never seen in untreated recipients. This crisis seemed to indicate an abortive homograft response (*3*).

We have since performed similar tests with C3H/HeJ mice, and in this case 6 of 15 splenectomized females rejected C3H/HeJ male skin grafts. Male grafts on the remaining 9 splenectomized females underwent a crisis before being finally accepted (Table 1). All the controls, whether untreated or sham-splenectomized, retained their grafts throughout the entire period of observation, more than 300 days. The difference between these results and those described earlier is probably attributed to immunological differences between the two C3H sublines.

It is not clear why splenectomy converts an ineffective or abortive homograft reaction to a fully implemented rejection. Splenectomy did not dramatically reduce the titers of demonstrable anti-H-Y in the serums of grafted females (*3*). The work by Streilein and Wiesner (*7*) on splenectomy in strains of mice that reject male skin showed that removal of the spleen caused accelerated rejection of male grafts. Furthermore, the reintroduction of spleen cells into such mice abrogated the effects of splenectomy on graft survival. Because the spleen is an abundant source of certain types of suppressor cells, cell-mediated suppression offers a likely hypothesis for further study in the H-Y homograft system.

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Avian Eggs: Thermoregulatory Value of Very High Near-Infrared Reflectance

Abstract. *Studies of the spectral reflectance of the eggs of 25 species of birds from nine families disclosed uniformly high reflectance (often above 90 percent) in the near infrared. This property is associated with the presence of the eggshell pigments protoporphyrin and the bilins. These pigments allow coloration for cryptic or other purposes with minimum solar heating, a combination not possible with the melanin pigments typical of vertebrates.*

Most ground-nesting birds utilize exposed nest sites. If they leave their eggs unattended, the clutch may be lost either to visual predators or to overheating by solar radiation. These two causes of death exert opposing selective pressures on eggshell pigmentation. A disruptive pigment pattern of brown, gray, and olive spots appears to provide cryptic protection against visual predators (*1*) for many species (*2*). However, heating by absorbed solar radiation is increased if the highly reflective (70 to 95 percent, depending on wavelength) shell is covered with dark pigment (*3*). The avian embryo is injured or killed by temperatures more than a few degrees above normal incubation values (*4, 5*), and so any increase in solar heating can be significant. Field investigators have found that overheating by solar radiation is a serious threat to the survival of unattended eggs (*5, 6*). Thus, selection due to predation appears to favor pigmentation, whereas selection resulting from solar heating appears to oppose pigmentation.

We have found that this conflict is minimized by eggshell pigments that absorb only visible radiation. The cryptic pigmentation thus does not significantly reduce the extremely high intrinsic near-infrared (near-IR) reflectance of eggshell (> 90 percent). Solar heating is thus minimized, because nearly half of the incident solar energy is in the near-IR. This result is not a unique freature of the cryptic eggs of ground-nesting birds; the bright blue eggs of some tree-nesting species also show high reflectance in the near-IR. Our absolute spectral reflectance (*7*) measurements, together with a sea-level solar spectrum (*8*) for comparison, are shown in Fig. 1.

The contrast between eggshell pigmentation and the typical gray to brown melanin pigmentation of vertebrates is illustrated in Fig. 1A. Note the spectacular enhancement of the near-IR reflectance of the eggs of Heermann's gull, *Larus heermanni* (*9*), compared to the melanin-pigmented adult feathers. The near-IR reflectance of the eggs exceeds 90 percent, even for the dark brown spots. The difference is entirely due to the absorption characteristics of melanin (*10*) versus the absorption characteristics of protoporphyrin and the biliverdin egg pigments (*11*). Figure 1B shows that the structure and index of refraction of unpigmented eggshell and feathers result in comparable near-IR reflectance.

The details of the spectral absorptance of an egg can significantly influence its cryptic value and the amount of solar radiation absorbed. Spectral absorptance can be affected by the amounts of the three principal egg pigments present [brown protoporphyrin, blue biliverdin IXa, and its green zinc chelate (*11*)] and possibly by the structure of the eggshell. Accordingly, we surveyed the eggs of 24 additional species of birds (*12*) to (i) determine the spectral reflectance of eggshell pigments in situ and (ii) determine whether high near-IR reflectance is a general property of avian eggs.

The curves for eggs of Heermann's gull (Fig. 1A) appear to be representative of protoporphyrin in situ. The curve for eggs of the tree-nesting American robin, *Turdus migratorius* (Fig. 1C), is representative of pure biliverdin IXa (*13*). The spectral reflectance of the brown-spotted blue eggs of the tree-nesting common crow, *Corvus brachyrhynchos* (Fig. 1C), shows the additive effect of combined pigments. No suitable specimens of blue-green eggs containing primarily the zinc chelate of biliverdin were available. Blue eggs show a transition to near-IR reflectance that is more rapid and occurs at a shorter wavelength than in reddish-brown eggs. This is the opposite of the published absorption curves of biliverdin and protoporphyrin in chloroform (*11*). The difference is apparently due to solvent effects and the great thickness of protoporphyrin deposits.

We examined cryptic eggs of 12 ground- or cliff-nesting larid species (including *L. heermanni*) and seven non-larid ground-nesting species to determine the generality of high near-IR reflectance (the species are listed in the legend to Fig. 1). The reflectance curves of normal brown-spotted eggs of most species fall within the stippled region in Fig. 1C. Little variation is evident in the near-IR. No obvious trends with latitude are evident. There are, however, consistent differences between some taxa. For example, the eggs of the ring-billed gull, *L. delawarensis*, are consistently light (reflectance, 90 to 92 percent in the near-IR), whereas the eggs of the herring gull, *L. argentatus*, nesting in the same colony, are consistently darker (reflectance, 82 to 85 percent). By far the lowest near-

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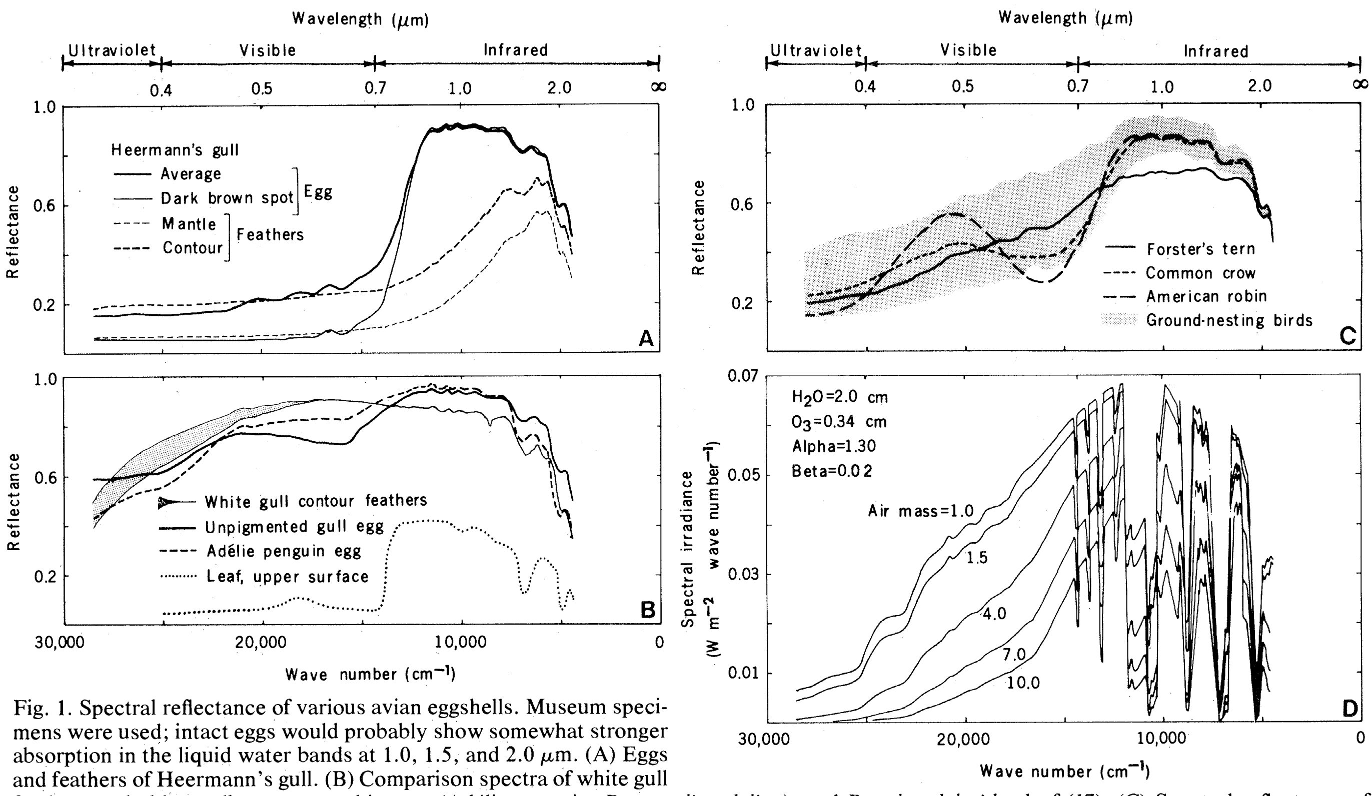

Fig. 1. Spectral reflectance of various avian eggshells. Museum specimens were used; intact eggs would probably show somewhat stronger absorption in the liquid water bands at 1.0, 1.5, and 2.0 μm. (A) Eggs and feathers of Heermann's gull. (B) Comparison spectra of white gull feathers, pale blue gull egg, pure white egg (Adélie penguin, *Pygoscelis adeliae*), and *Populus deltoides* leaf (*17*). (C) Spectral reflectance of assorted eggs. Ground-nesting birds that we studied included *Larus argentatus, L. canus, L. delawarensis, L. glaucescens, L. glaucoides, L. occidentalis, Rissa tridactyla, Sterna sandvicensis, S. anaethetus, S. albifrons, Catharacta maccormicki, Bartramia longicauda, Actitis macularia, Catoptrophorus semipalmatus, Charadrius vociferus, Recurvirostra americana, Himantopus mexicanus*, and *Coturnix c. japonica*. The blue eggs of the tree-nesting white-faced ibis, *Plegadis chihi*, and a brown-spotted blue egg of the cliff-nesting common murre, *Uria aalge*, gave reflectance curves similar to the blue eggs of the American robin and the brown-spotted blue eggs of the common crow. (D) Solar spectral irradiance at sea level (*8*) on a clear, dry day for various air masses for comparison with spectral absorptance. The air mass is approximately equal to the secant of the zenith angle of the sun; thus, air mass = 1 when the sun is directly overhead.

IR reflectance was found in the eggs of Forster's tern, *Sterna forsteri* (reflectance, 70 to 73 percent) (Fig. 1C). Perhaps this result is associated with the heavily vegetated nesting site used by Forster's tern. Further study of latitude and habitat correlations is necessary, since our sample size (usually three eggs selected visually as light, average, and dark examples of each species) was too small to permit precise conclusions. There can be considerable variation in the eggshell pigmentation of a species. The pigment patterns of spotted eggs, and the tendency of pigment to concentrate around the blunt pole, where measurements are most easily made, are also sources of variation. Further study should be based on a statistically adequate random sample of fresh material to accurately define means and variances in the eggshell pigmentation of a species.

The considerable thermoregulatory advantage of shell pigments that allow high near-IR reflectance can best be illustrated in terms of a comparison of the heating rate of unattended protoporphyrin- and hypothetical melanin-pigmented eggs of comparable cryptic value. For this comparison, we selected pairs of spectral reflectance curves for the eggs and adult feathers of Heermann's gull which matched in average visible reflectance (Fig. 1A). The effective solar absorptances (*14*), computed on the basis of the spectral irradiance curve with air mass = 1.5 (Fig. 1D) (*8*), are as follows: egg, 0.499; dark spot, 0.588; contour feathers, 0.655; and mantle, 0.813. Use of a visually equivalent amount of the feather melanin as the egg pigment would thus increase the solar heat load 31 to 38 percent, depending on the degree of pigmentation. Evaporative cooling of the egg is negligible, and heat production by the embryo is insignificant until late in development. Thus the equilibrium temperature of the egg is equal to the operative environmental temperature, T_e (*15*). On a warm, clear April day in the Heermann's gull breeding colony (*9*), air temperatures at egg height average 28° to 30°C, whereas the T_e of copper-lined eggs (*16*) in full sun ranges from 40° to 45°C and can reach 50°C. A 31 to 38 percent increase in the solar radiation absorbed by these eggs will increase T_e at least 3°C. One can calculate the length of time that an egg can be left safely in full sun (*15*) by assuming that the egg is initially exposed at 37°C (within the range observed around noon) and that the threshold for heat injury is 43°C (*4, 5*). When T_e = 45°C, the egg will reach 43°C in 36 minutes, but a hypothetical 3°C increase in T_e, resulting from substituting melanin for protoporphyrin as the egg pigment, reduces this time to 20 minutes. Differences of this order would appear to be of selective significance. Similar overheating can occur at much higher latitudes (*5*), so that this argument is quite general.

Blue and blue-green bilin pigments confer a similar reduction in solar heating, compared to the typical vertebrate blue formed by a Tyndall scattering layer over melanin. Since herons, ibis, and other large birds that lay blue eggs often use nest sites exposed to the sun, the high near-IR reflectance can be ecologically significant. Why their eggs are pigmented at all is less clear.

Spectral characteristics similar to the visible absorptance and near-IR reflectance of pigmented eggshells are known only in green vegetation (*17*), which must absorb radiation for photosynthesis and yet avoid excessive heat gain. Comparison of the eggshell curve in Fig. 1A and the curve for a leaf of the cottonwood, *Populus deltoides* (*17*) (Fig. 1B), shows the similarity. Although the near-IR reflectance of the leaf is less than 40 percent, the transmittance is about 50 percent so that the near-IR absorptance (absorptance is equal to 1 minus reflectance minus transmittance) of the leaf and eggshell are comparable ($\approx$10 percent). Some insects and certain hylid and centrolenid tree frogs (*1, 18*) have spectral reflectance matching that of green vegetation in the near-IR but presumably lack the transmittance. The enhanced near-IR reflectance reported for the fur of the red kangaroo, *Megaleia rufa* (*19*), involves an extended, gradual transition. The pigmented avian egg thus appears to be unique among animal surfaces in its combination of cryptic visual coloration and thermoregulatory near-IR reflectance.

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Myotonic Muscular Dystrophy: Altered Calcium Transport in Erythrocytes

Abstract. *Erythrocytes from patients with myotonic muscular dystrophy accumulate calcium at a significantly higher rate than normal controls do. This increased rate of net accumulation appears related to an enhanced permeability of the membrane to calcium, rather than to an impairment in its active outward transport.*

Myotonic muscular dystrophy (MyMD) is an inherited disease of man characterized by a variable clinical presentation which may include myotonia, weakness and atrophy of skeletal muscle, dysphagia and constipation, cardiac arrhythmias, testicular atrophy, hyperinsulinemia in response to a glucose load, cataracts, frontal balding, and rapid turnover of immunoglobulins (*1*). Although the specific inborn error of metabolism is not known, recent data suggest that the cellular plasma membranes in many organ systems are disturbed.

Electrophysiological studies of skeletal muscle have ascribed the myotonia of MyMD to alterations of muscle surface membrane (*2*). These data have been supported by the morphological demonstration through freeze fracture analysis of an increased number of intramembranous particles (*3*). Similar studies of other myotonic syndromes in man, such as myotonia congenita and drug-induced myotonia, have also localized the abnormality to surface membranes (*4, 5*). Although the myotonia of these latter conditions can be best explained by a decrease in chloride conductance, the myotonia of MyMD is not accompanied by any change in membrane resistance or conductance (*2*), and therefore an alternate mechanism must be investigated.

Previous studies have demonstrated that red blood cells of patients with MyMD exhibit definite membrane abnormalities, including increased membrane fluidity (*6, 7*), impaired protein phosphorylation (*8*), and abnormal shape when viewed through the scanning electron microscope (*9*). More recently, we have demonstrated an alteration in the calcium promoted potassium efflux in MyMD red blood cells exposed to iodoacetic acid and adenosine (*10*). Although these experiments only examined potassium movements, they did suggest a potential alteration in transmembrane calcium movements.

The availability of improved techniques for measuring calcium transport made possible our studies on the red blood cells of patients with MyMD. In comparison with normal controls, the red blood cells from six out of seven patients with myotonic dystrophy were observed to have a significantly increased rate of calcium accumulation and an increased rate of calcium efflux against an electrochemical gradient.

All studies were performed with heparinized blood from fasted subjects. The red cells were separated and washed with isotonic tetramethylammonium chloride (0.17*M*) adjusted with 5 m*M* tris buffer, *p*H 7.4, 20°C. Cells were washed three times with this solution for accumulation studies, six times for the measurement of calcium accumulation after incubation, and six times without incubation for the starting calcium values. The patient population consisted of seven males with moderately severe myotonic muscular dystrophy. Control samples were obtained from healthy males of similar ages. None were anemic. Both patient and control samples were ob-

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