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Insect micro-morphology and its applications

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Abstract
The study of form and structure of an insect at a microscopic level is known as insect micro-morphology. This review emphasizes some of the spectacular micro-structures present in insects namely gyroid photonic crystals in butterflies and weevils, brochosomes in leaf hoppers, neck region of dragonflies, labium of rove beetles, hypochaeta in forewings of fairyflies, hydrophobic structures in springtails, stridulatory organs in crickets, tymbal organs in cicadas, anti-reflective coatings in glass wing butterfly, facet lenses in robber fly, ocelli in halictid bees, reaction chamber in bombardier beetles and the keel of stick insects. The gyroid photonic crystals are essential for the reflection of colour in certain butterflies and weevils. The brochosomes secreted by the malphigian tubules of leaf hoppers covers their integument which acts as a protective covering. The further studies on the destruction of brochosomal layer would be helpful to maximize the parasitization and insecticidal efficiency. The knowledge on the similar other micro-structures might be worthy for species identification.

Keywords: Photonic crystals, coloration, brochosomes, malphigian tubules, hydrophobicity

Introduction

Photonic Crystals

The gyroid photonic crystals were first discovered by Yablonovitch, which was published in Scientific American magazine. These nano-structures selectively reflect parts of the visible spectrum and creates distinctive iridescent look. They interact with light to produce structural colour, they are distinct from dyes and pigments. These crystals are made out of chitin. The development of colour can be determined by two factors namely a) crystal size which makes up each scale b) the amount of chitin used to make up the entire crystal. Hence, the larger scales.

have larger crystalline structure and higher amount of chitin which reflects red light whereas smaller scales have smaller crystalline structure and lower amount of chitin which reflects blue light. The colour variations are generated by orienting the crystal structure at different angles which reflects different wavelengths of light. Today, they appeared in many areas of science, technology, medicine and as a product of nature in the biological world [1][11].

Atoms are arranged in straight rows in a 3-D periodic pattern to form crystal structure. A small part of the crystal that can be repeated to form the entire crystal. The study of science of light generation and detection through emission, transmission, modulation, signal processing, switching, amplification and sensing is called photonics. The gyroid membrane structure is occasionally found inside the cells. They have photonic band gaps that make them potential photonic crystals.

The photonic crystals are present in lycaenid butterflies like green hair streak (*Callophrys* sp.), Goodson's green streak (*Cyanophrys* sp.), Plumbeous hair streak (*Thecla* sp.) and nymphalid butterflies like blue morpho (*Morpho rhetenor),* Striped blue crow *(Euploea mulciber)* [1][5]. This photonic crystals are also present in coleopteran weevils like rainbow weevil, *Pachyrrhynchus congestus pavonius* [11]

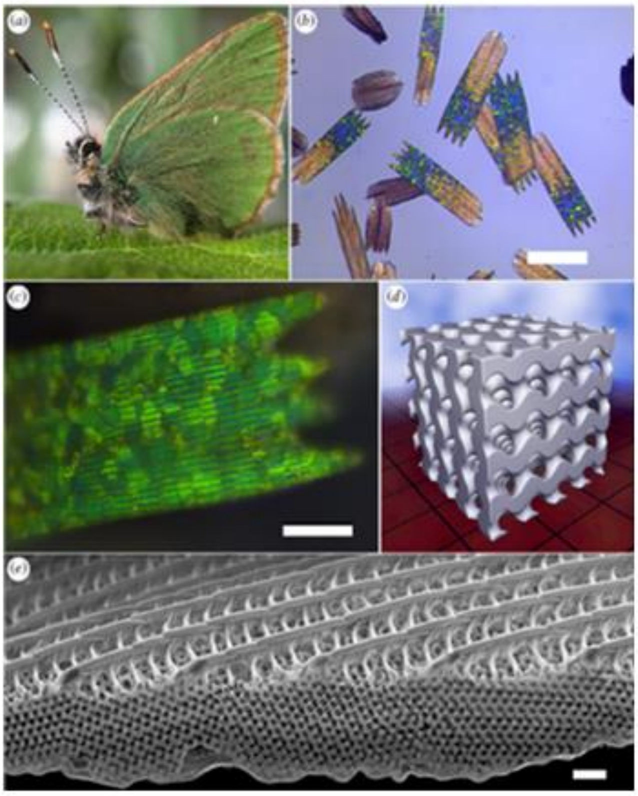


Fig 1: Photonic crystals in Lycaenid butterflies [1]

a. Green hair streak - *Callophrys* sp.
b. Image of both coloured and brown scales under lower magnification
c. Coloured wing scale showing characteristic green–yellow colour arises from many individual photonic crystal domains under higher magnification
d. Computer generated single gyroid model - 4×4×4 unit cells
e. SEM image of a coloured wing scale showing the ribbed upper surface, an undulating lower plate and five porous, single gyroid domains in different orientations.
Scale bars: (b, c and e) 100, 20 and 1 mm, respectively

Fig 2: Photonic crystals in Nymphalid butterflies [4]

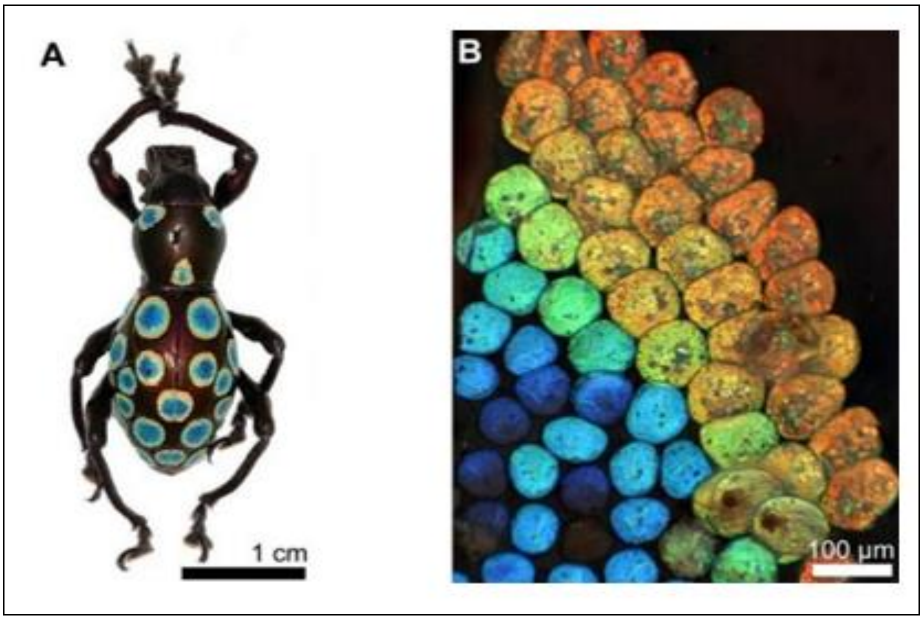


Fig 3: Photonic crystals in Rainbow weevil, *Pachyrrhynchus congestus pavonius* [11]

Applications

This photonic crystals are highly used in the physical science for the manufacturing of following materials

1. Silicon nano-photonic integrated circuits
2. Photonic bandgap fibers for precision surgery and cancer therapy
3. Photonic crystal enhanced light emitting diodes
4. Photonic crystal microwave cavities for high energy particle accelerators
5. Photo crystal optical fiber [4]

Brochosomes

Introduction

The brochosomes are microscale granules with nanoscale indentations. It looks like a soccer ball with 20 hexagonal and 12 pentagonal faces. The bottoms of the hexagonal and pentagonal surface compartments usually have openings leading into the central cavity. It is secreted by the leaf hoppers (Cicadellidae: Hemiptera) and found on their body surface and eggs. They are synthesized in the glandular segments of the Malpighian tubules of leafhoppers.

Brochosomes originates in Golgi complex and acquire their final shape before leaving secretory cells. It appears like a honeycomb structure and made out of proteins (brochosomins) and lipids [7][10].

The brochosomes was first described in 1952 with the aid of an electron microscope. The word brochosomes was described from Greek words *brochos* (mesh of a net) and *soma* (body), refers to the characteristic reticulated surface of the granules. They are having hollow spherical shape with 0.2-0.7 µm in diameter [13][15].

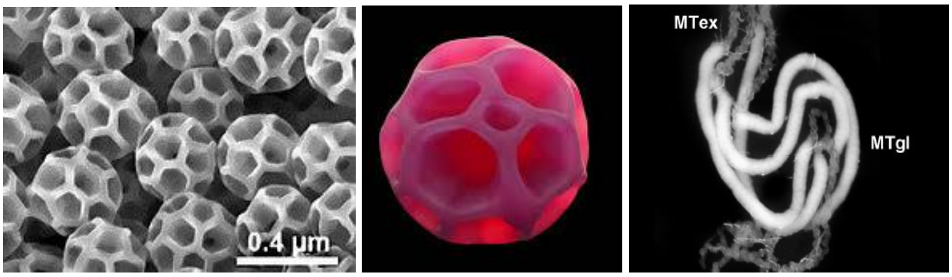


Fig 4: Structure of brochosome and malphigian tubule

Classification of brochosomes [9]

S. No.	Shape	Size
1.	Small spherical BS (♂)	0.3-0.7 µm
2.	Large spherical BS	0.7 -1.4 µm
3.	Irregular spherical BS	0.5- 1.1 µm
4.	Rod like BS (♀)	3.5- 11 µm length; 0.6- 1.2 µm width

Functional types of brochosomes [2]

1. Integumental brochosomes
2. Egg brochosomes

	Integumental brochosomes	Egg brochosomes
Use	Distributed across the integument	Placed onto egg nests made in plant tissues or directly on to eggs
Distribution	Most of the leafhoppers species	Certain genera of tribe *Proconiini* (*Oncometopia, Homalodisca, Molomea, Acrogonia)*
Produced by	Males, females and nymphs	Gravid females only
Function	Protection from water and excrement	Protection of eggs
Size	0.2-0.7 µm	1.0-20.0 µm
Shape	Spherical	Elongate, rarely spherical

Super hydrophobicity: water repellent properties

Rakitov (2002) conducted an experiment with two specimens A and B to study the water repellent properties of *Nionia palmeri*. The specimen A was poorly coated with brochosomes whereas specimen B was coated with thick orange brochosome coat. The result of an experiment revealed that the specimen B have got a rough hydrophobic surfaces.

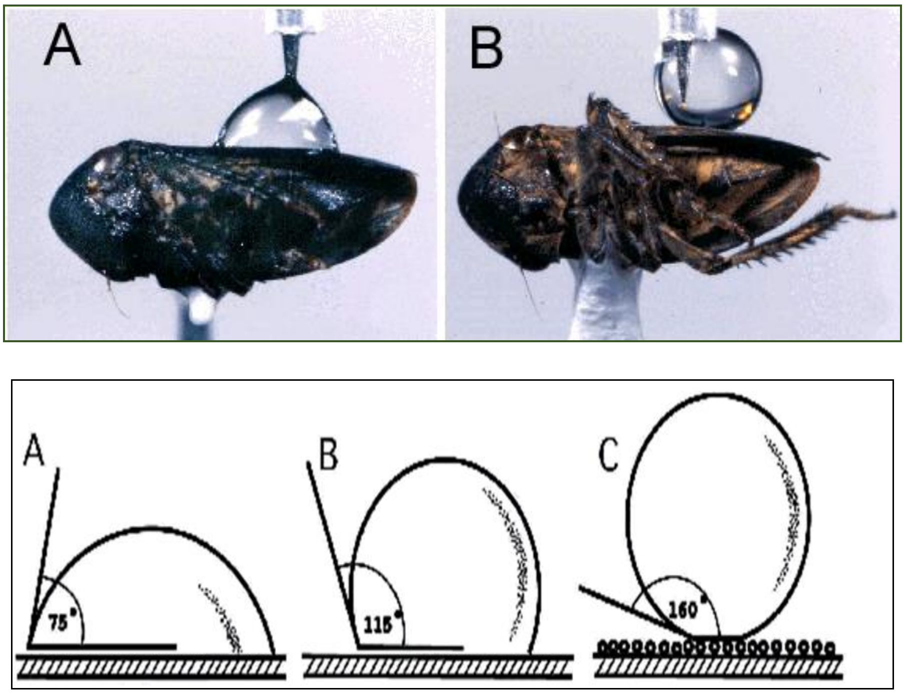


Fig 5: Super hydrophobicity nature of *Nionia palmeri* [10] (a) Smooth hydrophilic (b) Smooth hydrophobic (c) Rough hydrophobic

Also an experiment by Rakitov and Gorb, 2013 suggested that the brochosomes act as water-repellent and anti-adhesive protective barrier. The contact angle (CA) of water, diiodomethane, ethylene glycol and ethanol on detached wings of the leafhoppers like *Alnetoidia alneti*, *Athysanus argentarius* and *Cicadella viridis* was measured. Brochosome-coated integuments showed an average water CA of 165-172° whereas brochosome-removed integuments showed water CA of only 103-129°. The integuments were removed using peeling technique with a fluid called polyvinylsiloxane. Hence, a super hydrophobicity nature of an intact brochosome-coated integuments was proved.

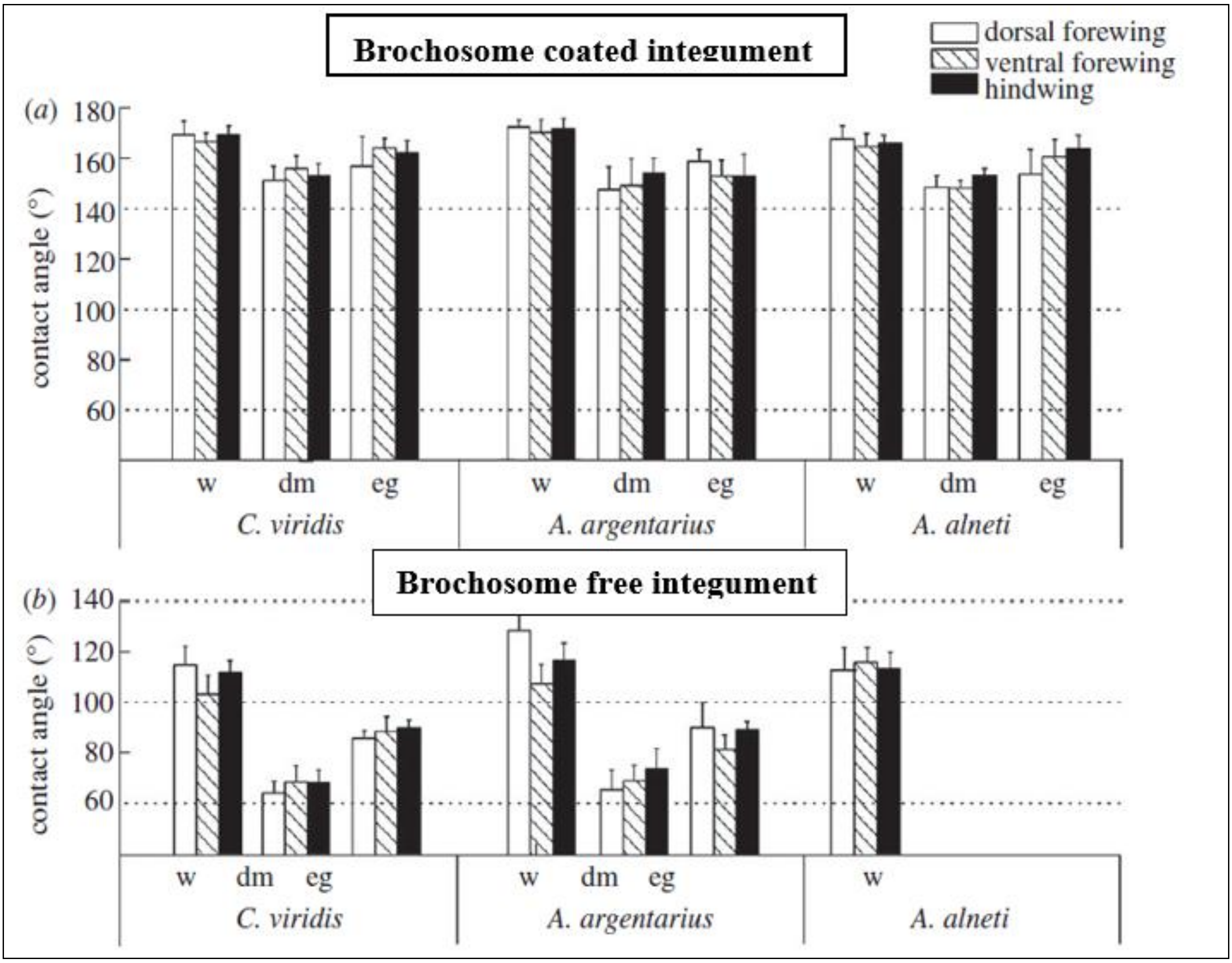


Fig 6: Contact angle measurement of *C. viridis, A. argentarius* and *A. alneti* [6]

Behaviors in leaf hopper
a. Anointing

Fig 7: Anointing behaviour in leaf hoppers

After each molt, most leafhopper species release droplets of the brochosome-containing fluid through the anus and actively spread them over the newly formed integument. The nymph using hind legs, picks up the single drop of the fluid exuded from the rear end and then smears it by kneading movements of all the legs on to the ventral part of the body.

During this process, they balances itself on the upper surface of the plant by rostrum and face whereas on the undersurface it hangs by its beak alone. Then, the insect returns to its normal posture and rests

b. Bathing:

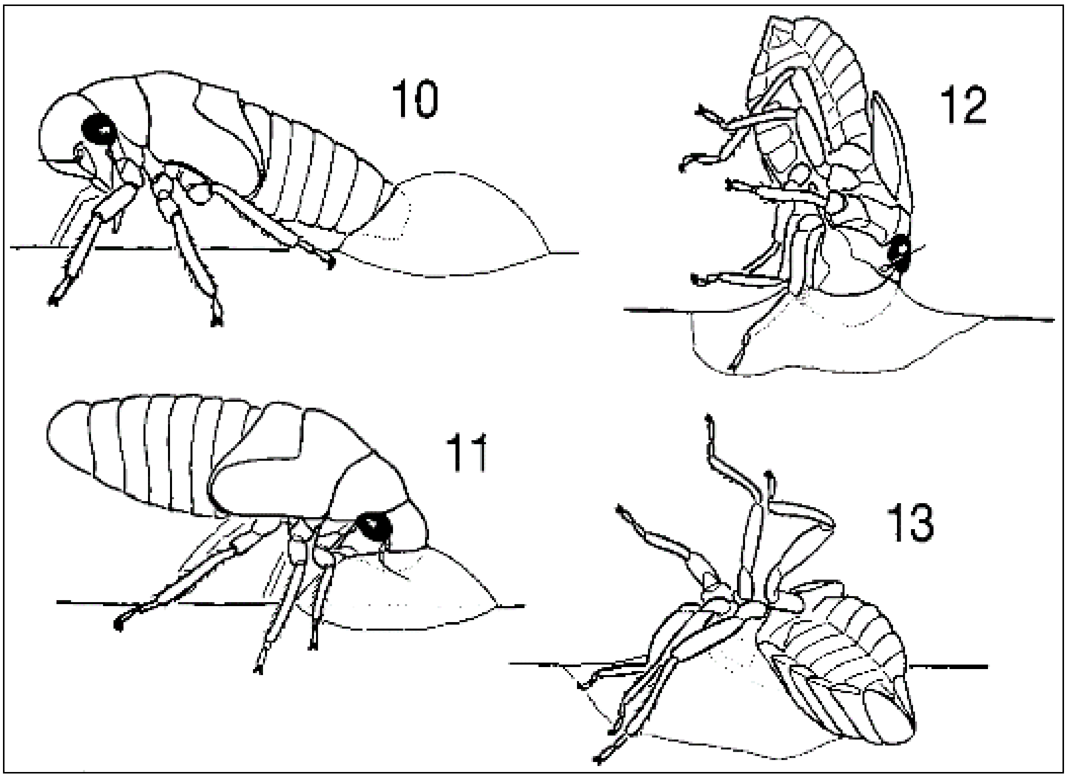


Fig 8: Bathing behaviour in the fifth instar nymph of *Oncopsis flavicollis* (L.)

A peculiar type of anointing is referred to as bathing, usually occurs in fourth instar and fifth instar nymphs after 90-180 minutes of moulting. The resting nymph suddenly starts to crawl and a few seconds later it exudes a single drop of fluid onto the plant surface. Thereupon, it turns around and dips its head into the drop. Later the nymph lays down on its side into the fluid and turns onto its back. This sequence of events recurs several times until the whole drop is spread onto the body. As a result the entire surface of the cuticle becomes wet with the secretion. Thereafter, the secretion will be dried up [8].

c. Grooming

The grooming is the hygienic procedure aimed to keep the body and appendages clean of contaminants. The main function of this behaviour is to manipulate the regular, continuous coat of dry brochosomes on the integument. Unlike anointing and bathing, grooming can be observed at any time [8].

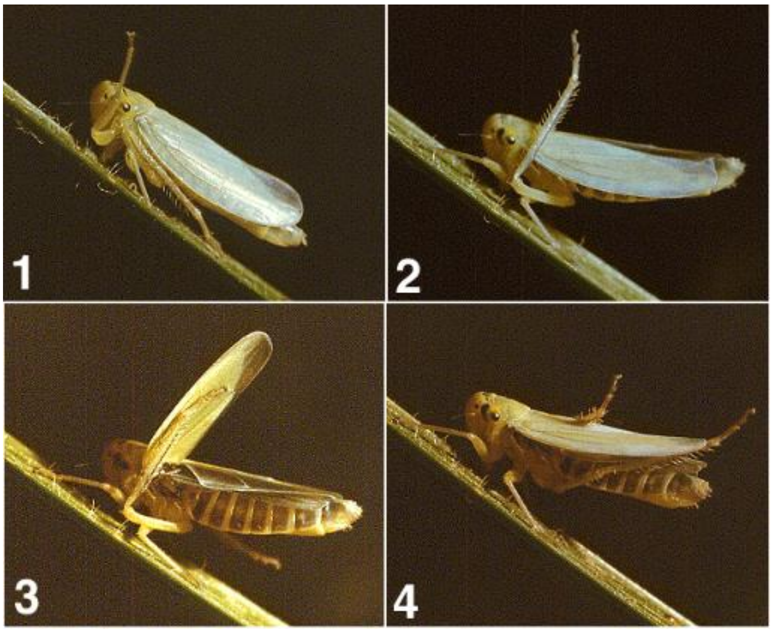


Fig 9: Grooming behaviour in *Cicadella viridis* (L.)

Powdering of egg brochosomes on oviposition sites:
Most leafhoppers insert their eggs into living plant tissues. In certain genera of tribe *Proconiini*, shortly before oviposition, a female places droplet of the brochosome suspension onto its forewings, where they dry as a pair of conspicuous white pellets. During oviposition, the female scrapes this white pellets with its hind tibiae onto the oviposition site making a pulverulent coat.

Fig 10: Brochosomes powdering in egg nests of *Homalodisca coagulata* [14]

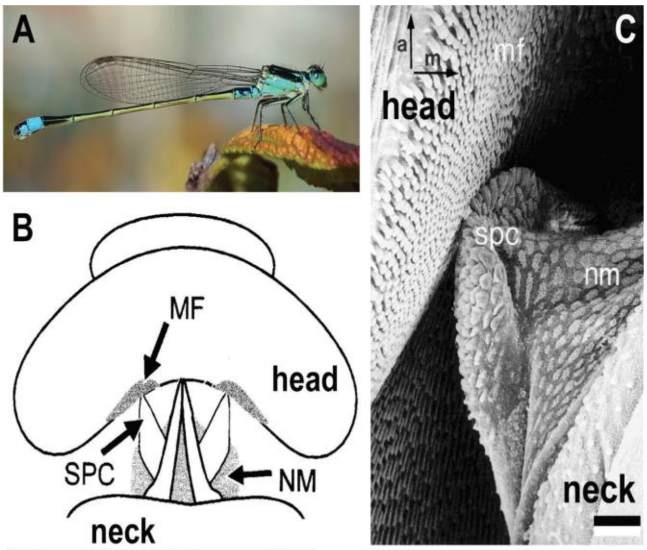
Functions of brochosomes

1. The leafhoppers protect their oviposition sites with brochosomes there by predation/ parasitization efficiency can be minimized.
2. The super hydrophobic nature of brochosomes repels away the water which in turn reduces the insecticidal efficiency.
3. It also gave protection against microbial pathogens, desiccation and UV light.

Applications

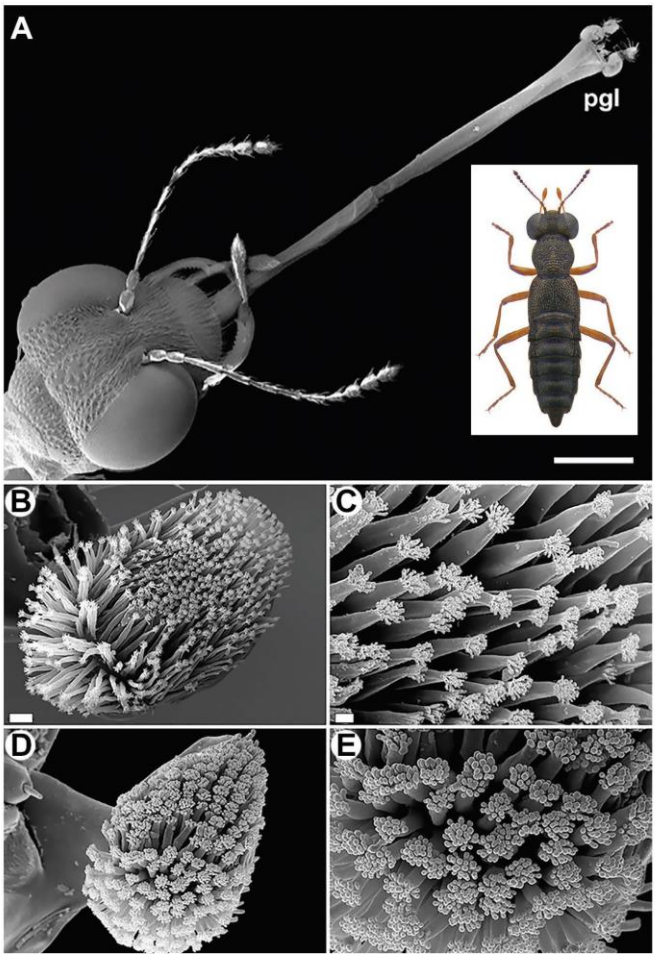
1. The termination of the brochosomal secretion in the leaf hoppers would be supportive in its effective management.
2. It helps in the taxonomic identification of species.

Neck Region of Dragonflies [12]



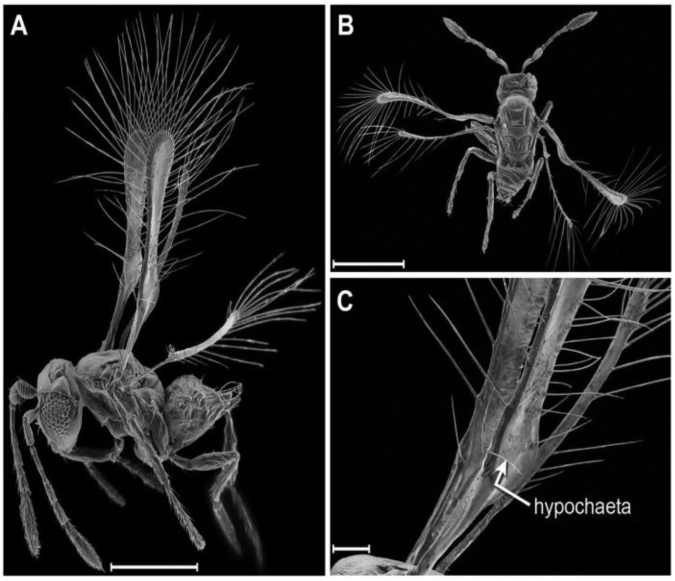
NM – Neck membrane
SPC – Postcervical sclerite
MF – Microtrichia field on back of head
Dragonfly necks are fragile and requires a reversible attachment system to secure them during high-intensity manoeuvers.

Labium of Rove Beetles [12]



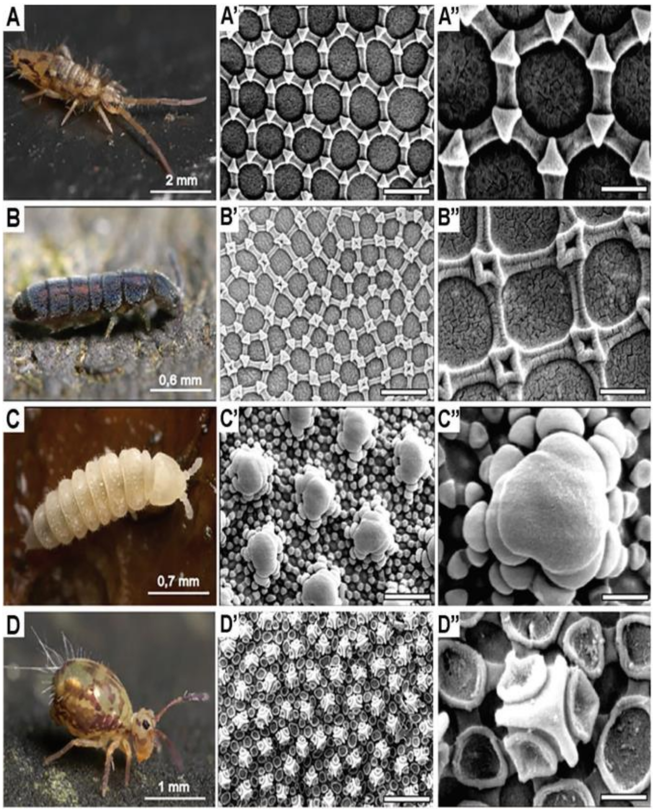
Rove beetles of the genus *Stenus* have extensible labium tipped with adhesive pads, The Paraglossae
B,C – Branching pattern of Paraglossae
(*Stenus clavicornis*)
D,E – Branching pattern of Paraglossae
(*Stenus fossulatus*)

Hypochaeta in Forewings of Fairyflies [12]



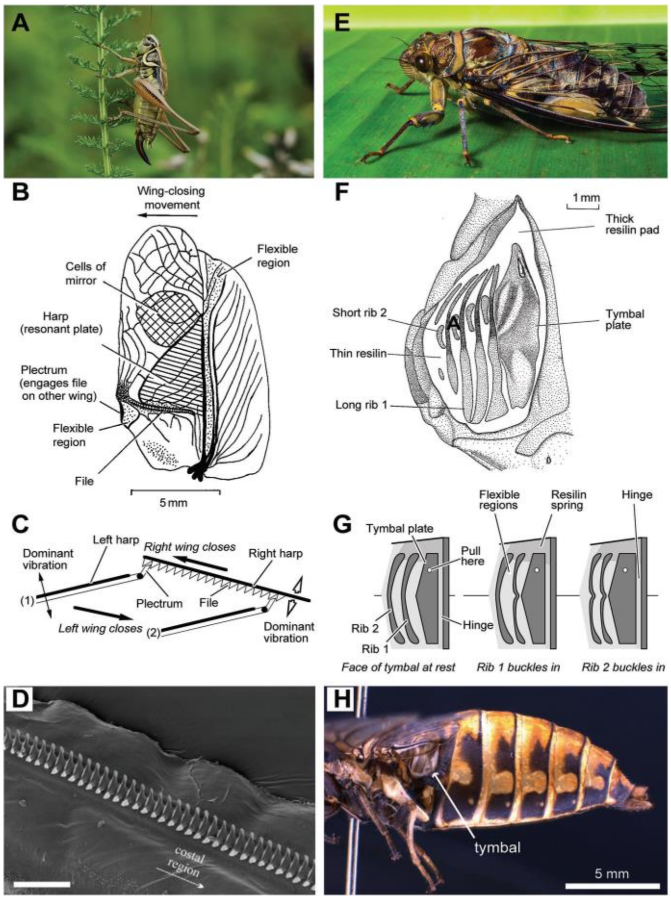
Fairyflies have unique bristle-based wings enabling efficient flight.
The forewings usually have hypochaeta.
These are small bristles (setae) which point distinctly backwards on the ventral surface of the wing membrane.

Hydrophobic Structures in Springtails [12]



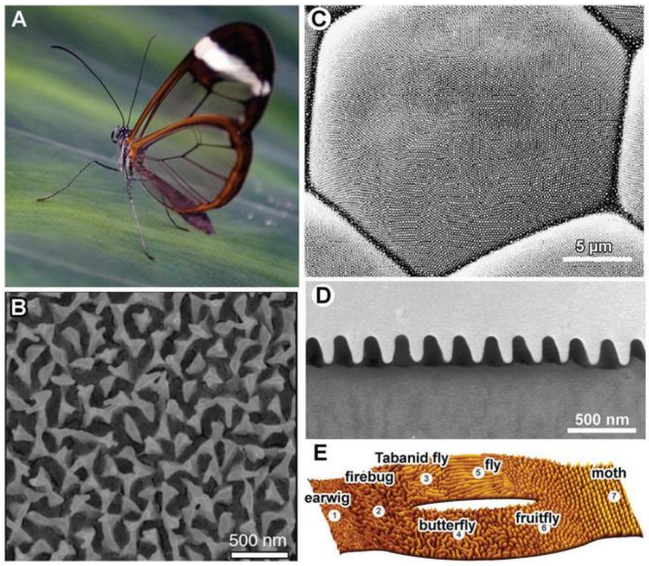
Spring tails are having variety of surface structures with hydrophobic properties
A. *Entomobrya intermedia*
B. *Vertagopus arboreus,*
C. *Kalaphorura burmeisteri*
D. *Dicyrtomina ornate*

Stridulatory and Tymphal Organs in Crickets and Cicadas [12]



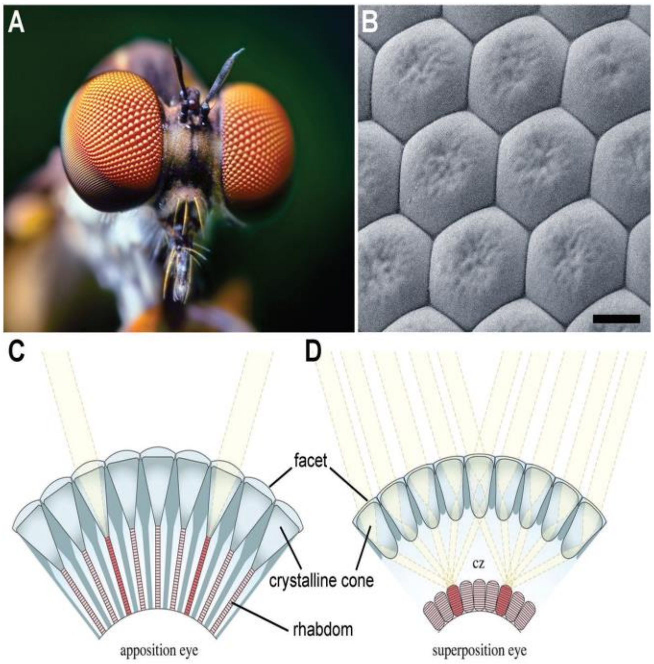
Crickets – Stridulation (Scraping a file along a ridged surface (Plectrum)
Cicadas – Tymbals (As the membrane's ribs buckle inward, clicks are created.

Anti-Reflective Coatings in Glass Wing Butterfly [12]



Insect wings and eyes carry nanostructured anti-reflection coatings to achieve near-perfect transparency.
E.g., Glass wing butterfly, *Greta morgana*
C. SEM image of cornea
D. TEM cross section of cornea

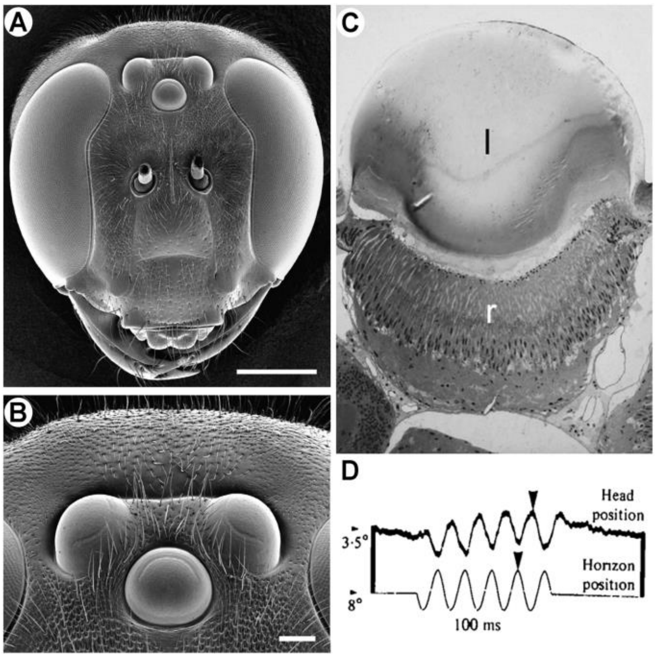
Facet Lenses in Robber Fly [12]



Insect eyes contains thousands of lenses that direct light towards a photoreceptive rhabdom

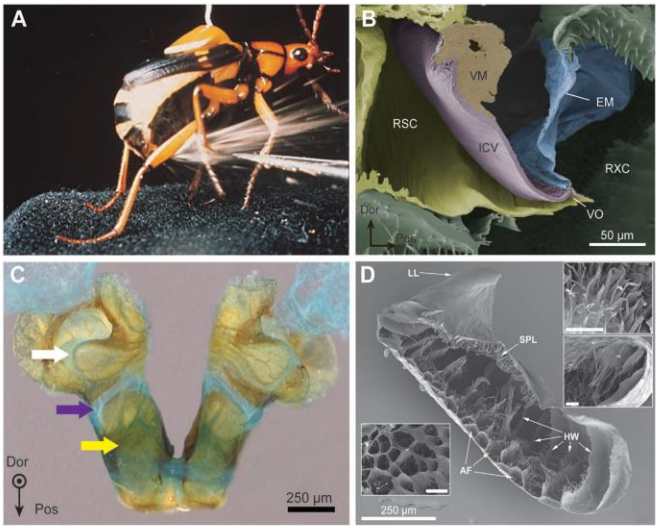
A. Robberfly showing regular arrangement of facet lenses.
B. Close up of facet lenses highlighting hexagonal shape.

Ocelli in Halicid Bees [12]



Ocelli are simple light detectors.
A,B – Head of nocturnal halictid bee
C – Median ocelli
l – lens
r – retinal layer

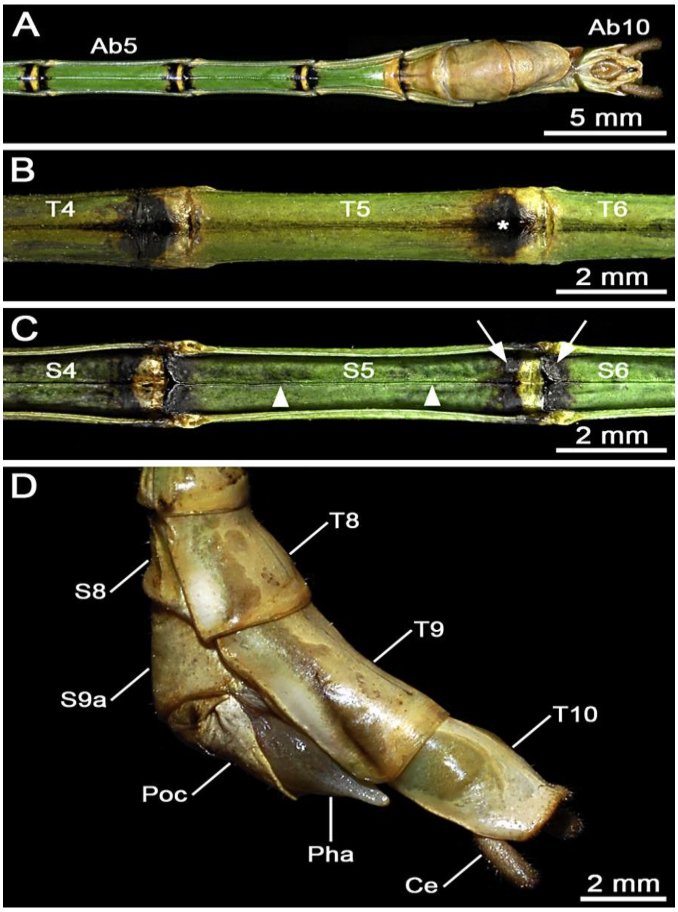
Reaction Chamber in Bombardier Beetles [12]



Specialized reaction chamber to store explosions

A. African bombardier beetle aiming its spray forward.
B. Inter-chamber valve leading from the reservoir chamber to the reaction chamber.
   Based on the contraction of valve muscles (VM), the valve opening (VO) opens and shuts passively by the elastic action of the resilin-containing expansion membrane (EM)
C. White arrow – reaction chamber Purple arrow – junction between reaction chamber and exit channel Yellow arrow – Exit channel

Keel in Stick Insects [3]



Male abdomen of *Hermarchus leytensis* (Phasmatodea)

A. Ventral view from segments 5 to 10
B. Detailed view of terga (T4–T6)
The characteristic black spot on the posterior end of the tergum
C. Detailed view of sterna (S4–S6) Longitudinal median keel (arrowheads) & the colouration of the boundary area b/n adjacent sterna with a yellowish brown stripe bordered in black (arrows)
D. Abdominal terga T8–T10, the 8^{th} abdominal sternum (S8), the 9^{th} abdominal sternum divided into an anterior (S9a) and a posterior (Poc) sternite, the prominent phallus (Pha), and the cylindrical cercus (Ce)

Conclusion

The photonic crystals in butterflies and weevils are responsible for the colour generation which also has its application in physical science. The future studies might be concentrated on the destruction of brochosomes in leafhoppers which inturn parasitization and insecticidal efficiency can be improved. The similar other micro-structures would also be helpful in species identification.

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