

Optothermal Detection of Infrared Radiation-Induced Absorption in Aqueous Solutions of Carbohydrates: Lactose and Corn Starch

Jan P. Favier* and Dane Bicanic

Laser Photoacoustic Laboratory, Department of Agricultural Engineering and Physics, Wageningen Agricultural University, Bomenweg 4, NL 6703 HD Wageningen, The Netherlands

Otto Dóka

Department of Physics, Pannon University of Agricultural Sciences, Vár 2, Mosonmagyaróvár, Hungary

Mihai Chirtoc

Institute for Isotopic and Molecular Technology, P.O. Box 700, 3400 Cluj-Napoca 5, Romania

Per Helander

AB Varilab, Fullerstavägen 32, S-14144 Huddinge, Sweden

The optothermal window method, a variant of photoacoustic spectroscopy, was used at 10 μm wavelength to explore its feasibility for direct investigation of aqueous solutions of lactose and corn starch. Present limits of detection attainable by this method are 0.19% (w/w) for lactose solution and 0.6% (w/w) for corn starch gel, respectively.

Keywords: *Photoacoustics; infrared spectroscopy; nondestructive analysis*

INTRODUCTION

Chemical–physical analyses are becoming steadily more important when evaluating the quality of raw agricultural products and foodstuffs (Alexander and Zobel, 1992; Belitz and Grosch, 1987; Chang *et al.*, 1995). Most of these techniques not only require some kind of sample treatment before detection is attempted but are also time consuming. The need for a rapid analysis while maintaining simultaneously sample integrity has therefore stimulated development of non-destructive inspection methods, among which are also various spectroscopic techniques (Chang *et al.*, 1995; Osborne *et al.*, 1993). In particular, the infrared (IR) region, with specific absorption frequencies and their band intensities uniquely characterizing chemical compounds, was utilized for qualitative and quantitative studies of foods (Belton and Tanner, 1983; Belton *et al.*, 1987; Kochhar and Rossell, 1989; Wilson and Belton, 1988).

As many foodstuffs contain water, which has a strong absorption in the IR, accurate quantitative spectroscopic measurements on such samples are not trivial. The use of traditional transmission cells for studies of pasty or viscous samples also causes problems associated with filling and cleaning procedures. These were partially alleviated by the introduction of the attenuated total reflection technique (ATR) (Belton *et al.*, 1987).

This paper exploits the prospects of the new technique, *i.e.*, that of optothermal window (OW), used in combination with a CO₂ laser (emitting between 1100 and 900 cm⁻¹) for quantitative IR analysis of lactose and corn starch in water. A peculiar feature of this new

technique, when compared to other methods, is the fact that the temperature variations induced in the sample by the absorption of radiation are sensed outside the area of the sample irradiated by the excitation source. Corn starch and lactose were selected as test samples because of their important role in foods (Belitz and Grosch, 1987). Starch is a major food constituent of foods and serves as a model for many foodstuffs. In the infrared corn starch has strong absorptions at 1681, 1053–952, and 855 cm⁻¹ (Kochhar and Rossell, 1989). On the other hand, lactose is an important constituent of milk and dairy products.

A sample of real milk was also investigated to validate the feasibility of the OW technique. Infrared analysis of milk is currently performed by determining content of lactose (1042 cm⁻¹), protein (1549 cm⁻¹), and fat (1745 cm⁻¹) using dedicated filter instruments (Biggs, 1979; Luinge *et al.*, 1993).

All OW studies described here were carried out only at a few discrete wavelengths of the laser. However, the evaluation of results obtained in this experiment is important to estimate the prospects of a new, experimental technique based on a combined use of continuous tunable (powerful) infrared diode lasers as the radiation source and the OW concept. Finally, incorporating this new concept into that of FTIR spectrometry for quick scanning of spectra is worth considering.

BASIC THEORETICAL CONCEPTS OF THE OW METHOD

The OW technique is a variant of photoacoustic spectroscopy (Helander, 1993; Bicanic *et al.*, 1995). The heat, produced in a condensed phase sample by the absorption of modulated frequency f (s⁻¹) radiation, diffuses into the disk supporting the sample. Expansion of the disk generates an acoustic wave which is detected

* Author to whom correspondence should be addressed (fax 31-317-484819; telephone 31-317-484947/484666; e-mail, Jan.PaulFavier@user.aenf.wau.nl).

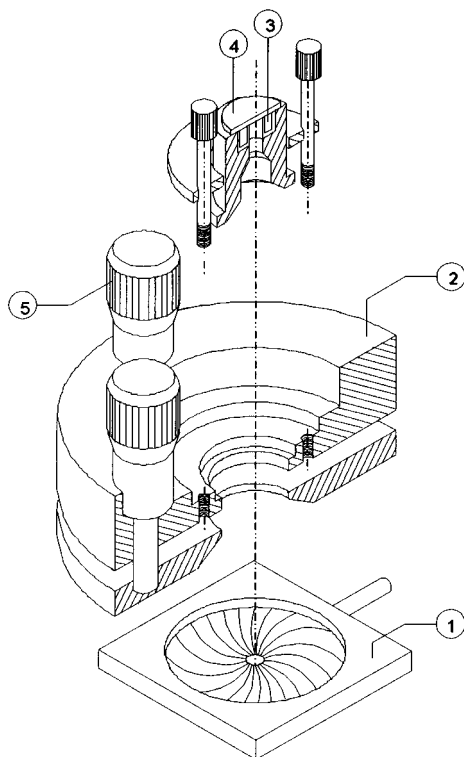


Figure 1. Heart of the experimental OW setup, displaying an adjustable diaphragm (1), gimbal mount (2), piezoelectric transducer (3), ZnSe disk (4), and adjusting micrometer screws (5).

by a piezoelectric transducer (PZT). A decisive parameter is the sample's thermal diffusion length (μ), defined as

$$\mu = \sqrt{\alpha/\pi f} \quad (1)$$

with α ($\text{m}^2 \text{s}^{-1}$) being the thermal diffusivity of the sample. Physically, μ is the distance across which the amplitude of a generated thermal wave is reduced to e^{-1} of its initial value. Only the heat originating from a layer of 1μ deep is detected by the PZT.

When the thickness of the sample is larger than its thermal diffusion length, the sample is termed as "thermally thick". For such a sample that, in addition, is in a good thermal contact with the disk, the normalized OW signal, $S(\beta)$ (Helander, 1993), is given by

$$S(\beta) = \frac{\sqrt{2\mu\beta}}{\sqrt{(\mu\beta)^2 + (\mu\beta + 2)^2}} \quad (2)$$

where β is the wavelength dependent absorption coefficient (m^{-1}) of the sample. In general, the OW signal decreases at higher frequencies; this is also true for the sensitivity of the PZT sensor. The absorption coefficient β of the sample can be obtained from eq 2 provided the OW cell is properly calibrated. In principle, recording the OW signal at varying wavelengths of incident laser radiation enables one to construct an absorption spectrum. The optical penetration depth must be larger than the thermal diffusion length, i.e., $\beta^{-1} > \mu$, if the OW spectrum is to coincide with the true absorption spectrum.

EXPERIMENTAL PROCEDURES

The OW experimental arrangement (Figure 1) comprised a

homemade c.w. CO_2 waveguide laser which was modulated (here 221 Hz) by a chopper (EG&G model 179). A 2 mm diameter diaphragm (1), was used to reduce the size of the unfocused laser beam, so it could pass from below (at a normal incidence) through a 20 mm diameter and 1.5 mm thick ZnSe disk (4) (Janos Technology, Inc.) with the annular PZT (3) (impedance 3 $\text{M}\Omega$ at 100 Hz) bonded to its bottom. To avoid Fabry-Perot resonances, the disk was provided with the antireflective coating on a side facing the incident laser beam. In order to prevent radiation striking the PZT directly, which would give rise to unwanted false OW signals, a reflective gold layer (5 μm thick) was deposited on the surface of the disk bonded to the PZT, in such a way as to leave a clear (uncoated), central circular area about 4 mm in diameter. A droplet of the sample (200 μL for lactose) or a small quantity of starch gel was deposited directly atop the disk. The strength of the preamplified OW signal (after current to voltage amplifier conversion) was measured by the lock-in amplifier (Stanford Research model SR850 DSP) and normalized to the incident intensity measured with a power detector (Spectra Physics model 407A) placed above the empty OW cell. The emission wavelength of the laser was verified by inserting a movable mirror to divert the laser radiation toward the spectrum analyzer.

The alignment of the OW cell (the disk and PZT assembly) was relatively easy to perform. With the empty OW cell in a gimbal mount (2) (Microcontrole), the lock-in signal was minimized (typically $0.7 \pm 0.4 \mu\text{V}$) by adjusting the micrometer (5) and maximizing the laser power throughput (typically 30–100 mW depending on the laser line) measured behind the cell. Both the alignment and the level of the OW signal were regularly checked between successive measurements as well as after completing the disk-cleaning procedure (simply wiping out the sample using cotton swabs and a paper towel).

RESULTS

Standard solutions varying from 2.5% to 20% (w/w) lactose (Merck CAS-No. 10039-26-6 7660) and from 2.5% to 10% (w/w) corn starch (Merck CAS-No. 9005-25-8 11686) were prepared by dilution in distilled water. The starch solutions were shaken vigorously after heating (30 s at 80 $^\circ\text{C}$) in a microwave oven; the gel was then formed on cooling down to room temperature.

Initially, the spectrum of 10% lactose was recorded with a FTIR spectrophotometer (Biorad BST-7, resolution 4 cm^{-1}) to determine appropriate excitation wavelengths of the CO_2 laser needed for OW studies. The maximal absorption of the lactose solution was found at 1041 cm^{-1} . The limit of detection (LOD), calculated as a ratio of 3 times the standard deviation obtained from the blank (water) and the slope of the fitted calibration curve, is 0.1% for this FTIR experiment. The spectra of 2.5% and 5% corn starch samples were recorded using a ZnSe ATR accessory. Difficulties were experienced when trying to evenly spread a layer of gel along the flat surface of the ATR crystal (the gel tends to fractionate easily). The FTIR-ATR spectra of the corn starch samples were only qualitative indicating the absorption peaks around 1053 cm^{-1} .

The calibration of the experimental setup was performed (Favier *et al.*, 1996) with distilled water because of its well-known optical and thermal properties. The 9P26 (1041 cm^{-1}) line of the CO_2 laser (nearly coinciding with the recommended analytical wavelength for lactose) was used for the calibration; the absorption coefficient β for water at this wavelength is $5.94 \times 10^4 \text{ m}^{-1}$ (Hale and Query, 1973). The calibration curve for lactose was linearized (eq 1), and the calculated absorption coefficients are shown in Figure 2; each data point is an average of four measurements.

The average regression coefficient of the calibration curve in Figure 2 is 0.9992, and the calculated LOD of

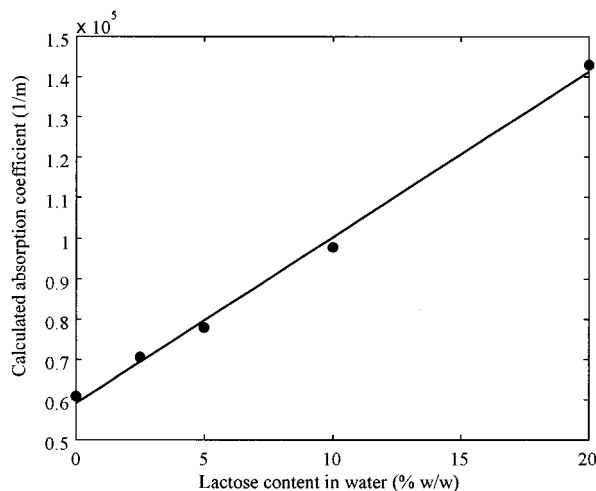


Figure 2. Absorption coefficient β for aqueous solutions containing a varying lactose content. The measurement was performed at the 9P26 (1041 cm^{-1}) line of the CO_2 laser.

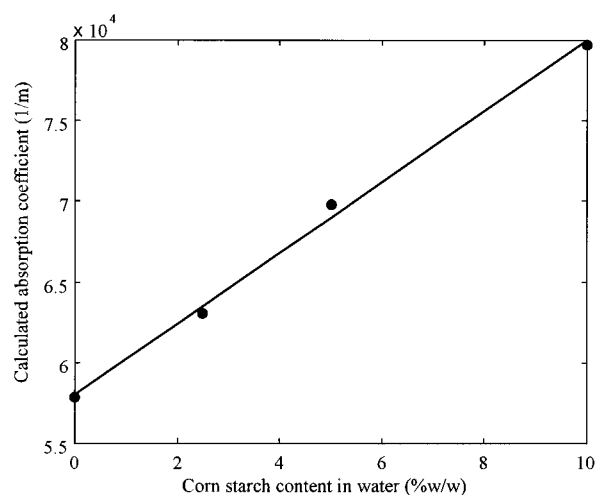


Figure 3. Absorption coefficient β for a varying content of corn starch in water. The measurement was performed at the 9P12 (1053 cm^{-1}) transition of the CO_2 laser.

lactose in water is 0.19% (w/w). The 0.4% relative error for distilled water (the ratio of the standard deviation $\sigma = 0.03$ to the normalized OW signal of 6.70) was higher than 0.2% ($\sigma = 0.01$ divided by 7.45) and 0.3% ($\sigma = 0.02$ divided by 7.89) obtained for 2.5% and 5% lactose solutions, respectively. The LOD of 0.19% (w/w) is substantially below the average lactose content found in milk. The straight line intercepts the y -axis at $5.92 \times 10^4\text{ m}^{-1}$ which is very close to the absorption coefficient of pure water at 1041 cm^{-1} . Finally, a commercial milk sample was used to test the feasibility of the OW technique. The 5.5% lactose content found in milk is 1% above the expected value, which is not surprising as no correction was made for the contribution to the OW signal due to absorption of protein and fat. For a comparison a FTIR apparatus is capable of 0.05–0.1% LOD for lactose (Luinge *et al.*, 1993) in milk.

The results of measurements on corn starch samples are shown in Figure 3; at the 9P12 (1053 cm^{-1}) line of the CO_2 laser, the LOD in water is 0.6% (w/w) and the regression coefficient is 0.999. The procedure in deriving these value was the same as described above for lactose. The intercept in Figure 3 gives $5.83 \times 10^4\text{ m}^{-1}$ (which corresponds well with the β of water at 1053 cm^{-1} (Hale and Querry, 1973)). The larger relative error (0.5–1%) for measurements of corn starch gels is ascribed to

the power instability of the laser. To check the reliability of the OW technique, the same samples were also examined at the 10P10 line of the CO_2 laser (953 cm^{-1}) where their absorption is expected to be negligible. The OW signal was found independent of the concentration, and the relative error was 0.6%. The difference between the OW signal strengths observed at 9P12 and 10P10 laser transitions is due to optical characteristics of the sample (gel) rather than to thermal ones.

CONCLUSIONS

The OW method with the CO_2 laser used here was used for quantitative and direct determination of lactose and corn starch contents in water. Presently attainable LOD's are 0.19% (w/w) for lactose and 0.6% (w/w) for corn starch gels. For the OW sensor described here, the physical condition (liquid or gel) of the sample is irrelevant, since a good thermal contact between sample and sensor is the only condition to be met.

Apparently the FTIR method is slightly more sensitive for determination of lactose than OW, although the relative error (0.02%) is comparable to that of the OW method. For precise determination of lactose in milk, the OW is at present inadequate due to the availability of only a limited number of wavelengths which precludes the application of multiple linear regression. For corn starch gels only qualitative studies could be made with FTIR-ATR. The effect of gel fractionation was less pronounced in the OW approach because the area that must be covered is much smaller.

Contrary to ATR and the enzymatic methods (used to determine incorporated starch), which both are very expensive (Biochemical Analysis), the low-cost OW device is easy to clean and moreover offers the possibility for on-line studies of optically opaque and thermally thick samples that are otherwise not accessible by other techniques.

At present, due to a limited tunability of the CO_2 laser, it is not possible to obtain the whole spectrum of the samples with the OW method. In the near future continuously tunable infrared diode lasers (gradually emerging on the market) are expected to provide power levels (strength of the OW signal is proportional to the power density) sufficient to generate an acceptable signal to noise ratio. Therefore, such a device when combined with the FTIR technique might be regarded as a new, candidate method for rapid analysis of realistic food samples throughout the entire infrared region. Additional potential practical applications of the OW method include the on-line control of processes (hydroponic growth) and the quality control in the food industry, for example, trans fatty acids content in margarine (Favier *et al.*, 1996) and phosphate sensor for soft drinks (Vonach *et al.*, 1995).

The performance of other thin IR transparent materials characterized by large values of thermal conductivity and thermal expansion coefficients might eventually surpass that of the ZnSe disk used in this experiment. Additional enhancement of the sensitivity is expected from a differential OW concept.

ACKNOWLEDGMENT

M.C. expresses gratitude to NWO (Dutch Organization for Scientific Research, The Hague) for a fellowship received. Credit is to B. van Veldhuizen (Department of Organic Chemistry, WAU), for advice and assistance

provided during FTIR experiments and to H. Boshoven for manufacturing components used in the experimental setup.

LITERATURE CITED

- Alexander, R. J.; Zobel, H. F. *Developments in Carbohydrate Chemistry*; American Association of Cereal Chemists: St. Paul, MN 1992.
- Belitz, H. D.; Grosch, W. *Food Chemistry*; Springer-Verlag: New York, Berlin, Heidelberg, 1987.
- Belton, P. S.; Tanner, S. F. Determination of the Moisture Content of Starch Using Near Infrared Photoacoustic Spectroscopy. *Analyst* **1983**, *108*, 591–596.
- Belton, P. S.; Saffa, A. M.; Wilson, R. H. Use of Fourier Transform Infrared Spectroscopy for Quantitative Analysis: A Comparative Study of Different Detection Methods. *Analyst* **1987**, *112*, 1117–1120.
- Bicanic, D.; Chirtoc, M.; Chirtoc, I.; Favier, J. P.; Helander, P. Photothermal Determination of Absorption Coefficients in Optically Dense Fluids: Application to Oleic Acid and Water at CO Laser Wavelength. *Appl. Spectrosc.* **1995**, *49*, 1485–1489.
- Biggs, D. A. Performance Specifications for Infrared Milk Analysis. *J. Assoc. Off. Anal. Chem.* **1979**, *62*, 1211–1214.
- Biochemical Analysis Food Analysis*; Boehringer Mannheim GmbH Biochemica: Germany 1986.
- Chang, S. K. C.; Holm, E.; Schwartz, J.; Rayas-Duarte, P. *Food. Anal. Chem.* **1995**, *67*, 127R–153R.
- Favier, J. P.; Bicanic, D.; Bovenkamp, P., van de; Chirtoc, M.; Helander, P. Detection of Total Trans Fatty Acids Content in Margarine: an Intercomparison Study of GLC, GLC + TLC, FT-IR and Photoacoustic Window (Open Photoacoustic Cell). *Anal. Chem.* **1996**, *68*, 729–733.
- Hale, G. M.; Querry, M. R. Optical Constants of Water in the 200-nm to 2000-nm Wavelength Region. *Appl. Optics* **1973**, *12*, 555–563.
- Helander, P. A Method for the Analysis of Photoacoustic and Photoacoustic Signals. *Meas. Sci. Technol.* **1993**, *4*, 178–85.
- Kochhar, S. P.; Rossell, J. B. Applications of Infrared Absorption Spectroscopy in Food Industry. *Spectroscopy* **1989**, *4*, 34–40.
- Luinge, H. J.; Hop, E.; Lutz, E. T. G.; Hemert, J. A.; Jong, E. A. M., de Determination of the Fat, Protein and Lactose Content of Milk using Fourier Transform Infrared Spectroscopy. *Anal. Chem. Acta* **1993**, *284*, 419–433.
- Osborne, B. G.; Fearn, T.; Hindle, P. H. *Practical NIR Spectroscopy with Applications in Food and Beverage Analysis*, 2nd ed.; Longman Scientific & Technical: Singapore, 1993.
- Vonach, R.; Kellner, R.; Lippitsch, M. A Phosphate Sensor for Soft-Drinks Based on IR Spectroscopy. Proceedings Eurofood VIII, Vienna, Austria, 1995.
- Wilson, R. H.; Belton, P. S. A Fourier-Transform Infrared Study of Wheat Starch Gels. *Carbohydr. Res.* **1988**, *180*, 339–344.

Received for review May 14, 1996. Revised manuscript received December 5, 1996. Accepted December 12, 1996.®

JF9603588

® Abstract published in *Advance ACS Abstracts*, February 1, 1997.