

Raman on ramen: Nutritional analysis and brand identification

Axell Rodriguez¹ | Valeryia Serada¹ | Patrick Stover² | Dmitry Kurouski^{1,2,3} 

¹Department of Biochemistry and Biophysics, Texas A&M University, College Station, Texas, USA

²Institute for Advancing Health Through Agriculture, Texas A&M University, College Station, Texas, USA

³Department of Biomedical Engineering, Texas A&M University, College Station, Texas, USA

Correspondence

Dmitry Kurouski, Department of Biochemistry and Biophysics, Texas A&M University, College Station, TX 77843, USA.

Email: dkurouski@tamu.edu

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Abstract

Precise and nondestructive assessment of food quality and its nutritional composition can be used to personalize nutrition, which, in turn, will improve well-being and help to prevent chronic diseases for millions of people around the world. Currently used methods for elucidation of nutritional composition of food are destructive, time, and labor consuming. In this study, we investigate the potential of Raman spectroscopy (RS), an emerging analytical technique, in a noninvasive, nondestructive, and a chemical-free analysis of the nutritional composition of ramen, one of the most popular foods in the world. We found that RS could be used to quantify the amount of carbohydrates, gluten, and lipids in ramen. In coupled to chemometrics, RS enables ~100% accurate differentiation between gluten-rich and gluten-free ramen. Furthermore, using RS, one can identify the brand and the country of origin of ramen. These findings suggest that Raman-based sensors can be used in daily life for quality control of consumed food in both restaurants and grocery stores.

KEY WORDS

gluten, PLS-DA, precision nutrition, Raman spectroscopy, ramen

1 | INTRODUCTION

People have different responses to foods and nutrients, which makes an ideal diet for one individual unhealthy for another person.^[1-3] Therefore, it is important to personalize a dietary intake. This emerging concept, known as precision nutrition, can be also used to prevent and treat chronic nutritional diseases. Precision nutrition stimulates the development of sensors that can quantify the amount of carbohydrates, proteins, and other macromolecules in food.^[4-6] Ideally, such sensors should be noninvasive, nondestructive, minimally laborious, and unexpensive.

Raman spectroscopy (RS) is an emerging analytical approach that fits all these requirements.^[4,7-10] The technique is based on the inelastic light scattering upon which photons emitted from the laser exchange energies

with the molecules in the sample.^[11,12] The energy exchange directly depends on the chemical groups present in the molecules.^[13-15] Therefore, collection of such inelastically scattered photons allows for probing chemical structure and composition of analyzed specimens.^[16-18] Our group previously shows that RS could be used to determine concentrations of proteins, carbohydrates, carotenoids, and fiber in intact potato tubers and corn kernels.^[19] Furthermore, RS was able to identify different varieties of both potatoes and corn and even predict geographical origin of their vegetation. Figueiredo and coworkers showed that RS could be used to identify four genotypes of Arabic coffee: one Mundo Novo line and three Bourbon lines with ~80% accuracy.^[20] Recently, Abreu and coworkers utilized RS to examine coffee quality affected by storage conditions and the duration of storage.^[21] It was found that changes in

kahweol could be used to predict quality of coffee beans and changes that took place upon their storage.^[22]

Expanding upon this, we investigate the extent to which RS can be used to analyze nutritional content of noodles, the major component of ramen, food that is broadly consumed in nearly all countries around the world.^[23] First appeared in China, ramen became very popular in Japan after the World War II due to the reduction in rice production and an increasing import of wheat and animal protein in the country.^[24] In 1958, Momofuku Ando, the Taiwanese-Japanese founder and chairman of Nissin Foods, invented instant ramen, which required addition of boiling water to make an approximation of the restaurant-served food.^[25] Simplicity of instant ramen made this food a Japanese cultural icon and one of the most consumed products in the world.^[24]

Ramen noodles are made of wheat flour, salt, water, and kansui, a mineral water that contains sodium and potassium carbonates, as well as a small amount of phosphoric acid.^[25,26] Although there are millions of different varieties and flavors of ramen, all noodles can be divided into two groups: fried (instant) and nonfried.^[24] The former ramen is typically stir-fried or dehydrated in oil heat, whereas the latter type of noodles is not preprocessed prior to packaging.^[24] Wheat is the gluten-rich food.^[27] Consumption of this protein can cause a severe immune reaction, as well as bloating and fatigue in a large group of people. Therefore, it becomes extremely important for gluten-intolerant people to control their food supplies.^[4–6] To address the dietary needs of gluten-intolerant people, rice rather than wheat flour is used to fabricate gluten-free ramen. One can expect that development of the portable Raman-based sensor that can quickly differentiate between gluten-rich and gluten-free ramen can be highly important for the individuals with gluten-intolerance. Therefore, we collected Raman spectra from gluten-rich and gluten-free ramen, as well as from noodles that were and were not fried before packaging. We investigate the extent to which RS can be used as a personal-, restaurant-, or a grocery store-based sensor for a quality control of food, as well as the detector that allows for quantification of carbohydrates, gluten, and fats in ramen.

2 | MATERIALS AND METHODS

2.1 | Materials

Ramen was purchased from local grocery stores in College Station, TX (Figure S1). Gluten and glyceryl tristearate were purchased from Sigma-Aldrich.

2.2 | RS

Raman spectra from all analyzed ramen were collected using a hand-held Resolve Agilent spectrometer equipped with 830-nm laser source (beam diameter ~ 2 mm). The following experimental parameters were used for all collected spectra: 1-s acquisition time, 495-mW power, and baseline spectral subtraction by device software. In total, 40–50 spectra were collected from each brand of ramen. Due to high fluorescence of gluten and glyceryl tristearate, Raman spectra from these foods were collected using a hand-held portable Rigaku Progeny ResQ spectrometer (Rigaku Analytical Devices, Inc. Wilmington, MA), equipped with a 1064 nm Nd:YAG laser. The following experimental parameters were used for both samples: 90-s acquisition time, 495-mW power, and baseline spectral subtraction by device software.

2.3 | Chemometrics

Spectral analysis was made in Matlab (Mathworks) equipped with PLS_Toolbox 9.0 (Eigenvector Research, Inc., Manson, WA). Before model-building, first derivative of the spectra was taken with polynomial order of 2 and derivative order of 1. Partial least squares discriminant analysis (PLS-DA) models were then built using the spectral data with classes assigned based on the brand of ramen. Following this step, the PLS_Toolbox 9.0 performed a cross-validation, where the order of the data was shuffled (classes are maintained), while portions of the data set were excluded, and the rest was used to rebuild the model. The resulted model then aimed to classify the excluded spectra. The permutation test was performed to determine the significance of the classes in the classification model by randomly and repeatedly reassigning data to different categories and then building new models. If the predictions were significant, models built from randomly classified data would perform poorly. All reported models were found to be significant ($\alpha = 0.05$) using 100 iterations of the permutation test. All reported accuracies of spectral prediction in this work are cross-validated results.

All reported PLS-DA models had nine latent variables: LV 1 (57.58%), LV 2 (23.00%), LV 3 (3.76%), LV 4 (2.22%), LV 5 (0.82%), LV 6 (1.42%), LV 7 (0.97%), LV 8 (0.50%), and LV 9 (0.63%).

For ANOVA, first derivative was taken from the spectra. Then, spectra were mean centered and area normalized.

3 | RESULTS AND DISCUSSION

Raman spectra acquired from gluten-rich and fried ramen exhibited vibrational bands that could be assigned to carbohydrates (441, 481, 580, 714, 857, 936, 1005, 1050, 1085, 1124, 1147, 1260, 1338, and 1384 cm^{-1}) (Figures 1 and S2 and Table 1). These bands primarily originated from C-O-C and C-O-H vibrations of sugars. We also observed CH and CH_2 bands at 1445 and 1458 cm^{-1} , which could not be assigned to the specific class of compounds (Table 1). We found that spectra acquired from gluten-rich and fried ramen possessed vibrational bands that could be assigned to gluten (1005 and 1656 cm^{-1}) (Figure 1). These bands originated from the aromatic rings and amide I vibrations of proteins, respectively. Finally, Raman spectra acquired from gluten-rich and fried ramen had bands that originate from the carboxyl group of lipids (1743 cm^{-1}) (Figure 1). These findings demonstrated that gluten-rich and fried ramen was dominated by carbohydrates with gluten and lipids present in these foods.

Raman spectra acquired from gluten-free and non-fried ramen exhibited vibrational bands that could be assigned to carbohydrates (441, 481, 580, 714, 857, 936, 1005, 1050, 1085, 1124, 1147, 1260, 1338, and 1384 cm^{-1}) (Figures 1 and S3 and Table 1). We also found that the intensities of aromatic ring of phenylalanine or tyrosine (1005 cm^{-1}), as well as amide I band of gluten (1656 cm^{-1}), were significantly lower in these spectra compared with the intensity of these bands in the Raman spectra acquired from the gluten-rich ramen. These results demonstrated that although gluten-free ramen, similar to gluten-rich and fried ramen, was dominated by carbs, these varieties of noodles possessed substantially lower amount of gluten. We also found that Raman spectra acquired from gluten-free and nonfried ramen did not exhibit vibrational bands that originated from lipids (1743 cm^{-1}), as well as had significantly lower intensities of CH and CH_2 vibrations compared with the intensities of these aliphatic vibrations in the Raman spectra acquired from gluten-rich ramen. Since CH and CH_2 groups are the major building blocks of fatty acids, these

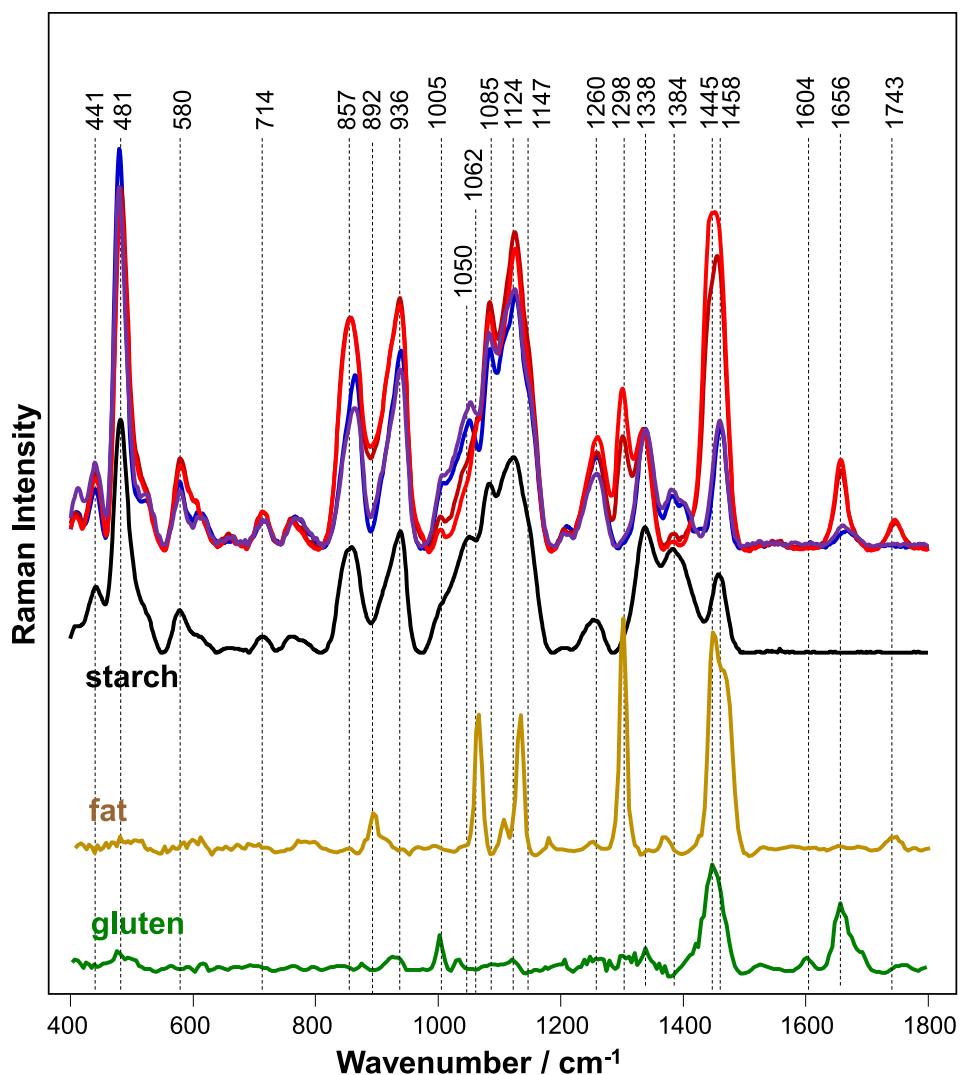


FIGURE 1 Representative Raman spectra of gluten-rich and fried (Gomtang, red and Paldo, maroon) and gluten-free and nonfried ramen (Koyo, blue and HakuBaku, purple). Raman spectra of starch (black), glyceryl tristearate (golden), and gluten (green). [Colour figure can be viewed at wileyonlinelibrary.com]

TABLE 1 Vibrational bands and their assignments for Raman spectra acquired from ramen.

Band	Vibrational mode	Assignment
1743	C=O stretching	Lipids ^[38]
1656	C=O stretching (amide I)	Proteins ^[27,39]
1604	Aromatic ring	Proteins ^[40]
1458	δ (CH) + δ (CH ₂)	Aliphatic ^[41]
1445	δ (CH) + δ (CH ₂)	Aliphatic ^[41]
1384	δ (C-O-H) - coupling of the CCH and COH deformation modes	Carbohydrates ^[41]
1338	δ CH ₂ bending vibration	Aliphatic ^[41]
1298	CH ₂ twisting	Lipids ^[38]
1260	δ (C-C-H) + δ (O-C-H) + δ (C-O-H)	Carbohydrates ^[41,42]
1147	ν (C-O-C), ν (C-C) in glycosidic linkage, asymmetric ring breathing	Carbohydrates ^[43]
1124	ν (C-O) + ν (C-C) + δ (C-O-H)	Carbohydrates ^[41]
1085	ν (C-O) + ν (C-C) + δ (C-O-H)	Carbohydrates ^[41]
1062	ν (C-O-C)	Lipids
1050	ν (C-O) + ν (C-C) + δ (C-O-H)	Carbohydrates ^[41]
1005	Phenylalanine ring stretching mode	Proteins ^[39]
936	δ (C-O-C) + δ (C-O-H) + ν (C-O) α -1,4 glycosidic linkages	Carbohydrates ^[41]
892	CH ₂ wagging	Lipids ^[38]
857	δ (C-C-H) + δ (C-O-C) glycosidic bond; anomeric region	Carbohydrates ^[41]
714	δ (C-C-O) related to glycosidic ring skeletal deformations	Carbohydrates ^[41]
580	δ (C-C-O) + τ (C-O)	Carbohydrates ^[41]
481	CCO and CCC deformations; related to glycosidic ring skeletal deformations δ (C-C-C) + τ (C-O) scissoring of C-C-C and out-of-plane bending of C-O	Carbohydrates ^[41]
441	Skeletal modes of pyranose ring	Carbohydrates ^[41]

spectroscopic changes confirm the absence of detectable quantities of fats in the nonfried ramen. These results demonstrated drastic differences between Raman spectra acquired from two major classes of noodles: gluten-free/nonfried and gluten-rich/fried ramen.

The question to ask is how accurate one can differentiate between gluten-rich vs gluten-free ramen using a hand-held Raman spectrometer. To answer this question, we utilized PLS-DA. Our results showed that RS enabled 100% accurate identification of gluten-rich/fried ramen and 99.6% accurate identification of gluten-free/nonfried noodles (Table 2). Thus, on average, RS could be used for nearly 100% accurate differentiation between gluten-rich versus gluten-free ramen.

One may question whether RS could be also used to quantify the amount of carbohydrates, gluten, and fats in ramen. In the acquired Raman spectra, we observed small variations in the intensities of the vibrational bands that can be assigned to carbohydrates, gluten, and fats. Therefore, one can expect that these spectroscopic differences could be used to quantify the amount of these important nutrients in noodles. To answer this question, we used ANOVA. We found that Lotus Foods and Koyo possessed significantly less carbohydrates than all other ramen, whereas Nissin and Nongshim exhibited the greatest amount of carbohydrates compared with all analyzed noodles. ANOVA of 1665 cm^{-1} band, which can be assigned to gluten, enabled quantitative assessment of gluten content in ramen. Specifically, we found that Lotus Foods possessed less gluten than all other ramen, whereas Mike's noodles had the highest amount of gluten compared with all gluten-free noodles. As expected, intensity of 1665 cm^{-1} band was high in all gluten-rich noodles. We also observed a drastic difference between the intensity of 1730 cm^{-1} band in all gluten-free (non-fried) and gluten-rich (fried) noodles. Our results also show that Gomtang and Nissin exhibit the highest amount of fats among all gluten-rich (fried) ramen. These results demonstrate that RS can be used to quantify the amount of important nutrients in ramen. These findings also showed that different varieties of ramen possess different amounts of carbohydrates, gluten, and fats. Thus, using a hand-held Raman scanner, one can verify the

	Accuracy, %	Gluten-free/nonfried	Gluten-rich/fried
Gluten-free/nonfried	99.6	292	0
Gluten-rich/fried	100	1	198

TABLE 2 Misclassification table of cross-validation for the PLS-DA model of gluten-rich/fried and gluten-free/nonfried ramen.

Abbreviation: PLS-DA, partial least squares discriminant analysis.

amount of these important nutrients in consumed food (Figure 2).

One may expect that Raman spectra of ramen could be used to identify brands and predict the

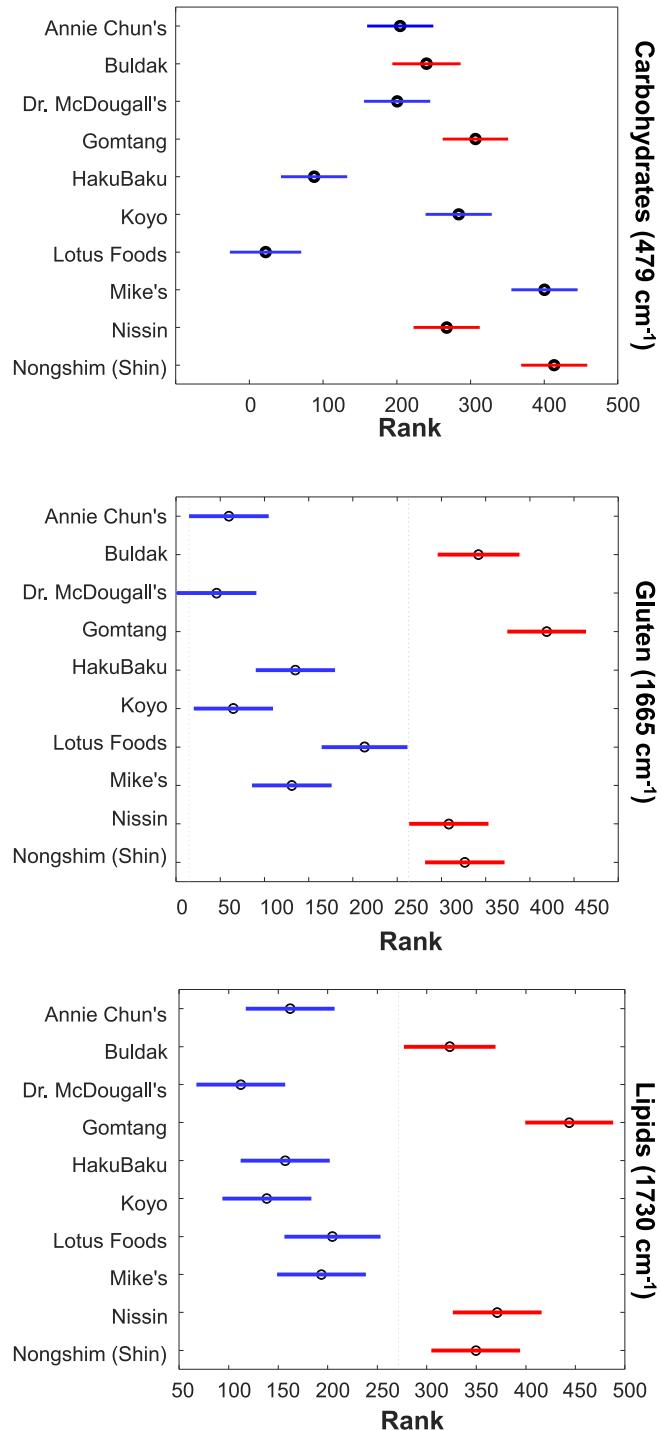


FIGURE 2 Means (circles) and confidence intervals for the intensities of 479 cm^{-1} (carbohydrates), 1665 cm^{-1} (gluten), and 1730 cm^{-1} (lipids) in the spectra acquired from gluten-rich/fried ramen (red) and gluten-free/nonfried noodles (blue). [Colour figure can be viewed at wileyonlinelibrary.com]

TABLE 3 Misclassification table of crossvalidation for the PLS-DA model of different brands of ramen.

	Accuracy, %	Annie Chun's	Buldak	Mr. McDougall's	Gomtang	HakuBaku	Koyo	Lotus foods	Mike's	Nissin	Nongshim
Annie Chun's	100.0	50	0	0	0	0	0	0	0	0	0
Buldak	68.0	0	32	1	4	1	0	0	1	0	0
Mr. McDougall's	64.0	0	1	32	0	0	6	0	1	1	0
Gomtang	92.2	0	8	0	47	0	0	0	0	2	2
HakuBaku	100.0	0	0	0	0	50	0	0	0	0	0
Koyo	86.0	0	1	12	0	0	43	0	0	0	0
Lotus Foods	100.0	0	0	0	0	0	0	43	0	0	0
Mike's	100.0	0	1	0	0	0	1	0	48	0	0
Nissin	100.0	0	0	0	0	0	0	0	50	0	0
Nongshim	92.0	0	5	1	0	0	0	0	0	0	46

Abbreviation: PLS-DA, partial least squares discriminant analysis.

country of origin of noodles. To answer this question, we performed PLS-DA analysis of brands and country of origin of ramen (Figure S4 and Tables 3 and 4). Our results show that RS enables on average 90% accurate identification of ramen brands. Some of the analyzed brands, such as Annie Chun's, HakuBaku, Lotus Foods, Mike's, and Nissin could be identified with 100% accuracy, whereas accuracy identification of Gomtang and Nongshim was 92%. Finally, we found that PLS-DA model enabled relatively low accuracy of identification of Mr. McDougall's and Buldak, 64% and 68%, respectively. Our results also showed that RS could be used to identify the country of origin of ramen (Figure S3 and Table 4). Specifically, Australian and Japan noodles could be identified using RS with 100% accuracy, whereas identification of Korean and American noodles could be achieved with 98% and 99% accuracies, respectively. ANOVA shows that on average, Australian ramen possesses significantly less carbohydrates than noodles from Japan, Korea, and USA (Figure 3). We also found that average gluten and fats increase from Australian noodles to ramen from USA and Korea, whereas the greatest amount of fats was observed in Japanese noodles. These results suggest that RS could be used to reveal the geographical origin of ramen.

These results are in a good agreement with experimental findings previously reported by our group^[19,28-30] and other^[31,32] research groups. These studies demonstrated that noninvasive and nondestructive nature of RS allowed for a highly accurate determination of the watermelon^[28] and tomato ripeness, elucidation of nutritional composition of banana fruits,^[31] potato tubers,^[30] peanuts,^[29] and corn kernels,^[19] as well as diagnostics of fungal,^[17,33] viral,^[10,34] and bacterial^[35-37] pathogens in plants. Thus, hand-held Raman scanners could be used to perform on-site analyses of food to reveal its nutritional profile and dietary suitability for different individual. One can also expect that such Raman sensors can ultimately enable determination of the geographical origin of food, as well as ensure the absence of pathogens in food supplies.

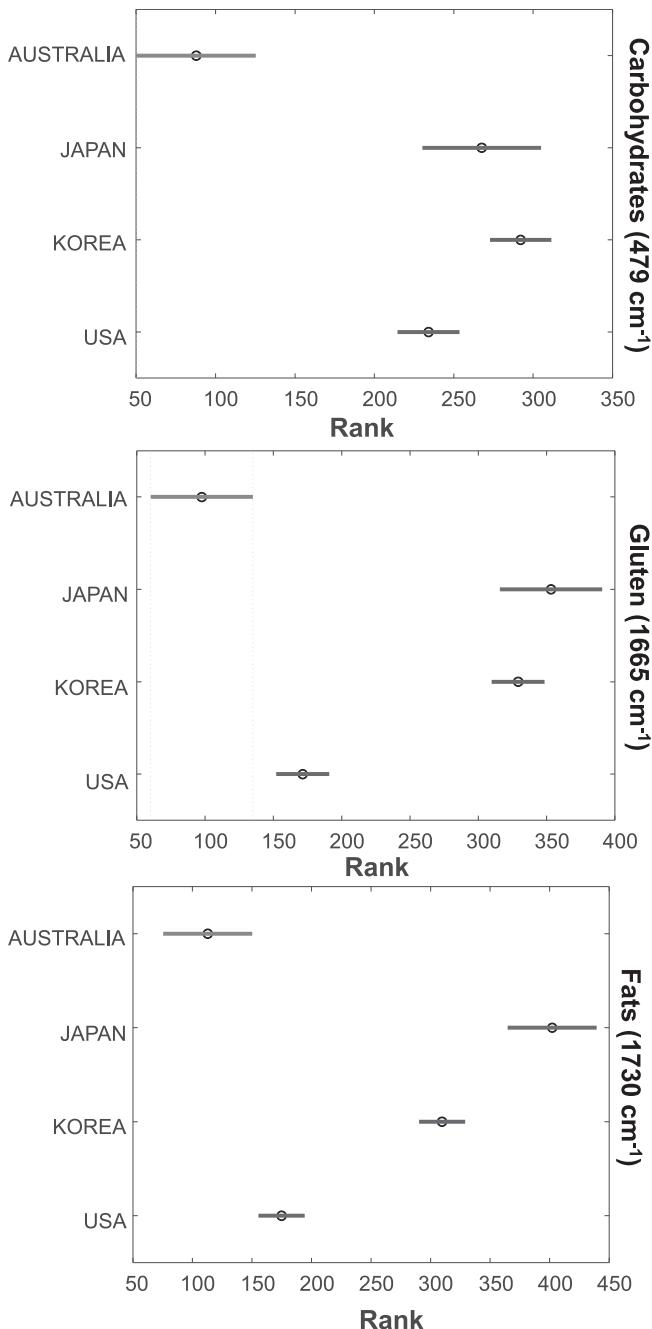


FIGURE 3 Means (circles) and confidence intervals for the intensities of 479 cm^{-1} (carbohydrates), 1665 cm^{-1} (gluten), and 1730 cm^{-1} (lipids) in the spectra acquired from Australian (green), Japanese (purple), Korean (blue), and American (red) noodles.

TABLE 4 Misclassification table of cross-validation for the PLS-DA model of ramen from Australia, Japan, Korea, and USA.

	Accuracy, %	Australia	Japan	Korea	USA
Australia	100	50	0	0	0
Japan	100	0	50	0	0
Korea	98.4	0	0	195	1
USA	99.4	0	0	3	192

Abbreviation: PLS-DA, partial least squares discriminant analysis.

4 | CONCLUSIONS

Our results show that RS can be used to differentiate between gluten-rich/fried and gluten-free/nonfried ramen with nearly 100% accuracy. Furthermore, using RS, one can quantify the amount of gluten in noodles. We also demonstrate that RS enables quantification of carbohydrates and lipids in ramen, as well as allows for identification of the brand and the country of origin of ramen. Considering noninvasive, nondestructive, and label-free nature of RS, one can expect that hand-held Raman sensors could be used in daily life for quality control of consumed food in both restaurants and grocery stores.

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COMPLIANCE WITH ETHICAL STANDARDS

CONFLICT OF INTEREST STATEMENT

None.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

ORCID

Dmitry Kurouski  <https://orcid.org/0000-0002-6040-4213>

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