

## Biomechanism of adhesion in gecko setae

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The study of the adhesion of millions of setae on the toes of geckos has been advanced in recent years with the emergence of new technology and measurement methods. The theory of the mechanism of adhesion by van der Waals forces is now accepted and broadly understood. However, this paper presents limitations of this theory and gives a new hypothesis of the biomechanism of gecko adhesion. The findings are obtained through measurements of the magnitude of the adhesion of setae under three different conditions, to show the close relationship between adhesion and status of the setae. They are reinforced by demonstrating two setal structures, follicle cells and hair, the former making the setae capable of producing bioelectrical charges, which play an important role in attachment and detachment processes. It is shown that the abundant muscular tissues at the base of the setae cells, which are controlled by peripheral nerves, are instrumental in producing the foot movement involved in attachment and detachment. Our study will further uncover the adhesion mechanism of geckos, and provide new ideas for designing and fabricating synthetic setae.

**gecko setae, adhesion, biomechanism, microstructure, bionics**

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The billions of years of evolution and competition for survival have produced a great diversity of animals. Their various attributes inspire the imagination and creativity of scientists wishing to understand and exploit natural phenomena. For example, insects, beetles, skinks, frogs and geckos can stick to, and climb on, smooth and rough surfaces very easily. Geckos are the most interesting because of their size. The largest geckos can weigh up to 300 grams and reach lengths of 350 mm, yet they are still able to run inverted and cling to smooth and rough walls. Understanding how to walk or climb like these animals has long been a human pursuit. Recently, scientists have discovered that the gecko's special ability lies in its feet, especially the very

fine hairs on its toes. There are billions of these tiny fibers that make contact with the surface, creating a significant adhesive force for firm attachment. The study of what underlies gecko adhesion is important in designing bionic setae, and tackling the attachment difficulties of wall-climbing robots.

There are many hypotheses about the adhesion mechanism of gecko setae, such as suction [1,2], microinterlocking [3], friction [3,4] and electrostatic attraction [5]. In 2000, Autumn *et al.* measured the force production by a single, isolated seta during attachment using a micro-machined, dual-axis, piezo-resistive cantilever [6,7]. They found that weak intermolecular forces were generated when the curled tips of setae were in close contact with the surface, suggesting that adhesion in geckos is the result of intermolecular

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forces. It was concluded that the attachment mechanism of gecko setae is a result of van der Waals forces, an explanation currently favored by many researchers [8–15]. However, in 2005, Huber *et al.* [16], on the basis of measurements of the adhesion force exerted by a single gecko spatula under various atmospheric conditions and surface chemistries, reported that humidity contributes significantly to gecko adhesion at a nanoscopic level. Autumn *et al.* also observed in their experiments that the adhesive force of gecko setae could only be detected when there was a sliding movement between the setae and the contacting surface [6], which is not perfectly consistent with the production of van der Waals forces. This sliding likely contributes to full contact between the setae and the surface. On the other hand, according to our early study on the gecko's foot movement, we hypothesize that the gecko's peripheral nerves and the muscular tissues on the base of the gecko's setae can modulate and control the setae when they attach or detach. Very likely, there are contractile proteins in setal cells, which can increase or decrease the van der Waals force of setae by changing the space between setae, thus realizing the attachment or detachment of setae. This raises the questions "Do forces other than van der Waals forces underlie the gecko's adhesive ability? Are there other factors involved in adhesion?" The answers to these questions will further uncover the adhesion mechanism of gecko setae.

Guo *et al.* [17] previously compared the setae of *Gekko gecko* (*G. gecko*) and *Gekko swinhonis* (*G. swinhonis*) employing scanning electron microscopy (SEM) and histological techniques. Here, under different conditions, tests were performed to measure the adhesive forces associated with a single toe of a gecko. These were performed for a non-anesthetized animal, an anesthetized animal, and a single dissected toe. The results do not fully support the conclusion of Autumn *et al.* as it is clear from the results of this work that biological factors play a part in the adhesion mechanism exploited by geckos. Our work provides a new hypothesis on the adhesion mechanisms of geckos, and offers new insights for designing synthetic gecko foot hairs.

## 1 Material and methods

### 1.1 Adhesion force measurements of a gecko's toe under three different conditions

Experiments were performed on 12 *G. gecko* obtained from Guangxi Zhuang Autonomous Region. Non-anesthetized and anesthetized geckos were used in equal number, and their weights were ( $79.6 \pm 6.3$ ) g and their full lengths were ( $159.5 \pm 1.3$ ) mm. The parallel and perpendicular hair forces of the 12 specimens (with the third toe of the left hind limb with nails removed prior to the tests) during attachment were measured with a self-made two-dimensional force sensor (with sensitivity of 5 mN and range of 0–4 N). The results were compared with the adhesion of corresponding

dissected toes (dissected and kept in a refrigerator for a week,  $n=6$ ). Real-time force data were recorded using a data acquisition system (AcqKnowledge V3.5.3, Biopac Systems Inc., USA) on a computer.

When measuring the toe adhesion of the non-anesthetized animal, only the third toe on its left hind limb was placed against a horizontal oriented glass slice that was fixed to the force sensor. The gecko was pulled backwards until the toe slipped off, the angle between the toe and contacting surface was kept within  $30^\circ$  [18]. To measure a single toe, the gecko was restrained by hand with the other toes held in a fixed position. Any trial in which the gecko struggled or moved its toe was excluded from the data set. The adhesion of the anesthetized animals and the dissected dry gecko toes was measured using the same equipment and technique.

### 1.2 SEM and histological observations

Three fresh gecko third toes and three dissected third toes were observed. Sample preparation and handling followed the procedures detailed in [19]. Toes were fixed with 10% formaldehyde solution, then dehydrated step by step with ethanol (70%, 80%, 90%, 95% and 100%), each step lasting about 24 h. After critical-point drying for 3 h (CPD030 Critical Point Dryer), the samples were gold-coated with a SCD005 Sputter Coater, then observed with a scanning electron microscope (FEI Quanta 200, Nihon Kohden, Japan).

Both longitudinal and transverse sections of gecko setae were prepared in this study using a microtome (Leica CM1900, Germany). Histological preparations were conducted following the methods in [20]. Toes were processed in the order of fixation, neutralization, water washing, dehydration, embedding, sectioning (10  $\mu\text{m}$  in thickness) and staining. The samples were observed and photographed with a Motic B series bio-microscope (Motic China Group Co., Ltd., China).

### 1.3 Modulation and control of the gecko's foot movement

Under the stereo microscope (Leica MZ95, Germany), three main peripheral nerves on the gecko's limb were exposed ( $n=4$ ). To observe the nerve innervations of the foot motion of *G. gecko*, we used a stimulator (YC-2-S, Chengdu Instrument Factory, China) separately on each of the three nerves with stimulation parameters of trains of unipolar rectangular pulse, amplitude of 0.1–0.3 V depending on a threshold value, duration of 1 ms, pulse interval of 20 ms, 20 pulses in a train and train interval of 60 s.

### 1.4 Testing of actins in gecko setal cells

There are extensive actins in animal's cells as a kind of

contractile protein that can make cells move. As a basic functional protein, its structure and function are highly conservative. We hypothesize that there are also actins in gecko setal cells and that they play a role in the attachment and detachment of setae. To verify this, firstly the actin gene sequence of the frog, bird and rat were compared, the polypeptide segment was worked out, and then an actin polyclonal antibody that had cross-reaction with the frog, bird and rat was made in the laboratory. With this antibody, western blotting was employed to test actins in the gecko setae. Furthermore, transverse and lateral resin sections of the gecko setae were made (2  $\mu\text{m}$  in thickness), and actins in the setae were fluorescent labeled with phalloidin.

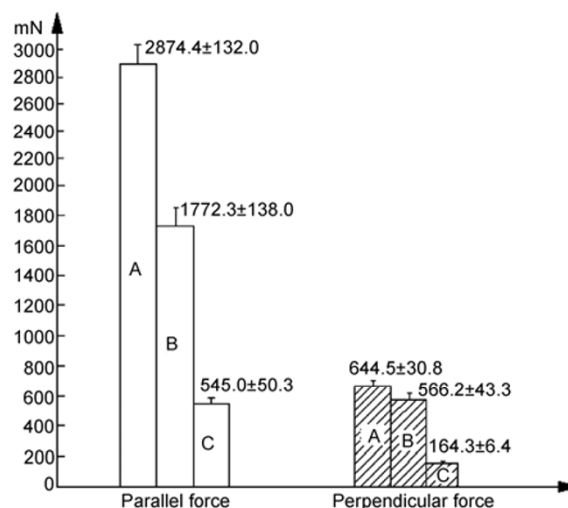
## 2 Results

### 2.1 Adhesion forces in two directions

The results demonstrated that adhesion forces of the dissected toes were reduced significantly in two directions compared with the forces measured in live animals, and the force was weaker in test 2 than in test 1 (Figure 1). According to the theory of Autumn *et al.*, van der Waals forces will be produced when the gap between the setae and the surface is as small as the atomic distance. The adhesive quality has a strong relationship with the fine structure of the setae tips, no matter if the toe is dead or alive. However, SEM observation of the fresh setae and the dissected dry setae (with dissected toes being kept in the refrigerator for a week) showed little morphological difference (Figure 2A and B). We therefore ask why the adhesive ability among setae in tests 1, 2 and 3 is so different. It seems that van der Waals forces are not the only element underlying the adhesion of the gecko's setae, and there must be other factors involved.

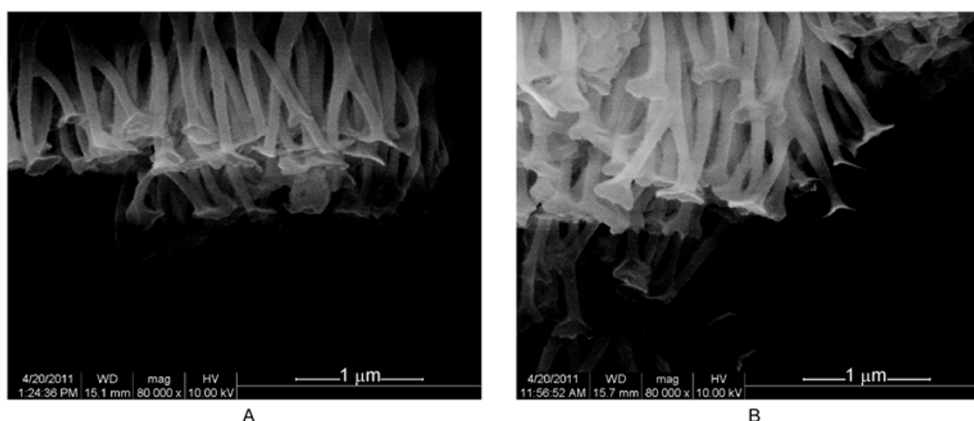
### 2.2 Two structures in setal development

Histological studies of *G. gecko* setae demonstrated that there are two structures during setal development. One is a

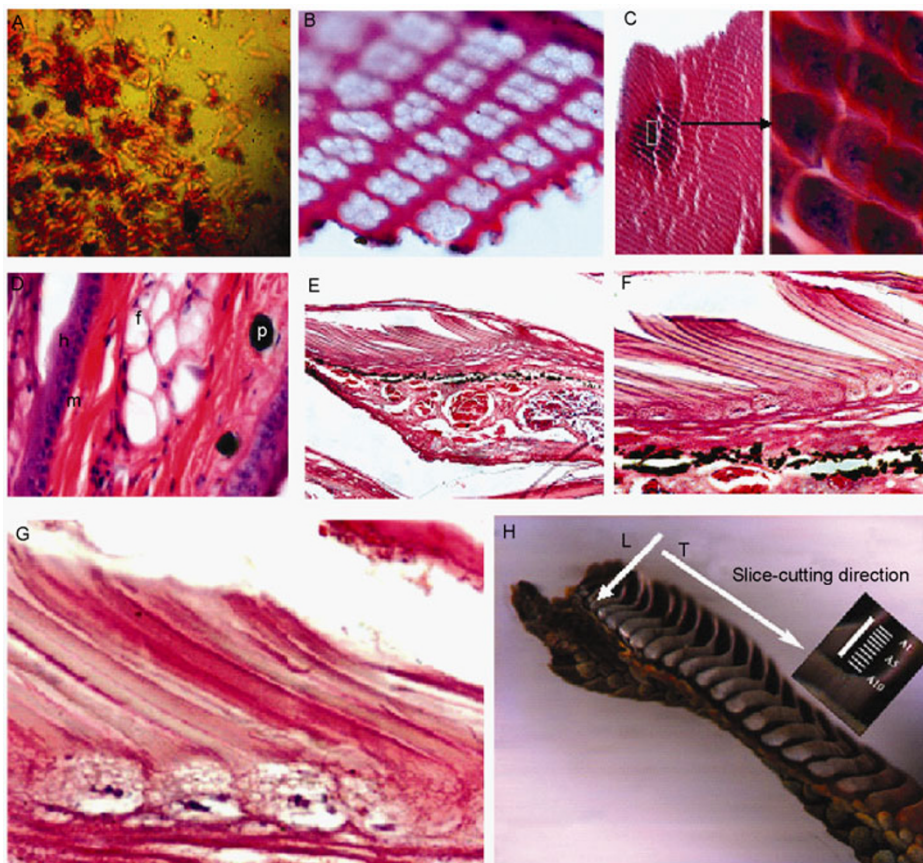


**Figure 1** Gecko's adhesion under three different conditions. A, Non-anesthetized animals. B, Anesthetized animals. C, Dissected dry toes.

follicle cell, with a nucleus located at the base of the hair. Figure 3A–D shows a series of transverse sections from the tip to the base of the setae. The sections through the tip of the setae have a large number of broken bent parts (Figure 3A); the sections through the middle of the setae showed cluster-like structures with four-six setae in each cluster, inside which were plentiful but not even contents (Figure 3B); the sections through the base showed the cell nucleus (Figure 3C), pigment cells, fat cells and muscular tissues at the base of the setae cells, but no gland cells (Figure 3D). The longitudinal series of sections from the tip to the base of the setae showed that the setae protrude from the cells on the surface of the dermal valvulae. These setae cells are arranged in order with the nucleus located at the base of the setae (Figure 3E–G). The other is a hair structure. As shown in Figure 4, setae are distributed on the dermal surface of the gecko's digital pad, just like "a branching out of the skin". Whatever the structure of gecko setae is, Alibardi [21,22] demonstrated that beta keratin is the main constituent of setae, other constituents need to be further studied. It



**Figure 2** Morphology of the fresh setae and the dissected setae. A, Fresh setae (80000 $\times$ ). B, Dissected setae (80000 $\times$ ).



**Figure 3** Transverse and longitudinal sections of the setae on the gecko's toe. A, Transverse section through the tip of the setae, 1000×. B, Transverse section through the middle parts, 1000×. C, Transverse section through the base of the setae (left, 100×; right, 1000×). D, Transverse section through the bottom of the setae, 1000× (f, fat cell; h, hair cell; m, muscle tissue; p, pigment cells). E–G, Longitudinal sections of the setae on the gecko's toe (200× (E), 400× (F), 1000× (G)). Below right, the cutting direction of the sections.



**Figure 4** Gecko's setae in the second development period.

was found in this study that the two hair structures existed in different periods of setal outgrowth, and that the follicle

cell is the origin of gecko setae but gradually degenerates during setal development. Figure 3C shows that both struc-

tures mainly exist in clusters and the setae with the cell nucleus at their base constitute about 10% of all setae observed.

### 2.3 Modulation and control of the gecko's foot movement

Our histological observations showed that the base of the setae cells have a plentiful supply of muscular tissues (Figure 3D) that can be modulated and controlled by the gecko's peripheral nerves to produce foot movements involved in attachment and detachment, such as adduction, abduction and rotation (Figure 5) [23].

### 2.4 Actins in the gecko setae

#### 2.4.1 Western blotting

Figure 6 is the Western blotting result. It is seen that there are actins in the gecko setae with molecular weight similar to that of actins in the frog and rat (42 kD).

#### 2.4.2 Fluorescent labeling

Figure 7 presents the fluorescent labeling results of actins in the gecko setae with phalloidin. It is seen that the positive staining cells have an annular distribution (Figure 7A) and are only located at the base of setae cells and not along the

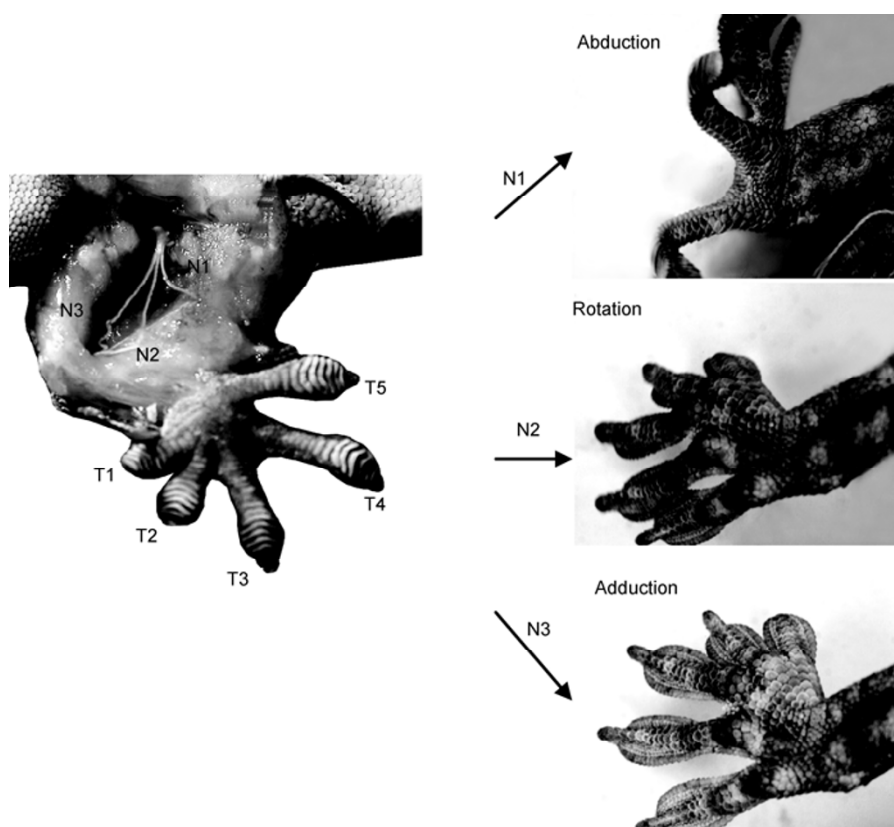
full length of setae (Figure 7A and B).

Figures 6 and 7 show that there are actins in the gecko setal cells and that their contraction and dilatation can change the spacing between setae, thus modulating the magnitude of the van der Waals force and controlling the adhesion force of setae.

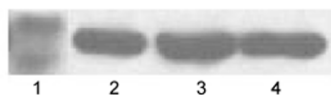
## 3 Discussion

Autumn *et al.* reached their conclusion only by measuring the force of an isolated gecko's mature seta, neither considering the effects of setal structure in different developmental periods on adhesive ability nor testing the adhesion of setae of live animals. Our study demonstrated the limitations of the theory of Autumn *et al.* by measuring setal adhesion under three different conditions. The experimental results showed that setal adhesion for the live animal was far greater than that for the dissected toe, and that there was a significant difference between the setae of anesthetized and non-anesthetized geckos. Therefore, in addition to van der Waals forces, there must be biological factors involved in gecko adhesion.

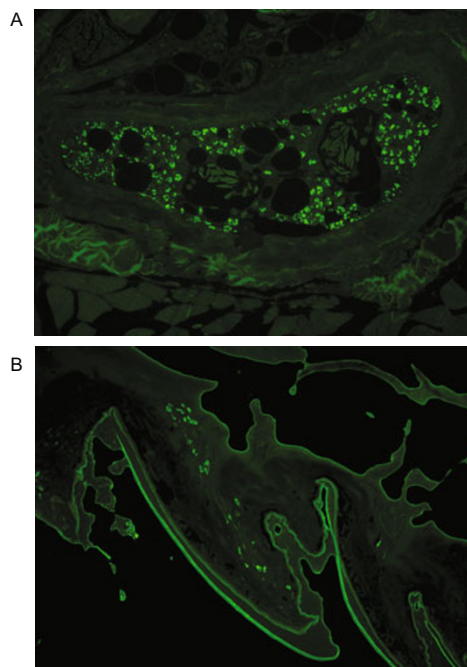
On the other hand, although current synthetic gecko setae possess some adhesive abilities, there are some difficult problems still to be tackled [24–26], such as how to easily



**Figure 5** Gecko's foot movements controlled by peripheral nerves. N1, peroneal nerve; N2, tibial nerve; N3, femoral nerve.



**Figure 6** Western blotting result. Lane 1, marker; lane 2, frog; lane 3, gecko; lane 4, rat.



**Figure 7** Fluorescent labeling of actins in the gecko setae. A, Transverse section of the setae (400 $\times$ ). B, Lateral section of the setae (400 $\times$ ).

switch to detachment from attachment, how to overcome the bending of hairs after they have been in contact with the opposite surface, and how to reduce environmental contamination, which will affect the application of synthetic hairs in an industrial situation. Why can more closely spaced, finer and more complicatedly branched gecko setae produce great adhesion without these deficiencies? How do geckos overcome these problems? If biological factors discussed here can be introduced into the design of synthetic hairs, their adhesive ability will be improved greatly, and their application will be broader and more efficient.

Our study has already demonstrated that there are actins at the base of the gecko setal cells, which encircle the setae. Recently, scientists have discovered that hair cells of some animal's inner ear, especially the stereocilia protruding from the top of the outer hair cells, have dense cores of the filamentous protein actin, which are functionally contractile. When stimulated by sound waves,  $\text{Ca}^{2+}$  enters the hair cells and interacts with the actin to cause contraction, leading to deformation of the hairs. Changes in the length and stiffness of the stereocilia, combined with the changes of beating frequency and strength, produced between the stereocilia and the tectorial membrane can tune the animal's acoustic

sensitivity. This demonstrates that deformations of stereocilia can not only transfer the energy passively but also actively mediate the process of energy transmission, thus enhancing the sensitivity and frequency selectivity of the tectorial membrane in response to the sound wave [27]. What makes gecko setae attach to and detach from the surface freely should be closely related to actins in the setal cells. The contraction and dilatation of actins can change the spacing between setae and the magnitude of van der Waals force, thus actively controlling the setae to attach or detach. At the same time, our work showed that the muscular tissues at the base of the setae can be modulated and controlled by the gecko's peripheral nerves to produce foot movements involved in attachment and detachment. These observations demonstrate that gecko setae can not only be innervated by the peripheral nerves and muscles at the bottom of the gecko's toe but also be actively mediated by actins in the setal cells, both ensuring that a gecko can walk freely and rapidly.

Our work has also shown that gecko setae are the protuberances from cells on the surface of dermal valvulae, and these cells are arrayed in order with the nucleus located at the base of the setae. Normally, there is electrical polarity between the interior (-) and exterior (+) cell membrane, and this polarity can be changed by external stimulation. For example, when a mechanical stimulus deforms the stereocilia located at the top of hair cells in an animal's auditory and vestibular system, the electrical polarity of the hair cell membrane depolarizes. This change is important for an animal to sense the external stimulus and transfer afferent signals. Another possible reason that gecko setae can attach to and detach from the surface easily is that as cell protuberances, the deformations of setae produce bioelectrical changes that play a role in gecko attachment and detachment. Presently, synthetic gecko setae possess some adhesive abilities but the hairs adhere to each other owing to the very thin pillars and very close spacing of pillars, which dramatically reduce adhesion [26]. Additionally, it is difficult for the synthetic setae to detach from the surface freely. All these problems hold back application of synthetic setae; however, it is hoped that they can be tackled if biologically modulating factors are brought into the design and fabrication of synthetic hairs, thus realizing the application of synthetic gecko setae in industry.

Our study has provided a new hypothesis for the mechanisms of gecko adherence, and will be of great help in uncovering the mature development of gecko setae and the adhesion mechanism. This work also provided new ideas for designing and fabricating artificial biomimetic setae as follows. First, the fine microstructure of the tips of gecko setae should be considered; second, the ideal surface material of synthetic setae should be elastic and be able to overcome the inter-hair adhesion; and third, the substrate material of synthetic setae should be electrosensitive with controllable plasticity.

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- 1 Gadow H. The Cambridge Natural History. Vol.8: Amphibia and Reptiles. London: McMillan & Co, 1901. 668
- 2 Gennaro J G J. The gecko grip. Natural History 78. New York, 1969. 36–43
- 3 Mahendra B C. Contributions to the bionomics, anatomy, reproduction and development of the Indian house gecko, *Hemidactylus flaviviridis Ruppell* Part II: The problem of locomotion. Proc Indian Acad Sci, 1941, 3: 288–306
- 4 Hora S L. The adhesive apparatus on the toes of certain geckos and tree frogs. J Proc Asiatic Society of Bengal, 1923, 9: 137–145
- 5 Schmidt H R. Zur Anatomie und Physiologie der Geckopfote Jena Z. Naturw, 1904, 39: 551
- 6 Autumn K, Liang Y A, Hsieh S T, et al. Adhesive force of a single gecko foot-hair. Nature, 2000, 405: 681–685
- 7 Autumn K, Sitti M, Liang Y A, et al. Evidence for van der Waals adhesion in gecko setae. Proc Natl Acad Sci USA, 2002, 99: 12252–12256
- 8 Gee H. Biomechanics: gripping feat. Nature, 2000, 405: 631
- 9 Desiraju G R. Chemistry beyond the molecule. Nature, 2001, 412: 397–400
- 10 Gorb S N, Beutel R G, Gorb E V, et al. Structural design and biomechanics of friction-based releasable attachment devices in insects. Integr Comp Biol, 2002, 42: 1127–1139
- 11 Bundle M W, Dial K P. Mechanics of wing-assisted incline running (WAIR). J Exp Biol, 2003, 206: 4553–4564
- 12 Wu P, Hou L H, Plikus M, et al. Evo-Devo of amniote integuments and appendages. Int J Dev Biol, 2004, 48: 249–270
- 13 Federle W, Endlein T. Locomotion and adhesion: dynamic control of adhesive surface contact in ants. Arthropod Struct Dev, 2004, 33: 67–75
- 14 Northen M T, Turner K L. A batch fabricated biomimetic dry adhesive. Nanotechnology, 2005, 16: 1159–1166
- 15 Yurdumakan B, Raravikar N R, Ajayan P M, et al. Synthetic gecko foot-hairs from multiwalled carbon nanotubes. Chem Commun, 2005, 30: 3799–3801
- 16 Huber G, Mantz H, Spolenak R, et al. Evidence for capillarity contributions to gecko adhesion from single spatula nanomechanical measurements. Proc Natl Acad Sci USA, 2005, 102: 16293–16296
- 17 Guo C, Wang W B, Yu M, et al. Comparative studies on the structure and adhesion of setae in *G. gecko* and *G. swinhonis*. Sci China Ser C-Life Sci, 2007, 50: 831–838
- 18 Autumn K, Dittmore A, Santos D, et al. Frictional adhesion: a new angle on gecko attachment. J Exp Biol, 2006, 209: 3569–3579
- 19 Cheng H, Sun J R, Li J Q, et al. Structure of the integumentary surface of the dung beetle *Copris ochus* Motschulsky and its relation to non-adherence of substrate particles (in Chinese). Acta Entomol Sin, 2002, 45: 175–181
- 20 Cheng H, Chen M S, Sun J R. Histological structure of the dung beetle *Copris ochus* Motschulsky integument (in Chinese). Acta Entomol Sin, 2003, 46: 429–435
- 21 Alibardi L. Ultrastructural autoradiographic and immunocytochemical analysis of setae formation and keratinization in the digital pads of the gecko *Hemidactylus turcicus* (Gekkonidae, Reptilia). Tissue Cell, 2003, 35: 288–296
- 22 Alibardi L, Toni M. Immunological characterization and fine localization of a lizard Beta-keratin. J Exp Zool B Mol Dev Evol, 2006, 306B: 528–538
- 23 Guo C, Dai Z D, Ji A H, et al. The modulation and control of the Gecko's foot movement. J Bionics Eng, 2005, 2: 151–156
- 24 Sitti M, Fearing R S. Nanomolding based fabrication of synthetic gecko foot-hairs. IEEE-NANO, 2002, 137–140
- 25 Sitti M, Fearing R S. Synthetic gecko foot-hair micro/nano-structures as dry adhesives. J Adhes Sci Technol, 2003, 18: 1055–1074
- 26 Geim A K, Dubonos S V, Grigorieva I V, et al. Microfabricated adhesive mimicking gecko foot-hair. Nature Mater, 2003, 2: 461–463
- 27 Pickles J O, Corey D P. Mechano-electrical transduction by hair cells. Trends Neurosci, 1992, 15: 254–259

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