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CARGILLE LABORATORIES
55 Commerce Road • Cedar Grove, New Jersey • 07009-1289 USA
Phone: 973-239-6633 Fax: 973-239-6096 www.cargille.com

MEASURING REFRACTIVE INDEX OF LIQUIDS
USING CARGILLE PRECISION SOLID REFRACTIVE INDEX STANDARDS
REFERENCE SETS M-7, M-18 AND M-25

The Becke Line Technique as descried in Cargille Technical Bulletin: Microscope Techniques, The Becke Line Method can be used with Cargille Reference Sets M-7, M-18 and M-25 to measure the refractive index of liquids of unknown refractive index. These sets contain solids of precisely known refractive index; the solids are in the form of -100 mesh grains, ideal for Becke Line determination and come with a table of "Precise Optical Values" (CARGILLE M-7 REFERENCE SET PRECISE OPTICAL VALUES). With practice the following method can measure the refractive index of liquids to better than ± 0.003 and usually better than ± 0.001, even though the intervals between the solid standards are normally ± 0.001 (see 5. and 6.)

PROCEDURE:

Read Technical Bulletin: MICROSCOPE TECHNIQUES, The Becke Line Method, and the cover sheet on the table of "Precise Optical Values".

Set up microscope with Köhler illumination or as in the Technical Bulletin preferably with white (incandescent) light and a removable 589 nm filter (see 2.), and some form of temperature monitoring (see 1.)

Make a succession of slides using the unknown liquid and solid standards by the Becke Line Method (see 3. and 4.) until either a solid is found that matches the refractive index of the liquid or the liquid appears to be between two solids of the closest interval.

If the liquid matches a solid, then look up the n_D (5893A) on the table of "Precise Optical Values" for that solid, and that value will be the refractive index of your liquid at the temperature of the slide (assuming refractive index change with temperature of the solid is negligible; see 1.). If the liquid appears to be between two solids you will obtain the refractive index of your liquid by interpolation: estimating how close in refractive index the liquid appears to be to both solids (see 5. and 6.).

NOTES:

1. TEMPERATURE The refractive index of a liquid will vary with temperature so it is important to know the temperature of the slide on your microscope stage. The change in refractive index of liquids with temperature is much greater than that of solids so even small changes in temperature will make a noticeable change in a liquids' refractive index. Work in a room of constant temperature and without drafts; the room temperature will then approximate the temperature of a slide on your microscope stage. A 3" mercury thermometer (graduated in 0.1°C) mounted near or taped to the microscope stage is a better indicator of stage temperature. The accuracy of your refractive index measurement will be further improved if you measure temperature using a digital thermometer with a micro-thermocouple in contact with the cover slip of your slide on the microscope stage. A small glass weight placed on the tip of the thermocouple on the cover slip will hold it in place and shield it from minor drafts.

The standard temperature for refractive index determinations is usually 25°C. To convert readings at other temperatures to 25°C you must use the temperature coefficient (the change in refractive index per 1°C) of the liquid. If you do not know the temperature coefficient of your liquid it is useful to know that most liquids have a temperature coefficient of about - 0.0004 per °C. Cargille Refractive Index Liquids have the temperature coefficient for each liquid on the label.

For example, Cargille Refractive Index Liquid Series A 1.498 has on the label: $(-)dn_D/dt = 3.85x\ 10^{-4}/°C$; this means that for each degree C (Celsius) increase in temperature , the refractive index will decrease by 0.000385. If your slide on the stage is 26°C and the liquid is Series A 1.498, then since Cargille Refractive Index Liquids are calibrated at 25°C, a refractive index reading of 1.4976 is what you will expect since 1.498000 – 0.000385 = 1.497615. If the liquid has an unknown temperature coefficient and your slide is at 26°C and your refractive index reading is 1.4976, you can estimate the refractive index at 25°C as 1.4976 – 0.0004 x (25 – 26) = 1.4980.

You can measure the temperature coefficient for a liquid by measuring the refractive index at two temperatures (for example: at 20 and 30°C) and dividing the difference in refractive index by the difference in temperature.

The temperature coefficients for solids are very small when compared to liquids so when you make readings using solids from Cargille Reference Sets M-7, M-18 or M-25 that are within a few degrees of the temperature at which the solids' refractive index values were read (listed on the table of "Precise Optical Values" under TEMP, °C and is usually 22°C), their change in refractive index due to temperature may be considered negligible and you can use the refractive index values for the solids directly from the table of "Precise Optical Values" listed under "n_D , 5893A" without making a temperature correction. For example, a temperature coefficient of 0.00001 is quite large for a solid, but even used at 30°C, the change in refractive index would be quite small: 30 - 22 = 8, 8 x 0.00001 = 0.00008. The temperature coefficients for most of the solid standards are listed in the table of "Precise Optical Values" under "TEMP COEF" (multiplied by 10^7). The difference between the temperature coefficient for ne listed under "e" on the table and the temperature coefficient for n_D is negligible.

2. <u>MONOCHROMATIC LIGHT</u>: Refractive index varies with wavelength; this variation is called "dispersion". In order to give a material a single refractive index value, sodium light (589 nm) is normally picked as the standard wavelength for refractive index measurements. When doing Becke Line using white light it is easy to be confused by the rainbow of light that surrounds the edge of the solid when the refractive index of the solid is close to that of the liquid as observed under the microscope. This rainbow is caused by the difference in dispersion between the solid and the liquid. Within the rainbow is the yellow-orange sodium wavelength. Using a 589 nm filter or using a sodium lamp will eliminate the dispersion rainbow and thus facilitate the reading and improve accuracy.

3. <u>PREPARATION OF SLIDES</u>: Prepare slides in the following manner: drop about five drops of liquid in the center of the slide; put a few grains of the solid in the center of the liquid; place a cover slip on top, and rock back and forth with the eraser end of a pencil to level the cover slip; if more liquid is needed to fill the space under the cover slip, add a few additional drops at the corners of the cover slip; then label the slide as to the liquid and solid used.

4. <u>ACCURATE READING OF BECKE LINE</u>: Since the Reference Set solids consist of fractured particles, you should look for a particle with a knife like edge to read the Becke Line, to avoid false lines caused by a particles' surface irregularities.

5. <u>ESTIMATION OF HOW CLOSE IN REFRACTIVE INDEX A LIQUID IS TO A SOLID</u>: You will notice that the closer the liquid is to the refractive index of the solid the less distinct will be the image of the solid, and using white light, the rainbow effect caused by the difference in dispersion between the solid and the liquid will become more intense.

Using white light, if the solid and liquid match or nearly match in refractive index for the sodium light wavelength (589 nm), raising the focus will create a pale blue line around the outside edge of the solid, and an orange line along the inside edge. This position of the pale blue line outside and the orange line inside is what you will find almost without exception because the dispersion of a liquid is almost always higher than a solid of the same refractive index; this is what you will find using Cargille Reference Set M-25 solids with Cargille Refractive Index Liquids. In a rare case when the dispersion of the liquid is less than the solid, the position of the orange and pale blue lines will be reversed. If the dispersions match, the particles of solid will virtually disappear in both sodium and white light because the refractive indices match at all wavelengths of light. When the refractive index at the sodium wavelength of the liquid is very close to the solid, you may find that the solid almost disappears when using a sodium light or 589 nm filter; in this case, white light slightly out of focus will make it easier to locate particles of solid in the field. The farther the liquid is from the refractive index of the solid, the more sharply defined will be the image of the solid; the solid will appear in higher relief, the Becke Line will be bolder and brighter, and using white light, the rainbow effect will tend towards red-brown and very pale blue lines, if the liquid is higher than the solid, or towards very pale yellow and gray-blue, if the liquid is lower that the solid; farther away still, the rainbow will become more gray and washed out.

You can express how close the refractive index of a liquid is to that of a solid standard by using a percentage of the difference in refractive index between one solid standard and the next, one solid being higher in refractive index than the liquid, and the other solid being lower. For example, you might make a slide of a liquid with the 1.68 solid and another slide with the 1.69 solid, and after examining the two slides you might say that the liquid appears to be 25% (of the difference between the solids) higher than 1.68 and 75% lower that the 1.69 solids.

Practicing with liquids of known refractive index, such as freshly opened Cargille Refractive Index Liquids, will help to clarify this technique. You will see that it is easy to differentiate between at least five liquids of different refractive indices that fall between two solid standards as in the following example:

TWO SOLIDS, A AND B, WITH "A" .01 LOWER THAN "B" IN REFRACTIVE INDEX (n_D) AS SEEN IN LIQUIDS OF VARIOUS n_D

* CONTRAST OF IMAGE (COI)
* APPEARANCE OF BECKE LINE (ABL)

* DIRECTION OF BECKE LINE (DBL) WHEN FOCUS IS RAISED – in or out of particle

Liquid	nD of Liquid		Solid A	Solid B
1.	Equal to A	COI	Image not Visible	Very bold
		ABL	Line not visible	Very bold
		DBL	Can not tell	In
2.	25% above A 75% below B	COI	Faint	Bold
		ABL	Faint	Bold
		DBL	Out	In
3.	50% above A 50% below B	COI	Fairly bold	Fairly bold
		ABL	Fairly bold	Fairly bold
		DBL	Out	In
4.	75% above A 25% below B	COI	Bold	Faint
		ABL	Bold	Faint
		DBL	Out	In
5.	Equal to B	COI	Very bold	Image not visible
		ABL	Very bold	Line not visible
		DBL	Out	Can not tell

* Using Sodium Light

6. EXAMPLE OF INTERPOLATION BETWEEN SOLIDS:

At 27°C the liquid appears to be 25% above the index of solid 1.68 lot C and 75% below 1.69 lot B. The temperature coefficient for the liquid is – 0.000385.

A. Look up n_D value for the two solids in the table of "Precise Optical Values":

Solid 1.68 lot C: n_D = 1.67827 at 22°C
Solid 1.69 lot B: n_D = 1.68880 at 22°C

B. Apply Temperature Coefficient for the solids to adjust refractive index to 27°C (this step usually skipped because of the negligible effect of temperature on solids). Look up the temperature coefficients for the solids in the table of "Precise Optical Values" under "TEMP COEF, e" (note: these are recorded x10^7).

Solid 1.68 lot C: Temp. Coef. = 0.0000002
Solid 1.69 lot B: Temp. Coef. = 0.0000080

temperature difference	temp. coef. of 1.68 solid times temp. diff.	n_D of 1.68 solid at 27°C	temp. coef. of 1.69 solid times temp. diff.	n_D of 1 .69 solid at 27°C
27	0.0000002	1.6782700	0.0000080	1.6888000
-22	x 5	+0.0000010	x 5	+0.0000400
5	0.0000010	1.6782710	0.0000400	1.6888400

C. Calculate n_D of the liquid at 27°C:

Difference in n_D of solids	Percent of difference that liquid is higher in n_D	n_D of liquid at 27°C
1.6888400	0.0105690	1.6782710
-1.6782710	25%	+0.0026422
0.0105690	0.0026422	1.6809132

D. Calculate n_D of the liquid at 25°C:

temperature difference	temp. coef. of liquid times temp. diff.	n_D of liquid at 25°C
25	-0.000385	1.6809132
-27	x- 2	+0.000770
-2	0.000770	1.6816832

E. Rounding off then: liquid n_D = 1.6817 at 25°C