Complete Proximate Analysis for Food Manufacturers

Abstract

Food manufacturing is an increasingly high-tech industry, with an ultimate focus on efficiency, throughput, scale, and automation. Food testing methods have lagged behind other technological improvements and, as a result, have become the bottleneck in the manufacturing process. This application note provides a background on historical test methods, and their modern alternatives which, in most cases, are not only faster and less labor intensive, but also provide more reliable results.

Introduction

There are many ways to measure components such as protein, moisture, fat, and ash in food samples. Traditionally, each component has been measured by a reference method that requires hours to perform. In recent decades, rapid alternatives such as near-infrared spectroscopy (NIR), bench top, time-domain nuclear magnetic resonance (TD-NMR), and even x-ray spectroscopy have gained widespread acceptance. This application note discusses the proximate analysis techniques that have traditionally been used, plus their rapid alternatives, with focus on special considerations for food manufacturers.

Crude Fat Analysis

For crude fat analysis, gravimetric wet chemistry techniques such as Soxhlet extraction and acid/base hydrolysis are commonplace. For this type of analysis, a sample is weighed, then placed in an apparatus where solvent can be used to dissolve and separate fat from non-fat components.

The fat-solvent mixture is then removed and the solvent is evaporated, typically with the aid of a heat source. The weight of the fat extract is compared to the initial weight of the sample and the crude fat content is calculated. Due to the nature of chemical extraction, and given the fact that the extract is assumed to be fat without being directly analyzed, it is critical that food manufacturers select the right extraction conditions for a particular sample matrix. A method that properly extracts fat for one sample type may over- or under-extract for another sample type. This is a significant potential source of error, especially for products such as processed foods that typically contain a wide variety of ingredients from many different plant and animal sources. Rapid alternatives such as time-domain nuclear magnetic resonance (TD-NMR) have been commercially available for decades, but most still require matrix-specific calibration which can become time consuming for manufacturers who regularly change formulations. The ORACLE™ universal fat analyzer is a unique bench-top NMR instrument that enables fat analysis of any sample, regardless of matrix, without calibration or method development. Unlike previous NMR fat analyzers, the ORACLE completely isolates and normalizes the fat signal from all other components, providing a uniform response that is not affected by the sample matrix. **Table 1** and **Table 2** show a direct comparison of results gathered on the ORACLE vs. NIR.

The average difference between the ORACLE and reference values was 0.03%, whereas the NIR exhibited a difference of 0.36%. Because the ORACLE does not require complex calibration and is not affected by optical properties such as color or texture, there are significantly fewer sources of error.

Table 1. ORACLE Fat Results Compared to Reference Values

Sample Type	Reference Value (AOAC 960.39)	ORACLE (AOAC 2008.06)	Difference (%)
Beef	26.56	26.55	0.01
Pork	22.30	22.30	0.00
Chicken	2.91	2.88	0.03
Turkey	1.00	1.03	0.03
Hot Dog	29.79	29.85	0.06

Table 2. NIR Fat Results Compared to Reference Values

Sample Type	Reference Value (AOAC 960.39)	NIR (AOAC 2007.04)	Difference (%)
Beef	29.30	29.99	0.69
Pork	22.25	21.99	0.26
Chicken	3.17	3.25	0.08
Turkey	1.48	1.89	0.41
Hot Dog	15.39	15.05	0.34

Moisture Analysis

For moisture analysis, loss-on-drying has traditionally been conducted in an air oven. The general principle is that samples are weighed before and after drying. The difference in weight, or loss-on-drying, is calculated as moisture content. With the right drying conditions, results can be very reliable, but test times can range anywhere from a few hours, to as much as 24 hours. The biggest risk with loss-on-drying is either under-drying the sample with insufficient time and temperature, or over-drying the sample which will cause non-moisture components to volatilize. In either case, the result is erroneous data which can lead to improper changes in formulations, or the need for a re-test which can delay production significantly.

Rapid alternatives for moisture analysis can be broken down into two main categories: direct and indirect. For direct rapid alternatives, moisture balances are commonplace. Moisture balances typically employ an infrared heat source and an integrated balance to directly monitor loss-on-drying. Indirect methods such as NIR and microwave spectroscopy rely on spectroscopic bands to correlate a signal to moisture.

NIR technology can provide results in less than a minute, but require constant calibration to maintain proper accuracy, so users need to keep reference method capabilities, such as an air-oven, or send regular samples to outside labs which tends to be costly.

The SMART Q™ and SMART 6™ moisture analyzers use a unique combination of technologies to reduce test times from four to eight hours in an air oven down to five minutes or less, while maintaining the same levels of accuracy and precision compared to an air oven. Other rapid analyzers take as long as 20 minutes to perform the same analysis, with added time necessary for cavity preheat or cool-down, which can bring total analysis time up to 30 minutes or more. The SMART Q is a direct loss-on-drying moisture analyzer that uses quartz-halogen infrared energy to rapidly dry samples. The SMART 6 uses a combination of quartz-halogen and microwave energy to provide even faster drying, with total test times of approximately two to three minutes. Both the SMART Q and the SMART 6 use optical temperature sensors to monitor the temperature of the sample and adjust power input accordingly. Other moisture balances use a simple thermocouple to measure the temperature of the air inside the cavity, which requires slower temperature ramps to avoid scorching or under drying samples. **Table 3** compares the results from a SMART Q, SMART 6, and air oven. **Figure 1** is a graphical representation of the data contained in **Table 3**, illustrating the accuracy and reproducibility of the SMART-series analyzers compared to reference results.

Table 3. Percent Moisture Comparison of Drying Methods for Snack Foods

Sample	Air Oven		SMART Q		SMART 6	
	Average	STDEV	Average	STDEV	Average	STDEV
Hot Cheetos	1.60	0.13	1.65	0.13	1.58	0.05
Ritz	2.67	0.05	2.67	0.09	2.60	0.06
Veggie Chips	2.83	0.06	2.84	0.11	2.91	0.10
Corn Tortillas	2.84	0.08	2.88	0.05	2.77	0.09
Pretzels	3.77	0.10	3.73	0.15	3.72	0.11
Nilla Wafers	3.98	0.17	3.94	0.09	3.91	0.07
Saltines	4.55	0.04	4.49	0.20	4.57	0.06

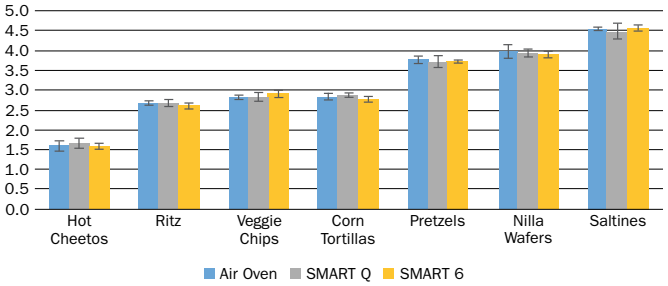

Figure 1. Percent Moisture Comparison of Drying Methods for Snack Foods

Protein Analysis

Protein content has traditionally been measured by the Kjeldahl method, which digests the sample and uses titration to determine nitrogen content. The underlying principle is that protein has a known amount of nitrogen, and this method relies on the assumption that the only major source of nitrogen in a sample is protein.

For food samples that do not have any non-protein nitrogen, Kjeldahl can yield satisfactory results, but it requires the use of hazardous reagents, takes hours to perform, and poses significant safety risks. Care must be taken with manufactured foods that contain non-protein nitrogen, which can be present in flavorings, stabilizers and curatives. Combustion analyzers are a rapid alternative that break samples down to their elemental form, then flow the combustion gas through a series of detectors to detect nitrogen, as well as hydrogen and carbon, depending on the instrument design. Unlike the aforementioned methods that convert nitrogen to a protein value, dye binding techniques, such as the Udy dye binding method and the Sprint® protein analyzer, utilize dye-binding technology and a colorimetric detector to bind and quantify protein directly. The Sprint protein analyzer uses a bright orange dye that becomes insoluble upon protein binding. At increasing protein concentrations, more dye becomes bound and falls out of solution, resulting in a decrease in absorbance of the dye. Because the Sprint measures actual protein and not nitrogen, it is impossible to generate false results due to non-protein nitrogen. **Table 4** is a comparison between the Sprint protein analyzer and Kjeldahl. The Sprint shows excellent repeatability with an average difference of less than 0.1% vs. Kjeldahl for meat samples.

Table 4. Comparison of Protein Content Between Sprint and Kjeldahl

Product	Sprint Replicates			Sprint Average	Kjeldahl Average	Diff.
	1	2	3			
Hot Dog	17.39	16.78	16.98	17.05	17.16	0.11
Sausage w Cheese	11.38	11.49	11.65	11.51	11.64	0.13
Sausage	15.47	15.39	15.42	15.43	15.31	0.12
Semi-dry Sausage	17.75	16.71	17.35	17.27	17.29	0.02
Beef Stick	28.27	27.82	27.61	27.90	28.08	0.18
Turkey Stick	34.01	33.67	34.04	33.91	33.89	0.02

The performance of the Sprint vs Kjeldahl for dairy samples is highlighted below, in **Table 5**. The average difference between the two methods is 0.04%.

Table 5. Accuracy of Sprint for Crude Protein in In-Process Yogurts, Permeates, and Retentates

	% Protein		
Sample	Sprint	Kjeldahl	Difference
Yogurt 1, In-Process	1.77	1.79	0.02
Yogurt 2, In-Process	2.49	2.47	0.02
Yogurt 3, In-Process	3.80	3.77	0.03
Yogurt 4, In-Process	4.21	4.33	0.09
Retentate 1	6.03	6.02	0.01
Retentate 2	9.54	9.64	0.10
Retentate 3	13.40	13.28	0.12
Permeate 1	0.18	0.20	0.02
Permeate 2	0.52	0.52	0.00
		Average	0.04

Bone Content Analysis

Bone content can be measured using a fairly wide variety of techniques, but the three most common methods all rely on the fact that bone is almost exclusively comprised of calcium. The first method is chemical titration, which utilizes an indicator dye system such as napthol blue and EDTA. The titration turns the dissolved sample from light pink to dark blue at the endpoint. To get calcium into solution, the bone-containing sample must be treated by boiling in hydrochloric acid, which poses considerable safety risks. Another method that requires the use of hazardous reagents is the determination of calcium via inductively coupled plasma (ICP) wherein the sample is digested in acid, then analyzed in an ICP instrument. Alternatively, bone content can be measured by ash analysis, wherein a sample is placed in a muffle furnace at a sufficient temperature to burn away all organic matter, leaving behind nothing but the mineral content (i.e. bone) of the sample. Ash analysis is straightforward, but can take eight hours or more to ash a sample in a traditional muffle furnace with ceramic crucibles. The Phoenix BLACK™ muffle furnace, when used in combination with quartz-fiber crucibles, optimizes airflow in and around the sample to reduce test time from eight hours to approximately 30 minutes. **Table 6** shows that percent bone, as determined via ash analysis, provides comparable results to the USDA bone method (calcium via ICP), but provides results in 30 minutes without the use of hazardous reagents.

Table 6. Bone Results for Mechanically Separated Chicken

MSC Sample	% Bone	USDA Bone	Difference
1	0.77	0.83	-0.06
2	0.70	0.76	-0.06
3	0.80	0.62	0.18
4	0.59	0.55	0.04
5	0.64	0.56	0.08
6	0.50	0.50	0.00
7	0.83	0.85	-0.02
8	0.85	0.88	-0.03

Multi-component Analyzers

While direct analysis is the best way to ensure the highest levels of accuracy and precision, there are times where speed and ease-of-use are a determining factor. There are a wide range of multi-component analyzers that utilize spectroscopy, such as near-infrared (NIR) Fourier-Transform Infrared (FT-IR) and even microwave spectroscopy. Spectroscopic instruments allow for very rapid analysis, but rely on unique signal profiles that must be calibrated to a primary reference method. Spectroscopic methods rely on complex optical trains that require constant re-calibration to maintain accuracy.

For food manufacturers interested in proximate analysis of raw and pre-blended meats, the ProFat™ meat analyzer uses well documented ratios of moisture to fat in meats to calculate fat content, based on direct moisture analysis. Once fat and moisture content are measured, the remaining protein and bone/ash levels can be determined. Altogether, the ProFat provides moisture, fat, protein, and ash values in approximately three minutes. **Table 7** shows a comparison of results from the ProFat compared to reference method values for moisture, fat, protein, and ash.

Conclusion

In the world of ever-increasing competition and the need for increasingly efficient production processes, the need to adopt faster, more accurate test methods has never been greater. With the right selection of rapid analysis methods, food manufacturers can reduce test times from hours or even days, down to minutes, not only speeding up test times, but enabling real-time feedback from the production line, which can be used for better process control.

Table 7. Accuracy of ProFat for Fat, Moisture, and Protein in Raw Meats and Blends

	% Fat			% Moisture			% Protein		
Sample	**ProFat**	**Soxhlet**	**Difference**	**ProFat**	**Oven**	**Difference**	**ProFat**	**Kjeldahl**	**Difference**
Beef, Fat Ground	29.04	29.22	0.18	55.02	54.93	0.09	14.91	14.91	0.00
Beef Ground	19.90	20.03	0.13	62.11	62.18	0.07	17.40	17.36	0.04
Beef, Lean Ground	28.42	28.38	0.04	54.04	53.98	0.06	16.34	16.33	0.01
Beef/Pork Blend	21.99	21.81	0.18	60.50	60.53	0.03	16.95	17.25	0.30
MSC, 11% Fat	11.14	11.11	0.03	70.46	70.49	0.03	17.18	17.13	0.05
MSC, 15% Fat	16.84	16.77	0.07	66.96	66.86	0.10	14.70	14.35	0.35
Pork, Ground	26.55	26.55	0.00	57.46	57.47	0.01	15.45	15.50	0.05
Turkey, 18% MDB	18.00	17.97	0.03	67.03	66.89	0.14	13.67	13.89	0.22

United States (Headquarters)
800-726-3331
704-821-7015
info@cem.com

France
33 (01) 69 35 57 80
info.fr@cem.com

Germany, Austria, Switzerland
(49) 2842-9644-0
info@cem.de

Ireland
+353 (0) 1 885 1752
info.ireland@cem.com

Italy
(39) 35-896224
info.srl@cem.com

Japan
+81-3-5793-8542
info@cemjapan.co.jp

United Kingdom
(44) 1280-822873
info.uk@cem.com

www.cem.com