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## Application of visible and near infrared hyperspectral imaging for non-invasively measuring distribution of water-holding capacity in salmon flesh

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#### ABSTRACT

Water-holding capacity (WHC) is a primary quality determinant of salmon flesh. One of the limiting factors for not having a direct measurement of WHC for salmon quality grading is that current WHC measurements are destructive, time-consuming, and inefficient. In this study, two hyperspectral image systems operated in the visible and short-wave near infrared range (400-1000 nm) and the long-wave near infrared range (897-1753 nm) were applied for non-invasive determination of four WHC indices, namely percentage liquid loss (PLL), percentage water loss (PWL), percentage fat loss (PFL), and percentage water remained (PWR) of salmon flesh. Two calibration methods of partial least square regression (PLSR) and least-squares support vector machines (LS-SVM) were applied, respectively, to establish calibration models of WHC indices based on the spectral signatures of salmon flesh, and the performances of these two methods were compared to determine the optimal spectral calibration strategy. The performances were also compared between two hyperspectral image systems, when full range spectra were considered. Out of 121 wavelength variables, only thirteen (PLL), twelve (PWL), nine (PFL), and twelve variables (PWR) were selected as important variables by using competitive adaptive reweighted sampling (CARS) algorithm to reduce redundancy and collinearity of hyperspectral images. The CARS-PLSR combination was identified as the optimal method to calibrate the prediction models for WHC determination, resulting in good correlation coefficient of prediction ( $r_P$ ) of 0.941, 0.937, 0.815, and 0.970 for PLL, PWL, PFL, and PWR analysis, respectively. CARS-PLSR equations were obtained according to the regression coefficients of the CARS-PLSR models and were transferred to each pixel in the image for visualizing WHC indices in all portions of the salmon fillet. The overall results show that the laborious, time-consuming, and destructive traditional techniques could be replaced by hyperspectral imaging to provide a rapid and non-invasive measurement of WHC distribution in salmon flesh.

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#### 1. Introduction

Fresh Atlantic salmon is a frequently consumed seafood product worldwide. Eating salmon can measurably improve people's health. Nowadays, consumer's expectation of salmon products with high quality and safety has increased. Superior salmon products should be manufactured to conform to consumers' expectations, leading to the success in the highly competitive salmon industry. Salmon received by the process industries shows extreme diversity, because they are harvested from different farmers who follow different farm systems and deal with multiple

\* Corresponding author. Tel.: +353 1 7167342; fax: +353 1 7167493. *E-mail address*: dawen.sun@ucd.ie (D.-W. Sun). *URLS*: http://www.ucd.ie/refrig, http://www.ucd.ie/sun (D.-W. Sun). breeds. It is difficult for the process industries to produce products with consistent quality that is critical for grading based on expected salmon palatability. Inconsistencies in quality would lead to the decline in customer satisfaction and subsequently in market shares. Consumers are willing to pay more for salmon products if high quality is guaranteed.

The measurement of the water-holding capacity (WHC) of muscle is a useful and effective way for describing quality changes in food and food products [1]. WHC is defined as the ability of muscle to retain water or resist water loss [2] and is of great significance for commercial value and consumer acceptance. The WHC of muscle to retain water is affected by several factors such as proteins oxidation, proteolytic activity of tenderising enzymes, and cross-linking of myofibrillar proteins [3]. The change of WHC to a large extent affects the texture of fish. Fish flesh usually becomes tougher when accompanied by a reduction of WHC [4]. Poor WHC will result in high drip and purge loss and is now of







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significant industry concern [5]. Therefore, it is critical to determine the WHC of salmon products throughout the entire process and management system to keep the products with superior quality and to understand how the different processing regimes influence the quality of the end product. Currently, the determination of WHC is commonly conducted by laborious and destructive techniques, such as gravimetric method [6], centrifugation method [2], filter paper wetness method [7], and cooking loss method [8]. However, these conventional methods to determine WHC are extremely time-consuming, which is a limitation for industrial online inspection. Therefore, there is a need for a rapid and nondestructive WHC evaluation method under industrial environment and the incorporation of such a method with the existing salmon inspection systems will bring in great benefits to the industry for efficient salmon product quality assessment.

Optical techniques are one of the most accessible ways instead of the destructive and time-consuming sensory and instrumental methods for predicting quality of food rapidly and non-invasively. Of the various non-invasive optical methods, near-infrared (NIR) spectroscopy has shown its ability to measure WHC of fish flesh [9] and is a potential method for on-line implementation. Indeed, visible and near-infrared (Vis-NIR) spectroscopy has also been intensively applied in quantitative evaluation of other quality attributes of salmon products, such as fat, moisture, and sodium chloride [10,11]. However, values of WHC vary along heterogeneous materials, such as salmon products. It is a difficult task to obtain the overall distribution of WHC for the salmon flesh at different positions using spectroscopy technique because of its small sample area (limited spatial information). In addition, traditional destructive techniques also cannot provide the WHC details for salmon flesh, but only its bulk value. The lack of knowledge on the concentration gradients and spatial distributions of WHC has seriously limited the ability of the salmon industry to provide consumers with products of consistent quality and safety. On the other hand, imaging techniques (generally refer to computer vision [71–73]) in the forms of monochromatic or colour images provide abundant information of a sample at a pixel-wise level and has been used for inspecting food quality [12,13]. However, they have limited capacity of inspecting constituent properties such as moisture, fat and protein, as they only work in the visible spectral range.

As an extension of both spectroscopy and imaging techniques, hyperspectral imaging technique has been widely accepted as a smart and promising analytical tool in quality evaluation and assurance of various agricultural and food products [14,15]. It can provide both spectral and spatial information of an object simultaneously, making hyperspectral imaging very powerful and advantageous in food quality control and assessment than conventional spectroscopic and imaging techniques [16]. Hyperspectral image data are made up of a series of congruent three dimensional "hypercube" (x, y,  $\lambda$ ), which contains twodimensional spatial information (x, y) as well as one-dimensional spectral information ( $\lambda$ ). These data provide a large amount of information and can be analysed to characterize a specific object more objectively and reliably. Moreover, the obvious advantage of hyperspectral imaging is the ability of visualization of quality attributes distribution, which is not obtainable by spectroscopy or conventional destructive methods. Recently, hyperspectral imaging technique has received considerable applications for food quality and safety assessment [17,18], such as beef [19], pork [20], lamb [21], prawn [22], egg [74], citrus [23], and cucumber [24]. In particular, endeavours of using hyperspectral imaging have been reported for salmon quality evaluation in recent years, such as fat [25], colour [26], NaCl content [27], ice fraction [28], moisture [29], and microbial spoilage [30]. However, no reports of using hyperspectral imaging to determine WHC of salmon flesh have been found.

Given the limited information on the usefulness of hyperspectral imaging systems to determine WHC in fish or fish products, the main aim of this study was to investigate the feasibility of hyperspectral imaging system in the visible and near-infrared spectral region of 400–1753 nm for predicting WHC distribution of salmon fillets. The specific objectives of the current work were to (1) acquire hyperspectral images of salmon fillets in the spectral range of 400–1753 nm; (2) extract spectral information and identify the optimal wavelengths that are most correlated to WHC prediction; (3) build a quantitative relationship between the WHC values and spectral information; (4) compare the prediction abilities of spectral ranges of 400–1000 nm and 897–1753 nm, respectively; and (5) display the WHC distribution in the salmon fillets by developing an image processing algorithm for visualizing WHC in all pixels of the image.

#### 2. Materials and methods

#### 2.1. Samples preparation

Salmon samples were prepared from local supermarkets in Dublin, Ireland. There were a total of eighteen farmed Atlantic salmon fillets (Salmo salar) originated from Norway (nine fillets) and Scotland (nine fillets). The salmon fillets were fresh and of superior quality. In order to establish a robust calibration model, a reasonable variation of attribute values should be guaranteed for the examined salmon samples. On the other hand, the WHC could vary within the same fillet and different WHC distributions could be found for the fillets with similar mean WHC values. Therefore, subsampling was conducted to obtain a wide span of WHC for the model calibration. In the laboratories of Food Refrigeration & Computerized Food Technology (FRCFT), University College Dublin (UCD), Ireland, each salmon fillet was first scanned by two hyperspectral imaging systems and then it was cut into several subsamples with a cuboid shape of  $1.5 \text{ cm} \times 1.5 \text{ cm} \times 1.0 \text{ cm}$  $(length \times width \times thickness)$  from different locations within the fillet. A total of 98 cubed samples were obtained from the eighteen fillets and were divided into a calibration set and a prediction set. In the separating process, all samples were first sorted according to their respective reference WHC values. One sample of every five samples was then selected into the prediction set, and remaining four samples were selected into the calibration set. As a result, 79 samples were used for the calibration and the remaining 19 samples were used for prediction. The reference WHC values of these cubed samples were then determined immediately after the acquisition of hyperspectral images using the centrifugation method, in which each sample was first pre-weighed and then placed in a plastic tube with a pre-weighed filter paper, and was then centrifuged (1500 g, 10 °C, 10 min) and reweighed. The time between imaging and WHC measurement for each cuboid subsample was less than three minutes. Four weight indices, namely the weight of sample (S), the weight of pre-weighed filter paper (V1), the weight of the wet paper (V2), and the weight of the wet paper after dried to constant weight (V3), were obtained from the centrifugation method. Four indices of WHC were determined in this study, which were the percentage liquid loss (PLL), percentage water loss (PWL), percentage fat loss (PFL), and percentage water remained (PWR). The first three indices were calculated based on S, V1, V2, and V3 according to the equations in Ref. [31], and the forth index was the percentage of water retained in the salmon flesh after centrifugation. Table 1 summarizes the variations in four WHC indices of the salmon samples. There were wide variations obtained in WHC indices, which was important to establish accurate and robust calibration models [32]. Key steps for the whole procedure are shown in Fig. 1.

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