

# External morphology and microstructure of the compound eye of fire ants, *Solenopsis invicta* Buren

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**Abstract** The external morphology of the compound eye of the winged female and male *Solenopsis invicta* Buren and its microstructure in light and dark adaptations were observed using scanning electron microscopy and optical microscopy. The results indicated that the compound eye located on the lateral side of its head, is the a shape of a half ellipsoid and composed of approximately 510 ommatidia in the female, and a near hemisphere with about 805 ommatidia in the male. The ommatidium was made of a corneal lens, crystalline cone, 8 to 9 retinula cells and basement membrane. The cornea was a colorless, transparent and double convex lens. The crystalline cone, with an inverted cone shape, was approximately 14.50 μm long, formed by four equal parts, and surrounded by many pigment granules. The rhabdom beneath the crystalline cone, was about 75.00 μm long, with a thicker middle part and thinner ends. More pigment granules were scattered in the distal and proximal ends and less in the middle, and the basement membrane was on the most bottom area of the ommatidium. The primary pigment cells moved horizontally along the crystalline cone from its distal to proximal end during dark adaptation or moved reversely during light adaptation. There was no significant difference between the pigment granule distribution and the structure of the crystalline cone between female and male ommatidium under the same light or dark adaptation. It is concluded that the fire ant compound eye is an apposition eye, whose light-tuning mechanism is accomplished by the change of crystalline cone and the movement of the pigment cells.

**Keywords** *Solenopsis invicta* Buren, compound eye, ommatidium, external morphology, microstructure

## Introduction

The red imported fire ant (RIFA), *Solenopsis invicta* (Buren) (Hymenoptera:Formicidae), is one of the worst invasive alien pest in the world because of its potential impact on public health, agriculture and wildlife. It first arose in the Parana River area, after which it has been found in many countries since 1930 and invaded China in 2003. Therefore, it is an extremely urgent matter to explore environmentally friendly and effective monitoring and control measures (Trager 1991; Allen et al., 1995; Vinson, 1997; Wojcik et al., 2001; Callcott,

2002; Xue et al., 2005).

The compound eye plays a very important role in an ant's life and behavior (Xu and Shen, 2000; Banks and Srygley, 2003; Harris et al., 2005). So far, much work has been done on its morphology, electrophysiology, behavior, etc. to elucidate its visual role in a variety of ants, but they only focused on its biology, regional forecast and the structure of the olfactory receptor in the RIFA because there have been only a few reports on them (Shoemaker and Ross, 1996; Gao and Luo, 2005; Xue et al., 2005; Fan et al., 2008; Chen et al., 2010). To clarify the fine structure similarities and differences between genders and their biological significance, especially in the female adult ants, and how to find a suitable new nest location, this paper observed the compound eye morphology and microstructure of the winged female and male adult ants (hereinafter referred to as female ants and male ants) and the impact of light and dark adaptations on them by using

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scanning electron microscopy and histological measures to provide a basis for the effective monitoring and control of RIFA, in particular, the new queen or reproductive ant.

## Materials and methods

### Materials

Fire ants were provided by the Guangdong Academy of Agricultural Plant Protection in China. In early July 2006, *Solenopsis invicta* Buren were collected from the Nansha District, Guangzhou City, China (N 22°48.096', E 114° 32.156').

### Methods

#### *SEM morphology of compound eye*

After being fixed and dehydrated, the samples were glued on the SEM sample stage by a double-sided tape with a conductive adhesive, according to different observation surfaces, and placed into the KYKY + SBC-12 ion sputtering coating machine to be sprayed with gold. Then, with a KYKY-2800 scanning electron microscope at an accelerated voltage of 30 kV, they were observed and photographed.

#### *Microstructure observation of compound eye*

The study set two treatments for the light and dark adaptations. In the light adaptation, the samples were placed in light for 3 h (light intensity about 1200 lx–1300 lx), and the head was cut and fixed in Bouin's fixative. In the dark adaptation test, insects were placed in the dark for about 3 h, while others were the same as stated above. By means of the paraffin embedding serial section technique, sections were sliced at a thickness of 4 µm and used for HE staining. Sections were observed, measured and photographs taken under Leica microscope.

## Results

### Compound eye morphology

#### *Compound eye of the female ants with the morphological characteristics*

The compound eye in the middle side of the head was a half ellipsoid and composed of about 510 ommatidia. The long axis radius was 640.1 µm, short axis radius was 350.1 µm and the protruding outside arc height was about 203.9 µm (Plate I: 1). The surface of the ommatidium was convex in different parts with different designs but the size was similar. The central region of tightly packed ommatidia, was a regular five-sided or hexagonal and the edge regions, where ommatidia were not closely arranged, were irregular five-sided or hexagonal (Plate I: 5). There were 2 to 6 hairs

sensory hairs in the compound eye of the central region near the dorsal area, which was perpendicular to the surface but a few micrometers/degrees from the base of the bend, close to the surface. The length and the diameter of the sensory hair were 17.5–90.2 µm and 2.2–4.3 µm, respectively (Plate I: 3).

#### *The male ants compound eye with the morphological characteristics*

The compound eye of the male contained about 805 ommatidia, more than that of the female. It occupied nearly half of the sphere, on both sides of the head, and the compound eyes were close to each other on the head. In addition, it was convex and the arc height was about 195.2 µm. Its long axis radius was 599.7 µm and short axis radius was 438.1 µm. The design and the order distribution of the ommatidia were similar to the female (Plate I: 2). The compound eye had more than two sensory hairs, perpendicular to the compound eye, with a small number of bent hooks at the end. The design of the sensory hair and the position was the same as that in the female ants. The sensory hair length was 17.5–27.9 µm, and the diameter was 1.4–2.5 µm. Compared with the female ants, they were significantly shorter (Plate I: 4).

### The internal microstructure of compound eye

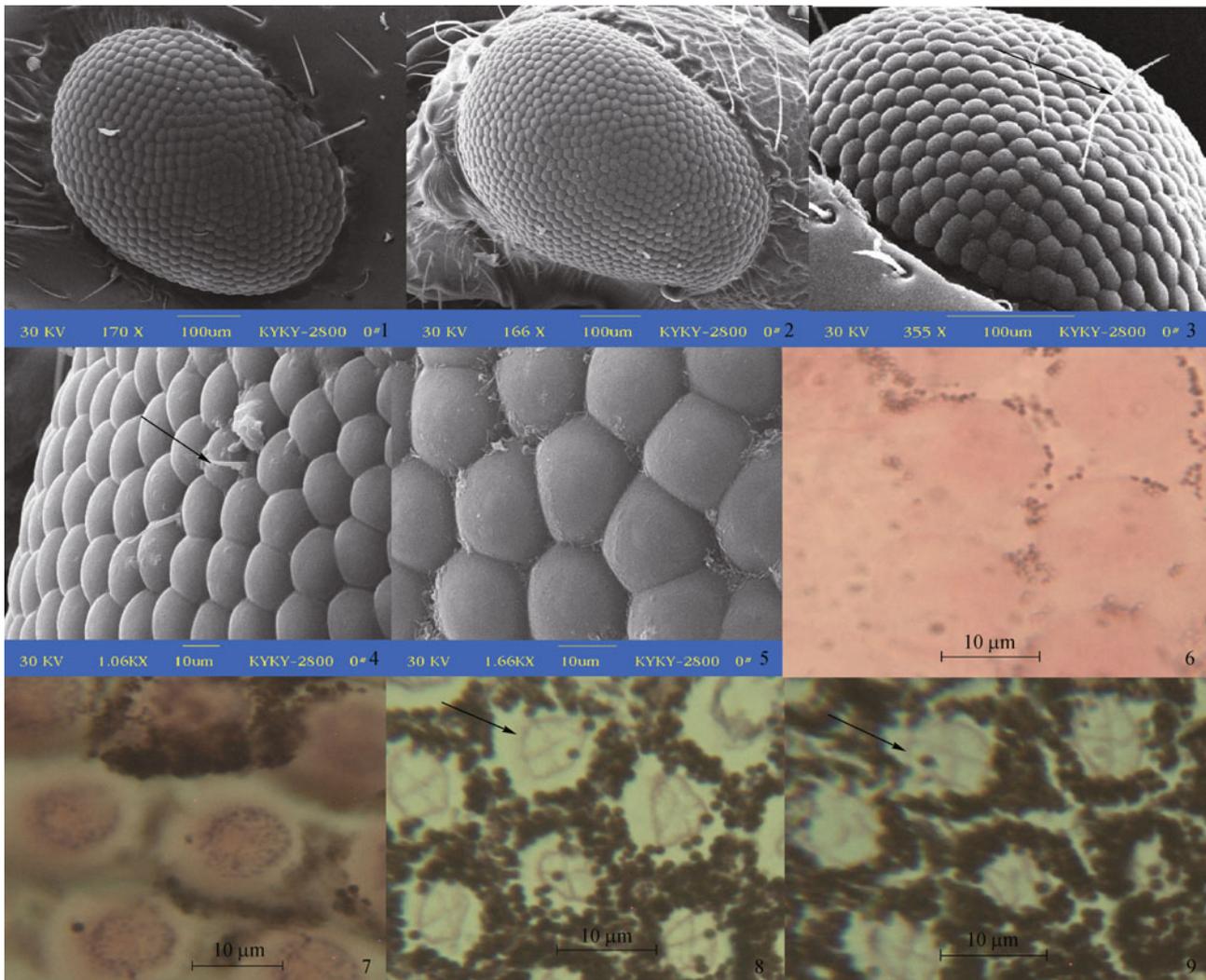
#### *The structure of ommatidium*

According to the depth of the transect and the longitudinal layer characteristics, it can be seen that the compound eye of fire ants is an apposition eye and the general structure of the ommatidium from the outside to the inside is comprised of the structure from the outside made of the following components (Fig. 1).

#### (1) Dioptic apparatus

It mainly consists of a cornea and a crystalline cone. The cornea is in the outermost layer of the ommatidium, with the inner edge being less prominent and the outer edge having the more prominent features of the larger double-convex lens with a multilayer sheet fibrous structure about 15.0 µm at the thickest place, which, in design, is different according to the different positions between the ommatidia and compound eyes, with an irregular cross-section. The cornea is divided into two obvious parts, the outer, which is lightly stained, and the inner layer adjacent to the part of crystalline cone, which looks dark stained on a light microscope. This finding indicates that the texture of the inner and outer cornea is heterogeneous (Plate I: 6).

The crystalline cone is a cone-shaped crystal, with a length of about 14.5 µm below the cornea and a distal end diameter of about 10.0 µm. It can be seen that the crystalline cone is divided into four equal parts from the four crystalline cone cells, surrounded by the primary pigment cells and a large number of pigment granules from the cross-section view (Plate I: 7, 8, 9).



**Plate I** External morphology, corneal lens and crystalline cone of the compound eye of fire ants, *Solenopsis invicta* Buren. 1: Compound eye of female ant; 2: Compound eye of male ant; 3: Sensory hair on female's eye; 4: Sensory hair on male's eye; 5: The rim area of compound eye; 6: Cross-section of the cornea; 7: Cross-section between the cornea and crystalline cone; 8: Cross-section of the crystalline cone in light adapted condition; 9: Cross-section of the crystalline cone in dark adapted condition.

## (2) Rhabdom

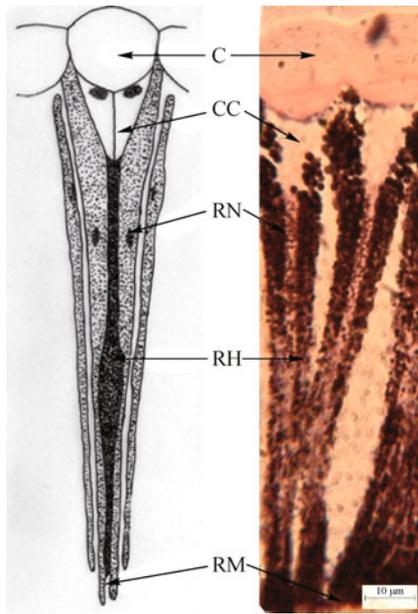
Rhabdom, as part of the photoreceptor, closely converges under the crystalline cone, with the small retinal cells from the center into the proximal part of the special combination of microvilli fusion and a large number of pigment granules around. The length is about 75.0 µm, and it is pink, as demonstrated by H E staining. From the longitudinal section, the middle rhabdom is deeper-dyed and significantly thicker than the other parts. In the same ommatidium, the rhabdom cross-section is different in design and sizes in different levels. The summaries from the outside of the fine structure of different levels are as follows.

The upper rhabdom segment was a circle at the cross-section. There were 8 or 9 larger reticular cells in each ommatidium, without trachea around the micro-segment and the microvilli was only a fractional part of it, therefore the

cross-sectional size of the rhabdom was small. In addition, it could be seen from the cross section that the deeper-colored part was the center rhabdomere (Plate II: 1).

Because the compound eye of the red imported fire ant was small, it was hard to clearly distinguish between the center rhabdomere and the edge rhabdomere through the cross section of the middle medial rhabdom segment. With only about 8 to 9 small retinal cells around it, the larger level of the cross-sectional area of the rhabdomere and microvilli were filled with most of the small retinal cells (Plate II: 2).

In the lower rhabdom segment, most of the area of each retinula cyton was occupied by the microvilli and a large number of pigment granules were distributed around the rhabdom. The retinula cyton was smaller than that in the middle or upside segment, and hence, this rhabdom cross-section was smaller (Plate II: 3).



**Figure 1** Longitudinal view of ommatidium of *Solenopsis invicta* Buren. C: cornea; CC: crystalline cone; RN: retinula cell nuclear; RH: rhabdom; BM: basement membrane.

### (3) Basement membrane

The basement membrane at the bottom of the ommatidium was similar to most insects with a fence-like structure. The more dense and dark pigment particles were distributed on both sides of it and diffused between the section of the lower rhabdom and the basement membrane by the thick trachea through the basement membrane into the retinal area. Based on previous studies, the basement membrane cannot only separate the ommatidia within the interior optic lobes but also probably have some functions of mechanical support and repair of the ommatidia. After the axon bundles of rhabdomeres go through the basement membrane, the number of axon bundles can converge to a larger bundle of axons into the optic nerve section (Plate II: 4).

### *Light and dark adaptation to the microstructure of compound eye*

Under different conditions of light and dark, there are some differences in the microstructure of the compound eye of red fire ants.

#### (1) Changes in the crystalline cone

From the crystalline cone cross-section, it could be seen that its circular cross-section was divided into four equal parts (Plate I: 8, 9). The longitudinal section showed that the crystalline cone was an inverted cone shape. Under dark adaptation, the crystalline cone length was about  $15.1\mu\text{m}$ , the width of the upper and bottom was about  $9.7\mu\text{m}$ , whereas in the light adaptation it was  $2.7\mu\text{m}$ . The crystalline cone length was about  $13.2\mu\text{m}$ , and the upper width and the bottom width were about  $9.5\mu\text{m}$  and  $2.2\mu\text{m}$ , respectively.

#### (2) Pigment granules' move

Under dark adaptation conditions, primary pigment cells moved along the horizontal and vertical lines to proliferate the color of the crystalline cone. There was lateral movement of primary pigment granules and they closed around the crystalline cone at the same time. Besides that, the secondary pigment particles showed horizontal proliferation, and was closed to the rhabdom to reduce the light absorption of the pigment granules. However, the primary pigment particles had horizontal and vertical expansions with the crystalline cone contraction, while the horizontal expansion of the secondary pigment cells was far away from the rhabdom in the light adaptation.

### *Microstructure of gender on the impact of the compound eye*

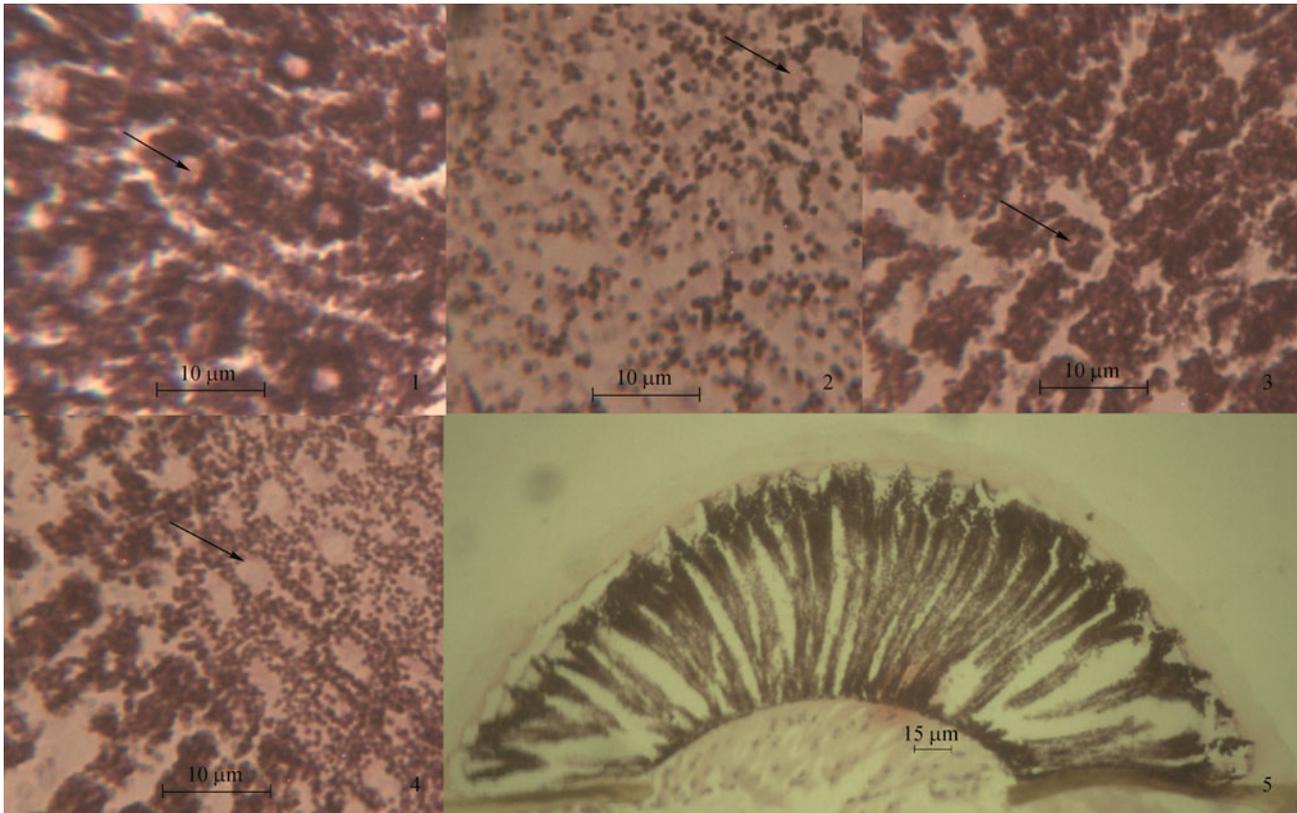
In the same adaptations to light and dark, there were no significant differences in the change in the microstructure of the compound eyes of the male and female ants.

## Discussion

We found that there were some obvious differences between female and male ants in the compound eye design and the number of ommatidium, demonstrating that species, living habitats and their behaviors, and sex were all responsible for it (Bernstein and Finn, 1970; Moser et al., 1999; Labhart, 2000). The underlying reason might be that the vision of the compound eye played different roles in the life cycle of both male and female ants. For instance, finding spouses is a common task during their nuptial flight. Females need to orient, locate and build a new nest against or near the bright reflected light of pools (Klotz et al., 2003). Moreover, like the desert ant *Cataglyphis bicolor*, the compound eye of RIFA also exhibit regional differentiation, hinting that the difference in the number and morphological characteristics of the ommatidium from different areas might be one reason for the changes in their internal functions (Meyer and Nassel, 1986). However, their light adaptation is worth investigating by monitoring and controlling RIFA by means of the difference and subsequent behaviors between males and females.

We could observe that there was a small amount of sensory hairs on the surface of the RIFA compound eye, and the number and size of the hairs on female ants were bigger and larger than those on male ants. The sensory hairs on or between ommatidia of the butterfly might be a pneumatic sensor to detect wind speed or to modulate the flight situation for bees (Neese, 1966; Feng et al., 1992). As the differences in the number and design of sensory hairs in connection with RIFA sex and their biological roles are unknown so far, they were likely to be employed to sense some environmental factors such as the direction and speed of wind, temperature, humidity, etc.

Additionally, the compound eyes of both female and male RIFA were a typically apposition, with no clear and transparent zone between the dioptric apparatus and retinal



**Plate II** Microstructure of the compound eye of fire ants, *Solenopsis invicta* Buren. 1: Cross-section of the distal end of rhabdom; 2: Cross-section of the middle of rhabdom; 3: Cross-section of the proximal end of rhabdom; 4: Cross-section of the basement membrane; 5: Longitudinal section of the whole compound eye.

cells. At the topmost layer, cornea in different depths, displayed different stained colors. The more outermost the layer was, the lighter the color would be, showing that the cornea, with a heterogeneous character of light conduction, played a stronger role in collecting incident rays. In the middle part, the pigment granules around the crystalline cone and the retinal pigment cells moved longitudinally from the outer to the inner or reversely with the changes in light and dark conditions. In the dark adaptation, the length and the diameter of the distal and proximal ends of the crystalline cone were both enlarged, conversely, they got shorter and thinner, and the distance of the proximal end of the crystalline cone and rhabdom was reduced by nearly one-fifth, showing a phenomenon similar to the *Camponotus* ant, which should be one of the mechanisms for the photoreceptor to tune the amount of light entering the rhabdom based on the changes in light and dark situations (Menzi, 1987).

In short, the regular change in the fine structure of the RIFA compound eye had a basis for some important vision-oriented behaviors, especially for female adults to find suitable locations in order to build a nest during their nuptial flight, and adaption to a strong light environment in the daytime or low light in the evening, which will probably help to find new, effective and environmentally friendly measures for monitoring and controlling the adults.

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