Distinguishing Microfossil Bio-Silica From Calcium Carbonate In Oblique Illumination

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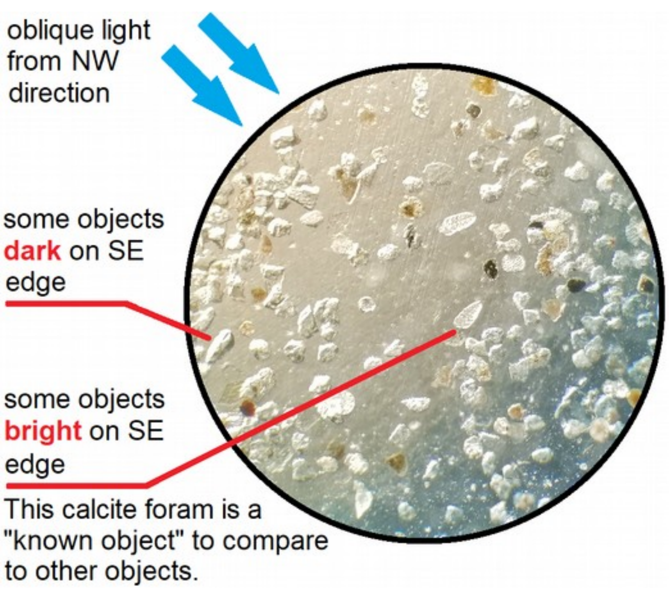
Introduction. This short note describes an optical trick I use to distinguish bio-silica microfossils from calcium carbonate microfossils without resorting to polarized light. The test uses directional shading of oblique illumination to show relative differences in index of refraction ("n" or "R.I."). The test relies upon the mounting medium having an R.I. between the items being compared. Common media such as Canada balsam and Norland 61 are suitable (Figure 1). High index diatom media are not suitable for this test, nor are mounts in air or water. The test consists of simply noting whether the unknown object has an R.I. less than or greater than the mounting medium. In other words: which side of the object has the shadow.

Figure 1. *Index of refraction of some mounting media and subjects. Items in red are considered in this article. Index of refraction values were tabulated from numerous sources, both online and in print.*

MOUNTING MEDIUM	Index of Refraction
air	1.00
water	1.33
glycerin jelly	1.443
Biological media: Euparal, Histoclear, Permount, Clearmount, etc	1.48 to 1.51
homogeneous immersion oil and glass slides	1.515
Canada balsam (variable)	1.52 to 1.54
Norland 61	**1.56**
Styrax	1.58
High index diatom media: Hyrax, Pleurax, Naphrax, Zrax	1.70 to 1.80
Realgar ☠	2.4

SUBJECT MATERIAL	Index of Refraction
Biological	
cytoplasm	1.35
cell walls	1.42
cell membrane (variable)	1.46 to1.60
Mineralogical	
Bio-silica (opal): sponge, diatom, radiolarian, ebridian, silicoflagellate, phytolith	**1.434**
Quartz	1.544, 1.553
Calcite: coccolith, foram, ostracod, some mollusc, echinoid, statoliths	1.486, **1.640**
Aragonite: some mollusc, including pteropods. some forams	1.530, 1.681, 1.685

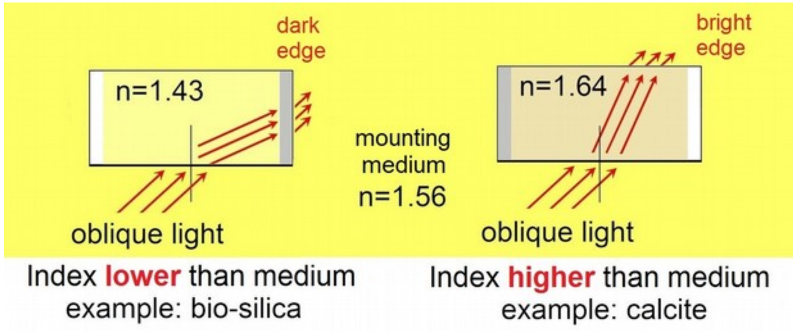
Background. Bio-silica (a form of opal) and calcium carbonate (usually calcite; sometimes aragonite) are materials often seen in microfossil shells. In axial transmitted light these materials look alike. They are colorless and transparent with medium to high optical relief. If you recognize the group to which your specimen belongs then, by extension, you already know its composition. If you don't recognize the type of fossil, the first step in identification is to determine what it is made of.



The Molecular Expressions microscopy website discusses R.I. in oblique illumination and offers an interactive tutorial (link). Try this experiment with the tutorial: drag the specimen R.I. slider from minimum to maximum and watch the eyepiece image change.

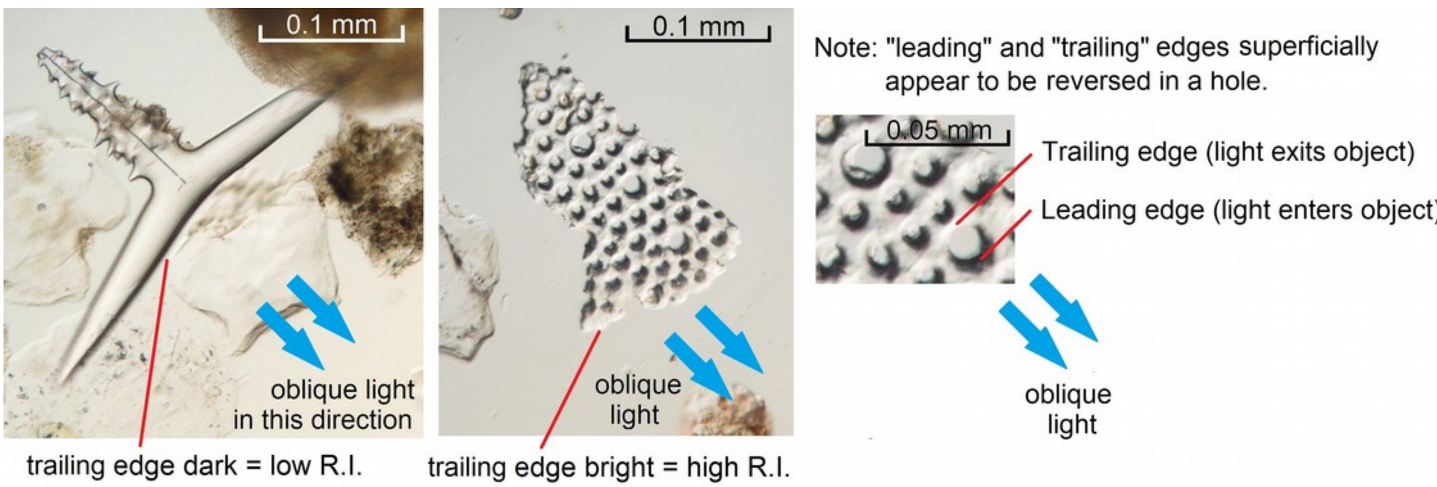
Test For Relative R.I. Figures 2, 3, 4 show how the test is set up and its results. The test depends upon both the direction of oblique light and the R.I. of the mounting medium, so I always observe a known object and note how the shadows fall before using the test on an unknown object (Figure 2). You don't need a decentering condenser iris for this test. You don't even need an oblique patch stop. It is enough to hold your fingers or a card below the condenser to block the light from one side.

Figure 2. Preliminary: *Establish oblique light. This overview shows the eyepiece image with a 4X objective.*



A **B**

Figure 3. The Theory. *This cross sectional schematic shows how light follows Snell's Law and refracts towards the perpendicular in the higher index component.* **A.** *Objects with a* ***low R.I.*** *are bright on the side where oblique rays enter the object and dark where oblique rays exit the object.* **B.** *Objects with a* ***high R.I.*** *have the opposite shading. They are bright where oblique rays exit the object.*



A **B** **C**

Figure 4. The Practice. *Photos were taken with a 10X objective. The mounting medium has n=1.57 (the old mountant Caedax whose R.I. is close to the currently available Norland 61). Sample and lighting are the same as Figure 2. The sample is from the Atlantic abyssal plain off Morehead City, North Carolina.* **A.** *A shadow along the trailing edge (the side where oblique light leaves the object) indicates an R.I. lower than the mounting medium. In this example we have bio-silica in the form of a sponge spicule.* **B.** *A bright trailing edge indicates an R.I. greater than the mounting medium. In this example we have a calcite shell fragment of a foram, possibly the planktic species* *Orbulina universa*. **C.** *This is a reminder that "leading" and "trailing" edges are with respect to the object, not holes in the object!*

Summary. You have an unrecognized microfossil whose composition you believe to be either bio-silica or calcium carbonate. Initiate oblique illumination. If the trailing edge of the specimen is dark, the material is bio-silica; if the trailing edge is bright, the material is calcium carbonate. This test can be adapted for other paired materials. It is not a general test for all materials, because most samples have multiple items in each R.I. category. If your material is an unknown mineral grain, this test alone won't identify it. The test *will* tell you if the grain has a low or high R.I. – the first piece of information to begin the identification process.

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