REPRODUCTIVE SEPARATION AND ISOLATING MECHANISMS BETWEEN SYMPATRIC DARK- AND LIGHT-PHASE WESTERN GREBES

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ABSTRACT.—Studies of dark- and light-phase Western Grebes (*Aechmophorus occidentalis*) during 1975–1977 revealed highly significant assortative mating by the color phases. Of 1,185 pair observations made during 2 yr in Utah, only 1.2% represented mixed pairs; the expected frequency of mixed pairs assuming random mating was 33%. Only two mixed pairs were noted in over 600 independent pair observations in California and Oregon. Mixed pairs with broods represented 0.25% of 766 broods surveyed. Nest initiation dates were significantly different between color phases in 1975 and 1976. Plumage development of captive chicks revealed striking differences; black crown feathers emerged 30–40 days later on light-phase chicks. Morphologically, only total culmen length for females differed significantly between color phases. Analysis of spatial distribution clearly indicates that light-phase birds are nonrandomly distributed among and within winter and summer flocks and among and within nesting colonies.

The data from Utah and California reveal that dark- and light-phase Western Grebes behave as separate biological species. Isolating mechanisms may involve a combination of inter-color-phase recognition and spatial segregation. *Received 8 September 1978, accepted 23 April 1979.*

THE two phenotypes of Western Grebe are classified as a single species (*Aechmophorus occidentalis*) by the American Ornithologists' Union Check-list Committee (1931, 1957) and are assumed to represent morphs (Mayr 1963: 151, Mayr and Short 1970). Storer (1965) and Lindvall (1976), however, observed assortative mating and found a combined total of 249 dark-phase pairs, 34 light-phase pairs, and 5 mixed pairs on Bear River Refuge in Utah.

The biological species concept is most commonly quoted as "groups of interbreeding natural populations that are reproductively isolated from other such groups" (Mayr 1969: 26). As Slobodchikoff (1976) points out, reproductive isolation is the key to this concept. Mayr (1951: 102, 1970: 18) recognized this problem and noted that incipient (new) species often have imperfect or incomplete isolating mechanisms leading to "occasional interbreeding of two otherwise well-delimited sympatric species." Bigelow (1965: 45) suggested that "overemphasis on 'interbreeding' seems to spring from the assumption that hybridization always tends to make gene pools of divergent populations more similar." Many authors, however, suggest that if hybrids are selected against, gene flow between closely related species may be an important factor in strengthening isolating mechanisms and leading to further divergence. Thus, speciation is regarded as essentially complete if, during sympatry, interbreeding is reduced to a level that prevents genetic swamping by the parent species (Mayr 1959, Bigelow 1965). Numerous accounts of hybridization are reported and discussed by Short (1969) and Mayr and Short (1970). Considering reports of assortative mating by dark- and light-phase Western Grebes, questions immediately arise: To what degree are the color phases of Western Grebe reproductively isolated? What are the behavioral mechanisms responsible for assortative mating, and are they being strengthened by selection? In order to answer these and other questions,

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The Auk 96: 573–586. July 1979

I designed a research plan to (1) determine the degree of reproductive separation, (2) analyze comparative ecological data, (3) identify and analyze isolating mechanisms, and (4) clarify the systematic relationship between dark- and light-phase Western Grebes.

George N. Lawrence (*in* Baird 1858: 894–895) originally described the phases as separate species, calling the dark form *Podiceps occidentalis* and the light form *Podiceps clarkii.* Mayr and Short (1970: 88) report the two color phases as "scattered polymorphism," and they are described by Storer (1965: 59): "There are dark-phase birds in which the bill is a rather dull greenish-yellow and the black of the crown extends below the eyes, the lores and the narrow line of bare skin extending from the eye to the gape; and there are light-phase birds which have orange-yellow bills and light faces—the black of the crown not reaching the lores or the eyes." Other plumage differences include whiter flanks and paler gray backs (for illustration see Storer 1965 and Ratti 1977).

Although published data on color phases are sparse, there is evidence suggesting clinal variation in the relative abundance of dark- and light-phase birds. Nero (*in* Palmer 1962: 96) found only 5 light-phase birds in a colony of 500 at Old Wives Lake, Saskatchewan; 2 of the 5 light-phase birds were paired. Storer (1965) reported a similar ratio of birds from that area. Bear River Refuge in Utah was reported (Storer 1965, Lindvall 1976) to have 12% light-phase birds (however, see results from this study). Dickerman (1973) recorded several populations in Mexico with light-phase birds being more abundant than dark-phase birds.

STUDY AREA AND METHODS

I studied Western Grebes on the Bear River Migratory Bird Refuge and at various locations in California and Oregon. Bear River Refuge is located 24 km west of Brigham City, Utah. The refuge contains 26,263 ha, 22,900 of which are open water. Prominent vegetation includes cattail (*Typha latifolia*), bulrushes (*Scirpus americana* and *S. paludosus*), saltgrass (*Distichlis stricta*), and pondweeds (*Potamogeton* spp.). The marsh is fed by the Malad and Bear rivers, and water is contained and controlled in five management units bordered by dikes. Marsh conditions vary from dry mud to 1 m of water; borrow ditches along dikes have numerous holes of greater water depth. The marsh is surrounded by cold shrub desert and has an average annual precipitation of 34 cm. Many additional details are provided by Behle (1958) and Williams and Marshall (1937).

I used three color-phase classifications in this study: light-phase, dark-phase, and intermediate. My classification of dark- and light-phase birds was consistent with that described by Storer (1965: 59) but was based primarily on facial pattern. Intermediate birds were those individuals whose black and white facial margin bisected the eye and in which the lores varied from white to dark gray. Storer (personal communication) classified seven facial types from museum specimens. Several of Storer's facial types differ by degrees of color (i.e. gray to black) and would be impossible to use while observing birds at a distance.

Two birds simply swimming or resting in close association were not classified as a pair. Birds were considered paired only when courtship communication was observed, primarily by "habit preening" (Storer 1963, 1969). Pairs consisting of two dark-phase birds will hereafter be referred to as *dark pairs,* two light-phase birds as *light pairs,* and a dark-phase bird paired with a light-phase bird as *mixed pairs.*

Surveys were conducted between 15 April and 1 August during the 1975 and 1976 seasons. An additional survey was conducted during peak activity in 1977. I conducted the surveys shortly after sunrise from a vehicle while slowly and systematically driving dike roads throughout the refuge; birds were observed through a 10× binocular and a 30× spotting scope. Because the same survey route was used each week, these survey observations contain an undetermined number of repeat observations, i.e. these combined data from Utah are not independent. In 1975, all birds that could be classified by color phase were recorded. Only paired birds and groups of five or more were recorded in 1976 and 1977. Records included location, date, general weather conditions, and color phase.

In January and June of 1977, I conducted field investigations in California, Nevada, and Oregon to obtain comparative data on flock composition by color phase and pair counts. Specific locations are listed in the Results section.

Searches for nests began around 1 June and continued into early September. Large nesting colonies were often detected by a sudden concentration of birds and by the vocalization that accompanies formation of colonies (Nuechterlein 1975). Many small colonies and isolated nests were located by systematic searches in a canoe along canals and marsh edges where emergent vegetation provided preferred nesting habitat. Low-level aerial surveys were conducted biweekly and proved successful, especially for more remote regions of the marsh that were not easily observed or searched. The color phase of birds attending nests was established prior to collecting additional data (for details see Ratti 1977). Clutch size was recorded for each identified nest; egg length and width were measured to ±0.1 mm using Vernier calipers. Initiation dates (first egg laid) were established by back-dating the age of eggs, using the methods reported by Westerskov (1950) and refined for Western Grebes by Nuechterlein (1975).

An estimate of production was obtained for each color phase by brood counts of free-swimming downy young along the same route used for weekly surveys. Brood counts were conducted at 10-day intervals and were analyzed by grouping alternate survey data to insure independent observations.

I obtained most of the birds used for morphological analysis by retrieving grebes found dead during routine travel on the marsh. An additional 15 birds were collected and provided data for electrophoretic analysis as well as morphology. All measurements were standardized according to Baldwin et al. (1931), with the following exceptions: (1) height of bill at nostrils was measured immediately posterior to the nostrils, and (2) length of closed wing flattened deviated, in that primary feathers were flattened against the measuring board.

A preliminary survey was conducted to determine if any electrophoretic variation existed between dark- and light-phase Western Grebes. Test samples were obtained from the Bear River Refuge. Additional details are presented by Ratti (1977). Buffer systems used are described by Ridgeway et al. (1970) and Markert and Faulhaber (1965). Specific details of the starch gel electrophoresis methodology employed are presented by May (1975). Methods of hatching and maintenance of captive birds are described by Ratti (1977).

RESULTS

Weekly surveys.—Between 6 April and 25 July 1975, I conducted 18 surveys averaging 208 independent observations. During the entire survey period, 3,736 dependent observations were accumulated. In June and July, when large numbers of grebes were present, the percentage of observed light-phase birds varied from 14 to 28%. An average of the weekly proportions yielded an estimate of 17.9% (±6.4 SD) light-phase birds. I classified 26 birds (0.7% of the total) as intermediate color phase. Total counts were not conducted in 1976 and 1977.

My observed frequency of intermediate-phase birds is consistent with that of Storer (1965). Storer (personal communication), however, reports greater frequency of intermediates among museum specimens. Light-phase birds were observed in the field that appeared to be intermediate-phase while feathers were relaxed, yet distinctly showed a white margin above the eye when crown and facial feathers were erected during behavioral interaction. Such individuals may be more difficult to classify accurately from museum specimens. Intermediate-phase birds accounted for less than 1% of over 3,500 independent observations in California, Oregon, and Nevada in 1977.

Confounding the problem was the existence of dark-phase birds with whitish lores in California. I classified these birds as "dark-phase" because the black of the crown extended below the eyes, and flank and bill colors were typical of dark-phase birds. Although this variant is very rarely observed in Utah, approximately 5% of the birds observed in California in January were classified as dark-phase birds with whitish lores. Only two such variants, however, were observed during June pair counts in California. Possibly the whitish lores on dark-phase birds are associated with winter plumage and the lores may darken in breeding plumage.

TABLE 1. Chi-square analysis of assortative mating by dark- and light-phase Western Grebes on the Bear River Migratory Bird Refuge, 1975, 1976, 1977.

Survey number	D × D	L × L	D × L	Corrected χ^2	Significance
1975					
1	4	4	0	—	0.014[a]
2	15	4	0	—	0.0003[a]
3	45	7	0	43.8	0.00001
4	16	2	0	—	0.006[a]
5	22	5	0	20.8	0.00001
6	26	2	0	14.9	0.0001
7	38	9	1	36.4	0.00001
8	41	9	2	34.5	0.00001
9	46	16	1	53.2	0.00001
10	29	4	2	14.8	0.0001
11	35	6	0	33.4	0.00001
12	41	12	0	47.4	0.00001
13	78	14	2	72.9	0.00001
14	54	17	1	61.6	0.00001
15	50	18	1	58.3	0.00001
Subtotal	*540*	*129*	*10*		
1976					
1	4	3	0	—	0.28[a]
2	6	4	0	—	0.0047[a]
3	13	3	0	—	0.0018[a]
4	17	8	0	20.6	0.00001
5	22	7	0	23.8	0.00001
6	45	9	1	42.1	0.00001
7	51	7	1	42.6	0.00001
8	61	15	0	69.8	0.00001
9	33	15	0	43.6	0.00001
10	38	14	1	43.5	0.00001
11	98	29	1	117.03	0.00001
Subtotal	*388*	*114*	*4*		
1977					
1	140	46	0	184.2	0.00001
GRAND TOTAL	1,068 (78%)	289 (21%)	14 (1%)		

[a] Fisher's exact test.

Assortative mating.—In 1975, I accumulated 719 observations of pairs; 570 (79.3%) were dark pairs, 135 (18.8%) were light pairs, 10 (1.4%) were mixed pairs, and 4 (0.5%) pairs had 1 intermediate-phase bird (either D × I or L × I). Of the 10 mixed-pair observations, seven were considered to be repeat observations of the same pair. A L ♀ × D ♂ pair was observed almost daily at the same location.

My observations of pairs in 1976 totaled 506; 388 (76.6%) were dark pairs, 114 (22.5%) were light pairs, 3 (0.6%) were mixed pairs, and 1 pair was D × I. Pairs containing an intermediate-phase bird were omitted from the analysis on assortative mating.

To test the statistical significance of assortative mating, each survey was subjected to Chi-square analysis. Table 1 reports on 27 surveys over 3 yr; each survey was tested separately to assure independence of observations. These data clearly indicate the low probability of observing a mixed pair.

Data on assortative mating from 11 July 1975, 12 July 1976, and 1 August 1977 were combined to estimate the expected percentage of mixed pairs assuming random mating. The three data sets represent the largest single survey totals for each year, and are assumed to be independent. I observed 316 dark pairs, 89 light pairs, and

TABLE 2. Assortative mating by dark- and light-phase Western Grebes observed in California and Oregon, June 1977.

Location	D × D	L × L	D × L	χ^2
OREGON:				
Malheur Lake	13	0	0	0.0
Upper Klamath Lake	18	17	1	32.2
CALIFORNIA:				
Lower Klamath Lake National Wildlife Refuge	32	8	1	34.4
Tule Lake National Wildlife Refuge	121	18	0	140.9
Goose Lake	17	299	0	321.1
Eagle Lake	60	1	0	62.5
TOTAL OBSERVED	261	343	2	
EXPECTED (POOLED)	213	296	97	

Pooled $\chi^2 = 591.1$; significance = $P < 0.001$

3 mixed pairs in these surveys. Chi-square analysis generated expected values of 247.1 dark pairs, 20.1 light pairs, and 140.8 mixed pairs ($P < 0.001$). Mixed pairs occurred at about 2% (3/140.8) of the frequency expected from random mating on Bear River Refuge during these 3 yr.

Field investigations in California and Oregon confirmed the degree of reproductive separation observed on Bear River Refuge in Utah. In June 1977, 606 independent observations of pairs were recorded. Only two mixed pairs were observed: a mixed pair feeding young on Upper Klamath Lake and a second pair attending a nest on Lower Klamath Lake. Data on pair counts from specific locations outside Utah, including observed and expected values, are presented in Table 2. These data indicate that mixed pairs occur at 2% of the expected frequency (2/97) assuming random choice of mates.

Nest initiation dates.—Initiation dates were determined for 70 nests of dark pairs and 39 nests of light pairs in 1975 and 103 nests of dark pairs and 68 nests of light pairs in 1976. Light-phase birds had an earlier mean initiation date than dark-phase birds in 1975 but a later mean date in 1976; the differences between color phases were highly significant (*t*-tests, $P < 0.001$; $\chi^2 = 32$, $P < 0.001$). Mean initiation dates were nearly identical between years for dark-phase birds: 21 July in 1975 and 22 July in 1976. Light-phase grebes, however, varied widely between years, with a mean initiation date of 29 June in 1975 and 28 July in 1976. Reproductive readiness is apparently signaled among members of individual flocks, which band together to form nesting colonies. Because flock composition is nonrandon (see below) and nesting by grebes is not highly synchronous at Bear River Refuge, it is not surprising to find differences in initiation dates.

Clutch size.—In 1975, clutch size was determined for 111 nests. Seventy nests attended by dark-phase birds contained an average of 2.53 eggs; 61.4% had 3 or more eggs. Light-phase nests had a lower average of 2.39 eggs for 41 nests, 46.4% containing 3 or more eggs. Chi-square analysis indicated the differences were not significant ($P = 0.11$).

Clutch size was recorded for 154 nests in 1976. These data indicate a reversal from 1975, in that light-phase birds had a higher mean clutch size of 2.44 eggs, compared to 2.31 eggs for dark-phase birds. Three or more eggs were found in

41.1% of the dark-phase nests and in 50% of the light-phase nests. Again, these differences were not significant ($P = 0.16$).

Egg length and width.—Measurements of egg length and width were taken on 173 eggs from dark-phase nests and 95 eggs from light-phase nests in 1975. Egg size did not differ significantly between color phases. The mean egg length was 57.6 mm (±2.2 SD) for dark-phase birds and 57.3 mm (±2.1 SD) for light-phase birds. The mean egg width was 38.8 (±1.1 SD) for dark-phase birds and 38.5 (±1.1 SD) for light-phase birds.

Brood counts and production estimate.—In 1975, I observed 507 Western Grebe broods, 349 with dark-phase adults and 158 with light-phase adults. The mean brood size was 1.85 for dark-phase birds and 2.11 for light-phase birds. In 1976, I observed 259 broods, 160 with dark-phase adults and 99 with light-phase adults. The mean size of dark-phase broods was 1.41 and light-phase broods averaged 1.53. Brood size ranged from 1 to 3 chicks for 99% of all broods surveyed. When I calculated the percentage of broods with 1, 2, or 3 chicks for each color phase, I discovered that light-phase pairs had 10–12% greater incidence of broods with 2 or 3 chicks for all surveys. Four brood surveys were conducted each season; each season's data were divided into two statistical tests to insure independent observations (for details see Ratti 1977). Chi-square analysis revealed differences at the 0.01 and 0.11 levels for 1975 but were clearly not significant for 1976 ($P = 0.42$, 0.37). Several mixed broods and mixed pairs with broods were observed. Of 766 broods surveyed in 2 seasons, 2 were attended by mixed pairs and 4 contained chicks of both color phases.

My observations indicate a short-term trend of proportionally greater production by light-phase birds relative to the entire population of grebes. Although light-phase birds represented approximately 18.6% of the population, their representation in pairing, nesting, and brood production gradually climbed to 33.6% (Fig. 1). Greater production by light-phase birds is also indicated by their larger mean size of broods. These data may indicate greater production by light-phase birds, reduced production by dark-phase birds, or a combination thereof. If the trend continues, the net result will be increased numbers and percentage of light-phase birds in the grebe population on the Bear River Refuge.

Spatial distribution of color phases.—From data collected during several months of field investigations, I suspected that spatial segregation existed between dark- and light-phase birds on the Bear River Refuge. Dickerman (1973) also reported that his series of specimens from Mexico "indicates there may be some geographic segregation of the phases." Consequently, data collection and analyses were designed to test the significance of spatial distribution of sympatric populations.

Chi-square analysis was employed to analyze flock composition by color phase. Each weekly survey was analyzed separately; flock size ranged from 3 to 97 individuals on the Utah study area. Nineteen flock composition surveys were conducted in 1976, and Chi-square tests for independence of phases revealed probability levels of $P < 0.05$ for 14 surveys, $P < 0.15$ for 2, and $P > 0.15$ for 3. These data clearly indicate that dark- and light-phase Western Grebes are not randomly distributed on the Bear River Refuge.

Spatial segregation of nesting birds was examined according to distribution among nesting colonies, within nesting colonies, and by nearest neighbor analysis of nests. A Chi-square test revealed no significant difference between the ratio of dark- and light-phase Western Grebes on the refuge for 1975 and 1976. Therefore, data on

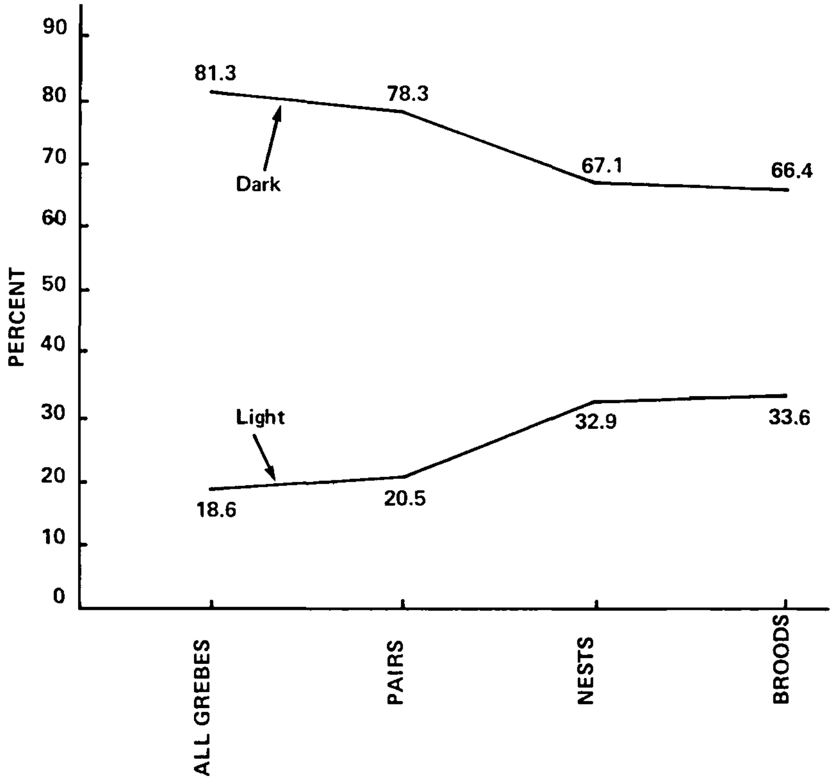

Fig. 1. Percent of dark- and light-phase representation for Western Grebe data on total numbers, pairs, nests, and broods on the Bear River Migratory Bird Refuge.

colony composition from both years were combined and analyzed. Dark- and light-phase Western Grebes were not randomly distributed among nesting colonies (Table 3). These data are consistent with those previously discussed concerning flock composition. One of the most striking examples of nonrandom distribution is demonstrated by colony number 1. Although light-phase birds represent less than 20% of the grebe population, 86% of the nests in colony number 1 were of light-phase birds.

Distribution of dark- and light-phase nests within colonies was analyzed by runs tests (Sokal and Rohlf 1969). I justified use of this test because the large, open-water colonies were generally "crescent shaped," with the length of the colony several times greater than the width. Runs tests determine whether or not the occurrence of a nest of one color-phase bird is a function of the color phase of a bird on the preceding nest. Data from this study were generated by systematically recording the color phase of incubating birds. Observations were conducted from a fixed observation point, beginning at one end of the nesting colony and proceeding in an arc (determined by the line of sight) through the colony. Therefore, data were recorded in a running series as each succeeding nest appeared in the line of sight (e.g. L L D L D D D D L . . .). Runs tests were conducted on five open-water nesting colonies. Two tests were highly significant ($P < 0.001$), indicating nonrandom distribution, and 3 were not significant ($P > 0.17$). Runs tests, however, are an extremely con-

TABLE 3. Chi-square analysis of colony composition for dark- and light-phase Western Grebe nests on the Bear River Migratory Bird Refuge, 1975 and 1976. Total grebe nests: dark = 374 (67%), light = 186 (33%).

	Number (% in parentheses)	
Colony number	Dark nests	Light nests
1	5 (14)	30 (86)
2	1 (25)	3 (75)
3	21 (100)	0 (0)
4	4 (50)	4 (50)
5	8 (47)	9 (53)
6	126 (76)	40 (24)
7	38 (49)	40 (51)
8	43 (74)	15 (26)
9	109 (75)	36 (25)
10	19 (68)	9 (32)
Raw Chi-square = 82.2; df = 9;	significance = 0.00001	

servative test and were utilized due to the lack of a more appropriate test. Observed segregation by color phases in nesting colonies closely resembled what Pielou (1961: 256) illustrated and described as "partly segregated."

Nearest neighbor analysis was conducted on five colonies located in emergent vegetation. Of 54 nests of dark-phase birds, 45 of the nearest neighbors were dark-phase. Of 35 nests of light-phase birds, 28 of the nearest neighbors were light-phase. These results are highly significant ($\chi^2 = 32.5$; $P < 0.001$) and more clearly demonstrate the nonrandom distribution of color phases within nesting colonies. The mean distance from the nest of a light-phase bird to the nearest light-phase neighbor was 6.8 m; dark to dark averaged 10.2 m ($P = 0.1$).

The significant and consistent evidence showing spatial segregation between color phases during summer months in Utah prompted investigation of spatial relationships on wintering grounds in California and Nevada (Table 4). Again, Chi-square analysis revealed nonrandom distribution of dark- and light-phase Western Grebes. Although light-phase birds accounted for only 12% of all grebe observations, several flocks (3, 4, 14, 15) contained 54–83% light-phase individuals (Table 4). All 2,373 observations are independent.

Runs tests were also conducted on nine flocks observed in California to test within-flock distribution; the results were consistent with those previously presented. Nonrandom distribution was significant ($P < 0.03$) for six of the nine flocks analyzed. General observations strongly supported the results of statistical tests; distribution of light-phase birds among and within flocks commonly appeared clumped.

Observations and analyses of spatial segregation received overwhelming support by the discovery of the light-phase population on Goose Lake during June 1977. Investigations at Goose Lake produced 819 independent observations, of which light-phase birds represented 93%. Goose Lake is in the approximate center of five lakes, all with large Western Grebe populations of predominantly dark-phase birds. Malheur Lake is 200 km to the northeast; Lower Klamath, Tule, and Clear lakes are within 160 km to the west; and Eagle Lake is 200 km south of Goose Lake.

Morphology.—Means and statistical tests of measurements taken from dark- and light-phase Western Grebes are presented by Ratti (1977). Only one measurement differed significantly between color phases: total culmen length in females averaged

TABLE 4. Chi-square analysis of flock composition for dark- and light-phase Western Grebes observed at various locations in California and Nevada, January 1977. Total grebes: dark = 2,098 (88.4%), light = 275 (11.6%).

Flock number[a]	Number (% in parentheses) Dark phase	Light phase
1	126 (85)	22 (15)
2	14 (70)	6 (30)
3	20 (46)	23 (54)
4	8 (40)	12 (60)
5	6 (75)	2 (25)
6	38 (100)	0 (0)
7	11 (92)	1 (8)
8	25 (93)	2 (7)
9	36 (92)	3 (8)
10	69 (97)	2 (3)
11	21 (100)	0 (0)
12	67 (100)	0 (0)
13	29 (83)	6 (17)
14	4 (17)	20 (83)
15	5 (42)	7 (58)
16	184 (95)	10 (5)
17	321 (99)	3 (1)
18	385 (93)	30 (7)
19	474 (82)	106 (18)
20	71 (99)	1 (1)
21	28 (100)	0 (0)
22	134 (92)	12 (8)
23	22 (76)	7 (24)

Raw Chi-square = 393.1; df = 22; significance = 0.00001

[a] Specific locations of each flock are reported by Ratti (1977).

3 mm smaller for light-phase birds than for dark-phase. All measurements of males revealed no difference between color phases. Six measurements were made for each sex, for a total of 12 cases. In eight cases, light-phase means were smaller. These data are difficult to interpret. Although only one of these is statistically significant, the general trend of smaller measurements of light-phase birds may have biological significance.

Electrophoresis.—Starch gel electrophoresis applied to tissue samples from dark- and light-phase birds revealed no differences between color phases (for illustration see Ratti 1977). A variant allele, however, was detected in muscle and liver alpha glycerophosphate dehydrogenase for both color phases. Two heterozygotes were observed in light-phase samples and one in dark-phase samples. Because of the excessively large sample sizes required, I did not attempt to investigate gene frequency of this variation for each color phase.

Phenotypic development.—Storer (1965, citing Dawson) was the first to report suspected dimorphism in the downy young of Western Grebes and suggested that this dimorphism might be related to dark and light color phases. Lindvall (1976) reported from field observations that chicks with dark-phase parents began to show black on the head and back at a much earlier age than light-phase young.

I studied development by periodic photographs of eight dark- and six light-phase birds hatched and raised in captivity (eggs were collected from nests where the color phase of incubating birds was known). Black crown color began to show on dark-phase chicks at 10–15 days of age. Dark-colored feathers on the crown of light-phase chicks did not emerge until 50–60 days of age. At 45 days of age, dark-phase birds

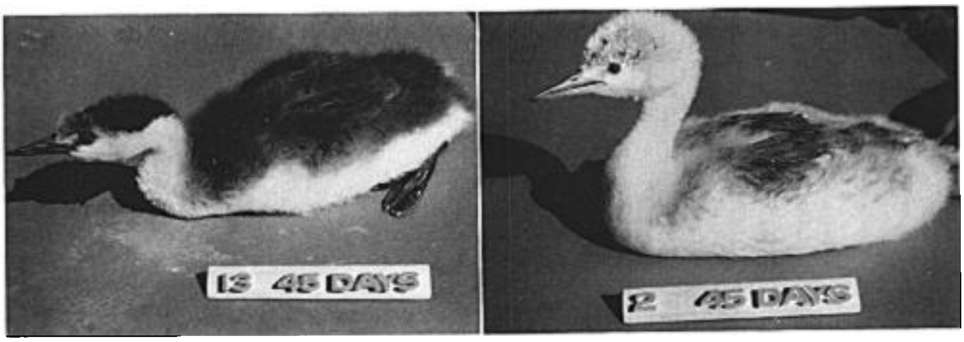

Fig. 2. Dark- (left) and light-phase (right) Western Grebe chicks at 45 days of age. Dark-phase chicks developed black crown feathers at an earlier age than light-phase chicks. Black back color also developed earlier on dark-phase birds, and, by comparison, light-phase birds generally appeared snowy white.

clearly showed black on the crown while light-phase birds had predominantly white crown feathers (Fig. 2). A similar pattern was observed for down feathers on the back and wings. Between 20 and 50 days of age, light-phase chicks appeared generally snowy white, while dark-phase chicks showed charcoal-colored backs and had a much darker appearance. Bill color was black for both color phases during the first 40 days, at which time the respective bill colors (previously described) slowly began to emerge. By 80 days of age, bill color closely resembled dark- and light-phase adults. All chicks had gray irides. The red color of adult irides did not appear in the first 80 days, and time of appearance was not established. Primary feathers emerged at approximately 40 days of age for each color phase. A full complement of primary feathers, which appeared suitable for flight, was acquired at approximately 70 days of age.

Captive birds were weighed daily, from hatching to 75+ days of age. These data were analyzed using Richard's (1959) growth model and the analysis is reported by White and Ratti (1977). No differences were detected between growth rate coefficients of dark- and light-phase birds.

DISCUSSION AND CONCLUSIONS

Systematic relationships.—Although Mayr's (1969) definition of species most heavily concentrates on interbreeding, avian systematists commonly consider geographic relationships, phenetic differences, and reproductive isolation as important components. Dark- and light-phase Western Grebes on the Bear River Refuge and several California marshes biologically function as separate species by these criteria. The morphs are sympatric, phenotypically different, and reproductively isolated. Although reproductive isolation is not entirely complete, the occurrence of hybridization is well within the limits observed for closely related sympatric avian species, especially gregarious forms. Consequently, the existence of occasional hybridization between dark- and light-phase Western Grebes is not evidence for conspecific status.

Both egg dumping and brood mixing could result in imprinting or conditioning by a given chick to an adult of the opposite color phase and thus increase the probability of that chick later seeking a mate of the opposite color phase. A similar hypothesis is presented by Prevett and MacInnes (1973) for hybridization between Canada Geese (*Branta canadensis hutchinsii*) and Snow (Blue) Geese (*Chen caeru-*

lescens). Finley (1907), Bent (1919), and Lindvall (1976) report dump nests for Western Grebes. Brood mixing is a common phenomenon among gregarious water bird species and requires little discussion here. Additional evidence that could be interpreted as either egg dumping or brood mixing is provided in this study. Only 1 nest with 5 eggs was recorded (none was recorded with more than 5), yet 3 broods with 5 chicks and 1 brood with 7 chicks were observed.

Although some variables are of greater biological significance than others, the differences observed for initiation dates, brood size, spatial segregation, morphology, and phenotypic development substantiate the data that indicate dark- and light-phase birds represent reproductively independent populations. I recommend resurrection of *A. clarkii* as a separate species from *A. occidentalis,* as originally described (Baird 1858: 894–895). Data are needed, however, on the biology of mixed pairs and intermediate-phase birds. Future study may show that such data are difficult to obtain due to their rare occurrence. In addition, data on assortative mating from other sympatric ranges, especially Mexico, should be obtained.

Isolating mechanisms.—Spatial segregation is apparent among and within flocks, among and within nesting colonies, by nearest neighbor analysis, and among breeding populations in general. On Bear River Refuge, light-phase birds are strongly attracted to each other, resulting in the highly significant nonrandom distribution of color phases. A similar trend was observed for dark-phase birds in the predominantly light-phase population at Goose Lake. Inter-color-phase avoidance is an alternative explanation, but the data and general observations fail to support this hypothesis. Spatial segregation, therefore, should not imply an inter-color-phase negative response. Although I commonly refer to "segregation," preferential association among members of the same color phase is an alternative and equally feasible explanation of nonrandom distribution. Spatial segregation may function as an effective isolating mechanism, because it increases the probability that a bird will be exposed to a mate of the same color phase at the onset of reproductive activity and pair formation.

Recognition of members of the same color phase, evidenced by spatial segregation, is a key factor to assortative mating. Dark- and light-phase birds clearly prefer to associate with individuals of the same color phase. Consequently, it is logical to conclude that color-phase recognition is important to mate selection. Although more data are needed, color-phase recognition is likely the result of imprinting and conditioning to the facial pattern of parents by chicks. Other isolating mechanisms, such as vocalization or minute behavioral differences, may be present; disclosure of these awaits further study.

Data on mixed pairs indicate that isolating mechanisms between dark- and light-phase Western Grebes are being strengthened by hybridization and the "test of sympatry" (Stebbins 1971: 99). Many mixed pairs may be unsuccessful nesters, for mixed pairs represent 1.2% of my observations on prenesting pairs but only 0.26% of pairs with broods. These data suggest that production by mixed pairs is extremely rare and that reproductive isolation between the color phases may be greater than indicated by data on assortative mating. An alternative explanation would be that the occurrence of mixed pairs is so infrequent that sampling error could account for the observed difference between the frequency of prenesting pairs and those with broods.

Evolutionary considerations.—Storer (1965) first hypothesized that isolated populations of Western Grebes diverged, with light-phase populations developing in the south and dark-phase in the north. Storer (personal communication) and Mayr and Short (1970) have also suggested that light- and dark-phase Western Grebes represent polymorphism. Mayr (1963) and Traylor (1966) report that polymorphism is best demonstrated by random mating or by both phenotypes occurring in a single nest. Dark- and light-phase Western Grebes exhibit an intensive degree of assortative mating, with hybridization occurring at about 2% of the expected frequency. Interbreeding between conspecific polymorphs has been shown to be considerably greater. Rockwell and Cooke (1977: 95), for example, estimated that mixed pairs of color phases of the polymorphic Snow Goose (*Chen caerulescens*) were 50% of the expected frequency. Mixed pairs of the polymorphic Parasitic Jaeger (*Stercorarius parasiticus*) occur at 87% of the expected frequency (calculated from O'Donald 1959). Consequently, the definition of polymorphs as a "single interbreeding population" (Mayr 1963: 150) provides a strong argument for rejection of this hypothesis for Western Grebes. If dark- and light-phase grebes are morphs, they represent a unique biological phenomenon. Examination of the literature on avian systematics indicates that no other broadly sympatric avian polymorphic species is known to exhibit such a low incidence of interbreeding.

Color phases of Western Grebe chicks cannot be identified accurately at hatching. Therefore, the best field indication of the composition of color types in a single nest is provided by brood surveys. Of 766 broods surveyed in two seasons, only 4 (0.5%) contained chicks of both color phases. These rare occurrences may indicate polymorphism, inter-color-phase egg dumping, or brood mixing.

Additional difficulties arise if we assume that Western Grebes were polymorphic prior to development of isolating mechanisms. Divergence by polymorphic populations is reported as sympatric speciation, a concept that is widely rejected by avian systematists. Hypothetical modes of sympatric speciation via polymorphism are presented by Maynard-Smith (1966) and Dickinson and Antonovics (1973). These examples describe morphs being selectively favored in different niches. Data from my study do not reveal differential niche utilization by color phases. Assortative mating by the polymorphic Snow Goose (Cooch and Beardmore 1959), however, indicates that such a mode of speciation is feasible, although genetic isolation by these geese has not reached a level to suggest speciation or strengthening of isolating mechanisms (Rockwell and Cooke 1977).

A mode of speciation that would be compatible with Storer's hypotheses of both polymorphism and geographic divergence is as follows: A large polymorphic population of Western Grebes may have become divided and geographically isolated. During isolation, selection pressures may have eliminated dark-phase morphs from one population and light-phase morphs from the other. When the populations expanded and became sympatric, isolating mechanisms had developed sufficiently to maintain reproductive isolation.

Morphologically and ecologically, dark- and light-phase Western Grebes are practically indistinguishable. Yet they are sympatric and reproductively isolated. These populations may represent one of the best avian examples of an exception to the competitive exclusion principle (Grinell 1917), with nearly identical utilization of resources in space and time. My observations lead me to suspect that if niche overlap is nearly complete, the situation is maintained due to populations being regulated below a level where resources are limiting; thus, there is little or no competition. An alternate hypothesis would be that secondary sympatry of the color phases may be relatively recent and that the process of competitive exclusion may be either unrecognized or in early stages.

Geographic variation.—Previous studies have reported clinal variation in abundance of dark- and light-phase Western Grebes, with dark-phase birds representing 80–90% of populations in California, Utah, and Canadian marshes. Consequently, the Goose Lake population appears to be an especially significant finding and possibly the largest concentration of light-phase birds in the United States and Canada. Thus, reports of clinal variation in abundance may need to be reconsidered.

Dickerman (1963, 1973) discussed several populations of Western Grebes from the Mexican Plateau and, from data that indicate the birds are smaller than northern populations, considers them a subspecies. Dickerman has suggested the subspecific name *A. o. clarkii,* which includes both color phases. This suggested classification may also require reevaluation in view of the data on reproductive separation between color phases.

ACKNOWLEDGMENTS

I thank John Kadlec and Keith Dixon for their advice and critical analysis throughout the research project. James Gessaman, Ivan Palmblad, and Gar Workman provided many helpful pre- and post-project suggestions. Don Sisson, Gary White, and Dave Otis assisted with statistical analyses and were essential to the study. Jan Peterson and Tom McCabe helped with several portions of the field work. An earlier draft of this manuscript was reviewed by Gary Schnell, Robert Selander, Charles Sibley, Robert Storer, and Milton Weller; their comments were a valuable contribution. Sincere thanks to Leslie Ratti and Alan Wentz for their helpful editorial suggestions.

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