

Dorsal Skin Color Patterns Among Southern Right Whales (*Eubalaena australis*): Genetic Basis and Evolutionary Significance

C. M. Schaeff, P. B. Best, V. J. Rowntree, R. Payne, C. Jarvis, and V. A. Portway

Distribution and inheritance of dorsal skin color markings among two populations of southern right whales (*Eubalaena australis*) suggest that two genes influence dorsal skin color. The grey-morph and partial-grey-morph phenotypes (previously known as partial albino and grey-blaze, respectively) appear to be controlled by an X-linked gene, whereas the white blaze appears controlled by an autosomal gene (recessive phenotype). Calving intervals, calf size, and length of sighting history data suggest that partial-grey-morph, white-blaze, and black cows experience similar levels of reproductive success. Grey-morph cows (X^gX^g) are rare or absent in the two populations, but this was not unexpected given observed population frequencies of grey-morph males (X^gY) and partial-grey-morph females (X^gX^g). The proportion of partial-grey-morph calves produced by black cows (X^eX^e) suggests that the reproductive success of grey-morph males was equal to that of black males, however, larger sample sizes are required to determine whether grey-morph males tend to have shorter sighting histories. The reproductive success of white-blaze males appeared similar to that of black males among whales off Argentina. There were significantly fewer white-blaze calves than expected off South Africa, which could be due to white-blaze males experiencing reduced reproductive success or to sighting biases that result in white-marked calves being misidentified as black calves. The relative frequencies of both types of dorsal color markings varied between the South African and Argentinian right whale populations, suggesting limited nuclear gene flow between these populations; analyses using other nuclear markers are under way to confirm the extent of gene flow.

Five dorsal skin color patterns have been documented among southern right whales (*Eubalaena australis*): black, white-blaze, grey-blaze, grey-and-white-blaze, and partially albinistic (Best 1990; Payne et al. 1983) (Table 1; Figure 1). Partially albinistic right whales are white as calves but darken and become grey or brownish-grey as they age (Best 1990; Payne et al. 1983; Figure 2). These calves lack the pink eyes associated with true albinism and have a scattering of black pigmentation which is usually distributed in a narrow transverse band posterior to the blow holes that extends partway down the side in the shoulder region. The marks on grey-blaze animals are also white in calves but darken as the animals age.

A number of white pigmentation patterns are associated with pathological conditions including dominant white spotting (homologous to *c-kit* in pigs) and steele (homologous to *kit* ligand in mice; see Silvers 1979 for review), which result in lower fertility and anemia, piebald and

spotch mutations in mice, which result in reduced survival (Goulding et al. 1993; Hosoda et al. 1994), and Chediak-Higashi syndrome (homologous to beige in mice and Aleutian disease in mink; Jackson 1997), which is associated with an increased susceptibility to infection. It is unclear what, if any, effect partial albinism or the other right whale pigment patterns have on an individual's viability.

A number of southern right whale populations have been studied intensely over the past two decades, but investigations of right whale color patterns have been hampered by an inability to determine many animals' sex. Methods do exist for sexing right whales in the field (e.g., adult-calf associations or observations of sex-specific morphology such as a penis or genital slits), however, because of the opportunistic nature of the methodology, many animals (especially males) remain unsexed even after years of behavioral work (Schaeff et al. 1993). Fortunately, recent advances in molecular biology have

From the Biology Department, American University, 4400 Massachusetts Avenue NW, Washington, D.C. 20016 (Schaeff, Jarvis, and Portway), Mammal Research Institute, University of Pretoria, Pretoria, South Africa (Best), and Whale Conservation Institute, 191 Weston Rd., Lincoln, Massachusetts (Rowntree and Payne). The collection of the South African samples was made possible through the financial support of the Foundation for Research Development and Earthwatch and the participation of volunteers S. Edwards, D. Petrucco, M. Whiston, C. Maul, N. Schlocker, L. Boast, D. Hauenstein, M. Ketting, J. Marnoch, B. Honchorek, P. Pond, O. Weisshuber, S. Ruegg, E. Welsh, S. Margison, S. Santra, K. Babiak, S. Jacobs, R. Mobbs, H. Achenbach, E. Lock, S. MacDonald, E. Dickens, D. Schmitz, C. Whitfield, M. Berkstresser, F. Frischknecht, B. Schnaar, and D. Shull is much appreciated. Invaluable field assistance was also provided by D. Reeb, M. Sanson, D. Kemp, D. Calder, R. Cloete-Hughes, K. Findlay, K. Calder, L. Staverees, M. Best, and C. du Plooy. We are grateful to all the graduate students, researchers, pilots of the Aeroclub of Trelew, friends and volunteers who assisted in the collection and analysis of aerial survey data of the right whales off Argentina. In Argentina, we also thank the Organismo Provincial de Turismo for permits to conduct the research, the Wildlife Conservation Society for use of the research station, and Estancia La Adela for use of their landing strip. We are especially grateful to Aerolineas Argentinas, the Fundacion A. Fortabat, S. Haney, Metcalf and Eddy, Inc., Minolta Inc., the National Geographic Society, People's Trust for Endangered Species, the Wildlife Conservation Society, and the World Wildlife Fund for support of research in Argentina. Financial support for the genetic analyses was provided by the American University and the Pittsburgh Zoo. Samples were collected under a permit issued to PBB by the South African Department of Environment Affairs and imported in accordance with U.S. regulations (NMFS #825432, NOAA #1019). We thank J. Seger for helpful discussions and comments on the manuscript. Address correspondence to C. M. Schaeff at the address above or e-mail: schaeff@american.edu.

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Table 1. Southern right whale dorsal color markings

Pattern	Description ^a
Black	Dorsal skin is black.
White-blaze	Animal has an unpigmented area with edges that are distinct and straighter than in partial-grey-morph; marks remain white through the animal's life.
Grey-morph ^{a,b}	Animals are mostly white as calves but their skin darkens with age and adults appear grey or brownish-grey. Grey-morphs tend to have scattered black spots that are often in a band behind and spread laterally over the shoulder.
Partial-grey-morph ^a	Animal has a mark that is white in calves but darkens with age; similar to grey-morph adults but with much less white. Markings are more complex and have a rounded edge compared to white blazes. Marks appear to be formed by many overlapping circles of white and sometimes there are circular inclusions of black within a partial-gray region.
Partial-grey-morph with a white-blaze	Partial-grey-morph animals with a white blaze; marks are usually associated.

^a Phenotypes defined previously (Best 1990; Payne et al. 1983) but with different nomenclature: grey-morph previously referred to as partial albino, partial-grey-morph as grey-blaze.

^b Grey-morph animals with white blazes also exist but are difficult to distinguish during aerial surveys from grey-morph animals that do not have a white blaze; for this study, category II includes all grey-morph animals.

resulted in the development of several methods for determining sex genetically [e.g., Southern hybridization with a Y-chromosome-specific probe, pDP1007 (Page et al. 1987); polymerase chain reaction (PCR) amplification of sex-specific products (Bérubé and Palsbøll 1996; Bradbury et al. 1990; Palsbøll et al. 1992)].

For this study, we collected skin biopsy samples from free-swimming right whales off South Africa and determined their sex genetically using a PCR-based, three-primer system for the ZFY/ZFX sex-chromosome-specific region (Bérubé and Palsbøll 1996). We analyzed the frequency of pigment patterns in right whale populations off South Africa and Argentina in conjunction with cow-calf transmission patterns to identify possible genetic mechanisms for the color types. The evolutionary significance of the color patterns was investigated by examining the reproductive success of males and females with different color markings.

We have changed the nomenclature for right whale dorsal skin color phenotypes to avoid confusion between white blazes that remain white throughout an animal's life and the marks that darken, and to pro-



Figure 1. Female right whale with both grey and white blazes with her grey-morph calf. (Photographer: K. Payne)

vide a more accurate label for the animals previously known as partially albinistic. We refer to animals that are born predominantly white but then darken to grey as grey-morph animals (previously partially albinistic), animals that have a white mark that darkens to grey as partial-grey-morphs (previously grey-blazed), and animals with a white mark that remains white throughout their life as white-blaze (no change in nomenclature). The marks on white-blaze and partial-grey-morph animals are generally very easy to distinguish (Table 1).

Materials and Methods

Sighting Data

Right whales can be individually identified by their callosity patterns, scars, and skin color markings (Best 1990; Kraus et al. 1986; Payne et al. 1983). Near-vertical aerial photographs were taken of right whales off the South African coast during helicopter surveys of the coastline each year from 1979 to 1996 [see Best (1990) for details]. Flights were directed primarily toward mother-calf pairs; other animals were photographed only when in groups with mother-calf pairs. Between 1995 and 1996, animals were photographed from boats prior to biopsy collection; all categories of animals were photographed during this period.

The population of right whales off Peninsula Valdés, Argentina, has been photographically surveyed from fixed-wing aircraft each year from 1971 through 1997 [see Payne et al. (1983) for details]. All an-

imals encountered during a survey were photographed.

Calf body length measurements were taken photogrammetrically from a helicopter off South Africa in 1988 and 1989 using a radar altimeter to establish range (Best and Rüther 1992).

Sexing

Right whales were sexed (1) by viewing their genital slits (females and males), (2) behaviorally (adults seen in associations with calves were assumed to be females), or (3) genetically (females and males; Schaeff and Best 1998).

Small skin samples were collected from free-ranging right whales using a Paxams dart gun loaded with lightweight polycarbonate darts. Darts were fitted with stainless steel tips designed to receive and secure small (0.5–1.0 g) skin samples and then float on the surface of the sea (Best et al., in preparation). Skin samples were stored in a preservative solution of supersaturated NaCl and 20% dimethylsulfoxide (DMSO) at 4°C.

DNA was extracted from the skin samples using lysis buffer and proteinase K, followed by phenol/chloroform extractions as described in Schaeff et al. (1993). The ZFY/ZFX sex-specific regions were then amplified by PCR using primers ZFYX0582F, ZFY00767R, and ZFX0785R (Berube and Palsbøll 1996). Approximately 50 ng of genomic DNA was used as template in each 20 μ l reaction (67 mM Tris-HCl, pH 8.8, 1.5 mM MgCl₂, 16 mM (NH₄)₂SO₄, 60 mM tetramethylammonium chloride, 200 mM dNTPs, 0.5 μ M each

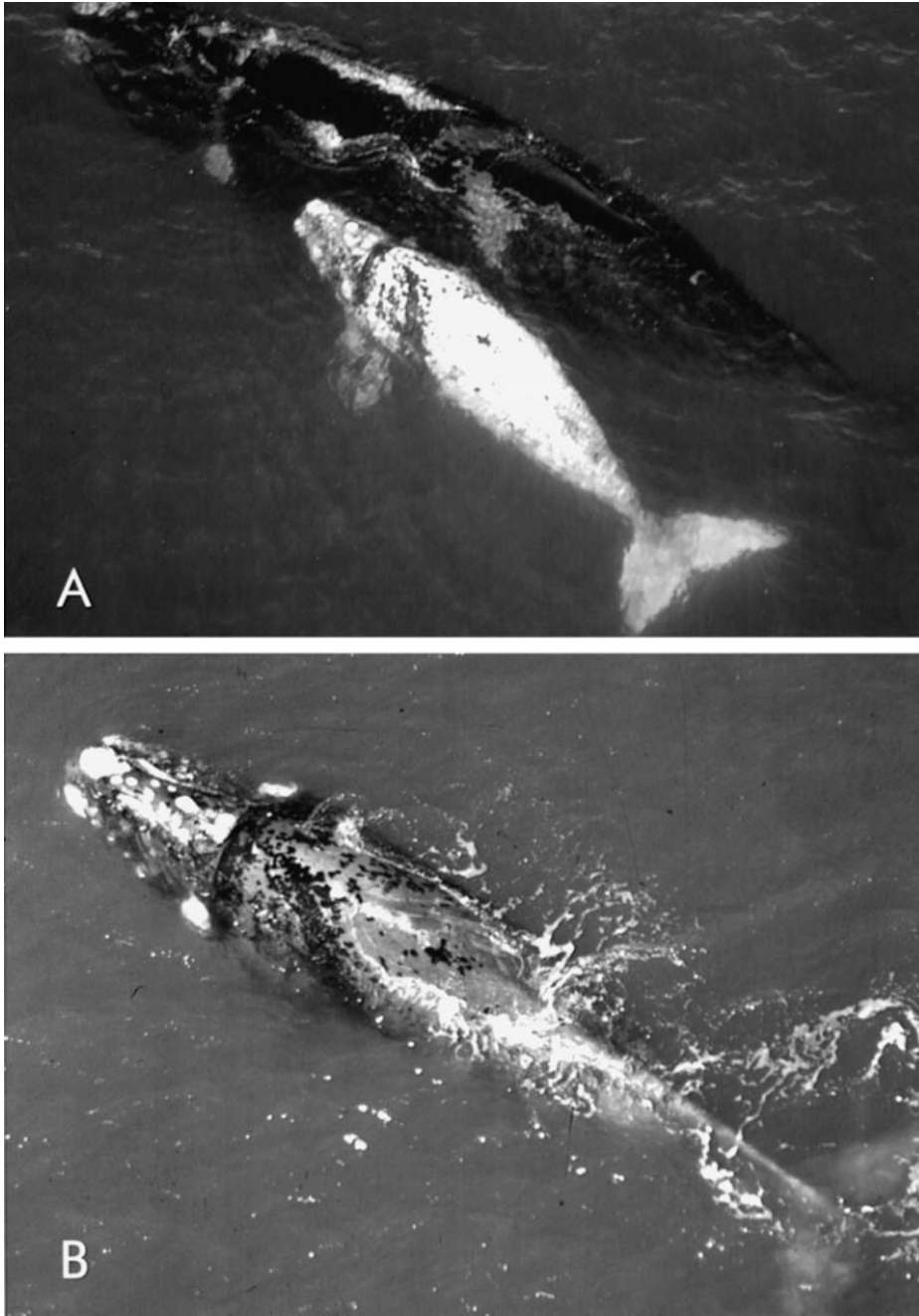


Figure 2. Grey-morph right whale photographed as (A) a calf and (B) a 2-year-old. (Photographers: G. Harris and R. Payne)

primer, and 0.4 units of BIOLASE DNA polymerase). Following denaturation at 94°C for 2 min, each reaction mixture was subjected to 34 cycles of 94°C for 60 s, 52°C for 60 s, and 72°C for 90 s. Negative controls (no template) were included in each experiment to detect any contaminating DNA. PCR products were sized by electrophoresis through a 3% NuSieve® gel. Each animal was sexed twice.

Modes of Inheritance

Most of the dorsal skin color patterns are easy to distinguish, including the marks

on white-blaze and partial-grey-morph calves; although the latter are white in calves, they are larger than the white blazes and the two have very distinctive shapes (Table 1; Figures 1 and 2). In contrast, grey-morph animals with white blazes exist, but the relative frequency of such animals remains uncertain due to the difficulty of detecting white blazes in young grey-morph animals in aerial photographs. Consequently, although six categories exist, we did not attempt to separate grey-morph animals with a white blaze from

those without, and hence used only five categories for this study (Table 1).

Using sighting data, the relative frequency of each color marking was determined for three groups of animals: cows, calves (animals in their first year of life), and noncalves (all animals sighted after their first year, including cows). Adult male frequencies were estimated from the noncalves data by assuming (1) a 1:1 sex ratio among noncalves and (2) that the color pattern proportions observed among known cows are representative of all noncalf females. Mode of inheritance was investigated by analyzing the distribution of marking patterns among animals of known sex and by analyzing skin color inheritance patterns among cow-calf pairs.

Evolutionary Significance

We examined the evolutionary significance of dorsal markings by comparing the reproductive success of animals with and without markings. Female reproductive success was assessed by comparing age at first calving, calf size, calving intervals, and length of sighting history. Right whales generally calve once every 3 years, but both shorter and longer intervals have been observed (Best 1990; Payne et al. 1990). It is difficult to separate longer calving intervals that result from increased time periods between pregnancies from those that result from a calf being produced during the intervening years but not seen. To compensate for this difficulty we calculated mean calving intervals for all interval data and also for 2- to 5-year intervals only.

Male success was estimated by comparing the frequency distribution of markings in calves with the frequencies expected based on noncalf frequencies and panmixic mating. For these calculations, all grey-morph adults were assumed to be males. Since some of these adults may be females, the predicted frequencies will provide maximum estimates of grey-morph male success. We also compared lengths of sighting histories of grey-morph and black males.

Results

Sexing

Skin samples were obtained from 97 animals including 60 with dorsal skin markings. Sampled whales included 25 animals (19 females and 6 males) whose sex was identified behaviorally and/or morphologically as well as genetically (Table 2). All

Table 2. Relative frequency of marks among genetically sexed animals

Marking	Males	Females
Black	18	19
White-blaze	8	6
Partial-grey-morph	0	28
Partial-grey-morph with white blaze	0	4
Grey-morph	14	0

genetic sex assignments agreed with those assigned in the field.

Modes of Inheritance

All five phenotypes were present in both populations (Table 3). The black phenotype was the most common in both populations, but the population off South Africa had a significantly lower proportion of black animals than did the population off Argentina (black versus nonblack, $G_1 = 62.72$, $P < .0001$).

If the dorsal markings are autosomal traits, then the marks should be equally common in males and females and marked calves should be produced by both marked and unmarked cows. If X-linked, we would expect the proportion of marked versus unmarked animals to differ between males and females, with males exhibiting the recessive phenotypes more frequently than females.

Among genetically sexed individuals, all partial-grey-morph animals were females, regardless of whether they had a white blaze as well ($N = 32$). Sighting data from Argentina indicated that partial-grey-morph animals (with and without white blazes) were approximately twice as common among cows (5.3%, $N = 340$) as among calves (3.3%, $N = 730$) and noncalves (2.8%, $N = 1048$); if calves and noncalves exhibit a 1:1 sex ratio, then these results are consistent with partial-grey-morph animals being female. Partial-grey-morph animals off South Africa were also approximately twice as common among cows as among calves (10.5%, $N = 351$, versus 5.8%, $N = 1026$) (data for noncalves not available).

All grey-morph animals sexed genetically were male ($N = 14$). The proportion of grey-morphs observed among calves and noncalves off Argentina were similar (Table 3; $G = 0.536$, $P = .464$; South African non-calf data not available), whereas grey-morph cows were rare or absent in both populations (Table 3). Grey-morph calves were produced by grey-morph cows and by partial-grey-morph cows with and without white blazes but not by black cows or

Table 3. Relative frequencies of dorsal skin color marks

Marking	Noncalves	Cows	Calves
A. Population off South Africa			
Black	N/A	84.3% ^a	89.3% ^b
White-blaze	N/A	4.8% ^a	1.5% ^b
Partial-grey-morph	N/A	9.1% ^a	5.6% ^b
Partial-grey-morph with white blaze	N/A	1.4% ^a	0.2% ^b
Grey-morph	N/A	0.3% ^a	3.5% ^b
B. Population off Argentina			
Black	93.1% ^c	92.0% ^d	93.7% ^e
White-blaze	2.8% ^c	2.6% ^d	2.0% ^e
Partial-grey-morph	2.6% ^c	4.7% ^d	3.2% ^e
Partial-grey-morph with white blaze	0.2% ^c	0.6% ^d	0.1% ^e
Grey-morph	1.3% ^{d,f}	0.0% ^d	1.0% ^e

^a $N = 351$.

^b $N = 1026$.

^c $N = 1048$.

^d $N = 340$.

^e $N = 730$.

^f One non-calf and one calf were grey-morphs that also had a white blaze.

white-blaze cows (Table 4). These patterns suggest that the partial-grey-morph and grey-morph phenotypes are controlled by one gene and the white-blaze phenotype by another. Given that the grey-morph trait has three phenotypes (grey-morph, partial-grey-morph, and black), one of which (partial-grey-morph) appears to be sex-specific, and given that males exhibited the grey-morph phenotype much more frequently than did females, this trait is probably X-linked, with genotypes $X^G X^g$ and $X^g Y$ producing the grey-morph phenotype, $X^G X^g$ producing the partial-grey-morph pheno-

type (no equivalent amount hemizygous males), and $X^G X^g$ and $X^g Y$ causing the black skin color.

Both males and females displayed white blazes including partial-grey-morph females (Tables 2 and 3) and grey-morph males (observed in Argentina population). Sighting data indicated that the proportion of white-blaze animals was similar among cows and noncalves off Argentina (Table 3). If noncalves exhibit a 1:1 sex ratio, then these results suggest that the white blaze occurs with equal frequency among both sexes. Both white-blaze and black calves were produced by cows with and without a white blaze (Table 4). These patterns are consistent with the white blaze being a simple autosomal trait.

If a simple genetic system is assumed, then it is possible to investigate whether the black or the white-blaze phenotype is dominant using the Hardy-Weinberg equation ($p^2 + 2pq + q^2 = 1$). This calculation requires that the population be in Hardy-Weinberg equilibrium (i.e., relative frequency of alleles remains the same from generation to generation; Ayala 1982). The relative frequency of white-marked animals differed significantly for cows and calves off South Africa (cows versus calves: $G = 16.107$, $P < .01$) indicating that the assumptions required for Hardy-Weinberg equilibrium were not met by this population. The relative frequencies among animals off Argentina did not differ significantly (cows versus calves: $G = 1.985$, $P = 0.32$; noncalves versus calves: $G = 2.68$, $P = 0.32$). For this reason, only the Argentina sample was used in this analysis.

If white blazes are the dominant phe-

Table 4. Cow-calf inheritance patterns for dorsal color markings

	Calves					
Cows	Black	White-blaze	Partial-grey morph	Partial-grey with white blaze	Grey-morph	Total
A. Population off South Africa						
Black	788	10	30	1	0	829
White-blaze	55	3	1	0	0	59
Partial-grey-morph	50	1	22	0	31	104
Partial-grey morph with white	8	1	3	1	2	15
Grey-morph	0	0	0	0	2	2
Total	901	15	56	2	35	1009
B. Population off Argentina						
Black	636	11	17	1	0	665
White-blaze	20	4	0	0	0	24
Partial-grey-morph	24	0	6	0	6	36
Partial-grey morph with white	4	0	0	0	1	5
Grey-morph	0	0	0	0	0	2
Total	684	15	23	1	7	730

notype, then at least half of the calves produced by white-blaze females should also be white-blaze. This prediction is not supported by cow-calf transmission patterns (cow_{white-blaze}/calf_{white-blaze} observed: 13.8%, $N = 29$; expected: $\geq 50\%$; $G = 9.01$, $P = .03$). In contrast, if the white blazes are recessive, then the proportion of white-blaze offspring produced by white-blaze cows should equal the probability that a male will pass on a recessive allele:

$$\text{cow}_{\text{white-blaze}}/\text{calf}_{\text{white-blaze}}: \\ \% \text{ expected} = q^2 + 0.5(2pq), \quad (1)$$

where q^2 is the population frequency of white-blaze adults. Similarly, if recessive, then the proportion of white-blaze calves produced by black cows is the probability that the cow is heterozygote and passes on her recessive allele times the probability that males will pass on a recessive allele:

$$\text{cow}_{\text{black}}/\text{calf}_{\text{white-blaze}}: \\ \% \text{ expected} = [q^2 + 0.5(2pq)] \\ \times [0.5(2pq)/(p^2 + 2pq)]. \quad (2)$$

Distribution of dorsal markers among cow-calf pairs off Argentina were consistent with both of these predictions (cow_{white-blaze}/calf_{white-blaze} observed: 13.8%, $N = 29$; if $q^2 = 3.0\%$, then expected: 17.2%; $G = 0.132$, $P = .717$; and cow_{black}/calf_{white-blaze} observed: 1.7%, $N = 713$ versus expected: 2.5%; $G = 1.22$, $P = 0.27$). Thus transmission patterns between cows and their offspring off Argentina are consistent with the white-blaze phenotype being recessive (ww) to the no-white-blaze (black) phenotype (Ww, or WW).

If the white-blaze trait is inherited independently of the grey-morph trait, then the ratio of partial-grey-morph and grey-morph animals with and without the white blaze should be the same as that for animals without the grey-morph trait (black) with and without the white blaze. Similarly the proportion of white-blaze animals with and without the grey- or partial-grey-morph should be the same as non-white-blaze animals (black) with and without the grey-morph trait. Distribution of markings among right whales off Argentina did not differ significantly from that expected if the grey-morph and white-blaze traits segregated independently (Calves: $G = 0.352$, $P = .55$; noncalves: $G = 1.75$, $P = .19$; cows: $G = 2.427$, $P = 0.12$; the expected frequencies of animals with both traits were very low for all three tests, $<1.0\%$ for

Table 5. Comparison of population parameters for right whales with different coloration patterns

	Dorsal skin color marking			
	Black	White-blaze	Partial-grey	Grey-morph
A. South African population				
Cows				
Calving interval (years)	3.6 ± 1.38 (539) ^a	3.8 ± 1.70 (37) ^a	3.3 ± 0.81 (69) ^a	n/a
Calf length (m)	3.1 ± 0.52 (473) ^b	3.1 ± 0.51 (30) ^b	3.2 ± 0.57 (66) ^b	n/a
Age at 1st calving (years)	6.87 ± 0.98 (131)	6.99 ± 0.80 (12)	7.42 ± 0.78 (5)	n/a
Sighting history (years)	n/a	8.25 ± 2.63 (4)	8.79 ± 2.17 (19)	n/a
Adult males				
Sighting history (years)	n/a	n/a	n/a	n/a
B. Argentinean population				
Cows				
Calving interval (years)	4.7 ± 2.53 (347) ^a	4.4 ± 2.29 (19) ^a	3.5 ± 1.15 (20) ^a	n/a
Calf length (m)	3.3 ± 0.71 (245) ^b	3.0 ± 0.41 (13) ^b	3.3 ± 0.83 (19) ^b	n/a
Age at 1st calving (years)	n/a	n/a	n/a	n/a
Sighting history (years)	11.26 ± 2.79 (19)	12.0 (1)	9.20 ± 1.79 (5)	n/a
Adult males				
Sighting history (years)	9.77 ± 5.29 (186)	11.67 ± 6.50 (6)	9.50 ± 4.93 (8)	n/a
	9.15 ± 5.35 (27)	6.50 ± 0.71 (2)	—	5.86 ± 4.02 (7)

Mean ± SD; numbers in parentheses are sample sizes.

^a All intervals.

^b 2- to 5-year intervals.

Statistics: calving intervals: all intervals: ANOVA: (partial-grey-morph/not, white-blaze/no white-blaze: a) $F_{1,1,1} = 0.758$, $P = .38$, Bonferroni/Dunn: all insignificant at 0.05 level; b) $F_{1,1,1} = 0.771$, $P = .38$, Bonferroni/Dunn: all insignificant at 0.05 level; 2- to 5-year intervals: a) $F_{1,1,1} = 0.179$, $P = .18$, Bonferroni/Dunn: all insignificant at 0.05 level; b) $F_{1,1,1} = 0.004$, $P = .95$, Bonferroni/Dunn: all insignificant at 0.05 level; calf length (black calves): a) ANOVA (length, month): partial-grey morph and black cows only (white-blaze cows, N too small to include) $F_{3,3} = 0.861$, $P = .46$; age at first calving: a) partial-grey morph versus white-blaze: t test: $t = 1.56$, $P = .13$; b) black versus partial-grey morph: t test: $t = .56$, $P = .13$; sighting history: a) cows: ANOVA (color) $F_2 = 1.741$, $P = .18$, Bonferroni/Dunn: all insignificant at 0.05 level; b) cows and adult males: ANOVA (sex, color) $F_{1,2} = 556$, $P = .57$, Bonferroni/Dunn: all insignificant at 0.05 level.

cows and calves, which increases the probability of type II errors) (South African data could not be analyzed because grey-morphs could not be distinguished from grey-morphs with white blazes).

We conclude that the two-gene model (X-linked grey-morph, white-blaze recessive autosomal) provides an excellent fit to all the available data for both southern right whale populations.

Evolutionary Significance

There was no significant difference in observed calving intervals for partial-grey-morph versus black cows within either population (all intervals or 2- to 5-year intervals only; Table 4). Mean calving intervals were shorter among cows off South Africa than those off Argentina, but inter-population differences in parameters such as calving intervals and age at sexual maturity are not valid because they assume that survey efficiency is comparable between populations, which is highly unlikely.

Among right whales off Argentina, partial-grey-morph cows tended to be younger at first calving than black cows, but the difference was not significant (Table 5). Data were unavailable for black cows off South Africa; mean age at first calving

among partial-grey-morph and white-blaze cows off South Africa did not differ (Table 5).

Black calves produced by partial-grey-morph and black cows had comparable mean body lengths (Table 5), as did partial-grey-morph and black calves produced by black cows (South Africa, partial-grey-morph calves: 6.66 ± 1.13 , $N = 7$ versus all-black calves: 6.90 ± 0.993 , $N = 133$; ANOVA: length and month measured, $F_{0,3} = 0.769$, $P = .51$; Argentina: data unavailable). Lengths of sighting histories were also similar for partial-grey-morph and black cows (Table 5; $t = 0.144$, $P = .886$); because grey-morph cows were very rare, there were insufficient data to include these cows in the analyses.

Grey-morph females (X^sX^s) result from matings between grey-morph (X^sX^s) or partial-grey-morph (X^cX^s) females and grey-morph males (X^sY). The number of grey-morph females expected in the population is the probability that a female will contribute a recessive allele times the probability that a male will contribute a recessive allele:

$$X^sX^s = [q_{\text{adult female}}^2 + 0.5(2pq)] \\ \times (0.5q_{\text{adult male}}). \quad (3)$$

Applying this formula, two of the 1009 right whale calves produced off South Africa between 1979 and 1992 should have been grey-morph females (one observed; $q_{\text{adult male}}$ estimated by $q_{\text{calf male}}$) and 0.2 of the 730 calves produced off Argentina between 1971 and 1990 should have been grey-morph females (none observed). South African sighting data indicated that partial-grey-morph animals occurred in groups with grey-morph animals as often as expected based on the relative frequency of the two types of animals in the population (mean number of animals observed in groups, excluding cow-calf pairs, was 2.96 ± 1.36 animals/group, $N = 122$, range 2–8 animals; groups with both a partial-grey-morph and grey-morph animal: observed 4/122 groups, expected 4.7/122; $G = 0.116$, $P = 0.73$).

The production of partial-grey-morph calves ($X^G X^s$) by black females ($X^G X^G$) indicates that grey-morph males ($X^s Y$) are fertile (Table 4). The expected proportion of partial-grey-morph calves produced by black cows equals the probability that a grey-morph male will contribute his X chromosome times his success in achieving fertilizations:

$$\frac{\text{cow}_{\text{black}}}{\text{calf}_{\text{partial-grey morph}}} \cdot \% \text{ expected} = (0.5q_{\text{adult male}})(s), \quad (4)$$

where s is the albino males' success in achieving fertilizations relative to that of black males. Among right whales off Argentina, the observed frequency of partial-grey-morph calves produced by black cows (i.e., black without white blaze: $X^G X^G$, W- and black with white blaze: $X^G X^G$, ww) was not significantly different from the number expected if grey-morph males experienced 100% reproductive success relative to black males (observed = 18/665 if $s = 100\%$, expected = 9/665, $G = 3.12$, $P = .08$). A population estimate for adult male grey-morph males was not available for right whales off South Africa; estimates based on $q_{\text{male calves}}$ indicated no difference between observed frequency (31/829) and that expected if males experienced 100% success relative to black males (29/829; $G = 0.069$, $P = .79$).

Among right whales off Argentina, mean length of sighting history was comparable for black males (9.15 ± 5.35 years, $N = 27$) and black females (9.77 ± 5.29 years, $N = 186$; t test: $t = 0.574$, $P = .57$). Grey-morph males exhibited shorter sighting histories (5.86 ± 4.02 years, $N = 7$) than did black males, but the difference was not significant given current sample sizes (t test: $t =$

2.29 , $P = 0.140$). Sighting histories of all animals with the grey-morph trait (male grey-morphs and female partial-grey-morphs, 7.80 ± 4.15 , $N = 15$) did not differ significantly from those of all black animals (male and female, 9.70 ± 5.29 ; $t = 1.35$, $P = .18$). Sighting history data were unavailable for male right whales off South Africa.

White-blaze and black cows exhibited similar calving intervals, mean calf body lengths, and length of sighting histories (Table 5). Nonetheless, the proportion of white-blaze animals was lower among calves than adults in both populations (South Africa: calves 1.7% versus cows 6.2%, $G = 16.107$, $P < .01$; Argentina: calves 2.1% versus cows 3.2%, $G = 0.985$, $P = .32$; versus noncalves 3.0%, $G = 1.00$, $P = .32$). The expected proportion of white-blaze calves (ww) produced by white-blaze cows (ww) equals the probability that a male will contribute a recessive allele times the males' success in achieving fertilization:

$$\frac{\text{cow}_{\text{white-blaze}}}{\text{calf}_{\text{white-blaze}}} \cdot \% \text{ expected} = [q^2 + 0.5(2pq)](s), \quad (5)$$

where s is the success of males that carry the recessive allele compared to that of males who do not carry the recessive allele (because data for noncalves were unavailable, q^2 for South Africa was calculated from cow frequencies; South Africa: if $s = 100\%$, $G = 9.154$, $P = .002$; if $s = 70\%$, $G = 4.19$, $P = .041$; if $s = 69\%$, $G = 3.36$, $P = .067$; Argentina: if $s = 100\%$, $G = 0.133$, $P = .72$). If the white blaze allele is completely recessive to the black allele, then although both heterozygous and homozygous recessive males contribute recessive alleles, only homozygous recessive males will experience any phenotypic effects. Since homozygous recessive animals are relatively rare in the population (South Africa: ww = 0.03 versus Ww = 0.346), the impact per ww male would have to be great to account for the observed reduction.

Note Added in Proof

Recent additions to the catalog of South African right whales (subsequent to the analysis presented in Table 3) indicate a total of three grey-morph individuals (presumably females) that have been photographed with a total of eight calves. Six of these calves are grey-morphs, but two (associated with two different adults) appear to be entirely black. The model developed

in this article explains grey-morph females as $X^s X^s$ homozygotes. Since such females would transmit an X^s chromosome to every one of their offspring, they should produce mainly partially grey-morph ($X^s X^G$) daughters and grey-morph ($X^s Y$) sons. (At low frequencies they would also produce $X^s X^s$ grey-morph females like themselves.) Such females are not expected ever to produce black offspring of either sex.

How can the two black offspring of grey-morph mothers be explained? There are several possibilities. In principle, better photographs might reveal small gray blazes on the poorly imaged extremities (or undersides) of the two apparently "black" calves, or these calves might not be offspring of the grey-morph individuals they were near when photographed. These artifactual explanations seem unlikely. Taking the observations at face value, models involving partial penetrance or X-inactivation can be constructed to explain how $X^s X^G$ females might occasionally exhibit grey-morph and/or black phenotypes. An even simpler model is one in which the South African population contains a third type of X chromosome, X^D , in which a region that includes the G locus is deleted. $X^s X^D$ females would be effectively hemizygous, like $X^s Y$ males, and would therefore be grey-morph, and some of their offspring would be black ($X^G X^D$) females.

Additional observations will be needed to distinguish among these alternative models. The discovery of black calves produced by grey-morph mothers does not warrant wholesale rejection of the basic sex-linked model for control of grey-morph and partial-grey-morph phenotypes, because the model gives such a good fit to the bulk of the data.

Discussion

Five dorsal skin color phenotypes are present in right whale populations off South Africa and Argentina (Best 1990; Payne et al. 1983; this study). Field and genetic data from these populations show similar general trends, suggesting that the modes of inheritance are the same in the two populations. The grey-morph trait observed among right whales (grey-morph, partial-grey-morph, and black phenotypes, previously known as partial albino, grey-blaze, and black, respectively; Best 1990; Payne et al. 1983) appears to be an X-linked gene with a female-specific partial-grey-morph phenotype resulting from the heterozygote genotype (hemizygous males do not exhibit the phenotype).

Since partial-grey-morph female calves have areas with black pigment and areas without pigment (similar to the lack of pigment observed in grey-morph calves), this phenotype may be the result of X-inactivation patterns (i.e., one of the female's two X chromosomes is deactivated in each cell, resulting in some cells exhibiting one phenotype and others the second phenotype—mosaicism or Lyon's hypothesis; Weaver and Hedrick 1992). The white blaze phenotype appears to be inherited independently of the grey-morph trait and to be a recessive autosomal trait. This two-gene model gives an excellent fit to all available data for both populations of southern right whales.

Although many skin color mutations are associated with pathological pleiotropism (Copeland et al. 1990; Goulding et al. 1993; Hosoda et al. 1994; Jackson 1997; Moller et al. 1996), we found no evidence to suggest that any of the dorsal skin color patterns observed in these right whales are correlated with a reduction in the reproductive success of cows. Calving intervals were similar for partial-grey-morph, white-blaze, and black cows in both populations suggesting that all cows are equally fertile. Calves produced by partial-grey-morph and black cows off South Africa had similar mean body lengths (white-blaze cows' data unavailable; Argentinian data unavailable) and the relative proportion of partial-grey-morph and white-blaze animals among calves and noncalves suggests that neither category of marked animals experienced increased juvenile mortality. In addition, partial-grey-morph, white-blaze, and black cows had sighting histories of similar length and exhibited similar ages at first calving. Small sample sizes for some categories in some of the analyses weakened the strength of the analyses. However, since the results from all five analyses are consistent with there being no difference in cows' fitness, there are ample data overall to support this hypothesis.

Grey-morph cows (X^sX^s) are rare and so are not included in the above analyses. Given the low population frequencies of the grey-morph (X^sY) and partial-grey-morph phenotypes ($X^G X^s$), few homozygote recessives are expected in either population. Hence it appears that grey-morph females are not missing but simply rare.

The occurrence of grey-morph females and the production of partial-grey-morph calves by black cows indicates that grey-morph males are fertile. As well, comparison of observed and expected frequencies of partial-grey-morph calves suggests

that grey-morph males are as successful as black males in achieving fertilizations with black cows. Grey-morph males may exhibit shorter sighting histories than do black males, but the difference was not significant given current sample sizes. Additional data are required to further investigate the impact of dorsal color patterns on male fitness.

The proportion of white-blaze animals is lower among calves than adults in both populations, significantly so among South African animals. Comparisons of observed and expected frequencies of white-blaze calves produced by white-blaze cows suggest that white-blaze males off South Africa may be less successful in achieving fertilizations than are black males ($s > 70\%$). In contrast, white-blaze and black males off Argentina appear to experience similar levels of success. In addition to differences in male reproductive success, some of the discrepancy in the proportions of white-blaze animals within and between populations could be the result of adults being scored as white-blaze due to scars they acquired since they were sighted as a calf, of white blazes on calves being missed if the blazes were positioned low on the back (South Africa), and of calves being less well photographed than cows—the backs of many calves cannot be seen clearly because the calves remain underwater (Argentina). Hence any potential correlation between the white-blaze dorsal mark and male reproductive success is difficult to judge.

Anomalously white individuals have been observed in 20 cetacean species (Fertl et al., in press; Hain and Leatherwood 1982), but southern right whales are the only species where such individuals are relatively common. A number of forms of partial albinism have been identified in other mammals, including three which may be X-linked: (1) the mottled gene series first described by Fraser et al. (1953), (2) Tietz's syndrome (Tietz 1963), and (3) a recessive X-linked trait described by Ziprkowski et al. (1962). However, none of these genes have patterns of inheritance and expression similar to those associated with grey-morph in right whales. For example, hemizygous mottled males are inviable, as are many of the homozygous mottled females. Both Tietz's syndrome and Ziprkowski's trait lack heterozygotic mosaicism and, in contrast to the situation in right whales, the white phenotype does not darken with age in any of these three forms of X-linked partial albinism. Finally, complete deafness is associated

with partial albinism in both Tietz's syndrome and Ziprkowski's trait (Searle 1968). Right whale courtship behavior is thought to be initiated by female vocalizations (Kraus 1991). Given the reproductive success of male grey-morphs (observed participation in right whale courtship groups and proportion of partial-grey-morph calves produced by black cows), it is unlikely that these males are suffering from severe deafness.

It is not clear why grey and partial-grey-morph right whales gradually darken with age. The dominant alleles of the roan (R) and ticking (T) genes cause dog coat colors to darken (Searle 1968), and temperature-dependent darkening is common in Himalayan mice, rabbits, and cats. As well, a slow spread of pigmentation from pigmented areas to nonpigmented ones has been observed in guinea pigs, cattle, sheep, pigs, and Dalmatians (Searle 1968). Additional data are required to further examine the issue in right whales.

Recently Portway et al. (1998) determined that despite the lack of physical barriers between geographic areas, the level of mtDNA gene flow is limited between the wintering populations of right whales off Argentina and South Africa. The different relative frequencies of both types of dorsal color markings observed in the two populations suggests that nuclear gene flow may also be limited. However, given the low frequency of the nonblack phenotypes in the populations, the usefulness of these markers for detecting gene flow is relatively limited. Analysis with a more sensitive marker, such as microsatellites, needs to be performed to estimate the extent of gene flow.

In addition to the two populations of southern right whales described in this study, all five phenotypes have been observed among southern right whales off Australia (Bannister J and Burnell S, personal communication) and New Zealand (Patenaude et al. 1998). In contrast, none of the northern right whales in the western North Atlantic display dorsal markings (Schaeff and Hamilton, in press) and only the white dorsal blaze has been observed among the North Pacific right whales (Herman et al. 1980; Rowntree et al. 1980). The failure to observe North Pacific animals with the grey-morph or partial-grey-morph phenotypes could be due to the small number of individuals sighted, a lower allele frequency in this population, or to the allele being absent. Observed differences in the frequencies of dorsal pigmentation patterns between the northern

and southern hemispheres suggest that gene flow across the equator may be limited and thus provides support for the hypothesis that northern (*E. glacialis*) and southern (*E. australis*) right whales are reproductively isolated (Braham and Rice 1984; Schaeff et al. 1991).

In addition to white dorsal blazes, many southern right whales have white ventral patches which are very similar in appearance to the dorsal blazes (Payne et al. 1983). It is not known how often the dorsal and ventral white marks occur together or whether they are controlled by the same gene. About 35% of right whales in the North Atlantic population have white ventral patches, but none have a dorsal blaze (Schaeff and Hamilton, in press). Without additional information it is also not clear whether the white ventral patches observed among northern and southern right whales are the same trait.

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Received June 29, 1998

Accepted December 31, 1998

Corresponding Editor: Stephen J. O'Brien