FoodTrack

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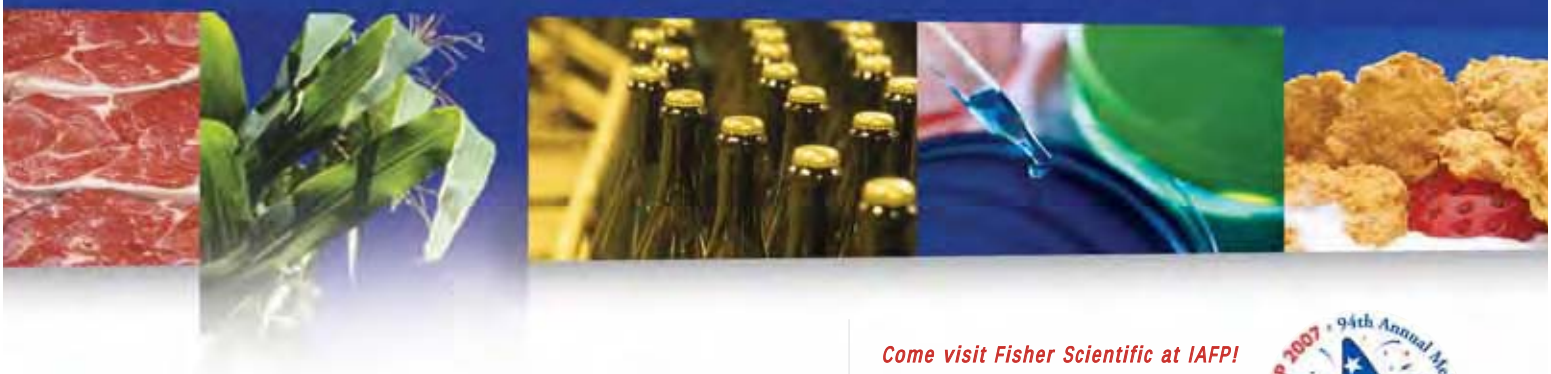

HIGHLIGHTS

- An ASTM-recognized electrode for measuring pHe in ethanol
- The world's smallest digital refractometer from Reichert
- Pathogen testing in fish—the importance of sample preparation

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pH Electrode for Determination of pHe of Ethanol

Thermo SCIENTIFIC

By Gayle Gleichauf, Thermo Scientific

Legislation and state-mandated programs such as EPAct, the Reformulated Gas Program, VEETC, and Renewable Fuel Standards are driving new demand for biofuels and alternate MTBE oxygenates. The specifications for fuel ethanol and denatured fuel ethanol for blending with gasolines are given in ASTM standards D5798 and D4806, respectively.

One specified measure of ethanol quality is its pHe value, as determined by ASTM standard D6423, the Standard Test Method for Determination of pHe of Ethanol, Denatured Fuel Ethanol, and Fuel Ethanol. The key to pHe measurement is the Thermo Scientific Orion Ross Sure-Flow combination pH electrode 8172BN (Fisher Cat. No. 13-642-243), which is the required electrode for the method.

Measuring Acid Strength

The ASTM D6423 method is a procedure to measure the relative acid strength of high ethanol content fuels containing about 70% or more ethanol. Acid strength is determined by measuring the pHe of fuel, which is similar but not directly comparable to the pH of a water solution. pHe is defined as "a measure of the acid strength of alcohol fuels defined by this apparatus and procedure."

Additionally, pHe is a good indicator of the corrosion potential of ethanol fuel. It is preferable to a total acidity measurement, which does not accurately account for different acid strengths. The pHe value will vary according to the fuel blend, stirring rate, and the amount of time the pH electrode is in the fuel.

Standard Electrode for pHe

ASTM has standardized on the use of the Thermo Scientific Orion Ross 8172BN pH electrode for pHe measurement, designating it as the necessary component for producing test results that are comparable from laboratory to laboratory. According to the standard, "Because the measurement is (of necessity) not made at equilibrium, it is essential that this exact electrode be used to ensure the reproducibility of results. Other electrodes (even those of similar design) will likely give different results under some or all conditions due to the use of a different size or type of glass membrane for the pH electrode, a different type of salt bridge junction, or other small differences, which may affect their nonequilibrium response."

Measurements in nonaqueous or low-water samples, such as high ethanol content fuels, typically lead to difficulties with unstable readings, long response times, and measurement errors. These symptoms are indicative of high sample resistance, bulb dehydration, contamination, and large, unstable junction potentials.

pHe is not quite the same as pH

Continued on back cover

Pocket Digital Refractometers Meet Diverse Application Needs in the Food Industry

Reichert Analytical Instruments

By Charles Smith, Reichert, Inc.

r² mini

Refractometers are increasingly popular quality-assurance tools in the food, beverage, ingredients, and flavors industries. They are used to determine the integrity and purity of raw materials that make up finished goods. They are also used to determine the concentration of dissolved solids in a solution. Refractometers ensure good, consistent quality of product and, more importantly, they help to control production costs, thus saving money and maximizing profits.

New Advances

The latest advances in these analytical instruments have improved the bottom line for the food industry. For nearly a century, traditional handheld (portable) refractometers relied entirely upon operator interpretation of an often "fuzzy" shadowline to obtain a concentration value for a given sample. The development of automatic digital handheld refractometers has greatly reduced operator error in this regard.

The *r²mini* Pocket Digital

As greater demands are placed upon the industry, production and quality-control managers are looking for a refractometer that offers even more capability, flexibility and value. The Reichert *r²mini* pocket digital refractometer was developed to meet these challenging requirements. The world's smallest, multiple-scale refractometer, it offers convenience and optimum performance. With one instrument, the operator can measure either the refractive index of a substance or the percent concentration expressed in Brix (sucrose).

What is refractive index? The refractive index is simply a physical property of a material, such as weight. It is, essentially, a measurement of the speed of light going through a sample. If the components that make up the sample are each slightly different, then the speed of light through that sample will change as well. Refractive index has also been described as the "unique fingerprint" of a sample.

The *r²mini* Is Used for a Variety of Applications:

Sugar and Sweetener Testing. Sugar and sweetener content is often the most critical, and expensive, component of food and beverage products. Maintaining proper concentration ensures consistent quality and reduces costs. Products are routinely monitored from the raw material stage, through production, to the final product. Brix is a sugar scale used to determine the total concentration of dissolved solids. Specifically, it is designed to measure sucrose content in a solution, but it is also used as a relative measurement of concentration of other types of sweeteners, such as High-Fructose Corn Syrups (HFCS). Measuring the concentration of dissolved solids with the *r²mini* refractometer will ensure that production yields are extended and profits are maintained. Sweetener is typically sold in a concentrated state. From that, the producer will reconstitute the sweetener into a lower concentration as part of the production of the food or beverage.

One problem that is frequently encountered during measurement is that food and beverage samples are often highly colored, and even opaque. Liquid sugars are often viscous. Certain samples such as fruit juices and concentrates, jams, and jellies have undissolved materials, such as pulp, which can affect the reading by blocking light in a manual-style handheld refractometer. The *r²mini* addresses this problem by measuring with reflected light, so the samples can be of any color or even opaque.

Flavorings, Oils, and Food Additives (Vitamins, Whey, Solidifiers, Emulsifiers). These components that make up a final product must be critically monitored and controlled. This is achieved by measuring the refractive index, to ultimately determine the purity of the product. If the measurement is slightly different than the expected and known value, then the product has been adulterated.

Fruit and Vegetable Ripeness. Many agricultural products are harvested when it is determined that a specific ripeness has been achieved. This ripeness is determined by taking the natural juices/sugars from the product and testing the Brix level (% solids). Wine producers know that it is time to harvest grapes when they reach a certain Brix or sugar level.

This is important, as the sugar level plays a major role in the fermentation process, impacting final product quality and consumer acceptance. The *r²mini* refractometer is portable and can be taken out into the field to do these measurements. It is rugged enough for factory use as well. For additional protection, it is IP65 rated for water resistance and is dustproof.

Biofuels. Biofuels are derived from biomass, or recently living organisms. They are a renewable energy source, unlike other resources such as petroleum and coal. Agricultural products grown for biofuel production include corn, soybeans, flaxseed, sugarcane, and palm oil. Vegetable oil is used to create biodiesel, while sugarcane and corn are used to create ethanol fuels. Ethanol fuel is the most common biofuel and manufacturers are now providing engines that can operate from it. Refractive Index measurements are important for testing crops during harvest, identifying for purity, and ongoing testing for monitoring of chemical reactions of biofuels in the production stage.

In Brazil, sugarcane is the predominant agricultural source for creating ethanol fuel; while in the U.S., corn has become the crop of choice. A recent issue of concern is the competition between the biofuel and food industries over these agricultural products. Because of rising demand and short supply, increased costs have been seen in both sugarcane and corn. Therefore, on both sides, the producers that purchase products derived from these crops want to know the purity of their purchase. A refractive index measurement can quickly determine the quality of the raw material and verify that the cost is justified. Ideal for this application, the *r²mini* measures both refractive index and % Brix.

An Indispensable Tool

With their many uses, pocket digital refractometers have become an indispensable tool in today's ever-changing food and beverage industry. Vital measurements such as refractive index and % Brix can now be performed quicker and easier, resulting in consistent quality of products and lower overall production costs.

Microbial Analysis and Isolation of Pathogens in Fish

brinkmann

For obvious consumer safety and health reasons, microbial analysis and isolation of pathogens is one of the most common quality-control measures employed in the food processing and packaging industry. Of particular importance is the sample preparation and homogenizing equipment used to prepare food samples for analysis. This article outlines the appropriate laboratory equipment, instrumentation, reagents, and methods required to effectively isolate, characterize, and identify *E. coli*, coliforms, *Listeria spp., Staphylococcus*, *Salmonella*, yeast, and mold.

Sample Preparation/Tissue Homogenization

This critical component of microbial analysis and isolation of select pathogens requires the use of robust homogenization and blending apparatus to ensure the complete breakdown of tissue and cellular material. Just as Enrichment (incubation) and Plating or Detection are considered vital to the process of food testing, the sample preparation and tissue homogenization process should be carefully considered as well. Raw samples of blue catfish were taken and analyzed for the presence of pathogens after 0, 2, 4, and 6 weeks of storage. A 10g representative sample was taken from the loin muscle of the fish (the thickest part of muscle) to prepare serial dilutions (10^{-1} – 10^{-3}) using sterile water as a diluent. The samples were homogenized for 60 seconds using a Seward Stomacher Lab Blender (Fisher Cat. No. 14-285-29). Total plate count, coliforms, *E. coli*, *Staphylococcus*, *Salmonella*, yeast and mold counts were then determined by the Grid-Membrane Filtration method (GMFM) (Peterkin et al., 1998). Next, the samples were homogenized (in 10mL volumes) using a Polytron Homogenizer (05-400-261) and a Brinkmann 7mm Easy Care Generator and passed through a 0.45mm grid membrane filter. After that, the filter was placed on a plate with media and incubated. *Listeria spp.* was determined by a qualitative method and the enrichment step was done with 10g of the sample added to 100mL of Demi Fraser broth. The solution was incubated at 30°C for 24 hours, followed by plating (0.1mL) in selective and differential media, ALOA (Agar Listeria Ottaviani & Agosti) at 37°C for 24 hours. ALOA is a prepared selective and differential medium for the isolation of *Listeria spp.* from foods for presumptive identification of *L. monocytogenes*.

Experimental Conditions

The differential activity is due to the presence in the medium of the chromogenic compound X-glucoside as a substrate for the detection of betaglucosidase enzyme, common to all *Listeria* species. The specificity is obtained by detecting the metabolism of a substrate by an enzyme (phospholipase) that is only present in the *L. monocytogenes* species. The combination of both subtracts allows the differentiation of *Listeria spp.* "non-monocytogenes" which develop blue colonies, from *Listeria monocytogenes*, which are surrounded by an opaque halo. ALOA allows differentiation of *L. monocytogenes*, even in the presence of a competitive flora (Microbiology International Co., 2000). Presumptive *Listeria* colonies from ALOA agar were identified using a gram staining technique followed by an API *Listeria* test (bioMerieux). The inoculum was prepared by suspending a few well-isolated colonies in 2mL suspension medium and the strips were placed in the incubation boxes with 3mL of distilled water. After that, 50C of bacterial suspension was distributed into the tubes and the incubation box was closed and incubated at 35ºC for 24 hours. Isolation, characterization and identification of *E. coli* and coliforms were done by using m-ColiBlue24 broth and m-Green YM broth for yeast and mold. For total plate count, *Staphylococcus* and *Salmonella* tests, m-plate count broth, m-*Staphylococcus* broth and MacConkey broth, respectively, were used. The Total Aerobic Plate Count (APC) plates were incubated at 37ºC for 48h, and the plates with *Staphylococcus* and *Salmonella* cultures were incubated at 35ºC for 48 hours. The yeast and mold cultures were also incubated for 48 hours at room temperature. Following incubation, the colonies were counted and data reported as a log CFU/g (colony forming units). Triplicate experiments were conducted, and each dilution was plated in duplicate.

Conclusion

Listeria, a well-known threat to consumer health and safety, is recognized as one of the primary pathogens contributing to food contamination. Proper sample preparation and tissue homogenization are the first step, and a key component, of microbial analysis and isolation of pathogens.

For information about Seward and Polytron sample preparation and tissue homogenization products, visit **www.fishersci.com** or contact your Fisher Scientific Sales Representative.

Courtesy of Brinkmann Instruments.

pH Electrode for Determination of pHe of Ethanol

Thermo SCIENTIFIC

Continued from front cover

Advantages of the 8172BN

The 8172BN electrode has many features that make it ideal for the pHe measurement of acid strength in high ethanol content fuels. First, the pH electrode membrane is constructed of a low-resistance glass.

This reduces unstable readings and drift caused by the higher resistance (lower conductivity) of the sample solution. Secondly, the glass body is resistant to attack from solvents. The standard test method calls for regular immersion in aqueous buffer to control dehydration of the glass-sensing membrane.

The refillable Sure-Flow reference prevents memory effects and is easily cleaned by briefly depressing the cap to flush the junction. The double junction prevents intrusion of the solvent into the reference electrode chamber, preventing bias in results due to contamination. The uniform flowrate of the junction provides a more stable junction potential. All of these advantages ensure that the Thermo Scientific Orion Ross Sure-Flow combination pH electrode (8172BN) is the electrode of choice for ASTM standard D6423, the standard test method for the determination of pHe of ethanol.

References:

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C. Clark Westcott. pH Measurements, Academic Press, 1978.

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For information about this electrode, visit **www.fishersci.com** or contact your Fisher Scientific Sales Representative.

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