Differential predation by mammals and birds: implications for egg-colour polymorphism in a nomadic breeding seabird

GUILLERMO BLANCO[1]* and MARCELO BERTELLOTTI[2]

[1]*Instituto de Investigación en Recursos Cinegéticos (CSIC-UCLM), Ronda de Toledo s/n, 13005 Ciudad Real, Spain*
[2]*Department of Applied Biology, Estación Biológica de Doñana (CSIC), Av. de Mª Luisa s/n, Pabellón del Perú, 41013 Sevilla, Spain*

Selection for crypsis in varying environments has long been established as the main evolutionary force promoting the huge variation in avian egg coloration. In several avian species, variation in egg coloration exists, but available information available on the relative success of these different colour morphs against predation is scarce. We investigated the value of eggshell coloration against mammal and avian predators in the South American Tern, *Sterna hirundinacea*. We found evidence of a relationship between particular eggshell ground coloration and success against predation, in different tern colonies, where strong selection was caused by single avian and mammalian predator species. Survival to hatching of eggs with greenish ground coloration was greater than in eggs of the remaining colours when a mammalian carnivore was present. This implies that the human visual system does not accurately represent predator perception but that, viewed through the predator's eyes, the conspicuous greenish eggs are well concealed. The rate of artificial nest predation by visually searching gulls was higher for eggs more conspicuous to the human eye than for eggs more closely resembling the nest substrate. The evolution of polymorphisms in eggshell ground colour may have resulted from differences in the type of predator present, and differences in choice of breeding site varying in the background substrate. The nomadic breeding behaviour of terns may imply that females differing in the frequency of alleles expressing particular egg coloration, selected for in particular environments, may eventually gather in some colonies, thus producing the observed intracolony variation in egg coloration. We hypothesise that egg colour variation could be maintained in the population by shifting peaks of predation impact in the different locations where colonies form, e.g. islands without mammalian predators vs. mainland sites. © 2002 The Linnean Society of London, *Biological Journal of the Linnean Society*, **75**: 137–146.

ADDITIONAL KEYWORDS: egg-colour polymorphism, predation, South American Tern, *Sterna hirundinacea*.

INTRODUCTION

Natural selection provides some traits with adaptive advantages against predation, exerting a strong influence on the life histories of birds (Ricklefs, 1969; Martin, 1995). Egg predation generally favours crypsis in egg colour, to resemble the coloration of nests and nesting places (Lack, 1968; Oniki, 1985; Solis & de Lope, 1995; Westmoreland & Kiltie, 1996). Interspecific egg coloration is therefore highly variable as eggs are laid on nests constructed from a great variety of materials, or on a variety of nesting sites in those species that lay eggs directly on the substrate (Collias & Collias, 1984). Within species, egg colour and pattern also vary individually (Lack, 1968; Kiff, 1991; Collias, 1993; Westmoreland & Kiltie, 1996). This variation has been suggested to have evolved as a counter-defence against conspecific or interspecific nest parasitism (Rohwer & Freeman, 1989; Soler & Møller, 1996), or to enhance individual egg recognition by adults nesting in dense aggregations (Buckley & Buckley, 1972a,b).

Among colonial birds, there are species where nest parasitism is uncommon, and pairs do not breed close

*Corresponding author. E-mail: gublanco@hotmail.com

enough together to confuse their nests. In such cases, intraspecific eggshell colour variability may be the result of local adaptations to the nesting conditions, namely: (1) the nature and colour of the nest materials or nesting substrate used by different individuals; and (2) the presence of particular nest predators (Lack, 1968). Egg polymorphism may be the result of differential selection for alleles that enhance egg crypsis against predators, because predator impact on a given prey-species should vary among areas with different predator assemblages (e.g. island vs. mainland). Ultimately, the degree of selection for a particular appearance should depend on the magnitude of the selective pressure through predation by different species with potentially different capability of prey detection (Endler, 1984). Information on the mechanisms that reduce nest predation, within the context of predator perception of different egg traits, however, is incomplete (Götmark, 1992).

Predators may find bird eggs by searching for breeding adults, nests or the eggs themselves. The variable impact of egg predators on prey populations is thought to be related to their degree of specialization in detecting eggs, depending on their size, perception and egg-searching strategies. The colour, marks and size of eggs may influence nest predation depending on the type of predator and the searching strategy adopted (Angelstan, 1986; Roper, 1992; Miller & Knight, 1993; Hogstad, 1995). Egg colour may offer a different value against predation by birds vs. mammals because of visual capacity and colour perception differences in these predators (Cornsweet, 1970; Jacobs, 1992). Other egg traits may also be important determinants of survival from predation. For example, odour or palatability of eggs (Cott, 1948, 1954) may offer contrasting advantages against avian vs. mammalian predators due to differences in the sensorial capabilities of these hunters.

Egg coloration could be selectively neutral if nest predators detect nests or incubating parents before detecting eggs (Götmark, 1992). Conversely, egg coloration may be especially relevant against predation in bird species with non-cryptic parents that lay eggs in open ground nests or directly on the bare ground. In the extreme case of highly visible nesting adults, colonial breeding may promote the evolution of mechanisms against predation not related to egg coloration because once the colonies are detected all eggs may be predated (Birkhead & Nettleship, 1995; Craik, 1997). In this case, predation may be independent of the characteristics of eggs, although its impact may depend on the number and size of specific predators acting on the colonies. Alternatively, individual variability of egg colour within the colony may offer differential selective advantages for particular eggs, even once the predators discover the colony.

In this study we investigated the value of specific colours and markings of eggs of colonial South American Terns, *Sterna hirundinacea*, against predation. Among terns, some species exhibit high variability in egg colour and markings (Buckley & Buckley, 1972a,b), which has been explained as enhancing individual egg recognition by adults without involving differential predation (Buckley & Buckley, 1972a). Contrary to other tern species, South American Terns do not nest in high enough densities to confuse their nests (unpubl. data). In addition, to our knowledge, conspecific and interspecific nest parasitism have not been documented in this species, although supernormal clutches due to egg displacement into neighbouring nests by flooding has been recorded (unpubl. data).

Terns, like a number of other seabirds, mainly build their nests and colonies in remote and inaccessible sites, presumably to deter mammalian predators which may destroy entire colonies (Birkhead & Nettleship, 1995; Craik, 1997). Nesting substrates are highly variable in South American and other tern species. This variation may also be high among years for particular individuals as a consequence of their nomadic breeding strategy, i.e. birds frequently shift colony sites between years (Burger, 1984; Visser & Petterson, 1994; Yorio *et al.*, 1998). In different colonies individuals are likely to be subjected to contrasting environmental conditions affecting selection for particular egg colours, including nest substrate and the guild of nest predators. If the need to be concealed from predators has forced the evolution of particular egg colours to resemble the nest background colour, we would expect egg colour variability within species nesting in substrates of variable background colour. By examining the relationships between egg trait variability, predator selection for particular egg traits and nest survival, we specifically tested the null hypothesis that egg pigmentation is neutral with respect to predation. We focused, in particular, on the different value of eggshell ground colour against predation by mammals and birds to explore the causes and consequences of intraspecific egg-colour polymorphism. We predict that different egg colours should have differential selective advantages against different predator species searching for eggs by using their visual capacity in the case of diurnal gulls, and a combination of different methods in the case of mammalian carnivores.

METHODS

QUANTIFICATION OF EGGSHELL AND NEST SUBSTRATE COLOUR

The variation in egg ground colour of South American Terns is sufficiently wide (from white to dark brown)
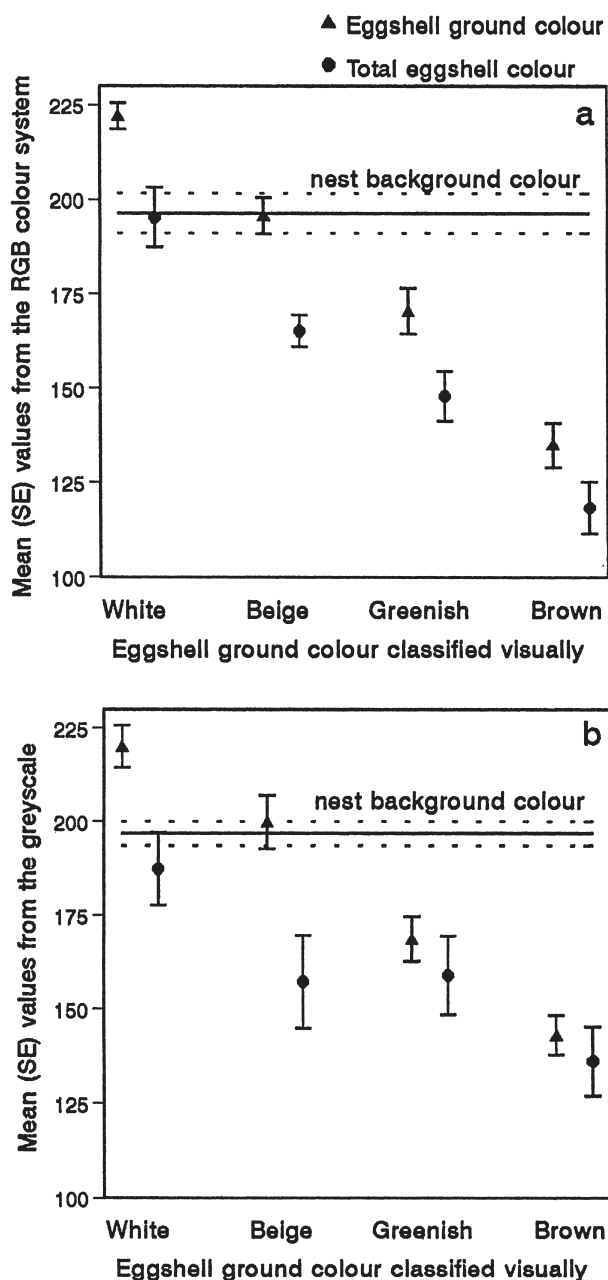
Figure 1. Mean ± SE values from the (A) RGB colour system and the (B) greyscale for South American Tern eggshell ground colours classified visually in relation to the nest background colour. Values for both eggshell ground colour and total eggshell colour including markings are shown.

visual classification of eggshell ground colour was sufficiently discriminative and objective, a sample of eggs from a colony on Punta Loma (42°49′S, 64°28′W), Chubut, Argentina, was photographed directly above nests with a digital camera with 1160×872 pixels of resolution and 256^3 colours. Photographed eggs were first classified into the considered colour categories, and then their images analysed using Adobe Photoshop software. Three 3×3 pixel subsamples of eggshell ground colour ($4.32\,mm^2$) selected at random in each egg were recorded as values in the RGB (red, green, blue) colour system. Mean values of RGB for the three samples of each photographed egg were obtained and tested by ANOVA for differences among the colours assigned visually (Villafuerte & Negro, 1998). We found that our visual classification of eggshell ground colour significantly discriminated egg colour in the blue ($F_{3,29} = 85.2$, $P < 0.001$), green ($F_{3,29} = 68.3$, $P < 0.001$) and red ($F_{3,29} = 14.5$, $P < 0.001$) spectra, and also in the average of the three colours ($F_{3,29} = 57.21$, $P < 0.001$, Fig. 1a). The objective quantification of eggshell ground colour was, therefore, consistent with our visual classification, suggesting that the human visual system is adequate to assess egg colour, at least when the variation in egg pigmentation is high (see also Collias, 1993; Westmoreland & Kiltie, 1996). The same analysis was repeated in the greyscale, given that the predators may have black and white vision. It was also found that eggshell ground colour significantly differed in the greyscale among the colours classified visually ($F_{3,29} = 36.94$, $P < 0.001$, Fig. 1b).

Our classification of egg ground colour, based on increasing pigmentation of eggs, was related to conspicuousness in the light, creamy-white colour of the nesting substrate. Eggs of ground colours equal or similar (white and beige) to the background colour (creamy-white) were presumably imitating the substrate and hence they were assumed to be more cryptic than eggs with greater pigmentation (greenish and brown). We tested this prediction by comparing objective values of the nest background colour (mean values of RGB and greyscale in three 60×60 pixel, i.e. $13.5\,cm^2$, subsamples around the eggs) and the eggshell colour for the sample of photographed nests. We tested eggshell ground colour and overall eggshell colour, including markings, separately. Overall eggshell colour was assessed by obtaining mean values in the RGB and greyscale for three eggshell areas of 60×60 pixels ($4.5\,cm^2$). Superficial egg markings consisted of a highly variable combination of different colours, mostly dark reddish brown, black and grey, that were impossible to categorize visually. We categorized them, however, according to their extent and distribution as: (1) homogeneous (homogeneously distributed small markings rarely forming blotches); (2) polarized (markings mostly concentrated at the larger

to allow visual classification into unambiguous categories (see also Buckley & Buckley, 1972a for *Sterna maxima*). We categorized eggshell ground colour, based on increasing pigmentation, as: (1) white, (2) beige, (3) greenish, or (4) brown. To assess whether our

end of the eggs forming a wreath or a cap); and (3) blotched (marks forming large blotches distributed throughout most of the eggshell surface). To reduce possible biases only one person (GB) categorized egg colour and marks.

DIFFERENTIAL PREDATION BY A MAMMALIAN CARNIVORE AND CLUTCH SURVIVAL

The value of egg colour against mammalian predation was studied in the Punta Loma colony by (1) comparing features of the available eggs in the colony vs. the eggs that were preyed upon, and (2) by assessing survival of marked clutches. In 1997, the colony of 3000 nests was located by following the continental coastline at the top of sedimentary cliffs. Nests were laid in relatively close aggregation (i.e. internest distance ranging from 15–100 cm), and were located in a band 3 m wide from the border of the cliffs and along 500 m of the coastline. Clutches were placed directly onto the rocky substrates and sediments (sand and small stones) of the same geological composition, and a unique light creamy-white colour. Nests were reduced to small depressions in the sediments, and contrary to other related species, South American Terns in the studied colony did not modify the colour of the nest substrate by adding small vegetation pieces, debris, molluscs shells, etc. No colony, concentration of gulls or other avian predators existed in the area (Yorio *et al*., 1998; pers. observ.).

We compared features of a sample of available eggs in the colony against eggs which were preyed upon previously and whose eggshells were collected during December 1997 and January 1998. Those eggshells had been collected, within the same area and in an area of ~3 ha adjacent to the colony, by searching during 12 days of the incubation period. The distinctive signs found on predated eggs (a small hole in the middle of the egg and marks of two canine teeth around the hole) suggest that the predator was a small to medium sized carnivore, probably Little Grison (*Galictis cuja*), a diurnal and nocturnal hunter (Blanco *et al*., 1999). Several individual predators acted simultaneously in the colony, as indicated by the high minimum number of eggs consumed daily, and the distant places where clumped eggshells of predated eggs were found (unpubl. data).

Eggshell ground colour and mark patterns were categorized visually as described above. Length and maximum breadth of eggs selected at random at the colony and eggs preyed upon, whose shape was intact, were measured with vernier callipers; egg volume was determined using the formula: volume = 0.51 × length × width^2 (Hoyt, 1979). To investigate the relationships between categorized egg features (colour and marks) and egg status (preyed upon vs. successful to hatch-

ing) we conducted chi-square tests. We assessed the difference in volume between preyed upon and successful eggs with a *t*-test. We evaluated the possibility of interactions between egg traits and egg status by means of a non-hierarchical fully saturated loglinear analysis. This procedure resembles analysis of variance in that total variance of frequency data is partitioned into different factors, making it possible to test for interactions. The test was run starting with the highest order interaction and proceeding backwards until all terms/interactions retained by the model reached significance ($P < 0.05$), so that dropping any of them would result in a significant lack-of-fit of the model.

To assess whether eggs of particular eggshell ground colour were predated more or less frequently than expected from their availability, we analysed the selection of egg features by means of the Savage selectivity index (Manly *et al*., 1993). This index is defined as $w_i = U_i/p_i$, where U_i is the proportion of each category of each trait within the group of eggs that were preyed upon and p_i is the proportion of eggs within that category within a random sample of recently laid eggs. This selectivity index varies from zero (0) (maximum negative selection) to infinite (maximum positive selection), one (1) being the central value defining the value expected by chance. We tested the statistical significance of this index by comparing the statistic ($w_i - 1)^2/se(w_i)^2$ with the corresponding critical value of a chi-squared distribution with one degree of freedom (Manly *et al*., 1993). We calculated the standard error of the index [$se(w_i)$] by $\sqrt{\{(1 - p_i)/(u_t \times p_i)\}}$, considering the null hypothesis that predators are selecting particular eggshell ground colour in proportion to availability, u_t being the total number of eggs preyed upon that were recorded. Eggs selected at random at the colony were sampled before the start of the regular collection of eggshells from predated eggs. This coincided in time with the laying peak in the colony, when the predators have already predated on eggs. The eggshells from predated eggs found before the assessment of colour and volume of the sample of random eggs were not considered in the selection analysis, although they were used to assess seasonal changes in the predation of each egg colour since the establishment of the colony. Our samples of available and subsequently predated eggs, therefore, were from the same statistical population.

To test if a spatio-temporal distribution pattern of clutches of particular colours existed, we assessed the colour in a sample of eggs from nests located close (<1.5 m) and far (>2 m) from the cliff border. This sampling allowed us to assess temporal and spatial distribution patterns of egg colours simultaneously, as first-laying pairs began to establish nests along the cliff border while pairs nesting later laid progressively

farther from the cliff border (unpubl. data), probably as an attempt to avoid terrestrial predation.

Clutch survival was assessed by marking 110 nests with small numbered sticks, in two nest patches that were visited, and non-visited, respectively. Nests were selected at random in relation to the colony edge and clutch size (85 of one egg, 24 of two eggs, and one of three eggs). Nests were checked at intervals varying from two to seven days (mean, 4 days) in the visited patch (75 marked nests). The non-visited patch (35 marked nests) was checked 21 days after nest marking (an interval equal to the incubation period) to document nest outcome, minimizing possible disturbance to the terns and cues for predators. Nests in the non-visited patch were used as controls to assess the potential influence of the investigator in the hatching success, and were not included in the analyses. The probability of a nest being successful, however, was not related to the frequency of visits to the nests, as assessed by comparing nest survival and daily clutch survival rates of visited vs. non-visited nests (Blanco *et al.*, 1999).

We used the Mayfield method (Mayfield, 1975) as modified by Johnson (1979) to estimate daily survival probabilities, and associated standard errors of clutches. We scored nests as either successful (producing one or more hatchlings), or failed (producing no hatchlings). We conducted comparisons among clutches with different egg features using the variance estimate for daily survival probability and hypothesis testing at $P = 0.05$ according to Hensler & Nichols (1981). Criteria for inclusion of a nest in a sample varied depending upon the analysis because data on egg features (e.g. colour, volume) and clutch survival were not recorded in all cases. In addition, several of the numbered sticks used to mark the nests were lost during the study. We only considered nests with one egg, or nests with two eggs of the same ground colour from the visited patch ($N = 52$), to avoid confusion between clutch survival and the survival of eggs of particular colours. We could not assess the value of different egg colour within clutches because of the low sample size. Sample sizes varied among analyses.

EXPERIMENTAL TEST OF EGGSHELL GROUND CRYPSIS AGAINST AVIAN PREDATION

To test the value of eggshell ground colour against avian predation, we conducted an experiment using South American Tern eggshells, previously predated by a mammalian carnivore in the Punta Loma colony (see above). We selected predated eggs with intact shape and filled them with a flour paste through the small hole left by the predator. Experimental eggs were placed on Vernaci Sudoeste Island (45°11′S, 66°31′W), San Jorge Gulf, Argentina, during December 1998. This island of 6.4 ha is located >600 m away from the mainland and does not hold mammalian predators. On the contrary, the island holds a colony of ~6500 breeding pairs of Kelp Gull, *Larus dominicanus* (Yorio *et al.*, 1998), a common predator of tern eggs and chicks. Such a large gull colony may impose an unrealistically strong predation pressure on artificial nests not attended by parents. The aim in designing this experiment, however, was not provide accurate estimates of predation rates comparable to those of real nests, but to assess differences in the value of variable egg ground colours against avian predation.

We selected 14 eggs of each eggshell ground colour (white, beige, greenish and brown) and a similar pattern of markings (homogeneous), and placed them among natural nests of South American Terns, which formed a small (about 20 pairs) and loose colony. The background substrate of the natural and experimental colonies consisted of a beach of small white fragments of mollusc shells discoloured by the sun, a bright white background substrate colour. We further standardized the white colour of the nest substrate by placing the experimental eggs in a depression made on the white layer of molluscs shells, avoiding remains of algae and large stones producing shadows. Experimental nests of one egg each (the modal clutch size) were distributed on a line parallel to the coastline and ~20 m away from the border of the gull colony. Eggs were separated from each other by a distance of 3 m, and were always ordered in the same sequence of eggshell ground colour, i.e. brown, beige, white, and greenish. The content of experimental nests was monitored one hour, one day and two days after their placement. A nest was considered as predated if the egg had disappeared or if gull bill-marks were found on the egg. The relationship between egg conspicuousness, as predicted by the increasing eggshell ground colour of experimental eggs on a white nesting substrate, and egg survival to predation (the minimum number of hours surviving to predation 1, 24 or 48 h after placement) was analysed with one-tailed Kendall rank order correlation due to the high frequency of tied data (Zar, 1984).

RESULTS

EGG CRYPSIS

The colour of the background nest substrate was similar for eggs classified visually in different eggshell ground colour categories, both in the RGB ($F_{3,29} = 0.67$, $P = 0.58$) and the greyscale ($F_{3,29} = 0.82$, $P = 0.50$), due to the same geological composition of the substrate in the colony where eggs were photographed (Punta Loma). However, the eggshell colour of South American

Table 1. Percentage of eggs of each ground colour available vs. preyed upon by a mammalian carnivore in Punta Loma colony. Values of w_i refer to the Savage selectivity index. The statistical significance of this index was tested by comparing an associated statistic (see Methods) with the corresponding critical value of a chi-square distribution with one degree of freedom, considering the null hypothesis that predators are selecting particular eggshell ground colour in proportion to availability

Egg colour	Preyed upon ($N = 319$)	Available ($N = 128$)	w_i	χ^2	P
White	17.6	15.6	1.13	0.17	0.680
Beige	37.9	31.3	1.21	2.45	0.118
Greenish	20.4	35.9	0.56	6.79	0.009
Brown	24.1	17.2	1.40	2.57	0.119

Tern eggs was variable enough to allow visual classification consistent with objective colour classification (see Methods and Fig. 1). Egg conspicuousness increased when egg ground colour pigmentation was high in relation to substrate background colour in Punta Loma. Eggs classified as beige mimicked substrate background colour in the RGB (t-test $t = 0.08$, $P = 0.94$, d.f. = 35), while eggs classified as white, greenish and brown differed significantly from the background colour ($t = 4.90$, $P < 0.001$ d.f. = 35, $t = 4.31$, $P < 0.001$, d.f. = 34 and $t = 11.07$, $P < 0.001$, d.f. = 36, respectively, Fig. 1a). Overall eggshell colour (including markings) showed a similar pattern of increasing conspicuousness: white eggs mimicked the background colour assessed in the RGB system ($t = 1.66$, $P = 0.87$, d.f. = 35, Fig. 1a), while beige, greenish and brown eggs increasingly contrasted with the nest background colour (beige: $t = 4.83$, $P < 0.001$, d.f. = 32; greenish: $t = 7.94$, $P < 0.001$, d.f. = 34; brown: $t = 13.1$, $P < 0.001$, d.f. = 36; Fig. 1a). Similar results were found when comparing values from the greyscale for both eggshell ground colour and overall eggshell colour (including markings) vs. nest background colour (Fig. 1b).

DIFFERENTIAL PREDATION BY A MAMMALIAN CARNIVORE AND CLUTCH SURVIVAL

Eggs that hatched and those preyed upon by a mammalian carnivore did not differ in marking pattern ($\chi^2_2 = 1.66$, $P = 0.44$) or volume (t-test $t = 1.17$, $P = 0.24$, $N = 277$), but they were different in eggshell ground colour ($\chi^2_3 = 9.48$, $P = 0.023$). In addition, the pattern of marks was associated with eggshell ground colour; eggs with light ground colours were also less pigmented in relation to superficial markings than dark ground colour eggs (Spearman rank correlation $r_s = 0.26$, $P < 0.00001$, $N = 473$). The relationship, however, between colour and egg status (successfully hatched vs. preyed upon) was not confounded by the covariation of egg colour and marks: a loglinear analysis showed that ground colour of eggs was independently linked to both superficial markings ($G = 43.44$, d.f. = 6, $P < 0.001$) and egg status ($G = 8.42$, d.f. = 3, $P = 0.038$). The three–way interaction and the interaction between marks and status were not significant ($G = 6.24$, d.f. = 6, $P = 0.39$ and $G = 1.45$, d.f. = 2, $P = 0.48$, respectively), the fit of the model being adequate (goodness of fit: $G = 6.23$, d.f. = 6, $P = 0.40$).

The analysis of the selection of egg ground colour revealed that the predator significantly and negatively selected (avoided) greenish eggs while eggs of other colours were preyed upon in proportion to their availability in the colony (Table 1). No eggshell ground colour was positively preferred by the predator, but greenish eggs were negatively selected.

Egg predation occurred since the first pairs established in the colony. This was probably because the predators had previous knowledge of the location of the abundant food source, as they had already preyed upon many eggs during the previous breeding season (author's pers. observ.). No seasonal trend was apparent, however, in the consumption of each egg colour when considering each day of eggshell collection ($\chi^2_{30} = 23.52$, $P = 0.79$). The distribution of colours of eggs predated before and after the beginning of regular removing of eggshells was compared, in order to assess seasonal changes in the predation of each egg colour since the establishment of the colony. The result was not significant, although the trend was that greenish eggs were predated more frequently at the end (24.0%, $N = 179$) than at the beginning (15.7%, $N = 140$) of the incubating period ($\chi^2_1 = 2.8$, $P = 0.09$ with Yates' correction). The distribution of clutches of different ground colour did not differ between early laying pairs nesting near to the cliff border and pairs laying later and farther from the cliff border ($\chi^2_3 = 2.77$, $P = 0.43$). Furthermore, the predators consumed clutches located throughout the colony.

Monitoring of marked clutches revealed that the percentage of nest losses due to mammalian predation

Table 2. Daily probabilities and associated standard errors of clutches surviving to predation by a mammalian carnivore in relation to egg ground colour in South American Terns in Punta Loma. Survival was assessed for the incubation period using the Mayfield method (Mayfield, 1975; Johnson, 1979)

Egg colour	Number of nests	Number of nest-days	Daily survival rate ± SE	% of losses
White	8	130.0	0.977 ± 0.013	37.5
Beige	18	271.5	0.974 ± 0.009	38.9
Greenish	18	337.0	0.994 ± 0.004	11.1
Brown	8	120.5	0.959 ± 0.018	62.5
Combined*	93	1336.0	0.968 ± 0.005	38.7

*Includes nests where egg colour was not recorded
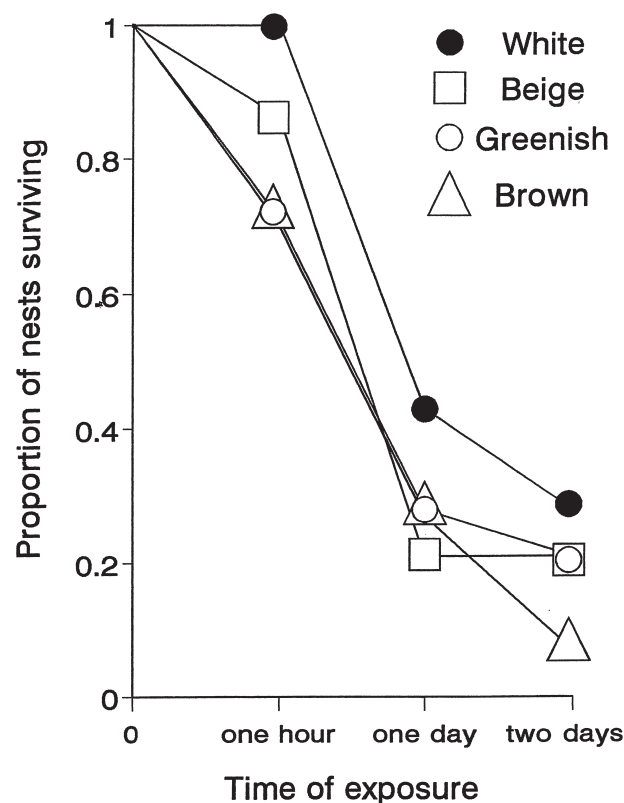
Figure 2. Proportion (%) of experimental nests of South American Terns surviving to predation by Kelp Gulls in relation to eggshell ground colour and time of exposure.

during incubation ranged between 11.1% (green eggs) and 62.5% (brown eggs; Table 2). Daily nest survival rates did not differ among nests with eggs whose colour (white, beige and brown) was not selected by the predator (see above; Hensler & Nichols test, all $P > 0.21$, Table 2). Nests with greenish eggs had a daily survival rate significantly higher than nests with beige (Hensler & Nichols test, $z = 1.916, P = 0.029$) and brown eggs ($z = 1.883, P = 0.030$). The difference was not significant when compared with nests with white eggs ($z = 1.237, P = 0.109$). To increase the power of the test according to Hensler & Nichols (1981), we compared survival of nests with eggs of negatively selected colours (greenish) and survival of nests with eggs whose ground colour were preyed upon in proportion to availability (white, beige and brown). Overall, nests with greenish eggs had a higher survival and a daily survival rate than nests with eggs of other colours (nest survival: $\chi_1^2 = 4.42, P = 0.035$ with Yates' correction; daily survival rate: Hensler & Nichols test, $z = 2.74, P = 0.0031$).

EXPERIMENTAL PREDATION BY GULLS

The proportion of experimental eggs surviving gull predation decreased with increasing exposure time in the colony (Fig. 2). Egg survival to predation, calculated as the minimum number of hours surviving to predation, decreased with increasing eggshell pigmentation as predicted by their increasing contrast against the substrate (i.e. conspicuosness increasing from white to brown; one-tailed Kendall tau = −2.59, $P = 0.00016, N = 56$). Eggs with white eggshell ground colour were the less predated while brown eggs were predated the most (Fig. 3).

DISCUSSION

Selection for crypsis in varying circumstances has been long established as the main evolutionary force promoting the high interspecific variation in avian egg coloration (Lack, 1968; Oniki, 1985; Westmoreland & Kiltie, 1996). The same process may be present within species, as individuals may be exposed to different environmental features such as the available nest substrate and the predator guild present in different areas (Lack, 1968), although this possibility has received scarce attention. In this study, evidence was found for different values of particular egg ground colours against predation in South American Tern
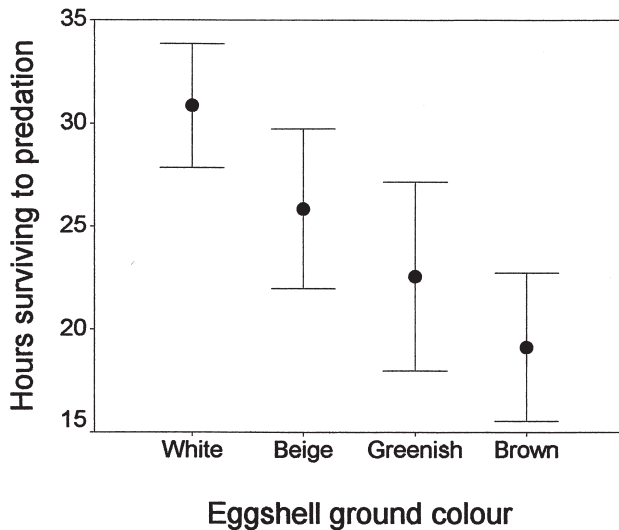
Figure 3. Mean ± SE minimum number of hours surviving to gull predation of experimental eggs of South American Terns in relation to increasing eggshell ground colour and predicted increasing contrast against the substrate (i.e. conspicuousness increasing from white to brown). Sample size is 14 eggs for each eggshell ground colour.

colonies, where strong selective predation pressure was caused by single avian and mammalian predator species.

The results suggest that greenish egg ground colour may offer a selective advantage against mammalian predation even under conditions of high breeding density and intense predation. The mechanism making greenish eggs more competitive against predation may be related directly to the greenish eggshell colour, which may be making eggs inconspicuous to predators. This implies that the human visual system does not accurately represent predator perception but that, viewed through the predator's eyes, the conspicuous greenish eggs are well concealed. These results agree with that of Westmoreland & Kiltie (1996), who found that a brighter egg phenotype from the human view had higher survival than more cryptic eggs in blackbirds (Icteridae). Ultraviolet vision of the predator may explain the differences in predation upon greenish eggs instead of eggs of other ground colours if different pigments in the eggshell (Kennedy & Vevers, 1975) make greenish eggs more concealed through the ultraviolet reflected. This possibility remains untested. Alternatively, a trait correlated with egg ground colour could explain the reduced predation against greenish eggs. Egg content or composition may differ providing green eggs with a particular flavour or odour conditioning its palatability (Cott, 1948; Dumbacher & Pruett-Jones, 1996). If greenish eggs are unpalatable they should be avoided over

palatable eggs when these are abundant, unpalatable prey may be consumed when the predator is hungry (Dumbacher & Pruett-Jones, 1996). This possibility remains untested, but it is in agreement with the hypothesis that the conspicuousness of eggs may be negatively correlated with their palatability, as Cott suggested after studying the preferences of cats, ferrets, mongooses, hedgehogs, rats and humans, used as tasters (Cott, 1954 and papers cited therein). Conversely, we found that the rate of artificial nest predation by visual searching gulls was higher for eggs more conspicuous to the human eye that for eggs apparently resembling the nest substrate. This suggests that eggshell coloration resembling the background substrate may be advantageous to eggs subjected to predation by visual searchers such as gulls.

Different egg ground colours provided different values against predation by single avian and mammalian predator species acting in two different colonies, where each other predator was absent. Other eggshell colours, superficial marks or a particular combination of these traits, and possibly other traits correlated with them, may offer advantages against predation caused by the same predators preying on eggs laid on different background substrates. In addition, other predator species with different visual and other sensory capabilities, including taste, or differing in size and abundance (and thus in the impact they cause) may impose a different pressure on eggs of variable colours. South American Terns nest in substrates of very different nature, including beaches of sand, stones, mollusc shells and on rocky cliffs and small islands with or without vegetation, and therefore the colour of the nesting substrate is also highly variable (i.e. from white to dark-brown; pers. observ.). In their variable nesting sites, terns may encounter different predator assemblages or particular predator species, which may exert a variable predation pressure depending on their abundance, size, and methods of searching for eggs. Approximately half (48%) of the known colonies of this species in the Atlantic coast of South America ($N = 42$) were located in remote islands without mammalian predators (Yorio et al., 1998; unpubl. data), where avian predators, especially gulls, may have been the main selective agents for egg crypsis, at least in ecological time. In fact, 75% and 23% of South American Tern colonies were, respectively, located on islands ($N = 20$) and mainland sites ($N = 22$) where breeding colonies of Kelp Gulls and/or other species of avian predators exist also (Yorio et al., 1998; unpubl. data).

Egg-colour polymorphism may reflect differences in frequencies of the alleles that code for the enzymes controlling the pigments deposited in the eggshell (Washburn, 1990; Collias, 1993). Females laying the

most cryptic eggs in relation to both the nesting substrate and the perception of eggs by particular predators should have higher fitness. Thus, eggshell ground colour may become polymorphic because different alleles for different colours have been selected for in different nesting environments varying in background substrate features and predation pressure. If eggshell coloration is heritable and constant throughout the life of particular females, as it occurs in some species showing egg colour polymorphism (Jensen, 1966; Washburn, 1990; Collias, 1993), selection for varying egg coloration may take place in different nesting locations differing in the degree to which selective factors constrain egg survival.

South American Terns share with other colonial terns the habit of switching nesting sites between years (Yorio *et al*., 1998; unpubl. data). This breeding strategy has been explained by the ability of terns to track the availability of food, concentrating in areas where aquatic productivity is high, but not to reduce predation, although predation may cause the abandonment of particular colonies or areas between years (Yorio *et al*., 1998; pers. observ.). This nomadic behaviour may imply that females differing in the frequency of alleles expressing particular egg coloration selected in particular circumstances may eventually gather in some colonies, then producing the observed intracolony variation in egg coloration. Egg colour variation, therefore, could be maintained in the population by shifting peaks of predator impact in the different locations where colonies form; e.g. islands without mammalian predators vs. mainland sites. In this scenario, selection for crypsis has been the most likely factor directing the evolution of egg-colour polymorphism in metapopulations of South American Terns, where individuals are subjected to variable environmental conditions regarding both food and predator dynamics. More research is needed on the possibility that intraspecific eggshell-colour polymorphism may have evolved as consequence of the different value of each egg colour against predation in varying circumstances.

REFERENCES

Angelstan P. 1986. Predation on ground nesting birds' nests in relation to predator densities and habitat edge. *Oikos* **47:** 365–373.

Birkhead TR, Nettleship DN. 1995. Arctic fox influence on a seabird community in Labrador: a natural experiment. *Wilson Bulletin* **107:** 397–412.

Blanco G, Yorio P, Bertellotti M. 1999. Effects of research activity on hatching success in a colony of South American Terns. *Waterbirds* **22:** 148–150.

Buckley FG, Buckley PA. 1972a. The breeding ecology of Royal Terns *Sterna* (*Thalaseus*) *maxima maxima. Ibis* **114:** 344–359.

Buckley FG, Buckley PA. 1972b. Individual egg and chick recognition by adult Royal Terns (*Sterna maxima maxima*). *Animal Behaviour* **20:** 457–462.

Burger J. 1984. Colony stability in Least Terns. *Condor* **86:** 61–67.

Collias EC. 1993. Inheritance of egg-color polymorphism in the village weaver (*Ploceus cucullatus*). *Auk* **110:** 683–692.

Collias NE, Collias EC. 1984. *Nest building and bird behavior*. Princeton: Princeton University Press.

Cornsweet TN. 1970. *Visual perception*. New York: Academic Press.

Cott HB. 1948. Edibility of the eggs of birds. *Nature* **161:** 8–11.

Cott HB. 1954. The palatability of eggs and birds: mainly based upon observations of an egg panel. *Proceedings of the Zoological Society of London* **124:** 335–463.

Craik C. 1997. Long-term effects of North American mink *Mustela vison* on seabirds in western Scotland. *Bird Study* **44:** 303–309.

Dumbacher JP, Pruett-Jones S. 1996. Avian chemical defense. *Current Ornithology* **13:** 137–174.

Endler JA. 1984. Progressive background matching in moths, and a quantitative measure of crypsis. *Biological Journal of the Linnean Society* **22:** 87–231.

Götmark F. 1992. Blue eggs do not reduce nest predation in the song thrush, *Turdus philomelos. Behavioural Ecology and Sociobiology.* **30:** 245–252.

Hensler GL, Nichols JD. 1981. The Mayfield method of estimating nesting success: a model, estimators and simulation results. *Wilson Bulletin* **93:** 42–53.

Hogstad O. 1995. Do avian and mammalian nest predators select for different nest dispersion patterns of Fieldfares *Turdus pilaris*? A 15-year study. *Ibis* **137:** 484–489.

Hoyt DF. 1979. Practical methods of estimating volume and fresh weight of bird eggs. *Auk* **96:** 73–77.

Jacobs GH. 1992. Ultraviolet vision in vertebrates. *American Zoologist* **32:** 544–554.

Jensen RAC. 1966. Genetics of cuckoo egg polymorphism. *Nature* **209:** 827.

Johnson DH. 1979. Estimating nest success: the Mayfield method and an alternative. *Auk* **96:** 651–661.

Kennedy GY, Vevers HG. 1975. A survey of avian eggshell pigments. *Comparative biochemistry and physiology.* **55:** 117–123.

Kiff L. 1991. The eggs came first. *Terra* **30:** 5–19.

Lack D. 1968. *Ecological adaptations for breeding in birds.* London: Methuen.

Manly BFJ, McDonald LL, Thomas DL. 1993. *Resource selection by animals*. London: Chapman & Hall.

Martin TE. 1995. Avian life history evolution in relation to nest sites, nest predation, and food. *Ecological Monographs* **65:** 101–127.

Mayfield HF. 1975. Suggestion for calculating nest success. *Wilson Bulletin* **87:** 456–466.

Miller CK, Knight RL. 1993. Does predator assemblage affect reproductive success in songbirds? *Condor* **95:** 712–715.

Oniki Y. 1985. Why robin's eggs are blue and birds build nests: statistical tests for Amazon birds. *Ornithological Monographs* **36:** 536–545.

Ricklefs RE. 1969. An analysis of nesting mortality in birds. *Smithsonian Contribution to Zoology* **9:** 1–48.

Rohwer FC, Freeman S. 1989. The distribution of conspecific nest parasitism in birds. *Canadian Journal of Zoology* **67:** 239–253.

Roper JJ. 1992. Nest predation experiments with quail eggs: too much to swallow? *Oikos* **65:** 528–530.

Soler JJ, Møller AP. 1996. A comparative analysis of the evolution of variation in appearance of eggs or European passerines in relation to brood parasitism. *Behavioural Ecology* **7:** 89–94.

Solis JC, de Lope F. 1995. Nest and egg crypsis in the ground-nesting Stone Curlew *Burhinus oedicnemus*. *Journal of Avian Biology* **26:** 135–138.

Villafuerte R, Negro JJ. 1998. Digital imaging for colour measurement in ecological research. *Ecology Letters* **1:** 151–154.

Visser JM, Petterson GW. 1994. Breeding populations and colony site dynamics of seabirds nesting in Louisiana. *Colonial Waterbirds* **17:** 46–152.

Washburn KW. 1990. Genetic variation in egg composition. In: Crawford RD, ed. *Poultry breeding and genetics*. Amsterdam: Elsevier Publishing Co., 781–804.

Westmoreland D, Kiltie RA. 1996. Egg crypsis and clutch survival in three species of blackbirds (Icteridae). *Biological Journal of the Linnean Society* **58:** 159–172.

Yorio P, Frere E, Gandini P, Harris G. 1998. *Atlas de la Distribución Reproductiva de Aves Marinas en el Litoral Patagónico Argentino*. Puerto Madryn: Fundación Patagonia Natural.

Zar JH. 1984. *Biostatistical analysis*. London: Prentice Hall.