" was the sire of "Chunjuan." As both sires were spotted seals, it logically follows that all three pups are indeed hybrids of gray and spotted seals (Table 4).

During the course of the current investigation, we have made interesting observations regarding the male competition that transpires between gray and spotted seals at the onset of the mating season. All the seals were in the same habitat. During the mating season, both male gray and spotted seals competitively pursued the female gray seals. Based on our preliminary results, it seemed that male gray seals were frequently bested in these competitions by their spotted counterparts. For instance, Benben, a female gray seal, was chased by a male spotted seal, leading to the birth of Chunjuan. However, not all mating activities involved interspecies coupling. We observed male gray seals mating with the female gray seal "Xiaoliu," who gave birth to a pup the next year. Unfortunately, the pup died shortly after birth due to drowning. It is regrettable that we were unable to determine the cause of the pup's death (whether it was accidental or related to fertility issues), nor

Table 2. Characterization of the 24 primer pairs employed in the present study (referred from Gao et al., 2020)

Primer pair	Size (bp)	Primer name	Primer sequence (5' to 3')	Repeat motif
PL7	280	Unigene_220913	CGAGTCCTCCCCTGTGTC TTCTCTCCTTTTCCCCCT	$(AACC)_6$
PL19	264	Unigene_03243	ACGCAAGCCTACTGAATG AGCACTGGTCCTCTGAAA	$(TA)_7$
PL31	224	Unigene_220914	AGTCCTGGATAACCAAACA GGACAGAGGTATTGAGGGT	$(GT)_7$
PL68	245	Unigene_23253	TGTTTGTTGAAAATCAGGATG CCTCTTACCCACTGCTTGT	$(TTC)_{21}$ $...(GA)_8$
SSR1	212	Unigene48751	TTCTTGGGAGGAAGAAGCAA AGCAGCTTCACTTCTAGGCG	$(GA)_6$
SSR2	336	Unigene2773	AGATTGCAGGTTCAGTTCCG CCATTTTCCCAACAGCATCT	$(CAA)_5$
SSR4	168	Unigene114136	GCTGAGATCTTTGCTTGCCT CGAAGGGAACTTTGAAGCAC	$(ACC)_5$
SSR5	121	Unigene114144	CGGGTTGCCTTTTCACTCTA AATTTCTTGGGAATGACCCC	$(TTG)_5$
SSR6	351	Unigene117519	TGGTAAAAGGTGGTCTTGCC GGCTCTGGTTTGCAGTTGTT	$(CGC)_5$
SSR7	338	Unigene99678	TGGGGAAAGCAAAAGGTATG CTGGGTTTGTCTGCACTGAG	$(GT)_6$
SSR8	196	Unigene98884	GCGTCCATCGATTTCTGTTT ACGTGACCTTGTTTTCTGGG	$(GT)_7$
SSR9	160	Unigene93881	AGGATGTCTGGGAGCCTCTT TTGACGCCCAATAGAAACCT	$(TA)_6$
SSR10	148	Unigene71588	TTGTGTCAGTGTTGAGGGTGA CCTGTAATGAAAACATTTTCCCC	$(GT)_7$
SSR11	131	Unigene115395	CCTGAGAAGATCCAAGTGAAGC GAGGACGAGGAGGAGGATG	$(CTC)_5$
SSR12	269	Unigene138597_gan_3	AGCTGCAGACGAAGTGGATT ATGGGACAAGAGAAAAGGGG	$(TTTG)_5$
SSR13	150	Unigene155319_gan_3	CCGGAGCCAAACATAGACTC CGTGGAAGGACCACATACCT	$(GGAT)_5$
SSR14	345	Unigene33747_gan_3	TCCCCAGAGACAACTCCATC CAATTGGCAACTTCTGCTCA	$(TAG)_7$
SSR15	158	Unigene39130_gan_3	ATTGAAGCCACGCAGAAACT AGGAGACACATTCCCATTGC	$(CAG)_5$
SSR16	100	Unigene39901_gan_3	TAACAAGAACCGAGAGCCCA CCCTGTGTGCAGATGCTTTA	$(CA)_6$
SSR17	184	Unigene44421_gan_3	TGCCAGCAATGAGACTGGTA TTCCTTGCAGTTTACTCTTCCA	$(AT)_6$
SSR18	154	Unigene50277_gan_3	CCGGAATTTCATGATTGGTC GTGCGTGTGCTTCCAGACT	$(GA)_6$
SSR19	141	Unigene56048_gan_3	TCAAGATGTTTGCTGAACGC GAGCAAGCAAAAAGAAACCG	$(AT)_{11}$
SSR20	102	Unigene63210_gan_3	TGCAAATACGTACACACCCA ACATGGGGGAAAAGCACATA	$(TA)_7$
SSR21	188	Unigene35031_gan_3	GGGAGTCCTGGGGGTTATTA GTGTGTGGGGGAGGAGAATA	$(TC)_7...(CT)_5$

Table 3. Microsatellite genotypes of seals used in the present study

Primer pair	Alleles (bp)																						Direct paternity determination
	Pups						Dam		Candidate sires														
	Buding		Chunjuan		Tangyuan		Benben		Ali		Lulu		Tangmu		Xiaowu		Halin		Nunu		Xiaohei		
PL7	273	273	273	277	273	277	273	273	273	277	273	273	273	277	273	277	273	273	273	273	273	273	Valid
PL68	233	254	236	257	236	257	233	236	218	257	248	257	248	257	257	254	230	233	233	233	233	236	Valid
SSR10	147	151	149	151	147	151	151	151	147	147	147	147	149	149	147	147	151	151	151	151	151	151	Valid
SSR11	129	129	129	129	129	151	129	154	129	129	129	151	129	129	129	151	129	129	129	154	129	129	Valid
PL19	262	284	260	284	262	284	262	284	260	284	260	284	260	284	257	284	262	284	262	284	262	284	Invalid
PL31	224	224	224	224	224	224	224	224	224	224	224	224	224	224	224	224	224	224	224	224	224	224	Invalid
SSR8	197	199	197	199	197	199	197	197	199	199	199	199	199	199	199	199	197	197	197	197	197	197	Invalid
SSR18	153	157	153	157	153	157	157	157	153	157	153	157	153	157	153	157	151	157	157	157	157	157	Invalid
SSR19	138	149	138	149	138	149	140	149	138	138	138	138	138	138	138	138	149	149	149	149	149	149	Invalid
SSR20	98	100	98	100	98	100	98	98	100	100	100	100	100	100	100	100	98	98	98	98	98	98	Invalid
SSR21	188	190	188	190	188	190	188	190	188	188	188	188	188	188	188	188	190	190	190	190	190	190	Invalid
SSR1	213	213	213	213	213	213	213	213	213	213	213	213	213	213	213	213	213	213	213	213	213	213	Invalid
SSR2	335	335	335	335	335	335	335	335	335	335	335	335	335	335	335	335	335	335	335	335	335	335	Invalid
SSR4	168	168	168	168	168	168	168	168	168	168	168	168	168	168	168	168	168	168	168	168	168	168	Invalid
SSR5	121	121	121	121	121	121	121	121	121	121	121	121	121	121	121	121	121	121	121	121	121	121	Invalid
SSR6	350	350	350	350	350	350	350	350	350	350	350	350	350	350	350	350	350	350	350	350	350	350	Invalid
SSR7	336	336	336	336	336	336	336	336	336	336	336	336	336	336	336	336	336	336	336	336	336	336	Invalid
SSR9	159	159	159	159	159	159	159	159	159	159	159	159	159	159	159	159	159	159	159	159	159	159	Invalid
SSR12	271	271	271	271	271	271	271	271	271	271	271	271	271	271	271	271	271	271	271	271	271	271	Invalid
SSR13	151	151	151	151	151	151	151	151	151	151	151	151	151	151	151	151	151	151	151	151	151	151	Invalid
SSR14	347	347	347	347	347	347	347	347	347	351	347	347	347	347	347	347	347	347	347	347	347	347	Invalid
SSR15	158	158	158	158	158	158	158	158	158	158	158	158	158	158	158	158	158	158	158	158	158	158	Invalid
SSR16	101	101	101	101	101	101	101	101	101	101	101	101	101	101	101	101	101	101	101	101	101	101	Invalid
SSR17	184	184	184	184	184	184	184	184	184	184	184	184	184	184	184	184	184	184	184	184	184	184	Invalid

Table 4. The result of paternity determination

Pup	Age	Dam (Gray seal)	Sire (Spotted seal)
Buding	4	Benben	Xiaowu
Chunjuan	1	Benben	Tangmu
Tangyuan	3	Benben	Xiaowu

Table 5. Birth information of seals in the Qujiang Polar Ocean Park

Species	Name	Delivery date (d/mo/y)	Result	Note
Gray seal	Xiaoliu	2/11/2016	Premature birth/dead	Chased by male gray seal during mating time
		29/11/2017	Premature birth/dead	Seen mating with gray seal
		14/2/2019	Eutocia/drowned	Chased by male gray seal during mating time
		7/12/2019	Premature birth/dead	
		17/12/2021	Premature birth/dead	Chased by male gray seal during mating time
	Benben	14/2/2018	Gave birth to Buding	Seen mating with spotted seal
		20/2/2019	Gave birth to Tangyuan	
		3/2/2021	Gave birth to Chunjuan	Chased by male spotted seal during mating time
Spotted seal	Chuangwei	24/1/2017	Eutocia/drowned	
		8/2/2019	Eutocia/drowned	

could we establish the identity of its sire. Thus, the incident did not yield any valuable information for evaluating the preference for xeno-breeding (Table 5).

Discussion

Husbandry and Breeding Background in the Present Study

The seals in the present study were all introduced from the wild or other aquariums to Qujiang Polar Ocean Park in 2012. They were kept free-range in the same enclosure, and no artificial breeding program was implemented. Mating behaviors were observed when seals became sexually mature. Several gray and spotted seals became pregnant, but they all failed to give birth successfully (Table 5). It was not until 2018 that the first pup was successfully born and survived. Regarding our present study, a total of three pups were born and survived, and they were all hybrids as determined by our results. Thus, our study provides evidence that a combination between a female gray seal and a male spotted seal was more successful in producing viable offspring. This is, to our knowledge, the first report of hybridization between gray and spotted seals.

The Theory of Hybridization

Hybridization is common in pinnipeds, especially in fur seals, and up to a 30% proportion of hybrids were reported in the *Arctocephalus* spp. population from subantarctic Macquarie Island (Lancaster et al., 2007). Hybrids are normally only a small portion of total offspring in genetically distinct sympatric populations, with most offspring being of pure lineage. While in fur seals, female mate choice is more influenced by male phenotype than genotype, females have some capacity to discriminate between males both within and between species based on phenotypic traits and are more likely to mate within their species (Goldsworthy et al., 1999). Also, there is a mechanism for species recognition that acts as a barrier to hybridization (Kingston & Gwilliam, 2007) because hybrids more likely have low fitness and reduced reproductive success (Mayr, 1963; Lancaster et al., 2007). However, the findings of our study are contradictory to the above research studies.

Other views hold that introgressive hybridization occurs when closely related taxa overlap in distribution, and this is often associated with historically isolated populations that come into contact as a result of anthropogenic disturbance. In these situations, hybridization is likely to occur if reproductive barriers are absent or if species recognition mechanisms are consequently hindered. An example of this is the genetic introgression of the endangered red wolf (*Canis rufus*) with coyotes (*Canis latrans*) (see explanation in Kingston & Gwilliam, 2007). Gray seals inhabit the subarctic area of the North Atlantic (Jefferson et al., 2015), whereas spotted seals inhabit the north and west segments of the North Pacific Ocean (Allen & Angliss, 2015). They are completely geographically isolated, and there is almost no chance for them to hybridize in the wild. Hybridization resistance mechanisms between them may lose efficacy to minimize potential cost. Thus, because of the human-created population overlap, hybridization occurred between the two species. Also, seals are highly seasonal breeding species (Atkinson, 1997). Gray seal mating often takes place in late February or early March (Harting, 1898), and spotted seal mating typically takes place between January and mid-April (Jefferson et al., 2015). This synchronism also provides support for their hybridization. These are some of the potential reasons that may cause successful hybridization between gray and spotted seals.

How Xeno-Breeding Overcame Conspecific Breeding

Xeno-breeding refers to the act of individuals mating with members of a different species of the opposite sex. Male gray seals lost when battling with male spotted seals in the present study which may be what prevented pure lineage offspring. For instance, male spotted seals were observed more eager to mate than male gray seals during mating season in the present study. In pinnipeds, male competitiveness, which is frequently correlated with body size, is regarded to be the main factor that influences successful mating (Haley et al., 1994; Lidgard et al., 2005; Thünken et al., 2011; Crocker et al., 2012). As was likewise the case for the male seals in the present study, gray seals (300 to 350 kg) clearly had a size advantage over spotted seals (85 to 110 kg) (Jefferson et al., 2015). Domination based on body size cannot therefore account for the current scenario. Another possible explanation is that large nutritional stores can be advantageous in endurance competition in addition to the benefits of large size during the competition process (Judge & Brooks, 2001). The dietary reserves present at the start of the mating season regulate the amount spent on reproduction, which could impact mating success (Crocker et al., 2012). So, male spotted seals may have relatively more energy reserves than male gray seals and thus have a more competitive edge in a mating competition.

Another possible explanation of how xeno-breeding can overcome conspecific breeding is female preference. Mammalian breeding systems were once thought to be dominated by males competing with one another for the opportunity to mate with submissive females (Amos, 2007). The males of the largest species dominating beaches and compelling the females of the smallest species to conceive hybrid offspring are well-known examples of how males vying for passive females can result in successful mating (Goldsworthy et al., 1999). However, we now know that females also play a significant role in successful mating (i.e., females decide with whom to mate; Amos, 2007). Twiss et al. (2006) discovered that a subset of female gray seals residing in the North Rona exhibited a tendency to seek out sires beyond the local male population's home range. This noteworthy observation might be indicative of an underlying female preference that could theoretically be linked to mate fidelity. Spotted seals are smaller than gray seals, and a male spotted seal might not be able to mount a gray seal female against her will. In light of this, we propose a potential mate choice based on the willingness of the female gray seals.

Mate choice depends on a range of phenotypic traits, including proportionate variations in body and flipper shape, pelage colors and pattern, vocalizations, and behavior, etc. (Goldsworthy et al., 1999). For example, vocal frequency can have a significant impact on the choice of mates during mating and other mating-related activities in cetaceans (Crossman et al., 2016). It has been established that mate preferences in other species (e.g., birds, terrestrial mammals, fish) are dependent, in part, on vocal behaviors (Miller, 1979; Robertson, 1996; Boughman & Wilkinson, 1998) and/or posture and facial traits (Ratciliffe & Grant, 1983; Gorb, 1998; Rowland, 1999). Scientists largely concur that vocal attraction is common in pinnipeds (Fitch et al., 2008). Sound transmissions are crucial to these species' reproductive communication. For instance, male gray seals actively compete for access to females using vocalizations (Jefferson et al., 2015). Spotted seals are typically silent but grow noisier during the breeding season as the males sing to attract females for mating (Zhang et al., 2016); therefore, it is possible that the female gray seal found the vocalizations of the spotted seals to be more attractive.

Alternatively, the interaction between gametes is also a potential reason that can affect successful fertilization. Sperm compete with one another to reach egg cells during fertilization. There is one exemplary case in mammals: the well-researched mouse t-haplotype. The t-haplotype is a genetic variation region of around 40 Mb that codes for a number of elements that lead to transmission ratio distortion, impairing sperm motility in heterozygous (t/+) males (Schimenti, 2000). Only t-sperm with the t/+ genotype exhibit a self-defense mechanism, expressing a dominant-negative protein kinase known as $SMOK^{TCR}$ and rescuing sperm motility specifically for t-sperm (Herrmann et al., 1999; Lyon, 2003). We can boldly hypothesize that because Benben, a female gray seal, mated with both male spotted seals and male gray seals, the spotted seal's genome may have contained unique gene pieces that altered gray seal's sperm activity simultaneously. Similar to the mouse t-haplotype sperm, sperm with these gene fragments can negatively impact other sperm (Amaral & Herrmann, 2021). These sperm are more adapt at binding to the egg cell to develop viable oosperm and express $SMOK^{TCR}$ that protects itself from harm. Similar gene pieces in spotted seals would significantly lessen the likelihood of gray seal sperm fusing with Benben's egg cell, creating hybrids.

Additionally, chemical communication between the female reproductive system and sperm allows for continued partner selection after mating (Fitzpatrick et al., 2020). In species that fertilize internally, females can continue cryptic female choice by manipulating the number of sperm or how well they swim by interacting with the female reproductive canal (Firman et al., 2017; Devigili et al., 2018). Mammalian sperm lack species specificity in response to chemoattractants, in contrast to marine invertebrates (Sun et al., 2003). For instance, Firman & Simmons (2015) discovered that eggs from house mice (*Mus domesticus*) were preferentially fertilized by sperm from less related males during in vitro fertilizations. They further suggested that these effects might be explained by either distinct chemoattractant reactions or direct interactions with gamete cell-surface proteins. The egg cell will emit chemical attractants that will hinder the travel of other sperm while helping certain sperm locate and unite with the egg cell more quickly (Ambs et al., 1999).

Individual Physical Functioning

In addition to the above-mentioned subjective factors like female preference and male competitiveness, a number of objective factors may also contribute to the failure of male gray seals. Sperm count has a substantial impact on the frequency of live births, and sperm counts below a particular threshold will result in male infertility (Bostofte et al., 1982). This suggests that the male spotted seals may have a higher volume of semen (or higher sperm count) than the competing gray seal males who did not father the offspring but did mate with the female gray seal. Alternatively, it is possible that candidate gray seals may not have produced sperm in normal quantities or may have struggled to complete intromission (Adler, 1969).

Conclusion

Our research may support a favored natural hybridization between female gray seals and male spotted seals when there were other adult male gray seals present in a mixed seal population under human care. As a result of competition with male spotted seals, male gray seals failed to produce offspring with female gray seals. To our knowledge, this is the first report of hybridization between gray seals and spotted seals, and this is also probably the first report of a cross-species breeding preference rather than conspecific breeding in mammals.

The present study discussed potential reasons for cross-species hybridization and its significance in advancing our understanding of evolutionary processes in pinnipeds, including reproductive isolation, female selection, and mating systems. Human activities have a significant impact on the environment, and it is widely acknowledged that alterations to environmental conditions, such as the melting of sea ice, may result in the mingling of species that were previously separated geographically, thereby increasing the probability of hybridization. It is noteworthy that if resulting hybrid offspring exhibit increased fitness, it can likely contribute to population growth. Against this backdrop, the findings of this study offer a unique opportunity to further probe the physiological sustainability of hybrid seals, which are unlikely to arise in natural settings, thus providing valuable insights into the biology of pinnipeds. It is essential to recognize, however, that the absence of well-designed studies aimed at monitoring the mating behaviors of captive seals impedes a more precise comprehension of the mechanisms of hybridization between gray and spotted seals. Empirical behavioral observations and genetic fitness studies must be pursued in-depth in order to explore and fully elucidate the intricacies of cross-species hybridization.

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