

Beyond the Microscope: The Power of High-Magnification Macro Photography

Microscopes have traditionally been the go-to tool for examining microscopic structures, delivering high magnification and allowing us to see fine details that would otherwise go unnoticed. However, photographing three-dimensional subjects, such as insect anatomy, poses several challenges with microscopes. The depth of field is often exceedingly shallow, reflective surfaces can be tricky to illuminate uniformly, and the working distances can be quite limiting. While focus stacking can be accomplished with a microscope and is commonly utilized, it proves to be more manageable in a macro photography setup.

This is where high-magnification macro photography comes into play. This hybrid method blends various macro techniques, including microscope optics, bellows extension, and reverse lenses, offering unique benefits. By using a high-resolution DSLR camera combined with bellows for precise control over focus, a dedicated tube lens, and high-magnification microscope objectives—or other similar techniques—you can achieve better control over lighting, depth of field, and focus stacking. This arrangement is perfect for capturing the intricate details of biological structures.

Importance of Vibration Control and Mitigating Diffraction Softening

At high magnifications, vibration and diffraction softening can greatly affect image sharpness and clarity. Even minor vibrations from mirror slap, external disturbances, or mechanical movement can lead to image blur, particularly with long exposures. To mitigate this, mirror-up mode was enabled, and a stable focusing rail was utilized to ensure precise movement between focus steps. Additionally, employing rear curtain sync with a flash reduces the effects of residual vibration by freezing motion at the end of the exposure.

Diffraction softening occurs when stopping down the aperture excessively, resulting in a loss of fine detail due to wave interference. This is especially significant in high-magnification setups, where light must pass through multiple optical elements. Wider apertures are favored over smaller ones since diffraction softening becomes more pronounced at very small apertures. Focus stacking allows the use of wider apertures while still achieving deep focus across the subject. Lenses typically perform optimally at one or two stops down from wide open, striking a balance between resolution and depth of field. Using high-quality objectives designed to minimize diffraction, combined with precise focus stacking, helped maintain image sharpness without sacrificing essential depth.

General Camera Setup & Imaging Workflow

In this series of images, a consistent high-magnification macro imaging workflow achieved the highest level of detail and clarity. All images were captured using a Nikon D850 camera in mirror-up mode to minimize vibrations. The optimization of shutter speed and flash settings reduced vibration effects, with rear curtain sync applied where appropriate.

Focus stacking was performed using Zerene Stacker software, and multiple flashes with

diffusion techniques controlled the lighting. Each setup included a focusing rail for precise incremental movement, allowing for extreme depth of field stacking at micron-level step sizes. Post-processing in Lightroom removed artifacts and adjusted contrast, with final sharpening applied using Topaz Photo AI. The following sections detail the specific setup variations for each subject.

Whole Insect Imaging: Pole Borer Beetle

One of the key advantages of macro photography is its ability to capture both full-body images and extreme close-ups with the same imaging setup. The image of the Pole Borer Beetle showcases the remarkable advantages of focus stacking and controlled lighting techniques in microscopic photography. These processes allow for a level of detail and clarity that typically cannot be achieved with a conventional stereo microscope, which often has limited depth of field and may miss intricate textures and features of the specimen. Additionally, controlled lighting plays a critical role in enhancing the visibility and contrast of the subject. By carefully manipulating light sources, photographers can highlight specific details, create dimensionality, and reduce shadows that could obscure important aspects of the beetle's morphology. The combination of these techniques results in a vivid, high-resolution image that captures the essence of the Pole Borer Beetle in a way that is both scientifically valuable and artistically impressive.

Camera Setup & Technique:

- Lens: Nikon 55mm AIS Micro f/2.8
- Extension: Nikon PK13 + PN11 Extension Tube (Total 80mm)
- Magnification: ~2x
- Aperture: f/5.6
- ISO 64
- Shutter: 5 sec
- Focus Stacking: Step size: 200 microns
- Number of steps: ~100
- Focusing Rail: WeMacro focusing rail

Bumble Bee Genitalia: Structural Complexity in Extreme Detail

The genitalia of bees exhibit remarkable specialization, playing a vital role in reproductive processes and assisting in distinguishing various species. The anatomy can include intricate features such as spicules, aedeagus, and modifications that are uniquely adapted for mating.

To capture the fine details of these structures, high-magnification macro photography is particularly effective. This technique enables the examination of minute features that are otherwise invisible to the naked eye. Controlled lighting is essential as it enhances contrast and reveals textures, ensuring that the details stand out clearly in the final images.

Multiple flashes were utilized during the photographic process to achieve optimal lighting

conditions. However, the specific number of flashes used was not documented. Adjusting the intensity and angle of the lighting can significantly impact the quality of the images obtained. Properly balancing these elements is crucial for effectively revealing the complexities of the bees' genital structures.

Camera Setup & Technique:

- Lens: Nikon AFD 24mm f/2.8 reversed on Nikon PB6 bellows
- Magnification: ~6x
- Aperture: f/4
- ISO: 125
- Shutter: 1 sec
- Focus Stacking: Step size: 40 microns
- Total steps: 47
- Focusing Rail: StackShot

Bumblebee Stinger: Defensive Adaptations

The bumblebee's stinger is a specialized defense adaptation that enables it to deliver venom when the bee perceives a threat. Thus, the bee protects itself and its colony. Unlike honeybees, whose barbed stingers remain embedded in their target, bumblebee stingers are smooth and do not become lodged, allowing them to sting multiple times if necessary. High-magnification macro photography significantly improves the study of bumblebee stingers compared to traditional methods like stereo microscopy, which has limitations. While stereo microscopes provide a three-dimensional view, they often restrict depth of field and can struggle with the fine details of minute structures. In contrast, macro photography allows for enhanced control over depth of field, enabling precise focus on the intricate details of the stinger.

Moreover, macro photography offers more flexibility in lighting, allowing researchers to illuminate features without causing glare or reflections that could obscure visibility. This is particularly important when studying small and complex structures. As a result, high-magnification macro photography stands out as an invaluable technique for capturing the complexities of bumblebee stingers. Compared to stereo microscopes and conventional microscopy, it offers a more thorough understanding of their roles in the bees' defensive strategies.

Note: The image was taken without rear curtain sync as part of an experiment to assess its impact on sharpness. While the results were acceptable, future high-magnification images will utilize rear curtain sync to minimize residual vibration effects.

Camera Setup & Technique:

- Microscope Objective: Nikon CFI Plan 10X Achromat Objective
- Tube Lens: Raynox DCR-150 on PB6 bellows

- ISO: 64
- Shutter: 1/250 sec (not rear-curtain sync)
- Focus Stacking: Step size: 5 microns
- Number of steps: 170
- Focusing Rail: WeMacro

Ground Beetle Leg and Tarsus: Adaptations for Locomotion

The leg and tarsus of a ground beetle are finely adapted structures that enable the insect to navigate various terrains, from rough surfaces to smooth plant stems. The specialized claws and adhesive pads provide both traction and the ability to cling to vertical surfaces.

To effectively image such structures, high magnification, deep focus stacking, and careful lighting control are necessary. A traditional microscope might not suffice due to its shallow depth of field and limited lighting options. The curved nature of the tarsus, combined with its fine setae and joint articulation, requires precise lighting to avoid harsh shadows while maintaining clarity in reflective areas. Moreover, focus stacking ensures that all critical structural details, including the claws and adhesive pads, remain in sharp focus.

Camera Setup & Technique:

- Microscope Objective: Nikon CFI Plan 10X Achromat Objective
- Tube Lens: Raynox DCR-150 on PB6 bellows
- ISO 64
- Shutter: 2 sec
- Focus Stacking: Step size: 5 microns
- Number of steps: 375
- Focusing Rail: WeMacro

Honey Bee Hindwing: Unlocking the Secrets of Flight

The hindwing of a honey bee plays a crucial role not only in flight but also in overall wing coordination. Positioned behind the forewing, the hindwings are typically smaller but are intricately shaped to enhance the bee's ability to maneuver and maintain stability in the air. Their design allows for rapid adjustments during flight, helping bees navigate complex environments while foraging for nectar or evading predators. The connection between the hindwing and forewing via the hamuli ensures that the bee can function as if it has a single, larger wing, optimizing lift and reducing drag. This synergy is essential for their agile flight patterns, which are necessary for their survival and efficiency in pollination.

Capturing the intricate details of the hamuli requires precise lighting control and an advanced imaging approach. Diffused lighting is essential to evenly illuminate the delicate wing structures without introducing harsh shadows that could obscure fine details. Since the hamuli are small

and arranged along the wing margin, achieving greater depth of field is crucial. Focus stacking allows for a fully detailed representation, ensuring that each hook-like structure remains sharp and visible. This technique highlights the essential role of the hamuli in connecting the forewing and hindwing, a key adaptation for efficient bee flight.

Camera Setup & Technique

- Microscope Objective: Nikon CFI Achromat LWD 20X objective,
- Tube Lens: Raynox DCR-150 on PB6 bellows
- ISO: 64
- Shutter: 1 sec
- Focus Stacking: Step size: 3 microns
- Number of steps: 82
- Focusing Rail: StackShot

The Fly's Compound Eye: Structure and Detail

The compound eye of a fly is an intricate and highly specialized visual system composed of thousands of individual photoreceptive units called ommatidia. Each ommatidium functions as a tiny independent eye, capturing a narrow section of the fly's surroundings. Together, these units create a wide field of view and enable the fly to detect rapid movement with remarkable precision. Unlike human vision, which relies on a single lens to focus images onto the retina, the fly's compound eye forms a mosaic of visual inputs, allowing for enhanced motion detection at the expense of fine resolution. The hexagonal arrangement of the ommatidia reduces gaps in vision, maximizing light capture and efficiency, making this structure particularly well-suited for a fast-moving insect.

A compelling example of macro-micro photography is the detailed close-up of a fly's compound eye. Traditional microscopes often struggle with uneven lighting and a shallow depth of field, making it difficult to capture the full structure. This imaging approach, however, provides precise control over illumination and focus stacking, ensuring an image that is both scientifically informative and visually striking.

Camera Setup & Technique:

- Microscope Objective: Nikon CFI Achromat LWD 20X objective,
- Tube Lens: Raynox DCR-150 on PB6 bellows
- ISO: 64
- Shutter: 1 sec
- Focus Stacking: Step size: 2 microns
- Number of steps: 597
- Focusing Rail: StackShot



Figure 1 - Pole borer beetle



Figure 2 - Bumblebee Genitalia



Figure 3 - Bumblebee Stinger



Figure 4 - Ground Beetle Leg and Tarsus

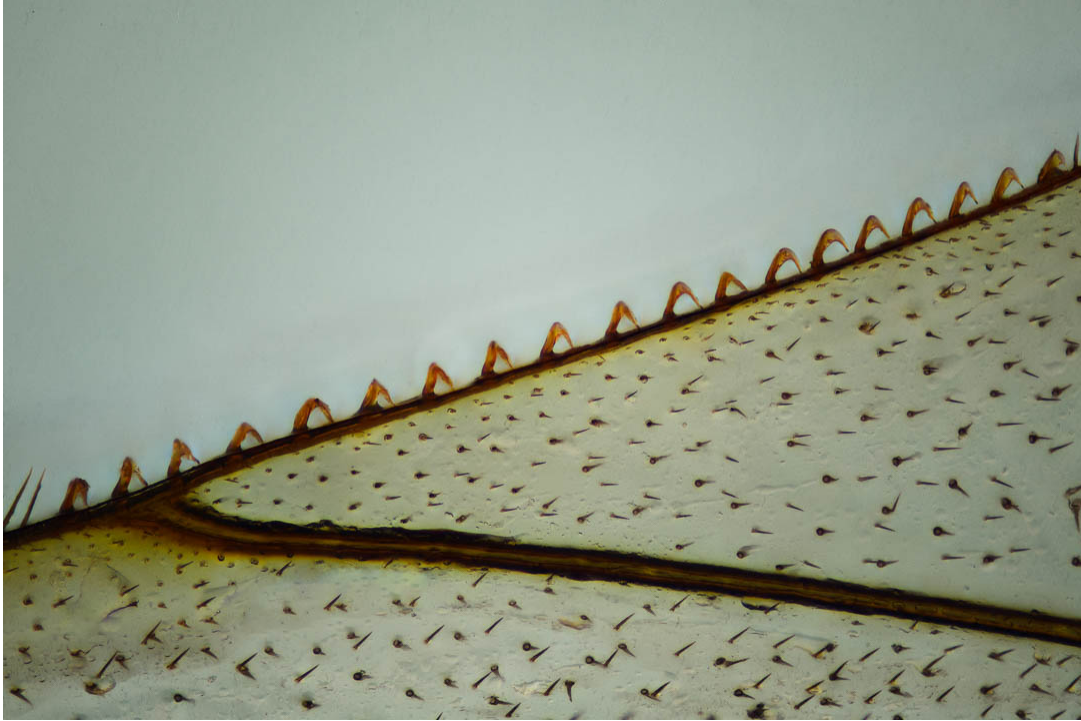


Figure 5 - Honey Bee Hindwing

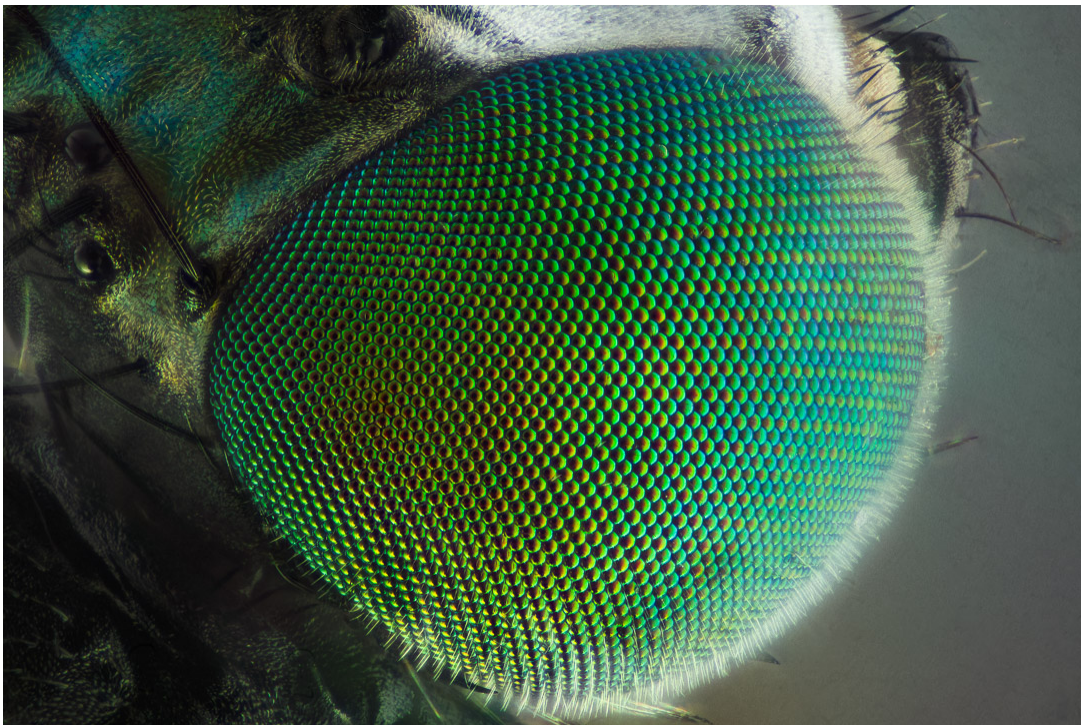


Figure 6 - Fly Compound Eye

Comments to the author Gedaliah Wolosh welcomed, email - gwolosh AT gmail DOT com.

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