

การใช้สเปกโทรสโกปีแบบอินฟราเรดย่านใกล้สำหรับการวิเคราะห์รำข้าวผสมรำข้าวหนึ่ง



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USE OF NEAR-INFRARED SPECTROSCOPY FOR ANALYSIS OF MIXED RICE
BRAN AND PARBOILED RICE BRAN

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การผลิตน้ำมันรำข้าวสามารถใช้รำข้าวหนึ่งหรือรำข้าวดิบ โดยรำข้าวหนึ่งมีสมบัติที่เป็นที่ต้องการมากกว่ารำข้าวดิบ เนื่องจากมีปริมาณน้ำมันสูงและมีค่ากรดต่ำกว่า จึงทำให้รำข้าวหนึ่งมีราคาสูงกว่า จากความแตกต่างด้านราคาของวัตถุดิบอาจเป็นเหตุจูงใจให้ผู้จำหน่ายวัตถุดิบผสมรำข้าวดิบในรำข้าวหนึ่ง ส่งผลให้วัตถุดิบมีค่ากรดสูงและมีปริมาณน้ำมันที่สกัดได้น้อยกว่าที่ควรจะเป็น ในปัจจุบันมีการใช้เทคนิค Near Infrared Spectroscopy (NIR) ในการวิเคราะห์องค์ประกอบของอาหาร ซึ่งสามารถวิเคราะห์ได้อย่างแม่นยำ รวดเร็ว ไม่ใช้สารเคมี และไม่ทำลายตัวอย่าง ในปัจจุบันมีฐานข้อมูล NIR ของรำข้าวดิบและรำข้าวหนึ่งแยกกัน งานวิจัยนี้จึงสนใจที่จะพัฒนาฐานข้อมูล NIR ของรำข้าวผสม โดยตัวอย่างที่ใช้ในการวิจัยเป็นรำข้าวผสมระหว่างรำข้าวดิบและรำข้าวหนึ่งที่แปรสัดส่วนรำข้าวดิบ: รำข้าวหนึ่ง 11 สัดส่วน ตั้งแต่ 0: 10 จนถึง 10: 0 จำนวน 10 สล็อต จากโรงสีในเขตพื้นที่จังหวัดชัยนาท พิจารณาครสวรรค์ พิชญ์โลก กำแพงเพชร และนครปฐม ตรวจวัดสมบัติของรำข้าวผสมโดยการใช้ NIR และศึกษาองค์ประกอบทางเคมี ได้แก่ ปริมาณความชื้น โปรตีน ไขมัน เส้นใยหยาบ กรดไฟติก ค่ากรด ความหนาแน่นรวมทั้งแบบเคาะและไม่เคาะ และค่าสี (ค่าดัชนีสีขาว) จากการศึกษพบว่า รำข้าวหนึ่งมีปริมาณไขมัน โปรตีน และเส้นใยหยาบสูงกว่ารำข้าวดิบ ในขณะที่ปริมาณความชื้น ค่ากรด กรดไฟติก ค่าดัชนีสีขาวและค่าความหนาแน่นรวมในรำข้าวดิบมีปริมาณสูงกว่าอย่างมีนัยสำคัญ ($p < 0.05$) และการติดตามสัดส่วนของรำดิบและรำหนึ่ง พบว่าทุกพารามิเตอร์สามารถใช้เป็นตัวชี้วัดสัดส่วนของรำดิบและรำหนึ่งได้ดี การวัดสมบัติ NIRS ของตัวอย่างใช้ข้อมูลการสะท้อนกลับในช่วงเลขคลื่น $12,500 \text{ cm}^{-1} - 3,600 \text{ cm}^{-1}$ และสร้างความสัมพันธ์ระหว่าง NIR spectrum กับสมบัติทางเคมีและกายภาพ โดยใช้ Partial least square (PLS) regression สร้างสมการสอบเทียบ ตรวจสอบสมการแบบใช้ชุดข้อมูลเดิม และแบบใช้ชุดข้อมูลภายนอกโดยใช้ตัวอย่างจำนวน 165, 55 และ 60 ตัวอย่าง ตามลำดับ ผลการศึกษาแสดงให้เห็นว่าสามารถใช้ข้อมูล Raw NIR spectrum ในการสร้างความสัมพันธ์กับปริมาณไขมันและค่าความหนาแน่นรวมแบบเคาะ ได้สมการทำนายที่มีค่า Coefficient of determination (R^2) เท่ากับ 0.98 และ 0.91 ตามลำดับ มีค่า Root mean square error (RMSE) เท่ากับ 0.36 และ 0.01 ตามลำดับ ความชื้น เส้นใยหยาบ และค่าสี (ดัชนีสีขาว) มีความสัมพันธ์กับข้อมูลสเปกตรัมที่แปลงด้วยวิธี Normalization ได้สมการที่ทำนายที่มีค่า R^2 เท่ากับ 0.91 0.91 และ 0.91 ตามลำดับ มีค่า RMSE เท่ากับ 0.13 0.36 และ 0.48 ตามลำดับ ค่ากรดและค่าความหนาแน่นรวมแบบไม่เคาะมีความสัมพันธ์กับข้อมูลสเปกตรัมที่แปลงด้วยวิธี Baseline ซึ่งได้สมการที่ทำนายที่มีค่า R^2 เท่ากับ 0.94 และ 0.92 ตามลำดับ มีค่า RMSE เท่ากับ 1.53 และ 0.01 ตามลำดับ ในขณะที่โปรตีนและกรดไฟติกมีความสัมพันธ์กับข้อมูลสเปกตรัมที่แปลงด้วยวิธี Smoothing moving average และ smoothing median filter ตามลำดับ ได้สมการที่ทำนายที่มีค่า R^2 เท่ากับ 0.92 และ 0.96 ตามลำดับ มีค่า RMSE เท่ากับ 0.20 และ 0.15 ตามลำดับ จากนั้นทดสอบความใช้ได้ของสมการแบบใช้ชุดข้อมูลชุดเดิม (internal validation) พบว่าการใช้ข้อมูล NIR สามารถทำนายปริมาณความชื้น โปรตีน ไขมัน เส้นใยหยาบ กรดไฟติก ค่ากรด ค่าความหนาแน่นรวมทั้งแบบเคาะและไม่เคาะ และค่าดัชนีสีขาวได้ดี โดยมีค่า R^2 ในช่วง 0.87 - 0.97 โดยการทำนายค่าความหนาแน่นรวมแบบเคาะมีค่า R^2 ต่ำที่สุด (0.87) ส่วนการทำนายปริมาณไขมันให้ค่า R^2 สูงที่สุด (0.97) และจากการทดสอบความใช้ได้ของสมการแบบใช้ชุดข้อมูลชุดภายนอก (external validation) พบว่าการใช้ข้อมูล NIR สามารถทำนายปริมาณไขมันได้ดีที่สุด รองลงมาคือ กรดไฟติก ค่าความหนาแน่นรวมแบบเคาะ เส้นใยหยาบ ค่าสี (ดัชนีสีขาว) โปรตีน และค่าความหนาแน่นรวมแบบไม่เคาะ ตามลำดับ ซึ่ง R^2 เท่ากับ 0.97, 0.89, 0.87, 0.86, 0.80, 0.78 และ 0.75 ตามลำดับ

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WIRONGRONG MAKSAWASD: USE OF NEAR-INFRARED SPECTROSCOPY FOR ANALYSIS OF MIXED RICE BRAN AND PARBOILED RICE BRAN. ADVISOR: ASSOC. PROF. JIRARAT ANUNTAGOOL, Ph.D., 84 pp.

Both rice bran and parboiled rice bran can be used for rice bran oil production. Parboiled rice is more desirable than rice bran due to its higher oil content and lower acid value compared to rice bran, hence higher price. The difference in price of raw materials may motivate suppliers to mix raw rice bran in parboiled rice bran. As a result, rice bran oil manufacturers frequently come across parboiled rice bran samples that have higher acid value and lower oil content. Currently, Near Infrared Spectroscopy (NIR) is widely used in rice bran oil production plant as a rapid and non-destructive method to analyze food components. At present, there are separate NIR databases for rice bran and parboiled rice bran. None of these two database can be used to accurately read or analyze the properties of mixed rice bran. Therefore, this research aimed to develop the NIR database of mixed rice bran. The research recruited 10 lots of rice bran and parboiled rice bran from the mills in Chai Nat, Phichit, Nakhon Sawan, Phitsanulok, Kamphaengphet and Nakhon Pathom. The mixed rice brans were prepared by mixing rice bran and parboiled rice bran in the 0: 10 to 10: 0 ratio. The properties of mixed rice bran were determined by using NIR spectroscopy, analyses of chemical composition and physical properties, e.g. the content of moisture, protein, fat, crude fiber, phytic acid, acid value, bulk density (tapped), bulk density (untapped), and color (whiteness index). The result showed that parboiled rice has higher protein and crude fiber content than rice bran, whereas rice bran had higher moisture content, acid value, phytic acid, whiteness index, and bulk density ($p < 0.05$). Measurement of NIRS properties of the samples was carried out in the range of $12,500 \text{ cm}^{-1}$ - $3,600 \text{ cm}^{-1}$ in diffuse reflection mode. The relationship between NIR spectrum and chemical - physical properties of the samples were done by using Partial least square (PLS) regression, with the sample data for calibration, internal validation, and external validation of 165, 55, and 60, respectively. The results showed that raw NIR spectral data could be used to correlate with fat content and bulk density (tapped) with the coefficient of determination (R^2) of 0.98 and 0.91, respectively, and root mean square error (RMSE) of 0.36 and 0.01, respectively. Moisture content, fiber, and whiteness index were correlated well with the pretreated spectra using normalization method with an R^2 of 0.91, 0.91, and 0.91, and RMSE of 0.13, 0.36, and 0.48, respectively. Acid value and bulk density (untapped) were correlated with the pretreated spectra using baseline method with an R^2 of 0.94 and 0.92, and RMSE of 1.53 and 0.01, respectively. Protein and phytic acid were correlated with the pretreated spectra using smoothing moving average and smoothing median filter method, respectively, with an R^2 of 0.92 and 0.96, and RMSE of 0.20 and 0.15, respectively. Internal validation showed that moisture content, protein, fat, fiber, phytic acid, acid value, bulk density (tapped), bulk density (untapped), and whiteness index could be predicted using NIRS readings with an R^2 of 0.87 to 0.97, where the R^2 for bulk density (tapped) prediction was the lowest (0.87) and the R^2 for fat prediction was the highest (0.97). Further confirmation using external validation samples showed that NIRS could be used to predict the properties of mixed rice bran in the descending order for R^2 ; fat content (0.97), phytic acid (0.89), bulk density tapped (0.87), fiber content (0.86), whiteness index (0.80), protein content (0.78), and bulk density untapped (0.75).

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CONTENTS

	Page
THAI ABSTRACT	iv
ENGLISH ABSTRACT.....	v
ACKNOWLEDGEMENTS	vi
CONTENTS.....	vii
LIST OF TABLES	i
LIST OF FIGURE.....	iii
CHAPTER I INTRODUCTION.....	1
CHAPTER II LITERATURE REVIEW	2
2.1 Rice bran	2
2.1.1 Raw rice bran.....	2
2.1.2 Parboiled rice bran.....	2
2.2 Near infrared spectroscopy	3
2.2.1 Infrared radiation.....	3
2.2.2. Principle of Near infrared spectrophotometer	4
2.2.3. The steps to analyze NIRS	5
2.2.3.2 Validation of the generated equations	6
2.2.4 Absorption process of Near infrared spectroscopy	6
2.2.4.1 Vibration of bonding	6
2.2.4.2 The absorbance process of NIRS	7
2.2.4.3 Patterns of interaction.....	11
2.2.5 Application of NIRS in food detection.....	12
CHAPTER III MATERIALS AND METHODS	15
3.1 Materials	15
3.1.1 Rice bran samples.....	15
3.1.2 Parboiled rice bran samples.....	15
3.2 Methods	16
3.2.1 Sample preparation.....	16
3.2.2 Chemical analysis.....	16

	Page
3.2.2.1 Moisture content.....	16
3.2.2.2 Protein content.....	16
3.2.2.3 Fat content	16
3.2.2.4 Crude fiber content.....	17
3.2.2.5 Acid value.....	17
3.2.2.6 Phytic acid content	17
3.2.3 Physical properties analysis.....	17
3.2.3.1 Color.....	17
3.2.3.2 Bulk density.....	17
3.2.4 FT-NIR analysis	17
3.2.5 Chemometric analysis of FT-NIR data	18
3.2.5.1 Predictive model construction	18
3.2.5.2 External validation	18
3.2.6 Statistical analysis	18
CHAPTER IV RESULTS AND DISCUSSION.....	19
4.1 Chemical composition and physical properties of mixed rice bran.....	19
4.1.1 Fat content	20
4.1.2 Acid value.....	20
4.1.3 Phytic acid.....	21
4.1.4 Protein content.....	25
4.1.5 Moisture content.....	25
4.1.6 Fiber content.....	25
4.1.7 Color (whiteness index).....	29
4.1.8 Bulk density.....	29
4.2 Relationship between chemical composition, physical properties and ratios of mix rice bran	33
4.3 FT-NIR analysis of mixed rice bran	37
4.3.1 FT-NIR spectra.....	37
4.3.2 Chemometric analysis of FT-NIR data	37

	Page
4.4 External validation.....	53
CHAPTER V CONCLUSIONS	63
REFERENCES	64
APPENDIX.....	69
APPENDIX A CHEMICAL ANALYSIS PROCEDURES	70
APPENDIX B CHEMICAL – PHYSICAL ANALYSIS.....	80
VITA.....	84



LIST OF TABLES

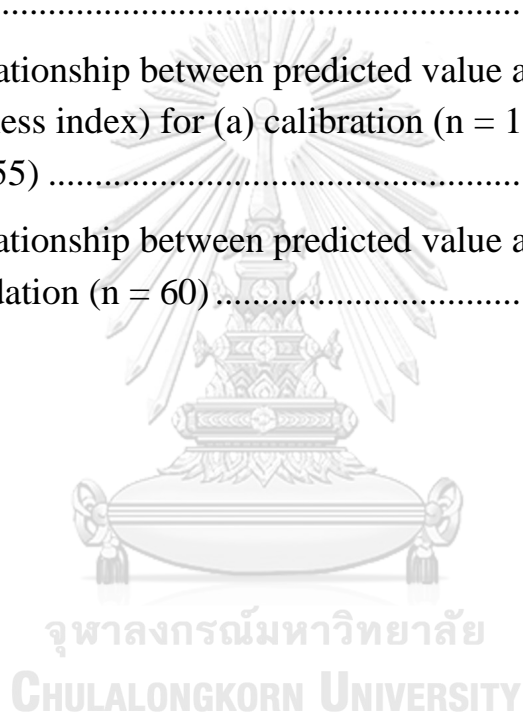
Table 2.1 Range of infrared radiation.....	3
Table 2.2 Classification of the near infrared spectroscopy range by type of energy level	3
Table 2.3 Classification of the near infrared spectroscopy range by type of band	4
Table 2.4 Positions of peak in the NIR spectrum relative to the various functional groups in the sample.....	9
Table 2.5 The peak position of important components in the sample.....	9
Table 2.6 Position of peak in the NIR spectrum correlates with the components in the product.....	10
Table 3.1 Rice mill area for rice bran samples.....	15
Table 3.2 Rice mill area for parboiled rice bran samples.....	16
Table 4.1 Chemical compositions and physical properties of 220 rice bran samples	19
Table 4.2 Fat content of mixed rice bran.....	22
Table 4.3 Acid value of mixed rice bran	23
Table 4.4 Phytic acid of mixed rice bran.....	24
Table 4.5 Protein content of mixed rice bran	26
Table 4.6 Moisture content of mixed rice bran	27
Table 4.7 Fiber content of mixed rice bran	28
Table 4.8 Color (whiteness index) of mixed rice bran	30
Table 4.9 Bulk density (tapped) of mixed rice bran.....	31
Table 4.10 Bulk density (untapped) of mixed rice bran.....	32
Table 4.11 Chemical composition and physical properties and ratios of mix rice bran	34

Table 4.12 Range of composition analysis in the sample used to develop PLS models	39
Table 4.13 PLS model statistics for mixed rice bran and parboiled rice bran	40
Table 4.14 Totals PLS model statistics for mixed rice bran and parboiled rice bran	49
Table 4.15 Chemical compositions and physical properties of 60 external rice bran samples	53
Table 4.16 Chemical analyzes and physical properties of external rice bran samples	55
Table 4.17 Chemical analyzes and physical properties of external parboiled rice bran samples	56
Table 4.18 Chemical analyzes and physical properties of external mixed rice bran samples	57
Table 4.19 Range of composition analysis for calibration set and the external validation set	58
Table 4.20 Test the accuracy of the equation by PLS model statistics for external validation set	59
Table B.1 Lightness of mixed rice bran	81
Table B.2 Redness of mixed rice bran.....	82
Table B.3 Yellowness of mixed rice bran	83

LIST OF FIGURE

Figure 2.1 Components of the NIR spectrophotometer	5
Figure 2.2 The steps to analyze NIRS	5
Figure 2.3 Changes in molecular bonds in various forms.....	7
Figure 2.4 Overtones in NIR spectrum.....	8
Figure 2.5 Patterns of NIR interaction (a) transmittance, (b) transreflectance, (c) reflectance, and (d) interactance.	11
Figure 4.1 Regression plot between chemical composition and physical properties and ratios of mixed rice bran and parboiled rice bran (a) moisture content, (b) protein content, (c) fiber content, (d) fat content, (e) acid value, (f) phytic acid (g) bulk density (tapped), (h) bulk density (untapped), and (i) whiteness index	35
Figure 4.2 Raw FT-NIR spectra of mixed rice bran and parboiled rice bran (wavenumber 12,500 - 3,600 cm^{-1})	37
Figure 4.3 Relationship between predicted value and reference value of fat for (a) calibration (n = 165) and (b) internal validation (n = 55)....	50
Figure 4.4 Relationship between predicted value and reference value of phytic acid for (a) calibration (n = 165) and (b) internal validation (n = 55)	50
Figure 4.5 Relationship between predicted value and reference value of acid value for (a) calibration (n = 165) and (b) internal validation (n = 55).....	50
Figure 4.6 Relationship between predicted value and reference value of protein for (a) calibration (n = 165) and (b) internal validation (n = 55)	51
Figure 4.7 Relationship between predicted value and reference value of bulk density (untapped) for (a) calibration (n = 165) and (b) internal validation (n = 55)	51

Figure 4.8 Relationship between predicted value and reference value of bulk density (tapped) for (a) calibration (n = 165) and (b) internal validation (n = 55)	51
Figure 4.9 Relationship between predicted value and reference value of moisture for (a) calibration (n = 165) and (b) internal validation (n = 55).....	52
Figure 4.10 Relationship between predicted value and reference value of fiber for (a) calibration (n = 165) and (b) internal validation (n = 55).....	52
Figure 4.11 Relationship between predicted value and reference value of color (whiteness index) for (a) calibration (n = 165) and (b) internal validation (n = 55)	52
Figure 4.12 Relationship between predicted value and reference value of external validation (n = 60).....	60



CHAPTER I INTRODUCTION

Both rice bran and parboiled rice bran can be used for rice bran oil production. Parboiled rice bran is more desirable than rice bran due to its higher oil content and lower acid value compared to rice bran, hence higher price. The difference in price of raw materials may motivate suppliers to mix raw rice bran in parboiled rice bran. As a result, rice bran oil manufacturers frequently come across parboiled rice bran samples that have higher acid value and lower oil content. Currently, Near Infrared Spectroscopy (NIRS) is widely used in rice bran oil production plant as a rapid and non-destructive method to analyze bran components. The method can be used to investigate several parameters; both quantitative and qualitative, within one scan. Application of NIRS in the agro-industry includes determination of various properties, such as total soluble solids and dry matter in mango fruit, soluble solids, titratable acidity, and lycopene content of tomatoes, dissolve solid and bulk density of pear, total amino acids in oilseeds, firmness and sugar content of apples, fat, nitrogen and moisture of cocoa powder, for examples. In rice grain, NIRS has been widely used to determine rice quality, for instance quality of flour, starch and whole grains. In fat or oil, NIRS has been used to determine oil content in plant such as sunflower seeds, palm, Chinese faba bean, cocoa bean, *Jatropha curcas* seed, peanut seed and rice bran. Form the literature reviewed, determination of mixed rice bran and parboiled rice bran using NIR spectroscopy is still scarce. At present, there are separate NIR databases for rice bran and parboiled rice bran. None of these two databases can be used to accurately read or analyze the properties of mixed rice bran. Therefore, this research aimed to develop the NIRS database for the assessment of chemical and physical properties of mixed rice bran and to determine the proportion of mixed rice bran in parboiled rice bran, as well as its properties.

CHAPTER II LITERATURE REVIEW

2.1 Rice bran

Rice bran used in the oil manufacturing process can be divided into raw rice bran and parboiled rice bran.

2.1.1 Raw rice bran

Raw rice bran, also called rice bran, is derived from the milling of raw rice paddy. It is a membrane, pericarp or arillus, necellus, aleurone layer, and germ layer (Houston, 1972). The chemical composition of rice bran depends on the part of rice bran, which is divided into 2 types; the bran derived from polishing brown rice of grain and the polish derived from whitening and polishing processes. The chemical composition of both types of bran is slightly different. Bran contains higher content of protein, fat, fibers, ash, minerals, and some vitamins than polish. Therefore, it is commonly used in crude oil extraction and protein extraction process (Luh, 1991).

2.1.2 Parboiled rice bran

Parboiled rice bran is derived from the milling of hydrothermally treated paddy. In the parboiling process, paddy is soaked in water until the seeds are saturated, then steamed until the grains are partially cooked. The parboiled rice is then dried and milled. The byproduct of this process is parboiled rice bran that has stable and strong structure because lipase and oxidase enzymes are destroyed (Luh, 1991). Parboiled rice bran usually yields more oil than rice bran in the oil extraction process. The oil content of parboiled rice bran is approximately 24% whereas that of rice bran is approximately 15% (Mali, 2001). Bhattacharya (2004) reported that the oil in rice grain diffuses from inside to the outer layer during steaming causing higher oil content in the parboiled rice bran. Parboiled rice bran also has darker color and higher protein content than rice bran. Phytic acid and phytate are generally found in the cotyledons of oil-producing plants (Shahidi, 1997). They can be degraded by soaking and heating. During soaking, phytase is activated and causes the hydrolysis of phytic acid (Lestienne *et al.*, 2005). Vidal-Valverde *et al.* (1994) reported that combined soaking and heating is more effective than soaking alone in reducing the level of phytic acid.

2.2 Near infrared spectroscopy

2.2.1 Infrared radiation

Electromagnetic radiation in the Infrared region can be subdivided into 3 ranges; near infrared (Near IR or NIR), mid infrared (Mid IR or MIR), and far infrared (Far IR) as shown in Table 2.1.

Table 2.1 Range of infrared radiation (Osborne *et al.*, 1993)

Range	Wavelength (nm)	Wave number (cm ⁻¹)	Interaction
Near infrared (Near IR, NIR)	600-2,500	14,000-4,000	Overtone, Combination
Mid infrared (Mid IR, MIR)	2,500-50,000	4,000-200	Basic vibration
Far infrared (Far IR)	50,000-1,000,000	200-10	Rotation

Infrared light distribution can be divided into two types, according to the energy level; short and long wavelength as shown in Table 2.2.

Table 2.2 Classification of the near infrared spectroscopy range by type of energy level (Barton II, 2002)

Wavelength (nm)	Range	Energy (kJ/mol)
800-1,100	Short wavelength NIR, SWNIR, Herschel region	~150-109
1,100-2,500	Long wavelength NIR, LWNIR	~109-48

Short wavelength NIR (800-1,100 nm) has higher energy and can penetrate into the mass well. In general, it can penetrate up to 1-2 cm. Therefore, it is often used in the short-wave analysis of specimens, for example shell fruit, especially fruits with thick shell (Barton II, 2002).

Long wavelengths NIR (1,100-2,500 nm) that has lower energy can penetrate up to 0.5-1 cm. Thus, it is suitable for common samples in both liquid and solid forms. Analyses using long wavelength NIR provide more chemical information than the short wavelength NIR because the first overtones and combination appear in this range of examination.

Table 2.3 shows classification of NIR according to band types. NIR having the wavelength between 800-1,200 nm will interact with the sample and cause second and third overtone and combination vibration, while that having the wavelength between 1,200-1,800 nm will interact with the sample and cause first overtones and combination vibration. NIR having the wavelength between 1,800-2,500 nm will interact with the sample and cause combination and second overtone vibration.

Table 2.3 Classification of the near infrared spectroscopy range by type of band (Chalmers, 2000)

Wavelength (nm)	Band
800-1,200	second overtone and third overtone, combination of XH (X=O, N, C)
1,200-1,800	first overtones, combination of XH
1,800-2,500	combination, second overtone of C=O

2.2.2. Principle of Near infrared spectrophotometer

Near infrared spectrophotometer consists of light source, monochromator, detector, and read out. During the operation, the light from light source is distributed and controlled by the monochromator to the wavelength required and passed to the sample. Then, the detector detects the amount of radiation that the sample transmits, reflects, interacts, or transfects. The signal is converted into a read out with the installed program in the form that the user can use (Figure 2.1). Finally, the NIR spectra are matched with the chemical analysis result of the same sample.

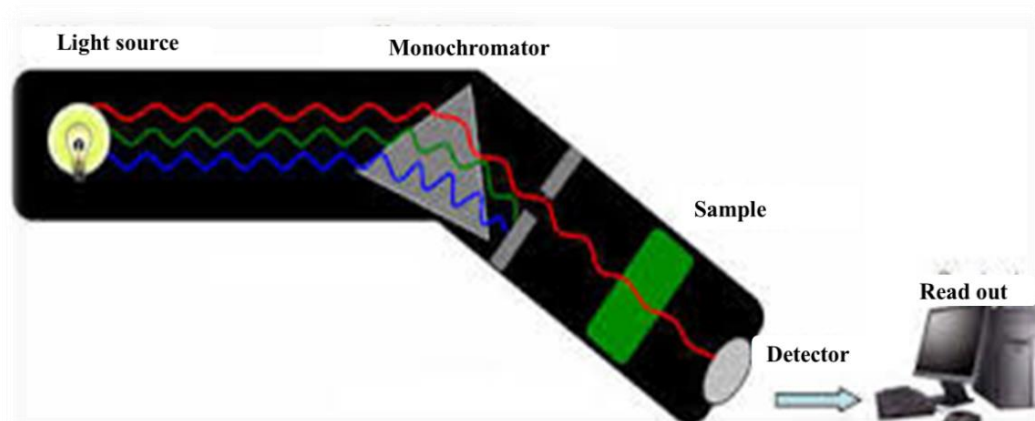


Figure 2.1 Components of the NIR spectrophotometer (Osborne *et al.*, 1993)

2.2.3. The steps to analyze NIRS

The analysis of NIRS data consists of two steps; first development of the calibration equation and second validation of the generated equations, as shown in Figure 2.2.

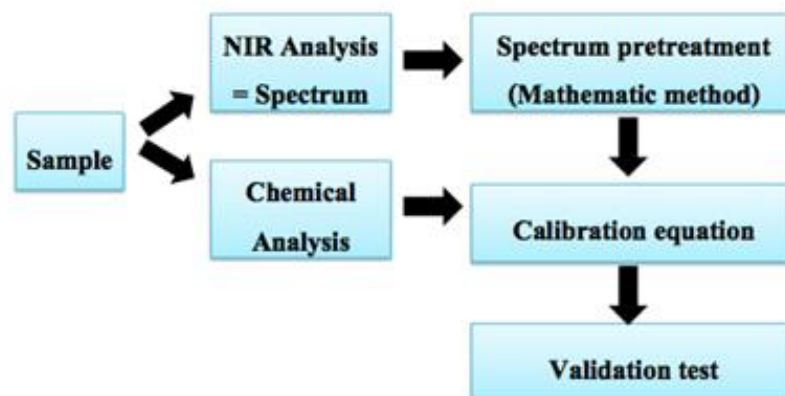


Figure 2.2 The steps to analyze NIRS

2.2.3.1 Development of the calibration equation

There are many steps to develop calibration equation. First, chemical analyses and NIRS analysis is carried out using the same set of samples. All analyses are performed at proximate time. Next, the spectra are pretreated so as to reduce the error of predicted value from the equation. Pretreatment methods can be one of the following: No treatment, Moving Average Smoothing, Gaussian Filter Smoothing,

Median Filter Smoothing, Savitzky–Golay Smoothing, Normalization, Gap Derivative, Gap Segment Derivative, S Golay 1st Derivative, S Golay 2nd Derivative, Baseline, and SNV. After that, the calibration equation is developed from NIR spectra (x axis) and dependent variables from standard values (y axis) together with appropriate regression in order to get the proper equation to predict qualitative unknown features.

2.2.3.2 Validation of the generated equations

To get precisely predicted quality value, it is necessary to validate the calibration equations in order to get an accurate equation. The accuracy of the equation depends on sufficient good sample quantity covering desired acceptable range of values, particularly the chemical composition of the sample. Therefore, sampling is a critical step in obtaining a good calibration equation to predict more accurate resulting from decrement of the error experimental value (Hruschka, 2001). Moreover, chemical analysis should also be considered as the standard method to get the right information because it has an effect on the accuracy of the created equation (Kawano, 1995).

2.2.4 Absorption process of Near infrared spectroscopy

NIR radiation in the electromagnetic wavelength ranges from 780 to 2,500 nm. When NIR is passed through sample, the samples' molecules will absorb the radiation. After that, bonds in the molecules will be vibrated at high frequencies. Each functional group of the molecules vibrates at different wavelength, resulting in excited vibration level from ground state. The excited molecules, then, return to ground vibration level due to released energy in the form of heat absorbance based on the Beer-Lambert rule. The intensity of released light energy from the sample molecules is proportional to the amount of chemical composition in the sample (Osborne *et al.*, 1993).

2.2.4.1 Vibration of bonding

Vibration of the functional groups in molecules can be divided into 2 types; stretching and bending. Stretching of the bond is a vibration that leads to change in the length between the bounded atoms. It can be classified into two sub-types; symmetric and asymmetric. Bending or deformation is a vibration that causes a change in the bonding angle. There are four sub-types of bending; scissoring, rocking, wagging,

and twisting. All of these may occur in the same plane (in-plane) or in different planes (out-of-plane) as shown in Figure 2.3.

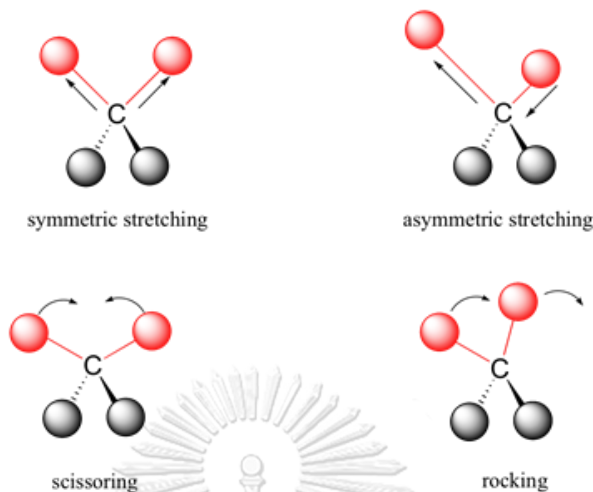


Figure 2.3 Changes in molecular bonds in various forms

The vibration of the bonding absorbs specific energy. Because there are many bonds in one molecule and each bonding pattern has many forms of vibration. As a result, the molecules will absorb NIR light at multiple wavelengths simultaneously that show in band or peak. Absorbance of NIR can be measured in two forms; transmittance and reflectance. Light intensity at each wavelength is shown at the horizontal axis and the vertical axis is the absorbance. The graph of the absorbance of the sample is called the NIR spectrum. Each molecule can absorb light at different wavelengths. Therefore, the spectrum is different. Organic compounds have X-H functional groups that can interact with NIR radiation and cause the molecules to vibrate from the ground vibration stage to excited stage. Consequently, overtone band and combination band are observed in the NIR spectra of organic compounds.

2.2.4.2 The absorbance process of NIRS

The absorbance process of NIRS involves the interaction between NIR and molecule which composes of hydrogen bonds (X-H), where X atom can be C, O, N, or S. In this interaction, molecules absorb NIR radiation and cause vibration of bond. The level of infrared absorption at different wavelengths will appear in the spectrum and can be further processed for quantitative and qualitative analyses.

Absorbance peak or band that appears in the NIR spectra is caused by absorption of energy. It results in the transition of

molecular energy from the ground vibration level to the excited vibration level.

The transition from $v = 0$ to $v = 2$ is called first overtone band.

The transition from $v = 0$ to $v = 3$ is called the second overtone band.

The transition from $v = 0$ to $v = 4$ is called the third overtone band.

The overtone bands have low intensity or low absorption compared to the peak. The first overtone band has higher intensity than the second overtone band and the third overtone band, respectively. The higher overtone bands cannot be observed because it has very low intensity (Karlberg, 2006). Figure 2.4 shows first overtone, second overtone, and third overtone in NIR spectrum. Table 2.4 shows positions of peak in the NIR spectrum relative to various functional groups in the sample. Table 2.5 shows the peak position of important components in the sample and Table 2.6 shows position of peak in the NIR spectrum that correlates with the components in the product.

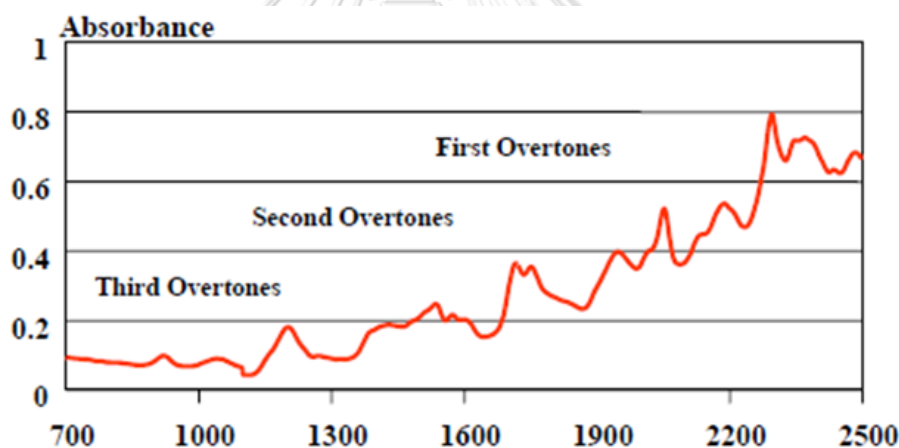


Figure 2.4 Overtones in NIR spectrum (Karlberg, 2006)

Table 2.4 Positions of peak in the NIR spectrum relative to the various functional groups in the sample (Osborne *et al.*, 1993)

Functional group	Band type	Wavelength range (nm)
H ₂ O (water)	Combination bands	1900–1950, 2250
H ₂ O (water)	1st Overtone	1400–1450
H ₂ O (water)	2nd and 3rd overtone	950–1000
R–NH ₂ (amino groups)	Combination bands	2150–2200
R–NH ₂ (amino groups)	1st Overtone	1450–1550
CHO (aldehyde group)	2nd and 3rd overtone	760–840, 970–1000
–COOH (carboxyl group)	Combination bands	2190–2220
–OH (hydroxyl groups)	Combination bands	1880–1910
–OH (hydroxyl groups)	Combination bands	2060–2090
–OH (hydroxyl groups)	1st Overtone	1410–1480
RCOOR' (ester groups)	2nd and 3rd overtone	920–945

Table 2.5 The peak position of important components in the sample (Shenk *et al.*, 2001)

Composition	Wavelength (nm)
Water (Moisture content)	1,940
Carbohydrate	2,100
Protein	2,180
Fat	2,310

Table 2.6 Position of peak in the NIR spectrum correlates with the components in the product

Wavelength (nm)	Bond vibration	Structure
1143	C-H str. 2 nd overtone	aromatic
1395	2xC-H str. + C-H def. combination	CH ₂
1410	O-H str. + 1 st overtone	ROH, oil
1440	O-H str. 1 st overtone	sucrose, starch
	C-H combination	CH ₂
1450	O-H str. 1 st overtone	starch, H ₂ O
1510	N-H str. 1 st overtone	protein
1520	O-H str. 1 st overtone	CONH ₂
	N-H str. 1 st overtone (intramol. H-bond)	ROH
1528	O-H str. 1 st overtone (intramol. H-bond)	starch
1540	O-H str. 1 st overtone (intramol. H-bond)	starch
1580	O-H str. 1 st overtone (intramol. H-bond)	starch, glucose
1725	C-H str. 1 st overtone	CH ₂
1765	C-H str. 1 st overtone	CH ₂
1900	O-H str. + 2xC-O str. combination	starch
1930	O-H str. + H-O-H def. combination	starch, cellulose
1940	O-H bend 2 nd overtone	H ₂ O
1960	O-H str. + O-H bend combination	starch
1980	N-H asym. str. + amide II* combination	protein, CONH ₂
2000	2x O-H def + C-O def. combination	starch
2050	N-H asym. str. + amide II* combination	protein
2055	N-H asym. str. + amide I* combination	protein
2060	N-H bend 2 nd overtone	protein
	N-H bend + N-H str. combination	protein
2070	O-H combination	oil
2080	O-H str. + O-H def. combination	ROH, sucrose, starch
2100	2xO-H str. + 2xC-O str. combination	starch
	C-O-O asym. str. 3 rd overtone	starch, cellulose
2132	N-H str. + C=O str. combination	amino acid
2140	C-H sym. def.	oil, NC=CH
2180	N-H bend 2 nd overtone	protein
	C-H str. + C=O str. combination	protein
	C=O str. + amide III* combination	protein
2252	O-H str. + O-H def. combination	starch
2276	O-H str. + C-C str. combination	starch
2300	C-H bend 2 nd overtone	protein
2310	C-H str. + C-H def. combination	CH ₂
	C-H bend 2 nd overtone	oil
2323	C-H str. + C-H def. combination	CH ₂ , starch
2380	C-H str. + C-C str. combination	oil
2461	C-H str. + C-C str. combination	starch
2470	C-N-C sym. str. 1 st overtone	protein
2488	C-H str. + C-C str. combination	starch, cellulose
2500	C-H str. + C-C str. combination	starch

str. = stretch, def. = deformation sym, sym. = symmetric, asym. = asymmetric amide I: C=O stretch. amide II: N-H in-plane bend, C-N stretch. amide III: C-N

Source: Osborne *et al.* (1993); Shenk *et al.* (2001); Williams and Norris (2001)

2.2.4.3 Patterns of interaction

There are four patterns of interaction, which are transmittance, reflectance, transfectance, and interactance.

1. Transmittance

Radiation impacts on one side of the sample and the detector measures the absorbance from the radiation passing through the sample on the opposite side, as shown in Figure 2.5 (a).

2. Transflectance

NIR radiates through the sample and impacts on non-absorbing object, such as ceramic, gold or aluminum sheets. The object then reflects the radiation back to the detector, as shown in Figure 2.5 (b).

3. Reflectance

Radiation impacts on the surface of the sample and may spread in a short time. Then, the detector measures the absorbance of the reflected radiation by, as shown in Figure 2.5 (c).

4. Interactance

This interaction occurs in the case of an optical fiber probe. The radiation comes from the outer ring of the probe and impacts on the sample. The reflected radiation is sent to the central detector of the optical fiber probe, as shown Figure 2.5 (d).

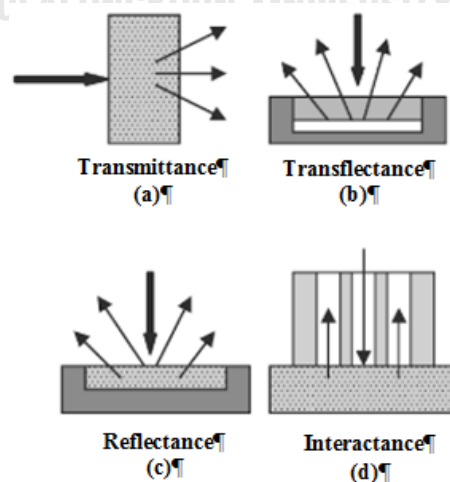


Figure 2.5 Patterns of NIR interaction (a) transmittance, (b) transflectance, (c) reflectance, and (d) interactance (Ozaki, 2006)

2.2.5 Application of NIRS in food detection

The NIR spectroscopy technique can be used to analyze the sample in both quantitative and qualitative ways in the agricultural and food products (Cen and He, 2007; Haughey *et al.*, 2013; Osborne *et al.*, 1993). A number of applications have been demonstrated; e.g. determination of total soluble solid and dry matter in mango fruit (Saranwong *et al.*, 2004), sugar content of grapefruit (Zude-Sasse *et al.*, 2002), soluble solids content, titratable acidity, and lycopene content of the tomatoes (Saad *et al.*, 2016), total dissolve solids and bulk density of pear (Liu *et al.*, 2008), total amino acids in oilseed leaves (Liu *et al.*, 2011), firmness and sugar content of apples (Lu *et al.*, 2000), fat nitrogen and moisture of cocoa powder (Veselá *et al.*, 2007), total anthocyanin content of red grape (Janik *et al.*, 2007), Epigallocatechin gallate, Epigallocatechin, Epicatechin gallate, Epicatechin of green tea (Chen *et al.*, 2009), reducing sugar and malvidin-3-glucoside of red wine (Fernández-Novales *et al.*, 2009), total phenolic compounds flavonoids content of coffee grounds (Páscoa *et al.*, 2013), the milk fat fatty acid profile of goats (Núñez-Sánchez *et al.*, 2016), and, lastly, dietary fibers extracted from lemon, orange and grapefruit seeds (Karaman *et al.*, 2017).

In rice grain, flour, starch and whole grains, NIRS had been widely used for the determination of rice quality such as starch and amylose content (Bao *et al.*, 2001), milled rice grades (Chen and Huang, 2010), amylose content in rice (Villareal *et al.*, 1994), moisture of paddy and brow rice (Kawamura *et al.*, 2003), moisture of wheat flour (Miralbés, 2004).

In fat or oil, NIRS had been widely used to determine oil content in plant such as sunflower oil (Picouet *et al.*, 2018), palm oil (Basri *et al.*, 2017), Chinese faba bean (Wang *et al.*, 2014), cocoa bean (Teye *et al.*, 2015), *Jatropha curcas* seed (Vaknin *et al.*, 2011), peanut seed oil (Bansod *et al.*, 2015) and rice bran (Bagchi *et al.*, 2016).

Delwiche *et al.* (1995) studied the use of NIR spectroscopy in reflectance mode in the wavelength range of 1,100-2,498 nm for determination of the protein content of several rice varieties. They showed that NIR spectra had good relationship with chemical values giving an R^2 of 0.98 and SEP of 0.1%. Delwiche *et al.* (1996) their subsequent study on the use of NIR spectroscopy in the wavelength range of 400-2498 nm for analysis of whole grain milled rice quality from 196 U.S. Rice samples showed that partial least square (PLS) was the most

suitable technique to determine amylose content in the range of 14 to 25% ($R^2 = 0.89$ and $SEP = 1.3\%$) and protein ($R^2 = 0.97$ and $SEP = 0.13\%$) in rice grain.

Himmelsbach *et al.* (2001) studied the use of fourier-transform NIR Raman spectroscopy in the wave number range 175-3,600 cm^{-1} for analysis of rice samples. They found that good relationship between NIR spectra vs. amylose in the range of 0.41-24.90% and NIR vs. protein in the range of 4.89-11.35% could be developed. The correlation between NIR and amylose content yielded R^2 , SEP, and bias of 0.985, 1.05%, and -0.006%, respectively, whereas the correlation between NIR and protein content yielded R^2 , SEP, and bias of 0.992, 0.138%, and -0.009%, respectively.

Williams and Sobering (1993) studied the use of NIR in transmittance and reflectance mode for determination of protein, oil, moisture content, and other properties in the several seeds. It was found that the use of both transmittance and reflectance NIRS technique could detect chemical composition of several seeds. But NIRS in the reflectance mode gave higher accuracy than the measurement in the transmittance mode. This study was consistent with Norris and Hart (1996) The measurement of NIRS in reflectance mode was popular for moisture analysis of grain and oil seed. Choosing the right measurement mode and the range of spectrum leads to appropriate and accurate analysis of certain data (Kawano, 1995).

Wang *et al.* (2006) used NIRS technique to determine fat content in brown rice grain, brown rice flour, milled rice grain, and milled rice flour. They found good relationship between NIR spectra vs. fat content. The result showed that R^2 of the calibration models were 0.79, 0.84, 0.89, and 0.91, respectively, and RMSE were 0.16, 0.14, 0.09, and 0.08%, respectively.

Bagchi *et al.* (2016) used NIR technique in the wavelength range of 400-2,500 nm to determine amylose and protein contents in 173 brown rice samples. Modification of partial least square (mPLSs) was used to develop calibration models. They found that using 173 brown rice samples gave calibration model with higher R^2 of 0.859 and 0.918 for amylose and protein contents, respectively. When using 86 rice bran samples, calibration models with lower R^2 of 0.567, 0.929, 0.552, 0.653, and 0.518 for protein, crude oil, moisture, ash, and fiber contents, respectively, were obtained.

Ferreira *et al.* (2013) studied the chemical composition of ground soybean using NIR technique in the reflectance mode in the wavenumber range of 4,000-10,000 cm^{-1} . Partial least square regression (PLSR) was used to develop calibration models. They found that R^2 of the calibration models for moisture, protein, fat, ash and carbohydrate content was 0.80, 0.81, 0.71, 0.63, and 0.50, respectively. Root Mean Square error of the prediction (RMSEP) for moisture, protein, fat, ash, and carbohydrate content was 1.55, 1.61, 1.20, 0.38, and 3.71, respectively. The following work of Ferreira *et al.* (2014) also studied the chemical composition of ground soybean using NIRS and Mid-infrared spectroscopy (MIRS) in the reflectance mode in the wavelength range of 1,250-2,500 nm and 2,500-25,000 nm, respectively. PLSR was used to develop calibration models. The result showed that both NIRS and MIRS could be used to predict the chemical composition of ground soybean. However, the use of NIRS could predict the fat content more accurately.

Finally, from the study of Fernández-Espinosa (2016), NIR spectra in the wavelength range from 1,000 to 2,300 nm in reflectance mode were selected to develop calibration models for moisture content, fat content, and acid value of olive fruits. PLSR was used to develop calibration models. The result showed that the R^2 of the calibration models for moisture content, fat content, and acid value was 0.88, 0.76, and 0.83, respectively. The RMSEP% of the calibration models for moisture content, fat content, and acid value was 4.98, 20.0, and 38.8, respectively.

Form the literature reviewed, determination of mixed rice bran and parboiled rice bran using NIR spectroscopy is still scarce. Therefore, as stated earlier, this research aimed to explore the possibility of using NIRS to determine the properties of mixed rice bran.

CHAPTER III

MATERIALS AND METHODS

3.1 Materials

3.1.1 Rice bran samples

Ten lots of freshly produced rice bran samples were collected from different rice mills in Phitsanulok, Phichit, Nakhon Sawan, Kamphaengphet, and Nakhon Pathom provinces. Table 3.1 shows the name of the rice mills and the area for each rice bran sample.

Table 3.1 Rice mill area for rice bran samples

Name of rice mill	Rice mill area
Phraya Phatthanarak Co., Ltd.	Phitsanulok province
Phichit Si Kasat Rice Co., Ltd.	Phichit province
Phichit Rom Charoen 2 Rice Co., Ltd.	Phichit province
Somthai Aree Chop Charoen Limited Partnership.	Phichit province
Kaoliao Pechpon Limited Partnership.	Nakhon Sawan province
Banphot Phisai Pechpon Rice Mill.	Nakhon Sawan province
Sawang Panit Rice Mill.	Khamphangphet province
Sai-ngam Charoen Chop Rice Mill.	Nakhon Pathom province

3.1.2 Parboiled rice bran samples

Ten lots of freshly produced parboiled rice bran samples were obtained from different rice mills in Phichit, Chainat, and Nakhon Sawan provinces. Table 3.2 shows the name of the rice mills and the area for each parboiled rice bran sample.

Table 3.2 Rice mill area for parboiled rice bran samples

Name of rice mill	Rice mill area
Pongpirod Rice Co., Ltd.	Phichit province
Sai-ngam Rice New Rice Mill.	Phichit province
Siam Tanyachat Co., Ltd.	Chai Nat province
Rongsifi Kasat Khathai Limited Partnership.	Chai Nat province
Kaew Sawang Rice Mill.	Chai Nat province
Kelyrti Khun Rice Mill. Limited Partnership.	Nakhon Sawan province
Rongsifi Damrong Charoen Chap Limited Partnership	Nakhon Sawan province
Sathit Sri Thong Rice Mill.	Nakhon Pathom province

3.2 Methods

3.2.1 Sample preparation

In each lot, freshly produced rice bran and parboiled rice bran were sieved through No. 20 mesh strainer (841 microns) to remove the husk and broken rice. The sample was then vacuum-packed in laminated aluminum foil bags (23 cm x 30 cm) and kept in a freezer until further analyses. Just before the analyses, rice bran and parboiled rice bran samples were mixed in 11 proportions ranging from 0: 10 to 10: 0 ratios (rice bran: parboiled rice bran).

3.2.2 Chemical analysis

The mixed rice bran samples were analyzed for chemical compositions as following:

3.2.2.1 Moisture content was determined following the method in AOAC (2012). The method is elaborated in appendix A.1.

3.2.2.2 Protein content was conducted using Kjeldahl method following the method in AOAC (2012). The method is elaborated in appendix A.2.

3.2.2.3 Fat content was determined by following the method in AOAC (2012). The method is elaborated in appendix A.3.

3.2.2.4 Crude fiber content was determined following the method in AOAC (2012). The method is elaborated in appendix A.4.

3.2.2.5 Acid value was determined following the method in AOCS (2004). The method is elaborated in appendix A.5.

3.2.2.6 Phytic acid content was determined following the method in K-PHYT (11/15, Megazyme, Ireland). The method is elaborated in appendix A.6.

The analyses were conducted in two replications.

3.2.3 Physical properties analysis

The mixed rice bran samples were analyzed for physical properties as following:

3.2.3.1 Color of the sample was measured using a Chroma meter (model CR-300 series, Konica Minolta, Tokyo, Japan) Hunter Color System (L^* , a^* and b^*). The whiteness index (WI) was calculated from L , a , and b using equation (1). The method is elaborated in appendix A.7.

$$\text{Whiteness index} = 100 - [(100 - L)^2 + a^2 + b^2]^{0.5} \quad (1)$$

Ten measurements were carried out per replicate.

3.2.3.2 Bulk density was determined following the method modified from Caparino *et al.* (2012). The method is elaborated in appendix A.8. Ten measurements were carried out per replicate.

The experiment was conducted in two replications.

3.2.4 FT-NIR analysis

The rice bran samples were analyzed using FT-NIR spectrometer (FT-NIR MPA, Bruker, Germany) in the diffuse-reflectance mode. Forty (40) g sample was filled in a quartz sample holder and scanned at 25 °C. All spectra were collected in the wavenumber range of 12,500 to 3,600 cm^{-1} (resolution: 16 cm^{-1} , number of sample scan: 32 scans). Six (6) spectra were collected on each sample. The spectra were then averaged to produce a single spectrum for each sample.

3.2.5 Chemometric analysis of FT-NIR data

3.2.5.1 Predictive model construction

The measured data and spectra were separated into 2 groups; the first group for calibration model (n=165) and the second group for internal validation (n=55). The ratio of calibration samples to validation samples was 3: 1, in which the minimum and maximum values were in the calibration samples. It means that the range of values for the validation set fell within the calibration set range for all parameters. Spectra were pre-treated with 12 methods which are Raw spectrum, Moving Average Smoothing, Gaussian Filter Smoothing, Median Filter Smoothing, Savitzky–Golay Smoothing, Normalization, Gap Derivative, Gap Segment Derivative, S Golay 1st Derivative, S Golay 2nd Derivative, Baseline, and SNV. Partial least square regression (PLS) was used to develop chemometric models using the Unscrambler-® X version 10.3 software package (CAMO, Norway) with full-spectrum analysis methods. Model performance was reported as the coefficient of determination (R^2) and Root Mean Square Error of Calibration (RMSEC) with each term calculated on the calibration set, Root Mean Square Error of Prediction (RMSEP), and bias (the average difference between modeled and reference values). The optimal model with lower RMSEC and higher R^2 was used to predict the sample properties in the validation set.

3.2.5.2 External validation

The external samples; ten rice bran samples and ten parboiled rice bran samples, were obtained from Thai Ruam Jai Vegetable Oil Co., Ltd. Ten mixed rice bran samples were prepared from rice bran and parboiled rice bran. All samples were subjected to NIRS analysis as well as analyses of chemical composition and physical properties as detailed in 3.2.2-3.2.4. All data from this set of samples (n=60) was used to validate the calibration model. The model performance was evaluated using the ability to predict from R^2 and RMSEP.

3.2.6 Statistical analysis

The data were analyzed by analysis of variance (ANOVA) with significance at $p \leq 0.05$. Duncan's multiple range tests (DMRT) were carried out for mean comparison. All statistical analyzes were performed using SPSS software (version 17, SPSS Inc., Chicago, USA).

CHAPTER IV

RESULTS AND DISCUSSION

4.1 Chemical composition and physical properties of mixed rice bran

Proximate compositions of 220 rice bran samples are shown in table 4.1. It was found that the major component in rice bran is fat which accounted for 19.2-31.8 % db, followed by protein content (14.7-18.4 % db), fiber content (9.1-14.0 % db), and moisture content (8.5-10.9 % wb). Rice bran also had the acid value and phytic acid in the 3.1-28.1 mg KOH/g oil and 4.8-8.2 g/100g bran, respectively. All rice bran samples had untapped bulk density in the 0.19-0.29 g/ml range, tapped bulk density in the 0.29-0.41 g/ml range, and whiteness index of 47.3-55.0.

Table 4.1 Chemical compositions and physical properties of 220 rice bran samples

Parameters	Range	Mean	SD
Moisture content (% wb)	8.5-10.9	9.8	0.4
Protein content (% db)	14.7-18.4	16.1	0.7
Fat content (% db)	19.2-31.8	25.4	2.3
Fiber content (% db)	9.1-14.0	11.6	1.2
Acid value (mg KOH/g oil)	3.1-28.1	12.1	6.2
Phytic acid (g/100g bran)	4.8-8.2	6.7	0.7
Bulk density (tapped) (g/ml)	0.29-0.41	0.36	0.02
Bulk density (untapped) (g/ml)	0.19-0.29	0.23	0.02
Whiteness index	47.3-55.0	50.5	1.6

Note: SD = standard deviation

Tables 4.2 to 4.10 show the chemical compositions and physical properties of 220 samples of rice bran samples.

4.1.1 Fat content

Table 4.2 shows the results of fat analysis. It was found that, from 10 lots of samples, rice bran had 22.4–25.5 % fat content whereas parboiled rice bran had 19.3–31.5 % fat content. It was observed that 8 out of 10 lots of parboiled rice bran contained higher fat content than rice bran. However, it excludes the parboiled rice bran sample in lot 4 and 5 which showed lower fat content than rice bran sample. This phenomenon could be due to the production of parboiled rice bran which involves soaking process before the steaming step. The long soaking process might cause fat to diffuse to the rice bran (Bhattacharya, 2004) and resulted in high fat content in the bran of parboiled rice. On the other hand, if soaking time is short, fat could diffuse to the outside part lesser. Parboiled rice bran from lot 4 and 5 could possibly come from the parboiling process with short soaking time. When rice bran was mixed with parboiled rice bran in various proportions, fat content decreased when rice bran proportions increased. In the research of Orthoefer (1996), it was illustrated that the fat content of parboiled rice bran increased 4-5 % when compared to that of rice bran.

4.1.2 Acid value

Table 4.3 shows the acid value of mixed rice bran samples. It was found that the samples in lot 1 to lot 10 had acid value in the 7.1–28.1 mg KOH/g oil range for rice bran and 3.1–5.4 mg KOH/g oil range for parboiled rice bran. Acid value is one of the key factors used to determine the quality of rice bran that is associated with its oil yield. In rice bran, lipase; a fat hydrolyzing enzyme, is usually present. The factors that affect lipase activity are storage temperature and time. Higher temperature can accelerate the reaction of lipase enzyme (Itthisoponkul, 2000). According to previous studies, the acid value of rice bran oil was higher than that of the oil from parboiled rice bran. Because the lipase enzyme in rice bran that hydrolyzes fat in the bran, which creates free fatty acids and glycerol, is destroyed during the parboiling process of rice. However, the results showed that, the acid value of rice bran in lot 9 and 10 was lower than other lots. It could be due to very efficient and timely management of the sample that did not facilitate the action of lipase enzyme. Thus, lower acid value of rice bran in lot 9 and 10 was observed. As expected, the acid value of mixed rice bran increased when rice bran proportion increased in every lot of samples. In the research of

Aizono (1973) who studied the characterization of lipase from rice bran of *Oryza sativa* (Japonica), the optimum temperature and pH of lipase were found to be 37°C and 7.5-8.0, respectively. The enzyme was reported to be destroyed when heated at 60°C for 15 minutes.

4.1.3 Phytic acid

Table 4.4 shows the phytic acid content of mixed rice bran samples. It was found that the samples in lot 1 to lot 10 had phytic acid in the 6.6-8.1 g/100 g bran range for rice bran and 4.8-6.3 g/100 g bran range for parboiled rice bran. Phytic acid is present in plants, especially in grains and beans. When phytic acid is bound with minerals, it is called phytate. Phytic acid and phytate can be degraded by heat. Thus, it can be used as an indicator for heat treatment in bran (Reddy *et al.*, 1982). In the research of Tanjor *et al.* (2016), it was stated that the phytate content was significantly decreased. In this study, the heat treatment can further reduce the amount of phytate by 28 percent. In the research of Lestienne *et al.* (2005), it was stated that the phytate content of cereals also decreased because phytase is activated and causes the hydrolysis of phytic acid during soaking process. Vidal-Valverde *et al.* (1994) reported that combined soaking and heating of lentils is more effective than soaking alone in reducing the level of phytic acid. In this study, the phytic acid content of rice bran was slightly higher than that of parboiled rice bran. When rice bran was mixed with parboiled rice bran in various proportions, phytic acid increased with increasing rice bran proportion.

Table 4.2 Fat content of mixed rice bran

Ratio of mixed rice bran (rice bran : parboiled rice bran)	Fat content (%db)									
	Lot 1	Lot 2	Lot 3	Lot 4	Lot 5	Lot 6	Lot 7	Lot 8	Lot 9	Lot 10
0:10	26.4±0.5 ^{Ae}	28.7±0.1 ^{Ac}	24.2±0.2 ^{Af}	19.3±0.2 ^{Fa}	23.4±0.1 ^{Fg}	29.9±0.7 ^{Ab}	25.9±0.1 ^{Ae}	27.6±0.3 ^{Ad}	31.5±0.1 ^{Aa}	27.8±0.2 ^{Ad}
1:9	26.1±0.2 ^{Ac}	28.5±0.1 ^{Abc}	23.8±0.0 ^{Abf}	20.9±0.6 ^{DEg}	23.8±0.0 ^{FFf}	29.5±0.0 ^{ABb}	25.6±0.1 ^{ABe}	27.5±0.0 ^{ABd}	31.6±0.2 ^{Aa}	27.5±0.1 ^{ABd}
2:8	26.2±0.3 ^{Ac}	27.6±0.3 ^{Cc}	23.7±0.1 ^{Bg}	21.2±0.1 ^{DEb}	24.0±0.1 ^{DEg}	28.8±0.2 ^{Bb}	25.3±0.2 ^{Bcf}	27.3±0.0 ^{ABcd}	30.7±0.0 ^{ABa}	26.8±0.4 ^{BCd}
3:7	25.8±0.3 ^{Ad}	28.1±0.2 ^{BCb}	23.5±0.1 ^{Bca}	20.6±0.1 ^{Ef}	24.2±0.2 ^{CDEe}	28.0±0.1 ^{Cb}	25.3±0.1 ^{Bce}	27.1±0.0 ^{ABc}	29.8±0.8 ^{Bca}	26.8±0.0 ^{Cd}
4:6	24.9±0.1 ^{Be}	27.5±0.1 ^{Cb}	23.5±0.2 ^{BCDg}	21.3±0.6 ^{Dh}	24.3±0.1 ^{CDf}	27.9±0.1 ^{Cb}	25.3±0.1 ^{BCDg}	27.0±0.1 ^{BCc}	29.5±0.3 ^{Ca}	26.2±0.1 ^{Cd}
5:5	24.9±0.3 ^{Be}	26.9±0.3 ^{Db}	23.1±0.1 ^{DEEg}	22.3±0.0 ^{Cg}	24.3±0.4 ^{CDa}	26.3±0.6 ^{DEb}	25.2±0.0 ^{CDd}	26.5±0.1 ^{CDbc}	28.8±0.1 ^{CDa}	25.4±0.1 ^{Dd}
6:4	24.6±0.2 ^{BCd}	26.4±0.2 ^{Db}	23.2±0.2 ^{CDEe}	22.2±0.1 ^{Cf}	24.5±0.1 ^{BCDd}	26.5±0.2 ^{Db}	25.0±0.1 ^{CDc}	26.4±0.1 ^{CDb}	28.4±0.0 ^{DEa}	24.4±0.3 ^{Ed}
7:3	23.9±0.6 ^{Ce}	25.5±0.3 ^{Be}	22.8±0.2 ^{EGFf}	23.1±0.1 ^{Bf}	24.8±0.1 ^{ABcd}	25.5±0.1 ^{FFbc}	24.9±0.1 ^{DEc}	26.1±0.4 ^{Db}	27.5±0.5 ^{FFa}	24.1±0.2 ^{Ed}
8:2	23.2±0.2 ^{Dde}	25.2±0.0 ^{Fb}	22.6±0.0 ^{BHe}	23.3±0.1 ^{Bde}	24.8±0.2 ^{ABb}	25.0±0.2 ^{Fb}	24.6±0.3 ^{FFbc}	26.1±0.0 ^{Da}	26.4±0.9 ^{Ca}	24.0±0.3 ^{Ed}
9:1	23.3±0.3 ^{CDde}	25.7±0.3 ^{Fa}	22.8±0.2 ^{FGe}	24.1±0.1 ^{Ac}	24.9±0.0 ^{ABb}	23.9±0.1 ^{Ccd}	24.5±0.2 ^{Fbc}	26.0±0.2 ^{Da}	25.7±0.8 ^{GHa}	23.1±0.4 ^{Fc}
10:0	22.7±0.0 ^{Ddef}	23.9±0.5 ^{Fcd}	22.4±0.1 ^{HFf}	24.1±0.4 ^{Ad}	25.1±0.2 ^{Aab}	23.5±0.4 ^{Gde}	24.3±0.0 ^{Fc}	25.5±0.4 ^{Fa}	24.7±0.1 ^{Hbc}	22.7±0.6 ^{Fef}

Note: The values in the table are shown in mean ± SD from the 2 replicates.

A, B, C... Mean values in a column with different superscripts are different significantly ($p \leq 0.05$)

a, b, c... Mean values in a row with different superscripts are different significantly ($p \leq 0.05$)

Table 4.3 Acid value of mixed rice bran

Ratio of mixed rice bran (rice bran : parboiled rice bran)	Acid value (mg KOH/ g oil)									
	Lot 1	Lot 2	Lot 3	Lot 4	Lot 5	Lot 6	Lot 7	Lot 8	Lot 9	Lot 10
0:10	3.4±0.0 ^{Ks}	3.7±0.0 ^{Lf}	4.9±0.1 ^{Lc}	5.2±0.2 ^{Kb}	4.4±0.1 ^{Ke}	3.8±0.1 ^{Kf}	5.4±0.0 ^{Ka}	3.1±0.1 ^{Kb}	5.1±0.0 ^{Hb}	4.6±0.1 ^{Ld}
1:9	5.5±0.0 ^{Lc}	6.1±0.0 ^{Lb}	6.2±0.1 ^{Hb}	7.2±0.0 ^{Ja}	6.2±0.1 ^{Lb}	5.3±0.1 ^{Ld}	7.2±0.0 ^{Ja}	4.7±0.1 ^{Lf}	5.5±0.1 ^{Gcd}	5.1±0.2 ^{Hs}
2:8	9.3±0.2 ^{Lb}	8.8±0.0 ^{Hc}	9.9±0.1 ^{Ga}	8.3±0.1 ^{Ld}	7.7±0.2 ^{Lc}	7.6±0.0 ^{Lc}	9.0±0.2 ^{Lc}	6.3±0.1 ^{Lf}	5.5±0.1 ^{Gs}	5.4±0.0 ^{Gs}
3:7	14.4±0.2 ^{Hs}	8.7±0.1 ^{Hd}	9.7±0.1 ^{Gc}	11.2±0.2 ^{Hb}	9.8±0.1 ^{Hc}	8.4±0.1 ^{Hc}	9.9±0.1 ^{Hc}	8.5±0.2 ^{Hde}	5.8±0.1 ^{Lf}	5.9±0.2 ^{Lf}
4:6	13.1±0.3 ^{Gb}	10.7±0.0 ^{Fd}	9.9±0.0 ^{Gc}	14.6±0.3 ^{Ga}	10.8±0.1 ^{Gd}	10.7±0.2 ^{Gd}	12.5±0.3 ^{Gc}	9.6±0.2 ^{Gc}	5.8±0.0 ^{Fs}	6.6±0.1 ^{Df}
5:5	16.4±0.4 ^{Fs}	10.2±0.1 ^{Gs}	13.8±0.3 ^{Fd}	15.9±0.1 ^{Fb}	12.9±0.1 ^{Fc}	13.1±0.4 ^{Fc}	14.9±0.3 ^{Fc}	11.8±0.2 ^{Ff}	6.0±0.1 ^{Lb}	6.2±0.1 ^{Lb}
6:4	18.2±0.4 ^{Es}	13.0±0.0 ^{Fd}	14.7±0.3 ^{Ec}	16.5±0.0 ^{Fb}	14.3±0.0 ^{Ec}	14.8±0.4 ^{Ec}	16.7±0.3 ^{Lb}	13.4±0.3 ^{Ld}	6.1±0.0 ^{DEc}	6.5±0.1 ^{DEc}
7:3	20.5±0.4 ^{Ds}	14.7±0.0 ^{Dc}	16.9±0.1 ^{Dc}	18.6±0.2 ^{Db}	15.9±0.3 ^{Dd}	16.5±0.4 ^{Dc}	18.4±0.3 ^{Db}	15.0±0.3 ^{Dc}	6.2±0.0 ^{Ds}	7.1±0.0 ^{Cf}
8:2	21.9±0.4 ^{Cs}	17.1±0.0 ^{Cd}	19.6±0.5 ^{Cb}	19.9±0.0 ^{Cb}	18.2±0.1 ^{Cc}	17.7±0.5 ^{Cd}	19.6±0.3 ^{Cb}	17.3±0.3 ^{Cd}	6.5±0.0 ^{Cf}	7.5±0.2 ^{Bc}
9:1	23.8±0.3 ^{Bs}	17.6±0.0 ^{Bs}	21.3±0.3 ^{Bc}	23.3±0.2 ^{Bs}	19.1±0.1 ^{Bc}	19.9±0.5 ^{Bd}	22.0±0.3 ^{Bb}	18.3±0.3 ^{Bf}	6.8±0.1 ^{Bb}	7.6±0.1 ^{Bi}
10:0	28.1±0.0 ^{As}	20.5±0.2 ^{As}	24.7±0.2 ^{Abc}	25.0±0.2 ^{Ab}	21.1±0.0 ^{As}	22.3±0.5 ^{Ad}	24.3±0.3 ^{Ac}	20.5±0.4 ^{As}	7.1±0.0 ^{As}	8.7±0.2 ^{Af}

Note: The values in the table are shown in mean ± SD from the 2 replicates.

A, B, C... Mean values in a column with different superscripts are different significantly ($p \leq 0.05$)

a, b, c... Mean values in a row with different superscripts are different significantly ($p \leq 0.05$)

Table 4.4 Phytic acid of mixed rice bran

Ratio of mixed rice bran (rice bran : parboiled rice bran)	Phytic acid (g/100g bran)									
	Lot1	Lot2	Lot3	Lot4	Lot5	Lot6	Lot7	Lot8	Lot9	Lot10
0:10	6.3±0.0 ^{6a}	6.3±0.1 ^{6a}	5.7±0.1 ^{1c}	5.1±0.1 ^{1e}	4.8±0.1 ^{1f}	6.3±0.1 ^{6a}	5.5±0.1 ^{7d}	6.2±0.1 ^{8a}	5.9±0.0 ^{9b}	5.4±0.1 ^{10d}
1:9	6.5±0.1 ^{7ab}	6.7±0.0 ^{7a}	5.9±0.1 ^{1Hc}	5.4±0.1 ^{1Hd}	5.1±0.1 ^{1He}	6.4±0.0 ^{6Gb}	5.8±0.2 ^{7e}	6.4±0.1 ^{8ab}	6.0±0.1 ^{9Ee}	5.6±0.0 ^{6d}
2:8	6.7±0.1 ^{8a}	6.6±0.1 ^{7a}	6.2±0.2 ^{9Hb}	5.5±0.1 ^{1Hde}	5.4±0.1 ^{1Ge}	6.6±0.0 ^{6Ea}	6.0±0.1 ^{7e}	6.7±0.0 ^{7a}	6.0±0.0 ^{9Ebc}	5.7±0.0 ^{6Gd}
3:7	6.7±0.0 ^{8b}	6.9±0.0 ^{7a}	6.3±0.1 ^{6d}	5.9±0.0 ^{6f}	5.8±0.0 ^{7a}	6.6±0.0 ^{6Ee}	6.4±0.0 ^{6d}	6.6±0.1 ^{8bc}	6.1±0.1 ^{9a}	5.9±0.1 ^{9Efs}
4:6	7.0±0.0 ^{9a}	7.0±0.2 ^{8a}	6.4±0.0 ^{6Gb}	6.4±0.1 ^{7b}	6.1±0.1 ^{7d}	6.9±0.1 ^{6a}	6.3±0.1 ^{6bc}	7.0±0.1 ^{8a}	6.2±0.0 ^{9cd}	5.9±0.1 ^{9Efa}
5:5	7.0±0.1 ^{9ab}	7.1±0.1 ^{8a}	6.7±0.1 ^{6Ee}	6.6±0.1 ^{6Ecd}	6.2±0.1 ^{7a}	6.9±0.1 ^{6b}	6.9±0.1 ^{6ab}	7.0±0.0 ^{8ab}	6.4±0.0 ^{9bd}	5.9±0.1 ^{9Efa}
6:4	7.3±0.1 ^{9ab}	7.3±0.1 ^{9a}	6.9±0.1 ^{6Ecd}	6.7±0.0 ^{6Ed}	6.8±0.1 ^{6d}	7.1±0.2 ^{6bc}	6.9±0.1 ^{6d}	7.3±0.0 ^{9ab}	6.4±0.1 ^{9Ca}	6.1±0.1 ^{9a}
7:3	7.3±0.1 ^{9b}	7.6±0.0 ^{9a}	7.2±0.2 ^{9Dd}	6.9±0.0 ^{6e}	6.8±0.1 ^{6c}	7.2±0.0 ^{6b}	7.3±0.2 ^{8b}	7.5±0.1 ^{9Dab}	6.6±0.1 ^{9Ac}	6.2±0.0 ^{9Dd}
8:2	7.5±0.0 ^{9ba}	7.7±0.0 ^{8Ca}	7.5±0.3 ^{9Cab}	7.2±0.0 ^{6cb}	7.2±0.1 ^{6b}	7.4±0.1 ^{8ab}	7.6±0.1 ^{9Aa}	7.6±0.1 ^{9Ca}	6.7±0.1 ^{9Ac}	6.3±0.1 ^{9Dcd}
9:1	7.6±0.1 ^{9ab}	7.9±0.1 ^{8a}	7.6±0.2 ^{9Aab}	7.5±0.2 ^{8b}	7.5±0.1 ^{8b}	7.5±0.1 ^{8b}	7.7±0.2 ^{9Aab}	7.8±0.1 ^{9Ab}	6.8±0.2 ^{9Ac}	6.4±0.1 ^{9ABd}
10:0	7.8±0.0 ^{9Ac}	8.1±0.1 ^{9Aa}	7.9±0.1 ^{9Abc}	7.9±0.1 ^{9Aa}	7.8±0.1 ^{9Ac}	7.7±0.0 ^{9Ac}	7.8±0.0 ^{9Aa}	8.0±0.0 ^{9Ab}	6.8±0.0 ^{9Ab}	6.6±0.1 ^{9Ac}

Note: The values in the table are shown in mean ± SD from the 2 replicates.

A, B, C... Mean values in a column with different superscripts are different significantly ($p \leq 0.05$)

a, b, c... Mean values in a row with different superscripts are different significantly ($p \leq 0.05$)

4.1.4 Protein content

Table 4.5 shows protein content of mixed rice brans. It was found that, from 10 lots of samples, rice bran had 14.7-15.2 % protein content while parboiled rice bran had 16.2-18.3 % protein content. In the steaming process of parboiled rice bran production, rice grains are partially cooked. Subsequent drying of partially cooked rice grain causes the grain to be stronger. As a result, the same degree of milling might result in parboiled rice bran with lower carbohydrate content, which yield parboiled rice bran with higher protein content. When rice bran was mixed with parboiled rice bran in various proportions, protein content decreased with increasing rice bran proportion.

4.1.5 Moisture content

Table 4.6 shows the results of moisture analysis. From 10 lots of samples, rice bran had 10.0-10.9 % moisture content whereas parboiled rice bran had 8.6-9.8 % moisture content. The drying step after the parboiling process caused parboiled rice bran to have lower moisture content than rice bran (Luh, 1991). The moisture content of both rice bran samples also varied in a narrow range. The moisture content of mixed rice bran increased with increasing rice bran proportion in every lot of samples, and still varied in a narrow range. This was beneficial to NIRS analysis as moisture consistency is one important factor that needs to be controlled (Ozaki, 2001).

4.1.6 Fiber content

Table 4.7 shows the fiber content of mixed rice bran samples. The samples in lot 1 to lot 10 had fiber content in the 9.2-10.0 % range for rice bran and 13.0-13.9 % ranges for parboiled rice bran. Steaming process of parboiled rice bran production causes rice grain to be stronger. Consequently, the same degree of milling might result in bran with lower carbohydrate content, which yield parboiled rice bran with higher fiber content. The fiber content of mixed rice bran decreased when rice bran proportion increased in every lot of experiments.

Table 4.5 Protein content of mixed rice bran

Ratio of mixed rice bran (rice bran : parboiled rice bran)	Protein content (%db)									
	Lot 1	Lot 2	Lot 3	Lot 4	Lot 5	Lot 6	Lot 7	Lot 8	Lot 9	Lot 10
0:10	17.2±0.0 ^{Acd}	17.0±0.2 ^{Acd}	17.1±0.2 ^{Acd}	18.3±0.2 ^{Aa}	17.5±0.1 ^{Ab}	17.0±0.1 ^{Ad}	17.3±0.1 ^{Abc}	16.7±0.1 ^{Ae}	16.4±0.2 ^{Af}	16.2±0.0 ^{ABf}
1:9	17.0±0.2 ^{Bcd}	16.8±0.0 ^{Ad}	17.2±0.2 ^{Abc}	17.8±0.1 ^{Ba}	17.4±0.0 ^{Ab}	16.9±0.1 ^{Ad}	17.0±0.1 ^{Bcd}	16.4±0.3 ^{Bde}	16.4±0.0 ^{Ae}	16.3±0.1 ^{Ae}
2:8	16.8±0.1 ^{Cb}	16.5±0.2 ^{Bc}	17.1±0.0 ^{Aa}	17.0±0.1 ^{CDa}	16.8±0.0 ^{Bb}	16.5±0.1 ^{Bcd}	16.6±0.0 ^{Bc}	16.3±0.1 ^{BCde}	16.3±0.1 ^{ABef}	16.1±0.1 ^{BCf}
3:7	16.4±0.1 ^{Dd}	16.2±0.1 ^{Cde}	16.6±0.1 ^{Bc}	17.2±0.1 ^{Ca}	16.9±0.1 ^{Bb}	16.4±0.0 ^{Bd}	16.3±0.0 ^{Dd}	16.3±0.0 ^{BCde}	16.1±0.0 ^{BCe}	15.9±0.0 ^{CDf}
4:6	16.5±0.1 ^{D,b}	16.1±0.1 ^{Cde}	16.3±0.1 ^{BCDcd}	16.9±0.3 ^{Da}	16.4±0.1 ^{Cbc}	16.4±0.0 ^{Bbc}	16.2±0.2 ^{D,bcd}	16.1±0.1 ^{CD,cd}	16.0±0.1 ^{CD,de}	15.8±0.2 ^{D,e}
5:5	16.1±0.1 ^{E,bcd}	16.1±0.2 ^{Cbc}	16.5±0.2 ^{BC,ab}	16.3±0.0 ^{E,ab}	16.2±0.2 ^{C,b}	16.1±0.0 ^{C,bcd}	15.8±0.2 ^{E,de}	16.0±0.1 ^{D,cd}	15.9±0.1 ^{DE,cd}	15.7±0.0 ^{DE,e}
6:4	15.9±0.0 ^{F, bc}	16.0±0.0 ^{CD,ab}	16.2±0.2 ^{CDa}	15.9±0.0 ^{F,bc}	16.2±0.1 ^{Ca}	15.7±0.1 ^{D,cd}	15.6±0.1 ^E	15.9±0.0 ^{D,bc}	15.8±0.1 ^{E,bcd}	15.6±0.1 ^{EF,de}
7:3	15.6±0.1 ^{G,cd}	15.7±0.0 ^{F, bc}	16.0±0.2 ^{DEa}	15.7±0.1 ^{F,bc}	15.6±0.1 ^{D,cd}	15.8±0.1 ^{D,bc}	15.8±0.0 ^{E,b}	15.6±0.1 ^{E,cd}	15.7±0.1 ^{EF,bc}	15.4±0.1 ^{FG,d}
8:2	15.6±0.0 ^{G,abc}	15.8±0.2 ^{DEa}	15.8±0.0 ^{E,ab}	15.3±0.1 ^{G,e}	15.6±0.2 ^{D,abc}	15.5±0.0 ^{F,cd}	15.3±0.0 ^{G,de}	15.5±0.0 ^{E,cd}	15.6±0.0 ^{F,bcd}	15.2±0.1 ^{GHe}
9:1	15.2±0.1 ^{H,bc}	15.3±0.1 ^{F,b}	15.4±0.1 ^{F,ab}	14.8±0.1 ^{H,d}	15.6±0.1 ^{Da}	15.4±0.1 ^{F,ab}	15.0±0.1 ^{H,c}	15.2±0.0 ^{F,de}	15.2±0.1 ^{G,bc}	15.2±0.1 ^{GHe}
10:0	15.2±0.0 ^{H,ab}	15.0±0.0 ^{G,de}	15.2±0.1 ^{Fa}	14.7±0.0 ^{H,g}	15.1±0.1 ^{E,bcd}	15.2±0.1 ^{G,abc}	14.7±0.0 ^{H,g}	14.8±0.1 ^{G,ef}	15.0±0.1 ^{H,cd}	15.1±0.1 ^{H,bcd}

Note: The values in the table are shown in mean ± SD from the 2 replicates.

A, B, C... Mean values in a column with different superscripts are different significantly ($p \leq 0.05$)

a, b, c... Mean values in a row with different superscripts are different significantly ($p \leq 0.05$)

Table 4.6 Moisture content of mixed rice bran

Ratio of mixed rice bran (rice bran : parboiled rice bran)	Moisture content (%wb)									
	Lot1	Lot2	Lot3	Lot4	Lot5	Lot6	Lot7	Lot8	Lot9	Lot10
0:10	8.6±0.1 ^{Hi}	9.2±0.1 ^{Fcd}	8.9±0.0 ^{He}	9.8±0.0 ^{Fa}	9.6±0.0 ^{Fab}	9.1±0.1 ^{Fd}	9.3±0.0 ^{Hi}	9.1±0.2 ^{Hd}	9.2±0.0 ^{Scd}	9.5±0.0 ^{Fb}
1:9	8.8±0.1 ^{Ge}	9.2±0.1 ^{Fcd}	9.2±0.2 ^{Gcd}	9.9±0.1 ^{Fa}	9.7±0.0 ^{Fb}	9.3±0.1 ^{Fcd}	9.3±0.0 ^{Hi}	9.1±0.0 ^{Hd}	9.4±0.0 ^{Fc}	9.6±0.0 ^{FFb}
2:8	8.9±0.1 ^{Ge}	9.4±0.1 ^{Ed}	9.4±0.0 ^{Fd}	10.0±0.0 ^{FFa}	9.7±0.1 ^{FFb}	9.5±0.0 ^{Fcd}	9.6±0.1 ^{Gbc}	9.4±0.2 ^{Gd}	9.4±0.1 ^{FFd}	9.7±0.0 ^{Dbc}
3:7	9.1±0.0 ^{Fe}	9.5±0.0 ^{Ed}	9.5±0.1 ^{FFd}	10.1±0.0 ^{DEa}	9.8±0.0 ^{DEbc}	9.6±0.1 ^{DEd}	9.9±0.1 ^{Fab}	9.6±0.2 ^{Fcd}	9.5±0.1 ^{Ed}	9.7±0.1 ^{DEcd}
4:6	9.3±0.1 ^{FFf}	9.5±0.1 ^{Ed}	9.5±0.0 ^{FFe}	10.2±0.1 ^{Da}	9.8±0.0 ^{DEc}	9.5±0.1 ^{Ed}	10.0±0.0 ^{FFb}	9.7±0.1 ^{FFcd}	9.6±0.1 ^{Dde}	9.8±0.0 ^{Cc}
5:4	9.4±0.1 ^{DEe}	9.6±0.1 ^{DEde}	9.6±0.2 ^{DEcde}	10.4±0.0 ^{Ca}	9.8±0.1 ^{DEb}	9.7±0.1 ^{CDbcd}	10.2±0.0 ^{DEa}	9.7±0.1 ^{FFb}	9.6±0.0 ^{Dcde}	9.8±0.0 ^{Cbc}
6:4	9.6±0.1 ^{CDe}	9.7±0.0 ^{CDde}	9.8±0.0 ^{DEe}	10.3±0.1 ^{Ca}	9.9±0.0 ^{CDbc}	9.8±0.1 ^{BCDcde}	10.4±0.2 ^{CDa}	10.0±0.0 ^{DEb}	9.7±0.0 ^{Cde}	9.8±0.1 ^{Cbed}
7:3	9.7±0.1 ^{Cd}	9.8±0.2 ^{Ccd}	9.8±0.1 ^{CDcd}	10.5±0.0 ^{Bab}	10.0±0.1 ^{BCc}	9.8±0.1 ^{BCcd}	10.6±0.1 ^{Ba}	10.3±0.1 ^{BCb}	9.7±0.0 ^{Cd}	10.0±0.0 ^{Bc}
8:2	9.8±0.0 ^{BCf}	9.9±0.1 ^{BCef}	9.9±0.0 ^{BCef}	10.6±0.1 ^{Ba}	10.0±0.0 ^{ABcd}	9.9±0.0 ^{ABde}	10.4±0.1 ^{CDb}	10.2±0.1 ^{CDc}	9.9±0.0 ^{Bef}	10.0±0.1 ^{Bde}
9:1	9.9±0.1 ^{ABb}	10.0±0.0 ^{ABb}	10.0±0.1 ^{ABb}	10.6±0.0 ^{Ba}	10.0±0.0 ^{ABCb}	10.0±0.1 ^{Ab}	10.5±0.1 ^{BCa}	10.5±0.2 ^{ABa}	9.9±0.0 ^{ABb}	10.1±0.1 ^{Ab}
10:0	10.1±0.1 ^{Ac}	10.1±0.0 ^{Ac}	10.1±0.1 ^{Ac}	10.7±0.0 ^{Ab}	10.1±0.0 ^{Ac}	10.1±0.0 ^{Ac}	10.9±0.1 ^{Aa}	10.7±0.1 ^{Ab}	10.0±0.0 ^{Ac}	10.1±0.0 ^{Ac}

Note: The values in the table are shown in mean ± SD from the 2 replicates.

A, B, C... Mean values in a column with different superscripts are different significantly ($p \leq 0.05$)

a, b, c... Mean values in a row with different superscripts are different significantly ($p \leq 0.05$)

Table 4.7 Fiber content of mixed rice bran

Ratio of mixed rice bran (rice bran : parboiled rice bran)	Fiber content (%)									
	Lot 1	Lot 2	Lot 3	Lot 4	Lot 5	Lot 6	Lot 7	Lot 8	Lot 9	Lot 10
0:10	13.0±0.1 ^{A,d}	13.6±0.2 ^{A,abc}	13.2±0.1 ^{A,c,d}	13.0±0.1 ^{A,d}	13.8±0.3 ^{A,ab}	13.2±0.3 ^{A,c,d}	13.4±0.1 ^{A,c,d}	13.9±0.1 ^{A,a}	13.4±0.2 ^{A,b,c,d}	13.8±0.2 ^{A,ab}
1:9	12.8±0.1 ^{A,c,d}	13.1±0.3 ^{B,abc}	12.7±0.2 ^{B,c,d}	12.5±0.2 ^{B,d}	13.6±0.3 ^{A,a}	13.1±0.2 ^{A,abc}	13.0±0.1 ^{A,b,c,d}	13.4±0.1 ^{B,ab}	13.0±0.1 ^{B,b,c,d}	13.0±0.2 ^{B,b}
2:8	12.2±0.1 ^{B,b,c}	12.4±0.1 ^{C,abc}	12.1±0.2 ^{C,c}	12.2±0.3 ^{B,C,b,c}	12.7±0.1 ^{B,C,abc}	12.6±0.3 ^{B,abc}	12.1±0.1 ^{B,c}	12.7±0.3 ^{C,ab}	12.7±0.3 ^{B,abc}	13.0±0.3 ^{B,a}
3:7	12.0±0.0 ^{B,C,c}	12.0±0.1 ^{CD,c}	11.9±0.2 ^{CD,c}	11.8±0.2 ^{C,c}	13.0±0.1 ^{B,a}	12.2±0.3 ^{B,C,b,c}	11.8±0.3 ^{B,c}	12.6±0.3 ^{CD,ab}	11.9±0.1 ^{C,c}	12.7±0.3 ^{B,ab}
4:6	11.6±0.1 ^{D,d}	12.3±0.1 ^{CD,ab}	11.8±0.1 ^{CD,abcd}	11.9±0.1 ^{C,abcd}	12.3±0.2 ^{C,a}	11.8±0.3 ^{CD,abcd}	11.8±0.3 ^{B,b,c,d}	12.3±0.3 ^{CD,ab}	11.7±0.3 ^{CD}	12.3±0.3 ^{C,abc}
5:5	11.8±0.3 ^{CD,ab}	11.8±0.1 ^{D,ab}	11.7±0.2 ^{D,ab}	11.7±0.3 ^{C,ab}	11.8±0.3 ^{D,ab}	11.8±0.2 ^{CD,ab}	11.7±0.1 ^{B,ab}	12.2±0.2 ^{D,a}	11.4±0.3 ^{C,b}	11.5±0.1 ^{D,b}
6:4	11.7±0.1 ^{CD,a}	11.2±0.2 ^{E,b,c,d}	10.8±0.2 ^{E,d}	10.8±0.2 ^{D,d}	11.4±0.1 ^{DE,abc}	11.5±0.1 ^{D,ab}	11.0±0.3 ^{CD}	10.9±0.2 ^{E,d}	11.6±0.1 ^{C,ab}	11.1±0.1 ^{E,c,d}
7:3	10.5±0.1 ^{E,c}	10.6±0.3 ^{F,c}	10.5±0.1 ^{E,c}	11.1±0.2 ^{D,ab}	11.1±0.2 ^{E,ab}	10.8±0.1 ^{E,abc}	10.4±0.1 ^{D,c}	11.2±0.3 ^{F,a}	10.8±0.2 ^{D,abc}	10.7±0.2 ^{F,b,c}
8:2	10.8±0.0 ^{F,ab}	10.7±0.2 ^{F,abc}	10.5±0.1 ^{E,c,d}	10.2±0.2 ^{E,d}	10.5±0.2 ^{F,b,c,d}	10.4±0.1 ^{F,c,d}	10.2±0.2 ^{DE,d}	10.1±0.2 ^{F,d}	10.6±0.2 ^{D,abc}	10.9±0.1 ^{F,a}
9:1	10.6±0.2 ^{E,a}	10.4±0.1 ^{F,ab}	9.9±0.2 ^{F,c}	10.2±0.2 ^{F,abc}	9.9±0.2 ^{G,c}	10.3±0.1 ^{F,ab}	10.2±0.2 ^{DE,abc}	10.1±0.2 ^{F,b,c}	10.4±0.1 ^{DE,ab}	10.3±0.1 ^{FG,ab}
10:0	9.8±0.2 ^{F,ab}	9.5±0.1 ^{G,b,c,d}	9.5±0.2 ^{F,b,c,d}	9.7±0.2 ^{F,abc}	9.3±0.1 ^{H,c,d}	10.0±0.1 ^{F,a}	9.8±0.2 ^{F,ab}	9.2±0.1 ^{G,d}	10.0±0.2 ^{E,a}	9.9±0.1 ^{G,ab}

Note: The values in the table are shown in mean ± SD from the 2 replicates.

A, B, C, ... Mean values in a column with different superscripts are different significantly ($p \leq 0.05$)

a, b, c, ... Mean values in a row with different superscripts are different significantly ($p \leq 0.05$)

4.1.7 Color (whiteness index)

Table 4.8 shows color of the samples in term of whiteness index. In lot 1 to lot 10, it was found that, the samples had whiteness index in the 49.3-53.1 range for rice bran and 48.2-54.9 ranges for parboiled rice bran. It was observed that 9 out of 10 lots of parboiled rice bran had lower whiteness index; darker color, than rice bran. However, it excludes the parboiled rice bran sample in lot 4 which showed higher whiteness index than rice bran sample. Generally, parboiled rice bran has darker color than rice bran. This is because the production of parboiled rice bran involves soaking process in the steaming step. During soaking, pigments in the layer of rice husk may spread to the layer of bran and causes the color of the bran to be darker. Moreover, Maillard reaction can occur in the heat treatment process leading to dark color development in the bran (Luh, 1991; Pillaiyar, 1990; Refai *et al.*, 1967). On the other hand, if soaking time is short, the pigments of husk could diffuse to the inside part lesser, resulting in lighter color. Parboiled rice bran from lot 4, with lighter color, could possibly come from the parboiling process with short soaking time. The result was consistent with the amount of fat content of parboiled rice bran from the same lot. When rice bran was mixed with parboiled rice bran in various proportions, whiteness index increased when rice bran proportions increased.

4.1.8 Bulk density

Tables 4.9 and 4.10 show the tapped bulk density and untapped bulk density of mixed rice bran samples. The result showed that, from 10 lots of samples, the samples had bulk density (tapped) in the 0.34-0.41 g/ml range for rice bran and 0.29-0.37 g/ml range for parboiled rice bran. The bulk density (untapped) of mixed rice bran samples was slightly lower; 0.23-0.29 g/ml range for rice bran and 0.19-0.26 g/ml range for parboiled rice bran. Lower bulk density, both tapped and untapped, of parboiled rice bran was due to higher fat content. The result was consistent with the amount of fat content in all samples. The bulk density of mixed rice bran increased when rice bran proportion increased in every lot of samples. Our result is consistent with that reported by Barberand Benedito de Barber (1980) in that the bulk density of rice bran was about 0.2-0.4 g/ml.

Table 4.8 Color (whiteness index) of mixed rice bran

Ratio of mixed rice bran (rice bran : parboiled rice bran)	Whiteness index									
	Lot 1	Lot 2	Lot 3	Lot 4	Lot 5	Lot 6	Lot 7	Lot 8	Lot 9	Lot 10
0:10	48.3±0.1 ^g	49.0±0.0 ^{ff}	51.2±0.0 ^{cc}	54.9±0.1 ^{aa}	52.5±0.0 ^{bb}	49.5±0.2 ^{ga}	50.4±0.2 ^{gd}	48.2±0.2 ^{gs}	47.4±0.2 ^{ha}	50.2±0.0 ^{ed}
1:9	48.4±0.2 ^{DEf}	49.0±0.0 ^{fe}	51.3±0.1 ^{cc}	54.2±0.3 ^{ba}	52.5±0.1 ^{bb}	49.3±0.0 ^{he}	50.4±0.2 ^{fgd}	48.3±0.0 ^{fgf}	47.5±0.1 ^{fs}	50.5±0.0 ^{DEd}
2:8	48.4±0.5 ^{DEg}	49.1±0.0 ^{ff}	51.6±0.2 ^{cc}	54.0±0.1 ^{ba}	52.5±0.4 ^{bb}	50.0±0.2 ^{ge}	50.6±0.1 ^{ffgd}	48.4±0.2 ^{fgs}	47.7±0.1 ^{ffh}	50.6±0.1 ^{Dd}
3:7	48.5±0.3 ^{DEf}	49.1±0.1 ^{ee}	51.7±0.2 ^{cc}	53.4±0.5 ^{ca}	52.4±0.1 ^{bb}	50.5±0.2 ^{ffd}	50.7±0.1 ^{fd}	48.7±0.2 ^{ffef}	48.0±0.1 ^{DEg}	50.7±0.2 ^{Dd}
4:6	48.8±0.0 ^{DEef}	49.1±0.0 ^{fe}	51.7±0.1 ^{cc}	52.4±0.2 ^{dac}	52.3±0.3 ^{cab}	50.3±0.6 ^{fd}	51.1±0.3 ^{cdcc}	48.8±0.0 ^{DEff}	48.4±0.2 ^{cdf}	51.1±0.2 ^{cc}
5:5	48.4±0.1 ^{DEd}	49.2±0.0 ^{dc}	51.8±0.1 ^{cc}	51.8±0.1 ^{ca}	52.4±0.1 ^{bb}	51.6±0.6 ^{cdab}	51.0±0.2 ^{db}	48.9±0.3 ^{cdcc}	48.4±0.3 ^{cd}	51.1±0.1 ^{cb}
6:4	49.3±0.1 ^{CDc}	49.2±0.0 ^{dc}	51.9±0.0 ^{cc}	51.4±0.2 ^{ffh}	52.5±0.1 ^{bb}	51.1±0.1 ^{DEb}	51.3±0.1 ^{BCdb}	49.2±0.3 ^{BCcd}	48.9±0.0 ^{fd}	51.3±0.0 ^{cb}
7:3	49.6±0.0 ^{BCc}	49.3±0.0 ^{CDcd}	52.1±0.0 ^{aa}	51.5±0.4 ^{fb}	52.6±0.0 ^{BCb}	51.4±0.1 ^{CDb}	51.4±0.1 ^{BCdb}	49.3±0.1 ^{BCcd}	48.9±0.4 ^{ABd}	51.4±0.0 ^{BCb}
8:2	50.3±0.1 ^{ABd}	49.3±0.0 ^{BCa}	52.1±0.1 ^{ABa}	50.9±0.2 ^{fc}	52.7±0.3 ^{Bc}	52.1±0.1 ^{BCa}	51.5±0.2 ^{BCb}	49.3±0.4 ^{BCa}	49.0±0.2 ^{ABa}	51.3±0.4 ^{CBc}
9:1	50.7±0.9 ^{Ac}	49.4±0.0 ^{ABa}	52.3±0.0 ^{Ab}	50.3±0.1 ^{gcd}	52.7±0.4 ^{Bc}	52.7±0.3 ^{ABa}	51.6±0.0 ^{ABb}	49.6±0.0 ^{ABde}	49.4±0.2 ^{Ac}	51.7±0.2 ^{ABb}
10:0	51.2±0.7 ^{Ac}	49.4±0.0 ^{ABef}	52.4±0.0 ^{Ab}	49.8±0.3 ^{gef}	53.1±0.0 ^{Ab}	53.0±0.0 ^{Ab}	51.9±0.1 ^{Ab}	49.9±0.2 ^{Ac}	49.3±0.1 ^{ABf}	52.0±0.1 ^{Ab}

Note: The values in the table are shown in mean ± SD from the 2 replicates.

A, B, C... Mean values in a column with different superscripts are different significantly ($p \leq 0.05$)

a, b, c... Mean values in a row with different superscripts are different significantly ($p \leq 0.05$)

Table 4.9 Bulk density (tapped) of mixed rice bran

Ratio of mixed rice bran (rice bran : parboiled rice bran)	Bulk density (tapped) (g/ml)									
	Lot 1	Lot 2	Lot 3	Lot 4	Lot 5	Lot 6	Lot 7	Lot 8	Lot 9	Lot 10
0:10	0.32±0.00 ^{g,ef}	0.29±0.00 ^{H,s}	0.36±0.00 ^{F,b}	0.36±0.00 ^{l,b}	0.35±0.01 ^{H,c}	0.32±0.01 ^{G,f}	0.37±0.00 ^{G,a}	0.33±0.00 ^{G,d}	0.33±0.00 ^{G,de}	0.33±0.00 ^{G,d}
1:9	0.33±0.01 ^{G,d}	0.31±0.00 ^{G,f}	0.36±0.00 ^{EF,bc}	0.36±0.00 ^{HL,b}	0.35±0.00 ^{GH,c}	0.32±0.00 ^{G,e}	0.38±0.00 ^{FG,a}	0.34±0.00 ^{FG,d}	0.33±0.00 ^{FG,a}	0.33±0.00 ^{FG,d}
2:8	0.34±0.00 ^{F,d}	0.32±0.00 ^{F,b}	0.36±0.00 ^{E,c}	0.37±0.00 ^{GH,b}	0.36±0.00 ^{FG,bc}	0.33±0.00 ^{F,g}	0.38±0.00 ^{FG,a}	0.34±0.00 ^{EF,de}	0.33±0.00 ^{FG,g}	0.34±0.00 ^{EF,ef}
3:7	0.34±0.00 ^{EF,de}	0.33±0.01 ^{EF,f}	0.36±0.00 ^{E,bc}	0.37±0.00 ^{FG,b}	0.36±0.00 ^{FG,c}	0.33±0.00 ^{F,f}	0.38±0.00 ^{EF,a}	0.35±0.01 ^{DE,d}	0.33±0.00 ^{EF,f}	0.34±0.00 ^{DE,ef}
4:6	0.35±0.00 ^{DEF,e}	0.33±0.00 ^{E,b}	0.36±0.00 ^{D,cd}	0.37±0.00 ^{EF,b}	0.37±0.01 ^{EF,bc}	0.35±0.00 ^{E,f}	0.39±0.00 ^{DE,a}	0.36±0.00 ^{D,de}	0.33±0.00 ^{DE,gh}	0.34±0.00 ^{CD,fg}
5:5	0.36±0.00 ^{DE,cd}	0.35±0.00 ^{D,de}	0.36±0.00 ^{D,c}	0.38±0.00 ^{DE,b}	0.38±0.00 ^{DE,b}	0.35±0.00 ^{D,cd}	0.39±0.01 ^{DE,a}	0.36±0.01 ^{D,c}	0.33±0.00 ^{DE,f}	0.34±0.00 ^{C,ef}
6:4	0.36±0.00 ^{CD,c}	0.36±0.00 ^{C,c}	0.36±0.00 ^{CD,c}	0.38±0.00 ^{CD,b}	0.38±0.01 ^{DE,b}	0.35±0.00 ^{DE,d}	0.39±0.00 ^{BCD,a}	0.38±0.00 ^{C,b}	0.34±0.00 ^{CD,e}	0.34±0.00 ^{CD,de}
7:3	0.37±0.01 ^{BC,c}	0.36±0.00 ^{BC,d}	0.37±0.00 ^{BC,d}	0.38±0.00 ^{BC,c}	0.38±0.00 ^{CD,b}	0.36±0.00 ^{BC,d}	0.39±0.00 ^{BC,a}	0.37±0.01 ^{C,c}	0.34±0.00 ^{BC,e}	0.35±0.00 ^{C,a}
8:2	0.38±0.00 ^{AB,b}	0.37±0.00 ^{B,c}	0.37±0.00 ^{AB,c}	0.38±0.00 ^{AB,b}	0.40±0.00 ^{B,a}	0.37±0.00 ^{B,c}	0.40±0.00 ^{B,a}	0.39±0.00 ^{BC,b}	0.34±0.00 ^{AB,e}	0.35±0.00 ^{B,d}
9:1	0.39±0.01 ^{A,a}	0.37±0.00 ^{B,b}	0.37±0.00 ^{AB,b}	0.39±0.00 ^{AB,a}	0.39±0.00 ^{BC,a}	0.36±0.00 ^{C,bc}	0.39±0.00 ^{B,a}	0.39±0.01 ^{AB,a}	0.34±0.00 ^{BC,d}	0.35±0.00 ^{B,c}
10:0	0.40±0.00 ^{A,c}	0.39±0.00 ^{A,c}	0.37±0.00 ^{A,f}	0.39±0.00 ^{A,d}	0.41±0.00 ^{A,a}	0.38±0.00 ^{A,e}	0.40±0.00 ^{A,b}	0.40±0.00 ^{A,c}	0.34±0.00 ^{A,b}	0.36±0.00 ^{A,g}

Note: The values in the table are shown in mean ± SD from the 2 replicates.

A, B, C... Mean values in a column with different superscripts are different significantly ($p \leq 0.05$)

a, b, c... Mean values in a row with different superscripts are different significantly ($p \leq 0.05$)

Table 4.10 Bulk density (untapped) of mixed rice bran

Ratio of mixed rice bran (rice bran : parboiled rice bran)	Bulk density (untapped) (g/ml)									
	Lot 1	Lot 2	Lot 3	Lot 4	Lot 5	Lot 6	Lot 7	Lot 8	Lot 9	Lot 10
0:10	0.20±0.00 ^{D,f}	0.22±0.00 ^{F,de}	0.25±0.00 ^{F,b}	0.26±0.00 ^{F,a}	0.23±0.00 ^{H,c}	0.19±0.00 ^{F,sh}	0.22±0.00 ^{H,cd}	0.19±0.00 ^{H,b}	0.20±0.00 ^{E,s}	0.22±0.00 ^{F,e}
1:9	0.20±0.01 ^{D,d}	0.22±0.01 ^{F,c}	0.25±0.00 ^{F,b}	0.27±0.00 ^{E,a}	0.23±0.00 ^{GH,c}	0.21±0.00 ^{E,d}	0.23±0.00 ^{G,c}	0.20±0.01 ^{G,d}	0.20±0.00 ^{F,d}	0.22±0.00 ^{F,c}
2:8	0.21±0.01 ^{D,ef}	0.22±0.00 ^{EF,cd}	0.26±0.00 ^{F,b}	0.27±0.00 ^{E,s}	0.23±0.00 ^{FG,c}	0.21±0.01 ^{DE,f}	0.23±0.00 ^{EFG,c}	0.20±0.00 ^{FG,f}	0.21±0.00 ^{D,f}	0.22±0.00 ^{E,de}
3:7	0.21±0.00 ^{D,f}	0.23±0.00 ^{DE,c}	0.26±0.00 ^{DE,b}	0.28±0.00 ^{D,a}	0.23±0.00 ^{DE,d}	0.21±0.00 ^{CDE,f}	0.23±0.00 ^{FG,d}	0.21±0.00 ^{EF,f}	0.21±0.00 ^{D,f}	0.22±0.00 ^{E,a}
4:6	0.22±0.01 ^{C,de}	0.23±0.01 ^{DE,c}	0.26±0.00 ^{DE,b}	0.27±0.00 ^{DE,a}	0.23±0.00 ^{EF,cd}	0.21±0.01 ^{CDE,fg}	0.23±0.00 ^{EF,cd}	0.22±0.00 ^{DE,fg}	0.21±0.00 ^{D,s}	0.22±0.00 ^{D,def}
5:5	0.22±0.01 ^{C,ef}	0.24±0.00 ^{CD,c}	0.26±0.00 ^{D,b}	0.28±0.00 ^{D,a}	0.23±0.00 ^{C,de}	0.22±0.00 ^{BCD,fg}	0.24±0.00 ^{CD,cd}	0.22±0.00 ^{BC,ef}	0.22±0.00 ^{C,s}	0.22±0.00 ^{C,ef}
6:4	0.22±0.00 ^{C,ef}	0.24±0.01 ^{CD,c}	0.26±0.00 ^{C,b}	0.28±0.00 ^{D,a}	0.23±0.00 ^{CD,de}	0.22±0.00 ^{BC,f}	0.23±0.00 ^{DE,d}	0.22±0.00 ^{CD,ef}	0.22±0.00 ^{AB,ef}	0.22±0.00 ^{C,ef}
7:3	0.24±0.00 ^{B,d}	0.25±0.00 ^{BC,c}	0.26±0.00 ^{C,b}	0.28±0.00 ^{C,a}	0.23±0.00 ^{B,s}	0.23±0.00 ^{AB,ef}	0.24±0.00 ^{CD,d}	0.23±0.00 ^{BC,e}	0.22±0.01 ^{B,f}	0.22±0.00 ^{B,ef}
8:2	0.24±0.01 ^{AB,d}	0.25±0.00 ^{ABC,c}	0.26±0.00 ^{C,b}	0.29±0.00 ^{BC,a}	0.23±0.00 ^{AB,fg}	0.23±0.00 ^{A,ef}	0.24±0.00 ^{BC,de}	0.23±0.00 ^{B,ef}	0.22±0.00 ^{AB,s}	0.22±0.00 ^{B,s}
9:1	0.24±0.00 ^{AB,d}	0.26±0.01 ^{A,c}	0.27±0.00 ^{B,b}	0.29±0.00 ^{B,a}	0.23±0.00 ^{AB,f}	0.23±0.01 ^{A,de}	0.24±0.00 ^{AB,d}	0.24±0.01 ^{A,d}	0.23±0.00 ^{AB,f}	0.23±0.00 ^{A,f}
10:0	0.25±0.01 ^{A,c}	0.26±0.00 ^{AB,c}	0.27±0.00 ^{A,b}	0.29±0.00 ^{A,a}	0.23±0.00 ^{A,ef}	0.24±0.00 ^{A,e}	0.24±0.00 ^{A,d}	0.25±0.00 ^{A,d}	0.23±0.00 ^{A,ef}	0.23±0.00 ^{A,f}

Note: The values in the table are shown in mean ± SD from the 2 replicates.

A, B, C... Mean values in a column with different superscripts are different significantly ($p \leq 0.05$)

a, b, c... Mean values in a row with different superscripts are different significantly ($p \leq 0.05$)

□

4.2 Relationship between chemical composition, physical properties and ratios of mix rice bran

Table 4.11 and Figure 4.1 show the results on physical properties and chemical composition of mixed rice bran and parboiled rice bran. The chemical composition and physical properties of mixed rice bran strongly related with rice bran-to-parboiled rice bran ratio, except for moisture content and whiteness index that varied in a narrow range, with R^2 in the range of 0.98 to 0.99. Therefore, all properties can be used as predictors for rice bran content in mixed bran. However, larger standard deviations were observed for acid value followed by fat content. This was due greatly to the variations in acid value and fat content of raw rice bran that was affected by the condition of storage and transportation of the samples. Raw rice bran contains active enzymes that continue to work until the point of analyses. The enzyme's activity depends on storage temperature and time. Any discrepancy in both factors can cause the acid value and fat content to change. The standard deviation of acid value and fat content grew larger as the proportion of rice bran increased. On the contrary, the standard deviation for phytic acid content, which is one indicator for heat treatment of rice bran, was quite narrow. This is because the insensitive nature of the substance with regard to time and temperature of storage. With its strong dependency on rice bran-to-parboiled rice bran ratio ($R^2 = 0.9991$), phytic acid content can be used as a good predictor for rice bran content in mixed bran.

Table 4.11 Chemical composition and physical properties and ratios of mix rice bran

Ratio of mixed rice bran (rice bran : parboiled rice bran)	Moisture (%wb)		Protein (%db)		Fat (%db)		Fiber (%db)		Acid value (mg KOH/g oil)		Phytic acid (g/100g bran)		Bulk density (tapped) (g/ml)		Bulk density (untapped) (g/ml)		Whiteness index	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
0:10	9.2	0.4	17.1	0.6	26.5	3.5	13.5	0.3	4.4	0.8	5.7	0.5	0.3	0.0	0.2	0.0	50.3	2.4
1:9	9.3	0.3	16.9	0.5	26.5	3.1	13.0	0.3	5.9	0.8	6.0	0.5	0.3	0.0	0.2	0.0	50.2	2.2
2:8	9.5	0.3	16.6	0.3	26.2	2.7	12.5	0.3	7.8	1.6	6.1	0.5	0.3	0.0	0.2	0.0	50.4	2.1
3:7	9.6	0.3	16.4	0.4	25.9	2.7	12.2	0.4	9.2	2.5	6.3	0.4	0.4	0.0	0.2	0.0	50.4	1.8
4:6	9.7	0.3	16.3	0.3	25.7	2.4	12.0	0.3	10.4	2.7	6.5	0.4	0.4	0.0	0.2	0.0	50.4	1.5
5:5	9.8	0.3	16.1	0.2	25.4	1.9	11.7	0.2	12.1	3.7	6.7	0.4	0.4	0.0	0.2	0.0	50.4	1.5
6:4	9.9	0.3	15.9	0.2	25.2	1.8	11.2	0.3	13.4	4.1	6.9	0.4	0.4	0.0	0.2	0.0	50.6	1.2
7:3	10.0	0.3	15.7	0.2	24.8	1.4	10.8	0.3	15.0	4.7	7.1	0.4	0.4	0.0	0.2	0.0	50.6	1.2
8:2	10.0	0.3	15.5	0.2	24.5	1.2	10.5	0.3	16.5	5.2	7.3	0.4	0.4	0.0	0.2	0.0	50.7	1.1
9:1	10.2	0.3	15.2	0.2	24.4	1.1	10.2	0.2	18.0	6.0	7.4	0.5	0.4	0.0	0.2	0.0	50.8	1.2
10:0	10.3	0.3	15.0	0.2	23.9	1.1	9.7	0.3	20.2	6.9	7.6	0.5	0.4	0.0	0.2	0.0	50.9	1.3

Note: The values in the table are shown in mean \pm SD from the 10 lots per ration.

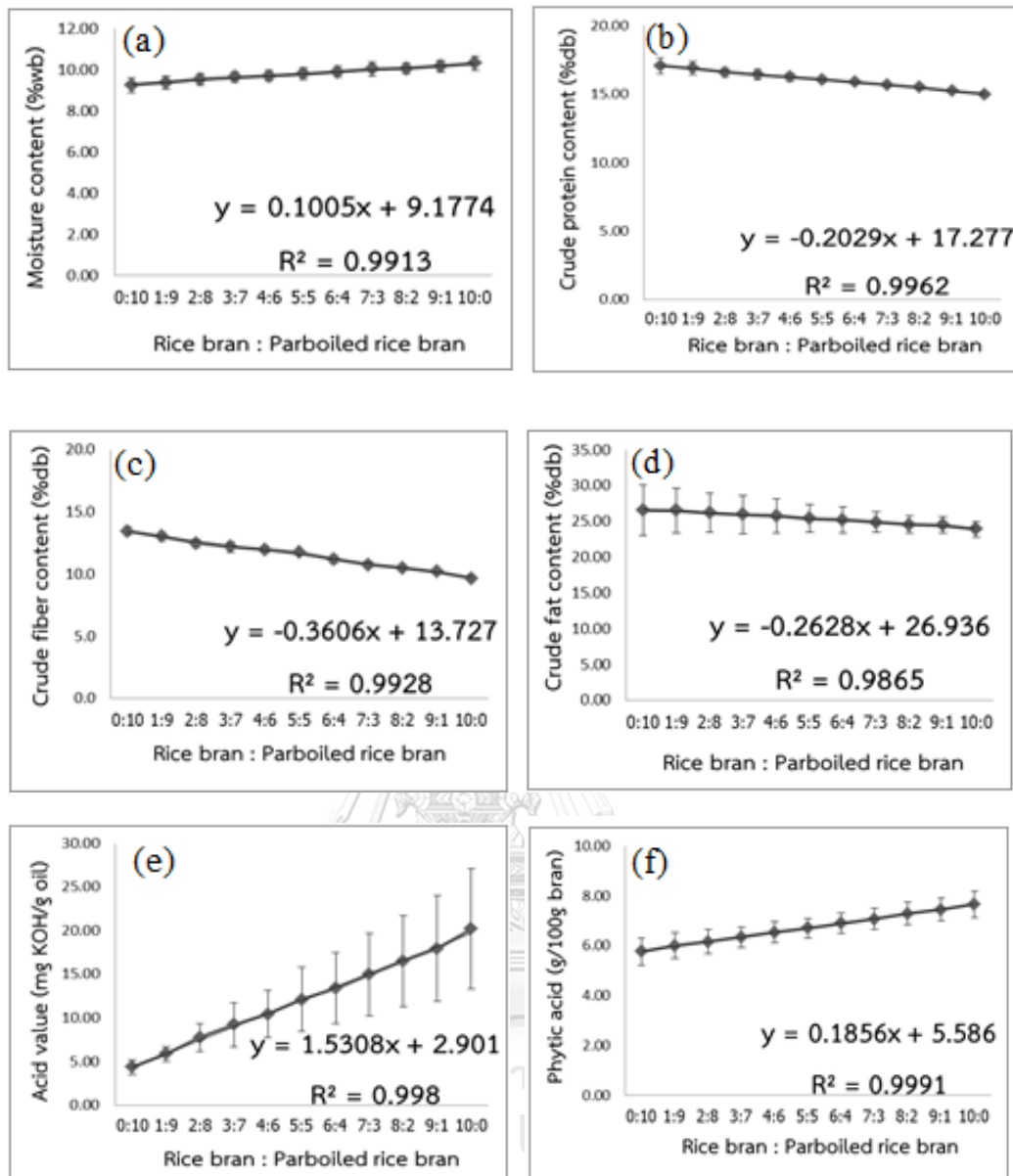


Figure 4.1 Regression plot between chemical composition and physical properties and ratios of mixed rice bran and parboiled rice bran (a) moisture content, (b) protein content, (c) fiber content, (d) fat content, (e) acid value, (f) phytic acid (g) bulk density (tapped), (h) bulk density (untapped), and (i) whiteness index

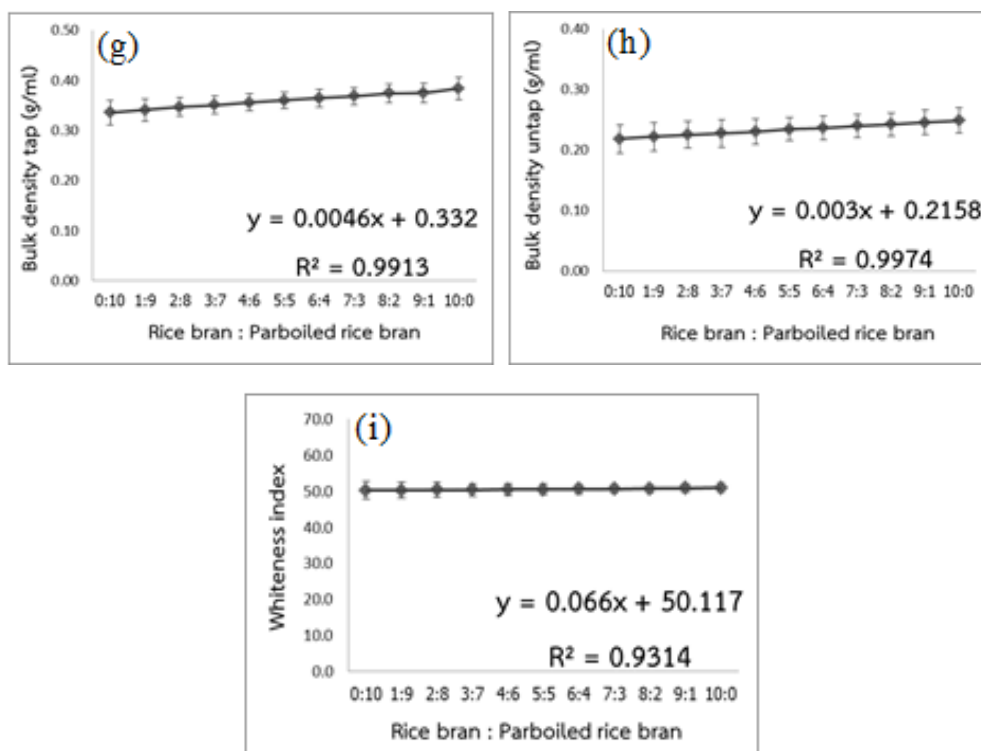


Figure 4.1 Regression plot between chemical composition and physical properties and ratios of mixed rice bran and parboiled rice bran (a) moisture content, (b) protein content, (c) fiber content, (d) fat content, (e) acid value, (f) phytic acid (g) bulk density (tapped), (h) bulk density (untapped), and (i) whiteness index (continued...)

4.3 FT-NIR analysis of mixed rice bran

4.3.1 FT-NIR spectra

The FT-NIR analysis of mixed rice bran samples were carried out for 220 rice bran samples. Sample spectra are presented in Figure 4.2 in the 3,600 to 12,500 cm^{-1} wavenumber region. The samples from the mixing between rice bran and parboiled rice bran in 11 proportions from 0: 10 to 10: 0 ratio were analyzed using FT-NIR spectrometer in the diffuse-reflectance mode.

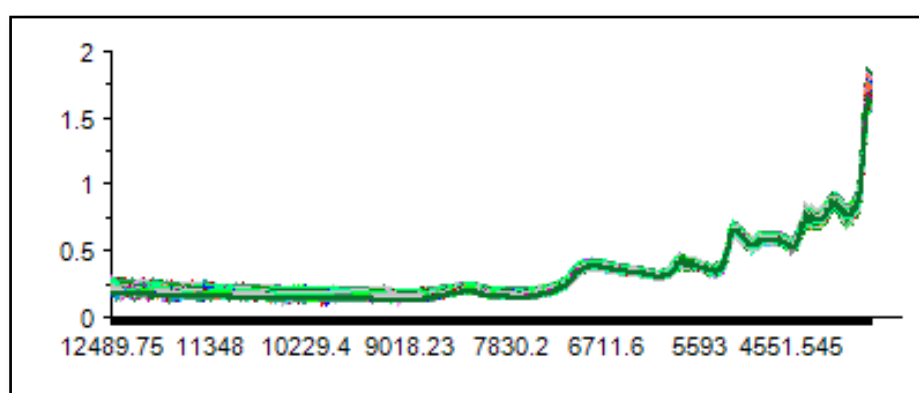


Figure 4.2 Raw FT-NIR spectra of mixed rice bran and parboiled rice bran (wavenumber 12,500 - 3,600 cm^{-1})

4.3.2 Chemometric analysis of FT-NIR data

Moisture content, protein content, fat content, crude fiber content, acid value, phytic acid, bulk density (tapped), bulk density (untapped), and color (whiteness index) were used to develop calibration models. Only 9 parameters could be used to develop good calibration models with R^2 greater than 0.91 for calibration set and 0.88 for validation set.

In the stage of model development, the samples were classified as the calibration set and the validation set. Table 4.12 shows the sample data range for the calibration set and the validation set. It is noted that the range of values for the validation set was within the range of the calibration set for all parameters. Small differences in range of minimum and maximum, mean and standard deviation between the calibration set and validation set indicated that both sets could represent

the analysis of chemical compositions and physical properties of rice bran samples.

For the modelling, the spectra were preprocessed by the combination of several mathematic treatments. The spectra obtained from FT-NIR and chemical analyses were pretreated by 12 mathematic methods: Raw spectrum, Moving Average Smoothing, Gaussian Filter Smoothing, Median Filter Smoothing, Savitzky–Golay Smoothing, Normalization, Gap Derivative, Gap Segment Derivative, S Golay 1st Derivative, S Golay 2nd Derivative, Baseline, and SNV. Calibration models were produced using partial least square (PLS) regression. The quality of the models was checked by calculation of the determination coefficient (R^2), root mean square error of calibration (RMSEC), root mean square error of prediction (RMSEP). Values closer to one for R^2 and low value for RMSEC indicate the good performance of the model for the prediction of the quality parameters of rice bran samples. Table 4.13 shows comparison of statistics obtained from the modelling of calibration and validation using different pretreatment methods. The summary of calibration and validation statistics for all factors are shown in Table 4.14. It was found that the treated FT-NIR spectra gave better models; higher R^2 and lower RMSEP, when compared to raw spectra.

From the result, raw NIR spectra were optimal for relationship development with fat and bulk density (tapped), whereas Median filter smoothing pretreatment method was optimal for relationship development with only phytic acid. Baseline method was optimal for relationship development with acid value and bulk density (untapped), while Moving Average Smoothing method was optimal for relationship development with only protein. In addition, Normalization method was optimal for that with moisture, crude fiber, and color (whiteness index).

Scatter plots for comparison of measured and predicted values for each parameter are shown in Figure 4.3-4.11. Good linearity and high R^2 indicated that the calibration equation could provide good prediction for fat, phytic acid, acid value, bulk density (untapped), protein, bulk density (tapped), moisture, fiber, and color (whiteness index). R^2 of calibration set was 0.98, 0.96, 0.94, 0.92, 0.92, 0.91, 0.91, 0.91, and 0.91, respectively, and RMSEC was 0.36, 0.15, 1.53, 0.01, 0.20,

0.01, 0.13, 0.36, and 0.48, respectively. Furthermore, R^2 of internal validation set was 0.97, 0.97, 0.93, 0.91, 0.92, 0.87, 0.92, 0.88, and 0.88, respectively, and RMSEP was 0.40, 0.13, 1.72, 0.01, 0.21, 0.01, 0.12, 0.41, and 0.56, respectively.

Table 4.12 Range of composition analysis in the sample used to develop PLS models

Parameter	Calibration set (n = 165)			Internal validation set (n = 55)		
	Range	Mean	SD	Range	Mean	SD
Moisture content (% wb)	8.5-10.9	9.8	0.4	8.9-10.9	9.8	0.4
Protein (% db)	14.7-18.4	16.1	0.7	14.7-18.2	16.1	0.7
Fat (% db)	19.2-31.8	25.3	2.3	20.5-31.6	25.4	2.3
Fiber (% db)	9.1-14.0	11.6	1.2	9.4-14.0	11.6	1.2
Acid value (mg KOH/g oil)	3.1-28.1	12.0	6.2	3.4-28.1	12.3	6.4
Phytic acid (g/100g bran)	4.8-8.2	6.7	0.7	5.1-8.1	6.7	0.7
Bulk density (tapped) (g/ml)	0.29-0.41	0.36	0.02	0.31-0.41	0.36	0.02
Bulk density (untapped) (g/ml)	0.19-0.29	0.23	0.02	0.19-0.29	0.23	0.02
Whiteness index	47.3-55.0	50.5	1.6	47.6-54.9	50.6	1.6

Note: n = number of samples, SD = standard deviation

Table 4.13 PLS model statistics for mixed rice bran and parboiled rice bran

Parameter	Pretreatment method	Factor	Calibration set			Internal validation set			
			RMSEC	SEC	R ²	RMSEP	SEP	Bias	R ²
Moisture content (%wb)	Raw spectrum	7	0.1309	0.1313	0.9069	0.1177	0.1163	-0.0238	0.9221
	Moving Average Smoothing	7	0.1307	0.1311	0.9072	0.1154	0.1171	0.0071	0.9251
	Gaussian Filter Smoothing	7	0.1308	0.1312	0.9070	0.1165	0.1163	-0.0166	0.9237
	Median Filter Smoothing	7	0.1308	0.1312	0.9070	0.1175	0.1163	-0.0232	0.9223
	Smoothing Savitzky–Golay	7	0.1306	0.1310	0.9073	0.1157	0.1171	0.0052	0.9247
	Normalization	7	0.1269	0.1273	0.9125	0.1174	0.1146	-0.0296	0.9225
	Gap Derivative	7	0.2246	0.2211	0.7360	1.3143	2.5535	1.2483	NA
Gap Segment Derivative	7	0.2205	0.2211	0.7360	1.3143	2.5535	1.2485	NA	
S Golay 1 st Derivative	7	0.2205	0.2211	0.7360	2.3143	2.5535	1.2483	NA	
S Golay 2 nd Derivative	7	0.2882	0.2890	0.5489	9.4523	16.9962	8.4098	NA	
Baseline		6	0.1367	0.1371	0.8985	0.1241	0.1229	-0.0237	0.9134
SNV		6	0.1341	0.1345	0.9023	0.1341	0.1345	0.0000	0.9023

Table 4.13 PLS model statistics for mixed rice bran and parboiled rice bran (continued...)

Parameter	Pretreatment method	Factor	Calibration set			Internal validation set			
			RMSEC	SEC	R ²	RMSEP	SEP	Bias	R ²
Protein	Raw spectrum	5	0.1962	0.1968	0.9240	0.2047	0.2045	-0.0289	0.9209
(%db)	Moving Average Smoothing	5	0.1958	0.1964	0.9243	0.2067	0.2037	-0.0445	0.9193
	Gaussian Filter Smoothing	5	0.1961	0.1967	0.9241	0.2050	0.2043	-0.0326	0.9206
	Median Filter Smoothing	5	0.1960	0.1966	0.9242	0.2049	0.2047	-0.0294	0.9207
	Smoothing Savitzky–Golay	5	0.1958	0.1964	0.9243	0.2064	0.2036	-0.0437	0.9196
	Normalization	4	0.1979	0.1985	0.9227	0.2135	0.2115	-0.0412	0.9139
	Gap Derivative	7	0.3995	0.4007	0.6850	10.7208	1.3730	-10.6341	NA
	Gap Segment Derivative	7	0.3465	0.3475	0.7630	3.2117	0.9061	-3.0836	NA
	S Golay 1 st Derivative	7	0.2303	0.2310	0.8953	4.9766	9.9560	4.9178	NA
	S Golay 2 nd Derivative	7	0.3995	0.4007	0.6850	10.7206	1.3730	-10.6339	NA
	Baseline	6	0.2127	0.2133	0.9107	0.1879	0.1901	0.0074	0.9333
	SNV	5	0.2090	0.2096	0.9138	0.1858	0.1927	0.0254	0.9348

Table 4.13 PLS model statistics for mixed rice bran and parboiled rice bran (continued...)

Parameter	Pretreatment method	Factor	Calibration set				Internal validation set			
			RMSEC	SEC	R ²	R ²	RMSEP	SEP	Bias	R ²
Fat (%db)	Raw spectrum	7	0.3588	0.3599	0.9757	0.3961	0.3984	-0.0322	0.9697	
	Moving Average Smoothing	7	0.3740	0.3752	0.9736	0.3982	0.4018	-0.0060	0.9694	
	Gaussian Filter Smoothing	7	0.3619	0.3630	0.9753	0.3954	0.3981	-0.0261	0.9698	
	Median Filter Smoothing	7	0.3623	0.3634	0.9752	0.3944	0.3976	-0.0202	0.9699	
	Smoothing Savitzky–Golay	7	0.3712	0.3723	0.9740	0.4023	0.4043	-0.0371	0.9687	
	Normalization	6	0.3702	0.3713	0.9741	0.4152	0.4188	-0.0141	0.9667	
	Gap Derivative	6	0.8698	0.8724	0.8571	35.0594	3.4531	-34.8920	NA	
	Gap Segment Derivative	7	0.7114	0.7136	0.9044	25.5245	3.0332	-25.3470	NA	
	S Golay 1 st Derivative	7	0.4657	0.4671	0.9590	2.5828	3.5863	1.4091	NA	
	S Golay 2 nd Derivative	7	0.8202	0.8227	0.8729	35.6679	3.3184	-35.5160	NA	
	Baseline	6	0.4506	0.4520	0.9616	0.4759	0.4763	-0.0616	0.9562	
	SNV	4	0.5246	0.5262	0.9480	0.5869	0.5918	-0.0247	0.9335	

Table 4.13 PLS model statistics for mixed rice bran and parboiled rice bran (continued...)

Parameter	Pretreatment method	Factor	Calibration set				Internal validation set			
			RMSEC	SEC	R ²	R ²	RMSEP	SEP	Bias	R ²
Fiber (%db)	Raw spectrum	6	0.3643	0.3654	0.9059	0.4225	0.4266	0.0070	0.8728	
	Moving Average Smoothing	6	0.3638	0.3649	0.9062	0.4275	0.4308	-0.0325	0.8698	
	Gaussian Filter Smoothing	6	0.3642	0.3653	0.9059	0.4235	0.4274	-0.0029	0.8722	
	Median Filter Smoothing	6	0.3637	0.3648	0.9062	0.4247	0.4292	0.0120	0.8715	
	Smoothing Savitzky–Golay	6	0.3646	0.3657	0.9057	0.4257	0.4275	-0.0424	0.8709	
	Normalization	6	0.3589	0.3600	0.9086	0.4105	0.4146	0.0092	0.8800	
	Gap Derivative	7	0.5360	0.5377	0.7962	9.7625	2.1389	-9.5297	NA	
	Gap Segment Derivative	7	0.5955	0.5974	0.7485	15.9969	1.4593	-15.9314	NA	
	S Golay 1 st Derivative	7	0.3688	0.3700	0.9035	4.4411	8.7463	4.2967	NA	
	S Golay 2 nd Derivative	7	0.5360	0.5377	0.7962	9.7624	2.1389	-9.5295	NA	
	Baseline	6	0.3597	0.3608	0.9082	0.4103	0.4200	0.0404	0.8801	
	SNV	5	0.3673	0.3685	0.9043	0.4295	0.4381	0.0363	0.8686	

Table 4.13 PLS model statistics for mixed rice bran and parboiled rice bran (continued...)

Parameter	Pretreatment method	Factor	Calibration set				Internal validation set			
			RMSEC	SEC	R ²	R ²	RMSEP	SEP	Bias	R ²
Acid value	Raw spectrum	7	1.5854	1.5902	0.9336	1.8266	1.9346	0.2541	0.9127	
(mg KOH/g oil)	Moving Average Smoothing	7	1.5889	1.5937	0.9333	1.9438	2.2175	0.5915	0.9052	
	Gaussian Filter Smoothing	7	1.5862	1.5910	0.9335	1.8771	1.9790	0.3275	0.9116	
	Median Filter Smoothing	7	1.5863	1.5912	0.9335	1.8616	1.9307	0.2543	0.9130	
	Smoothing Savitzky–Golay	7	1.5867	1.5916	0.9334	1.9401	2.2058	0.5812	0.9056	
	Normalization	7	1.6921	1.6973	0.9243	0.8881	2.1594	0.1994	0.8920	
	Gap Derivative	7	3.7131	3.7244	0.6356	36.9569	73.0455	35.9296	NA	
	Gap Segment Derivative	5	3.9261	3.9480	0.5905	65.4249	131.4713	65.0385	NA	
	S Golay 1 st Derivative	7	2.7220	2.7303	0.8042	52.0823	6.5963	-51.6705	NA	
	S Golay 2 nd Derivative	7	3.7131	3.7244	0.6356	36.9563	73.0441	35.9290	NA	
	Baseline	7	1.5257	1.5304	0.9385	1.7238	1.7786	0.2116	0.9254	
	SNV	7	1.7496	1.7550	0.9191	1.9838	2.0092	0.0967	0.9013	

Table 4.13 PLS model statistics for mixed rice bran and parboiled rice bran (continued...)

Parameter	Pretreatment method	Factor	Calibration set			Internal validation set			
			RMSEC	SEC	R ²	RMSEP	SEP	Bias	R ²
Phytic acid (g/100g bran)	Raw spectrum	7	0.1502	0.1507	0.9584	0.1292	0.1304	-0.0001	0.9682
	Moving Average Smoothing	7	0.1510	0.1514	0.9580	0.1311	0.1465	0.0361	0.9673
	Gaussian Filter Smoothing	7	0.1504	0.1508	0.9583	0.1287	0.1307	0.0083	0.9685
	Median Filter Smoothing	7	0.1500	0.1505	0.9585	0.1277	0.1289	-0.0007	0.9690
	Smoothing Savitzky–Golay	7	0.1511	0.1516	0.9579	0.1343	0.1506	0.0376	0.9657
	Normalization	6	0.1542	0.1547	0.9561	0.1313	0.1326	0.0026	0.9672
	Gap Derivative	7	0.4165	0.4178	0.6801	14.0705	28.3037	14.0066	NA
	Gap Segment Derivative	7	0.3525	0.3536	0.7709	5.8965	11.8086	5.8349	NA
	S Golay 1 st Derivative	7	0.2633	0.2641	0.8721	4.3091	0.6919	-4.2542	NA
	S Golay 2 nd Derivative	7	0.4165	0.4178	0.6801	14.0704	28.3035	14.0065	NA
	Baseline	7	0.1551	0.1556	0.9557	0.1290	0.1307	0.0069	0.9683
	SNV	6	0.1629	0.1634	0.9510	0.1517	0.1546	0.0122	0.9562

Table 4.13 PLS model statistics for mixed rice bran and parboiled rice bran (continued...)

Parameter	Pretreatment method	Factor	Calibration set			Internal validation set			
			RMSEC	SEC	R ²	RMSEP	SEP	Bias	R ²
Bulk density (tapped) (g/ml)	Raw spectrum	6	0.0071	0.0071	0.9104	0.0083	0.0087	0.0012	0.8724
	Moving Average Smoothing	6	0.0072	0.0072	0.9086	0.0087	0.0104	0.0032	0.8619
	Gaussian Filter Smoothing	6	0.0071	0.0071	0.9100	0.0083	0.0089	0.0017	0.8715
	Median Filter Smoothing	6	0.0071	0.0071	0.9101	0.8738	0.0086	0.0012	0.8738
	Smoothing Savitzky-Golay	6	0.0072	0.0072	0.9089	0.0088	0.0106	0.0033	0.8585
	Normalization	7	0.0074	0.0074	0.9037	0.0085	0.0089	0.0015	0.8675
	Gap Derivative	7	0.0152	0.0153	0.5886	0.5697	1.1481	0.5685	NA
	Gap Segment Derivative	7	0.0129	0.0129	0.7052	0.2312	0.4643	0.2296	NA
	S Golay 1 st Derivative	7	0.0116	0.0116	0.7612	0.0139	0.0140	-0.0005	0.6438
	S Golay 2 nd Derivative	7	0.0152	0.0153	0.5886	0.5697	0.0213	-0.0034	NA
	Baseline	6	0.0072	0.0072	0.9079	0.0083	0.0087	0.0013	0.8720
	SNV	6	0.0078	0.0078	0.8920	0.0091	0.0096	0.0015	0.8475

Table 4.13 PLS model statistics for mixed rice bran and parboiled rice bran (continued...)

Parameter	Pretreatment method	Factor	Calibration set			Internal validation set			
			RMSEC	SEC	R ²	RMSEP	SEP	Bias	R ²
Bulk density (untapped) (g/ml)	Raw spectrum	6	0.0064	0.0065	0.9166	0.0068	0.0069	-0.0005	0.9085
	Moving Average Smoothing	6	0.0064	0.0065	0.9165	0.0069	0.0070	-0.0004	0.9058
	Gaussian Filter Smoothing	6	0.0064	0.0065	0.9166	0.0069	0.0069	-0.0005	0.9075
	Median Filter Smoothing	6	0.0065	0.0065	0.9157	0.0069	0.0069	-0.0004	0.9075
	Smoothing Savitzky–Golay	6	0.0064	0.0065	0.9168	0.0070	0.0070	-0.0004	0.9051
	Normalization	6	0.0064	0.0064	0.9173	0.0071	0.0071	-0.0005	0.9019
	Gap Derivative	7	0.0112	0.0113	0.7470	0.2986	0.5992	0.2963	NA
	Gap Segment Derivative	7	0.0101	0.0101	0.7968	0.1014	0.1985	0.0973	NA
	S Golay 1 st Derivative	7	0.0092	0.0093	0.8285	0.0655	0.1272	0.0622	NA
	S Golay 2 nd Derivative	7	0.0112	0.0113	0.7470	2.9858	0.5992	0.2963	NA
Baseline		6	0.0062	0.0062	0.9221	0.0066	0.0067	0.0000	0.9137
SNV		5	0.0066	0.0066	0.9134	0.0072	0.0073	0.0002	0.8988

Table 4.13 PLS model statistics for mixed rice bran and parboiled rice bran (continued...)

Parameter	Pretreatment method	Factor	Calibration set				Internal validation set			
			RMSEC	SEC	R ²	R ²	RMSEP	SEP	Bias	R ²
Whiteness index	Raw spectrum	6	0.5023	0.5038	0.9007	0.5767	0.5821	-0.0021	0.8732	
	Moving Average Smoothing	6	0.5068	0.5083	0.8990	0.5763	0.5849	0.0355	0.5763	
	Gaussian Filter Smoothing	6	0.5029	0.5044	0.9005	0.5760	0.5815	0.0060	0.8736	
	Median Filter Smoothing	7	0.4924	0.4939	0.9046	0.5898	0.5959	0.0155	0.8674	
	Smoothing Savitzky–Golay	6	0.5022	0.5037	0.9008	0.5762	0.5860	0.0412	0.8735	
	Normalization	7	0.4759	0.4774	0.9109	0.5643	0.5690	-0.0225	0.8787	
	Gap Derivative	7	0.7077	0.7098	0.8029	11.6367	23.0888	11.3723	NA	
	Gap Segment Derivative	5	0.7405	0.7428	0.7842	6.7577	13.4794	6.6514	NA	
	S Golay 1 st Derivative	7	0.5578	0.5595	0.8776	5.1425	10.0551	4.9269	NA	
	S Golay 2 nd Derivative	7	0.7077	0.7098	0.8029	11.6367	23.0888	11.3723	NA	
	Baseline	7	0.4819	0.4833	0.9086	0.5646	0.5696	-0.0142	0.8785	
	SNV	5	0.5082	0.5097	0.8984	0.5725	0.5762	-0.0417	0.8751	

Table 4.14 Totals PLS model statistics for mixed rice bran and parboiled rice bran

Parameter	Preprocessing	Calibration (n = 165)			Internal validation (n = 55)			
		RMSEC	SEC	R ²	RMSEP	SEP	Bias	R ²
Moisture content	Normalization	0.1269	0.1273	0.9125	0.1174	0.1146	-0.0296	0.9225
Crude protein	Smoothing Moving Average	0.1958	0.1964	0.9243	0.2067	0.2037	-0.0445	0.9193
Crude fat	Raw spectrum	0.3588	0.3599	0.9757	0.3961	0.3984	-0.0322	0.9697
Crude fiber	Normalization	0.3589	0.3600	0.9086	0.4105	0.4146	0.0092	0.8800
Acid value	Baseline	1.5257	1.5304	0.9385	1.7238	1.7786	0.2116	0.9254
Phytic acid	Smoothing Median Filter	0.1500	0.1505	0.9585	0.1277	0.1289	-0.0007	0.9690
Bulk density tap	Raw spectrum	0.0071	0.0071	0.9104	0.0083	0.0087	0.0012	0.8724
Bulk density untap	Baseline	0.0062	0.0062	0.9221	0.0066	0.0067	0.0000	0.9137
Whiteness index	Normalization	0.4759	0.4774	0.9109	0.5643	0.5690	-0.0225	0.8787

Note: R² - determination coefficient, RMSEC, RMSEP - root mean square error of calibration and prediction,
SEC, SEP - standard error of calibration and prediction

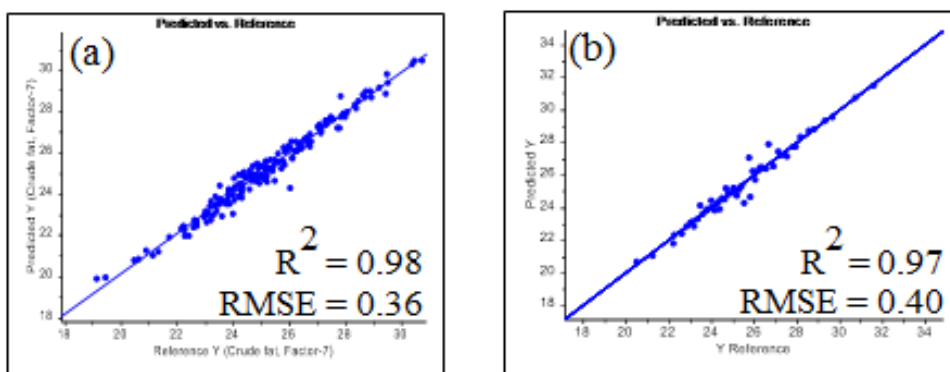


Figure 4.3 Relationship between predicted value and reference value of fat for (a) calibration (n = 165) and (b) internal validation (n = 55)

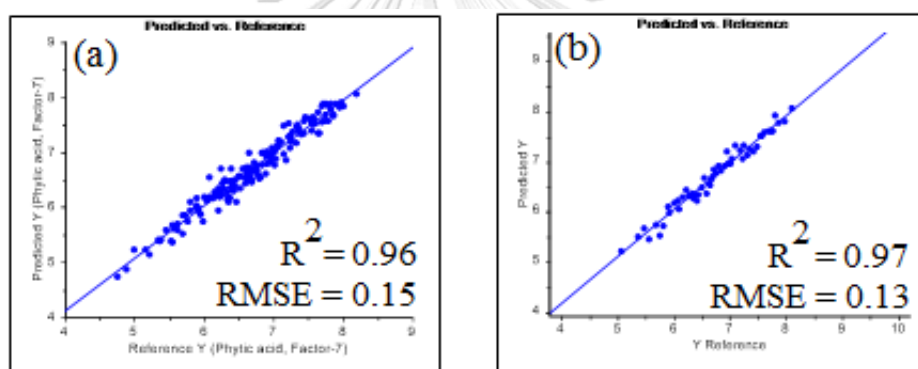


Figure 4.4 Relationship between predicted value and reference value of phytic acid for (a) calibration (n = 165) and (b) internal validation (n = 55)

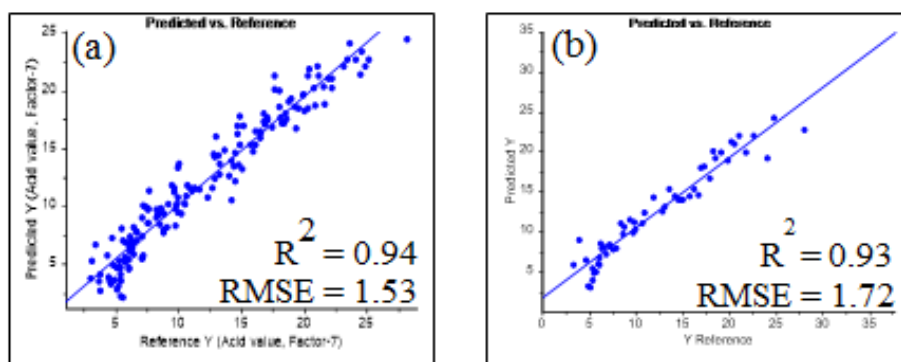


Figure 4.5 Relationship between predicted value and reference value of acid value for (a) calibration (n = 165) and (b) internal validation (n = 55)

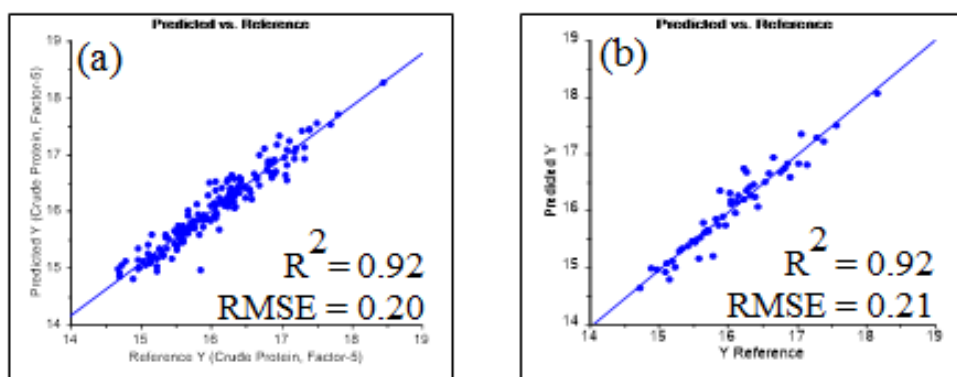


Figure 4.6 Relationship between predicted value and reference value of protein for (a) calibration (n = 165) and (b) internal validation (n = 55)

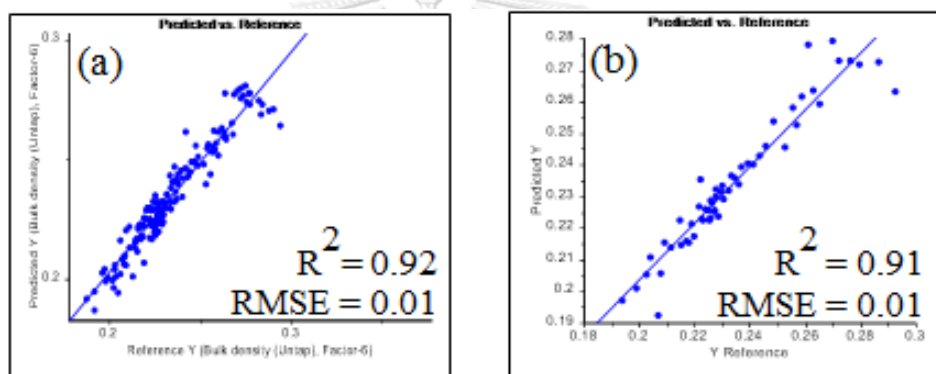


Figure 4.7 Relationship between predicted value and reference value of bulk density (untapped) for (a) calibration (n = 165) and (b) internal validation (n = 55)

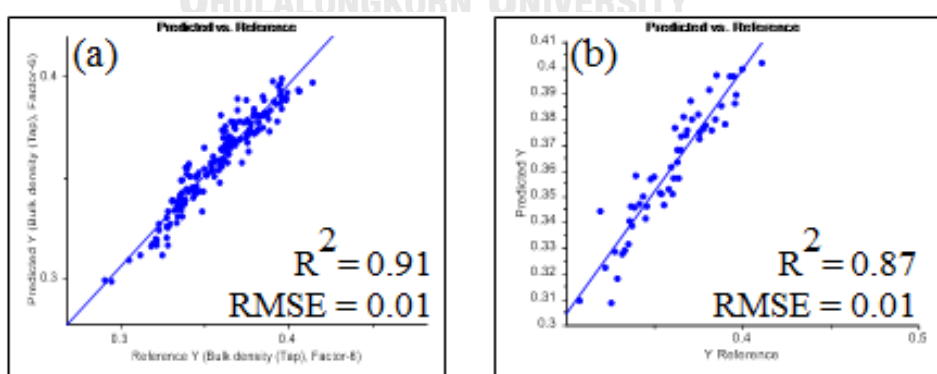


Figure 4.8 Relationship between predicted value and reference value of bulk density (tapped) for (a) calibration (n = 165) and (b) internal validation (n = 55)

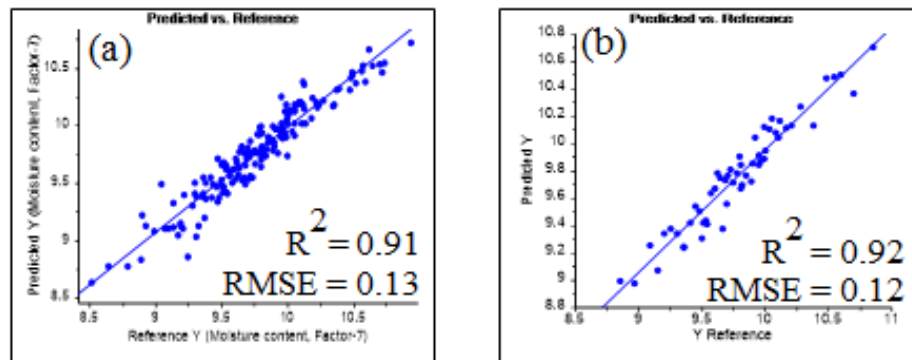


Figure 4.9 Relationship between predicted value and reference value of moisture for (a) calibration (n = 165) and (b) internal validation (n = 55)

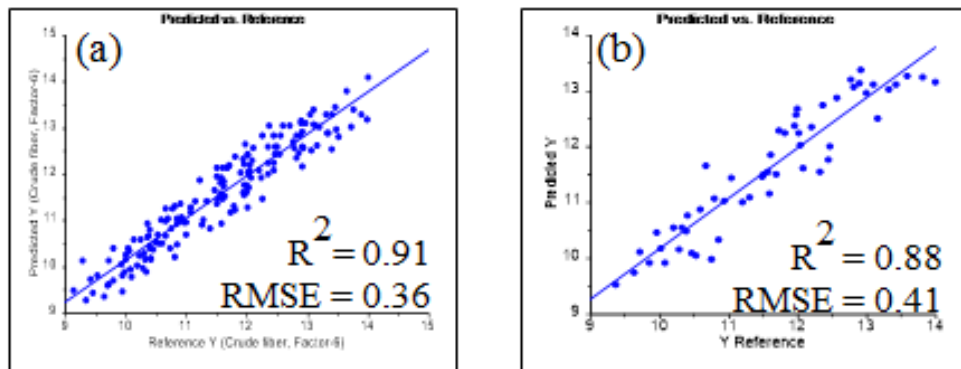


Figure 4.10 Relationship between predicted value and reference value of fiber for (a) calibration (n = 165) and (b) internal validation (n = 55)

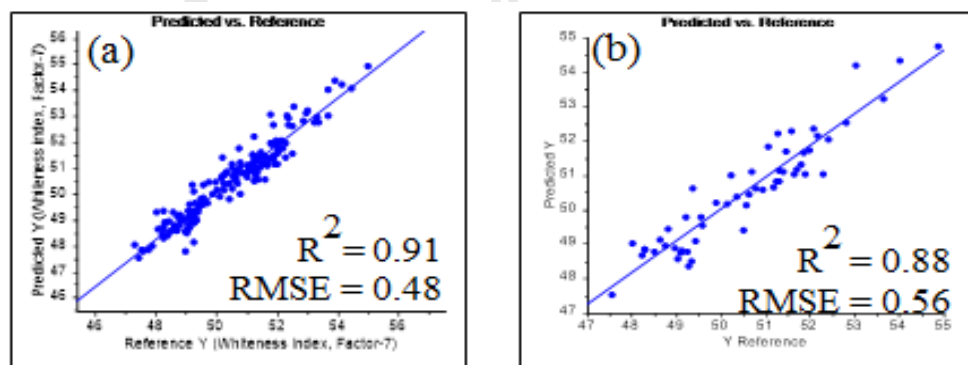


Figure 4.11 Relationship between predicted value and reference value of color (whiteness index) for (a) calibration (n = 165) and (b) internal validation (n = 55)

4.4 External validation

External rice bran samples from a different harvesting season were obtained and analysed in order to validate the calibration model developed earlier. Proximate compositions of 60 rice bran samples are shown in Table 4.15. It was found that the major component in rice bran is fat which accounted for 15.6-30.8 % db, followed by protein content (13.0-20.9 % db), crude fiber content (8.9-13.9 % db), and moisture content (9.2-11.4 % wb). Rice bran also presented the acid value in the 4.3-123.1 mg KOH/g oil range, 5.3-8.1 g/100g bran phytic acid. All rice bran samples had the untapped bulk density in the 0.18-0.26 g/ml range, the tapped bulk density in the 0.26-0.37 g/ml range, and whiteness index of 44.5-53.0.

Table 4.15 Chemical compositions and physical properties of 60 external rice bran samples

Parameters	Range	Mean	SD
Moisture content (% wb)	9.2-11.4	10.5	0.6
Protein content (% db)	13.0-20.9	15.6	2.0
Fat content (% db)	15.6-30.8	22.2	4.6
Fiber content (% db)	8.9-13.9	11.3	1.7
Acid value (mg KOH/g oil)	4.3-123.1	56.2	42.6
Phytic acid (g/100g bran)	5.3-8.1	7.0	0.8
Bulk density (tapped)	0.26-0.37	0.33	0.03
Bulk density (untapped) (g/ml)	0.18-0.26	0.23	0.02
Whiteness index	44.5-53.0	49.1	2.4

Note: SD = standard deviation

Tables 4.16 to 4.18 show the chemical compositions and physical properties of 60 samples of rice bran samples. It was observed that the external samples possessed chemical compositions and physical properties in close proximity with the rice bran samples previously acquired for NIR calibration model (Table 4.19), except for one rice bran

sample which contained very high acid value. The same samples were also subjected to FT-NIR analyses in the diffuse-reflectance mode.

The calibration and external validation statistics are shown in Table 4.20. It was found that the treated of best model could predict good models that gave high R^2 and low RMSEP. Scatter plots for relationship between predicted value and reference value of external validation for each parameter are shown in Figure 4.12. Good linearity and high R^2 indicated that the calibration equation could accurately be used for the prediction of crude fat, phytic acid, bulk density (untapped), protein, bulk density (tapped), crude fiber, and color (whiteness index). But high RMSEP and not available R^2 for moisture content and acid value are noted. This was because the range of moisture content and acid value of the external samples fell outside the range of value for the calibration model development. The model could still be successfully used to predict crude fat ($R^2 = 0.9714$) and phytic acid content ($R^2 = 0.8941$) of the external samples.

Table 4.16 Chemical analyzes and physical properties of external rice bran samples

Rice bran	Moisture (%wb)	Protein (%db)	Fat (%db)	Fiber (%db)	Acid value (mg KOH/g oil)	Phytic acid (g/100g bran)	Bulk density (tapped) (g/ml)	Bulk density (untapped) (g/ml)	Whiteness index
1	11.2±0.2	16.3±0.4	16.4±0.9	9.8±0.1	109.9±0.1	7.4±0.1	0.36±0.01	0.24±0.01	51.0±1.1
2	11.1±0.1	15.5±0.0	15.7±0.2	9.5±0.0	99.9±0.1	7.4±0.1	0.34±0.00	0.22±0.00	52.1±0.4
3	10.4±0.1	14.6±0.1	19.4±0.6	9.0±0.0	96.4±0.1	7.6±0.0	0.36±0.00	0.22±0.00	50.1±0.0
4	10.8±0.1	15.1±0.0	18.9±0.3	9.9±0.0	80.8±0.0	7.3±0.0	0.37±0.00	0.24±0.01	51.2±0.3
5	11.2±0.0	15.1±0.1	16.5±0.4	9.6±0.0	104.2±0.1	8.0±0.1	0.37±0.00	0.24±0.00	52.5±0.0
6	11.2±0.0	16.1±0.0	16.0±0.3	9.5±0.0	107.2±0.0	8.0±0.0	0.37±0.00	0.25±0.00	52.7±0.4
7	11.1±0.0	14.3±0.0	18.6±0.2	8.9±0.0	103.3±0.0	8.0±0.1	0.35±0.00	0.23±0.00	50.6±0.0
8	11.0±0.0	14.9±0.0	17.1±0.2	9.3±0.1	123.1±0.0	7.3±0.1	0.34±0.00	0.22±0.00	51.1±0.1
9	10.9±0.1	14.0±0.0	18.8±0.1	9.2±0.2	73.8±0.0	7.6±0.0	0.35±0.00	0.22±0.00	49.8±0.1
10	11.1±0.1	14.9±0.1	17.7±0.1	9.5±0.2	119.4±0.2	7.6±0.1	0.36±0.00	0.25±0.00	52.0±0.2

Note: The values in the table are shown in mean ± SD from the 2 replicates.

Table 4.17 Chemical analyzes and physical properties of external parboiled rice bran samples

Parboiled rice bran	Moisture (%wb)	Protein (%db)	Fat (%db)	Fiber (%db)	Acid value (mg KOH/g oil)	Phytic acid (g/100g bran)	Bulk density (tapped) (g/ml)	Bulk density (untapped) (g/ml)	Whiteness index
1	10.0±0.0	18.9±0.0	27.7±0.0	13.4±0.1	9.6±0.0	6.0±0.0	0.32±0.00	0.22±0.00	45.7±0.1
2	10.7±0.1	20.9±0.0	25.3±0.3	13.7±0.0	19.2±0.0	5.5±0.0	0.34±0.00	0.23±0.00	45.9±0.1
3	10.0±0.0	15.3±0.0	24.7±0.1	12.7±0.1	14.5±0.0	6.7±0.1	0.30±0.00	0.21±0.00	48.4±0.0
4	10.0±0.0	14.0±0.0	26.1±0.5	12.0±0.2	6.0±0.0	7.4±0.2	0.30±0.00	0.19±0.00	47.2±0.1
5	10.1±0.0	16.1±0.0	26.7±0.4	13.4±0.4	6.2±0.0	6.8±0.1	0.31±0.00	0.23±0.00	48.3±0.0
6	9.2±0.0	13.0±0.0	30.7±0.1	11.7±0.1	13.3±0.0	7.4±0.1	0.30±0.00	0.19±0.00	44.6±0.1
7	9.3±0.0	13.2±0.0	29.3±0.2	13.5±0.0	5.4±0.0	5.3±0.0	0.26±0.00	0.18±0.00	45.6±0.2
8	9.9±0.1	16.9±0.0	27.3±0.2	12.8±0.0	7.5±0.3	6.2±0.1	0.30±0.00	0.22±0.00	47.3±0.0
9	9.8±0.0	18.7±0.0	28.3±0.6	13.2±0.2	4.3±0.0	5.6±0.2	0.30±0.00	0.21±0.00	46.6±0.2
10	9.2±0.0	13.1±0.0	29.4±0.2	12.3±0.1	6.1±0.4	6.9±0.1	0.32±0.00	0.21±0.00	45.4±0.0

Note: The values in the table are shown in mean ± SD from the 2 replicates.

Table 4.18 Chemical analyzes and physical properties of external mixed rice bran samples

Ratio of mixed rice bran (rice bran : parboiled rice bran)	Moisture (%wb)	Protein (%db)	Fat (%db)	Fiber (%db)	Acid value (mg KOH/g oil)	Phytic acid (g/100g bran)	Bulk density (tapped) (g/ml)	Bulk density (untapped) (g/ml)	Whiteness index
0.5:9.5	10.2±0.1	18.7±0.0	26.2±0.1	13.9±0.0	16.0±0.0	6.2±0.0	0.33±0.00	0.22±0.00	45.8±0.2
1.5:8.5	10.8±0.0	20.3±0.1	22.9±0.2	13.9±0.0	31.9±0.0	5.9±0.1	0.35±0.00	0.24±0.00	47.4±0.3
2.5:7.5	10.1±0.1	14.7±0.1	22.5±0.2	11.8±0.0	34.9±0.0	7.0±0.0	0.32±0.00	0.22±0.01	49.4±0.3
3.5:6.5	10.3±0.1	14.5±0.0	23.0±0.0	11.4±0.0	32.9±0.0	7.4±0.2	0.32±0.01	0.22±0.00	48.8±0.1
4.5:5.5	10.6±0.1	15.7±0.1	21.1±0.3	11.9±0.0	50.7±0.0	7.5±0.3	0.35±0.01	0.25±0.01	50.9±0.3
5.5:4.5	10.5±0.1	15.0±0.0	21.7±0.1	10.8±0.0	66.2±0.0	7.7±0.0	0.33±0.00	0.23±0.00	50.5±0.1
6.5:3.5	10.6±0.0	14.0±0.0	21.8±0.1	10.8±0.0	69.7±0.0	7.1±0.1	0.31±0.00	0.22±0.00	48.9±0.1
7.5:2.5	10.6±0.1	15.7±0.1	19.1±0.3	10.4±0.0	94.2±0.1	7.1±0.0	0.33±0.00	0.25±0.00	51.1±0.1
8.5:1.5	10.7±0.0	15.0±0.1	19.8±0.2	10.1±0.0	64.1±0.0	7.6±0.1	0.36±0.00	0.26±0.00	50.5±0.0
9.5:0.5	11.2±0.1	14.80±1	17.2±0.2	9.9±0.0	114.1±0.2	7.5±0.0	0.35±0.00	0.25±0.00	51.8±0.0

Note: The values in the table are shown in mean ± SD from the 2 replicates.

Table 4.19 Range of composition analysis for calibration set and the external validation set

Parameter	Calibration set (n = 165)			External validation set (n = 60)		
	Range	Mean	SD	Range	Mean	SD
Moisture content (% wb)	8.5-10.9	9.8	0.4	9.2-11.4	10.5	0.6
Protein (% db)	14.7-18.4	16.1	0.7	13.0-20.9	15.6	2.0
Fat (% db)	19.2-31.8	25.3	2.3	15.6-30.8	22.2	4.6
Fiber (% db)	9.1-14.0	11.6	1.2	8.9-13.9	11.3	1.7
Acid value (mg KOH/g oil)	3.1-28.1	12.0	6.2	4.3-123.1	56.2	42.6
Phytic acid (g/100g bran)	4.8-8.2	6.7	0.7	5.3-8.1	7.0	0.8
Bulk density (tapped) (g/ml)	0.29-0.41	0.36	0.02	0.26-0.37	0.33	0.03
Bulk density (untapped) (g/ml)	0.19-0.29	0.23	0.02	0.18-0.26	0.23	0.02
Whiteness index	47.3-55.0	50.5	1.6	44.5-53.0	49.1	2.4

Note: n = number of samples, SD = standard deviation

Table 4.20 Test the accuracy of the equation by PLS model statistics for external validation set

Parameter	Preprocessing	Calibration			External validation			
		RMSEC	SEC	R ²	RMSEP	SEP	Bias	R ²
Moisture content	Normalization	0.1269	0.1273	0.9125	2.3271	0.5740	-2.2564	NA
Crude protein	Moving Average Smoothing	0.1958	0.1964	0.9243	0.9308	1.8767	0.9304	0.7769
Crude fat	Raw spectrum	0.3588	0.3599	0.9757	0.7654	0.4832	-0.5968	0.9714
Crude fiber	Normalization	0.3589	0.3600	0.9086	0.6271	1.0023	0.4452	0.8601
Acid value	Baseline	1.5257	1.5304	0.9385	60.6153	39.3051	-46.4228	NA
Phytic acid	Median Filter Smoothing	0.1500	0.1505	0.9585	0.2454	0.4809	0.2361	0.8941
Bulk density tapped	Raw spectrum	0.0071	0.0071	0.9104	0.0100	0.0000	-0.0100	0.8677
Bulk density untapped	Baseline	0.0062	0.0062	0.9221	0.0095	0.0156	0.0070	0.7481
Whiteness index	Normalization	0.4759	0.4774	0.9109	1.0751	1.0789	-0.1056	0.8042

Note: R² - determination coefficient, RMSEC, RMSEP - root mean square error of calibration and prediction,
SEC, SEP - standard error of calibration and prediction

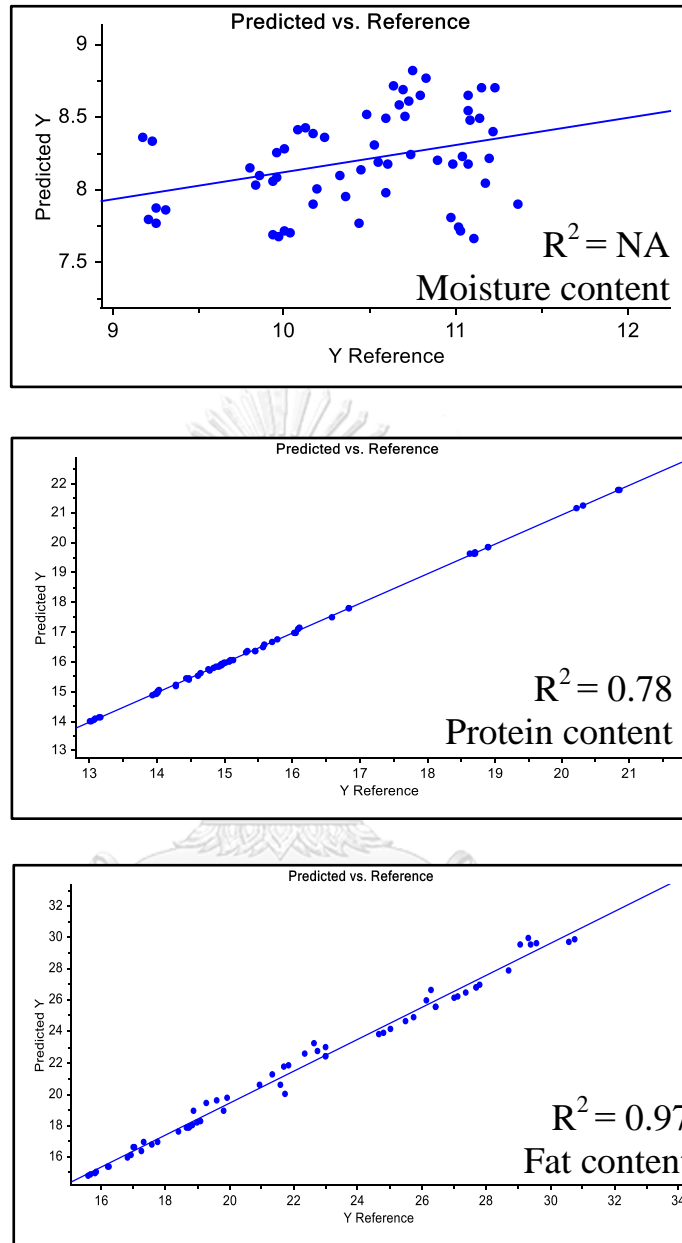


Figure 4.12 Relationship between predicted value and reference value of external validation (n = 60)

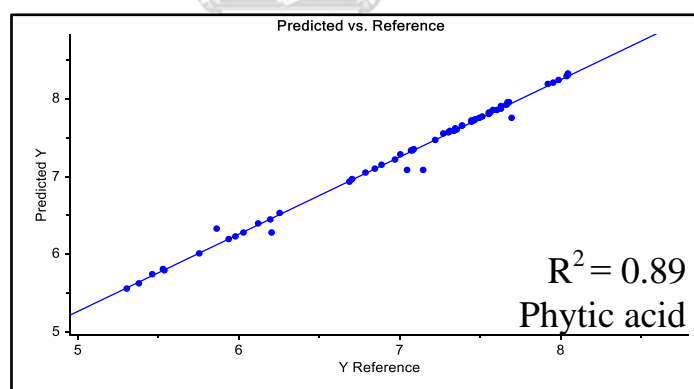
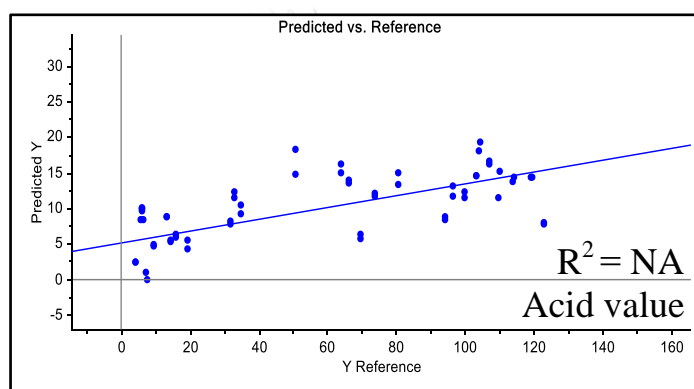
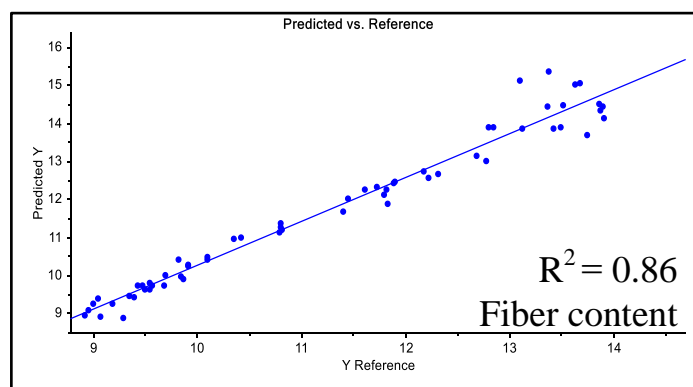


Figure 4.12 Relationship between predicted value and reference value of external validation (n = 60) (continued...)

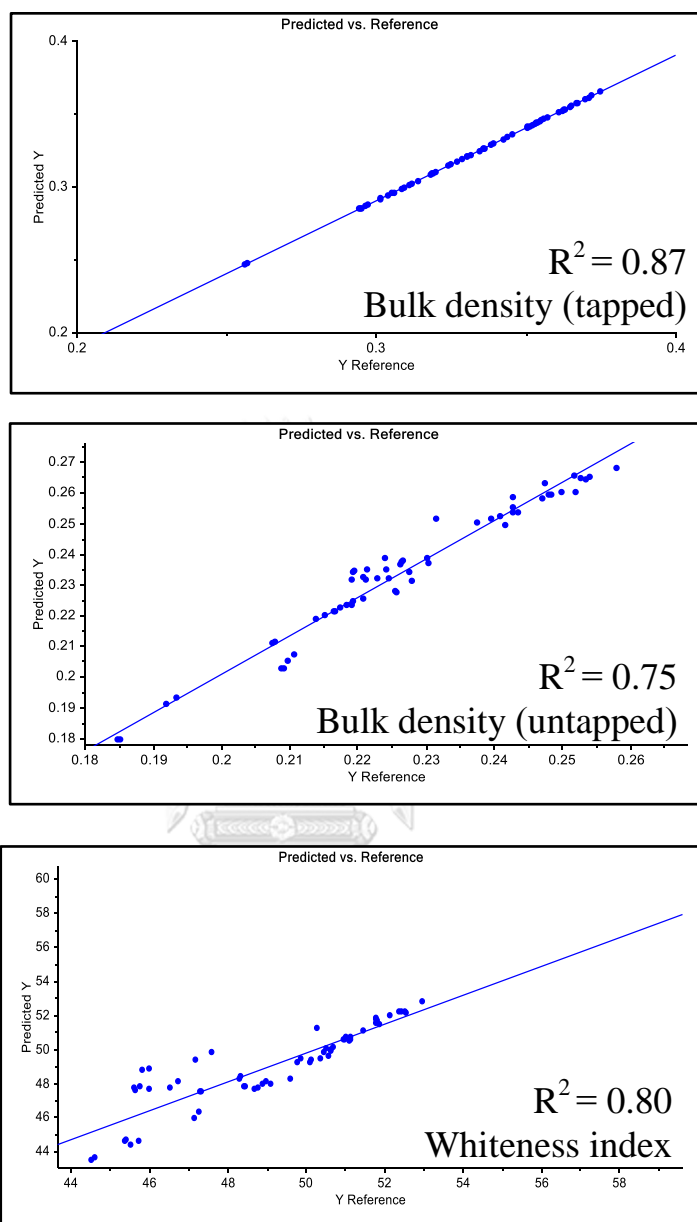


Figure 4.12 Relationship between predicted value and reference value of external validation (n = 60) (continued...)

CHAPTER V CONCLUSIONS

In summary, in terms of the composition of rice bran and parboiled rice bran, when parboiled rice bran was mixed with rice bran in various proportions, fat, protein, and fiber content were increased when parboiled rice bran proportion increased, whereas acid value, phytic acid, moisture content, color (whiteness index), and bulk density decreased. In the study of relationship between chemical compositions, physical properties and ratios of mixed rice bran, good R^2 could be developed in the following descending order: phytic acid, acid value, bulk density (tapped), protein content, fiber content, moisture content, bulk density (untapped), fat content, and color (whiteness index), respectively. All properties, except for moisture content, bulk density, and whiteness index, showed strong dependency on rice bran ratio. Therefore, they can be used as predictors for ratios of rice bran content in mixed bran.

From FT-NIR analyses, good calibration model between FT-NIR and properties could be developed in the following descending order: fat content, phytic acid, acid value, protein content, bulk density, moisture, crude fiber, and color (whiteness index), respectively. Therefore, FT-NIR can be used as a rapid and non-destructive tool to determine those properties of rice bran samples.

Lastly, the study revealed the accuracy of the calibration equations with regard to prediction of external samples from different harvesting season. The model could successfully predict fat content and phytic acid content in the external samples.

Suggestions

To produce good results, both rice bran and parboiled rice bran samples should be analyzed as soon as they are manufactured. The samples also need to be stored in freezing temperatures and vacuum packing environment. Finally, rice bran analysis by NIRS can be more efficient and accurate when the number of collected samples for calibration set increases. Variations of samples with regard to area of rice cultivation, season, and storage condition should also be taken into concerns.

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APPENDIX



จุฬาลงกรณ์มหาวิทยาลัย
CHULALONGKORN UNIVERSITY

APPENDIX A

CHEMICAL ANALYSIS PROCEDURES

A.1 Moisture content (AOAC, 2012)

Apparatus

1. Aluminum pan
2. Hot air oven (Binder, model ED/FD, Germany)
3. Weighing machine (4 digits) (Sartorius, BSA224S, Germany)
4. Desiccator

Procedures

1. The empty aluminum pans was heated at 105 °C until weight constant and cooled down in desiccator
2. Five gram of sample was weighed into pre-weighed aluminum pan and dried at 105 °C for 8 hours in a hot air oven
3. The heated aluminum pan and sample was removed to desiccator for cooling
4. The aluminum pan and sample was weighed after cooling
5. Drying is repeated until constant weight is achieved
6. Moisture content could calculate from equation A.1

$$\text{Moisture content (\% wet basis)} = \frac{W_2 - W_3}{W_2 - W_1} \times 100 \quad (\text{A.1})$$

W_1 : Constant weight (g) of aluminum pan

W_2 : Weight (g) of sample with aluminum pan before drying

W_3 : Weight (g) of sample with aluminum pan after drying

A.2 Protein content using Kjeldahl method (AOAC, 2012)

Apparatus

1. Filter paper (Whatman No.41)
2. Weighing machine (4 digits) (Sartorius, BSA224S, Germany)
3. Kjeldahl tube (Buchi, Switzerland)
4. Buchi digestion unit (Buchi, model K-424, Switzerland)
5. Buchi scrubber (Buchi, model B-414, Switzerland)
6. Distillation apparatus (Buchi, model B-324, Switzerland)

Reagent

1. Selenium mixture (Merck, Germany)
2. Sulphuric acid 98 % (QRëC®, New Zealand)
3. Sodium hydroxide (QRëC®, New Zealand)
4. Boric acid (Univar, Ajax Finechem, Australia)
5. Hydrochloric acid 37 % (QRëC®, New Zealand)
6. Mixed indicator solution: methyl red-methylene blue

Procedures

1. Zero point five gram of sample was weighed into filter paper (Whatman No.41) and moved to Kjeldahl tube
2. Add 5 gram of selenium mixture and 20 mL of sulphuric acid
3. Set a blank test following the method but used 1 mL of distilled water for substitute the sample
4. Place the tube on the Buchi digestion unit with Buchi scrubber and heat until to obtain clear red-brown solution
5. Cooled the sample to room temperature

6. Add 50 mL of 4 % (w/v) boric acid solution into 250 mL flask, add 2 drops of the indicator solution, mix and place flask under the condenser of the distillation apparatus with the distillation mode such as

- Distilled water: 50 mL
- 50% NaOH: 60 mL
- Distillation time: 5 min
- Steam: 100 %

7. Titrate the solution in the flask with 0.1 N HCl until obtained claret solution

8. Record the volume of 0.1 N HCl

9. Protein content is calculated from equation A.2.1 and A.2.2, respectively

$$\text{Total nitrogen (\% wet basis)} = \frac{(V-B) \times N \times 1.4}{W} \quad (\text{A.2.1})$$

V: Volume (mL) of 0.1 N HCl required for the sample titration

B: Volume (mL) of 0.1 N HCl required for the blank test

N: Normality factor of HCl solution

W: weight of sample (g)

$$\text{Protein content (\% wet basis)} = \text{Total nitrogen (\%)} \times 6.25 \quad (\text{A.2.2})$$

6.25: Multiply factor for used to obtain rice bran protein (%)

A.3 Lipid content (AOAC, 2012)

Apparatus

1. Filter paper (Whatman No.1)
2. Weighing machine (4 digits)
3. Extraction thimbles

4. Soxhlet
5. Rotary evaporator
6. Hot air oven
7. Desiccator

Reagent

1. Hexane (QRec®, New Zealand)

Procedures

1. Dried flat bottom flask at 105 °C in hot air oven for 1 hour or until constant weight and cooled down in desiccator
2. Weigh five gram of dried sample on filter paper and wrap
3. Move the filter paper (Whatman No.1) with sample into extraction thimble
4. Place the extraction thimble in the Soxhlet extraction and connected a weighed flat bottom flask containing 250 mL hexane
5. Connect the extractor to a reflux condenser
6. Extract fat for 4 hours at condensation rate of 5-6 drops/second
7. Evaporated the hexane by rotary evaporator
8. Dried flat bottom flask at 105 °C in hot air oven until constant weight, cooled down in desiccator and weigh
9. Lipid content could calculate from equation A.3

$$\text{Lipid content (\% dry basis)} = \frac{W_2 - W_3}{W_1} \times 100 \quad (\text{A.3})$$

W_1 : Weight (g) of sample

W_2 : Weight (g) of flat bottom flask with lipid

W_3 : Weight (g) of flat bottom flask without lipid

A.4 Crude fiber content (AOAC, 2012)

Apparatus

1. Büchner funnel
2. Crucible
3. Desiccator
4. Hot air oven
5. Muffle furnace
6. Weighing machine (4 digits)
7. Filter paper (Whatman No. 1 and 42)

Reagent

1. Sulphuric acid 98 % (QRëC®, New Zealand)
2. Sodium hydroxide (QRëC®, New Zealand)
3. Ethanol 95 %

Procedures

1. Weigh two gram of sample (without lipid) obtained from the determination of lipid content (W_1) into 600 mL beaker
2. Add 200 mL of 1.25 % sulphuric acid in the beaker
3. Heat at Boiling point for 30 minute (control volume of solution with boiling water)
4. Filter the content from point 3 through a Büchner funnel with a filter paper (Whatman No. 1)
5. Wash with boiling water until acid-free
6. Remove the content from point 5 into the original beaker and add 200 mL of 1.25 % Sodium hydroxide

7. Heat at Boiling point for 30 minute (control volume of solution with water)
8. Filter the content from point 3 through a Büchner funnel with a filter paper (Whatman No. 42)
9. Wash with boiling water until base-free
10. Wash twice with 25 mL of 95 % ethanol
11. Dried the filter paper with content (from 10.) at 105 °C until constant weigh (W_2)
12. Transfer the dried sample into crucible (pre-weigh)
13. Ashing in muffle furnace at 550 °C until obtain the white ash
14. Cool down the crucible in a desiccator and weight (W_3)
15. Crude fiber content could calculate from equation A.4

$$\text{Crude fiber content (\% dry basis)} = \frac{W_2 - W_3}{W_1} \times 100 \quad (\text{A.4})$$

W_1 : Weight (g) of sample

W_2 : Weight (g) of insoluble matter

W_3 : Weight (g) of ash

A.5 Acid value (AOCS, 2003)

Apparatus

1. Weighing machine (4 digits) (Sartorius, BSA224S, Germany)

Reagent

1. Ethanol (QRëC®, New Zealand)
2. Diethyl ether (QRëC®, New Zealand)
3. Phenolphthalein
4. Potassium Hydroxide or Sodium Hydroxide 0.1 N

Procedures

1. Weigh zero point five gram of sample (4 decimal places) put in an Erlenmeyer flask
2. Mix solvent (Ethanol: Diethyl ether = 1: 1) and add 50 ml of the solvent mixture to the sample
3. Phenolphthalein drops into the solution
4. Titrate the solution with 0.1 N KOH until obtained pink solution for 30 seconds
5. Record the volume of 0.1 N KOH
6. Acid value is calculated from equation A.5

$$\text{Acid value (mg KOH/g of sample)} = \frac{V}{W} \times 5.61 \quad (\text{A.5})$$

V: Volume (mL) of 0.1 N KOH required for the sample titration

W: weight of sample (g)

A.6 Phytic acid (K-PHYT,11/15, Megazyme, Ireland)

Apparatus

1. Vortex mixer
2. Heat water bath
3. Microfuge
4. Spectrophotometer
5. Weighing machine (4 digits) (Sartorius, BSA224S, Germany)

Reagent

1. Color reagent : Ascobic acid 10 w/v in Sulphuric acid 1 M mix with Ammonium molybdate 5 w/v (1: 5)
2. Trichloroacetic acid 50 w/v

3. Hydrochloric acid 0.66 M

4. Sodium hydroxide 0.75 M

Procedures

1. Weigh accurately 1 g of sample into a 75 mL glass beaker. Add 20 mL of hydrochloric acid (0.66 M), cover the beaker with foil and stir vigorously for a minimum of 3 h at room temperature (preferably overnight for convenience)

2. Transfer 1 mL of extract to a 1.5 mL microfuge tube and centrifuge at 13,000 rpm for 10 minutes, then immediately transfer 0.5 mL of the resulting extract supernatant to a fresh 1.5 mL microfuge tube and neutralise by addition of 0.5 mL of sodium hydroxide solution (0.75 M). Use the neutralised sample extract in the enzymatic dephosphorylation reaction

3. Separate (a) free phosphorus (b) total phosphorus. By (b) add phytase enzyme and alkaline phosphatase enzymes for all phytic acid hydrolyze and converted to inorganic phosphate, while (a) not fill the enzyme

4. Incubate in a water bath set at 40 °C for 15 min. After 15 min, stop the reaction by addition of: trichloroacetic acid (50 % w/v) 0.30 mL both (a) and (b)

5. Centrifuge the terminated reaction at 13,000 rpm for 10 min. Do not mix the tube after centrifugation

6. Transfer 1 mL of solution to determined colorimetric determination

7. Mix by vortex and incubate in a water bath set at 40 °C for 1 h. After 1 h, mix by vortex and then transfer 1 mL to a semi-micro cuvette and read the absorbance at 655 nm (A_{655}) within 3 h

8. Apply the absorbance values between (a) and (b). To find the total phosphorus content and the phytic acid from the standard graph

A.7 Color measurement (Whiteness index)

Apparatus

1. Chroma meter (model CR-300 series, Konica Minolta, Tokyo, Japan)
2. Weighing machine (4 digits) (Sartorius, BSA224S, Germany)

Procedures

1. 1.5 gram of rice bran into the sample cup
2. Use the cover to prevent any external light
3. Color measurement by Hunter Color System L*, a*, and b*
4. Whiteness index could calculate from equation A.7

$$\text{Whiteness index (WI)} = 100 - [(100 - L)^2 + a^2 + b^2]^{0.5} \quad (\text{A.7})$$

A.8 Bulk density measurement (Caparino et. al. 2012)

Apparatus

1. Cylinder
2. Weighing machine (4 digits) (Sartorius, BSA224S, Germany)

Procedures

1. Approximately 5 gram of rice bran sample was freely poured into a 25 ml glass graduated cylinder (readable at 1 ml)
2. the samples were repeatedly tapped manually by lifting and dropping the cylinder under its own weight at a vertical distance of 14 ± 2 mm high until negligible difference in volume, whereas untapped will not lifting and dropping the cylinder
3. Observe given the mass M and the apparent (tapped) volume V of the powder and weigh of the sample in cylinder
4. Bulk density could calculate from equation A.8

$$\text{Bulk density (kg/m}^3\text{)} = \frac{M}{V} \quad (\text{A.8})$$

W: weight of sample (g)

V: volume (mL) of cylinder



APPENDIX B
CHEMICAL – PHYSICAL ANALYSIS

B.1 Chemical – Physical analysis



Table B.1 Lightness of mixed rice bran

Ratio of mixed rice bran (rice bran : parboiled rice bran)	Lightness (L*)									
	Lot 1	Lot 2	Lot 3	Lot 4	Lot 5	Lot 6	Lot 7	Lot 8	Lot 9	Lot 10
0:10	50.5±0.1 ^{EF}	51.4±0.0 ^{EE}	53.34±0.0 ^{7c}	56.3±0.1 ^{Aa}	55.8±0.0 ^{Ab}	51.4±0.3 ^{Ge}	52.5±0.2 ^{7d}	49.5±0.2 ^{Gg}	49.6±0.2 ^{7g}	52.3±0.0 ^{Ed}
1:9	50.6±0.3 ^{DEa}	51.4±0.0 ^{EF}	53.45±0.2 ^{7c}	55.8±0.3 ^{Aa}	55.5±0.1 ^{ABbb}	51.0±0.1 ^{Gg}	52.5±0.2 ^{7e}	49.7±0.0 ^{Gi}	49.7±0.1 ^{Fi}	52.6±0.0 ^{DEd}
2:8	50.6±0.5 ^{DEe}	51.5±0.0 ^{DEd}	53.79±0.2 ^{7b}	55.7±0.2 ^{Aa}	55.3±0.4 ^{Baa}	51.9±0.3 ^{EEf}	52.8±0.1 ^{EEc}	50.0±0.1 ^{FGf}	49.9±0.2 ^{EEff}	52.7±0.1 ^{DEc}
3:7	50.7±0.3 ^{DEe}	51.5±0.1 ^{DEd}	54.09±0.3 ^{DEb}	55.0±0.4 ^{Baa}	54.5±0.1 ^{Cab}	52.5±0.3 ^{Ec}	52.9±0.1 ^{DEc}	50.6±0.1 ^{EEe}	50.2±0.1 ^{DEe}	52.8±0.2 ^{DEc}
4:6	51.1±0.0 ^{DEde}	51.5±0.0 ^{DEd}	54.06±0.1 ^{DEa}	54.3±0.2 ^{Ca}	54.5±0.3 ^{Ca}	52.4±0.7 ^{Ec}	53.3±0.3 ^{Cb}	50.5±0.1 ^{EEe}	50.7±0.2 ^{DEe}	53.3±0.2 ^{Cb}
5:5	50.6±0.1 ^{DEd}	51.7±0.0 ^{DEc}	54.20±0.1 ^{DEa}	53.7±0.0 ^{DEab}	54.1±0.2 ^{Ca}	53.8±0.6 ^{DEab}	53.3±0.3 ^{DEb}	50.8±0.3 ^{DEd}	50.7±0.3 ^{DEd}	53.3±0.1 ^{Cb}
6:4	51.6±0.1 ^{DEde}	51.7±0.0 ^{DEc}	54.34±0.0 ^{DEa}	53.5±0.1 ^{DEbc}	54.1±0.1 ^{Ca}	53.3±0.0 ^{DEc}	53.7±0.1 ^{Bbb}	51.3±0.4 ^{DEef}	51.2±0.0 ^{DEf}	53.5±0.0 ^{DEbc}
7:3	51.9±0.0 ^{DEc}	51.7±0.0 ^{DEc}	54.63±0.0 ^{DEca}	53.6±0.3 ^{DEb}	53.5±0.0 ^{DEb}	53.8±0.0 ^{DEb}	53.7±0.0 ^{DEb}	51.5±0.1 ^{BCcd}	51.2±0.4 ^{ABd}	53.7±0.0 ^{DEbc}
8:2	52.7±0.1 ^{ABd}	51.8±0.0 ^{DEe}	54.67±0.2 ^{ABa}	53.1±0.2 ^{DEcd}	53.0±0.3 ^{DEcd}	54.4±0.1 ^{BCa}	53.8±0.2 ^{Bbb}	51.6±0.3 ^{BCe}	51.3±0.2 ^{ABe}	53.5±0.4 ^{DEbc}
9:1	53.0±1.0 ^{A,d}	51.8±0.0 ^{ABef}	54.86±0.0 ^{ABab}	52.4±0.2 ^{DEdef}	52.6±0.5 ^{DEde}	55.0±0.3 ^{ABa}	54.1±0.0 ^{DEbc}	52.0±0.0 ^{ABef}	51.7±0.2 ^{Af}	54.0±0.2 ^{ABc}
10:0	53.6±0.7 ^{A,d}	51.8±0.0 ^{Afg}	54.97±0.0 ^{Aab}	52.1±0.3 ^{DEfg}	52.8±0.0 ^{DEe}	55.6±0.0 ^{Aa}	54.5±0.1 ^{ABc}	52.4±0.3 ^{Aef}	51.6±0.1 ^{ABg}	54.3±0.1 ^{A,c}

Note: The values in the table are shown in mean ± SD from the 2 replicates.

A, B, C... Mean values in a column with different superscripts are different significantly (p≤0.05)

a, b, c... Mean values in a row with different superscripts are different significantly (p≤0.05)

Table B.2 Redness of mixed rice bran

Ratio of mixed rice bran (rice bran : parboiled rice bran)	Redness (a [†])									
	Lot 1	Lot 2	Lot 3	Lot 4	Lot 5	Lot 6	Lot 7	Lot 8	Lot 9	Lot 10
0:10	1.7±0.0 ^{A3}	1.8±0.0 ^{A3}	0.3±0.0 ^{A3}	0.1±0.0 ^{A3}	0.5±0.1 ^{A3}	0.7±0.0 ^{A3}	0.8±0.0 ^{A3}	0.1±0.0 ^{A3}	1.3±0.0 ^{A3}	0.4±0.0 ^{A3}
1:9	1.6±0.1 ^{B3}	1.6±0.0 ^{B3}	0.3±0.0 ^{A3}	0.1±0.0 ^{M3}	0.5±0.0 ^{B3}	0.6±0.0 ^{B3}	1.0±0.0 ^{A3}	0.1±0.0 ^{B3}	1.2±0.0 ^{B3}	0.4±0.0 ^{A3}
2:8	1.4±0.0 ^{C3}	1.6±0.0 ^{B3}	0.4±0.0 ^{B3}	0.4±0.0 ^{A3}	0.6±0.0 ^{A3}	0.8±0.1 ^{A3}	1.0±0.0 ^{A3}	0.1±0.0 ^{A3}	1.2±0.0 ^{A3}	0.5±0.0 ^{B3}
3:7	1.4±0.0 ^{C3}	1.6±0.0 ^{B3}	0.4±0.0 ^{B3}	0.5±0.0 ^{A3}	0.7±0.0 ^{B3}	0.9±0.0 ^{A3}	1.1±0.0 ^{B3}	0.4±0.0 ^{B3}	1.1±0.0 ^{B3}	0.5±0.0 ^{C3}
4:6	1.3±0.0 ^{C3}	1.3±0.0 ^{B3}	0.5±0.0 ^{A3}	0.9±0.0 ^{B3}	0.9±0.0 ^{B3}	1.1±0.0 ^{B3}	1.1±0.0 ^{B3}	0.5±0.0 ^{B3}	1.0±0.1 ^{B3}	0.5±0.0 ^{C3}
5:5	1.3±0.0 ^{C3}	1.4±0.0 ^{C3}	0.5±0.0 ^{A3}	0.9±0.0 ^{B3}	1.0±0.0 ^{C3}	1.0±0.0 ^{C3}	1.1±0.0 ^{A3}	0.6±0.0 ^{A3}	1.0±0.0 ^{A3}	0.5±0.0 ^{A3}
6:4	1.3±0.0 ^{A3}	1.3±0.0 ^{B3}	0.5±0.0 ^{B3}	1.0±0.0 ^{A3}	1.2±0.0 ^{B3}	1.1±0.1 ^{B3}	1.2±0.0 ^{B3}	0.7±0.0 ^{B3}	0.8±0.0 ^{B3}	0.5±0.0 ^{B3}
7:3	1.1±0.0 ^{B3}	1.2±0.0 ^{A3}	0.6±0.0 ^{B3}	1.1±0.0 ^{C3}	1.0±0.0 ^{C3}	1.3±0.0 ^{A3}	1.3±0.0 ^{A3}	0.8±0.0 ^{C3}	0.9±0.0 ^{A3}	0.5±0.0 ^{A3}
8:2	1.1±0.0 ^{B3}	1.2±0.0 ^{A3}	0.7±0.0 ^{A3}	1.3±0.0 ^{B3}	1.2±0.0 ^{B3}	1.3±0.0 ^{B3}	1.3±0.0 ^{B3}	1.0±0.0 ^{B3}	0.9±0.0 ^{B3}	0.6±0.0 ^{B3}
9:1	1.0±0.0 ^{A3}	1.1±0.0 ^{A3}	0.6±0.0 ^{B3}	1.5±0.0 ^{A3}	1.4±0.0 ^{A3}	1.3±0.0 ^{A3}	1.3±0.0 ^{A3}	1.0±0.0 ^{A3}	0.8±0.0 ^{A3}	0.6±0.0 ^{A3}
10:0	0.9±0.0 ^{A3}	0.9±0.0 ^{A3}	0.7±0.0 ^{A3}	1.4±0.0 ^{A3}	1.4±0.0 ^{A3}	1.3±0.0 ^{A3}	1.5±0.0 ^{A3}	1.2±0.0 ^{A3}	0.8±0.0 ^{A3}	0.6±0.0 ^{A3}

Note: The values in the table are shown in mean ± SD from the 2 replicates.

A, B, C... Mean values in a column with different superscripts are different significantly ($p \leq 0.05$)

a, b, c... Mean values in a row with different superscripts are different significantly ($p \leq 0.05$)

Table B.3 Yellowness of mixed rice bran

Ratio of mixed rice bran (rice bran : parboiled rice bran)	Yellowness (b ⁺)									
	Lot 1	Lot 2	Lot 3	Lot 4	Lot 5	Lot 6	Lot 7	Lot 8	Lot 9	Lot 10
0:10	14.8±0.1 ^{D,c}	15.4±0.0 ^{F,a}	14.2±0.0 ^{G,a}	10.8±0.0 ^{F,i}	14.0±0.1 ^{E,f}	13.6±0.0 ^{G,s}	14.3±0.0 ^{F,d}	11.8±0.0 ^{H,b}	15.0±0.0 ^{H,b}	14.2±0.0 ^{I,a}
1:9	14.8±0.0 ^{D,c}	15.4±0.0 ^{F,a}	14.4±0.0 ^{F,d}	12.0±0.0 ^{E,i}	14.1±0.0 ^{E,f}	13.2±0.2 ^{H,s}	14.2±0.0 ^{G,e,f}	12.2±0.0 ^{G,b}	15.1±0.0 ^{G,H,b}	14.2±0.0 ^{H,c}
2:8	14.8±0.0 ^{D,b}	15.4±0.0 ^{F,a}	14.5±0.0 ^{F,c}	12.2±0.3 ^{E,s}	14.1±0.0 ^{E,d}	13.8±0.2 ^{G,e}	14.4±0.0 ^{F,e,d}	12.9±0.2 ^{F,f}	15.1±0.0 ^{F,G,b}	14.2±0.0 ^{H,c,d}
3:7	14.9±0.0 ^{D,b}	15.4±0.0 ^{F,a}	14.8±0.1 ^{E,b}	12.1±0.2 ^{E,f}	14.1±0.1 ^{D,d}	14.1±0.2 ^{F,d}	14.5±0.0 ^{E,c}	13.8±0.2 ^{E,e}	15.1±0.0 ^{F,F,b}	14.3±0.0 ^{G,e,d}
4:6	15.0±0.0 ^{B,c,b}	15.4±0.0 ^{F,a}	14.8±0.1 ^{E,c}	13.2±0.1 ^{D,f}	14.2±0.0 ^{D,e}	14.1±0.1 ^{F,F,e}	14.5±0.0 ^{E,d}	13.1±0.3 ^{F,f}	15.1±0.0 ^{D,E,b}	14.3±0.0 ^{F,d,e}
5:5	15.0±0.0 ^{B,c,b}	15.4±0.0 ^{F,a}	15.1±0.0 ^{D,b}	13.3±0.2 ^{D,f}	14.3±0.0 ^{B,C,d}	14.3±0.1 ^{D,E,f,d}	14.7±0.1 ^{D,c}	13.7±0.0 ^{E,e}	15.1±0.0 ^{D,b}	14.4±0.0 ^{F,d}
6:4	15.1±0.0 ^{A,B,b,c}	15.5±0.0 ^{B,a}	15.2±0.0 ^{C,a,b}	14.0±0.3 ^{C,e}	14.3±0.0 ^{B,C,d,e}	14.4±0.1 ^{D,E,d}	14.9±0.0 ^{C,c}	14.4±0.1 ^{D,d}	15.2±0.0 ^{C,b,c}	14.4±0.0 ^{D,d}
7:3	15.1±0.1 ^{A,B,a,b,c}	15.5±0.0 ^{B,C,a}	15.2±0.0 ^{C,a,b}	13.9±0.2 ^{C,f}	14.3±0.0 ^{B,e}	14.9±0.1 ^{B,b,c}	15.0±0.1 ^{C,b,c}	14.8±0.4 ^{CD,c,d}	15.2±0.0 ^{C,a,b}	14.5±0.0 ^{G,d,e}
8:2	15.1±0.0 ^{A,B,b}	15.5±0.0 ^{A,a}	15.5±0.0 ^{A,B,a}	14.6±0.1 ^{A,B,d}	14.3±0.0 ^{B,e}	14.7±0.1 ^{B,C,d}	15.0±0.0 ^{C,c}	14.8±0.1 ^{B,C,c}	15.2±0.0 ^{C,b}	14.4±0.0 ^{D,e}
9:1	15.1±0.1 ^{A,B,b}	15.5±0.0 ^{A,a}	15.4±0.0 ^{B,a,b}	14.3±0.4 ^{B,C,e}	14.3±0.0 ^{B,c}	14.5±0.0 ^{CD,c}	15.1±0.0 ^{B,b}	15.3±0.0 ^{A,B,a,b}	15.2±0.0 ^{A,a,b}	14.5±0.0 ^{B,c}
10:0	15.2±0.1 ^{A,b}	15.5±0.0 ^{A,a}	15.5±0.0 ^{A,a}	15.0±0.1 ^{A,c}	14.4±0.0 ^{A,e}	15.3±0.1 ^{A,b}	15.5±0.0 ^{A,a}	15.5±0.0 ^{A,a}	15.3±0.0 ^{A,b}	14.6±0.0 ^{A,d}

Note: The values in the table are shown in mean ± SD from the 2 replicates.

A, B, C... Mean values in a column with different superscripts are different significantly ($p \leq 0.05$)

a, b, c... Mean values in a row with different superscripts are different significantly ($p \leq 0.05$)

□

VITA

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