

Switchable reflector in the Panamanian tortoise beetle *Charidotella egregia* (Chrysomelidae: Cassidinae)

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(Received 29 April 2007; published 11 September 2007)

The tortoise beetle *Charidotella egregia* is able to modify the structural color of its cuticle reversibly, when disturbed by stressful external events. After field observations, measurements of the optical properties in the two main stable color states and scanning electron microscope and transmission electron microscope investigations, a physical mechanism is proposed to explain the color switching of this insect. It is shown that the gold coloration displayed by animals at rest arises from a chirped multilayer reflector maintained in a perfect coherent state by the presence of humidity in the porous patches within each layer, while the red color displayed by disturbed animals results from the destruction of this reflector by the expulsion of the liquid from the porous patches, turning the multilayer into a translucent slab that leaves an unobstructed view of the deeper-lying, pigmented red substrate. This mechanism not only explains the change of hue but also the change of scattering mode from specular to diffuse. Quantitative modeling is developed in support of this analysis.

DOI: 10.1103/PhysRevE.76.031907

PACS number(s): 42.66.-p, 42.70.Qs, 42.81.Qb

I. INTRODUCTION

Numerous living organisms rapidly alter their appearance in response to changes in the environment. The immediate threat of a predator attack, for instance, can trigger reactions that suddenly reveal brightly colored body parts as part of a “startle response.” This is very common among flying insects, which can hide and then suddenly display conspicuously patterned hindwings, in a strategy to confuse their predators and delay adverse actions [1].

More complex transformations are sometimes observed among terrestrial animals. One of the best known examples is the chameleon, some species of which (for instance *Chamaeleo chamaeleon*) are able to modify the coloration of their skin [2,3], partly according to their mood, partly as a cryptic adaptation to their surroundings. In that case, the change of color is explained by the migration and volume change of special cells (chromatophores) dispersed in the skin below the transparent outer layers. With reddish “erythrophores,” yellow “xanthophores,” and blue “iridophores,” the chameleon’s skin can display a large range of hues and color patterns by adjusting the absorption efficiency of each type of chromatophore. The presence of melanin in deeper layers also allows dark or light colors to be produced, according to the diffusive power given to the chromatophores. Another recently discovered reptile, the Kapuas mud snake (*Enhydris gyii*) from the Kalimantan area of the Borneo rainforest [4], switches from red-brown to white, but the underlying mechanism has not yet, to our knowledge, been fully elucidated. Numerous octopi, squid species, and cuttlefish

are able to change their skin color—most famously, species of the blue-ringed octopus, *Hapalochlaena spp.* The squid *Loligo pealei* is, likewise, able to modify its skin hue for camouflage and intraspecific communication [5]. A species of octopus was recently discovered in Indonesian waters that is not only capable of changing its skin color, but also seems to have evolved a very high degree of variability, allowing it to mimic the appearance of poisonous creatures, including their coloration patterns and body shapes [6].

Among insects, reversible changes of coloration have been described in several species, such as the phasmid *Carausius morosus* [7–9], the larvae of the dipteran *Corethra* [9–12], the grasshopper *Kosciuscola tristis* [13,14], a number of damsel flies [15–22], and some Coleoptera [23,24]. Most of these examples show only passive coloration changes due to diurnal changes of the atmospheric hygrometry. The cassidine beetles considered here are special because they undergo a more active change of color, controlled by the insect itself, as a reaction to events occurring in the environment. A review considering many cases of tortoise beetles (Chrysomelidae: Cassidinae) can be found in a paper by Jolivet [25].

The early work on cassidines led to the realization that the coloration change was structural, involving the selective reflection of light by a thick multilayer located in the outer layers of the transparent armor (see [26] for a brief sketch of the optical properties of periodic and chirped multilayer stacks and Bragg mirrors). Quantitative physics, however, has not yet contributed its share to the precise reverse engineering of this structure and the mechanism of coloration changes. The widespread assumption—the “hydraulic theory” [23,25,27]—is that the physical characteristics of the multilayer (refractive index and/or thickness of the layers)

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FIG. 1. (Color online) Adult *Charidotella egregia*. This leaf beetle (8 mm in size) is a representative of the large family Cassidinae. The insects in the family Cassidinae are often called “tortoise beetles,” because they have a wide, hard and strong armor, which includes the wing cases, the pronotum and the head cuticula. It extends widely, beyond the insect’s periphery, providing a very efficient shield against attempts to grip the insect when it is lying flat on a supporting leaf.

are modified under the pressure of a fluid, to produce the color changes. However, we will see that the color changing beetle under study here does not use this widely assumed mechanism.

The insect that is the subject of the present work (see Fig. 1) is *Charidotella egregia* (Boheman, 1855) (Coleoptera, Chrysomelidae, Cassidinae). Our choice of this insect species was influenced by the strong and rapid changes of appearance we observed it make and by the intriguing questions which these observations generated. One of these is that the color change not only affects the hue, but also the type of surface scattering: the appearance changes from a “metallic” sheen into a “matt,” or “diffusive,” aspect. This change, from specular reflection to wide-angle diffusion, calls for an improved analysis, which led us to need new, accurate optical measurements, scanning electron microscopy, detailed modeling of the structure and confirmation of the interpretation by numerical modeling. Since the effects observed only occurred with living animals and since these animals only feed on a single host plant, it was mandatory to make at least some of the observations and measurements in or near the insect’s natural habitat. The collection of specimens and many of the optical measurements were developed at the Smithsonian Tropical Research Institute, in the Republic of Panama.

II. OBTAINING SAMPLES

The golden tortoise beetle, *Charidotella egregia* is reported to feed specifically on the vine *Ipomoea lindenii* (Martens and Galeotti) [28], and its presence can often be detected by observing recent feeding damage on the leaves of this plant. The host-plant occurs abundantly near the summit of Cerro Galera, 6 km west of Panama City, where during the first week of September 2006, we easily collected 12 *Charidotella egregia* adults. We noted a significant number of larvae and pupae at the site as well, assuring us that the

population would not be adversely affected by the capture of a few individuals for our study.

Beetles were maintained in the laboratory in separate one-half pint plastic containers and provisioned daily with a fresh leaf of their host plant. The moisture in each container was maintained at a fairly high level, to prevent the leaves from desiccating rapidly and replicating as nearly as possible the hygrometry of the natural habitat. The insects survived well for weeks under these conditions.

III. APPEARANCE TRANSFORMATIONS

Charidotella egregia color change is observed primarily within the central part of its dorsal armor (“elytral and pronotal disks”), not in the flat, transparent, peripheral extension through which the colors and details of the leaf surface can be clearly observed. The transformation from gold to red can be initiated by nearly any sort of disturbance to the resting insect, such as touching the insect with a stylus strongly enough to displace the insect by a few millimeters. The intensity of the disturbance may be roughly equivalent to a blow from a bird during a predation act. After less than two minutes, the change from gold to red is complete. As the hue changes, the metallic sheen of the insect disappears and the surface under the transparent armor becomes more diffuse. Figure 2 shows the details of the appearance of the insect during this transformation. As can be seen, the color does not change uniformly. The change from gold to red first occurs at the periphery of the elytra and on the external and central parts of the prothorax, leaving zones of high reflectance. The low-saturated red appearance is what we term the “disturbed” state, reached when the insect has been disturbed in some way, while the gold appearance we term the “resting” state, the normal color of insects in repose, most often underneath their host plant’s newer leaves. The “resting” state was also observed in adults sitting on the top side of a leaf, in bright sunshine. Red, “disturbed” beetles return to their gold “resting” state after a long quiet stay on, or near, the food plant and are normally considerably less common than “resting” state individuals under natural conditions in the field. However, all individuals were once observed in the “disturbed” state during a heavy tropical rainstorm. Adults emerge from the pupa in the red state, and once mature, were observed at different times in both “disturbed” and “resting” states during copulation.

Another important observation, for the argument developed in the present paper is that the gold appearance is lost when the insect is dead and dry. The elimination of moisture from the cuticle leads to a reddish appearance close to (but not exactly the same as) the “red” color seen in the disturbed state of the beetle.

IV. OPTICAL PROPERTIES

Measuring the optical properties of a susceptible living animal is somewhat tricky. In the gold state, for instance, one could not manipulate the beetle (for instance, to hold it still under the probe of the spectrometer), as it can easily change state before, or while the measurement is being carried out.

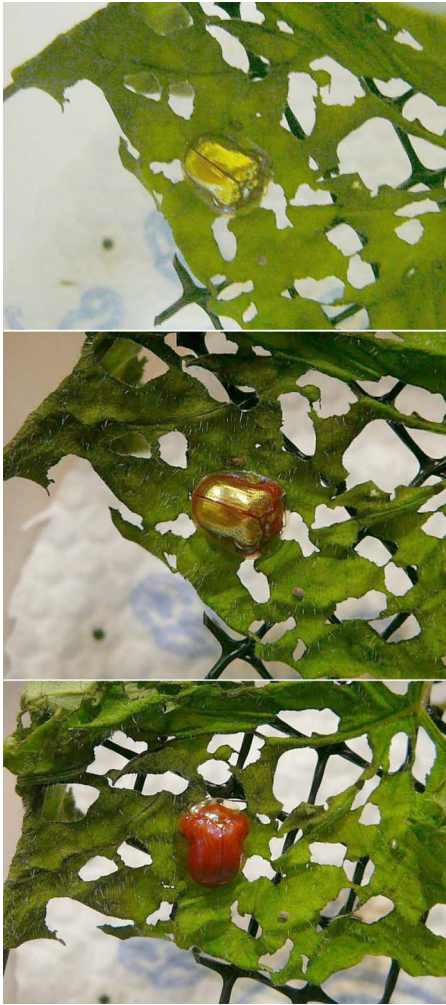


FIG. 2. (Color online) Adult *Charidotella egregia* switching from gold (above) to red (below). The timing of the whole process is variable, but a typical minimal conversion time is 1 min and 30 s. This transformation was triggered by touching the wing cases with a hard rod, plausibly simulating a missed attempt at predation.

Thus, in order to measure the reflectance spectrum of the gold state of *Charidotella egregia*, eight adults were installed in individual transparent containers, with a fresh leaf on the floor. In the morning, individuals were usually found resting in the gold state, some of them sitting still on the upper side of the leaf. The color of these individuals was measured directly inside their container without disturbance. An Avaspec 20048/2 fiber-optic spectrophotometer was used, with a bifurcated probe designed to collect the emergent light on the same path as the incident illumination, in an exact normal backscattering geometry. The probe containing the terminal parts of the optic fibers (a steel tube terminated by a lens) was passed through a fine textile net, shaped like a cone, attached to the periphery of the container top cover, so that it could be moved freely inside the container, without allowing the beetle to escape. This simple technique enabled us to take many spectra of the living insect in both states of coloration (gold and red) and to show intermediate steps in the color conversion.

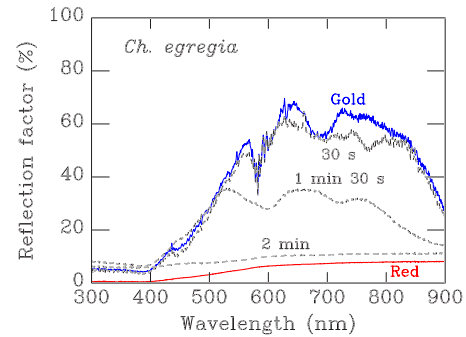


FIG. 3. (Color online) Reflection factor from the back of *Charidotella egregia*. The solid lines labeled “gold” and “red” are, respectively, the reflectance from the gold and red states. The dashed lines in between are intermediate reflectances, measured 30 s, 1 min and 30 s, and 2 min after applying a stressful disturbance. The measurement is performed in a backscattering geometry, in the direction of the normal to the cuticle surface.

Figure 3 shows typical results. The solid curves (labeled “gold” and “red”) correspond to well-defined gold and red stable states. “Gold” describes a state where the cuticle effectively contains a dielectric reflector [29], which gives the surface a metallic gold aspect. “Red” denotes a state where the surface has a diffusive red aspect that we consider to be the “disturbed” state, because its appearance is triggered by some external disturbance. The gold coloration is metallic, which means that the reflectance is high and that the scattering is predominantly specular. This suggests the presence of a multilayer, but in this case, the lack of color change with the incidence angle and the color desaturation more precisely indicates a broadband chirped dielectric mirror. The reflection factor remains high between 500 nm and 900 nm, which means that it covers the visible spectrum, with some near infrared, but lacks a blue component: hence the yellow hue of the “gold” color, which does not change with viewing angle (thus using a chirped geometry reminiscent of that occurring in *Chrysinia resplendens* [30], however with an isotropic material for each layer, so that the circularly polarizing properties of the latter does not occur). The spectral range is achieved by several reflection bands, which can be roughly located at the wavelengths 560, 640, and 810 nm. This, again, suggests that the gold dielectric mirror is chirped (with layer thicknesses varying with their depth in the cuticle) and that it can be seen as a stack of more elementary dielectric mirrors, each optimized for a specific wavelength. This is directly confirmed by nanomorphology, the subject of the next section. The red coloration appears to be spectrally unstructured and can be described as a vermilion hue, showing a uniform mixture of red, orange, and a weaker addition of yellow. This coloration results from incoherent scattering of nonabsorbed light: a process normally associated with the presence of pigments.

When the change from gold to red is activated by stressing the insect, the reflectance spectrum is progressively modified. The amount of the insect’s surface covered by the gold reflector diminishes, in favor of the red coloration. The curves shown in Fig. 3, labeled 30 s, 1 min and 30 s, and 2 min, correspond to reflection factors, measured at the cen-

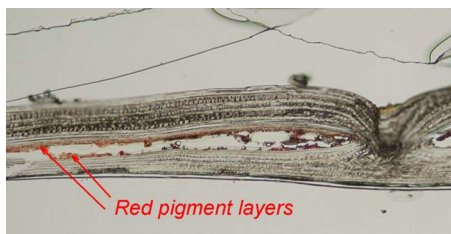


FIG. 4. (Color online) Cross section of the elytron of *Charidotella egregia*, indicating the location of the red pigment. The outer surface of the elytron is at the top of the section. The red contribution comes from a deep layer, well below the gold Bragg mirror (not perceptible at this scale).

ter of the remaining gold area, at the indicated time after the stress application. After 30 s, the reflectance is not significantly changed, but after 1 min and 30 s, we note both a significant reduction in the reflection intensity and a blueshift of the different components of the broadband spectrum (now perceived, respectively, near 520, 650, and 750 nm). We note that the blue intensity near 450 nm is not weakened, but the other components of the spectrum (greenish yellow, orange, and red) are reduced by nearly 50%. The reflection spectrum decreases significantly in the near infrared, but still extends over a large part of the visible range. The relative weight of the blue component increases because of the decrease of the yellow-red part, with the consequence that the color of the reflector is noticeably desaturated. Indeed, even with the naked eye, the coloration of the dielectric mirror is observed to turn from its initial gold color to a white silver hue, before disappearing. After about two minutes, the gold reflector has disappeared and the entire surface reaches a diffusive red color (a specular reflection generates highlights, i.e., sharp images of light sources, as on metallic surfaces, while a diffusive color does not).

Contrary to what could be expected from the “hydraulic” mechanism, the spectrum does not *shift* rigidly from yellow to red (something that would occur if, for some reason, the layers were only to increase their thickness or increase their average refractive index, as the result of a fluid injection). Rather, as the transformation proceeds, the high gold reflectance *disappears* and what remains is a broad, unstructured, red diffusion. The question is less about how the dielectric mirror is modified to shift the selection from a yellow to a red hue, than about how this mirror destroys itself, to leave a view of the red pigmentary material which is found beneath this multilayer. The location of this red layer is shown in Fig. 4, on a thin slice of an elytron cut along a cross section, and viewed with a transmission optical microscope. The elytron is hollow, and the red pigment lies at the edges of the cavity, well below the dielectric mirror that produces the gold reflectance.

In the following section, we bring information on the morphology of the cuticle to a resolution of a few nanometers, seeking to rationalize these observed optical properties.

V. NANOMORPHOLOGY

Physical coloration is basically due to the multiple scattering of light by an inhomogeneous medium. For red-orange

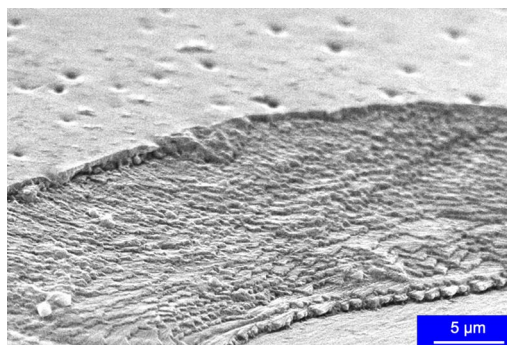


FIG. 5. (Color online) Scanning electron microscope view of the dry reflector, which produces the gold coloration. The obliquely broken exocuticle reveals the layered structure clearly, with layer thicknesses that increase proximally (i.e., towards the inside of the insect).

visible light, for instance ($\lambda=600$ nm), in a medium with an average refractive index $\bar{n}=1.5$, a typical distance between scatterers in the structure should be [31]

$$d \approx \frac{\lambda}{2\bar{n}} = \frac{600 \text{ nm}}{2 \times 1.5} = 200 \text{ nm}. \quad (1)$$

This distance decreases for shorter wavelengths or higher average refractive indexes. A resolution better than 10 nm is then required to study such structures and, consequently, we submitted transverse sections to a detailed examination with the scanning electron microscope (SEM) and the transmission electron microscope (TEM). The samples were prepared by cutting a piece of desiccated elytron in liquid nitrogen and covering it with a 15 nm thick gold layer. To begin, we searched for a spot showing a very oblique fracture. The idea of searching for the golden reflector with the SEM is not completely straightforward because, once desiccated for observation, the metallic gold appearance of the sample has effectively disappeared, which means that, at best, we can only expect to observe a “damaged” Bragg mirror. However, surprisingly, one finds this reflector apparently intact, in the form shown in Fig. 5. The topography of the terraces indicates that the exocuticle is a stack of hard layers with mechanically weaker junctions. The geometry of the structure provides a “chirped” multilayer stack, with layer thicknesses that decrease in the distal (outward) direction. Strangely enough, the number of periods is not always the same for samples taken from different locations of the insect’s armor: the number of periods actually varies from about 20 to 40, without greatly affecting the resulting visual appearance. In Fig. 6, the same type of multilayer is seen with the fracture normal to the cuticle surface. The chirped structure is confirmed, with a thickness variation such that the thicker layers have more than two times the thickness of the thinner layers. The fracture is highly irregular, which indicates that the material which forms the layers is, also, likely to be structured in the directions parallel to the multilayer’s interfaces. Such lateral structure is often encountered in the cuticle of Coleoptera [32]: for instance, the presence of parallel bars of chitin wrapped in proteins can explain the anisotropy of the

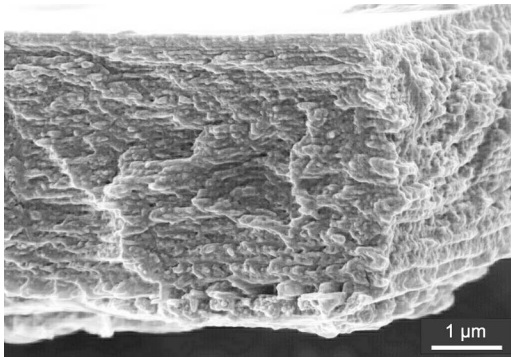


FIG. 6. In this sample for SEM, the fracture occurred normally, so that the thickness of the layers forming the gold reflector can be easily measured, in spite of the irregularity of the section surface.

layers, as needed to produce circularly polarized light from the reflection of unpolarized illumination, as observed in many Cetoniidae (see, for instance, Berthier [27], and references therein and [33]). However, in the actual state observed, nothing indicates that polarization effects are important in the case of *Charidotella egregia* and the structure giving rise to the fracture roughness is not related to any obvious regular organization.

The multilayer reflector shown in Fig. 6 has a rather simple, chirped, structure, which can be understood as a stack of three Bragg mirrors, assembled on top of each other. The first one, at the cuticle surface, contains 12 layers, with an average thickness of 186 nm. Assuming a refractive index $\bar{n}=1.5$ (slightly lower than that of chitin in order to account for some porosity), we can predict the spectral selectivity of this mirror: the dominant reflected wavelength is [31]

$$\lambda = 2\bar{n}d = 2 \times 1.5 \times 186 \text{ nm} = 558 \text{ nm}. \quad (2)$$

This is a yellowish-green mirror that accounts for the peak (near 560 nm) at the short-wavelength edge of the “gold” reflection band shown in Fig. 3. The next set of layers also contains about 12 layers, providing a stack with an average period of 213 nm. As before,

$$\lambda = 2\bar{n}d = 2 \times 1.5 \times 220 \text{ nm} = 660 \text{ nm} \quad (3)$$

and we are now in the red part of the visible spectrum. The second mirror is a rather thin (desaturated) red mirror, which can also explain the bump in the reflectance spectrum, near 640 nm in the reflection band in Fig. 3. Finally, the next eight layers in the proximal (inward) part of the cuticle show an average layer thickness of 270 nm, which corresponds to a dominant reflected wavelength of

$$\lambda = 2\bar{n}d = 2 \times 1.5 \times 270 \text{ nm} = 810 \text{ nm}, \quad (4)$$

at the edge of the infrared. This can be related to the broad component near 800 nm in the “gold” reflection band of Fig. 3. The simple analysis developed here shows that the cuticle multilayer seen at the surface of an elytron with electron microscopy has the dimensions required to explain the metallic reflection band observed in the “gold” state of the insect.

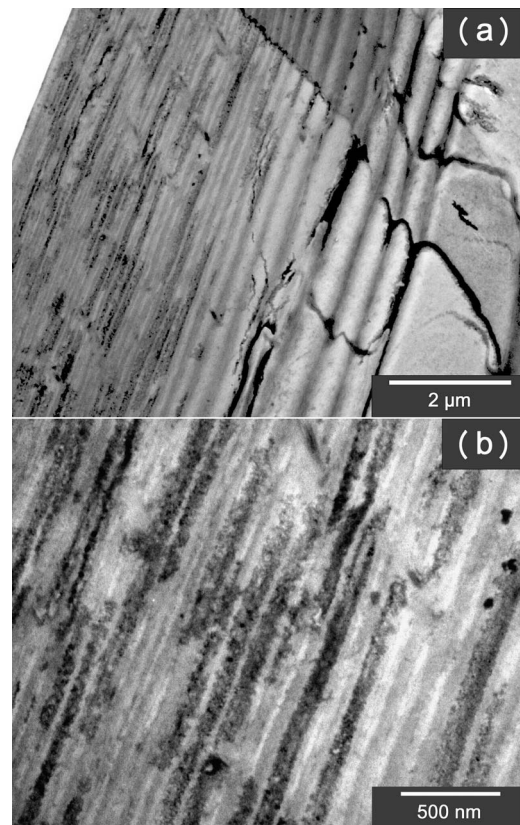


FIG. 7. Transmission electron microscope view of a thin slice of the cuticle of *Charidotella egregia*, showing irregularities in the material density, parallel to the layers. The finite-size flat zones oriented parallel to the surface can be described as porous patches distributed irregularly in each of the layers of the dielectric mirror.

TEM reveals much more of the dielectric mirror layers. Ultrathin sections of elytra, embedded in araldite, have then been cut in order to better observe the dry reflector cross section. Figure 7 shows the internal structure of the exocuticle at two different magnifications. At a magnification of about 10 000 times—Fig. 7(a)—the entire multilayer thickness is visible and each layer can easily be discriminated and characterized geometrically. At a larger magnification—Fig. 7(b)—the internal structure of the layers is revealed better. The density of the material in each of the layers can easily be perceived: the fact is that each layer clearly shows inhomogeneity, i.e., variation of density in the directions parallel to the layers. This can be best described as “porosity patches” lying within the layers of the dielectric mirror. The presence of these patches can explain the reversible destruction of the dielectric mirror, if we assume that the porous regions can rapidly switch between “humid” and “dry” conditions. All periods of the Bragg mirror actually contain two layers: the first is thick and takes most of the period thickness and varies in density because it contains the porosity patches; the second is much thinner and appears to be made of a homogeneously light material (as can be seen clearly in the images in Fig. 7) and constitutes the junction between the “patchy” slabs. The porosity patches have the thickness of the thick layer component, occur at random lateral locations in this layer, and assume a lateral size which varies between 300 nm and several μm .

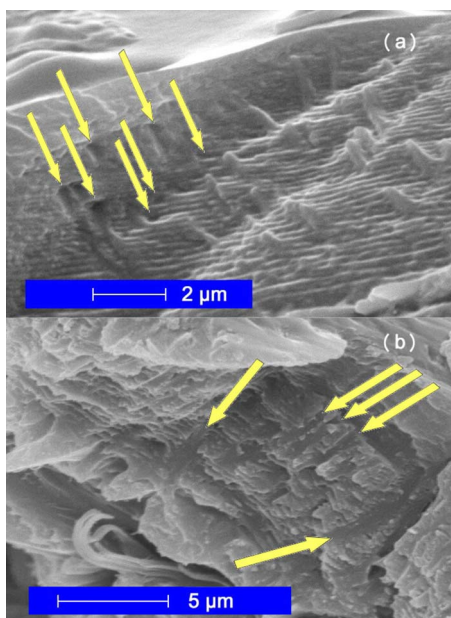


FIG. 8. (Color online) Scanning electron microscope view of a severely damaged part of the cuticle of *Charidotella egregia*, showing channels running through the optical exocuticle. It seems likely that these channels bring (or remove) fluid into (or from) the patches in the coloring multilayered exocuticle.

The presence of porosity patches gives a clue for understanding the physical mechanism of the color change of the insect. The two states of color can be related to the states of hygrometry of the exocuticle. The porous patches will show a different refractive index when the pores are filled with liquid or empty. The fluid transport to the porous patches requires a network of channels in the material which constitutes the elytron, including the very fine optically active exocuticle. Figure 8 confirms the presence of such a network. In Fig. 8(a), an oblique fracture of the exocuticle reveals these channels, indicated by arrows. Some of these channels manifest themselves as exiting cross sections which appear at various places on the irregular surface, while others that have been sectioned parallel to their axis appear as longitudinal grooves. The orientation of the channels is seen better in Fig. 8(b), where the optical exocuticle has been more severely damaged and several fracture orientations are present. Some channels run horizontally (parallel to the layers of the Bragg mirror), and vertically (normal to the multilayer interfaces).

The dissection of gold beetles (kept in this state by deep-freezing euthanasia) indicates that a similar optical structure can also be found closer to the red pigmentary layer, under the protection of the thick transparent shell which gives rigidity to the elytra. This film is susceptible of faster hydration and dehydration cycles, due to its proximity with the active part of the elytron structure.

For completeness, one should also mention that SEM pictures of the unbroken surface of the insect cuticle show microscopic openings which, to our knowledge, are not seen in other orders of insects (see Fig. 9). It is unclear whether these openings are actually connected to the network of channels or to the porosity patches, but we cannot rule out

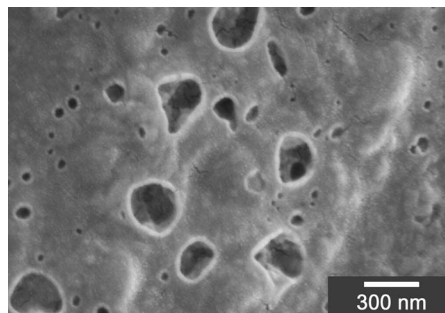


FIG. 9. Microwindows seen on the surface of unbroken elytra. These structures could accelerate exchanges of vapors between the inner volume of the optically active exocuticle and the outer atmosphere. However, the present study could not establish a relationship between these structures with the channels or the porosity patches.

the possibility that these openings help physiological fluid exchanges between the exocuticle and the atmospheric gases. Pores are however frequent and always present in Coleoptera, where they indeed serve several purposes, including giving a way out to defensive compounds produced by specific dermal glands.

VI. COLOR-SWITCHING MECHANISM

Developing a theory for color switching must account for a number of observations that are sometimes difficult to reconcile: (1) the insect can display two reversible states of coloration, a yellow (or more precisely “gold”) state and a red state; (2) the gold state emphasizes selective specular reflection, while the red state is bright, but manifests itself as a wide-angle diffusion, lacking metallic aspect; (3) the change of color occurs in patches which grow, following a well-defined sequence of areas on the dorsal cuticle; (4) the deepest layer inside the elytra is red; (5) the transformation from gold to red can be triggered by putting the insect under stress and takes about 1 min and 30 s; (6) the transformation from red to gold can be as fast as gold to red, but the start of the transformation cannot be triggered and is unpredictable.

Furthermore, it is observed that (7) naturally dead insects are red-brown when dry (this is the case of most specimens in the museum collections); (8) if quickly deep-frozen in the red state, the insect remains red when brought back (dead) to normal temperature; (9) if the insect is quickly deep-frozen while in the gold state, it turns brown when frozen, but reverts to gold when brought back (dead) to normal temperature; (10) subsequently, dead deep-frozen insects, returned to gold, very slowly lose this metallic appearance and turn red as they dehydrate. For some period of time (typically, a few hours), partially dehydrated specimens can still be forced back to gold if placed in a very humid environment (for instance in contact with wet pads). These last observations are very important as they distinguish the interpretation presented in this work (which associates the “gold” color with the moist state) from that conveyed by the widely assumed “hydraulic theory” (which explains that a “red” state is reached under fluid injection).

A. Hydraulic theory

The change of color from yellow (dominant wavelength 580 nm) to red (dominant wavelength 700 nm) can be understood as a change of the multilayer period thickness under the pressure of a fluid injected by the insect, as was proposed earlier for other tortoise beetle species [23,27]. The increase of the period a of a periodic multilayer with average refractive index \bar{n} results in a proportional increase of the dominant reflected wavelength, according to the expression [31]

$$\lambda = 2\bar{n}a,$$

so, a 20% increase of the wavelength means a 20% increase of the layer thickness: a very large change requiring a remarkable elasticity of the layers. Furthermore, we note that, in this explanation, the humid state, with fluid injected in the structure, is the long-wavelength coloration, that is, the red state. The golden or yellow state is, by contrast, the dry state. This, however, contradicts many observations, including the fact that dry, dead insects turn red, and that red dry insects resulting from deep-freezing of an insect in the golden state can be reverted to the metallic gold appearance by exposure to humidity.

Another point against this widely believed interpretation that an injected fluid increases the layers thickness and shifts the reflection band to the red is that it makes no distinction between specular reflection and diffuse scattering: the simple change of average refractive index and layer thickness does not change the translational symmetry of the Bragg reflector, meaning that we would expect a change from metallic gold to metallic red, instead of the diffuse red appearance which is actually observed.

B. Switchable mirror theory

In the present-work “switchable mirror” theory, the explanation is different: the dry specimen contains empty, porous patches, and this provides a set of scatterers that appear at random locations, with random lateral sizes, in the multilayer mirror structure. As the next section will show, these disordered scatterers lead to a “white” diffusion, which means that the optically active multilayer is now a translucent layer, revealing a view of the red substrate. If a fluid enters, the porous patches become filled and the refractive index of the inner layer in all periods becomes homogeneous. This rebuilds a near-to-perfect Bragg mirror, providing the gold coloration and, at the same time, restoring the specularly which leads to a metallic aspect. In this theory, the “gold” state is the humid state, while the “red” state is the dry state. This, evidently, fits the observations better.

The red “pigment” has been observed in transmission optical microscopy well under the layers of the Bragg mirror (see Fig. 4), which confirms that the red coloration does not originate from the same device that produces the gold color. The quality of the pigment may depend on its hydration state and on the hemolymph circulation in the bulk of the elytra, explaining that the red hue of a living disturbed insect is not exactly the same as the red-brown hue of a dead, dry insect. There is a possibility that the red pigment is brought in place by the hemolymph, but our preliminary observations indicate

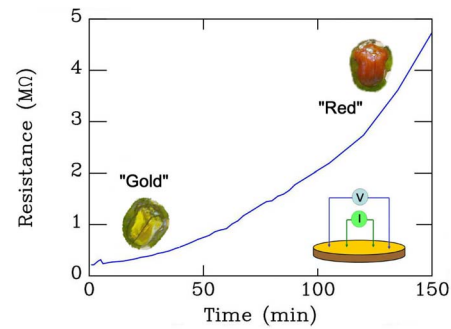


FIG. 10. (Color online) Variation of electrical resistance of an elytron of *Charidotella egregia* between the gold “(resting)” (humid) state and the red “(disturbed)” state, measured with a Kelvin probe. The four-point, or Kelvin, probe method is a common way to measure the resistance of a flat conductor. Two of the probes are used to source current and the other two probes are used to measure voltage. Using four probes eliminates measurement perturbations due to the probe resistance and the contact resistance between the metal probe and the sample.

that the hemolymph of the living insect is clear, so that we tend to believe that the hemolymph role here is only to bring the nutrients to the cells that produce and maintain the quality of the pigments.

The difference between the earlier “hydraulic theory” and the present “switchable mirror theory” then is the contrasting interpretations of the “gold” and “red” states in terms of their liquid contents. The hypothesis that the gold (dominantly yellow) state is associated with the presence of moisture, while the diffuse red appearance is a “dry” state can be tested by a measure of the electric resistance of an elytron removed from a specimen in the gold state, as a function of time in a dry atmosphere, letting the elytron turn passively into the red state. In this experiment, the resistance was measured as a function of time, using the Kelvin probe (four-probes) method, in order to avoid complications arising from non-ohmic contact resistances. The results in Fig. 10 show that as the elytron slowly changes from gold to red, as it loses its natural moisture, the elytron’s electrical resistance increases. Thus, this experiment further supports the observation that the humid state corresponds to the metallic gold appearance, while the dry state gives the diffusive red aspect.

We should not exaggerate the significance of this measurement, however, as the passive desiccation which takes place in this transformation and the resistance recording, deal with the whole elytron, and not solely the active optical layer. The time required to dry the whole elytron was much larger than the time required to make the gold reflector vanish, *in vivo*. This seems to indicate that an active mechanism must be present in the living insect to accelerate the desiccation of the optical layers without requiring the complete evacuation of the elytron. It was, unfortunately, not possible to conduct this experiment on a living specimen during a color change, because the time required for a sample setup under the Kelvin probe far exceeded the reaction time of the insect.

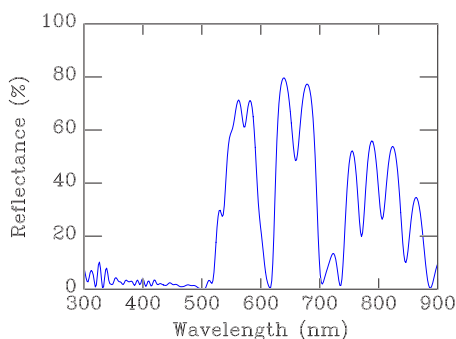


FIG. 11. (Color online) Transfer-matrix reflectance calculation, which describes the spectral reflection from a chirped photonic-crystal film which stacks three Bragg mirrors, selecting yellow, red, and infrared bands, as suggested by scanning electron microscopy data.

C. Numerical simulations in support of the switchable mirror theory

The “gold” state, then, is the humid state, in which the porous patches are filled with a fluid, which makes the wide, partially porous layer in the Bragg mirror optically homogeneous. This turns the complex structure into a one-dimensional chirped photonic crystal which provides both specularity (hence the metallic aspect) and the appropriate spectral selection to explain the gold color. Figure 11 shows the result of a numerical simulation for a structure that matches the layer description in Eqs. (2)–(4). The calculated structure stacks three Bragg mirrors on top of each other. The “distal” mirror (at the multilayer surface) has 12 periods of 186 nm (25% of this thickness is a light junction material with refractive index 1.35 and 75% is a higher-index material with a refractive index 1.55); the intermediate mirror has 12 periods of 200 nm, with the same refractive index distribution as the distal mirror; the proximal mirror (inside) has eight periods of 270 nm, again, with the same index distribution. A noticeable refinement in this calculation is the introduction of a pigmentary absorption for wavelengths shorter than 500 nm, in the form of an imaginary part in the dielectric constant ($\text{Im}[\epsilon]=0.05$). The calculation uses a transfer-matrix approach, that solves Maxwell’s equations with the full detail of vector multiple-scattering (for homogeneous isotropic layers, this reduces to one dimension, and 2×2 scattering matrices) [31]. As usual for unpolarized incident light, we average the transverse-electric and transverse-magnetic reflectances. The agreement is very good, except for the fact that the three mirrors’ contributions appear somewhat narrow: slight random deviations from the ideal layer thicknesses, however, could easily account for this minor discrepancy.

When the porous patches dry, their refractive index lowers and we see the formation of an unorganized distribution of extended scatterers that appear in the multilayer. This can actually destroy the Bragg mirror coherence and transform the optical reflecting layer into a translucent filter that gives a diffuse view on the red-pigmented underlayers. The material that makes up the rigid bulk of the elytron is indeed red, as can be seen when looking at the interior side of the elytron

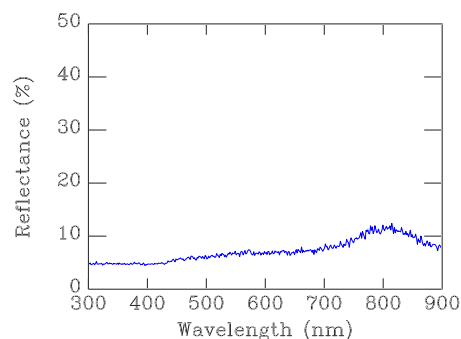


FIG. 12. (Color online) Numerical simulation which describes the reflectance of the chirped photonic-crystal film described in Fig. 11, perturbed by random inhomogeneities. When the refractive index of the layer’s porous part is allowed to fluctuate, the Bragg mirror is destroyed, and what remains is the effect of the pigmentary absorption. The incident light is unpolarized.

after mechanically removing the optically active layer. An indication for the reversible destruction of the Bragg mirror in presence of irregular dry porosity patches is provided by the model calculation shown in Fig. 12. In this model, the structure that provides (when perfect) the spectral reflectance in Fig. 11 is “corrupted” by random fluctuations of the refractive index, in the layers that contain the porous patches. In a given configuration, the refractive index of each layer is made to fluctuate between a maximum of 1.56 (the highest possible index of bulk chitin) and a minimum of 1.4, assuming, from inspection of the TEM images, that the volume occupied by the pores in the core of the layers [lighter areas in Fig. 7(b)] does not exceed 25%. The reflectance is averaged over a large number of random configurations (about 500, enough to stabilize the resulting reflectance spectrum). No correlation was introduced between successive layers, though this could be considered as a possible refinement of the model. As Fig. 12 shows, the bright “gold” reflection is destroyed, leaving only the reddish hue related to the pigmentary absorption of the substrate, included in the model.

VII. CONCLUSION

Charidotella egregia shows two states of coloration. The first one (resting state) can be described as providing a “metallic gold” appearance. It is generated by the multiple reflection and interference of light in a “chirped” multilayer (a stack of thin layers with thicknesses that increase with depth to produce a broadband reflector). The second one (disturbed state) can be described as providing a “diffuse red” of pigmentary origin.

Separately, such appearances are not unique in the insect’s world: there are a large number of documented examples of very conspicuous (often unpalatable) insects which are red in coloration and one can also find numerous examples of beetles displaying a metallic gold aspect [see, for instance the case of *Chrysina (Plusiotis) resplendens*, a beetle from Central America with a static gold metallic appearance, already considered—with erroneous conclusions about the origin of the coloration—by Michelson [34]]. What is outstand-

ingly peculiar in the case of *Charidotella egregia* is that it is able to actively and reversibly toggle between the gold and the diffuse-red states. The physical mechanism in use for this transformation involves a new type of chirped optical stack, made of laterally inhomogeneous porous layers (porous “by patches”), and a deeper-lying pigmentary red layer. When evacuated, the randomly distributed patches contribute to light scattering, turning the multilayer into a translucent cover that leaves a view on the red pigmentary layer. When filled with a liquid, the scattering power of the patches is drastically reduced, so that the light can start interferences in the chirped multilayer. The restoration of the lateral homogeneity favors a specular reflection and a metallic appearance. In the humid state, the surface multilayer then returns a large fraction of the incident light before it reaches the red pigments and a metallic gold appearance prevails.

From the point of view of biological evolution, the ultimate adaptive value of this transformation of appearance is far from being understood, largely because the observation of the insect, correlated with a large number of external circumstances (on the top of a leaf or underside, on a stem or on the ground, mating, hiding, finding a way to food, in presence or absence of a predator, in the sunshine, in dim light or under a heavy tropical rain, etc.), has not been frequent enough to provide reliable statistical data that can be used to answer the question. This is clearly a subject for further study.

From the physics and material science points of view, this natural structure seems to be very interesting: we have a surface which becomes a “metallic” reflector when it acquires moisture. We can refer to this behavior as “hygrochrome,” underlining the change of color with varying hygrometry. Tunable materials like electrochromic films (that

change color with varying applied electric fields) or thermochrome films (that change color with varying temperatures) all have a strong potential for applications in sensing or switching devices. The class of material evolved in *Charidotella egregia* could also find a number of uses in developing optical vapor sensors [35] or optical liquid gauges. Such optical sensors might be particularly appropriate, for instance, in detonating environments, when the use of classic electric probes would be too hazardous.

ACKNOWLEDGMENTS

This work was initiated and largely carried out during the stay of two of the authors (J.P.V. and J.M.P.) at the Smithsonian Tropical Research Institute (Panama). One of the authors (J.P.V.) acknowledges a travel grant from the Belgian National Fund for Scientific Research (FNRS). The study was also partly supported by the EU through FP6 BIOPHOT (NEST/Pathfinder) 012915 project and by the European Regional Development Fund (ERDF) and the Walloon Regional Government under the “PREMIO” INTERREG IIIa project. The authors acknowledge the use of Namur Interuniversity Scientific Computing Facility (Namur-ISCF), a common project between the FNRS and the University of Namur (Belgium). Two of the authors (J.P.V. and J.M.P.) acknowledge the logistic support supplied by the Smithsonian Tropical Research Institute in Panama. One of the authors (M.R.) was supported by the Belgian Fund for Industrial and Agonomic Research (FRIA). One of the authors (V.L.) was supported by the Belgian FNRS. The authors thank Professor Amand Lucas for many discussions and suggestions. The authors thank Marcos A. Guerra for providing Fig. 1 of this paper.

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- [26] A single slab of transparent material (Fabry-Pérot structure) with low refractive index selects specific colors in reflection, due to the interference of the waves multiply reflected from its surfaces. When many slabs are stacked on top of each other (but still separated by air or another material with a different refractive index), the resulting structure (a multilayer) also selects spectral bands for reflection, but much narrower and much more intense than for a single slab. Weak refractive in-

dex multilayer stacks are selective mirrors that reflect very saturated colors. When all the slabs have the same thickness we have a periodic multilayer, also called a Bragg mirror. If the thicknesses of the slabs change slightly from one slab to the next, we have a *chirped* multilayer, which is usually a high-efficiency broadband (wide-spectrum) reflector.

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