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Food Chemistry 96 (2006) 491–495
Food Chemistry
www.elsevier.com/locate/foodchem

Analytical, Nutritional and Clinical Methods

Nondestructive determination of water and protein in surimi by near-infrared spectroscopy

Musleh Uddin [a,*], Emiko Okazaki [a], Hideto Fukushima [a], Sandor Turza [b], Yamashita Yumiko [a], Yutaka Fukuda [a]

[a] *Division of Food Technology and Biochemistry, National Research Institute of Fisheries Science, 2-12-4 Fukuura, Kanazawa, Yokohama 236-8648, Japan*
[b] *Central Food Research Institute, Department of Analytics, H-1002 Budapest, Hungary*

Received 1 December 2004; received in revised form 9 March 2005; accepted 5 April 2005

Abstract

Nondestructive near-infrared spectroscopy (NIRS) between 400 and 1100 nm regions was employed directly on surimi using a surface interactance fibre optic accessory, to investigate the potential of NIRS as a fast method to determine water and protein contents. The reason why NIRS is well suited when assessing the presence of water or protein is due to the specificity of O–H and N–H bindings. At 980 nm only one broad peak in the original spectra can be seen due to the absorption of water since it contained nearly 80% of surimi. Predictive equations were developed using partial least squares (PLS) regression where excellent predictions for protein and water are noticed. Regression coefficients are higher than 0.98, errors are small and RPD value for protein is well over 8 and that for water is very close to it which can be used for any analytical purpose.

Keywords: Surimi; Protein; Water; NIRS; PLS

1. Introduction

"Surimi" is an intermediate fish product, used primarily to produce for the preparation of traditional gel food called "Kamaboko" and more recently used for the production of seafood analogs (fabricated food). It is defined as a refined fish protein product prepared by washing mechanically deboned fish to remove blood, lipids, enzymes and sarcoplasmic proteins, and as minced and washed fish flesh that has been stabilized by cryoprotectants (Vilhelmsson, 1997). Surimi technology has become one of the most important technologies in the area of the world food today. It gaining more prominence worldwide, because their protein quality, low fat content, and convenience for consumers. The surimi could be adjusted during mixing, to standard water content such as 78% as suggested by Lanier, Hart, and Martin (1991). This would require determination of water content of each lot of surimi prior to mixing which could cause a processing delay of up to 24 h. With surimi from fish such as arrowtooth flounder or pacific whiting, adding water alone would dilute the additive required to inhibit naturally occurring proteases (Porter, Koury, & Kudo, 1990). Additionally, for commercial application it may not always be feasible to adjust water to 78% since testable gels may not form with that much added water. Surimi gelation is associated with temperature-induced structural changes of protein; however, water which comprises about 73–80% of surimi plays a vital role in gel formation. Another key factor is optimum protein concentration for gel strength (Luo,

* Corresponding author. Tel./fax: +81 45 788 7664.
E-mail address: musleh@affrc.go.jp (M. Uddin).

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doi:10.1016/j.foodchem.2005.04.017

Kuwahara, Kaneniwa, Murata, & Yokoyama, 2001). To make a better gel, water and protein contents are critical; therefore, those are must need to adjust up to optimum level (Shindo, Ueshin, & Miki, 2000).

Using conventional methods such as oven drying or standard micro/macro-Kjeldahl procedure, determination of water and protein contents are laborious and time consuming. Near-infrared spectroscopy (NIRS) is nondestructive, fast and easy to implement where no reagents are required and no wastes are produce. Once calibrated, the NIRS is simple to operate. It is well suited for determining the major components of foods such as water, fat and protein (Osborne, Fearn, & Hindle, 1993). It has been widely used in the food industry, is based on the electromagnetic absorption of organic compounds (Misra, Mathur, & Bhatt, 2000; Svensson, Nielsen, & Bro, 2004; Uddin & Okazaki, 2004; Uddin, Ishizaki, Okazaki, & Tanaka, 2002). The reason why NIRS is well suited when assessing the presence of water or protein is due to the specificity of O–H and N–H bindings. The objective of the present study is to test if NIRS can be used to determine the water and protein contents of surimi. Previous studies have used NIRS to assess the protein and water content of whole fish with satisfactory results (Isaksson, Togersen, Iversen, & Hildrum, 1995; Pink, Naczk, & Pink, 1999). On that basis and due to the short sampling time, NIRS may be considered as a possible fast method for assessing water and protein contents in surimi and could be applied for on-line or at-line processing control.

2. Materials and methods

2.1. Surimi sample

Different lots of frozen SA grade walleye pollack *Theragra chalcogramma* surimi (Maruha Co., Tokyo, Japan) was thawed overnight at 5 °C. About 5–6 g of surimi was placed in a pre-weighed aluminum tray. The surimi was dried in an oven at 105 °C for 18 h. The percent weight of water was calculated from the difference between the beginning and ending mass of the surimi. Standard macro-Kjeldahl procedure (AOAC, 1990) was used for the determination of crude protein (CP) and the results are presented in Table 1.

Table 1
Characteristics of reference data for crude protein and water content in surimi

Constituent	*N*	Mean	SD	Maximum	Minimum	Units
Crude protein	110	13.96	1.35	16.17	11.98	g/100 g
Water	110	78.69	2.91	83.11	74.17	g/100 g

N, number of samples; SD, standard deviation.

2.2. NIR measurements

Surimi samples were scanned in the transmittance mode from 400 to 1100 nm in a scanning monocromator NIRSystems 6500 (NIRSystems, Silver Spring, MD, USA) at 2 nm spectral increments equipped with a surface interactance fibre optic accessory. The probe system consisted of two fibre-optic bundles, one for guiding monochromatic light to the probe head, and one for guiding the received light to a silicon detector. Within a 4 cm square probe face, seven quartz windows (1×20 mm) are fitted; windows are alternatively light exit ($n = 4$) and collection ($n = 3$) ports. Before spectra were measured on the fish, a reference spectrum was obtained by measuring the reflected radiation from a 5 cm diameter white ceramic plate. Excess surface water was removed from the surimi samples immediately prior to scanning by means of a paper tissue. Operation of the spectrophotometer and the collection of spectra were performed using the "VISION" software packages (Version 3.20, NIRSystems, MD, USA). The spectra were stored in optical density units $\log(1/T)$, where T represents the percent of energy transmitted. All of the subsequent operations were accomplished at 5 °C.

2.3. Multivariate-data analysis

Predictive equations were developed using partial least squares (PLS) regression. Leave-one-out cross validation (every calibration sample is a validation as well) was used to avoid over fitting of the equation. For CP multiplicative scatter correction (MSC) (Geladi, McDougall, & Martens, 1985) and Savitzky–Golay (SG) (Savitzky & Golay, 1964) second derivative with second order polynomial was used. The derivative window was set at 15 left and 15 right side points (30–30 nm) from the center point in the derivative window. For water the best results were achieved using SG second derivative with a second order polynomial and 8–8 left and right side points (16–16 nm). Spectral manipulations and PLS regressions were performed with the Unscrambler software (Version 8.05, Camo, NJ, USA). As an initial step, the 700–1098 nm wavelength range was used to establish calibrations and then this range was optimized using loading vectors of the regressions. Loading vectors were also used to interpret model structure on a factor to factor (latent variables in the model) basis. Calibration statistics included standard error of calibration (SEC), correlation coefficient (R), root mean square error of cross validation (RMSECV) and RPD. The optimum calibrations were selected on minimizing the RMSECV. Methods for PLS regression, validation and definitions for different errors measures can be found elsewhere (Martens & Naes, 1989; Naes, Isaksson, Fearn, & Davies, 2002).

3. Results and discussion

Fig. 1(a) shows the original near infrared spectra of all 110 surimi samples, while Fig. 1(b) displays the corresponding second derivative spectra of these samples. The original spectra look very smooth and have considerable baseline shifts (vertical offsets), and the only one broad peak can be seen at 980 nm, which is the absorption band for water (Downey, 1996; Osborne et al., 1993). To resolve broad peaks and eliminate baseline shifts second derivatives were used (Blazquez, Downey, O'Donnell, O'Callaghan, & Howard, 2004; Ding & Xu, 1999). This spectral treatment eliminated the offsets and made more peaks available to perform regressions as shown in Fig. 1(b). However, the parallel shifts of spectra seen in Fig. 1(a) still remains visible, especially at the 962 nm band (for second derivatives original positive peaks become negative), which is the same water band that appeared at 980 nm in the original spectra. This phenomenon is caused by scattering of samples. This scattering can be substantially reduced by, e.g., MSC as can be seen in Fig. 2(a) and (b). Now, the original spectra are spaced much closer to each other, for the variation due to scattering has been removed. Further evidence of that is clearly visible in Fig. 2(b), since the spectral variation at 962 nm has been greatly reduced. This removal can help achieve better calibrations in many instances (Davies & Grant, 1987; Steuer, Schulz, & Lager, 2001).

In our case, for crude protein, this spectral treatment helped reduce the model errors (SEC and RMSECV), thus increasing model reliability. On the other hand, for water calibrations MSC did not improve results, in fact with MSC they became a little worse. Regression results for crude protein and water are shown in Table 2. As can be seen results are excellent for crude protein and good for water. Regression coefficients are higher then 0.98, errors are small, and RPD value for CP is well over 8 and that for water is very close to it. This means that CP calibration for surimi can be used for any analytical purpose. As for water, the RPD is adequate for process control (Williams & Norris, 2001). The excellent fit of samples are again demonstrated in the scatter plots for CP and water as shown in Fig. 3 and 4. The samples

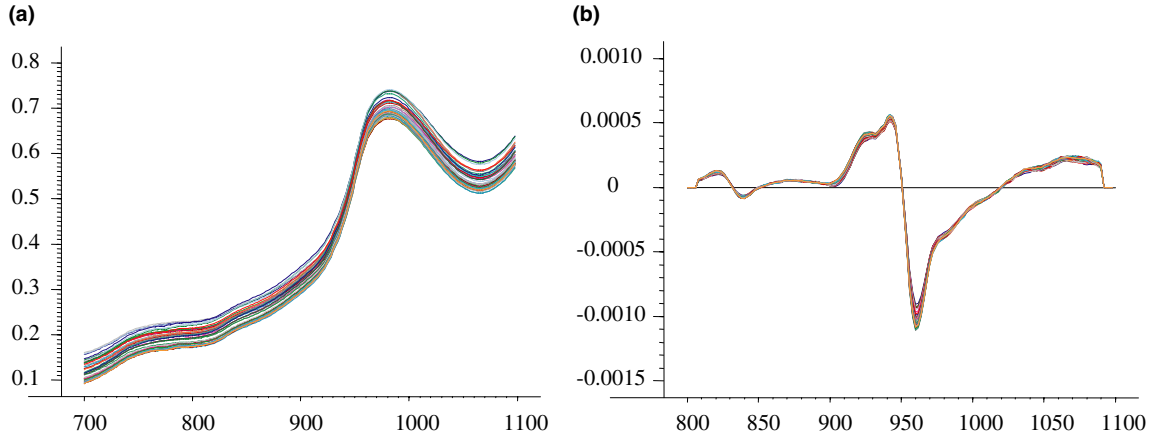

Fig. 1. Original transmission spectra of surimi samples in the 700–1098 nm wavelength range (a). Second derivative spectra of surimi samples in the 700–1098 nm wavelength range (b).

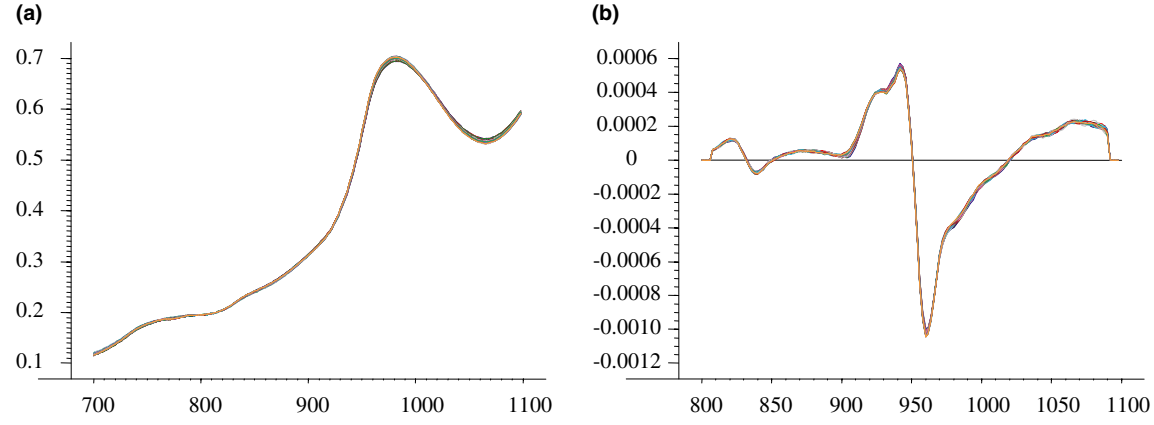

Fig. 2. MSC-treated spectra of surimi samples in the 700–1098 nm wavelength range (a). Second derivative of the MSC-treated surimi spectra in the 700–1098 wavelength range (b).

Table 2
Regression statistics for crude protein and water in surimi

Constituent	Spectral range (nm)	*N*	*F*	*R*	SEC (g/100 g)	RMSECV (g/100 g)	RPD
Crude protein	900–1100	110	4	0.982	0.11	0.13	10.38
Water	900–1100	110	4	0.978	0.30	0.38	7.63

F, number of factors in the model; *R*, multiple correlation coefficient; SEC: standard error of calibration; RMSECV, root mean square error of prediction; RPD, ratio of the standard deviation (SD) in the reference data for the validation set to the RMSECV.

(points) are very nicely positioned along the regression line.

As we have seen so far, the two models work wonderfully; however, it is also important to know why. Understanding model structure and finding wavelength regions relevant to the models are essential to find the best and most reliable solution. To do this loading vectors are analyzed for both models. Such vectors are displayed in Fig. 5 for CP. As we know (Table 2), four factors were needed for the model. These account for 99% and 99% of all spectral and chemical variability. Each factor explains a certain portion of the overall spectral and chemical information in the model. This information is given as percent explained variance for the spectral and chemical part of the model. Since, we have established a regression relationship between spectral

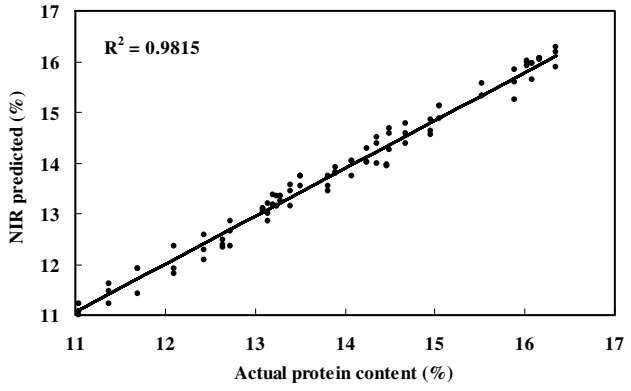

Fig. 3. Scatter plot for CP in surimi for the validation phase in the 900–1098 nm wavelength range. The range was determined by loading vector analysis.

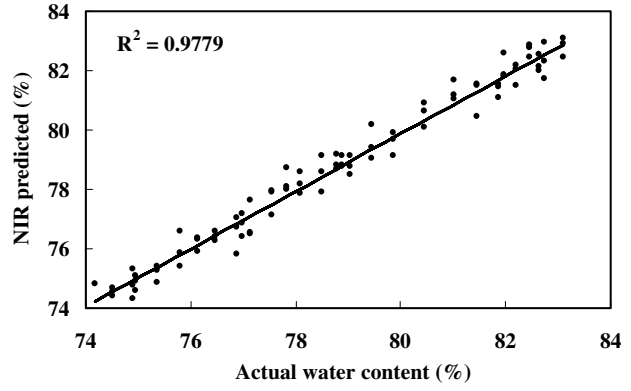

Fig. 4. Scatter plot for water in surimi for the validation phase in the 900–1098 nm wavelength range. The range was determined by loading vector analysis.

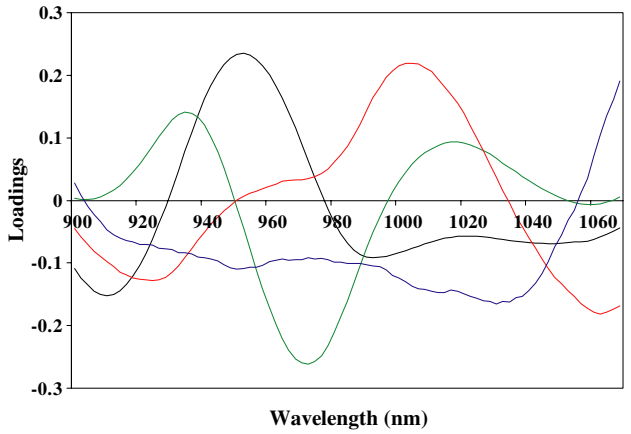

Fig. 5. Loading vectors for the CP model in the 900–1098 nm range. First factor (black line), second factor (green line), third factor (red line) and fourth factor (blue line).

data and chemical data (protein and water), explained variances are reported for spectral data and chemical data, respectively, for each model factor. By this, we can understand how much of the chemical information is explained by how much of the spectral information. This can be done on a factor-by-factor basis for every model. Earlier factors are more important to models as seen by the variances for the first and second factor. The first factor alone describes 69% and 80% of the model and the second one 27% and 13%, respectively, indicating they are the most relevant spectrally and chemically. The first factor (black line) has a negative peak at 910 nm, which is a protein absorption band (Osborne et al., 1993) and the second one (green line) has a peak at 970 nm where water has maximum absorption (Downey, 1996; Williams & Norris, 2001). This is quite understandable, since water and protein are major components of surimi. The remaining two factors are needed to improve results, though their contribution is much smaller, meaning that there are interactions and some other interfering "factors" which are modeled by those additional factors.

For water, on one hand, the situation is simpler, but on the other hand, it may be more complicated. The loading vectors in Fig. 6 show quite a different pattern and structure. Four factors describe the model a 100%, both spectrally and chemically, however, the first factor (black line) alone is responsible for 97% and 92% of all model variability. This factor has the most intensive

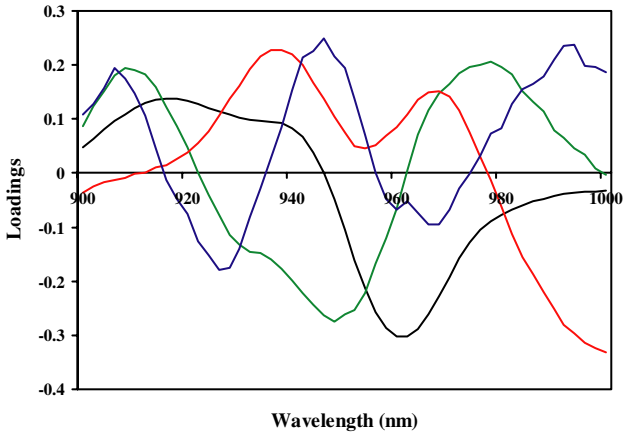

Fig. 6. Loading vectors for the water model in the 900–1000 nm range. First factor (black line), second factor (green line), third factor (red line) and fourth factor (blue line).

peak at 962 nm (Downey, 1996; Osborne et al., 1993; Williams & Norris, 2001), which is exactly the same position as described before for water (Fig. 1(b) and Fig. 2(b)). Therefore, this factor alone characterizes the model almost fully. This is logical, as the average water content is nearly 80%. The other factors are also relevant, because they possibly grab some information about water interactions through hydrogen bonding to other surimi components.

4. Conclusions

The study shows that NIRS is an obvious alternative to the time consuming conventional chemical analysis to determine the water and protein contents. An accuracy level of that obtained in the present work should have significant practical possibilities in the seafood industry.

Acknowledgements

The authors acknowledge Dr. Satoshi F. Noguchi and Dr. Yukio Asakura (Central Surimi Laboratory Co., Ltd.) for supplying surimi samples.

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