ASSESSMENT OF THE COLOUR DEVELOPMENT OF SHRIMP PASTE

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Abstract

This work is to investigate the quality improvement of Myanmar shrimp paste and to determine the most suitable colorant for shrimp paste from natural sources as well as synthetic products. Natural food colorant, anthocyanin was extracted from purple corn (Tatkon Township, Mandalay Region). Both purple corn-cob and kernel were used for natural colorant. Effects of temperature, extraction time and solvents on the yield percent of anthocyanin were studied. Prepared anthocyanin compounds (cob and kernel) were identified by Fourier Transformed Infrared (FT-IR) Spectroscopy and Ultraviolet Visible (UV/VIS) Spectroscopy. From UV spectrum, corn-cob and kernel extracts are anthocyanidins and anthocyanins. The FT-IR spectrum of extracted compound indicated for hydroxyl group, methoxy group, carbonyl group and secondary alcohol. For synthetic food colorant, secondary permitted food colorants are used to prepare the tertiary color for shrimp paste. Different permitted food dyes were mixed in different ratios to prepare color, suitable for shrimp paste. The color stability of the shrimp paste produced by using natural or synthetic colorants was assessed during one year storage period. Color No.359 was almost identical to natural shrimp paste colour of the better quality and popular one. Synthetic food colorant mixture (Color No.359) was observed to be more attractive to the consumers than anthocyanin extract, although both colorants lasted for 8 months. This research work will support the need of shrimp paste dealers to attract customers.

Keywords: Anthocyanin, shrimp paste, natural or synthetic colorants, purple corn.

Introduction

Myanma Ngapi (Shrimp Paste) is an important source of low cost dietary protein. Ngapi means “Pressed fish”, named by its traditional processing technique. It can be classified into two groups: nga-ngapi (for fish) and hmyin- or seinsa ngapi (for shrimp). Shrimp paste or shrimp sauce, is a common ingredient used in Southeast Asian and Southern Chinese cuisine (Myint Myint Sein, 1999).

Ngapi in Myanmar is known as terasi in Indonesia, kapi in Thai, Khmer and Lao, belacan (also spelled *belachan*, *blachang*) in Malay, mắm ruốc, mắm tép and mắm tôm in Vietnam, bagoong alamang in Philippines, haam ha/ha jeung and hom ha/hae ko in Chinese. Shrimp paste can be used in most meals in Myanmar, Laos, Thailand, Malaysia, Singapore, Indonesia and the Philippines. It is often an ingredient in dipping sauce for fish or vegetables. Its colour ranges from light pink to dark brown.

Shrimp paste varies in forms, some are almost liquid or liquid, some are smooth or other consistencies of paste and some are used as air-dried block forms or powder, depending on the region of the production, or the end product and its uses. At the end of the manufacturing process, shrimp paste is usually dried to reduce the moisture content and to produce a semi-solid product. The paste of the product needs only a little amount of salt and it has a strong umami taste. The drying process and reduction of moisture also increase the shelf life and flavour intensity of the product; complete drying would prevent rancidity of the product (Pérez-Villarreal. B. and Pozo R.,1990).

The color of shrimp varies from yellowish white to pinkish according to the season. During the hot season, the shrimp is pinkish and shrimp paste produced from this kind of shrimp is purple pink and its taste is more delicious than the rest. Most shrimp paste consumers prefer such kind of purple pink shrimp paste. The role of natural or synthetic colorant becomes important for yellowish white shrimp to meet customers’ demand.

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Materials and Method

Materials

Zea mays L. purple corn, purchased from Tatkon Township, Naypyidaw Region and *Mysis diluviana* shrimp from Naukmee Village, Pyapon Township were used for this research. The certified colors of commercial grade, manufactured by the Ram Ratna International Co., Ltd. Ram Ratna House, Victoria Mill Compound, PB Marg, Worli, Mumbai 400 013 (India) were used for synthetic color.

Extraction of Anthocyanin from Purple Corn-Cob and Kernel

The purple corns were threshed and corn-cobs were air-dried for five days. Dried purple corn-cobs were crushed before milling in a grinder. The milled corn-cobs were screened and then stored in a desiccator for the extraction of anthocyanin. Dry corn-cob granules (50 g) were mixed with 30 mL of 95 % ethanol in a beaker, and warmed in a water bath at 40 °C for 5 h. The mixture was filtered and the filtrate was put into a round-bottomed flask and then distilled. During distillation, the pressure of the solution was reduced to about 140 mmHg by using a vacuum pump. At this pressure, the boiling point of ethanol was reduced from 78.8 °C to 60 °C and ethanol was quickly evaporated. The ethanol was collected as condensate and anthocyanin was left in the flask with a small amount of ethanol. After air drying, the dried anthocyanin was weighed and stored for further analysis.

Purple corns were threshed and corn kernels were air-dried for five days and stored in a desiccator for the extraction of Anthocyanin 1 (c). The extractions were conducted as previously described and the results are shown in Tables 2, 3 and 4.

(a) **(b)** **(c)**

Figure 1: (a) Purple Corn
(b) Purple Corn Plant
(c) Purple Corn Maize

(a) **(b)**

Figure 2: (a) Purple Corn Cob (b) Extracted Anthocyanin from Purple Corn Cob

Effect of Extraction Temperature, Time and Solvents on the Yield of Anthocyanin

The extractions were conducted at different extraction temperature of 30 °C, 40 °C, 50 °C, 60 °C and 70 °C, with different extraction times of 3 h, 4 h, 5 h and 6 h at 60˚C, using different solvents such as distilled water, 95 % ethanol and ethanol acidified with 1 % citric acid. The results are shown in Tables 1, 2 and 3.

Table 1: Effect of Temperature on Extraction of Anthocyanin from Purple Corn

Sr. No	Extraction Temperature (°C)	Weight of Corn Cob/Kernel (g)	Anthocyanin Yield	
			Cob % (w/w)	Kernel % (w/w)
1.	30	50	0.5682	0.0825
2.	40	50	0.7218	0.0922
3.	50	50	0.8064	0.1143
4.	60*	50	0.9658	0.1592
5.	70	50	0.9826	0.1659

*60˚C is the most suitable extraction temperature.

Table 2: Effect of Extraction Time on Extraction of Anthocyanin from Purple Corn

Sr. No	Extraction Time (h)	Weight of Corn Cob/Kernel (g)	Anthocyanin Yield	
			Cob % (w/w)	Kernel % (w/w)
1.	3	50	0.7152	0.0967
2.	4	50	0.7966	0.1348
3.	5*	50	0.9658	0.1582
4.	6	50	0.9686	0.1631

*5 hr is the most suitable extraction time

Table 3: Effect of Solvents on Extraction of Anthocyanin from Corn Cob and Kernel

Sr. No	Solvents	Weight of Corn Cob/Kernel (g)	Anthocyanin Yield	
			Cob % (w/w)	Kernel % (w/w)
1.	Water	50	0.6532	0.0654
2.	95 % ethanol	50	0.9658	0.1582
3.	ethanol acidified with 1% citric acid *	50	0.9972	0.1865

* ethanol acidified with 1% citric acid is the most suitable solvent for extraction of anthocyanin.

Analysis of Extracted Anthocyanin

Identification of Anthocyanin by Ultraviolet Spectroscopy

Anthocyanin exhibits very high extinction coefficients between 270 - 280 nm and 465 - 560 nm wavelengths. Prepared Anthocyanins were identified by Ultraviolet Visible (UV/VIS) Spectroscopy [Figures 5 and 6 and Table 6].

Identification of Anthocyanin by Infrared (IR) Spectroscopy

Prepared anthocyanins (cob and kernel) were identified by Infrared (IR) Spectroscopy Figures 7 and 8, Tables 7 and 8.

Application of Prepared Anthocyanin on Shrimp Paste

About 100 g of shrimp from Pyapon Township was washed and weighed. Prepared natural food colorant, anthocyanin 0.01 % (w/w) was added to the shrimp before mixing with salt to prepare shrimp paste.

Preparation of Mixed Synthetic Food Colourants for Shrimp Paste

The most suitable food colorant for shrimp paste was produced by mixing 50 % of grape purple, 25 % of raspberry red and 25 % of bright red colors, known as color number 359.

Table 4: Properties of Certified Colors for Food Products

Sr. No.	Colour Name	Commercial Name	C.I. No*	Colour	pH	Density
1.	Bright red (E124)	Ponceau 4R	16255	Bright red colour	6-8	0.8
2.	Grape purple	(Brilliant Blue +Ponceau 4R) Blended Colour	75300 + 16255	Purple	6.5-8	0.7
3.	Raspberry red (E122)	Carmoisine	14720	red	6-8	0.7
4.	Strawberry red	Blended Colour	-	Strawberry	6-8	0.7

* C.I. No = Colour Index Number

Application of Synthetic Food Colourants on Shrimp Paste

About 100 g of shrimp from Pyapon Township, Naukmee village was washed, weighed and mixed with synthetic colorants. Each synthetic food colorant of 0.01 % (w/w) and 0.005 % (w/w) of prepared synthetic colorants mixture were used for coloring shrimp to produce shrimp paste.

(a) **(b)** **(c)**

Figure 3. (a) Mixed Permitted Color Solution for Shrimp Paste
(b) Suitable Permitted Color for Shrimp Paste
(c) Shrimp Paste with Certified Synthetic Colour

Results and Discussion

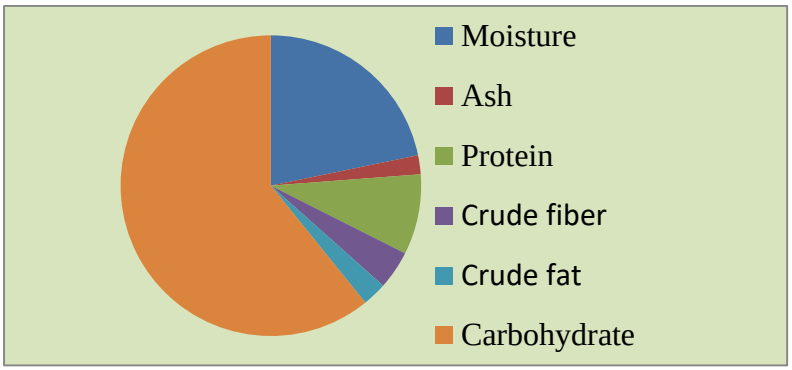

Figure 4: Nutritional facts of purple corn kernel

Nutritional values such as protein, ash, moisture content, crude fiber and crude fat (ether extract) of purple corn kernel were analyzed and results are shown in Figure (4).

The effects of extraction temperature, extraction time and nature of solvents on the yield of anthocyanin from purple corn cob and kernel are described in Tables (1) to (3) and. The most suitable extraction temperature was 60°C, extraction time was 5 hours and ethanol acidified with 1% citric acid was the best solvent for maximum yield of anthocyanin in this research work.

The UV spectrum of extracted anthocyanin from purple corn cob [Figure (5) and Table (6)] showed the flavonoid band I and band II in methanol at 531.2 nm and 278 nm. These absorption bands match with that of anthocyanins and anthocyanidins (Band I 465-560 nm; Band II 270-280 nm) (Markhan, 1982). Therefore, from UV spectrum, it can be concluded that corn cob extracts are anthocyanidins and anthocyanins.

The UV spectrum of extracted anthocyanin from purple corn kernel [Figure 6 and Table 6] showed the flavonoid band I and band II in methanol at 526.2 nm and 284.4 nm. These absorption bands match with that of anthocyanins and anthocyanidins (Band I 465-560 nm; Band II 270-280 nm) (Markhan, 1982). So, from UV spectrum, it can be said that corn kernel extracts are anthocyanidins and anthocyanins.

The FT-IR spectrum of extracted anthocyanin [Figure 7 and Table 7] indicated the stretching -OH group at 3392 cm^{-1} for the hydroxyl group. The absorption band at 2926 cm^{-1} due to CH_3, indicated the methoxy group. The absorption band at 1726 cm^{-1} is due to C=O, indicated

the carbonyl group. The stretching for C=C unsaturated cyclic ring is at 1631, 1514 cm^{-1}. The in-plane bending - OH at 1383, 1332 cm^{-1} was the hydroxyl group. The stretching C-O at 1267, 1211, 1076 and 1055 cm^{-1} is stretching C-O. The absorption band at 611 cm^{-1} detected the out-of-plane bending -CH.

The FT-IR spectrum of extracted compound [Figure 8 and Table 8] indicated the stretching-OH group at 3383 cm^{-1} for hydroxyl group. The absorption band at 2926, 2852cm^{-1} due to CH_3 indicated the methoxy group. The absorption band at 1718 cm^{-1} due to C=O, indicated the carbonyl group. The stretching for C=C unsaturated cyclic ring is at 1641 cm^{-1}. The in-plane bending -OH at 1408 cm^{-1} directed the hydroxyl group. The stretching C-O at 1269, 1078 and 1045 cm^{-1} is arc -OH and secondary alcohol. The absorption band at 570 cm^{-1} detected the out-of-plane bending -CH.

Shrimp paste prepared with natural and synthetic colorant was stored for one year and their color stability was assessed [Table 6]. Anthocyanin colorant gave delicate pink color although the color itself was bright. The shrimp paste using synthetic colorant was brighter than the common shrimp paste color. It is found that their color intensity was not suitable for shrimp paste. The color of shrimp paste using the synthetic color retained up to six months. There is neither color irregulation nor color changes of shrimp paste during six months. After six months, the shrimp paste with the synthetic pink color gradually changed into brown.

Shrimp paste sample with mixed synthetic food colourant known as colour number 359 (colourant mixture) is shown in Figure [3 (c)]. Most of the shrimp paste processors concluded that color No.359 is almost identical to the color of natural shrimp paste of better quality and popular one. Synthetic food colorant mixture (Color No.359) was observed to be more attractive to consumers than anthocyanin, although both colorants lasted for 8 months. These certified colorants are the ones that FDA Myanmar permitted for food preparation. These colors have no adverse effect on the human health. This research work gave useful information to improve the quality of Myanmar shrimp paste. This food permitted synthetic color from Ram Ratna International Co., Ltd. solved the problems for shrimp paste. This colour can be used to substitute the hazardous chemical dyes that were previously used in shrimp paste. This work also participates correct usage to produce the marine-based foods.

Table 5: Effect of Natural and Synthetic Color on Shrimp Paste

Sr. No.	Name of Colourant	Type of Colourant	Amount of Colourant % (w/w)	Colour of Shrimp Paste	Intensity of Colour
1.	Anthocyanin	Natural	0.01	Pinkish brown	Last for 8 months
2.	Raspberry red	Synthetic	0.01	Deep red	Last for 8 months
3.	Bright red	Synthetic	0.01	Bright red	Last for 8 months
4.	Strawberry red	Synthetic	0.01	Pinkish red	Last for 8 months
5.	Mixed colour suitable for shrimp paste	Synthetic	0.005	Purple pink	Last for 6 months

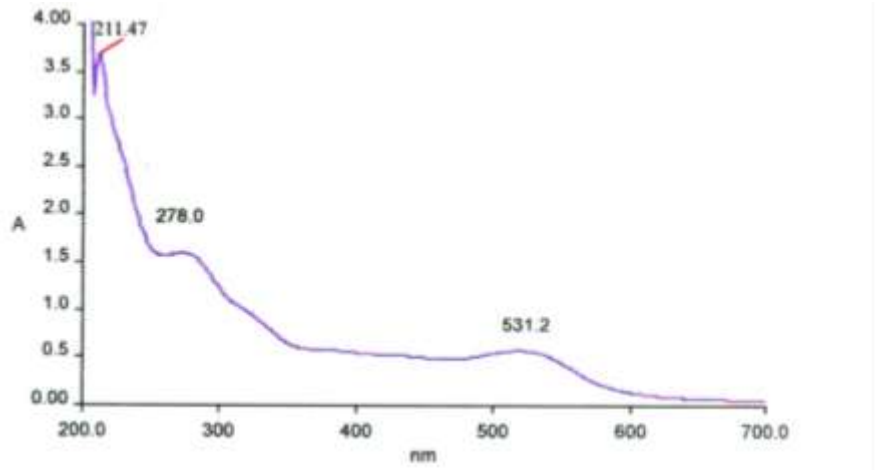

Figure 5: UV Spectrum of Extracted Anthocyanin (Corn Cob)

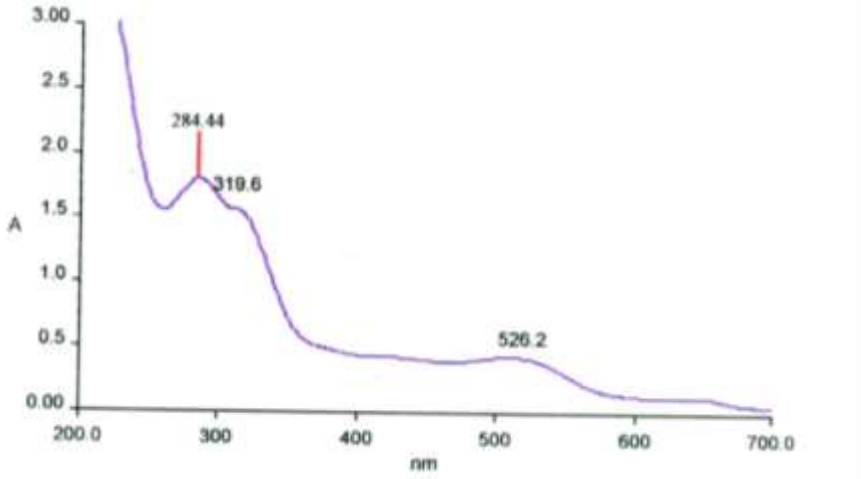

Figure 6: UV Spectrum of Extracted Anthocyanin (Corn Kernel)

Table 6: UV/VIS Data of Extracted Anthocyanin (Corn Cob & Corn Kernel)

Wave Number (nm)		**Assignment**	**Remarks**
Observed	**Literature***		
278	270-280	Band Π ππ*	Flavonoids compounds are anthocyanins and anthocyanidins (Conjugated double bonds are present)
531.2	465-560	Band I n-π*	
284.4	270-280	Band Π π -π*	Flavonoids compounds are anthocyanins and anthocyanidins (Conjugated double bonds are present)
526.2	465-560	Band I n-π*	

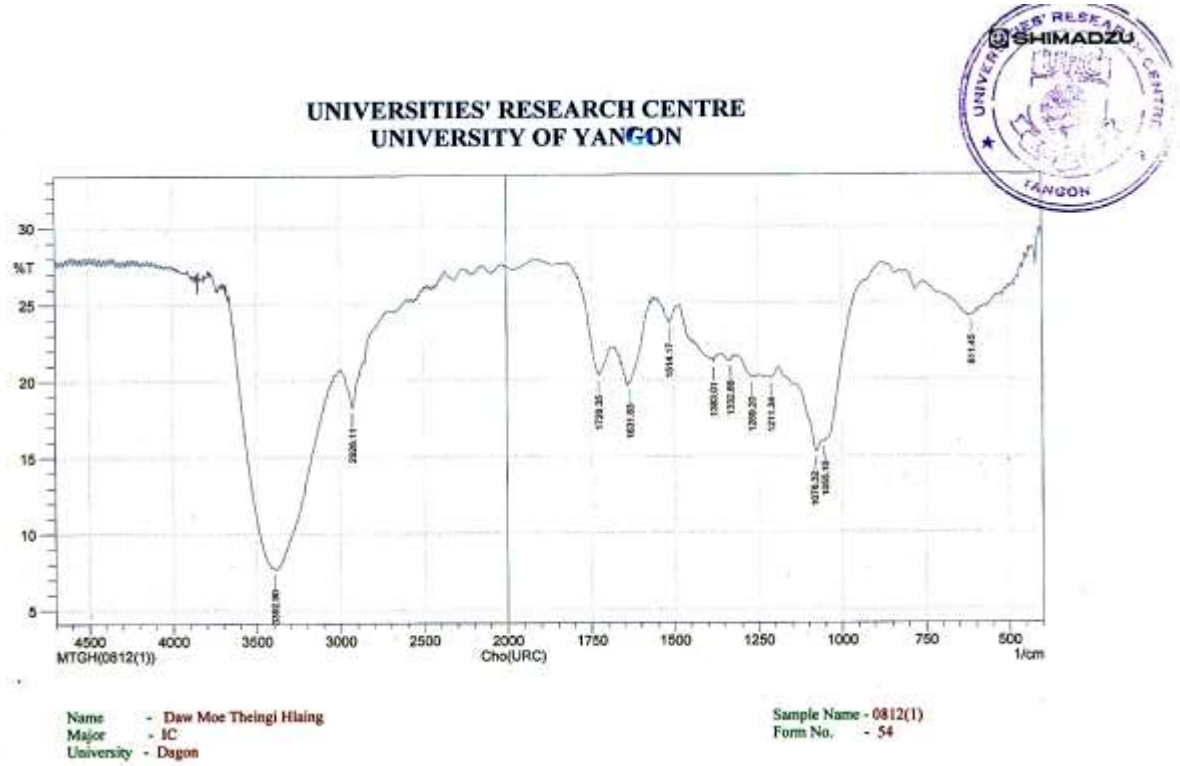

Figure 7: FT-IR Spectrum of Extracted Anthocyanin (Corn Cob)

Table 7: FT-IR Data of Extracted Anthocyanin (Corn Cob)

No.	Wave number, cm^{-1}		Functional Group	
	Observed	Literature*		
1.	3392	3550-3300	$\upsilon_{=OH}$	OH, stretching for alcohol or phenolic compound
2.	2926	~2800	$\upsilon_{=CH3}$	stretching vibration of methoxy group
3.	1726	~1700	$\upsilon_{C=O}$	stretching vibration of carbonyl group
4.	1631,1514	~1600	$\upsilon_{C=C}$	C = C stretching for unsaturated cyclic ring
5.	1383,1332	~1400	δ_{sCH}	C-H symmetric bending vibration
6.	1269,1211 1076,1055	1200-1000	$\upsilon_{c=O}$	stretching vibration of CO group
7.	611	900-450	δ_{CH}	C-H out of plane bending vibration

* Mohan 2000

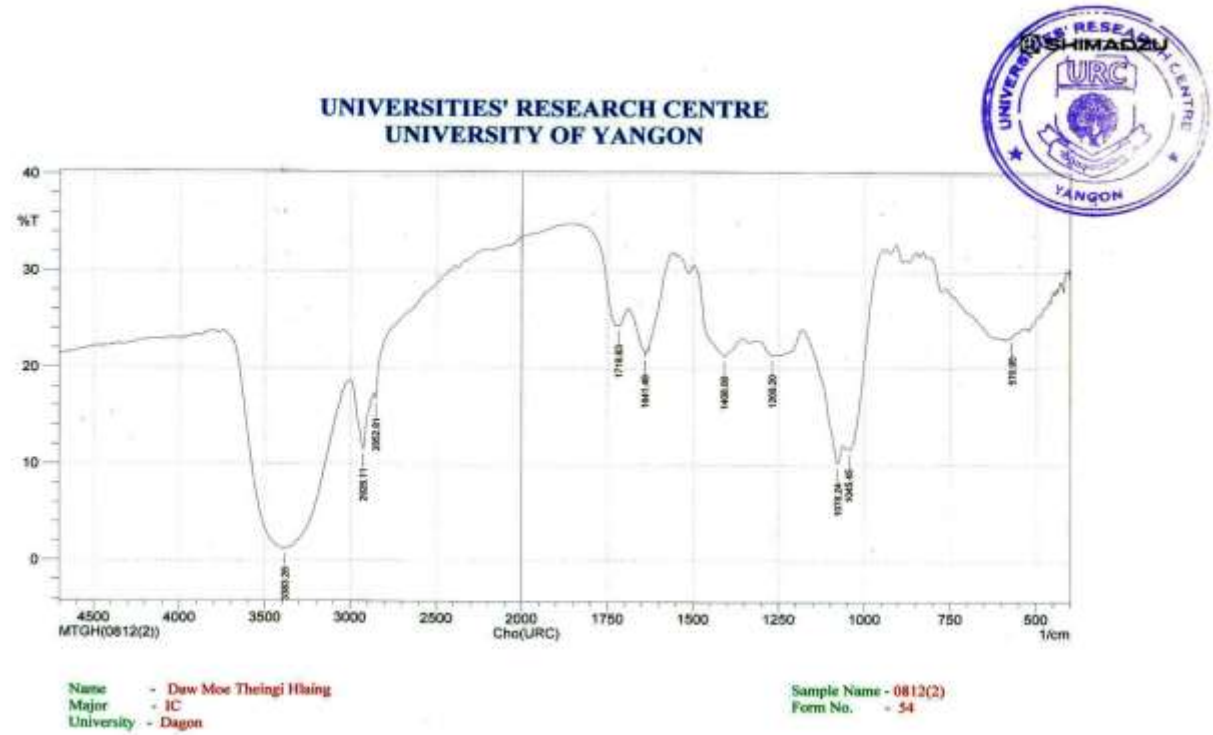

Figure 8: FT-IR Spectrum of Extracted Anthocyanin (Corn Kernel)

Table 8: FT-IR Data of Extracted Anthocyanin (Corn Kernel)

No.	Wave N		Functional group	
	Observed	Literature*		
1.	3383	3550-3300	$\upsilon_{=OH}$	OH, stretching for alcohol or phenolic compound
2.	2926,2852	~2800	$\upsilon_{=CH3}$	stretching vibration of methoxy group
3.	1718	~1700	$\upsilon_{C=O}$	stretching vibration of carbonyl group
4.	1641	~1600	$\upsilon_{C=C}$	C = C stretching for unsaturated cyclic ring
5.	1408	~1400	δ_{sCH}	C-H symmetric bending vibration
6.	1269,1078,1045	1200-1000	$\upsilon_{c=O}$	stretching vibration of CO group
7.	570	900-450	δ_{CH}	C-H out of plane bending vibration

* Mohan 2000

Conclusion

The IR spectra of extracted anthocyanin samples show the possible functional groups for characteristic absorption peaks of IR spectra. Based on these results, it can be concluded that the samples contain anthocyanin. Anthocyanins from purple corn are water-soluble pigments that may appear red, purple, or blue colours depending on pH.

The valuable anthocyanin extract can be used as natural colorant for shrimp paste. The optimum conditions for the extraction of anthocyanin from purple corn are, extraction temperature 60 °C and extraction time 5 h to give a yield of 0.9658 % w/w. The solvent which should be used is ethanol acidified with 1 % citric acid.

On the other hand, extracted anthocyanin gives light color on shrimp paste and the resultant color was not satisfactory for shrimp paste processors. The synthetic mixed colorant, color number 359 gave the most suitable color on shrimp paste. Both of their stabilities overcome the need of shrimp paste dealer.

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References

Faithong N., Benjakul S., Phatcharat S., Binsan W, 2010, (133-140) Chemical composition and antioxidative activity of Thai traditional fermented shrimp and krill products. Food Chemistry 119 (133–140)

FDA, 2001, Fish and Fishery Products Hazards and Controls Guidance, Scombrotoxin (Histamine Formation),3rd Edition

Hajeb. P, Jinap S.,2013, Umami taste components and their sources in Asian foods. Crit Rev Food Sci 8:1102–1115

Hajeb and Jinap, J Nutr Food Sci., 2012, Fermented Shrimp Products as Source of Umami in Southeast Asia

Haras, Alina, OvidiuToma, 2007, Determination of total proteins in some products

Harborne, J. B, 1973, Phytochemical Methods: A Guide to Modern Technique of Plant Analysis, Chapman and Hall, London

HPLC Basics, Fundamentals of Liquid Chromatography (HPLC), Courtesy of Agilent Technologies, Inc

Ishige N.,1993, Cultural aspects of fermented fish products in Asia. Fish fermentation technology, United Nations University Press

Malle and Poumeyrol, M.1989. A new chemical criterion for the quality control of fish; Trimethyl amine/Total volatile basic Nitrogen (%). Journal of food protection, 52(6):419-423

Markhan, K R. 1982, Techniques of Flavonoid Identification, Academic Press Inc., London New York

Mohan. J, 2000, Organic Spectroscopy: Principles and Applications, Narosa Publishing House, New Delhi

Myint Myint Sein, 1999, The mode of fermentation of shrimp paste, Chemistry, Ph D dissertation, Yangon University

Ogunwande, Dr. I.A., 2011, Colour Chemistry and Technology

Orella. C.J. and D.J. Kirwan, 1989, The Solubility of Amino Acids in Mixtures of Water and Aliphatic Alcohol Peralta E.M. et al, 2008, Food Chemistry 111,72-77

Pérez-Villarreal B. and Pozo R., 1990, Chemical composition and ice spoilage of albacore *(Thunnus alalunga) Journal of Food Science,* 55 678-682

Yoshida Y.1998, Umami taste and traditional seasonings. Food Rev Int 14: 213 246

http://purple-corn.html {Accessed August 2012}

www.purple-corn- drink html {Accessed August 2012}

Anthocyanin.htm {Accessed August 2012