Refractive Index (RI) and Brix Standards – Theory and Application

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1.0 Summary

The measurement of Refractive Index (RI) is an important parameter in a whole range of industries. There are several makes and models of refractometers available on the market, that vary in cost and complexity. The instruments fulfil to a varying degree, fitness for purpose requirements for various industries and various sample types. In some cases, RI measurement is heavily regulated, such as in the sugar industry and there have been a wide array of ASTM written standards produced for many applications. Irrespective of application, industry type, sample type, instrument used or target measurement uncertainty, high quality calibration standards are an imperative. These can also be used for quality control, method validation, analyst qualification or proefficiency testing. This paper details a brief explanation of the science of RI, how a typical refractometer works and presents some examples of instruments available in the marketplace. This is followed by narrative of the relevance of RI measurement and various ASTM standards that have been published depending on application.

The final section of the paper explains the reasons why high-quality liquid standards are necessary and then details specific RI standards available from Reagecon. It is important to point out, that there are other high quality manufacturers of such standards, but the Reagecon standards are presented here for example purposes, as these are most familiar to the author. In broad terms there are four main classifications of RI standards that include ICUMSA compliant and non-compliant sucrose standards expressed in Brix values, sucrose standards expressed as RI and solvent based standards expressed as RI.

It is hoped that this publication will provide the reader with a sound theoretical knowledge of RI measurement, and will provide a detailed knowledge of the metrology pertaining to RI and furnish the analyst with the knowledge and tools to procure and use RI or Brix Standards optimally.

2.0 Theory of Refractive Index (RI)

The speed of light travelling through a vacuum is always the same. When light moves through another medium, its speed slows down because the light is being constantly absorbed and re-emitted by atoms within the material through which the light is travelling. The ratio of the speed of light in a vacuum to the speed of light in another substance is defined as the Refractive Index for that substance. This relationship can be expressed as follows:

Fig. 1

$$RI = \frac{\text{speed of light in a vacuum}}{\text{speed of light in the substance measured}}$$

In terms of how a refractometer works, when light changes speed as it crosses the boundary from one medium to another, as well as its speed changing, its direction also changes. In other words – it is refractive. The relationship between the speeds of light through the two media, the angles of both incident and refraction and refractive index of the two media is expressed in a simple formula.

Fig. 2

$$\frac{V_A}{V_B} = \frac{\sin\theta_A}{\sin\theta_B} = \frac{\eta_B}{\eta_A}$$

On the basis of this formula, it is not necessary to measure the speed of light to determine its refractive index. Instead by measuring the angle of refraction and knowing the refractive index of the layer that is in contact with the sample – it is possible to measure the refractive index of the sample. Nearly every instrument on the market utilises this principle, although instruments may vary in optical design.

3.0 How a Refractometer Works

Samples with different refractive indices will produce different angles of refraction, bearing in mind that the angle of incidence and the refractive index of the prism are constant (Diagram 1).

A different sample in each glass exhibiting different refractive indices.

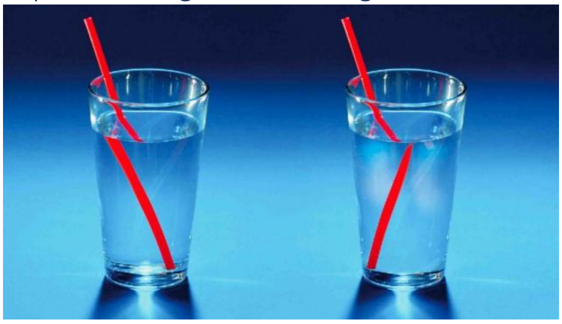

Diagram 1

The different angles of refraction in different samples will be reflected in a change in the position of the border line between the light and dark regions of the instruments.

By calibrating the instrument appropriately, the position of the border line can be used to determine the refractive index of any sample. More specifically, the sample is placed on the prism of the instrument, incident light is shone onto the prism, which is transmitted through the sample (See Diagram 2).

Light being transmitted through a sample placed on a refractometer prism.

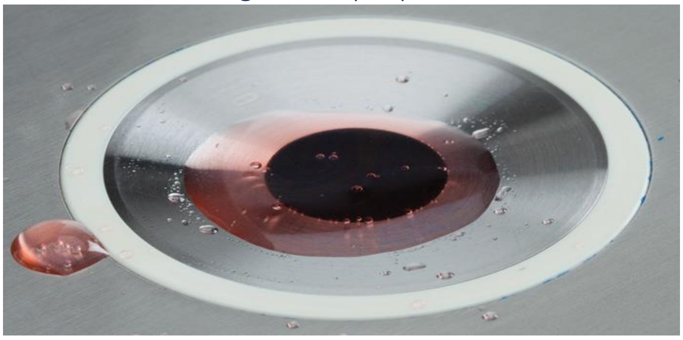

Diagram 2

This incident light is diffracted at the interface between the sample and the prism. The refractive index value is then calculated based on the relationship between the angle of incidence and the angle of diffraction for light hitting the boundary between the two media (See Diagram 3).

Comparison between angle of incidence and angle of refraction.

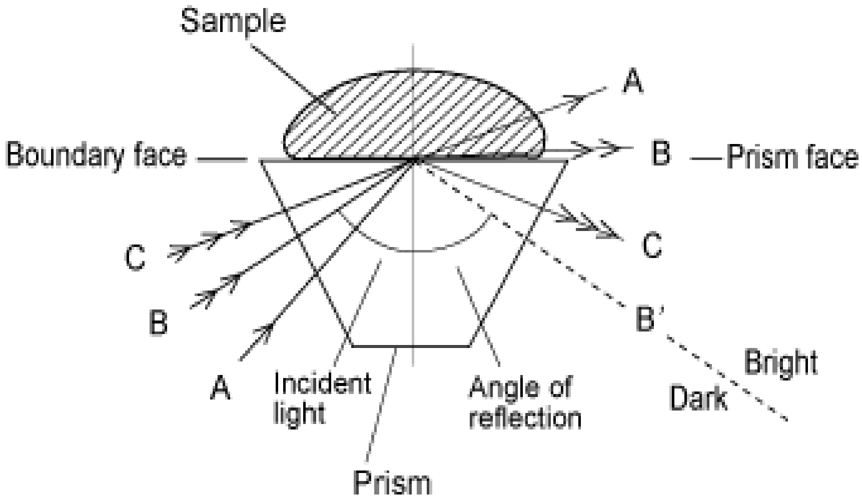

Diagram 3

However, accurate measurement is dependant, both on the temperature of the sample and the wavelength of the light used. The importance of both parameters will now be briefly explained, but in a lot of instruments the temperature is standardised to 20°C and the wavelength of the light is standardised at 589.3nm.

First of all, to deal with wavelength. In most sample types the refractive index will vary with wavelength. This variation is called dispersion and it is this phenomenon that causes white light moving through a prism to be refracted into the component colours of the rainbow. (Red, Orange, Yellow, Green, Blue, Indigo, Violet). For accurate measurement of refractive index, it is necessary to use monochromatic light and the commonest source of light used in refractometers is Sodium D Light at 589 nm, although this does not apply to ABBE Refractometers.

In terms of the temperature coefficient of variation, the speed of light is slower in a sample, than in a vacuum, due to absorption and emission of light by the atoms, as has been already stated. Since the density of a sample normally decreases as temperature increases, the speed of light through the sample will increase. Consequently, the refractive index will decrease. For water, this decrease in refractive index will be approximately 0.0001/°C. For certain other types of sample including liquid organics, the refractive index decrease will be approximately 0.0005/°C. Therefore, the management of temperature is critical for accurate measurement of refractive index.

All high-quality bench, on line and handheld refractometers where accurate measurement is required are equipped with temperature management systems. Refractive index results should always be reported at the temperature of measurement, which is normally either 20 or 25°C and the temperature of measurement is often denoted with the wavelength of the light used. Refractive Index measurement can be performed manually or automatically. A selection of widely available types of refractometer can be seen in Diagram 4.

A selection of widely available commercial refractometers.

Diagram 4

4.0 Why Measure Refractive Index

Refractive index is measured as part of the characterisation of liquid samples and is analogous to the way the measurement of melting point is used to characterise solid samples. Each substance has its own refractive index and its measurement may be of value in the following situations:

Firstly, to either identify or confirm the identity of a sample by comparing its refractive index to known values. Such known values can be derived experimentally, taken from the technical or scientific literature, or obtained from the manufacturer of the liquid. Secondly, Refractive index may be used to assess the purity of a sample by comparing the refractive index of the sample to the known value for the pure substance. Thirdly, Refractive index can be expressed in BRIX values for the measurement of the sugar content of sugar rich samples such as jams, jellies, syrups and fruit juices. The BRIX value correlates to the sugar concentration in the sample measured and is expressed as the number of grams of sucrose in 100 grams of sucrose/water solution. The relationship between the refractive index and BRIX is determined by the International Commission for Uniform Methods of Sugar Analysis (ICUMSA). Samples that have multiple ingredients such as salts and sugars (liquid sauces like soya sauce) can be measured for total percentage concentration of these substances using refractive index. The value can be expressed as BRIX or refractive index and the BRIX value can be read and interpreted as the total concentration of dissolved materials in the sample.

In general, though, Refractive Index measurement is a non-selective technique, because different solutions can have the same Refractive Index, is a fundamental property, and can be used to measure purity and concentration. Finally, Refractometers can be used to determine the concentration of solutions such as oil-based solutions, pharmaceuticals and heavy chemicals such as machining oil, cleaning solutions, glycols, Cesium Chloride, Sodium Hydroxide and others. Each solution has a correlation value between BRIX or refractive index and the concentration of solute can be established by comparing refractive index or BRIX to known concentrations of the solute using a standard curve. A conversion table can be established from the standard curve and several manufacturers of Refractometers input the conversion tables into the memory of their instruments and thus by measuring their refractive index can display concentration directly. In the market place, up to 80 different instruments are available with dedicated, fit for purpose concentration scales dedicated to specific end user needs and niches.

5.0 ASTM Standards

As well as several manufacturers offering quality control or calibration standards, dedicated for specific applications, there are several dedicated ASTM written standards available for various applications and industries. A selection of these are presented in Table 1.

ASTM Standards Dedicated for Various Applications

D542	**Index of Refraction of Transparent Organic Plastics**
D1045	Sampling and Testing Plasticizers Used in Plastics
D1218	Refractive Index and Refractive Dispersion of Hydrocarbon Liquids
D1416	Rubber from Synthetic Sources – Chemical Analysis
D1747	Refractive Index of Viscous Materials
D1807	Refractive Index and Specific Optical Dispersion of Electric Insulating Liquids
D3321	Use of Refractometer for Field Test Determination of the Freezing Point of Aqueous Engine Coolants
D4095	Use of the Refractometer for Determining Non-Volatile Matter (Total Solids) in Floor Polishes
D5006	Measurement of Fuel System Icing Inhibitors (Ether Type) in Aviation Fuels
D5775	Rubber from Synthetic Sources – Bound Styrene in SBR (styrene-butadiene copolymers)

Table 1

6.0 Why Use Refractive Index Standards

Understanding the theory of Refractive Index, the various instruments available and when and how to use these instruments is important. However, neither the test method or the instrument can be used in any meaningful way, without the availability of high-quality standards. From a metrological point of view, most workers have focussed on the use of RI standards for the calibration or re-calibration of the Refractometer. Few have focussed on an equally important, but much more widespread use of the standards as a control material for each test carried out in line with the principles of metrology and good laboratory practice. The advent of the ‘science of chemical metrology’ has given renewed emphasis to the use and necessity of control materials. The benefits of such controls cannot be overstated. As with all analytical standards or reference materials, RI standards should fulfil several criteria which are presented in Table 2.

Criteria That Refractive Standards Must Fulfill[1]

• **Provide traceability (needed to quote results in SI units)**
• **Demonstrate the accuracy of results**
• **Calibrate the equipment and methodology**
• **Monitor the user performance**
• **Validate the test**
• **Facilitate comparability, which is to ensure that when the correct procedures have been followed, the same analysis of the same materials will produce results that agree with each other whenever or wherever they are performed.**

Table 2

Such materials must also be able to fulfil the criteria required for quality control, accreditation and proficiency testing where appropriate. Standards and reference materials for RI should be produced and characterised in a technically competent matter, should be homogenous, stable, certified and have available a known uncertainty of measurement as a function of time. Production of primary standards rarely focuses on measurement uncertainty as a function of time that is realistic in the context of the production, transport and use of such standards under commercial or routine conditions. Such standards are rarely suitable for use as secondary or working standards.

Secondary or working standards need to be affordable, widely available and certified. The development, production, stability, assignation of uncertainty as a function of time and commercialisation of an extensive range of fit for purpose standards that includes refractive index standards has occupied the time and resources of the author's laboratory for several years[2]. In the context of normal commercial or routine use, RI standards or any other standards need to be rugged, have extensive shelf life, and be fit for purpose.

7.0 Applications of Refractive Index Measurement

In terms of markets and applications, these are wide and varied (see Table 3). They include the food industry, the manufacture and testing of both alcoholic and non-alcoholic beverages, a wide range of salts within the chemical industry, both in water based and petroleum-based chemicals. The measurement of Refractive Index has particularly wide application and relevance in the petrochemical industry, being relevant and useful for many types of products within that industry. Refractive Index is also very relevant in the sugar industry, both in processing of sugar beet and sugar cane.

Refractive Index, expressed in Brix units is a widely used measure of sugar content. In fact, as described earlier, ICUMSA, determines the relationship between Brix and sugar content and a whole host of Refractive Index and Brix Standards are made from the disaccharide sugar sucrose. Finally, the measurement of Refractive Index has huge relevance in the pharmaceutical, flavours, fragrances and perfume industries. For specific industries and applications within these industries, see Table 3.

Industrial Applications of RI or Brix

Industries	Applications
Food and Beverages	- Coffee Extract - Soy Bean Oils - Milk - Wine - Juice - Jam - Spirits
Sugar, Milling, Refining, Processing	- Molasses - Liquid Sugar - Cane Sugar Milling and Refining - Beetroot Sugar Milling and Refining
Water Based Chemicals	- Gels - Glycols - Resins - Polymers
Petroleum Based Chemicals	- Plastics - Solvents - Oil Based Plants - Resins
Petroleum Industry	- Additives - Lubricants - Fuel Oils - Paraffin, Waxes
Pharmaceutical Industry	- Toxicology Testing - Pharmacy Compounding - Drug Testing - EP and USP Test Methods
Flavour, Fragrance and Cosmetic	- Perfumes - Lemon Oil - Waxes - Creams

Table 3

8.0 Reagecon's Refractive Index and Brix Standards

Reagecon's standards fall into 4 different classifications (see Table's 4, 5, 6 and 7). To start with, we have Brix Standards which are comprised of ICUMSA compliant and ICUMSA non-compliant standards. Both categories are available in many different values, but differ in that the guidelines from ICUMSA, states that no preservatives can be added to Brix Standards, so as a result the ICUMSA compliant products have a shorter shelf life. Our range of ICUMSA compliant Brix Standards are also ISO 17034 accredited. Although these standards have a shelf life of 12 weeks as opposed to 6 weeks from our competitors, the non ICUMSA compliant standards have a shelf life of 12 months. All of the standards in both categories are expressed in Brix units.

Both of the other two categories of standards are expressed in Refractive Index Units. The first category is also sucrose based, have a twelve-month shelf life and are accredited to ISO 17025. The second category are solvent based Refractive Index standards, that have a higher upper range of up to 1.65808 units at 20°C and an extended shelf life of 24 months.

These products are accredited up to a Refractive Index value of 1.44193. Generally, the products are offered in 15ml dropper bottles that are offered singly or in a multipack of 6 bottles. The test result of both ranges is verified using a high-performance refractometer and the test is controlled using Certified Reference Materials. Customised pack options may be available upon request.

There are many good producers of RI standards, which are available on the market. Those available from Reagecon are presented here, simply, because these are the products that are most familiar to the author. We do believe, though, that the Reagecon list is comprehensive in terms of offering and specification and serve as a valuable template, to enable an analyst to know what to look for, when sourcing such standards in the marketplace.

Sucrose Standards (ICUMSA Compliant)
• **Primary level refractive index standards**
• **Values and temperature: 0, 5, 7, 10, 11.2, 11.5, 12, 12.5, 15, 20, 25, 30, 35, 40, 45, 50, 55 and 60% Brix @ 20C. *14.9, 19.4 and 23.8% Brix @ 20C also available**
• **Manufactured acc. to ICUMSA (International Commission for Uniform Methods of Sugar Analysis)**
• **Manufactured and certified in accordance with ISO 17034 requirements**
• **Extended shelf life of 12 weeks (competitors 6 weeks)**
• **Available in packs of 1 or 6 x 15ml dropper bottles**

Table 4

Brix Standards (Non- ICUMSA Compliant with Preservative)
• The most extensive range available in the market place
• Values and temperature: 0, 5, 7, 10, 11.2, 11.5, 12, 12.5, 15, 20, 25, 30, 35, 40, 45, 50, 55, 60 and 67.5% Brix @ 20C.
• Range with preservative and 12 months shelf life
• Tested and certified to ISO 17025 (values up to 60%)
• Available in packs of 1 or 6 x 15ml dropper bottles

Table 5

Refractive Index Standards – Sucrose Based
• Extensive range covers 1.34325 to 1.44193nD @ 20C
• Uncertainty of measurement up to ±0.00014nD
• Shelf life of 12 months
• Tested and certified ISO 17025 for the full range
• Available in packs of 1 or 6 x 15ml dropper bottles

Table 6

Refractive Index Standards – Solvent Based
• Extensive range covers 1.38779 to 1.65808nD @ 20C
• Uncertainty of measurement up to ±0.00014nD
• Shelf life of 24 months
• Tested and certified ISO 17025 up to 1.44193nD
• Available in packs of 1 or 6 x 15ml dropper bottles

Table 7

9.0 Features and Benefits of Reagecon's Standards

A summary of the features and benefits of Reagecon's Standards can be seen in Table 8.

Features and Benefits of Reagecon's Refractive Index Standards				
Range	Brix ICUMSA	Brix Non-ICUMSA	Refractive Index Sugar Based	Refractive Index Solvent Based
Accreditations	ISO 17025 for the whole range. ISO 17034 for Sucrose in Water (% Brix)	ISO 17025 up to 60% Brix	ISO 17025 for the whole range	ISO 17025 up to 1.44193$_n$D
Shelf-Life	12 weeks (6 weeks from competitors)	12 months	12 months	24 months
Traceability	Traceable to International Standards			
Applications	Suitable for use on any brand and type of refractometer			

Table 8

10.0 Conclusion

The measurement of refractive index is carried out in a wide range of industries as detailed in Section 3.0 of this publication. In such instances, critical decisions are made based upon these RI readings and so it is essential that analysts not only achieve the correct RI test results, but also prove the validity of these results.

Ultimately the provision of the correct result and proof of that correctness, is a direct function of the choice, quality and fitness for purpose of the standards used. The overall objective of this paper is to offer guidance on making the correct choice.

11.0 Bibliography

1. Barron, J., (1996) "Turbidity Standards and Reference Materials", Self-published, Reagcon Technical Paper, Available from the Reagecon Knowledge Site

2. Barron. J., Papacosta. K., (1997). Poster presentation, Pittcon Conference, New Orleans, USA