# **Aquatics under Polarization**

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A local roadside ditch is a rich source of aquatics possibly because it lacks predatory fish; the only predatory animals I have seen there are a few dragonfly larvae. Many of these aquatics, particularly the arthropods, make great subjects for polarized microscopy. I have an Olympus BH2 microscope fitted with, what Olymps calls, a Simple Polarizing Attachment consisting of an Intermediate Tube (BH2-KPA) with a 1.25x magnification factor that fits above the objectives and below the observation tubes. An upper polarizer (analyser) slides into the Intermediate Tube and a 2<sup>nd</sup> rotatable lower polarizer rests below the condenser. When crossed the two polarizers give total light extinction but birefringent materials glow white, referred to as 'plate out' in the images below. The upper polarizer has a slot for sliding in a compensator, or wave, plate. I have an Olympus AH-TP530-2 first order red 530nm plate. When inserted into the upper polarizer it often gives a strong magenta cast to the subject but also makes birefringent materials glow with bright colours as the lower polarizer is rotated, referred to as 'plate in'.

#### Protozoa

The only protozoa that I have found to show any enhancement of structures are *Paramecium* and some of the tests of certain Amoeba. *Paramecium* structures are best seen under DIC lighting but the odd specimen shows coloured inclusions under polarization (Fig. 1).



Fig. 1 Paramecium; plate in

A much greater effect is seen in certain Amoeba that utilize sand grains to construct a test (shell). Under brightfield illumination these tests resemble tiny clear glass vases but under polarized light the individual grains glow (Fig. 2).

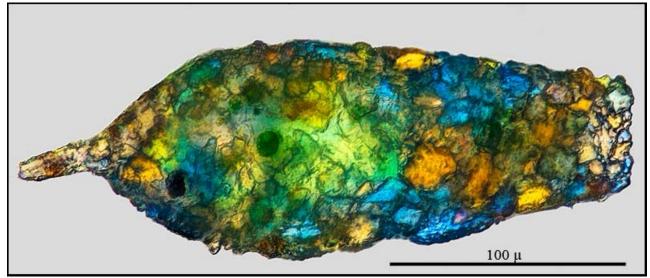


Fig. 2 Testate amoeba; plate in

This specimen has been identified by Ferry Siemensma, an expert on the group, as *Difflugia acuminata*, the light green 'blob' in the lower half of the test is the actual living amoeba.

### Arthropods 1 – Copepods

The musculature of these animals shows up really well under polarization. Figure 3 is the same specimen under different lighting – center is with the plate out, right and left with the plate in and with the lower polarizer at different positions. Figure 4 is a close-up of the cephalothorax showing the dorsal longitudinal muscles extending almost to the single eye.



Fig. 3 Copepod, center plate out, right & left plate in

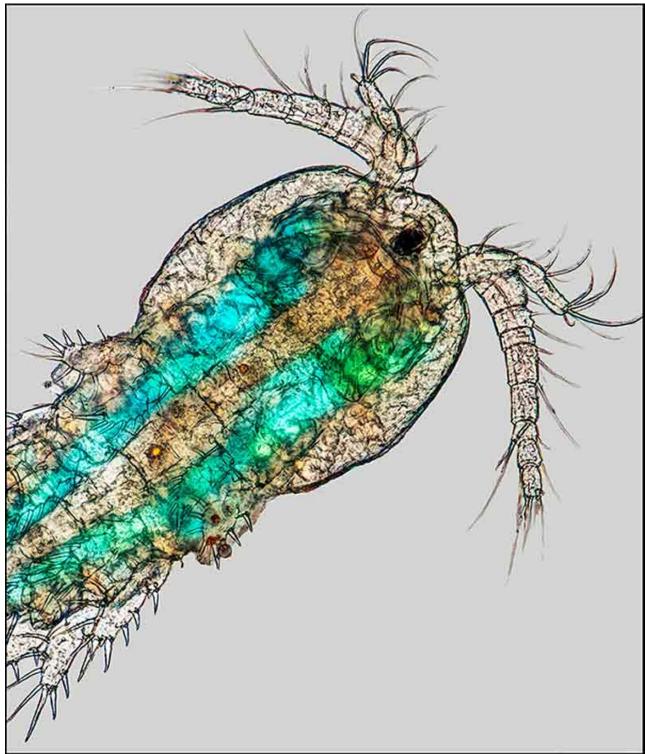


Fig. 4. Copepod; cephalothorax; plate in

Another specimen in side view (Figs. 5, 6).





Fig. 6. Copepod, plate in

#### Arthropods 2 – Freshwater Shrimp

These are common inhabitants of my local roadside ditch. Figure 7 shows the bilaterally flattened animal under reflected light.



Fig. 7. Freshwater shrimp

Under crossed polarizers the muscles in the bases of the antennae, the vertical muscles in the head that move the mouthparts, the dorsal muscles, and the muscles in the ventral appengages show as white (some colour in the dorsal muscles) in an otherwise dark animal (Fig.8).



Fig. 8. Freshwater shrimp; plate out

With the wave plate in the muscles take on various colours depending, I believe, on their orientation with regards to the polarized light. Note that the 1<sup>st</sup> 4 pairs of legs are pointed forward to assist in feeding while the more posterior legs are used for locomotion. Legs 1 & 2 have large grasping claws and large associated muscles (Fig. 9, seen in more detail in Figs. 10, 11, 12). Note the large vertical mouthpart-muscles in the head behind the eye (Fig. 10 and in close-up Fig.12).

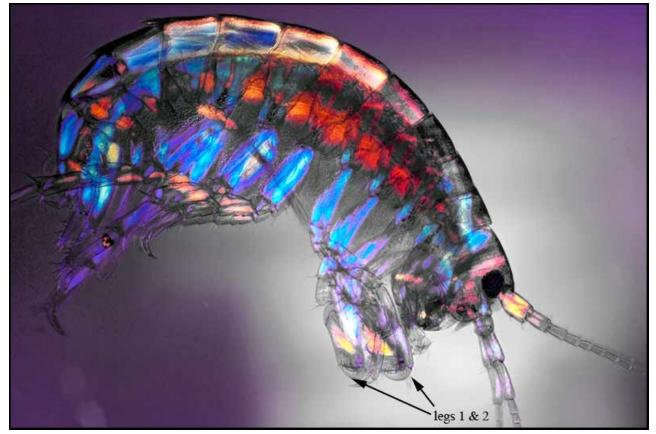


Fig. 9. Freshwater shrimp; plate in

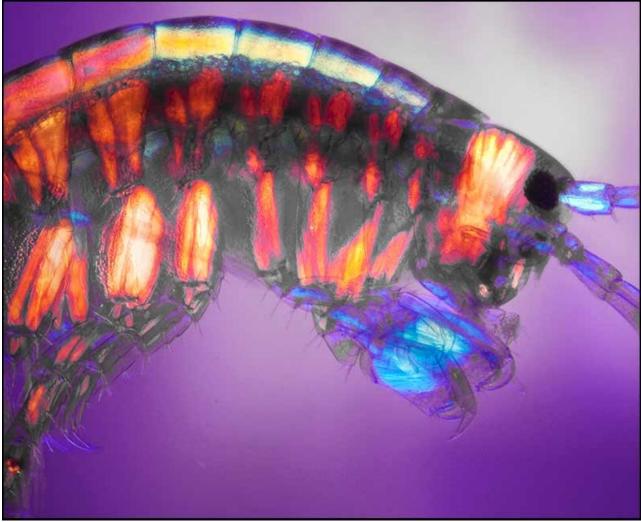


Fig. 10. Freshwater shrimp, anterior; plate in



Fig. 11. Freshwater shrimp, leg 1 modified for grasping food; plate in



Fig. 12. Freshwater shrimp, anterior musculature; plate in

## Arthropods 3 – Mayfly nymph

As with the other aquatic arthropods, the musculature is clearly defined under polarization. Figure 13 shows a mayfly nymph under reflected light and Fig. 14 shows the posterior end of the abdomen under polarization with the lower polarizer at different positions.



Fig. 13. Mayfly nymph, reflected light

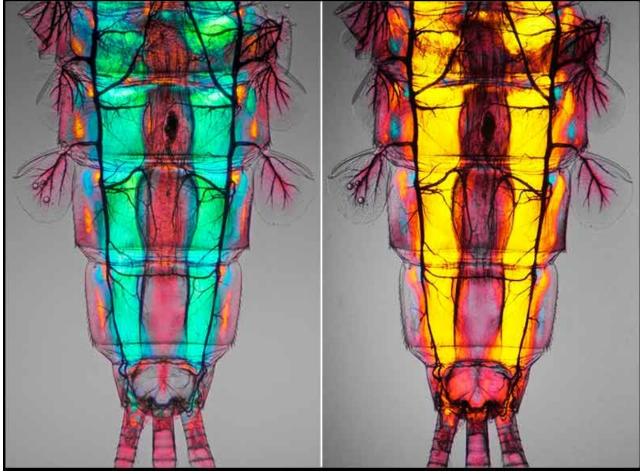


Fig. 14. Abdomen of mayfly nymph, polarized light;plate in.

Figure 15 is a close-up of a partial abdominal segment with muscles showing red, the exosketon blue and the trachea as black tubes (also seen in Fig.14).



Fig. 15. Partial abdominal segment of mayfly nymph, muscles red, exoskeleton blue, trachea black; plate in

#### Arthropods 4 – Chironomid midge, larva & pupa

Another abundant organism in the bottom sediment of the ditch. They are rather large insects for microscopic work, round in cross section with a bright red blood pigment, haemoglobin. Figure 16 shows a larva removed from its tube. Head on the right, ventral side is up, muscles in head and thorax showing as green/yellow, muscles in rest of body as dorsal and ventral bands. Dark center tube is gut with food. Even with a 2x objective (and a 1.25x intermediate tube lens + 2.5x relay lens) it is too long to get a full image on my D90 sensor.

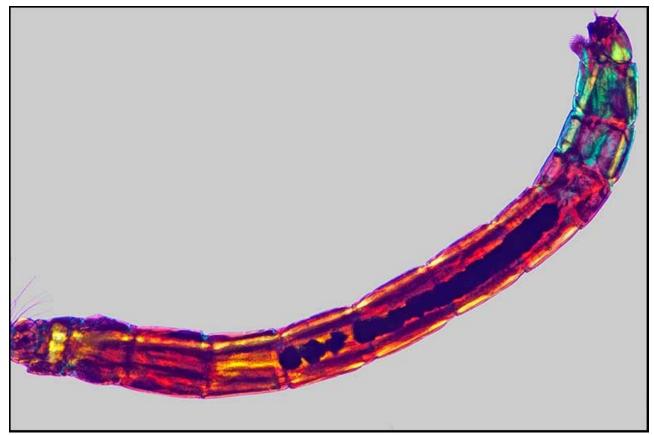
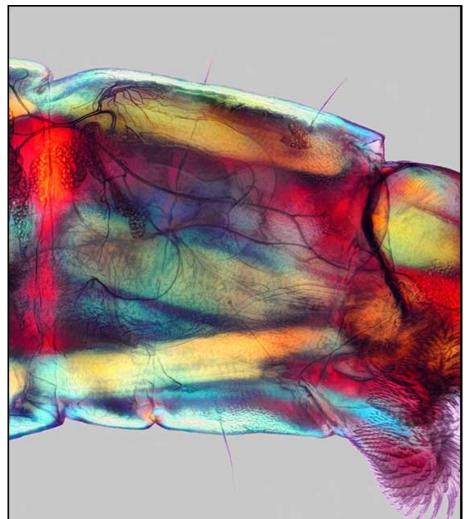


Fig. 16. Chironomid midge larva; plate in

Figure 17 is a close-up of the head and the 3 thoracic segments and Fig. 18 is a close-up of the 1<sup>st</sup> thoracic segment and the back of the head.



Fig. 17. Chiromid midge larva, head & thorax; plate in



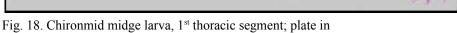


Figure 19 is a dorsal view of the last few abdominal segments showing the two bands, green, of dorsal muscles.

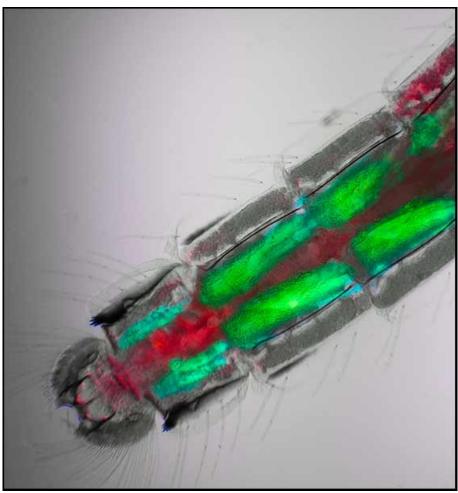


Fig. 19. Chironomid midge larva, last few abdominal segments; plate in

Figure 20 shows the anterior part of a pupa, the wing muscles showing green and the large lobe being the future wing of the adult.

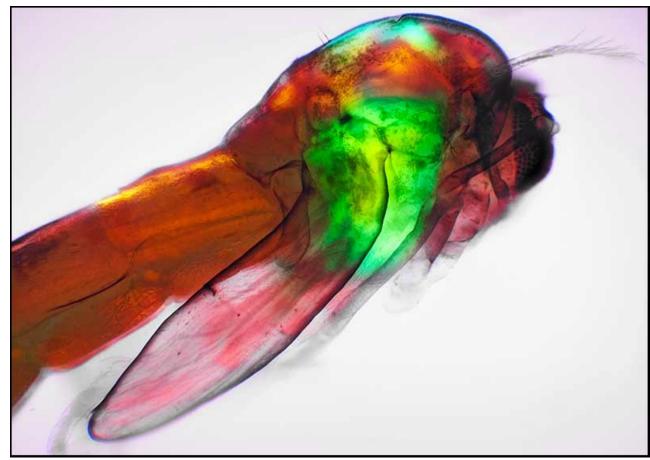


Fig. 20. Chironomid midge pupa showing wing muscles, green, and future adult wing; plate in

## **Microscope and Photographic Equipment**

My basic equipment is an Olympus BH2 with 2x, 4x, 10x, 20x, 40x, 60x, and 100x objectives; Olympus 2.5x NFK relay lens. I also have the components for Phase Contrast, DIC and Polarization. Camera is a Nikon D90 with Nikon PB-6 bellows; Nikon flash in place of Olympus' halogen lamp. For reflected light images I use Nikon CF objectives and El-Nikkor enlarging lenses.

Most images are stacks of several frames processed by Zerene Stacker.

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