Structural colours of nickel bioreplicas of butterfly wings

Tomas Tolениs\textsuperscript{a}, Stephen E. Swiontek\textsuperscript{b} and Akhlesh Lakhtakia\textsuperscript{b}

\textsuperscript{a}Center for Physical Sciences and Technology, Vilnius, Lithuania; \textsuperscript{b}Department of Engineering Science and Mechanics, Nanoengineered Metamaterials Group, Pennsylvania State University, University Park, PA, USA

**ABSTRACT**

The two-angle conformally evaporated-film-by-rotation technique (TA-CEFR) was devised to coat the wings of the monarch butterfly with nickel in order to form a 500-nm thick bioreplica thereof. The bioreplica exhibits structural colours that are completely obscured in actual wings by pigmental colours. Thus, the TA-CEFR technique provides a way to replicate, study and exploit hidden morphologies of biological surfaces.

**ARTICLE HISTORY**

Received 1 September 2015
Accepted 23 November 2015

**KEYWORDS**

Biomimetic optics; bioreplication; hidden structures; structural colour

1. Introduction

Humans have had a long-standing fascination with colours for both beauty and diverse functionalities, such as recognition and camouflage. Light either emanating from or scattered by an object has a physical spectrum that is captured by the eyes and interpreted by the brain. Colours of biological objects are very commonly due to chemical pigments and dyes \( (1) \). Other colours have purely physical origins based on commonplace optical phenomena such as scattering, diffraction and interference \( (2, 3) \). Often, complex colour patterns have both physical and chemical origins \( (4) \).

Susceptible to environmental insults through exposure to high temperature, moisture and sunlight, colours originating from chemicals alter and even fade over time \( (5) \). In contrast, colours originating from physical mechanisms are environmentally stabler. Since light rays do not carry information on the mechanism of colour production, our eyes cannot discriminate between colours based on their origin, tempting researchers to replace chemical colours by physical colours. An additional benefit may be the reduction in the release of hazardous volatile organic compounds during the manufacture and deployment of pigments and dyes.

Considerable progress has been made during the last two decades towards reproducing the iridescent blues of the wings of many butterfly species in the *Morpho* genus \( (6, 7) \). Although granules of pigments such as bioppterin are present in the scales of the wings of some *Morpho* species \( (8, 9) \), the iridescent blues are of physical origin. These colours result from a collaboration of three structural effects: multilayer interference, the diffraction-grating effect due to a periodic array of ridges on the scales on the wings, and the nonuniformity of the heights of those ridges \( (10) \).

In a biomimetic process to reproduce the structural features of the *Morpho* wing, first an irregularized surface grating is stamped on a resin film and then a periodic SiO\textsubscript{2}/TiO\textsubscript{2} multilayer is deposited on the grating \( (6) \). This process is suitable to colourize small surfaces, e.g. flakes that can be dispersed in paints, cosmetics and security inks \( (3, 7) \). In a bioinspired process that excludes both the surface grating and its irregularization, a periodic multilayer of two optically distinct polymers is cut up into yarn which is then embedded in a third polymer and flattened \( (11) \). The composite yarn has been woven into nonpigmented but structurally coloured fabrics that however lack the attractive iridescence of the *Morpho* blues \( (3) \).

Butterfly wings are so delicate that an actual wing is not used in either of the two foregoing processes. Yet the deposition of a nanoscale thin film on a butterfly wing ought to capture the surface structure of the wing. In a bioreplication \( (12) \) process, first a thin film of nickel was conformally deposited on the wing of a monarch butterfly (*Danaus plexippus*) and then the wing was plasma-ashed away, leaving behind a bioreplica \( (13) \). Whereas no pigmental contributions to the colours on the wing would be displayed by the nickel replica, some structural contributions to the wing colours should be. Multilayer interference would not contribute because the bioreplica is made of a single material, but surface gratings, whether strictly periodic or somewhat irregularly periodic, would.
Figure 1. Monarch (Danaus plexippus) butterfly. (The colour version of this figure is included in the online version of the journal.)

The nickel thin film was deposited using the conformally evaporated-film-by-rotation (CEFR) technique (13). The wing was mounted on a planar substrate, the substrate was affixed to a platform that could rotate rapidly about a central normal axis passing through it, and the entire assembly was installed in a vacuum chamber. Also present in the vacuum chamber was a metal boat containing nickel that could be evaporated by passing an electric current through the boat. After suitably depressurizing the vacuum chamber, nickel was evaporated. A \( \sim 250 \) nm thick columnar thin film of nickel condensed conformally on the wing. However, structural colours were not visually evident on the bioreplica.

Hence, we devised the two-angle CEFR (TA-CEFR) technique for better replication of the surface morphology. The nickel bioreplica of the \( D. \) plexippus hindwing turned out to display several colours dependent on the viewing angle. We report this development in this communication. Section 2 describes the sample preparation, the implementation of the TA-CEFR technique, and the imagery of the bioreplica. Section 3 presents and discusses the experimental results obtained. The communication ends in Section 4 with some concluding remarks.

2. Materials and methods

One notices from Figure 1 that the wings of a monarch butterfly are dominated by orange sections separated by blackish lines and possess blackish fringes with whitish spots. The orange hues are caused by mixtures of pigments called pterins and ommochromes (14) present in the scales that the wings comprise (4). The scales possess an array of long parallel ridges separated by crossribs, much like a ladder. These crossribs ought to function as surface gratings, but any consequent structural colours are completely overshadowed by the pigmental colours.

The dead \( D. \) plexippus specimen used by us was donated by Folk’s Butterfly Farm (Nescopeck, PA, USA). The left hindwing (Figure 2(a)) of the specimen was removed and was used as the sample. It was first washed with ethanol to remove particulates and then dried under the intense light of a tungsten light bulb. Next, the sample was mounted on a borosilicate glass substrate via copper tape (Uline, Pleasant Prairie, WI, USA). Finally, the entire assembly was mounted on a planar platform using Kapton® tape (Uline, Pleasant Prairie, WI, USA). This tape can withstand high temperature, produces negligible outgassing during the deposition and maintains its adhesive capabilities throughout that process.

The planar platform with the sample and the glass substrate was mounted on the axle of a computer-controlled stepper motor inside a custom-built vacuum chamber equipped to perform resistive -heating physical vapour deposition. The chamber contained a 3.2-mm wide tungsten trough (S4-.005W, R. D. Mathis, Long Beach, CA, USA) in which intertwined strands of nickel wire (99.6% pure, 0.5 mm diameter) were placed. The distance between the sample and the nickel strands was fixed at roughly 15 cm. The chamber was closed and the pressure was reduced to \( \sim 50 \) \( \mu \)Torr. The stepper motor was turned on to rotate the sample at 120 rpm about the axle. A second computer-controlled stepper motor then oriented the platform so that during deposition the collimated vapour of nickel would arrive at an angle \( \chi_v = 7^\circ \) with respect to the platform plane.

About 87-A current was then passed through the tungsten trough to generate the nickel vapour. A quartz crystal monitor mounted close to the sample was used to maintain the deposition rate at \( \sim 1 \) nm\( \cdot \)s\(^{-1}. \) The deposition lasted until a \( \sim 250 \) nm thick film of nickel was deposited. Then the current was switched off, the motors were stopped and the chamber was exposed to the atmosphere.

The foregoing process was repeated to deposit another columnar thin film of nickel on top of the first one. Whereas the thickness of the second thin film was also \( \sim 250 \) nm, it was deposited with the angle \( \chi_v = 20^\circ. \) Removal of the wing by plasma ashing was not undertaken as it is unnecessary for the optical investigation of the nickel bioreplica.

The bioreplica structure was analysed with a scanning electron microscope (SEM) (FEI Nova NanoSEM 630, Hillsboro, Oregon, USA). A small piece of the coated hindwing was cut off for analysis. Since the nickel film is conductive, no additional coating by gold (or some other metal) was necessary to prevent charging of the bioreplica by the impinging electrons during imaging.
Figure 2. Photographs of the left hindwing (a) before the implementation of the TA-CEFR technique, (b) after the first nickel thin film was deposited with the collimated vapour directed at angle $\chi_v = 7^\circ$, and (c) after the second nickel thin film was deposited with the collimated vapour directed at angle $\chi_v = 20^\circ$. (The colour version of this figure is included in the online version of the journal.)

Figure 3. Schematic of the system used for photographing structural colours. A CFL bulb was used to illuminate the bioreplica from a direction at 70$^\circ$ with respect to the normal to the mean plane of the bioreplica. A camera was mounted on a stepper motor. The angle $\psi \in [-40^\circ, 20^\circ]$ between the camera and the normal to the mean plane of the bioreplica was changed in steps of 10$^\circ$.

Structural colours were imaged by a Nikon D90 camera with a 105-mm lens. The schematic of the experimental set-up is shown in Figure 3. A 7-W 330-lumen compact fluorescent light (CFL) bulb was chosen as the light source since it is a common white-light source for indoor use. The bioreplica was mounted on a rotation stage, and the angle of incidence of white light was fixed at 70$^\circ$ with respect to the normal to the mean plane of the bioreplica. The angle $\psi \in [-40^\circ, 20^\circ]$ between the camera and the normal to the mean plane of the bioreplica was changed in steps of 10$^\circ$. At each value of $\psi \in \{-40^\circ, -30^\circ, \ldots, 10^\circ, 20^\circ\}$, the bioreplica was photographed with the CFL bulb turned on.

3. Results and discussion

The first deposition (with $\chi_v = 7^\circ$) would lead to the formation of highly slanted columns of nickel, whereas the second deposition (with $\chi_v = 20^\circ$) would lead to the formation of less slanted columns. This TA-CEFR method was devised to allow nickel vapour during the first deposition to penetrate deep into the butterfly wing and thus replicate the surface structure with higher fidelity during the second deposition.

After the first deposition, the native orange hues of the hindwing, although muted, were still evident through the nickel thin film, as captured by the photograph in Figure 2(b). After the second deposition, the orange hues were replaced by a whole gamut of colours that were obvious to the naked eye. These colours can be seen in the photograph in Figure 2(c). The colours were different from different viewing directions, as is characteristic of colours arising from diffraction by surface gratings (2, 3).

Figure 4 provides two SEM images of the nickel bioreplica. The wing is made up of scales of lateral

| $\psi$  | $\lambda_o$ (nm) for $m = -1$ | $\lambda_o$ (nm) for $m = -2$ |
|--------|----------------|----------------|
| 20$^\circ$ | 299 | 149 |
| 30$^\circ$ | 383 | 192 |
| 0$^\circ$ | 470 | 235 |
| $-10^\circ$ | 557 | 278 |
| $-20^\circ$ | 641 | 320 |
| $-30^\circ$ | 720 | 360 |
| $-40^\circ$ | 791 | 396 |

Note: Boldface values lie in the visible regime.
dimensions \(\sim 100 \mu m\). Each scale has parallel ridges spaced about 2.2 \(\mu m\) apart. The crossribs between the ridges are spaced about 500 nm apart. Diffraction gratings with periodicity of 500 nm can produce all visible colours via low-order diffraction. Using standard data for nickel (15), we determined that the specular reflectance of a 500-nm thick nickel film for normally incident light varies from about 50\% (at free-space wavelength \(\lambda_0 = 400 \text{ nm}\)) to about 70\% (at \(\lambda_0 = 800 \text{ nm}\)). Therefore, the nickel bioreplica can function as a reflection grating.

To observe all the colours exhibited by the bioreplica, a small piece of it was removed and mounted on a rotational stage. By fixing the angle of the light source at 70\(^\circ\) from the normal to the mean plane of the piece of the bioreplica and changing the angle of observation, colours were captured by the Nikon D90 camera (Figure 3). As \(\psi\) decreased from 20\(^\circ\) to –40\(^\circ\), the captured colours changed from blackish violet to blue to bluish green to greenish yellow to copper and finally to orangish red overlaid by violet and blue, as may be gathered from Figure 5.

In order to confirm that the captured colours were instigated by the crossribs of the wing scales replicated by the nickel film, the diffraction wavelength

\[
\lambda_\alpha(\psi) = \frac{d}{m} (\sin \psi - \sin \psi_{\text{ref}}), \quad m \in \{0, \pm 1, \pm 2, \ldots\},
\]

was calculated as a function of \(\psi\), with \(m\) as the diffraction order, \(d\) as the average spacing between two successive crossribs and \(\psi_{\text{ref}}\) as the value of \(\psi\) corresponding to specular reflection. We set \(d = 500 \text{ nm}\) according to measurements made on the SEM images and \(\psi_{\text{ref}} = 70^\circ\) according to the experimental set-up shown in Figure 3.

The diffraction wavelengths \(\lambda_\alpha(\psi)\) are listed in Table 1 for \(m \in \{-1, -2\}\). The boldface entries in this table lie within the spectral regime visible to most humans. Due to irregularities in the inter-crossrib spacing as well as the nonplanarity of the wing, every value of \(\lambda_\alpha(\psi)\) provided in Table 1 is not to be considered as precise, but as representative of a small range. When \(\psi = 20^\circ\), the nickel bioreplica looks blackish violet in Figure 5(a). The bluish colour in Figure 5(b) for \(\psi = 10^\circ\) is matched by \(\lambda_\alpha = 383 \text{ nm}\) for \(m = -1\), the bluish green colour in Figure 5(c) for \(\psi = 0^\circ\) by \(\lambda_\alpha = 470 \text{ nm}\) for \(m = -1\), the greenish yellow colour in Figure 5(d) for \(\psi = -10^\circ\) by \(\lambda_\alpha = 557 \text{ nm}\) for \(m = -1\), the copper colour in Figure 5(e) for \(\psi = -20^\circ\) by \(\lambda_\alpha = 641 \text{ nm}\) for \(m = -1\), the orangish red colour in Figure 5(f) for \(\psi = -30^\circ\) is matched by \(\lambda_\alpha = 720 \text{ nm}\) for \(m = -1\), and the violet overlay in the same figure by \(\lambda_\alpha = 360 \text{ nm}\) for \(m = -2\). Finally, the orangish red colour in Figure 5(f) for \(\psi = -40^\circ\) is overlaid by a blue colour that is matched by \(\lambda_\alpha = 396 \text{ nm}\) for \(m = -2\). Thus, the viewing-angle-dependent colours displayed by the bioreplica are structural colours produced by the diffraction of light by the (almost) periodically spaced crossribs.

### 4. Concluding remarks

The TA-CEFR technique was devised to coat nickel on the hindwing of a monarch butterfly and thereby replicate its surface morphology. Structural colours observed on the 500-nm thick nickel bioreplica were engendered by the periodic arrays of crossribs on the wing scales. These structural colours, which are completely hidden by pigmen-tal colours in the monarch-butterfly wings, thereby were uncovered. Thus, the TA-CEFR technique provides a way to replicate, study and exploit hidden morphologies of biological surfaces.

In closing, let us note that the physical basis of structural colour can also be exposed to varying degrees of success by other bioreplication techniques (12) such as the solution-based infiltration methods (16, 17), electroless deposition (18) and atomic layer deposition (19). Electro-
Figure 5. Colours of the nickel bioreplica captured by the camera at different values of the angle $\psi$: (a) $20^\circ$, (b) $10^\circ$, (c) $0^\circ$, (d) $-10^\circ$, (e) $-20^\circ$, (f) $-30^\circ$, and (g) $-40^\circ$. (The colour version of this figure is included in the online version of the journal.)
less deposition and infiltration methods are suitable for volumetric structures, whereas atomic layer deposition is suitable for areal structures, but none of these three techniques is likely to be industrially scalable. In contrast, by thickening the TA-CEFR nickel film via electroless deposition \(^{(20)}\), one could make robust moulds to stamp sheets and fabrics to exhibit structural colours. We note that a multi-angle CEFR technique may have to be devised for better bioreplication of certain specimen. Furthermore, the stamped sheet or fabric may be made of a carefully selected material so that functionalities in addition to structural colouration could be realized \(^{(21)}\).

**Disclosure statement**

No potential conflict of interest was reported by the authors.

**Funding**

This work was partially supported by the project Promotion of Student Activities [VP1-3.1-ŠMM-01-V-02-003] from the Research Council of Lithuania to TT. This project is funded by the Republic of Lithuania and European Social Fund under the 2007–2013 Human Resources Development Operational Programme’s priority 3. Additional funding was provided to AL and SES by the Charles Godfrey Binder Endowment at the Pennsylvania State University.

**References**

(1) Lee, D. *Nature’s Palette: The Science of Plant Colors*; University of Chicago Press: Chicago, 2007.
(2) Vigneron, J.-P.; Simonis, P. In *Advances in Insect Physiology. Vol. 38: Insect Integument and Colour*; Casas, J., Simpson, S.J., Eds.; Elsevier: Oxford, 2010; Chapter 5.
(3) Dushkina, N.; Lakhtakia, A. In *Engineered Biomimicry*; Lakhtakia, A., Martin-Palma, R.J., Eds.; Elsevier: Waltham, 2013; Chapter 11.
(4) Ghiradella, H. In *Advances in Insect Physiology. Vol. 38: Insect Integument and Colour*; Casas, J., Simpson, S.J., Eds.; Elsevier: Oxford, 2010; Chapter 4.
(5) Wicks Jr., Z.W.; Jones, F.N.; Pappas, S.P.; Wicks, D.A. *Organic Coatings: Science and Technology*, 3rd ed.; Wiley: Hoboken, NJ, 2007.
(6) Saito, A.; Miyamura, Y.; Ishikawa, Y.; Murase, J.; Akai-Kasaya, M.; Kuwahara, Y. *Proc. SPIE* **2009**, 7205, 720506.
(7) Saito, A.; Murase, J.; Yonezawa, M.; Watanabe, H.; Shibuya, T.; Sasaki, M.; Ninomiya, T.; Noguchi, S.; Akai-Kasaya, M.; Kuwahara, Y. *Proc. SPIE* **2012**, 8339, 83390C.
(8) Kumazawa, K.; Tabata, H. *Zool. Sci.* **1996**, 13, 843–847.
(9) Tabata, H.; Hasegawa, T.; Nakagoshi, M.; Takikawa, S.; Tsusue, M. *Experientia* **1996**, 52, 85–87.
(10) Kinoshita, S.; Yoshioka, S.; Kawagoe, K. *Proc. R. Soc. Lond. B* **2002**, 269, 1417–1421.
(11) Johara, K.; Yoshimura, M.; Tabata, H.; Shimizu, S. *Chem. Fibers Int.* **2000**, 50, 38–39.
(12) Pulsifer, D.P.; Lakhtakia, A. *Bioinsp. Biomim.* **2011**, 6, 031001.
(13) Pulsifer, D.P.; Lakhtakia, A.; Martín-Palma, R.J.; Pantano, C.G. *Proc. SPIE* **2011**, 7975, 797500.
(14) Davis, A.K.; Chi, J.; Bradley, C.; Alitzer, S. *PLoS One* **2012**, 7, e41323.
(15) Ordal, M.A.; Long, L.L.; Bell, R.J.; Bell, S.E.; Bell, R.R.; Alexander Jr., R.W.; Ward, C.A. *Appl. Opt.* **1983**, 22, 1099–1119.
(16) Li, B.; Zhou, J.; Zong, R.; Fu, M.; Bai, Y.; Li, L.; Li, Q. *J. Am. Ceram. Soc.* **2006**, 89, 2298–2300.
(17) Tan, Y.; Zang, X.; Gu, J.; Liu, D.; Zhu, S.; Su, H.; Feng, C.; Liu, Q.; Lau, W.M.; Moon, W.-J.; Zhang, D. *Langmuir* **2011**, 27, 11742–11746.
(18) Tan, Y.; Gu, J.; Zang, X.; Xu, W.; Shi, K.; Xu, L.; Zhang, D. *Angew. Chem. Int. Ed. Engl.* **2011**, 50, 8307–8311.
(19) Huang, J.; Wang, X.; Wang, Z.L. *Nano Lett.* **2006**, 6, 2325–2332.
(20) Sudagar, J.; Lian, J.; Sha, W. *J. Alloys Compd.* **2013**, 571, 183–204.
(21) Lakhtakia, A. *Bioinsp. Biomim. Nanobiomater.* **2015**, 4, 168–173.