FINE STRUCTURAL OBSERVATIONS RELATING TO THE PRODUCTION OF COLOR BY THE IRIDOPHORES OF A LIZARD, *ANOLIS CAROLINENSIS*

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ABSTRACT

This paper presents the results of light and electron microscopy done on iridophores in the dorsal skin of the lizard *Anolis carolinensis*. New fine-structural details are revealed, and their importance is discussed. Of some interest is the complex of filaments between crystalline sheets in the cell. It is proposed that this complex is involved in the arrangement of crystals into crystalline sheets, and that the crystal arrangement and spacing are critical for the production of the cells' blue-green color. Tyndall scattering and thin-film interference are discussed as possible explanations for iridophore color production in relation to the fine-structural data obtained.

INTRODUCTION

The dermal pigment system of *Anolis carolinensis* has been described before (1, 2, 55, 57). Briefly, it consists of an outer layer of xanthophores, with both pteridine and carotenoid granule types, an intermediate iridophore layer, two to four cells thick, and an inner melanophore layer. Vertically oriented melanophore arms from a single cell surround groups of iridophores, and send fine lateral projections between the outermost iridophores and the xanthophores and also external to the xanthophores. The movement of melanin granules, in response to hormones, lighting, or background, is responsible for the color changes seen in *Anolis* (23–26, 34, 35, 55). The animal is dark brown when the melanin granules are dispersed into the cellular extensions overlying the iridophore layer. By the retraction of granules into their cell centers, however, (aggregated state), the melanophores remove their opaque mask from over the iridophore layer, which is consequently exposed to the incoming light. The typical green coloration of the animal results from the selective reflection of blue-green wavelengths by the iridophores through the yellow “filter” of the xanthophore layer above them.

Studies of iridophores in the past have documented the absence of pigments in these cells (5, 15, 20, 36, 44). Instead, these studies have shown the iridophores to be packed with birefringent bundles of crystalline material arranged in a variety of species-specific patterns (7, 47, 51, 57). Most recently electron microscopy has further revealed that the crystals occur in groups of platelets, rodlets, or spicules of regular thickness, bounded by membranes, and stacked in parallel at regular distances from one another (1, 2, 6, 8, 9–11, 28–33, 49, 51, 53, 55). Some authors have discussed the physical implications of this precise organization in terms of the cells' ability to reflect specific wavelengths of light (see especially Land [36] and Denton and Nicol [15, 17]). No authors to date, however, have discussed the detailed ultra-
structural features of the cytoplasm of iridophores, features that are probably responsible for controlling the strikingly complicated and regular crystalline arrays that are doubtless critical in the production of structural color. While the iridophores in Anolis appear to remain passive in the color changes of this animal, those in other species (4, 6, 19, 21, 43, 49) are reported to undergo changes that are responsible for a color shift through the entire spectrum. The ultrastructural manifestations, mechanism, and generality of these changes have not been explored.

The purpose of this paper is to document the detailed morphological features of the static iridophores of Anolis as an approach to understanding the function and control of this “simple” iridophore. This study is preliminary to an experimental study of Anolis iridophores and to the study of more complicated systems in which color changes are reported to occur naturally in response to changes in ambient ions, hormones, neurosecretions, etc. It is hoped that the interpretation of events in iridophores will also yield a general understanding of the mechanisms involved in the control of cytoplasmic organizations observed in a wide variety of cell types.

Materials and Methods

Anolis carolinensis, obtained from Carolina Biological Supply Co., (Elon College, North Carolina), were killed by decapitation and pithing. Patches of back skin were peeled off of the animal, minced into small squares in saline with a clean, sharp razor blade, and subjected to one of four treatments.

(a) The skin pieces were fixed for 2 hr in 3% glutaraldehyde (in 1% phosphate buffer at pH 7.3), then washed in several changes of the same buffer for 1-2 hr, postfixed in 1% osmium tetroxide (in the same buffer) for ½ hr, and dehydrated either through an acetone series or an ethanol series followed by propylene oxide. After agitation overnight in Epon-Araldite embedding medium (5) diluted 1:1 with either ace tone (for the specimens dehydrated in acetone) or propylene oxide (for the specimens dehydrated in ethanol), they were transferred to pure embedding medium for 1 hr of agitation, then embedded in fresh medium and polymerized for 48 hr at 60°C. Silver or gold sections were cut on a Porter-Blum microtome, and either left unstained or stained for 5 min each with 2% aqueous uranyl acetate and Reynolds’s lead citrate (46). Microscopy was done with Hitachi HU-11C and Philips 200 electron microscopes. Measurements were made from plates or film negatives with the aid of a Nikon profile projector, Model 6C, equipped with a biaxial stage micrometer. The values obtained are accurate to within ±10% in all cases.

(b) The skin pieces were incubated for 16 hr in Difco TC Tyrode’s solution containing 0.15% collagenase (Worthington CLS, Worthington Biochemical Corp., Freehold, N. J.), 0.15% hyaluronidase (Worthington HSE, Worthington Biochemical Corp.), 5% bovine serum albumin (Sigma Fraction V, Sigma Chemical Co., St. Louis, Mo.), and 100 units (or mg) penicillin-streptomycin (Difco Laboratories, Inc., Detroit, Mich.) per ml of solution. At the end of this period the skin cells were freed from their digested connective tissue matrix by gentle pipetting, then pelleted for 30 sec at 3000 rpm in a table-top centrifuge. The supernatant was poured off and replaced by some fresh Tyrode’s solution mixture without enzymes. After two or three such washes, the cells were resuspended in 3% glutaraldehyde (in 1% phosphate buffer, pH 7.3). For some samples, further treatment was as in (a), with pelleting and resuspension at each step. Other samples were pelleted and embedded in 2% purified agar, then the cut-up agar pieces treated as in (a), except that the dehydration in 100% ethanol was extended to 2 days with frequent changes. Electron microscopy was done with Hitachi HU-11C and Philips 200 microscopes.

(c) The skin pieces were treated as in (b), but the glutaraldehyde-fixed cells were washed with phosphate buffer (1%, pH 7.3), then infiltrated with 30% glycerol over a ½ hr period. Portions of the centrifuged pellet were rapidly frozen onto gold grids in Freon cooled by liquid nitrogen. These samples were freeze-etched (42, 50). The 40% chronic acid- and distilled water-washed replicas were examined with Hitachi HU-11C and Philips 200 electron microscopes.

(d) Light microscopy was done on pieces of skin and isolated cells fixed in 3% glutaraldehyde (in 1% phosphate buffer, pH 7.3). Both transmitted light and epi-illumination were used to obtain comparisons between colors transmitted and reflected by iridophores. Corning filters of different colors were also used with both types of illumination: a CS5-56 filter was used as the blue filter; a CS2-58 as the red filter.

Results

I. Light Microscopy

Light microscopy of whole skin preparations (Figs. 1 a–c) shows raised, closely grouped scales covering the skin surface. Iridophores follow the contours of the scales and lie in the flat areas between scales. Where they are not covered by melanophore processes or by xanthophores (especially between scales, Fig. 1 b), the iridophores
FIGURE 1 Photographs of a piece of Anolis skin showing color-production in the scales (S). The skin was fixed in 3% glutaraldehyde and mounted in the fixative between slide and coverslip. Fig. 1 a, Epi-illumination, no filter. × 2000. High focus to show the green-to-blue reflections from the top surfaces of the scales. Note also a tinge of red in the reflection from the centers of the scales. Fig. 1 b, Epi-illumination, no filter. × 2000. Low focus to show iridophores, melanophores, and the purplish collagen matrix (C) between scales. Here the iridophores shown at thick arrows, unobscured by other pigment cells except for a few melanophores, appear much as do iridophores isolated enzymatically. Fig. 1 c, Transmitted illumination, no filter. × 2000. High focus to show the top surfaces of the scales with red iridophores and black melanophores. Fig. 1 d, Epi-illumination, blue filter (Corning CS5-56) over the light source. × 2000. High focus to show the blue-green reflections from the top surfaces of the scales. Fig. 1 e, Transmitted illumination, blue filter (Corning CS5-56) over the light source. × 2000. High focus to show complete nontransmission of blue wavelengths by the iridophores.
**Figure 2** Low-power view of a vertical section through the skin of *Anolis*, showing (from the top) epidermis (E), collagen (C), melanophore extensions (Me), and portions of three iridophores (I). The iridophores contain many rows of membrane-bounded spaces (G) where the reflecting crystals used to be. (They were dissolved away by the alkaline stain.) The large central nucleus (N) and numerous mitochondria (M) are also obvious. Less obvious, and unmarked (but see Figs. 14a and 15) are periodic densities along the crystalline rows and surface pits in the plasma membrane. × 12,300.

**Figure 3** As for Fig. 2, different area. Apparent here are slight variations of the parallel row pattern of crystal spaces. These are probably explainable in terms of obliquity of section and irregular cellular orientation (see text). Crystals appear to be grouped together into columns (Co), between which remarkable lattices of filaments (F) are apparent. × 17,300.
reflect blue-green light, predominantly. This is modified to green-yellow when xanthophores are present. By transmitted light, iridophores appear red (Fig. 1 c), since the shorter wavelengths have been selectively reflected back toward the light source. The ability of iridophores to selectively reflect light of short wavelengths is easily confirmed by the use of filters. In Figs. 1 d and 1 e, a blue filter was used. It is very apparent that in areas where iridophores are numerous, no blue light is transmitted (Fig. 1 d). The converse is true when a red filter is used.

Similar observations on freed iridophores show that these cells retain their ability to iridesce, and can, at least by this criterion, be considered healthy and structurally intact.

II. Electron Microscopy

A. General Morphology; The Crystals: Electron microscopy (Figs. 2 and 3) reveals that iridophores are large, flattened cells, lying with their long axes roughly parallel to the skin surface, and reaching a length of up to 20 µm. They have prominent nuclei, clusters of mitochondria, vesicles of many sizes, and elements of the endoplasmic reticulum and Golgi apparatus. In vertical section their cytoplasm is seen to be taken up almost entirely with small crystalline rods, approximately 1000 Å by 1700 Å, oriented in undulating rows inclined about 30°-50° to the skin surface (Figs. 2 and 4). The crystals are dissolved out by alkali since guanine is their major if not single component (unpublished results of this lab, using Sumner’s (52) method of extraction and UV spectrophotometric analysis of the extract); thus, only the spaces they leave behind are apparent in lead-stained material (Figs. 2, 3, 6, 8, 12-15). Contrary to what others have experienced (2, 48, 53), we have not found that any major loss of crystals occurs in the process of ultramicrotomy; crystals are regularly present in unstained silver-gold thin-sections (Fig. 4), and disappear only after lead staining. Freeze-etch preparations (Fig. 5) reveal the same overall cell architecture in three dimensions as seen in unstained thin sections in two dimensions (Fig. 4 b).

In sections parallel to the skin surface (Figs. 6 and 8), crystals (or the membrane-bounded spaces they leave behind), 1700 Å in diameter, are rounded in outline and grouped into sheets. Because the sheets are not precisely planar, any horizontal section cuts through parts of a given sheet, between which non-crystal-containing areas of cytoplasm appear. Figs. 6 a and 6 b show adjacent serial sections from the same specimen. The complementary crystal arrangement is striking, showing that the entire thickness of the crystalline sheet has been included in two sections. Partial obliquity of section, combined with irregular cellular orientation, could account for similar crystal groupings in Fig. 3. The freeze-etch preparation shown in Fig. 9 is also fractured in the plane parallel to the skin surface and shows particularly clearly the layering of crystalline sheets.

Close examination of intact crystals reveals an apparently layered substructure. Figs. 4 and 11 show the layering as differences in electron opacity, regularly aligned along the crystal rows. The layering could be due to artificial chipping out of

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**Figure 4** Enlarged areas of an unstained thin section through *Amia* skin. Here the dense intact crystals (G) are apparent and their substructural banding pattern (B) is shown. In Fig. 4 a, the crystals are cut in somewhat oblique section, and systems of parallel filaments (P) are seen to run between them. Fig. 4 b is a true vertical section showing the filaments end-on as point-densities (P): × 45,000.

**Figure 5** A freeze-etch replica of vertically fractured iridophores showing membrane-bounded crystals (G) in rows, filament cross-fractures (P) along crystalline rows, and surface pits (SP) in the plasma membranes (PM) of the cells. The circled arrow indicates the direction of platinum shadowing. × 27,000.

**Figure 6** Low-power views of adjacent serial sections cut parallel to the skin surface ("horizontal section"). Fig. 6 a shows closely grouped crystals (G) and filaments (P) along crystalline rows, and surface pits (SP) in the plasma membranes (PM) of the cells. The circled arrow indicates the direction of platinum shadowing. × 13,550.
FIGURE 7 Diagrammatic representations of a single crystal. Note the outer (0) and inner (I) unit membranes together making up the double membrane (DM) on the lateral margins of the crystal; the single unit membrane (SM) on the free face of the crystal; the cross-hatching (CII) of 190 Å periodicity between the outer and inner lateral membranes; the shadow (S) often seen at the margins of the crystal, probably representing the shadow of the crystal membrane included in the thickness of the section (but see explanation, Fig. 12). Fig. 7a, Vertical section of a crystal. Dotted outlines show crystals in segments of three adjacent crystalline rows, the vertical sections of crystalline sheets. Figs. 7b and 7c, single crystals cut vertically and horizontally, respectively, shown in three dimensions with the crystals removed to clarify the membrane structure around the crystal.

parts of the crystal during sectioning, but it is more likely to represent a pattern of crystallization or chemical inhomogeneity, or possibly a stacking of several crystalline platelets within a single crystal membrane, since it appears also in freeze-etch preparations (Fig. 10) of damaged iridophores, where the structure of the membranes surrounding the crystals may be disrupted enough to allow fracture planes through the crystal (stack) itself. The regularity and orientation of the pattern and its absence in precisely horizontal sections of crystals also indicate that it represents a true crystalline substructure. In vertical sections of crystals, the width of each layer in the crystalline substructure is approximately 156 Å (Fig. 11a). In oblique sections, the corresponding width increases to 353 Å (Fig. 11b). Calculation of the angle of obliquity of such sections with respect to the planes of the crystalline sheets using 156 Å/353 Å as the cosine of that angle, gives a value of 60°, corresponding closely to the images seen (Fig. 11b).

In sections tangential to the crystalline sheet, each crystal is bounded by what appears to be a double unit membrane, following precisely the (irregular) crystal contours (Fig. 8). Each membrane is 60 Å thick and the two membranes are separated by a dense 60 Å space. In vertical sections (Figs. 12 and 7), by contrast, there is a distinct asymmetry of the membrane construction. A doublet of unit membranes bounds each crystal on its lateral margins, that is, where it abuts adjacent crystals within a sheet. The outer of these two membranes of such adjacent crystals are closely apposed (but not fused). The dense material between the inner and outer of the two membranes often appears to be densely crosshatched with a periodicity of 120 Å. Only a single unit membrane bounds the crystal on its “free” face. This membrane appears to be continuous with both of the membranes on the lateral margins of the crystal.

Crystal spacing in iridophores is quite precise.

FIGURE 8 Enlarged area of a horizontal section through Anolis skin showing closely grouped crystal spaces (G) and filamentous cytoplasm (F) between the groups. The crystal spaces are bounded by a doublet of unit membranes (DM). × 31,500.

FIGURE 9 A freeze-etch replica of a horizontally-fractured iridophore showing clearly the layering of successive crystalline sheets (CS). The arrow indicates the direction of platinum shadowing. × 39,000.

FIGURE 10 A freeze-etch replica of a damaged iridophore, in which the membranes of some crystals have been broken open to allow fracture planes within the crystal lattice or stack (L). The large circled arrow indicates the direction of platinum shadowing. × 84,380.

FIGURE 11 Two sections through an unstained iridophore, showing intact crystals (G), whose banding patterns (B) differ with their orientation in the section. See text for interpretation of banded densities. Fig. 11a, vertical section. Fig. 11b, Oblique horizontal section. × 72,600.
Adjacent crystals within a row are only 100 Å apart, as measured between their outer membranes. The distance between rows, again measured between the outer membranes of the crystals in adjacent rows, is 740 Å. In sections tangential to crystalline sheets, the spacing of crystals is the same in all directions, and the intermembrane distance is 110 Å, on the average. The average dimensions of the crystal itself (within the crystal membrane) in vertical section through the center of the crystal are 1000 Å by 1700 Å.

B. THE FILAMENT NETWORK: On either side of each crystalline sheet (in sections oblique or parallel to the crystalline sheets, Figs. 3, 13, and 14) there is a filament lattice consisting of 75 Å filaments and many 75-80 Å cross-connections between the lattice and the crystal membrane and between elements within the lattice (Fig. 15).

The filaments stand out particularly well in preparations which have been soaked long enough to leach out granularities in the surrounding cytoplasm (Figs. 13 and 14). While filaments are often seen coursing in apparently random directions between crystal groups, the striking filament lattice seen in oblique sections (Figs. 3, 13, and 14) and the rows of point-densities seen along crystal rows in vertical sections (Figs. 2, 13, and 15) indicate a highly structured network in the intercrystalline zones. In keeping with this interpretation is the straightness of the filaments and their strict orientation in parallel with one another in the lattices (Figs. 3, 13, and 14). Filament spacings in the lattice are 140 Å. A synthesis of such images as Figs. 13, 14, and 15 is presented schematically in Fig. 17. There seem to be lattices of filaments above and below each crystalline sheet, with the filaments running at very shallow angles or parallel to the sheets and essentially in two parallel, mutually perpendicular arrays. There are also randomly placed short filament links connecting the lattices to one another and to the membranes of the crystals. Isolated 75 Å filaments course at odd angles through the lattices, parallel to the crystalline sheets. There seems to be no regularity in the relative axial orientations of filamentous components of successive filament lattices.

The same filament types and arrangements are present in isolated cells (Fig. 12), but for some reason they are less distinct. In freeze-etch preparations, only the "point-densities" (filament cross-sections) along the crystalline rows are clear (Fig. 5).

C. THE CELL SURFACE: Finally, the surface of iridophores is structurally complex. The cell membrane is irregularly pitted with pear-shaped, pinocytic-like vesicles which, however, do not appear to close up or move into the cell (Figs. 2 and 14 a). The pits are 800 Å by 600 Å in cross-section, and have a 300 Å diameter opening to the outside. Freeze-etch preparations show the identical structures in cross-fracture (Fig. 5), but add complementary face-on views of the pit openings in the split membrane (Fig. 16). In these preparations, the pits sometimes appear to be plugged. The function of the pits is unclear. Their distribution seems to be random over the cell.

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**Figure 12** Enlarged view of a vertical section through an isolated iridophore, showing the asymmetric disposition of the crystal membranes as seen in vertical section. On the lateral margins of the crystals, the membranes appear to be double (DM), while on the "free" surfaces of the crystals they are single (SM). Between the two membranes on the lateral margins of the crystal is a periodically banded matrix of medium density (MA). Within the crystal membranes is a frequently seen shadow (SH) of medium density, not dissolved out by lead staining. This probably represents a shadow of the membrane included in the thickness of the section (it has also been interpreted as noncrystalline purine material [6, 54]). The cytoplasm between crystalline sheets is filamentous, but the pattern of filament dispositions is not so clear as in the cells of the intact skin (Fig. 15). × 140,000.

**Figure 13** Oblique section through an iridophore, showing clearly the filament lattices (FL) and randomly coursing filaments (F) between crystal groups. Cross-sections of filaments in lattices (P) are apparent at the right of the figure, along the crystalline rows. × 33,700.

**Figure 14** Views of filament lattices (FL) in iridophores fixed with 3% glutaraldehyde made up in distilled water. Cytoplasmic granularities have been leached out by the treatment, exposing the lattices for better imaging. Surface pits (SP) in the plasma membrane are apparent in Fig. 14 a. 14 a, × 72,600. 14 b, × 53,300.

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FIGURE 15 Enlarged area of a vertical section through an iridophore in intact skin, showing the membrane-bounded crystal spaces (G), rows of point-densities (P) along the crystal rows, and fine filamentous connections (C) between neighboring rows of point-densities and between rows of point-densities and crystal membranes. X 78,750.

FIGURE 16 A freeze-etch replica of split plasma membranes (PM) of iridophores showing complementary aspects of surface pits (SP) in the membranes. Some of these pits appear to be plugged (shown by thick arrows). The circled arrow indicates the direction of platinum shadowing. X 64,000.

The structures most functionally significant for the iridophore are its purine crystals and filament lattices, which together constitute the bulk of its cytoplasmic contents.

The maintenance of organized parallel stacks of crystalline sheets has been elucidated from the evidence adduced in this paper. It is most easily ascribed to the complex filament arrays present between the sheets. These filaments resemble intercisternal filament systems seen in the Golgi complex (41, 56) when the ordering of the Golgi stacks is highly regular. Similarly, when vesicles of the endoplasmic reticulum are highly ordered into stacks, as is frequently the case in pancreatic cells, close examination reveals a filamentous network between the vesicles. All these lines of evidence point to the use of intracellular filament networks as scaffoldings wherever there is a requirement for precise ordering of cellular components. The labile character of the organization, at least in the pan-

1 Porter, K. R. Unpublished observation.
creas, points to a way in which the cell may control its functional architecture. One may speculate that similar orienting and polarizing structural complexes are present in all cells, but that their more diffuse, “relaxed” disposition in most cells has prevented their recognition to date in thin section preparations.

In the iridophore, the filament lattices close to the crystalline sheets (about 90 A away from the crystals), crossed by randomly coursing filaments, and attached to the membranes of the crystals by short filamentous connections might function to anchor each crystalline sheet. The cross-connecting filaments from the crystal membranes to the lattices on either side of the crystalline sheet would correspondingly lock the crystals into their sheet-like array by preventing their vertical movement in either direction. Similarly, the filaments connecting adjacent filament lattices would lock adjacent crystalline sheets into precise positions relative to one another, thus preventing random widening of the gap between sheets.

It is tempting to speculate on the mechanism of color production operating in Anolis iridophores, now that data on their ultrastructure are available. Since iridophores contain no pigment, some theory of structural color production resulting from refractive index inhomogeneities within the cells must be invoked (12, 15-17, 20, 22, 27, 36-40, 44). Iridophores are the principal site of wavelength selection and separation in Anolis skin. Even in isolation they are typically blue-green when viewed by reflected light, and red when viewed by transmitted light. The importance of melanophores and xanthophores in contributing to the over-all color of the animal (13, 55) is apparent, however, when isolated iridophores are incompletely separated from these surrounding pigment cells. Attached (underlying) melanophores are seen to decrease back-scattering of the red light transmitted through the iridophores by absorbing these long wavelengths of light. The melanophore-associated iridophores are consequently intensely blue-green as compared with fully isolated iridophores. Associated (overlying) xanthophores modify the reflected blue-green to green by providing a yellow filter that absorbs the shorter wavelengths.

It is the primary separation of wavelengths by the iridophores that is least understood. We make a few comments on this question below, based on the fine-structural observations presented in this paper.

Since the crystals in Anolis iridophores take the form of flattened cylinders whose largest dimensions are only half the length of the shortest visible wavelengths, Tyndall scattering, as has been suggested before (20), is an attractive hypothesis to explain their reflection of blue light. According to this theory, the electrons of particles (or optical inhomogeneities in a transparent medium) small relative to the wavelength of red light scatter incoming light in all directions. In such small...
reflection increases. The theory further predicts the reflected light will decrease as the angle of thin-film interference, such that the wavelength of wavelengths (iridescence) will occur as a result of the separation of incident wavelengths, an angular dispersion of which will be longer. For white light or any other mixture of species from the normal, the path length in the film obviously, for greater angles of incidence, measured within the film. The optical path length, in turn, varies with the angle of incidence of the light: obviously, for greater angles of incidence, measured from the normal, the path length in the film will be longer. For white light or any other mixture of incident wavelengths, an angular dispersion of wavelengths (iridescence) will occur as a result of thin-film interference, such that the wavelength of the reflected light will decrease as the angle of reflection increases. The theory further predicts that those wavelengths reinforced by reflection are cancelled by transmission. Thus, complementary colors are reflected and transmitted. A stacking of several thin films of equal optical thickness, but of alternating high and low refractive indices, further reinforces the color selection and increases the reflectivity over that from a single film. Up to 99% or even 100% reflectivity of very narrow wavelength bands can be achieved in this way (12).

In Anolis iridophores, the crystalline sheets could easily represent the films of high refractive index in a thin-film interference system. The intervening layers of filamentous cytoplasm would represent the alternating films of low refractive index. Clearly, as noted earlier, the colors seen in iridophores by reflected and transmitted light (Fig. 1) are complementary. The reflected wavelengths center around the blue-green for the iridophores alone, that is, around the wavelength range 4600 A–5500 A. Iridescence is detectable, but not striking; note the orange tinge in the central regions of the predominantly blue-green scales (Fig. 1 a).

Positive proof of the operation of thin-film interference in Anolis iridophores awaits spectral measurements of the reflected light and refractive index measurements of the “films” involved. Such measurements have been carried out in somewhat different iridophore systems (17, 96), and show that the hypothesis is reasonable and must be considered seriously for Anolis, too.

It is probable that if the interpretation of iridophore crystal stability is correct as presented in this paper, that is, that it is governed by a fairly rigid and complex filament network (a structured matrix) in Anolis iridophores, the filament organization should differ in the iridophores of animals whose color changes are at least partially due to iridophore changes. Here the cell should be able to control changes in filament organization in response to appropriate environmental influences, and through this, to control the crystal spacing and arrangement in the cell (and hence the color produced by the cell). A comparative survey of iridophores and experiments with cytochalasin B and other cell modifiers should answer this question, and are in progress in our laboratory at this time.

We are grateful to Drs. H. C. Berg, J. R. McIvor, L. A. Staehelin, and F. Rohrlich for the helpful comments and suggestions they offered in the course of the research for this paper.

This investigation was supported by Public Health
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